



TOWARDS TOKYO 2020: WHAT WILL CONTRIBUTE TO OPTIMAL OLYMPIC ATHLETE PERFORMANCE?

EDITED BY: Toby Mündel, Glen Davison, Hideaki Soya, Narihiko Kondo
and Matthew J. Barnes

PUBLISHED IN: Frontiers in Physiology and Frontiers in Psychology



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ISSN 1664-8714

ISBN 978-2-88963-885-7

DOI 10.3389/978-2-88963-885-7

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TOWARDS TOKYO 2020: WHAT WILL CONTRIBUTE TO OPTIMAL OLYMPIC ATHLETE PERFORMANCE?

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Citation: Mündel, T., Davison, G., Soya, H., Kondo, N., Barnes, M. J., eds. (2020). Towards Tokyo 2020: What Will Contribute to Optimal Olympic Athlete Performance?. Lausanne: Frontiers Media SA. doi: 10.3389/978-2-88963-885-7

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Discovery of Causal Paths in Cardiorespiratory Parameters: A Time-Independent Approach in Elite Athletes

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OPEN ACCESS

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 09 July 2018

Accepted: 25 September 2018

Published: 30 October 2018

Citation:

Młyńczak M and Kryštofiak H (2018)
Discovery of Causal Paths in
Cardiorespiratory Parameters:
A Time-Independent Approach in Elite
Athletes. *Front. Physiol.* 9:1455.
doi: 10.3389/fphys.2018.01455

Training of elite athletes requires regular physiological and medical monitoring to plan the schedule, intensity and volume of training, and subsequent recovery. In sports medicine, ECG-based analyses are well-established. However, they rarely consider the correspondence of respiratory and cardiac activity. Given such mutual influence, we hypothesize that athlete monitoring might be developed with causal inference and that detailed, time-related techniques should be preceded by a more general, time-independent approach that considers the whole group of participants and parameters describing whole signals. The aim of this study was to discover general causal paths among cardiac and respiratory variables in elite athletes in two body positions (supine and standing), at rest. ECG and impedance pneumography signals were obtained from 100 elite athletes. The mean heart rate, the root-mean-square difference of successive RR intervals (RMSSD), its natural logarithm (lnRMSSD), the mean respiratory rate (RR), the breathing activity coefficients, and the resulting breathing regularity (BR) were estimated. Several causal discovery frameworks were applied, comprising Generalized Correlations (GC), Causal Additive Modeling (CAM), Fast Greedy Equivalence Search (FGES), Greedy Fast Causal Inference (GFCI), and two score-based Bayesian network learning algorithms: Hill-Climbing (HC) and Tabu Search. The discovery of cardiorespiratory paths appears ambiguous. The main, still mild, rules best supported by data are: for supine - tidal volume causes heart activity variation, which causes average heart activity, which causes respiratory timing; and for standing - normalized respiratory activity variation causes average heart activity. The presented approach allows data-driven and time-independent analysis of elite athletes as a particular population, without considering prior knowledge. However, the results seem to be consistent with the medical background. Causality inference is an interesting mathematical approach to the analysis of biological responses, which are complex. One can use it to profile athletes and plan appropriate training. In the next step, we plan to expand the study using time-related causality analyses.

Keywords: athlete training adaptation biomarker, cardiac function, tidal volume, cardiorespiratory causality, elite athletes

1. INTRODUCTION

Elite athletes require regular physiological and medical evaluation and monitoring for proper planning of the schedule, intensity, and volume of training (Meeusen et al., 2013). Therefore, exercise scientists and sports physicians seek convenient biomarkers to evaluate the state of an athlete's body during training to monitor homeostasis, maximize effect, and avoid over-training (Wiewelhove et al., 2015).

Some of the most commonly used parameters are related to cardiac function. Heart rate monitoring is popular in sport and recreational activity, and widely used thanks to easy access to sophisticated tools, enabling beat-by-beat registration of electrocardiographic (ECG) signals and evaluation of heart rate variability (HRV) (Buchheit, 2014; Schmitt et al., 2015; Bellenger et al., 2016; Duking et al., 2016; Giles et al., 2016; Plews et al., 2017).

Methodical acquisition of RR intervals, performed during different training periods, provides a chance to discern the proper course of HRV changes under the influence of exercise training, and possibly to recognize anomalous patterns indicating poor post-exercise recovery, sustained fatigue, impaired adaptation, and development of over-training syndrome.

Despite wide access, practical application of HRV parameters in sports training monitoring remains limited. The seemingly simple phenomenon, related to autonomic nervous system activity, defies simple evaluation, because of many modifying factors. An additional problem is the selection of optimal HRV parameters. There is no clear consensus as to which are best in training response evaluation. Is a single parameter enough, or will a set be more effective? Should more advanced mathematical methods be used for optimal modeling? There is growing interest in this field and recent studies have identified new directions (Sala et al., 2016, 2017).

Because there is a complex relationship between heart rate and breathing, taking breathing activity into account seems relevant and necessary for proper analysis. (Grossman and Taylor, 2007; Gasior et al., 2016; Sobiech et al., 2017). The influence of inspiration and expiration is usually apparent in resting ECG as sinus respiratory arrhythmia (Larsen et al., 2010; Shaffer et al., 2014; McCraty and Shaffer, 2015). Bidirectional neural relationships between cardiac functioning and the respiratory processes have previously been presented. The cardiorespiratory coupling effect, where heartbeats seem to coincide with specific respiratory phases, has been tested recently (Penzel et al., 2016; Sobiech et al., 2017). The possible physiological mechanisms behind it appear to include increased sympathetic nervous activity, as well as changes in arterial blood pressure. The effect of baroreflex, with baroreceptors playing a crucial role in the adjustment of neural responses, was also extensively described (Reyes del Paso et al., 2013).

The studies present concerns about whether respiratory control should be conducted in the HRV analysis. For example, Saboul et al. (2013) found that the RMSSD index is uninfluenced by respiratory patterns, for both spontaneous and controlled breathing. Nevertheless, the relations are more evident when different mathematical analyses are performed.

Therefore, the identification of an optimal mathematical method has an essential role in the daily practice. Usually, in sports science research methodology, a parameter's value is a dependent variable of exercise stimuli (top-down approach). However, in practical applications, the strategy can be reversed, with the optimal parameter as the one which best indicates and describes adaptation status (bottom-up approach). The relation between perception and performance outcomes can be "correlated" with the results of the objective analysis. For instance, an over-training syndrome is a major issue that negatively affects an athlete's performance, but it is not always subjectively felt in the same way.

In that context, one should consider evaluating cross-dependencies or even causalities in recorded signals and calculated parameters. Causality is then the way to describe the relationships, also specifying the direction and the structure. Causal relations can be established as a directed acyclic graph (DAG), the type of Bayesian network, in which each node can represent a parameter, and each directed link has a probability measure. Also, the graph may be rewritten into structural equation models (SEM) to get coefficients, which express the strength of connections. For such DAGs, the do-calculus rules theorem was introduced to analyze the effects of interventions (Pearl, 1995).

Other frameworks to analyze different types of causalities in physiological signals have been also proposed (Javorka et al., 2016; Müller et al., 2016; Penzel et al., 2016). Non-linear approaches, assuming cardiorespiratory interactions, were developed (Jamsek et al., 2004; Lopes et al., 2011). Relatively newly recognized phenomena—phase synchronization between heartbeats and ventilatory signal, inverse respiratory sinus arrhythmia—have also been used (Bartsch et al., 2015; Kuhnhold et al., 2017; Mazzucco et al., 2017). Information domain applications for more than three signals were presented (Wejer et al., 2017). Granger-based causality was employed (Porta et al., 2017), and also tested along with coherence measure and cross-sample entropy (Radovanović et al., 2018). Cardiorespiratory coordination, proposed by Moser et al. (1995), defines the mutual influence of the onsets of cardiac and respiratory cycles on each other. Various methods were proposed to analyze this phenomenon (Riedl et al., 2014; Sobiech et al., 2017; Valenza et al., 2018).

We hypothesize, that the main athletic rationale for causal path discovery is to profile athletes within a new causal domain, plan training modifications—considered to be interventions done to found cause variables (Pearl, 2010), and track changes in objective cardiorespiratory responses. However, the methods mentioned in the previous paragraph are specifically intended to describe the systems' mutual temporal activity, not to propose directly the possible changes to training and causal-related parameters. We think that such approaches should be preceded by a more general one, which takes into account the parameters describing the entire segment of data, and tries to search for the structure of directional relationships.

Therefore, in this paper, we seek general time-independent causal paths between basic cardiac and respiratory variables in

TABLE 1 | The information of the set of participants evaluated after excluding those with too much signal distortion.

Group	Sport type	N		Body mass			Height		
		Female	Male	Min	Mean	Max	Min	Mean	Max
B	IB	4	21	61.0	82.6	104.2	170	193.2	208
	IIB	7	2	55.0	64.8	97.7	167	174.7	193
	IIIB	4	8	53.2	79.7	151.0	158	174.3	197
C	IC	1	4	55.1	71.7	85.2	169	176.0	190
	IIC	12	25	49.1	80.7	115.0	162	185.2	207
	IIIC	4	8	62.7	75.2	87.7	171	179.5	189

Despite the lack of distinction in the paper, the table is divided into types and groups of sports, for better insight; the sport types are defined according to Mitchell et al. (2005), where numbers refer to the static component of heart activity expressed as % of its maximal voluntary contraction (MVC)—low (I), medium (II), and High (III)—and letters to the dynamic component (e.g., % of $\dot{V}O_2\max$).

certain population, elite athletes, in two body positions (supine and standing) at rest.

2. MATERIALS AND METHODS

2.1. Subjects and Device

A group of 116 elite athletes (38 female; ages 24.4 ± 6.3) participated. Due to artifacts in signals gathered during examination, resulting from body movement and imprecise electrode mounting, data from 16 athletes could not be reasonably analyzed and were therefore excluded. The final study group consisted of 100 athletes (32 female; ages 24.6 ± 6.4).

The study was carried out at the National Centre for Sports Medicine in Warsaw during the routine periodic health evaluation and medical monitoring program, 3–4 months before the 2016 Olympic Games in Rio de Janeiro. The study group comprised athletes in sports of differing type and intensity. Data on sex, height, and body mass in specific sports are presented in Table 1.

The study, including the consent procedure, was approved by the Ethics Committee of Warsaw Medical University (permission AKBE/74/17). All participants were informed about the general aim of the measurements, though not about the importance of breathing activity (Mortola et al., 2016). Each subject had previously signed a consent form for the routine medical monitoring, which includes a statement of acceptance of the use of the results for scientific purposes.

Pneumonitor 2 was used to collect single-lead ECG signals (Lead 2), along with impedance pneumography (IP), which is related to respiratory activity (Młyńczak et al., 2017). The IP signal was measured using the tetrapolar method, with the specified electrode configuration (Seppa et al., 2013). Receiving electrodes were placed on the mid-axillary line at about 5th-rib level. Application electrodes were positioned on the same level on the insides of the arms. Standard Holter-type, disposable ECG electrodes were used. The sampling frequency was 250 Hz, sufficient in terms of heart rate variability analysis and over-sampled from a respiratory perspective (Task Force, 1996).

2.2. Protocol and Preprocessing

The measurements were performed in a diagnostic room designated for cardiological examinations. Since the periodic health evaluation and medical monitoring are performed frequently for Olympic-level athletes, the diagnostic room and the measurement procedure (very similar to that for resting ECG) were familiar to them.

Each athlete was asked to lie down on the diagnostic (ECG) couch. After attachment of the electrodes and passage of a 10-min stabilization phase, they were asked to remain supine and breathe freely (spontaneous breathing), and recording began. After 6 min, the athlete was asked to stand and again breathe freely while standing for another 6 min (Gilder and Ramsbottom, 2008; Sala et al., 2016). The duration of analysis (about 6 min for each body position) seems appropriate for characterizing cardiac and respiratory activities in a “single” measurement. This is consistent with the method used in other studies, with HRV measurement in the supine position followed by the standing position (Gilder and Ramsbottom, 2008; Sala et al., 2016). While this resembles the orthostatic maneuver, we took the latter only as an inspiration, performing the analysis for the entire supine and standing periods, i.e., without consideration of adaptation, recovery, etc. Physical data (height, body mass, and sex) were registered during the routine medical examination performed the same day.

Pre-processing of the obtained ECG signal consisted of non-linear detrending for baseline alignment and finding R peaks based on the Pan-Tompkins algorithm. Raw IP signals were pre-processed by removing the cardiac component from the IP signal by subtracting the noise component derived from least mean square adaptive filtration, then smoothing with a 400 ms averaging window (Młyńczak and Cybulski, 2017). Then, we detected and delimited breathing phases by applying an adaptive algorithm to the differentiated, flow-related signal. We did not carry out the calibration procedure to transform impedance values into volumes, instead assuming that impedance changes had reproduced the tidal volume signal in terms of shape since linear fitting provides the best agreement between IP and the reference, pneumotachometry (PNT) (Młyńczak et al., 2015).

We finally considered 10 cardiac and respiratory parameters, estimated for each participant, for the entire recording, separately for supine and standing:

- Mean heart rate (HR);
- Root-mean-square difference of successive RR intervals (RMSSD);
- Natural logarithm thereof (lnRMSSD);
- Mean respiratory rate (RR);
- ciRR—coefficient of variation of instantaneous breathing rate (iRR, calculated between inspiratory onsets);
- cInsT—coefficient of variation of the durations of the inspiratory phases (InsT);
- cExpT—coefficient of variation of the durations of the expiratory phases (ExpT);
- cInsV—coefficient of variation of the amplitudes of the inspiratory phases (InsV);

- cExpV—coefficient of variation of the amplitudes of the expiratory phases (ExpV); and
- Breathing regularity (BR), as described in formula (1) - tanh operations are added to ensure that a range of 0–100% is preserved).

$$BR = \left(100 - 20 \cdot \left(\tanh \frac{\sigma_{iRR}}{iRR} + \tanh \frac{\sigma_{InsT}}{InsT} + \tanh \frac{\sigma_{ExpT}}{ExpT} + \tanh \frac{\sigma_{InsV}}{InsV} + \tanh \frac{\sigma_{ExpV}}{ExpV} \right) \right) [\%] \quad (1)$$

The differences between body positions were assessed using the paired T or Wilcoxon rank tests (depending on the normality of the parameters, checked using the Shapiro-Wilk test; all with a significance level of $\alpha = 0.05$). Signal processing was performed using MATLAB software. Graphics and statistical inference were obtained using R software (R Core Team, 2018). The dataset of parameters and the R script are provided as **Data Sheet 1** and **Data Sheet 2**, respectively, to ensure reproducibility.

2.3. Time-Independent Causal Path Discovery

We started with the assumption that the general time-independent causality can be revealed only when the correlation appears meaningful. Therefore, we calculated the Bayesian correlation coefficient as a result of the multiplication of the slope coefficient from the linear model and the ratio of standard deviations of both vectors. Assuming

$$X = \alpha + \beta Y + \varepsilon \quad (2)$$

estimated using Bayesian approach, then

$$\text{cor}(X, Y) = \hat{\beta} \cdot \frac{\sigma(Y)}{\sigma(X)} \quad (3)$$

The significance was assumed when the maximum probability of effect $MPE > 0.9$ (from the estimation of the linear model) (Makowski, 2018).

Then, we studied all pairs using six techniques:

- The generalized correlations $r_{x|y}^*$ and $r_{y|x}^*$, with $|r_{x|y}^*| > |r_{y|x}^*|$ suggesting that y is more likely to be the “kernel cause” of x (though only when the p-value is significant) - equation (4) below implements the generalized correlation (Vinod, 2017) using the *generalCorr* R package (Vinod, 2018);
- Causal additive modeling, with *selGAM* pruning (Buhlmann et al., 2014), using the *CAM* R package (Peters and Ernest, 2015);
- Fast Greedy Equivalence Search for continuous variables (Ramsey, 2015) using the *rcausal* R package (Wongchokprasitti, 2017);
- Greedy Fast Causal Inference for continuous variables (Ogarrio et al., 2016) using the *rcausal* R package (Wongchokprasitti, 2017);

- Hill-Climbing—score-based Bayesian network learning algorithms—using the *bnlearn* R packaged (Scutari and Lebre, 2013); and
- Tabu Search, a modified hill-climbing algorithm able to escape local optima, using the *bnlearn* R package (Scutari and Lebre, 2013).

$$r_{y|x}^* = \text{sign}(r_{xy}) \cdot \sqrt{1 - \frac{E(Y - E(Y|X))^2}{\text{var}(Y)}} \quad (4)$$

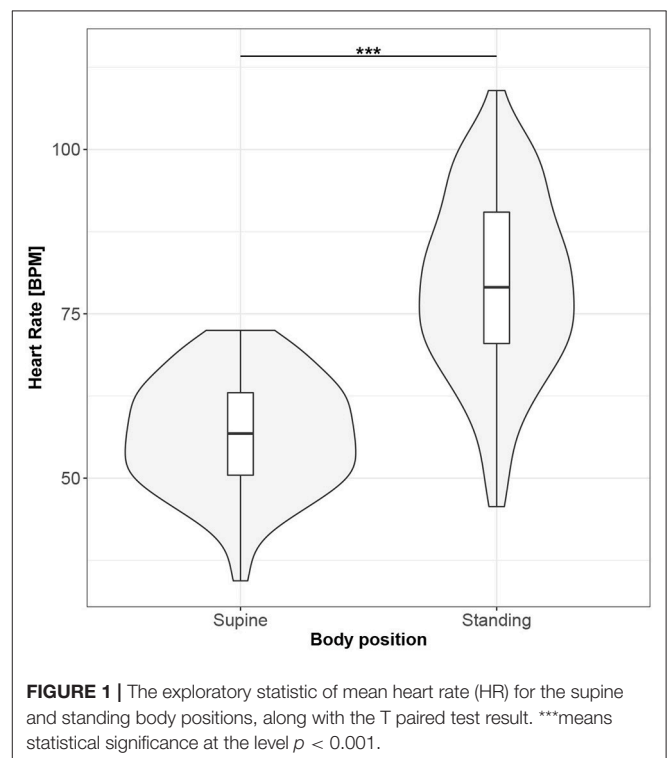
where r_{xy} is the Pearson’s correlation coefficient, *var* is variance, and the expression inside the square root is a generalized measure of correlation (GMC) defined in Zheng et al. (2012).

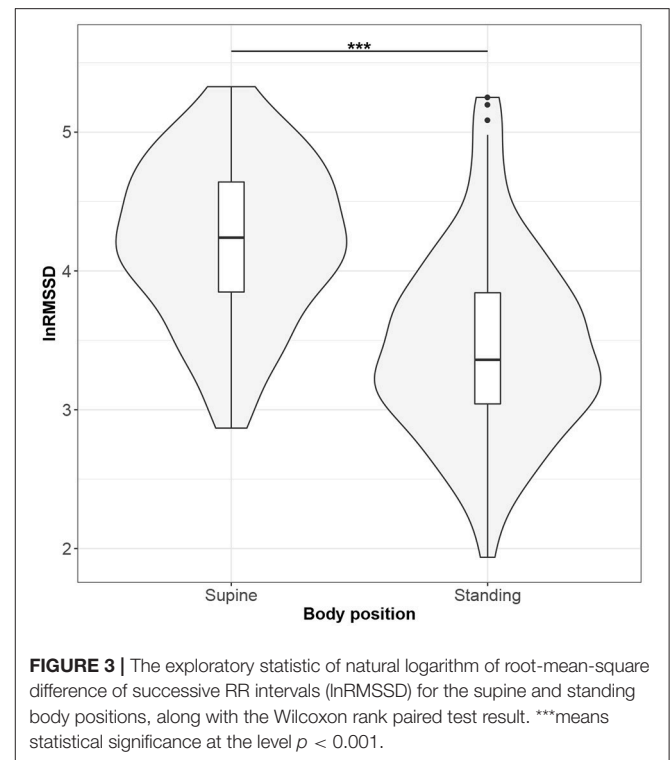
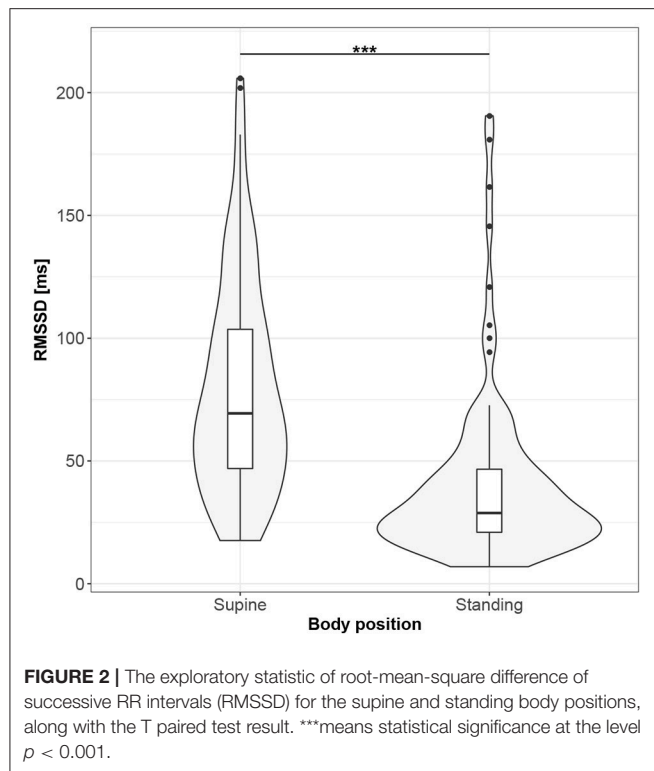
Finally, where possible, exploratory mediation analyses were conducted using the *medmod* R package (Selker, 2017). Sobel tests were performed to evaluate the significance of also considering the mediation effect, using the *powerMediation* R package (Qiu, 2018).

3. RESULTS

3.1. Statistics and Impact of Body Position

Figures 1–10 provide exploratory summaries (using violin and box plots) across body positions for all considered parameters, along with the paired test results. All parameters have statistically significant differences between body positions. As expected, HR was greater when standing; the reverse was true for RMSSD along with all respiratory parameters.





The significant Bayesian correlation coefficients are presented in **Table 2**. Results for supine are above the diagonal; for standing - below. Low correlations between cardiac and respiratory parameters (bottom left and top right corners of the table) suggested moderate connections between the analyzed parameters.

3.2. Causal Paths Discovery and Mediation Analysis

The discovered causal paths (ignoring the relationships between RMSSD and lnRMSSD, and between BR and its input coefficients) for the supine and standing positions are presented in **Figures 11–16**, separately for each of the considered methods.

Th connections between cardiac parameters seem equivocal because GC and CAM suggested the direction from RMSSD to HR for both body positions, the greedy algorithms (FGES and GFCI) could not determine the direction, while the Bayesian network methods (HC and Tabu) recommended assuming that the right direction is the opposite.

For respiratory parameters and supine body positions, 5 of the 6 methods showed that cInsT causes cExpT, and 5 out of 6 also that cInsV causes cExpV. Furthermore, all methods indicate that cExpV causes cExpT, and that cInsV causes cExpT.

The findings for the standing body position were different. Only 3 of the 6 methods confirmed the direction from cInsV to cExpV (2 were ambiguous). InsT seems to be connected much more weakly with ExpT. cInsV and cExpV appear not to cause cExpT to the same extent as for the supine position. Several loops are present between cInsV, cExpV, and ciRR. One should note

another connection, in which cInsV causes ciRR, indirectly or directly, via cExpV.

Three methods propose four connections between cardiac and respiratory parameters in supine body positions:

- cInsV \rightarrow RMSSD (CAM),
- cExpV \rightarrow RMSSD (CAM),
- HR \rightarrow cInsT (CAM and HC), and
- HR \rightarrow cExpT (Tabu).

These all indicate the occurrence of a relatively complex connection even in the most static cases. The relationships appear to be weak. Several paths can be created (in parallel); however, we think one of them may suggest the general direction for cardiorespiratory data during supine rest:

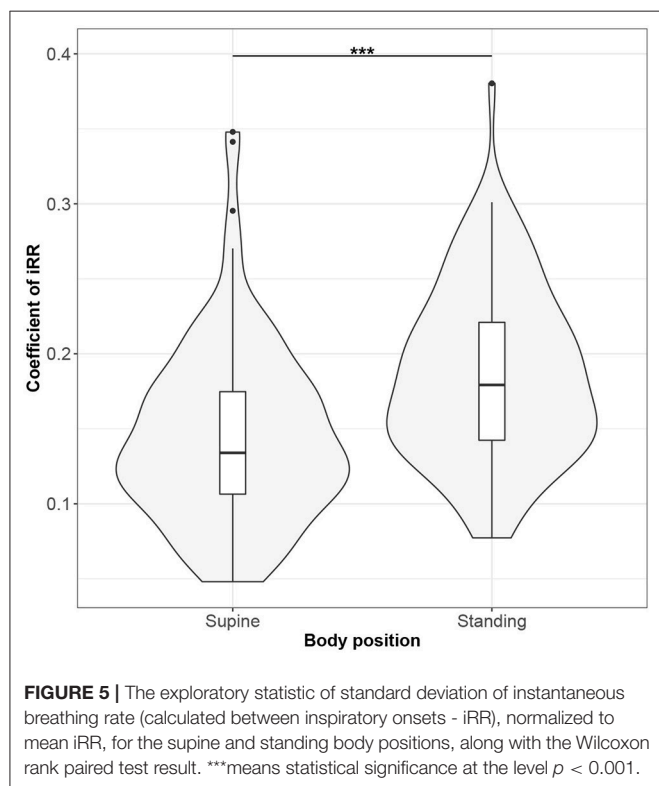
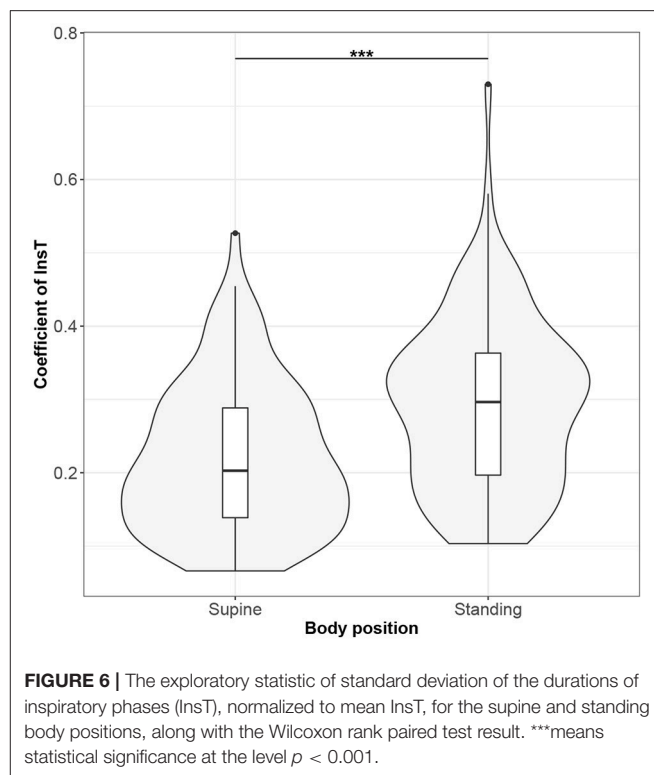
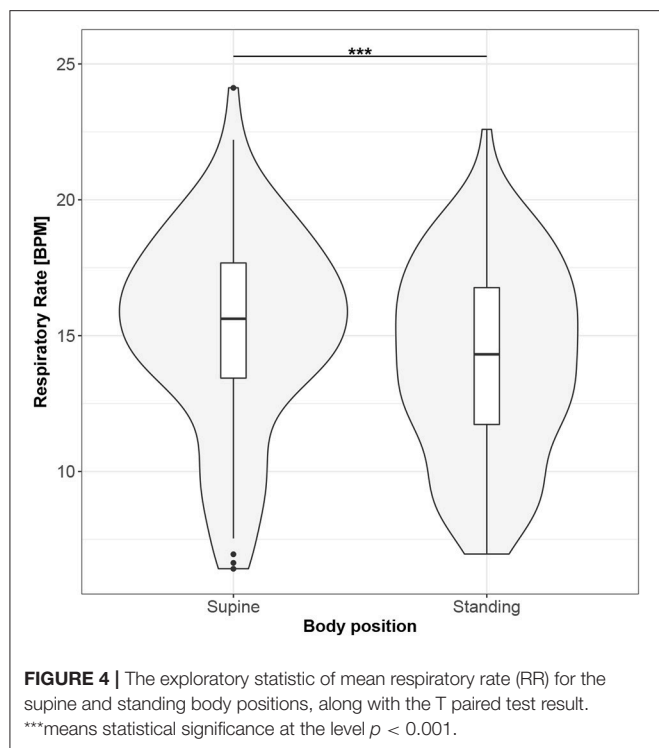
Tidal Volume \rightarrow Heart Activity Variation \rightarrow Average Heart Activity \rightarrow Respiratory Timing

Three methods (GC, HC, and Tabu) indicate the connection ciRR \rightarrow HR for the standing body position, implying the general rule for standing to be:

Normalized Respiratory Activity Variation \rightarrow Average Heart Activity

Finally, we set several paths to test for mediation effects:

- (1) RMSSD \rightarrow HR \rightarrow cInsT (supine),
- (2) HR \rightarrow cInsT \rightarrow cExpT (supine),
- (3) HR \rightarrow cInsT \rightarrow cInsV (supine),
- (4) cInsT \rightarrow ciRR \rightarrow HR (standing), and
- (5) cInsV \rightarrow ciRR \rightarrow HR (standing).



The Sobel p -values for the analyzed mediations are presented in Table 3. While none of the considered connections have a statistically significant mediation effect, the p -values

suggest tendency. It appears that the nature of these links is more non-linear, still being very light in terms of mutual correlations.

4. DISCUSSION

The main finding from our analysis is that, for the supine body position and in the elite athletes group, tidal volume seems to cause heart activity variation, then the latter causes average heart activity, which appears to affect the timing of inspiratory and expiratory phases. The relations are mild and this statement is not supported by all methods, which is not to say that any oppose it. For the standing body position, the causal relations are weaker. The most important remains that in which normalized respiratory activity variation causes average heart activity. On the other hand, for these conditions, more of the cross-correlations between cardiac and respiratory parameters were statistically significant.

This suggests the need to consider activity measures from both systems; however, in the common practice, only ECG analyses are usually carried out. The simplest, but still very informative, parameters of heart activity are mean heart rate and root-mean-square difference of successive RR intervals. The first enables study of the average value of the rhythm, while the other shows its diversity.

The concept of using HRV-related data in sports analysis has been already proposed in many applications, e.g.:

- Quantitative assessment of training load (Saboul et al., 2016),

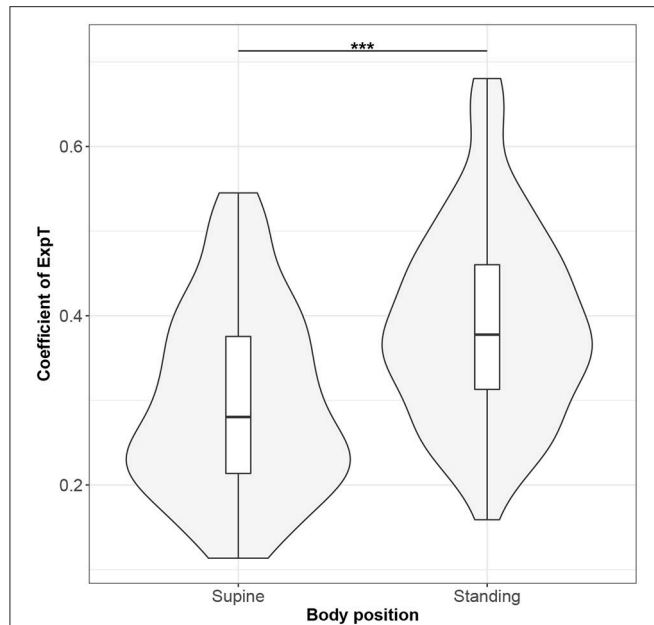


FIGURE 7 | The exploratory statistic of standard deviation of the durations of expiratory phases (ExpT), normalized to mean ExpT, for the supine and standing body positions, along with the Wilcoxon rank paired test result. ***means statistical significance at the level $p < 0.001$.

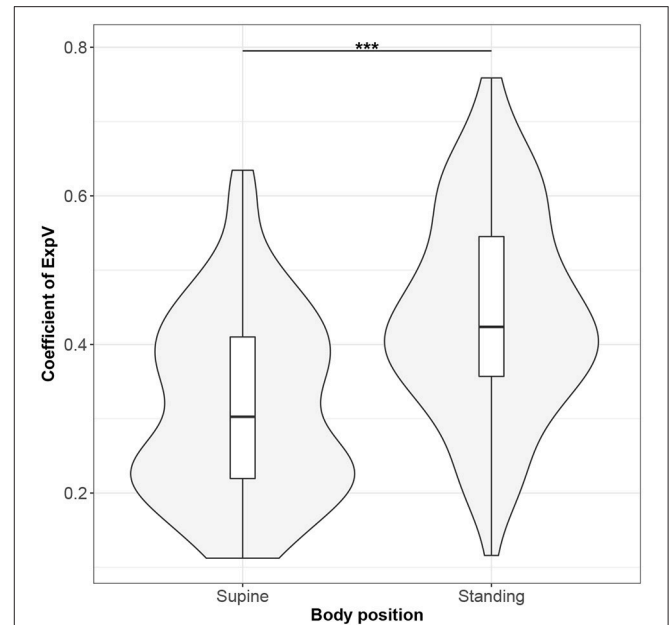


FIGURE 9 | The exploratory statistic of standard deviation of the amplitudes of expiratory phases (ExpV), normalized to mean ExpV, for the supine and standing body positions, along with the Wilcoxon rank paired test result. ***means statistical significance at the level $p < 0.001$.

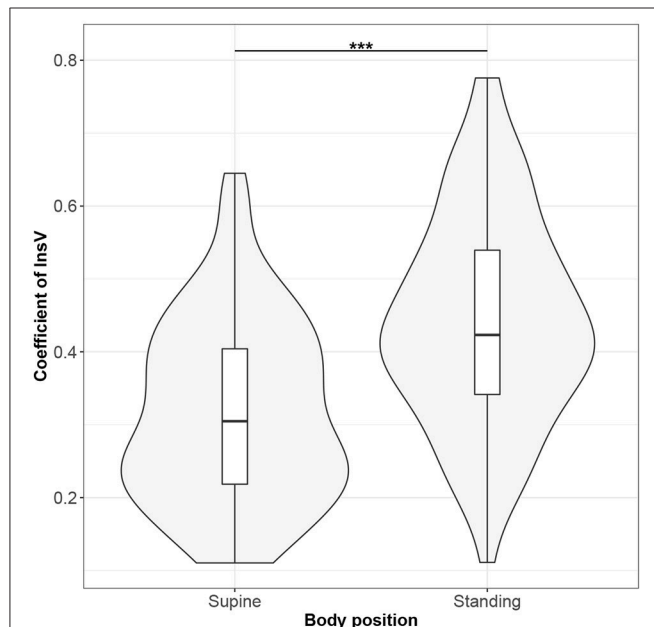


FIGURE 8 | The exploratory statistic of standard deviation of the amplitudes of expiratory phases (InsV), normalized to mean InsV, for the supine and standing body positions, along with the Wilcoxon rank paired test result. ***means statistical significance at the level $p < 0.001$.

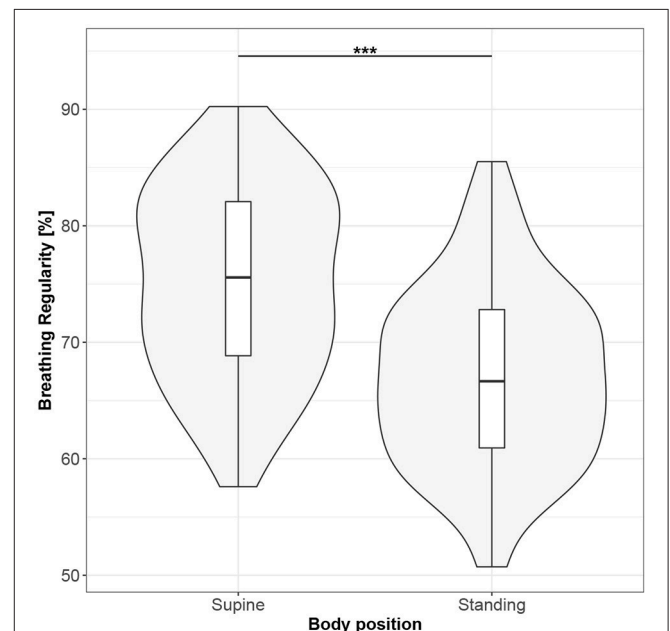


FIGURE 10 | The exploratory statistic of breathing regularity (BR) for the supine and standing body positions, along with the Wilcoxon rank paired test result. ***means statistical significance at the level $p < 0.001$.

- Monitoring of weekly HRV in futsal players during the preseason to evaluate high vagal activity (Nakamura et al., 2016),
- Progressive sympathetic predominance at peak training load as a performance prediction factor in recreational marathon runners (Tripodiadis et al., 2009),

TABLE 2 | Bayesian correlation coefficients calculated between parameters and presented when significant.

	HR	RMSSD	lnRMSSD	RR	ciRR	clnsT	cExpT	clnsV	cExpV	BR
HR	–	–0.36	–0.41			–0.17				
RMSSD	–0.47	–	0.95							
lnRMSSD	–0.51	0.91	–							
RR		0.14		–	–0.42	–0.14	–0.22			0.16
ciRR	0.22	–0.15	–0.17	–0.19	–	0.69	0.73	0.62	0.62	–0.81
clnsT	0.16	–0.17	–0.18		0.59	–	0.70	0.63	0.63	–0.84
cExpT	0.13		–0.16		0.66	0.39	–	0.62	0.61	–0.84
clnsV					0.51	0.36	0.44	–	0.96	–0.91
cExpV					0.56	0.40	0.48	0.90	–	–0.91
BR	–0.16		0.15		–0.79	–0.68	–0.73	–0.85	–0.88	–

Results for supine are above the diagonal; for standing-below.

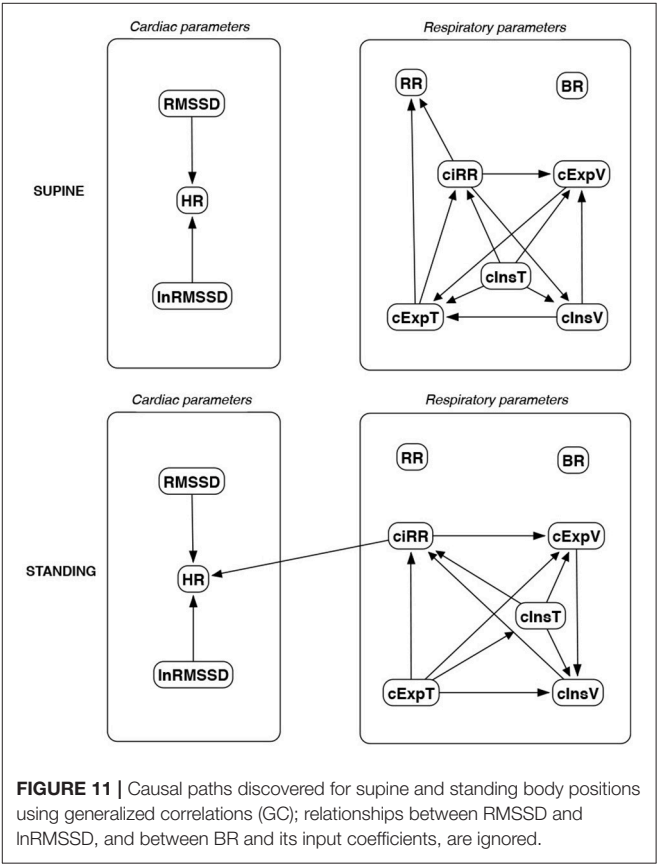
- Parasympathetic modulation elevation over a 24 h recording period induced by endurance and athletic activities (Vanderlei et al., 2008),
- Assessment of the parasympathetic tone resulting from training (Berkoff et al., 2007),
- Analysis of over-training syndrome (Dong, 2016), and
- Analysis of training adaptation (Plews et al., 2013).

However, in the presented works, the authors did not consider adding breathing activity to the analysis. Therefore, we performed the study with a device allowing recording of ventilation with minimal disruption of said activity. The Pneumonitor 2 was used to measure changes in thoracic impedance, which is related to changes in the amount of air in the lungs (Młyńczak et al., 2017). From that information, we estimated the average respiratory rate, along with five coefficients specifying the deviation of the respiratory rate, inspiratory and expiratory phase durations and amplitudes (related to volumes), allowing creation of a novel index, breathing regularity. Consequently, we parameterized cardiac and respiratory activity with indexes, which estimate a mean value and variation.

This approach connected with findings for supine body position create an interesting cardiorespiratory loop between systems. Several studies proposed to consider multi-directionality in the coupling of the cardiovascular and respiratory systems (Porta et al., 2013; Platisa et al., 2016; Radovanović et al., 2018). More importantly, the relation seems to combine (in terms of a specific mathematical framework) several physiological mechanisms. The first “arrow” indirectly describes the RSA phenomenon (Shaffer et al., 2014; McCraty and Shaffer, 2015). Some have already observed that the respiratory centers can modulate the frequency of the heart through the vagal sinus node intervention (Eckberg, 2009).

The last connection appears related to cardiorespiratory coupling, described by Sobiech et al. (2017) as a relation between the histograms of R peaks appearing before the inspiratory onset and of peaks appearing just after.

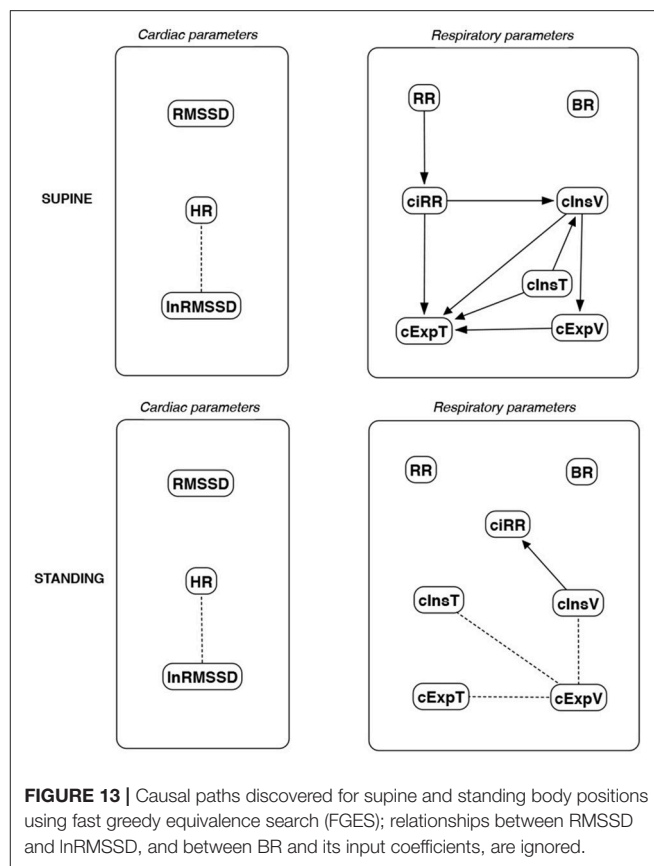
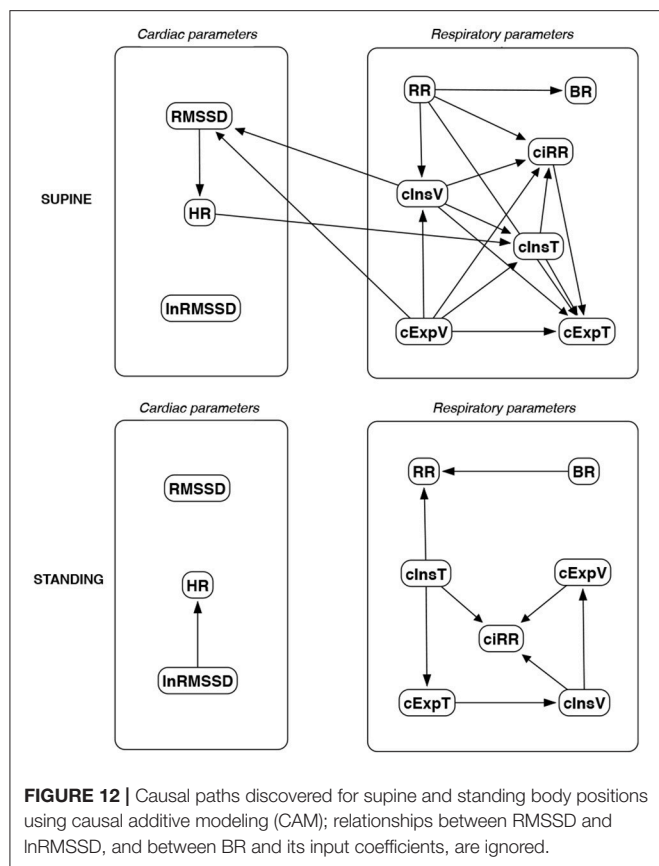
The lack of a direct “arrow” from heart activity variation, namely RMSSD in this study, to breathing corresponds with the



findings of Saboul et al. (2013) and Sala et al. (2016), wherein this index did not correlate with breathing, neither spontaneous nor controlled.

In that context, causality inference is a promising mathematical tool to expand the current analytical framework. Complex biological responses appear to be the right input, even when results are quite diverse.

All the techniques mentioned in the Introduction are based on pre-processed time series or beat-by-beat sequences. They



allow evaluation of different time resolutions, in order to focus on the specific condition and subject. We believe that this is the right approach, and that for a better grasp of how to prepare a sport-sensitive biomarker, it should be preceded by a more general approach. The approach would be time-independent, more holistic (considering a whole group of participants), and not based on prior medical knowledge. As the causality frameworks were originally introduced to deal with interventional variables, they can be even used for prediction, e.g., when several changes in a training program can be interpreted as changes to the system. As this is a retrospective study, it is also assumed that interventions are impossible, and the networks are created with no prior knowledge. Causal search relies on passive observation.

It appears debatable whether correct causal explanations can be chosen only by looking at observed data. On the other hand prior knowledge would enable acquisition of answers to causal questions without performing interventions (still difficult to manage from physiological perspective). Here, the context is different. We believe that causal analysis and cardiorespiratory relation can be a relevant supplement to already-established techniques and play the role of a biomarker (or its part) for establishing the state of the athlete. In this introductory approach, we assumed the single parameters describe specific subjects, and the analysis collects them all together.

The proposed protocol is inspired by the orthostatic maneuver, but the analysis for each positions is independent. Moreover, we hold that, from a causal discovery perspective, one can consider the entire segment of the signal, instead of subsections like the adaptation after standing up. Looking at the entire segment makes the analysis simpler from an operators perspective.

As the estimated causal structures seem mild, analyzing supine-to-standing changes would make the method more robust (by considering changes in intrathoracic pressure or differences in venous return characteristics). Radovanović et al. (2018) reported that even slight change of body position may change the direction of the relationships. Sobiech et al. (2017) also suggested that the results can be reliably analyzed only during static conditions and that the effect is the strongest in a resting state. However, in our opinion, the differences between body positions may have a significant impact on the analysis of cardiorespiratory data and should not be ignored. These differences, established for two body positions, may serve as an additional input for determination of the adaptation profile.

The effect observed for standing body position has already been studied by Sala et al. (2016), who analyzed on a basic level the effectiveness of assessment of the balance change in the autonomic system during active verticalization. They observed that this change should be stronger in athletes and assumed that it is associated with improved physical capacity. Apart from that,

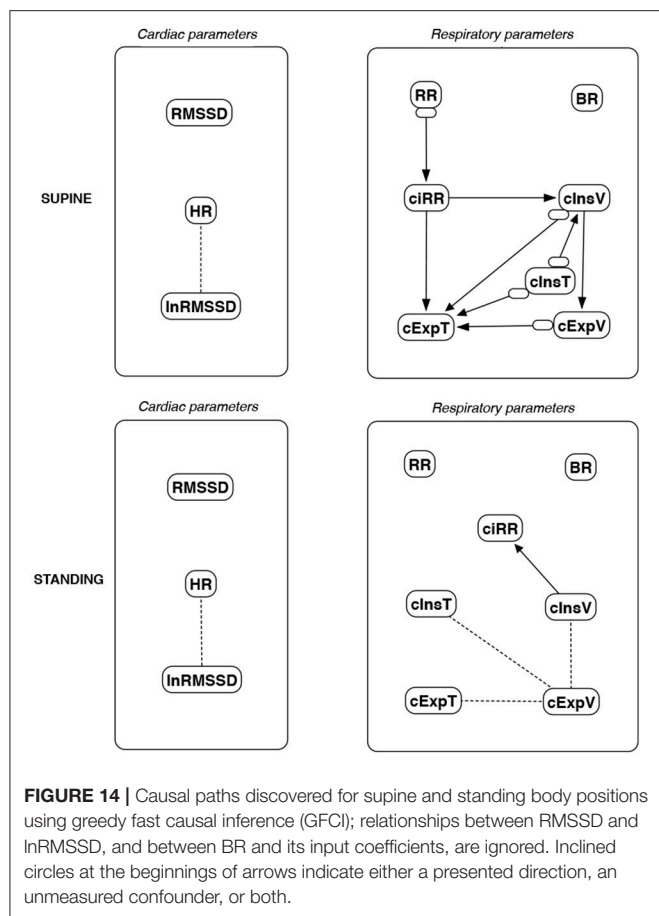


FIGURE 14 | Causal paths discovered for supine and standing body positions using greedy fast causal inference (GFCI); relationships between RMSSD and lnRMSSD, and between BR and its input coefficients, are ignored. Inclined circles at the beginnings of arrows indicate either a presented direction, an unmeasured confounder, or both.

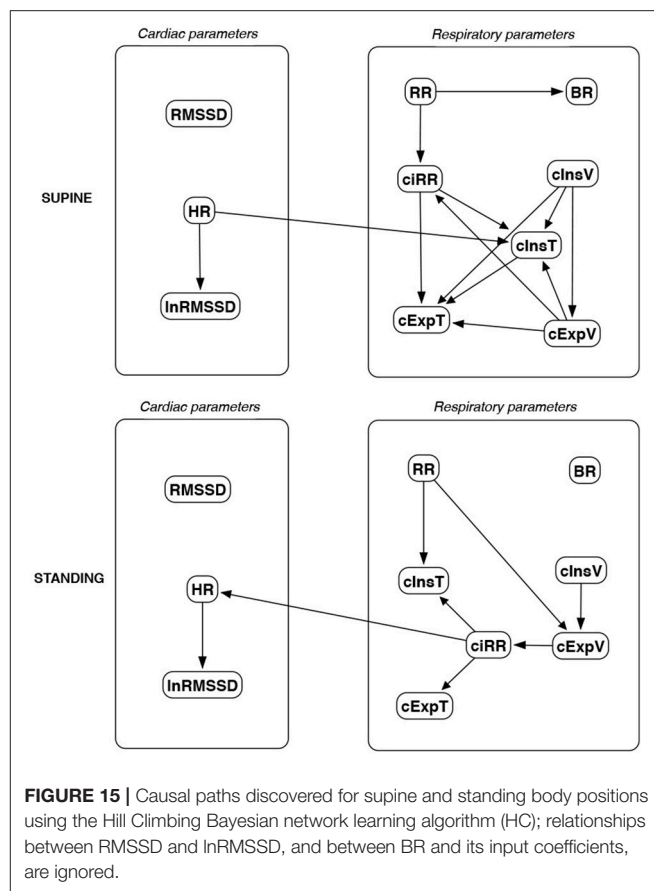


FIGURE 15 | Causal paths discovered for supine and standing body positions using the Hill Climbing Bayesian network learning algorithm (HC); relationships between RMSSD and lnRMSSD, and between BR and its input coefficients, are ignored.

increased coupling between heart period and arterial pressure in response to postural changes was reported in Silvani et al. (2017).

4.1. Use of Causal Discovery Framework

The findings presented in this study open up a novel domain of possible parameterization of physiological connections. We think the framework can be used to profile athletes, analyzing trends throughout the training schedule and testing changes in relations' direction and strength relative to improved adaptation, etc.

Causal inference, even if used to search for graphs between analyzed parameters, was originally proposed to evaluate the effects of interventions, dividing variables into interventional and observational sets. In both sports medicine and daily practice, modification of the training can be regarded as such an intervention.

The paths discovered by the various algorithms should be reconciled with medical background knowledge and, even more importantly, should be verified further in a prospective study. With so many possible interventions, the paths may reduce the complexity of test protocols: one might expect to influence heart rate and its variability by changing the depth, not the frequency, of breathing.

4.2. Limitations of the Study

There were only 116 participants, of whom 100 were ultimately considered. The study group appears to be heterogeneous, apart from the fact that the studies were conducted in the "hot period" 3–4 months before the Olympic Games, which may suggest a state of over-training (neither questionnaires nor objective data about it were collected). Also, the device was new for all subjects. These factors might influence the results, particularly the relatively high supine respiratory rates and standing heart rate. The existence of differences between sports would also affect the conclusions. The collection of only one observation per athlete precludes reproducibility analysis.

There were no control variables assumed in the protocol. One may consider a respiratory activity to be controlled during the measurement. However, the primary objective of the registration was to measure cardiorespiratory data without any restrictions to the respiratory activity. As this is a retrospective study, we could not affect it during the analysis. This may be one of the reasons why the studied relationships were so mild.

Measurements were carried out only once for each subject, and only at rest, not in a natural environment. It is necessary to perform comparative registrations on a unified group of athletes under laboratory conditions, during the preparatory period and just before the season, in combination with a psychometric questionnaire. The results of a study in which the registration

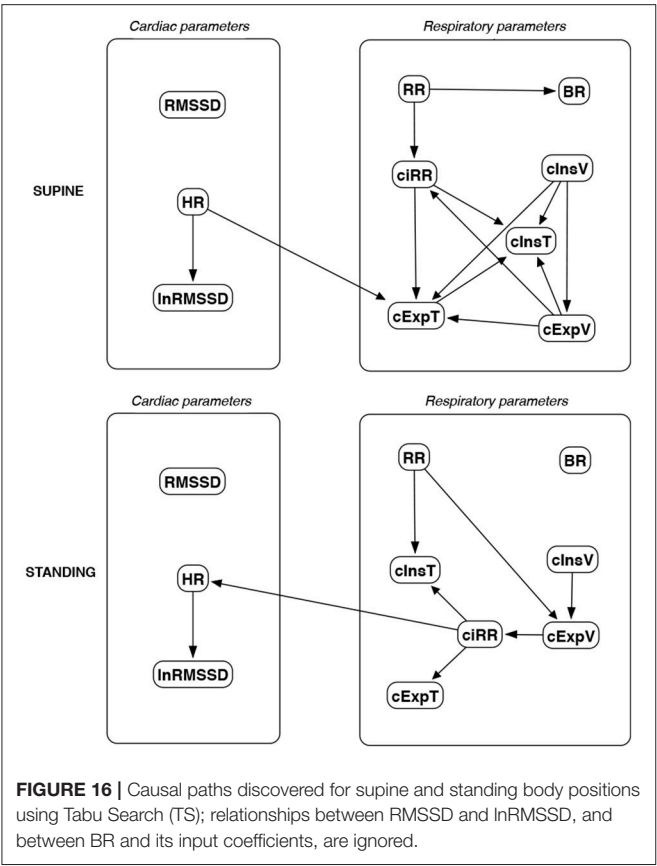


FIGURE 16 | Causal paths discovered for supine and standing body positions using Tabu Search (TS); relationships between RMSSD and lnRMSSD, and between BR and its input coefficients, are ignored.

Path	Position	Sobel's <i>p</i> -value
RMSSD → HR → cInsT	Supine	0.073
HR → cInsT → cExpT	— —	0.086
HR → cInsT → cInsV	— —	0.088
cInsT → cIRR → HR	Standing	0.105
cInsV → cIRR → HR	— —	0.058

could be performed outside the laboratory, during normal training, or even with 24 h Holter-based tracking of natural functioning, would yield more condensed findings.

Notably, the methods used to discover causal paths sense different aspects (the algorithms having different starting points) and can produce more liberal or conservative results. As only the “significant” pairs of causal-effect links were presented, possible bias cannot be evaluated directly. As the relevant analysis could not be performed for a few participants, we decided not to divide participants by sex or sport type.

4.3. Further Considerations

In the presented work, we use a data-driven and time-independent approach (on a large inter-individual scale). It is

considered as a starting point. As stated in the introduction, the presented approach seems to be an overview, to be performed before time-dependent methods which may allow the separate assessment of a single person, with finer time resolution. Therefore, as a next step, we plan to discover various non-linear parameters and techniques to confirm, expand and enhance the findings presented in this paper. Still, the main outcomes are coherent with the knowledge-based predictions and results reported, e.g., in Sobiech et al. (2017).

Eckberg showed that the RSA phenomenon is not only the effect of respiration on RR intervals (or heart rate in general) but might be also treated as the response of heart rate to the respiratory modulations resulting from arterial pressure changes mediated by baroreflex (Eckberg, 2009). Sobiech et al. (2017) suggested that arterial blood pressure is probably the driver (cause) of both cardiac and respiratory function. This requires further research with more modalities included in the analysis (Zhang et al., 2017).

Other important questions include: Are the cardiorespiratory connections (direction and strength) the same in different sports, or specific to each? What other factors, like sex, height, or body mass, confound the results? These questions are especially important not only for elite athletes, but also for subjects with abnormalities, and will be studied (Sharma et al., 2018).

5. CONCLUSIONS

Adding respiratory data to cardiac signals in sports medicine would provide better monitoring and evaluation of athletic training.

In the presented paper, we pose the hypothesis that average or diversity cardiac and respiratory parameters, describing overall characteristics of RR intervals and tidal volume curves, without consideration of the time resolution, are linked by causal paths. Furthermore, such an approach can precede more detailed, individual, time-series-related analysis.

Based on the data gathered from 100 elite athletes at rest in two body positions, the applied causal discovery frameworks suggested moderate connections. For supine, the general path led from tidal volume, through heart activity variation and average heart activity, to respiratory timing. For standing - from normalized respiratory activity variation to average heart activity. Different graphical structures and directions were observed for the two body positions, which may improve the resolution of the findings.

We think posterior-style causal inference and characterization may develop descriptions of cardiorespiratory connections and possibly distinguish between various groups of athletes. The method can be used to profile athletes, elaborate on modifications of their training schedules, and find objective ways to improve their competitive performance.

AUTHOR CONTRIBUTIONS

MM and HK worked on the conceptualization, investigation, methodology, project administration, validation, and writing and

reviewing. MM worked on data curation, formal analysis, and visualization. HK provided the resources.

ACKNOWLEDGMENTS

The authors thank Martin Berka for linguistic adjustments. MM gratefully acknowledges the Institute of Metrology and Biomedical Engineering of the Faculty of Mechatronics, Warsaw University of Technology, for supporting this work.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fphys.2018.01455/full#supplementary-material>

Data Sheet 1 | The data file with cardiac and respiratory parameters calculated from all participants for the two body positions.

Data Sheet 2 | The R script comprising all algorithms mentioned in the text, from loading of the above data to delivery of final outcomes.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Self-Reported Periodization of Nutrition in Elite Female and Male Runners and Race Walkers

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OPEN ACCESS

Edited by:

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 04 October 2018

Accepted: 16 November 2018

Published: 03 December 2018

Citation:

Heikura IA, Stellingwerff T and
Burke LM (2018) Self-Reported
Periodization of Nutrition in Elite
Female and Male Runners and Race
Walkers. *Front. Physiol.* 9:1732.
doi: 10.3389/fphys.2018.01732

Athletes should achieve event-specific physiological requirements through careful periodization of training, underpinned by individualized and targeted nutrition strategies. However, evidence of whether, and how, elite endurance athletes periodize nutrition is scarce. Accordingly, elite international female ($n = 67$) and male ($n = 37$) middle/long-distance athletes (IAAF score: 1129 ± 54 , corresponds to 13:22.49 [males] and 15:17.93 [females] in the 5000 m) completed an online survey (February–May 2018) examining self-reported practices of dietary periodization for micro (within/between-days), meso (weeks/months) and macro (across the year) contexts. Data are shown as the percentage of all athletes practicing a given strategy followed by the % of athletes reporting various beliefs or practices within this strategy. Differences according to sex, event (middle-distance [800 m/1500 m] vs. track-distance [3000 m–10000 m] vs. road-distance [marathon/race walks]), caliber (high [major championship qualifier] vs. lower), and training volume (low/moderate/high male and female tertiles) were analyzed using Chi-square test or Kruskal–Wallis Test and indicated statistically different when $p \leq 0.05$. Most athletes reported eating more on hard training days (92%) and focusing on nutrition before (84%; carbohydrate intake [63%] and timing [58%]) and after (95%; protein goals [59%], timing [55%], carbohydrate goals [50%]) key sessions. Road-distance were the most (62 and 57%), and middle-distance the least (30 and 30%) likely to train fasted ($p = 0.037$) or restrict carbohydrates periodically ($p = 0.050$), respectively. Carbohydrate intake during training (58% of total) was more common in males (79%; $p = 0.004$) and road-distance (90%; $p < 0.001$) than females (53%) or middle/track-distance (48 and 37%). Most athletes (83%) reported following a specific diet before and during race day, with half of the athletes focusing on carbohydrates. Nearly all (97%) road-distance athletes reported following a during-race nutrition plan (carbohydrates/fluids:89%). Only 32% reported taking advice from a dietitian/nutritionist. Based on our analysis: (1) Road-distance athletes periodize carbohydrate availability while track/middle-distance avoid low carbohydrate availability; (2) Middle-distance runners emphasize physique goals to guide their nutrition strategies; (3) Females seem to be more cautious of increasing energy/carbohydrate intake; (4) Among all athletes, nutrition strategies are chosen primarily to improve performance, followed by reasons related to physique, adaptation and health outcomes. Overall, these athletes appear to possess good knowledge of nutrition for supporting training and competition performance.

Keywords: nutrition periodization, elite athletes, endurance athletes, carbohydrate availability, questionnaire

INTRODUCTION

Despite decades of interest in the periodization of training, it is only recently that a holistic approach to periodization across a range of themes that affect competition preparation has been suggested (Burke et al., 2018; Mujika et al., 2018; Stellingwerff et al., 2018). In fact, the concept of integrating a periodized nutrition plan within the annual training program was formally proposed in a previous expert panel around nutrition for track and field athletes by Stellingwerff et al. (2007). The principles, practices and terminology around the periodization of nutrition have been summarized in several recent reviews (Jeukendrup, 2017; Burke et al., 2018; Stellingwerff et al., 2018). The underlying theme is that strategic and targeted nutritional interventions can be used to augment the outcomes of the various specific training cycles [micro (within-day to days), meso (several weeks) and macro (months to years)]. Thus, decisions on periodization of nutrition should be preceded by a thorough examination and understanding of the sport-specific (general) determinants of success as well as athlete-specific (individual) performance gaps, with strategies (including nutrition interventions) to address the gaps being integrated into the periodized training program (Stellingwerff et al., 2018).

A variety of aspects of nutrition can be periodized in support of different training goals, ranging from fundamental issues such as energy intake through to the more specialized and “fine tuning” aspects of supplement use (Jeukendrup, 2017; Stellingwerff et al., 2018). From a macro perspective, energy intake needs to be manipulated across, and within, training days according to fluctuations in the energy cost of the athlete's training program, as well as strategic integration of periods of alterations to energy balance to manipulate body mass/composition (Melin et al., 2018; Stellingwerff, 2018). Here, it is important to recognize that energy mismatches due to deliberate efforts to reduce body mass/fat content, or the failure to account for the energy cost of a heavy training loads for prolonged periods are likely to impair health and performance with both short- and long-term consequences (Mountjoy et al., 2018). At the other end of the spectrum, periodized use of supplements may range from the use of iron supplements to ensure adequate iron status during altitude training block (Mujika et al., 2018), to the use of performance aids such as caffeine, creatine, buffers (e.g., beta-alanine or bicarbonate) and nitrate/beetroot juice to practice intended competition strategies, or to provide support for targeted training sessions.

The periodization of macronutrients includes themes of meeting the specific fuel needs of training and competition sessions (micro-periodization: particularly in the case of carbohydrate [CHO], and perhaps, fat intake), practicing event nutrition strategies (meso-periodization: particularly CHO intake during longer events) and providing both an additional stimulus and the building blocks needed to optimize the synthesis of new proteins as part of the adaptation to training (micro-, meso-, and/or macro-periodization: e.g., protein intake). Current guidelines around protein intake for all types of athletes promote the regular intake of modest amounts (e.g., ~25 g every 3–4 h) of high quality protein over the day, including soon after the

completion of key training sessions (i.e., sessions of high intensity and/or high duration [>90 min]) (Phillips and Van Loon, 2011). Meanwhile, there may be advantages in increasing protein intake during periods of deliberate energy manipulation to achieve loss of body fat to assist with the maintenance of lean mass (Hector and Phillips, 2018). The amount and timing of CHO intake between and within days should track the substrate needs of training and events (Areta and Hopkins, 2018), particularly when it is important to optimize performance in competitions or key training sessions (Thomas et al., 2016). There may also be a need to undertake specific strategies to train the gut to tolerate increasing amounts of CHO and fluids during exercise (Cox et al., 2010; Jeukendrup, 2014), in preparation for race nutrition practices during prolonged events, particularly in hot environments, where intake during the event plays a major role in performance success. Taken together, there are numerous examples of macro-, meso-, and micro-periodization of nutrition to optimize training adaptation and/or acute sport performance.

Although CHO intake is mostly considered in relation to its role as a key fuel for the muscle and brain, the application of molecular techniques to investigate the muscle response to exercise has created interest in the effects of low CHO availability on cellular signaling and enhanced adaptation to endurance training. According to various reviews (Philp et al., 2011; Bartlett et al., 2015; Hearn et al., 2018; Impey et al., 2018), undertaking endurance exercise with an environment of low CHO availability during and/or after the session may upregulate the activity of key molecules in the adaptive responses to exercise, leading to an enhanced or prolonged adaptation period. Observations of increases in the acute response to exercise have led to studies of the chronic implementation of periodized CHO availability (i.e., integration of strategies of high CHO availability to “train hard” for optimized performance, and strategies of low CHO availability to “train smart” with enhanced adaptation) to test its effect on performance outcomes. It is important to note that it can be difficult to achieve the right balance between training quality and adaptation within a controlled laboratory study design (Yeo et al., 2008; Hulston et al., 2010). Accordingly, although a few studies have reported superior performance outcomes in cohorts of trained/well-trained individuals (Marquet et al., 2016a,b), the translation to elite athletes seems more difficult (Burke et al., 2017; Gejl et al., 2017). Nevertheless, the strategy has been integrated by some elite athletes (Stellingwerff, 2012) and recognized as an emerging concept in the most recent sports nutrition guidelines (Thomas et al., 2016). One challenge for athletes, coaches and sports scientists is understanding the meanings and nuances of different strategies to periodize CHO availability within training and competition preparation. However, this has been addressed in a recent commentary in which terminology, practices, mechanisms and evidence of different strategies have been summarized (Burke et al., 2018).

In recognizing the value of a periodized approach to sports nutrition, current guidelines also promote the importance of individualization, which is dependent upon the specific performance demands of the sport/event and the unique athlete response to the intervention. Factors that may influence the individualized implementation of a specific nutrition strategy

might include: (1) race distance (e.g., middle-distance athletes may not benefit from training the gut to consume liquids, while this is an important strategy for most road athletes); (2) event specific body composition norms; (3) training/event volume (e.g., higher volume training may require more emphasis on adequate energy/fuel availability); (4) training location (e.g., altitude/heat); and (5) time of the year (e.g., protocols to improve body composition and race performance might be emphasized closer to the competition season) (Stellingwerff et al., 2007; Stellingwerff, 2012; Melin et al., 2018; Stellingwerff et al., 2018). Therefore, it is expected that each athlete will have a unique and constantly changing periodized nutrition plan suited to their specific needs.

In parallel to the growing evidence base for the value of a periodized approach to nutrition, there is interest in understanding whether/how elite athletes practice these strategies within the real-world annual training/racing calendar. The available studies are limited to endurance and team sports athletes who have provided a snapshot of the micro- and meso-periodization of nutrition during training phases (Burke et al., 2003; Bradley et al., 2015; Naughton et al., 2016; Heikura et al., 2017a; Anderson et al., 2017a,b) or around competition (Stellingwerff, 2012, 2018).

More recently, we completed preliminary work to characterize self-reported approaches to periodization of nutrition over the annual training plan (Heikura et al., 2017b). Our pilot project captured an account of practices and the underlying rationale for nutritional periodization across the year (macro-periodization), with special consideration of various micro (between/within-day) and meso (various training/competition phases) cycles in 48 elite distance and middle distance track and field athletes. Having tested and updated this pilot study survey, in the current study we embarked on the investigation of the self-reported practices of dietary periodization across the annual training/racing calendar in a large cohort of world-class track and field endurance athletes. Our goal was to characterize periodized nutrition practices across the year (macro cycle), and during specific meso and micro cycles of training/racing in this group, with attention to the effects of sex, athlete caliber, event duration and volume of training on these practices.

MATERIALS AND METHODS

Study Design and Participants

Based on our pilot study using a similar survey self-reported approach (Heikura et al., 2017b) we further developed the current study's survey into an online tool consisting of variously themed questions around dietary micro-, meso-, and macro-periodization across the various annual training phases. Along with strategic changes to the survey, gained from insights from our pilot study, we also aimed for this to be completed by a larger (target = 100) and more internationally representative group of athletes. We recruited elite female and male middle/distance athletes using online advertisements as well as direct contacts (via email or word-of-mouth) to athletes, coaches, applied sports practitioners and national sporting organizations (in Canada,

United States, Australia, Japan, and Finland). To be included in the study, the athletes needed to be ≥ 18 years of age, currently and actively racing in the middle (800 m, 1500 m), distance (3000–10000 m) or road (half-marathon/marathon, 20 k/50 km race walk) events under the International Association of Athletics Federations (IAAF) and have a personal best of ≥ 1043 IAAF points (this corresponds to 5000 m time of 13:47.26 and 16:03.84 in males and females, respectively). Recruitment and completion of the surveys were completed between February 8 and May 21, 2018. The Ethics Committee of Australian Catholic University approved the study protocol which conformed to the Declaration of Helsinki.

The Survey

The survey consisted of an updated version of our pilot study based on our reflections on the responses from the original cohort and additional feedback from colleagues and athletes. Whereas the pilot survey included a total of 29 questions (7 main questions and 22 sub-questions), the updated survey was expanded to include a total of 59 questions (19 questions on training/racing characteristics, plus 13 main and 27 sub-questions around nutritional practices). The final version of the survey was built online using SurveyGizmo (Boulder, CO, United States). Skip logic was used, building a custom path through the questions according to the respondent's answers, for an improved participation experience (less confusion) and efficiency (less time to complete the survey).

The survey was completed anonymously, and an informed consent (a prerequisite for completing the survey) was completed as part of the online survey by all participants. The first part of the survey included background information, instructions, and general subject information. Thereafter, the athlete was asked to choose one of the two annual training periodization programs (track [e.g., 800–10000 m]) vs. road [e.g., the marathon and race walks] that best reflected his/her yearly program. Questions approached annual (macro) periodization of nutrition as a whole (*Part A: general principles of annual training/competition diet*) and as typically defined separate periodized training phases (meso cycles): *Part B: Base / endurance training phase*, *Part C: Main competition season* (i.e., several months in duration: track athletes) or *preparation for competition* (i.e., one or more weeks in duration: road athletes), and *Part D: Nutrition immediately before and on race day*. Part A also included questions on general nutrition principles (e.g., vegetarian, paleo, very high energy, low carb high fat, gluten free) in the overall diet or during specific time periods (e.g., altitude training or during return from illness/injury). Additionally, parts B, C, and D asked questions on training volume, key session and race frequency as well as number of race peaks. Across the survey and throughout this manuscript, hard training days were defined as “high volume and/or intensity days” and key sessions as “high intensity and/or high duration [> 90 min] sessions or serious gym sessions.” Fueling was defined as eating foods (CHO foods, protein foods, sports foods, etc.) before training. Fasted training was defined as “training first thing in the morning without having eaten any food or consumed any other carbohydrates, or training later in the day without having eaten any carbohydrates for at least 8 h prior.”

Within the survey, reminders of these terminology were included within each question that targeted nutrition in relation to these themes. Finally, the survey ended with an open, but optional, comment box.

It is important to note that the survey was purposely constructed to apply to the culture, practices and terminology used in endurance events in track and field. As such it is not directly applicable to other sports, and if used for other populations, even among endurance sports, it will need to be customized to the specific characteristics of these sports. A sample survey has been provided in the **Supplementary Material** to this paper.

Data Management and Statistical Analysis

The data were checked and cleaned by excluding duplicate responses (i.e., two responses from the same individual), responses that were clearly false or confusing, and responses from those that did not satisfy the requirement of ≥ 1043 IAAF points. The answers were classified into clusters for further analysis using groupings based on *sex* (male vs. female), *distance* (middle distance (MidD; 800, 1500 m) vs. track distance (TrackD; 3,000 m steeplechase to 10,000 m) vs. road distance (RoadD; marathon, race walks)), *sex-based training volume groupings* based on within sex tertiles of the entire data set (Females: low: ≤ 100 km/wk, moderate: 101–129 km/wk, high ≥ 130 km/wk; Males: low: ≤ 119 km/wk, moderate: 120–155 km/wk, high ≥ 156 km/wk) and athlete *caliber* (High: major championship (Olympics or World Championships) medalist, finalist and/or qualifier; Lower: the rest of the data set).

Data were first organized using Microsoft Excel, while further statistical analyses were conducted using SPSS Statistics 22 software (INM, Armonk, NY, United States). Data are presented as means \pm standard deviations (SD) and number (n) and percentage (%) of responses. Normality of continuous data was checked with the Shapiro–Wilk goodness-of-fit test. Student's t-test for independent samples was used to test for differences in age, training volume and IAAF scores between subgroups. For YES/NO answers, % was calculated from the n of the total sample; for sub-questions, % was calculated from the remaining n resulting after the main initial question. Chi-square test (X^2) for independence with Yates Continuity Correction along with phi effect size statistic were used to test for differences between subgroups. Where more than two subgroups were present, Kruskal–Wallis Test was used as a post hoc test. Across the paper, statistical significance is shown when $p \leq 0.05$. In addition to numerical outcomes, relevant quotes provided by athletes (in cursive) have been embedded in the results section to provide further qualitative insights into the topic in question. To aid in the interpretation of results from the lengthy survey, aggregation of consistent outcomes across the survey was undertaken by all authors upon visual inspection of figures to develop a series of themes.

RESULTS

General Questionnaire Outcomes

A total of 104 athletes (67 female and 37 male) from middle ($n = 27$), distance ($n = 34$), and road ($n = 43$) groups were

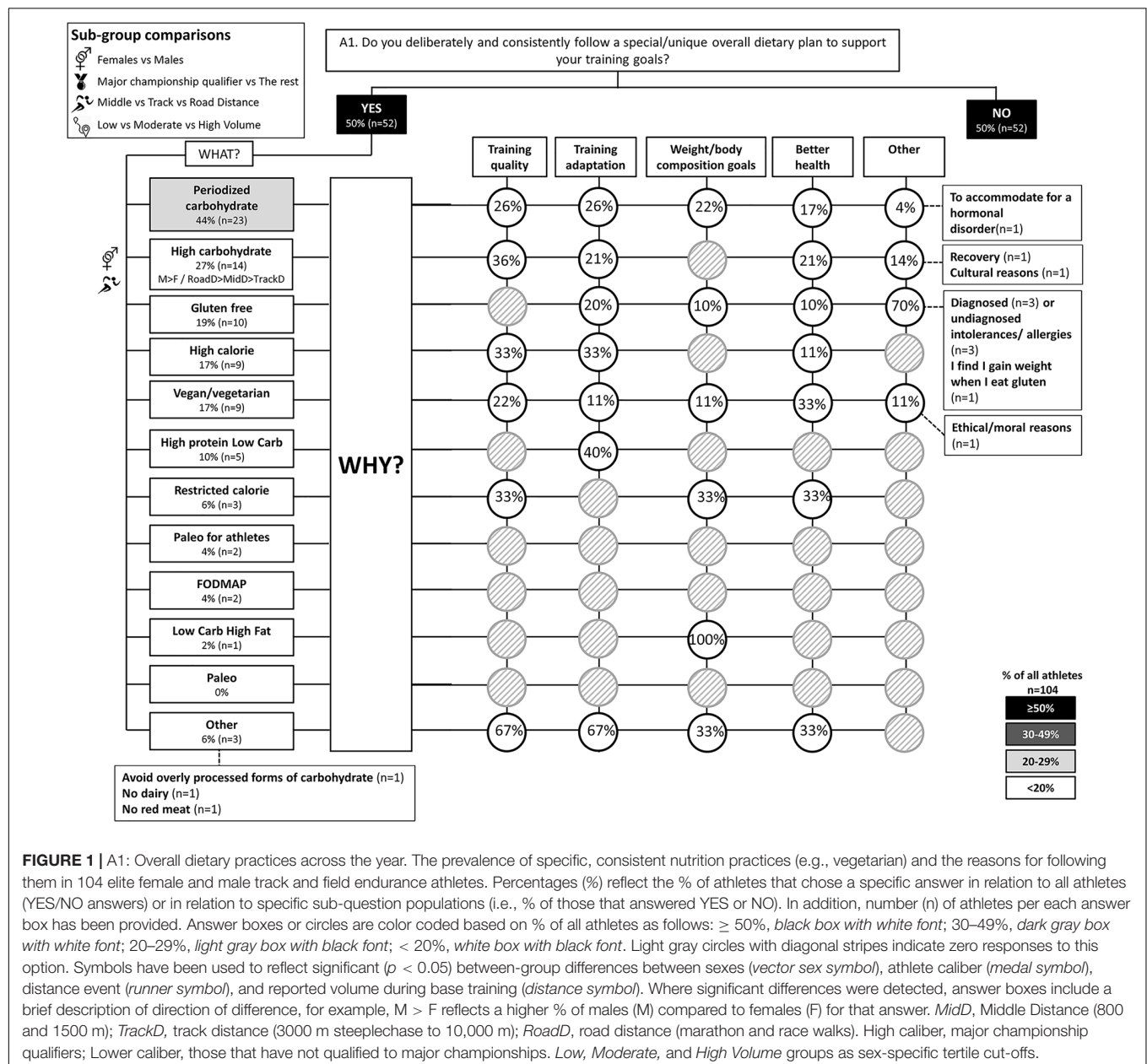
TABLE 1 | Participant background and characteristics of training and competition in elite middle/long-distance athletes (all, sex-based comparisons, distance-based comparisons).

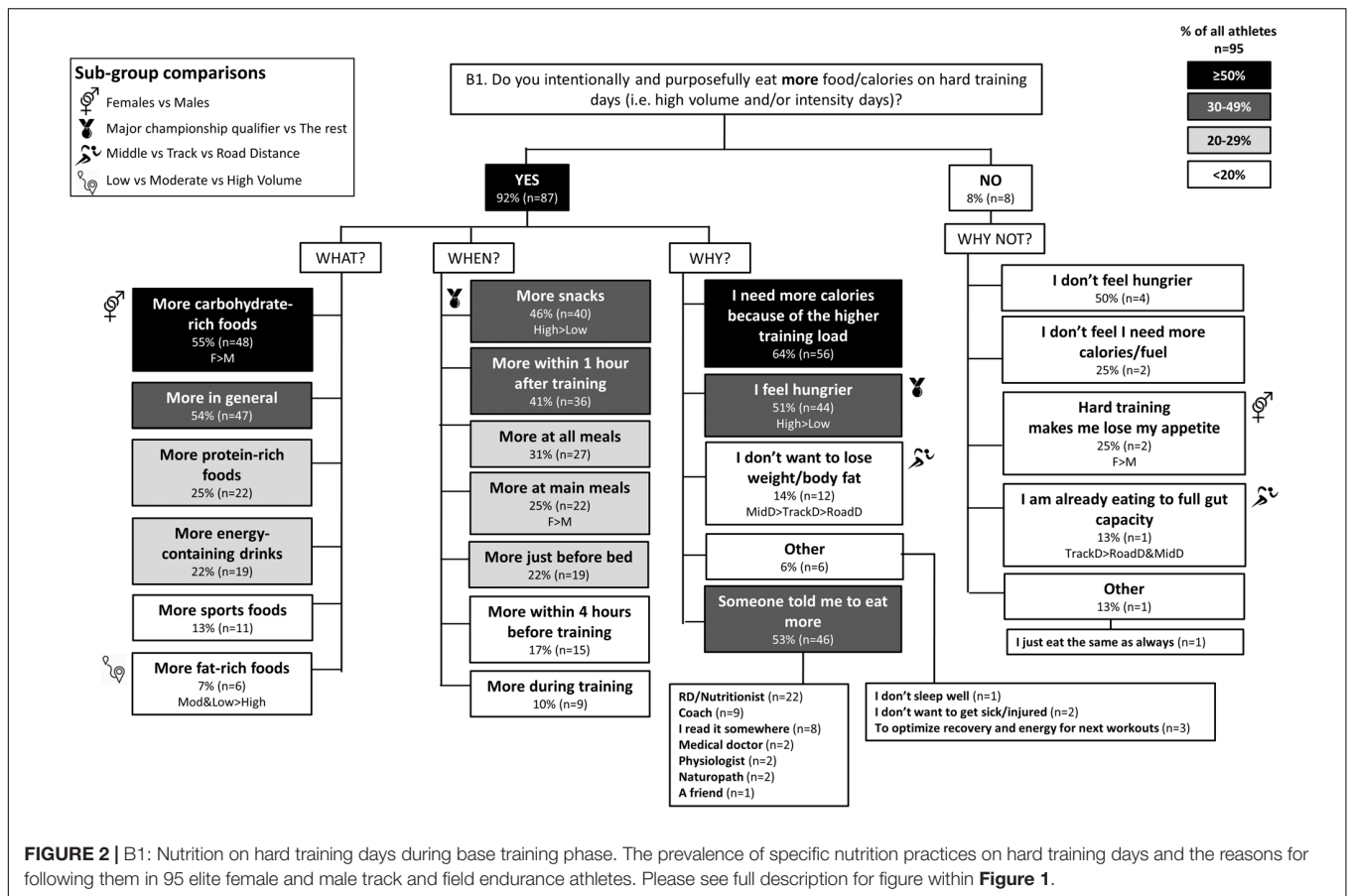
	All ($n = 104$)	Sex comparison		Distance comparison		
		Females ($n = 67$)	Males ($n = 37$)	MidD ($n = 27$)	TrackD ($n = 34$)	RoadD ($n = 43$)
Age (year)	29.2 \pm 5.8	28.9 \pm 5.8	29.6 \pm 5.9	26.5 \pm 3.6***	28.3 \pm 6.3 #	31.6 \pm 5.7
IAAF score	1129 \pm 54	1135 \pm 54	1119 \pm 54	1135 \pm 48	1119 \pm 45	1133 \pm 64
Training/competition background (years)	9.4 \pm 4.6	8.5 \pm 3.5L	11.2 \pm 5.8	7.1 \pm 3.2***	9.3 \pm 3.6	11.0 \pm 5.4
Base training phase						
Training volume (km/wk)	123 \pm 39	117 \pm 39L	135 \pm 36	92 \pm 23***	126 \pm 41\$	139 \pm 34
Number of key sessions per week	3.4 \pm 1.3	3.3 \pm 1.4	3.4 \pm 1.2	3.2 \pm 1.0	3.2 \pm 1.0	3.6 \pm 1.7
Competition season						
Season length (weeks)	9.3 \pm 3.8	9.3 \pm 3.7	9.5 \pm 4.2	10.9 \pm 1.6***	9.8 \pm 3.8 ##	3.8 \pm 3.3
Number of serious races within season	5.1 \pm 2.2	5.1 \pm 2.2	4.9 \pm 2.6	5.6 \pm 1.6*	5.2 \pm 2.6	3.0 \pm 2.1
Preparation for competition						
Season length (weeks)	4.5 \pm 2.6	4.2 \pm 2.7	4.9 \pm 2.6	N/A	3.3 \pm 1.8	5.1 \pm 2.8
Number of peaks per year	2.0 \pm 0.7	2.0 \pm 0.6	2.0 \pm 0.8	N/A	1.7 \pm 0.9	2.1 \pm 0.6
Number of serious races within a peak	1.7 \pm 0.9	1.9 \pm 0.9	1.5 \pm 1.0	N/A	2.0 \pm 1.3	1.6 \pm 0.8
Training volume during competition season/ preparation for competition (km/wk)	115 \pm 48	109 \pm 52	123 \pm 41	73 \pm 19*** aaa	118 \pm 46\$\$\$ aaa	137 \pm 47

Values are means \pm standard deviations. IAAF score, International Association of Athletics Federations scoring tables 2017; MidD, middle distance (800, 1500 m); TrackD, track distance (3000 m steeplechase to 10000 m); RoadD, road distance (marathon, race walks). N/A, not applicable (i.e., these questions were not part of the survey for this population). $Ep < 0.05$ significant difference between sexes; * $p < 0.05$, *** $p < 0.001$ significant difference between MidD and RoadD; # $p < 0.05$, ## $p < 0.01$, ### $p < 0.001$ significant difference between TrackD and RoadD; \$ $p < 0.05$, \$\$ $p < 0.01$, \$\$\$ $p < 0.001$ significant difference between MidD and TrackD; aaa $p < 0.001$ significant difference compared to base training phase.

included in the final analysis. Fifty-two were classified as high and 52 as lower caliber. Training volume groupings resulted in 33, 31 and 31 athletes classified as low, moderate and high volume, respectively. The majority of responses came from athletes born in United States (25%), Canada (21%), and Japan (13%), while the rest were from Australia and New Zealand (12%), the Nordic countries (10%), Western Europe (14%), South America (4%), and Africa (1%). Training/competition phase specific training and racing characteristics for all athletes pooled, and for specific sex- and event-subgroups, are shown in **Table 1**. It is worth noting that while the difference in training volume was significant between males and females, this difference is likely to disappear if training volume were to be assessed by minutes of total training. For example, most elite males complete

training at around 3:30 min/km pace while most elite females complete their training at around 4 min/km pace. Therefore, if this assumption was adjusted for in the results, the total training time for males (~472 min/week) and females (~468 min/week) would be almost equal. MidD and TrackD athletes showed meso-periodization of training volumes, whereby training volume was significantly less during the competition season compared to the base training phase (**Table 1**). This variation in training load was absent among RoadD, who reported equal training volumes between base training and preparation for competition. However it should be noted that for RoadD athletes, preparation for competition included the ~8 weeks before a key race, where training volumes might be maintained at a relatively high levels until ~2 weeks before the race. This is different to track





athletes, whose competition season may be extended over several weeks/months (10–11 weeks; **Table 1**) and where the athlete is likely to take part in frequent racing across this time period.

The results of the survey are summarized in the following figures: **Figure 1** (Overall dietary practices across the year); **Figure 2** (Eating on hard training days); **Figure 3** (Eating on easy training days); **Figure 4** (Fueling and recovery around key sessions); **Figure 5** (Training in the fasted state); **Figure 6** (Periodic CHO restriction); **Figure 7** (Ingestion of CHO during training); **Figure 8** (Major nutrition strategies implemented during competition season or preparation for competition); **Figure 9** (Nutrition in the 24–48 h before the race day); **Figure 10** (Nutrition on the race day); **Figure 11** (Nutrition during the race). Key themes that emerged from these data are now discussed.

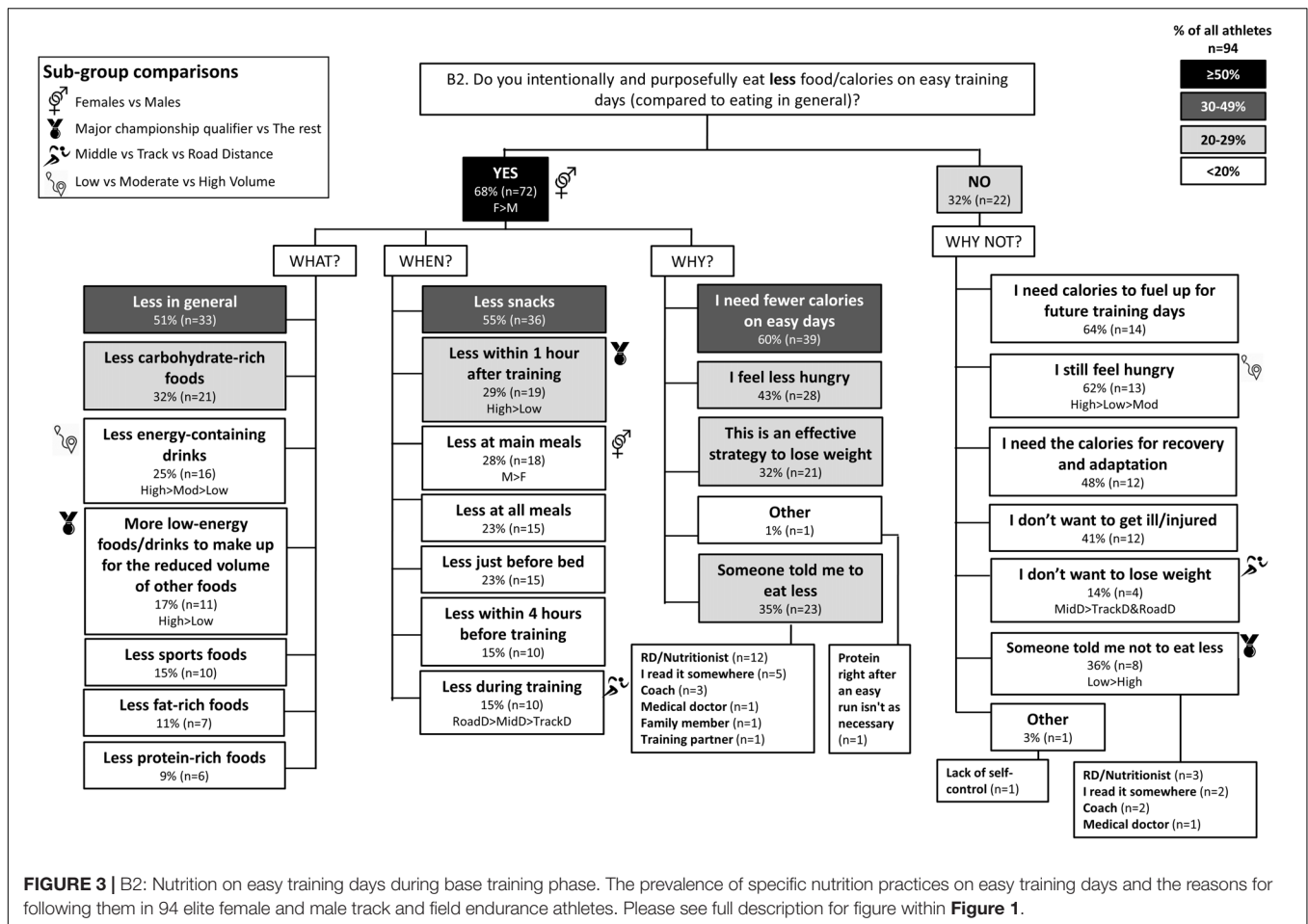
Theme 1: Road-Distance Athletes Utilize Much Greater Extremes of CHO Availability (Low to High) During Training Compared to Middle- or Track-Distance Athletes

A chronically high CHO diet was significantly more common among RoadD (46%) compared to TrackD [6%; $F(2) = 10.195$, $p = 0.009$] or MidD (10%) athletes (**Figure 1**). RoadD (62%) were

also more likely to train fasted during base training compared to MidD (30%; $p = 0.032$) (**Figure 5**). Similarly, purposeful CHO restriction was more popular among RoadD (57%) compared to MidD (30%; $p = 0.047$) (**Figure 6**). In terms of training with high CHO availability, ingestion of CHO during workouts in the base training phase was more common among RoadD (90%) compared to TrackD (37%) or MidD (48%; $p < 0.001$), mainly to practice race fueling [22% MidD vs. 38% TrackD vs. 67% RoadD, $F(2) = 6.534$, $p = 0.038$] (**Figure 7**). A higher number of RoadD (97%) and TrackD (85%) compared to MidD (57%) reported following a special diet in the 24–48h period preceding the race (MidD vs. RoadD, $p < 0.001$; MidD vs. TrackD, $p = 0.039$) (**Figure 9**). During this time, a low residue diet (i.e., low in fiber and whole foods) was more popular among RoadD (34%) compared to TrackD [5%; $F(2) = 8.546$, $p = 0.021$] or MidD (8%). Most RoadD (97%) compared to two-thirds of MidD (67%; $p = 0.009$) reported to follow a special diet on race day (**Figure 10**).

Theme 2: Middle-Distance Athletes Focus on Nutrition Strategies to Manipulate Physique

During the base training phase, MidD (32%) were more likely to report eating more on hard training days to prevent weight loss, compared to RoadD [5%; $F(2) = 7.181$, $p = 0.022$], with no



difference to TrackD (13%) (Figure 2). Furthermore, compared to TrackD (0%) and RoadD (0%), MidD (40%) were more likely to avoid eating less on easy training days because they did not want to lose weight [$F(2) = 8.512, p = 0.023$] (Figure 3). MidD (80%) were also more likely than TrackD (52%) or RoadD (46%) to focus on eating after key sessions to help retain/build muscle mass [$F(2) = 6.349, p = 0.042$ between MidD and RoadD] (Figure 4). MidD included individuals (19%) that had never heard of fasted training before [compared to 0% of TrackD and RoadD; $F(2) = 6.267, p = 0.044$]. During the competition season, MidD were more likely to focus on CHO restriction compared to RoadD [38% vs. 9%, $F(2) = 7.574, p = 0.023$] (Figure 8).

Theme 3: Females Are More Conscious of Intake of Extra Energy/CHO Than Males

A higher proportion of males than females [47% vs. 16%, $X(1) = 5.287, p = 0.022$] reported to follow a chronically high CHO diet (Figure 1). In addition, more females [79%; $X(1) = 7.412, p = 0.006$] than males (52%) reported eating less on easy days during the base training phase (Figure 3). CHO intake during training was more popular among males (79%) than females (53%; $p = 0.004$) (Figure 7). In the acute time

period preceding the race day, more males than females reported following a high energy diet [36% vs. 15%; $X(1) = 4.151, p = 0.042$] (Figure 9). Also, 76% females vs. 94% males [$X(1) = 4.187, p = 0.041$] follow a special diet on race day (Figure 10). However it should be noted that 65% of males, compared to 29% of females, identified themselves as RoadD, which has most likely influenced the outcomes.

Theme 4: Performance Is the Main Reason Behind Nutrition Strategies; Meanwhile Less Is Known About Nutrition for Adaptation

Overall, the quality of performance during training and racing seem to be the main driving factors behind the decision making when choosing a specific nutrition strategy. This theme is present in nutrition practices overall (Figure 1), as well as throughout specific micro, meso and macro levels of training and competition (Figures 2–11). Other common explanations for choosing a specific nutrition strategy were efforts to manipulate body composition (Figures 3–6), stay healthy/free of injuries (Figure 4), practicality (e.g., training before breakfast for time-management purposes; Figure 5), and because someone (usually a dietitian) told the athlete to do so. A substantial proportion

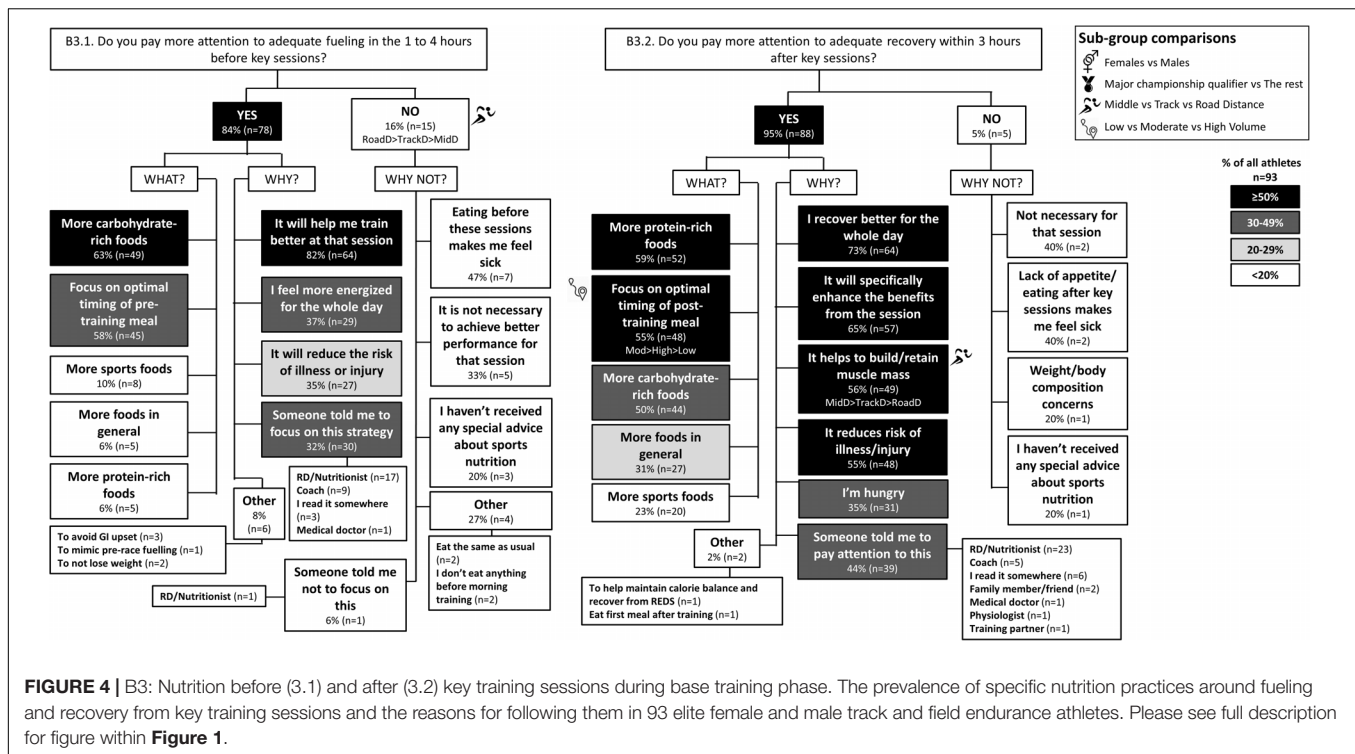


FIGURE 4 | B3: Nutrition before (3.1) and after (3.2) key training sessions during base training phase. The prevalence of specific nutrition practices around fueling and recovery from key training sessions and the reasons for following them in 93 elite female and male track and field endurance athletes. Please see full description for figure within Figure 1.

of athletes seemed to be unaware of the usefulness of specific strategies to enhance training adaptation via periodically training in the fasted state and/or restricting CHO intake around training sessions (Figures 5, 6).

Other Key Findings

Nearly half of the athletes (44%) reported following a periodized CHO diet over the annual training program (Figure 1), while ~one fifth of the athletes followed gluten free diets (19%) and vegan/vegetarian diets (17%). Even less common were diets emphasizing low CHO intake such as Paleo (0%), low CHO high fat diet (2%), restricted calorie (6%) and high protein low CHO (10%).

Overall, most athletes (92%) reported eating more on hard training days (Figure 2), including more CHO-rich foods (55%) and more in general (54%), while 68% reported adjusting nutrition intake to match lower energy expenditure on easy training days (Figure 3), including less in general (51%) and fewer CHO-rich foods (32%). Nutritional strategies to prepare for key training sessions were important to most (84%) athletes with key themes of the choice of CHO-rich foods (63%) and timing (58%). Meanwhile, 95% of athletes prioritized recovery after these sessions with key themes of choosing protein-rich (59%) or CHO-rich (50%) foods or considering the timing of intake (55%) (Figure 4). Fasted training (48%; Figure 5) and periodic CHO restriction (44%; Figure 6) were practiced by almost half of the athlete cohort, with the main rationale being weight loss (42 and 53%, respectively). More than half of the athletes (58%) reported consuming CHO during workouts, with the focus on key sessions 1–2 times a week (56%) and to maintain training intensity (74%)

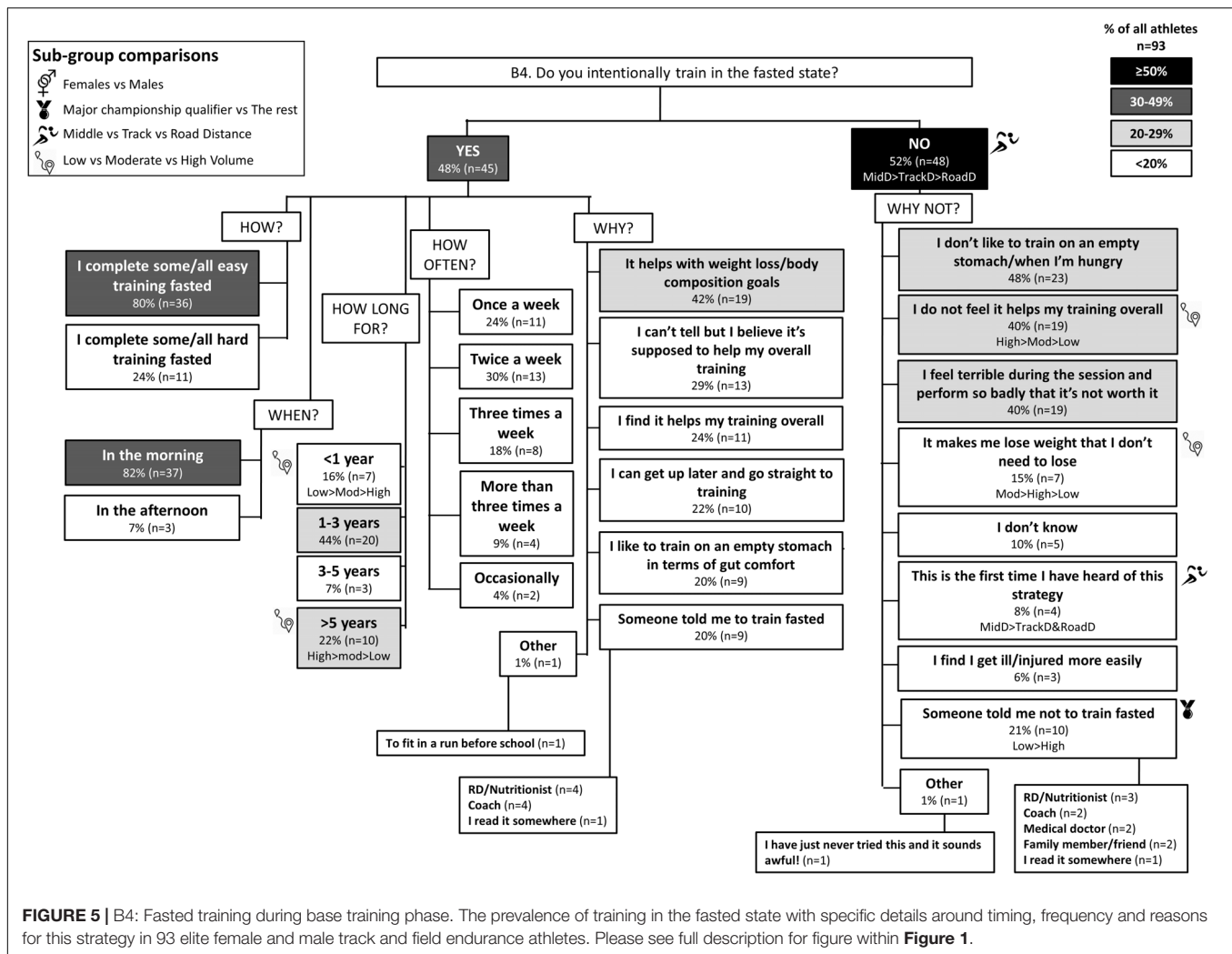
(Figure 7). Competition nutrition strategies focused mainly on adequate CHO and fluid intake before (Figure 9) and on race day (Figures 10, 11) as well as on low residue diet throughout this time period. A number of athletes further explained their dietary choices, A selection of noteworthy athlete quotes are in Table 2.

Sources of Information

One third (32%) of all athletes relied on a sports dietitian/nutritionist for nutrition advice. Of these, nearly half (43%) were MidD, while only 37% of TrackD and 20% of RoadD relied on this source of information [$X(1) = 9.751$, $p = 0.008$ between MidD and RoadD]. Other sources of information were less popular and included: coach (15%), read it somewhere (13%), training partner/a friend (5%), medical doctor (4%), physiologist (2%), naturopath (2%), and family member (1%), with no meaningful differences between subgroups.

DISCUSSION

This study aimed to characterize self-reported dietary periodization across macro (general practices across the annual cycle), meso (training and racing phases) and micro (between- and within-day) phases of training and competition in a large cohort of elite female and male middle- and long-distance runners and race walkers. We detected a number of key repeated themes across various levels of training periodization, including: (1) Road athletes reported different nutritional practices to middle- and track-distance athletes, by including strategies of training with both low and high CHO availability within the annual training plan; (2) Middle-distance athletes were the most



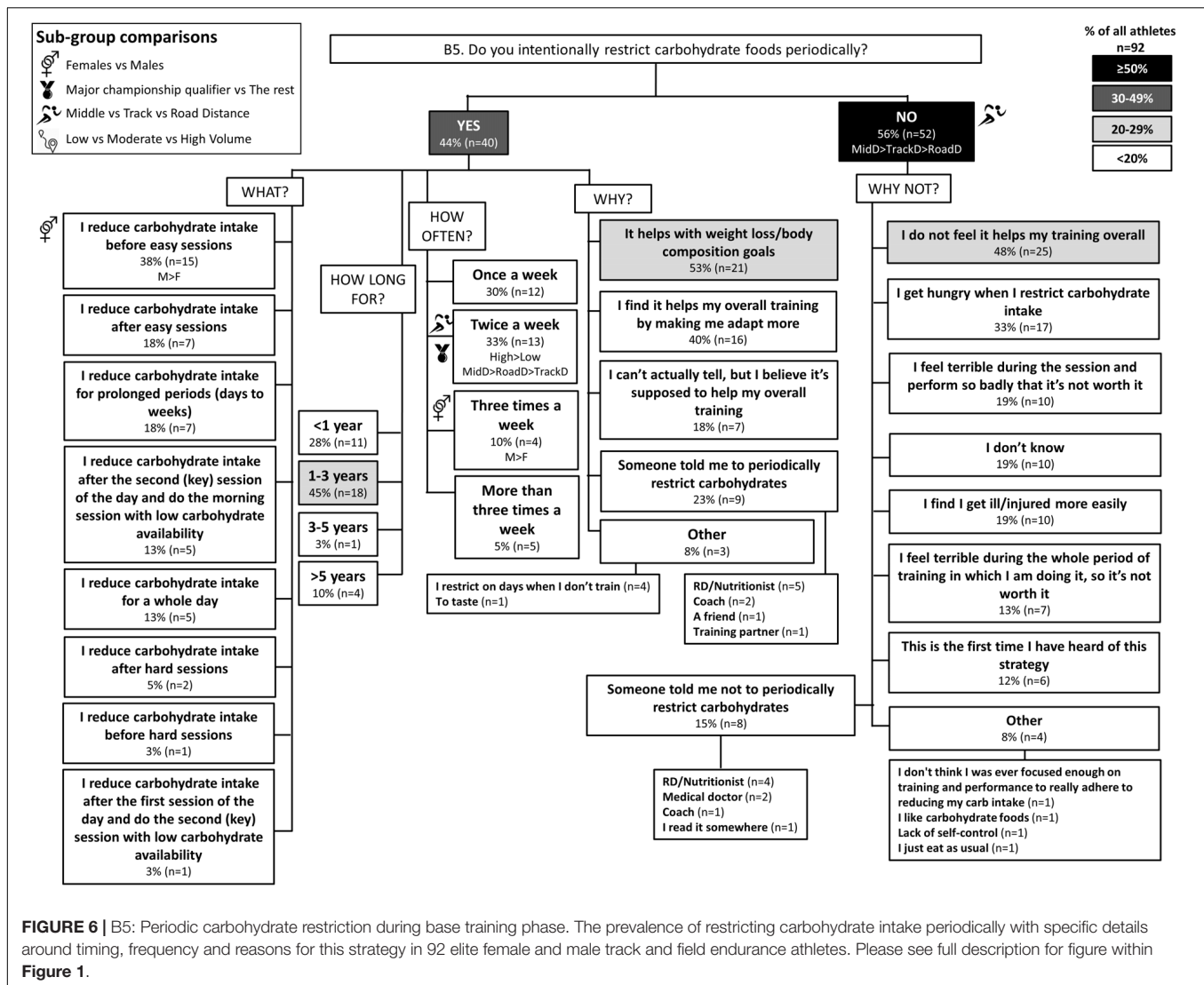
conscious about the effects of nutrition strategies on physique outcomes; (3) Females seemed to be more conscious of intake of extra energy/CHO compared to males; (4) Overall, training and race performance appeared key factors influencing nutrition choices, while themes such as body composition manipulation, health, and practicality were less important; (5) Many athletes within this cohort of high level athletes were unaware of the use of nutrition to manipulate training adaptations, or felt that there were side-effects or challenges that prevented their use.

Theme 1: Road-Distance Athletes Periodize CHO Availability Across the Year

Historically, nutrition guidelines for endurance athletes have focused on strategies to habitually achieve high CHO availability to support performance and recovery around training and races (Coyle, 1991). Protocols that supply sufficient CHO fuels to meet the demands of prolonged and/or high-intensity endurance sessions, such as consuming sufficient CHO to refuel glycogen stores prior to an event, including CHO loading for

events > 90 min (Hawley et al., 1997), a CHO rich meal in the hours before exercise (Coyle, 1991) or CHO intake during prolonged exercise according to the duration and mode of exercise (Stellingwerff and Cox, 2014) can enhance performance by ~2–3%. Contemporary recommendations support high CHO availability for competition, as well as for key training sessions in the athlete's program in which high-intensity performance needs to be completed at the highest quality possible, or in which race nutrition strategies need to be practiced. In the current study, 90% of road-distance athletes (marathon runners and race walkers) reported strategies of ingesting CHO during workouts in the base/endurance training phase (**Figure 7**), while 89% focus on CHO intake during racing (**Figure 11**). Indeed, it seems that this cohort of elite road-distance athletes are aware of, and aim to, follow current sports nutrition guidelines that emphasize optimal CHO intake around key training and racing (Thomas et al., 2016). On the contrary, and as expected, these strategies were less important for athletes competing in shorter distance events where endogenous CHO fuel stores are not limiting.

Meanwhile, more recent studies have focused on the adaptation and performance effects of strategically and

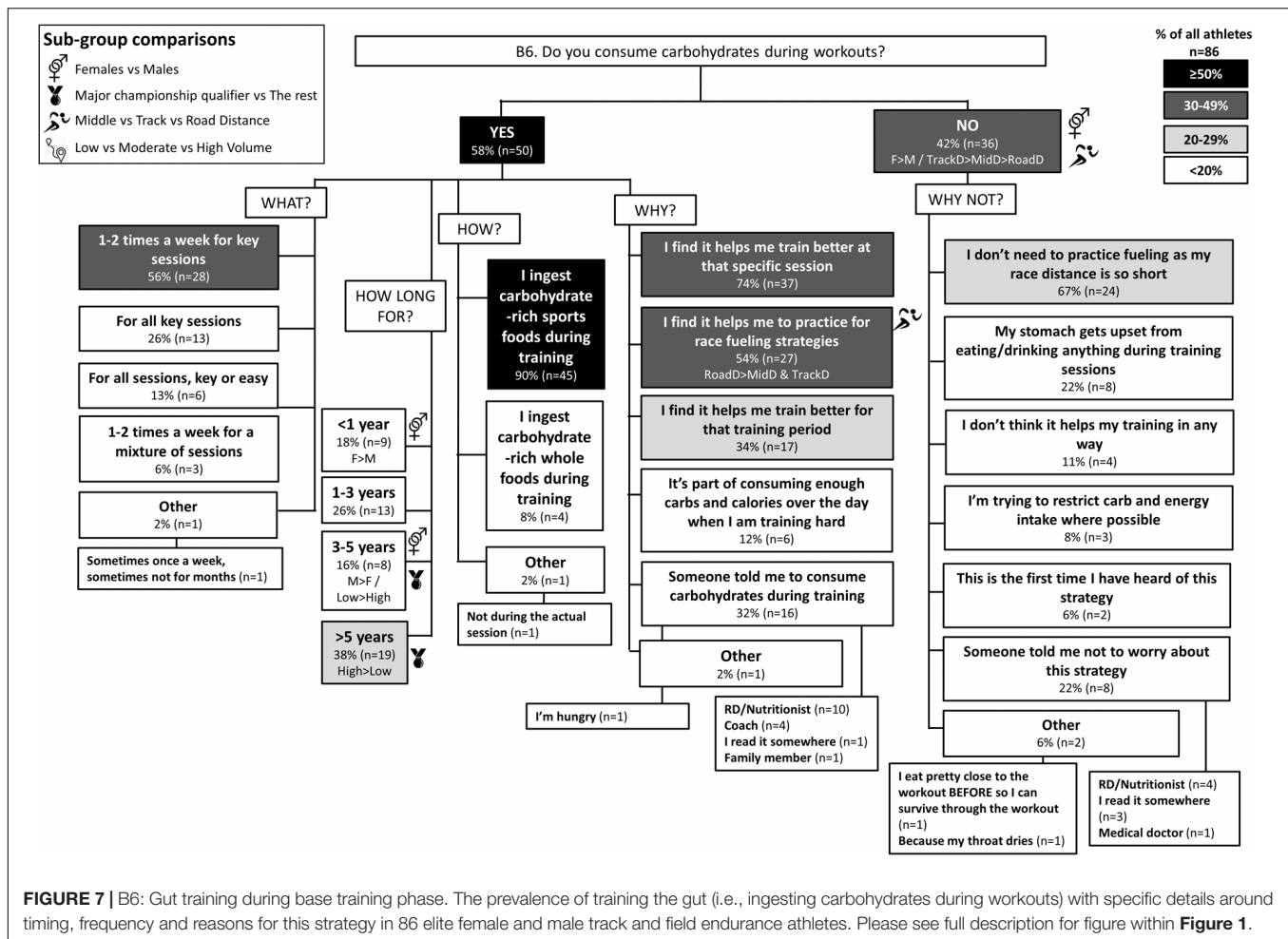


periodically implemented low CHO availability before, during, or after exercise (Bartlett et al., 2015; Hearnis et al., 2018; Impey et al., 2018). These studies suggest that occasional and strategic training with low CHO availability increases the cell signaling and gene expression responses that are usually seen after endurance training, thereby leading to further enhanced endurance capacity and performance. Possible strategies, as detailed in a recent commentary of definitions and proposed outcomes (Burke et al., 2018) include fasted training (Figure 5), CHO restriction between the first and the second session of the day (Figure 6), CHO restriction during prolonged exercise, and CHO-restricted recovery overnight. While these strategies and their potential outcomes are intriguing, studies in elite athletes have failed to show direct performance benefits (Burke et al., 2017; Gejl et al., 2017). Furthermore, studies on bone and iron health suggest these strategies may impair bone and iron metabolism, possibly leading into increased bone breakdown (Sale et al., 2015) and decreased iron levels (Badenhorst et al., 2015). Therefore, careful day-to-day periodization is likely

required, where low CHO availability is primarily scheduled around low intensity sessions (Hearnis et al., 2018). In the current study, 62% of road-distance athletes reported undertaking some training sessions in a fasted state, while this strategy was only half as popular among middle-distance athletes (30%; Figure 5). Weight loss was the most popular reason for training fasted (42% of athletes), while only 29% of those who practiced this believed that it might help with training adaptations.

Theme 2: Middle-Distance Athletes Focus on Optimal Physique

A more recent advancement in the field is periodization of body composition (Stellingwerff, 2018), which refers to the manipulation of body composition (via a mixture of nutrition and training strategies) for optimal health and performance. The underlying idea is that race weight should not be maintained year-round, as this is likely to require chronic periods of low energy availability (EA) and its related impairments of several



health and performance related measures (Mountjoy et al., 2018). Therefore, EA may need to be periodized across the year, with emphasis on higher EA levels during heavy training and altitude camps, and lower EA during lower training volumes and closer to the competition season. In addition to this macro and meso periodization of EA, emerging evidence suggests that within-day EA (micro level periodization) has also significant health consequences (Deutz et al., 2000; Fahrenholtz et al., 2018; Torstveit et al., 2018). Indeed, timing of energy intake around exercise (as opposed to “backend loading” with the majority of energy intake consumed in the evening) may be a powerful tool to manipulate physique while maintaining health. In the current study, middle-distance athletes reported more attention to the effects of nutrition strategies on physique outcomes; however, their chief focus was to build and maintain lean mass. For example, 40% of middle-distance athletes reported a maintenance of their food intake on easy training days to avoid weight loss (Figure 3). In addition, these athletes focused on nutritional support immediately after key workouts to maintain/build muscle mass (Figure 4). During the competition season, however, middle-distance athletes were more likely to report a reduction in CHO intake or use of CHO restriction strategies (Figure 8), which may reflect a relative reduction in training volume and/or

their efforts to reduce body mass to achieve an optimal race weight.

Theme 3: Females Are More Conscious of Intake of Extra Energy/CHO Than Males

Females and males have an equal ability for CHO storage and utilization during exercise if energy availability is adequate (Tarnopolsky et al., 2001; Wallis et al., 2006). However, female distance athletes tend to eat less CHO than males (Burke et al., 2001), although this difference is likely to disappear when CHO intake is adjusted to training volume (Heikura et al., 2017a), as recommended by current guidelines (Thomas et al., 2016). Regardless of equal (relative) energy and CHO needs for female and male athletes, dietary practices of females tend to be more cautious of extra energy/CHO intake. Indeed, females are more likely to suffer from eating disorders (Sundgot-Borgen and Torstveit, 2004). This may be due to a higher frequency of body image issues/concerns over body weight among female athletes (Martinsen et al., 2010). In the current study, we showed similar patterns of calorie/CHO awareness among elite distance athletes as has been previously reported in sub-elite athlete populations.

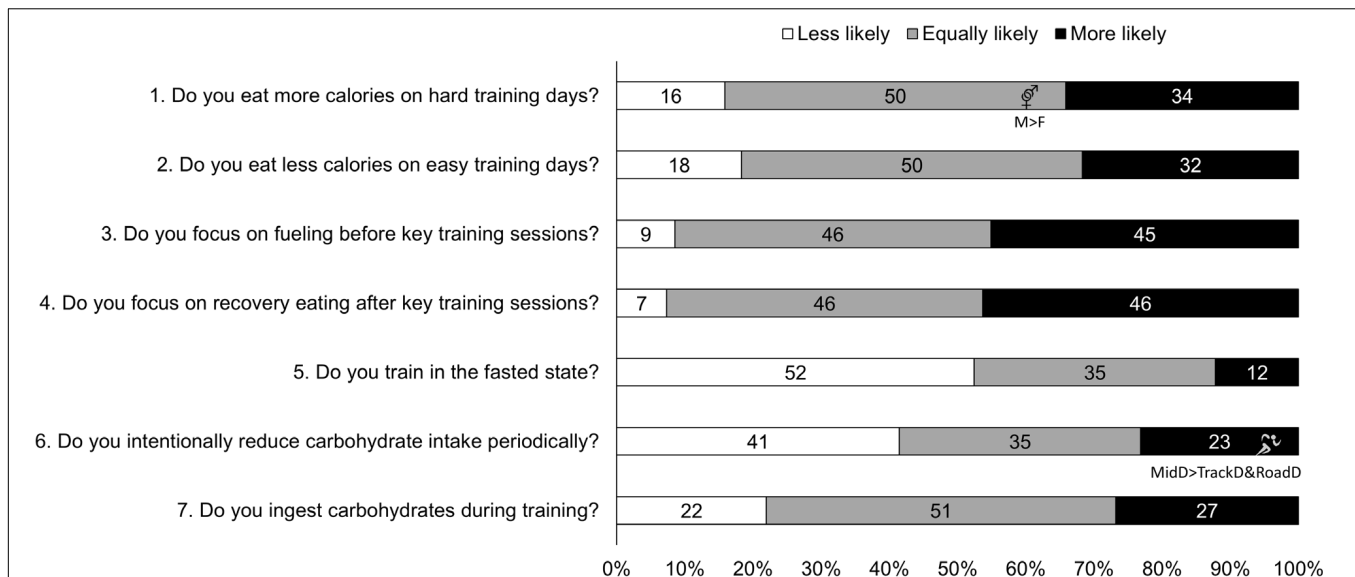


FIGURE 8 | C: Major nutrition strategies implemented during competition season and preparation for competition as compared to base/endurance phase. Answers to Part C: “Compared to nutrition during base/endurance training phase, how much do you focus on the following dietary strategies during competition season (track athletes) or preparation for competition (road athletes)?” Values are percentages of all athletes ($n = 83$): white bars, less likely; gray bars, equally likely; black bars, more likely to follow this strategy. Symbols have been used to reflect significant ($p < 0.05$) between-group differences between sexes (vector sex symbol), and distance event (runner symbol). Where significant differences were detected, the symbols are combined with a brief description of direction of difference, for example, M > F reflects a higher % of males (M) compared to females (F) for that answer. MidD, Middle Distance (800 and 1500 m); TrackD, track distance (3000 m steeplechase to 10,000 m); RoadD, road distance (marathon and race walks).

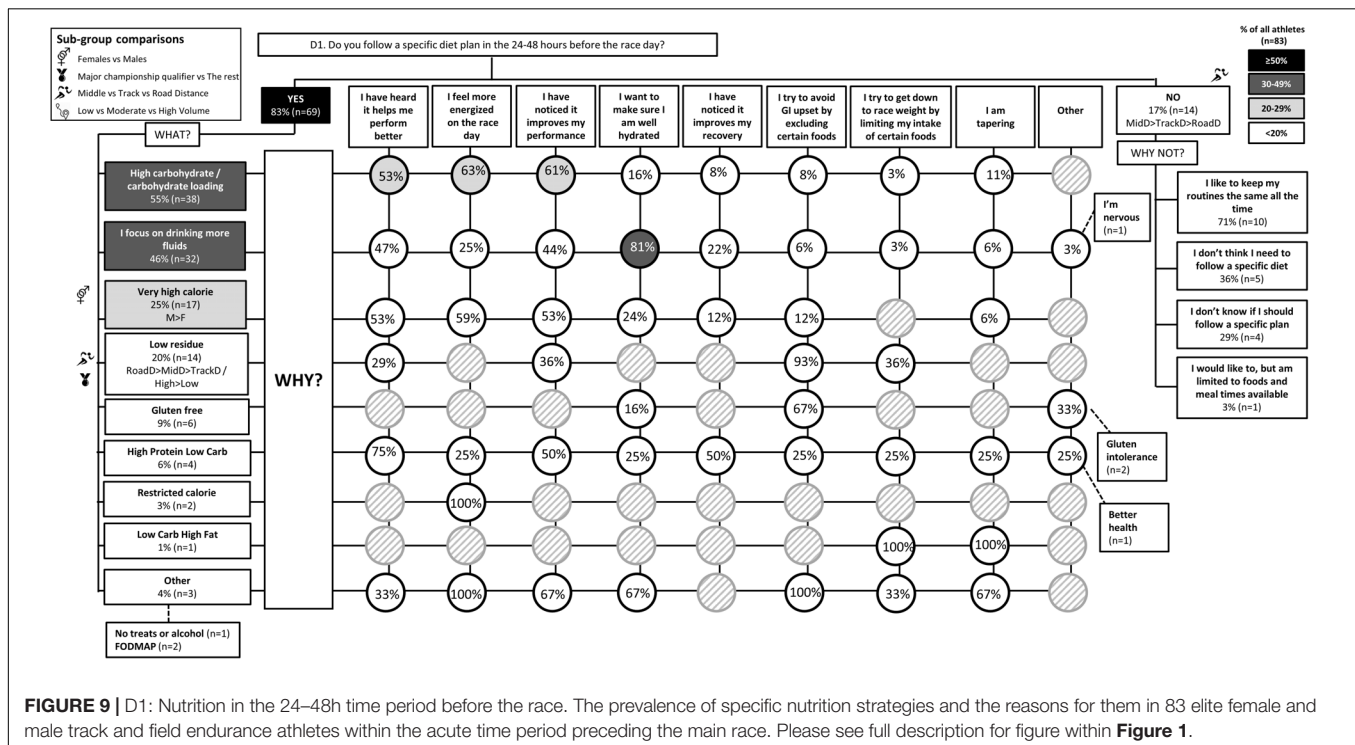


FIGURE 9 | D1: Nutrition in the 24–48h time period before the race. The prevalence of specific nutrition strategies and the reasons for them in 83 elite female and male track and field endurance athletes within the acute time period preceding the main race. Please see full description for figure within **Figure 1**.

Namely, male athletes were more likely to follow a chronically high CHO diet (**Figure 1**). In addition, a greater proportion of females (79%) than males (52%) reported eating less on easy days during the base training phase (**Figure 3**). Males were

also more likely to follow a high energy diet in the acute time period preceding the race day (**Figure 9**). Although qualitative, these outcomes suggest that female athletes may indeed be more concerned about consuming extra energy/CHO however whether

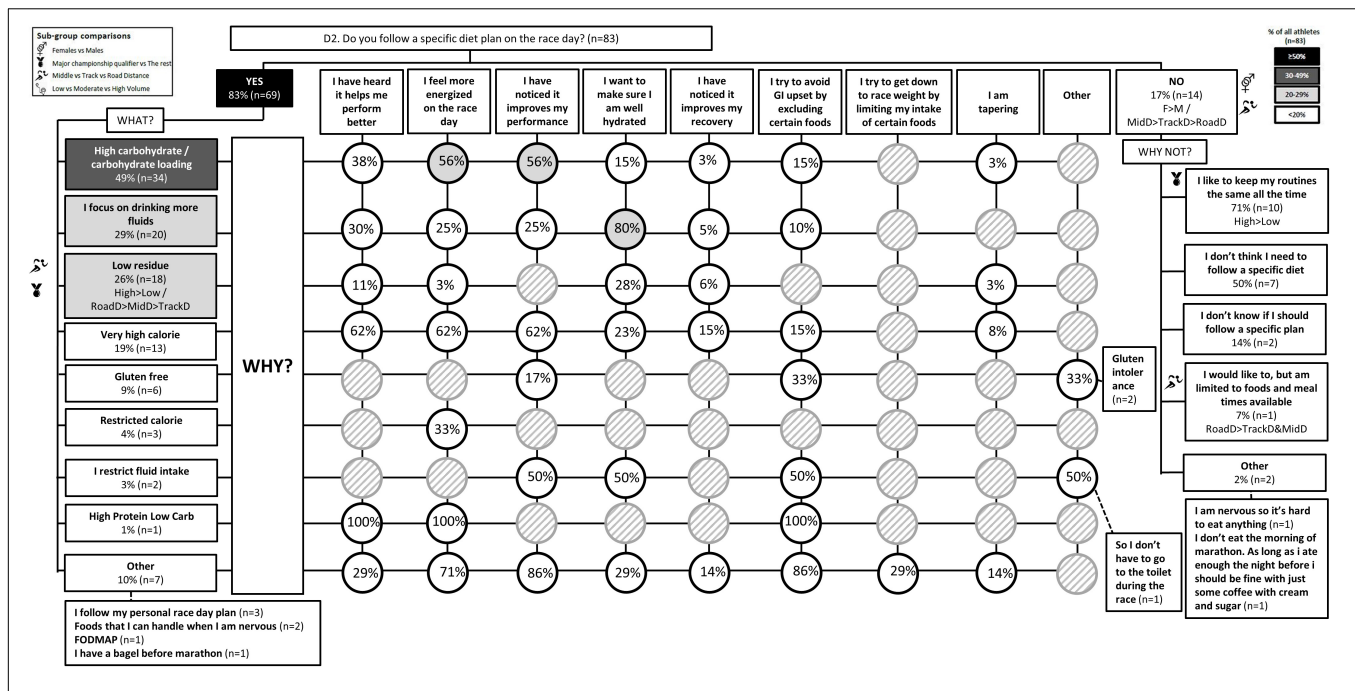


FIGURE 10 | D2: Nutrition on race day. The prevalence of specific nutrition strategies and the reasons for them in 83 elite female and male track and field endurance athletes on the day of the main race. Please see full description for figure within **Figure 1**.

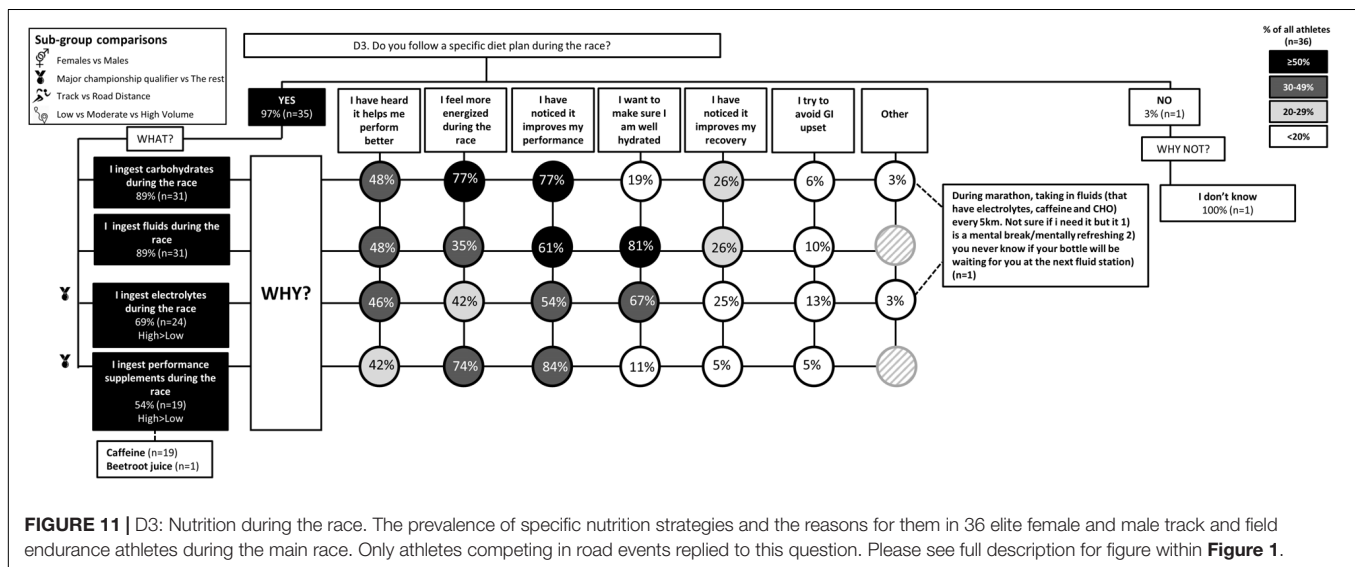


FIGURE 11 | D3: Nutrition during the race. The prevalence of specific nutrition strategies and the reasons for them in 36 elite female and male track and field endurance athletes during the main race. Only athletes competing in road events replied to this question. Please see full description for figure within **Figure 1**.

the reasons are justified due to a lower fuel requirement, or related to eating disorders/disordered eating, lack of knowledge, or other factors, cannot be concluded based on the current survey.

Theme 4: Nutrition Strategies Are Based on Performance Rather Than Adaptation

Contrary to previous guidelines (Coyle, 1991), more recent sports nutrition guidelines have incorporated the value of specialized strategies to optimize adaptations to training, noting that these

protocols may often be contradictory for acute performance outcomes or other health goals, and need to be carefully integrated into the various phases of the annual plan (Thomas et al., 2016). We were interested to identify whether these concepts were understood by elite athletes and used to inform their various nutrition strategies. Our results suggest that the most common nutrition strategies reported by this large cohort ($n = 104$) of elite track and field distance athletes (of whom 50% were qualifiers for World Championships and/or Olympic Games) were focused on performance enhancement during training (**Figures 2–4, 7**) and competition (**Figures 9–11**).

TABLE 2 | Selection of noteworthy athlete quotes regarding why they do, or do not, follow a specific nutrition strategy.

Nutrition strategy	Reason for following/not following this strategy
B3: Why do you focus on pre-key session fueling?	<i>"To help maintain calorie balance and recover from REDS."</i>
B4: Why do you not train in the fasted state?	<i>"That will put me so far behind in terms of energy intake which I can't afford." "I'm very lean to begin with - I don't think I'd make it through!" "I do not believe this approach is scientifically valid."</i>
B5: Why do you restrict carbohydrate intake periodically?	<i>"I think there is something to making your body more insulin sensitive." "It seems like a bad idea. My understanding is that the body uses carbs as primary fuel source."</i>
B1: Why do you eat more food/calories on hard training days?	<i>"I feel like I have earned it."</i>
B5: Why do you not restrict carbohydrate intake periodically?	<i>"I like carbohydrate foods." "Lack of self-control."</i>
D1: Why do you focus on drinking fluids in the 24–8 h before the race day?	<i>"I am nervous."</i>
D2: Why do you not follow a specific diet on the race day?	<i>"I am nervous so it's hard to eat anything."</i>

Meanwhile less was known about specific strategies to further stimulate cellular adaptations to exercise (Figures 5, 6). Indeed, many athletes lacked understanding of the periodization of strategies to train with low CHO availability, furthermore, others were either skeptical of their value, concerned about perceived or actual disadvantages particularly related to illness or injury, or practicing some aspects within their routines by accident. Since several outcomes identified the interest in using nutrition to manipulate body composition and/or to prevent illness/injuries, we conclude that the general priority for decisions around nutrition was performance > health > enhanced adaptation.

Limitations

It is important to note that the current study describes self-reported nutrition practices that are implemented across the training and competition year. We have previously shown that there is a discrepancy between general descriptions of practices (reflecting a macrocycle) and actual self-recorded intakes (collected across a micro cycle) in elite distance athletes (Heikura et al., 2017a). Indeed, it is possible that self-reports such as those found in the current study, reflect either what athletes aspire to achieve or perceive that they follow rather than actual behaviors. However, this potentially perceived versus actual mismatch would hypothetically be equivalent across the various sub-groups of athletes. Furthermore, our survey questions were qualitative (i.e., describing "high" or "low" intakes instead of specific amounts) and it is possible that these relative terms are interpreted differently by different individuals. Nevertheless, our survey was based on the learnings from a pilot study (Heikura et al., 2017b) and we are confident that the expanded and improved survey tool had greater precision and sensitivity,

along with more than 100 respondents, in detecting nutrition practices across all levels of training/racing periodization.

CONCLUSION

We characterized self-reported dietary periodization across macro (general practices across the annual cycle), meso (training and racing phases) and micro (between- and within-day) phases of training and competition in 104 elite female and male middle- and long-distance runners and race walkers (50% major championship qualifiers). Our key findings suggest that: (1) Road athletes train with both low and high CHO availability within the annual training plan, while track athletes are less likely to incorporate a large spectrum of CHO availability in their training; (2) Middle-distance athletes emphasize physique when choosing a nutrition strategy; and (3) Performance appears to be the key driving factor influencing nutrition choices, while themes such as body composition manipulation, health, and practicality are less important. Overall, our findings indicate that elite track and field distance athletes are aware of and report following the current sports nutrition guidelines in terms of high CHO availability around key training sessions and during racing. However, most of this cohort appears to be unaware of and/or unwilling to aggressively incorporate the more recent strategies of training with reduced CHO availability to support training adaptations.

ETHICS STATEMENT

This is a survey study which participants completed via an online survey tool. Consent to participate was completed via ticking in a box. The participants who proceeded to complete the survey were thus seen as consenting to participate in research.

AUTHOR CONTRIBUTIONS

IH, TS, and LB designed the study, developed the survey, recruited the participants, and prepared the manuscript. IH collected, organized, and analyzed the data. All authors approved the final manuscript.

ACKNOWLEDGMENTS

The authors would like to thank colleagues, coaches, and athletes for their assistance during the recruitment process. A special thank you goes to all athletes who participated in the study.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fphys.2018.01732/full#supplementary-material>

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Selective Influences of Maximum Dynamic Strength and Bar-Power Output on Team Sports Performance: A Comprehensive Study of Four Different Disciplines

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OPEN ACCESS

Edited by:

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 26 September 2018

Accepted: 05 December 2018

Published: 17 December 2018

Citation:

Loturco I, Suchomel T, James LP, Bishop C, Abad CCC, Pereira LA and McGuigan MR (2018) Selective Influences of Maximum Dynamic Strength and Bar-Power Output on Team Sports Performance: A Comprehensive Study of Four Different Disciplines. *Front. Physiol.* 9:1820. doi: 10.3389/fphys.2018.01820

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This study examined the selective influences of one-repetition maximum (1RM) values [assessed in the half-squat (HS)] and bar-power production [assessed in both HS and jump squat (JS) exercises] on the physical performance of male and female team sport athletes from four different sports. Three-hundred and three elite players (31 Olympians) from four different disciplines (47 male soccer players, 58 female soccer players, 28 male handball players, 58 female handball players, 49 male rugby players, and 63 male futsal players) participated in this study. The physical tests were performed over 2 consecutive days for soccer and rugby players, and in 1 day for the remaining athletes. On the first day, rugby and soccer athletes performed squat jumps (SJ), countermovement jumps (CMJ), and HS 1RM. On the second day, they executed HS and JS tests (to assess the maximum bar-power output) and the linear and change-of-direction (COD) speed tests. For the other players, the sequence of the measurements was the same; however, they did not perform the HS exercise. Athletes were separated, using a median split analysis, into two distinct groups, according to their bar-power output in both JS and HS exercises and their performance in HS 1RM. The magnitude-based inferences method was used to examine the differences between “higher” and “lower” performance groups. Overall, the bar-power outputs were better connected to improved acceleration, speed, and jump performance than the 1RM measures. From these findings, it is possible to infer that players able to produce higher bar-power outputs are likely to sprint faster and jump higher. Therefore, coaches involved in team sports are strongly encouraged to use the bar-power method to evaluate the athletic performance of their players.

Keywords: muscle power, optimal loads, straight speed, sprinting, explosiveness

INTRODUCTION

Strength and power capabilities play a key role in team sports performance. Several studies have shown that stronger and more powerful players of different sports are usually capable of accelerating faster, jumping higher, and changing direction more rapidly (Wilson et al., 1993; McBride et al., 2005; Newton et al., 2006; Loturco et al., 2016b; Freitas et al., 2018). In addition, research has indicated that even more specific sport tasks such as throwing, kicking, and tackling seem to be positively influenced by the individual ability to generate greater levels of force and power (Marques et al., 2007; Kilduff et al., 2008; Loturco et al., 2014, 2016a; Dello Iacono and Seitz, 2018; Loturco et al., 2018a). Therefore, coaches and sport scientists are constantly seeking better and more accurate methods to properly improve and assess neuromuscular function in top-level athletes.

The one-repetition maximum (1RM) test is one of the most widely used measurements in the field of sport science (Fleck, 1999; Channell and Barfield, 2008; Suchomel et al., 2016a). Through this test, coaches can determine the maximum load that a subject can move during a maximum-effort resistance exercise (Loturco et al., 2018c) and thereby prescribe relative loads [i.e., 1RM percentages (% 1RM)], according to the athlete's needs and objectives (e.g., strength or power development) (McMaster et al., 2013). Many studies have reported the effectiveness of 1RM-based training programs in improving the physical performance of team sport athletes. For example, Bogdanis et al. (2009) found significant increases in maximum and relative half-squat (HS) strength, change-of-direction (COD), vertical jumping, and sprinting abilities in senior soccer players who performed 6 weeks of HS training using loads ranging from 70 to 90% 1RM. Similarly, Appleby et al. (2012) showed that a long-term periodized training model with loads from 60 to 100% 1RM resulted in significant increases in body mass (BM), lean mass index, and upper-body strength in professional rugby union players. However, despite their popularity, some authors have raised concerns over the safety and usability of 1RM tests in professional sport settings (Chapman et al., 1998; Brown and Weir, 2001; Loturco et al., 2015d), where athletes regularly perform various concurrent and complementary activities, and time and resources are inherently limited (Bishop, 2008; Bishop et al., 2017; Freitas et al., 2018).

These issues are even more pronounced in large groups of individuals, which greatly compromise the use of 1RM measurements in team sport disciplines (Loturco et al., 2015d). To minimize these possible drawbacks and optimize performance gains, we proposed the use of an alternative training and testing strategy, based on barbell power production (Loturco et al., 2018c). In this regard, instead of considering only the "maximum mass" moved in a given exercise, the "bar-power approach" reflects, at the same time, the force and velocity applied to the barbell (Loturco, 2017; Loturco et al., 2018c). With this method, practitioners can safely determine the loads capable of maximizing bar-power output, using rapid incremental loading tests or instantaneously measuring the optimum bar-velocities (Loturco et al., 2015b, 2017d). To date, although a number of studies have confirmed the efficiency of the optimum power

loads (OPL) to improve the physical performance of team sport athletes, these investigations were executed with male players of specific sport disciplines (e.g., soccer and basketball) (Loturco et al., 2017a; Dello Iacono and Seitz, 2018; Freitas et al., 2018). Knowing more about the relationships between bar-power output and the athletic abilities of both male and female athletes of different sports may lead researchers to develop new studies regarding this topic, as well as stimulate coaches to implement this strategy in their professional practices. Moreover, the possibility of comparing the magnitude of these correlations with those related to more traditional performance measures (e.g., 1RM values) could also reinforce and support the use of the OPL in high performance sport.

As such, a recent study using a pooled sample of 61 elite athletes from four different sports (i.e., track and field, rugby sevens, soccer, and bobsled) compared these mechanical relationships, revealing that the bar-power outputs are more strongly associated with linear speed and vertical jump height than 1RM values (Loturco et al., 2018c). Nevertheless, a more comprehensive investigation is warranted by reporting these data in a more specific way (i.e., with subjects grouped on a sport-by-sport basis), involving male and female players of different field and court team sports (e.g., handball and futsal) and with additional performance outcomes (e.g., COD speed). An alternative strategy for estimating the influence of a given exercise on performance is examining the data provided by the median split analysis (Rampinini et al., 2007; Iacobucci et al., 2015). Based on this method, practitioners can group the athletes according to their physical skills, defining the lower and upper bounds of performances in a series of assessments. Under this rationale, it seems plausible to consider that superior levels of performance in two or more measurements might be closely interconnected, representing shared and direct relations between them (Loturco et al., 2017f).

Thus, the aim of this study was to test and compare the interconnection between bar-power output [collected in the HS and jump squat (JS) exercises] and 1RM values (collected in HS) and a variety of sport-specific performance measures (i.e., linear speed, COD, acceleration and jump abilities) in male and female elite players of four different sports (rugby, soccer, futsal, and handball).

MATERIALS AND METHODS

Participants

Three-hundred and three elite athletes (47 male soccer players, 58 female soccer players, 28 male handball players, 58 female handball players, 49 male rugby players, and 63 male futsal players) from four different sports participated in this study. The characteristics of the subjects are presented in **Table 1**. Male soccer players participated in the first division of the Paulista State Championship. Female soccer players participated in the first division of the Brazilian National Championship and won the 2017 Libertadores da America Cup. Male and female handball players participated in the first division of the Brazilian National Championships, comprising 39 (15 male and 24 female) athletes

TABLE 1 | Characteristics of the subjects (mean \pm standard deviation) of the four different sports disciplines.

	Male Soccer	Female Soccer	Male Handball	Female Handball	Rugby	Futsal
Age (years)	22.5 \pm 2.9	22.6 \pm 7.6	28.3 \pm 3.2	25.2 \pm 4.3	24.4 \pm 4.2	23.5 \pm 3.3
Weight (kg)	71.2 \pm 8.8	61.0 \pm 7.6	90.3 \pm 10.3	69.7 \pm 7.3	88.8 \pm 10.0	73.6 \pm 6.9
Height (cm)	177.1 \pm 7.6	166.4 \pm 6.9	188.3 \pm 4.6	173.4 \pm 5.8	179.1 \pm 6.1	176.3 \pm 5.7

of the Brazilian National Team, and 23 (11 male and 12 female) who participated at the Rio-2016 Olympic Games. Rugby players were members of the Brazilian National Team comprising nine athletes who participated in the rugby sevens tournament at the Rio-2016 Olympic Games. Finally, futsal players won the 2016 Brazilian National League. Therefore, we can confirm the high level of performance of the participants in this study. This study was carried out in accordance with the recommendations of the Anhanguera-Bandeirante Ethics Committee with written informed consent from all subjects. All subjects gave written informed consent in accordance with the Declaration of Helsinki. The protocol was approved by the Anhanguera-Bandeirante Ethics Committee.

Study Design

The athletes involved in this study were assessed during the competitive phase of the season and were well familiarized with testing procedures due to their constant assessments in our facilities. Physical tests were performed on 2 consecutive days for soccer and rugby athletes and 1 day for the other athletes. For rugby and soccer players, on day 1, squat jumps (SJ), countermovement jumps (CMJ), and a 1RM HS were performed. Meanwhile, on day 2, the maximum bar-power outputs in the HS and JS exercises, and linear and COD sprint tests were assessed. For the other sports players the sequence of tests was the same, but they did not perform the 1RM test or the assessment of bar-power outputs in the HS exercise. Participants were required to be in a fasting state for at least 2 h, avoiding caffeine and alcohol consumption for 24 h before the procedures. Prior to the tests, the athletes performed standardized warm-up protocols including general (i.e., running at a moderate pace for 10-min followed by active lower limb stretching for 3-min) and specific workouts (i.e., submaximal attempts at each tested exercise). Between each test, a 15-min rest interval was allowed, to explain the procedures and adjust the equipment.

Vertical Jumps

Vertical jump height was assessed using the SJ and CMJ. In the SJ, athletes were required to remain in a static position with a 90° knee flexion angle for \sim 2-s before jumping, without any preparatory movement. In the CMJ, athletes were instructed to execute a downward movement followed by complete extension of the legs and were free to determine the countermovement amplitude to avoid changes in jumping coordination. All jumps were executed with the hands on the hips and the athletes were instructed to jump as high as possible. The jumps were performed on a contact platform (Elite Jump®, S2 Sports, São Paulo, Brazil) that has previously been shown to be valid and reliable (Loturco

et al., 2017e). A total of five attempts were allowed for each jump, interspersed by 15-s intervals (Loturco et al., 2017e). The best attempts for the SJ and CMJ were used for the analyses.

Maximum Dynamic Strength Test in the Half-Squat Exercise

Maximum dynamic strength was assessed using the 1RM HS test as described previously (Brown and Weir, 2001). Prior to the test, subjects executed two warm-up sets, as follows: (1) five repetitions at 50% of the estimated 1RM and; (2) three repetitions at 70% of the estimated 1RM. A 3-min rest interval was provided between all sets. After 3 min, athletes started the test and were allowed up to five attempts to achieve their 1RM (i.e., maximum weight that could be lifted once using proper technique), which was measured to the nearest 1 kg (Brown and Weir, 2001). The test was performed using Smith-machine equipment (Hammer-Strength Equipment, Rosemont, IL, United States). Values were normalized by dividing the 1RM by the athletes' BM (i.e., relative strength = kg kg⁻¹).

Bar-Power Outputs in Jump Squat and Half-Squat Exercises

Maximum bar-power outputs were assessed in JS and HS, all performed on a Smith machine (Hammer Strength Equipment, Rosemont, IL, United States). Participants were instructed to execute three repetitions at maximal velocity for each load, starting at 40% of their BM in both exercises. In the JS, participants executed knee flexion until the thigh was parallel to the ground and, after the command to start, jumped as fast as possible without their shoulders losing contact with the bar. The HS was executed in a similar fashion to the JS, except that the subjects were instructed to move the bar as fast as possible without losing foot contact with the ground, keeping their heels on the floor. In both exercises, a load of 10% of BM was progressively added for each set until a clear decrement in mean power (MP), mean propulsive power (MPP), and peak power (PP) was observed (Loturco et al., 2018b). A 5-min rest period occurred between sets. To determine the power outputs, a linear position transducer (T-Force, Dynamic Measurement System; Ergotech Consulting S.L., Murcia, Spain) was attached to the Smith machine bar and values were automatically derived by the custom-designed software as follows: MP-value calculated during the entire concentric phase of each repetition; MPP – value calculated during the propulsive phase, defined as that portion of the concentric action during which the measured acceleration is greater than acceleration due to gravity; PP – the highest bar-power value registered at a particular instant (1-ms) during the concentric phase (Sanchez-Medina et al., 2010, 2014). The bar

position data were sampled at 1000 Hz. The maximum MP, MPP, and PP values obtained in each exercise were used for analysis. Values were normalized by dividing the absolute power by the athletes' BM (i.e., relative power = $W \text{ kg}^{-1}$) to produce more consistent relationships with athletic performance and allow for comparison with previous research (Cronin and Hansen, 2005; Cormie et al., 2007, 2010; Loturco et al., 2018c).

Linear Sprint Tests

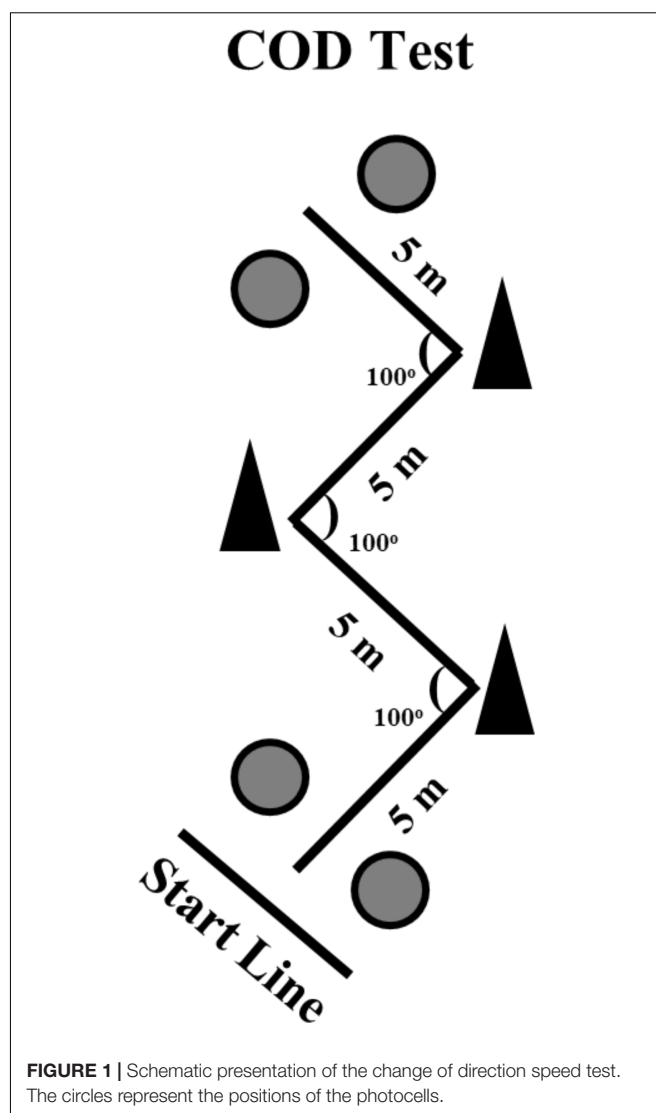
For the sprint test, rugby players performed a 40-m sprint test, whereas the other athletes sprinted over a total distance of 20-m. Four pairs of photocells (Smart-Speed, Fusion Equipment, Brisbane, QLD, Australia) were positioned at distances of zero, 5-, 10-, and 20-m along the sprinting course, and two additional pairs were placed at 30- and 40-m to assess rugby players. Sprint velocity (VEL) was calculated as the distance traveled over a measured time interval. The acceleration (ACC) capacity in the different distances (i.e., 0–5-, 5–10-, 10–20-, 20–30-, and 30–40-m) was calculated as the rate of change of velocity with respect to time. Athletes performed two sprints, interspaced by a 5-min rest interval, and the best attempt was retained for analysis.

Zig-Zag Change of Direction Speed Test

The Zig-zag COD test was performed on an indoor court and consisted of four 5-m sections (total 20-m of linear distance) marked with cones set at 100° angles (**Figure 1**) requiring the athletes to decelerate and accelerate as fast as possible around each cone. Two maximal attempts were performed with a 5-min rest interval between attempts. Starting from a standing position with the front foot placed 0.3-m behind the first pair of timing gates (Smart Speed, Fusion Equipment, Brisbane, QLD, Australia) (i.e., starting line), the athletes were instructed to complete the test as quickly as possible, until crossing the second pair of timing gates, placed 20-m from the starting line (Loturco et al., 2016c; Pereira et al., 2018). The fastest time from the two attempts was retained for further analysis.

Statistical Analyses

Data are presented as means \pm standard deviation. Data normality was tested using the Shapiro–Wilk test. Athletes were divided, using a median split analysis, into two groups according to their bar-power outputs in both exercises and HS 1RM (e.g., higher and lower JS MP, higher and lower HS MP, and higher and lower HS 1RM). The magnitude-based inferences method was used to analyze the differences between groups in the physical performance tests (Batterham and Hopkins, 2006). The magnitudes of the differences in the different performance variables were expressed as standardized mean differences [Cohen's d , effect size (ES)]. The smallest worthwhile change (SWC) was set by using the Cohen's principles for a small ES (i.e., 0.2) for each variable tested (Hopkins et al., 2009). To analyze the differences between groups, terms such as possibly and unclear were used if the 90% confidence limits (CL) crossed one or both SWC boundaries, respectively. Otherwise, if the CL did not cross SWC boundaries, the effect was inferred as probably. Additionally, the magnitudes of the standardized differences were



interpreted using the following thresholds: <0.2, 0.2–0.6, 0.6–1.2, 1.2–2.0, 2.0–4.0, and >4.0 for trivial, small, moderate, large, very large, and near perfect, respectively (Hopkins et al., 2009). The assessments used in this research presented good levels of absolute and relative reliability ($CV < 5\%$ and $ICC > 0.90$, for all tested variables) (Hopkins et al., 2009).

RESULTS

All data presented a normal distribution. **Table 2** shows the descriptive data of the vertical jumps, bar-power outputs in both JS and HS exercises, and 1RM in the HS exercise for the athletes of the different modalities assessed. **Table 3** demonstrates the results of the linear sprint and COD speed tests for the athletes of four different sports disciplines.

Figure 2 shows the Cohen's d for the comparisons between higher and lower 1RM and bar-power output groups in the SJ and CMJ height in the distinct groups of athletes. **Figure 3** depicts

the comparisons between higher and lower groups, divided based on their bar-power outputs and 1RM in the linear and COD speed tests in the athletes from the different sports disciplines. **Figure 4** demonstrates the comparisons of the acceleration results comparing higher and lower bar-power outputs and 1RM groups in the distinct groups of athletes.

DISCUSSION

This study examined the selective influences of 1RM values (assessed in HS) and bar-power production (assessed in both HS and JS exercises) on the physical performance of male and female team sport athletes of four different sports (rugby, soccer, futsal, and handball). The main results reported here are: (1) overall, the bar-power outputs (i.e., MP, MPP, and PP) were more connected to better performances in speed and power assessments (i.e., jump, linear sprint, and COD tests) than the 1RM values, and (2) the players able to generate greater levels of bar-power were, in general, able to sprint faster, jump higher, and change direction more quickly than their less powerful peers. This is the first study to show this connection for the bar-power approach in a comprehensive sample of elite team sport athletes of different team sport disciplines.

A previous investigation using the same statistical approach (i.e., median split analysis) showed similar trends in National rugby players (Loturco et al., 2017f). However, the previous study did not compare the possible influences of bar-power outputs and 1RM measures on athletic performance. Even so, in line with the current findings, the results indicated that players capable of generating more power in the JS were equally capable of performing better in jump, COD, and sprint tests. In contrast, also in line with our data, higher performances in HS were not connected to superior performance in any functional assessment. As such, the novelty of including the 1RM measurement in this research was not able to increase the selective influence of HS exercise on athletic performance of elite rugby players. Although it is clear from the literature that the maximum dynamic strength plays a critical role in rugby performance (Argus et al., 2012; Comfort et al., 2012; McMaster et al., 2014), at least in these specific motor tasks (i.e., jump, acceleration, high-speed, and COD efforts), the HS 1RM measurement was not sensitive enough to differentiate national team rugby players with distinct physical performance levels. These results partially confirm and extend previous observations showing that: (1) the 1RM values are less related to sprint and jump performance than the power-related variables (Baker and Nance, 1999; Cunningham et al., 2013; Loturco et al., 2018c), and (2) the HS exercise seems not to

TABLE 2 | Descriptive results of the vertical jumps, bar-power outputs, and one repetition maximum in the athletes of four different sports disciplines.

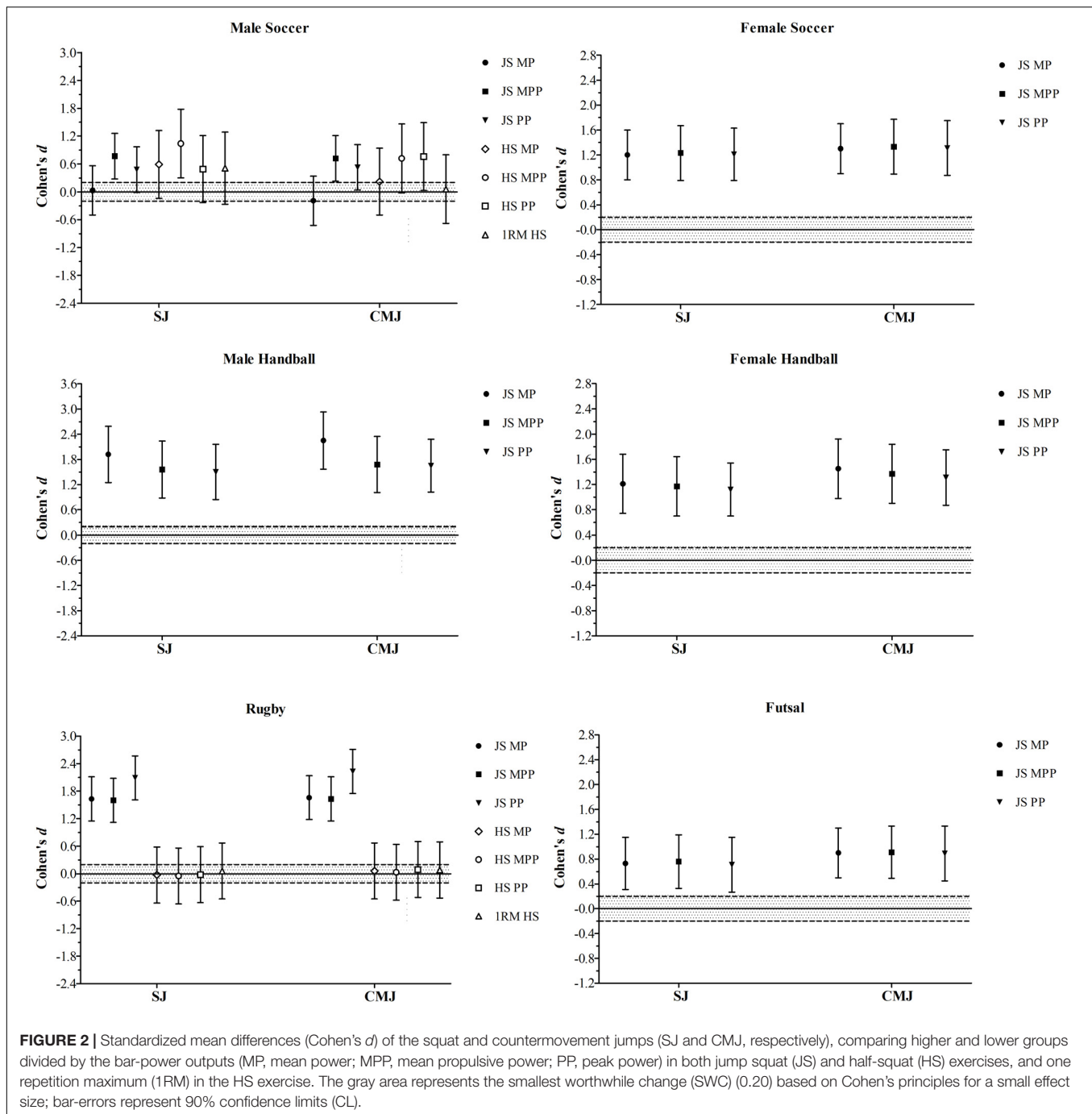
	Male Soccer	Female Soccer	Male Handball	Female Handball	Rugby	Futsal
SJ (cm)	39.68 ± 4.05	31.25 ± 4.37	37.75 ± 5.24	30.07 ± 4.37	40.76 ± 6.11	37.82 ± 7.10
CMJ (cm)	41.05 ± 4.74	31.81 ± 4.21	40.64 ± 6.53	30.86 ± 4.01	42.76 ± 6.14	38.50 ± 4.88
JS MP (W kg ⁻¹)	5.43 ± 0.83	5.10 ± 0.87	6.32 ± 1.33	5.31 ± 1.04	7.28 ± 1.34	6.44 ± 1.43
JS MPP (W kg ⁻¹)	8.08 ± 1.04	7.28 ± 1.25	8.62 ± 1.68	7.30 ± 1.42	10.40 ± 1.92	9.20 ± 2.04
JS PP (W kg ⁻¹)	17.34 ± 2.06	16.16 ± 2.77	19.05 ± 3.72	16.17 ± 3.15	23.55 ± 4.51	20.43 ± 4.53
HS MP (W kg ⁻¹)	5.39 ± 0.37	–	–	–	7.38 ± 1.49	–
HS MPP (W kg ⁻¹)	7.48 ± 0.86	–	–	–	9.46 ± 1.90	–
HS PP (W kg ⁻¹)	15.60 ± 2.00	–	–	–	20.81 ± 4.19	–
HS 1RM (kg kg ⁻¹)	1.82 ± 0.14	–	–	–	2.24 ± 0.30	–

Note: SJ, squat jump; CMJ, countermovement jump; JS, jump squat; HS, half-squat; MP, mean power; MPP, mean propulsive power; PP, peak power; 1RM, one-repetition maximum.

TABLE 3 | Descriptive results of the speed tests in the different distances tested in the athletes of four different sports disciplines.

	Male Soccer	Female Soccer	Male Handball	Female Handball	Rugby	Futsal
VEL 5-m (ms ⁻¹)	4.86 ± 0.25	4.35 ± 0.66	4.89 ± 0.33	4.62 ± 0.26	5.01 ± 0.31	4.81 ± 0.25
VEL 10-m (ms ⁻¹)	5.75 ± 0.19	5.14 ± 0.78	5.73 ± 0.29	5.29 ± 0.24	5.78 ± 0.28	5.68 ± 0.19
VEL 20-m (ms ⁻¹)	6.79 ± 0.22	5.96 ± 0.90	6.63 ± 0.28	6.06 ± 0.28	6.77 ± 0.31	6.61 ± 0.22
VEL 30-m (ms ⁻¹)	–	–	–	–	7.30 ± 0.33	–
VEL 40-m (ms ⁻¹)	–	–	–	–	7.64 ± 0.35	–
Zig-zag (ms ⁻¹)	3.37 ± 0.11	3.29 ± 0.11	3.54 ± 0.19	3.38 ± 0.15	3.63 ± 0.16	3.52 ± 0.11
ACC 0–5-m (ms ⁻²)	4.74 ± 0.50	3.96 ± 0.32	4.80 ± 0.67	4.29 ± 0.52	5.05 ± 0.63	4.64 ± 0.50
ACC 5–10-m (ms ⁻²)	1.26 ± 0.22	1.03 ± 0.16	1.16 ± 0.21	0.84 ± 0.18	1.04 ± 0.16	1.22 ± 0.22
ACC 10–20-m (ms ⁻²)	0.86 ± 0.09	0.61 ± 0.09	0.71 ± 0.11	0.55 ± 0.09	0.81 ± 0.10	0.74 ± 0.09
ACC 20–30-m (ms ⁻²)	–	–	–	–	0.47 ± 0.07	–
ACC 30–40-m (ms ⁻²)	–	–	–	–	0.30 ± 0.06	–

Note: VEL, velocity; ACC, acceleration.



be appropriate to predict or even monitor athletic performance in elite rugby players (Loturco et al., 2017f). Nonetheless, these data should be viewed with caution as previous research has suggested that enhanced force production (via the increased squat performance) might contribute to improved performance in professional rugby players (Comfort et al., 2012). Moreover, it has been reported that squat strength is strongly related to tackling ability in rugby league players (Speranza et al., 2015), an ability which was not measured in the current study. That said, in light of the above discussion, rugby practitioners are

encouraged to include loaded JS assessments in their testing routines, especially when assessing elite rugby players.

As observed in rugby, higher or lower HS 1RM performances appeared to have no influence on jump, speed, and acceleration capabilities in male soccer players. In contrast to rugby athletes, the soccer players with higher HS bar-power outputs, overall, performed better than their weaker peers in all functional assessments (Figures 2, 3, and 4). These data contradict previous research showing strong correlations of maximal squat strength with sprint performance and vertical jump height in elite soccer

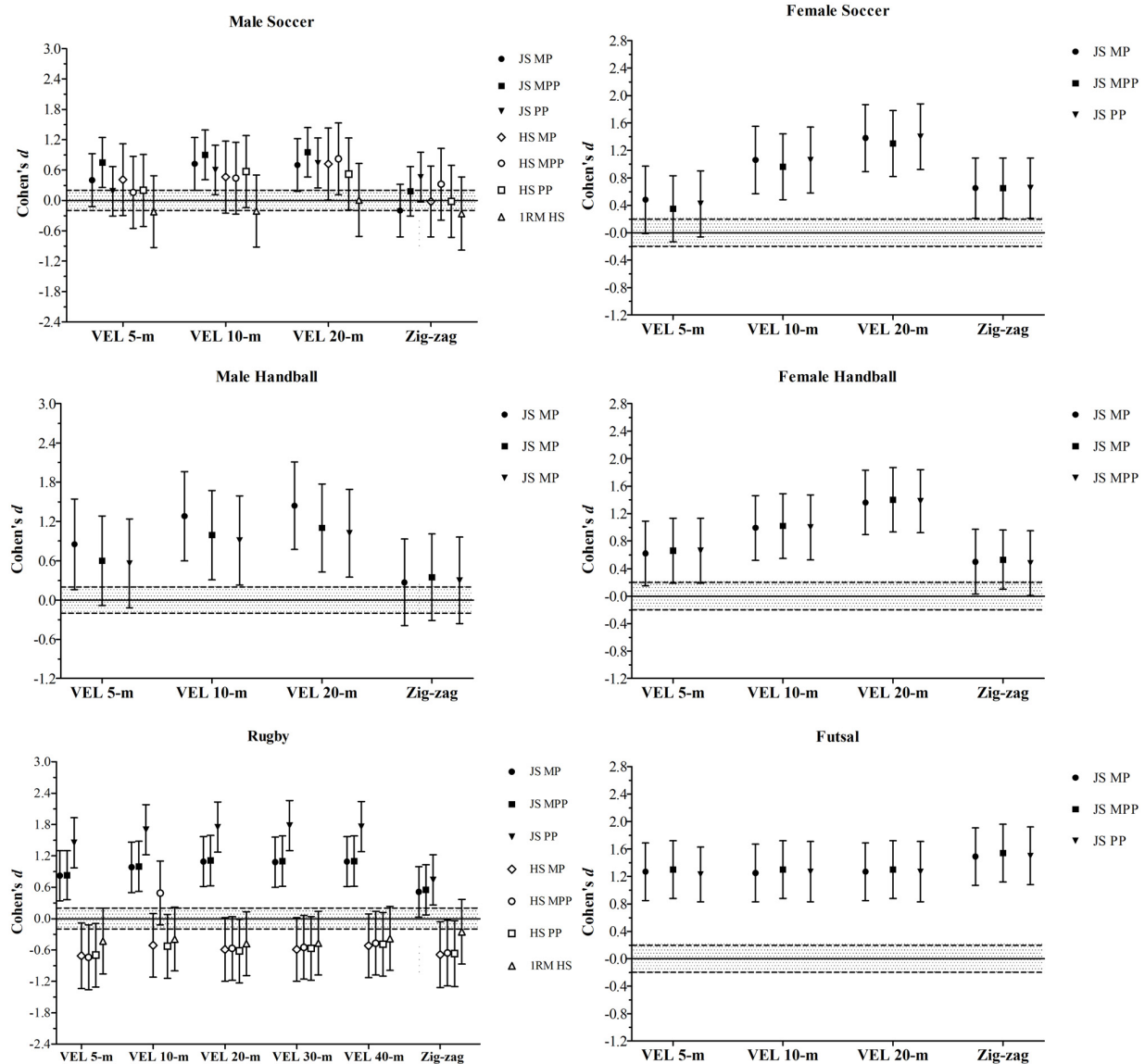


FIGURE 3 | Standardized mean differences (Cohen's *d*) of the sprint velocity (VEL) for the different distances tested and Zig-zag change of direction speed test, comparing higher and lower groups divided by the bar-power outputs (MP, mean power; MPP, mean propulsive power; PP, peak power) in both jump squat (JS) and half-squat (HS) exercises, and one repetition maximum (1RM) in the HS exercise. The gray area represents the SWC (0.20) based on Cohen's principles for a small effect size; bar-errors represent 90% CL.

players (Wisloff et al., 2004). To some extent, our results are similar to those of Requena et al. (2011), who found close relationships between traditional squat power output and sprint speed at 30- and 40-m. Although these authors also reported significant correlations between 1RM squat and sprint ability, only the power measures (i.e., maximal peak power and maximal average power) were significantly related to CMJ height (Requena et al., 2011). Nevertheless, in line with our findings, the associations between ballistic squats (i.e., loaded JS) and speed and jump variables were stronger than those detected for traditional squats. For many authors, the apparent superiority of JS over other resistance exercises to predict and improve

athletic performance may be due to its kinematic and kinetic features (Baker, 1996; Cormie et al., 2011; Suchomel et al., 2016b; Loturco et al., 2017f). Accordingly, it has been shown that some "mechanical similarities" (Loturco et al., 2016c) between JS and certain speed-power tasks may positively affect the specific training adaptations, thus increasing the transference effect of JS bar-power outputs to performance. Overall, these observations support and reinforce the use of loaded JS to both evaluate and improve physical qualities in male soccer players.

Despite the absence of HS assessments in the following groups (precluding comparisons between HS and JS exercises), female soccer players, male futsal players, and male and female

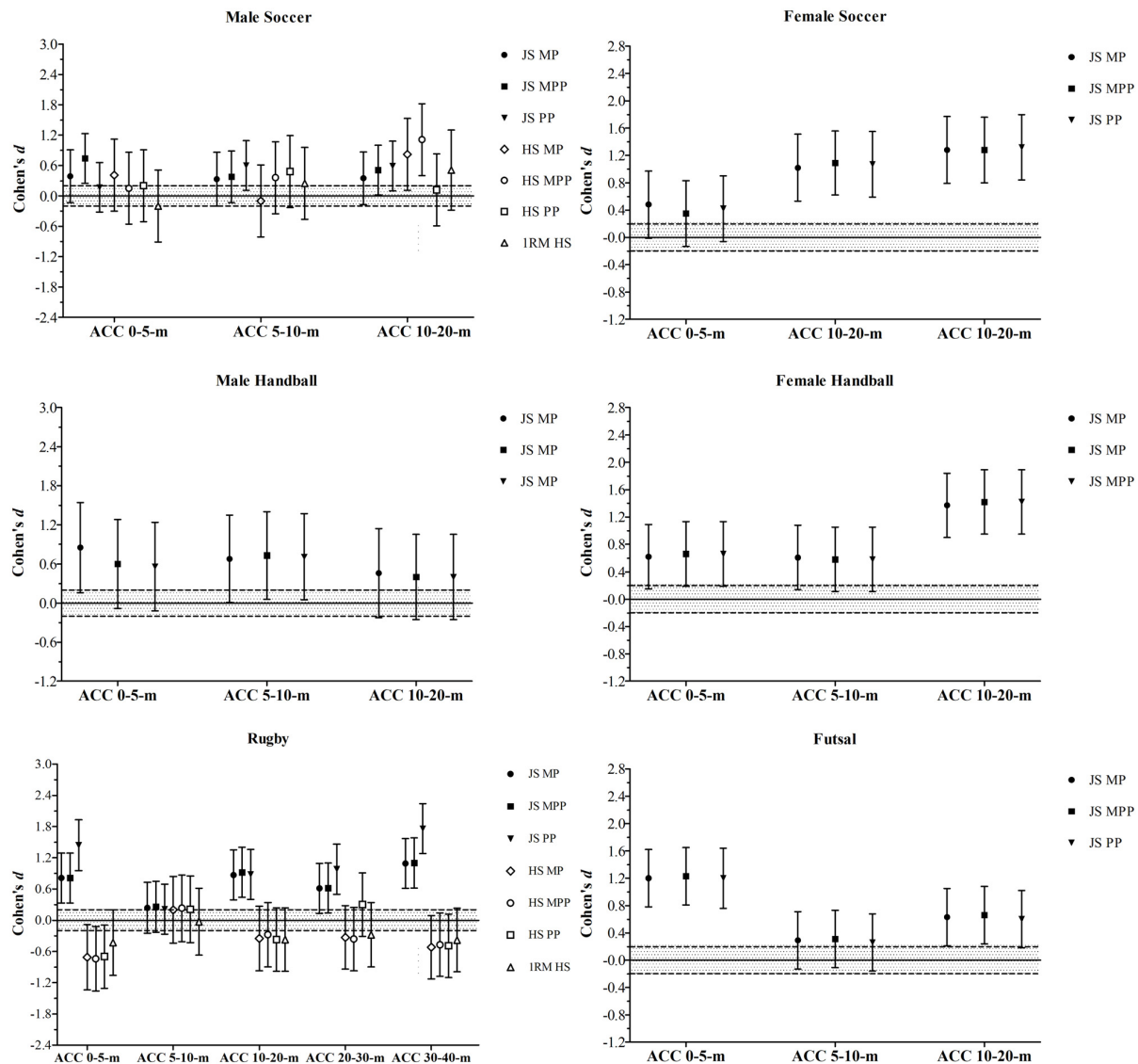


FIGURE 4 | Standardized mean differences (Cohen's *d*) of the sprint acceleration (ACC) for the different distances tested, comparing higher and lower groups divided by the bar-power outputs (MP, mean power; MPP, mean propulsive power; PP, peak power) in both jump squat (JS) and half-squat (HS) exercises, and one repetition maximum (1RM) in the HS exercise. The gray area represents the SWC (0.20) based on Cohen's principles for a small effect size; bar-errors represent 90% CL.

handball players with greater measures of bar power-output also perform better in both SJ and CMJ tests. These results are in accordance with those reported in several other studies and, as aforementioned, are likely related to the mechanical resemblances between loaded and unloaded vertical jumps (Figure 2) (Cronin and Hansen, 2005; Moir et al., 2005; McBride et al., 2010; Janssen et al., 2012; Loturco et al., 2015a, 2017b, 2018c). As a consequence, athletes able to generate higher levels of power during loaded JS (using light to moderate loads) can be expected to produce higher levels of power under unloaded jumping conditions (SJ and CMJ), and are also likely to jump higher (Loturco et al., 2015a). The same holds true for acceleration and speed capabilities (Figures 3 and 4), which have been shown to be strongly related to the JS maximum

power output (Cronin and Hansen, 2005; Loturco et al., 2015a, 2017c). In fact, when a subject executes a loaded JS, they have to jump lifting up the whole mechanical system (i.e., weighted barbell + body mass), providing measurements automatically adjusted by the BM (Loturco et al., 2017c). Therefore, a greater JS performance might also indicate an increased ability to overcome the inertia and accelerate the body quickly and effectively, which is essential to achieve higher velocities over short distances (Cronin and Hansen, 2005; Loturco et al., 2015c; Kale and Acikada, 2016). Research by Cormie et al. (2011) supports this, stating that ballistic JS “circumvents any deceleration phase by requiring subjects to accelerate throughout the entire range of motion to the point of projection” (i.e., takeoff), being “more sport-specific for a vast number of sports.” Another advantage of

using loaded JS to assess a number of strength-power variables is related to its high degree of reliability (Moir et al., 2005), achieved without the need to perform familiarization sessions, supporting the suitability of the tests for monitoring physical performance team sport athletes (who regularly perform many concurrent activities, within a congested schedule of engagements) (Loturco et al., 2015d; Freitas et al., 2018). Together, these data strongly support the notion that JS performed with a load that maximizes power output is one of the best methods to assess and improve physical performance in professional athletes from a wide variety of sports (Baker, 1996; Loturco et al., 2017c, 2018c).

A particular aspect of the current investigation is the lack of consistency in the outcomes related to COD performance across the examined sports (**Figure 3**). Briefly, COD speed can be characterized as a multifaceted ability, which relies on a series of different and multiple technical and physical aspects (e.g., stride adjustments, foot placement, straight speed, leg muscle qualities, etc.) (Young and Farrow, 2006; Brughelli et al., 2008; Hewit et al., 2012). This well-documented complexity could have affected the performance obtained by some athletes during the Zig-zag test (Little and Williams, 2005; Pereira et al., 2018), making this maneuver more convenient for assessing (for example) futsal players than male soccer players (Nimphius et al., 2010; Chaouachi et al., 2012). Indeed, previous research showed that the reduced pitch dimensions and more frequent turnovers during futsal match-play (compared to soccer), in both attacking and defending actions, support the development of higher coordinative skills in futsal players (Benvenuti et al., 2010). Although these differences were found in “reactive COD tasks,” these sport-related characteristics (and competences) may have influenced our results. However, these are only speculations and further work is needed to identify the most relevant factors for COD performance. Thus, this study confirms and strengthens previous conclusions, highlighting the necessity to create and adopt more effective training strategies to properly evaluate and develop COD ability in elite team sport athletes (Nimphius et al., 2010; Young and Farrow, 2006; Hewit et al., 2012).

In summary, this research shows that the bar-power approach is a useful method to assess team sport players, due to its close connection to acceleration, speed, and jumping abilities. These data are similar to those reported in a recent investigation, indicating that the bar-power outputs are more strongly associated with speed-power performances in elite athletes from four different sports than 1RM measurements (Loturco et al., 2018c). Therefore, as previously suggested, the possibility of using a range of loads which optimize the force and velocity applied to the barbell simultaneously [instead of only considering the maximum mass moved during a maximum effort (i.e., 1RM)] might better reflect the physical abilities and technical skills

required in team-sport-tasks (Loturco, 2017; Loturco et al., 2018c). Finally, it is essential to emphasize that this work is inherently limited by its cross-sectional design, precluding inferences about causality. Nonetheless, our findings are strongly supported by a series of studies which has already demonstrated the effectiveness of the OPL (directly assessed on the barbell) to acutely or chronically improve performance in elite and sub-elite team sport athletes (Loturco et al., 2015d, 2016c; Dello Iacono and Seitz, 2018; Freitas et al., 2018).

CONCLUSION

The bar-power approach is a practical and useful strategy to assess the physical performance of elite team sport players. Similar to previous findings (Loturco et al., 2018c), also in highly trained athletes, the bar-power output seems to be closely related to a series of athletic capabilities, which are recognized to play an important role in team sports performance, especially when considering the decisive game actions (Faude et al., 2012; Povoas et al., 2012; Ross et al., 2014). As described in other sport disciplines (Loturco et al., 2018c), it is likely that the opportunity to use measurements which consider, at the same time, the force and velocity applied to the barbell may have contributed to the stronger connections observed between bar-power variables and acceleration, speed, and jump qualities (when compared to the 1RM measures). Despite the lack of consistency and uniformity among the outcomes related to COD performance across the examined sports (which appears to be commonplace in COD studies) (Brughelli et al., 2008), it is possible to infer from these findings that players able to produce higher bar-power outputs are more prone to sprint faster and jump higher. From a general perspective, these “interconnections” are still more pronounced when the outcomes are directly collected from the loaded JS. Future studies should be conducted to test the causality between the variables reported here, as well as to search for more precise and consistent predictors of COD speed.

AUTHOR CONTRIBUTIONS

IL, LP, and MM: designed the work. IL, CA, and LP: data acquisition. IL, TS, LJ, CB, CA, LP, and MM: analysis and interpretation of data. IL and LP: drafting first version of the work. IL, TS, LJ, CB, CA, LP, and MM: critically revising the work. IL, TS, LJ, CB, CA, LP, and MM: final approval of the version to be published. IL, TS, LJ, CB, CA, LP, and MM: agreed to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work were appropriately investigated and resolved.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

The handling Editor declared a past co-authorship and collaboration with one of the authors MM.

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Post-exercise Hot Water Immersion Elicits Heat Acclimation Adaptations in Endurance Trained and Recreationally Active Individuals

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OPEN ACCESS

Edited by:

Toby Mündel,
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United Kingdom

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 01 November 2018

Accepted: 06 December 2018

Published: 18 December 2018

Citation:

Zurawlew MJ, Mee JA and
Walsh NP (2018) Post-exercise Hot
Water Immersion Elicits Heat
Acclimation Adaptations in Endurance
Trained and Recreationally Active
Individuals. *Front. Physiol.* 9:1824.
doi: 10.3389/fphys.2018.01824

Hot water immersion (HWI) after exercise on 6 consecutive days in temperate conditions has been shown to provide heat acclimation adaptations in a recreationally active population. Endurance athletes experience frequent, sustained elevations in body temperature during training and competition; as a consequence, endurance athletes are considered to be partially heat acclimatized. It is therefore important to understand the extent to which endurance trained individuals may benefit from heat acclimation by post-exercise HWI. To this end, we compared the responses of eight endurance trained and eight recreationally active males (habitual weekly endurance exercise: 9 h vs. 3 h) to a 6-day intervention involving a daily treadmill run for 40 min (65% $\dot{V}O_{2max}$) in temperate conditions followed immediately by HWI (≤ 40 min, 40°C). Before (PRE) and after the intervention (POST), hallmark heat acclimation adaptations were assessed during a 40-min treadmill run at 65% $\dot{V}O_{2max}$ in the heat (33°C , 40% RH). The 6 day, post-exercise HWI intervention induced heat acclimation adaptations in both endurance trained and recreationally active individuals. Training status did not significantly influence the magnitude of heat acclimation adaptations from PRE to POST (interactions $P > 0.05$) for: the reduction in end-exercise rectal core temperature (T_{re} , mean, endurance trained -0.36°C ; recreationally active -0.47°C); the reduction in resting T_{re} (endurance trained -0.17°C ; recreationally active -0.23°C); the reduction in T_{re} at sweating onset (endurance trained -0.22°C ; recreationally active -0.23°C); and, the reduction in mean skin temperature (endurance trained -0.67°C ; recreationally active -0.75°C : PRE to POST $P < 0.01$). Furthermore, training status did not significantly influence the observed reductions in mean $\dot{V}O_2$, mean metabolic energy expenditure, end-exercise physiological strain index, perceived exertion or thermal sensation (PRE to POST $P < 0.05$). Only end-exercise heart rate was influenced by training status ($P < 0.01$, interaction); whereby, recreationally active but not endurance trained individuals experienced a significant reduction in end-exercise heart rate from PRE to POST ($P < 0.01$). In summary, these findings demonstrate that post-exercise HWI presents a practical strategy to reduce thermal strain during exercise-heat-stress in endurance trained and recreationally active individuals.

Keywords: heat, acclimation, hot water, thermal strain, training, running

INTRODUCTION

Exercise in the heat increases physiological strain, attenuates exercise capabilities and increases susceptibility to exertional heat illness and the potentially fatal, exertional heat stroke (Young et al., 1985; Binkley et al., 2002; Racinais et al., 2015). In the early twentieth century, pioneering research on fatal heat stroke in the South African gold mines highlighted a high mortality in the first four shifts worked by miners under high heat exposure; particularly in those native to cold dry areas (Cluver, 1932; Dreosti, 1935). To mitigate the risk of heat stroke, miners acclimatized by gradual introduction to the unfavorable working conditions underground (Cluver, 1932). Current recommendations are for athletes, military personnel and others in occupations involving high heat exposure to complete a period of heat acclimation prior to competing or operating in the heat. Heat acclimation typically involves exercising in the heat on 5–14 occasions for >60-min, where core body temperature and skin temperature are elevated and perfuse sweating is initiated (Taylor, 2014; Periard et al., 2015). The adaptive responses to exercise-heat-acclimation include, but are not limited to: an earlier onset of cutaneous vasodilatation and sweating; an increase in sweating rate; a reduction in resting and exercising core body temperature; a reduction in cardiovascular strain and skin temperature; that in turn, improve thermal comfort and enhance endurance performance in the heat (Gagge et al., 1967; Lorenzo et al., 2010; Taylor, 2014).

Routine endurance training performed in temperate conditions, which elevates body temperature and initiates perfuse sweating, shares common adaptive responses to exercise-heat-acclimation such as; an earlier onset and an increase in sweating rate, a reduction in core temperature and a reduction in cardiovascular strain during exercise-heat-stress; which in turn, improves endurance performance in the heat (Piwonka et al., 1965; Strydom et al., 1966; Gisolfi and Robinson, 1969; Shvartz et al., 1977). As such, endurance trained individuals are considered to be partially heat acclimatized (Piwonka et al., 1965; Strydom et al., 1966; Gisolfi and Robinson, 1969). By the same token, its long been considered that endurance trained individuals have less adaptation potential and require fewer exercise-heat-exposures to achieve a plateau in heat acclimation responses, compared with untrained individuals (Pandolf et al., 1977; Shvartz et al., 1977). For example, following constant work rate heat acclimation, trained individuals acquired smaller thermal benefits during exercise-heat-stress than untrained individuals (Shvartz et al., 1977). Soldiers of the highest aerobic fitness required only four exercise-heat-acclimation exposures to achieve a plateau in the reduction of end-exercise rectal core temperature (T_{re}); whereas, soldiers with the lowest aerobic fitness required eight exercise-heat-acclimation exposures (Pandolf et al., 1977). A limitation of these studies, reporting smaller and more rapid adaptations to heat acclimation in trained individuals, is that the observed plateau in heat acclimation adaptations may simply represent habituation to the constant exercise-heat-stress; resulting in a decline in the adaptation stimulus (Taylor, 2014). Recent studies that have maintained the endogenous thermal stimulus during controlled

hyperthermia heat acclimation demonstrate comparable thermal and cardiovascular adaptations in endurance trained (Neal et al., 2016) and recreationally active individuals (Gibson et al., 2015).

Despite compelling evidence that exercise-heat-acclimation alleviates thermal strain and improves performance in the heat (Nielsen et al., 1997; Lorenzo et al., 2010), only 15% of athletes competing at the 2015 World Athletics Championships in the heat and humidity of Beijing heat acclimatized as part of their preparation (Periard et al., 2017). One possible explanation is that athletes consider their high level of fitness confers adaptations similar to heat acclimatization (Piwonka et al., 1965; Strydom et al., 1966; Gisolfi and Robinson, 1969); so they favor natural heat acclimatization in the few days preceding competition and prioritize other strategies to combat the heat such as fluid replacement and pre-cooling (Periard et al., 2017). Another explanation is that conventional exercise-heat-acclimation protocols can be costly, impractical and may interfere with an athlete's training and taper: exercise-heat-acclimation typically involves access to an environmental chamber and precise control over exercising core temperature during endurance exercise. The completion of alternative heat acclimation methods, such as post-exercise sauna bathing (Scoon et al., 2007) and hot water immersion (HWI) (Zurawlew et al., 2016) have received increasing interest of late (Casadio et al., 2017). These methods are; accessible, time efficient, simple to administer and minimize disturbances to training and tapering. Recently, HWI after exercise in temperate conditions on 6 consecutive days initiated hallmarks of heat acclimation in recreationally active individuals (Zurawlew et al., 2016, 2018). Heat acclimation adaptations to post-exercise HWI included reductions in; T_{re} at rest, T_{re} at sweating onset and T_{re} during exercise-heat stress; in turn, restoring endurance performance in the heat to the level observed in temperate conditions (Zurawlew et al., 2016). Similar to controlled hyperthermia heat acclimation, post-exercise HWI ensures a maintenance of the daily thermal stimulus for adaptation (daily ΔT_{re} ; $\approx 2.1^\circ\text{C}$), since the termination of the HWI relies primarily on participants removing themselves due to thermal discomfort (Zurawlew et al., 2016, 2018).

It remains unknown whether heat acclimation adaptations to post-exercise HWI in a recreationally active population translate to an endurance trained population. As such, the aim of the current study was to compare the adaptation responses of endurance trained and recreationally active individuals following post-exercise HWI. We hypothesized that HWI after submaximal exercise in temperate conditions on 6 consecutive days would induce comparable heat acclimation adaptations in endurance trained and recreationally active individuals.

MATERIALS AND METHODS

Participants

In accordance with previously defined classifications (De Pauw et al., 2013), eight endurance trained males (runners, $n = 6$ and triathletes, $n = 2$; age: 25 ± 4 years; body mass: 69 ± 4 kg; self-reported weekly endurance exercise: 9 ± 3 h;

$\dot{V}O_{2\max}$: $68 \pm 6 \text{ mL} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$) and eight recreationally active males (age: 21 ± 3 years; body mass: $71 \pm 9 \text{ kg}$; self-reported weekly endurance exercise: $3 \pm 1 \text{ h}$; $\dot{V}O_{2\max}$: $54 \pm 6 \text{ mL} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$), participated in the study. All participants provided written informed consent to participate, were healthy, non-smokers, free from any known cardiovascular or metabolic diseases and were not taking any medication. Additionally, all participants had not been exposed to hot environmental conditions in the past 3 months and were not regular hot bath or sauna users. The study received local ethical approval and was conducted in accordance with the Declaration of Helsinki (2013).

Study Design

A mixed-methods (between and within) repeated measures (PRE to POST) design was used to assess the effect of training status on heat acclimation adaptations. Endurance trained and recreationally active participants completed a 40-min submaximal treadmill run at 65% $\dot{V}O_{2\max}$ in the heat (33°C , 40% relative humidity; RH) before (PRE) and after (POST) heat acclimation, as described previously (Zurawlew et al., 2016). Heat acclimation involved a daily 40-min submaximal treadmill run at 65% $\dot{V}O_{2\max}$ in temperate conditions (19°C), followed by a ≤ 40 -min HWI (40°C water) on 6 consecutive days, as described previously (Zurawlew et al., 2016).

Preliminary Measurements

In temperate conditions (19°C), a continuous incremental exercise test on a motorized treadmill (HP Cosmos Mercury 4.0, Nussdorf-Traunstein, Germany) assessed $\dot{V}O_{2\max}$, as previously described (Fortes et al., 2013). The interpolation of the running speed– $\dot{V}O_2$ relationship determined a running speed that elicited 65% $\dot{V}O_{2\max}$. This speed was verified during steady state exercise with a 60-s expired gas sample collected by Douglas bag method, 30-min after the $\dot{V}O_{2\max}$ test. This individualized running speed was used during the submaximal exercise in both the experimental trials and the daily intervention.

Experimental Trials

Participants were instructed to refrain from any exercise 24-h prior to, and on the day of experimental trials. In addition, participants were instructed to refrain from alcohol, caffeine or tobacco and to complete a diet diary 24-h prior to PRE. Twenty-four hours prior to POST, participants were instructed to replicate this food and fluid intake. On the morning of experimental trials, participants arrived at the laboratory fasted and were provided with a standardized breakfast (0.03 MJ kg^{-1}) and a bolus of water equivalent to $7 \text{ mL} \cdot \text{kg}^{-1}$ of body mass. Following a 20-min seated rest in temperate conditions (19°C), dressed in a T-shirt, running shorts, socks and shoes, a venous blood sample was taken without stasis. A pre-exercise nude body mass was taken using a digital platform scale (Model 705; Seca, Hamburg, Germany) after voiding. A urine sample was provided and analyzed for urine specific gravity to confirm that participants were hydrated (<1.03) (Armstrong, 2005) using a handheld refractometer (Atago Uricon-Ne refractometer, NSG Precision cells, Farmingdale, NY, United States). If participants

did not meet the hydration criteria they were provided with a 500-mL bolus of water and urine specific gravity was reanalyzed; exercise began only when urine specific gravity <1.03 ($n = 1$). Participants were instrumented for the exercise protocol, then rested in a temperate laboratory to establish baseline measures prior to beginning the exercise.

Dressed in running shorts, socks and shoes the participant entered the environmental chamber ($33 \pm 0^\circ\text{C}$, $40 \pm 4\%$ RH; Delta Environmental Systems, Chester, United Kingdom) and completed a submaximal treadmill run (40-min, 65% $\dot{V}O_{2\max}$, 1% gradient). T_{re} , skin temperatures and heart rate (Polar FT1, Polar Electro, Kempele, Finland) were monitored continuously and local forearm sweat rate was measured every 20-s for the first 15-min of exercise. Physiological strain index (PhSI) was calculated, as previously described (Tikuisis et al., 2002). Expired gas samples (60-s) were collected by Douglas bag method to assess for $\dot{V}O_2$ and respiratory exchange ratio (RER) immediately prior to the 10th, 20th, 30th, and 40th min of exercise. Metabolic energy expenditure was calculated using $\dot{V}O_2$ and RER as described (Nishi, 1981). Rating of perceived exertion (RPE) (Borg, 1970) and thermal sensation (Hollies and Goldman, 1977) were recorded every 10-min of exercise. On completion of the exercise protocol, participants exited the environmental chamber and rested in temperate conditions, dressed in running shorts, socks, and shoes for 15-min. To estimate whole body sweat rate (WBSR), participants towel dried and provided a nude body mass following the seated rest. Participants were then provided with water equivalent to sweat losses and were free to leave the laboratory when $T_{re} \leq 38.5^\circ\text{C}$.

Post-exercise Hot Water Immersion Intervention

Post-exercise HWI heat acclimation was completed on 6 consecutive days, as previously described (Zurawlew et al., 2016). During the intervention, participants were instructed to reduce their normal endurance exercise volume by that completed during the intervention in the laboratory and to consume their normal diet and fluid intake, including caffeine and alcohol (≤ 3 units per day). Participants arrived at the laboratory each day between 0600-h and 1000-h. A heart rate monitor and a rectal thermistor were fitted and the participant rested in temperate conditions (19°C) for 15-min. Following the seated rest, dressed in shorts, socks and trainers, in a hydrated state (urine specific gravity < 1.03), participants completed a 40-min submaximal run (65% $\dot{V}O_{2\max}$, 1% gradient) on a motorized treadmill in temperate conditions (19°C). Within the first 20-min of exercise, participants consumed a bolus of water ($5 \text{ mL} \cdot \text{kg}^{-1}$ of body mass). Following exercise, participants undertook a ≤ 40 -min HWI (40°C), immersed to the neck dressed in shorts (2–3 min transition time). Immersion in hot water was terminated either at 40 min, when participants removed themselves due to thermal discomfort or when T_{re} exceeded the institutional ethical cut off (39.9°C). Following removal from the hot water, participants rested in temperate laboratory conditions, dressed in shorts for 15-min without fluids. Following which, participants towel dried and a nude body mass was recorded and adjusted for fluid

intake as a measure of WBSR. Participants were free to leave the laboratory when $T_{re} \leq 38.5^{\circ}\text{C}$.

Measurement and Instrumentation

Body Temperatures

T_{re} was measured using a flexible, sterile rectal thermistor (Henleys Medical Supplies Ltd., Herts, United Kingdom), self-inserted 10 cm beyond the rectal sphincter and recorded using a data logger (YSI model 4000 A, YSI, Dayton, OH, United States). An area under the curve (AUC) analysis was performed on T_{re} (time T_{re} was $> 38.5^{\circ}\text{C}$) during each post-exercise HWI exposure to assess for cumulative hyperthermia, as previously described (Cheuvront et al., 2008). Skin temperatures were measured during experimental trials using insulated thermistors (Grant EUS-U, Cambridge, United Kingdom) secured on the right side of the body at four locations: chest at a midpoint between the acromion process and the nipple; the lateral mid-bicep; the anterior mid-thigh; and, lateral calf and recorded using a data logger (Grant SQ2020, Cambridge, United Kingdom). Mean skin temperature (T_{sk}) was calculated from the four sites using a weighted equation (Ramanathan, 1964).

Sweating Responses

Changes in dry nude body mass estimated WBSR during experimental trials and post-exercise HWI heat acclimation exposures. Dew point hygrometry measured local forearm sweat rate during the experimental trials, as previously described (Fortes et al., 2013). The individual relationships between local forearm sweat rate and T_{re} were used to calculate the onset of sweating (Cheuvront et al., 2009).

Blood Sample Collection and Analysis

During experimental trials, prior to exercise a venous blood sample (6 mL) was collected into an EDTA vacutainer (BD, Oxford, United Kingdom) without stasis from an antecubital vein, following a 20-min seated rest to stabilize body fluids. Hemoglobin concentration ($\text{g} \cdot \text{dL}^{-1}$; Hemocue, Sheffield, United Kingdom) in duplicate and hematocrit (%) in triplicate (capillary tube method) were immediately assessed from aliquots of whole blood. The change in plasma volume was estimated by correcting the initial plasma volume at PRE for the percentage change in plasma volume at POST, as previously described (Dill and Costill, 1974).

Statistical Analysis

A sample size calculation (G*Power 3.1.2), using an alpha level of 0.05, power of 0.80 and a strong correlation of 0.7, was performed using data from a study comparing heat acclimation responses in endurance trained and untrained individuals (Shvartz et al., 1977). For a two-way (group \times time) repeated measures ANOVA, a sample size of eight participants per group was calculated to detect a significant difference in the magnitude of reduction in end-exercise T_{re} ($\Delta 0.3^{\circ}\text{C}$), between endurance trained and untrained individuals following heat acclimation. All data were checked for normality and sphericity and statistical significance was accepted at $P < 0.05$. Two-way repeated measures analysis of variance (ANOVA) with

Greenhouse Geisser correction to the degrees of freedom (where necessary) were used to assess for main effects, i.e., differences between groups (endurance trained vs. recreationally active) and changes from PRE to POST during the experimental trials and from day 1 to day 6 of the intervention, as well as interaction effects (group \times time). Bonferroni-adjusted pairwise comparisons were used where appropriate to determine where differences occurred. Independent t -tests assessed for differences in total HWI time and total AUC between endurance trained and recreationally active. The magnitude of effect was reported using Cohen's d , where 0.2, 0.5, and 0.8 represent small, medium and large effects, respectively (Cohen, 1988). Pearson's correlations were used to determine the strength of the relationship between training status or aerobic fitness (habitual weekly endurance exercise and $\dot{V}\text{O}_{2\text{max}}$) and the reduction in end-exercise T_{re} and heart rate, and between the thermal stimulus (total AUC) during the heat acclimation intervention and the reduction in end-exercise T_{re} and heart rate. Data are presented as mean \pm standard deviation (SD) and were analyzed using SPSS version 24 (IBM Corporation, Armonk, NY, United States), or GraphPad Prism Version 5.02 (GraphPad Software Inc., La Jolla, CA, United States).

RESULTS

Intervention

All participants completed a 40-min treadmill run at 65% $\dot{V}\text{O}_{2\text{max}}$ followed by HWI (≤ 40 min) on 6 consecutive days. Compared to their typical endurance exercise volume, during the intervention, weekly endurance exercise volume was unchanged for endurance trained (-1 ± 2 h; $P > 0.05$) and increased in recreationally active individuals ($+2 \pm 0$ h; $P < 0.01$). During the 6-day intervention, HWI duration increased from day 1 to day 6 ($P < 0.05$); ensuring a maintenance of the endogenous stimulus for adaptation, with a similar AUC and end-HWI T_{re} between day 1 and day 6 ($P > 0.05$; **Table 1**). In addition, total immersion time ($P = 0.08$, $d = 1.0$) and total AUC ($P = 0.08$, $d = 0.8$) during the 6-day intervention tended to be greater in endurance trained than recreationally active individuals (**Table 1**).

Experimental Trials

No significant interaction effects (group \times time; $P > 0.05$) demonstrate that training status did not influence the observed adaptations to the 6-day post-exercise HWI intervention, for measures of: end-exercise T_{re} (**Figure 1A**); resting T_{re} ; T_{re} at sweating onset; ΔT_{re} during exercise; end-exercise T_{sk} ; end-exercise $T_{re}-T_{sk}$ gradient; end-exercise PhSI; end-exercise RPE, end-exercise thermal sensation, mean $\dot{V}\text{O}_2$, mean RER or mean metabolic energy expenditure (**Table 2**). Training status did not relate strongly to the magnitude of thermal adaptation; since the reduction in end-exercise T_{re} during exercise-heat-stress was not strongly correlated with either habitual endurance exercise volume ($r = 0.35$, $P > 0.05$) or $\dot{V}\text{O}_{2\text{max}}$ ($r = 0.29$, $P > 0.05$). In endurance trained individuals, a larger thermal stimulus during the HWI intervention (total AUC $63\text{--}320^{\circ}\text{C min}^{-1}$) was strongly associated with a larger reduction in end-exercise T_{re}

TABLE 1 | The influence of 40-min submaximal running at 65% $\dot{V}O_{2\max}$ in temperate conditions followed by post-exercise hot water immersion in 40°C water on thermoregulatory variables, heart rate and immersion time in endurance trained and recreationally active participants.

	Endurance trained		Recreationally active	
	Day 1	Day 6	Day 1	Day 6
Submaximal exercise				
End-exercise T_{re} (°C)	38.37 ± 0.48	38.27 ± 0.43	38.34 ± 0.32	38.22 ± 0.23
End-exercise heart rate (beats \min^{-1})*	147 ± 13	144 ± 10	150 ± 9	144 ± 9
Hot water immersion				
End-immersion T_{re} (°C)	39.44 ± 0.44	39.36 ± 0.31	39.15 ± 0.18	39.21 ± 0.20
Immersion time (min)**	35 ± 8	40 ± 0	28 ± 5	40 ± 1
<i>n</i> completing 40-min immersion	5 of 8	8 of 8	0 of 8	7 of 8
Submaximal exercise and hot water immersion				
WBSR ($L \cdot h^{-1}$)* #	1.08 ± 0.34	1.25 ± 0.26	0.72 ± 0.17	0.95 ± 0.18
AUC (°C \cdot min $^{-1}$)	33 ± 24	29 ± 15	18 ± 7	20 ± 7

T_{re} , rectal temperature; AUC, area under the curve. * $P < 0.05$ and ** $P < 0.01$, main effect of time. # $P < 0.05$, main effect of group. Data displayed as mean ± SD.

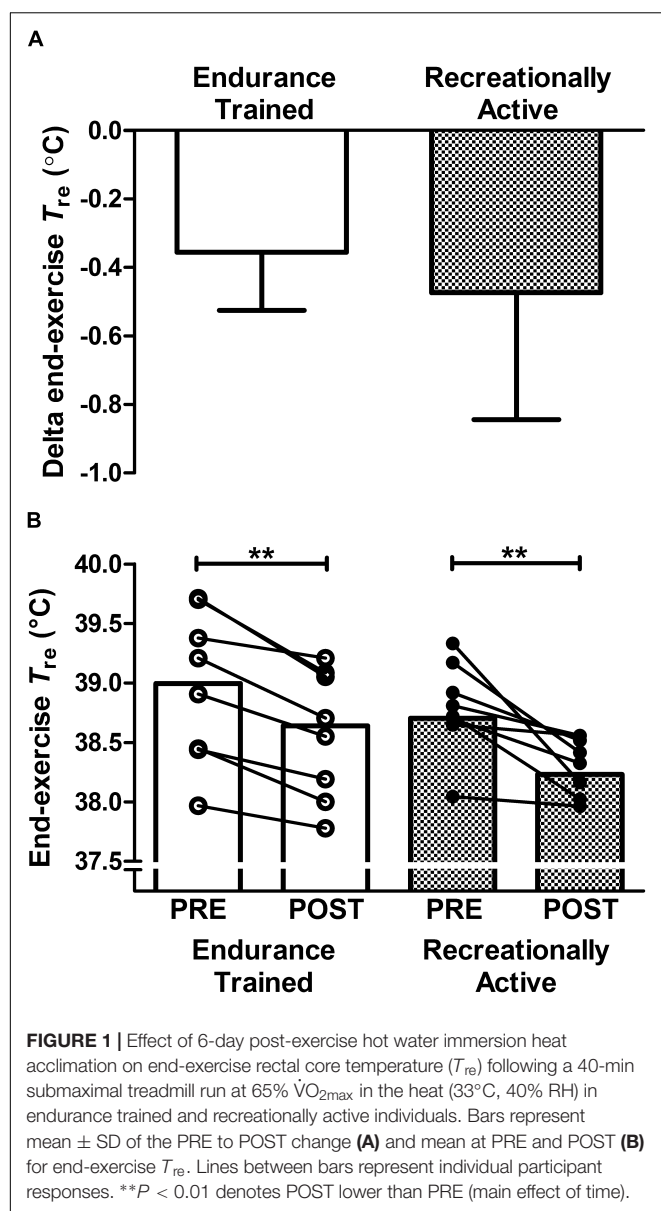
($r = -0.71$; $P < 0.05$). Moreover, post-exercise HWI reduced thermal strain during exercise-heat stress in all 16 participants, supported by a main effect of time (PRE vs. POST) for end-exercise T_{re} (PRE; $38.85 \pm 0.49^\circ\text{C}$, POST; $38.43 \pm 0.42^\circ\text{C}$, $P < 0.01$, $d = 0.9$; **Figure 1B**); albeit, one recreationally active participant experienced only a 0.08°C reduction in end-exercise T_{re} . Contrary to the notion that the most highly trained would benefit the least from the HWI intervention, the most accomplished endurance trained participant, an international marathon runner ($\dot{V}O_{2\max}$: $81 \text{ mL} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ and road half-marathon PB: 66 min) experienced a meaningful reduction in end-exercise T_{re} during exercise-heat-stress (PRE; 38.94°C , POST; 38.62°C).

Recreationally active individuals experienced a smaller thermal stimulus during the HWI intervention (total AUC $58\text{--}197^\circ\text{C} \cdot \text{min}^{-1}$) than endurance trained individuals, since they terminated HWI sooner due to thermal discomfort (**Table 1**). The thermal stimulus during the post-exercise HWI intervention was not strongly related to the reduction in end-exercise T_{re} in recreationally active individuals ($r = 0.12$, $P > 0.05$). As such, there appear to be other drivers contributing to the observed adaptations in recreationally active individuals, beyond the total AUC. An interaction effect (group \times time; $P < 0.01$) was observed for end-exercise heart rate, with a significant reduction from PRE to POST in recreationally active (PRE; $178 \pm 12 \text{ beats} \cdot \text{min}^{-1}$, POST; $163 \pm 9 \text{ beats} \cdot \text{min}^{-1}$, $P < 0.01$, $d = 1.4$), but not endurance trained individuals (PRE; $167 \pm 15 \text{ beats} \cdot \text{min}^{-1}$, POST; $163 \pm 16 \text{ beats} \cdot \text{min}^{-1}$, $P > 0.05$, $d = 0.2$; **Table 2**). Correlations suggest that the decrease in end-exercise heart rate during exercise-heat-stress after the HWI intervention was relatively strongly related to habitual exercise volume ($r = 0.68$, $P < 0.01$; **Figure 2A**) and aerobic fitness ($\dot{V}O_{2\max}$; $r = 0.57$, $P < 0.05$; **Figure 2B**); whereby, those with a higher habitual exercise volume and aerobic fitness demonstrated a smaller reduction in end-exercise heart rate after the 6-day post-exercise HWI intervention. In contrast, the thermal stimulus during the HWI intervention (total AUC) was not strongly related to the decrease in end-exercise heart rate during exercise-heat-stress ($r = 0.14$, $P > 0.05$).

Other hallmark heat acclimation adaptations were achieved following post-exercise HWI (main effect of time, PRE vs. POST, $n = 16$), including reductions in: resting T_{re} (PRE; $36.91 \pm 0.31^\circ\text{C}$, POST; $36.71 \pm 0.35^\circ\text{C}$, $P < 0.01$, $d = 0.6$); T_{re} at sweating onset ($P < 0.01$, $d = 0.6$); ΔT_{re} during exercise ($P < 0.05$, $d = 0.5$); end-exercise heart rate ($P < 0.01$, $d = 0.7$); T_{sk} ($P < 0.01$, $d = 0.8$); $T_{re}-T_{sk}$ gradient ($P < 0.05$, $d = 0.3$); PhSI ($P < 0.01$, $d = 0.9$); RPE (“fairly hard” to “fairly light,” $P < 0.05$, $d = 0.7$); thermal sensation (“hot” to “uncomfortably warm,” $P < 0.01$, $d = 1.0$); mean $\dot{V}O_2$ ($P < 0.01$, $d = 0.2$) and mean energy expenditure (PRE; $1037 \pm 160 \text{ W}$, POST; $1003 \pm 160 \text{ W}$, $P < 0.01$, $d = 0.2$; **Table 2**). No main effect for time (PRE vs. POST; $n = 16$) was observed for WBSR ($1.02 \pm 0.35 \text{ L} \cdot \text{h}^{-1}$ to $1.07 \pm 0.33 \text{ L} \cdot \text{h}^{-1}$, $P > 0.05$, **Table 2**) or mean RER ($P > 0.05$) and the relative changes in plasma volume were not significantly different in endurance trained ($4 \pm 8\%$) or recreationally active participants ($3 \pm 7\%$; $P > 0.05$, $d = 0.6$). There was no main effect for training status (endurance trained vs. recreationally active), for measures of resting T_{re} , T_{re} at sweating onset, mean exercising RER and measures taken at end-exercise including: T_{re} ; heart rate; T_{sk} ; $T_{re}-T_{sk}$ gradient; RPE and thermal sensation ($P > 0.05$). However, ΔT_{re} during exercise, end-exercise PhSI, WBSR, mean $\dot{V}O_2$ and mean metabolic energy expenditure were greater in endurance trained compared with recreationally active individuals ($P < 0.05$).

DISCUSSION

The present study sought to compare heat acclimation adaptations in endurance trained and recreationally active individuals after a 6-day post-exercise HWI intervention. In agreement with our hypothesis, the new and noteworthy finding is that HWI brought about comparable heat acclimation adaptations in endurance trained and recreationally active individuals. Hallmark heat acclimation adaptations, observed in endurance trained and recreationally active individuals, included reductions in resting T_{re} and reductions in: end-exercise T_{re} ; end-exercise PhSI; T_{re} at sweating onset and T_{sk}



during exercise-heat-stress. Furthermore, training status did not significantly influence observed reductions in thermal sensation or RPE during exercise-heat-stress after the HWI intervention. The observed benefits were achieved by exposure to a large thermal stimulus for adaptation during the daily heat acclimation sessions (change in $T_{re} \approx 2^\circ\text{C}$; $T_{sk} = 40^\circ\text{C}$); despite no significant changes in WBSR or plasma volume.

The heat acclimation benefit of the HWI intervention for endurance trained individuals is emphasized by the association between the thermal stimulus (total AUC $^\circ\text{C} \cdot \text{min}^{-1}$) and the reduction in end-exercise T_{re} during exercise-heat-stress ($r = -0.71$); whereby, thermal strain during exercise-heat-stress was reduced most in endurance trained individuals who experienced the greatest thermal stimulus during the HWI

intervention. Contrary to the notion that the most highly trained individuals would benefit the least (Pandolf et al., 1977; Shvartz et al., 1977), our most accomplished endurance performer, an international marathon runner, experienced a meaningful reduction in end-exercise T_{re} after the HWI intervention (0.32°C). Nevertheless, endurance trained individuals tended to require a greater thermal stimulus during the HWI intervention to achieve a similar reduction in thermal strain as recreationally active individuals. This was likely a consequence of the endurance trained individuals' partial heat acclimatization status and associated increased heat tolerance (Piwonka et al., 1965; Strydom et al., 1966; Gisolfi and Robinson, 1969; Selkirk and McLellan, 2001). The responsible mechanism(s) for the reduction in resting T_{re} with post-exercise HWI (Zurawlew et al., 2016) and exercise-heat-acclimation (Tyler et al., 2016) require elucidation. Reductions in resting T_{re} have been related to a lowering of metabolic rate in seasonal heat acclimatization (Buguet et al., 1988) and to endurance training adaptations (Baum et al., 1976). The coupling of the reduction in resting T_{re} (-0.20°C) and T_{re} at sweating onset (-0.22°C) likely accounts for the further reduction in thermal strain during exercise heat stress after the HWI intervention (end exercise $T_{re} -0.42^\circ\text{C}$): heat loss via sweating and cutaneous vasodilation are initiated at lower thermoregulatory thresholds after heat acclimation (Buono et al., 1998).

As a consequence of their habitual exercise training, endurance trained individuals are considered to be further along the heat adaptation continuum than recreationally active individuals; reducing their adaptation potential (Taylor, 2014). It's perhaps not surprising then, and in keeping with exercise-heat-acclimation findings (Shvartz et al., 1977), that a greater reduction in exercising heart rate was observed in recreationally active than endurance trained individuals after the HWI intervention (-15 vs. -4 beats $\cdot \text{min}^{-1}$ in endurance trained). Indeed, the magnitude of the reduction in heart rate was associated with habitual endurance exercise volume ($r = 0.68$) and aerobic fitness ($r = 0.57$); whereby, heart rate was reduced most in those completing less habitual endurance exercise and those with lower aerobic fitness (Figure 2). Likely, the passive heat stimulus during HWI elicited the notable reduction in cardiovascular strain in recreationally active individuals in the present study; in agreement with the findings of others (Brebner et al., 1961; Brazaitis and Skurvydas, 2010). It's unlikely that the reduction in heart rate was due to the daily exercise as we have previously shown no improvements in cardiovascular fitness (i.e., no reduction in heart rate or $\dot{V}O_2$) in recreationally active individuals who performed the same daily exercise intervention followed by a thermoneutral bath (Zurawlew et al., 2016). The more notable reduction in heart rate in recreationally active individuals after the HWI intervention likely relates to alterations in cardiac autonomic regulation (Periard et al., 2016); adaptations already possessed by the endurance trained individuals (Carter et al., 2003). We show no obvious plasma volume expansion and a similar widening of the $T_{re}-T_{sk}$ gradient in recreationally active and endurance trained after the intervention; both mechanisms are often posited to reduce cardiovascular strain with heat acclimation (Periard et al., 2015).

TABLE 2 | Effect of 6-day post-exercise hot water immersion heat acclimation on thermal, cardiovascular, metabolic, and perceptual responses at rest and to 40-min submaximal treadmill running at 65% $\dot{V}O_{2\max}$ in the heat (33°C, 40% RH) in endurance trained and recreationally active participants.

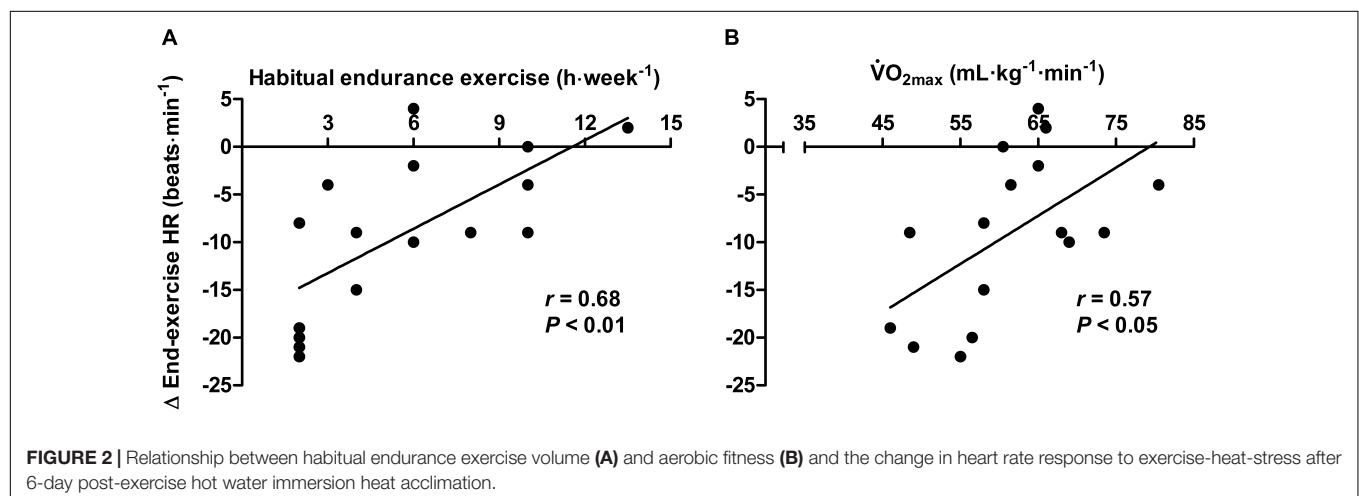
	Endurance trained	Recreationally active
T_{re} at sweating onset (°C)**	-0.22 ± 0.24	-0.23 ± 0.29
ΔT_{re} during exercise (°C)* ##	-0.19 ± 0.35	-0.25 ± 0.27
End-exercise T_{sk} (°C)**	-0.67 ± 0.38	-0.75 ± 0.70
End-exercise $T_{re}-T_{sk}$ gradient (°C)*	0.31 ± 0.42	0.27 ± 0.62
WBSR ($L \cdot h^{-1}$)##	0.13 ± 0.02	-0.03 ± 0.25
End-exercise heart rate (beats \cdot min $^{-1}$)**	-4 ± 5	-15 ± 7 ††
End-exercise PhSI (0–10)** #	-1 ± 1	-1 ± 1
Mean $\dot{V}O_2$ ($L \cdot min^{-1}$)** ##	-0.1 ± 0.1	-0.1 ± 0.2
Mean metabolic energy expenditure (W)** ##	-28 ± 31	-40 ± 50
End-exercise RPE (6–20)*	-1 ± 1	-2 ± 3
End-exercise thermal sensation (1–13)**	-1 ± 1	-1 ± 1

T_{re} , rectal temperature; T_{sk} , mean skin temperature; WBSR, whole body sweat rate; PhSI, physiological strain index; RPE, rating of perceived exertion. Data displayed as mean \pm SD of the PRE to POST change. * $P < 0.05$ and ** $P < 0.01$, denotes POST different than PRE (main effect of time). # $P < 0.05$ and ## $P < 0.01$, denotes endurance trained different than recreationally active (main effect of group). †† $P < 0.01$, denotes POST lower than PRE within group (post hoc time effect).

Hallmark adaptations to the heat have long been considered to include an expansion in resting plasma volume (Greenleaf et al., 1983) and an increase in WBSR during exercise-heat-stress (Wyndham and Strydom, 1969). Corroborating our recent work in recreationally active individuals (Zurawlew et al., 2016, 2018), the current findings demonstrate that post-exercise HWI also reduces thermal strain during exercise-heat-stress in endurance trained individuals; despite no obvious increase in plasma volume or WBSR. It's noteworthy that a recent meta-analysis highlighted the rather modest and variable plasma volume expansion ($+4 \pm 5\%$) and increase in WBSR ($+5 \pm 11\%$) in short-term heat acclimation studies (<7 exposures) (Tyler et al., 2016). Typically, >7 exercise-heat-acclimation exposures are required

to initiate an increase in WBSR; but even then, these responses are highly variable ($+29 \pm 29\%$) (Tyler et al., 2016). The semi-recumbent body position and hydrostatic forces during HWI may maintain central vascular volume and in-turn reduce fluid regulatory stress and the stimulus for plasma volume expansion (Nagashima et al., 1999; Bradford et al., 2015). However, the absence of an expansion in plasma volume may be associated with errors in estimating relative changes in plasma volume using hemoglobin and hematocrit; therefore, future research should verify this finding using tracer techniques. Immersing the skin in hot water has been shown to reduce sweat gland activity and the stimulus for an increase in sweating (Hertig et al., 1961; Brebner and Kerslake, 1968). However, a more likely explanation for the lack of an increase in WBSR with HWI is that the decrease in T_{re} at sweating onset (-0.22°C) was offset by the decrease in resting T_{re} (-0.20°C). No increase in WBSR during exercise-heat-stress after the HWI intervention may provide additional thermoregulatory and performance benefits during exercise-heat-stress, by constraining dehydration and preserving central blood volume (Montain and Coyle, 1992). Heat acclimation by post-exercise HWI may limit the “wasteful overproduction of sweat” (Mitchell et al., 1976); particularly important in high humidity conditions when evaporative heat loss is limited and when sweat may drip from the skin. Notwithstanding, we recognize that the relatively modest exercise-heat stress for experimental trials (65% $\dot{V}O_{2\max}$, 33°C, 40% RH) may have masked an increase in WBSR (Poirier et al., 2015); as such, studies should investigate the influence of heat acclimation by post-exercise HWI on WBSR during a more uncompensable exercise scenario.

Despite evidence that exercise-heat-acclimation alleviates thermal strain and improves performance in the heat (Nielsen et al., 1997; Lorenzo et al., 2010), practical barriers limit athlete engagement with current exercise-heat-acclimation recommendations (Tyler et al., 2016; Casadio et al., 2017; Periard et al., 2017). As such, there has been an increasing interest of late in practical heat acclimation methods; including, training in temperate conditions whilst wearing additional clothing (Stevens et al., 2018) and post-exercise HWI



(Zurawlew et al., 2016, 2018). Exercise in temperate conditions wearing additional clothing may provide the necessary elevations in core and skin temperature for adaptation (Dawson et al., 1989; Ely et al., 2018). However, a recent field study in triathletes showed that training in temperate conditions (18°C) wearing additional clothing was not an effective heat acclimation strategy (Stevens et al., 2018). For endurance trained athletes residing and training in temperate conditions, the current findings support the recommendation that incorporating a hot bath (lasting up to 40 min) in the post-exercise washing routine represents an effective and accessible heat acclimation strategy to prepare for competition in the heat. Taking a hot bath after temperate exercise limits interference with an athlete's training and taper and does not require access to an environmental chamber or precise control over exercising core temperature. The current findings (delta end exercise T_{re} -0.42°C), and our previous work (Zurawlew et al., 2016, 2018), show that the magnitude of adaptations following post-exercise HWI compare favorably with exercise-heat-acclimation interventions, as reported in a recent meta-analysis (delta exercise core temperature -0.34°C) (Tyler et al., 2016). Therefore, the findings from the current study, considered alongside the extant literature, do not support the notion that exercise-heat-acclimation evokes superior adaptation and should be recommended in favor of passive heat acclimation (Periard et al., 2016; Tyler et al., 2016). Notwithstanding, future studies should directly compare post-exercise HWI and exercise-heat-acclimation and confirm whether the observed adaptations in endurance trained individuals translate to improved aerobic performance; as was previously observed in recreationally active individuals (Zurawlew et al., 2016). A small handful of studies provide evidence of heat acclimation with repeated HWI alone (without prior exercise) (Brebner et al., 1961; Bonner et al., 1976; Brazaitis and Skurvydas, 2010); as such, studies may wish to compare the efficacy of HWI with and without prior exercise. However, unpublished observations in our laboratory show a larger thermal stimulus (daily AUC $^{\circ}\text{C} \cdot \text{min}^{-1}$) for post-exercise HWI than for the HWI alone strategies used in two of these studies (Brebner et al., 1961; Bonner et al., 1976). Moreover, we have concerns regarding participant safety and tolerance to the unpleasantly high water temperature (44°C) used in another of these studies (Brazaitis and Skurvydas, 2010). On the one hand,

studies should determine whether meaningful heat acclimation can be achieved by fewer and/or shorter post-exercise HWI exposures. On the other hand, mindful of safety and practical constraints, exploring whether the observed adaptations can be further augmented by increasing the intensity of the prior exercise, or increasing the number and/or duration of HWI exposures requires investigation. Finally, studies should also determine the rate of decay of heat acclimation after the 6-day post-exercise HWI intervention and whether the adaptations observed herein translate to females.

CONCLUSION

Hot water immersion after exercise in temperate conditions on 6 consecutive days reduced thermal strain during exercise-heat-stress in endurance trained and recreationally active individuals. For high level athletes residing and training in temperate conditions, incorporating a hot bath in the post-exercise washing routine represents an effective heat acclimation strategy to prepare for major competition in the heat.

AUTHOR CONTRIBUTIONS

NW had primary responsibility for the final content. NW, JM, and MZ were involved in the conception of the project and development of the research plan. MZ led the data collection. NW, JM, and MZ performed the data analysis, interpreted the data and prepared the manuscript.

ACKNOWLEDGMENTS

We would like to thank the following people for their valuable assistance with data collection: Claire Potter, Tom Ibbitson, David Harding, Liam Renton, Michael Gregson, Jonathan Donoghue, Lauren Casling, Benjamin Price, Thomas Storer, Harry Brown, Michael Williams, George Hunt, Lucy Beighton, Jason Edwards, and Kevin Williams. We are also indebted to the participants for their time and co-operation.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Long-Haul Northeast Travel Disrupts Sleep and Induces Perceived Fatigue in Endurance Athletes

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OPEN ACCESS

Edited by:

Toby Mündel,
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Reviewed by:

Melissa Skein,
Charles Sturt University, Australia
Greg Atkinson,
Teesside University, United Kingdom

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 19 October 2018

Accepted: 06 December 2018

Published: 20 December 2018

Citation:

Stevens CJ, Thornton HR,
Fowler PM, Esh C and Taylor L (2018)
Long-Haul Northeast Travel Disrupts
Sleep and Induces Perceived Fatigue
in Endurance Athletes.
Front. Physiol. 9:1826.
doi: 10.3389/fphys.2018.01826

Introduction: Long-haul transmeridian travel is known to cause disruptions to sleep and immune status, which may increase the risk of illness.

Aim: This study aimed to determine the effects of long-haul northeast travel for competition on sleep, illness and preparedness in endurance athletes.

Methods: Twelve trained (13.8 ± 3.2 training h/week) masters (age: 48 ± 14 years) triathletes were monitored for sleep (quantity via actigraphy and quality via self-report), mucosal immunity (salivary immunoglobulin-A) and stress (salivary cortisol) as well as self-reported illness, fatigue, recovery and preparedness. Baseline measures were recorded for 2 weeks prior to travel for all variables except for the saliva samples, which were collected on three separate days upon waking. Participants completed normal training during the baseline period. Measures were subsequently recorded before, during and after long-haul northeast travel from the Australian winter to the Hawaiian summer, and in the lead up to an Ironman 70.3 triathlon.

Results: All comparisons are to baseline. There was a *most likely* decrease in sleep duration on the over-night flight (-4.8 ± 1.2 h; effect size; $\pm 90\%$ confidence limits = 3.06; ± 1.26) and a *very likely* increase in sleep duration on the first night after arrival (0.7 ± 1.0 h; 1.15; ± 0.92). After this time, sleep duration returned to baseline for several days until it was *very likely* decreased on the night prior to competition (-1.2 ± 1.0 h; 1.18; ± 0.93). Nap duration was *likely* increased on the first day after arrival (36 ± 65 min; 3.90; ± 3.70). There was also a *likely* increase in self-reported fatigue upon waking after the first night in the new destination (1.1 ± 1.6 AU; 0.54; ± 0.41) and there were three athletes (25%) who developed symptoms of illness 3–5 days after arrival. There were no changes in sleep quality or mucosal measures across study.

Discussion: Long-haul northeast travel from a cool to a hot environment had substantial influences on sleep and self-reported fatigue, but these alterations had returned to pre-departure baseline 48 h after arrival. Endurance athletes undertaking similar journeys may benefit from optimizing sleep hygiene, especially on the first 2 days after arrival, or until sleep duration and fatigue levels return to normal.

Keywords: air travel, hot environment, sleep, immunity, illness, fatigue, salivary IgA, salivary cortisol

INTRODUCTION

Insufficient sleep and/or illness can disrupt an athlete's training, competition and performance-recovery (Pyne et al., 2005; Rae et al., 2017). Athletes are susceptible to reductions in sleep duration and quality during training and proximal to competition, especially the night prior to competition (Juliff et al., 2015; Roberts et al., 2018), albeit subject to high inter- and intra-individual variability (Nedelec et al., 2018). Athletes often relocate from various global locations (e.g., home training base, pre-competition or holding-camp) to compete at major competitions, meaning many athletes will relocate several times across numerous continents before a competition. Sleep loss has consistently been reported during long-haul travel in economy class (Fowler et al., 2015, 2017), likely due to the cabin conditions, including the uncomfortable sleeping position (Roach et al., 2018). Light, noise and the timing of meals and stopovers may also contribute to sleep disruption on a long-haul flight. Airliner cabin conditions can increase the risk of illness, particularly drying of the respiratory epithelium due to the low humidity, close contact with fellow infectious travelers and exposure to their re-circulated air (Schwellnus M. P. et al., 2012b; Svendsen et al., 2016). Moreover, athletes could be at greater risk of travel induced illness compared to the general population, since prolonged exercise and intensified training are known to suppress mucosal immunity, increasing the risk of upper respiratory tract infections (URTI) (Walsh et al., 2011). Moreover, insufficient sleep itself increases susceptibility to respiratory infections and airborne viruses (Prather et al., 2015; Prather and Leung, 2016).

Long-haul transmeridian air-travel can elicit jet-lag symptomatology, predominately due to misalignment between body clock time (as indicated by the circadian rhythm in body temperature or melatonin) and local time at the new location (Reilly et al., 2005; Forbes-Robertson et al., 2012). Jet-lag mediated sleep disruption is not uncommon, with delayed sleep onset and early awakening common after eastward and westward travel, respectively (Thun et al., 2015; Fowler et al., 2017). Such responses can temporarily reduce facets of performance (Fullagar et al., 2015), with emerging evidence suggesting that direction of travel (e.g., eastward travel eliciting a greater detrimental performance effect than westward) is important (Fowler et al., 2017). Other detrimental physiological and perceptual responses are also seen (Fullagar et al., 2015). While several studies have characterized sleep responses to east and westward travel (Thun et al., 2015; Fowler et al., 2017), there is a lack of research on long-haul travel that causes little disruptions to the sleep-wake cycle (i.e., little jet-lag), where fatigue from the travel itself can be studied without influences from large shifts in time-zone.

Factors associated with the destination may also have a negative influence on sleep and illness risk upon arrival. The environmental conditions, food, and exposure to different pathogens upon arrival were purported as reasons for the twofold to threefold increase in the incidence of all illness in professional rugby union players following international travel (Schwellnus M. et al., 2012a). A large climatic contrast between the place of departure and destination is also suggested to be a risk factor for illness (Schwellnus M. et al., 2012a) and perhaps sleep disruption. For example, given the circadian influence of a reduction in body temperature on sleep onset (Wyatt et al., 1999), exposure to and/or exercise in the heat, particularly in the afternoon/evening, may negatively affect sleep (Buguet, 2007). Lastly, athletes training/competing in the heat may be at increased risk of heat-related illness (Racinais et al., 2015), especially if they are coming from a cooler climate and have not been able to completely heat acclimatize (Heathcote et al., 2019). Hence, athletes traveling from a cool to a hot environment may be at increased risk of sleep disruption, lowered immunity and illness.

Therefore, the aim of the current study was to determine the effects of long-haul northeast travel from a cool (Winter of New South Wales, Australia) to a hot environment (Summer of Hawaii, United States) for competition on sleep, mucosal immunity and stress, as well as illness, fatigue, recovery and preparedness in endurance athletes. We hypothesize that the travel will; (i) decrease sleep duration on the day of the flight; (ii) decrease salivary IgA (sIgA) concentration on arrival; and (iii) negatively influence self-reported fatigue, recovery and/or preparedness on arrival, compared to baseline.

MATERIALS AND METHODS

Participants

Twelve masters level triathletes (age: 48 ± 14 years, height: 172 ± 11 cm, body mass: 72 ± 11 kg) volunteered to participate in the study. The athletes had a mean weekly training duration of 13.8 ± 3.2 h (range = 10.4–20.2 h) across the first 3 weeks of May 2017 (i.e., the last month before competition but prior to commencing a taper on the day of departure). The sample included both 'trained' and 'well-trained' athletes according to published guidelines (De Pauw et al., 2013). Training was completed on the mid-north coast of Australia, predominantly during early mornings of the autumn season (10–15°C). Hence, the athletes were not seasonally acclimatized, however, they did perform three to five training sessions in

additional clothing (Stevens et al., 2017) within 4 weeks of the race. The protocol was approved by the Southern Cross University Human Research Ethics Committee. All subjects gave written informed consent in accordance with the Declaration of Helsinki.

Design

This prospective cohort study monitored sleep (quality and quantity), mucosal markers of immunity (sIgA) and stress (salivary cortisol [sCort]) as well as self-reported symptoms of illness, fatigue, recovery and preparedness before and after long-haul transmeridian travel from the Australian Winter to the Hawaiian Summer in preparation for the 2017 Hawaii Ironman 70.3 triathlon. An experimental schematic of the timeline of the measures is illustrated in **Figure 1**. **Figure 2** illustrates the mean, maximum and minimum ambient temperatures, as well as the mean daily humidity experienced by the athletes, in the days prior to travel and after arrival in the new destination.

Travel

The travel involved 22.6 ± 2.4 h of door-door travel time including 11.5 h of flight time. All athletes traveled in economy class for all flights. The journey involved;

- (1) Road transfer from individual homes (15:00–17:00 AEST) to Coffs Harbour airport, Australia (0.1–0.5 h)
- (2) Flight to Sydney, Australia (1.25 h)
- (3) Flight to Honolulu, United States (9.5 h)
- (4) Flight to Kona, United States (0.75 h)
- (5) Road transfer from Kona airport to accommodation in Waikoloa, United States (0.75 h; arriving between 15:30 and 17:00 HST).

Stopovers in Sydney and Honolulu ranged from 2 to 5 h depending on the airline, but all athletes experienced the same flight duration (and hence very similar routines on the flights themselves in terms of meals and light exposure). Eight of the athletes departed on the 29th, three departed on the 28th and one departed on the 27th of May, 2017, arriving at the accommodation on the same day that they departed due to the shift in time zone (−22 h).

Measures

Sleep

Sleep was monitored using self-report diaries and wrist activity monitors (wActiSleep+, Actigraph, FL, United States) for 3 weeks (15th May to the 4th June 2017). According to previously described methods, data from the sleep diaries and activity monitors were used to determine when participants were awake and asleep (Sargent et al., 2016). All time was scored as wake unless: (i) the sleep diary indicated that the participant was lying down attempting to sleep and (ii) the activity counts from the monitor were sufficiently low to indicate that the participant was immobile (i.e., where the weighted activity count for an epoch fell below the defined threshold). When these two conditions were satisfied simultaneously, time was

scored as sleep. This scoring process was conducted using the corresponding software (Actilife, version 6.13.3, Actigraph, FL, United States) and Cole-Kripke algorithm, which has been validated for use in adults (Cole et al., 1992). The following variables were derived from the sleep diary and activity monitor data; sleep duration (h:min): the amount of time spent in bed asleep and sleep efficiency (%): sleep duration expressed as a percentage of time in bed. Participants also self-reported nap duration (min; there was no minimum) and sleep quality (where 1 = very poor, 2 = poor, 3 = fair, 4 = good, 5 = very good). All sleep on the flight(s) was scored via actigraphy as ‘night time sleep’ i.e., participants were instructed to exclude this from their self-reported nap duration.

Mucosal

The sIgA and sCort were determined from saliva samples analyzed with an IPRO cube reader (IPRO Interactive, Wallingford, United Kingdom), that has been deemed valid ($r = 0.93$) and reliable (ICC = 0.89, CV = 9.4%) for the measurement of sIgA (Coad et al., 2015) and valid ($r = 0.52$) and reliable ($r = 0.69$) for measurement of sCort (Fisher et al., 2015). All samples were collected upon waking within a 1-h window for each individual (with the exception of race day, where wake time was 3:30 AM) and prior to any exercise, food or fluid ingestion. An oral swab was placed on the top of the tongue, and when an adequate volume of saliva (0.5 mL) was collected, the volume-indicator turned blue. Swabs were then placed into a buffer solution provided by the manufacturers, and two drops of the buffer-saliva mixture were placed on the dual-cassette test strip. Following a 10-min incubation period, the test strip was placed in the IPRO to determine raw concentrations of sIgA ($\mu\text{g mL}^{-1}$), and sCort (ng mL^{-1}). As per **Figure 1**, saliva samples were collected on three occasions at baseline in the week prior to travel, and another sample was collected on the day of the travel. Further, samples were collected on the day after arrival and then on a daily basis until the day following the event. The mucosal data excluded one participant who reported illness in the days prior to the flight, which persisted throughout the flight and for 2 days after arrival. A further three participants were also removed due to inadequate sample preparation/technical problems.

Self-Report

The athletes self-reported several measures on a daily basis upon waking throughout the study period. This included symptoms of illness via the athlete illness questionnaire (Matthews et al., 2010), training load via session-rating of perceived exertion (Foster et al., 2001) as well as measures of fatigue (“rate your fatigue over the last 24 h”; 0 = no fatigue, 10 = as bad as you can imagine); recovery (“how well recovered are you?”; 0 = very poorly, 10 = very well); physical preparedness (“how physically ready are you for strenuous exercise?”; 0 = not ready at all, 10 = totally ready), and; mental preparedness (“how mentally ready are you for strenuous exercise?”; 0 = not ready at all, 10 = totally ready).

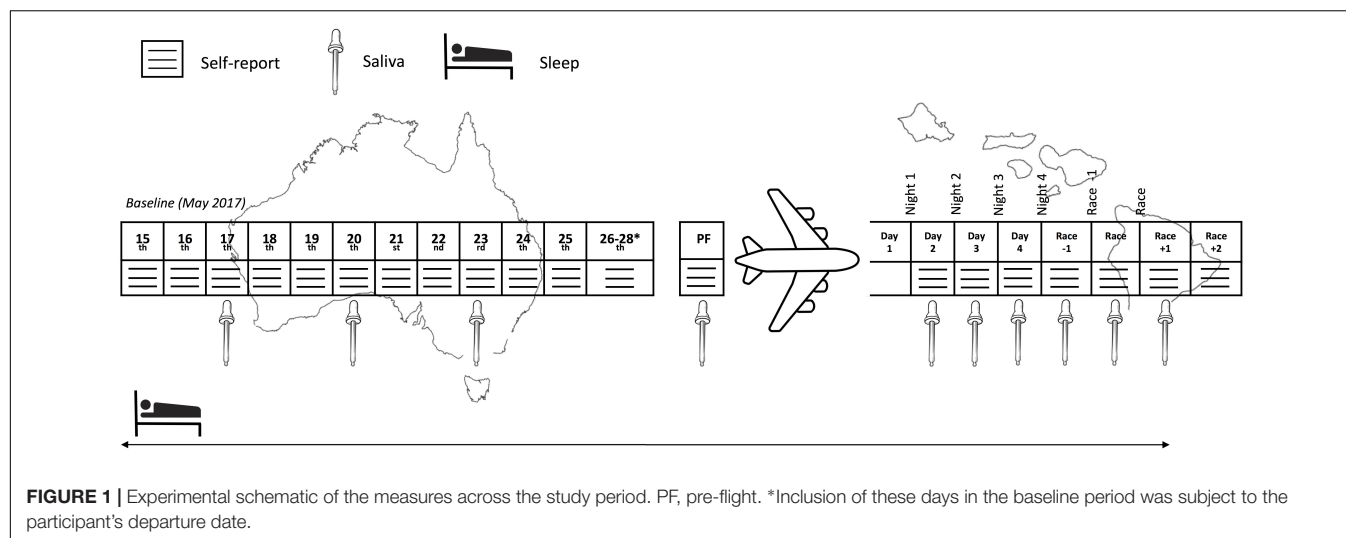


FIGURE 1 | Experimental schematic of the measures across the study period. PF, pre-flight. *Inclusion of these days in the baseline period was subject to the participant's departure date.

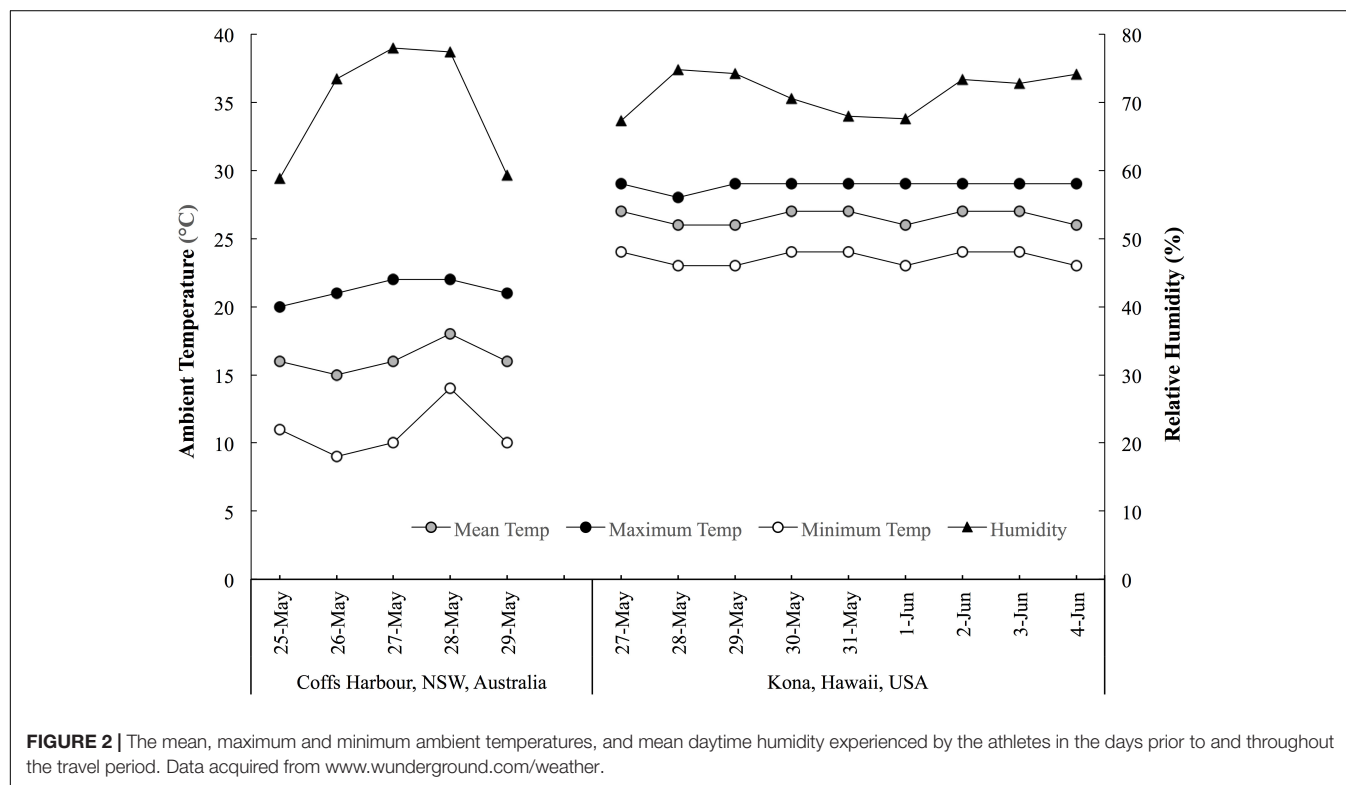


FIGURE 2 | The mean, maximum and minimum ambient temperatures, and mean daytime humidity experienced by the athletes in the days prior to and throughout the travel period. Data acquired from www.wunderground.com/weather.

Data Analysis

For the purposes of analysis, the following periods/days were defined as per **Figure 1**; *Baseline*: Two-week period immediately prior to the travel, where athletes lived in their usual home environment and continued with routine training; *Pre-flight*: The day/night prior to travel; *Flight*: The data recorded during the international flight; *Days/Nights 1 to 4*: First 4 days/nights after arrival; *Race -1*: Day/night prior to the race; *Race*: Day/night of the race; *Race +1*: Day following the race; *Race +2*: Two days following the race. For the mucosal markers, both the raw data and

the change (delta; Δ) from the individual's baseline were determined.

Statistical Analysis

Measurements are presented as mean \pm standard deviation (SD) and were analyzed using a non-clinical magnitude-based inference approach (Hopkins et al., 2009). The data were log-transformed (i.e., $100 \times$ natural log) and the magnitudes of the changes between trials were expressed as standardized differences (effect sizes; ES) with 90% confidence limits (CL). The criteria used for interpreting the magnitude of the ES were: ≤ 0.2

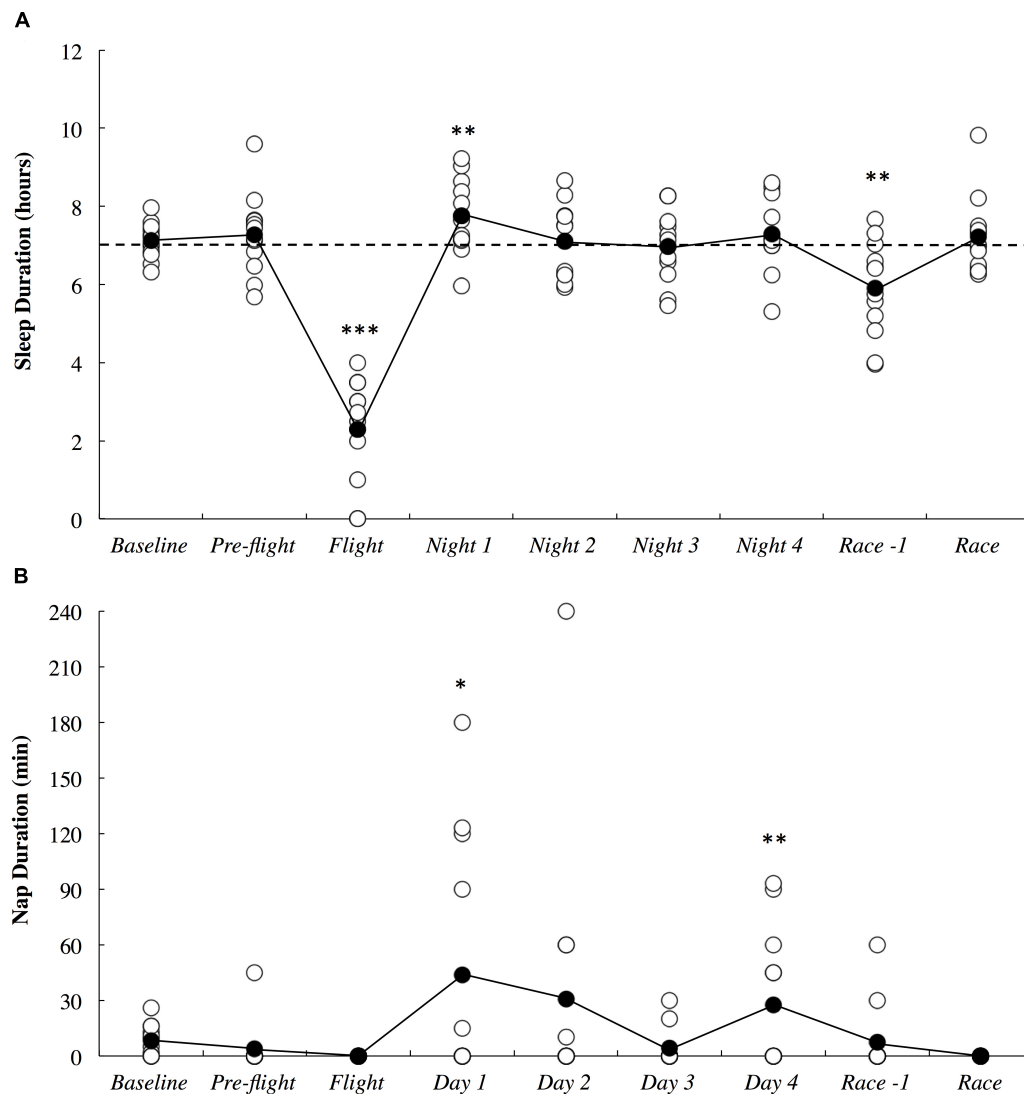
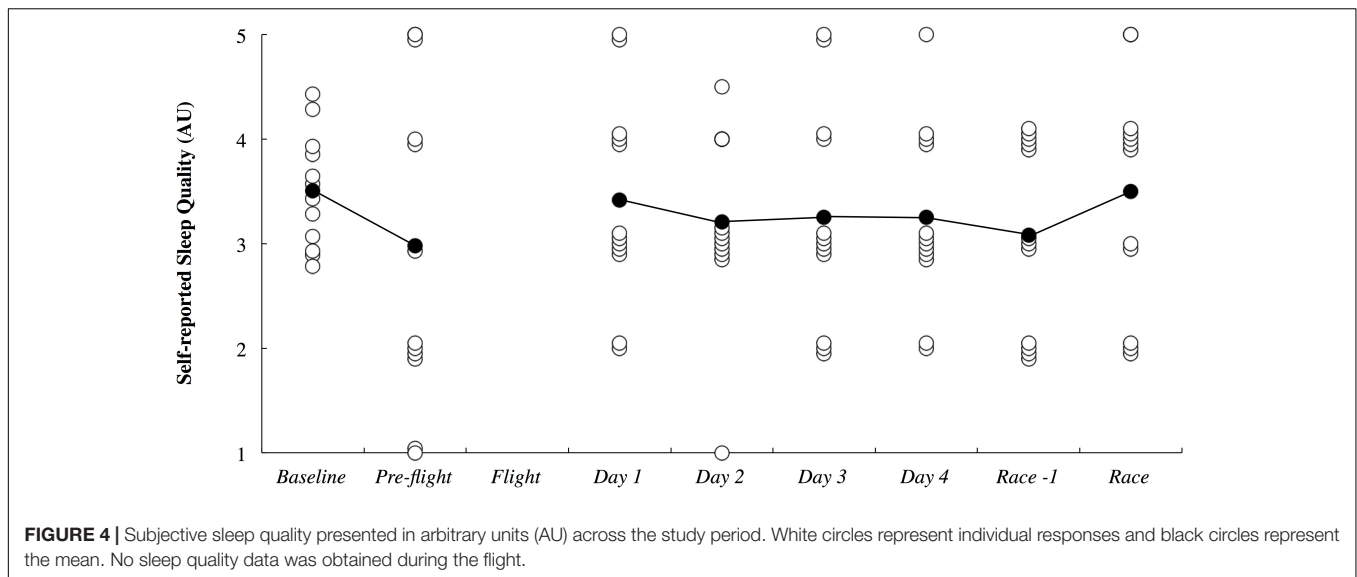


FIGURE 3 | Objective sleep duration (A) and subjective nap duration (B) across the study period. White circles represent individual responses and black circles represent the mean. Substantial differences compared to baseline are denoted as **likely*, ***very likely* and ****most likely*. The dotted line represents the minimum sleep duration recommended for adults.

(trivial), >0.2 (small), >0.6 (moderate), >1.2 (large) and >2.0 (very large) as described previously (Hopkins et al., 2009). If the 90% CL overlapped positive and negative trivial ES values then the effect was deemed unclear. The quantitative chances of differences being substantial were assessed qualitatively as follows: 75–95% (*likely*); 95–99% (*very likely*); $>99\%$ (*most likely*) as described previously (Hopkins et al., 2009) and determined using a published spreadsheet (xPostOnlyCrossover.xls) available online (Hopkins, 2017).

Relationships between athletes' sleep duration and each outcome measure (mucosal measurements and self-report responses) were assessed using linear mixed models. In these models, sleep duration (minutes) was included as the predictor variable (fixed effect), and separately, mucosal and self-report responses were included as the outcome measure. Using a

random intercept and slope design, athlete identification and the corresponding outcome variable were included as random effects. The relationship between sleep duration and the outcome variables were standardized by multiplying the final model slope by $2 \times$ the within-subject standard deviation that were obtained using a mixed model reliability analysis with a random effect for athlete identification (Higham et al., 2014). This method results in the expected change in the outcome measure from a typically low (-1 SD) to a typically high value ($+1$ SD) (Hopkins et al., 2009). This effect (expressed as a SD) was then converted to an ES using the between-subject standard deviation (obtained from the mixed model reliability analysis), that were categorized using the ES magnitude thresholds as described previously and were also interpreted using the magnitude-based inference approach as stated above. These analyses were



performed using customized R Studio statistical software (V 1.1.453), and packages including lme4, lmerTest and emmeans were used.

RESULTS

The objective sleep duration and self-reported nap duration are illustrated in **Figure 3**. When compared to baseline, sleep duration was *most likely* lower during the international flight ($3.06; \pm 1.26$) and *very likely* higher on Night 1 ($1.15; \pm 0.92$). Sleep duration returned to baseline thereafter until it was *very likely* lower on the night prior to competition ($1.18; \pm 0.93$). Compared to baseline, nap duration was *likely* higher on day 1 ($3.90; \pm 3.70$) and *very likely* higher on day 4 ($2.13; \pm 1.77$). Self-reported sleep quality is illustrated in **Figure 4**. There were no changes in subjective sleep quality or objective sleep efficiency compared to baseline. No sleep occurred during either of the short domestic flights.

The absolute and delta sIgA and sCort concentrations across the study period are illustrated in **Figure 5**. There were no substantial differences in sIgA and sCort responses compared to baseline on any day throughout the study period.

There were three athletes (25%) who developed symptoms of illness 3–5 days after arrival in the new destination. Athlete A developed ‘severe’ upper respiratory symptoms that persisted for 5 days (including race day). Athlete B developed ‘minimal’ upper respiratory symptoms that persisted for 1 day. Athlete C developed ‘minimal’ symptoms of chest infection and ‘moderate’ symptoms of headache which both persisted for 3 days.

Subjective measures of fatigue, recovery and preparedness are illustrated in **Figure 6**. All comparisons are made compared to baseline. Fatigue ratings were *likely* higher upon waking after the first night in the new destination ($0.54; \pm 0.41$), *likely* lower on the day of the race ($0.78; \pm 1.27$), *very likely* higher on the day after the race ($1.58; \pm 0.96$) and *likely* higher 2 days after the race ($0.83; \pm 1.13$). Recovery ratings were *likely* higher on the day of

the race ($1.67; \pm 2.94$), were *most likely* lower the day after the race ($2.21; \pm 0.81$) and were *likely* lower 2 days after the race ($1.14; \pm 1.00$). Physical preparedness was *most likely* lower the day after the race ($2.64; \pm 0.90$) and was *likely* lower 2 days after the race ($1.09; \pm 0.92$). Mental preparedness was *likely* higher on the day of the race ($1.25; \pm 1.89$), *most likely* lower on the day after the race ($1.91; \pm 0.83$) and was *likely* lower 2 days after the race ($1.03; \pm 0.88$).

There were meaningful associations between sleep duration and various self-reported measures. Specifically, sleep had a *very likely* association with both fatigue ($0.92; \pm 0.55$) and perceived sleep quality ($0.84; \pm 0.64$). There was also a *likely* negative association between sleep duration and recovery ($-1.04; \pm 1.21$). Associations between sleep duration and sIgA and sCort were unclear.

The session RPE training load data is illustrated in **Figure 7**. All comparisons are made to baseline measurements. All of the athletes had a rest day on day 1, and as such, training load was *most likely* lower on this day ($2.65; \pm 0.48$). Training load was *very likely* lower on day 2 ($5.30; \pm 3.34$) and day 3 ($1.78; \pm 1.07$) until it was *most likely* lower on day 4 ($2.22; \pm 1.03$) and the day before the race ($5.98; \pm 2.76$). Training load was *most likely* higher on the day of the race ($4.32; \pm 0.80$) and *most likely* lower on both days following the race when no training was completed ($2.65; \pm 0.48$).

Total Hawaii Ironman 70.3 race duration for male (range h:min: 4:59–6:41) and female (7:42–8:30) athletes show some variance. These durations do not securely reflect the training status of the athletes, due to the extreme heat and arduous terrain of this event.

DISCUSSION

The current study demonstrated that long-haul northeast travel from a cool to a hot environment had *most likely* and *likely* negative influences on sleep and self-reported fatigue, respectively, in masters level endurance athletes. Sleep duration

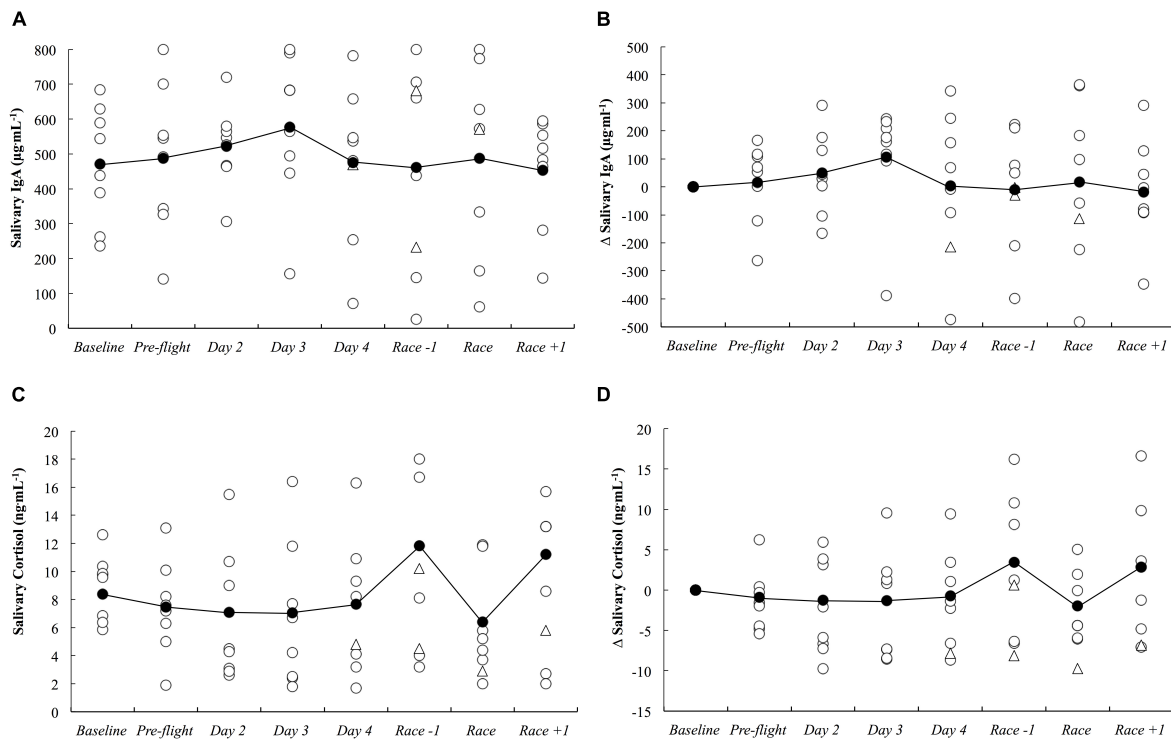


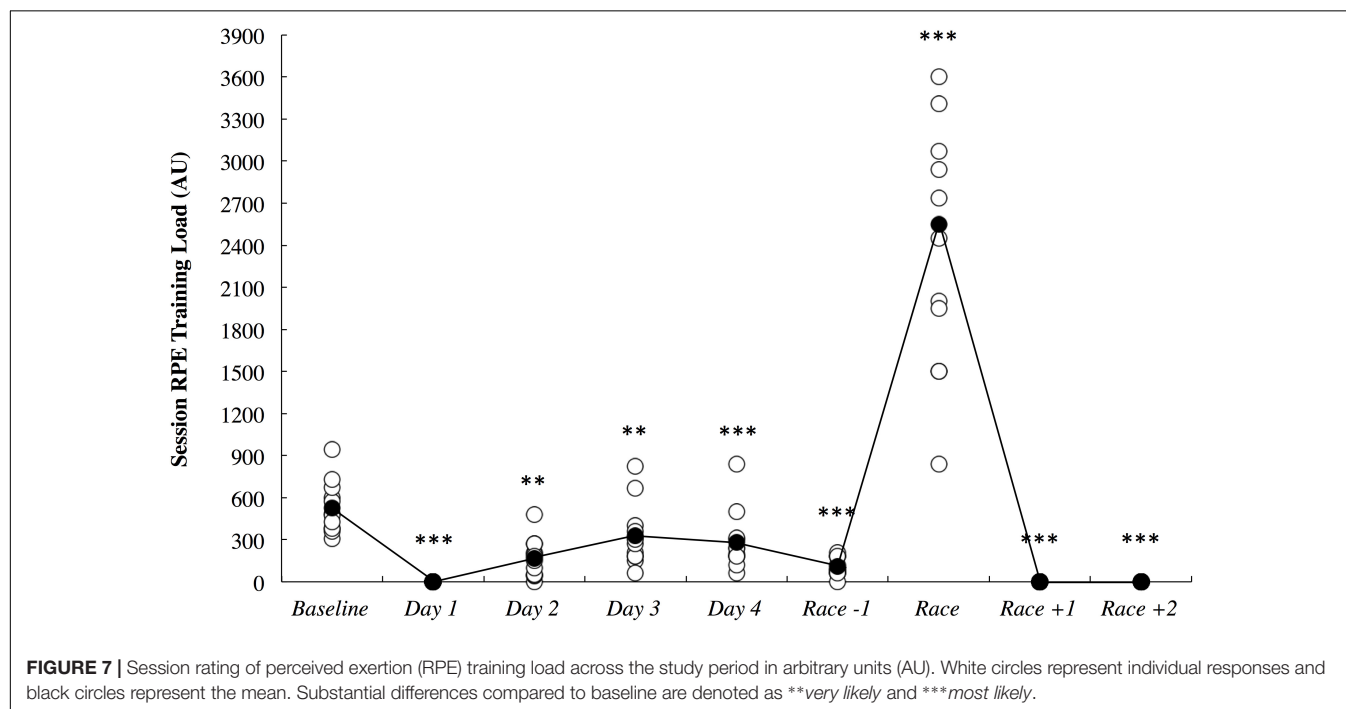
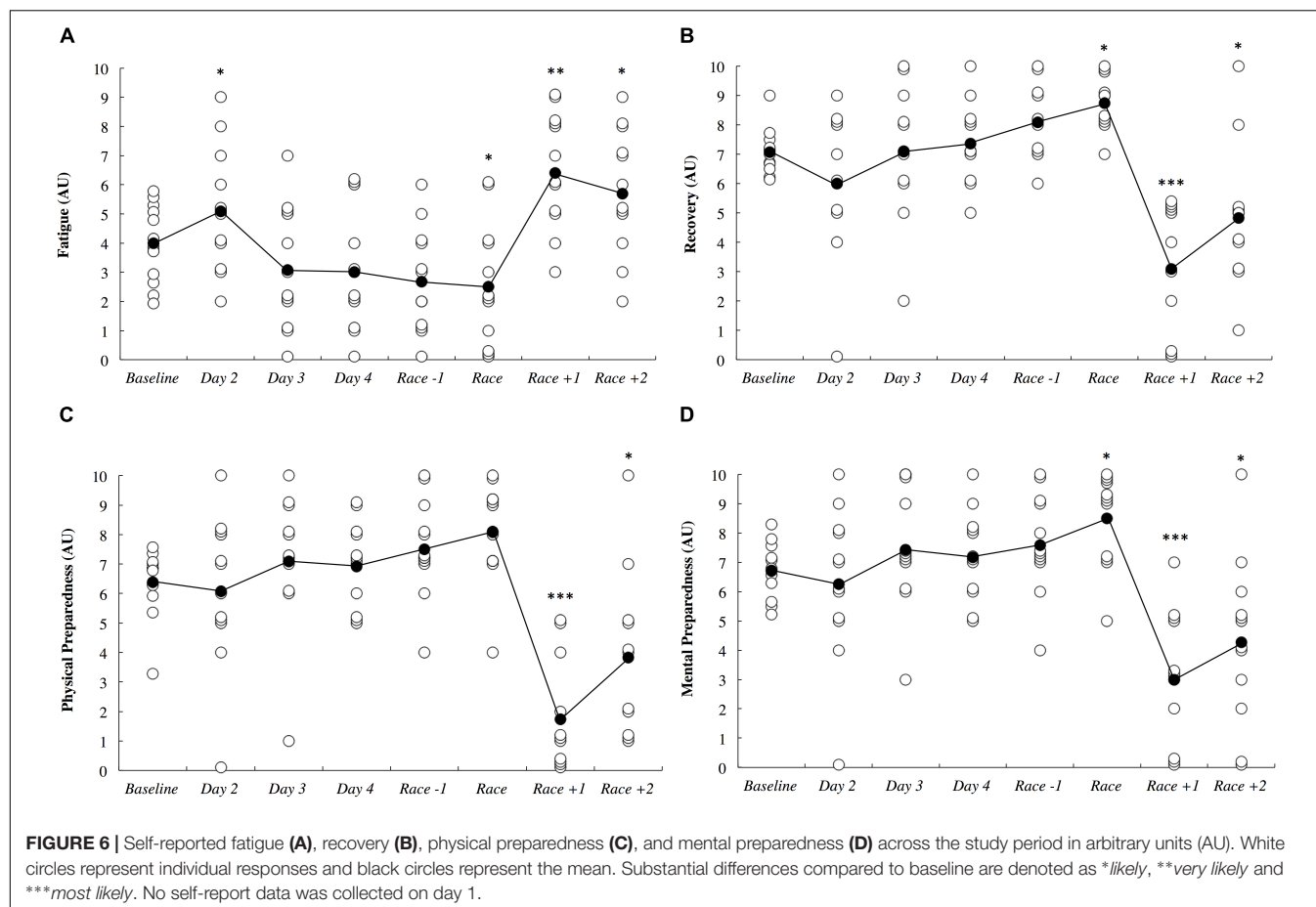
FIGURE 5 | Salivary IgA concentration as absolute (A) and delta (B) values, and salivary cortisol concentration as absolute (C) and delta (D) values across the study period. White circles represent individual responses, white triangles represent individual responses when they reported illness symptoms and black circles represent the mean.

was *most likely* decreased on the overnight flight, and to compensate, nap duration was *likely* increased on the first day, and sleep duration was *very likely* increased on the first night after arrival (Figure 3). Upon waking after the first night, the athletes reported *likely* greater fatigue (Figure 6A). Three athletes (25%) developed symptoms of illness 3–5 days after arrival at the destination, but there were no changes in mucosal markers of immunity or stress throughout the study period (Figure 5). As such, hypothesized ‘i’ and ‘iii’ are accepted, and hypothesis ‘ii’ is rejected. On the day of competition (Hawaii Ironman 70.3), fatigue, recovery and mental preparedness were *likely* improved (Figure 6) corresponding with the tapering training program (Figure 7), but sleep duration was *very likely* decreased on the night prior to the competition. Overall, these findings are important and useful for practitioners when planning competition that involves international long-haul travel. In particular, interventions are may assist in reducing sleep disruption during travel and the night prior to competition, together with minimizing the risk of illness.

Sleep duration was dramatically lower on the overnight international flight compared to a normal night of sleep when the athletes were in their own homes (Figure 3A). This observation may have been caused by the uncomfortable upright seating/sleeping position (Roach et al., 2018), light, noise and the unusual environment and routine experienced on the flight. The *very likely* increased sleep duration observed on the first night after arrival is a common occurrence due to a greater homeostatic

sleep pressure as a result of reduced sleep duration on the flight (Fowler et al., 2015; Fullagar et al., 2016). On average, however, more sleep duration was lost during the flight (4.8 h) compared to what was regained through daytime napping (0.7 h) and sleeping (0.7 h) on the first day/night after arrival. These athletes may have had difficulty catching up on the sleep lost on this first night due to: (i) additional daytime napping interrupting evening sleep; (ii) additional evening socializing associated with staying in an overseas resort with friends and family; (iii) early awakening due to the change in time-zone (–22 h) and; (iv) scheduled morning training at 07:00. Hence, the athletes may have found it difficult to get to sleep any earlier than normal in the new environment, while still having morning training demands (albeit slightly later than usual) on each day after arrival. Relative to the American National Sleep Foundation, several of the athletes slept less than the recommended minimum duration of 7 h for adults (see Figure 3A) both at baseline and throughout the study period (Hirshkowitz et al., 2015). For these athletes, sleep education and sleep hygiene interventions may be beneficial before traveling, while the major challenge for athletes already sleeping 7 h and above would be compensating for sleep lost upon arrival.

The athletes reported *likely* increased fatigue on the morning after the first night in the new destination, possibly due to the lack of sleep on the flight, combined with the difficulty of compensating for lost sleep on the first night in the new environment. Living in the hotter environment may have also contributed to the additional fatigue, as a large amount of time



was spent outside in the heat (away from air conditioning). Both sleep duration, and self-reported fatigue had returned to baseline by the second night and third morning after arrival, respectively. More generally, high levels of fatigue and low levels of recovery were associated with increased sleep duration across the study period, as identified via linear mixed models. Therefore, the first 48 h after long-haul travel is perhaps a key time to optimize sleep hygiene practices in athletes traveling for competition. Sleep could also be positively influenced by employing napping to further compensate for the air-travel induced decrements in total sleep duration. While some of the athletes napped on the day of arrival (5/12) and the day after arrival (4/12), this strategy could have been beneficial for all athletes. Indeed, evidence suggests that naps timed appropriately after previous exercise and taken later in the day (13:00–15:00) may positively influence an athletes willingness to engage in further physical and mental efforts (Bonnar et al., 2018). Further, findings regarding professional rugby league athletes demonstrated napping did not impede night time sleep, rather increasing total sleep quality and quantity (Thornton et al., 2017). Other acute sleep hygiene strategies (e.g., appropriate lighting, electronic device availability, cool room temperature, ear plugs, eye masks, etc.) have had varying effects on sleep quality and duration in athletes (Bonnar et al., 2018) and could be employed on an individual basis as needed.

While others have demonstrated significant associations between a habitual sleep duration of <6 h and an increased risk of the common cold (Prather et al., 2015), we observed no changes in mucosal markers of sIgA or sCort (Figure 5) on day 2 onward following the overnight travel (sleep duration on the flight was <4 h in all athletes). Indeed, associations between sleep duration and sIgA and sCort were unclear within the present data. However, the mucosal data may be confounded by the much lower training loads completed upon arrival compared to baseline (Figure 7), as the intense and prolonged training completed at baseline is known to reduce sIgA (Mackinnon and Hooper, 1994) and hence, sIgA may be naturally increased under the lighter training loads after the travel, making it difficult to identify lower sIgA as a risk factor for URTI at this time. Further, saliva samples could not be obtained at the usual time of day on day 1 due to logistical constraints of being on the airplane/in the terminal, meaning it is unknown if the mucosal responses were altered upon waking/immediately after the international flight. Nevertheless, three athletes (25%) developed symptoms of illness (URTI or chest infection), including one athlete for whom severe symptoms persisted for 5 days (including the day of competition). This illness frequency distribution is similar to what has been reported previously in those who slept an average of 5–6 h (29%) and 6–7 h (23%) across a 7-day period (Prather et al., 2015). The *likely* – *very likely* differences in sleep and fatigue identified in this manuscript all occurred in the first 2 days after arrival, before any symptoms of illness were reported. Therefore, the illnesses that were self-reported were not impacting on the major outcomes of this investigation. However, the illnesses experienced after this time would have affected the self-report measures, but evidently, not by enough to affect the outcomes of the study (i.e., there were no other *likely*, *very likely* or *most likely* changes

compared to baseline in the negative direction), and as such the participants that reported illness were not removed from analysis of the self-report data.

Fatigue, recovery and mental preparedness were *likely* improved compared to baseline on the morning of competition (Figure 6), which may be due to the tapering training program (Figure 7) and other individual race day preparation strategies that may have been used by the athletes (e.g., pre-race routines). It is not surprising that all of these measures, as well as physical preparedness, were *very likely* deteriorated in the days following the 5- to 8-h triathlon. It should be noted that sleep duration was also *very likely* lower on the night prior to competition (Figure 3A), perhaps a result of the required early wake up time on this day (3:30 AM), combined with feelings of anxiety (Juliff et al., 2015). Hence, the sleep hygiene practices discussed above may also be beneficial for athletes to implement on the night prior to competition. There is evidence in the current study that some of the athletes increased their nap duration in the days prior to competition (Figure 3B), perhaps in preparation for a lack of sleep on the night prior to the race.

It is important to identify the limitations of the current study. Firstly, the baseline measures were taken during normal training weeks, when some fatigue was present. Other studies have established baseline sleep and mucosal measurements during rest weeks (Coad et al., 2016), although this was not possible in the current design and with triathletes, who generally train year round. Saliva samples were analyzed with a portable point of care device, which had lower (but acceptable) reliability compared to laboratory techniques (Coad et al., 2015; Fisher et al., 2015). Further, saliva and self-report data were not obtained upon waking from the overnight flight due to logistical constraints, which potentially coincided with the time when immunity, stress and fatigue were challenged the most. Two different airlines were used by the athletes when flying from Sydney to Honolulu, which created a small variance in the stopover duration at Sydney airport, however, the flight duration was the same (and hence there were very similar routines on the flights themselves in terms of meals, light exposure and physical activity). It should also be noted that self-reported symptoms of illness do not necessarily reflect illness as diagnosed by a physician or detected by the presence of a pathogen (Gleeson, 2007). Finally, the travel fatigue and the hotter environment could not be isolated in the current study, but they represent two combined contextual factors that athletes may face when traveling to a hot environment for competition.

CONCLUSION

Long-haul northeast travel from a cool to a hot environment had *most likely* and *likely* negative influences on sleep and self-reported fatigue, respectively, but these alterations had returned to pre-departure baseline within 48 h after arrival. This travel appeared not to challenge immunity or physiological stress, when quantified using mucosal markers of sIgA and cortisol. Nevertheless, three athletes (25%) did self-report symptoms of illness 3–5 days after arrival. Endurance

athletes undertaking similar journeys may benefit from optimizing sleep hygiene, especially on the first 2 days after arrival, or until sleep duration and fatigue levels return to normal.

AUTHOR CONTRIBUTIONS

CS collected the data and wrote the manuscript. CS, HT and CE analyzed the data. All authors developed the research question and study design. All authors provided critical feedback and revisions to the manuscript drafts.

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FUNDING

The authors acknowledge Southern Cross University and the ASPETAR Qatar Orthopaedic and Sports Medicine Hospital for funding the research.

ACKNOWLEDGMENTS

The publication of this article was funded by the Qatar National Library.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Renal Hemodynamics During Sympathetic Activation Following Aerobic and Anaerobic Exercise

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OPEN ACCESS

Edited by:

Matthew J. Barnes,
Massey University, New Zealand

Reviewed by:

Daniel Gagnon,
Université de Montréal, Canada
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University of Tsukuba, Japan

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 27 October 2018

Accepted: 21 December 2018

Published: 10 January 2019

Citation:

Schlader ZJ, Chapman CL,
Benati JM, Gideon EA, Vargas NT,
Lema PC and Johnson BD (2019)
Renal Hemodynamics During
Sympathetic Activation Following
Aerobic and Anaerobic Exercise.
Front. Physiol. 9:1928.
doi: 10.3389/fphys.2018.01928

We tested the hypotheses that prior aerobic (Study 1) or anaerobic (Study 2) exercise attenuates the increase in renal vascular resistance (RVR) during sympathetic stimulation. Ten healthy young adults (5 females) participated in both Study 1 (aerobic exercise) and Study 2 (anaerobic exercise). In Study 1, subjects completed three minutes of face cooling pre- and post- 30 min of moderate intensity aerobic exercise ($68 \pm 1\%$ estimate maximal heart rate). In Study 2, subjects completed two minutes of the cold pressor test pre- and post- the completion of a 30 s maximal effort cycling test (Wingate Anaerobic Test). Both face cooling and the cold pressor test stimulate the sympathetic nervous system and elevate RVR. The primary dependent variable in both Studies was renal blood velocity, which was measured at baseline and every minute during sympathetic stimulation. Renal blood velocity was measured via the coronal approach at the distal segment of the right renal artery with pulsed wave Doppler ultrasound. RVR was calculated from the quotient of mean arterial pressure and renal blood velocity. In Study 1, renal blood velocity and RVR did not differ between pre- and post- aerobic exercise ($P \geq 0.24$). Face cooling decreased renal blood velocity ($P < 0.01$) and the magnitude of this decrease did not differ between pre- and post- aerobic exercise ($P = 0.52$). RVR increased with face cooling ($P < 0.01$) and the extent of these increases did not differ between pre- and post- aerobic exercise ($P = 0.74$). In Study 2, renal blood velocity was 2 ± 2 cm/s lower post- anaerobic exercise ($P = 0.02$), but RVR did not differ ($P = 0.08$). The cold pressor test decreased renal blood velocity ($P < 0.01$) and the magnitude of this decrease did not differ between pre- and post- anaerobic exercise ($P = 0.26$). RVR increased with the cold pressor test ($P < 0.01$) and the extent of these increases did not differ between pre- and post- anaerobic exercise ($P = 0.12$). These data indicate that 30 min of moderate intensity aerobic exercise or 30 s of maximal effort anaerobic exercise does not affect the capacity to increase RVR during sympathetic stimulation following exercise.

Keywords: renal vascular resistance, cold pressor test, face cooling, Doppler ultrasound, exercise recovery

INTRODUCTION

Blood pressure is often reduced for up to 60 min following an acute bout of dynamic exercise (Halliwill et al., 2013, 2014). In healthy untrained adults, this post-exercise hypotension is caused by reductions in total peripheral resistance (TPR) that are not fully offset by increases in cardiac output (Meade et al., 2018). Renal, splanchnic and cutaneous vascular resistances return to pre-exercise levels within ~20 min following exercise (Pricher et al., 2004; Wilkins et al., 2004). Thus, the lower TPR is due to persistent vasodilation in the muscle vasculature following exercise (Halliwill et al., 2013, 2014).

Orthostatic tolerance, defined as the ability to maintain blood pressure during orthostasis (Schlader et al., 2016b), is often impaired following dynamic exercise (Halliwill et al., 2014). This is caused by a relative inability to maintain stroke volume during orthostasis, which is likely due to inadequate increases in resistance in the muscle vasculature, thereby promoting pooling of blood in this vascular bed (Halliwill et al., 2013, 2014). That said, orthostasis also provokes sympathetically mediated increases in resistance in the splanchnic (Rowell et al., 1972; Jarvis et al., 2012) and renal (Bakris et al., 1986; Hirsch et al., 1989; Minson et al., 1999) vasculatures. These vasculatures each receive 20–30% of cardiac output and contribute significantly to blood pressure regulation during orthostasis (Rowell et al., 1972; Hirsch et al., 1989). For instance, pharmacological redistribution of blood flow away from the splanchnic vasculature by ~140 mL/min (or ~3% of cardiac output) improves orthostatic tolerance (Jarvis et al., 2012). Moreover, orthostatic stress reduces renal blood flow by a similar magnitude (~170 mL/min) (Hirsch et al., 1989). Thus, despite resistance in these visceral vascular beds returning to pre-exercise levels shortly after exercise (Pricher et al., 2004), it may be that prior exercise attenuates the ability to increase vascular resistance during sympathetic activation. If this were the case, the renal and/or splanchnic vasculatures could contribute to post-exercise orthostatic intolerance. However, the effect of prior exercise on the hemodynamic response to sympathetic stimulation in one of these visceral vasculatures is unknown.

With this background, the purpose of this study was to test the hypothesis that prior aerobic exercise attenuates the increase in renal vascular resistance (RVR) during sympathetic stimulation. The extent of post-exercise orthostatic intolerance is influenced by the intensity of the exercise (Mündel et al., 2015). Moreover, there is indirect evidence that high intensity (mostly anaerobic) exercise elicits a greater incidence of pre-syncope (i.e., the onset of syncopal signs and symptoms) during orthostasis compared to moderate intensity (primarily aerobic) exercise (Halliwill et al., 2014). Furthermore, reductions in renal blood flow can be maintained for up to 60 min following high intensity exercise (Suzuki et al., 1996), which is suggestive of sustained alterations in renal vascular control. Thus, we also conducted a separate study that tested the hypothesis that prior anaerobic exercise attenuates the increase in RVR during sympathetic stimulation.

MATERIALS AND METHODS

Ethics Statement

This study was approved by the Institutional Review Board at the University at Buffalo and conformed to the standards set by the Declaration of Helsinki, except for registration in a database. Before completing any study related activities, each subject was fully informed of the experimental procedures and possible risks before giving informed, written consent.

Subjects

Ten healthy young adults (five females) participated in both Study 1 (aerobic exercise) and Study 2 (anaerobic exercise). Only four subjects completed both Studies. Thus, these Studies were considered independent of one another. The subject characteristics were as follows. Study 1 – age: 23 ± 2 years, height: 169 ± 11 cm, weight: 67.1 ± 14.8 kg; Study 2 – age: 23 ± 3 years, height: 171 ± 11 cm, weight: 69.5 ± 14.5 kg. Subjects were physically active, non-smokers, not taking medications, and reported to be free from any known cardiovascular, metabolic, renal, or neurological diseases. Female subjects were not pregnant, which was confirmed via a urine pregnancy test, and self-reported to be normally menstruating. For both Studies, subjects visited the laboratory on two occasions. Visit one was a screening and familiarization visit, and visit two was the experimental trial. For visit two, subjects arrived at the laboratory having refrained from strenuous exercise, alcohol and caffeine for 12 h, and food for 2 h, and were instructed to be well hydrated. Females were tested throughout their menstrual cycle. This was deemed acceptable because of the repeated measures design and that all experimental testing was conducted on the same day, with shifts in menstrual cycle hormones between the pre- and post-exercise data collection periods likely being minimal.

Instrumentation and Measurements

Height and weight were measured with a stadiometer and scale (Sartorius Corp. Bohemia, NY, United States). Heart rate was continually measured via a 3-lead ECG (DA100C, Biopac Systems, Inc., Goleta, CA, United States) during experimental testing pre- and post-exercise. During exercise, heart rate was measured using a wireless heart rate monitor (Polar, Kempele, Finland). Beat-to-beat blood pressure was measured via the Penaz method (Finometer Pro, FMS, Amsterdam, Netherlands). These data were confirmed intermittently via auscultation of the brachial artery by electrophygmomanometry (Tango M2, SunTech Raleigh, NC, United States). In most instances no corrections were necessary, except in one subject in Study 2. In this subject, blood pressure data obtained via auscultation of the brachial artery were used. Stroke volume was estimated from the beat-to-beat blood pressure waveform using Modelflow (Wesseling et al., 1993) ($n = 9$ in Study 2, see above). The partial pressure of end-tidal carbon dioxide (PETCO₂) was measured via capnography (Nonin Medical, Inc., Plymouth, MN, United States). Due to technical issues, PETCO₂ was measured in only eight subjects in Study 1. Renal blood velocity was assessed in the distal segment of the right renal artery during

the same phase of the respiratory cycle via Doppler ultrasound (GE Vivid 7 Dimension, Chicago, IL, United States) (Momen et al., 2004; Wilson et al., 2007; Drew et al., 2013; Patel et al., 2013). The coronal approach was utilized using a phased-array transducer with a 2.5–3.5 MHz pulsed frequency with subjects in the left lateral recumbent position. The focal zone was set to the artery's depth, and the transducer was held in the same location for all measurements (Momen et al., 2004; Wilson et al., 2007; Drew et al., 2013; Patel et al., 2013). All measurements and analyses were obtained by the same sonographer (JMB). The transducer location was marked with indelible ink during pre-exercise data collection, which ensured that during post-exercise data collection renal blood velocity measurements were made in the same anatomical position. The insonation angle was always $<60^\circ$ and was the same pre- and post-exercise. Using this approach, the within-subject test-retest coefficient of variation for mean renal blood velocity for the sonographer was $3.9 \pm 2.4\%$. Mean renal blood velocity was indexed by the time-averaged maximum velocity from the envelope of the velocity waveform. At each measurement period, mean renal blood velocity, peak systolic blood velocity and end diastolic blood velocity were measured and averaged over 2–4 cardiac cycles (Momen et al., 2003). Given the depth of the renal artery, it is not possible to accurately measure artery diameter. However, the diameter of the renal artery does not change during pharmacologically induced renal vasoconstriction (Marraccini et al., 1996). Thus, changes in renal blood velocity were interpreted to reflect changes in renal blood flow, as has been done previously (Wilson et al., 2007; Patel et al., 2013; Drew et al., 2017). Renal blood velocity was obtained in all subjects, except one subject in Study 2, where a post-exercise Doppler ultrasound image was not able to be obtained. Therefore, in Study 2 renal blood velocity data are presented as $n = 9$.

Experimental Protocol

Study 1 – Effect of Prior Aerobic Exercise

Following instrumentation, subjects assumed the supine position. Following 10 min of quiet rest, baseline measurements were taken over the next 5 min. At the end of this period, face cooling commenced. Face cooling was achieved by placing a flexible bag of ice water (0°C) directly on the forehead, eyes, and cheeks for 3 min. The volume of the ice water was 2.5 L. Based on previous work from our laboratory (Schlader et al., 2016a, 2018; Johnson et al., 2018), 3 min of face cooling was deemed sufficient to elicit sympathetically mediated increases in vascular resistance and blood pressure, which occurs subsequent to stimulation of trigeminal afferents (Schuitema and Holm, 1988). Moreover, a similar face cooling procedure has been shown to elicit increases in RVR as measured using Doppler ultrasound (Patel et al., 2013). Renal blood velocity data were collected at baseline, each minute of face cooling, and 60 s following face cooling (recovery). These face cooling procedures were completed prior to and following 30 min of aerobic exercise on a treadmill (Quinton Instruments, Seattle, WA, United States) at a speed and grade that elicited a heart rate of 134 ± 3 bpm ($68 \pm 1\%$ estimate maximal heart rate). All data were collected in a temperature-controlled laboratory ($24 \pm 2^\circ\text{C}$, $20 \pm 5\%$ relative humidity). The time delay between

the end of exercise and the start of the 10 min post- aerobic exercise baseline period was <5 min. This delay was necessary to accommodate re-instrumentation following exercise.

Study 2 – Effect of Prior Anaerobic Exercise

Following instrumentation, subjects assumed the supine position. Following 10 min of quiet rest, baseline measurements were taken over the next 5 min. At the end of this period, the cold pressor test commenced. The cold pressor test was achieved by submerging the subject's right hand in agitated ice water (0°C) up to the wrist for 2 min. This two min cold pressor test elicits sympathetically mediated increases in vascular resistance and blood pressure (Victor et al., 1987; Cui et al., 2010), which occurs subsequent to stimulation of nociceptors (Kregel et al., 1992). Moreover, the cold pressor test has been shown to elicit increases in RVR as measured using Doppler ultrasound (Patel et al., 2013). Renal blood velocity data were collected at baseline, each minute of the cold pressor test, and 60 s following the cold pressor test (recovery). These cold pressor test procedures were completed prior to and following a 30 s Wingate Anaerobic test on a cycle ergometer (Monark 894E, Sweden). This test utilizes mostly anaerobic fuel sources (Beneke et al., 2002). Following a 5 min self-selected warmup, the Wingate Anaerobic Test consisted of a maximal effort against a resistance of 7.5% total body mass following a 3 s unweighted acceleration phase. All data were collected in a temperature-controlled laboratory ($23 \pm 2^\circ\text{C}$, $21 \pm 13\%$ relative humidity). The time delay between the end of exercise and the start of the 10 min post- anaerobic exercise baseline period was <5 min. This delay was necessary to accommodate re-instrumentation following exercise.

It should be noted that Study 1 and Study 2 were initially designed to test different hypotheses than those of the present studies. As a result, the two Studies used different sympathoexcitatory stimuli and a mostly different cohort of subjects. Despite these differences, however, the stimuli used in Study 1 (face cooling) and Study 2 (cold pressor test) elicit sympathetically mediated increases in RVR (Patel et al., 2013) and the time course of testing both pre- and post-exercise was identical between the two Studies. Thus, the data obtained from these Studies were deemed appropriate to test our hypotheses. That said, given the differences in study design, no direct comparisons were made between the two Studies.

Data and Statistical Analyses

All non-ultrasound data were sampled continuously at 1000 Hz via a data acquisition system (Biopac MP150, Goleta, CA, United States) in both Studies. These data were binned as a 60 s average at the end of the baseline data collection period, and during and following face cooling (Study 1) or the cold pressor test (Study 2) the non-ultrasound data were binned as a 30 s average every 60 s. Cardiac output was calculated as the product of stroke volume and heart rate, while TPR was calculated as the quotient of mean arterial pressure and cardiac output. RVR was estimated as the quotient of mean arterial pressure and mean renal blood velocity.

To isolate the effect of prior exercise on the responsiveness to face cooling (Study 1) or the cold pressor test (Study 2),

data were analyzed as the absolute change from baseline. These data were analyzed using two-way repeated measures ANOVA (time \times exercise). When an ANOVA revealed a significant main effect or interaction, *post hoc* Sidak test pairwise comparisons were made. Absolute data at baseline pre- and post-exercise were analyzed using paired *t*-tests. All data were analyzed using Prism software (Version 7, GraphPad Software Inc., La Jolla, CA, United States). *A priori* statistical significance was set at $P \leq 0.05$ and actual *P*-values are reported where possible. Data are reported as mean \pm SD.

RESULTS

Study 1 – Effect of Prior Aerobic Exercise

Heart rate was 9 ± 9 bpm higher following exercise ($P < 0.01$), but stroke volume did not differ between pre- and post-exercise ($P = 0.28$, **Table 1**). As a result, cardiac output was 1.3 ± 1.9 L/min higher following exercise ($P = 0.03$, **Table 1**). Mean arterial pressure, systolic blood pressure, and diastolic blood pressure were not different between pre- and post-exercise ($P \geq 0.08$, **Table 1**). However, TPR was 3.1 ± 4.5 mmHg/L/min lower following exercise ($P = 0.03$, **Table 1**). PETCO₂ did not differ between pre- and post-exercise ($P = 0.26$, **Table 1**). Mean renal blood velocity, peak systolic renal blood velocity, end diastolic renal blood velocity, and RVR did not differ between pre- and post-exercise ($P \geq 0.24$, **Table 1**).

Face cooling decreased heart rate both pre- and post-exercise ($P < 0.01$) and the magnitude of this decrease was greater post-exercise at 1 and 3 min of face cooling ($P \leq 0.04$, **Figure 1A**). Stroke volume was not affected by face cooling ($P = 0.14$) and there was no effect of prior exercise ($P = 0.55$, **Figure 1B**). Cardiac output decreased during face cooling ($P < 0.01$), but the magnitude of this reduction did not differ between pre- and post-exercise ($P = 0.36$, **Figure 1C**). TPR increased with

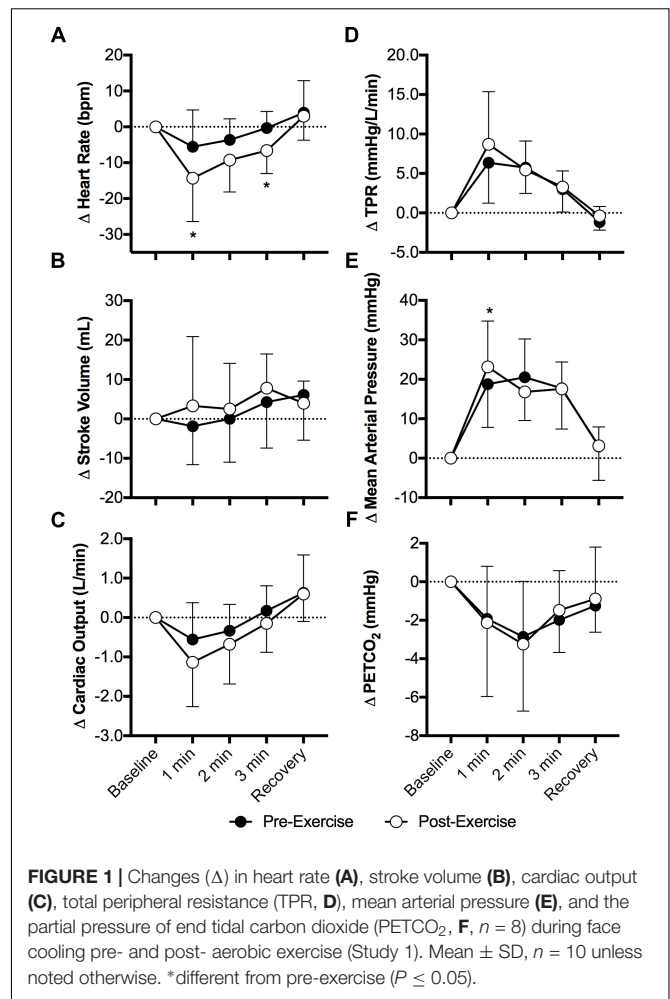


FIGURE 1 | Changes (Δ) in heart rate (**A**), stroke volume (**B**), cardiac output (**C**), total peripheral resistance (TPR, **D**), mean arterial pressure (**E**), and the partial pressure of end tidal carbon dioxide (PETCO₂, **F**, $n = 8$) during face cooling pre- and post-aerobic exercise (Study 1). Mean \pm SD, $n = 10$ unless noted otherwise. *different from pre-exercise ($P \leq 0.05$).

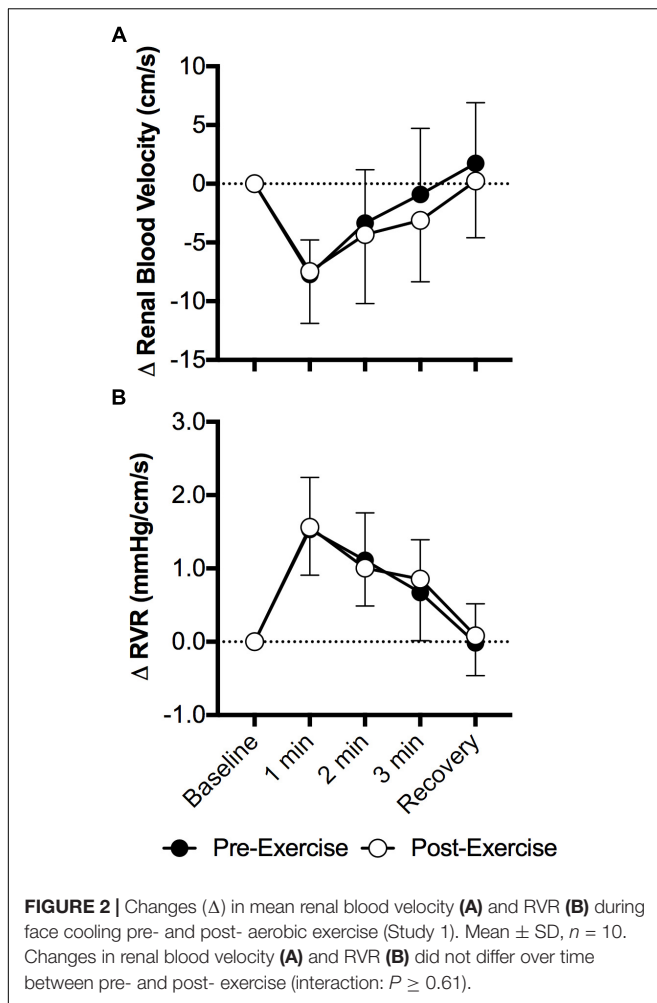
face cooling ($P < 0.01$) and the magnitude of the increase did not differ between pre- and post-exercise ($P = 0.47$, **Figure 1D**). Mean arterial pressure increased with face cooling both pre- and post-exercise ($P < 0.01$) and the magnitude of this increase was 4 ± 8 mmHg greater at 1 min post-exercise ($P = 0.05$, **Figure 1E**). Changes in systolic and diastolic blood pressure mirrored that of mean arterial pressure, with systolic and diastolic blood pressures increasing with face cooling ($P < 0.01$) and the magnitude of these increases being greater at 1 min post-exercise ($P \leq 0.05$). PETCO₂ decreased during face cooling ($P < 0.01$) and the extent of these reductions did not differ between pre- and post-exercise ($P = 0.93$, **Figure 1F**).

Face cooling decreased mean renal blood velocity ($P < 0.01$) and the magnitude of this decrease did not differ between pre- and post-exercise ($P = 0.52$, **Figure 2A**). Changes in peak systolic and end diastolic renal blood velocity mirrored that of mean renal blood velocity, with peak systolic and end diastolic renal blood velocity decreasing with face cooling ($P < 0.01$) and the magnitude of these decreases did not differ between pre- and post-exercise ($P \geq 0.27$). RVR increased with face cooling ($P < 0.01$) and the extent of these increases did not differ between pre- and post-exercise ($P = 0.74$, **Figure 2B**).

TABLE 1 | Hemodynamics at baseline pre- and post-aerobic exercise.

	Pre-exercise	Post-exercise	<i>P</i> -value
Heart rate (bpm)	60 \pm 8	68 \pm 7	< 0.01
Stroke volume (mL)	83 \pm 16	87 \pm 23	0.27
Cardiac output (L/min)	4.9 \pm 1.0	6.2 \pm 2.2	0.03
Mean arterial pressure (mmHg)	85 \pm 12	83 \pm 9	0.28
Systolic blood pressure (mmHg)	118 \pm 17	111 \pm 13	0.08
Diastolic blood pressure (mmHg)	65 \pm 8	64 \pm 8	0.50
TPR (mmHg/L/min)	17.8 \pm 3.5	14.7 \pm 4.4	0.03
PETCO ₂ (mmHg) ($n = 8$)	41 \pm 3	40 \pm 1	0.26
Mean renal blood velocity (cm/s)	34 \pm 5	34 \pm 9	0.31
Peak systolic renal blood velocity (cm/s)	64 \pm 10	62 \pm 15	0.24
End diastolic renal blood velocity (cm/s)	19 \pm 3	20 \pm 6	0.26
RVR (mmHg/cm/s)	2.6 \pm 0.5	2.6 \pm 0.9	0.47

TPR: total peripheral resistance, PETCO₂: partial pressure of end tidal carbon dioxide, RVR: renal vascular resistance. Data are presented as Mean \pm SD, $n = 10$ unless noted otherwise.



Study 2 – Effect of Prior Anaerobic Exercise

Heart rate was 26 ± 12 bpm higher following exercise ($P < 0.01$), but stroke volume did not differ between pre- and post-exercise ($P = 0.43$, Table 2). As a result, cardiac output was 2.3 ± 2.3 L/min higher following exercise ($P < 0.01$, Table 2). Mean arterial pressure, systolic blood pressure, and diastolic blood pressure were not different between pre- and post-exercise ($P \geq 0.29$, Table 2). However, TPR was 5.2 ± 10.2 mmHg/L/min lower following exercise ($P = 0.05$, Table 2). PETCO₂ was 6 ± 4 mmHg lower following exercise ($P < 0.01$, Table 2). Mean renal blood velocity was 2 ± 2 cm/s lower post-exercise ($P = 0.02$, Table 2). Peak systolic renal blood velocity was higher post-exercise (by 9 ± 2 cm/s, $P < 0.01$), but end diastolic renal blood velocity was 3 ± 4 cm/s lower post-exercise ($P < 0.01$, Table 2). RVR was not different between pre- and post-exercise ($P = 0.08$, Table 2).

Heart rate increased during the cold pressor test both pre- and post-exercise ($P < 0.01$) and the magnitude of this increase was attenuated post-exercise at 1 min of the cold pressor test ($P = 0.04$, Figure 3A). Stroke volume (Figure 3B) and cardiac output (Figure 3C) were not affected by the cold pressor test

TABLE 2 | Hemodynamics at baseline pre- and post- anaerobic exercise.

	Pre-exercise	Post-exercise	P-value
Heart rate (bpm)	61 \pm 5	87 \pm 13	< 0.01
Stroke volume (mL) ($n = 9$)	85 \pm 22	86 \pm 16	0.43
Cardiac output (L/min) ($n = 9$)	5.2 \pm 1.6	7.8 \pm 2.5	< 0.01
Mean arterial pressure (mmHg)	82 \pm 13	82 \pm 13	0.50
Systolic blood pressure (mmHg)	111 \pm 13	114 \pm 17	0.29
Diastolic blood pressure (mmHg)	63 \pm 15	64 \pm 13	0.41
TPR (mmHg/L/min) ($n = 9$)	17.7 \pm 6.7	12.1 \pm 6.0	0.05
PETCO ₂ (mmHg)	38 \pm 4	33 \pm 4	< 0.01
Mean renal blood velocity (cm/s) ($n = 9$)	34 \pm 7	32 \pm 7	0.02
Peak systolic renal blood velocity (cm/s) ($n = 9$)	66 \pm 13	77 \pm 17	< 0.01
End diastolic renal blood velocity (cm/s) ($n = 9$)	19 \pm 5	15 \pm 3	< 0.01
RVR (mmHg/cm/s) ($n = 9$)	2.4 \pm 0.5	2.6 \pm 0.7	0.08

TPR: total peripheral resistance, PETCO₂: partial pressure of end tidal carbon dioxide, RVR: renal vascular resistance. Data are presented as Mean \pm SD, $n = 10$ unless noted otherwise.

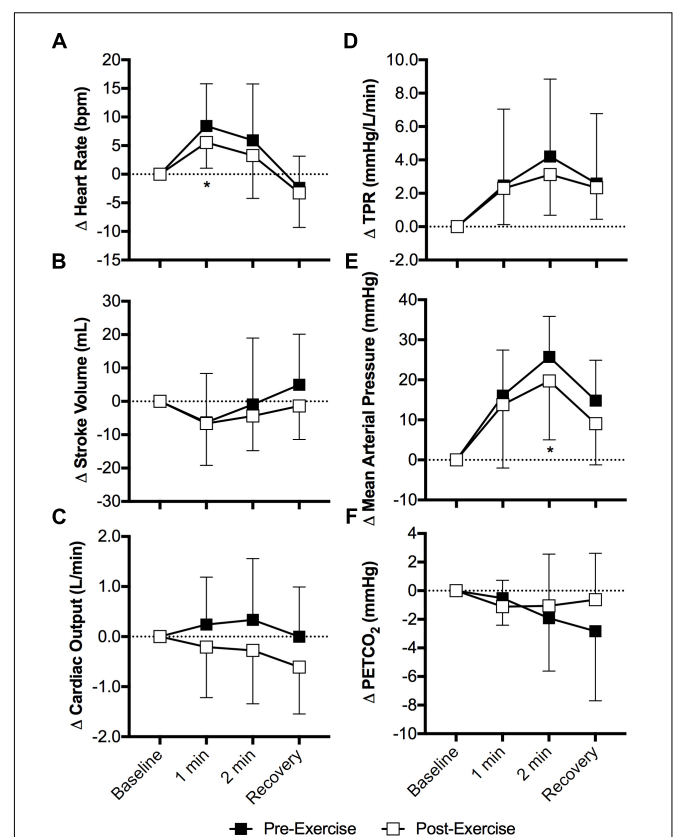


FIGURE 3 | Changes (Δ) in heart rate (A), stroke volume (B, $n = 9$), cardiac output (C, $n = 9$), TPR (D, $n = 9$), mean arterial pressure (E), and the PETCO₂ (F) during the cold pressor test pre- and post- anaerobic exercise (Study 2). Mean \pm SD, $n = 10$ unless noted otherwise. *different from pre-exercise ($P \leq 0.05$).

($P \geq 0.10$) and there was no effect of prior exercise ($P \geq 0.14$). TPR increased during the cold pressor test ($P < 0.01$) and the magnitude of the increase did not differ between pre- and

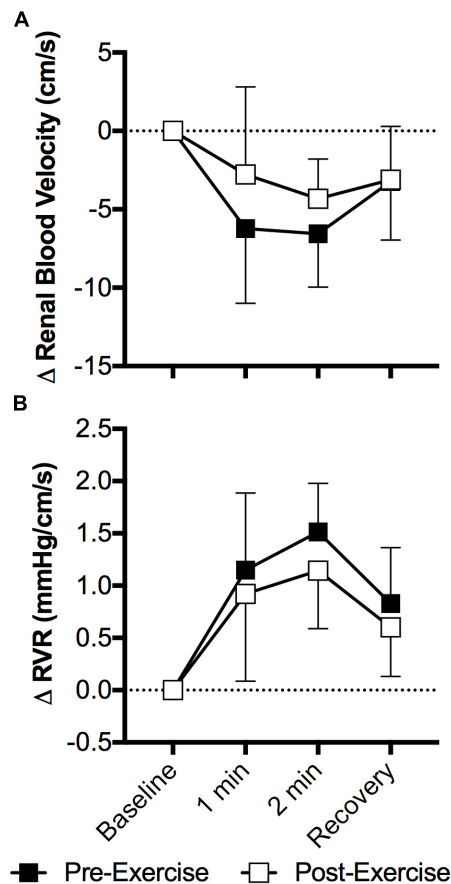


FIGURE 4 | Changes (Δ) in mean renal blood velocity (A) and RVR (B) during the cold pressor test pre- and post- anaerobic exercise (Study 2). Mean \pm SD, $n = 9$. Changes in renal blood velocity (A) and RVR (B) did not differ over time between pre- and post- exercise (interaction: $P \geq 0.39$).

post-exercise ($P = 0.68$, **Figure 3D**). Mean arterial pressure increased during the cold pressor test both pre- and post-exercise ($P < 0.01$) and the magnitude of this increase was 6 ± 10 mmHg lower at 2 min post-exercise ($P = 0.05$, **Figure 3E**). Both systolic and diastolic blood pressure increased during the cold pressor test ($P < 0.01$) and the magnitude of these changes did not differ between pre- and post-exercise ($P \geq 0.20$). PETCO₂ did not change during the cold pressor test ($P = 0.20$) and there was no effect of prior exercise ($P = 0.41$, **Figure 3F**).

The cold pressor test decreased mean renal blood velocity ($P < 0.01$) and the magnitude of this decrease did not differ between pre- and post-exercise ($P = 0.26$, **Figure 4A**). Changes in peak systolic and end diastolic renal blood velocity mirrored that of mean renal blood velocity, with peak systolic and end diastolic renal blood velocity decreasing during the cold pressor test ($P < 0.01$) and the magnitude of these decreases did not differ between pre- and post-exercise ($P \geq 0.10$). RVR increased with face cooling ($P < 0.01$) and the extent of this increase did not differ between pre- and post-exercise ($P = 0.12$, **Figure 4B**).

DISCUSSION

Contrary to our hypotheses, our data demonstrate that prior aerobic or anaerobic exercise does not influence the extent of increases in RVR during sympathoexcitatory stimuli. Specifically, we have identified that 30 min of moderate intensity aerobic exercise that invoked $\sim 70\%$ of age-predicted maximum heart rate did not affect the post-exercise renal vascular response to face cooling, a stimulus known to elicit sympathetically mediated increases in vascular resistance (Fisher et al., 2015; Schlader et al., 2016a, 2018; Johnson et al., 2018), including in the renal vasculature (Patel et al., 2013) (**Figure 2**). In a second study we found that the completion of a 30 s Wingate Anaerobic Test also did not affect the post-exercise renal vascular response to the cold pressor test, a stimulus known to increase vascular resistance secondary to sympathetic activation (Victor et al., 1987; Cui et al., 2010), including the renal vascular bed (Patel et al., 2013) (**Figure 4**). Collectively, these data indicate that prior moderate intensity aerobic exercise or brief maximal effort anaerobic exercise, to the extent studied in the present investigations, does not affect sympathetically mediated renal vasomotor responsiveness.

Prior Aerobic Exercise Does Not Affect the Renal Vascular Response to Face Cooling (Study 1)

Orthostatic tolerance is often impaired following moderate intensity aerobic exercise (Halliwill et al., 2014). These observations are likely explained by a relative inability to increase vascular resistance during orthostasis (Lacewell et al., 2014). This reduces venous return, thereby compromising stroke volume and the ability to adequately maintain cardiac output (Krediet et al., 2004). The attenuated increase in vascular resistance is likely due to changes in the muscle vasculature, such that for a given sympathetic stimulus the increase in muscle vascular resistance is blunted (Halliwill et al., 1996a). However, to the best of our knowledge, a role for alterations in renal vascular control has not been discounted, despite that this vasculature contributes to blood pressure regulation during orthostasis (Bakris et al., 1986; Hirsch et al., 1989; Minson et al., 1999). Data in rats indicate that the relationship between blood pressure and renal sympathetic nerve activity is shifted downward following aerobic exercise (Miki et al., 2003). In humans, aerobic exercise induced elevations in RVR return to pre-exercise levels shortly after exercise (Pricher et al., 2004). This is supported by our data such that renal blood velocity and our estimate of RVR did not differ at baseline pre- versus post- aerobic exercise (**Table 1**). Based on the data presented herein (**Table 1**) and previous findings in humans (Pricher et al., 2004), together with the aforementioned study in rats (Miki et al., 2003), we speculate that a given level of renal sympathetic activity likely results in a higher RVR following aerobic exercise compared to before exercise. The reason for this is unknown, but may be related to the balance of circulating vasoactive factors, which likely more readily favor vasoconstriction in the renal vasculature. For instance, vasopressin and aldosterone both increase with

moderate intensity aerobic exercise (Convertino et al., 1981; Freund et al., 1991). These vasoactive hormones could raise vascular resistance independent of sympathetic activation, particularly in the renal vasculature (Schmid et al., 1974; Schmidt et al., 2006). Notably, this contention is speculative. Therefore, more research is required to understand how prior exercise may affect the neuro-hormonal balance underlying the control of renal blood flow.

Our data also demonstrate that the magnitude of increases in RVR during face cooling are not affected by prior aerobic exercise (Figure 2). To the best of our knowledge, these findings are novel. Our data can likely be explained by the findings that the gain of the relation between renal sympathetic nerve activity and blood pressure are not affected by prior aerobic exercise (Miki et al., 2003). This suggests that the shift in the relationship between blood pressure and renal sympathetic nerve activity observed in rats (Miki et al., 2003), does not affect the capacity of the renal vasculature to respond to a sympathoexcitatory stimulus. Notably, the maintenance of blood pressure during orthostasis is dependent upon increasing sympathetic nerve activity (Cooke et al., 2009). Therefore, our data might suggest that an inability to vasoconstrict the renal vasculature following aerobic exercise is unlikely to contribute to post-exercise orthostatic intolerance.

It is important to note that our aerobic exercise paradigm did not elicit the expected post-exercise hypotension (Table 1). Thus, in the present study the expected reductions in TPR were offset by increases in cardiac output, which was almost entirely due to elevations in heart rate (Table 1). The reason for this observation is likely due to the duration and intensity of the aerobic exercise. In the present study subjects exercised at ~70% of age-predicted maximum heart rate for 30 min, which is estimated to elicit 50–60% maximal oxygen uptake (Lounana et al., 2007). This exercise intensity and duration is slightly lower and shorter than that commonly used to study post-exercise hypotension (e.g., Halliwill et al., 1996a,b; Pricher et al., 2004; Wilkins et al., 2004; McCord et al., 2006). Furthermore, the magnitude of hypotension following aerobic exercise is dependent on the total work completed, and is not necessarily dependent on the exercise intensity or duration *per se* (Jones et al., 2007). Thus, we speculate that if our aerobic exercise was higher in intensity or longer in duration, post-exercise hypotension would have been observed. Importantly, however, we do not believe the lack of post-exercise hypotension following aerobic exercise invalidates our finding that the increase in RVR during sympathetic activation was not affected by prior aerobic exercise. For instance, impairments in blood pressure regulation during orthostasis have been observed following high intensity exercise, despite that hypotension was not observed before orthostasis (Lacewell et al., 2014; Sieck et al., 2016). It is possible that this prior observation is a function of the exercise intensity (discussed below), but it may also indicate that a hemodynamic or sympathetic stimulus (e.g., orthostasis) is required before alterations in physiological function can be observed (Halliwill et al., 2014).

Although it was not a primary outcome of our study, the greater fall in heart rate with face cooling following aerobic exercise is interesting (Figure 1). Face cooling stimulates cold afferents downstream of the trigeminal nerve,

which simultaneously stimulates both the sympathetic and parasympathetic nervous systems (Schlader et al., 2016a). As observed in the present study (Figure 1), these reductions in heart rate do not significantly compromise cardiac output (Schlader et al., 2016a). Thus, blood pressure rises secondary to sympathetically mediated increases in vascular resistance (i.e., TPR) (Fisher et al., 2015; Schlader et al., 2016a) (Figure 1). The increase in heart rate during exercise is mediated by both the withdrawal and activation cardiac parasympathetic and sympathetic activity, respectively (White and Raven, 2014). The opposite occurs following aerobic exercise, as heart rate is restored toward pre-exercise levels (White and Raven, 2014). The findings presented herein suggest that the sensitivity to a parasympathetic stimulus is enhanced following moderate intensity aerobic exercise. We speculate that this is due to the greater prevailing parasympathetic drive during recovery from aerobic exercise and/or a greater end organ (cardiac) responsiveness, the latter of which could be related to the higher heart rates following aerobic exercise. Importantly, our findings are suggestive of the potential utility of face cooling as a tool to probe parasympathetic nervous system function (or dysfunction) (Johnson et al., 2017, 2018).

Prior Anaerobic Exercise Does Not Affect the Renal Vascular Response to the Cold Pressor Test (Study 2)

The extent of post-exercise orthostatic intolerance is influenced by the intensity of the exercise (Mündel et al., 2015). To our knowledge, however, a study comparing prior aerobic and anaerobic exercise on post-exercise orthostatic tolerance has not been reported. That said, it is often speculated that high intensity (anaerobic) exercise elicits a greater incidence of pre-syncope (~73% of the observations) during orthostasis compared to moderate intensity (aerobic) exercise (~42% of the observations) (Halliwill et al., 2014). The mechanisms underlying such potential differences in orthostatic tolerance are likely the same between aerobic and anaerobic exercise, but they probably differ in magnitude (Halliwill et al., 2014). For instance, the magnitude of decreases in muscle vascular resistance post-exercise are likely greater and the extent by which muscle vascular resistance is increased during orthostasis is likely attenuated following anaerobic exercise compared to following aerobic exercise. That said, a potential contribution for alterations in cerebral blood flow regulation, occurring subsequent to hypocapnia (Lacewell et al., 2014; Sieck et al., 2016) and/or changes in autoregulation (Ogoh et al., 2007), is also possible. Nevertheless, a potential contribution for the renal vasculature in post-anaerobic exercise orthostatic intolerance has not been formally considered. As described above, a sympathetically mediated increase in RVR is an important contributor to blood pressure regulation during orthostasis (Bakris et al., 1986; Hirsch et al., 1989; Minson et al., 1999). Increases in RVR during exercise are intensity dependent (Grimby, 1965; Castenfors, 1977), and the resulting reductions in renal blood flow can be maintained for up to 60 min following high intensity exercise (Suzuki et al., 1996). This is supported by our data such that renal blood velocity was lower and estimated

RVR was higher ($P = 0.08$) following anaerobic exercise (Table 2). These sustained reductions in renal perfusion observed following anaerobic exercise differ from what happens following moderate intensity exercise (Table 1) (Pricher et al., 2004). The reasons for these differences can likely be explained by the heightened sympathetic activation (Kenney and Zappe, 1994; Suzuki et al., 1996) and/or greater concentrations of circulating vasoactive hormones (e.g., vasopressin, aldosterone, etc.) (Convertino et al., 1981; Freund et al., 1991) following exercise, all of which are known to increase with increased exercise intensity. The renal circulation is also sensitive to changes in arterial carbon dioxide, such that hypocapnia reduces renal sympathetic nerve activity (Shirahata et al., 1985) and decreases RVR (Norman et al., 1970; Sharkey et al., 1998). Thus, we speculate that the moderate hypocapnia observed following anaerobic exercise in our study (Table 2) likely helped to maintain renal perfusion, such that if hypocapnia were not present the reductions in renal blood velocity (and increases in RVR) would have been greater. However, a role for changes in arterial carbon dioxide on renal vascular control following exercise remains to be fully elucidated.

Our data also demonstrate that the capacity to increase RVR during the cold pressor test is not affected by prior anaerobic exercise (Figure 4). As described above, it is likely that these findings can be explained by data demonstrating that the gain of the relation between renal sympathetic nerve activity and blood pressure are not affected by prior aerobic exercise (Miki et al., 2003). Thus, the data presented herein further this concept to anaerobic exercise such that, despite the relative vasoconstricted state, the capacity to increase vascular resistance in the renal vasculature during sympathetic stimulation was unaffected by prior anaerobic exercise.

Methodological Considerations

There are a few methodological considerations that warrant discussion. First, we did not directly measure renal blood flow. Rather, we measured renal blood velocity using Doppler ultrasound. This enabled quantification of dynamic changes in an index of renal blood flow. This was deemed ideal to discern the effect of prior exercise on the renal vascular response to acute sympathetic stimulation, which would have been virtually impossible due to the relatively short data collection periods if we had used para-aminohippuric acid clearance, a traditional method for estimating renal blood flow in humans (Beierwaltes et al., 2013). That said, there are limitations associated with using Doppler ultrasound. For instance, renal blood flow is a function of artery diameter and blood velocity. Given the depth of the artery, it is not possible to accurately measure renal artery diameter using ultrasound. The diameter of the renal artery does not change during pharmacologically induced renal vasoconstriction (Marraccini et al., 1996). Thus, in the present study changes in renal blood velocity were interpreted to reflect changes in renal blood flow, as has been done previously (Wilson et al., 2007; Patel et al., 2013; Drew et al., 2017). However, it is acknowledged that we did not measure volumetric renal blood flow. Moreover, due to potential differences in the insonation location and/or angle, it is possible that the test-retest reliability of the Doppler ultrasound measurement of renal blood velocity

is poor. To overcome this limitation, controls were put in place to ensure our insonation location and angle were the same pre- and post- exercise. Furthermore, our data during face cooling and the cold pressor test were primarily analyzed as the absolute change from baseline. Nevertheless, conclusions associated with baseline measurements (e.g., Tables 1,2) should be made with caution. However, it is notable that the 6% reduction in renal blood velocity observed following anaerobic exercise (Table 2) is outside of the variation in the measurement of our sonographer (~4%). Second, the data presented in Studies 1 and 2 were obtained from two independent studies that used different sympathoexcitatory stimuli and a different cohort of subjects. Given these differences in study design, direct comparisons between the two Studies were not made. Therefore, the direct comparative effects of aerobic versus anaerobic exercise on the renal vascular response to sympathetic stimulation remains unknown. Third, we did not directly measure any indices of sympathetic activation (e.g., muscle sympathetic nerve activity) and it is not possible to measure renal sympathetic nerve activity in humans. Therefore, it is unknown if the magnitude of the whole-body and/or renal sympathetic response invoked by face cooling (Study 1) and the cold pressor test (Study 2) was the same pre- versus post-exercise. Fourth, exercise induced body fluid losses and/or elevations in body temperature may play a role in post-exercise hypotension and orthostatic intolerance (Halliwill et al., 2013, 2014; Meade et al., 2018). However, we did not measure any aspects of body fluid status (e.g., changes in body weight or plasma volume) or body temperature (e.g., core temperature). Thus, a potential contribution of these factors to the renal vascular response following exercise remains unknown. Fifth, we tested both males and females and we did not control for menstrual cycle phase in our female subjects. Notably, we are underpowered to conduct a formal analysis between males and females. There is some evidence that the incidence of post-exercise orthostatic intolerance is lower in females (Halliwill et al., 2014). Thus, it is possible that the renal vascular response to sympathetic stimulation following aerobic or anaerobic exercise may differ between males and females, and across the menstrual cycle. Finally, we tested the renal vascular response to hypertension-invoking sympathetic stimuli in the supine position. This enabled the measurement of dynamic changes in RVR using Doppler ultrasound. However, whether our findings would differ if they were obtained during orthostasis or with unloading of the baroreceptors, as occurs with lower body negative pressure, is unknown.

Perspectives

Our studies provide unique insights into the recovery of the cardiovascular and renal systems after aerobic or anaerobic exercise. This is important because the post-exercise recovery period presents a key window of opportunity that may be used to promote training adaptations (Luttrell and Halliwill, 2015). This may be particularly important as it relates to plasma volume, whereby post-exercise hypotension appears to be a key determinant of training invoked plasma volume expansion (Hayes et al., 2000). Moreover, our studies also have the potential to inform the development of countermeasures to protect against

syncope following exercise. Collectively, our data indicate that an inability to increase resistance in the renal vasculature following aerobic or anaerobic exercise is unlikely to contribute to post-exercise orthostatic intolerance. Thus, interventions aimed toward augmenting RVR post-exercise are unlikely to be effective at alleviating the incidence of orthostatic intolerance following aerobic or anaerobic exercise. Rather, such interventions should selectively target the muscle vasculature (McCord et al., 2008) or venous return more generally, as can be augmented by respiratory impedance (Lacewell et al., 2014).

CONCLUSION

The present study demonstrates that 30 min of moderate intensity aerobic exercise or 30 s of maximal effort anaerobic

exercise does not affect the capacity to increase RVR during sympathetic stimulation following exercise.

AUTHOR CONTRIBUTIONS

ZS, CC, NV, and BJ conceptualized the studies. CC, JB, EG, NV, and PL collected the data. ZS, CC, JB, EG, and PL analyzed the data. ZS, CC, NV, and BJ contributed to data interpretation. ZS drafted the manuscript. All authors approved the finalized manuscript.

ACKNOWLEDGMENTS

We would like to thank the subjects for participating in our study.

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Conflict of Interest Statement: PL is a consultant for Mindray North American Ultrasound.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Monitoring Training Loads and Perceived Stress in Young Elite University Athletes

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OPEN ACCESS

Edited by:

Matthew J. Barnes,
Massey University, New Zealand

Reviewed by:

Alexandre Moreira,
University of São Paulo, Brazil
Carl Foster,
University of Wisconsin–La Crosse,
United States

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 09 October 2018

Accepted: 11 January 2019

Published: 29 January 2019

Citation:

Hamlin MJ, Wilkes D, Elliot CA,
Lizamore CA and Kathiravel Y (2019)
Monitoring Training Loads
and Perceived Stress in Young Elite
University Athletes.
Front. Physiol. 10:34.
doi: 10.3389/fphys.2019.00034

With increased professionalism in sport there has been a greater interest in the scientific approach to training and recovery of athletes. Applying appropriate training loads along with adequate recovery, is essential in gaining maximal adaptation in athletes, while minimizing harm such as overreaching, overtraining, injury and illness. Although appropriate physical stress is essential, stress for many athletes may come from areas other than training. Stress may arise from social or environmental pressure, and for many athletes who combine elite athletic training with university study, academic workloads create significant stress which adds to the constant pressure to perform athletically. This research aimed to determine if subjective stressors were associated with counterproductive training adaptations in university athletes. Moreover, it aimed to elucidate if, and when, such stressors are most harmful (i.e., certain times of the academic year or sports training season). We monitored subjective (mood state, energy levels, academic stress, sleep quality/quantity, muscle soreness, training load) and objective (injury and illness) markers in 182 young (18–22 years) elite athletes over a 4-year period using a commercially available software package. Athletes combined full-time university study with elite sport and training obligations. Results suggest athletes were relatively un-stressed with high levels of energy at the beginning of each university semester, however, energy levels deteriorated along with sleep parameters toward the examination periods of the year. A logistical regression indicated decreased levels of perceived mood (0.89, 0.85–0.94, Odds Ratio and 95% confidence limits), sleep duration (0.94, 0.91–0.97) and increased academic stress (0.91, 0.88–0.94) and energy levels (1.07, 1.01–1.14) were able to predict injury in these athletes. Examination periods coincided with the highest stress levels and increased likelihood of illness. Additionally, a sudden and high increase in training workload during the preseason was associated with an elevated incidence of injury and illness ($r = 0.63$). In conclusion, young elite athletes undertaking full-time university study alongside their training and competition loads were vulnerable to increased levels of stress at certain periods of the year (pre-season and examination time). Monitoring and understanding these stressors may assist coaches and support staff in managing overall stress in these athletes.

Keywords: student-athletes, academic stress, athletic performance, injury, athlete monitoring, illness, sport training

INTRODUCTION

The last 30 years has seen an increase in professionalism in sport and with that has come greater interest in the scientific approach to training and recovery of elite athletes. For athletes, the balance between stress and recovery is crucial for improving sport performance (Kellmann, 2010). On the one hand, adequate physical stress is required in the form of training load which produces fatigue resulting in adaptation of the various bodily systems (Smith, 2003). On the other hand, recovery from training stress is also important if fatigue is to be overcome, adaptation optimized and subsequent performance enhancement realized. Because training, and therefore adaptation, and subsequent performance will be compromised if this balance is not maintained, monitoring of stress and recovery in athletes is vital.

In addition to training loads, elite athletes typically encounter stress from other sources such as social, work-related, lifestyle and athlete-coach relationships. Pioneering work by Pierce and Stratton (1981), suggest that young athletes experience the highest stress when they perform poorly, make mistakes, and when they perceive pressure from parents, coaches, and teammates (Pierce and Stratton, 1981). Athletes who are also involved in university study are very prone to study-related stressors such as coursework demands, study/life balance, and financial strain (Stallman and Hurst, 2016).

When stress (psychological, academic, training, or performance-related) overloads an athlete's stress-coping ability, the susceptibility to performance decrement increases, as does the risk of injury and illness. The "Stress and Injury" model proposed by Williams and Andersen has been used to explain this relationship (Andersen and Williams, 1988; Williams and Andersen, 1998). According to the model the stress response increases general muscle tension in the body, which can result in reduced motor coordination and flexibility, both of which can influence fatigue. The model also suggests that stress may diminish the visual field, thereby reducing visual attention which may decrease the ability to use relevant peripheral information (Williams and Andersen, 1998). There is a strong body of evidence indicating that an increase in psychosocial stress also increases injury risk in athletes (Williams, 2001; Galambos et al., 2005; Pensgaard et al., 2018), and reducing such stress (via stress-management interventions) decreases the likelihood of injury (Perna et al., 2003). Similarly, chronic high training load stress (Drew and Finch, 2016) or sudden and severe increases in training load stress over a short period of time (Gabbett et al., 2014; Hulin et al., 2014, 2015) can result in significantly higher risk of injury.

Excessive stress (training- and non-training related) not only increases the risk of injury but also the development of acute illness (Walsh et al., 2011b; Gleeson and Pyne, 2016) as well as the risk of overtraining or burnout (Kellmann et al., 2018). Fry et al. (1991) suggested the relationship between health and loading/stress can be viewed as a continuum where load/stress and recovery are the two competing factors ultimately influencing health (Fry et al., 1991). The theory suggests training (and non-training) related loads create stress on the athlete which shifts the athletes psychological and physical well-being along a continuum

that advances from homeostasis to acute fatigue, over-reaching, overtraining, subclinical changes (tiredness, lethargy, etc.), clinical symptoms (compromised immunity, influenza, etc.) and illness (or injury). The ideal amount of stress should progress the athlete from the area of homeostasis on the continuum into the area of acute fatigue or over-reaching. However, when adequate recovery is provided, the process is reversed, resulting in adaptation and restoration of homeostasis at a higher level of fitness. Too much stress or inadequate recovery will prohibit adaptation, leading the athlete into the unhealthy and potentially harmful end of the continuum. Monitoring of training- and non-training related stress can therefore enhance the understanding of the training and stress response and help prevent the risk of maladaptation to training which may result in illness or injury (Foster, 1998; Halson, 2014).

Athletes experience stress and subsequent fatigue on a regular basis, yet it is a complicated process (Naokes, 2012), which can follow an individualized pattern unique to each athlete (Mann et al., 2014). Thus monitoring the individual stress response to training and competition is necessary to maintain the unique balance required for homeostasis in each athlete. Such monitoring can take the form of subjective and objective measures used to indicate training load (training volume/duration, rating of perceived exertion, GPS, etc.) and stress/fatigue (perceptual wellness scales, biochemical markers, immunological markers, sleep quantity and quality, etc.) (Halson, 2014). Since it is impractical and expensive to monitor large numbers of athletes in the lab, many coaches and trainers have adopted subjective measurement systems to monitor the stress and fatigue of their athletes (Saw, 2015; Nässi et al., 2017). The subjective reporting of training load, perceived stress, and psychological mood states can be a reliable indicator of training load (Robson-Ansley et al., 2009), and can be more responsive to tracking the training response than objective measures (Saw et al., 2016).

The Lincoln University Sports Scholarship program in New Zealand supports approximately 100 athletes each year in 8–10 major sports. Given that these athletes also undertake university study, any accumulation of unmanaged stress may result in injury, illness or a number of other adverse effects. Time away from training or competition due to illness or injury can elicit major consequences, including rehabilitation costs or adverse social, psychological and economic impacts. Having the athlete's welfare in mind, a monitoring program was developed to monitor subjective measures of stress and fatigue.

The primary objective of this research was to examine the subjective measures that contribute to the overall stress among young elite athletes in a university educational environment. A secondary objective was to investigate the relationship between subjective measures of stress (including training load) and injury or illness.

MATERIALS AND METHODS

Subjects

The perceived stress, training loads and injury/illness incidence were retrospectively investigated from 2014 to 2017 in 182

TABLE 1 | Characteristics of athletes.

	<i>n</i>	Weekly training duration (min)	Weekly training load (arbitrary units)
Male	132	280 ± 184	1557 ± 1046
Female	50	258 ± 175	1486 ± 1040
Rugby	60	294 ± 165	1596 ± 973
Netball	13	247 ± 159	1690 ± 1108
Hockey	35	226 ± 161	1319 ± 972
Cricket	21	219 ± 138	1378 ± 820
Basketball	20	268 ± 169	1536 ± 1083
Rowing	10	295 ± 227	1928 ± 1529
Athletics	4	531 ± 255	1721 ± 874
Football	8	279 ± 195	2037 ± 1611
Other sports	11	142 ± 87	970 ± 585

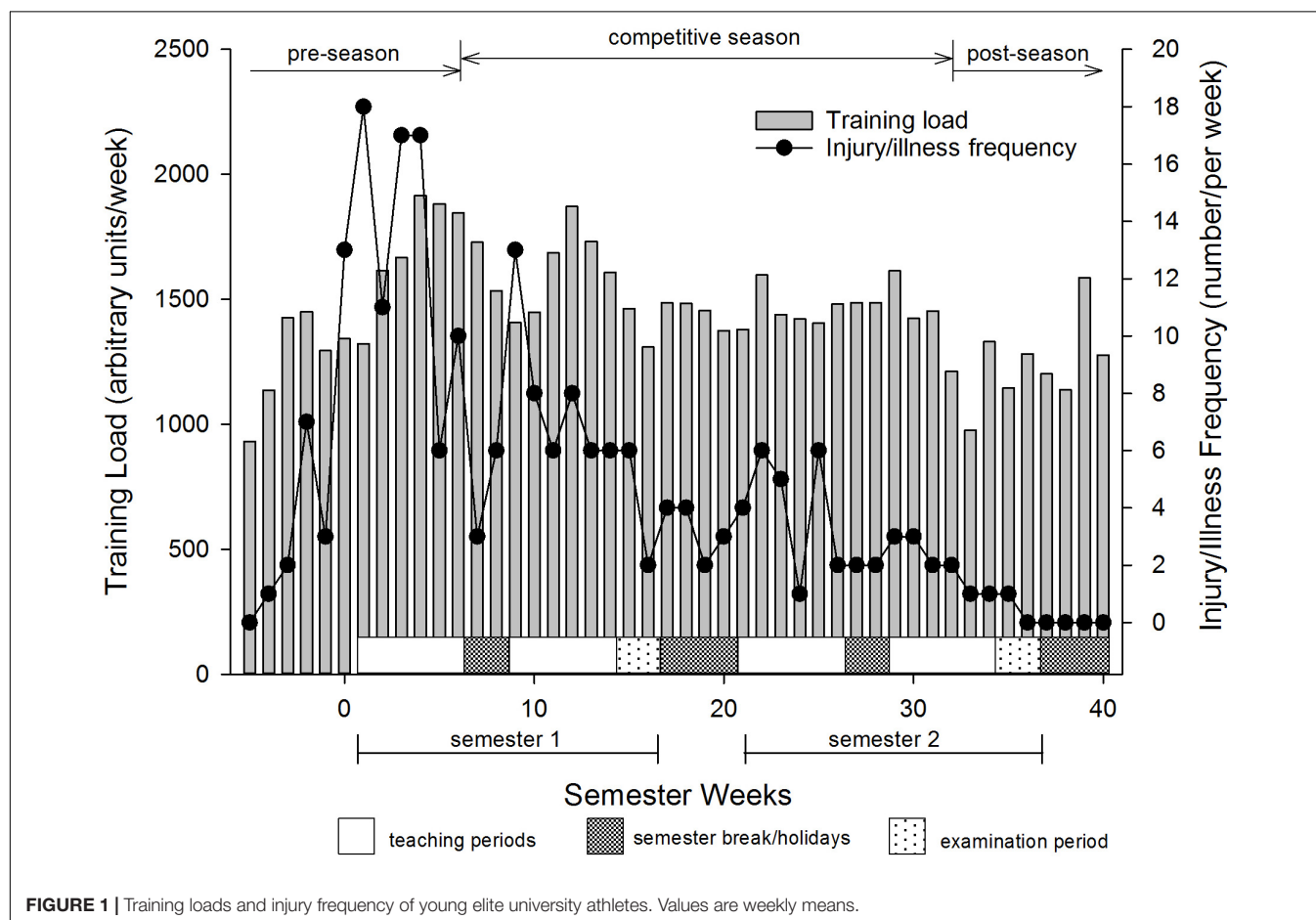
Data are mean ± SD. Training load = training intensity (measured via rating of perceived exertion) × training duration (minutes). Other sports included athletes like cyclists, triathletes, throwers, etc.

young athletes during their time at university (approximately-February to October over 4 years). Athletes were involved in a university sport scholarship program where athletes received nutritional, psychological, and medical advice along with individualized training. All participants were young elite

athletes (18–22 years old) selected for age-group provincial or national representative honors. This study was carried out in accordance with the recommendations of the Lincoln University Human Ethics Committee. All subjects gave their written informed consent in accordance with the Declaration of Helsinki. The protocol was approved by the University's Human Ethics Committee (Reference No. 2018-01). Participant characteristics are presented in **Table 1**. The wide cross-section of athletes sampled caused difficulty in splitting the year of competition into appropriate training phases, however 75% of the athletes represent sports that are played in winter and have a similar training and competitive season. Thus, we have classified the data into the following phases; pre-season (up to and including week 6 of semester 1); competitive season (weeks 7 to 32); and post-season (week 33+) (see **Figure 1**).

Study Design

This longitudinal retrospective cross-sectional study used a commercially available software system (Health and Sport Technologies, Ltd., trading as Metrfit, Millgrange, Greenore, Co., Louth, Ireland) to collect training data along with subjective feelings of stress, fatigue, academic pressure, mood, sleep quality/quantity as well as clinically-derived incidence of injury or illness in athletes during their time at university. The data was



collected using the Metrifit phone application 3–4 weeks prior and then throughout the athlete's academic year (two semesters). Each semester comprised of 12-weeks of teaching, 1 week of study break, followed by a 2-week final examination period to close the semester. Each semester is interrupted mid-way with a 2-week holiday break. There is a 4-week break between semester 1 and 2, and a summer holiday of 15 weeks prior to the start of the next university year in February. Most students spend holidays (mid and end-of-semester) away from university, for example, returning home to spend time with their families or traveling.

Training

Every year, individualized training programs were developed by the strength and conditioning staff at the university for each athlete, depending on the type of athlete, their competitive season and injury status. In most weeks, athletes would have at least three training sessions, one sport-specific skills session and one practice game or competition. Athletes recorded their daily training information including type, duration and intensity of training. The intensity of training was estimated using a modified 10-point scale (Foster et al., 2001). Previous research by our group (Hamlin and Hellemans, 2007) and others (Eston and Williams, 1988; Impellizzeri et al., 2004; Gabbett and Domrow, 2007), support these effort ratings as reliable indicators of exercise intensity.

The training load (internal training load) was calculated as the product of volume (duration of training) and intensity (subjective rating of training intensity) as proposed by Foster et al. (2001). It is well-documented that subjective measures (mood disturbance, perceived stress, sleep disruption, etc.) consistently show superior responsiveness to training compared to objective measures (Verde et al., 1992; Coutts et al., 2007; Saw et al., 2016). Unfortunately many existing subjective questionnaires (e.g., Recovery Stress Questionnaire for Athletes (Kellmann and Kallus, 2001), Daily Analysis of Life Demands of Athletes (Rushall, 1990), and Multi-Component Training Distress Scale (Main and Grove, 2009) are long with numerous questions making them time-consuming and complicated and not fit for purpose in a practical setting. Because of this, the Lincoln University Sport Scholarship program decided to incorporate elements of established measures into our own customized, brief, easy-to-use, self-report measure. For this study we asked a series of questions used successfully in a number of other studies (Hamlin and Hellemans, 2007; Hamlin et al., 2017) which were modeled on previous research (Mackinnon and Hooper, 1996; Killen et al., 2010). The questions used in the phone App were based on a five-point Likert scale to record athletes subjective ratings of mood (1 = very stressed, 2 = quite stressed, 3 = slightly stressed, 4 = little stress, 5 = no stress), sleep quality (1 = poor, 2 = below average, 3 = normal, 4 = good, 5 = very good), energy levels (1 = extremely low, 2 = very low, 3 = low, 4 = normal, 5 = high/excellent), muscle soreness (1 = extremely sore, 2 = very sore, 3 = quite sore, 4 = mild soreness, 5 = no soreness), and academic pressure (1 = academic pressure high, 2 = academic pressure building, 3 = heavy academic day, 4 = normal academic pressure, 5 = no academic pressure). In the phone App, athletes had to move an electronic slider (which was initially situated on the far left of the screen, or at number "1" for each question) to

the appropriate perceived subjective rating for the day for that question. Athletes also recorded their perceived sleep duration in hours and minutes. The Metrifit software also produces a calculated variable called the Readiness to Train (RTT) score. This variable gives a score out of 100, that is thought to represent the overall stress in the athlete and the estimated ability of the athlete to be ready to train (100 = fully fresh with no fatigue and optimally prepared to train). The RTT score uses the athlete's subjective measures of mood state, sleep quality, energy level, muscle soreness, academic stress and then applies a weighting appropriate to each subjective measure's influence on performance and recovery and calculates the RTT. The exact weighting and algorithm used is considered intellectual property (IP) by the software owners and subjective to IP laws. All athletes were given clear instructions on how to use the Metrifit system which included a 2-h training session around understanding the data required by the system and how to enter the data using the Metrifit App Interface on each student's phone. Athletes were encouraged to use the software to input data daily and they received text message reminders on their mobile phones if data entry was missed.

It is important to not only focus on current training regimes, but also what athletes have previously completed in terms of preparation for training. Previous work suggests a sharp increase in current training (acute training load), without the appropriate preparation (chronic training load), can result in injury (Gabbett, 2016). We therefore calculated the acute:chronic workload which gives an estimate of the preparedness of athletes to handle increases in workload stress using an exponentially weighted moving average (EWMA) as proposed by Williams et al. (2017). The calculation is as follows:

$$EWMA_{\text{today}} = \text{Load}_{\text{today}} \times \lambda_a + ((1 - \lambda_a) \times EWMA_{\text{yesterday}}) \quad (1)$$

Where λ_a is a value between 0 and 1 representing the degree of decay, which assigns a lower weighting for older observations. The λ_a was calculated as:

$$\lambda_a = 2/(N + 1) \quad (2)$$

Where N is the chosen time decay constant in days, which was selected as 1-week (to represent acute workload over the last 7 days) and 4-weeks (representing chronic workload over the last 28 days). After arbitrarily recording the first observation in the dataset as the first observation, the above formula was used to calculate the average acute and chronic workloads for each week for all subjects combined. The acute:chronic ratio was then calculated by dividing the acute workload by the chronic workload (Williams et al., 2017).

Injury and Illness

The Metrifit system allows the entry of injury and illness data by the athlete, coach or medical staff. For this study we have used the injury definition of Timpka et al. (2014) which states an injury is a physical complaint or observable damage to body tissue produced by the transfer of energy experienced or sustained by an athlete during training or competition, regardless of whether medical attention was received. In this study most injuries (70%)

were diagnosed by a registered physiotherapist or medical doctor. Injuries were counted only once and any re-injury of a previous injury was not included in the data. Injuries were grouped by anatomical location, and nature of injury (e.g., strain, sprain, rupture, etc.) according to current guidelines (Timpka et al., 2014). We also categorized injuries according to the occasion (i.e., match, training), and whether the injury mechanism was via contact or not. Illness was defined as a physical or psychological complaint or manifestation by an athlete not related to injury, causing an impairment in competition or training regardless of whether the athlete received medical attention (Timpka et al., 2014).

Statistical Analysis

Changes in the mean of the variables and standard deviations representing the between-and within-subject variability were estimated using a mixed modeling procedure (Proc Mixed) in the Statistical Analysis System (Version 9.3, SAS Institute, Cary, NC, United States). Chances that the true effects were substantial were estimated when a value for the smallest worthwhile effect was entered into the calculation. We chose 0.20 standardized units (representing change in mean divided by the between-subject SD at baseline) as the smallest worthwhile change (Cohen, 1988). To make inferences about the true (population) uncertainties in the estimate of change were presented as 90% confidence intervals and as likelihoods that the true value of the effect was increased, decreased or trivial. The descriptors: increased, trivial or decreased were used to describe the direction of the change. Where the confidence interval spanned all three possibilities (increased, trivial, and decreased), the result was deemed unclear. In all other cases, such as no overlap, or an overlap between two possibilities (trivial and increased, or trivial and decreased) a clear result was achieved. The magnitude or probability of the change was assessed using a qualitative scale defined as: <0.5%: almost certainly not; <5%: very unlikely; <25%: unlikely/probably not; 25–75%: possibly, possibly not; >75%: likely, probably; >95%: very likely; and >99.5%: almost certainly.

Team training loads and the incidence of injury were analyzed in SAS using the PROC CORR procedure to determine the association between training load and injury prevalence. The weekly training data varied considerably between different stages of training (e.g., the start of the training year compared to the rest of the training year), therefore, the aggregated weekly results were analyzed during what was believed to be the pre-season for most athletes (e.g., up to and including week 6 of semester 1; i.e., weeks -5 to -6) and the competitive season (weeks 7–32). Since this was aggregated data grouped by week, the results could then be applied to the training group in general.

Individual subjective measures (mood state, sleep quality/duration, energy levels, academic stress), along with injury data were modeled together using a single logistic regression model with a binomial distribution (injured, not injured) and logit link function. These data were analyzed in SAS using the PROC LOGISTIC procedure. The summary statistic used for assessing the adequacy of the fitted model (goodness of fit) was the likelihood ratio chi-square. Odds ratios (and 95% confidence limits) were calculated to determine whether changes

in subjective measures increased (or decreased) the odds of injury. Unlike the training data, the subjective data represented individuals, therefore the results can be applied to the individual rather than the training group.

Illness data was fitted with a non-linear regression equation (Peak, Gaussian, 3 Parameter [$f = a \cdot \exp(-0.5 \cdot ((x-x_0)/b)^2)$] to smooth the individual illness frequencies and identify periods of highest illness counts.

RESULTS

Training Duration and Load

The training loads for the athletes over the academic year are shown in **Figure 1**. Five weeks prior to starting university the athletes were completing approximately 931 ± 710 (mean \pm SD) arbitrary units (au) of training load per week (~ 3.3 h in duration) which increased substantially over the next 8–9 weeks to peak at 1916 ± 1229 (an increase of 106%). Training loads were then maintained at about this level throughout semester 1 apart from slight reductions during semester holidays (students not required to stay on campus to train) and examination periods. The overall average training load was lower ($0/18/82$, chances of positive/trivial/negative differences in training load; $p = 0.001$) in semester two (1409 ± 952 au) compared to semester one (1594 ± 1079 au).

Subjective Markers

At the start of each year, athlete's perceived energy levels started to decline and only recovered during breaks spent away from university life (**Figure 2A**). Athletes perceived their energy levels to be lowest during the first semester examination period. Perceived levels of muscle soreness were highest at the beginning of the year, particularly just prior to the start of the teaching. Muscle soreness gradually increased throughout the duration of each semester, but recovered during the mid and end-semester holiday breaks.

Athletes were unstressed at the start of each semester as indicated by their relatively high mood scores (1 = stressed, 5 = unstressed) (**Figure 2B**). However, as the semester progressed perceived mood scores deteriorated and only recovered back to baseline levels during the holiday breaks. Mood scores were lowest during the two examination periods at the end of each semester, particularly in semester one. Sleep quality mirrored mood scores such that athletes perceived their quality of sleep was highest in the periods away from university and lowest during the examination periods. The sleep quality data was reinforced by the sleep duration data that showed athlete's sleep duration tended to increase when away from university. Perceived academic stress was at its highest during the examination periods occurring at the end of each semester (**Figure 2C**).

A relationship was observed between subjective measures of mood, energy, academic stress, sleep duration and the odds of injury, such that lower levels of mood, sleep duration, and academic stress or increased levels of energy were able to predict injury (**Table 2**). The model was successful at fitting the data as evidenced by the likelihood ratio $\chi^2 = 31.76$ with 3 degrees

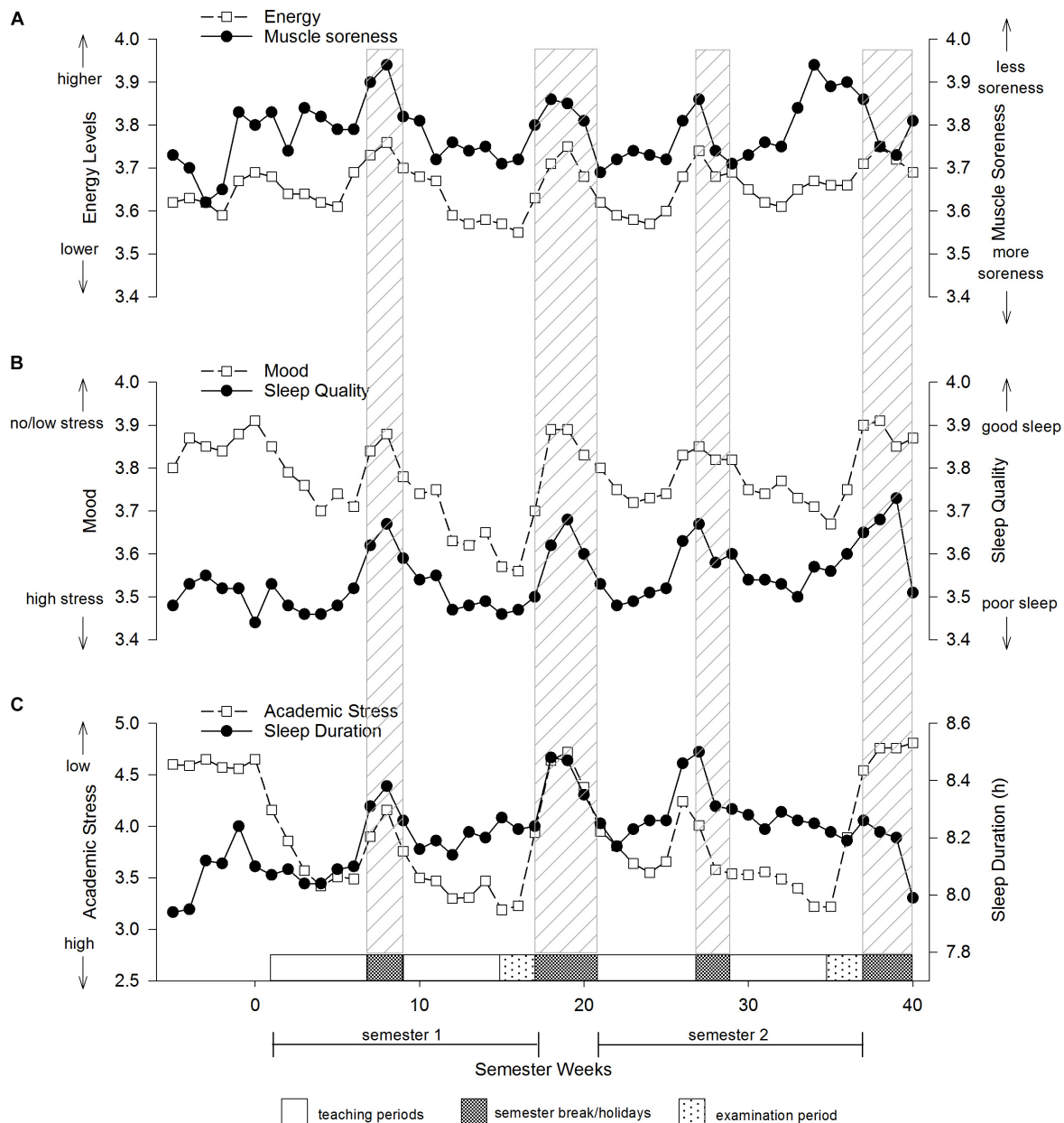


FIGURE 2 | Subjective measures of young elite university athletes. **(A)** Energy and muscle soreness, **(B)** mood state and sleep quality, **(C)** academic stress and sleep duration. Values are weekly means.

of freedom, $p < 0.001$. Converting the odds ratio to percent change [$1 - (\text{OR}) \times 100$] we found that every unit decrease in mood was associated with a 10.8% increase in the odds of incurring an injury. Similarly, each unit decrease in sleep duration and academic pressure was associated with a 5.9 and 9.0% increase in the odds of getting injured, respectively. Sleep quality was not associated with the odds of injury. The final regression model was $(-1.2523 - 0.1137 \times \text{mood} + 0.0056 \times \text{sleep quality} - 0.0605 \times \text{sleep duration} + 0.0706 \times \text{energy} - 0.0947 \times \text{academic})$.

Similar to the separate subjective measures, the aggregated RTT score was lowest during the examination periods,

particularly in semester 1 ($64 \pm 15\%$ at week 16, mean \pm SD), but recovered to baseline levels (approximately 74–75%) during times when students were away from the university (Figure 3).

Injury and Illness

The overall incidence of injury and illness for the 45 weeks that athletes recorded their data was 15.6 ± 3.9 injuries per 1000 training hours (mean \pm 90% CL). Incidence of injury was higher over the pre-season [up to week 6 (30.7 ± 3.9 injuries per 1000 training hours)], than at any other time during the rest of the year (10.3 ± 1.9 injuries per 1000 training hours), 0/2/98, chances of

TABLE 2 | Odds ratios of psychological variables as risk factors for injury in young elite university athletes.

Factor	Odds ratio	95% confidence limits
Mood	0.89*	0.85 to 0.94
Energy	1.07*	1.01 to 1.14
Sleep quality	1.01	0.96 to 1.06
Sleep duration	0.94*	0.91 to 0.97
Academic stress	0.91*	0.88 to 0.94

*Odds of injury substantially related to factor.

positive/trivial/negative differences in injury incidence; $p = 0.01$). The majority of injuries sustained over the 4 years were to the ankle and knee which made up almost 30% of all injuries and illness (Table 3). Injuries to the head, shoulder/clavicle, lumbar spine/lower back, thigh, lower leg, and groin were also common. Over half of the injuries sustained over the 4 years were muscle strains and joint sprains (Table 4). The most common illness was lower respiratory tract infection mainly from influenza (Table 3). Illnesses accounted for approximately 14% of the loss of training and playing days and were most notable during times of highest stress which was just before and during semester 1 examinations (Figure 3).

Effect of Training Load on Injury Incidence

The sudden increase in workload as indicated by the higher acute:chronic workload ratio over the first 9 weeks of pre-season was associated with an increased incidence of injury and illness ($r = 0.63$). During the competitive season, however, increases in training loads resulted in no further increases in the incidence of injury or illness (Figures 1, 4).

DISCUSSION

The monitoring of athletes has become an important area in sport science, not only because it is the aim of coaches and trainers to give athletes the best care and support, but it is also important to protect athletes as much as possible from any harm or unwanted consequences of training. This research highlights periods within the academic year when athletes undertaking university study are likely to be influenced by increased stress which is associated with increased risk of injury and illness. Our data shows obvious cyclical effects whereby athlete's subjective measures of stress increased steadily during the semester to reach a nadir during the examination period, after which stress was reduced during the semester break/holiday period (Figure 2).

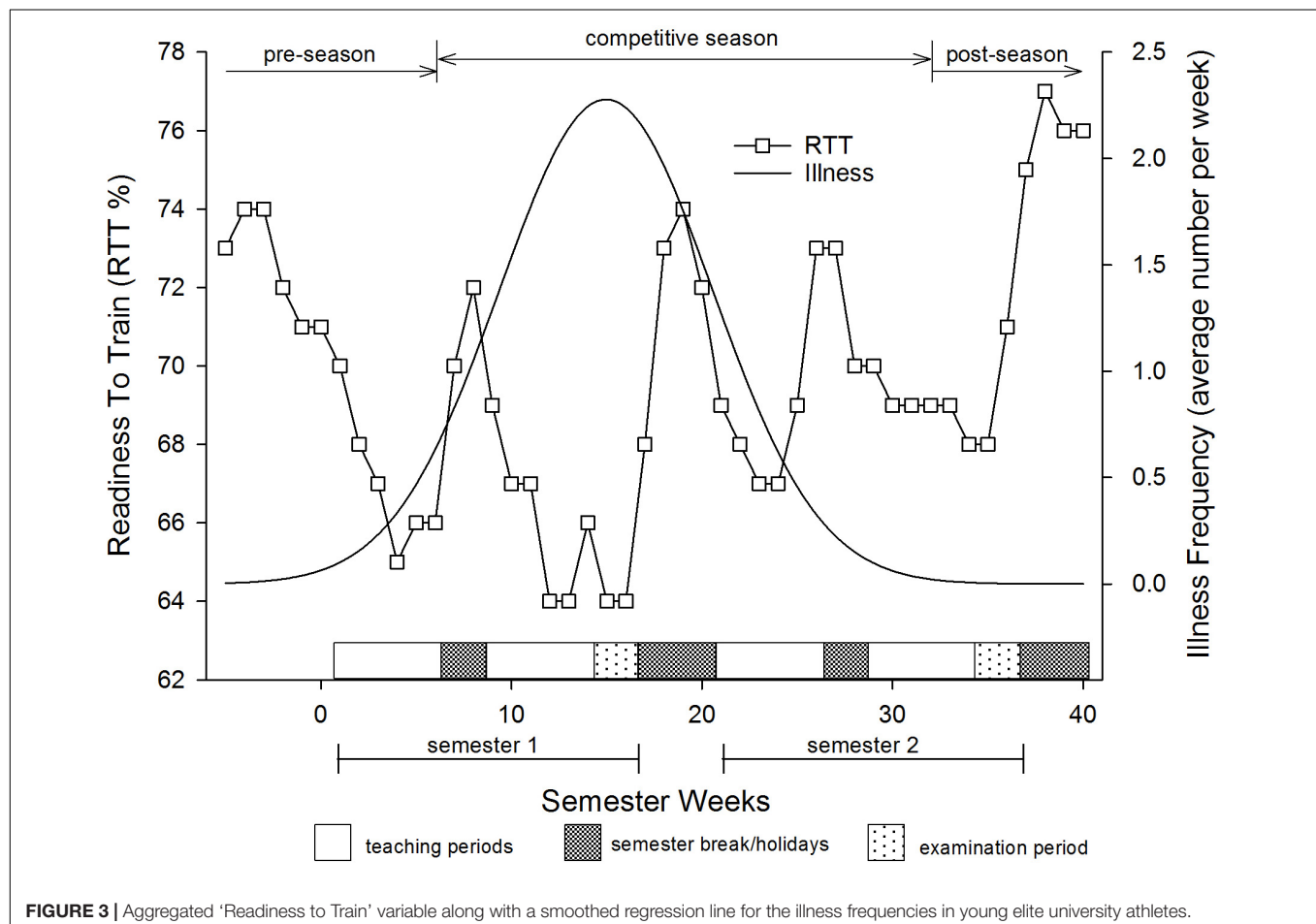
**FIGURE 3 |** Aggregated 'Readiness to Train' variable along with a smoothed regression line for the illness frequencies in young elite university athletes.

TABLE 3 | Location of injury and illness sustained by young elite athletes over 4 years at university.

	<i>n</i>	%
Head (including concussion)	15	5.8
Face (including eye, ear, nose)	5	1.9
Shoulder/clavicle	19	7.3
Neck/cervical spine	0	0
Abdomen	3	1.2
Thoracic spine/upper back	6	2.3
Lumbar spine/lower back	14	5.4
Sternum/ribs	4	1.5
Elbow	3	1.2
Upper arm	1	0.4
Finger	4	1.5
Hand	6	2.3
Wrist	5	1.9
Thumb	4	1.5
Thigh	16	6.2
Lower leg	11	4.2
Hip	4	1.5
Groin	11	4.2
Knee	39	15.2
Ankle	36	13.9
Foot/toe	8	3.2
Other	9	3.5
Upper respiratory tract	7	2.7
Lower respiratory tract	19	7.3
Other illness	10	3.9
Total	259	100

Data are frequencies of injuries and illness and % of total over 4 years.

TABLE 4 | Type and cause of injury sustained by young elite athletes over 4 years at university.

Type of injury	<i>n</i>	%
Strain/muscle rupture/tear	78	30.1
Sprain (injury of joint and/or ligament)	64	24.7
Ligamentous rupture	5	1.9
Concussion	16	6.2
Contusion/hematoma/bruise	6	2.3
Fracture (traumatic)	9	3.5
Fracture (stress)	4	1.5
Dislocation/subluxation	7	2.7
Laceration/abrasion/skin lesion	10	3.9
Other	24	9.3
Cause of injury		
Contact	101	39.0
Non-contact	110	42.5
Other/missing data (i.e., viral, bacterial)	48	18.5

Data are frequencies of injuries and % of total injuries over 4 years.

The overall incidence of injury/illness was ~16 injury/illness per 1000 training hours over the whole academic year. Since 13% of these were illnesses (mostly influenza) the actual injury rate was slightly lower at 14 injuries per 1000 training hours. This incidence represents the number of injuries that occurred

as a result of training and competition combined and is higher than what has been found in football (soccer) (8.0/1000 training hours) (Ekstrand et al., 2011) or rugby league (6.9/1000 training hours) (Killen et al., 2010), but is similar to some earlier research on rugby players (12.4/1000 training hours) (Sparks, 1985). Evidence indicates that stress plays a major part in the etiology of injury (Williams and Andersen, 1997; Rogers and Landers, 2005). Therefore reducing unwanted stress may help reduce the incidence of injury, particularly during the pre-season period in the athletes of this study.

In this study we observed a relationship between a number of subjective measures and odds of injury. In particular, this study showed that mood, sleep duration (but not quality) and academic pressure were the strongest contributors to injury. These findings corroborate previous work by Galambos et al. (2005) who found subjective measures (mood disturbance and increased perceived life stress) were able to predict injury in elite athletes. Moreover, results from this study highlight the importance of measuring subjective stress variables in elite athletes at university. Indeed, a large majority of the models in the sport injury literature suggest that sport injuries result from an accumulation of not just physical but psychosocial stressors (Pensgaard et al., 2018).

Probably the most influential theory on the relationship between psychosocial stress and injury is the early work of Andersen and Williams (1988) who outlined a stress-injury model that suggested athletes who accumulate stress levels that overcome their stress-coping abilities are unable to relax which subsequently alters the athletes attentional ability (Williams et al., 1986). Compromised attention may then result in a failure to detect vital clues about the athletes body or the environment and/or increases in their muscle tension (Nideffer, 1983), thereby disturbing motor coordination and increasing the risk of injury (Andersen and Williams, 1988). Andersen and Williams' theory implies, that if situational demands exceed an athlete's coping ability, elevated stress levels will result. The stress response is heightened further if the athlete perceives the consequences of their performance will detrimentally affect the athlete's sports career or self-esteem (Andersen and Williams, 1988). Indeed, the athletes involved in this study were under considerable stress over and above what normal university students might encounter. The sport scholarship athletes must not only pass their courses throughout their scholarship tenure, but they must also perform to high expectations in the gym (meet strength and conditioning targets) and on the sports field (be selected for certain development or representative teams which compete at the highest level for this age group). This increased stress, particularly at the beginning of the year, in the athletes of this study may have led to the higher rates of injury.

A regression analysis showed that the individual subjective measures (mood state, academic stress, etc.) were related to the risk of injury. When employing an aggregated measure of several subjective variables in the regression model (RTT score), a stronger relationship with the risk of injury emerged (OR = 0.58, CL 0.47 to 0.71). Perhaps analyses using an aggregate variable comprised of a number of subjective measures (e.g., academic, mood, sleep, energy levels) may be better suited to monitoring

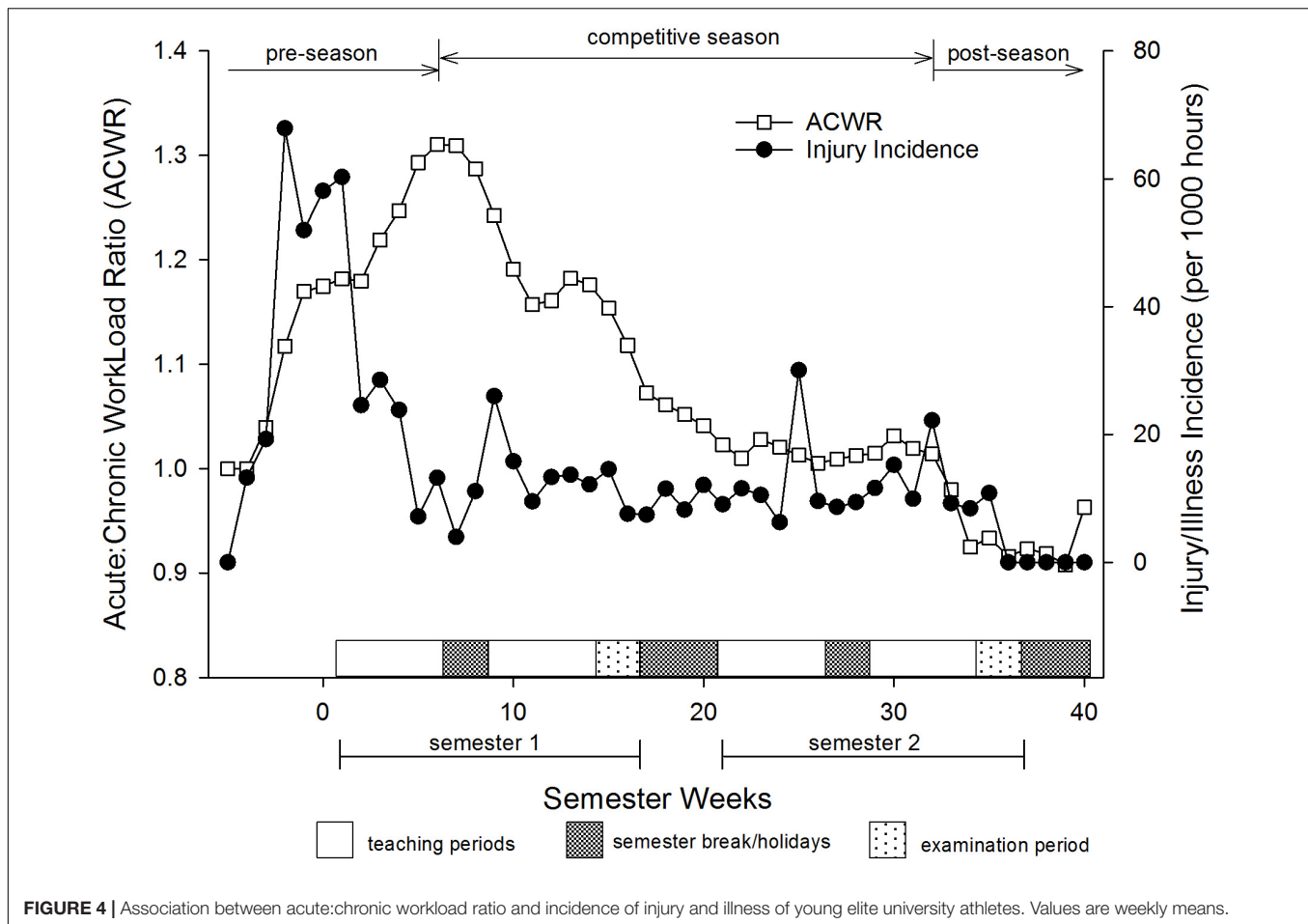


FIGURE 4 | Association between acute:chronic workload ratio and incidence of injury and illness of young elite university athletes. Values are weekly means.

overall subjective levels of athlete stress than individual subjective measures, particularly when determining relationships with the risk of injury.

Interestingly an increase in subjective feelings of energy was associated with a 7.3% increase in the odds of injury (Table 2). While this seems counter-intuitive, since injury is normally associated with negative emotional states (Kolt and Kirby, 1994), more recent research suggests injury may also be associated with positive emotional states (Hanin, 2000). It has been theorized that being successful or feeling energized may result in complacency, which might lead to a decrease in alertness and a subsequent increased risk of injury (Devonport et al., 2005). On the other hand, being overconfident may also prevent utilization of all resources during activity, resulting in underperformance and injury. Overconfidence may result in risk-taking behavior during training or competition which could also result in increased injury risk. Jones et al. (2017) noted this unexpected relationship and suggested that when athletes perceive themselves to have high energy levels (low fatigue) they may train or play at higher intensities, thereby increasing the forces and strains involved during exercise resulting in higher risk of injury (Jones et al., 2017). Whatever the cause of this association, coaches, and support staff need to be vigilant that athletes follow appropriately-prescribed training loads (even if

they have low fatigue levels and are feeling highly energized) in order to avoid injury.

Our study also found that high acute:chronic team training workloads were associated with an increased risk of injury. High acute:chronic workload, particularly at the beginning of the year (during the first 7 weeks where athletes are in their pre-season period), increased the incidence of injury and illness almost fourfold compared to the rest of the year (Figure 4). Gabbett and Domrow (2007) also found increased workload toward the beginning of the training season resulted in increased injury prevalence (Gabbett and Domrow, 2007). Previous research suggest inadequate pre-season training or low off-season aerobic fitness increased athlete's risk of injury during the pre-season (Gabbett and Domrow, 2005). The reduction in injury and illness despite the increase in workload during the semester (from weeks 7 onwards) would suggest the high injury/illness spike during weeks 1–7 may be due to insufficient preparation prior to commencing pre-season training. Indeed, the highest muscle soreness levels of the year also occurred during this period which would indicate physical unpreparedness of the athletes. These data suggest there is a critical window during the pre-season when coaching and support staff need to be attentive to avoid unwanted illness and injury.

Previous researchers have suggested acute:chronic workloads within the range of approximately 0.8–1.3 represent the ‘sweet spot’ where training load is high enough to result in adaptation but not too high as to cause a heightened risk of injury (Blanch and Gabbett, 2016; Gabbett, 2016). These authors suggested an acute:chronic workload above 1.5 increased the risk of injury in athletes. The average weekly acute:chronic workload in our athletes was consistently below 1.5 (**Figure 4**) and yet our athletes still incurred injuries. The differences in athletes between studies may account for the higher injury incidence at a lower acute:chronic workload, although Blanch and Gabbett (2016) data was based on team sport athletes who make up a large proportion of the athletes in this study. However, most of the research by Blanch and Gabbett (2016) and Gabbett (2016) was on professional elite athletes who probably have a greater training base than our athletes and can sustain a larger increase in the acute:chronic workload before injuries occurred. Other studies investigating injury prevalence in collision sports suggest the introduction of contact drills and skills into training may increase the risk of injury (Gabbett and Domrow, 2005), however, this is unlikely in this study, since such drills and skills were not introduced until mid-way through the start of semester one when injury incidence was reducing.

The most common injuries tended to be joint injuries (shoulder, knee and ankle made up 36.4% of all injuries) that occurred predominantly during the pre-season period. All athletes were given off-season programs, therefore it was not a matter of inadequate information causing the injuries, but perhaps a lack of motivation. A possible solution might be to incorporate positive reinforcement to encourage athletes to comply with their pre-season training program (e.g., fewer sport scholarship ‘chores’ to attend to if athletes meet certain fitness targets). Perhaps the challenge of a running time trial on their first week back of semester might also encourage maintenance of fitness standards over the off-season break. Perhaps we should anticipate a larger decline in fitness over the off-season and cater for this by having a ‘home-based preseason’ warm-up period which would prepare athletes prior to coming to campus for their actual preseason training. An additional approach could be to introduce stress management skills along with muscle relaxation and attentional awareness techniques that may help reduce stress and thereby vulnerability to injury (Olmedilla-Zafra et al., 2017).

This study found that in times of high stress, illness rates increased substantially, particularly during the winter semester (**Figure 3**). This trend was most obvious during the end of semester one leading into the first examination period of the year. Periods of heavy training have been linked to depressed immunity and subsequent illness (Walsh et al., 2011a). However, the most dangerous period for illness in our athletes was at a time when training load was relatively moderate (**Figure 3**), suggesting training stress may not be the sole culprit behind the increased prevalence of illness; a finding common in the literature (Fricker et al., 2005; Veugelers et al., 2016). Previous research indicates that impaired immune function may also be related to sleep deprivation (Simpson et al., 2017). Indeed, an earlier study reported subjects with less than 7 h sleep were almost three times more likely to develop influenza than those

sleeping eight or more hours (Cohen et al., 2009). It is also well-known that academic stress (academic tests and papers, etc.) is positively correlated with the occurrence of illness in university students (Lesko and Summerfield, 1989). We postulate that the combined effects of oncoming winter, along with regular training with low sleep quality and reduced sleep duration and high academic stress may have acted to push the athlete along the health continuum away from homeostasis and toward maladaptation thereby suppressing the immune response in our athletes resulting in an increased likelihood of developing illness over the semester one examination period.

This study has three key limitations that should be noted. Firstly, the use of ‘bespoke’ subjective questions used in the phone App questionnaire in this study suggests that results should be considered speculative until substantiated by vigorous validity and reliability testing. Secondly, this research relied on the timely and correct entering of accurate data by the athletes and any deviation from this practice may have corrupted the data. Thirdly, the participants of this study are all young people engaging in elite sports programs and university study, therefore, the results of this research may only apply to this cohort and may not be generalizable.

This is not the first study to investigate the association between subjective markers and injury/illness in athletes. However, several factors make this study unique, including the collection of long-term (over 4 years) subjective data, which provide an overall impression of the change in these measures during the academic year for elite athletes. This study also collected clinically-diagnosed illness and injury data allowing us to investigate the links between injury/illness and subjective levels of stress. Finally, the dataset itself, using young elite athletes undertaking university study is relatively distinctive. Although the study encompasses a broad spectrum of athletes from a wide variety of sports, the findings are quite clear. Athletes undertaking academic workloads in addition to their normal physical training and competition stresses are vulnerable at certain times of the year to increased stress (pre-season and examination time). These results have implications for not only the athletes, but also their coaches, administrators, and other support staff (athletic and academic). The implications are that they may better understand that certain clusters of subjectively-reported stressors can trigger higher amounts of stress, which can lead to increased risk of injury and illness. Moreover, the results suggest adopting a stress-reduction program, particularly prior to the pre-season and examination periods, might help to prevent issues from arising, or help to efficiently mitigate and manage those that do arise.

DATA AVAILABILITY STATEMENT

The dataset for this manuscript is not publicly available because of commercial sensitivity in regards to the software system used in collecting the data (Health and Sport Technologies, Ltd., trading as Metrfit, Millgrange, Greenore, Co., Louth, Ireland). Requests to access the datasets should be directed to MH mike.hamlin@lincoln.ac.nz.

AUTHOR CONTRIBUTIONS

MH conceptualized and designed the study. MH and DW assisted in the planning and acquisition of data. MH, DW, CE, CL, and YK helped with the analysis and interpretation of the data, critically revising the manuscript, and adding important intellectual content. All authors gave approval for the final version of this manuscript to be published and agreed to be accountable for all aspects of the work.

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FUNDING

The Department of Tourism, Sport and Society, Lincoln University, New Zealand, funded the open access fee for this article.

ACKNOWLEDGMENTS

The authors thank the athletes and their coaches for their assistance.

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Conflict of Interest Statement: YK was employed by company Sports Doctors, Christchurch, New Zealand.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Cardiorespiratory Temporal Causal Links and the Differences by Sport or Lack Thereof

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OPEN ACCESS

Edited by:

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 21 October 2018

Accepted: 16 January 2019

Published: 05 February 2019

Citation:

Młyńczak M and Kryštofiak H (2019)
Cardiorespiratory Temporal Causal
Links and the Differences by Sport or
Lack Thereof. *Front. Physiol.* 10:45.
doi: 10.3389/fphys.2019.00045

Fitness level, fatigue and adaptation are important factors for determining the optimal training schedule and predicting future performance. We think that adding analysis of the mutual relationships between cardiac and respiratory activity enables better athlete profiling and feedback for improving training. Therefore, the main objectives were (1) to apply several methods for temporal causality analysis to cardiorespiratory data; (2) to establish causal links between the signals; and (3) to determine how parameterized connections differed across various subgroups. One hundred elite athletes (31 female) and a control group of 20 healthy students (6 female) took part in the study. All were asked to follow a protocol comprising two 5-min sessions of free breathing - once supine, once standing. The data were collected using Pneumonitor 2. Respiratory-related curves were obtained through impedance pneumography, along with a single-lead ECG. Several signals (e.g., tidal volume, instantaneous respiratory rate, and instantaneous heart rate) were derived and stored as: (1) raw data down-sampled to 25 Hz; (2) further down-sampled to 2.5 Hz; and (3) beat-by-beat sequences. Granger causality frameworks (pairwise-conditional, spectral or extended), along with Time Series Models with Independent Noise (TiMINo), were studied. The connections enabling the best distinctions were found using recursive feature elimination with a random forest kernel. Temporal causal links are the most evident between tidal volume and instantaneous heart rate signals. Predictions of the “effect” variable were improved by adding preceding “cause” samples, by medians of 20.3% for supine and 14.2% for standing body positions. Parameterized causal link structures and directions distinguish athletes from non-athletes with 83.3% accuracy on average. They may also be used to supplement standard analysis and enable classification into groups exhibiting different static and dynamic components during performance. Physiological markers of training may be extended to include cardiorespiratory data, and causality analysis may improve the resolution of training profiling and the precision of outcome prediction.

Keywords: granger causality framework, athlete training adaptation biomarker, cardiac function, tidal volume, elite athletes

1. INTRODUCTION

Comprehensive monitoring and testing of homeostatic processes, fitness level, fatigue, adaptation and recovery appears crucial for sports medicine practitioners to identify optimal training schedules, establish sufficient training loads and promote desirable progress and competitive performance (Meeusen et al., 2013; Halson, 2014; Coutts et al., 2017; Kellmann et al., 2018; Schneider et al., 2018).

The advance of medical devices and even wearable sensors makes it easy to quantify outputs. Training load indicators may come from the training set-up itself, from training equipment, accelerometers, etc. (Cardinale and Varley, 2017). The training schedule may be established objectively, and competitions produce a wealth of performance metrics.

The problem is in quantifying and analyzing the input information. Homeostasis is a capacious term. Fitness level and fatigue are largely subjective. Adaptation and recovery can be estimated, but usually only regarding a specific parameter. It appears there is no holistic framework (Heidari et al., 2018).

One of the commonly used methods in daily practice is cardiac monitoring (Buchheit, 2014; Schmitt et al., 2015; Bellenger et al., 2016; Duking et al., 2016; Giles et al., 2016; Plews et al., 2017). Average heart rate and many heart rate variability parameters have been proposed to describe the resting, exercise and recovery states of the heart, to assess the training load (Saboul et al., 2016), to evaluate high vagal activity (Nakamura et al., 2016), to predict performance (Triposkiadis et al., 2009), to test the heart activity changes induced by endurance and athletic activities (Berkoff et al., 2007; Vanderlei et al., 2008), and to analyze over-training syndrome (Dong, 2016) or training adaptation (Plews et al., 2013).

Still, there are many doubts about implementing heart activity parameters, due to various studies yielding discordant results, using different courses of analysis or even over-interpreting (Schneider et al., 2018). Therefore, separate heart activity data can be used only for a few aspects of sports medicine.

The concept of network physiology is widely accepted (Bartsch et al., 2015), as cardiac parameters may be influenced by many factors, e.g., environmental, anatomical, physiological, psychological, demographic, etc. (Sandercock et al., 2005; Fatissou et al., 2016). There is also no clear consensus as to which coefficients are best in training response evaluation (Sala et al., 2017).

One testable combination is that of breathing and heart activity. The relationship between heart rate and ventilation is well described, but still very complex. The effect of breathing phasing is usually apparent in resting ECG as sinus respiratory arrhythmia (Larsen et al., 2010; Shaffer et al., 2014; McCraty and Shaffer, 2015). The alternate cardiorespiratory coupling, in which heartbeats seem to coincide with specific respiratory phases due to increased sympathetic nervous activity and changes in arterial blood pressure, has also been tested (Penzel et al., 2016; Sobiech et al., 2017). On the other hand, the baroreflex seems to adjust neural responses and affect both heart and respiratory activity (Reyes del Paso et al., 2013).

Separate use of both signals would not reveal significant information nor improve study resolution. Several parameters,

like RMSSD, might be uninfluenced by tidal volume pattern, for both spontaneous and controlled breathing (Saboul et al., 2013), still being under the effect of respiratory rate (Schipke et al., 1999). Therefore, many methods of mutual signals analysis have been considered. Assessed frameworks have included time-, frequency- or information-domain parameterization; temporal, phase or causal relations; etc. (Jamšek et al., 2004; Lopes et al., 2011; Riedl et al., 2014; Gasior et al., 2016; Javorka et al., 2016; Müller et al., 2016; Kuhnhold et al., 2017; Sobiech et al., 2017; Wejer et al., 2017).

In our previous work, we tried to add another domain of interest, time-independent causality (Młyńczak and Krysztofiak, 2018). Our discoveries suggested different paths for lying supine (from tidal volume, through heart activity variation and average heart activity, to respiratory timing) than for standing (from normalized respiratory activity variation to average heart activity).

In a traditional approach, such a graph of connections is treated as an input. We proposed a different context: from a graph, one can indicate which interventions (changes in training schedule) may be applied to expect specific results. This is related to the “bottom-up” strategy of thinking about designing the training based on the optimal parameter that describes and shows adaptation status, not the inverse (Młyńczak and Krysztofiak, 2018).

Going into greater detail, the next step is temporal causality analysis. Granger-based causality or transfer entropy are the two most important methods (Faes et al., 2013; Porta et al., 2017; Valenza et al., 2018). In general, regardless of the details in various formulations, they may cover different aspects of relations between two time series, except for linear Gaussian processes, when they can be considered equivalent (Porta and Faes, 2016). The novel frameworks also include the generalization of the basic concept, in which the effect of zero-lag can also be considered and evaluated. Causal inference related to Time Series using Restricted Structural Equation Models (TiMINo) has also been introduced (Peters et al., 2013).

As the methods mentioned above can produce some parameters and indices, we hypothesize that they can be used to improve athlete profiles and analyze trends during a training period.

Therefore, the main objectives were:

- to apply several methods for temporal causality analysis to cardiorespiratory data;
- to establish causal links between the collected cardiac and respiratory signals; and
- to determine how parameterized connections differed across groups depending on the static and dynamic activity component during performance, or control.

2. MATERIALS AND METHODS

2.1. Subjects and Device

A group of 100 elite athletes practicing different sports (31 female; mostly overlapping with the previous paper (Młyńczak and Krysztofiak, 2018), the difference in 5 subjects is coming from the need to ensure the stationarity of the signal for sufficient

temporal causality analysis) and 20 healthy students (treated as a control group; 6 female) took part in the study, carried out at the National Centre for Sports Medicine in Warsaw during the routine periodic health evaluation and medical monitoring program, 3–4 months before the 2016 Olympic Games in Rio de Janeiro.

The sport types are defined according to Mitchell et al. (2005), where numbers refer to the static component of heart activity expressed as % of its maximal voluntary contraction (MVC):

- Low (I-29 subjects);
- Medium (II-42); and
- High (III-29);

and letters to the dynamic component (e.g., % of VO_{2max}) occurring during competition:

- Low (A-5 subjects);
- Medium (B-45); and
- High (C-50).

The demographic descriptive statistics of the athletes are summarized in **Table 1**.

The procedure was approved by the Ethics Committee of Warsaw Medical University (permission AKBE/74/17). All participants were informed about the general aim of the measurements, and each athlete had previously signed a general consent form for the routine medical monitoring (consisting of a statement of acceptance of the use of the results for scientific purposes). The students provided separate written consent.

The data were collected using our device, Pneumonitor 2 (Młyńczak et al., 2017). This is the academically-developed prototype, intended for conducting research and teaching. Respiratory-related curves were obtained through impedance pneumography, along with a single-lead ECG (lead 2). The impedance data were measured using the tetrapolar method, with the electrode configuration proposed by Seppa et al. (2013). Standard Holter-type, disposable ECG electrodes were used. Both signals were sampled at 250 Hz. Task Force (1996) stated it is the smallest sufficient in terms of R peak finding and heart rate variability analysis and over-sampled from a respiratory perspective.

2.2. Protocol and Pre-processing

The measurements were performed in a diagnostic room designated for cardiological examinations. All participants were asked to follow a protocol comprising:

- attachment of the electrodes,
- 10-min stabilization phase,
- 5-min session of spontaneous breathing while lying supine, and
- 5-min session of spontaneous breathing while standing.

None knew the impact of breathing and position on the study outcomes. The protocol is inspired by the orthostatic maneuver; however, we did not take into account several sub-periods (adaptation, recovery, etc.), but rather performed the analysis for the entire supine and standing segments.

ECG signals were processed by non-linear detrending for baseline alignment. Consecutive R peaks in each signal were then found based on the Pan-Tompkins algorithm. On the respiratory side, raw IP signals were first pre-processed by smoothing with a 1 s averaging window (Młyńczak and Cybulski, 2017). Then, inspiratory and expiratory phases were detected from the differentiated, flow-related signal. We did not transform impedance into the volume, instead assuming that impedance changes reproduce the tidal volume signal in terms of shape (linear fitting provides the best agreement between IP and the reference, pneumotachometry) (Młyńczak et al., 2015). Therefore, no calibration was performed.

Next, three kinds of dataset (to assess the reliability of several temporal causality methods when applied to cardiorespiratory data) were calculated for each participant in each body position:

1. tidal volume (TV) + instantaneous respiratory rate (iRR) + and instantaneous heart rate (iHR), all down-sampled to 25 Hz (for the temporal causal analysis of signals);
2. the same data further down-sampled to 2.5 Hz (for the spectral causal analysis of signals); and
3. lengths of consecutive RR intervals + TV-related impedance amplitude + iRR + breathing phase, the last three measured at the R peaks (for the causal analysis of beat-by-beat sequences; 1 is an arbitrary value standing for inspiratory phase, -1 for expiratory, and 0 for pause).

The iRR was calculated by estimating intervals between consecutive inspiratory onsets, then interpolating to the initial sampling frequency. In the same manner, the iHR was calculated by estimating intervals between successive R peaks, then interpolating to fit the number of samples for TV data.

The first down-sampling was intended to reduce computational complexity (deep embedding in terms of samples), the second - to analyze the proper sub-range of frequencies. Both were accompanied by suitable low-pass filtering. All analyses were carried out in MATLAB.

The stationarity analyses were performed on the signals from the first dataset using augmented Dickey-Fuller Test (in R, using *tseries* package, Trapletti and Hornik, 2018). As the null hypothesis is that the time series has a unit root, we considered the data to be stationary when $p < 0.05$.

2.3. Causality Analysis

Four methods of causality analysis were studied:

- Granger causality;
- Spectral Granger causality;
- Extended Granger causality; and
- Time Series Models with Independent Noise (TiMINo).

First three techniques are based on the Granger concept of causality intended for time series. The main idea of this approach is that X can be treated as a “cause” of Y if taking previous X values along with Y ones enables preparing the model, which predicts the next Y values better than by only taking previous Y values. The efficiency of the prediction can be parameterized by the variance of the differences between predicted and actual Y values. So-called G-causality combines the variances of two

TABLE 1 | The demographic summary of the study group; the sports types are defined according to Mitchell et al. (2005) and the description in Materials and Methods.

Group (sport type)	N		Height [cm]				Body mass [kg]			
	Female	Male	Min	Mean	SD	Max	Min	Mean	SD	Max
Control	6	14	157.0	176.0	7.5	186.0	52.0	66.5	9.9	91.0
IIIA	0	5	168.0	176.8	9.2	189.0	68.0	80.8	10.2	95.0
IB	4	20	170.0	193.5	11.2	208.0	61.0	82.9	12.9	104.0
IIB	7	2	167.0	174.7	9.4	193.0	55.0	65.0	15.0	98.0
IIIB	4	8	158.0	174.3	12.0	197.0	53.0	79.8	32.6	151.0
IC	1	4	169.0	176.0	9.3	190.0	55.0	71.8	13.0	85.0
IIC	11	22	162.0	185.3	12.8	207.0	49.0	81.2	17.9	115.0
IIIC	4	8	171.0	179.5	6.4	189.0	63.0	75.4	6.3	88.0

models with or without including X, and can be calculated with the equation (1):

$$GC_{x \rightarrow y, p} = \ln \left(\frac{\text{Var}(\hat{y} \parallel y, p)}{\text{Var}(\hat{y} \parallel y + x, p)} \right) \quad (1)$$

where p is the model order, and the arrow presents tested direction.

It appears that the G-causality parameter has F-distribution and the statistical test can be proposed to assess the significance of the predictability improvement when using X along with Y. When p -value is lower than an arbitrarily adopted threshold at the level of 0.05, X can be considered to be a “Granger-cause” of Y (Granger, 1980; Barnett and Seth, 2014).

The first approach, pairwise-conditional one (because of multivariate data), was applied to the first dataset and based on the MVGC framework (Barnett and Seth, 2014). It uses VAR modeling, and the best model order is chosen based on the automatically-established Bayesian Information Criterion (BIC), as presented in the **Supplementary Material S1**.

The second approach, pairwise-conditional spectral analysis, was applied to the second dataset and also uses MVGC framework (Barnett and Seth, 2014). The possibility of Granger analysis in the frequency domain is based on the concept of cross-power spectral density decomposition and utilizing the generalized definition of G-causality. The process (and also the justification of the usage of F-statistics for a conditional case) is described in detail in Geweke (1982) and Barnett and Seth (2014).

The third approach, extended Granger causality framework, is built on the same concept; however, differently as in the original case, it also takes into account zero-lag, instantaneous effects. This assumption came from physiological analyses, when phenomena take place within the same cycle, for cardiorespiratory conditions mostly affected by the parasympathetic system. The implementation is presented in detail by Schiatti et al. (2015). We applied the method to the third dataset, with an arbitrarily-established maximum lag of 4 (as R peak is a trigger it usually covers from the middle to the full respiratory cycle).

The last approach, Time Series Models with Independent Noise (TiMINo), is based on generalized additive models (GAMs) as an extension of the Structural Equation Model

framework to time series data (Peters et al., 2013). Not like it is in the Granger definition, which exploits the residual variance, TiMINo models require independent residual time series. Another aspect is the model class restriction to additive noise ones. As described by Peters et al. (2013) both lagged and instantaneous effects can be found and so-called unfaithful feedbacks between the time series may be also deduced. Peters et al. (2013) showed that when the data are causally insufficient or the proposed model is misspecified, this method will avoid incorrect answers. Additionally, this method is not built on the asymmetry of time direction but rather considers identifiability emerging from restricted structural equation models (Peters et al., 2013). Similarly, this approach was also applied to the third dataset, with a maximum lag of 4.

Then, we calculated the G-causality values with the first and third methods and determined the peak amplitude and frequency of spectral G-causality with the second method. All values were stored only when statistically significant. Granger causality methods were implemented in MATLAB; to ensure reproducibility, the code used to calculate G-causalities from a hypothetical dataset, along with accompanying p -values, is provided as **Supplementary Material S1**. The G-causality values, along with demographic and descriptive information about the subjects, are provided as **Supplementary Material S2**.

From the G-causality, one can calculate the prediction improvement in the case where the cause variable is assumed in the model, based on Equation (2):

$$\text{Prediction Improvement} = 100 \cdot e^{G\text{-causality}} - 100 \quad [\%] \quad (2)$$

The fourth method, TiMINo, was used to view the problem in a completely different context. We established causal links for the entire athlete and control groups and separately for both body positions, according to the TiMINo results.

The quantitative data (from the first three methods and from each connection, except for breathing phases and tidal volume for the third approach, which are dependent) enabling the best distinction between groups and between different competitive levels of activity components were found using Recursive Feature Elimination (RFE) with a random forest algorithm used to produce a model and estimate performance (10-fold cross-validation was implemented). The exploratory accuracy and

Cohen's Kappa (without dividing the data into training and testing subsets) were calculated.

TiMINo, exploratory statistical analysis, and RFE were performed in R (R Core Team, 2018). The relevant code is provided as **Supplementary Material S3**.

The entire flow of the analysis is presented in **Figure 1**.

3. RESULTS

3.1. General Findings

Granger causality analysis is originally intended for stationary data, therefore augmented Dickey-Fuller stationarity analysis was performed. It showed that:

- 2 TV-related signals during supine body position,
- 4 TV-related signals during standing body position,

- 1 iHR signal during supine body position, and
- 4 iHR signals during standing body position

can be labeled as non-stationary; however, it occurred jointly in no case.

The sample signals acquired for both supine and standing body positions were presented in the **Figure 2**.

All the Granger-based methods revealed that iHR seems to cause TV. This can be due to the applied convention that inspiration causes the respiratory-related signal to increase, and expiration to decrease. Therefore, at the instantaneous peak of heart rate during inspiration, the phase of iHR seems to precede that of TV. This is further debated in the Discussion section.

Apart from the spectral analysis, iRR appeared causally independent of iHR and TV, which may suggest that the heart activity is more related to the depth of breathing than to the rate.

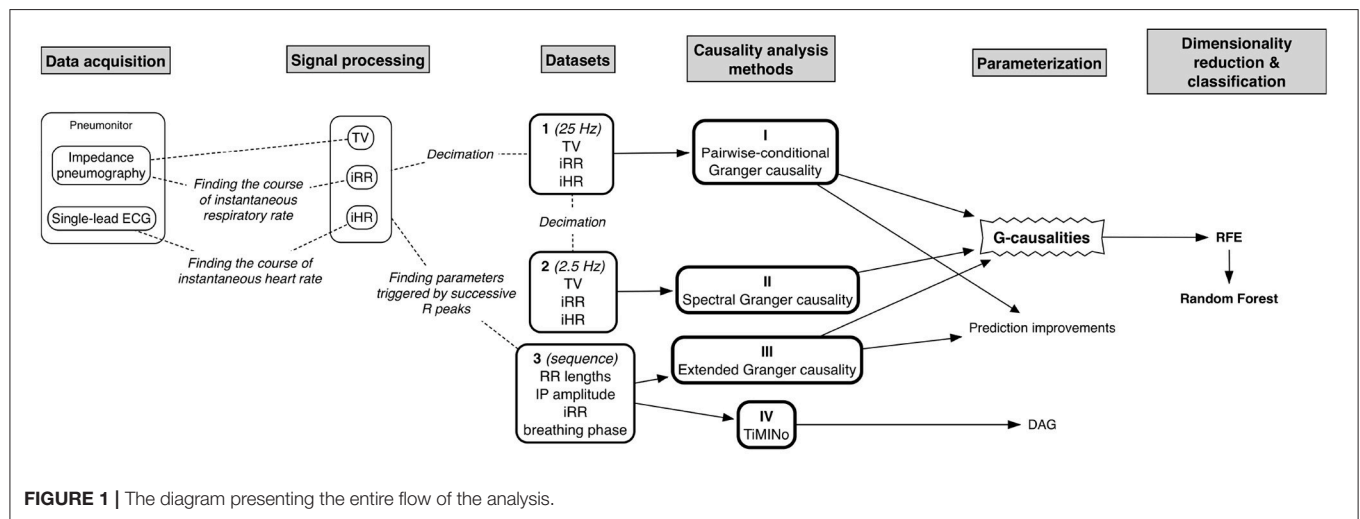


FIGURE 1 | The diagram presenting the entire flow of the analysis.

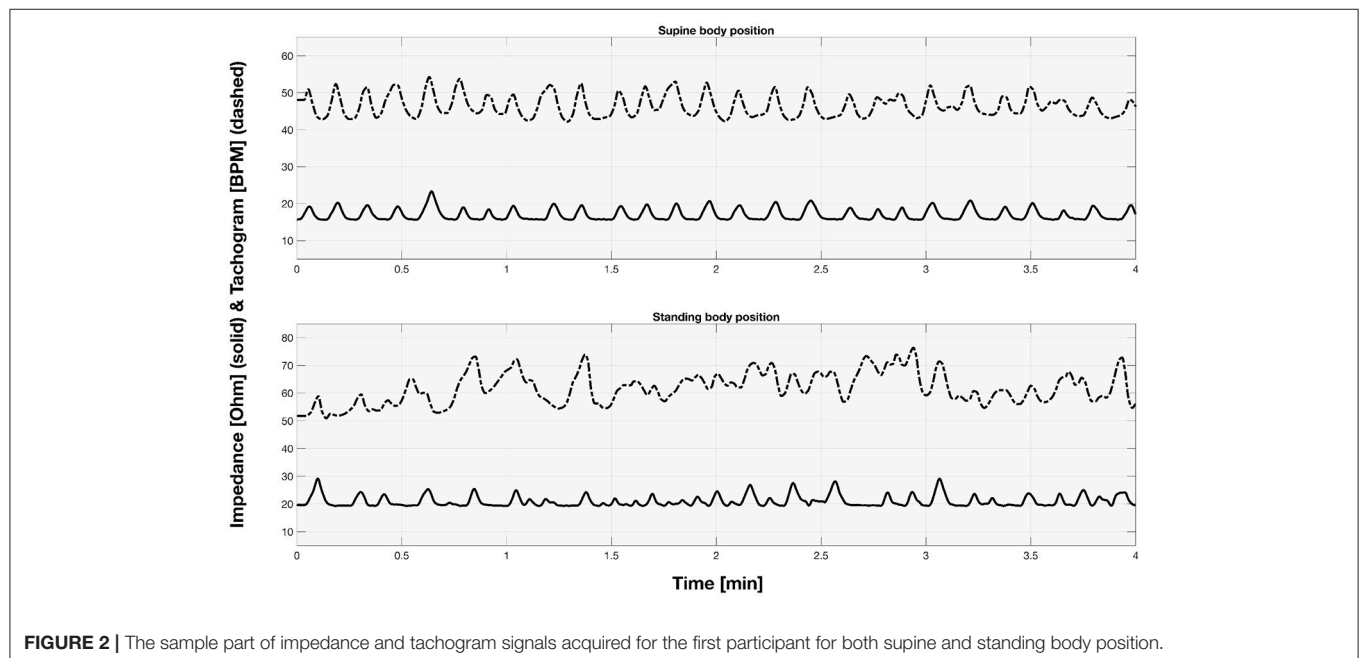


FIGURE 2 | The sample part of impedance and tachogram signals acquired for the first participant for both supine and standing body position.

3.2. Pairwise-Conditional Granger Causality

The first method showed iHR causing TV changes, more for the supine body position (2.5% median prediction improvement; only 11 results out of 120 were statistically insignificant) than for standing (median of 1.7%; 20 insignificant results). The summary is stored in **Table 2**. For supine, the median G-causality for the athletes was greater than for the control group, but insignificantly so ($p = 0.33$ for the Wilcoxon rank test). The medians increased slightly, still insignificantly, for sports types with greater dynamic components (**Figure 3**; $p = 0.81$ for the Kruskal-Wallis test). For standing, the medians increased insignificantly for sports types with greater static components (**Figure 4**; $p = 0.12$ for Kruskal-Wallis). It is worth noting that the modest improvements are mainly caused by a relatively high sampling rate (from the perspective of the Granger causality framework). Inducing Granger causality appears quite insufficient when the sampling rate is so high compared to the physiological activity changes.

3.3. Pairwise-Conditional Spectral Granger Causality

The median peak frequency at which iHR influences TV was identified as 0.23 Hz for lying supine (median G-causality equaled 0.82, vs. 0.27 for the opposite direction, only 3 results statistically insignificant) and 0.19 Hz for standing (median G-causality of 0.68, vs. 0.14 for the opposite, 15 results insignificant). The second method also showed that iHR and TV seem to cause iRR at 0.04 Hz on average, for both supine and standing. All directions are summarized in **Table 3**.

3.4. Extended Granger Causality

The third method confirmed the direction to be from the lengths of successive RR intervals to the amplitudes of impedances at the R peaks. The median prediction improvement was 20.3% for supine (only 5 insignificant) and 14.2% for standing (also 5 insignificant). The summary appears in **Table 4**. For supine, the median G-causality value for the sports group was greater than for the control ($p = 0.042$ for Wilcoxon). Medians increased

insignificantly for sports types with greater static components (**Figure 5**; $p = 0.86$ for Kruskal-Wallis). For standing, the median G-causality value for the sports group was greater than for the control ($p = 0.042$ for Wilcoxon). Medians increased significantly for sport types with greater static components (**Figure 6**; $p = 0.028$ for Kruskal-Wallis). Pair-wise, *post-hoc* Wilcoxon rank test indicated that significant differences appear between mild- and high-static-component groups ($p = 0.048$), and between moderate- and high-static-component groups ($p = 0.048$).

The level of prediction improvement for the third approach, compared to the first, shows that Granger causality is more reliable for physiological beat-by-beat parameterizations than for raw signals sampled at 25 Hz.

3.5. TiMINo

As expected for physiological data, many results of TiMINo analysis were left unidentified due to unfulfilled model

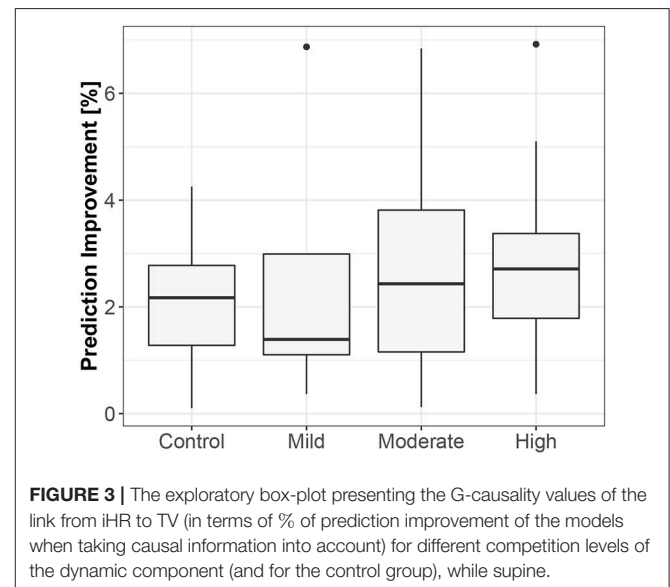


TABLE 2 | The summary of prediction improvements (PI) for all considered directions for the first approach (raw signals sampled at 25 Hz): 1. TV; 2. iRR; 3. iHR; NA, not assigned, if statistically insignificant (the more NAs, the more uncertain the link).

Body position	Link	Mean PI	SD PI	Median PI	IQR PI	NA count
Supine	1 → 2	0.24	0.12	0.19	0.19	69
	1 → 3	0.84	1.80	0.55	0.47	29
	2 → 3	0.58	0.43	0.41	0.60	30
	2 → 1	0.37	0.36	0.25	0.33	63
	3 → 1	2.57	1.48	2.53	2.03	11
	3 → 2	0.25	0.12	0.20	0.18	80
Standing	1 → 2	0.28	0.16	0.24	0.13	69
	1 → 3	0.47	0.40	0.37	0.38	46
	2 → 3	0.64	0.45	0.49	0.58	36
	2 → 1	0.45	0.33	0.34	0.31	58
	3 → 1	1.70	1.28	1.67	1.51	20
	3 → 2	0.21	0.10	0.17	0.10	99

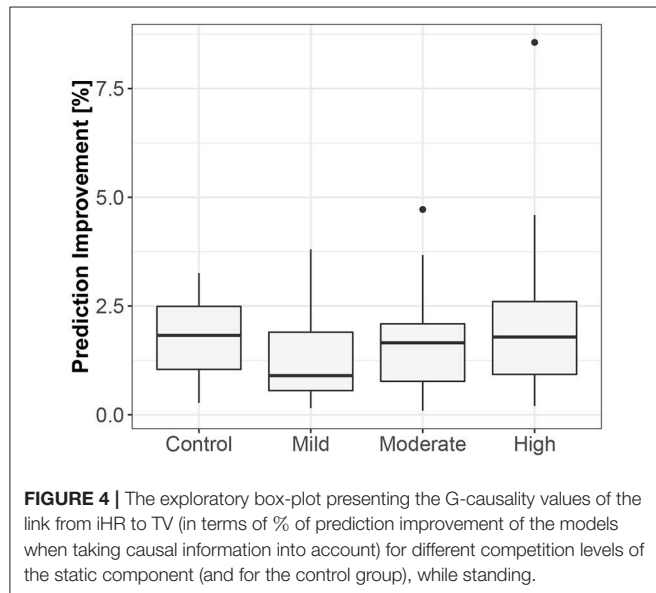
Bold values represent the most prominent and discussed link in the text.

assumptions (independence of residuals or model complexity too high for the amount of data available). The outputs suggested mostly that breathing phases cause the changes in lengths of consecutive RR intervals (29% for supine athletes, 21% for standing athletes, 40% for supine controls and 20% for standing controls). In this context, TiMINo seems to favor the respiratory sinus arrhythmia effect, no matter which convention of respiratory curve presentation is applied. Other connections are present, but only for a few cases, and only for athletes. The graphical summary is presented in **Figure 7**.

3.6. Distinguishing Sports or Lack Thereof

Recursive feature elimination suggested that 14 variables form the best set for distinguishing athletes and non-athletes ($83.3 \pm 3.9\%$ accuracy). The top five of these are:

- frequency at the peak of G-causality (2nd approach) from iHR to TV, while standing;



- G-causality value from the 1st approach, from TV to iRR, while standing;
- G-causality value from the 3rd approach, from lengths of consecutive RR intervals to breathing phases at R peaks, while standing;
- G-causality value from the 3rd approach, from lengths of consecutive RR intervals to TV-related impedance amplitudes at R peaks, while standing; and
- G-causality value from the 3rd approach, from lengths of consecutive RR intervals to TV-related impedance amplitudes at R peaks, while supine.

Another set was identified as the best for differentiating between moderate and high dynamic components during competition (we neglected low-dynamic-component-participants as there were only 5 subjects in this group ($63.1 \pm 14.6\%$ accuracy):

- frequency at the peak of G-causality (2nd approach) from iRR to iHR, while standing;
- G-causality value from the 3rd approach, from iRR at R peaks to breathing phases at R peaks, while supine;
- G-causality value from the 3rd approach, from iRR at R peaks to the breathing phase at R peaks, while standing;
- G-causality value from the 3rd approach, from TV-related impedance amplitude at R peaks to lengths of consecutive RR intervals, while standing;
- G-causality value from the 1st approach, from iRR to iHR, while standing; and
- G-causality value from the 1st approach, from iHR to TV, while supine.

Finally, a set of 30 variables appears best for analysis of various levels of the static component ($51.8 \pm 12.3\%$ accuracy), of which the top five are:

- G-causality value from the 3rd approach, from lengths of consecutive RR intervals to breathing phases at R peaks, while supine;
- G-causality value from the 3rd approach, from breathing phases at R peaks to iRR at R peaks, while supine;

TABLE 3 | The summary of medians and IQRs for peak G-causality and frequency for all considered directions for the second, spectral approach (raw signals sampled at 2.5 Hz); 1. TV; 2. iRR; 3. iHR; G, G-causality; f, frequency in Hz; NA, not assigned, if statistically insignificant (the more NAs, the more uncertain the link).

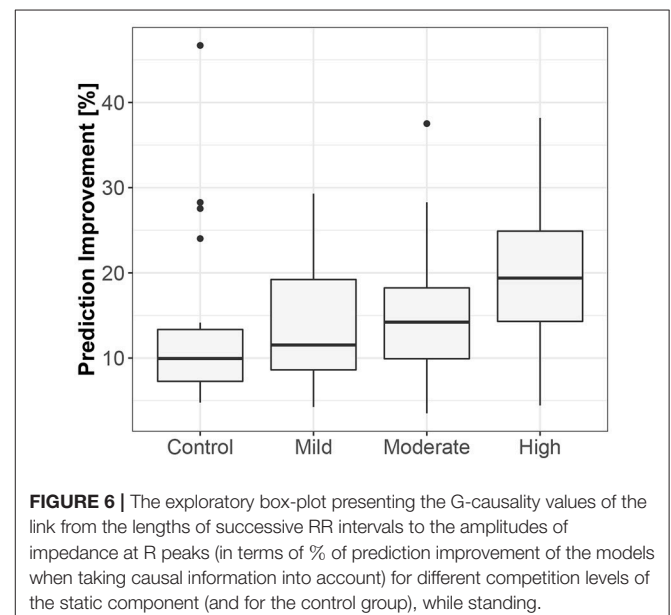
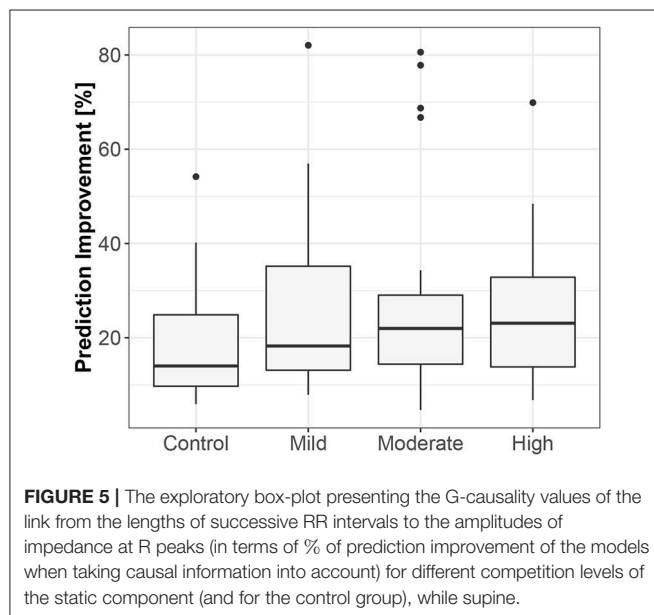
Body position	Link	Median G	IQR G	Median f	IQR f	NA count
Supine	1 → 2	0.50	0.71	0.04	0.01	2
	1 → 3	0.27	0.37	0.10	0.06	2
	2 → 3	0.10	0.14	0.07	0.09	28
	2 → 1	0.04	0.04	0.21	0.12	63
	3 → 1	0.82	0.65	0.23	0.12	3
	3 → 2	0.23	0.49	0.04	0.01	7
Standing	1 → 2	0.45	0.65	0.04	0.02	4
	1 → 3	0.14	0.20	0.06	0.04	15
	2 → 3	0.14	0.20	0.07	0.03	15
	2 → 1	0.06	0.06	0.18	0.08	40
	3 → 1	0.68	0.68	0.19	0.09	3
	3 → 2	0.22	0.31	0.04	0.02	12

Bold values represent the most prominent and discussed link in the text.

TABLE 4 | The summary of prediction improvements (PI) for all considered directions for the third approach (beat-by-beat sequences); 1. Lengths of consecutive RR intervals; 2. TV-related impedance amplitude at R peaks; 3. iRR at R peaks; 4. Breathing phases at R peaks; NA, not assigned, if statistically insignificant (the more NAs, the more uncertain the link).

Body position	Link	Mean PI	SD PI	Median PI	IQR PI	NA count
Supine	1 → 2	24.44	16.48	20.33	17.32	5
	1 → 3	7.41	6.96	5.92	2.49	92
	1 → 4	16.71	14.09	11.34	14.08	29
	2 → 3	9.32	7.73	7.05	4.41	88
	2 → 4	69.15	39.76	56.99	54.43	2
	3 → 4	6.06	3.35	4.58	4.53	102
	2 → 1	19.42	25.74	12.60	11.81	40
	3 → 1	7.51	4.21	6.52	5.54	97
	4 → 1	11.58	8.95	8.16	7.04	66
	3 → 2	6.22	2.58	5.66	2.56	90
	4 → 2	34.50	21.88	33.56	26.08	9
	4 → 3	7.43	5.95	5.61	2.37	78
Standing	1 → 2	15.37	8.40	14.18	12.15	5
	1 → 3	4.34	2.81	3.49	1.89	105
	1 → 4	10.36	9.72	7.24	6.87	54
	2 → 3	6.63	3.28	4.90	3.85	89
	2 → 4	68.28	32.13	61.62	43.59	0
	3 → 4	8.05	8.69	4.84	3.88	103
	2 → 1	10.11	9.75	6.75	6.92	51
	3 → 1	10.91	12.58	5.72	9.61	97
	4 → 1	8.07	6.58	5.72	4.56	74
	3 → 2	5.85	3.58	4.18	3.82	100
	4 → 2	31.62	22.67	30.53	31.71	7
	4 → 3	5.73	3.22	4.40	3.52	93

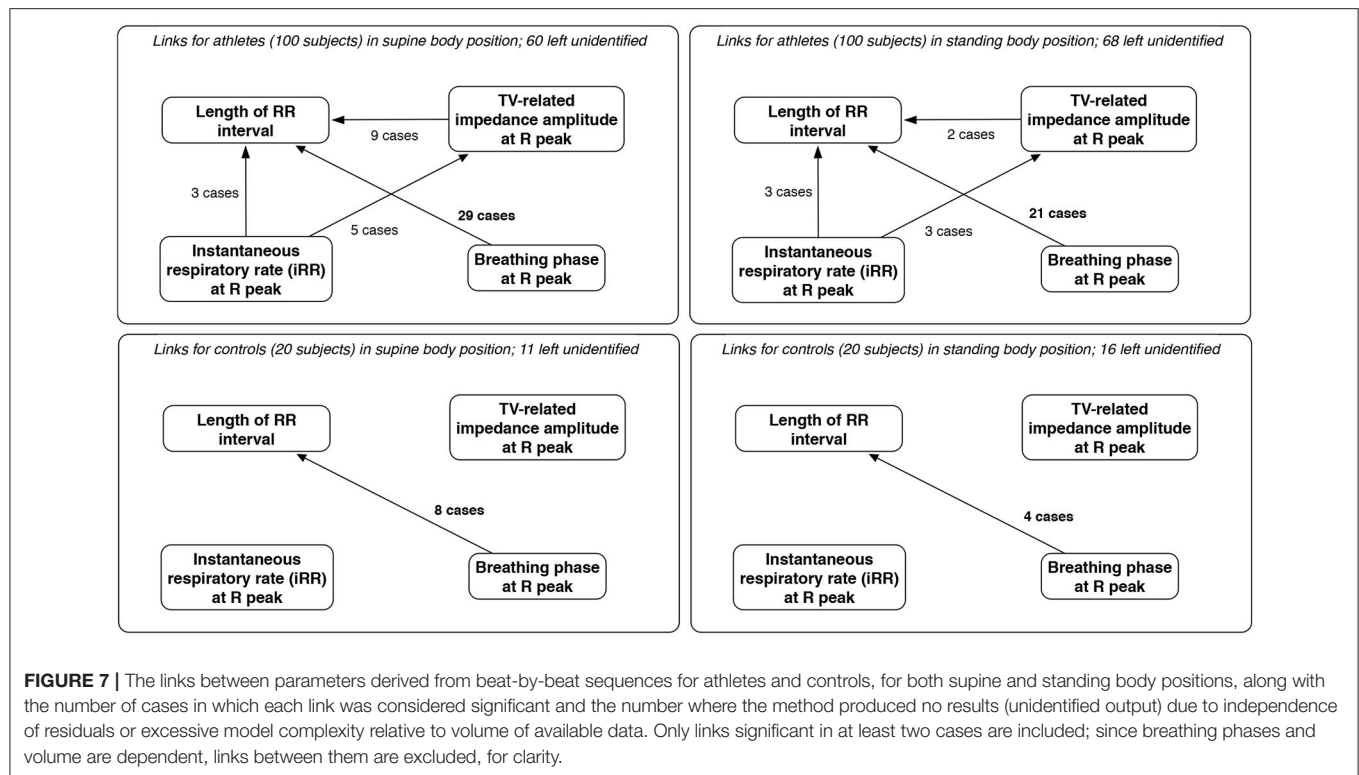
Very high numbers for the connections between 2 and 4 are ignored, due to their intrinsic relationship. Bold values represent the most prominent and discussed link in the text.



- peak amplitude of G-causality (2nd approach) from iHR to iRR, while standing;
- peak amplitude of G-causality (2nd approach) from iRR to iHR, while standing; and
- frequency at the peak of G-causality (2nd approach) from iRR to TV, while supine.

4. DISCUSSION

The main finding of our analysis is that the instantaneous heart rate (iHR) signal is causally related to the tidal volume (TV) signal. The Granger methods showed that iHR caused TV



changes, in both time and spectral domains and for both raw signals and beat-by-beat sequences.

As it is incoherent with the respiratory sinus arrhythmia effect (which is expected to have the largest impact, particularly during static supine; respiratory centers modulate the frequency of the heart through the vagal sinus node intervention (Eckberg, 2009) and the observations being the basis of the clinical autonomic screening tests (CARTs), we hypothesize that this is mainly due to the definition of the Granger causality—the cause should be before its effect. Relatively similar signal shape for RR intervals seems to appear before tidal volume, as the heart rate peak occurs during inspiration. In other words, the phase of iHR seems to precede that of TV when inspiration is presented as an increase of the signal's value. A visualization of this remark is presented in **Figure 8**.

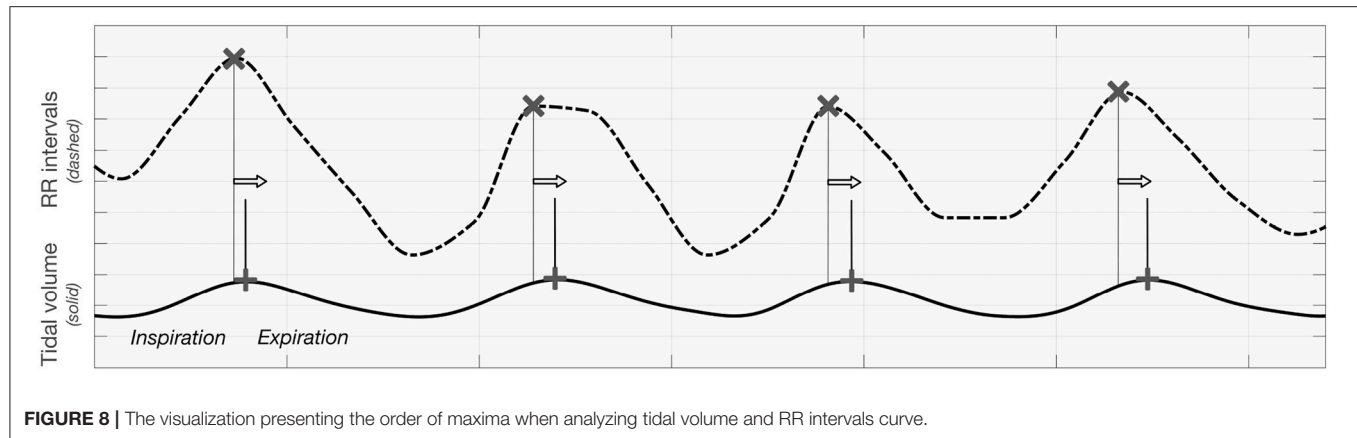
Another method, Time Series Models with Independent Noise (TiMINo), suggested differently that breathing phases recorded at consecutive R peaks cause the lengths of RR intervals. As from the definition it is not based on the asymmetry of time, the RSA phenomenon is emphasized and confirmed (Shaffer et al., 2014; McCraty and Shaffer, 2015). An opposite effect, described by Sobiech et al. (2017), in which R peaks occur at strict intervals before the inspiratory onset, requires different mathematical approaches and different data preparation process.

The concept of analyzing both signals and sequences of parameters triggered by the heart activity with Granger methods is considered as a “test” of information capacity that can be extracted. The first approach is therefore inspired by recurrent, or even convolutional, deep learning methods, in which raw time

series can be used as an input for the analysis. The results showed, however, that for signal-related analysis high computational complexity may distort the final inference. This is also most likely due to the redundancy that complicates the VAR modeling. Apart from that, the spectral analysis seems more proper when the intervals between the samples are equal.

We supposed that said relationships can be quantitatively parameterized to add extended data for sports training scheduling and monitoring. Koenig and Thayer (2016) found substantial differences between sexes regarding autonomic control of the heart. So why not explore the possible differences across sports, the levels of specific components during competition, static and dynamic conditions, body positions, etc.?

Therefore, we believe causality analysis may become a valuable and practical tool for trainers and physicians, as a method that not only finds or confirms causal connections and their directions, but can also parameterize them. As Schneider et al. (2018) wrote, training context is key. In our opinion, there are two steps to unlocking its promises. The first is to supplement the standard heart activity parameters in order to assess trends and competitive performance. Greenham et al. (2018) in their meta-analysis summarized that many biological and biochemical biomarkers varied with training intensity but not with performance. Several, like neutrophils, glutamine, urea and the testosterone/cortisol ratio, may be used to track performance. However, such approaches need not be individual. Cardiorespiratory data and analysis could be an objective addition, even more robust when combined with causality



inference (it should not be used independently when the reported accuracies remain far from sufficient). This should be studied further.

The second is to use newfound knowledge of causal structures to better predict the effects of interventions (establishment of sufficient training load) to determine optimal training schedules (Pearl, 2010). This suggests the need to consider activity measures from both systems together. All may be performed inside or outside the laboratory; our prototype, Pneumonitor 2 (which can measure changes in thoracic impedance, which is related to changes in the amount of air in the lungs, Młyńczak et al., 2017), appears appropriate for that task, because the results suggested that the depth of breathing is more important than its rate.

This is also indirectly connected with the statement of Fossion et al. (2018), that homeostasis may be quantified using time-series analysis, which might offer several explanations for physiological mechanisms. Use of a portable device allows accounting for various conditions, e.g., body positions. From the analysis, we discovered that the relations between cardiac and respiratory parameters are quite similar for both analyzed body positions. However, the prediction improvement associated with adding a cause parameter was lower for standing than for supine (which is expectable, but can still serve as an additional input). We did not note the effect described by Radovanovic et al. (2018), who reported that even a slight change of body position may change the direction of the cardiorespiratory relationships; however, it merits further study.

Another interesting outcome is the frequency for which the peak G-causality value occurred: about once every 5 s. This is longer than a typical cardiac interval, but could be related to mean breathing rate. This would show a mechanism in which the frequency of breathing serves as a trigger for the causal process even if the depth is more directly related with changes in RR intervals.

Also, as many effects may occur too quickly for the measurement system to track at current time resolution, considering zero-lag elements in the model is in our opinion a crucial step in similar physiological research. This is why (Schiatti et al., 2015) introduced the apparatus for extended Granger causality. Beyond the traditional set of connections, a matrix

of instantaneous effects may be prepared. In the analysis, the framework showed the significance of zero-lag effects, not only between durations and amplitudes but also between durations and breathing phases at R peaks (this was not explicitly presented in the text for clarity). The approach seems to also be tied to the kernel regression criteria, which can be exploited for causality inference (Zheng et al., 2012; Vinod, 2016).

Another commonly used measure to assess direction-sensitive connections is the transfer entropy, which can consider equivalent to Granger causality approach for linear Gaussian processes (Schreiber, 2000; Porta and Faes, 2016). The simple implementation of the method based on mutual information distance or generalized correlation sum is written in *TransferEntropy* R package (Mount et al., 2016). Similar to extended Granger causality, the concept was presented earlier by Faes et al. (2013), who introduced so-called compensated transfer entropy, which also includes zero-lags elements into consideration. Instantaneous transfer entropy has also been already used by Valenza et al. (2018) for physiological analyses. Nevertheless, we decided not to incorporate the method in the analysis as there is no output value for transfer entropy such as prediction improvement or *p*-value, that can be easy to interpret for physicians.

4.1. Limitations of the Study

The study included only 100 athletes, who formed a heterogeneous group, unevenly distributed in relation to the Mitchell et al. (2005) division. All were studied in the “hot period” 3–4 months before the Olympic Games, which may suggest a state of over-training. Therefore, the findings should be compared with another similar procedure.

As cardiorespiratory parameters and relations (in general) are affected by a number of factors, e.g., age, gender, and levels of physiological or psychological stress, they might have been considered as both confounders and even direct cause variables (Schulz et al., 2015; Widjaja et al., 2015). However, we decided not to include them for clarity and due to the lack of psychologically-oriented questionnaires gathered from athletes. Also, the control group did not answer any questions, so it is not possible to evaluate the differences in their physical preparation

relative to the athletes (beyond the fact that no students reported professional participation in sports).

Also, the R peak locations are determined at the 250 Hz sampling frequency, so the uncertainty in the location of the R peaks is of 4 ms, which may affect the estimates of G-causality computed using such a tachogram.

The collection of only one observation per subject precludes reproducibility analysis. Also, measurements were carried out for a single protocol in an atypical environment. The results of registrations performed outside the laboratory, during normal training, or even with 24 h Holter-based tracking, would yield more condensed and more general findings.

Moreover, the classical Granger causality framework is a linear approach. Several nonlinear generalizations would better fit cardiorespiratory signal specifications. Segments of registrations where analyzed whole, so only a single coefficient was estimated per segment.

Accuracy analysis of the use of causal parameters to distinguish groups was illustrative, not conclusive: there are too little data and the groups are unbalanced. However, one can conclude, those causal parameters may be treated as additional information to standard parameterization, where the accuracies are too weak for them to be used independently.

We did not assume any control variables in the protocol. As this is a retrospective study, we cannot change the respiratory protocol after registration, and also we cannot introduce any interventions to evaluate its effect).

Additionally, our modeling is based on the signals, which cover end-organ responses modulated by multiple levels of complex mechanisms (Dampney, 2015). However, in this study, we attempted a data-driven approach, without including prior knowledge (Młyńczak and Krysztofiak, 2018). Also, one should be aware of the very possible collinearities between X and Y in the original Granger's formulation. This is why we tried to analyze both directions of possible connections and a possible reason for why the causal structure strengths were estimated as mild.

Also, the analysis of two body positions (particularly standing) without segmenting the signals into sub-periods may cause that the results do not cover our mechanisms and processes, and their changes in time. Our choice is, however, dictated by the need to maintain the appropriate signal length for Granger causality approaches.

Finally, we focused only on the data types which can be registered using Pneumonitor 2. However, as the presented protocol was "static," adding different modalities appears relatively simple. For example, Sobiech et al. (2017) suggested that arterial blood pressure is probably the driver (cause) of both cardiac and respiratory function.

4.2. Considerations for Further Studies

The discussion identified several issues for further study:

- how would the accuracy of athletes profiling increase with the addition of causal parameters to standard cardiorespiratory data?
- how would causal links and their strengths differ during natural activity of the subject?

- how coherent are causal parameters for a specific participant in comparable conditions? (reproducibility analysis)
- how would adding restricted breathing to the protocol affect the causal parameters?
- how would the causal links be changed or emphasized with the addition of an arterial blood pressure signal (Silvani et al., 2017; Zhang et al., 2017)?
- can the causal analysis be made more specific and more robust (free or insensitive to collinearity) with model terms conditioned on covariates or even with the addition of a non-linear kernel of Granger-like analysis?
- can the DAG structures be confirmed with a prospective study, which assumes sufficient perturbations and interventions on the cause variable?

One could also evaluate different methods, e.g., based on directional coherence analysis. Schäck et al. (2018) proposed a novel method, robust time-varying generalized partial directed coherence (rTV-gPDC), which carries information about the non-linear connectivity structure using a piecewise linear time-varying moving-average (TVMA) model. It is worth investigating in the presented contexts because the approach assumes a model which is non-linear and which, even more importantly, may adapt over the course of measurement.

5. CONCLUSIONS

Physiological markers of training performance may be not only biochemical- or cardiac-, but also cardiorespiratory-related. Besides temporal-, spectral-, or information-domain approaches, causal link analysis using pairwise-conditional standard, spectral or extended Granger causality or TiMINo frameworks may introduce new contexts, make the inference more robust and improve result resolution.

We proposed a protocol for elite athletes and controls inspired by the orthostatic maneuver, consisting of free breathing while resting supine and while standing, and took into account various forms of registered data: (1) raw signals sampled at 25 Hz, (2) raw signals sampled at 2.5 Hz, and (3) beat-by-beat sequences of cardiac and respiratory parameters.

Based on the data gathered from 100 elite athletes and 20 students included in the control group, we found that temporal causal links are the most evident between the tidal volume signal and the instantaneous heart rate curve (RR intervals or tachogram), and that adding a "cause" variable may improve the prediction of the "effect" variable by 20.3% (median) for supine body positions. While the same causal directions are suggested for standing, the complexity seems higher, as the improvement falls to 14.2% (median). The causal link structures and directions can be parameterized and enable distinguishing athletes from non-athletes with 83.3% accuracy on average. The classification of static and dynamic components can probably be supplemented with causal parameters; however, this requires further investigation and confirmation.

In our opinion, the presented approaches would extend the set of techniques used for profiling training trends by connecting cardiorespiratory data with other psychological information.

AUTHOR CONTRIBUTIONS

MM and HK worked on the conceptualization, investigation, project administration, validation, writing and reviewing. MM worked on data curation, methodology, formal analysis, and visualization.

ACKNOWLEDGMENTS

We thank Martin Berka for linguistic adjustments. MM gratefully acknowledges Warsaw University of Technology and the Institute of Metrology and Biomedical Engineering (at WUT) for supporting this work from the resources for young researchers and the statutory resources, respectively.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fphys.2019.00045/full#supplementary-material>

Supplementary Material S1 | The Matlab script used for calculating G-causalities.

Supplementary Material S2 | The data comprising all calculated G-causalities (columns) for all considered participants (rows); NA - not assigned (when statistically insignificant); stored as an RData file (ready to load into R for analysis).

Supplementary Material S3 | The R script consisting of the TiMINo analysis, exploratory data visualization and recursive feature elimination.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Efficacy of Heat Mitigation Strategies on Core Temperature and Endurance Exercise: A Meta-Analysis

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OPEN ACCESS

Edited by:

Toby Mündel,
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Reviewed by:

Tatsuro Amano,
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equally to this work

Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 03 November 2018

Accepted: 21 January 2019

Published: 13 February 2019

Citation:

Alhadad SB, Tan PMS and Lee JKW
(2019) Efficacy of Heat Mitigation
Strategies on Core Temperature and
Endurance Exercise: A Meta-Analysis.
Front. Physiol. 10:71.
doi: 10.3389/fphys.2019.00071

Background: A majority of high profile international sporting events, including the coming 2020 Tokyo Olympics, are held in warm and humid conditions. When exercising in the heat, the rapid rise of body core temperature (T_c) often results in an impairment of exercise capacity and performance. As such, heat mitigation strategies such as aerobic fitness (AF), heat acclimation/acclimatization (HA), pre-exercise cooling (PC) and fluid ingestion (FI) can be introduced to counteract the debilitating effects of heat strain. We performed a meta-analysis to evaluate the effectiveness of these mitigation strategies using magnitude-based inferences.

Methods: A computer-based literature search was performed up to 24 July 2018 using the electronic databases: PubMed, SPORTDiscus and Google Scholar. After applying a set of inclusion and exclusion criteria, a total of 118 studies were selected for evaluation. Each study was assessed according to the intervention's ability to lower T_c before exercise, attenuate the rise of T_c during exercise, extend T_c at the end of exercise and improve endurance. Weighted averages of Hedges' g were calculated for each strategy.

Results: PC ($g = 1.01$) was most effective in lowering T_c before exercise, followed by HA ($g = 0.72$), AF ($g = 0.65$), and FI ($g = 0.11$). FI ($g = 0.70$) was most effective in attenuating the rate of rise of T_c , followed by HA ($g = 0.35$), AF ($g = -0.03$) and PC ($g = -0.46$). In extending T_c at the end of exercise, AF ($g = 1.11$) was most influential, followed by HA ($g = -0.28$), PC ($g = -0.29$) and FI ($g = -0.50$). In combination, AF ($g = 0.45$) was most effective at favorably altering T_c , followed by HA ($g = 0.42$), PC ($g = 0.11$) and FI ($g = 0.09$). AF (1.01) was also found to be most effective in improving endurance, followed by HA (0.19), FI (-0.16) and PC (-0.20).

Conclusion: AF was found to be the most effective in terms of a strategy's ability to favorably alter T_c , followed by HA, PC and lastly, FI. Interestingly, a similar ranking was observed in improving endurance, with AF being the most effective, followed by HA, FI, and PC. Knowledge gained from this meta-analysis will be useful in allowing athletes, coaches and sport scientists to make informed decisions when employing heat mitigation strategies during competitions in hot environments.

Keywords: thermoregulation, aerobic fitness, heat acclimation, heat acclimatization, pre-exercise cooling, fluid ingestion

INTRODUCTION

Exercising in the heat often results in elevation in body core temperature (T_c). This is the cumulative result of more heat being produced by the working muscles than heat loss to the environment coupled with hot and/or humid environmental conditions (Berggren and Hohwu Christensen, 1950; Saltin and Hermansen, 1966). Studies have shown that an accelerated increase in T_c could impair both exercise performance (i.e. time trial) and exercise capacity (i.e., time to exhaustion) (Galloway and Maughan, 1997; Parkin et al., 1999). In ambient temperatures of 4°, 11°, 21°, and 31°C, a compromise in endurance capacity due to thermoregulatory stress was already evident at 21°C (Galloway and Maughan, 1997). Parkin et al. (1999) found that time to exhaustion was longest when cycling in ambient temperatures of 3°C (85 min), followed by 20°C (60 min) and 40°C (30 min).

Elite athletes, however, cannot avoid competing in the heat since a majority of high-profile international sporting events are often held in warm conditions. The 2008 Summer Olympics in Beijing was held in average ambient conditions of 25°C with 81% relative humidity. Similarly, the 2010 Youth Olympic Games in Singapore had temperatures reaching 31°C with relative humidity between 80 and 90%. The upcoming 2020 Olympics held in Tokyo's hot and humid summer period could potentially expose athletes to one of the most challenging environmental conditions observed in the modern history of the Olympic Games, with temperatures upwards of 35°C and above 60% relative humidity. Therefore, athletes have to learn to adapt and perform in these unfavorable environments and whenever possible, incorporate mitigation strategies to counter the negative effects of heat strain to augment performance and health.

Exercise tolerance in the heat can be affected by multiple factors such as the attainment of a critically high T_c (Gonzalez-Alonso et al., 1999b), cardiovascular insufficiency (Gonzalez-Alonso and Calbet, 2003), metabolic disturbances (Febbraio et al., 1994b, 1996; Parkin et al., 1999) and reductions in central nervous system drive to skeletal muscle (Nybo and Nielsen, 2001; Todd et al., 2005). Indeed, a high T_c represents one of the key limiting factors to exercise tolerance in the heat. The development of hyperthermia has been associated with alterations in self-pacing strategies in exercise performance trials or earlier voluntary termination during exercise capacity trials (Nielsen et al., 1993; Gonzalez-Alonso et al., 1999a,b).

In order to optimize exercise tolerance in the heat, exercising individuals often employ strategies to alter T_c . There are various ways in which this can be done, such as aerobic fitness (AF) (Nadel et al., 1974; Cheung and McLellan, 1998b), heat acclimation/acclimatization (HA) (Nielsen et al., 1993; Cotter et al., 1997), pre-exercise cooling (PC) (Gonzalez-Alonso et al., 1999a,b; Cotter et al., 2001) and fluid ingestion (FI) (Greenleaf and Castle, 1971; McConell et al., 1997). These strategies have shown to be effective in improving exercise tolerance in warm conditions through various processes that include alterations in heat dissipation ability, cardiovascular stability and adaptations and changes to the body's heat storage capacity.

Being able to objectively rank these heat mitigation strategies in order of their efficacy will be particularly useful for an athlete preparing to compete in the heat. This knowledge will also be beneficial for coaches, fitness trainers and backroom staff to discern when they consider heat mitigation in warm, humid conditions. With limited amount of time and resources, an evidence-based approach to quantify the efficacy of various heat mitigation strategies will allow selection of the most effective strategy to optimize performance and health and determine the priority in which these strategies should be employed. Furthermore, no comparison of the effect of different heat mitigation strategies have been presented using a meta-analysis thus far.

Therefore, the purpose of this review was to objectively evaluate the efficacy of various heat mitigation strategies using Hedges' g . Each study was analyzed in terms of the degree to which (i) T_c was lowered at the start of exercise; (ii) the rise of T_c is attenuated; (iii) T_c is extended at the end of exercise to safe limits (McLellan and Daanen, 2012) and (iv) endurance are improved. The weighted averages of Hedges' g (Hopkins et al., 2009) were then calculated, and the various heat mitigation strategies ranked in order of effectiveness in terms of both affecting T_c measurements and endurance.

MATERIALS AND METHODS

Search Strategy

A computer-based literature search was performed using the following electronic databases: PubMed, SPORTDiscus and Google Scholar. The electronic database was searched with the following keywords: "fitness," "training," "heat acclimation," "heat acclimatization," "precooling," "pre-cooling," "cold water immersion," "cold air," "cold room," "cold vest," "cold jacket," "ice vest," "cold fluid," "cold beverage," "neck collar," "neck cooling," "ice slurry," "ice slush," "fluid ingestion," "fluid intake," "water ingestion," "water intake," "fluid replacement," "rehydration," "thermoregulation," "core temperature," and "heat mitigation." Searches were systematically performed by combining the keywords and using Boolean operators "AND" and "OR" to yield the maximum outcome of relevant studies. Where applicable, we applied filters for language (English) and species (Human). In addition, a manual citation tracking of relevant studies and review articles was performed. The last day of the literature search was 24 July 2018.

Inclusion and Exclusion Criteria

Studies were screened and included if they met the following criteria: (i) they investigated the effect of a heat mitigation strategy on T_c in an exercise context; (ii) they were conducted in warm or hot ambient conditions of more than 20°C; and (iii) they included a control condition or a pre-intervention and post-intervention assessment. Studies were excluded based on the following criteria: (i) they reported the use of pharmacological agents to alter T_c due to ethical issues and dangers involved with its use; (ii) they were review articles, abstracts, case studies and editorials; (iii) they involved combined use of different methods; and (iv) they involved children or the elderly.

Data Extraction

The following data were extracted: participant characteristics, sample size, ambient conditions, exercise protocol, intervention method, exercise outcome and T_c measurements. T_c measurements included the type of T_c measure used, T_c at the beginning of exercise, rate of rise of T_c and T_c at the end of exercise. In studies where mean and standard deviation of T_c were not reported in the text, the relevant data was extracted using GetData Graph Digitiser (<http://getdata-graph-digitizer.com>). In the event that pertinent data were not available, the corresponding authors of the manuscripts were contacted. Studies with missing data that could not be retrieved or provided by the author were excluded from the meta-analysis.

Data Analysis

In the event that rate of rise of T_c was not provided in the study, it was calculated as the difference between the T_c at the end of exercise and T_c at the beginning of exercise divided by the time taken to complete the task. When studies only reported standard errors, standard deviations were calculated by multiplying the standard error by the square root of the sample size.

Standardized mean differences (Hedges' g) and 95% confidence intervals (CIs) were also calculated for each study. This was derived using the mean T_c differences divided by the pooled standard deviation either between the control and intervention groups or between the pre-intervention and post-intervention states. A bias-corrected formula for Hedges' g for all studies was used to correct for positive and small sample bias (Borenstein et al., 2009). Weighted average of Hedges' g for each heat mitigation strategy was calculated and presented in a forest plot. A combined weighted average of Hedges' g values across all three phases for each strategy's effect on altering T_c and on endurance was also calculated, and used as the basis for ranking. The magnitude of the Hedges' g -values were interpreted as follows: <0.20 , trivial; 0.20 – 0.49 , small; 0.50 – 0.79 , moderate; and ≥ 0.80 , large.

RESULTS

Search Results

The initial identification process yielded 5159 references and after removing duplicates and screening for title and abstract, 229 full texts were obtained. Of these, based on the assessment of study relevance and the inclusion and exclusion criteria, 118 were found to be relevant and therefore included in the analysis. The number of studies found for each heat mitigation strategy is as follows: AF ($n = 22$), HA ($n = 35$), PC ($n = 42$), and FI ($n = 24$) (Figure 1). It should be noted that AF studies may incorporate effects of HA due to the environmental conditions that the AF studies are carried out in. To separate these effects, training periods for "within subjects" AF studies included were conducted at temperatures of 30°C and below. No separation based on temperature was determined for "between subjects" studies as no training was carried out for the subjects prior to the exercise test. Characteristics of the selected studies are summarized in Tables 1–4.

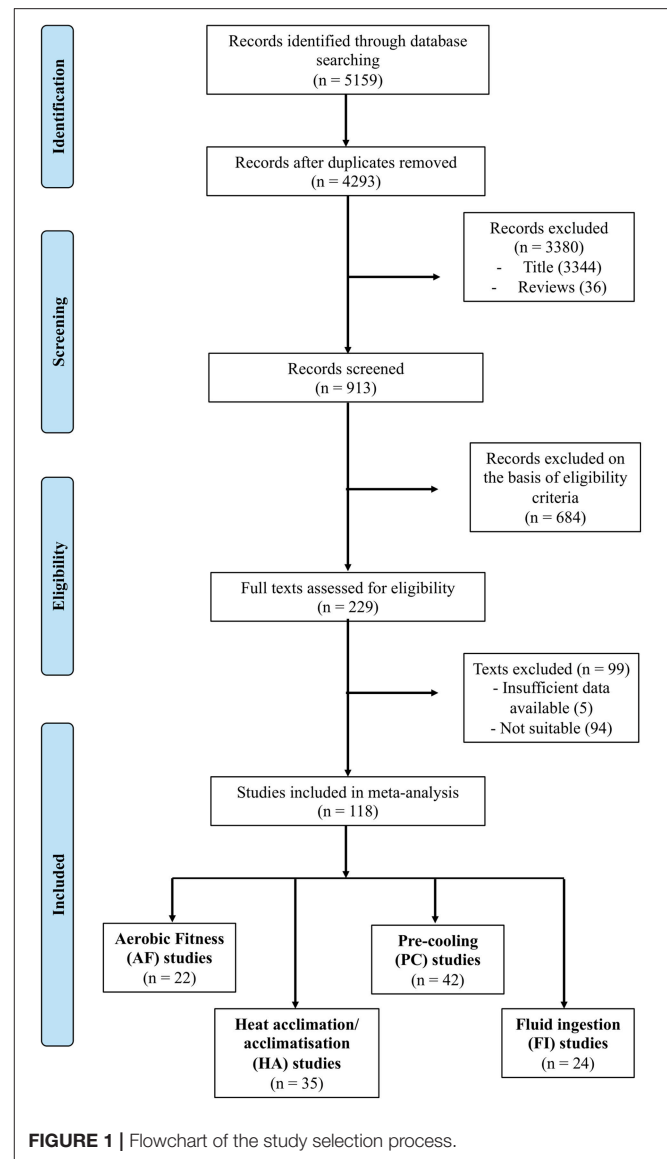


FIGURE 1 | Flowchart of the study selection process.

Effect of Heat Mitigation Strategies on T_c

PC was found to be the most effective in the lowering of T_c before exercise (Hedge's $g = 1.01$; 95% Confidence Intervals 0.85 – 1.17 ; Figure 2). A moderate effect on lowering of T_c before exercise was observed for HA (0.72 ; 0.58 to 0.86) and AF (0.65 ; 0.46 to 0.85) while FI (0.11 ; -0.08 to 0.31) only exhibited a trivial effect on lowering T_c before exercise.

Rate of rise of T_c during exercise was most attenuated by FI (0.70 ; 0.46 to 0.94), followed by HA (0.35 ; 0.19 to 0.50). AF (-0.03 ; -0.24 to 0.18) showed a trivial effect on the rate of rise of T_c while PC (-0.46 ; -0.63 to -0.28) did not appear to be as effective in lowering the rate of rise of T_c .

AF (1.11 ; 0.71 to 1.51) exhibited a large effect on extending the limit of T_c at the end of exercise. However, HA (-0.28 ; -0.52 to -0.04), PC (-0.29 ; -0.44 to -0.14), and FI (-0.50 ; -0.74 to -0.27) did not seem as effective in extending the T_c limit at the end of exercise.

TABLE 1 | Summary of aerobic fitness studies.

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Mora-Rodriguez et al., 2010	36°C 25% RH 2.5 m/s airflow	10 untrained 10 trained	EPW: Cycle at 40, 60 or 80% VO ₂ peak, equaled by total work	–	–	T _{re}	Utr: 37.6 ± 0.2°C Tr: 37.4 ± 0.2°C (S)	–	–
Ichinose et al., 2005	30°C 50% RH	9	EPW: 20 min cycle at pretraining 70% VO ₂ peak under isosmotic conditions	Cycle at 60% VO ₂ peak at 30°C, 50% RH for 1 hr/day for 10 days	–	T _{oes}	Before: 36.68 ± 0.15°C After: 36.53 ± 0.18°C (S)	Before: 5.31 ± 1.17°C/h After: 4.74 ± 0.97°C/h (CAL)	–
Selkirk and McLellan, 2001	40°C 30% RH <0.1 m/s wind speed	6 untrained (low BF) 6 untrained (high BF) 6 trained (low BF) 6 trained (high BF)	EC: Treadmill walking at 3.5 km/h to exhaustion	–	Longer exercise times in T _{low} vs T _{low} and T _{low} vs. T _{high} (S)	T _{re}	Utr _{low} : 37.19 ± 0.20°C T _{low} : 37.02 ± 0.20°C Utr _{high} : 37.26 ± 0.37°C T _{high} : 37.10 ± 0.22°C (NS)	Utr _{low} : 1.20 ± 0.34°C/h T _{low} : 1.27 ± 0.10°C/h Utr _{high} : 1.24 ± 0.19°C/h T _{high} : 1.55 ± 0.15°C/h (CAL)	Utr _{low} : 38.58 ± 0.47°C T _{low} : 39.48 ± 0.02°C Utr _{high} : 38.78 ± 0.59°C T _{high} : 39.22 ± 0.22°C (S)
Periard et al., 2012	40°C 50% RH 4.1 m/s convective airflow	8 untrained 8 trained	EC: Cycle to exhaustion at 60 & 75% VO ₂ max	–	No influence on times to exhaustion	T _{re}	Utr _{H60%} : 37.0 ± 0.3°C T _{H60%} : 36.9 ± 0.2°C Utr _{H75%} : 37.1 ± 0.3°C T _{H75%} : 36.8 ± 0.3°C (REQ)	–	Utr _{H60%} : 39.4 ± 0.4°C T _{H60%} : 39.8 ± 0.3°C Utr _{H75%} : 38.8 ± 0.5°C T _{H75%} : 39.3 ± 0.6°C (NS)
Cheung and McLellan, 1998a	40°C 30% RH <0.1 m/s wind speed	7 moderately fit 8 highly fit	EC: Treadmill exercise at 3.5 km/h, 0% grade in a euhydrated state to exhaustion	–	No influence on tolerance time	T _{re}	MF: 36.93 ± 0.27°C HF: 36.85 ± 0.22°C (NS)	MF: 1.14 ± 0.29°C/h HF: 1.21 ± 0.27°C/h (CAL)	MF: 38.77 ± 0.27°C HF: 39.15 ± 0.18°C (NS)
Ichinose et al., 2009	25°C 45% RH	11	EPW: Cycle at 50% VO ₂ max for 30 min	Cycle at 60% VO ₂ max for 60 min/day, 4–5 days/week over 3 menstrual cycles at 30°C, 45% RH	–	T _{oes}	Before: 37.27 ± 0.33°C After: 37.07 ± 0.20°C (S)	Before: 0.68 ± 0.81°C/h After: 0.80 ± 0.52°C/h (CAL)	–
Cheung and McLellan, 1998b	40°C 30% RH <0.1 m/s wind speed	8	EC: Treadmill heat stress test in a euhydrated state to exhaustion	Treadmill walk for 1 h, 6 days/week at 60–65% VO ₂ max for 2 weeks in a normothermic environment	No influence on tolerance time	T _{re}	Before: 37.08 ± 0.24°C After: 36.93 ± 0.34°C (NS)	Before: 1.04 ± 0.34°C/h After: 1.07 ± 0.30°C/h (CAL)	Before: 38.70 ± 0.37°C After: 38.61 ± 0.25°C (NS)

(Continued)

TABLE 1 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Wright et al., 2012	40°C 30% RH <0.1 m/s wind speed	11 untrained 12 trained	EC: Treadmill walk at 4.5 km/h, 2% incline to exhaustion	–	Longer time to exhaustion (S)	T _{re}	–	Utr: 1.25 ± 0.20°C/h Tr: 1.14 ± 0.28°C/h (NS)	Utr: 39.0 ± 0.3°C Tr: 39.7 ± 0.3°C (S)
Takeno et al., 2001	30°C 50% RH	5	EPW: 30 min cycle at 60% VO ₂ peak	Cycle at 60% VO ₂ peak for 60 min/day, 5 days/week for 2 weeks at atmospheric pressure	–	T _{oes}	Before: 37.0 ± 0.2°C After: 36.8 ± 0.2°C (S)	Before: 2.6 ± 1.0°C/h After: 2.6 ± 0.6°C/h (CAL)	–
Stapleton et al., 2010	30°C 15% RH	10	EPW: 60 min cycle at a constant rate of heat production	Aerobic and resistance training for 8 weeks	–	T _{oes}	Before: 37.10 ± 0.28°C After: 36.95 ± 0.24°C (S)	Before: 0.68 ± 1.8°C/h After: 0.56 ± 0.16°C/h (S)	–
Lim et al., 2009	35°C 40% RH	9 normal training 9 increased training	EC: Treadmill run at 70% VO ₂ max to exhaustion	NT: Routine training program for 14 days IT: 20% increase in training load for 14 days	–	T _{gl}	Before _{NT} : 36.68 ± 0.32°C After _{NT} : 36.70 ± 0.41°C Before _{IT} : 36.98 ± 0.46°C After _{IT} : 37.11 ± 0.39°C (NS)	Before _{NT} : 3.48 ± 0.96°C/h After _{NT} : 2.88 ± 1.14°C/h Before _{IT} : 3.42 ± 1.20°C/h After _{IT} : 3.48 ± 1.26°C/h (CAL)	–
Ho et al., 1997	36°C 20% RH	6 young sedentary 6 young fit	EPW: 20 min cycle at 35% VO ₂ peak	–	–	T _{oes}	Sedentary: 37.1 ± 0.2°C Fit: 36.9 ± 0.2°C (NS)	–	–
Shvartz et al., 1977	23°C dry bulb 16°C wet bulb <0.2 m/s wind speed	7 untrained 7 trained	EPW: 60 min bench stepping at 41 W	–	–	T _{re}	Utr: 36.9 ± 0.19°C Tr: 37.1 ± 0.31°C (NS)	Utr: 1.0 ± 0.37°C/h Tr: 1.0 ± 0.42°C/h (CAL)	–
Cramer et al., 2012	24.5°C 0.9 kPa RH 1.3 m/s air velocity	10 unfit 11 fit	EPW: 60 min cycle at 60% VO ₂ max or to produce metabolic heat of 275 W/m ²	–	–	T _{re}	Unfit _{60%} : 37.40 ± 0.22°C Fit _{60%} : 37.09 ± 0.20°C Unfit _{BAL} : 37.43 ± 0.25°C Fit _{BAL} : 37.14 ± 0.23°C (S)	Unfit _{BAL} : 0.93 ± 0.40°C/h Fit _{BAL} : 0.95 ± 0.33°C/h (CAL)	–

(Continued)

TABLE 1 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Shvartz et al., 1974	21.5°C dry bulb 17.5°C wet bulb	5	EPW: 60 min bench-stepping at 85% VO ₂ max	Bench-stepping for 60 min/day for 12 days	–	T _{re}	Before: 37.4 ± 0.3°C After: 37.2 ± 0.2°C (S)	–	–
Ikegawa et al., 2011	30°C 50% RH	7	EPW: 30 min cycle at 65% VO ₂ peak in a euhydrated state	Cycle for 30 min/day for 5 days	–	T _{oes}	Before: 36.74 ± 0.32°C After: 36.50 ± 0.16°C (S)	Before: 3.18 ± 0.83°C/h After: 3.06 ± 0.49°C/h (CAL)	–
Yamauchi et al., 1997	23°C 60% RH	5 untrained 6 trained	EPW: 30 min cycle at 80W	–	–	T _{sym}	Utr: 36.71 ± 0.22°C Tr: 36.50 ± 0.15°C (NS)	–	–
Yamazaki et al., 1994	25°C 35% RH	8 untrained 9 trained	EPW: 30 min cycle at 35% VO ₂ max	–	–	T _{oes}	Utr: 37.06 ± 0.30°C Tr: 37.02 ± 0.23°C (NS)	–	–
Gagnon et al., 2012	42°C 20% RH 1 m/s air speed	8 untrained 8 trained	EPW: 120 min cycle at 120 W with fluid replacement	–	–	T _{oes}	Utr: 36.96 ± 0.25°C Tr: 36.69 ± 0.25°C (NS)	Utr: 0.68 ± 0.30°C/h Tr: 0.82 ± 0.34°C/h (CAL)	–
Merry et al., 2010	24.3°C 50% RH 4.5 m/s wind velocity	6 untrained 6 trained	EPW: 40 min cycle at 70% VO ₂ peak in a euhydrated state	–	–	T _{rec}	Utr: 36.88 ± 0.26°C Tr: 36.56 ± 0.29°C (REQ)	–	–
Shields et al., 2004	32°C 32% RH	7	EPW: 45 min cycling at 40% VO ₂ peak	Exercise at 50% VO ₂ reserve for 40 min/day for 3 days per week, over 12 weeks	–	T _{oes}	Before: 37.00 ± 0.27°C After: 36.88 ± 0.25°C (REQ)	Before: 0.69 ± 0.65°C/h After: 0.64 ± 0.89°C/h (REQ)	–
Smoljanic et al., 2014	25°C 37% RH	7 fit 7 unfit	EPW: Run for 60 min at 60% VO _{2max} , followed by run at fixed metabolic heat production of 640 W	–	–	T _{re}	–	Fit _{60minrun} : 1.23 ± 0.37°C/h Unfit _{60minrun} : 0.90 ± 0.30°C/h (S) Fit _{fixedmetheatprod} : 0.86 ± 0.26°C/h Unfit _{fixedmetheatprod} : 0.92 ± 0.32°C/h (NS)	–

RH, relative humidity; EC, exercise capacity; EP, exercise performance; EPW, exercise performance at a fixed workload; S, significant; NS, not significant; CAL, calculated values; REQ, requested values; T_{re}, rectal temperature; T_{oes}, oesophageal temperature; T_{gi}, gastrointestinal temperature; T_{sym}, tympanic temperature; Utr, untrained subjects; Tr, trained subjects; BF, body fat; NT, normal training; IT, increased training; MF, moderately fit subjects; HF, highly fit subjects; AF, aerobic fitness.

In combination, AF was found to be the most effective at favorably altering T_c (0.45; 0.32 to 0.59), followed by HA (0.42; 0.33 to 0.52), PC (0.11; 0.02 to 0.19) and FI (0.09; -0.03 to 0.13) (Figure 3).

In addition, AF studies included both longitudinal and cross-sectional studies. We sought to determine if there was an effect on T_c variables when comparing “between subjects” and “within subjects” studies. We found that effect sizes were comparable with “between subjects” AF studies (0.45; 0.28 to 0.61) and “within subjects” AF studies (0.38; 0.14 to 0.61). The large overlap in CIs suggest that the inclusion of both study types did not have significantly different effects on T_c variables.

Effect of Heat Mitigation Strategies on Endurance

Of the 118 articles selected and used for analysis of the strategies based on effects on T_c , 45 studies also included measurements of endurance. The number of studies for each heat mitigation strategy is as follows: AF ($n = 5$), HA ($n = 7$), PC ($n = 24$), and FI ($n = 9$).

We observed that AF was the most effective in improving endurance (1.01; 1.40 to 0.61), followed by HA (0.19; -0.16 to 0.54), FI (-0.16; -0.53 to 0.22), and PC (-0.20; -0.56 to 0.17) (Figure 4).

DISCUSSION

This meta-analysis aimed to evaluate the efficacy of different heat mitigation strategies. Our main findings suggest that AF was most effective in altering T_c , followed by HA, PC and FI. A secondary objective was to evaluate the effect of these strategies on endurance. We observed that aerobic fitness was again the most beneficial, followed by heat acclimation/acclimatization, fluid ingestion and pre-cooling. It is noteworthy that the ranking of the effectiveness of the heat mitigation strategies on favorably altering T_c is similar to their effectiveness in improving endurance (Table 5).

Aerobic Fitness

Individuals with a higher aerobic fitness have been shown to have a lower pre-exercise T_c at rest (Selkirk and McLellan, 2001; Mora-Rodriguez et al., 2010). Aerobic fitness also enhances heat dissipation by lowering the threshold T_c at which both skin vasodilation and sweating occur (Nadel et al., 1974; Ichinose et al., 2009). Kuwahara et al. (2005) found that sweat rates of trained individuals were significantly higher than that of untrained individuals over a 30 min cycling exercise and that the onset of sweating occurred earlier on in the exercise as well. Higher aerobic fitness has also shown to cause an increase in skin blood flow (Fritzsche and Coyle, 2000). The combination of these two effects will lower T_c by enhancing heat dissipation during exercise in the heat. In addition, a greater aerobic fitness elicits a higher T_c attained at the end of exercise (Cheung and McLellan, 1998b; Selkirk and McLellan, 2001). This is corroborated by studies in marathon runners, where highly aerobically trained individuals were able to tolerate greater end T_c without any pathophysiological effects (Maron et al., 1977; Byrne et al., 2006).

However, it should be noted that the ability to extend the limit of T_c at the end of exercise may pose as a double-edged sword, as highly motivated individuals may continue to exercise past the limits of acceptable T_c which could cause higher rates of exertional heat related illnesses occurring.

Heat Acclimation/Acclimatization

Heat acclimation/acclimatization refers to the physiological adaptations that occur as a result of prolonged, repeated exposure to heat stress (Armstrong and Maresh, 1991). It is noteworthy that the magnitude and duration of the heat acclimation/acclimatization protocols are important considerations in the development of the above physiological adaptations (Tyler et al., 2016). Previous meta-analysis and studies have shown that effects on cardiovascular efficiency and T_c may be achieved in protocols lasting less than 7 days, while thermoregulatory adaptations and improvements in endurance capacity and performance may require up to 14 days. For the benefits to be maximized, protocols longer than 2 weeks may also be considered (Armstrong and Maresh, 1991; Pandolf, 1998; Tyler et al., 2016). Heat acclimation/acclimatization has been shown to effectively reduce pre-exercise body temperature (Nielsen et al., 1993; Cotter et al., 1997). The physiological adaptations also observed include decreased heart rate (Harrison, 1985; Lorenzo and Minson, 2010), increased cardiac output (Harrison, 1985; Nielsen, 1996) and plasma volume (Mitchell et al., 1976; Lorenzo and Minson, 2010). Most significantly, cutaneous vasodilation occurs at a lower T_c threshold, together with an increase in skin blood flow (Roberts et al., 1977). The onset of sweating also occurs at a lower T_c threshold, resulting in increased sweat rates during exercise (Cotter et al., 1997; Cheung and McLellan, 1998a). Taken together, this helps to reduce the rate of rise of T_c during exercise due to increased cardiovascular efficiency and heat dissipation mechanisms.

However, for tropical natives, heat acclimatization does not lead to more efficient thermoregulation. In a study by Lee and colleagues (Lee et al., 2012), military soldiers native to a warm and humid climate were asked to undergo a 10 day heat acclimatization programme. Although there was an increase in work tolerance following acclimatization, no significant cardiovascular or thermoregulatory adaptations were found. These observations could suggest that thermoregulatory benefits of heat acclimatization are minimized in tropical natives, possibly due to the “partially acquired heat acclimatization status from living and training in a warm and humid climate” (Lee et al., 2012). Alternatively, thermoregulatory benefits from heat acclimatization may also be minimized in tropical natives due to modern behavioral adaptations such as the usage of air conditioning in living spaces and the avoidance of exercise during the hottest periods of the day that reduce the environmental heat stimulus experienced (Bain and Jay, 2011). In addition, evaporative heat loss through sweating is compromised with high relative humidity and therefore results in a higher rate of rise of T_c during exercise (Maughan et al., 2012).

It is also noteworthy that heat acclimation/acclimatization encompasses aerobic fitness as well. In most protocols, there

TABLE 2 | Summary of heat acclimation/acclimatization studies.

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
LOW HUMIDITY (<50% RH)									
Lorenzo and Minson, 2010	38°C 30% RH	12	EP: 1 h cycling time trial	Two 45 min exposures to 40°C, 30% RH conditions for 10 days	Higher power output (S)	T _{re}	Before: 37.1 ± 0.3°C After: 37.0 ± 0.4°C (REQ)	–	Before: 39.5 ± 0.3°C After: 39.4 ± 0.7°C (NS)
Cheung and McLellan, 1998a	40°C 30% RH <0.1 m/s wind speed	7 moderately fit 8 highly fit	EC: Treadmill walk at 3.5 km/h, 0% grade in a euhydrated state to exhaustion	1 h exposures to 40°C, 30% RH conditions for 5 days/week for 2 weeks	No influence on tolerance time	T _{re}	Before _{MF} : 36.93 ± 0.27°C After _{MF} : 36.96 ± 0.28°C Before _{HF} : 36.85 ± 0.22°C After _{HF} : 36.74 ± 0.19°C (NS)	Before _{MF} : 1.14 ± 0.29°C/h After _{MF} : 1.08 ± 0.25°C/h Before _{HF} : 1.21 ± 0.27°C/h After _{HF} : 1.25 ± 0.20°C/h (CAL)	Before _{MF} : 38.77 ± 0.27°C After _{MF} : 38.79 ± 0.31°C Before _{HF} : 39.15 ± 0.18°C After _{HF} : 39.14 ± 0.21°C (NS)
Nielsen et al., 1993	40°C 10% RH	8	EC: Cycling at approximately 50% VO ₂ max to exhaustion	90 min exposures to 40°C, 10% RH conditions for 9–12 days	Increase in endurance time (S)	T _{oes}	–	–	Before: 39.8 ± 0.4°C After: 39.7 ± 0.4°C (NS)
Horstman and Christensen, 1982	45°C dry bulb 23°C wet bulb	6 men 4 women	EPW: 120 min cycle at 40% VO ₂ max	2 h exposures to 45°C dry bulb, 23°C wet bulb conditions for 11 days	–	T _{re}	–	Before _{men} : 1.5 ± 0.5°C/h After _{men} : 0.8 ± 0.2°C/h (NS) Before _{women} : 1.4 ± 0.4°C/h After _{women} : 0.5 ± 0.0°C/h (S)	–
Weller et al., 2007	46.1°C dry bulb 17.9% RH	8 in RA ₁₂ 8 in RA ₂₆	EPW: 60 min treadmill walk at 45% VO ₂ peak	100 min exposures to 46.1°C, 17.9% RH conditions for 10 days	–	T _{re}	Before ₁₂ : 37.20 ± 0.27°C After ₁₂ : 36.95 ± 0.22°C Before ₂₆ : 37.27 ± 0.15°C After ₁₂ : 37.00 ± 0.13°C (S)	Before ₁₂ : 1.39 ± 0.41°C/h After ₁₂ : 1.17 ± 0.37°C/h Before ₂₆ : 1.42 ± 0.28°C/h After ₁₂ : 1.16 ± 0.21°C/h (CAL)	–
Shvartz et al., 1977	23°C dry bulb 16°C wet bulb <0.2 m/s wind speed	7 untrained 7 trained	EPW: 60 min bench stepping at 41 W	3 h exposures to 39.4°C dry bulb, 30.3°C wet bulb conditions for 8 days	–	T _{re}	Before _{unr} : 37.1 ± 0.31°C After _{unr} : 36.7 ± 0.20°C Before _{tr} : 36.9 ± 0.19°C After _{tr} : 36.7 ± 0.13°C (S)	Before _{unr} : 1.0 ± 0.37°C/h After _{unr} : 1.0 ± 0.27°C/h Before _{tr} : 1.0 ± 0.42°C/h After _{tr} : 0.9 ± 0.21°C/h (CAL)	–

(Continued)

TABLE 2 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Febbraio et al., 1994a	40°C 20% RH	13	EPW: 40 min cycle at 70% VO ₂ max	90 min exposures to 40°C, 20% RH conditions for 7 days	–	T _{re}	Before: 37.2 ± 0.4°C After: 36.8 ± 0.4°C (NS)	Before: 3.8 ± 0.8°C/h After: 3.6 ± 0.8°C/h (CAL)	–
Beaudin et al., 2009	24°C 30% RH	8	EC: Incremental cycling to exhaustion	2 h passive exposures to 50°C, 20% RH conditions for 10 days	–	T _{oes}	Before: 37.57 ± 0.23°C After: 37.32 ± 0.14°C (S)	–	–
Magalhaes Fde et al., 2006	40°C 32% RH	6	EPW: 60 min cycle at 50% VO ₂ peak	1 h exposures to 40°C, 32% RH conditions for 9 days	–	T _{re}	Before: 37.2 ± 0.2°C After: 37.0 ± 0.2°C (S)	Before: 0.94 ± 0.16°C/h After: 0.88 ± 0.27°C/h (NS)	–
Armstrong et al., 1985	40.1°C 23.5% RH	9	EPW: 90 min treadmill walk at 5.6 km/h, 6% grade with a high or low sodium diet	90 min exposures to 40.1°C, 23.4% RH conditions for 8 days	–	T _{re}	Before _{low} : 37.44 ± 0.66°C After _{low} : 37.05 ± 0.30°C (S) Before _{high} : 37.25 ± 0.72°C After _{high} : 36.97 ± 0.45°C (NS)	Before _{low} : 0.85 ± 0.48°C/h After _{low} : 0.74 ± 0.26°C/h Before _{high} : 0.93 ± 0.59°C/h After _{high} : 0.79 ± 0.36°C/h (CAL)	–
Watkins et al., 2008	39.5°C 27% RH	10	EPW: 30 min cycle at 75% VO ₂ peak	30 min exposures to 39.5°C, 27% RH conditions for 7 days	–	T _{re}	Before: 37.2 ± 0.2°C After: 37.0 ± 0.2°C (S)	Before: 1.8 ± 0.9°C/h After: 1.8 ± 0.5°C/h (CAL)	–
Burk et al., 2012	42°C 18% RH	21	EC: Treadmill walk at 60% VO ₂ peak to exhaustion	Two 50 min exposures to 42°C, 18% RH conditions for 10 days	Increase in endurance time (S)	T _{re}	Before: 37.2 ± 0.2°C After: 37.0 ± 0.2°C (S)	Before: 1.7 ± 0.4°C/h After: 1.0 ± 0.3°C/h (CAL)	Before: 39.7 ± 0.4°C After: 39.7 ± 0.4°C (NS)
Hodge et al., 2013	35.3°C 40.2% RH	8	EPW: 90 min treadmill walk at 40% VO ₂ max	90 min exposures to 35.3°C, 40.2% RH conditions for 8 days	–	T _{re}	Before: 37.1 ± 0.3°C After: 36.8 ± 0.4°C (REQ)	Before: 1.8 ± 0.3°C/h After: 0.7 ± 0.4°C/h (REQ)	–
Magalhaes Fde et al., 2010	40°C 45% RH	9	EPW: 90 min treadmill run at 50% maximal power output	90 min exposures to 40°C, 45% RH conditions for 11 days	–	T _{re}	Before: 37.43 ± 0.17°C After: 37.26 ± 0.18°C (REQ)	Before: 1.05 ± 0.29°C/h After: 1.03 ± 0.23°C/h (REQ)	–
Racinals et al., 2012	44°C 44% RH	18	EPW: 30 min treadmill walk at 5 km/h, 1% grade	Football training in 38–43°C, 12–30% RH conditions for 6 days	–	T _{re}	Before: 37.37 ± 0.17°C After: 37.26 ± 0.23°C (REQ)	Before: 1.18 ± 0.51°C/h After: 1.24 ± 0.62°C/h (REQ)	–

(Continued)

TABLE 2 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Best et al., 2014	35°C 40% RH	7	EPW: 60 min cycle at 70% VO _{2max}	60 min cycling at 70% VO _{2max} in 35°Cm, 40% conditions for 6 days	–	T _{re}	–	–	Before: 39.1 ± 0.3°C After: 38.7 ± 0.3°C (S) (Graph)
Dileo et al., 2016	45°C 20% RH	10	EC: Ramped running protocol until volitional fatigue	2 × 45 min periods cycling at 50% VO _{2max} in 45°C, 20% RH conditions for 5 days	–	T _{re}	Before: 36.9 ± 0.2°C After: 36.7 ± 0.2°C (NS) (Graph)	–	Before: 38.9 ± 0.6°C After: 38.7 ± 0.4°C (S)
Flouris et al., 2014	40°C 20% RH	10	EPW: Cycle at fixed rates of metabolic heat production equal to 300, 350 and 400 W/m ² , for 30 min each	90 min cycling at 50% VO _{2peak} in 40°C, 20% RH for 14 days	–	T _{re}	Before: 37.0 ± 0.2 °C After: 36.7 ± 0.1°C (S) (Graph)	–	–
Gibson et al., 2015	40°C 28% RH	24	EPW: 30 min running at 9 km/h and 2% elevation	FIXED protocol: 90 min of cycling at 50% VO _{2peak} in 40°C, 39% RH ISOCONT: Cycle at 65% VO _{2peak} until T _{re} of 38.5°C reached ISOPROG: Cycle at 65% VO _{2peak} until T _{re} of 38.5°C reached for first 5 days, (then until 39°C for last 5 days). STHA – Protocol above for 5 days LTHA – Protocol above for 10 days	–	T _{re}	Before (FIXED): 37.2 ± 0.4°C Before (ISOCONT): 37.1 ± 0.2°C Before (ISOPROG): 36.9 ± 0.4°C STHA – Before (FIXED): 36.9 ± 0.4°C Before (ISOCONT): 37.0 ± 0.2°C Before (ISOPROG): 36.7 ± 0.4°C (S) LTHA – Before (FIXED): 36.9 ± 0.4°C Before (ISOCONT): 37.0 ± 0.2°C Before (ISOPROG): 36.8 ± 0.3°C (S)	Before (FIXED): 2.35 ± 0.87°C/h Before (ISOCONT): 3.21 ± 0.6°C/h Before (ISOPROG): 2.97 ± 0.4°C/h STHA – After (FIXED): 2.49 ± 1.13°C/h After (ISOCONT): 2.77 ± 0.71°C/h After (ISOPROG): 2.87 ± 0.49°C/h LTHA – After (FIXED): 2.39 ± 0.94°C/h After (ISOCONT): 2.56 ± 0.75°C/h After (ISOPROG): 2.82 ± 0.78°C/h	–
Racinais et al., 2015b	34°C 18% RH	9	EP: 43.3 km cycling time trial	4 h exposures to 34°C, 18% RH conditions for 2 weeks	Faster time trial (S)	T _{re}	–	–	Before: 40.2 ± 0.4°C After: 40.1 ± 0.4°C
HIGH HUMIDITY (> 50% RH)									
Cotter et al., 1997	39.5°C 59.2% RH	8	EPW: 70 min cycle at 50% peak aerobic power	70 min exposures to 39.5°C, 59.2% RH conditions for 6 days	–	T _{ac}	Before: 36.83 ± 0.05°C After: 36.62 ± 0.05°C (S)	–	–

(Continued)

TABLE 2 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Fuji et al., 2012	37°C 50% RH <0.2 m/s wind speed	10	EPW: 75 min cycle at 58% VO ₂ peak	Four 20 min exposures to 37°C conditions for 6 days	–	T _{Oes}	Before: 36.6 ± 0.1°C After: 36.4 ± 0.2°C (S)	–	–
Buono et al., 1998	35°C 75% RH	9	EPW: 2 h exercise bouts of either a treadmill walk at 1.34 m/s, 3% grade or a cycle at 75 W	Either treadmill walking at 1.34 m/s, 3% grade or cycling at 75 W in 35°C, 75% RH conditions	–	T _{re}	Before: 37.0 ± 0.3°C After: 36.7 ± 0.4°C (S)	Before: 1.0 ± 0.2°C/h After: 0.8 ± 0.3°C/h (CAL)	–
Lee et al., 2012	32°C dry bulb 70% RH 400 W/m ² solar radiation	18	EPW: Three 60 min marches on the treadmill at 4 km/h, 0% gradient in Skeletal Battle Order (SBO) or Full Battle Order (FBO)	Outdoor route marches at 4 km/h in 29°C, 80% RH conditions for 10 days	–	T _{gi}	BeforeSBO: 37.2 ± 0.3°C AfterSBO: 37.0 ± 0.3°C BeforeFBO: 37.1 ± 0.4°C AfterFBO: 37.0 ± 0.3°C (NS)	BeforeSBO: 0.4 ± 0.2°C/h AfterSBO: 0.4 ± 0.2°C/h BeforeFBO: 0.4 ± 0.2°C/h AfterFBO: 0.5 ± 0.2°C/h (CAL)	–
Kotze et al., 1977	32.2°C wet bulb 33.9°C dry bulb 0.4 m/s wind velocity	4	EPW: 4 h block stepping at an external workload after receiving placebo	4 h exposures to 32.2°C wet bulb, 33.9°C dry bulb conditions for 10 days	–	T _{re}	Before: 37.5 ± 0.2°C After: 37.1 ± 0.2°C (CAL)	Before: 0.5 ± 0.1°C/h After: 0.3 ± 0.1°C/h (CAL)	–
Kobayashi et al., 1980	33.5°C 60% RH	5	EPW: 60 min cycle at 60 to 70% VO ₂ max	100 min exposures to 45 to 50°C, 30 to 40% RH conditions for 9 days	–	T _{re}	Before: 37.4 ± 0.2°C After: 37.0 ± 0.4°C (S)	Before: 2.0 ± 0.4°C/h After: 2.2 ± 0.5°C/h (CAL)	–
Saat et al., 2005	31.1°C 70% RH	16	EPW: 60 min cycle at 60% VO ₂ max	60 min exposures to 31.1°C, 70% RH conditions for 14 days	–	T _{re}	Before: 37.35 ± 0.34°C After: 37.14 ± 0.32°C (NS)	–	–
Patterson et al., 2004	39.8°C 59.2% RH	6	EPW: 90 min cycle at ~44% W _{peak}	90 min exposures to 40°C, 60% RH conditions for 16 days	–	T _{Oes}	Before: 36.97 ± 0.20°C After: 36.74 ± 0.14°C (REQ)	Before: 1.27 ± 0.15°C/h After: 1.04 ± 0.31°C/h (REQ)	–
Garrett et al., 2009	35°C 60% RH	10	EPW: 90 min cycling at 40% peak power output	90 min exposures to 40°C, 60% RH conditions for 5 days	–	T _{re}	Before: 37.05 ± 0.37°C After: 36.95 ± 0.26°C (REQ)	Before: 1.03 ± 0.41°C/h After: 0.90 ± 0.31°C/h (REQ)	–

(Continued)

TABLE 2 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Garrett et al., 2012	35°C 60% RH	8	EPW: 10 min rowing at 30% peak power output, followed by 10 min rowing at 60% peak power output	90 min exposures to 39.5°C, 60% RH conditions for 5 days	–	T _{re}	Before: 37.33 ± 0.16°C After: 37.28 ± 0.28°C (REQ)	Before: 2.04 ± 0.82°C/h After: 1.38 ± 0.98°C/h (REQ)	–
James C. A. et al., 2017	32°C 60% RH	10	EP: 5 km running time trial	90 min exposures to 37°C, 59% RH conditions for 5 days	Faster time trial time (S)	T _{re}	Before: 36.97 ± 0.33°C After: 36.83 ± 0.32°C (S)	–	–
James et al., 2018	32°C 60% RH	9	EP: 5 km running time trial	90 min exposures to 37°C, 60% RH conditions for 5 days	Faster time trial time (S)	T _{re}	Before: 37.12 ± 0.22°C After: 37.03 ± 0.23°C (NS)	Before: 5.41 ± 0.91°C/h After: 5.56 ± 0.25°C/h (CAL)	Before: 39.34 ± 0.3°C After: 39.16 ± 0.44°C (S)
Willmott et al., 2016	30°C 60% RH	14	EP: 5 km running time trial	STHA: 45 min cycling at 50% VO _{2peak} at 35°C, 60% RH once for 4 days TDHA: 45 min cycling at 50% VO _{2peak} at 35°C, 60% twice daily for 2 days	No influence on time trial time.	T _{re}	STHA – Before: 37.5 ± 0.4°C After: 37.3 ± 0.3°C (NS) TDHA – Before: 37.4 ± 0.3°C After: 37.3 ± 0.2°C (NS) (Graph)	Before: 38.69 ± 0.38°C After: 38.53 ± 0.45°C (NS) TDHA – Before: 38.59 ± 0.37°C After: 38.52 ± 0.5°C (NS) (Graph)	STHA – Before: 38.69 ± 0.38°C After: 38.53 ± 0.45°C (NS) TDHA – Before: 38.59 ± 0.37°C After: 38.52 ± 0.5°C (NS) (Graph)
Brade et al., 2013	35°C 60% RH	10	EPW: 70 min repeat sprint protocol	32–48 min cycling exposure at 35°C, 60% RH conditions for 5 days	No influence on performance	T _{gi}	Before: 37.0 ± 0.4°C After: 36.9 ± 0.3°C	Before: 1.54 ± 0.48°C/h After: 1.37 ± 0.36°C/h (CAL)	Before: 38.8 ± 0.4°C After: 38.5 ± 0.3°C
Zimmermann et al., 2018	35°C 50% RH	8	EP: 800 kJ cycling time trial	60 min cycling at 50% VO _{2peak} at 35°C, 49% RH conditions for 10 days (5 days on, 2 off, 5 days on)	Faster cycling time	T _{gi}	Before: 36.9 ± 0.3°C After: 36.7 ± 0.4°C	Before: 3.23 ± 1.31 °C/h After: 3.57 ± 1.04°C/h	Before: 39.0 ± 0.8°C After: 38.9 ± 0.5°C

RH, relative humidity; EC, exercise capacity; EPW, exercise performance at a fixed workload; S, significant; NS, not significant; CAL, calculated values; REQ, requested values; Graph, graph-extracted values; T_{re}, rectal temperature; T_{oes}, oesophageal temperature; T_{ac}, auditory canal temperature; T_{gi}, gastrointestinal temperature; MF, moderately fit subjects; HF, highly fit subjects; STHA, Short term Heat acclimation and acclimatization (HA); TDHA, Twice daily HA; LTHA, Long term HA.

TABLE 3 | Summary of pre-event cooling studies.

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
COLD WATER IMMERSION									
Kay et al., 1999	31.4°C 60.2% RH	7	EP: 30 min self-paced cycling time trial	CON: 30 min rest INT: Whole body water immersion for 58.6 min	Greater distance covered (S)	T _{re}	–	–	CON: 38.7 ± 0.3°C INT: 38.4 ± 0.5°C (NS)
Booth et al., 1997	32°C 60% RH	8	EP: 30 min running time trial	CON: No cooling INT: Cold water immersion for 60 min before exercise	Greater distance covered (S)	T _{re}	CON: 37.4 ± 1.1°C INT: 36.7 ± 0.3°C (S)	–	CON: 39.6 ± 0.6°C INT: 38.9 ± 0.6°C (NS)
Tsuji et al., 2012	37°C 50% RH	10	EC: Cycle at 50% VO ₂ peak to exhaustion	CON: 25 min immersion in 35°C water INT: 25 min immersion in 18°C water	Longer time to exhaustion (S)	T _{oes}	CON: 36.9 ± 0.3°C INT: 36.1 ± 0.3°C (S)	–	–
Gonzalez-Alonso et al., 1999b	40°C 19% RH	7	EC: Cycle at 60% VO ₂ max to exhaustion	CON: 30 min immersion in 36°C water INT: 30 min immersion in 17°C water	Longer time to exhaustion (S)	T _{oes}	CON: 37.4 ± 0.3°C INT: 35.9 ± 0.5°C (S)	CON: 3.7 ± 0.1°C/h INT: 4.0 ± 0.1°C/h (CAL)	CON: 40.2 ± 0.3°C INT: 40.1 ± 0.3°C (NS)
Yeargin et al., 2006	27°C	15	EP: 2 mile time trial	CON: No cooling (mock treatment) INT: 12 min immersion in 14°C water during recovery	Shorter run time (S)	T _{re}	CON: 37.82 ± 0.54°C INT: 37.39 ± 0.77°C (S)	–	CON: 38.87 ± 0.50°C INT: 38.59 ± 0.58°C (S)
Barr et al., 2011	49°C 12% RH	8	EPW: 20 min treadmill walk at 5 km/h, 7.5% grade	CON: No cooling INT: 15 min hand/forearm immersion during recovery	–	T _{gi}	CON: 38.3 ± 0.2°C INT: 38.0 ± 0.2°C (S)	CON: 2.7 ± 0.8°C/h INT: 2.4 ± 1.1°C/h (CAL)	–
Wilson et al., 2002	21.3°C 22.4% RH	8	EPW: 60 min cycle at 60% VO ₂ max	CON: 30 min immersion in 35°C water INT: 30 min immersion in 18°C water	–	T _{re}	CON: 36.81 ± 0.25°C INT: 36.14 ± 0.51°C (S)	–	–
Smith et al., 2013	21.6°C 20% RH	10	EC: Incremental treadmill protocol beginning at 2.7 km/h, 10% grade	CON: No cooling INT: 24 min immersion in 23°C water	Shorter time to exhaustion (S)	T _{gi}	CON: 37.1 ± 0.4°C INT: 36.6 ± 0.3°C (S)	CON: 2.0 ± 1.1°C/h INT: 1.2 ± 1.4°C/h (CAL)	CON: 37.6 ± 0.4°C INT: 36.9 ± 0.3°C (S)
Duffield et al., 2010	33°C 50% RH	8	EP: 40 min cycling time trial	CON: No cooling INT: 20 min lower body immersion in 14°C water	Greater mean power (S)	T _{re}	CON: 37.6 ± 0.3°C INT: 37.7 ± 0.3°C (REQ)	–	CON: 39.0 ± 0.4°C INT: 38.9 ± 0.3°C (REQ)
Siegel et al., 2012	34.0°C 52% RH	8	EC: Treadmill run at first ventilatory threshold to exhaustion	CON: No cooling INT: 30 min immersion in 24°C water	Longer time to exhaustion (S)	T _{re}	CON: 37.11 ± 0.28°C INT: 37.14 ± 0.34°C (REQ)	CON: 2.88 ± 0.96°C/h INT: 2.28 ± 1.56°C/h (CAL)	CON: 39.48 ± 0.36°C INT: 39.48 ± 0.34°C (NS)
Hasegawa et al., 2006	32°C 80% RH	9	EPW: 60 min cycle at 60% VO ₂ max	CON: No cooling INT: 30 min immersion in 25°C water	–	T _{re}	CON: 37.36 ± 0.15°C INT: 36.80 ± 0.30°C (REQ)	CON: 1.76 ± 0.21°C/h INT: 1.85 ± 0.48°C/h (REQ)	–

(Continued)

TABLE 3 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Castle et al., 2006	34°C 52% RH	12	EPW: 40 min intermittent cycling sprint protocol	CON: No cooling INT: 20 min immersion in 18°C water	More work done (S)	T _{re}	CON: 37.5 ± 0.1°C INT: 37.1 ± 0.1°C (S)	CON: 2.3 ± 0.3°C/h INT: 2.0 ± 0.4°C/h (CAL)	CON: 39.0 ± 0.1°C INT: 38.4 ± 0.1°C (S) (Graph)
Clarke et al., 2017	32°C 47% RH	8	EPW: 90 min treadmill run at 65% VO ₂ max	CON: 60 min rest INT: 60 min immersion in 20°C water	–	T _{re}	CON: 36.7 ± 0.3°C INT: 35.7 ± 0.9°C (S) (Graph)	CON: 1.5 ± 0.3°C/h INT: 2.1 ± °C/h (CAL)	CON: 38.9 ± 0.5°C INT: 38.8 ± 0.5°C (NS) (Graph)
Lee et al., 2018	32°C 47% RH	8	EPW: 90 min treadmill run at 65% VO ₂ max	CON: 60 min rest INT: 60 min immersion in 20°C water	–	T _{re}	CON: 36.7 ± 0.3°C INT: 35.7 ± 0.9°C (S) (Graph)	CON: 1.56 ± 0.45°C/h INT: 2.15 ± 0.72°C/h (S)	CON: 38.9 ± 0.5°C INT: 38.9 ± 0.5°C (Graph)
Skein et al., 2012	31°C 33% RH	10	EPW: 50 min self-paced intermittent sprint exercise protocol	CON: 15 min rest INT: 15 min immersion in 10°C water	Longer total sprint time (S)	T _{gl}	CON: 37.3 ± 0.2°C INT: 36.8 ± 0.4°C (S) (Graph)	–	CON: 38.9 ± 0.5°C INT: 38.7 ± 0.7°C (NS) (Graph)
Stevens et al., 2017	33°C 46% RH	9	EP: 5 km self-paced running time trial	CON: No cooling INT: 30 min immersion in 23–24°C water	Faster running time (S)	T _{re}	CON: 37.3 ± 0.3°C INT: 36.7 ± 0.4°C (S) (Graph)	CON: 3.8 ± 0.3°C/h INT: 4.7 ± 0.3°C/h (CAL)	CON: 38.9 ± 0.3°C INT: 38.6 ± 0.4°C (S) (Graph)
COLD AIR EXPOSURE									
Lee and Haymes, 1995	24°C 51–52% RH	14	EC: Treadmill run at 82% VO ₂ max to exhaustion	CON: 30 min rest in a 24°C, 53% RH room INT: 33 min rest in a 5°C, 68% RH room	Longer time to exhaustion (S)	T _{re}	–	CON: 3.86 ± 0.51°C/h INT: 3.76 ± 0.54°C/h (CAL)	CON: 38.02 ± 0.46°C INT: 37.86 ± 0.53°C (NS)
Olshewski and Bruck, 1988	18°C 50% RH	6	EC: Cycling with a constant increase in workload to exhaustion	CON: No cooling INT: Double cold air exposure before starting exercise	Longer time to exhaustion (S)	T _{oes}	–	–	CON: 38.94 ± 0.34°C INT: 38.64 ± 0.27°C (S)
COLD VEST OR ICE VEST									
Stannard et al., 2011	24–26°C 29–33% RH	8	EP: 10 km running time trial	CON: Wearing a t-shirt INT: Wearing a cooling vest for 30 min before time trial	No influence on run time	T _{gl}	CON: 37.7 ± 0.72°C INT: 37.3 ± 0.73°C (NS)	–	–
Armstrong et al., 2004	32°C 50% RH	17	EP: 5 km running time trial	CON: Wearing a t-shirt INT: Wearing an ice vest for 38 min before time trial	Shorter run time (S)	T _{oes}	CON: 37.4 ± 0.4°C INT: 37.1 ± 0.5°C (S)	–	CON: 39.8 ± 0.4°C INT: 39.7 ± 0.4°C (REQ)
Kenny et al., 2011	35°C 65% RH	10	EPW: 120 min treadmill walk at 3 miles/h, 2% grade	CON: NBC suit without ice vest INT: NBC suit with ice vest	–	T _{oes}	CON: 36.88 ± 0.13°C INT: 36.94 ± 0.25°C (NS)	CON: 1.08 ± 0.22°C/h INT: 0.90 ± 0.24°C/h (CAL)	–

(Continued)

TABLE 3 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Bogard et al., 2010	29.3°C 80% RH	8	EPW: 60 min cycle at 65% VO ₂ peak	CON: No cooling INT: Wearing an ice vest for 45 min before exercise	–	T _{re}	CON: 37.0 ± 0.2°C INT: 37.1 ± 0.2°C (NS)	CON: 2.1 ± 0.54°C/h INT: 2.0 ± 0.54°C/h (CAL)	–
Barr et al., 2011	49°C 12% RH	8	EPW: 20 min treadmill walk at 5 km/h, 7.5% grade	CON: No cooling INT: Wearing an ice vest for 15 min during recovery	–	T _{gi}	CON: 38.3 ± 0.2°C INT: 38.2 ± 0.1°C (NS)	CON: 2.7 ± 0.8°C/h INT: 2.7 ± 0.4°C/h (CAL)	–
Quod et al., 2008	34.3°C 41.2% RH	6	EP: 40 min cycling time trial	CON: No cooling INT: Wearing a cooling jacket for 40 min before exercise	No influence on cycling time	T _{re}	–	–	CON: 39.6 ± 0.4°C INT: 39.7 ± 0.5°C (REQ)
Brade et al., 2014	35°C 60% RH	12	EPW: 70 min repeat sprint protocol	CON: No cooling INT: Wearing a cooling jacket for 30 min before exercise	No influence on performance	T _{gi}	CON: 37.0 ± 0.4°C INT: 36.9 ± 0.3°C	CON: 1.6 ± 0.3°C/h INT: 1.7 ± 0.3°C/h (CAL)	CON: 38.9 ± 0.3°C INT: 38.9 ± 0.5°C
Castle et al., 2006	34°C 52% RH	12	EPW: 40 min intermittent cycling sprint protocol	CON: No cooling INT: Wearing an ice vest for 20 min before exercise	More work done (S)	T _{re}	CON: 37.5 ± 0.1°C INT: 37.3 ± 0.1°C (NS)	CON: 2.3 ± 0.3°C/h INT: 2.3 ± 0.5°C/h (CAL)	CON: 39.0 ± 0.1°C INT: 38.8 ± 0.2°C (NS)
Faulkner et al., 2015	35°C 51% RH	10	EPW: 1 h cycling time trial at 75% W _{max}	CON: No cooling INT _{COLD} : Wearing a frozen cooling garment for 30 min before exercise INT _{COOL} : Wearing a cooling garment saturated in 14°C water for 30 min before exercise	Faster time trial for COLD (S) No influence on performance for COOL	T _{gi}	CON: 36.7 ± 0.4°C INT _{COLD} : 36.5 ± 0.3°C INT _{COOL} : 36.7 ± 0.6°C (NS)	CON: 1.9 ± 0.3°C/h INT _{COLD} : 2.2 ± 0.2°C/h INT _{COOL} : 1.9 ± 0.4°C/h (CAL)	CON: 38.6 ± 0.5°C INT _{COLD} : 38.7 ± 0.4°C INT _{COOL} : 38.6 ± 0.5°C (NS)
COLD FLUID INGESTION									
Byrne et al., 2011	32°C dry bulb 60% RH 3.2 m/s air velocity	7	EP: 30 min self-paced cycling time trial	CON: 37°C fluid INT: 2°C fluid	Greater distance covered (S)	T _{re}	–	–	CON: 38.6 ± 0.5°C INT: 38.1 ± 0.3°C (NS)
Lee et al., 2008	35.0°C 60% RH	8	EC: Cycle at 65% VO ₂ peak to exhaustion	CON: Warm drink (37°C) INT: Cold drink (4°C)	Longer time to exhaustion (S)	T _{re}	CON: 36.8 ± 0.3°C INT: 36.4 ± 0.3°C (S)	CON: 3.0 ± 0.2°C/h INT: 2.9 ± 0.2°C/h (REQ)	CON: 39.4 ± 0.4°C INT: 39.5 ± 0.4°C (REQ)
ICE SLURRY INGESTION									
Siegel et al., 2012	34.0°C 52% RH	8	EC: Treadmill run at first ventilatory threshold to exhaustion	CON: Warm fluid (37°C) INT: Ice slurry mixture (–1°C)	Longer time to exhaustion (S)	T _{re}	CON: 37.11 ± 0.28°C INT: 36.70 ± 0.31°C (REQ)	CON: 2.88 ± 0.96°C/h INT: 3.60 ± 1.20°C/h (CAL)	CON: 39.48 ± 0.36°C INT: 39.76 ± 0.36°C (S)

(Continued)

TABLE 3 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Siegel et al., 2010	34.0 ± 0.2°C 54.9 ± 5.9% RH	10	EC: Treadmill run at first ventilatory threshold to exhaustion EP: Perform a set amount of work in as fast a time as possible	CON: Cold water (4°C) INT: Ice slurry (−1°C)	Longer time to exhaustion (S)	T _{re}	CON: 36.87 ± 0.11°C INT: 36.55 ± 0.16°C (REQ)	CON: 3.00 ± 0.72°C/h INT: 3.24 ± 0.48°C/h (CAL)	CON: 39.05 ± 0.37°C INT: 39.36 ± 0.41°C (S)
Stanley et al., 2010	34°C 60% RH	10	EP: Perform a set amount of work in as fast a time as possible	CON: Cold liquid beverage (18.4°C) INT: Ice-slush beverage (−0.8°C)	No influence on cycle time	T _{re}	CON: 37.4 ± 0.2°C INT: 37.0 ± 0.3°C (S)	–	CON: 39.1 ± 0.4°C INT: 39.0 ± 0.5°C (NS)
Yeo et al., 2012	28.2°C wet bulb globe temperature	11	EP: 10 km outdoor running time trial	CON: Ambient temperature drink (30.9°C) INT: Ice slurry (−1.4°C)	Faster performance time (S)	T _{gi}	CON: 37.2 ± 0.3°C INT: 36.9 ± 0.3°C (REQ)	–	CON: 39.8 ± 0.4°C INT: 40.2 ± 0.6°C (S)
Brade et al., 2014	35°C 60% RH	12	EPW: 70 min repeat sprint protocol	CON: No cooling INT: Ice slurry (0.6°C)	No influence on performance	T _{gi}	CON: 37.0 ± 0.4°C INT: 36.9 ± 0.4°C	CON: 1.6 ± 0.3°C/h INT: 1.8 ± 0.3°C/h (CAL)	CON: 38.9 ± 0.3°C INT: 39.0 ± 0.4°C
Burdon et al., 2013	32°C 40% RH	10	EP: 4 kJ/kg BM cycling time trial	CON: Thermoneutral drink (37°C) INT: Ice slurry (−1°C)	Improved cycle time	T _{re}	CON: 36.9 ± 0.2°C INT: 36.8 ± 0.3°C (NS)	CON: 5.3 ± 0.1°C/h INT: 6.2 ± 0.2°C/h (CAL)	CON: 38.7 ± 0.1°C INT: 38.7 ± 0.3°C (NS)
Gerrett et al., 2017	31°C 41% RH	12	EPW: 31 min self-paced intermittent running protocol	CON: Water (23°C) INT: Ice slurry (0.1°C)	No influence on distance covered	T _{gi}	CON: 37.2 ± 0.2°C INT: 36.7 ± 0.4°C (S)	CON: 3.3 ± 0.2°C/h INT: 3.7 ± 0.3°C/h (CAL)	CON: 38.9 ± 0.3°C INT: 38.6 ± 0.3°C (NS)
James et al., 2015	32°C 62% RH	12	EC: Running with increase workload till exhaustion	CON: No cooling INT: Ice slurry (−1°C)		T _{re}	CON: 37.21 ± 0.31°C INT: 36.94 ± 0.31°C (S)	CON: 1.11 ± 0.29°C/h INT: 1.38 ± 0.26°C/h (NS)	CON: 39.03 ± 0.45°C INT: 38.96 ± 0.55°C (NS)
Stevens et al., 2016	33°C 46% RH	11	EP: 5 km self-paced running time trial	CON: No cooling INT: Ice slurry (−1°C)	No influence on running time	T _{re}	CON: 37.2 ± 0.4°C INT: 36.9 ± 0.3°C (S)	CON: 4.4 ± 0.2°C/h INT: 4.9 ± 0.2°C/h (CAL)	CON: 39.12 ± 0.25°C INT: 39.04 ± 0.28°C (NS)
Takeshima et al., 2017	30°C 80% RH	10	EC: Cycle at 55% peak power output to exhaustion	CON: No cooling INT: Ice slurry (−1°C)	Longer run time (S)	T _{re}	CON: 37.5 ± 0.3°C INT: 37.1 ± 0.2°C (S)	CON: 2.0 ± 0.2°C/h INT: 2.1 ± 0.2°C/h (CAL)	CON: 39.2 ± 0.3°C INT: 39.2 ± 0.3°C (NS)
Zimmermann and Landers, 2015	33°C 60% RH	9	EPW: 72 min intermittent sprint protocol	CON: Water (25°C) INT: Ice slurry (−0.5°C)	No influence on performance	T _{gi}	CON: 36.7 ± 0.4°C INT: 36.0 ± 0.4°C (S)	–	CON: 38.2 ± 0.4°C INT: 37.8 ± 0.4°C (NS)
Zimmermann et al., 2017a	35°C 50% RH	10	EPW: 60 min cycling at 55% VO _{2peak}	CON: Water INT: Ice slurry	–	T _{gi}	CON: 36.7 ± 0.3°C INT: 36.2 ± 0.1°C (S)	CON: 1.3 ± 0.3°C/h INT: 1.5 ± 0.1°C/h (CAL)	CON: 38.0 ± 0.3°C INT: 37.7 ± 0.2°C (S)
Zimmermann et al., 2017b	35°C 50% RH	10	EP: 800 kJ cycling time trial	CON: Water INT: Ice slurry	No influence on cycling time	T _{gi}	CON: 37.1 ± 0.4°C INT: 36.4 ± 0.4°C (S)	CON: 1.8 ± 0.3°C/h INT: 2.5 ± 0.2°C/h (CAL)	CON: 39.0 ± 0.5°C INT: 39.0 ± 0.4°C (NS)

RH, relative humidity; EC, exercise capacity; EP, exercise performance; EPW, exercise performance at a fixed workload; S, significant; NS, not significant; CAL, calculated values; REQ, requested values; Graph, graph-extracted values; T_{re}, rectal temperature; T_{oes}, oesophageal temperature; T_{gi}, gastrointestinal temperature; CON, control; INT, intervention.

TABLE 4 | Summary of fluid ingestion studies.

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
EUHYDRATED STATE WITH LOW FLUID/AD LIBITUM vs. HIGH FLUID INTAKE									
Marino et al., 2004	31.3°C 63.3% RH 2 m/s wind speed	8	EC: Cycle at 70% peak power output to exhaustion EP: 80km cycling time trial	CON: Fluid replacement equal to half the sweat rate INT: Fluid replacement equal to sweat rate CON: Fluid ingested to replace 33% of weight lost INT: Fluid ingested to replace 100% of weight lost	No influence on cycling time No influence on cycling time	T _{re}	CON: 38.7 ± 0.4°C INT: 38.6 ± 0.5°C (REQ)	–	CON: 39.0 ± 0.4°C INT: 38.8 ± 0.6°C (NS)
Dugas et al., 2009	33°C 50% RH	6	EPW: 2h cycle at a power output equal to 62–67% maximal oxygen consumption	CON: Small (50%) fluid replacement INT: Large (80%) fluid replacement	–	T _{re}	CON: 36.8 ± 0.1°C INT: 36.9 ± 0.2°C (NS)	–	CON: 39.2 ± 0.5°C INT: 38.9 ± 0.4°C (NS)
Montain and Coyle, 1992a	33°C 50% RH 2.5 m/s wind speed	8	EPW: 2h cycle at 60% VO ₂ peak	CON: 50% fluid replacement INT: 100% fluid replacement	–	T _{oes}	CON: 37.01 ± 0.20°C INT: 37.01 ± 0.26°C (REQ)	CON: 0.60 ± 0.14°C/h INT: 0.47 ± 0.18°C/h (REQ)	–
McConell et al., 1997	21°C 43% RH	7	EPW: 2h cycle at 60% VO ₂ peak	CON: 50% fluid replacement INT: 100% fluid replacement	–	T _{re}	CON: 37.2 ± 0.2°C INT: 37.1 ± 0.2°C (REQ)	CON: 0.8 ± 0.3°C/h INT: 0.7 ± 0.1°C/h (REQ)	–
Bardis et al., 2017	AD: 31.4 ± 0.5°C PD: 31.7 ± 0.4°C (NS) 6.4 m/s	10	EPW: 3 sets of 5 km cycling at 50% maximal power output followed by 5 km cycling all out at 3% grade (Total 30 km)	CON: <i>ad libitum</i> water intake INT: Fluid ingested to replace 100% of fluid lost via sweating	Faster cycling speed (S)	T _{gi}	CON: 37.4 ± 0.1°C INT: 37.6 ± 0.2°C (NS) (Graph)	–	CON: 38.7 ± 0.4°C INT: 38.4 ± 0.4°C (S) (Graph)
James L. J. et al., 2017	34°C 50% RH 0.3–0.4 m/s	7	EPW: 15 min cycling performance test	CON: Fluid replacement to induce 2.5% body mass loss INT: Fluid replacement to replace sweat loss	More work completed (S)	T _{gi}	CON: 37.0 ± 0.2°C INT: 37.2 ± 0.3°C (Graph)	CON: 6.8 ± 1.8°C/h INT: 4.4 ± 2.3°C/h (CAL)	CON: 38.7 ± 0.5°C INT: 38.3 ± 0.5°C
Périard et al., 2014	37°C 33% RH	10	EPW: 20 min tennis match	CON: <i>ad libitum</i> water intake INT: Fluid ingested to match 70% of sweat loss	–	T _{re}	CON: 37.8 ± 0.3°C INT: 37.7 ± 0.3°C (NS) (Graph)	CON: 4.8 ± 1.75°C/h INT: 4.5 ± 2.0°C/h (CAL)	CON: 39.4 ± 0.5°C INT: 39.2 ± 0.6°C (NS)
EUHYDRATED STATE WITH NO FLUID vs. HIGH FLUID INTAKE									
Marino et al., 2004	31.3°C 63.3% RH 2 m/s wind speed	8	EC: Cycle at 70% peak power output to exhaustion	CON: No fluid replacement INT: Fluid replacement equal to sweat rate	Longer time to exhaustion (S)	T _{re}	CON: 38.8 ± 0.4°C INT: 38.6 ± 0.5°C (NS)	–	CON: 39.2 ± 0.4°C INT: 38.8 ± 0.6°C (NS)
Hargreaves et al., 1996	20–22°C	5	EPW: 2h cycle at 67% VO ₂ peak	CON: No fluid ingested INT: Ingestion of fluid to prevent loss of body mass	–	T _{re}	CON: 36.7 ± 0.2°C INT: 36.7 ± 0.4°C (NS)	CON: 0.9 ± 0.3°C/h INT: 0.6 ± 0.3°C/h (CAL)	–

(Continued)

TABLE 4 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Armstrong et al., 1997	33°C 56% RH 0.1 m/s air speed	10	EPW: 90 min treadmill walk at 5.6 km/h, 5% grade	CON: No water intake INT: <i>ad libitum</i> water intake	–	T _{re}	–	CON: 0.7 ± 0.2°C/h INT: 0.6 ± 0.2°C/h (CAL)	–
Robinson et al., 1995	20°C 60% RH 3 m/s air speed	8	EP: 60 min cycle to achieve greatest possible distance	CON: No fluid ingested INT: Ingestion of fluid to replace approximate sweat loss	Less distance covered (S)	T _{re}	CON: 36.8 ± 0.3°C INT: 36.5 ± 0.6°C (NS)	–	CON: 38.6 ± 0.6°C INT: 38.1 ± 0.6°C (NS)
Fallowfield et al., 1996	20°C	8	EC: Treadmill run at 70% VO ₂ max to exhaustion	CON: No fluid ingested INT: Fluid replacement before and during exercise	Longer time to exhaustion (S)	T _{re}	–	–	CON: 38.8 ± 1.1°C INT: 39.1 ± 0.6°C (NS)
Coso et al., 2008	36°C 29% RH 1.9 m/s airflow	7	EPW: 120 min cycle at 63% VO ₂ max	CON: No fluid ingested INT: Ingestion of mineral water	–	T _{re}	CON: 37.6 ± 0.3°C INT: 37.6 ± 0.3°C (NS)	CON: 0.9 ± 0.2°C/h INT: 0.6 ± 0.2°C/h (CAL)	–
Cheung and McLellan, 1997	40°C 30% RH	8	EC: Either a light (3.5 km/h, 0% grade) or a heavy (4.8 km/h, 4% grade) treadmill walk to exhaustion	CON: No fluid replacement INT: Fluid replacement	Longer time to exhaustion (S) for light exercise	T _{re}	CON _{light} : 36.89 ± 0.29°C INT _{light} : 36.85 ± 0.28°C (NS) CON _{heavy} : 36.88 ± 0.21°C INT _{heavy} : 36.94 ± 0.27°C (NS)	CON _{light} : 1.19 ± 0.46°C/h INT _{light} : 1.15 ± 0.32°C/h (CAL) CON _{heavy} : 1.88 ± 0.32°C/h INT _{heavy} : 1.76 ± 0.42°C/h (CAL)	CON _{light} : 38.74 ± 0.68°C INT _{light} : 38.90 ± 0.40°C (NS) CON _{heavy} : 38.71 ± 0.43°C INT _{heavy} : 38.69 ± 0.62°C (NS)
Munoz et al., 2012	33°C 30% RH	10	EP: 5 km running time trial	CON: No rehydration INT: Oral rehydration	No influence on performance time	T _{re}	CON: 37.78 ± 0.41°C INT: 37.57 ± 0.31°C (NS)	–	CON: 39.19 ± 0.45°C INT: 38.97 ± 0.36°C (NS)
Kay and Marino, 2003	33.2°C 63.3% RH	7	EP: 60 min cycle to achieve greatest possible distance	CON: No fluid ingested INT: Fluid ingested to prevent any change in body mass	No influence on distance cycled	T _{re}	–	–	CON: 38.9 ± 0.5°C INT: 38.7 ± 0.4°C (NS)
Dugas et al., 2009	33°C 50% RH	6	EP: 80 km cycling time trial	CON: No fluid ingested INT: Fluid ingested to replace 100% of weight lost	No influence on cycling time	T _{re}	CON: 36.8 ± 0.2°C INT: 36.9 ± 0.2°C (NS)	–	CON: 39.2 ± 0.4°C INT: 38.9 ± 0.4°C (NS)
Hasegawa et al., 2006	32°C 80% RH	9	EPW: 60 min cycle at 60% VO ₂ max	CON: No water intake INT: Water ingestion at 5 min intervals	–	T _{re}	CON: 37.37 ± 0.15°C INT: 37.37 ± 0.16°C (REQ)	CON: 1.77 ± 0.22°C/h INT: 1.39 ± 0.27°C/h (REQ)	–

(Continued)

TABLE 4 | Continued

Study	Ambient conditions	N =	Exercise protocol	Intervention method	Exercise outcome	T _c measure	T _c before	T _c rate of rise	T _c end
Gagnon et al., 2012	42°C 20% RH 1 m/s air speed	8 untrained 8 trained	EPW: 120 min cycle at 120 W	CON: No fluid replacement INT: Fluid replacement	–	T _{oes}	CON _{UT} : 37.23 ± 0.57°C INT _{UT} : 36.96 ± 0.25°C	CON _{UT} : 0.74 ± 0.28°C/h INT _{UT} : 0.70 ± 0.18°C/h	–
Montain and Coyle, 1992b	33°C 50% RH 2.5 m/s wind speed	8	EPW: 2 h cycle at a power output equal to 62–67% maximal oxygen consumption	CON: No fluid replacement INT: Large (80%) fluid replacement	–	T _{oes}	CON: 36.99 ± 0.36°C INT: 37.01 ± 0.26°C (REQ)	CON: 0.84 ± 0.24°C/h INT: 0.47 ± 0.18°C/h (REQ)	–
McConell et al., 1997	21°C 43% RH	7	EPW: 2 h cycle at 60% VO ₂ peak	CON: No fluid replacement INT: 100% fluid replacement	–	T _{re}	CON: 37.1 ± 0.2°C INT: 37.1 ± 0.2°C (REQ)	CON: 1.0 ± 0.2°C/h INT: 0.7 ± 0.1°C/h (REQ)	–
Wall et al., 2015	33°C 40% RH 32 km/h	10	EPW: 25 km cycling time trial	CON: No fluid replacement INT: 100% fluid replacement	No influence on cycling time	T _{re}	CON: 37.1 ± 0.2°C INT: 37.0 ± 0.2°C (NS)	CON: 2.6 ± 0.5°C/h INT: 2.49 ± 0.53°C/h (CAL)	CON: 38.9 ± 0.3°C INT: 38.7 ± 0.3°C (S)
Wittbrodt et al., 2015	32°C 65% RH	12	EPW: 50 min cycling at 60% VO ₂ peak	CON: No fluid intake INT: 100% fluid replacement	–	T _{re}	CON: 37.0 ± 0.3°C INT: 36.8 ± 0.8°C (NS)	CON: 1.4 ± 0.7°C/h INT: 1.0 ± 1.3°C/h (CAL)	CON: 38.2 ± 0.5°C INT: 37.6 ± 0.7°C (S)
Trangmar et al., 2015	35% 50% RH	8	EC: Cycling at 60% VO ₂ max until volitional exhaustion	CON: No fluid intake INT: Fluid intake to replace body mass loss	Shorter exercise duration (S)	T _{gi}	CON: 37.4 ± 0.1°C INT: 37.3 ± 0.1°C (NS)	–	CON: 38.7 ± 0.1°C INT: 38.2 ± 0.2°C (S)
HYPOHYDRATED STATE WITH NO FLUID vs. HIGH FLUID INTAKE									
Armstrong et al., 1997	33°C 56% RH 0.1 m/s air speed	10	EPW: 90 min treadmill walk at 5.6 km/h, 5% grade	CON: No water intake INT: <i>ad libitum</i> water intake	–	T _{re}	–	CON: 1.2 ± 0.2°C/h INT: 0.7 ± 0.2°C/h (CAL)	–

RH, relative humidity; EC, exercise capacity; EPW, exercise performance at a fixed workload; S, significant; NS, not significant; CAL, calculated values; REQ, requested values; Graph, graph-extracted values; T_{re}, rectal temperature; T_{oes}, oesophageal temperature; T_{gi}, gastrointestinal temperature; CON, control; INT, intervention.

is some form of training in the simulated laboratory settings or in the natural environmental settings. Few studies have attempted to separate the effects of heat acclimation from aerobic fitness. A study by Ravanelli et al. (2018) showed that a greater maximum skin wittedness occurred at the end of aerobic training in temperate conditions (22°C, 30% relative humidity), and this was further augmented by heat acclimation in a hot and humid condition (38°C, 65% relative humidity). This suggests that studies that include aerobic training in the heat acclimation/acclimatization protocols may have had their thermoregulatory effects augmented. However, as there have been few studies that have isolated the effects of heat acclimation/acclimatization from aerobic training or compared exertional vs. passive exposure to heat in heat acclimation/acclimatization protocols, it would be difficult to isolate the effects of heat acclimation/acclimatization from aerobic fitness.

Pre-exercise Cooling

The main intention of pre-exercise cooling is to lower T_c before exercise to extend heat storage capacity in hope to delay the onset of fatigue and in this review, we have observed pre-exercise cooling to be most effective in this aspect compared to the other heat mitigation strategies. For comprehensive reviews on pre-exercise cooling (see Marino, 2002; Quod et al., 2006; Duffield, 2008; Jones et al., 2012; Siegel and Laursen, 2012; Wegmann et al., 2012; Ross et al., 2013). The various pre-exercise cooling methods include cold water immersion (Booth et al., 1997; Kay et al., 1999), cold air exposure (Lee and Haymes, 1995; Cotter et al., 2001), cold vest (Arngrimsson et al., 2004; Bogerd et al., 2010), cold fluid ingestion (Lee et al., 2008; Byrne et al., 2011), and ice slurry ingestion (Siegel et al., 2010; Yeo et al., 2012).

Largely, the methods above have been shown to be effective in lowering T_c pre-exercise, which could consequently reduce thermal strain and therefore enhance endurance performance. Apart from lowering T_c pre-exercise, ice slurry ingestion has shown to increase T_c at the end of exercise. In both laboratory and field studies, T_c was higher at the end of exercise with ice slurry. In the laboratory study by Siegel et al. (2010) oesophageal temperature was higher by 0.31°C, and in the field study by Yeo et al. (2012), gastrointestinal temperature was higher by 0.4°C with the ingestion of ice slurry. Siegel et al. (2010) suggested that the ingestion of ice slurry may have affected thermoreceptors present causing a “physiologically meaningful reduction in brain temperature.” In addition, ice slurry ingestion may have potentially attenuated any afferent feedback that would have resulted in central reduction in muscle activation, allowing tolerance of a greater thermoregulatory load (Lee et al., 2010).

In addition, practitioners should consider the magnitude of pre-exercise cooling strategies being employed. Large volumes of ice slurry/cold water ingestion may blunt heat loss pathways by limiting sweat gland activity. This would reduce evaporative heat loss which may counteract to cause a greater heat storage and higher T_c during exercise which would be unfavorable (Ruddock et al., 2017). However, it should be noted that this potentially negative effect of ice slurry/cold water ingestion may be a greater concern in dry environments as compared to

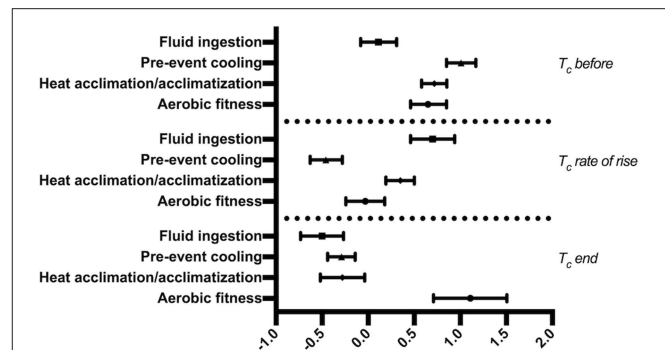


FIGURE 2 | Forest plot of Hedges' g weighted averages of heat mitigation strategies effect on T_c at different points.

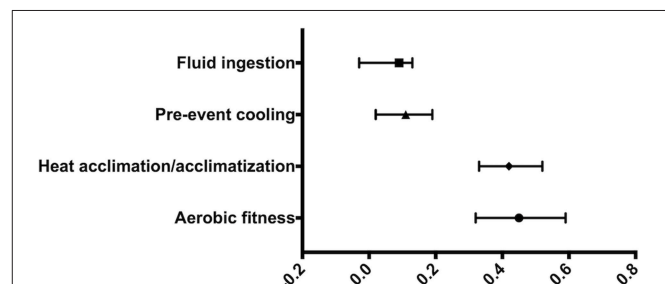


FIGURE 3 | Forest plot of combined Hedges' g weighted averages of heat mitigation strategies.

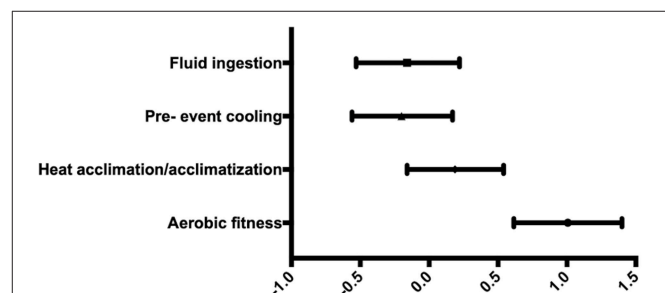


FIGURE 4 | Forest plot of Hedges' g weighted averages of heat mitigation strategies on endurance.

humid environments. In hot and humid environments, despite reductions in evaporative heat loss potential, actual evaporation may not be reduced, and ice slurry/cold water ingestion would still be beneficial in reducing body heat storage. This is due to the attainment of the maximum evaporation potential anyway, and any additional sweat generated would drip off the skin in hot and humid environments (Jay and Morris, 2018). Numerous studies also support the effectiveness of pre-exercise ice slurry/cold water ingestion in lowering T_c and demonstrate that this profile is continued during exercise (Lee et al., 2008; Siegel et al., 2010, 2012; Byrne et al., 2011; Yeo et al., 2012).

TABLE 5 | Ranking of heat mitigation strategies based on Hedges' g weighted averages.

	Combined Hedge's g weighted averages effect on T_{c}	Rank	Combined Hedge's g weighted averages effect on performance and/or capacity	Rank
Aerobic Fitness	0.45	1	1.01	1
Heat acclimation/acclimatization	0.42	2	0.19	2
Pre-exercise cooling	0.11	3	-0.20	4
Fluid ingestion	0.09	4	-0.16	3

The effectiveness of pre-cooling as a strategy in altering T_{c} may be limited as it is mostly done acutely before exercise. As such, its benefit may not be able to be sustained throughout the exercise duration. To counteract this limitation, considerations can be made to consider per/mid-exercise cooling. Whilst not discussed in the present meta-analysis, previous reviews have shown that per/mid-exercise cooling may be as effective in enhancing exercise performance in hot environments (Bongers et al., 2015, 2017).

Fluid Ingestion

Fluid ingestion is a common strategy used to reduce thermoregulatory strain in the heat. Many studies have shown that when fluid is ingested during exercise, exercise capacity and performance are enhanced (Fallowfield et al., 1996; Cheung and McLellan, 1997; Marino et al., 2004). A more controversial issue is the optimal amount of fluid to be consumed during exercise. Two dominant viewpoints exist—the first is that athletes should prevent fluid loss of $>2\%$ body mass (Sawka et al., 1985; Montain and Coyle, 1992a; Sawka and Coyle, 1999; Casa et al., 2010), while the other recommends drinking *ad libitum* (Noakes, 1995; Beltrami et al., 2008; Lee et al., 2011) due to an increased prevalence of exercise associated hyponatremia, commonly referred to as water intoxication (Noakes, 1995). Even in warm conditions where sweat rates are high, the behavioral drive to ingest fluids could exceed the physiological sweat loss (Lee et al., 2011).

This review analyzed the effects of a (i) low fluid/*ad libitum* vs. high fluid intake and (ii) no fluid vs. high fluid intake on T_{c} . All participants began exercise in a euhydrated state. Dugas et al. (2009) found that *ad libitum* drinking while cycling replaces approximately 55% of fluid losses., while Daries et al. (2000) found that *ad libitum* drinking during a treadmill run replaces approximately 30% of fluid losses. Hence in this evaluation, a fluid intake trial replacing closest to $\sim 45\%$ of fluid losses was chosen to represent the low fluid/*ad libitum* condition. It should also be stated that the results in trials in which the control state was no fluid intake may have exaggerated the results of fluid ingestion seen in this meta-analysis. This is especially so when we consider that it is impractical during a competition event to avoid drinking. As such, future hydration studies should consider avoiding a “No fluid” control state.

Ideally, individuals should begin their exercise in a euhydrated state. This could be achieved by drinking 6 mL of water per kg body mass for 2–3 h pre-exercising in a hot environment (Racinais et al., 2015a). During exercise, fluid is largely

lost through sweating. Sweat rates may vary depending on individual characteristics, environmental conditions and heat acclimation/acclimatization status (Cheuvront et al., 2007). Practitioners should therefore consider determining their sweat rate prior to exercising in a hot environment to determine the amount of rehydration or fluid intake that is necessary to reduce physiological strain and optimize performance, without increasing body weight. Considerations can also be made to include supplementation with sodium (Casa, 1999; Sawka et al., 2007) and glucose (von Duvillard et al., 2007; Burke et al., 2011).

PRACTICAL IMPLICATIONS

Logically, employing a combination of all the different heat mitigation strategies would be most beneficial in extending an athlete's heat storage capacity and in optimizing exercise performance in the heat. However, due to time and resource constraints, it may not be practical for athletes and coaches to employ all these strategies for competition. By knowing which heat mitigation strategy is most effective, an informed decision can be made. Strategies such as aerobic fitness and heat acclimation/acclimatization have to be conducted months and weeks respectively before competition in order to reap its benefits. On the other hand, strategies such as pre-exercise cooling and fluid ingestion can be done immediately before or during competition. Practicality and comfort should be the main focus when deciding which heat mitigation strategy to employ. For example, pre-exercise cooling methods such as cold water immersion may be effective in lowering T_{c} before exercise begins. However, it may be cumbersome to set up a cold water bath especially during outdoor field events. Furthermore, being immersed in a cold water bath may be an uncomfortable experience for some athletes, and may cool the muscles prior to the event and hence is not practical to be used prior to competition (Quod et al., 2006; Ross et al., 2013). It is noteworthy that there could be inter-individual differences when employing each of these heat mitigation strategies. Athletes and coaches are advised to experiment with these strategies during training before deciding on the appropriate strategy to employ during competition. Finally, the importance of the usage of heat mitigation strategies when competing in hot and humid environments cannot be stressed enough. From this meta-analysis, we have shown that aerobic fitness is the most effective heat mitigation strategy. However, this does not understate the importance of a combination of heat mitigation strategies, nor does it reflect that should an athlete be aerobically fit,

other heat mitigation strategies are not necessary. In the 15th International Association of Athletics Federations (IAAF) World Championships held in Beijing (China), mean and maximal temperatures were anticipated to be 26° and 33°C respectively, with relative humidity of ~73%. Despite the expected hot and humid conditions, only 15% of athletes reported having specifically prepared for these conditions. Of these, females and athletes with previous history of exertional heat illnesses (EHI) were more likely to adopt heat mitigation strategies (Périard et al., 2017). Although <2% experienced EHI symptoms, athletes should be more aware of the potential benefits of using one or more heat mitigation strategies in the lead up to competitions in hot and humid environments. As global temperatures continue to rise, the importance of such heat mitigation strategies in enhancing performance and in reducing the likelihood of EHI cannot be understated.

LIMITATIONS

The methodology of using a meta-analysis to evaluate effectiveness of different strategies is not without limitation. Publication and language restriction bias may have affected the number of studies that could be included in the analysis. As such, care was taken to ensure to control for such biases, such as a manual tracking of review articles to ensure that studies that were relevant but that did not show up in the initial search of the databases could be included as well. The heterogeneity of the included studies was also controlled for by statistical analysis. In addition, due to the practical difficulty in blinding the participants to the heat mitigation strategy being employed, any beneficial effect arising from the placebo effect could not be eliminated.

This meta-analysis also did not include behavioral alterations that could be undertaken as a mitigation strategy against exertional heat stress. Taking regular breaks during exercise is an effective way to minimize heat strain by preventing an excessive rise of T_{c} and increasing exercise tolerance in the heat (Minett et al., 2011). Individuals should also avoid exercising during the hottest part of the day. Alternatively, several shorter sessions of exercise can be performed rather than having a single long session, to reduce hyperthermia, while maintaining the quality of the exercise session (Maughan and Shirreffs, 2004). When exercising in the heat, an important consideration is to ensure that the material in the clothing does not prevent the

evaporation of sweat from the skin (Maughan and Shirreffs, 2004). Furthermore, black and dark-colored clothing absorb more heat and should not be worn when exercising in the heat. For a review of the thermal characteristics of clothing (see Gonzalez, 1988; Parsons, 2002). One reason for the exclusion is that there is often time pressure to complete a task or race as fast as possible and/or in certain attire that does not permit behavioral alteration during competitions. There are also few studies that looked at the effect of behavioral alterations on endurance that fulfilled our inclusion criteria, which did not allow for the calculation of an effect size to compare effectively with the other heat mitigations strategies.

Although these limitations should be accounted for, this is the first meta-analysis to compare several different heat mitigation strategies and their effects on T_{c} and endurance. As such, this meta-analysis could provide the information necessary to allow for more informed decision making by coaches, athletes and sports scientists during exercise in hot and/or humid environments.

CONCLUSION

In conclusion, aerobic fitness was found to be the most effective heat mitigation strategy, followed by heat acclimation/acclimatization, pre-exercise cooling and lastly, fluid ingestion. The similarity in ranking between the ability of each heat mitigation strategy to favorably alter T_{c} and affect endurance suggest that alteration of heat strain may be a key limiting factor that contributes to endurance. This analysis has practical implications for an athlete preparing for competition in the heat and also allows coaches and sport scientists to make a well-informed and objective decision when choosing which heat mitigation strategy to employ.

AUTHOR CONTRIBUTIONS

SA and PT realized the research literature. SA, PT, and JL contributed to the writing of the manuscript.

FUNDING

Funds received for open access publication fees were obtained from Defence Innovative Research Program Grant No. 9015102335, Ministry of Defence, Singapore.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Transcranial Direct Current Stimulation With Halo Sport Enhances Repeated Sprint Cycling and Cognitive Performance

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OPEN ACCESS

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 23 July 2018

Accepted: 30 January 2019

Published: 14 February 2019

Citation:

Huang L, Deng Y, Zheng X and
Liu Y (2019) Transcranial Direct
Current Stimulation With Halo Sport
Enhances Repeated Sprint Cycling
and Cognitive Performance.
Front. Physiol. 10:118.
doi: 10.3389/fphys.2019.00118

The present study investigated the effects of transcranial direct current stimulation (tDCS) using the Halo Sport device on repeated sprint cycling ability and on cognitive performance. In this triple-blind, randomized, sham-controlled study, nine physically active participants received either a placebo stimulation (Sham) or real stimulation (Halo) for 20 min. Participants then performed 5 × 6-s sprints interspersed with 24 s of active recovery on a cycle ergometer. Peak and mean power output were measured for each sprint. In addition, cognitive performance in terms of reaction time (RT) and accuracy (ACC) was assessed via Stroop test pre- and post-stimulation. There was a significant interaction for mean power output [$F(4,32) = 2.98$, $P = 0.03$]. A main treatment effect was observed in all of the repeated sprints apart from the initial one. Halo did not affect RT in either the congruent or incongruent condition but did increase ACC in the incongruent condition [$F(1,8) = 10.56$, $P = 0.012$]. These results suggest that tDCS with the Halo Sport system is able to enhance aspects of sprint cycling ability and cognitive performance.

Keywords: tDCS, Halo Sport, exercise performance, cognition, Stroop test

INTRODUCTION

Non-invasive electrical brain stimulation is an emerging technique that claims to improve training effects and boost exercise performance. The rationale for such effects is based on the ability of the stimulation to safely modulate brain excitability and functional plasticity (Angius et al., 2017). The Halo Sport device is a commercial system that consists of a headset similar to conventional headphones. Halo Sport uses transcranial direct current stimulation (tDCS) in which weak direct currents (DC) below 2–3 mA is delivered for a period of minutes over the scalp through surface electrodes, termed primers, with the intention of inducing changes in both sides of the motor cortex.

The primary motor cortex (M1) is a complex network of interconnected localized groups of neurons with similar inputs and outputs, aimed to control movements (Schieber, 2001). The role of the M1 is to generate neural impulses that control the execution of movement (Moscatelli et al., 2016a). It is claimed that Halo Sport produces changes in motor cortex excitability. Therefore, Halo Sport may improve exercise performance. One possible mechanism is that the electrical stimulation induces increases in intracortical facilitation and motor cortex excitability, allowing motor-cortex

neurons to build neural connections more easily, enhancing motor drive to the muscles (Hornyak, 2017).

Halo Sport has been used in training and competition, but its effects on physical performance remain elusive. Early studies investigated the effect of tDCS on physical performance using single joint isometric exercise (Cogiamanian et al., 2007). However whole-body exercise better represents real sporting competition than single joint exercise and therefore cycling performance is likely to be more suitable for assessing the ergogenic effect of tDCS. Anodal tDCS applied to M1 of healthy volunteers has been reported to enhance cycling performance (Okano et al., 2015; Vitor-Costa et al., 2015; Angius et al., 2016) and similar effects may be expected for the Halo Sport device. However, no study to date has examined whether Halo Sport applied over the motor cortex is able to enhance cycling performance.

Excellence in sport performance requires not only physical and motor capabilities, but also sensory-cognitive skills (Moscatelli et al., 2016b). Halo Sport is thought to act as a central nervous stimulant, and it may affect cognitive and psychomotor functioning during exercise. To date, no studies have examined the effect of Halo Sport on cognition. Anodal tDCS applied to the dorsolateral prefrontal cortex (DLPFC) of healthy volunteers has been reported to enhance the cognition (Dockery et al., 2009; Stone and Tesche, 2009; Zaehle et al., 2011) and similar effects may be expected for the Halo Sport device. Moreover, other studies have reported that tDCS is a central nervous stimulant and has positive effects on cognitive functioning by affecting perception and attention (Shin et al., 2015). This finding could suggest that the performance-enhancing effects of tDCS are due to altered central nervous system function, possibly related to the attenuation of central fatigue effects (Vitor-Costa et al., 2015).

The primary aim of the present study was to examine the effects of Halo Sport on repeated sprint and cognitive performance. It was hypothesized that Halo Sport would improve repeated cycle sprint performance and cognitive function.

MATERIALS AND METHODS

Participants

Participants were deemed eligible using the following criteria: (1) age between 18 and 30 years; (2) males; (3) no diagnosis of neurological, or psychiatric disorders; (4) no history of drug or alcohol abuse; (5) not enrolled in another trial involving weight training; and (6) being physically active (practicing physical activities at least three times a week for at least 6 months; Vitor-Costa et al., 2015). Nine males (age, 20 ± 1.2 years; height, 176.8 ± 6.6 cm; mass, 73.1 ± 6.5 kg) volunteered to participate in the exercise trial. All participants were fully informed of the nature and possible risks of the study before giving written consent. The local ethical committee of Shanghai University of Sport approved the experimental protocol.

Study Design

This study was a single blinded, randomized, placebo-controlled, crossover study with a repeated measures design. The subjects visited the laboratory twice. Written informed consent was obtained from all participants before study enrolment. On the day of the experiment, participants were asked to go to the toilet and empty their bladder, then they had their body mass and height measured. Subjects were seated in a comfortable chair for the cognitive tasks (Stroop tasks). Stroop tasks consisted of one practice trial and one baseline (Stroop pre). Following that, participants received a 20-min Halo Sport session either with (Halo) or without (Sham) electrical current delivered to the primers. During stimulation, subjects were seated, closed eyes, kept the same posture and quiet. All subjects received all stimulation conditions. The two experimental trials were separated by 5 days in a counterbalanced order, and conducted at the same time of day to eliminate any effects of circadian variations. At the end of the Halo Sport session, the subjects walked to a cycle simulator and started the exercise protocol. The cycle sprint exercise was based on a previously reported reliable protocol (McGawley and Bishop, 2006). Briefly, after a 5-min warm-up on a calibrated Monark cycle ergometer, the participant was then required to pedal at 50 rpm before being given a verbal countdown to commence a 6-s maximal sprint effort with a resistance of 10% of body mass applied to the front wheel. Five 6-s sprints were completed, with 24 s of unloaded pedaling between each effort. The peak power output and mean power output were recorded in each 6-s loaded sprint. Finally, the Stroop tasks were repeated (Stroop post). The protocol is shown in Figure 1.

Halo Sport Procedures

Halo Sport is a commercial tDCS device made by[®] Halo Neuroscience (San Francisco, CA, United States). In addition,[®] Halo Neuroscience provided permission for their name and equipment to be used in this publication in our study. Halo Sport is designed as a self-contained headset similar in appearance to audio headphones. Three studded foam electrodes termed primers ($24 \text{ cm}^2/\text{primer}$), which are wetted prior to use, make the electrical contact with the head. As with normal headphones, Halo Sport needs to be positioned over the vertex of the head. In this position, the primers lie across the top of the head, spanning from ear to ear, with the aim of stimulating both sides of the motor cortex. The electrodes are connected to a continuous current electric stimulator, driven by a Lithium-ion (LiPo) cell (36 V). The maximum energy output was 2.2 mA and was controlled by the Halo application which was set using an iPhone or iPad.

Participants reclined in a chair, in resting state. The Halo Sport headset was correctly positioned on the head of subjects, and the electrical current was ramped up to 2.0 mA over the course of 30 s. In the active Halo Sport group, the current intensity was maintained at this level for 20 min, whereas in the sham Halo Sport group it was ramped-down after 30 s. This stimulation



FIGURE 1 | Time line of one experimental trial. Stroop fam, familiarization trials; Stroop pre, Stroop task at baseline; Stroop post, Stroop test after Halo Sport stimulation.

procedure is similar to that used in previous studies of tDCS (Gandiga et al., 2006).

Stroop Task

The Stroop test is a classical assessment that measures multiple aspects of cognitive function, including information processing speed, sustaining attention, interference, and inhibition. It is also a neuropsychological assessment that is recommended in research regarding exercise and cognition (Chang et al., 2015). It is sensitive to interference and the ability to suppress an automated response. The Stroop task was programmed and performed on E-prime 1.0 software (Psychology Software Tools, Pittsburgh, PA, United States). This task consisted of two conditions. The congruent condition included three Chinese color words (i.e., 绿 for green, 蓝 for blue, and 红 for red) that were displayed in the same color (e.g., “green” displayed in a green font), whereas the incongruent condition included the same three color names but each was displayed in a different color (e.g., “green” displayed in a blue or red font). Subjects had to identify the display color of the word, and the reaction time (RT) and accuracy in doing so was recorded.

Each trial included a fixed cross presented on the center of the screen with 500 ms, followed by a stimulus that was also presented for 500 ms. Participants performed two blocks of 120 trials consisting of congruent trials (trials, $n = 60$) and incongruent trials (trials, $n = 60$) presented in a random order. To avoid the participants’ expectation to stimuli, the interval between the fixed cross and the stimulus presentation was randomly varied between 300 and 800 ms, and the inter-stimulus interval (ISI) was 1500 ms. The RT and accuracy (ACC) were recorded to evaluate Stroop performance. In addition, we used the “interference index” in the Stroop effect as one index to evaluate Stroop performance. The “interference index” was calculated via RT of the incongruent condition minus RT of the congruent condition.

Statistical Analyses

Statistical analyses were conducted with SPSS v. 20 (IBM, United States). Alterations in peak power output, mean power output, RT, ACC and interference index of Stroop effect were assessed via two-way (treatment \times time) repeated-measures ANOVAs. Significant main or interaction effects were followed by appropriate *post hoc* analyses with LSD. Between-stimulation differences in mean peak power output and mean power output were analyzed using a paired-sample *t*-test. The magnitudes of differences in the changes in mean peak power output, in mean power output, and in interference index of Stroop effect between treatments were calculated

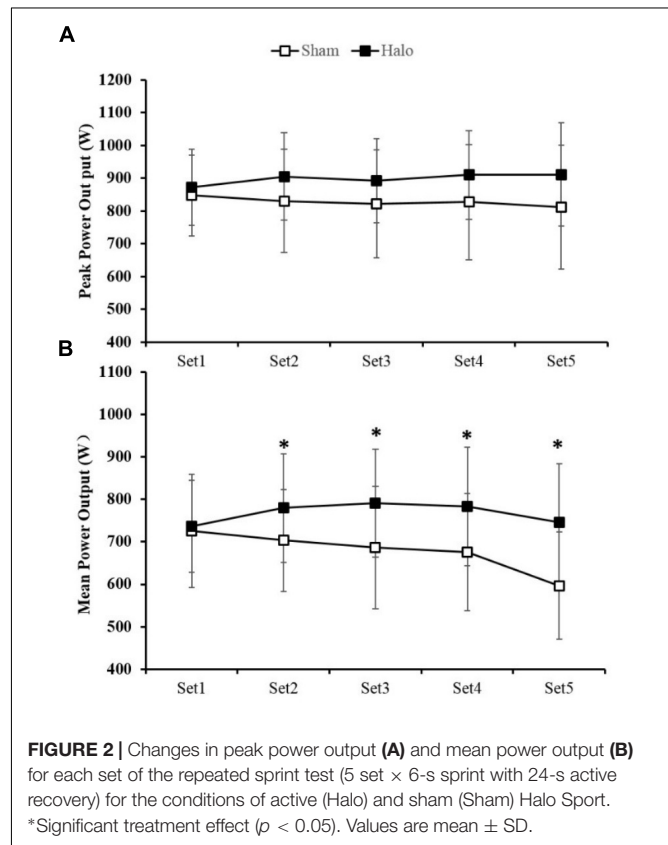


FIGURE 2 | Changes in peak power output (A) and mean power output (B) for each set of the repeated sprint test (5 set \times 6-s sprint with 24-s active recovery) for the conditions of active (Halo) and sham (Sham) Halo Sport. *Significant treatment effect ($p < 0.05$). Values are mean \pm SD.

as Cohen’s effect size (ES). The criteria to interpret the magnitude of ES were as follows: <0.2 , trivial; 0.2 – 0.5 , small; 0.5 – 0.8 , moderate; and >0.8 , large (Cohen, 1992). Data are presented as mean \pm SD. Statistical significance was accepted at $P < 0.05$.

RESULTS

Repeated Sprint Ability Peak Power Output

Figure 2A summarizes changes in peak power every sprint set for each treatment. A 2×5 mixed ANOVA revealed that there was no significant interaction for peak power output [$F(4,32) = 0.91$, $P = 0.47$]. A trend for greater mean peak power output following Halo Sport was observed (Sham: 827.8 ± 145.3 W; Halo: 898.3 ± 116.3 W; $P = 0.07$). Compared with the Sham group, Halo Sport

stimulation showed a moderate effect on mean peak power output ($ES = 0.53$).

Mean Power Output

Figure 2B shows changes in mean power every sprint set for each treatment. A 2×5 mixed ANOVA revealed that there was a significant interaction for mean power output [$F(4,32) = 2.98$, $P = 0.03$]. A main treatment effect was observed in Set 2 (Sham: 703.4 ± 128.5 W; Halo: 779.8 ± 128.1 W; $P < 0.05$), 3 (Sham: 686.9 ± 154.5 W; Halo: 791.5 ± 127.4 W; $P < 0.05$), 4 (Sham: 676.1 ± 147.8 W; Halo: 783.5 ± 139.0 W; $P < 0.05$) and 5 (Sham: 596.6 ± 134.8 W; Halo: 745.3 ± 139.1 W; $P < 0.05$). Compared with Sham group, Halo Sport stimulation showed a moderate effect on mean power output ($ES = 0.60$).

Stroop Test Reaction Time

For the incongruent condition, a 2×2 mixed ANOVA revealed that there was a significant main effect of RT [$F(1,8) = 17.68$, $P = 0.003$, **Table 1**], with shorter RTs after stimulation. However, main effects for treatment and the interaction of treatment by time were not significant [$F(1,8) = 0.047$, $P = 0.83$]. For the congruent condition, we also found a significant main effect of RT, $F(1,8) = 5.69$, $P = 0.04$, with, again, shorter times observed in the post-stimulation test. However, main effects for treatment and the interaction of treatment by time were not significant, $F(1,8) = 0.48$, $P = 0.51$ (**Table 1**).

Accuracy

For the incongruent condition, a 2×2 mixed ANOVA revealed that there was a significant interaction for ACC [$F(1,8) = 10.56$, $P = 0.01$, **Table 1**]. ACC was significantly decreased after stimulation in the Sham group (Pre: 0.91 ± 0.05 ; Post: 0.88 ± 0.06 ; $P < 0.05$). In the Halo group, ACC was significantly increased after stimulation (Pre: 0.87 ± 0.07 ; Post: 0.92 ± 0.05 ; $P < 0.05$). However, for the congruent condition, a 2×2 mixed ANOVA revealed that there was a significant main effect of ACC ($F = 9.59$, $P = 0.015$, **Table 1**), where an increase in ACC was observed in the post-stimulation test. However, main effects for treatment and the interaction of treatment by time were not significant, $F(1,8) = 0.96$, $P = 0.36$.

Stroop Effect

With respect to the “interference index” in the Stroop effect, no significant interaction was found (**Table 1**). Compared with pre-stimulation, Sham showed a trivial effect ($ES = 0.13$), and Halo showed a small effect ($ES = 0.43$).

DISCUSSION

This is a novel study to show the effects of tDCS using the Halo Sport device on repeated sprint cycling ability and on cognitive performance. We found that tDCS with the Halo Sport device improved repeated sprint cycling power output and Stroop performance.

Interest in the possible ergogenic effect of non-invasive brain stimulation is growing and whilst there are a number of studies looking at tDCS there are few reports specifically concerning the Halo Sport device. Early studies investigated the effect of tDCS on physical performance using single joint isometric exercise (Cogiamanian et al., 2007). However whole-body exercise better represents real sporting competition than single joint exercise and therefore cycling performance is likely to be more suitable for assessing the ergogenic effect of tDCS. Of those studies that have examined the effect of tDCS on physical performance in cycling, the evidence is inconsistent (Angius et al., 2015, 2016; Okano et al., 2015; Vitor-Costa et al., 2015; Barwood et al., 2016). Okano et al. (2015) reported that 2 mA for 20 min of anodal tDCS targeting the temporal cortex enhanced maximal power output by about 4%. On the other hand, using similar methodology as Okano et al. (2015); Barwood et al. (2016) observed that following 20-min of anodal tDCS at 1.5 mA over the left temporal cortex, 20 km cycling time trial performance was unaffected. In addition, they also found no effect of 20-min of tDCS at 2.0 mA on exercise performance in the heat. Such inconsistencies indicate that the effects of tDCS may be dependent on a range of factors including experimental environment, stimulation duration and intensity, and electrode configuration and position on the head. The Halo Sport device is one commercial form of tDCS and any effects it produces may be affected by such factors (Angius et al., 2017).

The present study is the first to provide evidence that Halo Sport is able to improve cycling performance. We found that 20 min of stimulation at 2mA with Halo Sport significantly enhanced the mean power output during cycling sprints. In

TABLE 1 | The reaction time, interference index, and accuracy rate of the Stroop test.

		Sham group		Halo group	
		Pre	Post	Pre	Post
Reaction time	Incongruent (ms)	636.78 \pm 54.65	602.21 \pm 51.04 [#]	652.96 \pm 72.18	613.65 \pm 65.81 [#]
	Congruent (ms)	581.21 \pm 21.78	568.39 \pm 28.78 [#]	586.53 \pm 32.17	565.35 \pm 39.73 [#]
Interference index (ms)		38.89 \pm 43.95	33.82 \pm 30.64	66.43 \pm 48.95	48.30 \pm 33.14
Accuracy rate	Incongruent	0.91 \pm 0.05	0.88 \pm 0.06 [#]	0.87 \pm 0.07	0.92 \pm 0.05 ^{**}
	Congruent	0.95 \pm 0.03	0.95 \pm 0.03	0.93 \pm 0.05	0.96 \pm 0.02 [#]

Data are presented as mean \pm SD. Pre, pre-stimulation; Post, post-stimulation; *, significant difference with Sham, #, significant difference with Pre.

previous work, it has been reported that 2 mA of stimulation for 20 min targeting the motor cortex bilaterally of tDCS enhanced muscle power in lower limb exercise (Lattari et al., 2017). Therefore, the ability of Halo Sport to enhance cycling performance may be related to the increases in lower limb muscle power during cycling. The precise mechanism through which Halo Sport improves exercise performance is unknown. Previous studies suggested that the performance-enhancing effects of tDCS are due to altered central nervous system function, possibly related to the attenuation of central fatigue effects (Vitor-Costa et al., 2015). In the present study, we observed that Halo Sport was able to improve cognitive test. Cognitive decrease is related to central fatigue (Meeusen, 2014), therefore our finding indirectly evidence that the ability of Halo Sport to enhance cycling performance may be related to inhibit central fatigue. One possible mechanism is that the electrical stimulation induces increases in intracortical facilitation and motor cortex excitability, allowing motor-cortex neurons to build neural connections more easily, enhancing motor drive to the muscles, increasing power output of cycle and metal performance, improving cycling performance (Hornyak, 2017).

Moreover, Tanaka and Watanabe (2012) developed a neural circuit for the action of this facilitatory pathway. First, sensory input from the peripheral system to M1 reduces motor output (supraspinal fatigue), and a neural pathway that interconnects the spinal cord, thalamus, secondary somatosensory cortex, medial insular cortex, posterior cingulate cortex, anterior cingulate cortex, premotor area, supplementary motor area (SMA), and primary motor cortex constitutes the inhibition system. Then, a facilitation system increases motor output from M1 to overcome the existing supraspinal fatigue. A re-entrant neural circuit that bridges the limbic system, basal ganglia, thalamus, orbitofrontal cortex, prefrontal cortex, anterior cingulate cortex, premotor area, SMA, and primary motor area represents the facilitation system. Motivational input to this system enhances SMA activity, and subsequently, motor cortex enhances motor output to the peripheral system (Vitor-Costa et al., 2015). Thus, the output (exit of information from the motor cortex to the corticospinal pathways and, consequently, motoneurons) from M1 is regulated primarily by the balance between inhibition and facilitation, leading us to speculate that Halo Sport has a facilitatory effect for increasing power output of cycling. This hypothesis needs to be evaluated in future studies.

To our knowledge, this is the first study to observe that Halo Sport can enhance cognitive performance. It is difficult to compare our findings with those of previous non-invasive neuromodulation studies on cognitive function. Anodal tDCS applied to the DLPC of healthy volunteers has been reported to enhance the executive function of cognition (Dockery et al., 2009; Stone and Tesche, 2009; Zaehle et al., 2011). The executive function is one aspect of cognition, and it generally consists of mental-set-shifting, information updating, and inhibition of prepotent responses (Miyake et al., 2000; Hofmann et al., 2012). Fregni et al. (2005) reported that anodal tDCS of the prefrontal cortex enhanced ACC of 3-back which is a test for information updating performance. They proposed that tDCS

could improve the information updating performance aspect of executive function. In the present study, we found similar results to those of Fregni et al. (2005). Following Halo Sport stimulation over both sides of the motor cortex, the ACC enhancement of Stroop incongruent trial in the Halo group cannot be accounted for by slowed responses, as response times were not changed by stimulation. These results showed that Halo Sport stimulation leads to an enhancement of Stroop performance. Stroop test is a classic task for inhibition of prepotent responses. Therefore, Halo Sport has a positive effect on executive function. These results indicate that Halo Sport may be useful for enhancing all types of exercise in which concentration, RTs, and technical/tactical skills have a major influence on both physical and mental performance, such as cycling/mountain biking, skiing, most ball game and so on.

In the present study, we have only shown the positive effect of Halo Sport on Stroop performance, but the mechanisms behind this phenomenon are unknown. Milham et al. (2003) reported that during the Stroop task, the DLPC is the primary region involved in the implementation of top down attention control. Additionally, according to Krompinger and Simons (2011) the DLPC resolves conflicts that occur during information processing of incongruent stimuli during the Stroop task. Therefore, the Stroop performance is related to activation of the DLPC. Moreover, previous work indicates that bilateral stimulation of the motor cortex induces widespread changes in functional connectivity, in particular with the prefrontal cortex, and the primary and secondary motor cortices (Sehm et al., 2012). Anatomically, M1 is located next to the SMA. The activation of M1 might affect the active SMA, whose functions are considered to plan the movement and make the decision about when to start an action (Nachev et al., 2008; Coull et al., 2016). And previous studies have shown that SMA might work with dorsal anterior cingulate cortex (dACC) to process the cognitive interference, which is produced by conflict conditions of Stroop task (Liu et al., 2004; Deng et al., 2018). DACC sends the signal on cognitive interference to DLPFC, which would participate in resolving the cognitive interference (MacDonald et al., 2000; Liu et al., 2004). In the present study, the improved Stroop performance may be due to Halo Sport increasing the activation of the DLPC in addition to both sides of the motor cortex. Further studies are needed to clarify the effects of Halo Sport on brain activity.

CONCLUSION

tDCS with the Halo Sport device improved repeated sprint cycling power output and Stroop performance. These results indicate that Halo Sport may have the potential to enhance performance across a wide range of exercise activities that entail both physical and cognitive demands.

ETHICS STATEMENT

This study was carried out in accordance with the recommendations of human experimental guidelines, the

local ethical committee of Shanghai University of Sport. All participants were fully informed of the nature and possible risks of the study before giving written consent. The local ethical committee of Shanghai University of Sport approved the experimental protocol.

AUTHOR CONTRIBUTIONS

XZ and LH conceived the study. YL supervised the study. XZ and YD designed the experiments. XZ and LH carried out the experiments. XZ and YD analyzed the data. XZ and LH wrote

the manuscript. All authors approved the final version of the submitted manuscript.

FUNDING

The authors would like to acknowledge supports for the study from the National Natural Science Foundation of China (31701044, 31701041); the Shanghai City Committee of Science and Technology Key Project (No. 17080503200); China Postdoctoral Science Foundation funded project (Grant No. 2017M610266).

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- Conflict of Interest Statement:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Sprinting After Having Sprinted: Prior High-Intensity Stochastic Cycling Impairs the Winning Strike for Gold

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OPEN ACCESS

Edited by:

Toby Mündel,
Massey University, New Zealand

Reviewed by:

Chris R. Abbiss,
Edith Cowan University, Australia
Carl Paton,
Eastern Institute of Technology,
New Zealand

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 02 September 2018

Accepted: 28 January 2019

Published: 14 February 2019

Citation:

Etxebarria N, Ingham SA,
Ferguson RA, Bentley DJ and
Pyne DB (2019) Sprinting After
Having Sprinted: Prior High-Intensity
Stochastic Cycling Impairs
the Winning Strike for Gold.
Front. Physiol. 10:100.
doi: 10.3389/fphys.2019.00100

Bunch riding in closed circuit cycling courses and some track cycling events are often typified by highly variable power output and a maximal sprint to the finish. How criterium style race demands affect final sprint performance however, is unclear. We studied the effects of 1 h variable power cycling on a subsequent maximal 30 s sprint in the laboratory. Nine well-trained male cyclists/triathletes ($\dot{V}O_{2peak}$ 4.9 ± 0.4 L·min⁻¹; mean \pm SD) performed two 1 h cycling trials in a randomized order with either a constant (CON) or variable (VAR) power output matched for mean power output. The VAR protocol comprised intervals of varying intensities (40–135% of maximal aerobic power) and durations (10 to 90 s). A 30 s maximal sprint was performed before and immediately after each 1 h cycling trial. When compared with CON, there was a greater reduction in peak ($-5.1 \pm 6.1\%$; mean \pm 90% confidence limits) and mean ($-5.9 \pm 5.2\%$) power output during the 30 s sprint after the 1 h VAR cycle. Variable power cycling, commonly encountered during criterium and triathlon races can impair an optimal final sprint, potentially compromising race performance. Athletes, coaches, and staff should evaluate training (to improve repeat sprint-ability) and race-day strategies (minimize power variability) to optimize the final sprint.

Keywords: repeated sprints, stochastic cycling, peak power, race profile, triathlon

INTRODUCTION

Modern road cycling events held in large cities commonly use closed circuit criterium style cycle courses during major competitions such as a World Cup, World Championship, or Olympic Games. Consequently these race-courses often consist of repeat laps of numerous tight and technical corners that result in multiple rapid accelerations and decelerations (Menaspa et al., 2017), often close to and above maximal aerobic power (Etxebarria et al., 2014b). The combination of geographical, technical and tactical characteristics of these cycling races can elicit a variable power profile (Ebert et al., 2006; Bernard et al., 2009). Other Olympic events resembling multiple high intensity efforts before a final sprint include the scratch and also the points race in track cycling or even the ~ 1 h cycle section of modern draft-legal Olympic distance triathlon races. However, it is unclear how the modern race settings affect the

physiological and performance abilities of the athletes compared to the traditional 'out and back' style road cycling courses. Furthermore, it is worth exploring how well athletes are prepared physically to suit modern race demands, and also track cycling events such as the scratch and points race.

Despite the relatively extensive analysis of multiple-stage road cycling races, little is known about the shorter road cycle races such as time trial events, with no previous study to our knowledge having investigated the effects of highly variable power output cycling on the ability to generate a short-term maximal sprint. Criterion style courses characterized by a mass start, frequent tight corners and bunch riding, can result in highly variable intensity cycling exercise that increase the physiological demands compared with less variable non-drafting or time trial events (Etxebarria et al., 2013). Moreover, the > 40 min race performance of closed circuit technical courses are best associated with the cyclist's ability to generate high power output over efforts less than 2 min long, showcasing the importance of short-term power output for these endurance events (Babault et al., 2018). These type of races often contain breakaways that demand a sustained high intensity burst, often preceded by multiple high intensity efforts (Abbiss et al., 2013), and followed by a final decisive sprint (Peiffer et al., 2018). Similarly, many track cycling events have a pattern of multiple high intensity efforts throughout, and are often contested in a sprint to the finish line. Multiple sprints without adequate recovery in between lead to lowered repeat sprint ability (Gaitanos et al., 1993), and this could be detrimental to a rider's final sprint where the race is typically decided.

Cycling power profiles can be readily assessed with use of a power-meter in training and during competition, however, simplified summaries of power analysis do not often reflect the demands of the session (Passfield et al., 2017). The highly variable power output of criterium-style races include multiple 10–30 s high intensity efforts and ~12% of the time spent at exercise intensities above $8 \text{ W}\cdot\text{kg}^{-1}$ (Ebert et al., 2006), during which the physiological demands differ markedly from time trial style cycling (Etxebarria et al., 2014c). Furthermore, draft legal Olympic distance triathlon races can induce high intensity power outputs for a substantial time (~15%) including frequent high intensity efforts spread intermittently throughout (Bernard et al., 2009). These reoccurring high intensity efforts often exceed supra-maximal intensities ranging 100 to 140% of maximal aerobic power (Etxebarria et al., 2014b). With frequent high intensity efforts, the physiological demands of city-based certain road cycling events and draft legal triathlon are similar (Ebert et al., 2006).

The physiological demands imposed by different cycling strategies including constant and variable power cycling have been studied in a laboratory setting by implementing variable power protocols with smaller variability (Palmer et al., 1997; Lepers et al., 2008; Suriano et al., 2010) than that observed in some contemporary cycling races (Ebert et al., 2006; Bernard et al., 2009). The duration of the treatment protocols implemented (variable vs. constant) also differ substantially between studies: from ~30 min (Bernard et al., 2007; Suriano et al., 2007; Lepers et al., 2008; Thomas et al., 2012) to ~2 h

(Palmer et al., 1999) and use a diverse range of performance and outcome measures. These methodological differences between studies investigating constant and variable cycling limit the transfer of the findings to actual sporting performances. A recent laboratory-based protocol with a range of power variations showed substantially greater physiological demands during variable power cycling compared to a sustained effort matched for mean power output (Etxebarria et al., 2013). However, no cycling performance outcome measures were reported and it is unclear how sprint ability would be affected by multiple intermittent high intensity efforts.

Many of these road (and selected track) cycling races are decided in a bunch-sprint after a multiple of high intensity efforts, raising the need to conserve repeat sprint-ability to minimize fatigue before the final sprint that can last ~20 s or more (Peiffer et al., 2018). As a secondary performance measure for Olympic distance triathlon, the impairment of a maximal cycling sprint could translate to accumulated fatigue before the subsequent and decisive 10 km run. Therefore, the aim of this study was to compare the effect of 1 h cycling at variable power (simulating real world competition demands) vs. constant power output, matched for time and mean power output, on the ability to generate maximal power during a subsequent 30 s sprint. This information will inform the preparation for, and tactics employed during, different cycling and triathlon events.

MATERIALS AND METHODS

Subjects

Nine well-trained male triathletes and cyclists (age: 30 ± 7 year; stature: 1.79 ± 0.05 m; body mass: 74.3 ± 5.3 kg; $\dot{V}O_{2\text{peak}}$ $4.9 \pm 0.4 \text{ L}\cdot\text{min}^{-1}/66.0 \pm 3.9 \text{ mL}\cdot\text{kg}^{-1} \text{ min}^{-1}$, mean \pm SD) completed the preliminary testing and experimental cycle trials in a single group cross-over design. All subjects had at least 3 years of training and racing in cycling and triathlon events. In the 24 h prior to each laboratory visit subjects were required to abstain from any physical exercise, caffeine and alcohol intake and replicate the same dietary practice. The study was approved by the Loughborough University Ethics Advisory Committee and followed the guidelines of the Declaration of Helsinki. All subjects provided written informed consent after explanation of the study protocols and experimental procedures.

Procedures

All participants reported to the laboratory on three separate occasions. The first visit involved performing an incremental exercise test to determine $\dot{V}O_{2\text{peak}}$ followed by a 30 min break and a 30 s maximal sprint familiarization trial. The subsequent two visits comprised of a 1 h cycle at either variable (VAR) or constant power (CON) cycling, in a randomized counterbalanced order, with a 30 s maximal sprint just before and closely after the 1 h cycle. The two 1 h experimental cycle trials were performed on two subsequent occasions and at least 5 days apart. The preliminary $\dot{V}O_{2\text{peak}}$ test consisted of a progressive incremental ramp test on an SRM cycle ergometer (SRM Ergometer with integrated SRM Training System, Science

version, Jülich, Germany) following a 10 min warm up at 100 W. The starting power output for the maximal test was between 160 and 180 W, depending on the level of training and experience of subjects. Increments of 5 W every 15 s were employed during the maximal progressive test to ensure exhaustion was reached after approximately 10 min. This protocol allows for a higher maximal aerobic power for a similar $\dot{V}O_{2\text{peak}}$ than a more traditional 3 min incremental stage protocol (Bishop et al., 1998). Pedal cadence was freely chosen and maintained at a constant rate. Maximal aerobic power was defined as the mean of the highest consecutive power values recorded during the test for a 1 min period. Participants performed a 30 s maximal sprint familiarization trial, 30 min after the $\dot{V}O_{2\text{peak}}$ test.

Upon arrival for the CON or VAR trial subjects performed a 10 min warm up at 100 W followed by a 30 s maximal sprint from a stationary start before the 1 h cycle trials. Due to the lack of information in criterium style races, the mean intensity for both cycle trials was set at 60% maximal aerobic power, similar to the intensity observed during draft legal triathlon races (Le Meur et al., 2009). The CON trial involved cycling for 1 h at a constant power equivalent to 60% maximal aerobic power. The power variations during VAR were characterized by intermittent efforts of different intensities and durations: 10 s at 135%, 40 s at 110%, 90 s at 85%, 20 s at 130%, and 30 s at 120% of maximal aerobic power (Figure 1). This protocol was designed to replicate a generic power profile typically experienced during criterium races and the cycle section of triathlon races, and similar to the Beijing Olympic test event (Bernard et al., 2009). Subjects self-selected their preferred pedal cadence during the first trial and were required to replicate this during the second trial. A minute after terminating the 1 h cycle test subjects performed another 30 s maximal sprint. Participants were allowed to drink a maximum of 750 mL of water during the 1 h cycling trials.

Body mass was recorded on the first visit to the laboratory using an electronic scale (Seca 770, GMBH & Co., Germany). Peak heart rate (HR_{peak}) was recorded during the $\dot{V}O_{2\text{peak}}$ incremental exercise test using a telemetry system (Polar Electro iS610, Oulu, Finland). A 25 μL capillary blood sample taken from a finger-tip within 30 s of the end of the maximal test and analyzed for blood lactate concentration (B_{La}) using an automated blood lactate analyzer (Biosen C-Line, Southam, United Kingdom). During the $\dot{V}O_{2\text{peak}}$ test expired air was sampled continuously for CO_2 and O_2 content and volume on a gas analysis system with a day-to-day reliability of 2.3% (Oxycon Pro Jaeger, Höchberg, Germany). Respiratory gas exchange variables ($\dot{V}O_{2\text{peak}}$, $\dot{V}CO_2$, \dot{V}_E) were sampled every 15 s, the mean of the highest consecutive four readings taken as the $\dot{V}O_{2\text{peak}}$ value.

Power output during the $\dot{V}O_{2\text{peak}}$ incremental exercise test, 30 s maximal sprints and 1 h cycling trials were measured on an electromagnetically-braked cycle ergometer, hyperbolic mode. During the sprint test, the ergometer was set in open test mode. The cycle-ergometer set up was individualized, mimicking each participant's bike set up on their own bike. Cycling data were downloaded to a computer and analyzed with SRM software (v6.40.05, Schoberer Rad Meßtechnik, Germany). The SRM power-meter was zeroed before each trial by recording the zero

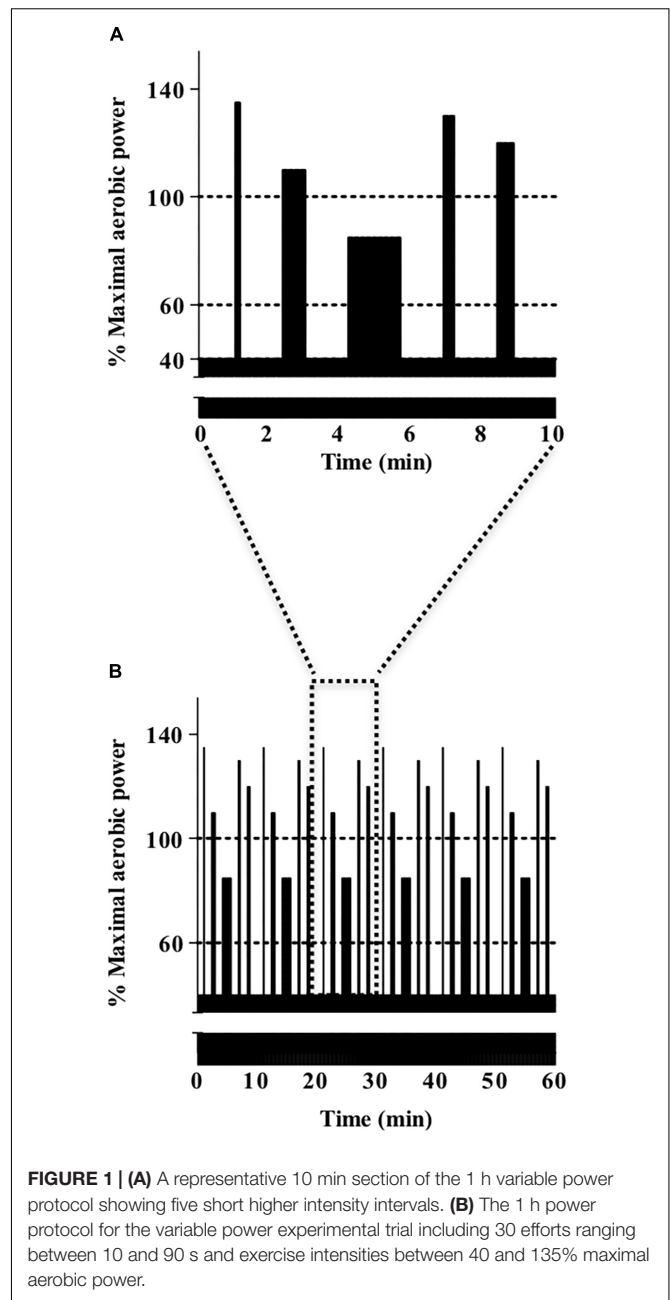


FIGURE 1 | (A) A representative 10 min section of the 1 h variable power protocol showing five short higher intensity intervals. **(B)** The 1 h power protocol for the variable power experimental trial including 30 efforts ranging between 10 and 90 s and exercise intensities between 40 and 135% maximal aerobic power.

offset without any force/load on the cranks. During the $\dot{V}O_{2\text{peak}}$ test power output was sampled at 1 Hz. During the 30 s maximal sprint tests data was sampled at 0.5 Hz. Peak power and peak pedal cadence were defined as the highest power and cadence recorded by the cycle ergometer during each 30 s effort. Mean power was defined as the average of the power outputs during the 30 s all out efforts. Time to peak power was measured from the start of an effort to the time when the subject reached peak power.

Statistical Analyses

Data modeling involved point estimation of peak and mean power response to stochastic and steady-state cycling protocols,

and interval estimates of the uncertainty about the value of these parameters. A statistical approach using magnitude-based inferences and precision of estimation was used to determine practical/clinical significance of effects (Hopkins, 2017). Mean effects of the variable and constant power strategies and their 90% confidence limits (CL) were estimated via the unequal-variances *t*-statistic computed for change scores between pre- and post-tests of the two groups. Each subject's change score was expressed as a percentage of baseline score via analysis of log-transformed values, in order to reduce bias arising from non-uniformity of error. The magnitude of difference between the two groups was expressed as a standardized effect size. The criteria to interpret the magnitude of effects were: <0.2 trivial, 0.2–0.6 small, 0.6–1.2 moderate, 1.2–2.0 large, and >2.0 very large (Hopkins, 2000).

For mean power output, we estimated the smallest worthwhile effect in this cohort of well-trained (but not elite) cyclists as $0.5 \times 2 \times 2.5 = 2.5\%$ using the method outlined by Paton and Hopkins (2001) where 0.5 is the default smallest worthwhile proportion of the typical within-subject variability in time-based performance tasks or events (Hopkins et al., 1999), 2.0% is the estimated typical within-subject variability (% coefficient of variation) of well-trained road cyclists (Malcata and Hopkins, 2014), and 2.5 is the constant for conversion of performance time to power output (Paton and Hopkins, 2001; Malcata and Hopkins, 2014). When the 90% CL concurrently crossed the thresholds for the smallest meaningful decrement and improvement, the effect was deemed unclear. Standardized scores for correlation were interpreted according to a scale of magnitudes: <0.1 trivial, 0.1–0.3 small, 0.3–0.5 moderate, 0.5–0.7 large (Hopkins et al., 2009). A correlation was deemed unclear if the confidence interval spanned both -0.1 and $+0.1$ values. A sample size of 11 subjects was deemed appropriate in a single group cross-over design assuming a smallest worthwhile difference in mean power output of 2.5%, a typical error of 2.0%, and type I and II errors of 5 and 25% respectively. Descriptive data are reported as mean \pm standard deviation (SD).

RESULTS

Physiological and Performance Characteristics

The well-trained nature of the subject cohort was indicated by the values of maximal aerobic power and mean power output. Maximal aerobic power was 389 ± 32 W (mean \pm SD) and peak B_{La} at the end of the test was 12.4 ± 2.3 mmol L^{-1} with a HR_{peak} of 189 ± 9 b min^{-1} . Mean power output during the 1 h cycle was 233 ± 19 W for CON and 234 ± 20 W during VAR for all participants. The higher variability of the 1 h VAR protocol was indicated by a coefficient of variation (%CV) in power of 50% with an SD in power output of 117 ± 9 W. In contrast, the %CV during the 1 h CON protocol was only 9% with an SD in power output of 22 ± 5 W. Mean pedal rate was 94 ± 4 rev. min^{-1} and 95 ± 4 rev. min^{-1} for CON and VAR, respectively. There were trivial differences between CON and VAR in mean power and pedal rate.

Variable Versus Constant Cycling

The variability in power output during VAR hindered the ability to produce short-term maximal power output after 1 h of cycling compared to the constant power trial. Peak power during the 30 s sprint prior to the 1 h trials was similar: 866 ± 134 W (mean \pm SD) for CON and 869 ± 137 W for VAR. There was a small difference in the change of peak power output between VAR and CON (-0.45 ± 0.37 ; standardized difference \pm 90% CL) (Figure 2A). The 1 h VAR cycling decreased peak power output ($-5.1 \pm 6.1\%$; % difference \pm 90%CL) and mean power output ($-5.9 \pm 5.2\%$) generated during the post-trial 30 s sprint (Table 1). Mean power output during the 30 s sprint prior to the 1 h cycle trials were also similar for CON (567 ± 73 W; mean \pm SD) and VAR (560 ± 72 W). There was a small difference in the change of mean power output between VAR and CON (-0.33 ± 0.37 ; standardized difference \pm 90%CL). After the VAR trial the mean power output was also lower by $\sim 6\%$ to 526 ± 71 W (mean \pm SD) with only a trivial change after CON to 565 ± 66 W (Figure 2B).

Time to peak power output and peak cadence before and after the 1 h cycle remained very similar for both CON (10.9 ± 2.0 vs. 10.9 ± 2.9 s and 126 ± 14 vs. 126 ± 6 rev. min^{-1}) and VAR (10.4 ± 2.3 vs. 10.7 ± 3.4 s and 128 ± 12 vs. 123 ± 9 rev. min^{-1}). A moderate relationship ($r = -0.53$, -0.83 to 0.03 90% confidence interval) between the relative maximal power output ($W \cdot kg^{-1}$) and the decrease in mean power output during the 30 s sprint was evident after the 1 h VAR condition. Similarly, there was a moderate relationship ($r = -0.65$, -0.83 to 0.05 , 90% confidence interval) between triathletes/cyclists who had higher relative PPO ($W \cdot kg^{-1}$) during the 30 s sprint before the 1 h VAR having a larger decrease in PPO afterward.

DISCUSSION

Variable power cycling resembling criterium-style racing (bunch riding) and some track cycling events such as the scratch or points race, reduced end maximal sprinting capacity by $\sim 6\%$. This decrease in the ability to produce maximal power output during the latter stages of criterium-style races would be detrimental in the fight for a final sprint, often the way the race is decided, negatively impacting the final outcome of the race. The $\sim 6\%$ decrease in 30 s peak and mean power output we observed at the end of the 1 h VAR protocol could translate to a reduced ability to fight for the top positions in a world championship or Olympics Games when races can be decided in the last 20–25 s of the race. Both road and track cycling events such as the scratch and points race contain multiple high intensity efforts during the event, given the interplay of technical and tactical factors.

The investigation of variable and constant power cycling required development of a suitable laboratory-based cycling protocol with acceptable content and face validity. The relative mean power output for the 1 h cycle trial in this study ($\sim 60\%$ maximal aerobic power) is comparable to that described in criterium style races by Ebert et al. (2006). The frequent high intensity efforts featured in our 1 h VAR protocol by the numerous supra-maximal efforts ($>$ maximal aerobic power)

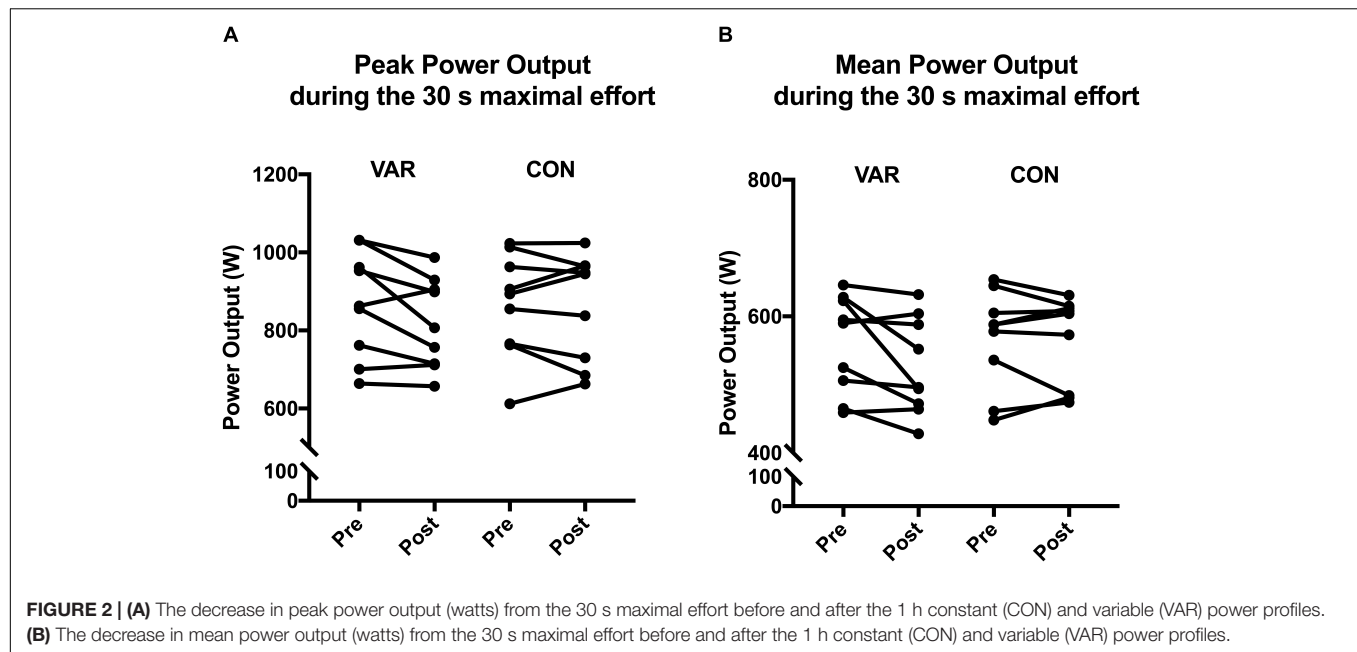


TABLE 1 | Differences in peak power, mean power, peak pedal cadence and time to peak pedal cadence during the 30 s maximal sprint before and after 1 h constant and variable power cycling.

	% Change after 1 h trial (± 90% CL)		% Difference (± 90% CL)	Standardized difference (± 90% CL) qualitative inference
	Constant power	Variable power		
Peak power output (W)	−0.5 ± 6.4	−5.6 ± 7.3	−5.1 ± 6.1	−0.33 ± 0.37, small
Mean power output (W)	−0.3 ± 5.4	−6.1 ± 8.6	−5.9 ± 5.2	−0.45 ± 0.37, small
Peak pedal cadence (rpm)	−0.1 ± 10.7	−4.1 ± 10.8	−4.0 ± 8.7	−0.61 ± 1.25, unclear
Time to peak power (s)	−2.0 ± 38	1.0 ± 38	3.0 ± 17	0.08 ± 0.48, trivial

simulated the power fluctuations and duration of effort observed in real-world cycling performances. This study overcomes some of the shortcomings of previous studies implementing short 30 min protocols (Lepers et al., 2008; Theurel and Lepers, 2008) or had not included multiple short (10–30 s) high intensity efforts (Suriano et al., 2010) common in criterium races. Furthermore, the power distribution implemented during the variable power protocol in the present study is similar to the protocol resembling the cycle section of draft legal triathlon races (Etzebarria et al., 2013). Consequently, the decrement in peak and mean power output during a maximal sprint in the present study is representative in terms of content and face validity, and applicable to these sporting situations. More specifically, slower swimmers who do not make the leading pack (during subsequent cycling) and tend to increase their power output during the latter stages of the cycling section to breach the gap (Vleck et al., 2008), might be negatively affected by the higher cycling power output and compromise their chance to ‘get into’ the race or keep being a contender for the top positions.

The greater decrease in peak power output in the 30 s maximal sprint after VAR presumably reflects accumulated fatigue from the repeated high intensity peaks observed in races. Fatigue caused by repetitive intermittent and high

intensity exercise is influenced by a combination of metabolic (Westgarth-Taylor et al., 1997; Stepto et al., 2001; Allen et al., 2008) and neuromuscular factors (Gandevia, 2001). Variable power cycling has also different muscle recruitment patterns (Palmer et al., 1999; Suriano et al., 2010) and metabolic responses (Palmer et al., 1999) than cycling at constant power. Variable power cycling that includes supra maximal intensities decrease maximal voluntary contraction torque and activation (Billaut et al., 2006; Theurel and Lepers, 2008) but not when lower (60 to 90% maximal aerobic power) exercise intensities are employed (Lepers et al., 2008). The evidence presented in this study should inform coaches and athletes of the areas to focus their training on when competing under criterium style race demands, repeat sprint-ability, and power variability.

Variable and constant power cycling appear to yield a similar decrease in total glycogen (Brickley et al., 2007). However, cycling at variable power elicits a greater level of glycogen depletion in type II muscle fibers (Palmer et al., 1999; Suriano et al., 2010), the same pool of muscle fibers that would have been targeted during a 30 s maximal sprint. Athletes with a higher PPO are likely to have a higher percentage of these fast-twitch fibers. Consequently these athletes are more likely to fatigue after several sprints, inducing a greater drop-off in PPO after VAR, which an

outcome we observed in this study. Therefore, constant power cycling is likely to spare higher levels of PCr (non-oxidative path) for the post-trial 30 s effort as high intensity efforts rely on non-oxidative ATP resynthesis pathways (Gastin and Lawson, 1994; Bogdanis et al., 1996). The high intensity efforts involved in VAR induce three times the blood lactate concentration at the end of the 1 h protocol compared with CON (Etxebarria et al., 2013). The selective depletion of glycogen during variable power cycling and reported higher glucose oxidation (Palmer et al., 1999) could be the explanation why fluctuating power output is detrimental to end race performance and/or the early stages of the subsequent running section in triathlon (Etxebarria et al., 2014a). Future studies should investigate relationships between patterns of glycogen depletion, power profiles, and potentially dietary manipulation as a strategy for improving race performance.

Given the wide range of cycling intensities coupled with frequent changes in pace (similar to those experienced in criterium and triathlon), the deleterious effects of variable power cycling on short-term maximal power generation capacities, are applicable to real-world race situation. The ~6% reduction in the ability to generate power after variable power cycling could have negative implications for the late stages of a criterium style cycle race. A geographically (hills) and technically (multiple corners) challenging cycling course in which major competitions are race under could translate to early fatigue and loss of medal hope. The cycling course for the Tokyo 2020 Olympics and Paralympics will start in the metropolitan area and already described as 'startingly testing course' for the road cycling and time trial events¹. Similarly, the triathlon course for the cycling section is based on a 5 km closed circuit course with several 360 and 180 degree turns in each lap². Therefore, athletes could benefit from using a similar interval training protocol to the VAR intervention in to gain specific adaptations to race demands during the Tokyo games.

Further research is needed to investigate the mechanisms explaining this greater reduction in peak and mean power output during a 30 s all-out effort after a relatively short (~60–90 min), variable power cycle bout. However, there are several specific training strategies that could help in promoting specific adaptations to technical courses inducing variable power output such as high intensity interval training (Etxebarria et al., 2014a) and improving cycling technical competency (Babault et al., 2018). These strategies are especially important for triathletes, who do not spend as much time on the bike to develop bunch-riding and technical skills as specialist cyclists do.

Practical Applications

A diminished ability to generate peak power outputs could lead to a variety of detrimental race situations for athletes, including missing an attacking opportunity (defensive shortcoming) or failing to create a breakaway to establish a leading gap (attacking shortcoming) over an opponent or group of opponents. Coaches

and athletes should consider the 1 h sport-specific cycling protocol as a useful training option for cyclists to prepare for races. This type of training should increase the ability to produce repeat sprints with small reductions in peak power toward the end of road races, as well as track cycling events such as the scratch or the points race. The more powerful cyclists were most vulnerable to losing their sprint-ability after fluctuating power cycling in this study – these athletes could benefit from further developing their aerobic capacity.

The cycling demands during major competitions such as the Olympic Games and World Championships are often dictated by other competitors' tactics and performances, and/or the technical nature of the course that induce high intensity efforts and multiple changes in cycling pace. Improving cycling skills in preparation for highly technical courses may be advantageous to limit abrupt decelerations and consequent sharp accelerations out of corners. These skills would enable athletes to sustain a higher velocity for less power produced, increased control for peloton management and 'saving the legs' for a final sprint. The aim of athletes competing in criterium style cycling courses should be to combine increased sprint-ability, minimizing power variability, promoting recovery between sprints, and optimizing technical skills.

CONCLUSION

Cycling at race-specific variable power output for 1 h appears to decrease the ability to generate short-term (30 s) maximal power output compared to cycling at constant power for the duration. Athletes, coaches, and staff should evaluate training and race-day strategies to better maintain the final sprint or end spurt.

ETHICS STATEMENT

The study was approved by the ethics committee for human research at Loughborough University.

AUTHOR CONTRIBUTIONS

NE, SI, and RF devised the study design. NE conducted the data collection and processing. All authors participated in interpretation of the data, preparation of the written manuscript, and read and approved the final manuscript.

FUNDING

This research project was funded by Loughborough University.

ACKNOWLEDGMENTS

The authors thank all the subjects for their time and effort, the English Institute of Sport in Loughborough, United Kingdom for supporting this project and especially Jill Stanley for her support in the Sports Physiology laboratory at Loughborough University.

¹<https://www.cyclingweekly.com/news/racing/olympics/olympic-cycling-race-route-357988>

²<https://tokyo2020.org/en/games/sport/olympic/triathlon/>

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Inconsistency in the Standard of Care—Toward Evidence-Based Management of Exertional Heat Stroke

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OPEN ACCESS

Edited by:

Toby Mündel,
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Reviewed by:

Jason Kai Wei Lee,
DSO National Laboratories, Singapore
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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 01 November 2018

Accepted: 28 January 2019

Published: 18 February 2019

Citation:

Hosokawa Y, Nagata T and
Hasegawa M (2019) Inconsistency
in the Standard of Care—Toward
Evidence-Based Management
of Exertional Heat Stroke.
Front. Physiol. 10:108.
doi: 10.3389/fphys.2019.00108

Tokyo 2020 Summer Olympics are projected to experience environmental heat stress that surpasses the environmental conditions observed in the Atlanta (1996), Athens (2004), Beijing (2008), and Rio (2016) Summer Olympics. This raises particular concerns for athletes who will likely to be exposed to extreme heat during the competitions. Therefore, in mass-participation event during warm season, it is vital for the hosting organization to build preparedness and resilience against heat, including appropriate treatment, and management strategies for exertional heat stroke (EHS). However, despite the existing literature regarding the evidence-based management of EHS, rectal thermometry and whole-body cold-water immersion are not readily accepted by medical professionals outside of the sports, and military medicine professionals. Current Japanese medical standard is no exception in falling behind on evidence-based management of EHS. Therefore, the first aim of this paper is to elucidate the inconsistency between the standard of care provided in Japan for EHS and what has been accepted as the gold standard by the scientific literature. The second aim of this paper is to provide optimal EHS management strategies that should be implemented at the Tokyo 2020 Summer Olympics from organizational level to maximize the safety of athletes and to improve organizational resilience to heat. The risk of extreme heat is often neglected until a catastrophic incidence occurs. It is vital for the Japanese medical leadership and athletic communities to re-examine the current EHS management strategies and implement evidence-based countermeasure for EHS to expand the application of scientific knowledge.

Keywords: rectal thermometry, cold water immersion, pre-hospital care, medical control, exertional heat illness, exertional heat illness treatment

INTRODUCTION

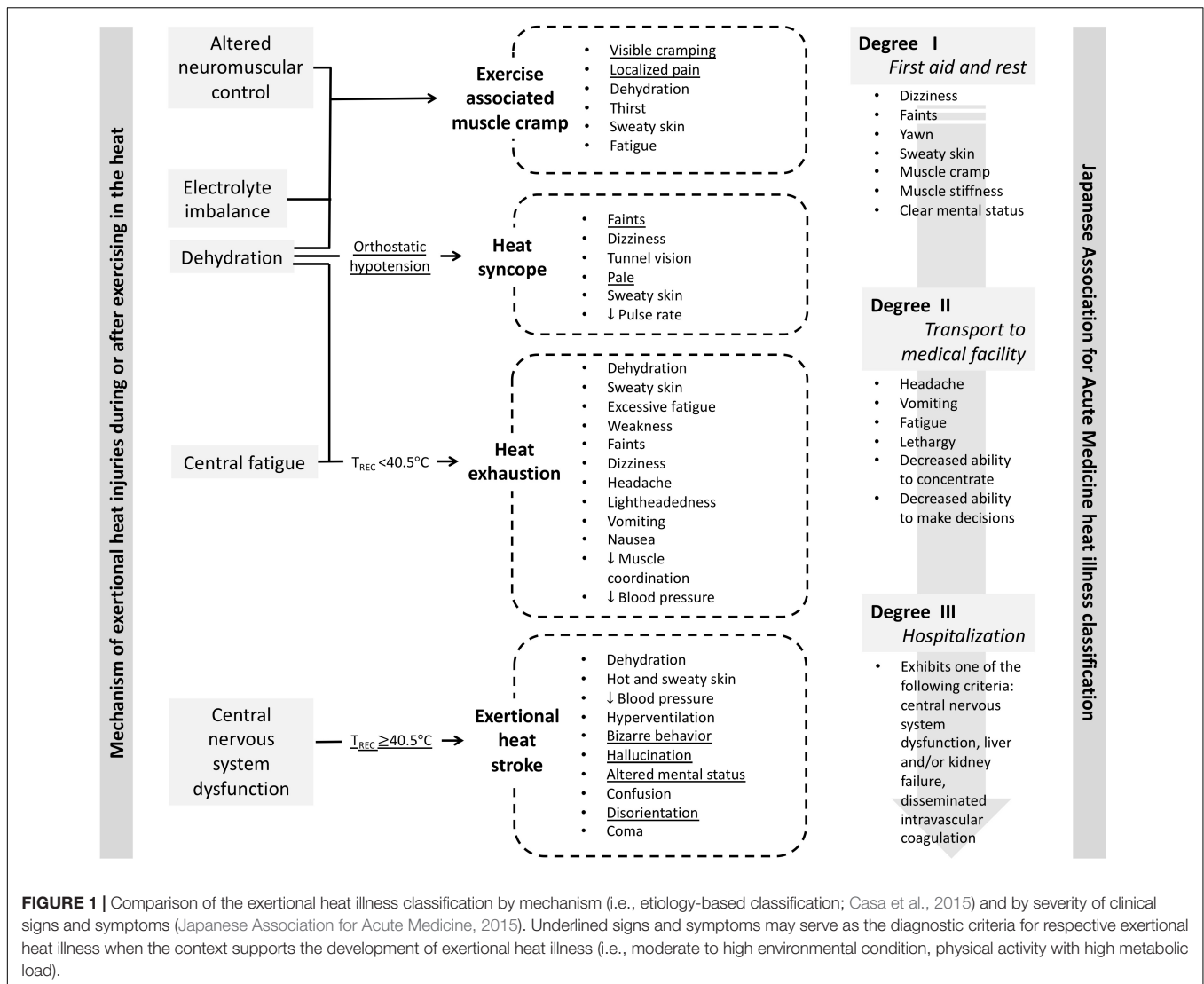
Tokyo 2020 Summer Olympics are projected to experience environmental heat stress that surpasses the environmental conditions observed in the Atlanta (1996), Athens (2004), Beijing (2008), and Rio (2016) Summer Olympics (Kakamu et al., 2017). This raises particular concerns for athletes who may have their competitions scheduled during the day time when the exposure to extreme heat is likely. Given that Summer Olympic sports include many outdoor endurance events (e.g., marathon, race walk, and triathlon), it is evident that these athletes may particularly be at risk for exertional heat illness (Bergeron, 2014). Therefore, in mass-participation event during warm season, it is vital for the hosting organization to build preparedness and resilience against heat, including appropriate treatment and prevention strategies for exertional heat illness. According to press releases from Japan Meteorological Agency (2018) and Fire and Disaster Management Agency (2018), Japan experienced the warmest recorded summer in 2018, doubling the number of heat related injury emergency room visits (2018, 7.5 per 10,000 per population; 2015–2017, 4.2 ± 0.2 per 10,000 population) and fatalities (2018, 1.3 per 1,000,000 populations; 2015–2017, 0.6 ± 0.2 per 1,000,000 population) compared to the previous 3-year average. Although these reports do not distinguish exertional heat illness and classic heat illness (i.e., non-exertional), the need to reexamine current medical practice surrounding heat related illness is heightened. In particular, management of exertional heat stroke (EHS), the most severe form of exertional heat illness, should be reviewed by all medical providers since inappropriate care may lead to death when data otherwise suggests 100% survival rate, if treated properly (Demartini et al., 2015). A plethora of studies suggest that determining factor for EHS prognosis is the duration of hyperthermia ($\geq 40^\circ\text{C}$) incurred by the patient. Therefore, accurate assessment of internal body temperature (e.g., rectal temperature), rapid cooling of the EHS patients using whole-body cold-water immersion, and continuous monitoring of internal body temperature to prevent overcooling are vital in optimizing their survival. Continuous monitoring also helps determine the duration of sustained hyperthermia, which is highly correlated to injury severity (Heled et al., 2004). Despite the existing literature regarding the evidence-based management of EHS, rectal thermometry, and whole-body cold-water immersion are not readily accepted by medical professionals outside of the sports and military medicine professionals. Furthermore, since majority of medical professionals rarely encounter patients who become ill due to over-exertion in the heat, many clinicians have poor understanding of EHS (Casa et al., 2005). Current Japanese medical practice is no exception in falling behind on evidence-based management of EHS (Japanese Association for Acute Medicine, 2015). Therefore, the first aim of this paper is to elucidate the inconsistency between the standard of care provided in Japan for EHS and what has been accepted as the gold standard by the scientific literature. The second aim of this paper is to provide optimal EHS management strategies that should be implemented at the Tokyo 2020 Summer Olympics from organizational level to maximize

the safety of athletes and to improve organizational resilience to heat.

COMPARISON OF EXERTIONAL HEAT STROKE TRIAGE

Coordination of medical care at athletic events poses unique challenges to the event host and medical providers due to the limited resources available on-site (Adams et al., 2018). Therefore, establishing a evidence-based consensus among medical providers should be of the utmost importance in order to minimize unnecessary hospital transfers and confusion among group of medical providers who are working together for the first time (i.e., volunteers). Scientific literature suggests that the key paradigm for EHS treatment and survival is to *cool first transport second* (Casa et al., 2015). This order of triage may be unique to EHS, as most catastrophic injuries that require medical attention are *transported first*. Consequently, it may be counter-intuitive for many medical providers to treat (i.e., cool) the EHS patient before sending to a hospital, unless an EHS-specific protocol is pre-established (Belval et al., 2018).

In Japan, the paradigm for successful EHS treatment and survival is not well accepted due to two major barriers faced by the medical system. First, in order to perform *cool first* on EHS patients, accurate internal body temperature must be measured rectally to determine the need to cool and the end point of cooling at pre-hospital triage (Casa et al., 2015). However, the current common practice by the emergency medical technician is to use axillary or tympanic membrane temperature (Japanese Association for Acute Medicine, 2015), which provide inaccurate estimate of internal body temperature in exercising individuals (Casa et al., 2007a; Ganio et al., 2009; Taylor et al., 2014; Morán-Navarro et al., 2018). Axillary thermometry is an inexpensive method of internal body temperature assessment and is commonly used to assess pyrexia. Despite its convenience, it has been shown to demonstrate lower value than rectal temperature by mean bias of -0.94 to -1.25°C during indoor exercise in the heat (Ganio et al., 2009). This deviation becomes even larger to a mean bias of -2.07 to -2.58°C when the exercise is conducted outdoors (Casa et al., 2007a). It is evident that extent of bias in axillary temperature to diagnose EHS is too large that it may underestimate the level of hyperthermia and may preclude the patient from receiving the appropriate care. The lower value in axilla than rectal thermometry is likely due to the influence from sweaty skin, which is inherent to someone who has been exercising in thermal environment. Tympanic membrane thermometer is also a common type of thermometer used daily by non-medical and medical personnel. This method is thought to receive less influence from the surface body temperature compared to the axillary method. However, a narrow physical space for measurement (i.e., tympanic membrane), difficulty in capturing the line of sight to the tympanic membrane, influence from earwax buildup, and the estimation of temperature using infrared radiation from the tympanic membrane makes this method vulnerable for invalid measures (Ganio et al., 2009). Consequently, although narrower range than the axillary method,



tympenic membrane temperature resulted in a mean bias of -0.67°C and -1.00°C during indoor and outdoor exercise, respectively (Casa et al., 2007a; Ganio et al., 2009). A separate study revealed much narrower range of mean bias (0.10 – 0.20°C) using a tympanic membrane thermometer during exercise in the heat (Morán-Navarro et al., 2018); however, the exercise-induced hyperthermia observed in this study was moderate ($38.3 \pm 0.9^{\circ}\text{C}$) compared to the previous studies ($\approx 40.0^{\circ}\text{C}$), (Casa et al., 2007a; Ganio et al., 2009) suggesting that the deviation in tympanic membrane temperature becomes larger as the internal body temperature increases. Notwithstanding the evidence, the lack of personnel who are trained to take rectal temperature at pre-hospital settings in Japan inevitably delays the EHS diagnosis until the patient is transported to the hospital (Japanese Association for Acute Medicine, 2015). Japanese medical textbooks have also long used the terms heat stroke and heat syncope interchangeably, which also did not help establishing a standardized definition of heat related injuries.

As a result of limited ability to diagnose EHS based on rectal temperature and lack of standardized definition to describe heat related injuries Yasuoka et al. (1999), proposed the assessment of heat illness by the severity of signs and symptoms that is classified into Degree I (mild), II (moderate), and III (severe). This classification was readily adopted by the Japanese emergency medical community since similar severity category was used to describe burn injuries and heat failure. Degree I heat illness is considered benign and self-resolving with rest in cool area, external body cooling, oral rehydration, and sodium supplementation. Clinical symptoms classified under Degree I heat illness include dizziness, faints, yawn, sweaty skin, muscle cramp, muscle stiffness, and clear mental status. Degree II heat illness is considered more severe than Degree I, and its clinical signs and symptoms include headache, vomiting, fatigue, lethargy, decreased ability to concentrate, and decreased ability to make decisions. Japanese Association for Acute Medicine (JAAM) suggests that patients with Degree II heat illness should be transported to medical facility to restore normal

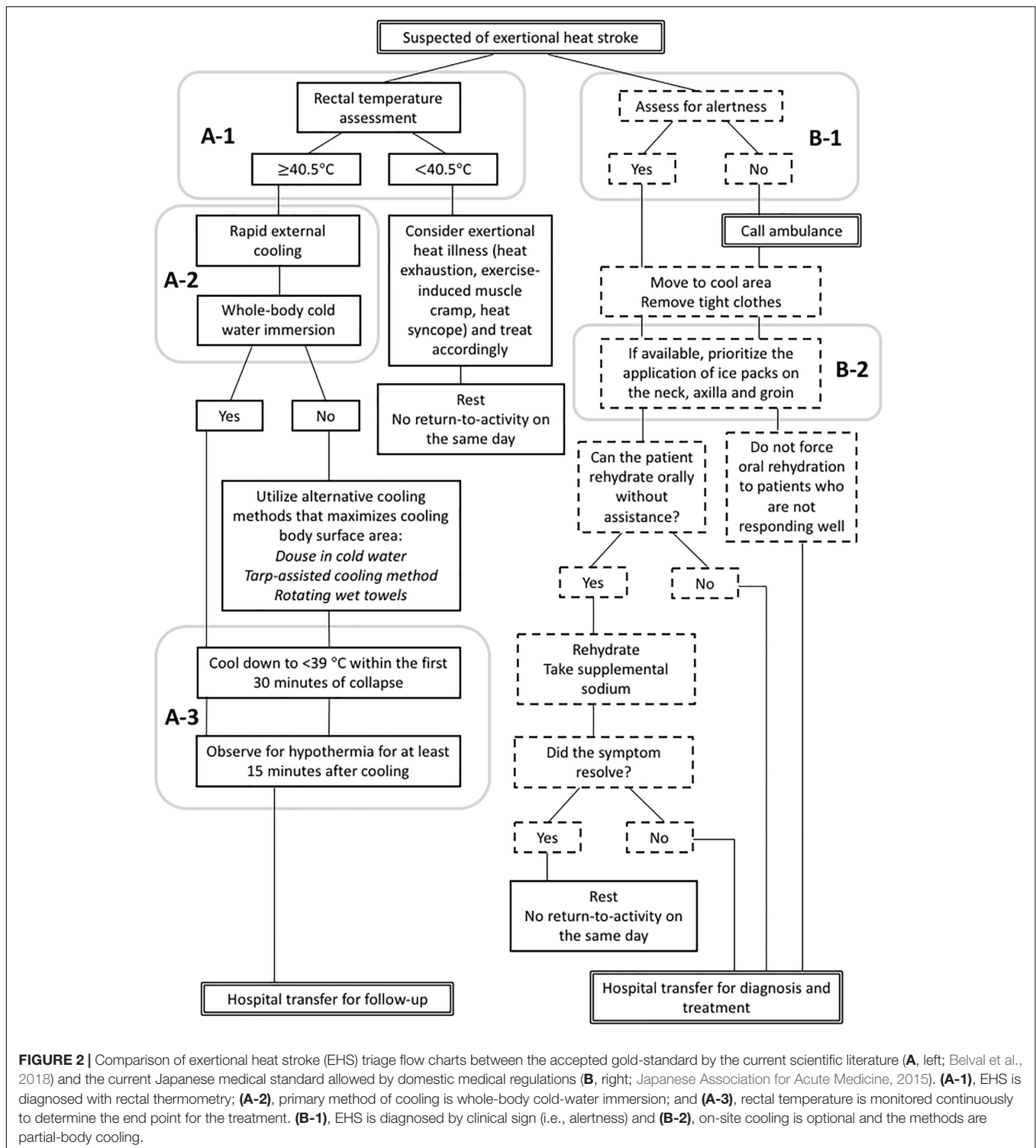


FIGURE 2 | Comparison of exertional heat stroke (EHS) triage flow charts between the accepted gold-standard by the current scientific literature (A, left; Belval et al., 2018) and the current Japanese medical standard allowed by domestic medical regulations (B, right; Japanese Association for Acute Medicine, 2015). (A-1), EHS is diagnosed with rectal thermometry; (A-2), primary method of cooling is whole-body cold-water immersion; and (A-3), rectal temperature is monitored continuously to determine the end point for the treatment. (B-1), EHS is diagnosed by clinical sign (i.e., alertness) and (B-2), on-site cooling is optional and the methods are partial-body cooling.

body temperature and hydration status (Japanese Association for Acute Medicine, 2015). The most severe form of heat illness is classified as Degree III, which requires hospitalization. The clinical manifestation of Degree III heat illness includes central nervous dysfunction, liver and kidney failure, and disseminated intravascular coagulation (DIC). The heat illness

severity classification was later validated by Okudera et al. (2002) by retrospectively examining the medical data from patients who were admitted the emergency department due to heat illness. It is important to note that organ failures and DIC are the outcomes of EHS that were *not* treated properly (Casa et al., 2015; Stearns et al., 2016), and they can be prevented

if early recognition of hyperthermia and aggressive cooling are implemented (Casa et al., 2007b). In other words, unless the pre-hospital medical standards for EHS changes to allow rectal thermometry and cooling before transport, the classification system is opening a room for EHS prognosis that may be debilitating or in the worst case, fatal. **Figure 1** compares the exertional heat illness classification by mechanism (i.e., etiology-based classification) and by severity of clinical signs and symptoms (i.e., JAAM classification). While mechanism based-classification allows clinicians to look for presence (or absence of) distinct clinical presentations, symptom-based classification without rectal thermometry is subjective, which opens a window for overlooking EHS (**Figure 1**).

The second barrier to *cool first transport second* comes from the hesitancy among medical professionals to cool EHS patients using whole-body cold-water immersion due to the fear of inducing shock (Japanese Association for Acute Medicine, 2015). Although JAAM recognizes the importance of aggressive cooling in their Heat Illness Treatment Guideline 2015 (Japanese Association for Acute Medicine, 2015), it does not outline the specific method for cold water-immersion. Instead, clinicians choose to apply ice packs on major arteries and use alcohol wipes to induce evaporative cooling for EHS patients, both of which have demonstrated inadequate cooling rates for EHS treatment (Casa et al., 2007b). Literature suggest that cooling modality for EHS treatment should have a cooling rate of $0.15^{\circ}\text{C min}^{-1}$, which can be achieved using cold water-immersion of $1\sim 20^{\circ}\text{C}$ (Casa et al., 2007b; McDermott et al., 2009). Other methods such as ice packs covering the body ($\approx 0.03^{\circ}\text{C min}^{-1}$) and fanning ($\approx 0.05^{\circ}\text{C min}^{-1}$) have very limited ability to cool exercise-induced hyperthermia in a timely manner. Since the severity of organ damage and chance of survival are dictated by the duration of sustained internal body temperature above 40°C (Casa et al., 2007b), the ability to cool the patient before transport will directly impact the chance of survival from EHS. Inconvenience from cold water immersion (i.e., wet skin, limited access to supplemental treatments) should not outweigh the need to protect organs from sustained hyperthermia (Casa et al., 2007b). Although the chance is very unlikely, cardiopulmonary arrest case that warrant automated external defibrillator (AED) application would be managed, similarly regardless of the use of cold water immersion; patient should be removed to a dry area, have the contact area for AED pads dried, and promptly begin cardiopulmonary resuscitation and apply AED (Casa et al., 2007b). **Figure 2** compares the common paths of EHS triage with and without the rectal thermometry and whole-body cold-water immersion as accepted methods.

EXERTIONAL HEAT STROKE MANAGEMENT STRATEGIES FOR THE TOKYO 2020 SUMMER OLYMPICS

Recent documents (Japan Medical Association, 2015; Matsumoto, 2018) have reiterated the importance of the

use of rectal thermometry and cold water immersion for EHS diagnosis and treatment; however, due to the aforementioned barriers, they have yet to become the standard of care in Japan. One of the solutions to overcome these barriers before the Tokyo 2020 Summer Olympics is to implement a medical control system that allow trained professionals (e.g., physician, nurse, emergency medical technician, and athletic trainer) to implement an EHS specific pre-hospital care, allowing the recognition and treatment of EHS *before* arriving to the hospital (Belval et al., 2018). Since the number of medical professionals who are trained to use of rectal thermometry and cold water immersion for EHS patients is currently limited, the Summer Olympic Games can turn into a great opportunity for Japanese medical professionals to receive training in these methods and support a paradigm shift in the outdated EHS diagnosis and treatment methods used in Japan. In the context of mass sporting, establishing the chain of commands ahead of the time to coordinate not only the on-site medical personnel but also the local hospital network, event officials, and security becomes vital in ensuring that all parties involved in the care of the collapsed athlete are following the same protocol (Adams et al., 2018).

In addition to the proper pre-hospital care for EHS, there should be pre-established criteria to modify, discontinue, or cancel competitions to safeguard athletes especially with the growing prevalence of extreme heat days exceeding $\geq 28^{\circ}\text{C}$ in wet bulb globe temperature (WBGT) (American College of Sports Medicine Armstrong et al., 2007). Since external factors, such as the number of preliminary matches that must be completed within the fixed number of days, sponsorship, and primetime hours for broadcasting will likely influence the competition hours for televised sporting events, lack of flexibility to adjust the competition time freely will greatly affect the ability to make modifications to the existing schedule. Nevertheless, event organizer should strive to avoid the time of extreme heat according to the historical climate data. Furthermore, there should be an action plan for environmental heat hazards that outlines (1) the upper WBGT threshold to modify, discontinue or cancel events, (2) the chain of command about who makes the final decision to modify, discontinue or cancel events, and (3) specific actions that need to be followed once an event is modified, discontinued, or canceled. Similar to the medical control system for EHS pre-hospital care, emergency action plan for environmental heat hazard should be shared among all parties involved in the event. Moreover, simulating the worst case possible will help elucidate the extent (or lack) of resources available to handle such an emergency, and will facilitate discussions to improve organizational resilience to heat by addressing specific scenarios of anticipated heat hazards (Adams et al., 2018).

CONCLUSION

In conclusion, an evidence-based countermeasure for EHS has not been implemented in Japan and strong leaderships from medical and athletic communities are warranted to reduce the risk of EHS at the Tokyo 2020 Summer Olympics.

The need to implement evidence-based management of EHS has long been overlooked due to the unfamiliarity of the condition by most healthcare providers. It is critical that medical community recognize the shortcoming of their current practice and implement what is proven to be best in the current literature.

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AUTHOR CONTRIBUTIONS

YH created the main conceptual ideas for the paper. All authors conducted a thorough review of the existing literature and contributed to the manuscript writing and review process.

Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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The Relationship Between Self-Efficacy and Aggressive Behavior in Boxers: The Mediating Role of Self-Control

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OPEN ACCESS

Edited by:

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Specialty section:

This article was submitted to
Movement Science and Sport
Psychology,
a section of the journal
Frontiers in Psychology

Received: 27 July 2018

Accepted: 22 January 2019

Published: 19 February 2019

Citation:

Chen X, Zhang G, Yin X, Li Y,
Cao G, Gutiérrez-García C and Guo L
(2019) The Relationship Between
Self-Efficacy and Aggressive Behavior
in Boxers: The Mediating Role
of Self-Control.
Front. Psychol. 10:212.
doi: 10.3389/fpsyg.2019.00212

Aggressive behavior has been one of the core issues in sports psychology, whereas boxers' aggressive behavior has received limited attention. Although some literature reported that self-efficacy is related to aggressive behavior, the mechanism whereby self-efficacy affects aggressive behavior remains unclear. The present study investigated the relationship between self-efficacy and aggressive behavior, as well as the effect of self-control as a mediating factor. This study uses the Self-efficacy Scale for Athletes, the Self-control Questionnaire for Athletes, and the Buss-Perry Aggression Questionnaire. This relationship is explored through self-reported measures from $N = 414$ Chinese professional boxers, $n = 243$ were male and $n = 171$ were female, the average age was $M = 17.72$ years ($SD = 3.147$), the participants, the average number of years of exercise was $M = 3.89$ years ($SD = 2.734$); Results showed that male boxers reported greater aggression than female boxers; It was found that the self-efficacy and self-control improved as age of the participants increased; The higher the level of competition, the higher levels of self-efficacy and self-control; Self-efficacy was negatively related with aggressive behavior and positively correlated with self-control. Self-control was also negatively correlated with aggressive behavior among boxers. Self-control had a full mediating effect on the relationship between self-efficacy and aggressive behavior.

Keywords: self-efficacy, self-control, aggressive behavior, mediating effect, boxer

INTRODUCTION

Athletes abide by the Olympic motto, "faster, higher, stronger" in their arduous training and competitions, and they strive to maintain an optimal performance state (United States Olympic Committee [USOC], 1993). While rigorously improving themselves and pursuing optimal performance, they may encounter various psychological problems. Aggressive behavior in athletes is one of the core issues concerning the *International Society of Sports Psychology* (Tenenbaum et al., 1997). Most studies have shown that there is a significant negative correlation between aggressive behavior and sports performance (McCaw and Walker, 1999; Shafizadeh, 2008). Most of the existing research focused on

ordinary people's aggressive behavior; and the research on aggressive behaviors in sporting contexts focused on Football athletes (Krenn and Meier, 2018), Basketball athletes (Carleton et al., 2016), Rugby athletes (Kerr, 2018), Ice hockey athletes (Cusimano et al., 2016), Baseball athletes (Krenzer and Splan, 2018), etc. However, there is a lack of attention to the aggressive behavior of combat athletes, such as Boxer, Kickboxing, Muay Thai, etc. Furthermore, unlike group ball sports, Boxing is a combat sport that places two individuals in intense one-on-one physical and mental competition where physical injury or even death (British Medical Association Scientific Department, 1993) is a distinct possibility. Therefore, this study focuses on aggressive behavior of boxers, and investigates whether there are differences between different levels of boxers' aggressive behaviors.

In the social learning theory, Bandura (1973) attributed aggressive behavior to a wide variety of social phenomena. Anderson and Bushman (2002) defined aggressive behavior as exerting intentionally inflicts substantial harm to another individual, and the formation of aggressive behavior is a cognitive process in which personal and environmental factors influence each other. In the field of sports, previous research suggests that malicious aggressive behavior in sporting competitions can lead to anti-social behavior in athletes (Rowe, 1998; Kavussanu et al., 2013). As such, aggressive behavior remains to be one of the most critical issues in many disciplines. Bandura (1997) found that reducing aggression and violence in adolescents requires very strong self-efficacy, and most researchers believe that self-efficacy can protect the physical and mental health of individuals (Benight and Bandura, 2004; Luszczynska et al., 2009). Self-efficacy is defined by Bandura (1977, 1986) as the degree of one's feelings about one's ability to accomplish goals. Some studies have shown that self-efficacy is related to aggressive behavior (Pompili et al., 2007; Piko and Pinczés, 2014; Allen et al., 2018). Few studies have been explicitly conducted to assess the association of self-efficacy with aggressive behavior (Wu et al., 2015; Brubacher et al., 2016). Additionally, prior studies have neglected to explore the intrinsic mechanism of self-efficacy that affects aggressive behavior. Therefore, this study surveyed Chinese professional boxers to shed some light on the impact of self-efficacy on aggressive behavior and of the paths of such effects. This provides theoretical guidance for the prevention and intervention of aggressive behavior in athletes.

Self-Efficacy and Aggressive Behavior

According to social cognitive theory, aggressive behavior is caused by the cognitive bias of the aggressor, and low expectations of one's ability and performance may lead to anti-social behavior (Ganooverway et al., 2009). Specifically, reducing aggressive and violent behavior requires for a strong sense of self-efficacy (Bandura, 1994; Bandura, 1997). Studies have found that self-efficacy in emotion regulation effectively regulated the externalization of aggression and criminal behavior, and that self-efficacy in emotion regulation was negatively related with aggressive behavior (Caprara et al., 2010; e.g., Wu et al., 2015). In addition, studies in different research areas have shown that self-efficacy predicted aggressive behavior (Buser et al., 2015). Particularly, in sports, researchers have found that self-efficacy

acted as a psychological mechanism that triggers aggressive behavior and that the use of exercise intervention may improve self-efficacy and then reduce aggressive behavior (Khademi Mofrad and Mehrabi, 2015). In other words, an increase in self-efficacy resulted in a decrease of aggression (e.g., Buser et al., 2015). The sense of self-efficacy in sports refers to the individual's belief of sports ability, and it is the individual's assessment about whether he or she could use his or her own abilities or skills to complete the tasks in sports, in other words, it can refer to a subjective judgment that controls one's sports behaviors and competence (Wei et al., 2008). Based on the considerations aforementioned, we assume that self-efficacy helps athletes to deal with negative emotions effectively and to restore the balance of physical and psychological states, subsequently inhibiting aggressive behavior: Boxers with higher self-efficacy should display lower aggression levels. Thus, the first hypothesis (H1) states that self-efficacy is a significant negative predictor of aggressive behavior.

Self-Control and Self-Efficacy

Self-regulation theory proposes that self-control is influenced by self-efficacy (e.g., Bandura, 1977). In general, self-efficacy is an important factor that influences an individual's ability to exert self-control and it can further explain the ability to exhibit self-control (Graham and Bray, 2015). Baumeister (2002) found that self-control depends on an individual's self-control resources, while self-efficacy complements such resources by acting as a positive emotion. Numerous studies have shown a positive association between self-efficacy and self-control, and that self-efficacy can significantly predict self-control (Hamedani, 2013; Au, 2015; Huang and Yang, 2015). In sports, the theory of challenge and threat posits that under a challenging situation, athletes may exhibit a higher self-efficacy and sense of control, hold approach achievement goals, and perform better in competitions (Jones et al., 2009). On the one hand, Chinese researchers have found that self-efficacy and self-control in athletes influence each other (Li et al., 2013). On the other hand, in boxing, high self-efficacy and high self-control ability are prerequisites for boxers to excel (Ba and Zhu, 2008). Athletes' self-control refers to the individuals' ability to overcome or change internal reactions, suppress impulses, and interrupt impulsive behavior response trends, such as changing and adjusting behaviors, thoughts, emotions, and habituation (Li and Zhang, 2011). So, does a boxer's self-efficacy have a positive effect on self-control? There are currently limited academic research results on this. Therefore, our second hypothesis (H₂) states that self-efficacy and self-control of boxers are positively correlated.

Self-Control and Aggressive Behavior

Several studies have suggested the potential role of self-control in aggressive behavior (DeWall et al., 2011). Self-control can improve the ability to adapt to the living environment (Coyne and Wright, 2014). Most researchers have found a significant negative correlation between self-control and aggressive behavior (White and Turner, 2014; Pung et al., 2015; Meldrum et al., 2016). They have also found that self-control training can reduce an individual's level of aggression (Denson, 2013;

Duckworth et al., 2014; Wang et al., 2017). Researchers have observed that individuals with reduced self-control were likely to exhibit more aggressive behavior (Teng et al., 2014; Osgood and Muraven, 2016). In sports, self-control plays a critical role in the performance of athletes and is decisive in the control of aggressive behavior (Dorris et al., 2012). However, mixed findings also existed. For example, it has been found that individuals who engage in self-control display greater aggression than those who do not (Dewall et al., 2007; Stucke and Baumeister, 2010). The general theory of crime is of the view that all delinquencies and problem behaviors are due to low self-control (Gottfredson and Hirschi, 1990). By improving the internal psychological characteristics of self-control, individuals can reduce aggressive and problem behaviors (e.g., Gottfredson and Hirschi, 1990; Xin et al., 2007). Several research have shown that among different groups, self-control played a mediating role between other psychological traits and aggressive behavior (Li et al., 2017; Song et al., 2017; e.g., White and Turner, 2014; Zhang, 2016). Therefore, there is still a lack of research on the relationship between self-control and aggressive behavior in sports; does self-control also play a mediating role between self-efficacy and aggressive behavior in boxers? In view of the lack of research on the interactions between self-efficacy, self-control, and aggressive behavior, Based on the above literature, this study proposed our hypothesis H3 and H4: Self-control would be negatively correlated with aggressive behavior, self-control would play a mediating role in the relationship between self-efficacy and aggressive behavior.

MATERIALS AND METHODS

Participants

This study adopted cluster sampling and selected boxers from the Chinese national boxing team and boxing teams of Sichuan, Chongqing, Guizhou, and Yunnan provinces as participants to complete a survey questionnaire. Permission was obtained from the University's Human Research Ethics Committee. Prior to answering the items, participants read information about the purpose of the study, implications of participation, and data protection. The information stressed that participation was completely voluntary and anonymous. A total of 450 questionnaires were distributed and $N = 414$ valid ones were returned (92% response) rate. Among the participants, $n = 243$ were male (58.7%) and $n = 171$ were female (41.3%). According to Chinese technical classification of athletes, $n = 255$ participants were classified as Level 3 athletes (54.3%), $n = 57$ as Level 2 athletes (13.8%), and $n = 64$ as Level 1 athletes (15.5%), and 68 athletes at the Master Level or above (16.4%), the higher the level in turn (Gazette of the State Council of the People's Republic of China, 1995). Their average age was $M = 17.72$ years ($SD = 3.147$), and their average experience in training was $M = 3.89$ years ($SD = 2.734$). Sixty-nine participants were younger than 16 years. Written and informed consent was obtained from the parents/legal guardians of all non-adult participants.

Procedure

Written and informed consent was obtained from the parents/legal guardians of all non-adult participants. The participants in this study were all national boxers who were strictly trained before the survey was conducted. After receiving informed consent from the management, coaches, and athletes of the national team and other sports teams, the questionnaire was distributed to teams at the provincial level or above. The questionnaires were distributed at the National Olympic Training Center, Beijing Sport University, Qujing in Yunnan, Qingzhen in Guizhou, Shapingba in Chongqing, and Liangshan in Sichuan between the 12th of June and the 8th of July, 2017. The instructions were explained in detail and example questions were provided to the participants, who were asked to read the questionnaire carefully and answer according to their actual circumstances. The questionnaire took about 20 min to complete.

Measures

This study employed the surveys. Specifically, the following three questionnaires were included in the study, a total of questions that took minutes to complete by boxer participants.

Self-Efficacy Scale for Athletes

Based on self-efficacy theory and related theories and knowledge about competitive sports, Wei et al. (2008) compiled a self-efficacy scale for athletes in heavy athletics sport. This scale included 15 items, such as "I can keep my mind clear and focused during the competition." Each item was measured by a five-point scale (1 = never been like this; 5 = always so). A higher score indicates a higher self-efficacy. The one-dimensionality of the scale was proved by a confirmatory factor analysis (CFA): $\chi^2/df = 1.086$, RMSEA = 0.014, TLI = 0.996, GFI = 0.976, NFI = 0.969, CFI = 0.997, IFI = 0.997. The factor loadings (regression coefficients) of the items ranged from $\alpha = 0.355$ to $\alpha = 0.690$. The internal consistency of the scale was good (Cronbach's $\alpha = 0.91$).

Self-Control Questionnaire for Athletes

Li and Zhang surveyed 820 professional athletes in China and created a self-control scale for athletes. Their questionnaire contains 24 items, which are scored on a five-point scale, ranging from "1 = not at all" to "5 = very much"; higher scores indicate a better self-control (item example: "In order to complete the training task, I can endure extreme fatigue."). In a previous study, the scale provided a reference for athlete selection (e.g., Li and Zhang, 2011). A confirmatory factor analysis proved the one-dimensionality of the scale $\chi^2/df = 1.220$, RMSEA = 0.023, TLI = 0.975, GFI = 0.954, NFI = 0.910, CFI = 0.982, IFI = 0.982. The factor loadings of the items ranged between $\alpha = 0.380$ and $\alpha = 0.652$. The internal consistency of the scale was good (Cronbach's $\alpha = 0.85$).

Buss-Perry Aggression Questionnaire

We used the modified Chinese version (Yang and Wang, 2012) of the Aggressive Behavior Questionnaire developed by Buss and Perry (1992) to assess adolescent aggressive

behavior. The questionnaire consists of four dimensions. The physical aggression CFA results were good: $\chi^2/df = 1.762$, RMSEA = 0.043, TLI = 0.975, GFI = 0.943, NFI = 0.972, CFI = 0.987, IFI = 0.987. The verbal aggression CFA results were good: $\chi^2/df = 1.520$, RMSEA = 0.035, TLI = 0.988, GFI = 0.965, NFI = 0.986, CFI = 0.995, IFI = 0.995. The anger CFA results were good: $\chi^2/df = 1.105$, RMSEA = 0.016, TLI = 0.997, GFI = 0.972, NFI = 0.984, CFI = 0.998, IFI = 0.998. The hostility CFA results were good: $\chi^2/df = 1.196$, RMSEA = 0.022, TLI = 0.993, GFI = 0.957, NFI = 0.974, CFI = 0.996, IFI = 0.996. The smallest item factor loadings was $\alpha = 0.461$, the highest $\alpha = 0.709$. A representative item was “Once in a while I can’t control the urge to strike another person.” The questionnaire contains 29 items in total. Items were scored on a five-Likert scale, ranging from 1 = very non-compliant to 5 = very much in line, with higher scores meaning more aggressive behavior. The higher the total score, the higher the level of aggressive behavior in the athlete. The Cronbach’s alpha coefficients for the physical aggression (nine items), verbal aggression (five items), anger (seven items), and hostility (eight items) dimensions were 0.816, 0.755, 0.809, and 0.802, respectively. The Cronbach’s alpha coefficient for the total scale was 0.92 indicating high reliability. The Chinese version of this scale had a Cronbach alpha of 0.94 (e.g., Yang and Wang, 2012).

Data Analysis

This study used SPSS 21.0 for statistical analysis (including correlation analysis, regression analysis, and variance analysis) and AMOS 20.0 for constructing models and conducting path analyses. Following the two-step procedure recommended by Gerbing and Anderson (1988), this study tested the measurement model before construction. We used CFA for data analysis to confirm the differences between the factors. If the discriminant validity of the variables was good, we would initiate the structural model analysis as the next step.

RESULTS

Common Method Bias

There is a risk of common method bias by collecting data using questionnaires; thus, this study adopted the method proposed by previous researchers (Zhou and Long, 2004; Xiong et al., 2012) to

control for common method bias. Harman’s single factor test was applied to test for common method bias (Podsakoff et al., 2012). The results showed that there were 16 factors with an eigenvalue greater than 1 and the first factor had an explanatory variance of 21.52%, which is lower than the threshold of 40%, indicating that the common method bias was not significant.

Self-Efficacy, Self-Control, and Aggressive Behavior: Group Differences

The mean total score for self-efficacy was 3.47, SD = 0.62, indicating that the overall self-efficacy score of this sample of Chinese boxers was generally high. As shown in **Table 1**, this study found the mean self-efficacy score was significantly different between boxers of different technical grades ($F = 9.423$, $p < 0.001$; i.e., Master or above > Level 3, Level 1 > Level 3, Level 2 > Level 3). A regression analysis found that as the age of the boxers increased, the level of self-efficacy also increased ($\beta = 0.224$, $p < 0.001$), and that as the boxers’ number of years of training increased, the level of self-efficacy also increased ($\beta = 0.230$, $p < 0.001$).

The mean total score for self-control was 3.69, SD = 0.62, indicating that the overall self-control score was generally high. As shown in **Table 1**, the results show there were significant differences between self-control in boxers belonging to the four different technical grades ($F = 10.710$, $p < 0.001$; i.e., Master or above > Level 3, Level 1 > Level 3, Level 2 > Level 3). A regression analysis found that the higher the age of boxers, the higher the level of self-control ($\beta = 0.188$, $p < 0.001$), and that as the boxers’ number of years of training increased, the level of self-control also increased ($\beta = 0.202$, $p < 0.001$).

The mean total score for aggressive behavior was 2.23, SD = 0.57, indicating that the overall aggression level of the Chinese boxers was generally low. This study indicates the aggression levels in males and females were significantly different, with males having higher aggression levels than females ($T = 2.830$, $p < 0.01$). Physical aggression levels were significantly different in male and female boxers, with males having higher aggression levels than females ($T = 3.408$, $p < 0.01$). There were significant differences with regard to physical aggression in boxers belonging to the four different technical levels of classification (Master or above, $M = 1.75$, SD = 0.64; Level 1, $M = 1.94$, SD = 0.67; Level 2, $M = 1.85$, SD = 0.66; Level 3,

TABLE 1 | Differences in self-efficacy between boxers of different sports levels.

	Level	M	SD	F	Comparison between groups
Self-efficacy	Master or above	3.75	0.64	9.423***	Master or above > Level 3, Level 1 > Level 3, Level 2 > Level 3
	Level 1	3.55	0.62		
	Level 2	3.57	0.61		
	Level 3	3.33	0.59		
Self-control	Master or above	3.82	0.48	10.710***	Master or above > Level 3, Level 1 > Level 3, Level 2 > Level 3
	Level 1	3.76	0.44		
	Level 2	3.88	0.42		
	Level 3	3.57	0.44		

* indicates $p < 0.05$, ** indicates $p < 0.01$, and *** indicates $p < 0.001$.

$M = 2.05$, $SD = 0.63$, $F = 4.384$, $p < 0.001$). A regression analysis found that as the boxers' number of years of training increased, the lower the physical aggression levels ($\beta = -0.122$, $p < 0.05$). Male and female boxers showed significant differences in verbal aggression, with females having higher verbal aggression levels than males ($T = 2.807$, $p < 0.01$). As shown in **Table 2**.

Self-Efficacy, Self-Control, and Aggressive Behavior: Correlations

As shown in **Table 3**, self-efficacy was positively correlated with self-control, self-efficacy was negatively related to aggressive behavior, and self-control was correlated negatively with aggressive behavior. The significant correlations between the variables in this study provided a basis for subsequent testing of mediating effects. Therefore, hypotheses H_1 , H_2 , and H_3 were confirmed.

The Mediating Effect of Self-Control

Structural equation modeling (SEM) was used to analyze the relationships among the variables with self-efficacy as the exogenous ("independent") variable and aggressive behavior as the endogenous ("dependent") variable. Physical aggression, verbal aggression, anger, and hostility were observational variables in the model, and gender was a control variable in the model. According to the testing procedure of mediating effects (Wen et al., 2004; Preacher et al., 2006), the direct effect of self-efficacy on aggressive behavior was to be tested first, followed by the fitness of the model and the significance of each path coefficient after adding in the mediating variable. The fitness indicators of the SEM direct effect analysis results were as follows: $\chi^2/df = 2.637$, RMSEA = 0.063, TLI = 0.967, CFI = 0.989, GFI = 0.992, NFI = 0.983, IFI = 0.989. The direct effect path coefficient of self-efficacy on aggressive behavior was significant ($\beta = -0.36$, $p < 0.001$).

Self-control was placed as a mediating variable between self-efficacy and aggressive behavior in boxers and the fitness results shown in **Figure 1** were as follows: $\chi^2/df = 3.115$, RMSEA = 0.072 (90% CI for RMSEA = 0.043, 0.102), TLI = 0.960, CFI = 0.983, GFI = 0.982, NFI = 0.975, IFI = 0.985. The path coefficients between self-efficacy and self-control ($\beta = 0.62$, $p < 0.001$), self-control and aggressive behavior ($\beta = -0.65$, $p < 0.001$), and gender and aggressive behavior ($\beta = -0.11$, $p < 0.01$), were

significant. However, after adding the mediating variable, the path coefficient between self-efficacy and aggressive behavior turned from significant ($\beta = -0.36$, $p < 0.001$) to non-significant ($\beta = 0.09$, $SE = 0.043$, $p > 0.05$). Therefore, the results indicate that self-control had a full mediating effect on the relationship between self-efficacy and aggressive behavior among boxers, thus confirming hypothesis H_4 of this study.

DISCUSSION

The results showed that boxers with high self-efficacy exhibited less aggressive behavior than those with low self-efficacy. Self-control played a mediating role in the relationship between self-efficacy and aggressive behavior in boxers, further revealing the mechanism behind the relationship.

Group Differences in Self-Efficacy, Self-Control, and Aggressive Behavior

The results of this study indicated that there were significant gender differences in aggressive behavior and physical aggression in boxers, with men showing greater aggressive behavior than women; however, verbal aggression was higher in women than men. Different gender depended to a large extent on differences in individual traits, while it also confirmed prior findings (Coulomb-Cabagno and Rasclé, 2006; Maria et al., 2006; Wu and Jiang, 2012). Some scholars believe that there are many reasons for the gender differences in aggressive behaviors, which are the result of the combination of multiple factors (Carol, 1974). Some studies have found that boys are more likely to face risk factors of attack development, such as nervous system dysfunction, difficult temperament, impulsivity, and learning disabilities (Gorman-Smith and Loeber, 2005; Lahey et al., 2006). What's more, studies have also shown that men are more likely to develop physical aggression due to this different level of emotional arousal (Knight et al., 2010). Men are more aggressive than women in exercise or in daily life (Card et al., 2008; Guerra et al., 2011).

There were no significant gender differences in self-efficacy and self-control. Instead, both the number of years of training and age were significant positive predictors, and the higher the technical grade, the higher the scores on the two variables. These results were in line with previous studies (Vaughn et al., 1984; Zhu and Pan, 2004; Cheng et al., 2010). In the part of demographic difference in self-control and self-efficacy, I have added: as the boxer's age and the years of exercise increases, the individual's psychological traits tend to be a process of continuous improvement of mental health; it is obvious that the physical and mental health of boxers have improved a lot, and then the level of self-efficacy and self-control of boxers has increased significantly, which is also in line with the basic laws of human evolution theory.

Direct Influence of Self-Efficacy on Aggressive Behavior

Correlation analyses showed that self-efficacy was negatively correlated with aggressive behavior in boxers. This is consistent

TABLE 2 | Comparison of different gender boxers in Aggressive behavior, Physical aggression, and verbal aggression.

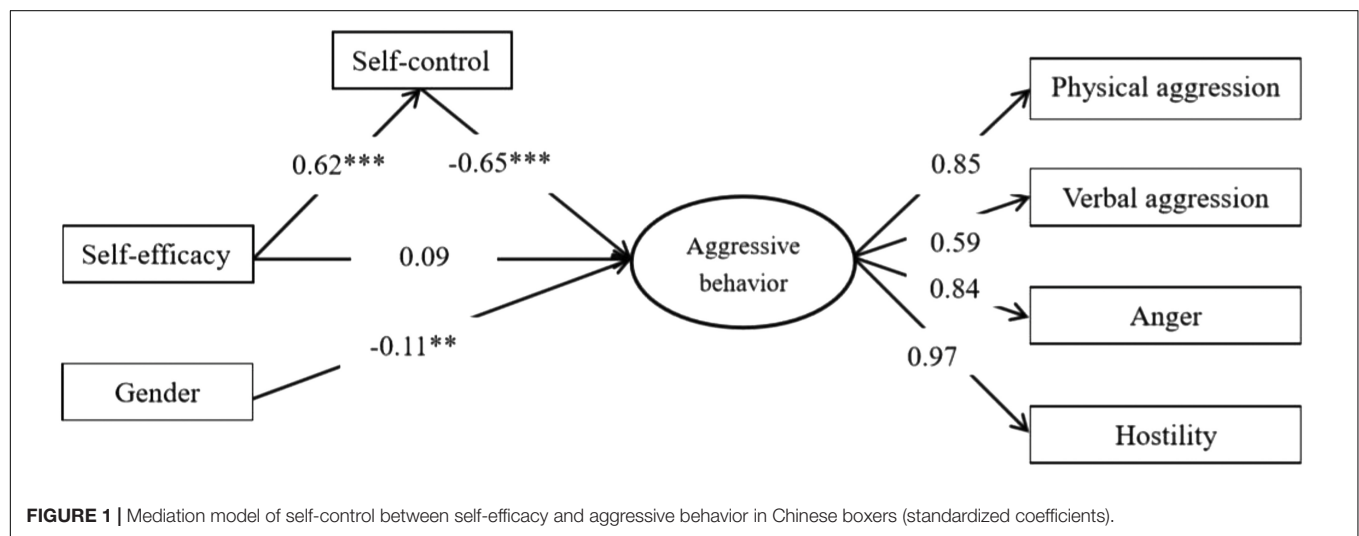
	Gender	<i>M</i>	<i>SD</i>	<i>T</i>	Comparison between groups
Aggressive behavior	Male	2.32	0.55	2.830*	Male > Female
	Female	2.16	0.58		
Physical aggression	Male	2.08	0.62	3.408*	Male > Female
	Female	1.86	0.66		
Verbal aggression	Male	2.55	0.67	2.807*	Female > Male
	Female	2.75	0.66		

* indicates $p < 0.05$, ** indicates $p < 0.01$, and *** indicates $p < 0.001$.

TABLE 3 | Relationships between Demographic Variables, Self-efficacy, Self-control, Aggressive Behavior, and Physical Aggression.

	<i>M</i>	<i>SD</i>	1	2	3	4	5	6	7	8	9	10
1. Age	17.72	3.15	–									
2. Number of years of exercise	3.89	2.73	0.836***	–								
3. Competitive level	4.03	1.23	0.720***	0.696***	–							
4. Self-efficacy	3.47	0.62	0.224***	0.230***	0.245***	–						
5. Self-control	3.69	0.46	0.188***	0.202***	0.216***	0.618***	–					
6. Aggressive behavior	2.23	0.57	–0.119*	–0.089	–0.101*	–0.313***	–0.602***	–				
7. Physical aggression	1.95	0.64	–0.148**	–0.122*	–0.159**	–0.230***	–0.522***	0.843***	–			
8. Verbal aggression	2.63	0.74	–0.013	–0.006	0.024	–0.226***	–0.350***	0.749***	0.478***	–		
9. Anger	2.27	0.74	–0.108*	–0.078	0.037	–0.255***	–0.515***	0.844***	0.600***	0.548***	–	
10. Hostility	2.27	0.66	–0.097*	–0.065	0.088	–0.325***	–0.567**	0.866***	0.624***	0.596***	0.631***	–

* indicates $p < 0.05$, ** indicates $p < 0.01$, and *** indicates $p < 0.001$.



with previous studies (Buser et al., 2015; Brubacher et al., 2016; e.g., Khademi Mofrad and Mehrabi, 2015). Social cognitive theory and self-efficacy theory propose that as self-efficacy increases, aggression levels of individuals decrease, and the expectation value of their own performance and ability rises. This creates a positive attitude in individuals. The results of this study to a certain extent support the views of social cognitive theory and self-efficacy theory, that self-efficacy influences aggression levels in boxers. Most empirical studies have found that self-efficacy can not only affect the individual's psychological state (Bandura, 1986; Crick and Dodge, 1994) but also effectively deter and reduce the occurrence of aggressive behavior and violence (Caprara et al., 2010). Researchers have also found that among different groups, individuals with high self-efficacy generally exhibited lower aggression levels and were physically and mentally healthier (Brubacher et al., 2016; e.g., Khademi Mofrad and Mehrabi, 2015). With regard to boxing, improving self-efficacy in boxers is not only necessary for performance in competition but also a protective factor in preventing physical and mental health problems. If the level of self-efficacy declines in boxers, it will inevitably increase the likeliness of their aggressive behavior. This study confirms the relationship between

self-efficacy and aggressive behavior, whereby self-efficacy is a significant negative predictor of aggressive behavior in boxers.

Self-Control as a Mediator Between Self-Efficacy and Aggressive Behavior

This study showed that in boxers, self-efficacy was positively correlated with self-control and self-control was negatively correlated with aggressive behavior. This is consistent with previous studies (Hamama and Ronen-shenhav, 2012; Hamedani, 2013; Eingar and Steinhart, 2017). Self-regulation theory and theory of Challenge and Threat States in Athletes propose high self-efficacy and high self-control are important factors for winning at critical moments in sports competitions and they constrain and promote each other (Hamedani, 2013; Li et al., 2013). Meanwhile, this study was also concerned with the deeper mechanisms of self-efficacy and self-control in boxing and offers specific explanations of the theories of self-regulation and challenge and threat states in athletes. Among boxers, self-efficacy and self-control are not only important indicators for selection but also a key factor for competitive performance. Both play a positive

role in the development of the physical and mental health of boxers.

In this study, the intrinsic relationship between self-control and aggressive behavior in boxers validates the integrative cognitive model of aggression. Several studies have shown that self-control has the ability to change and constrain aggressive behavior (Denson et al., 2011; Sofia and Cruz, 2015). As a major applied and practical implication, integrating strategies and skills to strengthen self-control capacity and resources (e.g., Duckworth et al., 2014) would certainly be a useful and potential way to tackle the problem of aggression in sport competition, particularly under the stress and pressure situations occurring in sport competition. In intense boxing competitions, boxers who display high self-control are able to reduce impulsivity and maintain rational thinking, which helps them win the game. In addition, if they consider the long-term consequences of their behavior, they will achieve the purpose of reducing aggressive behavior. Therefore, this study establishes the mechanisms behind self-efficacy, self-control, and aggressive behavior.

After confirming the reverse predictive effect of self-efficacy on aggressive behavior, we introduced a mediating variable and explored the mechanism behind the effect. Results indicated that self-control had a full mediating effect in the relationship between self-efficacy and aggressive behavior. From the perspectives of self-regulation theory and the general theory of crime, individuals can rely on their levels of self-control to regulate their own behavior and deter the occurrence of criminal and problem behaviors (Bandura, 2012). By examining the mediating role of self-control in boxers, self-regulation theory and the general theory of crime are further reinforced. We have also found that these two theories are also applicable in boxing. Researchers from different fields have also found that self-control acted as a mediator between other variables and aggressive behavior (Shepperd et al., 2015; Li et al., 2017; e.g., White and Turner, 2014). The same result has been observed in the field of sports (e.g., Zhang, 2016). Combining these results, this study proposes that the self-efficacy should be a focus in competitive training, and that effective training methods should be adopted to improve the ability of self-control, in order to prevent boxers from engaging in aggressive behavior in high-pressure training or competitions. Hence, self-control plays a mediating role between self-efficacy and aggressive behavior in boxers.

Limitations

Although this study contributes to explore the mechanisms of self-efficacy and aggressive behavior, as well as the mediating role of self-control in that relationship, it has two limitations. Firstly, a cross-sectional design was used in this study, which rendered it impossible to make causal inferences. Future studies should adopt a longitudinal design to better understand the process whereby self-efficacy influence aggressive behavior. Secondly, we focused on the role of self-control in the relationship between self-efficacy and aggressive behavior, but there are still many other important mediating variables to be explored, such as personality traits or self-esteem, which should be also explored in future

research. Whether the results of this study can be generalized to other sports remains to be verified. Despite these limitations, the results of this study help us understand the intrinsic relationship between self-efficacy and aggressive behavior to a certain extent as well as its possible causes and mechanisms in the context of Chinese culture. The exploration of the variables and processes involved in this meta-theory, as well as attempted to test its major assumptions, can add a fruitful avenue toward the development and advancement of knowledge about the processes involved in aggression in sport.

CONCLUSION

Male displayed more aggressive behavior than female. As age increased, the levels of self-efficacy and self-control also increased. The higher the competitive level, the higher the self-efficacy and self-control; and the higher the number of years of training, the higher the self-efficacy and self-control. Self-efficacy was negatively correlated with aggressive behavior and positively correlated with self-control. Self-control also negatively correlated with aggressive behavior. Furthermore, self-control had a full mediating effect between self-efficacy and aggressive behavior in the current study.

In summary, training boxers' self-efficacy and self-control in daily training and competition reduces their aggressive behavior. Then, boxers' self-efficacy, self-control and aggressive behavior should be considered as the key indicator of psychological selection, and the mechanism of psychological selection of boxers should be improved. Therefore, this new conceptual framework can be a valuable and novel perspective for future research of aggression in applied fields such as sport, providing possible targets for intervention and forming a basis for further research.

ETHICS STATEMENT

This study was carried out in accordance with the recommendation of the University's Human Research Ethics Committee with written informed consent from all subjects. All subjects gave written informed consent in accordance with the Declaration of Helsinki. The protocol was approved by the Southwest University's Human Research Ethics Committee.

AUTHOR CONTRIBUTIONS

XC, GZ, XY, YL, GC, CG-G, and LG conceived the study, interpreted the data, drafted and revised the work, approved the final version of the manuscript to be published, and agreed to be accountable for all aspects of the work.

FUNDING

This work was supported by the Fundamental Research Funds for the Central Universities of China (SWU1709116) and the Sports Scientific Research Project of Chongqing in 2016 (B201622).

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Considerations When Assessing Endurance in Combat Sport Athletes

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OPEN ACCESS

Edited by:

Toby Mündel,
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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 07 November 2018

Accepted: 18 February 2019

Published: 05 March 2019

Citation:

Barley OR, Chapman DW,
Guppy SN and Abbiss CR (2019)
Considerations When Assessing
Endurance in Combat Sport Athletes.
Front. Physiol. 10:205.
doi: 10.3389/fphys.2019.00205

Combat sports encompass a range of sports, each involving physical combat between participants. Such sports are unique, with competitive success influenced by a diverse range of physical characteristics. Effectively identifying and evaluating each characteristic is essential for athletes and support staff alike. Previous research investigating the relationship between combat sports performance and measures of strength and power is robust. However, research investigating the relationship between combat sports performance and assessments of endurance is less conclusive. As a physical characteristic, endurance is complex and influenced by multiple factors including mechanical efficiency, maximal aerobic capacity, metabolic thresholds, and anaerobic capacities. To assess endurance of combat sports athletes, previous research has employed methods ranging from incremental exercise tests to circuits involving sports-specific techniques. These tests range in their ability to discern various physiological attributes or performance characteristics, with varying levels of accuracy and ecological validity. In fact, it is unclear how various physiological attributes influence combat sport endurance performance. Further, the sensitivity of sports specific skills in performance based tests is also unclear. When developing or utilizing tests to better understand an athletes' combat sports-specific endurance characteristic, it is important to consider what information the test will and will not provide. Additionally, it is important to determine which combination of performance and physiological assessments will provide the most comprehensive picture. Strengthening the understanding of assessing combat sport-specific endurance as a physiological process and as a performance metric will improve the quality of future research and help support staff effectively monitor their athlete's characteristics.

Keywords: physiological assessment, performance monitoring, measurement precision, biology of combat sports, sports specificity, aerobic capacity

INTRODUCTION

Combat sport is a term used to describe a wide range of competitive contact sports typically involving physical combat where the winner is determined by specific criteria depending on the rules of the sport. Combat sports have a large public following with sports such as boxing and mixed martial arts (MMA) having millions of followers and approximately 20% of summer Olympic

medals available in combat sports such as boxing, judo and taekwondo (Franchini et al., 2012; James et al., 2016b; Reale et al., 2016). Combat sports can be categorized as grappling, striking or mixed style sports. Grappling sports involving gripping, throwing, ground combat, chokeholds and joint locks (Ratamess, 2011), while striking sports incorporate skills ranging from only punches to combinations of punches, kicks, knees, and elbows (Rodrigues Silva et al., 2011). Mixed style combat sports involve both grappling and striking, thus requiring a diverse skill-set (Tack, 2013). The rules vary between combat sports resulting in different methods of victory and competition durations, which in turn results in many possible competitive styles, even within the same sport (Tack, 2013). In several combat sports, specific techniques or positions are worth a set number of points and successfully applied techniques are then subsequently totalled to determine victory should an opponent not be defeated via a submission, knock-out (KO) or technical knock-out (TKO; Balmer et al., 2005; Ratamess, 2011). In contrast, some sports utilize an ever-evolving and more subjective scoring system for bouts that are not ended by a submission, KO or TKO. For example, the current scoring system applied to both amateur and professional bouts in several combat sports is referred to as the “10-Point Must System,” whereby judges subjectively decide the winner of the round awarding 10 points and the opponent 9 or less. These scoring systems result in diverse methods of victory in combat sports, and thus the term “performance” is complicated. Indeed, overcoming an opponent through submission in the first minute of an event or on points after fifteen minutes of fighting would both be examples of successful competitive performances but achieved through very different physical characteristics, skill sets and tactics. Thus, for coaches and physical conditioning support personnel assessing such physical characteristics is important for optimizing athlete development, competition tactics and preparations for competition. As a result, a large body of literature has been developed around understanding the physical and physiological characteristics of combat sport athletes (Chaabene et al., 2018; James et al., 2016b).

Combat sports are physically demanding, requiring a diverse physical and physiological profile to be successful in competition (Kraemer et al., 2001; Ratamess, 2011; Bridge et al., 2014; Franchini et al., 2014; Chaabene et al., 2015). Striking movements such as punches and kicks require explosive strength and power (Loturco et al., 2014; House and Cowan, 2015), while grappling movements can require a greater emphasis on isometric and concentric strength (Ratamess, 2011; James et al., 2016b). Additionally, combat sports are comprised of many different sports-specific movements which will influence the physical load. For instance, sports such as boxing and judo exert a greater demand on the upper limbs whilst taekwondo exerts a greater demand on the lower limbs (Bridge et al., 2014; Franchini et al., 2014; Chaabene et al., 2015). Even differences in equipment requirements may influence the physical demands of the sport, such as the use of a kimono in Brazilian Jiu-Jitsu and judo increasing the use of the forearm muscles (Andreato and Branco, 2016). The specific skills and rulesets of a combat sport will significantly influence the energy cost of competition (Crisafulli et al., 2009; Andreato and Branco, 2016; Hausen et al., 2017).

However, combat sports do not typically involve a single execution of one particular technique but instead involve repeated executions interspersed with lower intensity actions (Rodrigues Silva et al., 2011; Franchini et al., 2013; Andreato et al., 2015; Miarka et al., 2015a,b). The high-intensity repeat-effort nature of combat sports typically results in a large aerobic response during exercise as demonstrated by athletes reaching near maximal heart rates and oxygen consumption ($>90\%$ of maximum HR and $\text{VO}_{2\text{max}}$) during simulated competition (Crisafulli et al., 2009; Doria et al., 2009; Campos et al., 2012). Additionally, the high-intensity component of combat sport competitions induces significant anaerobic strain with research observing high levels of blood lactate ($>12 \text{ mmol.L}^{-1}$) following competition (Bouhlef et al., 2006; Hanon et al., 2015). This makes endurance an important characteristic of success in competitive combat sports (Amtmann and Berry, 2003; La Bounty et al., 2011; Ratamess, 2011; Lenetsky and Harris, 2012). However, endurance is a difficult concept to define as it is influenced by a wide range of physiological, psychological and biomechanical factors (Abbiss and Laursen, 2005). For the purposes of this review endurance will be defined as the ability to maintain a high-intensity or repeated efforts over longer exercise durations. The work:rest ratios of high-intensity efforts during competition vary, thus resulting in different endurance profiles between combat sports (Del Vecchio et al., 2011; Rodrigues Silva et al., 2011; Andreato et al., 2015). It is also important to consider the total duration of events in combat sports, with some sports having a single round lasting 10 min and others involving up to twelve 3 min rounds. Furthermore, it is possible that different regulations in performance enhancing drugs and weight cutting practices may also influence performance in different combat sports and levels of competition. These differences create complications when attempting to compare results acquired from exercise testing of athletes from different types of combat sports, especially Olympic and professional ones (Andreato and Branco, 2016). Additionally, the rules of many combat sports are somewhat unique and allow for an athlete to win before the allotted competition time, which makes the total duration highly variable. Indeed, it has been reported that over half of all MMA fights in the Ultimate Fighting Championship® (UFC®) are ended within the first round (Del Vecchio and Franchini, 2013). However, it is not uncommon for fights to last the entire allocated duration (Miarka et al., 2015b). These diverse requirements, due to both the physical requirements and varied length of competition, result in athletes requiring several well-developed physical characteristics including strength, power, agility and endurance on top of technical skill and tactics to be successful (Amtmann and Berry, 2003; La Bounty et al., 2011; Ratamess, 2011). While there are a wide range of potential tools for support staff to use to assess an athlete's relevant physical characteristics, what approaches are most relevant to combat sports success currently are not clear, especially in the case of combat sports endurance related performance (Chaabene et al., 2018). As such, the purpose of this review is to provide a critical appraisal of the current methods of assessing physical capacities in combat sports with a focus on endurance ability in an effort to help develop best practice guidelines.

CURRENT PRACTICES IN ASSESSING PHYSICAL CHARACTERISTICS IN COMBAT SPORTS

It may not be possible to develop a single test that simultaneously assesses all factors of all combat sports performance under controlled conditions. As a result, the best available approach is to assess individually characteristics integral to competitive success, such as strength, power, agility and endurance (La Bounty et al., 2011; Ratamess, 2011; James et al., 2016b) with some insight into the underlying physiology to lesser or greater degree. Effectively identifying and evaluating such characteristics is essential to inform training interventions, nutritional demands, talent identification, and design the optimal competition strategies for athletes. When selecting a test to assess a characteristic in combat sports athletes, it is important to differentiate between tests where the primary outcome measure is an indicator of physiological capacity or performance. Tests of performance are primarily used to simulate competition in a controlled manner as opposed to assessing the function of a physiological system (Bassett and Howley, 2000; Currell and Jeukendrup, 2008). When evaluating the body of research investigating key physical characteristics in combat sports, it is apparent that the employed methods range from physiological assessments with low sports specificity to performance assessments with high sports specificity (Franchini et al., 2011; Chaabène et al., 2012; James et al., 2016b). Thus assessments seeking to isolate an underlying physical characteristic with low sports specificity used across combat sports include: one repetition maximum (1RM) testing (Franchini et al., 2007; Garthe et al., 2011; James et al., 2016b), maximal isokinetic strength assessment (Timpmann et al., 2008), counter-movement and squat jumps (Fogelholm et al., 1993; Garthe et al., 2011; Ouergui et al., 2014; James et al., 2016b; Barley et al., 2018b), 40-m sprint (Garthe et al., 2011), 30 s continuous jump (Čular et al., 2018), repeated contractions on an isokinetic dynamometer (Moore et al., 1992; Kraemer et al., 2001; Oopik et al., 2002; Timpmann et al., 2008; Barley et al., 2018a), Wingate testing (Fogelholm et al., 1993; Artioli et al., 2010; Mendes et al., 2013; Durkalec-Michalski et al., 2014; Ouergui et al., 2014), various repeat-sprint tests (Barley et al., 2018b; James et al., 2018) and maximal aerobic capacity testing (Guidetti et al., 2002; Ravier et al., 2006; Franchini et al., 2011; Bruzas et al., 2014; Reljic et al., 2015; James et al., 2016b). As the spectrum of physical assessments shifts toward a higher level of sport specificity assessment, examples include exercise circuits (i.e., burpees, press-ups, and sports-specific skills such as throws or strikes) (Smith et al., 2000; Franchini et al., 2005, 2007, 2011; Hall and Lane, 2001; Artioli et al., 2010; Chaabène et al., 2012; Villar et al., 2016; Sant'Ana et al., 2017) and simulated combat with a live opponent (Yang et al., 2018) (Table 1). Although the use of such testing modalities appears sound, critical evaluation of the ecological validity of the test or physiological characteristic involved is required, including an understanding of the precision to detect small but important changes.

There is a belief that when assessing a performance characteristic, the assessment should relate as closely as possible

to the sport itself. However, there can be a trade-off between precision of measurement for the physical characteristic and maintaining sporting relevance. While some assessment methods of physical characteristics closely relate to competitive success (James et al., 2016a,b), the relationship for others is much less clear (James et al., 2016b). For example, greater levels of strength and power have been linked to a higher competitive level in combat sports (James et al., 2016a,b) and to greater punching force (Loturco et al., 2014; House and Cowan, 2015). In fact, it appears that the body of research assessing strength and power in combat sports athletes is robust (Loturco et al., 2014; House and Cowan, 2015; Iermakov et al., 2016; James et al., 2016a,b). In contrast, the methods of assessing combat sports-specific endurance are highly varied in the literature and much less robust (Chaabène et al., 2018). This is likely due to the complex nature of endurance as a physical characteristic underpinning combat sport performance, making it far more complicated to assess. Indeed, it is acknowledged that the demand on the aerobic system varies depending on intensity and competition length, with sports involving multiple rounds such as boxing, kickboxing and MMA placing a greater strain on the aerobic system (Smith, 1998; La Bounty et al., 2011; Rodrigues Silva et al., 2011; Alm and Yu, 2013; Del Vecchio and Franchini, 2013; Chaabène et al., 2015). Additionally, sports with single rounds likely require a significant aerobic contribution (Chaabène et al., 2012; Ratamess, 2011; Franchini et al., 2014). However, it is important to note that endurance is influenced by many more factors than just aerobic capacity (Coyle, 1999; Bassett and Howley, 2000; Aziz et al., 2007; Buchheit, 2008; Aguiar et al., 2016). The repeated high-intensity efforts involved require competitive athletes to have well developed strength-endurance, efficiency and anaerobic capacities alongside a capacity to rapidly recover (Coyle, 1999; Ratamess, 2011; Bridge et al., 2014; Chaabène et al., 2015; Salci, 2015). Thus, to better understand endurance ability in a combat sport athlete it is important to consider all factors relevant to the individual combat sport's-specific endurance. Given the lack of clarity in the literature regarding the assessment of endurance relevant to combat sports, the following sections seek to provide recommendations for methods of evaluating characteristics relevant to endurance ability in combat sports athletes.

ASSESSING ENDURANCE IN COMBAT SPORTS ATHLETES FROM A PERFORMANCE PERSPECTIVE

When assessing endurance performance, the overall duration and intermittent nature of the specific combat sport should be taken into account (Del Vecchio and Franchini, 2013). Combat sports with multiple rounds can involve greater than 30 min of high-intensity intermittent activity (Guidetti et al., 2002; Andreato and Branco, 2016). Additionally, combat sport bouts involve the use of a wide range of sports-specific skills that can also influence the physical requirements which may be difficult to replicate under controlled conditions (Bridge et al., 2014; Franchini et al., 2014; Chaabène et al., 2015; Andreato and Branco, 2016). Whilst it

TABLE 1 | Common methods to assessment physical capacities in combat sports athletes.

Assessment	Physical capacity assessed	Reference within combat sports	Does the assessment involve sports-specific skills	Primary outcome variable	Lower body or upper body engaged	Repeat effort	Combat sport the test can be used for
One repetition maximum testing	Strength	Franchini et al., 2007	No	Weight lifted	Upper or lower body	No	Generic
Maximal isokinetic strength assessment	Strength	Barley et al., 2018a	No	Torque generated	Upper or lower body	No	Generic
Counter-movement and squat jumps	Power	Barley et al., 2018a	No	Jump height, and force generated	Lower body	No	Generic
30-s continuous jump	Anaerobic power and capacity	Čular et al., 2018	No	Number of jumps and height of jumps	Lower body	Yes	Generic
Repeated contractions on an isokinetic dynamometer	Repeat-effort endurance	Barley et al., 2018a	No	Torque generated and number of contractions	Upper or lower body	Yes	Generic (depending on effort-relief intervals)
Wingate anaerobic assessment	Power and anaerobic capacity	Ouergui et al., 2014	No	Peak power, average power and fatigue index	Upper or lower body	No (repeated Wingate protocols can be designed)	Generic
Maximal aerobic capacity testing	Aerobic capacity and continuous effort endurance ability	Bruzas et al., 2014	Possibly (in most cases no)	Maximal oxygen consumption and workload achieved	Upper, lower or whole body	Depends on the protocol	Longer duration combat sports (i.e., multiple rounds)
Special Judo fitness test	Repeat-effort endurance and anaerobic capacity	Franchini et al., 2011	Yes	Index (calculated by heart rate and number of throws)	Whole body	Yes	Judo
Karate-specific aerobic test	Repeat-effort endurance and aerobic capacity	Chaabène et al., 2012	Yes	Time to exhaustion	Whole body	Yes	Karate
Repeat sled-push test	Repeat-effort endurance	Barley et al., 2018b	No	Average run speed, total test time and peak sprint test	Whole body	Yes	Mixed martial arts
Repeated sprint ability test	Repeat-effort endurance	James et al., 2018	No	Mean sprint time	Whole body	Yes	Mixed martial arts (effort-relief interval adjustments could make the test apply to other sports)
Taekwondo anaerobic test	Anaerobic power and capacity	Sant'Ana et al., 2014	Yes	Number of repetitions, test time and kick force	Whole body	Yes	Taekwondo
Specific jiu-jitsu anaerobic performance test	Repeat-effort endurance and anaerobic capacity	Villar et al., 2016	Yes	Number of repetitions	Whole body	Yes	Jiu-jitsu

is generally understood that utilizing sports-specific assessments is ideal (Müller et al., 2000), developing a sports-specific and scientifically valid assessment of endurance for all combat sport athletes is difficult. This is due to the high degree of variation in the physiological demands, sports-specific skills and competitive approaches both between and within combat sports (Franchini et al., 2011; Chaabène et al., 2012).

The variation between and within combat sports complicates the assessment of endurance in such athletes. As a result, researchers examining endurance capacity in combat sport athletes have utilized a range of assessment methods. These include circuits of activities conducted in a manner that reflect the required physical and physiological load of the specific sport, and in many cases, including sports-specific skills such as strikes and throws to mimic the performance aspects required (Smith et al., 2000; Hall and Lane, 2001; Franchini et al., 2005, 2007, 2011; Artioli et al., 2010; Chaabène et al., 2012; Villar et al., 2016; Sant'Ana et al., 2017) (**Table 1**). In locomotion sports such as cycling or running, the goal of any assessment is to evaluate such locomotion as this is the context in which competitive performance occurs. But in sports such as combat sports, where locomotion is not the sole objective, there are many goals and measures of possible performance. Specifically, the ability to continually attack and defend effectively against an opponent during later rounds is essential to victory (Miarka et al., 2015b). The ability to continue to execute sports-specific skills over longer periods of time despite fatigue has been assessed in judo (Franchini et al., 2011), karate (Chaabène et al., 2012), boxing (Smith et al., 2000), taekwondo (Sant'Ana et al., 2014) and Brazilian jiu-jitsu (Villar et al., 2016) (**Table 1**). These tests all involve the repeated execution of one or more sports-specific skill for an allocated time or until volitional fatigue, with varying measurements recorded. Simulation-style tests such as these provide valuable information on the fatigue induced by such sports-specific drills. There remain however many combat sports that do not have sports-specific performance tests available, and future research should aim to address this gap in available methodology. While it is important to consider that the potential limitations of such testing methods have not been completely explored, these include aspects related to the precision of *in situ* aerobic capacity measurement, lactate and ventilatory threshold identification and more general instances of measurement and repeatability of test performance. The special judo fitness test has undergone reliability testing as well as physiological examination (Franchini et al., 2009, 2011). Indeed, ventilatory gas analysis identified that $28.2 \pm 2.9\%$ of energy requirement were aerobic during this test (Franchini et al., 2011). An issue with this and many other performance based tests is that they are unable to assess important physiological characteristics such as thresholds, efficiency of motion or others likely to be relevant to performance. Regardless, typical tests of repeatability are derived from locomotion tasks, such as repeated-sprint ability, where the relationship between fatigue and changes in biomechanics is better understood (Morin et al., 2006). However, in combat sports the changes in technical ability resulting from fatigue and how important such changes are to competitive success are not clear. Given the highly technical nature of combat

sports it is plausible that fatigue-induced reductions in skill would have an even greater impact on competitive success than in locomotion based sports. Current sports-specific protocols do not comprehensively monitor impairments in skill which could result in missing important information that would plausibly have a substantial influence on performance during real competition. To better understand this future research should investigate the specific kinematic changes in combat sports techniques resulting from fatigue.

Repeat-effort ability is regarded as essential in a wide range of sports outside of those centered on combat. As a result there is a substantial body of literature that examines the repeat-effort ability of athletes (Bishop et al., 2001). While assessments of repeat-effort ability will have a similar basic structure, they can vary in a range of ways, including the duration of efforts, the recovery duration and number provided, and the modality of exercise (Bishop et al., 2001; Aziz et al., 2007). Previously reported testing protocols have involved repeated sprints on foot (Zagatto et al., 2009; James et al., 2018), on a cycle ergometer (Bishop et al., 2001), upper-body ergometer (Mendes et al., 2013), or pushing a sled (Barley et al., 2018b). Generic running tests such as the 30–15 intermittent fitness test are common examples of repeat-effort running tests commonly used in the field (Aziz et al., 2000; Buchheit, 2008; James et al., 2018), although the work-rest ratios are unlikely to be reflective of combat sports competitions. Many repeat-effort assessments however, only measure the high-intensity component (i.e., the sprint duration) while neglecting the low-intensity recovery portion, which potentially results in missing important information (Balsom et al., 1992; Spencer et al., 2005; Barley et al., 2018b; James et al., 2018). For example, when assessing repeated sprints on a cycle ergometer it is common for the tester to require the athlete to maintain at least 60 rpm during the recovery period. Yet it is very often unreported whether this protocol factor was adhered to and thus, although seeking to standardize recovery, the potential insight for the efficacy of an athlete's recovery process is lost.

Combat sports athletes require the ability for continual movement about the competitive arena for positioning an opponent and an ability for sustained upper-body isometric and dynamic contractions. While some of the repeat-effort data collected using common methods could elucidate this performance aspect, it is important to consider that sustained upper body isometric and dynamic contractions are not reflected easily in most repeat-effort testing as the methods used do not require significant strength, and therefore would not likely translate to combat sports. Previous research has tried to mitigate this issue with varying degrees of ecological validity by requiring athletes (judokas) to perform a series of unopposed throws of an opponent in their weight category between brief sprints (Franchini et al., 2011) or, alternatively, to push a weight sled (body mass relative) maximally for a specific distance (Barley et al., 2018b). However, these methods require further investigation to determine their applicability to their respective sports considering the aforementioned limitations of such protocols. Additionally, it is also important to consider if factors such as the level of competition or biological sex will influence the best practices for assessing physical

capacities in combat athletes, which should be a topic of future research.

ASSESSING ENDURANCE IN COMBAT SPORTS ATHLETES FROM A PHYSIOLOGICAL PERSPECTIVE

Repeat-effort ability is maintained by a complicated relationship between anaerobic and aerobic metabolism, with the anaerobic system being mostly important in high-intensity performance and the aerobic system being important to recovery between efforts (Bishop et al., 2011; Girard et al., 2011). The initial high-intensity effort will be heavily reliant on anaerobic metabolism with increasing aerobic contribution as more efforts are completed (Girard et al., 2011). However, even in the final efforts of repeat-sprint protocols the majority of energy can still be yielded anaerobically, though to a lesser extent (Girard et al., 2011). A study by McGawley and Bishop (McGawley and Bishop, 2015) observed significant aerobic contribution in the final sprint of a 5 × 6-s maximal sprint protocol. As such, the high-intensity components of repeat-effort sports will be significantly influenced by an athlete's anaerobic capacity even as the competition duration extends (Girard et al., 2011). Indeed, repeat-effort performance is likely to be heavily influenced by the accumulation of metabolic by-products, limitations in energy supply and neural fatigue (Girard et al., 2011). This is supported by studies that observe significant increases in blood metabolites during combat sports competitions (Bouhlef et al., 2006; Hanon et al., 2015) and the maximal cardiac response associated with combat sports competitions (Crisafulli et al., 2009; Hausen et al., 2017). As a result, common assessments of anaerobic capacity such as a Wingate or maximal accumulated oxygen deficit (MAOD) assessment will likely have important implications to combat sports endurance ability (Vandewalle et al., 1987; Faude et al., 2009; Bishop et al., 2011). The relationship between anaerobic capacity and competitive level in combat sports athletes has been observed in previous research (James et al., 2016b). However, the ability to recover from such high-intensity efforts during the low-intensity components will be driven primarily by the aerobic system to buffer hydrogen ion concentration and enhance phosphocreatine (PCr) regeneration (Balsom et al., 1992; Girard et al., 2011). This has been confirmed by previous research investigating the energy demands in taekwondo athletes during combat simulation (Campos et al., 2012). As such, greater aerobic fitness will likely improve repeat-effort ability in combat sports competitions by increasing oxygen availability, improving lactate removal and enhancing PCr regeneration (Tomlin and Wenger, 2001; Bishop et al., 2011). Increased aerobic fitness will also induce many physiological adaptations which could aid in combat sports endurance such as increased mitochondrial respiratory capacity, faster oxygen uptake kinetics, accelerated post-effort muscle re-oxygenation rate, improved lactate and ventilatory thresholds and a greater $\text{VO}_{2\text{max}}$ (Bishop et al., 2011). However, it is important to remember that the relationship between maximal aerobic capacity and combat sports competitive level, or even

repeat-effort performance in general does not appear to be linear (Bishop et al., 2011; Girard et al., 2011; Bridge et al., 2014; James et al., 2016b). We postulate that at the higher levels of combat sport competition there is a diminishing rate of return in the gross markers of aerobic capacity and adaptation. In fact, previous research has found sports-specific aerobic training in judo athletes to not improve $\text{VO}_{2\text{max}}$ but to significantly affect ventilation thresholds, heart rate and VO_2 recovery (Bonato et al., 2015). As such, other markers of aerobic fitness such as metabolic thresholds, economy, oxygen kinetics and the power output associated with $\text{VO}_{2\text{max}}$ could more closely relate with fatigue development during repeat-effort exercise as observed in combat sports (Faude et al., 2009; Bishop et al., 2011). Due to the aforementioned differences between intermittent and continuous exercise, the ecological validity would increase if practitioners were to evaluate such factors during intermittent exercise protocols (Drust et al., 2000; Koralsztein and Billat, 2000) particularly with world class athletes. Further research is required to better understand exactly which markers of both anaerobic and aerobic fitness best relate to combat sports endurance.

CONCLUSION

Combat sports are popular, physically demanding sports with a diverse competitive cohort around the globe. With a developing body of research, it is important to critically examine the current practices and how these may be best applied by the practitioner in the monitoring of athlete adaption process. Such a body evidence will not only help inform the assessment of endurance in combat sports athletes, but also the development of physical capacities which is a topic that needs further investigation. The current assessments of characteristics important for competitive combat sports performance, particularly those involved with endurance require further evaluation to determine their efficacy. This is important as many of the current methods may not be accurately assessing endurance ability or may lack the sensitivity to detect any changes. Endurance is a complicated characteristic comprised of many factors and as such cannot be comprehensively evaluated with a single test. A combination of assessments designed to simulate aspects of performance (in particular, repeat-effort ability) and others designed to better understand the underlying physiology will provide the most complete picture of combat sport endurance ability. However, while there is research investigating what physiological markers are important for combat sport athletes further research is needed to understand the relevant importance of variables such as metabolic thresholds and oxygen kinematics. When designing a test to simulate combat sport-specific endurance there are many things to consider, including how to induce a comparable physical and physiological load to competition alongside carefully choosing what activities will be included in the testing with respect to their ecological and scientific validity. Future research should investigate the potential impact that fatigue may have on combat sports-specific techniques and how such changes may influence assessments of endurance. Developing a better understanding the issues presented throughout this

review will improve researchers' ability to accurately assess characteristics relevant to combat sports performance, alongside allowing coaching staff to make appropriate training decisions and more effectively monitor the impact of such decisions.

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AUTHOR CONTRIBUTIONS

OB and SG conceptualized the review topic and design. All other authors contributed to the crafting and editing of the manuscript.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Age-Related Changes in Para and Wheelchair Racing Athlete's Performances

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OPEN ACCESS

Edited by:

Matthew J. Barnes,
Massey University, New Zealand

Reviewed by:

Mike James Price,
Coventry University, United Kingdom
Maha Sellami,
Qatar University, Qatar

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 11 October 2018

Accepted: 25 February 2019

Published: 19 March 2019

Citation:

Schipman J, Gallo P, Marc A, Antero J, Toussaint J-F, Sedeaud A and Marck A (2019) Age-Related Changes in Para and Wheelchair Racing Athlete's Performances. *Front. Physiol.* 10:256. doi: 10.3389/fphys.2019.00256

During the last decades, all para-athletes with disabilities have significantly increased their performance level due to technological progress and human investment, through better training or recovery protocols, medical care and nutritional monitoring. Among these elements, the athlete's age is one of the determining factors in performance. The aim of this study was to investigate the effect of age on maximal performances for para-athletes and wheelchair racing athletes, scaled on able-bodied records. We collected 53,554 results including athlete's best performance of the year, event, age and disability classification from the International Paralympic Committee competitions between 2009 and 2017 for both female and male para-athletics and wheelchair racing disciplines for a total of 472 sport events in Track and Field (considering each impairment type for each event) and gathered the all-time able-bodied records from the International Association of Athletics Federations (IAAF) at the end of 2017. Maximal performance by age was fitted with the Moore function for each para-athletics and wheelchair racing event. This study finds a similar age-related pattern in maximal performance among para-athletes and wheelchair racing athletes. The age at peak performance varies according to sex, impairment type and event and increases gradually from sprint to endurance events. The best Top 100 performances include a large age range suggesting that performance has probably not been optimized yet for most elite para-athletes and wheelchair racers. The next Paralympic Games of Tokyo 2020 and Paris 2024 will certainly offer exceptional performance.

Keywords: peak age, impairment and disability, track and field, paralympic games, classification

INTRODUCTION

There are a number of unanswered questions relating to the components of performance in Paralympic sport. For example, how age affects para-athletic performance for different types of impairment? Is there an age difference at peak performance between para-athletes and able-bodied athletes? At the age of 19 after his gold medal, the blade runner Jonnie Peacock said at the

Paralympic Games of London in 2012:” I lost my leg aged at 5... Now I’m 1.9 s behind Usain Bolt.” This comment questions the physical capacities of the para-athletes and the technological contribution of prostheses on the evolution of performance (Jones and Wilson, 2009; Weyand et al., 2009; Willick and Lexell, 2014; Dyer, 2015a; Baker, 2016). Since the first 1960 Paralympic Games, an increasing number of athletes with physical, visual or intellectual impairments have participated to elite para-athletic competitions (Dyer, 2015b; Fagher et al., 2016). Scientific and technological progress, such as prosthetic equipment, contributed to para-athletic promotion and the improvement of their performances (Lepers et al., 2012; Dyer, 2015b; Grobler et al., 2015). Indeed, beyond the impairment type, para-athletic performance is a complex process including both intrinsic parameters such as genetics, morphology (height and mass) or age and extrinsic factors such as environmental conditions (temperature, humidity, pollution), training methods, nutrition or technology (Berthelot et al., 2015; Blauwet et al., 2016).

In this framework, age is a major determinant of the performance in able-bodied athletes. Previous studies investigated the effect of age for different maximal physical performances (Moore, 1975; Baker and Tang, 2010; Guillaume et al., 2011; Berthelot et al., 2012; Allen and Hopkins, 2015; Marck et al., 2017b; Lepers et al., 2018; Marc et al., 2018) and showed a similar age-related pattern for maximal physical performances in Track and Field, swimming disciplines, or in tennis performance for high level athletes (Guillaume et al., 2011; Berthelot et al., 2012; Marck et al., 2017b). Maximal performance gradually increases with age until it reached a peak around 25–30 years according to the type of sport event. Then, it exponentially declines due to the aging process (Marck et al., 2017b). All the physiological systems involved in locomotion are intimately linked in the process of development and aging. During childhood and adolescence, there is an increase in muscle mass and strength. After maturity, skeletal muscle aging is characterized by a progressive loss of muscle mass. It is estimated that muscle mass losses are in the range of 0.02 to 3.3% per year (Mitchell et al., 2012; McGregor et al., 2014). These alterations of muscle with age remain intimately related to the degradation of the other systems with which it constantly interacts (Mitchell et al., 2012). This loss of mass can vary greatly from one muscle to another and the muscle mass loss in the legs may be higher compared to the arms (Lepers et al., 2012; Mitchell et al., 2012). This could impact the slope of performance decline that illustrates athlete’s impairment.

In para-athletics, no study has reported age related performance determinants. They only focused on technology (Weyand et al., 2009; Cooper and De Luigi, 2014), biomechanical analyses (Frossard, 2012; Beck et al., 2016), or incidence of injuries (Gawronski et al., 2013; McNamee et al., 2014; Fagher et al., 2016). Furthermore, the performance gap between able-bodied athletes and para-athletes, though well perceived in the daily life, has not been precisely quantified in all Track and Field events.

The present study characterizes the age-related changes in maximal performance and estimates the peak age for 47 para-athletics and wheelchair racing events, with a total of 472 event classes (that considers all impairment types) for both female and male para-athletes. These are scaled to the all-time able-bodied record performance. We also compare the patterns of performance decline for the para-athletes and wheelchair racing athletes and determine the classes contributing to the maximal performance of each event.

MATERIALS AND METHODS

Data Collection

Data (53,554) including athlete’s best performance of the year, event, age, and disability classification were collected for all International Paralympic Committee (IPC¹) competitions from 2009 to 2017 for 47 female and male para-athletes and wheelchair events (see details in **Table 1**) incorporating a total of 472 sport classes. Before 2009, data were unavailable online. These performances were achieved by 7,231 athletes: 3,500 male para-athletes, 1,348 female para-athletes, 1,650 male wheelchair athletes, and 733 female wheelchair athletes.

For the able-bodied maximal performance, the all-time world records by event at the end of 2017 were collected on the International Association of Athletics Federation’s website².

Study Design

For running events, racing times were converted to average speed in meters per second (ms^{-1}). All performances were analyzed according to age (in years), speed (in ms^{-1}) or meters (m) for throwing and jumping events and classes of disability. For males and females in all events, a unique maximal

¹www.paralympic.org

²www.iaaf.org

TABLE 1 | Number of performances and age indicators by event for para-athletes (PA) and wheelchair racing athletes (WCA) included in the database.

Event /Categories	Para-athletes male	Para-athletes female	Wheelchair athletes male	Wheelchair athletes female
100 m	3885	2115	1872	810
200 m	3271	1725	1690	723
400 m	2529	1003	1981	799
800 m	1299	224	1553	654
1 500 m	1458	320	1169	458
5 000 m	618	65	634	211
10 000 m	164	/	109	/
Marathon	319	50	601	174
Discus	2038	936	1969	935
Shot put	2256	1150	2086	1135
Javelin	1639	671	1550	856
Long jump	2110	1061	/	/
Triple jump	282	10	/	/
High jump	387	/	/	/

performance was selected among all individuals for each age. To compare the age-related pattern in performance between para-athletes and wheelchair racing athletes, two main categories were allocated: PA (Para-athletes) and WCA (Wheelchair athletes). Impairments types were regrouped according to IPC competition classification. For PA, these were; athletes with visual impairment (VI): T11-T12-T13-F11-F12-F13, athletes with cerebral palsy (CP): T35-T36-T37-T38-F35-F36-F37-F38, athletes with upper limb disabilities (UL): T45-T46-T47-F45-F46-F47, athletes with lower limb disabilities (LL): T42-T43-T44-F42-F43-F44 and athletes with intellectual disabilities (ID): T20-F20. In addition, for WCA, the classifications were: athletes with cerebral palsy in a wheelchair (CPW) T33-T34-F31-F32-F33-F34, athletes with tetraplegia disabilities (TD): T51-T52-F51-F52 and athletes with paraplegia disabilities (PD) T53-T54-F53-F54-F55-F56-F57-F58.

Characterization of the Age-Performance Relationship

To characterize age-related changes in maximal performance, the data were fitted with the Moore equation, which is a double exponential function (simple inverted U-shaped) initially developed on the athletes running speed-age relationship (Moore, 1975).

$$\text{Eqn1} : P(t) = a \left(1 - e^{-bt}\right) + c \left(1 - e^{-dt}\right) \text{ with } a, b, c, d > 0$$

$P(t)$ is the performance (t the time), a and c are scaling parameters, b and d are the characteristic times of the exponential growth and decline, respectively. For an estimated performance P , the model can be described as the sum of two von Bertalanffy's growth functions (VBGF): $P(t) = A(t) + B(t)$ where $A(t)$ is the increasing exponential process (first VBGF) and $B(t)$ the decreasing exponential process (the second VBGF is modified with $d > 0$). The Moore equation (Eqn 1) allows the estimation of the age (in years) at peak performance. These coefficients are determined using a least-square non-linear regression (Marck et al., 2017b). The quality of each fit was estimated by the coefficient of determination R^2 and the Root Mean Square Error (RMSE) (see **Supplementary Table S1**).

To quantify the gap between PA, WCA and able-bodied athletes, all datasets were scaled by maximal able-bodied performance of the event using the following formula:

$$\text{Eqn2} : \text{Scaled performance} = \frac{\text{performance}}{\text{maximalable} - \text{bodiedperformance of the event}}.$$

For each event, the exact peak age was computed and corresponded to the age when the performance was maximal. All performances [speed (m/s) or meter (m)] were reported in percentages (%) in order to compare the events.

Distribution of Age

In a complementary approach of the characterization of the age-related performance patterns, this study represented the age of the 100 best performances for PA and WCA by event using

the heat map function of Matlab software. This visualization enhances a better visibility of the peak and range of athlete distribution using a density scale [scale: 0 (low density) to 15 (high density)].

Distribution by Class for Best Performances

To assess the impact of each impairment type to the best performance, the number of sporting classifications within the top 100 best performances of each event were calculated as a percentage (%) for each sex.

All analyses were performed using the Matlab (MathWorks Inc.,) 2017b 9.3.0 software.

Ethics Statement

This study was designed and monitored by the IRMES (Institut de Recherche bio-Médicale et d'Epidémiologie du Sport) scientific committee. It used a research protocol qualified as non-interventional, in which "...all acts are performed in a normal manner, without any supplemental or unusual procedure of diagnosis or monitoring." (Article L1121-1 of the French Public Health Code).

RESULTS

Age at Peak Performance

In all Track and Field events, the age-performance relationship showed a similar pattern for both PA and WCA categories in female and male athletes. There was a gradual progression of the best performances up to a peak and thereafter performance progressively declined. The age of the estimated peak performance varied according to the event (**Figures 1–3**; details in **Supplementary Table S1**).

In male PA sprinting events, the age of the estimated peak ranged from 24.0 years in the 100 m to 20.8 years in the 400 m (**Figures 1, 2**). In endurance events, the estimated peak was 23.2 years for the 800 m and 33.0 years for the marathon (**Figure 1**). In throwing events, the age of the estimated peak varied from 24.1 years for javelin to 29.3 years for shot-put; in jumping events from 23.2 for triple jump to 26.0 years for high jump. In male WCA events, the estimated peak occurred later and ranged from 29.0 years in the 100 m to 35.6 years for the marathon (**Figures 1, 2**).

In female PA sprinting events, the estimated peak ranged from 24.8 years in the 100 m to 23.3 years in the 400 m (**Figures 1, 3**). In endurance events, no relation occurred between the age of the estimated peak performance and distance. In the marathon, the estimated peak was 18.1 years (**Figure 1**). In throwing events, the age of the estimated peak varied from 26.9 years for javelin to 33.4 years for shot-put; in jumping events from 22.8 for triple jump to 26.4 years for long jump.

Similar to male, the peak performance among female WCA ranged from 23.6 years in the 100 m to 31.9 years for the marathon (**Figures 1, 3**). In throwing events, the study showed a later peak compared to PA, with an estimated peak of 33.6 years for discus and 38.3 years for shot-put.

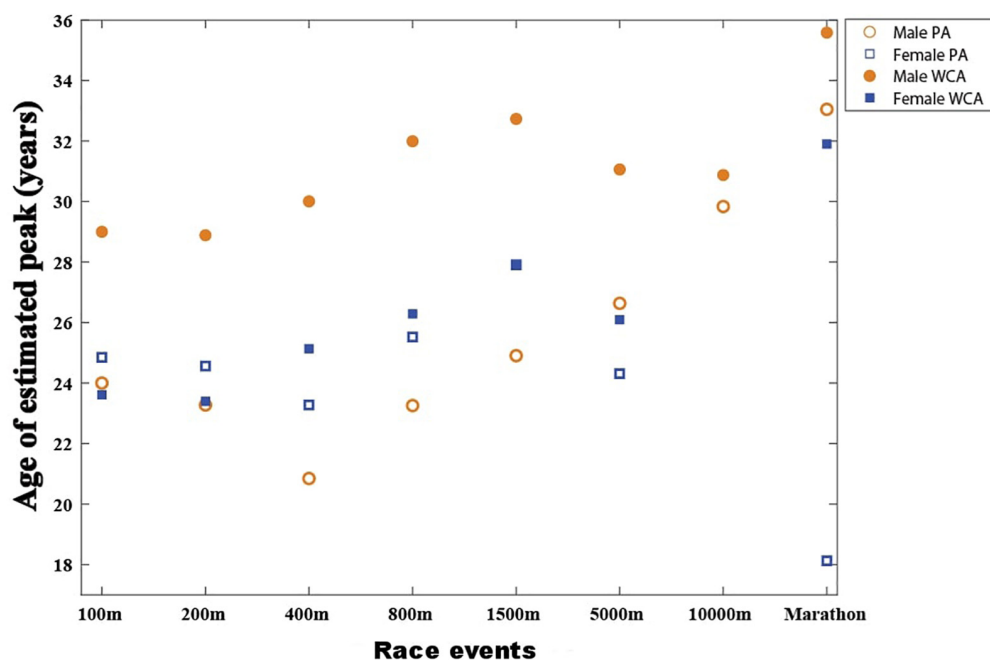


FIGURE 1 | Age of estimated peak performance with the Moore equation in race events for male para-athletes (PA: ○) and male wheelchair racing athletes (WCA: ●); for female para athletes (PA: □) and female wheelchair racing athletes (WCA: ■).

Performance Differences

In sprinting events, both female and male PA were faster than WCA. The difference decreased with distance and maximal performances were similar in the 400 m track event. For both sexes, maximal performances were lower than able-bodied best performances. In endurance events, PA were slower than WCA with the gap increasing as distance increased. In comparison to the best able-bodied athletes, WCA were faster (from +10.3% for female and +10.7% for male in the 800 m to +53.4% and +57.5% in the marathon) while PA were slower. In all throwing events, both female and male PA reached better performances than WCA, though all maximal performances were lower than able-bodied best performances (from −14% for male PA discus to −58% for female wheelchair javelin) (see details in Supplementary Table S1).

Age-Range for Optimal Performance

For both sexes, the age-range of the top 100 best performances was determined (Figures 4A,B). The findings showed a widespread age distribution from 20 to 60 years old and indicated that the density of performance was maximal around a peak age between 20 to 30 years old dependent upon event and sexes.

Class of Disabilities and Optimal Performance

All classes of disabilities were represented within best PA performances (Figures 5A,B). In WCA events for both sexes, the vast majority of the top 100 performances were achieved by athletes from the paraplegia impairments types.

Among female PA, athletes with VI were represented in the top 100 for all events. They represented 78% (400 m) to 85% (100 m) of top 100 best results in sprinting events and 34% for the shot-put events. Athletes with an ID were mostly represented in middle-distance events (31% for the 800 m; 52% for the 1500 m) and shot-put. Athletes with UL were mostly represented in sprinting events (13% in the 400 m; 18% in the 200 m), in javelin (39%) and long jump (27%). Athletes with a LL in long-jump (12%) and throwing events (10% in javelin; 18% in discus). Finally, CP were represented in throwing events (6% in discus; 17% in shot-put) and mostly absent from the other events.

The male distribution of the top 100 was similar to female distribution. CP athletes were represented by 5% in javelin and 35% in discus but mostly absent from the other events. Similarly, athletes with a LL were 22% in javelin and 54% in discus, 13% in long jump, 45% in high jump, 32% in the 200 m, and 18% in the 400 m. UL athletes were represented in all events (7% in discus; 55% in triple jump). Athletes with an ID were mostly represented in middle- and long-distance running events (7% in the 10,000 m; 25% in the 1500m), in shot-put (23%), long jump (25%) and triple jump (9%). Male VI athletes were represented in the top 100 of all events (up to 71% in the 10,000 m).

DISCUSSION

The Age at Maximal Peak Performance

Age-related changes in maximal performances for both PA and WCA revealed a similar pattern. The age at peak performance varied depending on sex, impairment classification and event.

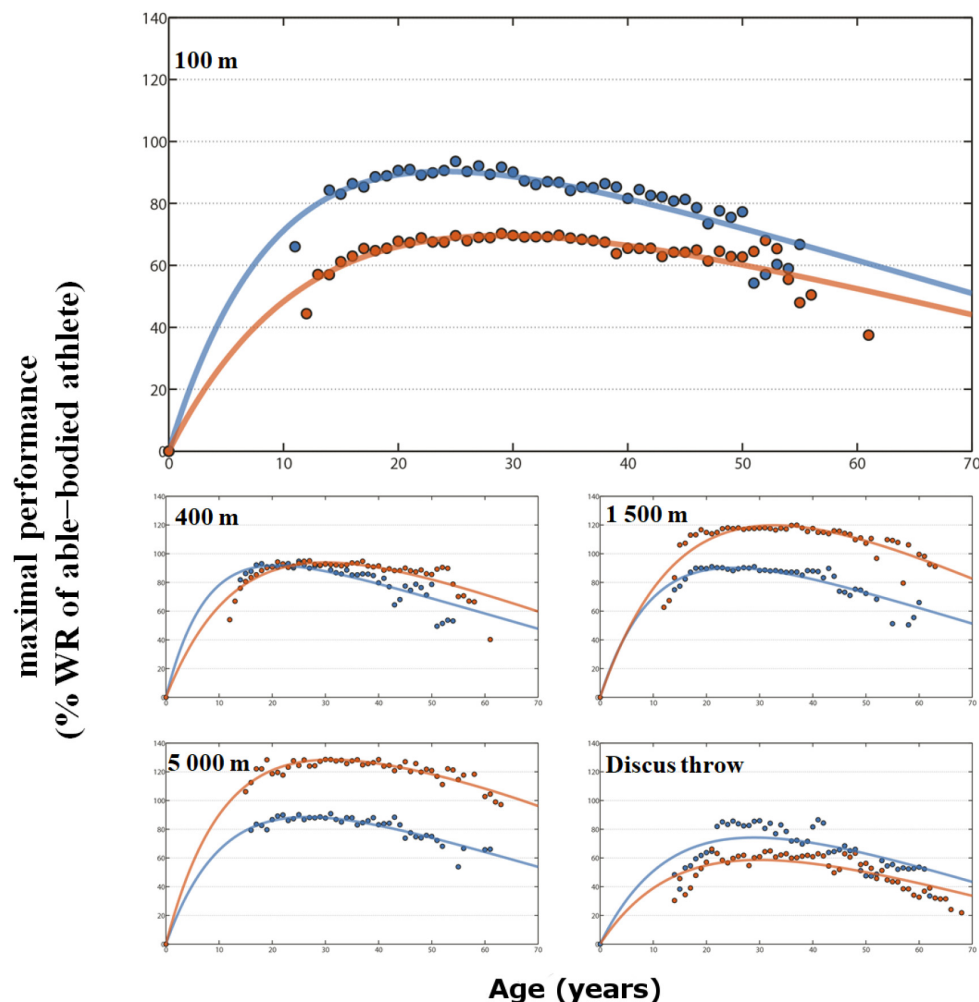


FIGURE 2 | Envelope curve in % of maximal able-bodied performance for male para-athletes (blue line) and wheelchair racing athletes (red line) by age on 100 m, 400 m, 1500 m, 5000 m and discus throw. Peak age and R^2 in **Supplementary Table S1**.

Such an age difference may depend on the “mosaic” of aging processes that do not homogeneously alter the organism but that similarly impacts each type of impairment.

The collected data on 53,554 PA and WCA performances showed a similar pattern of progression in the performance-age relationship for all events, with an initial gradual increase in performance until reaching the peak, then a gradual decrease in maximum performance with age. The maximal PA and WCA performances fitted with the Moore equation showed that the respective determination coefficients (R^2) of this equation were well adjusted to the maximum age-related performances (**Figures 2, 3**). Following this similar age-related performance pattern illustrated in many sport events and different species (Guillaume et al., 2011; Berthelot et al., 2012; Marck et al., 2017b; Marc et al., 2018), the para-athlete age-related performances confirmed the reliability and the robustness of the Moore equation (Eqn 1).

In male sprinting events (**Figures 1, 2**), PA peak age was similar to able-bodied athletes and increases in endurance events

(Moore, 1975; Berthelot et al., 2012; Marck et al., 2017b; Marc et al., 2018). It is well known that the aging process involves physiological and psychological changes, whether structural or functional (Mitchell et al., 2012; McGregor et al., 2014). The number of muscle cells and of motor units decreases sharply with age (Faulkner et al., 2007). However, fast type II fibers (favored in sprint races) are more prematurely altered than slow type I fibers (favored in endurance racing). The increase in peak performance with the event duration and distance could therefore be an explanation for PA male results. For female PA (**Figures 1, 3**), the appearance of a plateau and a young age for marathon peak may be mainly related to a low number of participants (50 archived performances only, vs. 319 in male PA marathon or 2115 in women PA 100 m).

Furthermore, at the individual scale, the origin and development of the disability, either born with it or acquired later in life, is also an element in the age at peak performance or the appearance of not clear peak, which needs to be

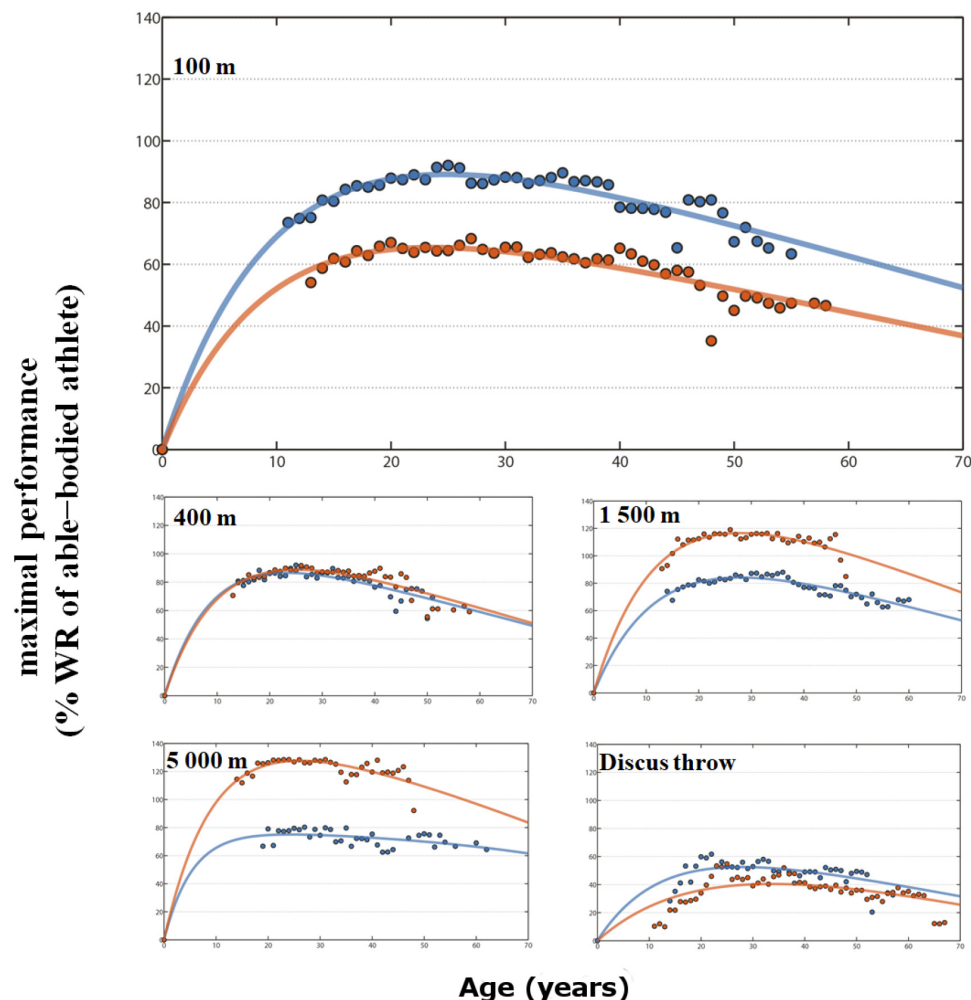


FIGURE 3 | Envelope curve in % of maximal able-bodied performance for female para-athletes (blue line) and wheelchair racing athletes (red line) by age on 100m, 400m, 1500m, 5000m and discus throw. Peak age and R^2 in **Supplementary Table S1**.

investigated. Indeed, PA seem to start their career younger due to the innate origin of the disability (Ravensbergen et al., 2018), while WCA begin their career and peak at a later age because of an impairment acquired during the adolescence or the young adult age (Ravensbergen et al., 2018). Depending upon the disability or the technical equipment, an adaptation period, in order to acquire an optimized techno-physiological interaction, may contribute to an older age at peak performance.

In some events, WCA age-related performance curves did not show a discernible peak performance. Power of the upper arms, muscle strength of the elbow extensors, muscle endurance of brachial triceps (Mitchell et al., 2012) and push angle (Lepers et al., 2014) may contribute to this observation. Such an element certainly increases the inter-individual variability for the age at peak performance.

The Top 100 performances included a large age range from 15 to 55 years (**Figure 4**) revealing that performance is not yet optimized for most of the elite PA and WCA. For the best male

and female able-bodied athletes, the age range varies from 21 to 36 years for the 100 m to 20 to 38 years for the marathon (Marc et al., 2018), which indicates a tighter age-performance range. These results could be explained by the fact that able-bodied athletes who have competed since the first Olympic Games in 1896 represent an accumulated pool of 30,535 able-bodied athletes over 100 years including the 2016 Rio Olympic Games by comparison, the first Paralympic Games held in 1960 only giving 56 years of data on 12,752 para-athletes. Therefore, these lower numbers of participants can also be attributed to disability-induced barriers to mobility. Para-athletes may experience difficult interactions with their social and physical environments which leads to limitations in their activities of daily life, and restrictions in competition participation. Depending on the degree of functional loss, the para-athletes are impacted differently by contextual, environmental and personal factors (Fellinghauer et al., 2012). In addition, such a large age-band might be typical of para-athletes and reflect the performance of those born without impairment who often are

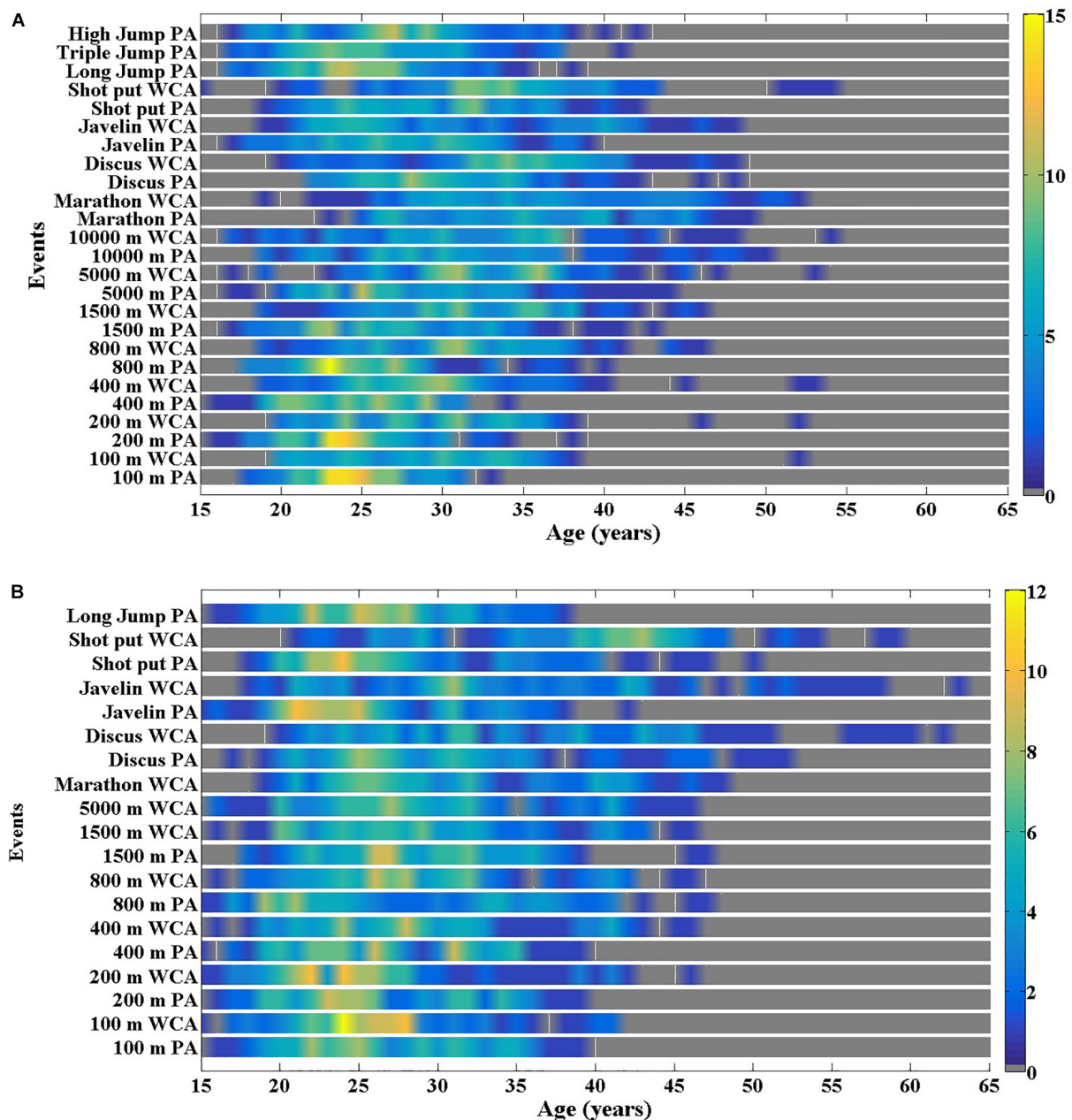


FIGURE 4 | Density distribution of the 100 best performances by age for male (A) and female (B) for para-athletes (PA) and wheelchair racing athletes (WCA).

athletes with accidental or progressive impairments appearing during their life.

For the best performances, only a few classes contributed to the maximal performance (Figure 5). For WCA, in both male and female events, the fastest athletes were in the PD class. Among paraplegic people, muscle strength in the upper extremities and respiratory function are comparable to that of the able-bodied population (Haisma et al., 2006a). In tetraplegic people, muscle strength varies greatly and respiratory function is considerably reduced relative to the values in an able-bodied population (Haisma et al., 2006b). This provides support for

the lack of representation from this group in the 100 best performances achieved by the WCA. Among PA, the distribution seemed more heterogeneous. Overall, from 100 to 400 m, only male PA with a VI, LL and UL represented the best performances, whereas the female PA were predominantly in the VI category. Females with LL were less represented in sprint events. This could be explained by the fact that the mechanical properties of carbon prostheses may not be adapted yet to female specificities (such as developed strength or anthropometric factors – height or BMI (Sedeaud et al., 2014). With a constant stiffness, the lower strength produced by female PA could

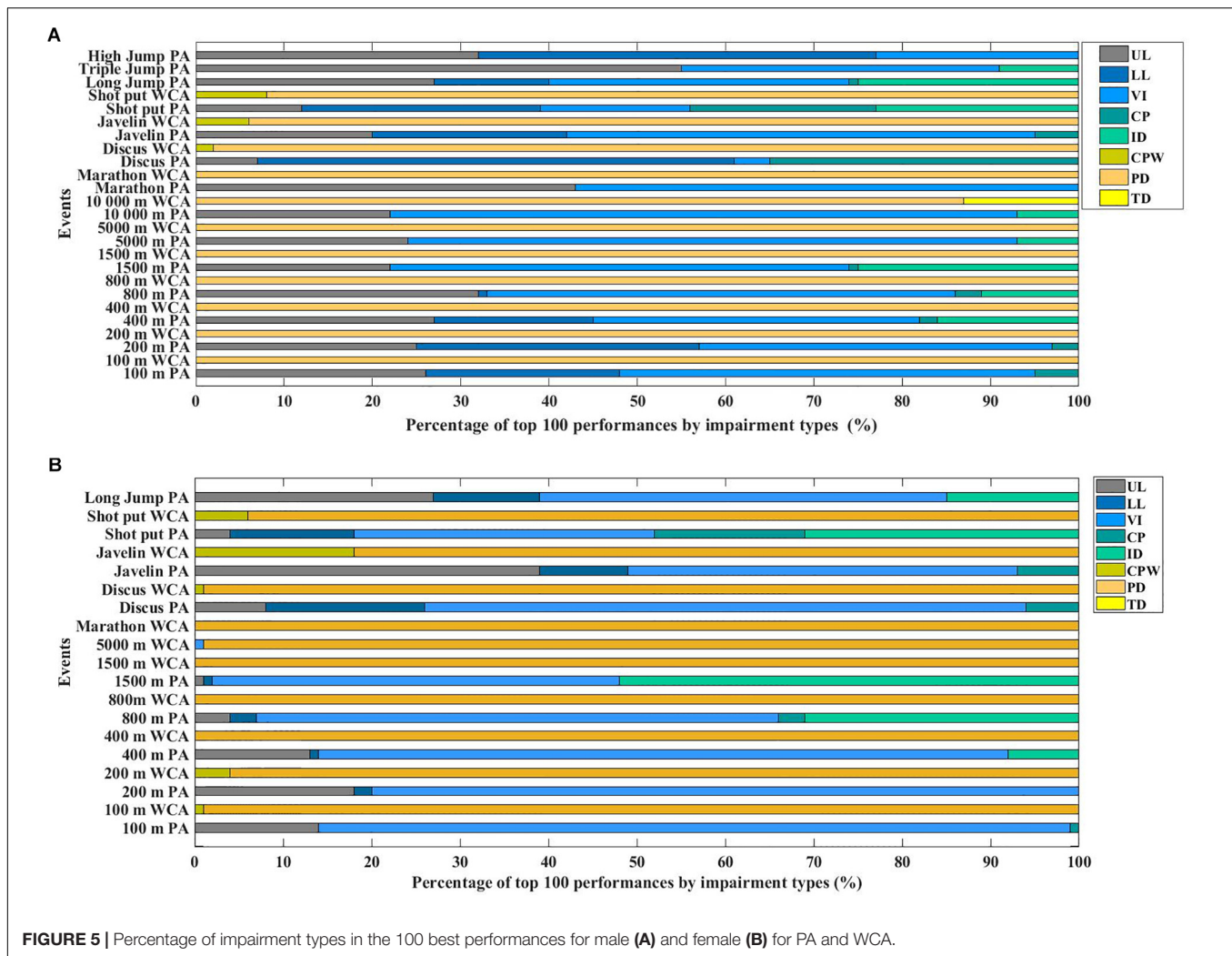


FIGURE 5 | Percentage of impairment types in the 100 best performances for male (A) and female (B) for PA and WCA.

impact the prosthesis reaction, which depends on the magnitude of the applied force (Beck et al., 2016) thereby reducing the generated speed.

The Performance Levels

Large differences remain between PA, WCA and able-bodied athlete's performances, related to biomechanical properties, difference in equipment and sample size. In male and female sprint events, the best PA performances were 5 to 9% under the able-bodied world record. When race distance increases, so does this gap with a larger difference in females.

The PA's maximal performances have come closer to the able-bodied world records but do not surpass them (Grobler et al., 2015). Nevertheless, maximal performances of able-bodied athletes have been plateauing for three decades and now seem to have reached their upper limits (Marck et al., 2017a). Similarly, the rate of progression of WCA seems to have considerably slowed down, at least through the observation of world records or the best performances such as in the Oita marathon (Lepers et al., 2012). WCA gap gradually increased with the race distance. The best WCA performances from

800 m to the marathon were, respectively, 10 to 57% over the able-bodied world record. These increases demonstrate that WCA in endurance events are comparable to hand cycling events where technological and strategic contributions are different compared to able-bodied athletes (Lepers et al., 2014). Indeed, WCA are able to coast for recovery or energy conservation (Cooper and De Luigi, 2014), whereas able-bodied runners must keep expending precious energy even during downhill sections.

Technological advances will undoubtedly increase the performance levels and at the same time improve the quality of the wheelchair or prosthesis use in the daily life. However, the environment and economic situation could be less favorable to the improvement of such innovations and may even play a crucial role in the regression of maximal performances in both able-bodied and disabled athletes due to travel, physical and financial difficulties. In this context, it would be important to continue to develop policies that increase and promote physical activity and sport for the beneficial effects on health, such as a decreased risk of chronic diseases and an improved quality of life (Global Recommendations on Physical Activity for Health, 2010),

particularly for people with physical disabilities (de Hollander and Proper, 2018).

CONCLUSION

Para-athletes and wheelchair athletes display an age-related pattern in maximal performances, similar to able-bodied athletes. The age at peak performance increases gradually from sprinting to endurance events for para-athletes. The Top 100 best performances include a large age range suggesting that performance has probably not yet been optimized for most elite para-athletes and wheelchair racers. The Paralympic Games of Tokyo 2020 and Paris 2024 will certainly offer exceptional performances which can still be improved upon for most of the elite wheelchair racing and para-athletes. Further studies will contribute to increasing knowledge about age-related changes and the origin of the impairment in para-athletes and wheelchair racing athletes.

DATA AVAILABILITY

Publicly available datasets were analyzed in this study. This data can be found here: www.paralympic.org.

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AUTHOR CONTRIBUTIONS

JS, PG, AnM, J-FT, AS, and AdM designed the research. JS, PG, AnM, AS, and AdM performed and analyzed the research. JS, PG, AnM, JA, J-FT, AS, and AdM wrote the manuscript. All authors read and approved the final manuscript.

ACKNOWLEDGMENTS

The authors thank Stacey Johnson for proofreading the manuscript, the INSEP teams and Arnaud Litou for their full support, Sandra Mauduit, Julien Héricourt, and Oliver Deniaud from the French disabled sports federation (FFH), the French Paralympic and Sports Committee (CPSF). In addition, the authors thank all the curators of the websites.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fphys.2019.00256/full#supplementary-material>

TABLE S1 | Coefficients values, fitting indicators and age at the peak performance for all track and field events.

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- Conflict of Interest Statement:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.
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Mechanical Work and Physiological Responses to Simulated Flat Water Slalom Kayaking

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OPEN ACCESS

Edited by:

Glen Davison,
University of Kent, United Kingdom

Reviewed by:

Firat Akca,
Ankara University, Turkey
Andy Galbraith,
University of East London,
United Kingdom

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 20 November 2018

Accepted: 28 February 2019

Published: 20 March 2019

Citation:

Macdermid PW, Osborne A and
Stannard SR (2019) Mechanical Work
and Physiological Responses
to Simulated Flat Water Slalom
Kayaking. *Front. Physiol.* 10:260.
doi: 10.3389/fphys.2019.00260

The purpose of this study was to assess the physical work demand in relation to metrics of force and subsequent physiological response to a simulated flatwater slalom competition. Eight New Zealand team members completed a standard incremental step-test to ascertain power: oxygen consumption relationship. This was followed by a simulated race run where breath-by-breath analysis along with force and power data logged at 50 Hz to determine stroke length, impulse, peak force, time to peak force, and rate of peak force per stroke. Physiological response to negotiating a flatwater slalom course was greater than straight-line paddling (36.89 ± 2.01 vs. 32.17 ± 1.97 ml·kg⁻¹·min⁻¹, $p = 0.0065$) at the same power output. Mean power output for the duration of the simulated race (91.63 ± 7.19 s) was 203.8 ± 45.0 W, incurring an oxygen deficit of 1.386 ± 0.541 L·min⁻¹ translating to an overall anaerobic contribution of $32 \pm 18\%$ and aerobic contribution of $68 \pm 18\%$. Moderate to strong relationships between time duration and stroke peak force ($R^2 = 0.354$, $R^2 = 0.485$) and rate of peak force development ($R^2 = 0.345$, $R^2 = 0.426$) but not for stroke length ($R^2 = 0.022$, $R^2 = 0.012$), impulse ($R^2 = 0.088$, $R^2 = 0.097$) or time to peak force ($R^2 = 0.001$, $R^2 = 0.0001$) for left and right strokes, respectively. The number of propulsive (<0.6 s) strokes outweighed turning/driving (>0.6 s) strokes with a ratio of 94:6%. Longer stroke duration was significantly correlated to greater impulse ($R^2 = 0.507$, $p < 0.0001$) and time to peak force ($R^2 = 0.851$, $p < 0.0001$), but a lower rate of force development ($R^2 = 0.107$, $p < 0.0001$). The results show that a flatwater slalom under simulated race conditions entails initial supra-maximal (anaerobic) work rate with a subsequent transition to one associated with maximal aerobic capacity. Inability to sustain work done and the subsequent decline in peak force and force profile per stroke requires further research regarding strategies to enhance performance.

Keywords: canoeing and kayaking, intermittent, field testing and monitoring, oxygen deficit, Slalom canoe racing

INTRODUCTION

International Canoe Slalom races are defined by the rules of the International Canoe Federation (ICF), which in turn determines the technical demands and physical environment of competition, and thus the cognitive and physiological stresses on athletes taking part.

Currently, the rules require athletes to race over a predetermined course, changing from race to race, on a section of river involving natural and artificial obstacles, where the length ranges from

200 to 400 m down the center line of the river. Courses involve sequences of two suspended poles referred to as gates (width per gate of 1.2–4.0 m). Each course consists of 18–25 gates, of which six must be negotiated in an upstream direction, and a finish sprint of 15–25 m. Typically, a competent paddler will take 90-s (range: 75–95-s) to complete the course with the fastest total time winning [time penalties are added for hitting (2-s) and missing (50-s) per gate].

Presently, published physiological or physical descriptions of slalom during or following race or simulated race runs are limited (Baker, 1982; Zamparo et al., 2006). These observations include blood lactate values of members of the British team during the 1981 world championships (Baker, 1982). These being 16.2 ± 1.2 , 13.1 ± 2.1 and 12.2 ± 1.8 mM for K1M, C1M and K1W categories respectively, taken 5 min following competition. Later studies (Zamparo et al., 2006; Manchado-Gobatto et al., 2014; Messias et al., 2015a,b) report lower post-competition lactate values [mean 7.98 ± 1.6 mM, range: 5.6–10.3 mM in Italian national team members, samples 5 min post-competition (Zamparo et al., 2006)], and suggest a lower intensity, a less capable cohort in the latter study, or a change in emphasis/strategy for success in the sport between studies. Changes in technical regulations may explain this, although there is no information available regarding the work demand or stroke kinetics during competition to corroborate such statements. However, such data is available for flatwater straight-line kayaking, where elite male paddlers typical propulsive stroke have durations of ~ 0.44 s, generate peak forces of ~ 375 N and an impulse of 109 N·s (Baker, 1998; Sperlich and Baker, 2002). Information of this nature taken from slalom paddlers during racing or simulated racing would enhance understanding of the sport whilst being invaluable in the monitoring of athlete development.

Observations of cardiovascular stress during slalom canoeing, peak and mean heart rates are reported to be 184 ± 8 and 173 ± 14 bpm during race simulation (Messias et al., 2015b) and as such, the work rate performed in a slalom race has been considered moderate to vigorous in intensity (Sigmund et al., 2016) with aerobic-anaerobic contribution reported as 24.9% for alactic, 29.9% anaerobic lactic, and 45.2% aerobic (Zamparo et al., 2006).

To our knowledge, there is only one published empirical study indicating metabolic cost during real or simulated competition (Zamparo et al., 2006) who compared all-out flatwater effort with a simulated slalom competition. This work used breath-by-breath gas exchanges and post-exercise blood lactates to estimate aerobic and anaerobic components using a three term model to calculate total metabolic energy (Wilkie, 1981). Results highlighted similarity in aerobic and anaerobic contribution (50–50%, respectively) between conditions even though total energy expenditure was about 30% greater during the all-out flatwater test. Despite the acknowledgment of the importance of power-meters (Sperlich and Klauck, 1992; Messias et al., 2018) and the recent validation of devices capable of recording power output during kayaking (Macdermid and Fink, 2017), no published description of the physical work demand produced by paddlers during course negotiation has occurred.

Thus, the primary aim of this study was to assess the physical work demand and subsequent physiological response to a simulated flatwater slalom competition. Additional aims included a description of stroke kinetics in relation to competition timecourse and stroke length, and a comparison of straight-line and slalom paddling. We hypothesize that: straight-line paddling at the same work rate will trigger smaller physiological response and thus appear more efficient. The simulated race energy distribution will involve maximal demand from both anaerobic during the early stages and aerobic during the latter half but intermittent periods of anaerobic respiration will occur. The ability to generate force and thus work done will decline with time but is dependent on the aim of the stroke. As such, turning strokes will involve greater application of force over prolonged periods resulting in high impulse but low power outputs while propulsive strokes will have low impulse and high power outputs.

MATERIALS AND METHODS

Participants

Eight competitive slalom kayakers who formed part of the New Zealand Slalom development team (mean \pm SD, height: 173 ± 4 cm, mass: 65.8 ± 6.0 kg, $\dot{V}O_{2max}$ 46.7 ± 4.9 ml·kg⁻¹·min⁻¹) were recruited to participate in this study. Prior to their involvement, all participants provided written consent in accordance with the requirements of the University Human Ethics Committee and the Declaration of Helsinki.

Testing

On arrival at the venue (Centennial Lagoon, Palmerston North, New Zealand) all participants were weighed with and without kayaking clothing and measured (cm). Participants were instructed on the course of gates to be negotiated (**Figures 1A,B**) in order to mentally plan their route as is typical within competition and training (MacIntyre and Moran, 2007). Once they had deemed sufficient knowledge of the sequence of gates they were fitted with a portable gas analyzer (K42b, COSMED, Rome, Italy).

While participants used their own kayaks, all used the same kayak paddles which included a straight carbon fiber power meter kayak paddle shaft (One Giant Leap, Gisborne, New Zealand) with Naja Maxi elite blades attached (GalaSport, Hrádek, Czechia). Inexperience of some participants ($n = 3$) in using the blades chosen was considered, however all participants were comfortable by the time of any testing. Blade angles in the shaft were adjusted to that preferred and normally used by the participant under normal training and racing conditions.

Following a 5 min warm-up period consisting of straight-line paddling, interspersed with participants preference for turning manoeuvres, performed at an intensity ≤ 50 W and acting as a re-familiarization with the test equipment. Participants performed a sub-maximal incremental step-test over a predetermined course (**Figure 1A**) in order to obtain the Power: $\dot{V}O_2$ relationship (Medbo et al., 1988). Data taken from a 5 Hz GPS unit

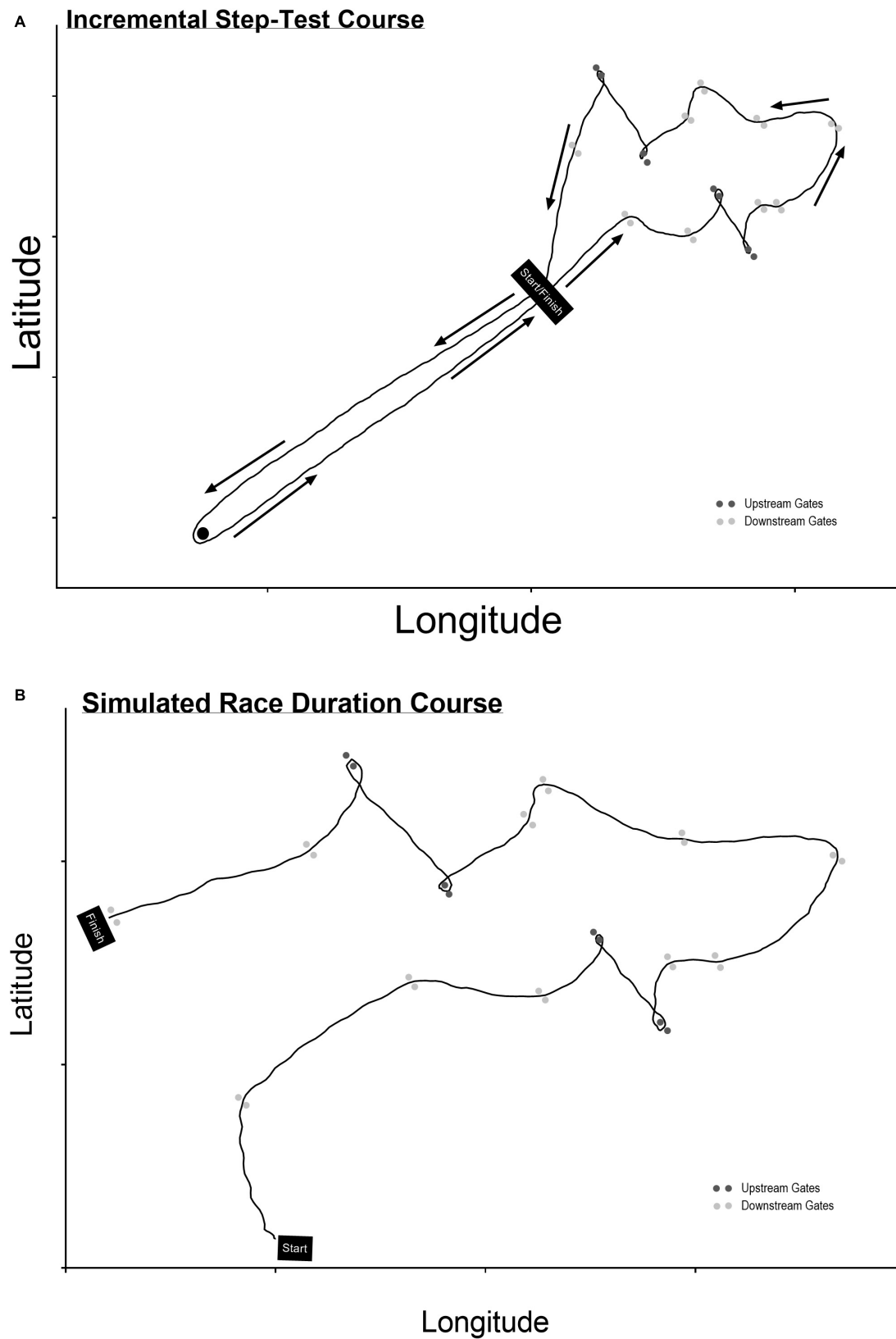


FIGURE 1 | The courses for the **(A)** incremental step-test and **(B)** race duration test. The line is taken from a participant's high-speed GPS unit.

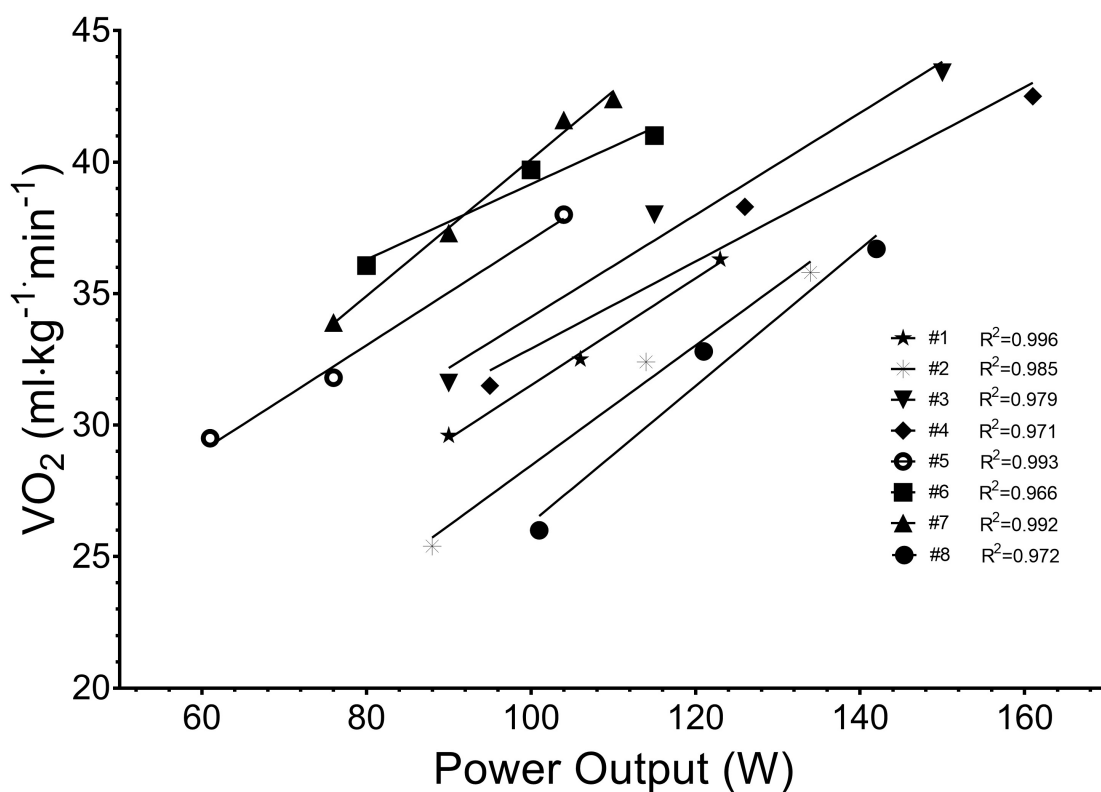


FIGURE 2 | Submaximal field test data for each participant used to determine paddler economy and power: $\dot{V}O_2$ relationship.

(GPSports, Ltd., Canberra, ACT, Australia) device showed the course length to be 325.3 ± 2.0 m ($n = 24$), while data from the Garmin 920 XT sports watch used to log power data and heart rate provided a length of 291.5 ± 3.8 m ($n = 24$). The course included four upstream gates, 11 downstream gates, inclusive of eight major turning manoeuvres. This test consisted of three incremental bouts of exercise, ~ 3 min duration at a pre-determined power output, based on fitness, and displayed as average lap power on a portable device (Garmin 920XT) at the front of the kayak cockpit to ensure correct participant work rate. Power output data was logged (1 Hz) throughout the trial and averaged along with $\dot{V}O_2$ data (sampled breath-by-breath) over

the final 2 min of each stage (Medbo et al., 1988). This enabled an individual power: $\dot{V}O_2$ relationship to be formed whilst kayaking and including manoeuvres typical in slalom, and where data could be extrapolated to predict O_2 demand (Medbo et al., 1988; Gastin, 2001) during the competition simulation test described below.

Supplementary material – time line video of the protocol used
doi: 10.6084/m9.figshare.7376783.

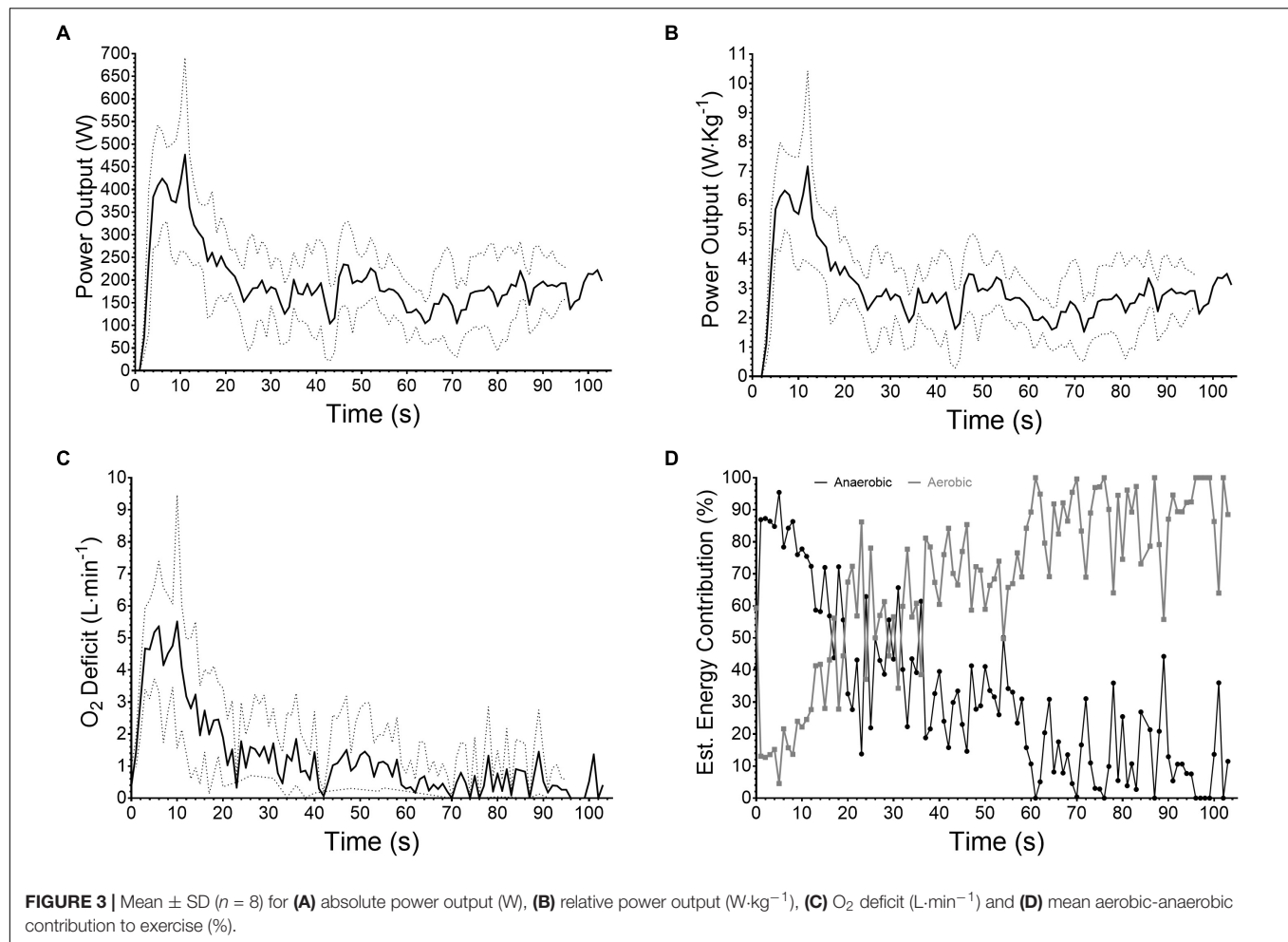
After a 15 min active break which involved 10–12 min of flatwater paddling at an overall intensity deemed light but inclusive of non-monitored short duration high-intensity efforts, followed by a period of rest as per competition race procedures, participants performed one simulated race run (Figure 1B). Throughout, participants wore the automated, portable gas analyzer (breath-by-breath analysis) and used the kayak power meter (data logging 50 Hz). The sequence of gates used was the same as the incremental test previously described (Figure 1A), but differing in the start and finish position. Data taken from 5 Hz GPS unit (GPSports, Ltd., Canberra, ACT, Australia) device showed the course length to be 188.4 ± 2.2 m ($n = 8$).

Participants were instructed to approach the start as per a normal slalom race where athletes work at supra-maximal intensities whilst accelerating to their preferred speed (Zamparo et al., 2006).

The kayak power meter logs force and power data for both left and right shafts separately at 50 Hz. This can be transferred to a standard personal computer as a csv. file and processed using

TABLE 1 | Mean \pm SD and range for data taken from the submaximal (1–2)-maximal (3–9) component of testing.

	Mean (SD)	Range
(1) $\dot{V}O_{2@90W}$ (ml·kg ⁻¹ ·min ⁻¹)	31.63 (5.09)	23.67–37.73
(2) Economy@90W (W·L ⁻¹ ·min ⁻¹)	45.23 (8.62)	38.5–64.4
(3) Peak power (W)	606 (169)	418–824
(4) Peak power (W·kg ⁻¹)	8.78 (2.26)	6.26–12.19
(5) W_{max} (W)	203.2 (33.2)	151–303
(6) W_{max} (W·kg ⁻¹)	3.07 (0.69)	2.38–4.32
(7) $\dot{V}O_{2max}$ (L·min ⁻¹)	3.085 (0.328)	2.699–3.559
(8) $\dot{V}O_{2max}$ (ml·kg ⁻¹ ·min ⁻¹)	46.68 (4.85)	39.3–51.8
(9) Maximum heart rate (bpm)	178 (18)	149–197



MATLAB R2016a. On a stroke by stroke basis data were analyzed (Macdermid and Fink, 2017) for: (A) Stroke length (s), defined as the time taken from data onset (when the drive side blade triggers a force threshold > 2 N and is a > 10 number points from the end of the previous stroke) to data offset (when the drive side blade triggers a force threshold < 2 N); (B) Impulse ($\text{N}\cdot\text{s}$), the area under the force curve per stroke; (C) Peak force (N), the maximum force reached during each stroke; (D) Time (s) to peak force, the time from stroke onset to the time of maximum force. Subsequently, the data for the left and right shafts were combined and analyzed for total power output at a sample rate of 50 Hz and averaged over 1 s epochs to coincide with gas analysis data. Additional analysis separated data based on forward propulsive paddling or a turning stroke.

Physiological variables continuously sampled throughout the test included, breath-by-breath expired air in order to calculate $\dot{V}\text{O}_2$ ($\text{L}\cdot\text{min}^{-1}$) averaged every second. The $\dot{V}\text{O}_2$ and power output data, used in conjunction with the linear relationship data established via the incremental test was used to estimate O_2 deficit and subsequently energy system contribution to exercise based on the following calculations:

$$\text{O}_2 \text{ demand} = mx + b$$

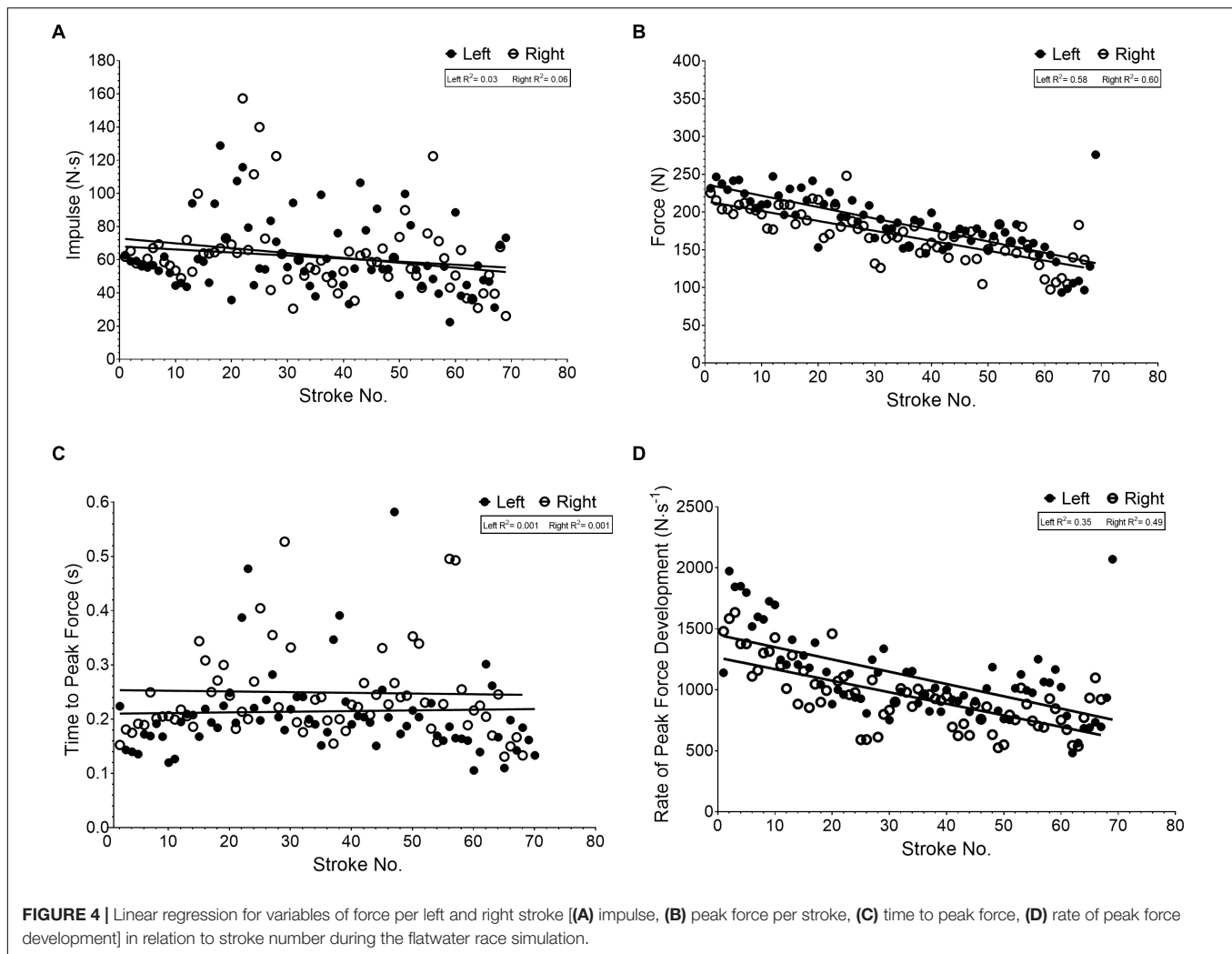
where, m is the slope and b is y-intercept of the power: $\dot{V}\text{O}_2$ relationship determined in the incremental test, and x is the 1st smoothed average of power output measured during the field test.

$$\text{O}_2 \text{ deficit} = \text{O}_2 \text{ demand} - \text{Actual } \dot{V}\text{O}_2.$$

Statistical Analyses

Data recorded throughout the trials were transmitted to a conventional PC and processed with the Garmin Training Centre, One Giant Leap analysis application¹ and Cosmed Omnia software provided with the relevant hardware. Descriptive data [mean, standard deviation (SD)] were calculated for all dependant variables. Strokes were categorized as forward (propulsive) or turning via frequency distribution based of stroke length duration. All comparison used a paired t -test (GraphPad Prism, Version 7.0) while relationships were determined using linear regression statistics. Significance was set at $p < 0.05$ while classification regarding strength of relationship used Cohen system (Cohen, 1988).

¹http://analysis.onegiantleap.co.nz/#/?_k=3rhtaa



RESULTS

Participant characteristics obtained from the submaximal and maximal components of the testing are presented in **Table 1**. The submaximal tests were performed at work rates specific to participants' state of fitness (**Figure 2**), and included a straight-line and slalom course component (**Figure 1**) typical of what occurs during this type of training. Significant difference for $\dot{V}O_2$ (32.17 ± 1.97 vs. 36.89 ± 2.01 ml·kg⁻¹·min⁻¹, $p = 0.0065$) but not mean power (107.2 ± 16.4 vs. 106.4 ± 15.9 W, $p = 0.171$) were shown when comparing straight-line paddling with the slalom course components respectively.

Mean \pm SD time for the simulated race equaled 91.63 ± 7.19 s and this required a mean power output of 203.8 ± 45.0 W or 3.07 ± 0.63 W·kg⁻¹. Stroke rate was 85.6 ± 58.3 spm of which 94% were forward and 6% turning strokes. Physiological variables associated with these work characteristics for the flatwater slalom equaled to rate of oxygen consumption of 2.629 ± 0.498 L·min⁻¹ (mean \pm SD), oxygen deficit 1.386 ± 0.541 L·min⁻¹, aerobic contribution $68 \pm 18\%$, anaerobic contribution $32 \pm 18\%$, and HR 170 ± 2 bpm. Absolute power output, relative power

output, O₂ deficit and the reliance of anaerobic metabolism decreased over time (**Figures 3A–C**), while aerobic metabolisms contribution increased (**Figure 3D**).

Separate analysis for left and right stroke data presented as mean \pm SD for stroke length (0.499 ± 0.184 vs. 0.462 ± 0.213 s, $p = 0.184$), impulse (61.6 ± 21.7 vs. 62.6 ± 23.6 N·s, $p = 0.785$), peak force (183.9 ± 39.8 vs. 169.9 ± 33.2 N, $p = 0.0002$), time to peak force (0.215 ± 0.098 vs. 0.249 ± 0.0114 s, $p = 0.079$), and rate of peak force development (1098 ± 339 vs. 942 ± 265 N·s⁻¹, $p < 0.0001$) provides descriptive kinetics. Linear regression of this data (**Figures 4A–D**) identified significant negative correlations between stroke number (left and right) and peak force ($R^2 = 0.354$, $p < 0.0001$ and $R^2 = 0.485$, $p < 0.0001$, **Figure 4B**) and rate of peak force development ($R^2 = 0.3449$, $p < 0.001$ and $R^2 = 0.4257$, $p < 0.001$, **Figure 4D**), a weak relationship between impulse ($R^2 = 0.088$, $p = 0.014$ and $R^2 = 0.097$, $p = 0.009$, **Figure 4A**), and no relationships for time to peak force ($R^2 = 0.001$, $p = 0.837$ and $R^2 = 0.0001$, $p = 0.860$, **Figure 4C**) or stroke length ($R^2 = 0.022$, $p = 0.220$ and $R^2 = 0.012$, $p = 0.370$). Tests to determine whether the slopes and intercepts (i.e., strategy) are different between participants

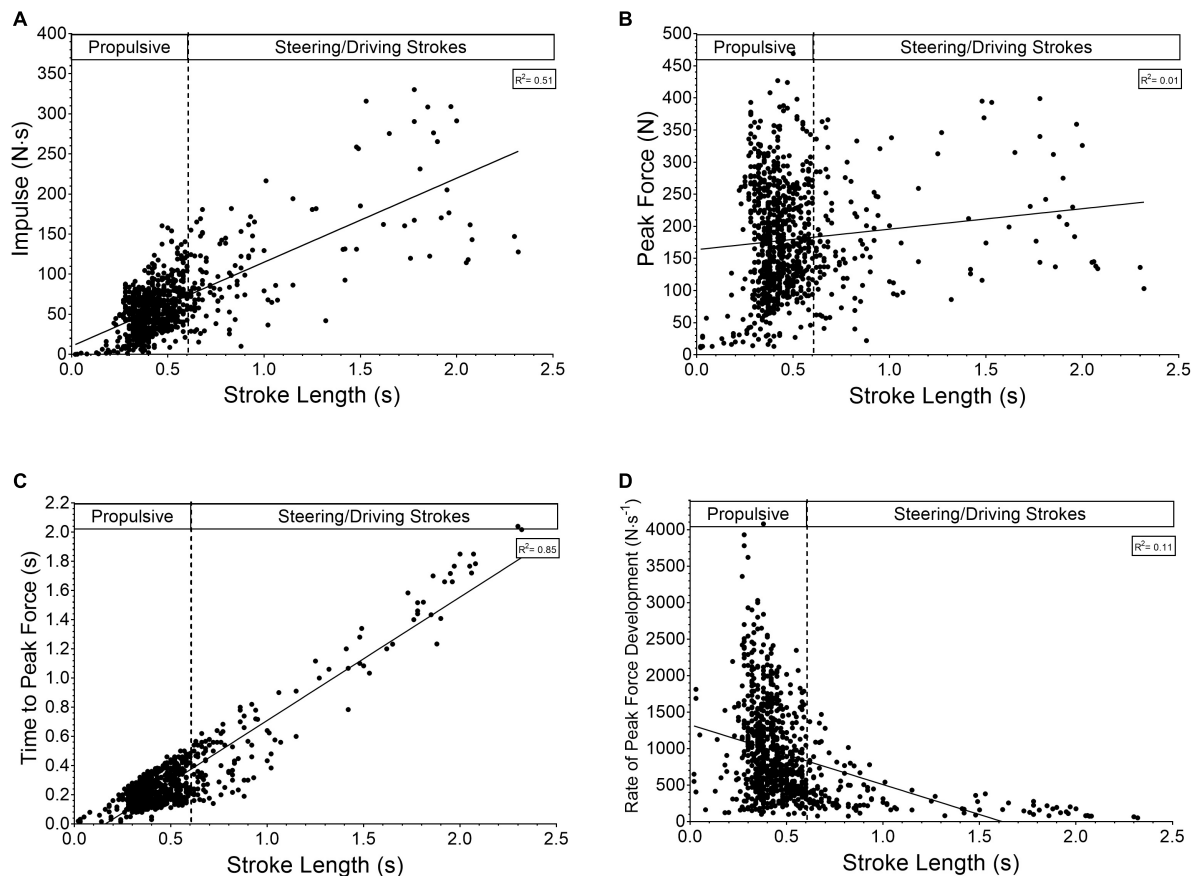


FIGURE 5 | Relationship between stroke length and (A) impulse, (B) peak force, (C) time to peak force, and (D) rate of peak force development.

for strokes indicated a significantly different slope for peak force ($p < 0.0001$) and rate of peak force development ($p < 0.0001$) but not stroke length ($p = 0.9811$), impulse ($p = 0.1857$) or time to peak force ($p = 0.9402$). Where possible intercepts were identified as significantly different ($p < 0.0001$) for stroke length, impulse and time to peak force. Comparisons between performance time and slope ($R^2 = 0.4016$, $p = 0.0916$; $R^2 = 0.4150$, $p = 0.0847$) and y-intercept ($R^2 = 0.3571$, $p = 0.1177$; $R^2 = 0.3613$, $p = 0.1150$) for peak force and rate of peak force development over the simulated competition run showed moderate-strong relationships, though not significant.

Further analysis of variables of force based around stroke length indicates that strokes of longer duration have greater impulse ($R^2 = 0.507$, $p < 0.0001$; **Figure 5A**), vary little with regards to peak force magnitude ($R^2 = 0.012$, $p = 0.0009$; **Figure 5B**), take longer to reach peak force ($R^2 = 0.851$, $p < 0.0001$; **Figure 5C**), with a lower rate of force development ($R^2 = 0.107$, $p < 0.0001$; **Figure 5D**).

DISCUSSION

This investigation set out to describe the work requirement of a simulated flatwater slalom in the field with reference to physical

and physiological demand. In agreement with our hypothesis the main findings were: (a) Paddling in a straight-line with no turning strokes is more efficient than negotiating a course of slalom gates; (b) Forward propulsive strokes are key to simulated race distance on flatwater; (c) Flatwater slalom is dominated by a preponderance to anaerobic respiration in the early stages and during key aspects of competition while high-intensity aerobic respiration dominates overall; (d) Driving/turning stroke force profiles differ to propulsive strokes; (e) Performance is related to the peak force, its rate of development and the corresponding slope of fatigue as the competition progresses.

The introduction of a kayak paddle shaft enabling real time work rate monitoring (Macdermid and Fink, 2017) and subsequently its use in the field, enables valuable monitoring opportunities for athletes, coaches and sports scientist alike. The data presented within **Figure 2** and **Table 1** display athlete characteristics traditionally gained from a laboratory (Zamparo et al., 2006), which exclude sport specific actions related to steering, balance, and proprioception.

The participants used for this study were all part of the New Zealand development squad from which two members qualified for world cup semi-finals during the year of testing. Even though physiological variables assessed were taken from participants paddling on water, over a pre-determined slalom

course, maximal oxygen consumption was similar to those previously reported during laboratory assessments (Pendergast et al., 1979; Vaccaro et al., 1984; Zamparo et al., 2006; Tremblay et al., 2012).

Determining the power:oxygen consumption relationship enabled athlete economy assessment, but primarily was used to estimate energy system contribution as previously reported in other sports (Macdermid and Stannard, 2012). Due to the two basic components of the test used it was possible to compare straight-line paddling with the askew nature of negotiating slalom gates. Paddling at the same work rate over the two components led to a ~15% increase in physiological demand, i.e., the oxygen consumption over the given epoch at the specific work rate. Care must be taken when interpreting such results as time course may influence the results as the slalom section followed the straight-line component. However, this is unlikely due to the sub-maximal intensities used and is more likely a result of extra physical demand. A result of turning, expressed through longer strokes and a resultant increase in stroke impulse (Figure 5A) typical of turning/driving after periods of deceleration. As such, participants have to work harder to maintain the same power outputs when exercise is intermittent in nature compared to continuous. Future research could use such methods to explore the efficacy of training methods and long-term athlete development concerning physical, technical, and physiological capability along with the ensuing performance. Similarly, changes to equipment design (boats or paddle blades) could be assess physiological response to set work rates and ratified through additional performance testing.

In setting out to simulate a race, participants were asked to approach it with a similar strategy to that which they would typically use. The supra-maximal power outputs presented (Figures 3A,B) implies this occurred, and is associated with a large oxygen deficit (Figure 3C) and thus anaerobic contribution (Figure 3D) to exercise (Gastin, 2001). This is important for a sport that has a large dependency on cognitive function (MacIntyre and Moran, 2007) and where supra-maximal intensities have been shown to impair such performance (Isaacs and Pohlman, 1991) along with physical capabilities (Mendez-Villanueva et al., 2008). As such, paddlers must employ a strategy based on an understanding of physical and physiological capability in order to perform manoeuvres demanded by the course set. Failure to judge this correctly will lead to increased chance of incurred time related and/or time-penalty mistakes. It is likely that our participants deemed the technical demand of flatwater slalom easier than a world-class course and as such work rate was higher throughout. However, further research needs to corroborate this, along with optimal pacing strategies in regards to physical and physiological capabilities and the performance outcome. With this in mind, we advocate physical and physiological testing be performed in the athletes normal sport specific environment so data can be used for training prescription and related to real world performance.

Similar duration events, excluding the same level of technicality, have been shown to commence with supra-maximal work rates as shown in Figures 3A,B and likewise

settle quickly to a more sustainable level (Craig and Norton, 2001). This level separating the heavy-severe domain of exercise intensity has been termed critical power (Hill, 1993) and relates to an exercise intensity similar to that associated with maximal oxygen consumption capability (Vanhatalo et al., 2007). This supports the oxygen deficit profile presented (Figure 3C) where it drops to a sustainable level between 20 and 30 s and is negligible by 60 s, emphasizing the importance of a large component or requirement for a high aerobic capacity combined with the already established high anaerobic capability.

Variables of force (Impulse, peak force, time to peak force, rate of peak force development, Figure 4) per stroke over the whole simulated race run showed a different pattern (excluding rate of peak force development, Figure 4D) to power output. This data suggests that participants technique with regards to impulse and time to develop peak force remained similar throughout (Figures 4A,C) while the magnitude of peak force decreased linearly (Figure 4B). This could highlight specific aspects in need of developing within this cohort such as an ability to sustain greater peak forces per stroke and can now be monitored through technological advances.

Classifying strokes based on time-force thresholds and using frequency analysis highlights a preponderance for propulsive strokes during flatwater slalom. The relationship between propulsive and steering or driving strokes (Figures 5A–D) suggests that those used to manoeuvre the boat require similar peak forces but developed over greater time and thus resulting in greater impulse. These movements are usually associated with reductions in speed or maintaining position in the water. Further research into their nature regarding muscular contraction type, could enhance training methodology within the sport. Additionally, an increase in such movements over a competition run would likely see a competitors average power output decrease suggesting that the ratio of propulsive strokes to steering/driving stroke could be a good indicator of performance or technical capability. Again, it is likely that the 94:6 stroke ratio obtained in this study would differ during actual racing on a white-water course due to the increased demand of steering the boat, but this needs corroborating.

CONCLUSION

The results of the present study indicates that a flatwater slalom under simulated race conditions has a power profile dependent upon the course set, but typically evolves around an initial supra-maximal (anaerobic) work rate with a subsequent transition to one associated with maximal aerobic capacity. Turning, accelerating or decelerating manoeuvres occurring throughout a competition require high forces generated over a significantly longer time period than forward propulsive strokes, while the ability to generate peak force decreases over the competition period. As such, athletes need to gauge fatigue levels and subsequent effort afforded to propulsion as the competition run progresses in order to sustain turning manoeuvres essential to negotiating the course.

Practical Implications

The following are practical recommendations for athletes and coaches working with slalom kayak athletes:

- The use of a powermeter kayak shaft provides athletes, coaches and support staff quantifiable sport specific data with regards to physical, physiological and technical competency not currently available to slalom kayakers.
- Athlete performance plans need to plan to improve physical, physiological, and technical aspects of paddling that enables maintenance of peak force(s) and its rate of development over a competition run. This requires development of both anaerobic and high intensity aerobic capability combined with aspects of specific (on-water) strength.

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Conflict of Interest Statement: AO was employed by company High Performance Sport NZ and Canoe Slalom NZ. All other authors declare no competing interests.

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Can Match-Mimicking Intermittent Practice Be Used as a Simulatory Training Mode of Competition Using Olympic Time Frame in Elite Taekwondo Athletes?

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Edited by:

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 03 December 2018

Accepted: 25 February 2019

Published: 20 March 2019

Citation:

Chuang S-J, Sung Y-C, Chen C-Y,
Liao Y-H and Chou C-C (2019) Can
Match-Mimicking Intermittent
Practice Be Used as a Simulatory
Training Mode of Competition Using
Olympic Time Frame in Elite
Taekwondo Athletes?
Front. Physiol. 10:244.
doi: 10.3389/fphys.2019.00244

Aim: The physiological realities between Taekwondo (TKD) simulation kicking training and TKD competition according to the Olympic time frame remain unclear. The purpose of this study is to establish an Olympic match-simulated kicking model and compare its effects with real TKD competition on physiological challenges and hormonal responses during serial matches in elite athletes.

Method: Sixteen elite TKD athletes randomly were assigned into either a TKD match-simulated kicking group (TMSK; $N = 8$, age: 21.3 ± 0.2 years) or a simulated TKD competition group (STC; $N = 8$, age: 21.6 ± 0.5 years). Both groups performed either simulated kicking or TKD competitions in the same time-course order, and all physiological parameters and blood sampling time-points were identical between groups. The heart rate (HR) and rating of perceived exertion (RPE) were recorded during each match-simulated kicking and TKD competition session. Blood samples were obtained before competition (Pre-Comp.), after competition—in this case meaning four consecutive matches (End-Comp.), and 24 h after the first match (Next day) for determination of biomarkers of muscle damage (myoglobin and CK), hematological profiles, and hormonal profiles (testosterone and cortisol).

Results: The responses of HR, RPE, and blood lactate levels during the consecutive testing sessions showed no differences between TMSK and STC. The changes in CK and myoglobin were greater in STC ($p < 0.05$), and a greater decrease in red blood cell (RBC) loss was observed in the STC group ($p < 0.05$). Compared with TMSK, the inflammatory state, reflected by the ratios of neutrophils-to-lymphocyte (NLR) and platelets-to-lymphocyte (PLR), was higher in STC ($p < 0.05$). Moreover, the catabolic state (cortisol/testosterone) was greater in STC than in TMSK ($p < 0.05$).

Conclusion: We demonstrated that, compared with TMSK, the STC produced greater muscle damage, inflammatory responses, and catabolic stress in the Olympic competition time frame in elite male TKD athletes. Although TMSK is capable of eliciting similar physiological challenges as TKD competition, the muscle damage and hormonal profiles provoked by TMSK were not comparable to TKD competition. Our findings provide

science-based data and better understanding for coaches, athletes, and sports scientists to develop TKD-specific training programs for Olympic preparation.

Keywords: high-intensity interval exercise, heart rate, muscle damage, inflammation, full-contact competition

INTRODUCTION

Among competitive sports, combat/martial art sports are unique and contain several specific features, including short periods with extremely high intensity, intermittent exercise patterns, and frequent physical contact/impacts during competition events. Taekwondo (TKD), a traditional Korean martial art combat sport, has evolved into a modern Olympic discipline (Bridge et al., 2009). TKD is an intensive sport with a high demand for anaerobic energy (~30% of total metabolic work during combat) and is accompanied by tremendous muscle injury or soreness from intense eccentric contractions (Bridge et al., 2009; Crisafulli et al., 2009; Campos et al., 2012; Lopes-Silva et al., 2018) during athletic practice and competitions (Balnave and Thompson, 1993; Kwok, 2012). Furthermore, according to the Olympics and World Taekwondo (WT) regulations, TKD combats are comprised of at least three rounds of 2 min combat and 1 min break between each round (World Taekwondo, 2018), and the athletes may have to fight in consecutive schedules for at least 4–5 matches within a single day to be qualified to compete for the final championship. Consequently, the accumulative physiological fatigue during competitions is extreme in TKD athletes.

For coaches and athletes in combat sports, integrating tasks that represent formal competitive conditions into a regular training program is a common way to promote the transfer of combat skills from training to competition. To enhance practice, TKD athletes need to complete representative learning tasks for simulating key aspects of competition (Araújo et al., 2007). Accordingly, athletes not only have to perform TKD-specific skills and conditioning training but also have to participate in regional tournaments. However, these reality simulated training regimens (team combat practice, friendly match/competition, and regional tournament) possibly cause varying levels of sport injuries due to the frequent physical contact between athletes (Kazemi et al., 2005; Covarrubias et al., 2015; Hammami et al., 2018), because injuries are an inherent risk due to the nature of combat sports. These negative impacts during training may thus result in short-term training cessation and subsequently perturb athletic performance and competition preparations (Eston et al., 2003).

On the other hand, during intensified competitions or intense training, exercise-induced muscle damage has been associated with decreases in muscle strength, impaired muscle contractile functions, and impaired muscle recovery capacity (Knitter et al., 2000; Kendall and Eston, 2002), which may further impair subsequent competition performance. In addition, exercise-induced stress also markedly increases systematic inflammatory responses (Chou et al., 2018), cell damage (Chou et al., 2018), and catabolic/anabolic hormonal balance (Brownlee et al., 2005; Pilz-Burstein et al., 2010) in both general populations and

combat sport athletes. Hence, it would be critical to develop practical training models with less physical contact while still being capable of mimicking the physiological reality of international competition and minimizing injury risks. It has also been shown that TKD combat simulation is not able to perfectly mimic TKD competition. During combat simulation, TKD athletes displayed similar exchange time, shorter preparation time, and a longer exchange preparation ratio compared to TKD competition using time-motion analysis (Hausen et al., 2017). Nevertheless, it is still little known whether competition-simulated exercise with less direct physical contact could completely reproduce the physiological responses of competition in addition to bring the benefit of lower injury risk.

To date, there is still very limited number of studies comparing the differences between simulated-kicking/fight-training and TKD competition in elite TKD athletes (Hausen et al., 2017; Maloney et al., 2018). One recent psychological study reported that the emotional and cognitive demands of competition cannot be simulated by fighting in training (Maloney et al., 2018), and Tayech et al. suggest that the anaerobic intermittent kick test can be used as a reliable and specific test for evaluating anaerobic power in TKD athletes (Tayech et al., 2019). Thus, it is still unclear whether a TKD-specific simulated training mode would be capable of replicating the physiological challenges of combat during intensive competitions. Moreover, to our knowledge, there is no study that has developed a low-impact fighting simulation training model and examined the relevant physiological variables in accordance with the Olympic competition time frame.

In TKD, kick techniques are one of the primary attacking strategies (Pieter and Heijmans, 1997; Whang et al., 1999) and are very frequently used in competitions (Matsushigue et al., 2009; Kwok, 2012). According to the literature, for both medalists and nonmedalists, the main attack actions used during TKD combat are kicks (approximately 98% of overall attacks per match), such as roundhouse kick (63–69%), sidekick (6–7%), or reverse sidekick (1–2%) (Kwok, 2012; Bridge et al., 2014). Several studies have pointed out that the work-to-rest ratio (WRR) varied from 1:1 to 1:9 due to the level of games (Matsushigue et al., 2009; Santos et al., 2011; Campos et al., 2012; Del Vecchio et al., 2016). These varied WRRs across studies also create difficulties to reconstruct an appropriate simulation model for this combat sport. Although Matsushigue et al. (2009) reported a 1:1 WRR during a TKD match with 2-min rounds, the authors used Songahm Taekwondo competitions, which have very different rules compared to World Taekwondo. On the other hand, it has to be noted that the TKD athletes still have to perform high-intensity avoiding/dodging or other defending movements when they are not attacking, which may lead to an underestimation of real working time during combat. Considering these aspects, we reconstructed a competition-simulated kick model (TMSK) consisting of the most common kick techniques with a WTT of 1:1 (continuous 10-s

kicking attacks) in order to elicit sufficient physiological stress and exercise intensity during the simulation. Consequently, we here developed a TKD-specific 2-min intermittent exercise model consisting of continuous 10-s shifting kicks (sidekick/reverse side kick/roundhouse kick)/10-s low intensity bouncing (1:1 WRR), attempting to reconstruct the physiological challenges during TKD combat, and compared this simulated intermittent kick model with a real TKD combat match. In this regard, we hypothesized that the TKD match-simulated kicking (TMSK) model would elicit comparable physiological stress and hormonal responses to the extent of TKD competition with an Olympic time frame. Therefore, the purpose of this study is to compare the effects of high-intensity intermittent match-simulated kicking and real TKD matches on the cardiopulmonary demands, physiological challenges, muscle-damaging biomarkers, and anabolic/catabolic hormonal responses in elite combat TKD athletes.

MATERIALS AND METHODS

Participants

Sixteen elite TKD athletes (mean \pm S.E.M.; age: 22.0 ± 0.3 years, height: 177.0 ± 1.8 cm, body weight: 71.0 ± 2.2 kg, BMI: 23.0 ± 0.4) participated in this study, and they were weight-matched and randomly assigned into either TMSK ($n = 8$) or STC ($n = 8$). All participants hold black belts, classifying in the National Division I category. The weight class of participants was classified based on Olympic weight class rules as follows: ≤ 58 kg ($n = 2$), >58 to ≤ 68 kg ($n = 4$), >68 to ≤ 80 kg ($n = 6$), >80 kg ($n = 4$). All combat matches were conducted by matching players with opponents at the similar skill levels in the same weight categories. Participants were free of musculoskeletal injuries and cardiovascular/metabolic disorders, determined by screening with a self-screening health questionnaire. This study was carried out in accordance with the recommendations of the Institute Review Board (IRB) of the University of Taipei with written informed consent from all subjects. All subjects gave written informed consent according to the Declaration of Helsinki.

Experimental Design

A randomized and weight-matched study was used. During the trial day, all participants had the same meals. Before the first match, the participants consumed a breakfast provided by the researchers (energy: ~ 530 kcal; carbohydrate: 62%; fat: 20%; protein: 18%). Standardized snacks (energy: ~ 400 – 500 kcal; carbohydrate: 61–83%; fat: 5–20%; protein: 9–18%) were given immediately after each match, and dinner (energy: $\sim 1,100$ kcal; carbohydrate: 57%; fat: 25%; protein: 18%) was given after the last match (Match #4). The participants were requested to consume the provided food within 30 min to ensure consistency.

Match-Simulated Kicking Mode and Simulated Competition

Participants in STC fought against an opponent in their matched weight category, which was based on Olympic weight regulation

and equal experience to ensure the intensity of competition. The competition was performed in an 8 m \times 8 m rubber mat octagonal competition area according to WT regulations and rules. Each match was composed of three rounds of 2 min and two breaks of 1 min. The consecutive competition (STC)/simulated matches (TMSK) were performed at 8:30 h (Match #1), 10:30 h (Match #2), 14:00 h (Match #3), and 16:30 h (Match #4) (Figure 1A). Participants wore the whole set of electronic protectors (2.7 kg; Dae do International, Barcelona, Spain) during simulated competition. For the TMSK, the athletes wore the whole set of regular protectors (2.1 kg; Adidas Double-d Martial Arts, Émerainville, France) during simulation, and the extra weight vest (0.6 kg) was used to balance the difference between the electronic and normal protectors. To simulate TKD match, subjects continuously kicked a soft target (Figure 1B) for 10 s as many times as possible and rested actively by low intensity bouncing for 10 s (1:1 WRR) between each trial of kicking to simulate a high-intensity interval pattern such as what would occur in competition. As sidekick/reverse sidekick/roundhouse kick are the primary choice of attack and accounted for 72–76% of the total attacks (Kwok, 2012; Bridge et al., 2014), subjects performed three different kicking techniques in sequence in a simulation based on the time frame of an Olympic competition schedule. The heart rate was measured using a wireless heart rate monitor (Polar® RS800CX™; Polar Electro Inc., Lake Success, NY, USA) during exercise and rest for both STC and TSMK groups.

Measurement of Exercise Intensity and Fatigue

To monitor heart rate, a wireless heart rate monitor was attached to participants and the heart rate (HR) was periodically recorded during the experiment procedure. Blood lactate was measured from fingertip blood samples immediately post-match using a lactate meter (Edge Blood Lactate Monitoring System, ApexBio Inc., Taipei City, Taiwan). Borg rating of perceived exertion (RPE, 6–20 scale) was used to monitor exercise intensity, and the participants were familiarized with subjective scale prior to trial. During the STC or TMSK, the RPE rating was recorded at baseline, Round 1 (R1), Round 2 (R2), and Round 3 (R3) of each match.

Analyses of Muscle Damage Biomarkers, Hematological Profiles, and Hormonal Levels

The 10-ml venous blood samples were collected in a tube coated with EDTA before Match #1 (Pre-Comp.), after completion of Match #4 (End-Comp.) and at 24 h after the first match (Next day). An automated hematology analyzer (Sysmex XT-2000, Sysmex Corp., Kobe, Japan) was used to analyze hematological profiles, including neutrophils, white blood cells (WBC), red blood cells (RBC), lymphocytes, and platelets, according to the manufacturer's instructions. Both the neutrophils-to-lymphocytes ratio (NLR) and the platelet-to-lymphocytes ratio (PLR) have been clinically recognized as systemic inflammatory markers (Turkmen et al., 2013; Balta et al., 2016). Creatine

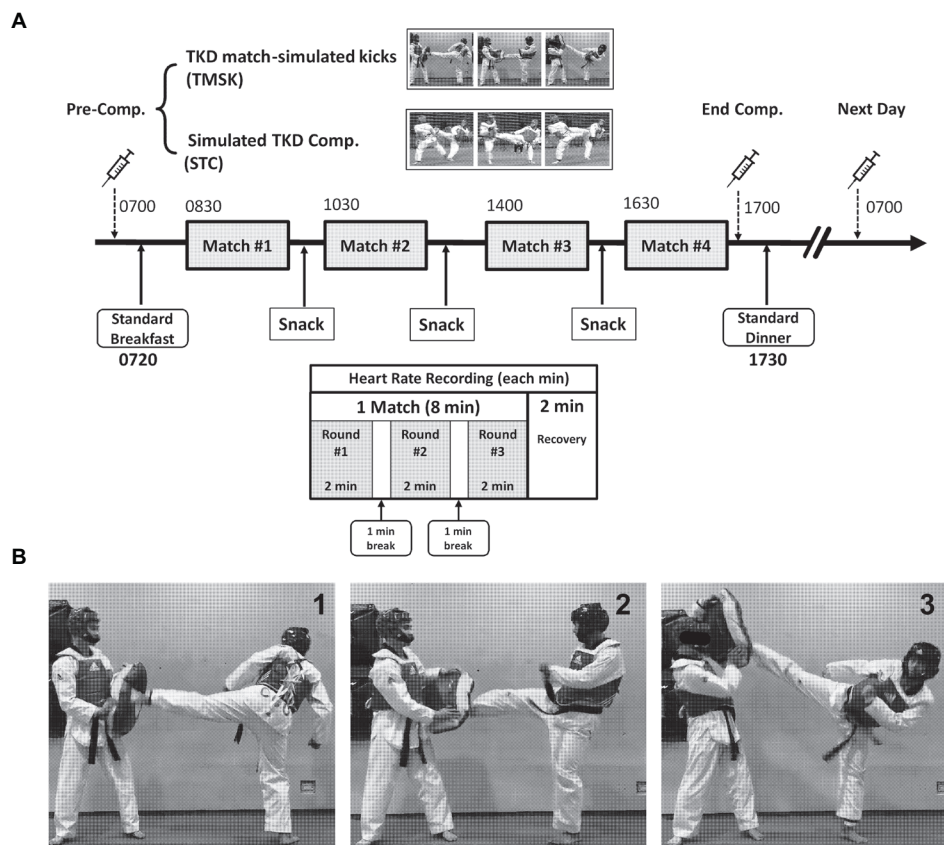


FIGURE 1 | Experiment design and procedure of match in accordance with current Olympic TKD competition time frame. **(A)** Four consecutive combat matches were performed at 8:30, 10:30, 14:00, and 16:30. The blood samples were drawn before matches (7:00), after the completion of the last match (17:00), and on Next day (07:00; 24 h after the first match). **(B)** Three different kicks were used in the intermittent interval exercise (10-s kicking with full effort and 10-s bouncing jumping break) in the present study, and the movements used to reconstruct real TKD competition during the TMSK, including (1) sidekick, (2) reverse side kick, and (3) roundhouse kick. The individuals in this manuscript gave written informed consent to use and publish these image details. TMSK: TKD-match simulated kicks; STC: simulated TKD competition.

kinase (CK) and myoglobin are commonly used indicators of muscle damage (Balnave and Thompson, 1993; Serrao et al., 2003). The concentration of plasma CK was determined using an LX-20 clinical chemistry analyzer (intra-assay CV = 7.5%; Beckman, Brea, CA, USA). Serum myoglobin was determined using radioimmunoassay by test kit (intra-assay CV = 1.5%; Daiichi Radioisotope Laboratory Ltd, Tokyo Japan). The circulating testosterone and cortisol levels were measured using commercially available enzyme-linked immunosorbent assay (ELISA) kits (testosterone: intra-assay CV% = 11.95%; #582701; cortisol: intra-assay CV% = 8.35%; #500360; Cayman Chemical Co., Ann Arbor, MI, USA), and the optical density of ELISA analyses was determined using a TECAN Genios reader (Salzburg, Austria) according to the manufacturer's instructions.

Statistical Analysis

All data were expressed as mean \pm standard error of mean (Mean \pm S.E.M.). Data were analyzed and depicted by SPSS 16.0 software (SPSS, Chicago, IL, USA) and GraphPad Prism 5.0

(GraphPad Software Inc., La Jolla, CA, USA), respectively. Prior to performing statistical analysis, all data were examined for normality of distribution. The heart rate, blood lactate, and blood biomarkers (e.g., creatine kinase, myoglobin, hematological profiles, hormonal responses, etc.) were analyzed using two-way mixed analysis of variance (2-way ANOVA; treatment group \times time) with repeated measures to determine the intra- and inter-group differences. The effect size was calculated by Cohen's *f*. When significance was found, *post hoc* testing (Tukey's multiple comparisons test) was used to analyze the difference. The alpha level was set at 0.05 ($p \leq 0.05$) for statistical difference.

RESULTS

Physiology Response Measuring Heart Rate, RPE, and Lactate Levels

Figure 2 shows the changes in heart rate (Figure 2A), blood lactate (Figure 2B), and RPE (Figure 2C) during four TKD

matches and the simulation. For heart rate, there was a significant main effect of time ($F(44, 616) = 172.1$, $p < 0.001$, $\eta^2 = 0.924$) but not mode and interaction (**Figure 2A**). There was no effect of mode and interaction observed in RPE. Consistent with heart

rate, RPE increased with time ($F(12, 168) = 114.8$, $p < 0.001$, $\eta^2 = 0.901$) (**Figure 2B**). For lactate, as shown before in RPE, there was a significant main effect of time ($F(4, 56) = 90.42$, $p < 0.001$, $\eta^2 = 0.849$) but not of mode or interaction (**Figure 2C**).

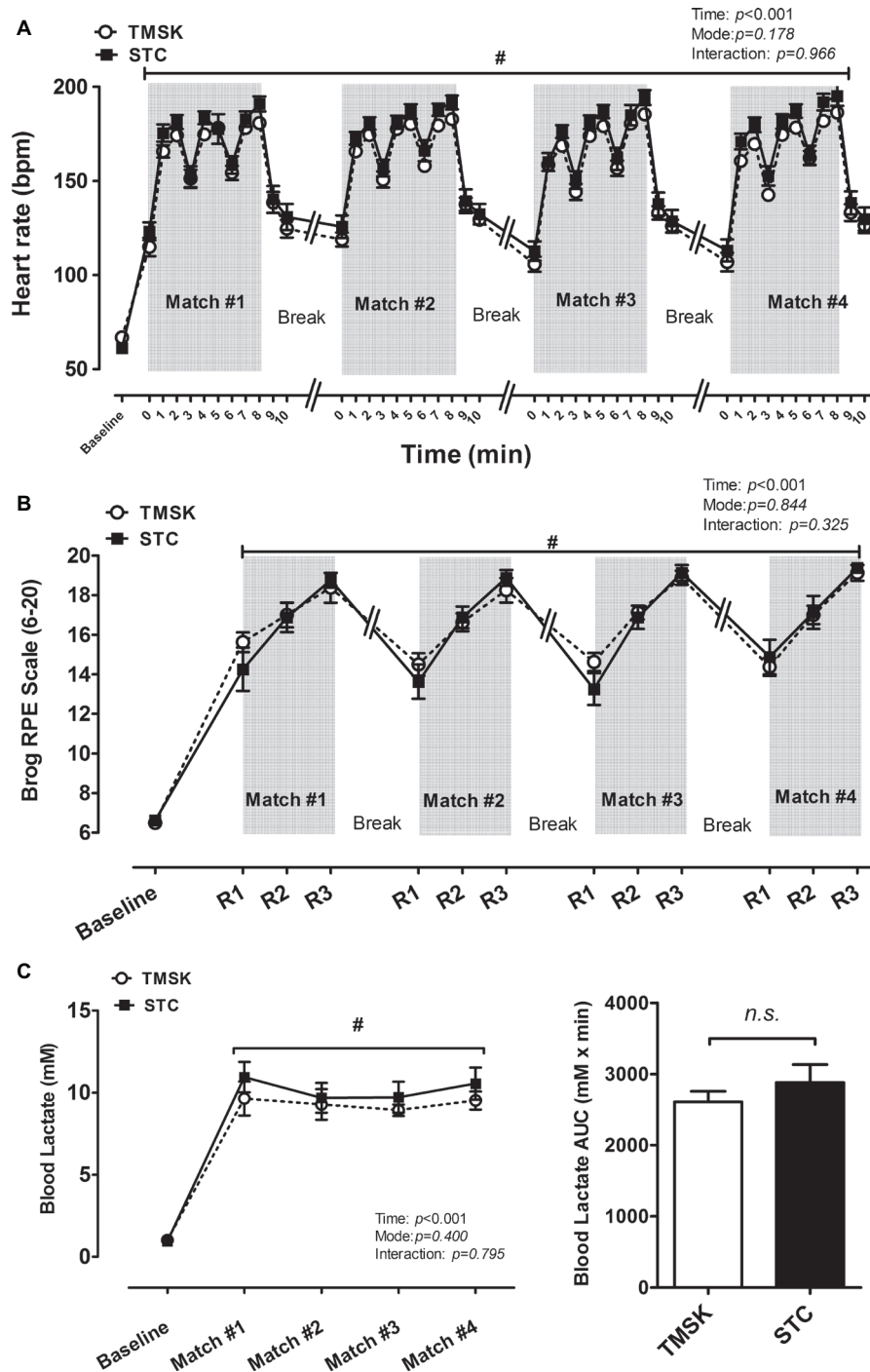


FIGURE 2 | The physiological responses during real combat match and simulation kicking exercise. **(A)** heart rate, **(B)** blood lactate, and **(C)** subjective Borg RPE. # denotes significant difference compared to baseline values ($p < 0.05$). TMSK: TKD-match simulated kicks; STC: simulated TKD competition.

Muscle Damage and Intravascular Hemolysis

Figure 3 shows the effect of TKD combat matches and the kicking simulation on muscle damage (**Figures 3A,B**) and intravascular hemolysis (**Figures 3C,D**). For serum myoglobin and CK levels, there was a significant main effect of time (myoglobin: $F(1, 14) = 28.31, p < 0.001, \eta^2 = 0.657$; CK: $F(2, 28) = 15.59, p < 0.001, \eta^2 = 0.429$), mode (myoglobin: $F(1, 14) = 17.14, p < 0.001, \eta^2 = 0.550$; CK: $F(1, 14) = 7.568, p = 0.016, \eta^2 = 0.351$), and interaction (myoglobin: $F(1, 14) = 19.19, p < 0.001, \eta^2 = 0.565$; CK: $F(2, 28) = 10.50, p < 0.001, \eta^2 = 0.336$). *Post hoc* analyses showed significant increases in myoglobin in STC at End-Comp (95% CI: 157.2–361.0, $p < 0.001$) and significant increases in CK level in STC at Next day (95% CI: 471.8–1,447, $p < 0.001$). Intravascular hemolysis can be measured by the decrease in red blood cells. For hematocrit and hemolysis, there was a significant main effect of time (hematocrit: $F(2, 28) = 19.20, p < 0.001, \eta^2 = 0.578$; hemolysis: $F(2, 28) = 14.58, p < 0.001, \eta^2 = 0.510$), mode (hematocrit: $F(1, 14) = 11.69, p = 0.004, \eta^2 = 0.852$; hemolysis: $F(1, 14) = 18.73, p < 0.001, \eta^2 = 0.628$), and interaction (hematocrit: $F(2, 28) = 13.60, p < 0.001, \eta^2 = 0.493$; hemolysis: $F(2, 28) = 13.95, p = 0.002, \eta^2 = 0.499$). *Post hoc* analyses showed significant decreasing in STC in hematocrit (End-Comp: 95% CI: –8.584 to –2.641, $p < 0.001$; Next day: 95% CI: –6.984 to –1.041, $p = 0.005$) and in hemolysis (End-Comp: 95% CI: –10.44 to –4.337, $p < 0.001$; Next day: 95% CI: –7.525 to –1.425, $p = 0.002$).

Hematological Profiles and Inflammatory Parameters

Figure 4 represents the changes of WBC (**Figure 4A**), neutrophils (**Figure 4B**), lymphocyte (**Figure 4C**), NLR ratio (**Figure 4D**), platelet (**Figure 4E**), and PLR ratio (**Figure 4F**) during matches or kicking simulation. For WBC, there was

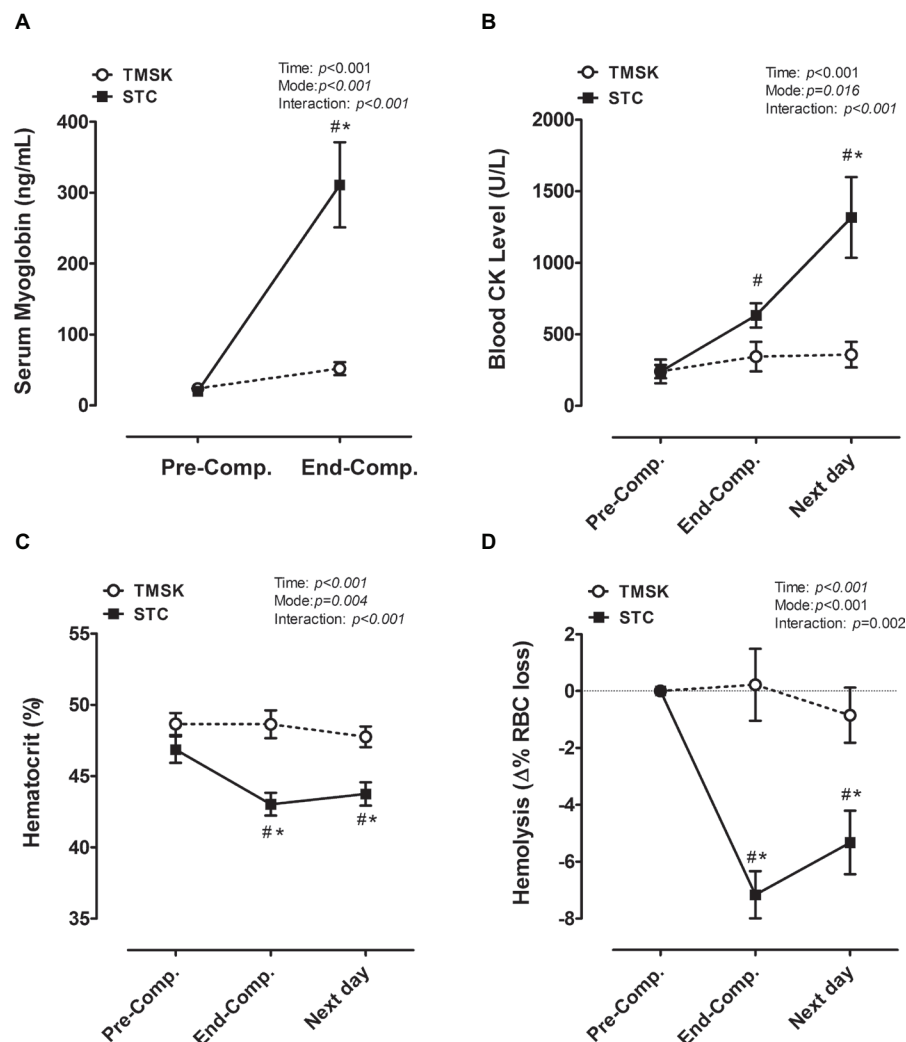
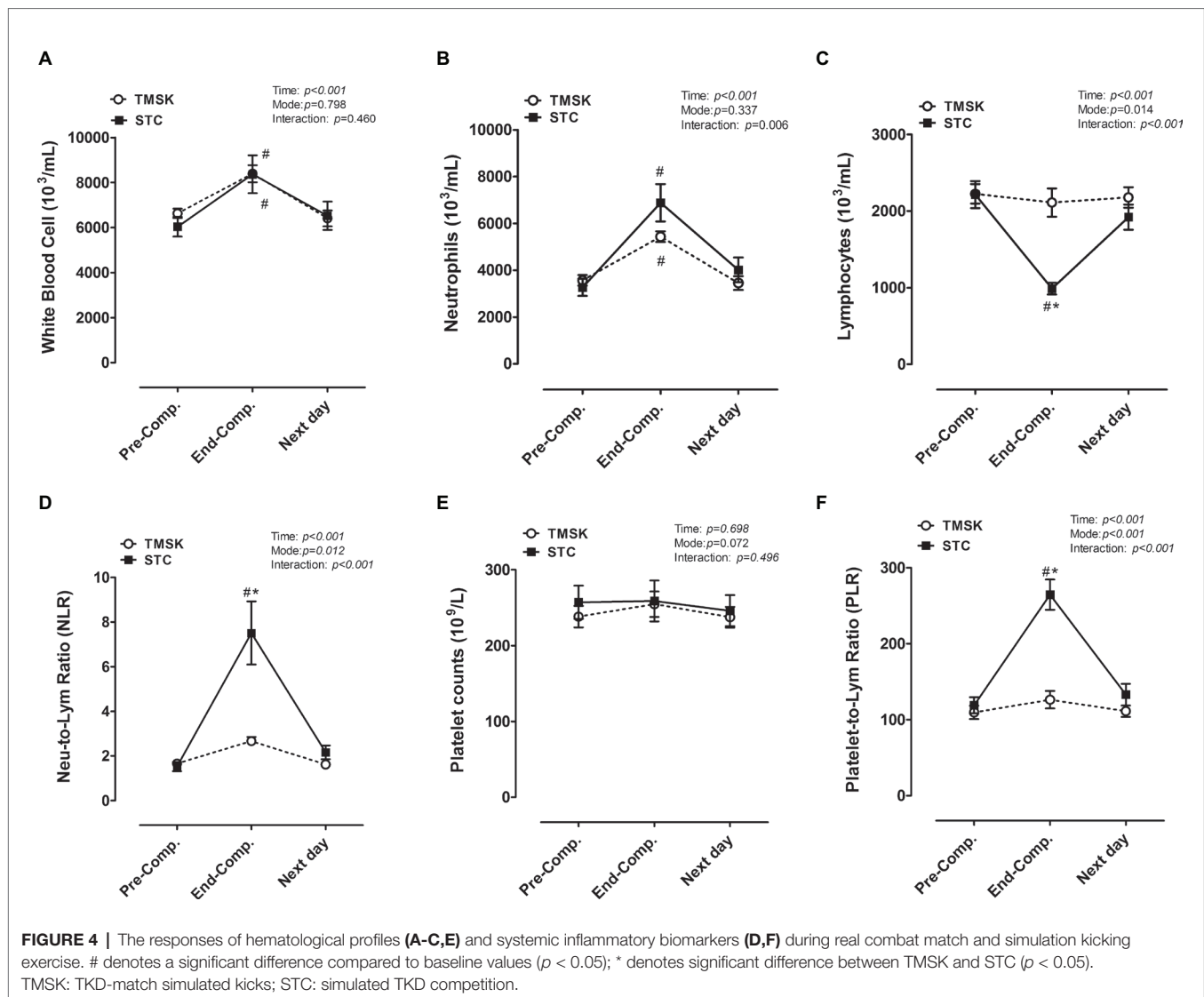


FIGURE 3 | The responses of muscle-damaging blood biomarkers and status of hemolysis during real combat match and simulation kicking exercise. **(A)** Serum myoglobin, **(B)** blood CK, **(C)** hematocrit, **(D)** hemolysis. # denotes the significant difference compared to baseline values ($p < 0.05$); * denotes a significant difference between TMSK and STC ($p < 0.05$). TMSK: TKD-match simulated kicks; STC: simulated TKD competition.



a significant main effect for time ($F(2, 28) = 29.89$, $p < 0.001$, $\eta^2 = 0.681$) but not mode or interaction (**Figure 4A**), and both STC and TMSK increased WBC (TMSK: 95% CI: $-2,740$ to -787.9 , $p < 0.001$; STC: 95% CI: $-3,308$ to $-1,357$, $p < 0.001$) at End-Comp., and the values returned to baseline at Next day. There was a significant main effect for time ($F(2, 28) = 72.48$, $p < 0.001$, $\eta^2 = 0.838$) and interaction ($F(2, 28) = 6.100$, $p = 0.006$, $\eta^2 = 0.303$) but not mode in neutrophils, and both STC and TMSK increased neutrophils (TMSK: 95% CI: $-2,742$ to -999.3 , $p < 0.001$; STC: 95% CI: $-4,481$ to $-2,739$, $p < 0.001$) at End-Comp., and the values returned to baseline at Next day (**Figure 4B**). For lymphocytes, significant main effects for time ($F(2, 28) = 19.03$, $p < 0.001$, $\eta^2 = 0.576$), mode ($F(1, 14) = 7.959$, $p = 0.014$, $\eta^2 = 0.474$), and interaction ($F(2, 28) = 13.39$, $p < 0.001$, $\eta^2 = 0.489$) were observed. Only STC significantly decreased lymphocytes at End-Comp. (95% CI: 855.9 – $1,600$, $p < 0.001$) below the level of Pre-Comp., and the lymphocytes was significantly lower in STC than TMSK at End-Comp. (95%

CI: $-1,647$ to -600.5 , $p < 0.001$) (**Figure 4C**). For the NLR ratio, a significant main effect for time ($F(2, 28) = 26.79$, $p < 0.001$, $\eta^2 = 0.657$), mode ($F(1, 14) = 8.271$, $p = 0.012$, $\eta^2 = 0.369$), and interaction ($F(2, 28) = 12.99$, $p < 0.001$, $\eta^2 = 0.481$) was observed. Only STC significantly increased NLR at End-Comp. (95% CI: -182.6 to -108.6 , $p < 0.001$) above the level of Pre-Comp., and the NLR was significantly higher in STC than TMSK at End-Comp. (95% CI: 93.34 – 183.4 , $p < 0.001$) (**Figure 4D**). The platelet counts did not show any significant differences during TKD matches, and there were no differences between the two groups (**Figure 4E**). For the PLR ratio, a significant main effect of time ($F(2, 28) = 31.72$, $p < 0.001$, $\eta^2 = 0.694$), mode ($F(1, 14) = 20.23$, $p < 0.001$, $\eta^2 = 0.575$), and interaction ($F(2, 28) = 20.06$, $p < 0.001$, $\eta^2 = 0.589$) was observed. Only STC significantly increased PLR at End-Comp. (95% CI: -182.6 to -108.6 , $p < 0.001$) above the level of Pre-Comp., and the PLR was significantly higher in STC than TMSK at End-Comp. (95% CI: 93.34 – 183.4 , $p < 0.001$) (**Figure 4F**).

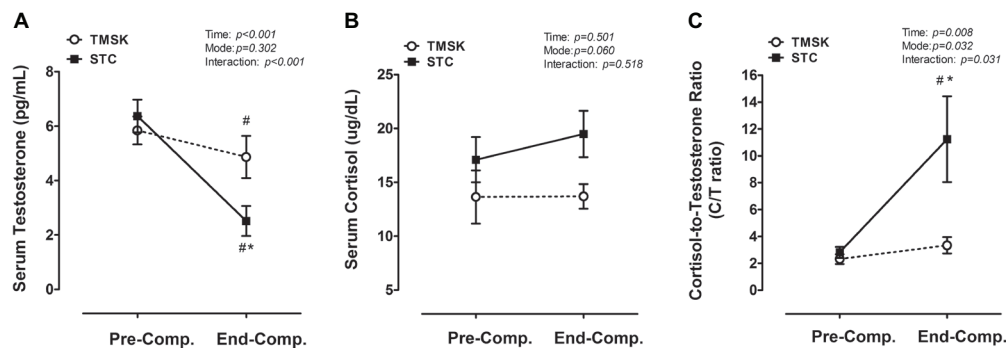


FIGURE 5 | The responses of anabolic and catabolic hormones and balance during real combat match and simulation kicking exercise. **(A)** Serum testosterone, **(B)** serum cortisol, and **(C)** C/T ratio. # denotes the significant difference compared to baseline values ($p < 0.05$); * denotes significant difference between TMSK and STC ($p < 0.05$). TMSK: TKD-match simulated kicks; STC: simulated TKD competition.

Anabolic/Catabolic Hormone Responses to Taekwondo Matches

Figure 5 indicates the changes of circulating testosterone (Figure 5A), cortisol (Figure 5B), and systemic catabolic index presented by C/T ratio (Figure 5C) during simulated competition or kicking simulation. For serum testosterone, a significant main effect for time ($F(1, 14) = 127.3$, $p < 0.001$, $\eta^2 = 0.901$) and interaction ($F(1, 14) = 45.31$, $p < 0.001$, $\eta^2 = 0.764$), but no effect for mode was observed. *Post hoc* analyses revealed that TMSK (95% CI: -1.735 to -0.2151 , $p = 0.012$) and STC (95% CI: -4.617 to -3.098 , $p < 0.001$) significantly decreased serum testosterone during competition/simulation, and TMSK further significantly reduced at end-comp (95% CI: -4.423 to -0.2792 , $p = 0.024$) (Figure 5A). After competition/simulation, there was no effect of time, mode, or interaction in serum cortisol (Figure 5B). For the C/T ratio, there was an effect for time ($F(1, 14) = 9.417$, $p = 0.006$, $\eta^2 = 0.402$), mode ($F(1, 14) = 5.701$, $p = 0.032$, $\eta^2 = 0.347$), and interaction ($F(1, 14) = 5.770$, $p = 0.031$, $\eta^2 = 0.29$). *Post hoc* analyses indicated that STC significantly increased C/T ratio (95% CI: 2.958 – 13.88 , $p = 0.003$), and the C/T ratio was significantly higher in STC than TMSK at End-comp. (95% CI: 2.362 – 13.44 , $p = 0.004$) (Figure 5C).

DISCUSSION

The aim of the present research was to compare the effects of TMSK and TKD competition matches on physiological challenges, exercise-induced muscle damage, and hormonal responses in elite combat TKD athletes. To our knowledge, this study is the first to compare the effect of kicking simulation and simulated competition using an Olympic competition time frame on physiological challenges. The primary findings of this study were that (1) both TKD competition (STC) and kicking simulation (TMSK) were high-intensity intermittent exercise and elicited comparable challenges in heart rate, perceived exertion, and lactate responses; (2) compared with TMSK, the STC produced markedly greater muscle damage and inflammatory responses in the Olympic competition time frame; (3) furthermore, the

STC provokes a relatively stronger systemic catabolic stress, reflected by an increase in the ratio of cortisol to testosterone after the consecutive combat matches, whereas there were no such response observed in TMSK group. Here, we demonstrated that the match-simulating kicking practice was capable of eliciting comparable physiological challenges as that of TKD competition, and no obvious exercise-induced muscle damage was observed. However, we found significant differences in systemic inflammatory and catabolic stresses between the combat and simulated kicking exercise models. Our present findings, therefore, provide a better understanding of the physiological diversities between match-based simulation training and combat competition for Olympic Taekwondo preparation.

Physiological Characteristics of TMSK and STC

The data of physiological responses demonstrated that TMSK elicited a similar heart rate, perceived exertion rating, and lactate response compared to the STC (Figures 2A–C), indicating that both TMSK and STC can be recognized as high-intensity intermittent exercise (HIIE). Our results thus suggest that the match-simulated kicking exercise (TMSK) was capable of imitating and reconstructing physiological challenges and energy demands during TKD combat in an Olympic competition time frame. To our knowledge, this is the first study using a simulated kicking model combined with a schedule containing consecutive competitions (four simulations/matches in one day) alongside techniques for investigating relevant physiological responses according to Olympic TKD regulations. Previous studies regarding TKD training or competition are mostly based on technical/tactical training, physiological responses, or nutrient supplementations applying 1–4 simulation or combat matches (Bridge et al., 2009; Bridge et al., 2011; Casolino et al., 2012; Lopes-Silva et al., 2015; Chou et al., 2018), but only a few of these studies used the exact time frame of current Olympic competition regulations to investigate the changes in the relevant physiological biomarkers (Hausen et al., 2017). Some discrepant results from different studies might be explained by the amount of simulation/competition and

different WRRs. Particularly, in the present study, we used here a longer period of simulation kicks with 1:1 WRR (attack time lasted for 10 s), which might result in higher accumulative physiological stress on athletes. In addition, the current Olympic TKD regulations have continued to be modified and updated (e.g., duration of combat round and between-round rest, contest area specifications, scoring regulations, etc.) (World Taekwondo, 2018), thus the investigations of physiological responses during combat using the most updated international rules is warranted for assisting in the preparation of a development plan for this sport.

In the Olympics, athletes must perform 4–5 consecutive competitions in 1 day to proceed to the final competition, and a classical match includes 3–4 rounds with 1-min intervals (i.e., three regular rounds plus one golden round if necessary) (World Taekwondo, 2018). Following the Olympic competition time frame, we herein observed that the heart rate, RPE, and blood lactate level were dramatically increased from Round 1 to Round 3 during each match in both STC and TMSK modes (maximal HR ranged from 181 to 195 bpm; maximal RPE ranged from 18.8 to 19.4; maximal lactate ranged from 8.94 to 10.95 mM), indicating that the physiological stresses also increased progressively across matches in both experimental modes. Our findings indicate that the TMSK-induced HR, PRE, and lactate responses are comparable to our present simulation combat mode (**Figures 2A–C**) and previous TKD studies using simulation combats (Campos et al., 2012; Lopes-Silva et al., 2015; Hausen et al., 2017). For example, the peak heart rate and peak blood lactate gradually increased from 181 to 189 bpm and from 8.4 to 12.3 mM after each round during a consecutive 3-round simulation TKD combat, respectively (Hausen et al., 2017). Furthermore, Lopes-Silva et al. (2015) also reported a gradually increasing peak heart rate and RPE from 177 to 188 bpm and from 14 to 18, respectively. These findings suggest that TMSK elicited adequate physiological challenges to the extent of simulated combat competitions. However, the peak heart rate (increasing from 175 to 187 bpm) and blood lactate (increasing from 7.5 to 11.9 mM) responses appear to be slightly lower in competitions (Bridge et al., 2009) than those of simulated combat. One possible explanation for the differences between combat simulation and real competition might be the preparation time and exchange time/preparation time ratio (ET:PT ratio) (Bridge et al., 2009; Hausen et al., 2017). In combat simulation, athletes spend less time in preparation, and the ET:PT ratio is also lower, indicating that athletes may attack more in the simulation. In addition, the slightly higher physiological stress (heart rate, RPE, and blood lactate) may be contributed by the 4-match simulation of 1:1 WRR used in present study, as previous studies used from 1 to 3 matches with different WRRs. Consequently, in the present study, TMSK is capable of providing comparable similarities in intensity and physiological stress as STC in terms of international-level TKD combats.

TKD Competition Induced More Tissue Damages Than Simulated Kicking Exercise

In competition, in addition to attacks, the TKD athletes themselves must also perform defensive skills and dodge, thus

TKD athletes need to use some parts of the body to resist the opponent's attack to prevent the opponent from scoring. Both attacking and resisting cause the body to hit a hard part or be hit by a hard part (joints, bones, etc.), thus causing more marked tissue damages. As a consequence, the nature of TKD combat may result in server muscle damage from the direct contacts during the matches (Bridge et al., 2009; Campos et al., 2012; Kwok, 2012). Singh et al. compared the effects of physical contact-induced muscle soreness on exercise performance during team sports, and they found that impact contact caused greater negative effects on subsequent performance with even stronger muscle discomfort/damage (Singh et al., 2011). In the present study, we observed that, compared to STC, the exercise-induced increasing myoglobin and CK were significantly lower in TMSK, suggesting that the simulated kicking did not provoke the same degree of muscle damage as TKD competition under comparable exercise intensity (**Figures 3A,B**). According to our present findings, the greater muscle damage in STC is probably due to direct physical contacts during attack and defense. Our observations are in line with the study by Takarada et al. that the blood muscle damage markers (myoglobin and CK) are closely associated with the number of tackles in a competitive rugby match (Takarada, 2003). Furthermore, mechanical trauma during exercise such as repeated physical impacts occurring at long-distance running is a major cause of intravascular hemolysis (Miller et al., 1988; Janakiraman et al., 2011). Although the frequency of direct physical contact during TKD combat may not be as high as long-distance running, each impact by defending or attacking during TKD combat can be much greater than each foot impact during long-distance running, which might lead to a substantial increase in intravascular hemolysis in STC group (**Figure 3D**). These results therefore suggest that, in the present study, the soft-target kicking simulation exercise yielded relatively lower tissue damages (i.e., muscle damage and hemolysis) compared with TKD matches, which might thereby prevent the increase in exercise-induced inflammation.

Diverse Responses of Systemic Inflammation and Catabolic Stress Between TMSK and STC

Numerous studies have revealed that exhaustive exercise or eccentric exercise can lead to myofibrillar disruptions and initiate the inflammatory response (Tidball, 1995; Suzuki et al., 1999; Nosaka et al., 2002; Sayers and Clarkson, 2003). Neutrophils are rapidly infiltrated into damage tissue after exercise-induced muscle damage and evoke initial local inflammation (Kanda et al., 2013). Moreover, the NLR and PLR are widely applied to reflect the systemic inflammatory status in athletes and patients (Turkmen et al., 2013; Lou et al., 2015; Liao et al., 2016; Chen et al., 2017). In this study, we found the NLR and PLR were significantly higher in STC than in TMSK following the consecutive tasks, indicating that the STC provoked even greater systemic inflammatory responses (**Figures 4D,F**). In addition, we also observed that the STC but not TMSK significantly decreased the number of circulating lymphocytes

(**Figure 4C**). This was in agreement with previous studies that the lymphocyte cell viability markedly decreased after exercise (Pedersen and Toft, 2000; Fisher et al., 2011). It has to be noted that only STC provoked a marked increase in muscle damage, hemolysis, and inflammatory responses after successive tests (**Figures 3A,B**), which is consistent with the finding by Singh et al. that only direct full contact produced a significant inflammatory response during exercise (Singh et al., 2011). As a result, these data support the postulation that the lymphocytes might be more susceptible to cytotoxic agents released from damaged muscle during post-exercise recovery, thereby changing the balance of immune cells' profiles. However, the precise physiological mechanisms underlying the inflammatory responses to the consecutive full-contact taekwondo combat warrant further investigations.

On the other hand, the ratio between cortisol and testosterone (C/T ratio), the two primary catabolic and anabolic hormones, has been used to determine the degree of systemic catabolic stress in response to exercise (De Luccia, 2016). Here we observed that the C/T ratio dramatically increased following consecutive TKD competitions, but the C/T ratio was sustained at a relatively lower level in participants with simulated intermittent kicking exercise (**Figure 5C**). The present finding revealed clear accumulative systemic catabolic stress caused by the consecutive TKD competitions but not by the simulated kicking exercise. Corresponding to previous studies (Suzuki et al., 1999; Bishop et al., 2003; Davison and Gleeson, 2006), the circulating ratio between cortisol and testosterone markedly rose in response to strenuous exercise. Additionally, the circulating acute muscle damage biomarkers have been reported to be associated with catabolic endocrine profiles in team sport competitions (McLellan et al., 2010; Thorpe and Sunderland, 2012), therefore the elevated muscle damage appears to, at least in part, account for the STC-induced catabolic state after the consecutive TKD combats.

In this study, although our present findings revealed similar physiological and subjective responses (e.g., heart rate, lactate, and RPE) to the exercise loads between TMSK and STC modes, we are aware that other studies have done similar work and are based on closer WRRs (e.g., range of 1:3–1:5) of the observed ones in the modality to determine the influence on internal load and neuromuscular load responses in TKD athletes (Matsushigue et al., 2009; Santos et al., 2011; Campos et al., 2012; Del Vecchio et al., 2016; Sant'Ana et al., 2017). Additionally, the heart rate and blood lactate responses seem slightly lower during international competitions (Bridge et al., 2009). Hence, there still exists a certain degree of discrepancy to the external load actually observed during taekwondo competitions in this study, and this limitation might require further that studies apply even closer WRRs to investigate these relevant internal/external parameters using the Olympic time frame.

PRACTICAL PERSPECTIVES

Taekwondo competition results in considerable physiological challenges plus tissue damage (i.e., muscle damage and hemolysis)

through physical contact. Based on the finding that the TMSK mode yields less tissue damage, TKD coaches and conditioning specialists can incorporate TMSK into daily and regular training programs (e.g., strength training or drill/tactical/skill practice) to synergistically optimize the athlete's physical condition and to help athletes to familiarize themselves with competition arrangement. Moreover, the marked increases in muscle damage and catabolic stress remained at high levels for at least 24 h after the competition, indicating that the coaches and sport scientists must take this into consideration for developing proper post-exercise recovery strategies (e.g., nutritional intervention, recovery modality, alternative approach, etc.) for these athletes. Our present data not only deliver the basic information about physical condition following competition but also provide a new conditioning training protocol complying with the Olympic competition time frame.

CONCLUSION

The simulation protocol (TMSK) used in the present study elicited comparable physiological challenges to taekwondo competition but did not induce catabolic stress to the same extent as competition during the consecutive competitions under an Olympic time frame in elite TKD athletes. Although TMSK produced similar heart rate and lactate as real TKD competition, the muscle damage, inflammatory responses, and catabolic stress brought about by the simulated kicking exercise were relatively lower compared to the real TKD competition. The fundamental information provided by the present study may help coaches, athletes, and sport scientists to develop a more realistic TKD-specific training program and post-competition/training recovery strategy for Olympic preparation.

AUTHOR CONTRIBUTIONS

C-CC, Y-HL, and Y-CS conceived and designed the experiments. Y-HL, C-YC, and Y-CS performed the experiments. Y-HL, S-JC, and C-CC analyzed the data. S-JC, Y-CS, Y-HL, and C-YC wrote the paper. Y-HL, C-CC, and Y-CS contributed reagents, materials and analysis tools.

FUNDING

We also thank the partial support provided by the Ministry of Science and Technology (MOST 107-2410-H-227-005-MY2 and MOST 107-2410-H-845-022-), Taiwan. The funding institute played no role in this study and the preparation of manuscript.

ACKNOWLEDGMENTS

We deeply appreciate all the efforts from all the participants in this study.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Nicotine Supplementation Does Not Influence Performance of a 1h Cycling Time-Trial in Trained Males

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OPEN ACCESS

Edited by:

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University of Canberra, Australia

Reviewed by:

Stephen Cheung,
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Andrew Renfree,
University of Worcester,
United Kingdom

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 30 October 2018

Accepted: 06 March 2019

Published: 26 March 2019

Citation:

Mündel T, Houltham SD,
Barnes MJ and Stannard SR (2019)
Nicotine Supplementation Does Not
Influence Performance of a 1h Cycling
Time-Trial in Trained Males.
Front. Physiol. 10:292.
doi: 10.3389/fphys.2019.00292

The use of nicotine amongst professional and elite athletes is high, with anecdotal evidence indicating increased prevalence amongst cycling sports. However, previous investigations into its effects on performance have not used high-validity or -reliability protocols nor trained cyclists. Therefore, the present study determined whether nicotine administration proved ergogenic during a ~1 h self-paced cycling time-trial (TT). Ten well-trained male cyclists (34 ± 9 years; 71 ± 8 kg; $\dot{V}O_{2\max}$: 71 ± 6 ml \cdot kg $^{-1}$ \cdot min $^{-1}$) completed three work-dependent TT following ~30 min administration of 2 mg nicotine gum (GUM), ~10 h administration of 7 mg \cdot 24 h $^{-1}$ nicotine patch (PAT) or color- and flavor-matched placebos (PLA) in a randomized, crossover, and double blind design. Measures of nicotine's primary metabolite (cotinine), core body temperature, heart rate, blood biochemistry (pH, HCO_3^- , La^-) and Borg's rating of perceived exertion (RPE) accompanied performance measures of time and power output. Plasma concentrations of cotinine were highest for PAT, followed by GUM, then PLA, respectively ($p < 0.01$). GUM and PAT resulted in no significant improvement in performance time compared to PLA (62.9 ± 4.1 min, 62.6 ± 4.5 min, and 63.3 ± 4.1 min, respectively; $p = 0.73$), with mean power outputs of 264 ± 31 , 265 ± 32 , and 263 ± 33 W, respectively ($p = 0.74$). Core body temperature was similar between trials ($p = 0.33$) whilst HR averaged 170 ± 10 , 170 ± 11 , and 171 ± 11 beats \cdot min $^{-1}$ ($p = 0.60$) for GUM, PAT, and PLA, respectively. There were no differences between trials for any blood biochemistry (all $p > 0.46$) or RPE with mean values of 16.7 ± 0.9 , 16.8 ± 0.7 , and 16.8 ± 0.8 ($p = 0.89$) for GUM, PAT, and PLA, respectively. In conclusion: (i) nicotine administration, whether via gum or transdermal patch, did not exert an ergogenic or ergolytic effect on self-paced cycling performance of ~1 h; (ii) systemic delivery of nicotine was greatest when using a transdermal patch; and (iii) nicotine administration did not alter any of the psycho-physiological measures observed.

Keywords: smokeless tobacco, stimulant, performance, competitive, athlete, doping, WADA

INTRODUCTION

Presently, the use of nicotine or nicotine-containing substances is not banned by the World Anti-Doping Agency (WADA). Yet, use of nicotine or nicotine-containing substances amongst elite and professional athletes is high and increasing. For example, cross-sectional, self-report data indicate a 25–35% prevalence of smokeless tobacco use, whilst data from anti-doping urine analyses display a detection of nicotine or its metabolites in 23–36% of samples (see Mündel, 2017 for review).

Following this, WADA placed nicotine on its Monitoring Program (World Anti-Doping Agency [WADA], 2012) to further detect patterns of use to determine whether it should be upgraded to the List of Prohibited Substances.

Anecdotal reports indicate an increased prevalence of nicotine use in cycling sports. Through its psychostimulatory and sympathomimetic properties, nicotine exerts psychological and physiological effects that should be nootropic and ergogenic (Mündel, 2017). To date, eight studies have assessed performance using cycling protocols in response to consumption of nicotine or smokeless tobacco. Three studies identified an ergogenic effect (Mündel and Jones, 2006; Johnston et al., 2018; Zandonai et al., 2018) whilst the remaining five found no effect, ergogenic or ergolytic (Baldini et al., 1992; Pysny et al., 2015; Fogt et al., 2016; Zandonai et al., 2016; Mündel et al., 2017). However, the protocols used have minimal validity (time-to-exhaustion, 30 s Wingate, and incremental maximal tests) and together with the untrained, non-cyclist cohorts used reduce the reliability of these performance tests, thereby limiting the smallest worthwhile effect that can be detected (i.e., sensitivity; see Currell and Jeukendrup, 2008 for review).

The 40 km time-trial (TT) in cycling is often viewed as the blue riband event, featuring in national, international (e.g., Grand Tours), world and Olympic championships. Coyle et al. (1991) demonstrated that in well-trained and familiarized male cyclists, 40 km TT performance is highly correlated to a self-paced, 1-h laboratory cycle ergometer test. Furthermore, this simulated cycling TT has demonstrated high reliability, especially when trained, and familiarized cyclists are used as participants (Jeukendrup et al., 1996). Therefore, the primary purpose of the present study was to determine whether nicotine proved ergogenic when using a protocol and participants that have demonstrated high validity and reliability, in order to be able to translate these results to competitive cyclists and other endurance athletes.

Nicotine is delivered via different routes using a variety of products, which apart from affecting ease-of-use, can result in different nicotine bioavailability and pharmacokinetics (Mündel, 2017). Therefore, over-the-counter products such as nicotine gum, transdermal patches, inhalers and sublingual tablets will vary in their delivery of nicotine and their subsequent systemic effect due to differences in absorption etc. (Mündel, 2017). For example, use of nicotine gum results in an earlier but lower peak blood concentration of nicotine than a transdermal patch, with the former more appropriate for an acute delivery of nicotine (Mündel, 2017). To our knowledge, no previous investigation has determined any differential effect between nicotine delivery systems on exercise performance. Therefore, this was the secondary purpose of the present study.

MATERIALS AND METHODS

Ethics Statement

The study was approved by the Central Regional Health and Disability Ethics Committee (CEN/08/09/056), and conformed to the standards set by the latest revision of the *Declaration*

of Helsinki, except for registration in a database, with each participant providing informed, written consent.

Participants

Ten well-trained male cyclists (mean \pm standard deviation age: 34 ± 9 years, body mass: 71 ± 8 kg) volunteered to participate in this study. All participants were competing at a club or national level on a regular basis and maintained a weekly training volume of more than 200 km. According to De Pauw et al. (2013), our participants were classified as performance levels 3/4, or a trained/well-trained participant group due to their peak aerobic power (346 ± 46 W and 4.9 ± 0.5 W \cdot kg $^{-1}$) and peak rate of O₂ consumption ($\dot{V}O_2$ peak, 5.0 ± 0.6 L \cdot min $^{-1}$, and 71 ± 6 mL \cdot kg $^{-1}$ \cdot min $^{-1}$). All participants were non-smokers, and did not habitually use any form of nicotine administration.

Experimental Overview

All participants attended the laboratory on five occasions: (1) preliminary submaximal and maximal tests, (2) experimental familiarization, and (3–5) experimental trials. The three experimental trials were completed in a randomized, crossover, double blind design. All visits were separated by 7 days, conducted at the same time of day (± 1 h), following >24 h of dietary and exercise control, with participants also having refrained from alcohol and caffeine during this period. All exercise was on an electromagnetically braked cycle ergometer (Lode Excalibur, Netherlands) with participant-specific set up for the seat, handle bars and pedals, which was maintained constant for each trial within a participant. All testing was conducted in a temperate laboratory environment (18–22°C) with a fan-generated airflow of 19 km \cdot h $^{-1}$ facing participants.

Preliminary Testing and Familiarization

Following body mass (Jandever, Taiwan) and height (Seca, Germany) measurements, participants began a submaximal test that consisted of four consecutive 5-min power outputs: 100, 150, 200, and 250 W, at a self-selected but constant cadence. Following 5 min active recovery and 5 min inactive recovery, a ramp protocol was used to determine $\dot{V}O_2$ peak. Work rate began at 100 W and consisted of a linear increase at 40 W \cdot min $^{-1}$ until volitional fatigue. Expired gases were collected continuously (VacuMed Vista Turbofit, United States) for the determination of ventilation and O₂ uptake ($\dot{V}O_2$). Following this, a linear relationship between the mean rate of $\dot{V}O_2$ during the last 2 min of each submaximal stage and power output was determined and used to calculate a power output which would elicit 80% of $\dot{V}O_2$ peak for each participant for the remaining TTs.

The familiarization trial was undertaken to ensure participants were accustomed to the experimental procedures and to minimize learning effects. This trial replicated entirely the experimental trial outlined below.

Dietary and Exercise Control

Participants were asked to refrain from exercise between 24 and 48 h prior to each experimental trial. Twenty-four hours prior to each experimental trial, participants attended the laboratory

to complete a standardized training ride 60 min in duration at a fixed power output that elicited $\sim 60\%$ $\dot{V}O_2$ peak. Participants were then provided with a standardized snack ($1 \times$ Sanitarium UP&GO, New Zealand: 823 kJ providing 30.3 g carbohydrate, 8.3 g protein, and 3.8 g fat), and recorded their diet during the 24 h period prior to the first experimental trial. This diet was replicated for each subsequent experimental trial, and in order to further minimize variation in pre-trial metabolic state a standardized meal ($1 \times$ Sanitarium UP&GO, New Zealand: 823 kJ providing 30.3 g carbohydrate, 8.3 g protein and 3.8 g fat, and $1 \times$ One Square Meal, New Zealand: 1450 kJ providing 45.1 g carbohydrate, 8.4 g protein, and 11.7 g fat) was consumed 3 h prior to arriving at the laboratory for the experimental trial, after which no food was consumed. Fluid was encouraged and *ad libitum* until 3 h prior to the experimental trial.

Nicotine/Placebo and Temperature Pill Administration

Approximately 10 h prior to each experimental trial, a staff member not involved with the research project placed a patch on the participant between the right shoulder blade and the spine. The patch was either a nicotine patch (7 mg 24 h^{-1} , Habitrol, Novartis, New Zealand) or a placebo patch (orthoptic eye patch 63.5 mm \times 45.7 mm, Nexcare, 3M, New Zealand). Participants were also given a factory-calibrated temperature-sensing radio pill (CorTempTM, HQ Inc., United States) to ingest at this time. For most, this occurred ~ 1 h before each participant went to bed. Approximately 10 h later, at 40 min prior to the beginning of the trial, the same independent staff member handed participants a piece of gum to chew for 30 min. The gum was either nicotine gum (Nicorette 2 mg, Johnson & Johnson, New Zealand) or a placebo gum (Juicy Fruit, Wrigley Corp, IL, United States). Participants were asked to chew the gum as directed by the manufacturer; briefly, this involved participants chewing the gum until the flavor became strong (~ 1 min), then placing against the cheek until the flavor disappeared (~ 2 min). This process was repeated until 30 min had elapsed. Participants were not aware of the research hypotheses, and were informed that the purpose of the study was to investigate the timing of nicotine administration, hence they would be administered three of the following four options: (i) PAT-GUM, PAT-PLA, PLA-GUM, PLA-PLA. Following the third experimental trial, participants were fully de-briefed. The independent staff member was only aware that they were administering intervention A (PAT), B (PLA patch), C (GUM), or D (PLA gum) with results remaining blinded to the authors until data collection was complete, after which disclosure was made.

Experimental Procedure

Following the pre-trial control described above, participants arrived at the laboratory and were checked that they still had the radio pill in their gastro-intestinal tract. A blood sample was obtained from the antecubital vein (see below), following which participants changed into their cycling shorts and top, shoes and socks. They then received their chewing gum and rested seated for 30 min before another blood sample was

obtained. Participants then completed 3 min cycling at each of 100, 150, and 200 W, to allow sufficient warm-up. Immediately on completion of the 200 W bout, the ergometer was set to linear mode based on the formula of Jeukendrup et al. (1996), where participants were required to complete an individualized set amount of work (996 ± 132 kJ) as quickly as possible, which was calculated as the equivalent of 60 min of cycling at 80% $\dot{V}O_2$ peak. Participants were notified of their progress at each 20% of the total work completed, with no other feedback provided. A 7% glucose polymer drink was provided to the participants at a rate of 100 ml every 20% of work completed and was required to be ingested within the time taken to complete 20% of work; this drink minimized the likelihood of dehydration or hypoglycemia influencing the results, and mimics competition. Immediately following the self-paced TT, participants began a 5-min cool-down (100 W) before a final blood sample was obtained.

Measurements taken during the final 2 min of each 20% work completed included heart rate (Polar Vantage XL, Polar Electro), gastro-intestinal body temperature (T_{gi}), Borg's rating of perceived exertion (RPE) measured using the 15-grade scale, from 6 to 20 (Borg, 1970), and work completed.

Blood Sampling and Analyses

Venous blood samples were obtained from an antecubital vein into two vacutainer tubes (Becton-Dickinson, Plymouth, United Kingdom), one 4 ml containing lithium heparin and one 4 ml containing clot activator. Following inversion, the tube containing clot activator was allowed to clot at room temperature for 30 min before being centrifuged (Eppendorf, Hamburg, Germany) at 4°C for 10 min at 805 g. Serum was removed, aspirated into 500 μl aliquots and frozen at -80°C for later analyses using high-performance liquid chromatography (HPLC). The tube containing lithium heparin was analyzed within 30 min for determination of pH, bicarbonate and lactate via an automated analyzer (Radiometer, Brønshøj, Denmark).

Due to nicotine's tendency to fluctuate and relatively short half-life, cotinine, its major metabolite with a longer retention time is preferred, especially for anti-doping purposes (Dhar, 2004; Mündel, 2017). Sample preparation, extraction and analysis by HPLC were based on previous methodology (Massadeh et al., 2009) and performed in duplicate. The HPLC system (Shimadzu Prominence 20 Series) consisted of a DGU-20AS Prominence degasser, SIL-20AC Autosampler, SPD-M20A Diode array detector and a CTA-20A column oven with a Phenomenex Luna 5 μ C18 (2) 100A 150 mm \times 4.6 mm column attached. Operating conditions were as per the method used by Massadeh et al. (2009) except for the column, with a limit of detection for cotinine of $7.8\text{ ng} \cdot \text{mL}^{-1}$.

Data and Statistical Analyses

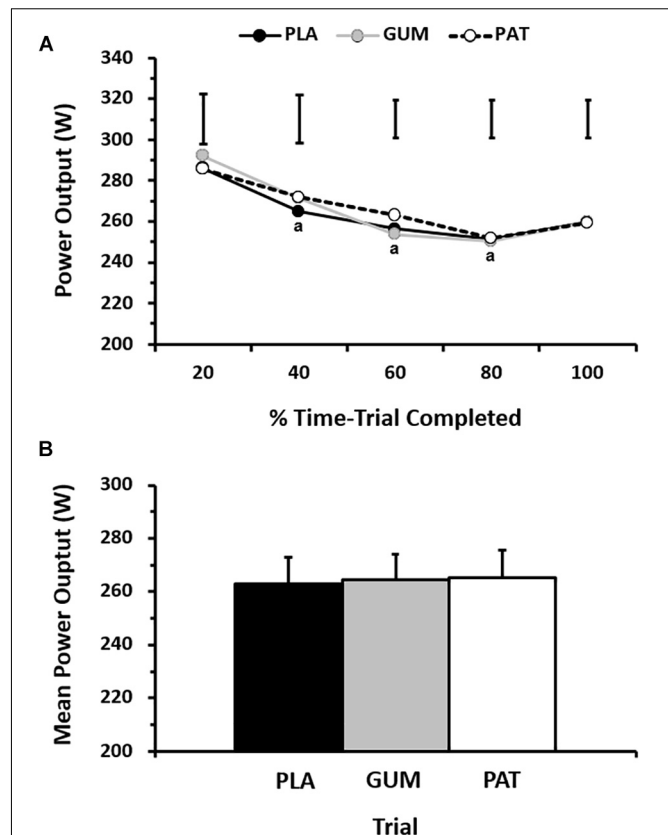
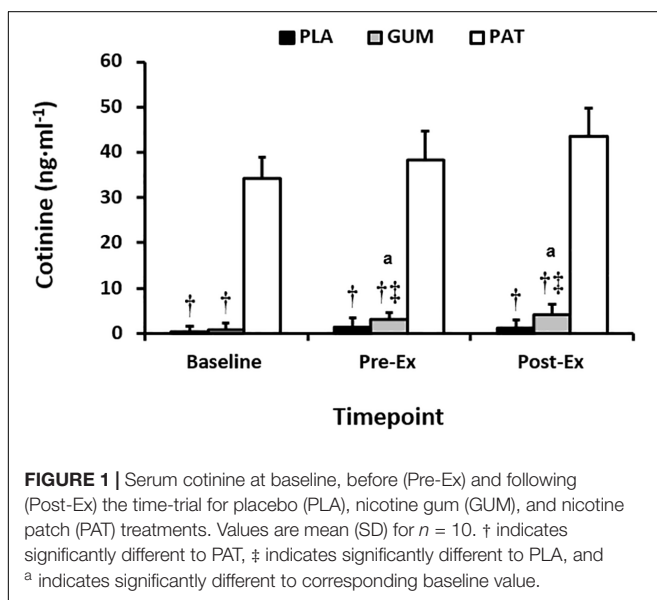
All descriptive and statistical analyses were performed with SPSS software for windows (IBM SPSS Statistics 20, NY, United States). Descriptive values were obtained and reported as means and standard deviation (SD) unless stated otherwise. Levene's test was used to ensure data did not differ substantially

from a normal distribution. Data were analyzed using two-way (treatment \times time) ANOVA for repeated measures. Sphericity was assessed and where the assumption of sphericity could not be assumed, adjustments to the degrees of freedom were made ($\epsilon > 0.75$, Huynh-Feldt; $\epsilon < 0.75$, Greenhouse-Geisser). Where main or interaction effects occurred, *post hoc* pairwise analyses were performed using a paired samples *t*-test (Bonferroni correction where relevant), with statistical significance set at $P \leq 0.05$. Partial eta-squared (η_p^2) is reported as a measure of effect size, with demarcations of small (<0.09), medium (>0.09 and <0.25), and large (>0.25) effects, respectively (Cohen, 1988). This combination of statistical significance and effect size provided an indication of the likelihood of committing a Type I (i.e., $P \leq 0.05$ but $\eta_p^2 < 0.09$) or II (i.e., $P < 0.10$ but $\eta_p^2 > 0.25$) error. The typical error of measurement as a coefficient of variation (CV) between trials was calculated according to Hopkins (2000). Finally, we sought to determine whether [cotinine] was associated with exercise performance and body mass, using Pearson's correlation coefficient to describe the form and strength of bivariate association for absolute values.

RESULTS

Treatment Verification

Plasma cotinine concentrations can be seen in **Figure 1**. Main effects of treatment ($p < 0.01$, $\eta_p^2 = 0.85$) and time ($p = 0.03$, $\eta_p^2 = 0.31$) but no interaction ($p = 0.27$, $\eta_p^2 = 0.14$) were observed, such that the magnitude in concentrations were attained in the following order: PAT > GUM > PLA, and concentrations increased above baseline for GUM whilst concentrations remained constant for PAT and PLA. Participants reported no adverse effects with overnight exposure to nicotine via the transdermal patch or through chewing gum.



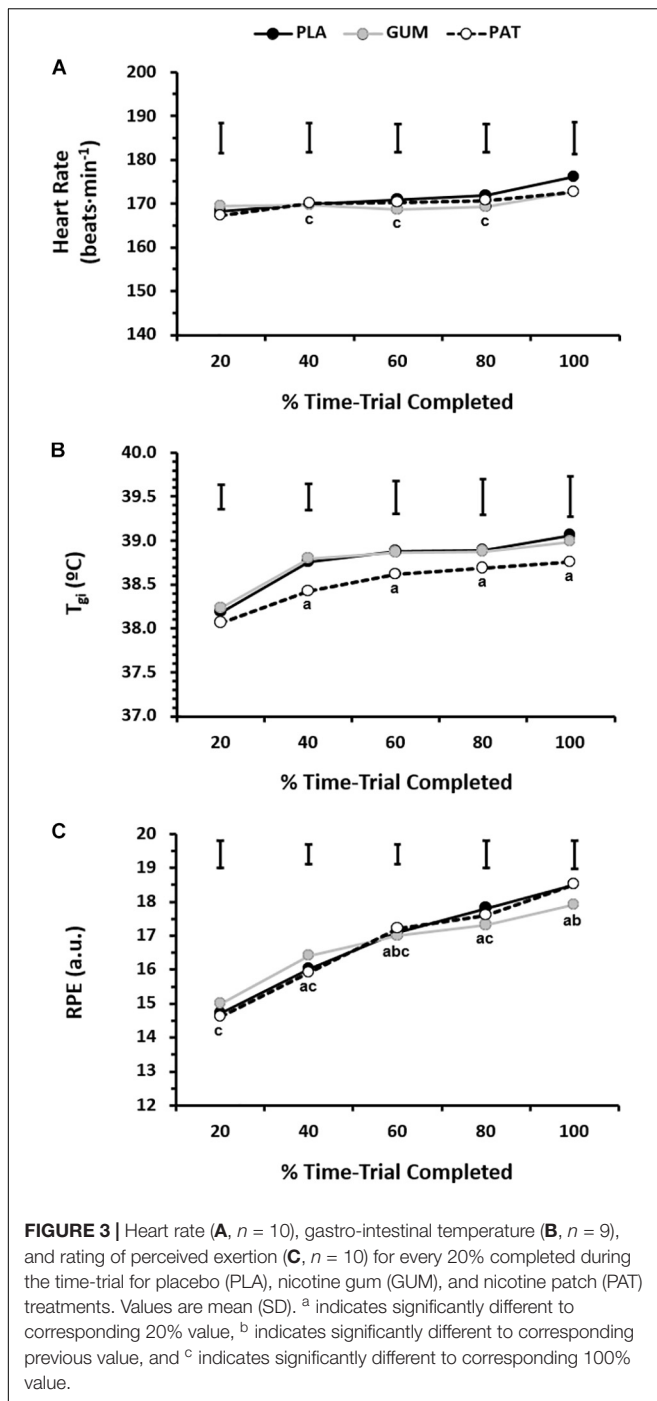
Time-Trial Performance

Mean power output between treatments and the profile over time can be seen in **Figure 2**. The self-paced power output profile was similar between treatments ($p = 0.71$, $\eta_p^2 = 0.04$) but changed over time ($p < 0.01$, $\eta_p^2 = 0.65$) such that power output decreased with time before a characteristic end-spurt; however, this was not dependent on treatment (interaction: $p = 0.27$, $\eta_p^2 = 0.15$). Time to complete the set work was similar between treatments ($p = 0.73$, $\eta_p^2 = 0.03$) with performance times of 63.3 ± 4.1 min, 62.9 ± 4.1 min, and 62.6 ± 4.5 min for PLA, GUM and PAT, respectively. This corresponded to mean power outputs of 263 ± 33 , 264 ± 31 , and 265 ± 32 W, respectively ($p = 0.74$, $\eta_p^2 = 0.03$).

When viewing the performance trials by completion order, the typical error of measurement as a CV between trials was $2.5 \pm 1.2\%$. By comparison, the change in performance time due to treatments was $-0.6 \pm 4.4\%$ (GUM) and $-1.0 \pm 4.8\%$ (PAT).

Physiological and Perceptual Responses

The responses for heart rate, core temperature and perceived exertion can be seen in **Figure 3**. The heart rate response was similar between treatments ($p = 0.60$, $\eta_p^2 = 0.06$) but changed over time ($p = 0.01$, $\eta_p^2 = 0.51$) such that heart



rate was maintained (~ 170 beats \cdot min $^{-1}$) until it increased (~ 174 beats \cdot min $^{-1}$) with the end-spurt; however, this was not dependent on treatment (interaction: $p = 0.19$, $\eta_p^2 = 0.16$). The core temperature response was similar between treatments ($p = 0.32$, $\eta_p^2 = 0.15$) but changed over time ($p = 0.01$, $\eta_p^2 = 0.57$) such that core temperature increased until 40% before reaching a relative plateau, with end-exercise values of $38.9 \pm 0.7^\circ\text{C}$; however, this was not dependent on treatment (interaction: $p = 0.72$, $\eta_p^2 = 0.05$). The RPE was similar between treatments

($p = 0.89$, $\eta_p^2 = 0.01$) but changed over time ($p < 0.01$, $\eta_p^2 = 0.77$) such that RPE increased progressively from 14.7 ± 1.1 (a.u.) to 18.3 ± 1.2 (a.u.); however, this was not dependent on treatment (interaction: $p = 0.15$, $\eta_p^2 = 0.17$).

Blood Biochemical Responses

The responses for pH, bicarbonate and lactate can be seen in Table 1. The pH was similar between treatments ($p = 0.99$, $\eta_p^2 < 0.01$) but changed over time ($p < 0.01$, $\eta_p^2 = 0.73$) such that pH was decreased following exercise; however, this was not dependent on treatment (interaction: $p = 0.80$, $\eta_p^2 = 0.05$). The bicarbonate response was similar between treatments ($p = 0.46$, $\eta_p^2 = 0.09$) but changed over time ($p < 0.01$, $\eta_p^2 = 0.95$) such that bicarbonate was decreased following exercise; however, this was not dependent on treatment (interaction: $p = 0.78$, $\eta_p^2 = 0.05$). The lactate response was similar between treatments ($p = 0.89$, $\eta_p^2 = 0.01$) but changed over time ($p < 0.01$, $\eta_p^2 = 0.90$) such that lactate decreased from baseline to pre-exercise and then increased following exercise; however, this was not dependent on treatment (interaction: $p = 0.36$, $\eta_p^2 = 0.12$).

Correlation Analyses

In absolute terms, the [cotinine] correlated with performance time ($r = 0.63$, $p = 0.05$) and body mass ($r = -0.36$, $p = 0.05$) for PAT, but not for GUM ($r = 0.04$, $p = 0.92$ and $r = -0.25$, $p = 0.29$, respectively).

DISCUSSION

The present study sought to determine whether nicotine administration, delivered acutely via gum or more sustained via transdermal patch, proved ergogenic during a 1 h self-paced cycling TT in trained males. We used a protocol and participants that have demonstrated high validity and reliability, thereby maximizing sensitivity, and therefore these results should be applicable not only to competitive cycling but wider endurance sports/athletes. The important results are that (1) nicotine administration, regardless of delivery method, did not exert any effect (beneficial or detrimental) on exercise performance, (2) systemic delivery of nicotine was greater when using a transdermal patch than gum, and (3) nicotine administration did not affect any of the perceptual or physiological measures observed.

Individual but Not Group Performance Is Affected by Nicotine

Several previous studies have observed a performance benefit of 7–17% when nicotine or smokeless tobacco is administered (Mündel and Jones, 2006; Johnston et al., 2018; Zandonai et al., 2018), although others have found no effect (Baldini et al., 1992; Pysny et al., 2015; Fogt et al., 2016; Zandonai et al., 2016; Mündel et al., 2017). Although all of these studies have used cycling protocols (time-to-exhaustion, 30 s Wingate, and incremental maximal tests), these are known to have poor reliability and/or validity and none have used trained cyclists.

TABLE 1 | Measures of venous pH, bicarbonate (HCO_3^-), lactate (La^-) for placebo (PLA), nicotine gum (GUM), and nicotine patch (PAT) treatments.

	PLA			GUM			PAT		
	Baseline	Pre-Ex	Post-Ex	Baseline	Pre-Ex	Post-Ex	Baseline	Pre-Ex	Post-Ex
pH (a.u.)	7.57 (0.07)	7.59 (0.08)	7.49 (0.06) ^{ab}	7.58 (0.08)	7.59 (0.05)	7.50 (0.09) ^{ab}	7.56 (0.08)	7.58 (0.07)	7.51 (0.06) ^{ab}
HCO_3^- (mmol · l ⁻¹)	29.8 (2.3)	29.1 (1.1)	21.1 (2.4) ^{ab}	29.6 (2.2)	30.0 (1.9)	20.9 (3.3) ^{ab}	30.6 (2.5)	30.5 (2.6)	22.0 (2.9) ^{ab}
La^- (mmol · l ⁻¹)	1.3 (0.9)	1.1 (0.2) ^a	5.0 (1.6) ^{ab}	1.1 (0.5)	0.9 (0.3) ^a	5.4 (1.8) ^{ab}	1.2 (0.3)	1.1 (0.5) ^a	4.9 (1.7) ^{ab}

Values are mean (SD) for $n = 9$.

^aSignificant difference to corresponding Baseline time-point.

^bSignificant difference to corresponding Pre-Ex time-point.

In the present study the typical error of measurement (CV) between trials was ~3%, whilst the changes in performance attributable to either nicotine intervention was $\leq 1\%$. At an individual level, nicotine improved performance times nine times (GUM: $-5.6 \pm 0.8\%$, PAT: $-4.8 \pm 3.3\%$) compared to a detriment eleven times (GUM: $+2.8 \pm 1.3\%$, PAT: $+2.8 \pm 2.1\%$) when compared with PLA. Furthermore, nineteen of the twenty intervention trials (95%) resulted in parallel treatment outcomes i.e., both nicotine treatments collectively increased or decreased performance in the same individuals. Thus, our results indicate that in well-trained cyclists the effect of nicotine on performance is dichotomous with the effect direction dependent on the individual.

Route of Nicotine Administration Affects Systemic Delivery

We observed that the magnitude of systemic nicotine delivery, as measured by nicotine's major (70–80%) metabolite cotinine, was a function of route of administration (Figure 1). Whilst this is consistent with the known absorption pharmacokinetics and bioavailability of buccal versus the more sustained transdermal administration (Mündel, 2017), it is surprising that concentrations were so low with GUM (mean $< 5 \text{ ng} \cdot \text{ml}^{-1}$). We (Mündel et al., 2017) and others (Johnston et al., 2018) have reported cotinine concentrations of 10–45 $\text{ng} \cdot \text{ml}^{-1}$ following administration via chewing 2 mg gum for 20 min or dispersible 5 mg sublingual strips, respectively. In the present study, of the 90 serum cotinine sample results returned, 16 (18%) were values below the limit of detection i.e., $>0 < 7.8 \text{ ng} \cdot \text{ml}^{-1}$, the vast majority occurring during GUM. Consequently, it appears as though buccal absorption was not maximized and thus nicotine-rich saliva was swallowed, with subsequent first-pass metabolism. Therefore, we cannot exclude the possibility that the pharmacologic effects varied between PAT and GUM. Another explanation could be an insufficient time for the conversion of nicotine to cotinine, however this appears less likely due to the known rates of absorption and metabolism (see Benowitz et al., 2009).

Many over-the-counter products (especially pharmaceutical), require consideration whether a fixed or individualized (e.g., to body mass) dose should be administered. Results from the present study support an individualized approach if a systemic concentration determines the resultant pharmacologic effect, as lower [cotinine] was correlated with a higher body mass. Importantly, however, absolute [cotinine] correlated with

absolute performance time, indicating that high(er) systemic concentrations of nicotine resulted in an *impaired* performance. At low(er) doses nicotine proves nootropic/ergogenic whilst at high(er) doses it does not (Perkins et al., 1994; Poltavski et al., 2012; Mündel et al., 2017), this dose-response relationship due to nicotine's stimulant (low-dose), and depressant/relaxant (high doses) effects (Ashton and Stepney, 1982; Lester et al., 1988).

Nicotine Does Not Influence Perceptual and Physiological Responses

Nicotine exerts psychostimulatory effects via increased mesolimbic dopamine, and a sympathoadrenal effect through release of the catecholamines (Mündel, 2017). However, no effects of either nicotine treatment were observed on the physiological and perceptual variables measured in the current study (Figure 3 and Table 1). This may be partly due to the self-paced nature of the exercise protocol, such that these measures reflect the relative effort and intensity of exercise i.e., power output. It has been argued previously (Mündel and Jones, 2006) that when sympathetic output is high during prolonged or high-intensity exercise, the peripheral effects of nicotine might be attenuated and the current results support this. Nevertheless, it can be seen (Figure 3 and Table 1) that the trained participants in the current study were likely close to their maximum capacity; heart rates were maintained high, by the end of exercise perception of effort was close to “Extremely Hard,” considerable hyperthermia was evident despite the temperate environment, and a reduction in bicarbonate due to a lactic acidosis had occurred.

Considerations

Mündel (2017) proposed that in order to better interpret future results on nicotine and smokeless tobacco administration during exercise, a rigorous experimental design, for example a double-blind, placebo-control protocol with manipulation check are necessary. This is only the second study to address these shortcomings (Johnston et al., 2018), whilst the current study is the first to have sufficiently considered criterion validity of the laboratory performance test or how expert performers might respond. No study has investigated how the female response to exercise differs from men when administered nicotine or smokeless tobacco. Given that women metabolize nicotine faster than men, with this further accelerated in those taking

estrogen-containing oral contraception (Benowitz et al., 2006), this provides a worthwhile avenue for investigation.

Many competitive sporting events, especially endurance-related, take place in warm-to-hot environments/climates. As a systemic vasoconstrictor nicotine causes cutaneous vasoconstriction, decreased skin temperature, and systemic vasoconstriction (Roth et al., 1944; Eckstein and Horsley, 1960; Benowitz et al., 1982). When combined with exercise this raises a safety concern for its use during exercise/sport with heat stress where cutaneous vasodilation and sweating are the primary routes of heat loss, potentially placing athletes at greater risk of developing a heat illness. This warrants further investigation, particularly as participants in the current study reached a T_{gi} of $\sim 39^{\circ}\text{C}$ in a temperate environment.

Finally, it is worth considering the anti-doping stance. The half-life of nicotine is 1–2 h, which is why cotinine is favored as a biomarker for nicotine intake, particularly as urine samples, as its metabolism is far slower than nicotine (half-life of ~ 16 h) with reduced daily fluctuation (Benowitz et al., 2009; Mündel, 2017). Therefore, it would be worthwhile comparing the detection of this WADA-monitored substance during the peri-exercise period between blood (serum) and urine indices.

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AUTHOR CONTRIBUTIONS

TM conceived the work, obtained the funding, analyzed and interpreted the data, and wrote the first draft of the manuscript. TM, SH, and SS designed the work. All authors acquired the data and contributed to manuscript revision, read, and approved the submitted version.

FUNDING

This study was funded by grants from the World Anti-Doping Agency (09D8TM) and Massey University Research Fund (RM13226). SH was supported by a Vacation Studentship Award from The Physiological Society.

ACKNOWLEDGMENTS

Partial results of this study were presented as a poster at the 57th Annual Meeting of the American College of Sports Medicine in Baltimore, MD, United States. The authors would like to thank Michelle McGrath for performing magic with the HPLC analysis.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Keeping Athletes Healthy at the 2020 Tokyo Summer Games: Considerations and Illness Prevention Strategies

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Keeping athletes healthy will be important for optimal athletic performance at the 2020 Tokyo Summer Olympic and Paralympic Games. Athletes will be exposed to several stressors during the preparatory and competition phases of the Summer Games that have the potential to depress immunity and increase illness risk. This mini-review provides an overview on effective and practical stressor-specific illness prevention strategies that can be implemented to maintain and protect the health of Olympic and Paralympic athletes.

OPEN ACCESS

Edited by:

Glen Davison,
University of Kent, United Kingdom

Reviewed by:

Arwel Wyn Jones,
University of Lincoln, United Kingdom
Helen Hanstock,
Mid Sweden University, Sweden

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 03 November 2018

Accepted: 27 March 2019

Published: 17 April 2019

Citation:

Keaney LC, Kilding AE, Merien F
and Dulson DK (2019) Keeping
Athletes Healthy at the 2020 Tokyo
Summer Games: Considerations
and Illness Prevention Strategies.
Front. Physiol. 10:426.
doi: 10.3389/fphys.2019.00426

Keywords: Olympic, Paralympic, illness, strategies, health, stressors

INTRODUCTION

Acute illness is one of the single biggest factors that can prevent athletes from successful performance at pinnacle events (Ray Smith and Drew, 2016). Upper respiratory tract symptoms (URTS) are the most common illness reported by elite athletes at the Olympic and Paralympic Games (Derman et al., 2013, 2017; Engebretsen et al., 2013; Soligard et al., 2017). Common URTS include sore throat, headache, runny nose and coughing (Walsh et al., 2011b). URTS can negatively impact training availability, reduce exercise performance, and can even result in athletes missing a major competition (Gleeson and Pyne, 2016). The importance of maintaining athlete health has been highlighted by studies demonstrating that World and Olympic winning medal athletes experience fewer URTS than less successful athletes (Ray Smith and Drew, 2016; Svendsen et al., 2016). As such, keeping athletes healthy in preparatory and competition phases of the 2020 Tokyo Olympic and Paralympic Games (referred to as “Summer Games” in this mini review) will contribute toward optimal performance. The purpose of this mini review is to describe how stressors impact upon an athlete’s risk for illness and to identify which athletes may be more susceptible to illness at the Summer Games, along with outlining strategies to maintain and protect athlete health.

PREPARATORY PHASE OF THE SUMMER GAMES

Keeping athletes healthy in the lead up to the Summer Games is important for optimal performance. In elite track and field athletes, having fewer illnesses (and injuries) and completing more than 80% of planned trainings in the 6-months prior to a major event increases the likelihood of achieving pre-defined performance goals (Ray Smith and Drew, 2016). Nevertheless, maintaining

athlete health during the preparatory phase may prove to be a challenging task, as both Northern and Southern hemisphere athletes will be exposed to different stressors that can challenge immunity and increase URTS risk, including seasonal specific stressors, limited ultraviolet B exposure (UVB), heat acclimation and travel.

Seasonal Specific Stressors

Seasonal Influenza

It is well established that the incidence of influenza exhibit seasonal fluctuations, with peak incidence occurring during the winter months (Doyle and Cohen, 2009). Therefore, compared to Northern hemisphere athletes, athletes residing in the Southern hemisphere will be at an increased risk for infectious URTS episodes during the preparatory phase of the Summer Games.

Strategies to minimize seasonal influenza risk

- **Vaccination:** Advise athletes to have the influenza vaccine in autumn (April) before the influenza season as it usually takes 5–7 weeks to take effect (Schwellnus et al., 2016). Administration of the vaccine should occur during a non-competition period, or at least 2 weeks prior to competition to allow time for the development of a specific adaptive immune response and any potential side effects (Daly and Gustafson, 2011). There may be some benefit in performing moderate intensity exercise prior to vaccine administration as it has been shown to facilitate vaccine efficacy (Edwards et al., 2007, 2012) and reduce adverse reactions (Lee et al., 2018). This adjuvant strategy seems to be most successful in immunocompromised individuals (e.g., elderly) (Ranadive et al., 2014), therefore it is reasonable to suggest it may be worthwhile in elite athletes. It is considered a harmless strategy where the potential benefits could be crucial to preparation, however, further research on athletes is required to determine optimal protocols.
- **Hygiene:** Maintain good hygiene (refer to **Table 1** for guidelines).
- **Illness monitoring:** Monitor illness to enable early detection and application of appropriate illness prevention strategies. Use the Jackson Common Cold Scale (Jackson et al., 1958) to monitor athlete illness. Consider monitoring household illness by adding a question alongside the Jackson Common Cold Scale (Keaney et al., 2018). Household illness monitoring is a promising strategy, although, further research in this area is required.
- **Probiotic supplementation:** Supplement throughout the preparatory phase (3 months) (refer to **Table 1** for further guidelines).
- **Zinc acetate supplementation:** Supplement athletes experiencing acute URTS with zinc acetate lozenges (75 mg/day) to decrease the duration of URTS (Note: Zinc must be taken <24 h after onset of URTS and can be taken for 1–2 weeks) (Maughan et al., 2018). Excessive zinc supplementation (>150 mg/day) should be avoided as it can impair immune cell functions.

Cold Environmental Conditions

Upper respiratory tract symptoms can result from infectious (viral, bacterial, or fungal etiology) or non-infectious and inflammatory (e.g., caused by allergies, asthma and trauma to respiratory epithelial membranes) causes (Gleeson and Pyne, 2016). Southern hemisphere athletes training during winter will be exposed to cold dry air. Inhalation of cold dry air can damage airway epithelium and lead to non-infectious URTS episodes (Koskela, 2007). Athletes with asthma and allergies may be at higher risk for URTS, as winter training has been shown to increase URTS incidence among individuals with these conditions (Hyrkäs et al., 2014; Gleeson and Pyne, 2016).

Strategies to minimize cold air mediated non-infectious URTS

- **Diagnose asthma and allergies:** Administer the validated questionnaire Allergy Questionnaire for Athletes (AQUA) to identify athletes with asthma and allergies (Bonini et al., 2009). Confirm the diagnosis with a physician.
- **Control asthma and allergies:** Ensure appropriate therapeutic control of asthma and allergies and comply to World Anti-Doping Agency (WADA) regulations (Helenius and Haahntela, 2000).
- **Protect airways:** When practical, take extra precautions to avoid inhalation of cold dry air (below 0°C). For example, train indoors or for outdoor training use facial masks to protect airways (Walsh, 2018). It is unknown if facial mask reduce URTS incidence, however, they can attenuate cold air exercise-induced asthma which is known to elicit non-infectious URTS episodes (Beuther and Martin, 2006).

Summer Allergies and Asthma

Northern hemisphere athletes will be exposed to environmental factors and high periods of allergen load during the preparatory phase of the Summer Games, including heat and humidity, pollen, grasses, weed, mold, and dust. During exercise, high ventilation rates combined with increased exposure to environmental factors and allergens can exacerbate asthma and allergies. Prevalence of asthma and allergy is high in elite athletes, especially endurance athletes (Silva and Moreira, 2017). Exacerbations of asthma and allergies may elicit non-infectious URTS, such as runny nose, repetitive sneezing, and coughing, all of which can disrupt training and performance (Gleeson and Pyne, 2016).

Strategies to minimize summer allergy and asthma mediated non-infectious URTS

- **Diagnose and control asthma and allergies** (see section “Strategies to Minimize Cold Air Mediated Non-infectious URTS” for details).
- **Allergen avoidance:** When practical, avoid exposure to allergens (e.g., clean room and change bed lining regularly to reduce house dust mite exposure. Follow pollen forecasts and consider adapting training venues and training schedules during high pollen periods, etc.) (Silva and Moreira, 2017).

Low Ultraviolet B Exposure

Southern hemisphere athletes will be at an increased risk for Vitamin D (VD) deficiency as it is more prevalent during winter when UVB exposure and endogenous synthesis of VD are low (Backx et al., 2017). Previous research suggests that up to 50% of athletes could be considered to have an inadequate VD status, during winter training months (He et al., 2013, 2016). Some Northern hemisphere athletes may also be at risk for VD deficiency, such as indoor athletes, athletes with dark skin tone, athletes who live and train in northern latitudes ($<30^\circ$ or $>70^\circ$) and athletes residing in countries with a poor summer season with limited sun exposure (i.e., sun exposure <20 min/day) (He et al., 2016). VD deficiency appears to be an important determinant of URTS risk in athletes (He et al., 2013, 2016). Evidence suggests an optimal serum 25(OH)D of 75 nmol/L may enhance immunity and prevent URTS (He et al., 2016).

Strategies to Maintain VD Levels

- Vitamin D recommendations for Southern hemisphere and at-risk Northern hemisphere athletes: There may be some benefit in measuring athletes serum 25(OH)D concentration, to allow more targeted VD supplementation. As outlined in recent guidelines, athletes with serum 25(OH)D concentrations <75 nmol L⁻¹ should be supplemented with 2000–4000 IU VD₃/day (Owens et al., 2018). However, the measurement of serum 25(OH)D is not always feasible (cost approximately US\$255 per athlete) and may not be the most appropriate measure of an athlete's VD status (Allison et al., 2018; Owens et al., 2018). Therefore, rather than measuring serum 25(OH)D, the most practical approach may be to supplement all Southern hemisphere and at-risk Northern hemisphere athletes with 1000 IU VD₃/day (comply to WADA anti-doping regulations) (He et al., 2016). There is some risk for toxicity when supplementing with exogenous VD, however, previous reports suggest 1000 IU VD₃/day is a safe dosage (He et al., 2016).
- General VD guidelines for Northern hemisphere athletes: Aim to acquire 15 min of non-protected (i.e., no sunscreen) sun exposure per day (He et al., 2016).

Heat Acclimation

The Summer Games are expected to be hot ($>30^\circ\text{C}$) and humid ($>70\%$ relative humidity). Therefore, heat acclimation (HA) will be an integral component of the preparatory phase. Exercise immunology research suggests that heat does not pose a challenge to the immune system. Indeed, performing a one-off bout of exercise in hot conditions [$28\text{--}38.7^\circ\text{C}$, $45\text{--}76\%$ relative humidity (RH)] does not appear to exacerbate exercise induced immune perturbations, compared to temperate conditions (Mitchell et al., 2002; McFarlin and Mitchell, 2003; Niess et al., 2003; Laing S. et al., 2005; Laing S.J. et al., 2005). Similarly, HA has been shown to have negligible effects on immunity. For example, no change in white blood cell counts (Willmott et al., 2016) or inflammatory cytokines (Amorim et al., 2011; Barberio et al., 2015) has been demonstrated following HA. However,

the current limitation to the HA studies discussed is that illness reports were not measured alongside immune markers. Therefore, it remains unclear how HA may impact upon athletes URTS risk. Consideration may want to be given to the acclimation status of athletes performing HA. During the preparatory phase, it is possible that acclimation status will differ between Northern and Southern hemisphere athletes; athletes residing in the Northern and Southern hemisphere will likely be seasonally acclimated and unacclimated, respectively. Future studies should assess the baseline acclimation status of athletes engaging in HA to understand whether it is associated with URTS risk.

Strategies to Maintain Athlete Health During HA

The health status of athletes should be considered when implementing HA. Athletes experiencing illness symptoms should not participate in HA as it may exacerbate illness (Casadio et al., 2017).

- Hygiene: Maintain good hygiene (e.g., remove wet clothing and have a warm shower immediately following HA sessions) (refer to **Table 1** for further guidelines).
- Training load and recovery management: Heat stress adds to athletes overall training load. Carefully manage training load when training with additional heat (Walsh, 2018). Ensure adequate recovery between HA sessions, particularly if HA protocols involve prolonged exercise (≥ 90 min) as it can cause more severe immune perturbations than shorter duration exercise (<60 min) (Diment et al., 2015).
- Daily wellness monitoring and management: Monitor wellness to understand how individual athletes tolerate HA. A customized psychometric questionnaire (Hooper and Mackinnon, 1995) utilizing Likert scales can be used to assess indicators of wellness (e.g., sleep quality, stress, fatigue, mood, and muscle soreness) (Buchheit et al., 2013; Gallo et al., 2017). In addition to monitoring wellness during the HA period, the questionnaire should be administered during normal training weeks to establish baseline wellness data. High stress/anxiety levels and sleep deprivation have been linked to increased URTS incidence (Cohen et al., 1991, 2009). Over the HA period, apply strategies listed in **Table 1** (i.e., minimize stress and anxiety and improve sleep). If athletes wellness scores are substantially reduced consider adjusting the HA protocol (e.g., reduce load) (Schwellnus et al., 2016).
- Carbohydrate (CHO) intake: Maintain day-to-day CHO availability over HA period, aim for $>50\%$ daily energy intake as CHO (Walsh, 2018).
- Hydration: Permissive dehydration is often used during HA sessions to accelerate the adaptation process (Garrett et al., 2014). Exercising in a dehydrated state does not appear to cause further exacerbation of immune perturbations, compared to euhydrated exercise (Svendsen et al., 2014; Killer et al., 2015). Therefore, permissive dehydration can be used during HA as it is unlikely to impair immunity. However, during recovery from HA, fluid replacement

should be prioritized, as per current rehydration guidelines (Thomas et al., 2016).

- Probiotic supplementation: Begin supplementation at least 2-weeks before the HA block is due to commence and supplement throughout HA (refer to **Table 1** for guidelines).

Long Haul Travel

During the preparatory phase, many Southern and Northern hemisphere athletes will undertake international travel to competition, heat camps and the Summer Games itself. Travel has been identified as a prominent risk factor for URTS. Increased URTS incidence and severity has been demonstrated in team sport athletes traveling to international destinations that were >5 or 11 time zones, respectively (Haywood et al., 2014; Fowler et al., 2016). Similarly, in elite endurance athletes, air travel was found to significantly increase URTS susceptibility (Svendsen et al., 2016). The current limitation to the studies discussed is that immune markers were not measured alongside illness reports. Nevertheless, in the general population simulated long haul travel has been found to induce transient immune changes that may contribute to increased URTS susceptibility (Wilder-Smith et al., 2012). Contrastingly, a recent study in master-level athletes showed that long haul travel did not impair mucosal immune responses (Stevens et al., 2018). Further research on elite athletes is required to elucidate how travel impacts upon the immune system and subsequently URTS risk.

Strategies to Maintain Athlete Health During Long Haul Travel

- Travel vaccines: Consult a physician and update athlete and support staff vaccines.
- Hand hygiene: Apply alcohol-based hand gel after touching potentially contagious objects. For example, hand gel should be used after handling airport plastic security screening trays as a recent study identified that they have the highest frequency of respiratory viruses, compared to other airport surfaces (e.g., toilets, handrails) (Ikonen et al., 2018).
- Avoid ill people: If possible, seat athletes away from ill passengers. Increased risk for infection transmission has been associated with sitting within two rows of a contagious passenger for >8 h (Mangili and Gendreau, 2005). If it is not possible to change seats, athletes should wear a disposable face mask (Walsh et al., 2011a).
- Hydration: Encourage athletes to drink plenty of water to keep well hydrated and potentially prevent mucosal membranes from drying out.
- Optimize sleep hygiene: Pre-departure, improve sleep quantity and quality (refer to **Table 1** for guidelines). After long haul travel, greatest sleep disruption has been reported in the first 48 h (Stevens et al., 2018). Optimize sleep hygiene (e.g., electronic device availability, cool room temperature, caffeine, ear plugs, eye masks, etc.) to improve sleep on the first 2 days after arrival (Stevens et al., 2018).

- Recovery: Avoid flying on the same day as competition or intensive training, delay travel until at least the subsequent day (Svendsen et al., 2016).
- Probiotic supplementation: Begin supplementation at least 2-weeks before scheduled travel (refer to **Table 1** for guidelines).

COMPETITION PHASE OF THE SUMMER GAMES

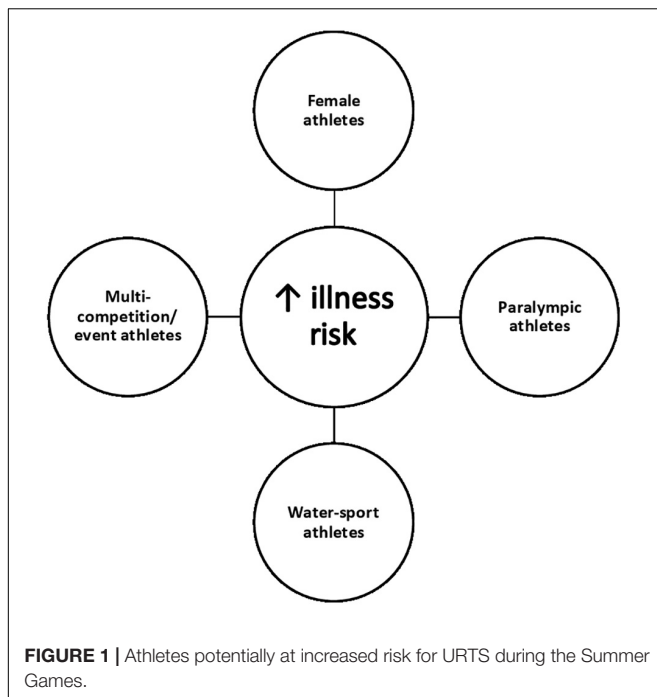
Stressors Associated With the Summer Games

During the Summer Games, both Northern and Southern hemisphere athletes will be exposed to a range of stressors. Such stressors include, intensive competition, hot and humid environmental conditions, dehydration, psychological stress, and sleep deprivation (Keaney et al., 2018; Walsh, 2018). The effect of these stressors on immunity and illness risk has been summarized in recent reviews (Keaney et al., 2018; Walsh, 2018; Williams et al., 2018). Previous studies have tended to examine each stressor in isolation when in reality athletes will be simultaneously exposed to all stressors at the Summer Games. The synergism of these stressors could potentially have a compounding effect on immunodepression, resulting in higher incidence of illness than if each stressor were applied alone. Further research is needed to understand how multiple stressors affect immunity and illness risk.

At the Summer Games, medals are often won by the smallest of margins, so even a mild illness could negatively affect results. To keep athletes healthy and minimize the potential immunodepression evoked by Summer Games stressors, athletes should consider adhering to the five key illness prevention strategies listed in **Table 1**. These strategies have been selected on the assumption that Summer Games athletes will adhere to fundamental principles of nutrition and sport science (e.g., macro- and micro-nutrient intake, hydration, recovery protocols, training load management, etc.). Illness prevention strategies should not replace fundamentals, but work alongside them to keep athletes healthy. In addition to these strategies, other reviews exist which provide detailed recommendations on avoiding infection and maintaining immune health in athletes (Schwellnus et al., 2016; Walsh, 2018).

Athletes at Increased Risk for Illness During the Summer Games

With the aim of protecting the health of athletes, the International Olympic Committee (IOC) monitored illness incidence at the London (2012) and Rio (2016) Olympic and Paralympic Games (Derman et al., 2013, 2017; Engebretsen et al., 2013; Soligard et al., 2017). At previous Summer Games 5–14% of athletes experienced at least one illness, with the highest incidence of illness affecting the respiratory tract (Derman et al., 2013, 2017; Engebretsen et al., 2013; Soligard et al., 2017). IOC reports demonstrated that the illness rates varied considerably between gender and sports



(Derman et al., 2013, 2017; Engebretsen et al., 2013; Soligard et al., 2017). As summarised in **Figure 1**, it appears that some athletes may be more susceptible to illness during the Summer Games, namely: (1) Female athletes; (2) Paralympic athletes; (3) Water-sport athletes; and (4) Multi-competition/event athletes (i.e., athletes who compete on > 1 day) (Derman et al., 2013, 2017; Engebretsen et al., 2013; Soligard et al., 2017).

Female Athletes

Data obtained at the London and Rio Summer Games demonstrated significantly higher (40–60%) illness incidence in female compared to male athletes (Engebretsen et al., 2013; Derman et al., 2017; Soligard et al., 2017). In agreement with these findings, longitudinal studies have shown that female athletes tend to be at increased risk for URTS (Gleeson et al., 2011; He et al., 2014) and experience URTS episodes for a longer duration than male athletes (He et al., 2014). Sex differences in immune variables may explain the higher illness susceptibility observed in female athletes. Differences in immune responses between males and females have largely been attributed to sex hormones and their inherent immune modulatory functions (Klein and Flanagan, 2016). Furthermore, increased URTS susceptibility in female athletes may be associated with low energy availability (LEA). Higher rates of LEA have been demonstrated in female compared to male athletes (Logue et al., 2018), and LEA has been identified as a key risk factor for illness in Olympic-level female athletes (Drew et al., 2017).

Strategies to maintain female athlete health

- **Diagnose and treat LEA:** In the preparatory phase, identify female athletes with LEA using the validated questionnaire Low Energy Availability in Females Questionnaire (LEAF-Q) (Melin et al., 2014). Athletes with LEA should

work closely with a nutritionist to ensure daily energy intake matches training and competition demands (Logue et al., 2018).

- **Supplementation:** At present, a number of supplements have been proposed to alter specific aspects of the immune system and reduce athletes URTS risk (Maughan et al., 2018). However, few supplements have convincing evidence supporting their use. Currently, probiotic (refer to **Table 1** for guidelines), vitamin C (0.25–1.0 g/day) (Hemilä and Chalker, 2013) or quercetin (1 g/day) (Somerville et al., 2016) are the most promising supplements in this area, although further research is needed to determine how the combined use of these supplements influence URTS risk. Athletes need for these supplements should be assessed on an individual case-by-case basis, based on several factors (e.g., URTS history, sport, nutrient status, etc.). Supplements to be used at the Summer Games should be piloted (for acceptance/compliance/safety) in an off season/preparatory phase. Ensure selected supplements are batch tested and comply to WADA regulations.

Paralympic Athletes

Paralympic athletes appear to be more susceptible to illness than able-bodied athletes. Paralympic athletes suffered almost double the amount of URTS than able-bodied athletes during previous London and Rio Summer Games (Paralympics: 12–14% vs. Able-bodied: 5–7%) (Derman et al., 2013, 2017; Engebretsen et al., 2013; Soligard et al., 2017). It is difficult to ascertain why Paralympic athletes are at a heightened risk for illness, as research on Paralympic sport is limited compared to investigations of able-bodied athletes (Van Rensburg et al., 2018). Illness risk will differ between Paralympic athletes based on their disability type. Paralympic athletes with spinal cord injuries have altered autonomic control and immunity, and impaired immune function has been cited as the main reason for increased illness susceptibility in this population (Leicht et al., 2013). In addition, the use of wheelchairs by Paralympic athletes likely increases infection transmission risk, as wheelchairs pick up and carry high numbers of bacteria. Indeed, at the Rio Paralympics, the highest illness incidence rate was reported in wheelchair fencing, while wheelchair basketball was only behind Paralympic swimming in terms of illness sustained (Derman et al., 2017).

Strategies to maintain paralympic athlete health

- **Hygiene:** Wheelchair athletes should regularly disinfect wheelchairs, wear gloves, ensure good hand hygiene, and avoid self-inoculation by touching eyes, nose, and mouth (Walsh, 2018).
- **Supplementation** (see section “Strategies to Maintain Female Athlete Health” for details).

Water-Sport Athletes

Athletes involved in Water-sports may be at an increased risk for illness during the Summer Games. At previous Summer Games, the IOC identified the top 5 sports with the highest illness incidence; water-sports accounted for 2 out of 5 (sailing and synchronized swimming) and 4 out of 5 (diving, open water

TABLE 1 | Summary of five key illness prevention strategies that athletes should consider adhering to during the Summer Games.

Strategy	Proposed rationale	Practical recommendations	References
1. Hygiene practices	Minimize risk for infection transmission	<ul style="list-style-type: none"> - Hand hygiene: Wash hands regularly (rub hands with soap >20 s and dry hands thoroughly with clean towel) and carry alcohol-based hand gel - Clean sporting equipment and clothing regularly - Isolate sick athletes and support staff (e.g., move out roommates) - Avoid self-inoculation by not touching eyes, nose, and mouth - Avoid shaking hands with other athletes and support personal - Where possible, avoid crowded areas, sick people, young children (if avoidance is not possible, wear facial masks) 	Schwellnus et al., 2016 Keaney et al., 2018; Walsh, 2018
2. Maintain day-to-day CHO availability	Preventing low CHO availability may minimize the exercise induced rise in stress hormones (cortisol and catecholamines) which in turn may attenuate immune perturbations	<ul style="list-style-type: none"> - Total CHO intake should match daily training and competition requirements - Athletes engaging in prolonged continuous exercise or high intensity intermittent team sport exercise should aim to consume 30–60 gCHO/h 	Burke et al., 2011; Berman et al., 2017
3. Probiotic supplementation	Probiotics may help to reduce the incidence, severity, and duration of URTS	<ul style="list-style-type: none"> - Type: Non-refrigerated (travel friendly), multi-strain probiotic combining <i>Lactobacillus</i> and <i>Bifidobacterium</i>, ensure selected probiotic complies with WADA anti-doping regulations - Dosage: 1×10^9 colony forming units per day - Timing: Commence probiotic supplementation at least 2 weeks before traveling to Tokyo, to allow adequate time for colonization - Potential side effects: In first 2 weeks athletes may experience gastrointestinal issues (e.g., stomach rumbles, increased flatulence), athletes experiencing these symptoms should take their probiotic on an empty stomach. If side effects persist (>2 weeks) try reducing the dosage by half and gradually increase dosage as symptoms ease 	Pyne et al., 2015; Williams et al., 2018
4. Minimize stress and anxiety	Stress and anxiety are risk factors for illness. Management of stress and anxiety may lower risk for URTS	<ul style="list-style-type: none"> - Identify athletes with high anxiety and stress using validated questions, e.g., Depression, Anxiety, Stress Scale (DASS-21), Recovery Stress Questionnaire (REST-Q-Sport-52) - Monitor stress and anxiety using a wellness questionnaire (refer to section “Strategies to Maintain Athlete Health During HA” for details) - Consult a psychologist to provide education around anxiety and stress management techniques - Mindfulness practice (refer to section “Strategies to Maintain Multi-Competition/Event Athlete Health” for details) 	Schwellnus et al., 2016; Drew et al., 2017; Walsh, 2018
5. Improve sleep	Sleep deprivation is a risk factor for illness. Improving sleep may reduce URTS risk	<ul style="list-style-type: none"> - Use objective (e.g., wrist actigraphy) or subjective (e.g., questionnaire) methods to identify sleep deprived athletes (<7 h per night) athletes - Aim for a minimum 8 h of quality sleep per night - Apply sleep hygiene strategies to optimize sleep quantity and quality [e.g., maintaining a regular bed and wake time, ensuring a quiet, cool, and dark bedroom environment (19–22°C), avoidance of stimulants (e.g., caffeine) prior to sleep, avoidance of light-emitting technology devices in the 30 min prior to sleep] 	Fullagar et al., 2015; O'Donnell and Driller, 2017

marathon, canoe slalom, and synchronized swimming) sports at the London (Engebretsen et al., 2013) and Rio Olympics (Soligard et al., 2017), respectively. Similarly, at the Rio Paralympics, Para-Swimming was the sport with the second highest illness incidence

(Derman et al., 2017). There are two likely factors underpinning increased illness susceptibility in this population: (1) chlorine exposure for pool-athletes; and (2) water quality issues for open-water athletes. Airway disorders, including asthma and

rhinitis are prevalent in pool-athletes and are often attributed to chlorine and chlorine by-products causing airway changes (Škrjat et al., 2018). As such, asthma and allergy mediated non-infectious URTS may explain why pool-sport athletes appear to be at an increased risk for illness. Alternatively, for open-water sport athletes, particularly at the Rio Summer Games, reports suggest that water quality issues (i.e., contamination with bacteria and viruses) were the primary cause of higher illness incidence (Keith, 2017).

Strategies to maintain water-sport athlete health

- Diagnose and control asthma and allergies (see section “Strategies to Minimize Cold Air Mediated Non-infectious URTS” for details).
- Supplementation (see section “Strategies to Maintain Female Athlete Health” for details).

Multi-Competition/Event Athletes

Multi-competition/event athletes may be at an increased risk for illness. Indeed, data obtained at previous Olympic games demonstrated that of the top 5 sports with the highest illness incidence the majority were multi-competition/event sports [5/5 in London: Athletics, Beach VB, Football, Sailing, and Synchronized Swimming (Engebretsen et al., 2013)] [4/5 sports in Rio: Diving, Canoe Slalom, Equestrian and Synchronized Swimming (Soligard et al., 2017)]. It is unclear why these athletes are more susceptible to illness, nonetheless it may be explained by the psychological element of having to mentally prepare for multiple events. Recent research suggests that mental state influences immunity, for example, state-anxiety and perceived psychological stress before exercise has been shown to influence immune responses to a greater extent than exercise itself (Edwards et al., 2018). In addition, a significant association between mental health (i.e., perceived stress and depression) and illness incidence has been demonstrated in athletes preparing for the Rio Olympics (Drew et al., 2017). Further studies should explore mental state to better elucidate how it affects athletes' immunity and URTS risk.

Strategies to maintain multi-competition/event athlete health

- Manage stress and anxiety (refer to **Table 1** for guidelines).
- Mindfulness practices: Mindfulness interventions such as meditation, breathing awareness, walking and yoga have the potential to alleviate psychological stress and anxiety. Recent studies have reported significant improvements to athletes' mental state with 4–6 weeks of mindfulness training (Ajilchi et al., 2019; Chen et al., 2019). Furthermore, in wheelchair basketball players,

8 weeks of mindful meditation utilizing a smart phone app attenuated the rise in cortisol associated with a competition period (MacDonald and Minahan, 2018). However, immune responses did not appear to be influenced (MacDonald and Minahan, 2018). It is currently unclear if mindful training influences URTS risk in athletes, nevertheless in the general population a reduction in URTS incidence has been demonstrated following 8 weeks of mindful meditation (Barrett et al., 2012). Mindfulness training appears to be a promising strategy for athletes, although further investigation is warranted. Athletes planning to use mindfulness interventions at the Summer Games should pilot and optimize practices in an off season/preparatory period.

- Supplementation (see section “Strategies to Maintain Female Athlete Health” for details).

CONCLUSION

It is apparent that athletes will be exposed to various stressors during both the preparatory and competition phases of the Summer Games. Athletes residing in the southern hemisphere appear to be at increased risk for illness during the preparatory phase, while female, Paralympic, water-sport and multi-competition/event athletes may be more susceptible to illness during the competition phase of the Summer Games. To maintain athlete health, illness prevention strategies should be targeted to stressors and at-risk athletes. Keeping athletes healthy will contribute to optimal Olympic and Paralympic athletic performance. While the considerations and strategies outlined in this mini review are targeted for the Summer Games, many could be used for other major competitions and as such should be considered for future sporting success.

AUTHOR CONTRIBUTIONS

All authors were involved in the conception of the manuscript. LK drafted the manuscript. FM, AK, and DD critically revised the manuscript and approved the final version to be published.

ACKNOWLEDGMENTS

None. The authors received no external financial support to aid the writing of this mini review.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Ambient Conditions Prior to Tokyo 2020 Olympic and Paralympic Games: Considerations for Acclimation or Acclimatization Strategies

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OPEN ACCESS

Edited by:

Toby Mündel,
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Hidekazu Otani,
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Medical Center, United States
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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 04 December 2018

Accepted: 26 March 2019

Published: 24 April 2019

Citation:

Gerrett N, Kingma BRM, Sluiter R
and Daanen HAM (2019) Ambient
Conditions Prior to Tokyo 2020
Olympic and Paralympic Games:
Considerations for Acclimation or
Acclimatization Strategies.
Front. Physiol. 10:414.
doi: 10.3389/fphys.2019.00414

The Tokyo Olympics and Paralympic games in 2020 will be held in hot and humid conditions. Heat acclimation (in a climatic chamber) or heat acclimatization (natural environment) is essential to prepare the (endurance) athletes and reduce the performance loss associated with work in the heat. Based on the 1990–2018 hourly meteorological data of Tokyo and the derived wet bulb globe temperature (WBGT) (Liljegren method), Heat Index and Humidex, it is shown that the circumstances prior to the games are likely not sufficiently hot to fully adapt to the heat. For instance, the WBGT 2 weeks prior to the games at the hottest moment of the day (13:00 h) is $26.4 \pm 2.9^\circ\text{C}$ and $28.6 \pm 2.8^\circ\text{C}$ during the games. These values include correction for global warming. The daily variation in thermal strain indices during the Tokyo Olympics (WBGT varying by 4°C between the early morning and the early afternoon) implies that the time of day of the event has a considerable impact on heat strain. The Paralympics heat strain is about 1.5°C WBGT lower than the Olympics, but may still impose considerable heat strain since the Paralympic athletes often have a reduced ability to thermoregulate. It is therefore recommended to acclimate about 1 month prior to the Olympics under controlled conditions set to the worst-case Tokyo climate and re-acclimatize in Japan or surroundings just prior to the Olympics.

Keywords: Olympics, thermal strain, WBGT, temperature, humidity, heat index, humidex

INTRODUCTION

The Tokyo 2020 Olympics are expected to be the hottest ever with daily air temperatures exceeding 30°C and wet bulb globe temperature (WBGT) (>28 during the hottest part of the day), exceeding that of previous games (Kakamu et al., 2017). These conditions expose athletes to extremely challenging conditions in which one cannot perform optimally. It is undisputed that heat acclimation or acclimatization, hereafter collectively referred to as HA, brings about physiological

adaptations in humans to lessen the thermal strain as a result of enhanced thermoregulatory capabilities. And since precooling opportunities are limited in most sports (Bongers et al., 2017), the athletes strongly depend on HA to prepare for the environmental conditions. Athletes and their support teams have two options for HA protocols: acclimation or acclimatization. Acclimation is the physiological or behavioral changes that reduces the strain or enhances the endurance of the strain brought about by artificial exposure to the climatic conditions (IUPS Thermal Commission, 2001). This requires the use of climatic chambers, which are expensive, require trained professionals and are not widely accessible; questioning the feasibility of this protocol for all athletes. Alternatively, they can adopt an acclimatization protocol whereby physiological or behavioral changes that reduces the strain arise from exposure to a natural climate (IUPS Thermal Commission, 2001). Namely, athletes may travel to Japan prior to the Olympics and adapt to the local climate naturally but the ambient conditions prior to the Olympic games may not reflect the actual conditions expected during the games. Both techniques have advantages and disadvantages and which protocol to adopt should be a key consideration for athletes and their support teams.

The reported physiological adaptations to HA protocols include a lower resting and exercising core body temperature (T_c), a lower heart rate, a higher sweat rate (SR) alongside a diluted sweated ion concentration, expanded plasma volume and a lower T_c for the onset of sweat production and vasodilation (Sawka et al., 2001). The physiological adaptations that occur serve to attenuate the thermal strain and can also improve athletic performance in warm-hot conditions (Lorenzo et al., 2010; Garrett et al., 2012; Racinais et al., 2015). Périard et al. (2015) summarized the time course of various physiological adaptations; suggesting that the adaptation process begins with the first exposure and complete HA occurs after approximately 14 days. Full adaptation for reduced HR, core and skin temperatures taking about 7 days and the longest requirement for full adaptation is about 2 weeks for SR responses. This information is generally based on studies that utilized acclimation protocols and only a few studies used heat acclimatization. A recent study showed that athletes from countries with a warm climate performed better than athletes from countries with a cold climate during the Marathon des Sables (Ioannou et al., 2018). To our knowledge no study has directly compared heat acclimation with heat acclimatization. Although there is some evidence that tolerance to higher T_c is greater in those acclimatized over several weeks compared to those acclimated to 1–2 weeks (Sawka et al., 2001). However, the difficulty in making direct comparisons is substantial. It remains unclear which technique offers the best advantage for complete HA.

For athletes traveling far, jet lag, travel fatigue and adjustments of circadian rhythms also must be considered. In this sense it may be appealing to travel to Japan several weeks prior to the games in order to adjust circadian rhythms and heat adapt to the local climate using heat acclimatization. However, there is a considerable risk that the local climate prior to the games is not stressful enough to elicit the necessary adaptations. The Tokyo

2020 Olympics are scheduled 24th July until 9th August with the month of August having the highest ambient temperature conditions. The Paralympics are scheduled from August 25th to September 6th. Japan has distinct seasons so it is highly probable that the environmental temperatures in June and July are lower than that expected during the games. For optimal heat adaptation, it is recommended that the environmental conditions (i.e., temperature and humidity) should be similar to, or higher than that expected during competition (Taylor et al., 1997). Additionally, there is some evidence that the physiological adaptations for rectal temperature and heart rate are not evident when sufficient recovery is not available during a HA protocol (Daanen et al., 2011). To ensure optimal heat adaptations, the conditions expected during the Tokyo 2020 Olympics games should ideally be utilized during a HA protocol with relatively low thermal strain after the heat exposure to allow for sufficient recovery. The aim of this paper is to describe the ambient conditions prior to and during the Olympic and Paralympic games based on meteorological data from the past 28 years that include the effects of global warming. Using this information, we can describe the conditions expected during the games and assess the risk of incomplete adaptations through heat acclimatization.

MATERIALS AND METHODS

Meteorological data were derived from the Japan Meteorological Agency. Data were collected specifically from Tokyo ward (i.e., the city) of Tokyo prefecture; where most of the events will take place and includes the location of the Olympic village and the Olympic stadium. Some events are scheduled in neighboring prefectures to Tokyo such Kanagawa (sailing), Shizuoka (cycling), Ibaraki (football), Yokohama (football, baseball, and softball), and Saitama (football, basketball, and golf) which have similar environmental conditions to Tokyo. Some football events are also scheduled for prefectures north of Tokyo in Hokkaido (Sapporo; 830 km), Miyagi (Rifu; 310 km), and Fukushima (Fukushima; 240 km) prefecture, with the former having considerable cooler environmental conditions than Tokyo prefecture. For succinctness, the data presented only include meteorological data from Tokyo ward (Tokyo prefecture).

Hourly data were collected from the period of June 1 to September 6 (final day of the Paralympics) from 1990 to 2018. Meteorological data is available from 1989 but due to inconsistent gaps in data sampling during the year of 1989 we decided to exclude this year from our analysis. The data collected included temperature ($^{\circ}\text{C}$), dew point temperature ($^{\circ}\text{C}$), relative humidity (%), solar radiation (MJ/m^2), and wind speed (m/s).

To determine the ambient conditions in Tokyo on the lead up to and during the Olympic and Paralympic games we calculated the number of days prior to the start of the Tokyo 2020 games. For example, June 1st is identified as day -53 ; being 53 days before the start of the Olympics. July 24th is the official start date and as such is coded as 0 and ends on August 9th, coded as Day 16. The Paralympics are scheduled from August 25th until September 6th which are coded as Day 32 to Day 44. These data

were plotted alongside single meteorological data (temperature and relative humidity) and biometeorological indices [Heat Index (HI), Humidity Index (Humidex) and the WBGT].

Meteorological and Biometeorological Indices

Acclimation protocols are conducted in controlled climatic conditions based on temperature and relative humidity and thus these were included in our analysis. Biometeorological indices are generally better predictors of thermal strain than single meteorological variables and thus we used the meteorological data to calculate the following: Humidex, HI and WBGT [see the following papers for more detailed descriptions of these indices (Budd, 2008; Blazejczyk et al., 2012)]. Humidex uses air temperature and air vapor pressure to reflect the perceived temperature (Ho et al., 2016) and was calculated using the following equation:

$$\text{Humidex} = T + 0.5555 \cdot (vp - 10)$$

Where,

$$vp = 6.11 \cdot e^{\left(5.417753 \cdot \left(\frac{1}{273.16}\right) \cdot \left(\frac{1}{(273.16 + td)}\right)\right)}$$

Where, td is dew point temperature (in °C) and e is Euler's number.

The Humidex values are categorized as: 20–29 = *no discomfort*, 30–39 = *some discomfort*, 40–45 = *great discomfort*; *avoid exertion*, and ≥ 46 = *Dangerous*; *possible heat stroke*.

Heat Index is somewhat similar in that it as it combines air temperature and relative humidity to determine an apparent temperature, which indicates how hot it feels (Rothfus, 1990). HI was calculated using the following equation:

$$\begin{aligned} HI = & -42.379 + 2.04901523 \cdot T + 10.14333127 \cdot RH \\ & - 0.22475541 \cdot T \cdot RH \\ & - 0.00683783 \cdot T \cdot T - 0.05481717 \cdot RH \cdot RH \\ & + 0.00122874 \cdot T \cdot T \\ & \cdot RH + 0.00085282 \cdot T \cdot RH \cdot RH - 0.00000199 \cdot T \cdot T \cdot \\ & RH \cdot RH \end{aligned}$$

Where, RH is relative humidity (%) and T is temperature (°F). The temperature data from the Japan Meteorological Agency was converted from °C into °F to calculate the HI .

The output value from the HI calculation is categorized with the following descriptors: 27–32 = *Caution* (fatigue is possible with prolonged exposure and/or physical activity), 32–41 = *Extreme Caution* (sunstroke, muscle cramps, and/or heat exhaustion possible with prolonged exposure and/or physical activity), 41–54 = *Danger* (sunstroke, muscle cramps, and/or heat exhaustion likely, heat stroke possible with prolonged exposure and/or physical activity), ≥ 54 *Extreme Danger* (heat stroke or sunstroke likely).

As Japanese summers are known to be especially hot and humid, the Humidex and the HI were considered appropriate

for inclusion and are common to the United States and Canada, respectively. WBGT was also chosen as it is more internationally recognized and used by many sporting organizations and thus familiarity is greater. WBGT was calculated using the following equation:

$$\text{WBGT } (^{\circ}\text{C}) = 0.7 \cdot \text{wetbulb} + 0.2 \cdot T_{\text{globe}} + 0.1 \cdot T_{\text{drybulb}}$$

Where, wetbulb is the wet-bulb temperature, T_{globe} is the globe temperature and T_{drybulb} is the dry bulb temperature.

WBGT was calculated from meteorological data using the Liljegren method (Liljegren et al., 2008) using the Heat stress package in R developed by Meteo Suisse¹. Data for solar radiation collected from the Japan Meteorological Agency was measured in MJ/m²/hr and converted by a factor of 0.0036 to W/m².

Different sports governing bodies define the recommendations for activities at various WBGT ranges. As a general summary, the following recommendations are provided for continuous activity and competition for WBGT <18.3°C is “Generally safe/normal activity”; 18.4–22.2°C “risk of exertional heat stroke and heat illness; high risk individuals should be monitored or not compete”; 22.3–25.6°C “risk for all competitors increases”; 25.7–27.8°C “risk for unfit, non-acclimatized individuals is high”; 27.9–30.0°C “cancel level for exertional heat stroke risk”; 30.1–32.2°C “cancel or stop practice and competition”; $\geq 32.3^{\circ}\text{C}$ “Cancel exercise” (based on ASCM position stand, Armstrong et al., 2007). The category ranges and the associated risks are reduced in those who are fit, low risk and acclimatized individuals engaging in training or non-continuous activity.

The time schedule for Tokyo 2020 Olympics had not been announced at the time of writing this paper, therefore we assumed a time span of 08:00–21:00 h based on the three previous Olympics (Rio, London and Beijing), thus excluding night time temperatures. Data were averaged during this time period. To determine the worst-case scenario, we also averaged the data between 12:00 and 15:00 h. Data are also plotted for every hour of the day, averaged from day 0–16 and from day 32–44 for the start and end of the Olympics and Paralympics, respectively.

The average values over the 1990–2018 periods may not reflect the actual situation during the Tokyo 2020 Olympics due to numerous factors such as year-to-year variations, urban heat island effect and climate change. Using the Hothaps database (Kjellstrom et al., 2009, 2014) we extrapolated the yearly changes in WBGT, HI, and Humidex values to the year 2020 to correct for global warming. Hothaps monitors the effects of climate change worldwide and estimates the increase in meteorological and biometeorological indices per decade. The data generated were specific to Tokyo prefecture. Temperature, dew point temperature, and WBGT are estimated to have the following yearly increases: 0.038, 0.009, and 0.025°C, respectively. The collected data are based over 28 years with 2004 as the central year. Taking 2004 as the central year we then applied a correction of 0.61°C for temperature, 0.14°C for dew point temperature and 0.40°C for WBGT and applied this to the collected data (average

¹<https://rdrr.io/github/anacv/HeatStress/>

of the 1990–2018 period) to get the best estimate for the Tokyo 2020 Olympics. The temperature increase during the 1990–2018 period includes the urban heat island effect. The population in Tokyo prefecture increased from approximately 11.9 million in 1990 to 13.2 million in 2010². Assuming a similar exponential growth, the number of inhabitants is predicted to increase to 15.2 million in 2020. If we take the urban heat island effect ($2.01 \log P - 4.06$) (Oke, 1973), this results in 0.16°C increase in ambient temperature of the city center from the 1990–2018 average to 2020. Thus, the linear extrapolation of 0.61°C in temperature may slightly underestimate the real warming due to non-linear growth of the Tokyo population.

The probability of WBGT on the days prior to the Olympic games being similar to the expected conditions during the Olympic games for different time periods (08:00–11:00, 12:00–15:00, and 16:00–19:00 h) were calculated. To generate probability curves, we used the 50th and 95th percentile of the WBGT of the specific day prior to the Olympics and compared this to the actual percentiles during the Olympic period. In

this way we avoided the skewness (-1.05) of the WBGT data during the Olympic period. Although some skewness exists in the data (from day 10 onward) the variation for the parameters (temperature, relative humidity, HI, Humidex, and WBGT) is expressed as standard deviation.

RESULTS

24-h Meteorological and Biometeorological Indices During the Olympics

Hourly ambient temperature, relative humidity, WBGT, HI, and Humidex for the Olympic period (24th July until 9th August) are shown in **Figures 1A–D**. Solar radiation was slightly skewed so descriptive data (minimum, median, maximum, first, and third quartile) are shown in **Table 1**. The ambient temperature is lowest during the night time at approximately 05:00 h and begins to rise from 07:00 h, peaking to $31.3 \pm 3.1^\circ\text{C}$ at 14:00 h. As expected the relative humidity peaks when the ambient temperature is lowest

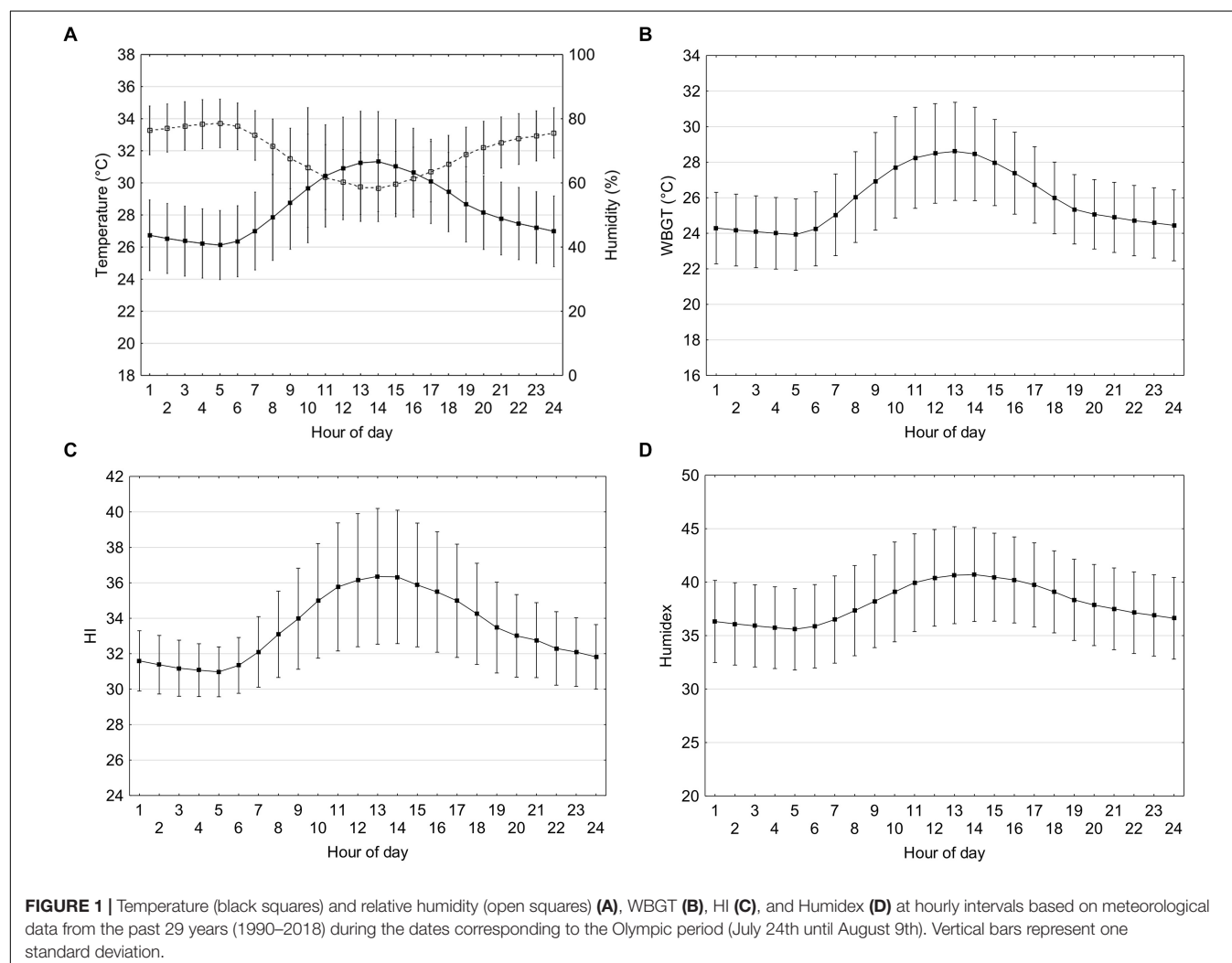


TABLE 1 | Minimum, first quartile, median, third quartile, and maximum solar radiation at hourly intervals based on meteorological data from the past 29 years (1990–2018) during the dates corresponding to the Olympic period (July 24th until August 9th) and Paralympic period (August 25th until September 6th).

	Solar radiation (W/m ²) Olympic period					Solar radiation (W/m ²) Paralympic period				
	Minimum	First quartile	Median	Third quartile	Maximum	Minimum	First quartile	Median	Third quartile	Maximum
05:00	0	0	0	3	14	0	0	3	6	22
06:00	0	19	42	69	122	0	9	39	72	158
07:00	0	58	131	206	328	0	26	106	192	411
08:00	0	111	233	392	553	3	49	194	358	569
09:00	8	153	353	539	711	3	68	286	506	733
10:00	0	194	483	678	853	3	85	331	619	869
11:00	0	236	561	783	942	4	95	408	703	961
12:00	0	286	636	839	953	2	115	450	728	1000
13:00	0	281	608	825	942	3	109	456	747	981
14:00	0	242	550	764	900	3	99	419	689	914
15:00	0	225	469	650	797	3	82	367	592	817
16:00	0	153	331	489	614	2	60	253	419	631
17:00	0	97	189	306	422	0	34	150	264	444
18:00	0	42	86	139	225	0	15	69	122	217
19:00	0	8	17	25	67	0	3	14	25	47
20:00	0	0	0	0	3	0	0	0	0	3
21:00–04:00	0	0	0	0	0	0	0	0	0	0

around 05:00 h at $79 \pm 8\%$ and as temperature rises the relative humidity declines to $58 \pm 10\%$ at the hottest part of the day and rises thereafter.

The WBGT is lowest during the night time at approximately 05:00 h (23.9 ± 2.0) and begins to rise from 06:00 h, peaking to $28.6 \pm 2.8^\circ\text{C}$ at 13:00 h. The Humidex is lowest during the night time at approximately 05:00 h (35.6 ± 3.8) and begins to rise from 06:00 h, peaking to 40.6 ± 4.5 at 13:00 h. HI also follows a similar pattern, with the lowest during the night time at approximately 05:00 h (31.0 ± 1.4) and begins to rise from 06:00 h, peaking to 36.4 ± 3.8 at 13:00 h. Solar radiation on a cloudless sky peaks at 917 W/m^2 at the hottest part of the day (12:00 h).

24-h Meteorological and Biometeorological Indices During the Paralympics

Hourly ambient temperature, relative humidity, WBGT, Humidex, and HI during the Paralympic period (August 25th to September 6th) are shown in **Figures 2A–D** and solar radiation in **Table 1**. The ambient temperature is lowest ($25.0 \pm 2.3^\circ\text{C}$) during the night time at approximately 05:00 h and begins to rise from 07:00 h, peaking to $29.7 \pm 3.4^\circ\text{C}$ at 13:00 h. Relative humidity peaks when the ambient temperature is lowest around 05:00 h at $78 \pm 9\%$ and as temperature rises the relative humidity declines to $59 \pm 13\%$ at the hottest part of the day and rises thereafter.

The WBGT is lowest during the night time at approximately 05:00 h ($22.8 \pm 2.2^\circ\text{C}$) and begins to rise from 06:00 h, peaking to 27.0 ± 2.9 at 13:00 h. The Humidex is lowest during the night time at approximately 05:00 h (33.5 ± 4.1) and begins to rise from 06:00 h, peaking to 37.8 ± 4.6 at 13:00 h. HI also follows a similar

pattern, with the lowest during the night time at approximately 05:00 h (30.6 ± 1.4) and begins to rise from 06:00 h, peaking to 33.4 ± 3.5 at 13:00 h. Solar radiation on a cloudless sky peaks at 1000 W/m^2 at the hottest part of the day (12:00 h).

Meteorological and Biometeorological Indices Prior to and During the Olympics and Paralympics (08:00–21:00 h)

Based on previous Olympics games, the typical competing period was between 08:00 and 21:00 h. The ambient temperature, relative humidity, WBGT, Humidex, and HI on the days prior to the Olympics during this time period are shown in **Figures 3A–D**. This data is based on hourly data averaged between 08:00 and 21:00 h when it is expected that the games will take place. Fifty-three days prior to the Olympics the ambient temperature is $22.9 \pm 3.1^\circ\text{C}$ and continues to rise to a plateau during the Olympics period, after which temperature starts to drop. Relative humidity reaches its peaks and plateaus much more quickly (by day -40) and remains high throughout the Olympic and Paralympic period. For the Olympic period the average temperature and relative humidity is $29.7 \pm 3.1^\circ\text{C}$ and $65 \pm 11\%$. The Paralympic period has an average temperature of $28.2 \pm 3.2^\circ\text{C}$ and relative humidity of $65 \pm 13\%$.

The WBGT at day -53 is $19.9 \pm 2.8^\circ\text{C}$ and rises steadily until day -1 followed by a plateau until day 17. The average WBGT during the Olympic period is $27.0 \pm 2.8^\circ\text{C}$. After this period, the WBGT slowly starts to decline. The average WBGT for the Paralympics is $25.4 \pm 3.0^\circ\text{C}$. Humidex at day -53 is 25.9 ± 3.9 and rises steadily until day 2 followed by a short plateau until day 19 and slowly declines thereafter. The average Humidex during

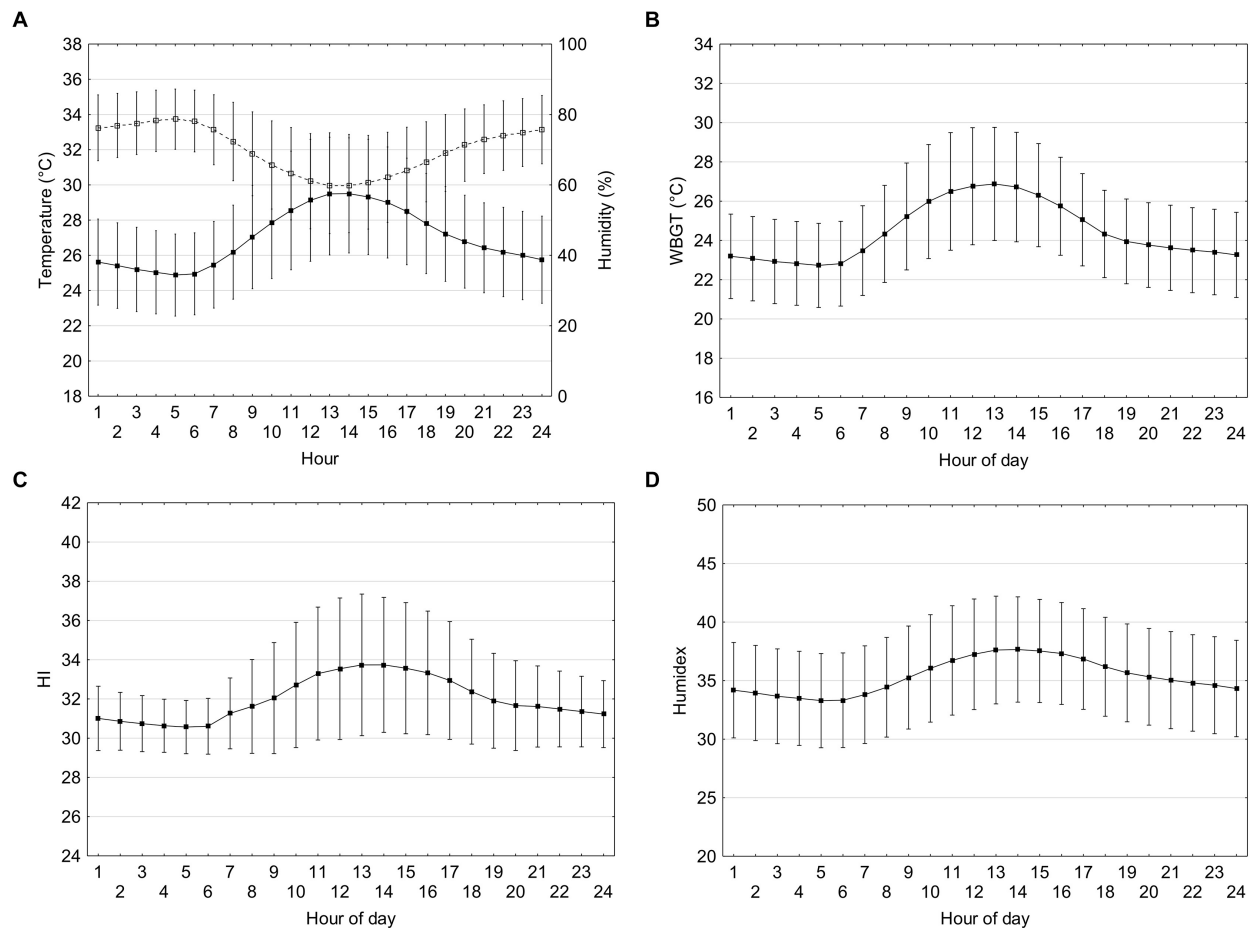


FIGURE 2 | Temperature (black squares) and relative humidity (open squares) (A), WBGT (B), HI (C), and Humidex (D) at hourly intervals based on meteorological data from the past 29 years (1990–2018) during the dates corresponding to the Paralympic period (August 25th until September 6th). Vertical bars represent one standard deviation.

the Olympic period is 39.3 ± 4.3 and 36.5 ± 4.5 during the Paralympic. HI at day -53 is 28.8 ± 1.8 and rises steadily until day 3 followed by a short plateau until day 15 and thereafter declines. The average HI during the Olympic and Paralympic periods are 34.8 ± 3.4 and 33.0 ± 3.3 , respectively.

Meteorological and Biometeorological Indices Prior to and During the Olympics and Paralympics (12:00–15:00 h)

The hottest period of the day is between 12:00 and 15:00 h and may represent the worst-case scenario during the Olympic and Paralympic games. The ambient temperature, relative humidity, WBGT, Humidex, and HI on the days prior to the Olympics during this time period are shown in **Figures 4A–D**.

The ambient temperature rises from $24.3 \pm 3.2^\circ\text{C}$ on day -53 until approximately day -3 where the temperature peaks and plateaus, declining slowly from day 17 onward. The relative humidity rises from $50 \pm 14\%$ on day -53 and rises to a long plateau on day -38 with small variations during the Olympic and Paralympic period. The temperature and RH during the

Olympic period between 12:00 and 15:00 h are $31.3 \pm 3.1^\circ\text{C}$ and $59 \pm 10\%$. The temperature and RH during the Paralympic period are $29.5 \pm 3.4^\circ\text{C}$ and $60 \pm 13\%$.

The WBGT at day -53 is $21.5 \pm 2.7^\circ\text{C}$ and rises steadily until day 7 followed by a plateau until day 17, declining thereafter. The average WBGT during the Olympic and Paralympics periods are $28.4 \pm 2.8^\circ\text{C}$ (5th and 95th percentile: 23.1 and 31.7, skewness -1.05) and $26.8 \pm 3.0^\circ\text{C}$ (5th and 95th percentile: 21.6 and 30.6, skewness -0.4). Humidex at day -53 is 27.2 ± 4.0 and rises steadily until day -1 followed by a short plateau until day 15, declining thereafter. The average Humidex during the Olympics and Paralympics are 40.7 ± 4.4 and 37.8 ± 4.5 , respectively. HI at day -53 is 27.8 ± 1.8 and rises steadily peaking at day 11, declining thereafter. The average HI during the Olympics and Paralympics are 36.1 ± 3.7 and 33.8 ± 3.6 , respectively.

Chance of Successful Acclimatization

The probability of WBGT on the days prior to the Olympic games being similar to the expected conditions during the Olympic games were calculated and plotted in **Figure 5**.

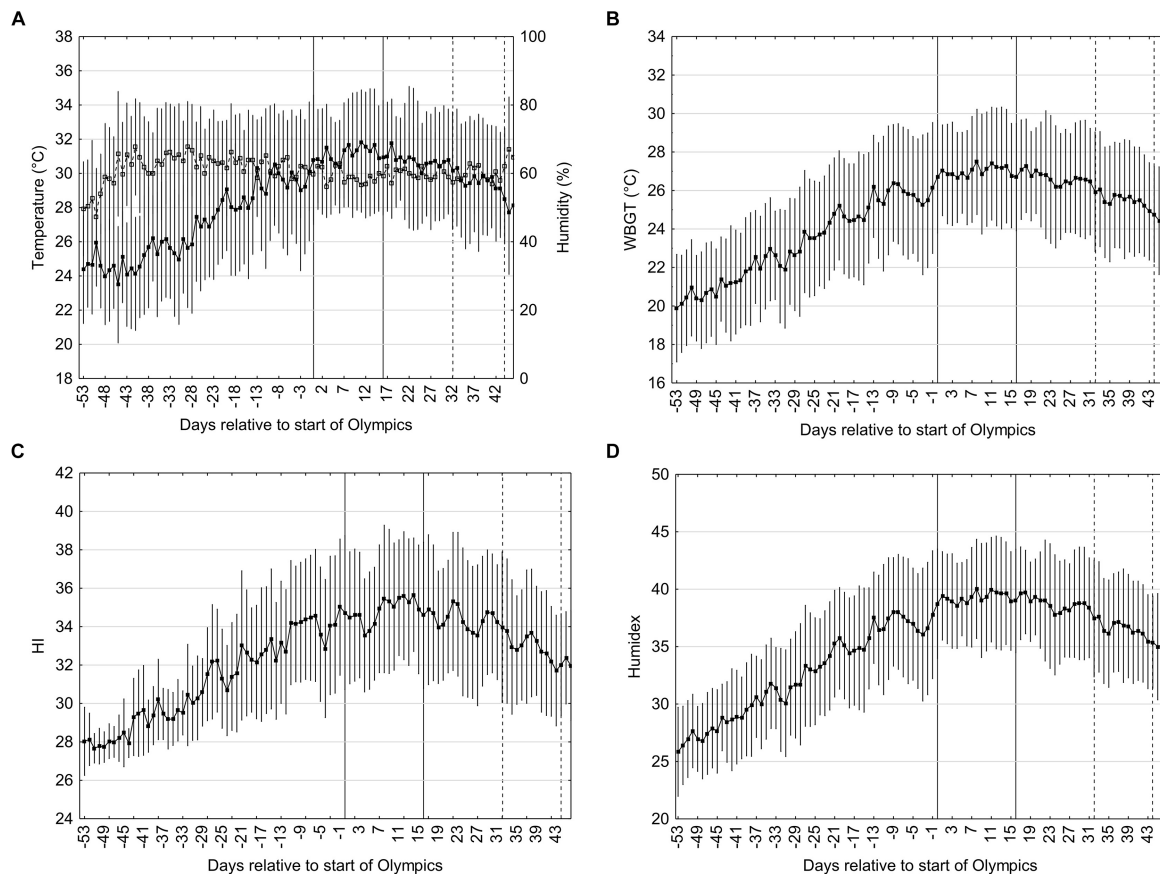


FIGURE 3 | Temperature (black squares) and relative humidity (open squares) **(A)**, WBGT **(B)**, HI **(C)**, Humidex **(D)** on the days prior to the start of the Olympics and Paralympics. Data is the mean (\pm SD) during the time period of 0800–2100 h obtained from hourly metrological data from the past 29 years (1990–2018). The Olympics period is coded on day 0 to 16 and the Paralympics on day 32 and 44; the former is in between the solid lines and the latter between the dotted lines. Vertical bars represent one standard deviation.

Using the 50th percentile for 08:00–11:00, 12:00–15:00, and 16:00–19:00 h, generated the following respective logistic equations:

$$\text{probability (\%)} = \frac{1}{1 + e^{-0.08} (0 - 8.1)}$$

$$\text{probability (\%)} = \frac{1}{1 + e^{-0.08} (0 - 1.1)}$$

$$\text{probability (\%)} = \frac{1}{1 + e^{-0.08} (0 - 13.3)}$$

Where, e is Euler's number.

Using these equations, it can be calculated that 14 days prior to the Olympic games at 08:00–11:00 h there is a 38%, chance of WBGT being similar to that expected during the Olympic games. Between 12:00–15:00 and 16:00–19:00 h the chance of similar conditions during the Olympic games are 23 and 49%, respectively.

To account for the maximum temperatures (i.e., the 95th percentile), the curve shifts upward and to the left. Thus, 14 days

prior to the Olympics there is 87% chance of the expected WBGT occurring at 08:00–11:00 h and 78% at 12:00–15:00 h. The chance of the conditions being similar to those during the games between 16:00 and 19:00 h is 92%. The probability curves for the 95th percentile for 08:00–11:00, 12:00–15:00, and 16:00–19:00 h generated the following respective logistic equations:

$$\text{probability (\%)} = \frac{1}{1 + e^{-0.08} (0 - 37.4)}$$

$$\text{probability (\%)} = \frac{1}{1 + e^{-0.08} (0 - 29.8)}$$

$$\text{probability (\%)} = \frac{1}{1 + e^{-0.08} (0 - 45.1)}$$

DISCUSSION

The first purpose of this study was to describe the ambient conditions during the Tokyo 2020 Olympic and Paralympic games based on meteorological data from the past 28 years

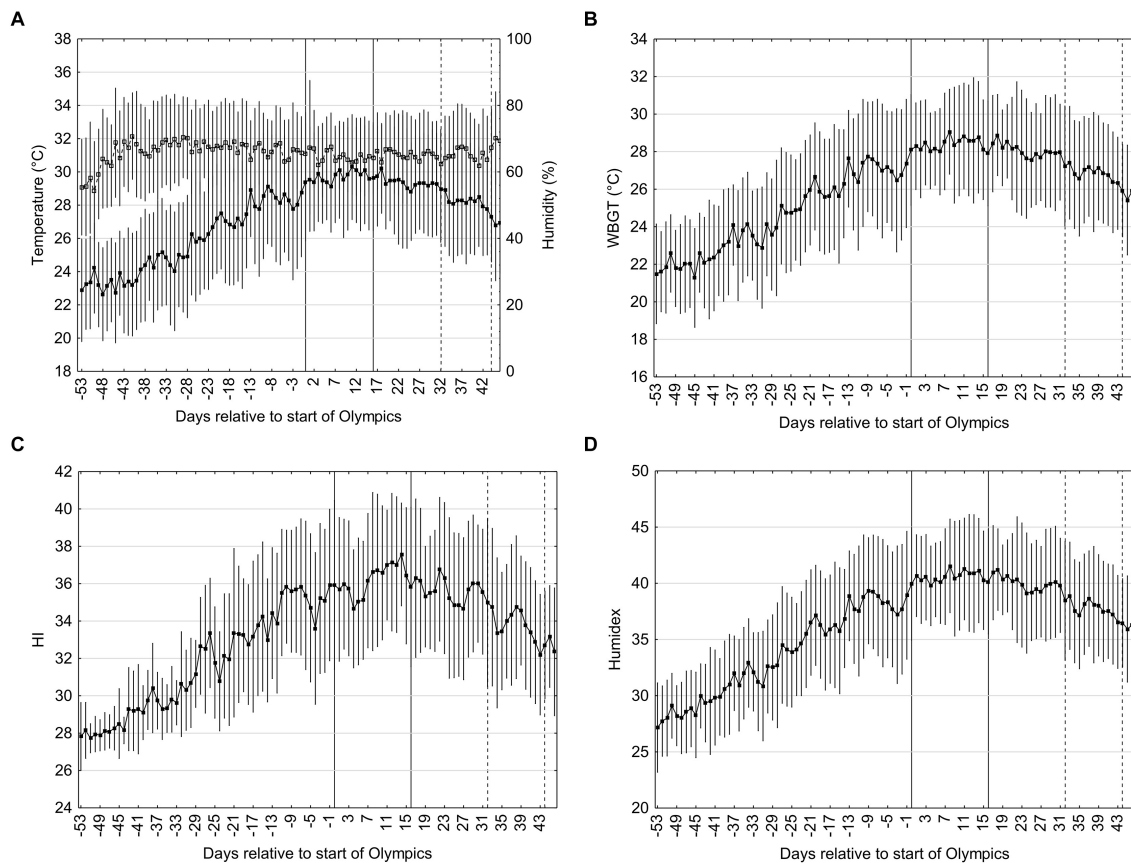


FIGURE 4 | Temperature (black squares) and relative humidity (open squares) (A), WBGT (B), HI (C), Humidex (D) on the days prior to the start of the Olympics and Paralympics. Data is the mean (\pm SD) during the time period of 12:00–15:00 h obtained from hourly metrological data from the past 29 years (1990–2018). The Olympics period is coded on day 0 to 16 and the Paralympics on day 32 and 44; the former is in between the solid lines and the latter between the dotted lines. Vertical bars represent one standard deviation.

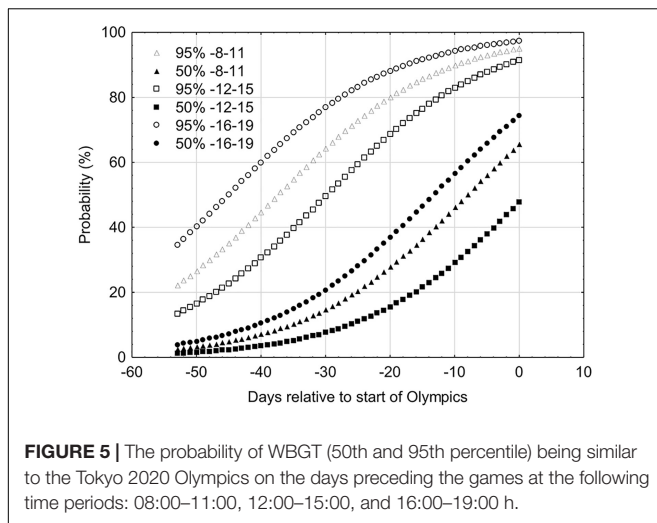
in Tokyo city corrected for global warming. Hourly data for ambient temperature and relative humidity and three indices of heat strain (WBGT, Humidex, and HI) were calculated for both the Olympic and Paralympic periods. Using this data, we can describe the expected conditions for the upcoming games and highlight average and peak conditions. Furthermore, the second purpose of this study was to describe the ambient conditions on the days prior to the start of the Olympic games and assess the risk of incomplete adaptations through heat acclimatization. Athletes, coaching staff and sports governing bodies will find the descriptive data presented useful for knowing the expected environmental conditions and for designing optimal preparation strategies.

Hourly Meteorological and Biometeorological Indices During the Games

The average estimated WBGT values during the Tokyo 2020 Olympics games are $27.0 \pm 2.8^\circ\text{C}$ averaged over the day and $28.4 \pm 2.8^\circ\text{C}$ over the hottest period of the day (Figures 3, 4, respectively). The latter values are in the 27.9 – 30°C “cancel

level for exertional heat stroke risk” range for continuous activity/competition which may be relevant for endurance based events such as the marathon (Armstrong et al., 2007). According to the ACSM guidelines (Armstrong et al., 2007) for non-continuous activity, which may be more relevant for team based intermittent events, the risk to the athlete is lower when heat acclimatized and events canceled when the WBGT exceeds 32.3°C . Kakamu et al. (2017) calculated that on average 49 days in Japan exceed the 27.9°C threshold. However, this may be an overestimation as the WBGT estimation of Kakamu et al. (2017) was based on the Australian Bureau of Meteorology (ABM) model and may show considerable deviations from the real WBGT in particular in cloudy situations (Grundstein and Cooper, 2018). We used a method that has been shown to estimate WBGT from meteorological data with greater precision (Liljegren et al., 2008). Given that the variation in the data is large the chances of the WBGT exceeding 27.9°C is high (85%), whilst the chances for exceeding the threshold for 32.3°C are only 15% for the hottest part of the day (WBGT $28.4 \pm 2.7^\circ\text{C}$).

During the Olympic games the average daily Humidex and HI are 39.3 ± 4.3 and 34.8 ± 3.4 , respectively. If athletes are competing during the hottest period of the day (12:00–15:00 h)



the Humidex and HI values of 40.7 ± 4.4 and 36.1 ± 3.7 could be expected. According to the guidelines for Humidex this would expose individuals to “some discomfort” during most of the day but “great discomfort” during the hottest periods, with the recommendation to “avoid exertion.” For HI this would expose athletes to conditions categorized as “Extreme caution” with sunstroke, muscle cramps and/or heat exhaustion possible with prolonged exposure and/or physical activity. The heat strain indices, Humidex and HI, specify “danger” levels at values ≥ 46 and 41–54, respectively. **Figures 1C,D** shows that whilst these levels may not be attained during the hottest part of the day (Humidex; 40.7 ± 4.4 and HI; 36.1 ± 3.7), the standard deviations are considerable so there is a possibility that those conditions could be exceeded during the Olympics and Paralympics. Kosaka et al. (2018) measured the ambient conditions along the course of the marathon route during July and August, with either 06:30 h or 07:30 h start times. The best- and worst-case scenario for temperature and relative humidity were $26.3 \pm 0.85^\circ\text{C}$ and $51.8 \pm 3.4\%$ and $34.3 \pm 1.8^\circ\text{C}$ and $47.4 \pm 5.6\%$, respectively. Furthermore, no cloud cover alongside high ambient temperature and relative humidity during the hottest period of the day (see **Table 1**) can add considerable thermal challenges and negate performance (Otani et al., 2019). The worst-case scenario during the Olympics could be solar radiation of $855\text{--}917\text{ W/m}^2$ between 12:00 and 15:00 h. The evidence to date suggests that athletes and coaches may consider preparing for the worst-case scenario; heat adaptation strategies, pre- and per-cooling, clothing, reducing time outdoors, and strategies to promote behavioral thermoregulation should be considered.

The Paralympic games are scheduled August 25th until September 6th; 16 days after the Olympics whereby the ambient temperatures have begun to decline. Nevertheless, the average daily temperature and relative humidity during the Paralympic games are $28.2 \pm 3.2^\circ\text{C}$ and $65.0 \pm 12.7\%$ and can be expected to reach $29.5 \pm 3.4^\circ\text{C}$ and $59.6 \pm 13.0\%$ during the hottest periods of the day. Solar radiation could be as high as $914\text{--}1000\text{ W/m}^2$ during the hottest period of the day when there is minimal cloud cover. These conditions still impose a considerable

thermal challenge and even more so as some Paralympic athletes have impaired thermoregulatory functions (Price, 2006; Griggs et al., 2015). There is less evidence for the application of heat strain indices in special populations and sporting events for disabled athletes (Girard, 2015). Guidelines are available from some sports governing bodies for para-athletes employing similar or lower thresholds for the suspension of play compared to able-bodied athletes. The International Tennis Federation for example suspends play when $\text{WBGT} \geq 28^\circ\text{C}$. During the Paralympic period the estimated daily WBGT is 25.4 ± 3.0 and $26.8 \pm 3.0^\circ\text{C}$ during the hottest periods of the day. Given the variation in the data the possibility of conditions exceeding this threshold is likely. The average daily HI and Humidex are 33.0 ± 3.3 and 36.5 ± 4.5 , respectively. If athletes are competing during the hottest period of the day (12:00–15:00 h) the Humidex and HI values of 37.8 ± 4.5 and 33.8 ± 3.6 could be expected. For Humidex “some discomfort” would be expected and the HI would vary between “caution” to “extreme caution.” However, we could not find any information about whether these categories are appropriate for special populations. Given the diverse health conditions of Paralympic athletes and the elevated risk of heat related illness amongst those with impaired thermoregulatory functions (e.g., spinal cord injured individuals) the thermal challenge still exists and should be prepared for.

Heat Acclimate or Heat Acclimatize?

Based on the expected environmental conditions described previously it is evident that the conditions will impose thermal challenges and all athletes are advised to take additional precautions to ensure safe and optimal performances. A highly effective method to prepare for the environmental conditions can be achieved through heat acclimation or heat acclimatization protocols. The former involves creating an artificial environment usually in a climatic chamber and exposing oneself to similar conditions expected during the games for a given period of time. This can typically be achieved over 5–14 days with approximate daily exposures of 2 h/day. To magnify the thermal stress, Taylor et al. (1997) recommended using a dry bulb temperature at least equivalent to the highest anticipated and where possible elevate this a further $5\text{--}10^\circ\text{C}$; with 40°C as the upper limit for humid conditions ($>60\%$ RH). Based on these recommendations and the reported meteorological data the environmental conditions employed in an acclimation protocol should be >31 and $\leq 40^\circ\text{C}$ with $>60\%$ RH. Recommended WBGT values to use for optimal HA are not widely reported but based on the expected WBGT values during the Olympic games a $\text{WBGT} \geq 28.4^\circ\text{C}$ should be considered. Recently there has been less focus on the environmental conditions in which these acclimation sessions are completed but rather using ambient conditions sufficient enough to raise core temperature $>38.5^\circ\text{C}$ quickly and keeping it elevated for 60 min (Garrett et al., 2012; Gibson et al., 2015; Neal et al., 2016). Whilst this technique of controlled hyperthermia focuses on the core temperature responses rather than the ambient conditions, conditions which restrict heat loss (i.e., hot and humid) will result in a faster elevation in core temperature and require a lower metabolic heat production to maintain an elevated

core temperature $>38.5^{\circ}\text{C}$. The physiological adaptations that accompany controlled hyperthermia are reported to occur more quickly and more completely than standard HA protocols (Taylor et al., 1997; Périard et al., 2015; Daanen et al., 2018). However, heat acclimatization has been favored in the literature for inducing physiological and psychological adaptations specific to the conditions where the event will take place (Hellon et al., 1956; Edholm, 1966; Périard et al., 2015; Racinais et al., 2015). Previous studies have shown an improved thermal tolerance in acclimatized compared to acclimated individuals, although direct comparisons are difficult as conditions were not similar between groups (Edholm, 1966; Sawka et al., 2001; Ioannou et al., 2018). Furthermore, the impact of solar radiations and air flow are often unaccounted for in acclimation protocols as they are difficult to replicate in climatic chambers but they have a large impact on overall heat stress in the field (Saunders et al., 2005). If the controlled hyperthermia protocol is used for HA, it has to be realized that it requires considerable exercise intensities to achieve the target core temperatures if the athlete chooses to acclimatize in the Tokyo climate about 2–3 weeks prior to the Olympics. The WBGT 2 weeks prior to the games at the hottest moment of the day (13:00 h) is only $26.4 \pm 2.9^{\circ}\text{C}$ compared to $28.6 \pm 2.8^{\circ}\text{C}$ during the games. The high exercise intensity may interfere with the tapering protocol that is generally used to recover from extensive exercise in the period prior to the Olympics (Mujika and Padilla, 2003).

Traveling to Japan several days to weeks prior to the Olympic games to acclimatize naturally to the environmental conditions is an attractive, feasible and alternative strategy to acclimation. It may be particularly advantageous when access to climatic chambers are not possible and also provides an opportunity for the adjustment of circadian rhythms if traveling far to Japan. The heat strain indices (temperature, WBGT, Humidex, and HI) are all considerably lower prior to the Olympics. The indices rise on the days leading up to the Olympics; peaking and plateauing during the Olympic games. Under these circumstances the thermal stress prior to the Olympics may be too low to achieve optimal adaptations (Taylor et al., 1997) or lead to exercise intensities interfering with tapering. The advice to use an acclimatization strategy may only be applicable if one is certain of stable and similar conditions prior to the event itself. Assuming that the environmental conditions required for successful acclimatization are equal to, or above, the conditions expected during competition (Taylor et al., 1997). **Figure 5** provides some indication of whether full acclimatization can occur. A 14-day HA strategy is often suggested for full adaptations, in similar environmental conditions (Taylor et al., 1997; Périard et al., 2015). If an athletes' scheduled competition is between 12:00 and 15:00 h and they employ a 14-day acclimatization strategy in Tokyo at the same time of day then at the start of heat acclimatization there is a 23% chance (50th percentile) of the conditions between similar to the Olympic games. This means that there is a strong risk (77% chance) of the WBGT being less than that expected during the Olympic games. Throughout the 14-day acclimatization protocol this chance reduces to 53% of the WBGT being less than that expected during the Olympic games. If we consider the 95th percentile, i.e., as an

indicator of the maximum heat strain during that day, then the chances during a 14-day acclimatization protocol of WBGT being less than the Olympics ranges from 8 to 22%. Schasfoort (2014) compared the meteorological conditions 2–3 weeks prior to and during the 1996, 2000, 2004, and 2008 Olympics and 12 other major sports events. Thermal stress was lower during the 2000 and 2004 Olympics prior to the games, but not for 1996 and 2008. In particular the 2012 UEFA European Soccer Championships in Ukraine (maximum temperature prior: 21.4°C , during: 26.3°C) and the 2011 Men's Hockey Champions Trophy in India (prior: 25.1°C ; during 29.6°C) showed significantly lower thermal stress prior to than during the games. The data indicates that there is some risk of incomplete adaptation if an acclimatization protocol in Tokyo is employed.

Figure 5 provides useful information to the organizing committee of the Olympic games. The probability of conditions reaching the 50th percentile for WBGT by day 0 is only 48% for events scheduled between 12:00–15:00 and 66 and 75% for events scheduled between 8:00–11:00 and 16:00–19:00 h. Such information may be pertinent for the organization committee of the Olympics to schedule high risk heat stress events, such as the marathon, toward the end of the Olympic games to allow athletes more opportunity to acclimatize naturally. When the event schedules are announced, **Figures 1–5** will provide useful advice on the ideal times to acclimatize to the heat. It is expected that prolonged and sustained high intensity efforts will be held early in the morning ($<09:00$ h) where the WBGT is $<26^{\circ}\text{C}$ (**Figure 1B**). **Figure 4B** shows that if 2 weeks prior to the Olympic games, the average WBGT is $>26^{\circ}\text{C}$ 2 weeks prior to the Olympic games. Based on our calculations if athletes choose to travel to Japan and acclimatize naturally during the 2 weeks preceding the games then the chances of WBGT being $>26^{\circ}\text{C}$ averages 67% from 12:00 to 15:00 h. Alternatively, when events are scheduled during the hottest periods (12:00–15:00) during the Olympics then sub optimal acclimatization conditions should be expected due to insufficient heat stress in the weeks prior to the event. In such situation's athletes/coaching staff may consider supportive strategies to facilitate heat acclimatization. Post-exercise hot water immersion has recently showed some positive results to facilitate physiological adaptations to heat (Zurawlew et al., 2016). As long as core body temperature is monitored to ensure athletes are working within safe boundaries then overdressing during training may also be administered (Ely et al., 2018; Willmott et al., 2018).

Studies show the physiological and performance benefits of training in the heat for competition in temperate conditions (Shvartz et al., 1979; Lorenzo et al., 2010; Corbett et al., 2014). To the authors knowledge there are no acclimation studies employing environmental conditions lower than those used during a heat stress test. Indeed, a review based on approximately twenty-one heat acclimation studies indicated that the conditions used for acclimation were the same or higher than those imposed during a heat stress test (Daanen et al., 2018). Karlsen et al. (2015) did, however, utilize natural acclimatization ($34 \pm 3^{\circ}\text{C}$, $18 \pm 5\%$) and tested performance in conditions that were, perhaps by chance rather than design, lower than the

conditions used during a heat stress test in a climatic chamber ($44 \pm 3^\circ\text{C}$, $44 \pm 5\%$). Performance during a cycling time trial relative to performance in an unacclimatized state, improved by 11 ± 8 and $5 \pm 4\%$ after 6 and 13 days (respectively) of natural acclimatization. Without a comparison it is difficult to determine whether HA in environmental conditions below the expected competition conditions provides a sufficient stimulus to negate detriments to performance in the heat; more importantly, whether the adaptations are optimal. This uncertainty elevates the risk of incomplete adaptations if an acclimatization strategy in Japan is used. Paralympic athletes on the other hand stem to benefit from the higher ambient conditions prior to the Paralympic games. For the hottest parts of the day the WBGT is 26.8 ± 3.0 and $28.4 \pm 2.8^\circ\text{C}$ during the Paralympic and Olympics, respectively. It may be feasible to acclimatize in Japan prior to the Paralympic games but athletes and coaches should fully consider the safety implications of training in the heat especially for those with impaired thermoregulatory abilities that results from their impairment and/or medication.

An important challenge is incorporating the high physical demands of a HA protocol within an athletes busy training schedule. The effects of HA begin to decay very quickly, with most physiological adaptations generally disappearing in 28 days (Williams et al., 1967; Armstrong and Maresh, 1991; Daanen et al., 2018). It is therefore advisable to HA as close as possible to competition or to maintain the acclimation status using several short periods of heat re-acclimation (Casadio et al., 2017; Daanen et al., 2018). There is some research to suggest that following a decay, the time period for regaining the previously attained physiological adaptations is reduced (Givoni and Goldman, 1973; Pandolf et al., 1977; Weller et al., 2007; Ashley et al., 2015) and one study even found that supercompensation existed in the physiological adaptations (Saat et al., 2005). Thus, it is possible to complete a full HA protocol 1 month prior to competition and then complete a shorter re-HA protocol just prior to competition. Within the context of Tokyo 2020 Olympics, traveling to Japan several days-to-weeks to acclimatize naturally to the environment imposes a risk of incomplete adaptations as the data presented clearly indicates lower ambient conditions prior to the Olympic games. If athletes have access to a climatic chamber and can complete a controlled hyperthermia acclimation strategy then this may allow athletes to acclimate fully prior to the games. Athletes can then incorporate their tapering phases within the decay period. They can travel to Japan with sufficient time for circadian adjustments and re-acclimatize in

Japan, where the chances of the conditions being similar to competition are greater.

Limitations

We aimed to estimate the ambient conditions in Tokyo city (Tokyo prefecture) during the 2020 Olympic games based on metrological data from the past 28 years. Climate forecasting is difficult and our analyses are estimations. The variation in the data may be larger than we present, especially as climate change has resulted in more extreme weather occurrences and challenges future forecasting. We aim to provide an estimation that considers the urban heat island effect, Tokyo's growing population and climate change but there are numerous factors uncontrolled for within our estimation.

To summarize, we provided an overview of the predicted environmental conditions prior to and during the Tokyo 2020 Olympic and Paralympic games. It is evident that there is a considerable thermal challenge and athletes and coaching staff are advised to prepare for these conditions. Whilst acclimatization is a feasible strategy, data are presented to assess the probability of the conditions prior to the Olympics being lower than during the Olympics. This information can be extremely useful in the planning and designing of strategies to allow the athletes to best prepare for the environmental conditions of the Tokyo 2020 Olympic and Paralympics games.

AUTHOR CONTRIBUTIONS

NG, RS, BK, and HD were involved in the conceptual ideas, data collection, analysis, interpretation, and manuscript presentation.

FUNDING

This study was supported by ZonMw (Project: Thermo Tokyo: Beat the heat), Netherlands Organisation for Scientific Research (NWO) (Project: Citius, Altius, Sanius), and Heatshield, under EU Horizon 2020 grant agreement No 668786.

ACKNOWLEDGMENTS

The authors thank Dr. Tatsuro Amano (Niigata University, Japan) for his assistance with translating the Japanese Meteorological Agency website.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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The Efficacy of Ingesting Water on Thermoregulatory Responses and Running Performance in a Warm-Humid Condition

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OPEN ACCESS

Edited by:

Gary W. Mack,
Brigham Young University,
United States

Reviewed by:

Zachary Schlader,
University at Buffalo, United States
Kei Nagashima,
Waseda University, Japan
Keiji Hayashi,
University of Shizuoka, Japan

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 03 November 2018

Accepted: 11 April 2019

Published: 07 May 2019

Citation:

Che Muhamed AM, Yusof HA,
Stannard SR, Mündel T and
Thompson MW (2019) The Efficacy
of Ingesting Water on
Thermoregulatory Responses
and Running Performance in a
Warm-Humid Condition.
Front. Physiol. 10:507.
doi: 10.3389/fphys.2019.00507

The understanding that fluid ingestion attenuates thermoregulatory and circulatory stress during exercise in the heat was based on studies conducted in relatively dry (~50% RH) environments. It remains undetermined whether similar effects occur during exercise in a warm and more humid environment, where evaporative capacity is reduced. Nine well-trained, unacclimatised male runners were randomly assigned to perform four experimental trials where they ran for 60 min at an intensity of 70% $\dot{V}O_{2\max}$ followed by an incremental exercise test until volitional exhaustion. The four trials consisted of non-fluid ingestion (NF) and fluid ingestion (FI) in a warm-dry (WD) and warm-humid condition (WH). Time to exhaustion (TTE), body temperature (T_b), whole body sweat rate, partitioned calorimetry measures, heart rate and plasma volume were recorded during exercise. There was no significant difference in T_b following 60 min of exercise in FI and NF trial within both WD ($37.3^{\circ}\text{C} \pm 0.4$ vs. $37.4^{\circ}\text{C} \pm 0.3$; $p > 0.05$) and WH conditions ($38.0^{\circ}\text{C} \pm 0.4$ vs. $38.1^{\circ}\text{C} \pm 0.4$; $p > 0.05$). The TTE was similar between FI and NF trials in both WH and WD, whereas exercise capacity was significantly shorter in WH than WD (9.1 ± 2.8 min vs. 12.7 ± 2.4 min, respectively; $p = 0.01$). Fluid ingestion failed to provide any ergogenic benefit in attenuating thermoregulatory and circulatory stress during exercise in the WH and WD conditions. Consequently, exercise performance was not enhanced with fluid ingestion in the warm-humid condition, although the humid environment detrimentally affected exercise endurance.

Keywords: fluid ingestion, thermoregulation, circulation, relative humidity, running exercise

INTRODUCTION

Recent evidence demonstrates a reduced physical capacity of trained individuals during exercise in the heat as relative humidity (RH) increases (Maughan et al., 2012; Moyon et al., 2014; Che Muhamed et al., 2016). This observation is attributed to a greater thermoregulatory and circulatory strain (Moyon et al., 2014; Che Muhamed et al., 2016), as the evaporative capacity of the environment (E_{\max}) reduces, leading to a decline in sweating efficiency and increased area of

skin wettedness and the subsequent rise in core temperature (Candas et al., 1979; Frye and Kamon, 1983; Alber-Wallerström and Holmér, 1985).

Numerous recommendations encourage athletes to ingest sufficient fluid in order to prevent a loss of >2% of their body weight during exercise (Sawka et al., 2007, 2015). This recommendation was based on laboratory studies demonstrating that a body mass loss of more than 2% impairs aerobic exercise performance in a thermally stressful environment (Montain and Coyle, 1992; Sawka et al., 2015). On the other hand, numerous publications have refuted the claim that aerobic exercise performance is degraded with increased (>2%) dehydration (Noakes, 2007; Goulet, 2011, 2013; Wall et al., 2013). A meta-analysis by Goulet (2011) had highlighted that exercise-induced dehydration by up to 4% of body weight loss does not alter exercise performance. Instead, drinking according to the dictate of thirst will potentially improve exercise performance (Noakes, 2007). While the debate on the efficacy of fluid ingestion in minimizing the decline of aerobic exercise performance in ambient heat continues, the value of fluid ingestion during humid conditions is not well understood.

Early reports of an attenuation in hyperthermia with fluid ingestion are based on studies conducted in a hot and relatively dry (~50% RH) environment with the large E_{\max} allowing for efficient sweat evaporation (Montain and Coyle, 1992; Below et al., 1995). By contrast, in a humid environment with a lower E_{\max} , sweat will drip off the skin instead of evaporating (Candas et al., 1979; Frye and Kamon, 1983; Alber-Wallerström and Holmér, 1985) and so “ineffective” fluid loss through sweating is much greater. Therefore, the proportion that fluid ingestion contributes to thermoregulation via increasing sweating rate would also be less. These observations on the sweating response and sweating efficiency might question the efficacy of fluid ingestion during prolonged exercise in a warm-humid condition. To date, Kay and Marino (2003) have reported that self-paced exercise performance in a hot-humid environment was not improved with fluid ingestion. In addition, Lee et al. (2010), had reported that fluid ingestion did not alter thermoregulatory responses during field-based experiments among military personnel. Therefore, the purpose of this study was to investigate the efficacy of ingesting plain water in attenuating the thermoregulatory and circulatory stress during prolonged moderate-high intensity exercise under a warm-dry and warm-humid environment and its consequences on running performance.

MATERIALS AND METHODS

Participants

Nine well-trained, unacclimatised male runners who regularly participated in middle and long distance running events volunteered for this study. The subjects' characteristics were (mean \pm SD): age, 32 ± 4 years; height, 183 ± 6 cm; weight, 72 ± 4 kg; body surface area, 1.9 ± 0.1 m²; percent body fat, $11 \pm 6\%$; $\text{VO}_{2\max}$, 62 ± 5 ml kg⁻¹ min⁻¹. Participants were briefed on the experimental protocol and testing procedures

before providing written informed consent to participate. The University of Sydney Human Ethics Committee approved the experimental protocol for this study (Ref. No: 99/05/46), which conformed to the current Declaration of Helsinki guidelines. All experiments were conducted in a purpose-built environmental chamber during the autumn and winter season in Sydney, Australia. The average outdoor temperature during the experimental trials was 20 to 22°C in autumn and 7 to 10°C during winter. None of the participants were accustomed to exercise in the conditions simulated in this study.

Preliminary Testing and Familiarization

Each participant visited the environmental chamber and was familiarized with the exercise protocol, equipment, and measurement procedures used in the study. This was followed by a preliminary testing session during which anthropometric measures, including body height, weight, composition, and surface area were determined. Body surface area was calculated using the method of DuBois and DuBois (1916). Percent body fat was measured using the hydrodensitometry underwater weighing technique described by Siri (1961).

Each participant then performed a running economy test followed by a maximal exercise test. The running economy test required each subject to run at four submaximal velocities of 10, 12, 14, and 16 km h⁻¹, respectively, for 4 min per stage. On completion of the submaximal running, participants engaged in an active recovery, where they walked for 5 min at a speed of 5 km h⁻¹. This was followed by a graded exercise test to determine maximal oxygen uptake ($\text{VO}_{2\max}$), where the participant ran at a fixed speed of 12 km h⁻¹ with the treadmill gradient elevated by 2% every 2 min until volitional fatigue was attained. Oxygen consumption and heart rate were taken throughout the exercise duration. Post-exercise, a linear regression line was plotted between submaximal steady state oxygen consumption and treadmill velocity to determine the participant's running speed that elicited an intensity of 70% $\text{VO}_{2\max}$, which was used for the individual's experimental trials for each subsequent testing session. The preliminary and familiarization sessions were conducted in a thermoneutral environment (20°C, 40% RH).

Prior to reporting to the laboratory for testing, each participant was reminded to refrain from heavy exercise and alcohol consumption the day before testing and to avoid caffeine consumption for 12 h before the test. They were also told to maintain a similar training routine throughout the duration of the study. Participants were asked to keep a 24 h food diary before testing and replicate their diet before subsequent visits to the laboratory. To ensure that participants were euhydrated at the onset of exercise, they were asked to ingest 6 ml of water per kg lean body mass at 2 h intervals (excluding when asleep) during the day before the test as well as the morning of testing.

Experimental Protocol

Each participant ran for 60 min at a speed eliciting an intensity of 70% $\text{VO}_{2\max}$. Immediately thereafter, participants continued running at the same velocity while the treadmill gradient was elevated by 2% every 2 min until volitional exhaustion, defined as the point at which participants could no longer maintain the

pace of the treadmill, upon which the test was terminated. During the 60 min submaximal exercise, the treadmill was briefly stopped (1 min) at 30 and 60 min to allow time for the determination of the participant's body mass. The air speed set for each trial was matched to the individual running speed and thus simulated the effect of air resistance on a calm day outdoors. Each test session was separated by a week apart to minimize acclimation effects as well as to provide adequate recovery.

The experiment started with participants being randomly assigned to perform the running exercise without ingesting water (non-fluid ingestion trial; NF) across two environmental conditions of warm-dry (WD: 30°C and 24% RH) and warm-humid (WH: 30°C and 71% RH). Following the NF trial, participants were randomly assigned to the same exercise regimen while being allowed to ingest water (fluid ingestion trial; FI) across the WD and WH conditions. The NF trials enabled the determination of each individual subject's sweat rate within the specific environmental condition for the calculation of the fluid volume needed to replace 80% of sweat loss during the FI trial.

At least 1 week elapsed between each of the subject's four scheduled visits to the laboratory, in order to minimize any residual effects from the previous visit. To avoid any psychological apprehension, subjects were not informed of the environmental condition on the day of the test.

Fluid Ingestion Protocol

In the FI trial, water amounting to 80% of total sweat loss from the NF trial was equally divided into five aliquots and provided to participants during submaximal exercise, starting at 10 min and followed at every 10 min interval with the final ingestion at 50 min. The volume of fluid ingestion in this study which was based on 80% of total sweat loss was about 1 L (ranged between 1.12 and 0.96 L). This amount of fluid ingestion was consistent with the water requirements for an hour of running exercise at a similar metabolic rate in a hot and humid condition as previously described by Sawka and Pandolf (1990). The temperature of the water ingested at each time interval was maintained at 15°C.

Measurements

As an index of core temperature, rectal temperature (T_{re}) was measured by a thermistor probe (YSI 400 series; Mallinckrodt Medical, St. Louis, MO, United States) inserted 12 cm beyond the anal sphincter and data were recorded on a portable data logger (T-logger; The University of Sydney, Sydney, NSW, Australia). Skin temperature was measured at four different sites (left shoulder, left chest, right mid-thigh, and right mid-shin) using thermistor probes (YSI 409 Series). Rectal and skin temperatures were sampled at 1 min intervals. Weighted mean skin temperature (\bar{T}_{sk}) was calculated from the four sites (Ramanathan, 1964). Mean body temperature (T_b) was calculated using the equation developed by Colin et al. (1971) as $T_b = 0.79 (\bar{T}_{re}) + 0.21 (\bar{T}_{sk})$. Both rectal and skin thermistor probes used in the study were calibrated before and after the study in a water bath with temperature ranging from 15 to 50°C; calculated accuracies were ± 0.05 and ± 0.01 , respectively.

Expired respiratory gas was obtained using the Douglas bag method at rest and at every 10 min intervals of the submaximal

exercise and analyzed for fractions of O_2 and CO_2 concentration using gas sensors and analyzers (O_2 analyzer, Ametek S-3A/I and CO_2 analyzers, Ametek CD-3A, Applied Electrochemistry Ametek Inc., Thermox Instruments Division). The gas analyzers were calibrated with known calibration gasses prior to each testing session.

Cardiac output during the steady state exercise phase was determined at 10, 30 and 60 min by the CO_2 rebreathing method (Collier, 1956). Stroke volume was then calculated using the Fick equation. Heart rate was continuously monitored telemetrically via a Polar transmitter-receiver (Polar Vantage XL, Polar Electro, Kempele, Finland) and recorded at rest, during submaximal exercise at 5 min intervals and at volitional exhaustion.

Whole body sweating rate (WBSR) during steady state exercise was calculated as the difference in pre and post 60 min exercise nude body weight with correction for respiratory moisture loss (Mitchell et al., 1972) and incorporating volume of fluid consumed. Participant's perception of the difficulty of the exercise effort was recorded based on the 6–20 point RPE scale (Borg, 1982) and was recorded at 10 min intervals.

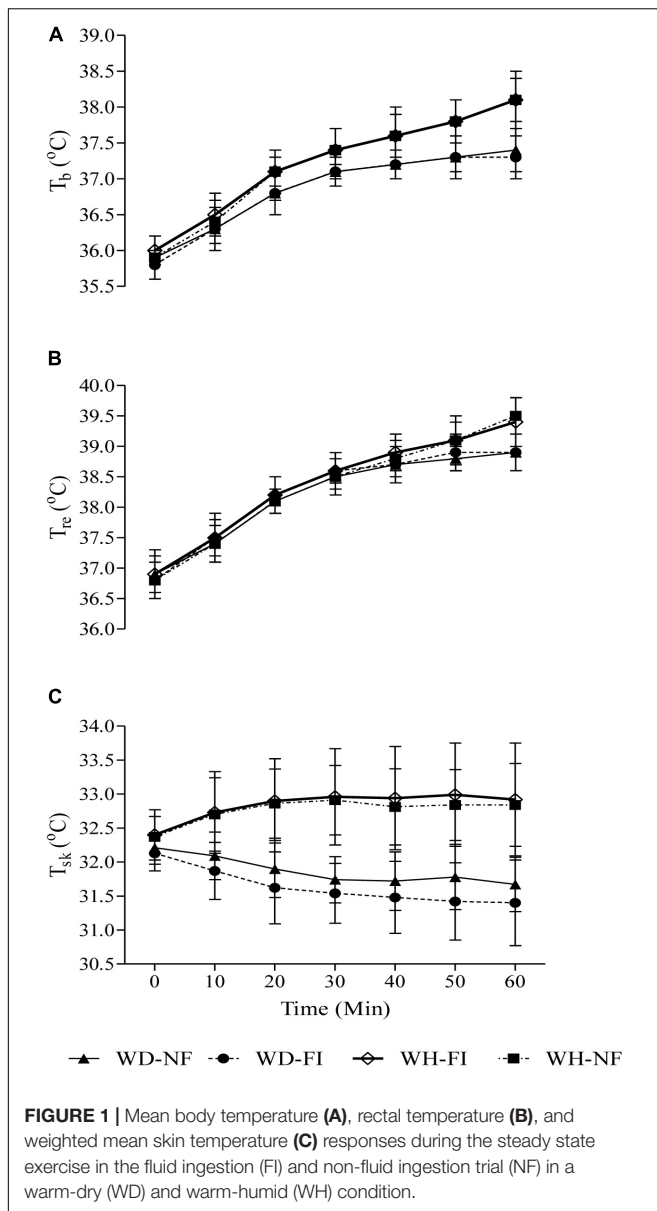
A partitionial calorimetry software program developed by Atkins and Thompson (2000) was used to estimate the magnitude and avenue of body heat gain and heat loss during each of the prolonged exercise bouts. Partitionial calorimetry calculations were based on measures taken within the last 30 min of submaximal exercise of each trial. Evaporative heat loss in $W m^{-2}$ was estimated from the classic body heat balance equation of $E = M - W$ (Eres + Cres) – R – C – S, where M is metabolic heat production, W is external work, Eres + Cres is heat transfer via the respiratory tract, R is radiative heat loss, C is convective heat loss, and S is body heat storage. S was estimated using the following equation: $[3474 \times wt \times (T_b \text{ final} - T_b \text{ initial}) \times t^{-1}] \times A_D^{-1}$ ($W m^{-2}$), where 3474 is the average specific heat of body tissue ($J kg^{-1} ^\circ C^{-1}$), wt is body mass (kg), T_b is mean body temperature ($^\circ C$), t is exercise time (sec), and A_D is body surface area (m^2). Estimation of tissue heat conductance (K) was derived from the method of Davies (1979) as $K = H_{sk} / [(T_{re} - \bar{T}_{sk}) * A_D]^{-1}$ ($W m^{-2} ^\circ C^{-1}$), where H_{sk} is the heat dissipated from the skin (E + R + C), T_{re} is rectal temperature, \bar{T}_{sk} is mean skin temperature, and A_D is the DuBois body surface area in m^2 . Convective and radiative heat loss in $W m^{-2}$ were estimated using the equations found in Fanger (1970) and McIntyre (1980), respectively.

Blood Sampling and Analysis

Venous blood was sampled at rest and 10, 30, and 60 min of submaximal exercise and were immediately stored for analysis of hematocrit and hemoglobin (CIBA-Corning 800 Series). Analysis for hematocrit percentage and hemoglobin concentration were analyzed in triplicate and used to estimate percentage changes in resting plasma volume based on the method of Dill and Costill (1974).

Statistical Analysis

A two way (humidity x fluid treatment) repeated measures analysis of variance (ANOVA) was used to compare the differences between the means of the data measured in this study.



If a primary significant difference was observed, the *post hoc* Tukey's paired *t*-test with a Bonferroni correction for multiple comparisons was used to detect where the differences occurred. A Huynh-Feldt correction was applied to adjust the degrees of freedom when the test of sphericity was significant. Statistics were analyzed using SPSS statistical software (V22.0, Chicago, IL, United States), with statistical significance accepted at an α level of 0.05. Data reported are presented as means \pm SD.

RESULTS

Steady-State Exercise

Responses of T_{re} , T_{sk} and T_b can be seen in **Figure 1**. ANOVA revealed no significant main effect of fluid ingestion on T_{re} , T_{sk}

or T_b (all $p > 0.2$), however, a main effect of humidity was observed for all (all $p < 0.02$) during the steady state exercise. At the end of steady state exercise, a significantly higher T_b was recorded in WH as compared with WD for both FI ($38.0^\circ\text{C} \pm 0.4$ vs. $37.3^\circ\text{C} \pm 0.3$; $p < 0.01$) and NF trials ($38.1^\circ\text{C} \pm 0.4$ vs. $37.4^\circ\text{C} \pm 0.3$; $p < 0.01$).

WBSR differed between fluid trials as a function of ambient humidity (fluid ingestion \times ambient humidity: $p < 0.01$), such that NF in WD resulted in similar WBSR during exercise when compared with FI ($24.5 \text{ g min}^{-1} \pm 2.3$ vs. $24.3 \text{ g min}^{-1} \pm 2.3$, respectively; $p = 0.6$), whilst in WH WBSR was significantly higher in FI compared to NF ($26.1 \text{ g min}^{-1} \pm 4.8$ vs. $22.7 \text{ g min}^{-1} \pm 3.4$, respectively; $p = 0.01$). Percent body weight deficit was significantly larger in the NF as compared with the FI trial ($p < 0.01$), within both WD (2.0 ± 0.2 vs. 0.4 ± 0.1 , respectively) and WH environments (1.8 ± 0.3 vs. 0.6 ± 0.2), although ambient humidity had no effect ($p = 0.6$). The change in PV (measured between 10th and 60th minutes) was similar between FI and NF trials ($2.1 \pm 1.8\%$ vs. $4.2 \pm 3.0\%$, respectively; $p = 0.1$) although the change in PV differed as a consequence of ambient humidity ($2.0 \pm 1.4\%$ vs. $4.2 \pm 2.9\%$, respectively; $p = 0.04$).

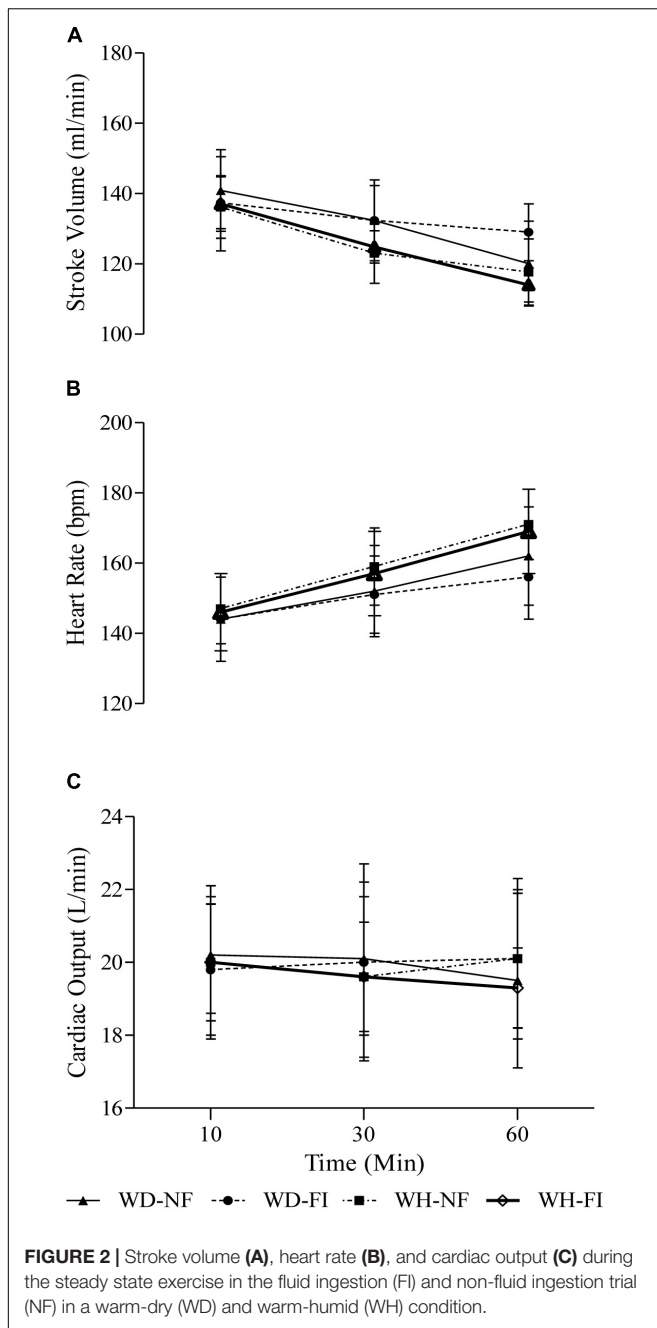
There was no significant effect of fluid ingestion on RPE during exercise ($p = 0.4$), however, ambient humidity significantly affected RPE ($p < 0.01$). Participants reported exercise as being harder to perform in the WH environment as compared with WD, respectively, for both NF (16 ± 2 vs. 14 ± 2 , respectively) and FI trials (15 ± 2 vs. 13 ± 2 , respectively).

ANOVA revealed a significantly higher S during NF than FI ($6 \pm 7 \text{ W m}^{-2}$; $p = 0.03$) and WH than WD ($22 \pm 9 \text{ W m}^{-2}$; $p < 0.01$). M was significantly higher during WH than WD ($23 \pm 27 \text{ W m}^{-2}$; $p = 0.03$) with no effect of fluid ingestion ($p = 0.3$), whilst E was significantly higher during FI than NF ($14 \pm 15 \text{ W m}^{-2}$; $p = 0.03$) with no effect of ambient humidity ($p = 0.2$). K and $C + R$ were both higher during WH than WD (22 ± 12 and $25 \pm 8 \text{ W m}^{-2}$, respectively; both $p < 0.01$) with no effect of fluid ingestion (both $p > 0.2$).

The circulatory responses can be seen in **Figure 2**. ANOVA revealed a significant effect of fluid ingested ($p = 0.01$) and ambient humidity ($p < 0.01$) on heart rate, such that on average FI reduced HR by $2 \pm 2 \text{ beats min}^{-1}$ whilst on average the WH environment increased HR by $7 \pm 5 \text{ beats min}^{-1}$. Stroke volume was lower during WH than WD by $7 \pm 5 \text{ ml}$ ($p < 0.01$) with no effect of fluid ingested ($p = 0.7$). No effects of fluid ingestion or ambient humidity were observed on cardiac output (both $p > 0.5$).

Graded Exercise to Exhaustion

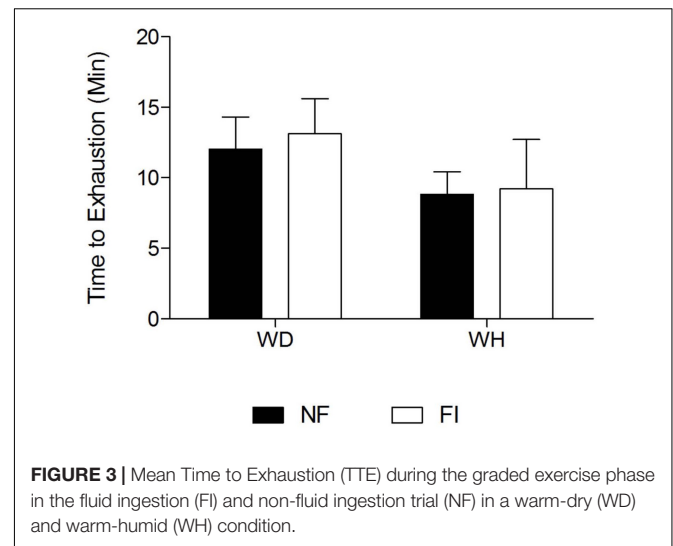
Fluid ingestion did not have any effect on the time to exhaustion within both conditions when compared to the no fluid ingestion trial as presented in **Figure 3**. The exercise capacity during the graded exercise test within both NF and FI trials was significantly shorter in WH than WD ($9.1 \pm 2.8 \text{ min}$ vs. $12.7 \pm 2.4 \text{ min}$, respectively; $p = 0.01$). At exhaustion, T_{re} was not significantly different between the FI and the NF or WD and WH trials (global mean $39.3 \pm 0.5^\circ\text{C}$; all $p > 0.2$). Similarly, T_{sk} (global mean $32.1 \pm 1.2^\circ\text{C}$; all $p > 0.1$) and heart rate (global mean $182 \pm 1 \text{ beats min}^{-1}$; all $p > 0.5$).



min^{-1} ; all $p > 0.3$) were also not significantly different between any condition.

DISCUSSION

To our knowledge, this is the first study to directly examine the efficacy of fluid ingestion in attenuating thermoregulatory strain during prolonged exercise in two different environments: warm-dry (WD) and warm-humid (WH). This study was designed to simulate real outdoor running where runners would experience a high level of air velocity during their



intense running exercise, thus promoting convective cooling. The main finding of this study was that fluid ingestion during exercise in warm-humid and warm-dry conditions did not attenuate hyperthermia during exercise. Consequently, the time to exhaustion in the subsequent graded exercise session was not improved during the FI trial. Under this current scenario, fluid ingestion was seen not to provide any beneficial effect on thermoregulatory and circulatory responses and consequently on exercise performance.

This study had also postulated that the level of percent body mass deficit did not influence the magnitude of physiological strain during submaximal exercise and running performance during the graded exercise test. In the present experiment, the percent body mass deficit during the no fluid (NF) trial, in the WD and WH condition was 2 and 1.8%, respectively.

Therefore, the current study provides evidence that thermoregulatory responses during the submaximal exercise could have been influenced by the level of RH as previously reported (Maughan et al., 2012; Moyon et al., 2014; Che Muhamed et al., 2016) and not associated with the level of dehydration as advocated in numerous publications (Noakes, 2007; Goulet, 2011, 2013; Wall et al., 2013). This current observation is contrary to earlier observations conducted in drier heat that fluid ingestion attenuates hyperthermia during exercise in a heat stressful condition (Montain and Coyle, 1992; Below et al., 1995; Armstrong et al., 1997; Sawka et al., 2012).

The findings of the present study provide new insight in better understanding the efficacy of fluid ingestion on thermoregulatory responses in two different heat stress conditions (i.e., warm-dry and warm-humid). Numerous earlier guidelines on fluid ingestion (Casa et al., 2000; Sawka et al., 2007, 2015) do not specifically comment on the efficacy of fluid ingestion in attenuating heat strain under different thermal profiles. The present study demonstrated that in the humid condition with a reduced E_{max} , there is an increase in the amount of sweat dripping off the skin instead of

being evaporated (Candas et al., 1979; Frye and Kamon, 1983; Alber-Wallerström and Holmér, 1985). Therefore, the notion that fluid ingestion during exercise heat stress will attenuate hyperthermia by enhancing sweat evaporation does not take place in a humid condition. Instead, the physical characteristics of the environment would dictate the efficacy of fluid ingestion in attenuating hyperthermia. This was evident in the current study where a higher WBSR recorded in the humid condition did not result in a lowering of skin temperature that would have increased the gradient between the core and the skin, thus promoting heat transfer and lowering body heat storage.

Instead, skin temperature remained elevated in the humid condition due to the lower E_{\max} . The current observation that fluid ingestion was not effective in attenuating thermal strain and improving endurance performance was consistent with several earlier studies. Kay and Marino (2003) reported that water ingestion failed to improve self-paced exercise performance in a hot-humid environment. In addition, several other field-based studies conducted in hot-humid environments had reported no association between fluid intake and percent dehydration with core temperature response (Byrne et al., 2006; Lee et al., 2010). This further supports numerous reports that have proposed that a certain amount of dehydration is tolerable during running exercise where thermoregulatory function is not significantly impaired until a critical level of dehydration has accrued. Davies and Thompson (1986) recorded a 5.5% body weight deficit at the end of 4 h strenuous running exercise with core temperature at no point exceeding 39.3°C. Based on this evidence it is suggested that the level of dehydration may have a relatively small influence on core temperature during exercise if the subjects are euhydrated at the onset of exercise. Several more recent findings have consistently demonstrated that a body mass loss of up to 4% has been well tolerated during prolonged exercise and did not appear to have any detrimental effect on exercise performance (Zouhal et al., 2011; Del Coso et al., 2014; Hoffman and Stumpfle, 2014).

Guidelines on fluid ingestion during exercise have been formulated mainly based on the notion that dehydration impairs endurance exercise performance (Casa et al., 2000; Sawka et al., 2007, 2015) as dehydration level exceeds 2% of body mass (Montain and Coyle, 1992; Below et al., 1995; Ebert et al., 2007; Merry et al., 2010; Sawka et al., 2012; Cheuvront and Kenefick, 2014). On the contrary, several other researchers have consistently shown that in well-trained cyclists who were euhydrated at the onset of exercise and went on to cycle for up to 60 min, their cycling performance was not compromised within an ambient temperature of up to 33°C and 60% RH despite their dehydration level reaching 4% (Noakes, 2007; Goulet, 2011, 2013; Wall et al., 2013).

The level of plasma volume in this study was well maintained during exercise despite the difference in dehydration rate between the NF and FI trial. This observation is consistent with several earlier studies that have shown plasma volume can be partially defended at an even larger percentage of body weight deficit during intense (65–75% $\dot{V}O_2$ max) running exercise (Costill et al., 1970; Sawka et al., 1980; Kolka et al., 1982;

Gass et al., 1983; Davies and Thompson, 1986). The magnitude of dehydration in the present study was much smaller than these earlier studies that have reported a stable plasma volume. For instance, Costill et al. (1970), Sawka et al. (1980), and Kolka et al. (1982) recorded a stable plasma volume during exercise at an even higher percent body weight deficit of 4%, 4.8–6.8% and 7%, respectively. The ability to defend the level of plasma volume in light of increasing dehydration level has been associated with the participants euhydration level at the onset of exercise as well as the ability to shift body water interstitially. In addition, the release of water from glycogenolysis, metabolic water production and the redistribution of water from inactive skeletal muscle has also been reported to assist in maintaining plasma volume during exercise (Pivarnik et al., 1984).

CONCLUSION

In conclusion, the current study has demonstrated that the efficacy of fluid ingestion in attenuating thermoregulatory and circulatory stress during prolonged exercise is potentially dependent on the physical characteristics of the environment. Runners who started exercise in a euhydrated state did not gain any ergogenic benefit for the 1 h of exercise from fluid ingestion. Runners in our study who had experienced dehydration ranging from 1.8% to 2.0% did not improve their performance. This was in contrast with the well-established notion that dehydration of up to 2% of body weight loss will increase thermoregulatory and circulatory stress leading to impairment in endurance exercise performance (Casa et al., 2000; Sawka et al., 2007, 2015). Instead, our finding was in agreement with earlier reports of Kay and Marino (2003) as well as Lee et al. (2010) which failed to observe any ergogenic benefit of fluid ingestion on exercise performance in humid heat.

This study highlights the need to consider the characteristics of the environment in evaluating the efficacy of fluid ingestion in attenuating thermoregulatory and circulatory strain during exercise and its implications on exercise performance. Future recommendations relating to fluid ingestion during exercise should now discuss the impact of environmental conditions.

ETHICS STATEMENT

The University of Sydney Human Ethics Committee approved the experimental protocol for this study (Ref. No: 99/05/46), which conformed to the current Declaration of Helsinki guidelines.

AUTHOR CONTRIBUTIONS

ACM, SS, and MT involved in the design and implementation of the study. TM and HY assisted the corresponding author in the data analysis. All authors contributed in preparing the manuscript with HY preparing the figures.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

The reviewer ZS declared a past co-authorship with one of the authors TM to the handling Editor.

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Nine-, but Not Four-Days Heat Acclimation Improves Self-Paced Endurance Performance in Females

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OPEN ACCESS

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 04 December 2018

Accepted: 16 April 2019

Published: 16 May 2019

Citation:

Kirby NV, Lucas SJE and
Lucas RAI (2019) Nine-, but Not
Four-Days Heat Acclimation Improves
Self-Paced Endurance Performance
in Females. *Front. Physiol.* 10:539.
doi: 10.3389/fphys.2019.00539

Although emerging as a cost and time efficient way to prepare for competition in the heat, recent evidence indicates that “short-term” heat acclimation (<7 days) may not be sufficient for females to adapt to repeated heat stress. Furthermore, self-paced performance following either short-term, or longer (>7 days) heat acclimation has not been examined in a female cohort. Therefore, the aim of this study was to investigate self-paced endurance performance in hot conditions following 4- and 9-days of a high-intensity isothermic heat acclimation protocol in a female cohort. Eight female endurance athletes (mean \pm SD, age 27 ± 5 years, mass 61 ± 5 kg, $\text{VO}_{2\text{peak}}$ 47 ± 6 ml·kg⁻¹·min⁻¹) performed 15-min self-paced cycling time trials in hot conditions (35°C, 30%RH) before (HTT1), and after 4-days (HTT2), and 9-days (HTT3) isothermic heat acclimation (HA, with power output manipulated to increase and maintain rectal temperature (T_{rec}) at $\sim 38.5^\circ\text{C}$ for 90-min cycling in 40°C, 30%RH) with permissive dehydration. There were no significant changes in distance cycled ($p = 0.47$), mean power output ($p = 0.55$) or cycling speed ($p = 0.44$) following 4-days HA (i.e., from HTT1 to HTT2). Distance cycled ($+3.2\%$, $p = 0.01$; $+1.8\%$, $p = 0.04$), mean power output ($+8.1\%$, $p = 0.01$; $+4.8\%$, $p = 0.05$) and cycling speed ($+3.0\%$, $p = 0.01$; $+1.6\%$, $p = 0.05$) were significantly greater in HTT3 than in HTT1 and HTT2, respectively. There was an increase in the number of active sweat glands per cm² in HTT3 as compared to HTT1 ($+32\%$; $p = 0.02$) and HTT2 ($+22\%$; $p < 0.01$), whereas thermal sensation immediately before HTT3 decreased (“Slightly Warm,” $p = 0.03$) compared to ratings taken before HTT1 (“Warm”) in 35°C, 30%RH. Four-days HA was insufficient to improve performance in the heat in females as observed following 9-days HA.

Keywords: heat acclimation, acclimatization, thermoregulation, female, women, exercise physiology, sports performance

Abbreviations: %HR_{max}, percentage of age-estimated maximum heart rate; η_p^2 , partial eta-squared; AUC, area under the curve; BSA, body surface area; HA, heat acclimation sessions; HIIT, high-intensity interval training; HR, heart rate; HTT, time trial in hot conditions; IUD, intrauterine device; NBM, nude body mass; OCP, oral contraceptive pill; SR_{BSA} , estimated sweat rate relative to body surface area; STHA, short-term heat acclimation; $\text{Sweat Loss}_{\%BM}$, estimated sweat loss as a percentage of body mass; T_{rec} , rectal temperature; T_{sk} , weighted mean skin temperature; $\text{VO}_{2\text{peak}}$, maximal aerobic capacity; W, watts.

INTRODUCTION

Hot ambient temperatures and elevated humidity are known to negatively impact endurance exercise performance (Tattersson et al., 2000; Périard et al., 2011). Heat acclimation is an effective strategy to drive favorable physiological adaptations, thereby reducing athletic performance impairments caused by these challenging environments (Sawka et al., 2011; Périard et al., 2015; Racinais et al., 2015). Heat acclimation typically consists of repeated daily heat stress exposures, with exposure durations commonly lasting between 60 and 90 min. Traditionally, 10 days of heat exposure are undertaken to elicit the heat acclimation phenotype and improve endurance performance in the heat (Armstrong and Maresh, 1991; Lorenzo et al., 2010; Sawka et al., 2011), though 75–80% of physiological adaptations occur in the first 4–7 days of heat acclimation in male cohorts (Pandolf, 1998; Shapiro et al., 1998). Based on this, Garrett et al. (2009) first demonstrate meaningful performance improvements following just 5 days of isothermic heat acclimation, termed “short-term heat acclimation” (STHA).

STHA has since been defined as being <7 days in length (Garrett et al., 2011), and is promoted as a cost and time efficient option for athletes preparing for competition in the heat. Successful STHA lasting 4–7 days in male cohorts has been well documented (Petersen et al., 2010; Fujii et al., 2012; Chen et al., 2013; Garrett et al., 2012, 2014; Best et al., 2014; Costa et al., 2014; Gibson et al., 2015; Mee et al., 2015; Racinais et al., 2015; Guy et al., 2016; James et al., 2016; Willmott et al., 2016). However, few studies to date have examined STHA effects in female cohorts. It was initially shown that there were no sex differences (4 females vs. 4 males) in adaptations to 10-days heat acclimation when aerobic fitness and surface area to mass ratios were matched (Avellini et al., 1980). However, Mee et al. (2015) more recently reported a significant sex difference in the time course of heat acclimation (i.e., 5- vs. 10-days), challenging past assumptions. In this study Mee et al. (2015) found that following STHA females did not exhibit a lower resting core temperature, or an attenuated rise in core temperature and heart rate (HR) when exercising at a fixed workload, critical requirements in demonstrating the heat acclimation adaptation. However, the male cohort successfully attained these adaptations following the same protocol. Mee et al. (2018) then demonstrated that a longer daily heat exposure (achieved via 20-min of sitting in sauna suits in 50°C, 30%RH immediately before 90-min isothermic heat acclimation) successfully induced heat adaptations in a female cohort following STHA (5-days). The authors therefore concluded that females require either a longer daily heat exposure (Mee et al., 2018), or a greater number of heat exposures (Mee et al., 2015) to elicit favorable physiological adaptations.

It is unclear if females can achieve meaningful performance improvements following STHA or if a longer heat acclimation period is needed. Sunderland et al. (2008) reported a 33% improvement in distance run during a repeated shuttle run performance test following STHA in a female cohort as well as a reduced rate of rise in rectal temperature (T_{rec}). The authors attributed performance improvements to the high-intensity

intermittent exercise performed during the acclimation sessions, a strategy implemented by Pethick et al. (2018) to successfully induce plasma volume expansion in a female cohort following 5-days high-intensity heat acclimation. Therefore, the addition of high-intensity interval training (HIIT) may offer a means to increase effectiveness of STHA in a female cohort. However, the performance test employed by Sunderland and colleagues was a time to exhaustion trial, which is a less reliable test and subject to greater variation than self-paced performance tests (Hopkins et al., 2001; Borg et al., 2018). Self-paced performance outcomes and their improvements following heat acclimation have only been documented in male (Garrett et al., 2012; Keiser et al., 2015; Racinais et al., 2015; Guy et al., 2016; Wingfield et al., 2016) or mostly male (Lorenzo et al., 2010) cohorts, and remain to be investigated in females. Such information is timely for female athletes competing at upcoming international competitions in hot climates, such as the 2020 Olympic Games in Tokyo and the 2019 IAAF World Athletics Championships in Doha. Currently, female athletes must either depend on conflicting literature or fill knowledge gaps with information inferred from male cohorts.

Therefore, the aim of the present study was to investigate self-paced endurance performance in hot conditions following 4-days (STHA), and 9-days of a high-intensity isothermic heat acclimation protocol in a female cohort. It was hypothesized that females would not exhibit performance improvements in self-paced exercise following STHA, as previous studies indicate that 4-days of 90-min heat acclimation is unlikely to be a sufficient stimulus for the thermoregulatory and cardiovascular adaptations necessary for performance improvements in the heat. We further hypothesized that performance improvements in power output and time trial distance would occur following 9-days heat acclimation.

MATERIALS AND METHODS

General Overview and Design

This study was approved by the University of Birmingham Ethics Committee, and conformed to the standards set by the Declaration of Helsinki 2013. All participants were informed of the experimental procedures and possible risks involved in the study before their written consent was obtained. Each participant completed a general exercise questionnaire and a menstrual cycle questionnaire (detailing the day their menstrual cycle commenced, premenstrual symptoms, and contraceptive medication or devices) to ascertain what phase of their cycle they were in for each time trial. All experimental procedures were completed in the environmental chamber (TIS Services, Hampshire, United Kingdom) in the School of Sport, Exercise and Rehabilitation Sciences building at the University of Birmingham. Participants performed all heat acclimation and testing sessions at the same time of day (± 2 h), and at similar times to their normal training sessions so as not to disrupt their normal circadian rhythms (Reilly and Brooks, 1986). This included mornings, afternoons, or evenings. Participants were familiarized with the 15-min time trial performance tests in cool conditions (15°C, 30% RH) on three occasions, with the

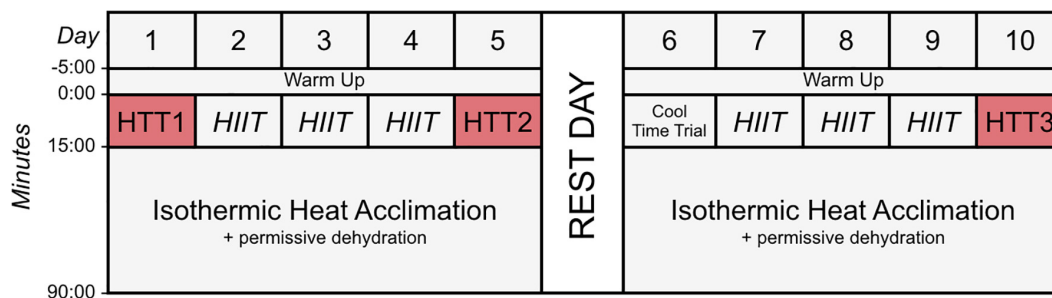


FIGURE 1 | Schematic diagram of the time trials and heat acclimation sessions (HA). Time trials were conducted in hot conditions [HTT; 35°C, 30% relative humidity (RH)], before HA (HTT1), after 4-days (HTT2) and after 9-days (HTT3) HA. On days 2–4 and 7–9, participants completed 15-min of high-intensity intervals (HIIT), where maximum effort was given for 15-s, with 45-s of active recovery. Participants then undertook 75-min of isothermic heat acclimation (where exercise intensity was manipulated to increase and maintain rectal temperature at ~38.5°C; 40°C, 30%RH) with permissive dehydration. There was one rest day following 5-days HA. Cool Time Trial refers to a 15-min cycling time trial in cool conditions (15°C, 30% RH), which was part of a larger dataset that are not reported herein.

final occasion 48 h prior to beginning the protocol. Participants performed a 15-min cycling time trial in hot (35°C, 30% RH) conditions pre-acclimation, following 4-days (STHA), and following 9-days isothermic heat acclimation (HA). An overview of the hot time trials and HA sessions are displayed in **Figure 1**. This experiment was conducted in the United Kingdom during the months of February, April, May, and June, when mean ambient temperatures were below 20°C (exclusive of 3 days where the mean daily temperatures were 23, 24, and 27°C, respectively). The protocol was performed in addition to normal training sessions (i.e., weight training and normal conditioning such as swimming and running). Participants' activity was not restricted, except on the day prior to (no exhaustive exercise) or the day of (no other activity) time trials in hot conditions (HTTs). Participants were asked to refrain from alcohol and overly strenuous exercise outside of the laboratory 48 h before time trials.

Participants

Eight recreational endurance athletes aged 21–35 years volunteered for and completed this study. An additional participant volunteered, but dropped out due to relocation

after preliminary testing and was not included in the results. All participants were familiar with competitive, race-style endurance events, and trained 5 ± 1 days per week, averaging 9 ± 4 h of weekly endurance exercise training. Participants were eumenorrheic or using various forms of hormonal contraceptives (**Table 1**) and did not report any negative premenstrual symptoms that may have affected performance during time trials (Giacomoni et al., 2000). Participants had not previously undergone a heat acclimation protocol and had not been in hot conditions for the past 2 months. Participants also completed an incremental ($20 \text{ W} \cdot \text{min}^{-1}$ stages) exercise test on a cycle ergometer (Sport Excalibur, Lode, Groningen, Netherlands) to determine maximal aerobic capacity ($\text{VO}_{2\text{peak}}$), with expired air (Vyntus CPX, Jaeger, Wuerzburg, Germany) and HR (Polar Electro, Kempele, Finland) measured continuously. Personal characteristics are summarized in **Table 1**.

Heat Acclimation Sessions

A combination of HIIT, permissive dehydration and isothermic heat acclimation (Garrett et al., 2014; Sunderland et al., 2008) was used to construct a “high intensity” HA protocol. Participants

TABLE 1 | Participants' personal characteristics.

Participant	Age (years)	Height (cm)	$\text{VO}_{2\text{peak}}$ ($\text{ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$)	Body mass (kg)	Menstrual cycle/contraceptive	Day of menstrual cycle or pill taking phase on HTT1
1	32	168	40	54	OCP (Cilest)	15
2	28	168	45	59	Implant	N/A
3	25	176	43	65	Implant	N/A
4	23	165	43	55	EU	23
5	35	173	42	69	EU	17
6	32	172	53	63	IUD Coil	N/A
7	23	165	53	61	OCP (Yasmin)	11
8	21	174	54	61	Implant	N/A
Mean	27	170	47	61		

OCP, oral contraceptive pill user (pill brand); IUD Coil, copper coil intrauterine device; EU, eumenorrheic natural cycle.

voided their bladder upon arrival to the laboratory to provide a urine sample. Towel-dried, nude body mass (NBM) was recorded to 0.1 kg using digital scales (Seca 877, Seca, Hamburg, Germany) before and immediately after each session to estimate sweat loss. Conditions during HA sessions were set to 40°C, 30%RH with a fan-generated airflow of $\sim 3 \text{ m} \cdot \text{second}^{-1}$ facing participants. All heat acclimation sessions and time trials were completed using a cycle ergometer (Velotron, Racermate Inc., Seattle, WA, United States), which was calibrated according to manufacturer instructions for the chosen temperature and confirmed to exhibit $<1\%$ deviation from calibration settings before each use. Following a 5-min, self-selected warm-up, participants completed 15-min of high-intensity intervals, where participants were asked to give maximum effort for 15-s, with 45-s of active recovery. The aim of the high-intensity intervals was to rapidly increase T_{rec} . This was followed by an additional 75-min of continuous cycling at an intensity manipulated with the aim to further increase T_{rec} and maintain it at $\sim 38.5^\circ\text{C}$ (Patterson et al., 2004; Garrett et al., 2012), totalling 90-min HA plus 5-min warm up. On days that hot time trials (HTT) preceded HA sessions, the HTTs were used in place of the high-intensity intervals. On these test days, the temperature of the environmental chamber was immediately increased to 40°C, 30%RH following the time trial. There was one rest day following 5-days HA. Cool Time Trial refers to a 15-min cycling time trial in cool conditions (15°C , 30% RH), which was part of a larger dataset that are not reported herein. Power output, HR, and T_{rec} across HA sessions during STHA (days 1–4) and days 5–9 are depicted in **Figure 2**. Ratings of perceived exertion (RPE; Borg, 1982), thermal sensation, and thermal comfort were recorded at 15-min intervals during HA sessions. Participants were instructed to refrain from fluid consumption as much as could be tolerated during HA sessions to induce the added stressor of dehydration (permissive dehydration; Garrett et al., 2014). Fluid consumed ($295 \pm 235 \text{ ml}$ each session) was recorded by weighing water bottles to 0.001 L (Oertling, United Kingdom) before and after HA sessions, and was considered in the calculations of total body sweat loss. Heat acclimation involved 9 consecutive

days of HA sessions, except for 1 day of rest following STHA (**Figure 1**).

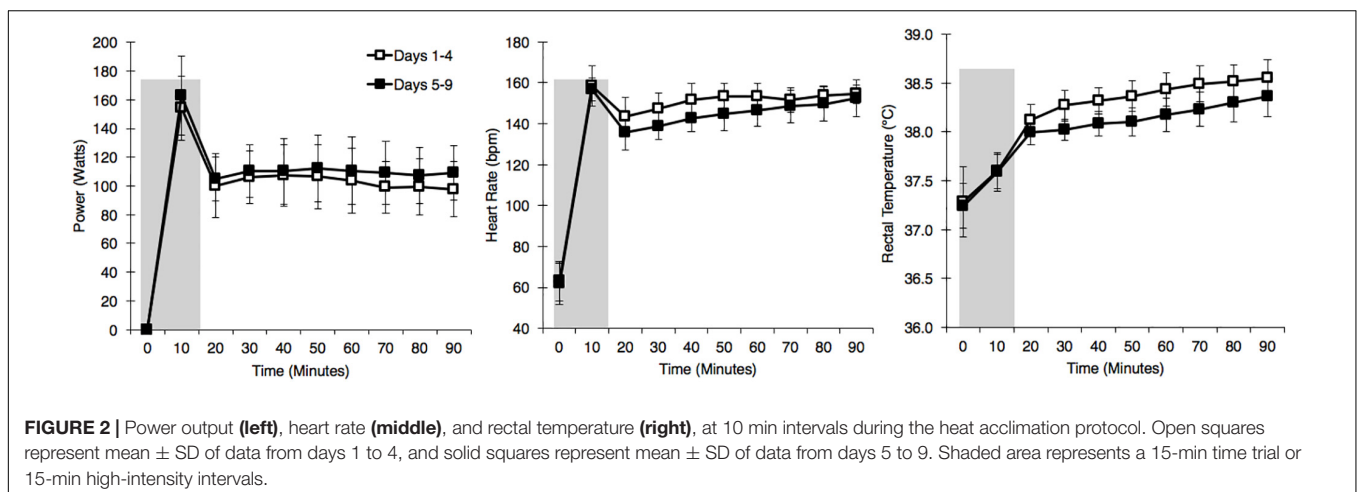
Hot Time Trials

Time trials were performed in hot conditions (35°C , 30%RH with a fan-generated airflow of $\sim 3 \text{ m} \cdot \text{second}^{-1}$ facing participants) on the 1st day of HA (Day 1; HTT1), and following 4-days HA (Day 5; HTT2), and 9-days HA (Day 10; HTT3). Participants were instructed to maintain normal hydration before each HTT, which was verified with a urine osmolality value of $\leq 700 \text{ mOsm} \cdot \text{kg}^{-1}$ (Sawka et al., 2007). Participants lay supine for 10 min of stabilization at room temperature prior to each trial to collect resting measures of T_{rec} and blood lactate.

Participants entered the environmental chamber and commenced a 5-min warm up at a self-selected pace, before completing a 15-min, self-paced cycling time trial. Power output and distance cycled were recorded continuously by the Velotron Coaching Software. Participants were aware of the time elapsed, as displayed by a stop-clock mounted to the handles of the cycle ergometer, however, they were blinded to any other physiological or performance feedback (i.e., HR, power output, distance cycled, etc.). Participants were given equal verbal encouragement by the same researchers at similar time points during the HTT. Free drinking was permitted during HTTs. RPE, blood lactate and sweat gland activity were recorded immediately following the HTTs. Ratings of thermal comfort and thermal sensation were reported inside the environmental chamber, preceding the warm-up for HTTs, as well as immediately after. Following HTTs, participants completed 5 min of self-paced active recovery before proceeding with the acclimation session for that day.

Measures

Urine osmolality was measured prior to each experimental session to assess hydration (Osmocheck, Vitech Scientific Ltd., West Sussex, United Kingdom). T_{rec} was measured using a rectal thermistor inserted 10 cm past the anal sphincter prior to beginning each experimental session (Mon-a-Therm, Covidien,



Mansfield, MA, United States). Weighted mean skin temperature (T_{sk}) was recorded using skin thermistors (Squirrel Thermal Couples, Grant Instruments, Cambridge, United Kingdom) attached to four sites: the mid-point of the right pectoralis major (T_{chest}), midpoint of the right biceps brachii (T_{arm}), right rectus femoris (T_{thigh}), and right gastrocnemius lateral head ($T_{lower\ leg}$). Skin and rectal thermistors were connected to a Squirrel Data Logger (Squirrel 2020 series, Eltek, Ltd., United Kingdom) and were recorded at 30-s intervals throughout HA sessions and HTTs. HR (Polar Electro, Kempele, Finland) was also recorded throughout each session. Power output and distance cycled were recorded by the Velotron Coaching Software (Velotron CS 2008, RacerMate Inc., Seattle, WA, United States). Blood lactate measures were taken from a finger-tip blood sample and immediately analyzed using a Lactate Plus analyzer (Lactate Plus, Nova Biomedical, Waltham, MA, United States). Active sweat glands were quantified using a modified-iodine paper technique with computer aided analysis (Gagnon et al., 2012). Samples were collected from the dorsal side of the thickest segment of the forearm. Thermal sensation and thermal comfort ratings were measured using 13-point and 10-point scales, respectively, which were modified from scales used by Gagge et al. (1967).

Data Analysis

Mean T_{rec} for the final 75 min of the session, which followed the 15-min high-intensity intervals, is represented by T_{rec75} . Maximum T_{rec} recorded during the session ($Max\ T_{rec}$) was used to calculate T_{rec} increase from rest (ΔT_{rec}). T_{sk} was calculated as a weighted average according to Ramanathan (1964):

$$T_{sk} = 0.3 \cdot (T_{chest} + T_{arm}) + 0.2 \cdot (T_{thigh} + T_{lower\ leg})$$

Estimated sweat rate relative to body surface area (SR_{BSA}) was calculated from changes in NBM pre- to post-session with considerations of water consumed body surface area [(BSA); calculated using the formula derived by Du Bois and Du Bois, 1916] and normalized for exercise time:

$$\text{Estimated sweat loss (g)} = (\text{pre-trial NBM} - \text{post-trial NBM}) + (\text{water bottle pre-trial} - \text{water bottle post-trial})$$

$$BSA(m^2) = 0.007184 \cdot (\text{height}^{0.725} \cdot \text{body mass}^{0.425})$$

$$SR_{BSA} (g \cdot h^{-1} \cdot m^{-2}) = (\text{estimated sweat loss}) \cdot (1\ h \cdot \text{exercise time}^{-1})^{-1} \cdot (BSA)^{-1}$$

Two values were obtained for measurements of resting blood lactate and an additional two values were obtained for blood lactate immediately following HTTs. The results were averaged to yield a single value for each time point (pre- and post-trial). Extreme outliers falling outside the physiological range were

excluded, and only the rational value was used (Goodwin et al., 2007; $n = 3$ incidences).

Power output (watts) was recorded each second during HTTs, and an average of each minute's power output was used to calculate area under the curve (AUC; Pruessner et al., 2003). AUC was also calculated for T_{rec} (recorded at 30-s intervals) during HTTs. All data were analyzed using SPSS statistical software (SPSS version 24.0.0, SPSS, Chicago, IL, United States). To assess performance and physiological differences during HA days 1–4 vs. days 5–9, a mean value was calculated for each participant across the aforementioned days, and analyzed using a repeated-measures one-way analysis of variance (ANOVA). Mean performance values during HTTs (i.e., power output and speed), AUC comparisons (power output and T_{rec}), distance cycled, and physiological measures between HTT1, HTT2, and HTT3, were also analyzed using a repeated-measures one-way ANOVA. Additionally, 1 min averages of power output were analyzed using a two-way repeated-measures ANOVA (3 HTT \times 15 time points). Normality of the data was assessed using Mauchly's test of sphericity, and Greenhouse–Geisser corrections were applied where assumptions of sphericity were violated. When a significant main effect was found, Bonferroni-corrected *post hoc* comparisons were made. Main effect sizes for both one-way and two-way ANOVAs were calculated using partial eta-squared (η_p^2), with $\eta_p^2 > 0.06$ representing a moderate difference and $\eta_p^2 > 0.14$ representing a large difference (Cohen, 1988). To assess ordinal data (i.e., RPE, thermal sensation and thermal comfort) differences during HA days 1–4 vs. days 5–9, and between HTT1, HTT2, and HTT3, Friedman's test was performed with *post hoc* analysis by Wilcoxon sign-rank tests. Absolute data are expressed as mean \pm standard deviation (SD) and mean within-subject differences are presented with 95% confidence limits (mean difference, 95% CL: lower limit, upper limit). Significance was set at $p < 0.05$ for each analysis. A power analysis indicated that eight participants were a sufficient sample size to detect an 8–10% difference in power output during time trial performance (as observed by Lorenzo et al., 2010; Keiser et al., 2015). This analysis used an accepted parameter of power ($\beta \geq 0.80$) at an α level of 0.05.

RESULTS

Heat Acclimation Sessions

Mean T_{rec75} (-0.2°C , $[-0.1, -0.3]$; $p < 0.01$) peak T_{rec} (-0.1°C , $[-0.1, -0.2]$; $p = 0.01$), and peak T_{sk} (-0.4°C , $[-0.1, -0.7]$; $p = 0.01$) were lower during HA sessions on days 5–9 as compared to HA sessions on days 1–4. Mean HR and percentage of age-estimated maximum heart rate (%HR_{max}) ($-5\ \text{beats} \cdot \text{minute}^{-1}$, $[-1, -10]$; $p = 0.03$, and -3% $[-1, -5]$; $p = 0.02$, respectively) were also lower during HA sessions on days 5–9 as compared to HA sessions on days 1–4. These physiological changes were present in spite of a significantly higher workload (i.e., power output) on days 5–9 as compared to HA sessions on days 1–4 ($-9\ \text{W}$, $[-3, -14]$, $p = 0.01$).

Participants' mean RPE, thermal sensation, and thermal comfort ratings across all HA sessions were not different ($p > 0.05$) and equalled 15 ± 2 ("Hard"), 10 ± 1 ("Hot"), and 5 ± 2 ("Uncomfortable"), respectively. There were no changes in ΔT_{rec} or sweat loss ($p > 0.05$). Results of HA sessions are summarized in **Table 2**.

Hot Time Trials

There was a large ($\eta_p^2 = 0.55$) and significant ($p < 0.01$) main effect of acclimation on distance cycled during time trials in hot conditions (**Figure 3**). *Post hoc* analysis indicated that distance increased by 3.2% (+240 m, [+70, +420]; $p = 0.01$) from HTT1 to HTT3, and by 1.8% (+140 m, [+10, +270]; $p = 0.04$) from HTT2 to HTT3 (**Table 3**). There was no difference in distance cycled from HTT1 to HTT2 (+100 m, [-100, +300]; $p = 0.47$).

These results were matched by the comparison of minute averages of power output. A two-way ANOVA yielded a large

($\eta_p^2 = 0.59$) and significant main effect of acclimation ($p < 0.01$), but no significant time-condition interaction ($p = 0.20$; **Figure 4**). *Post hoc* analysis of condition indicated that mean power output across 15-min increased by 8.1% (+14 W, [+4, +24]; $p = 0.01$) from HTT1 to HTT3, and by 4.8% (+9 W, [0, +18]; $p = 0.05$) from HTT2 to HTT3 (**Table 3**). There was no difference in mean power output from HTT1 to HTT2 (+5 W, [-6, +16]; $p = 0.55$). Additionally, AUC calculated from minute averages of power output yielded a large ($\eta_p^2 = 0.58$) and significant ($p < 0.01$) main effect of acclimation. *Post hoc* comparisons revealed that power output AUC during HTT3 was greater than in HTT1 (+7.6%, [+2.1, +12.8]; $p = 0.01$) and showed a trend toward increases from HTT2 (+4.4%, [-0.4, +7.7]; $p = 0.07$). Power output AUC was not different between HTT1 and HTT2 (+3.2%, [-3.0, +7.0]; $p = 0.53$).

There was a large ($\eta_p^2 = 0.57$) and significant ($p < 0.01$) main effect of acclimation on mean cycling speed during time trials in hot conditions. *Post hoc* analysis indicated that mean cycling speed increased by 3.0% (+0.9 km · h⁻¹, [+0.2, +1.7]; $p = 0.01$) from HTT1 to HTT3, and by 1.6% (+0.5 km · h⁻¹, [0, +1.1]; $p = 0.05$) from HTT2 to HTT3 (**Table 3**). There was no difference in distance cycled from HTT1 to HTT2 (+0.4 km · h⁻¹, [-0.4, +1.2]; $p = 0.44$).

Mean ($p = 0.63$), peak ($p = 0.97$), and change ($p = 0.46$) in T_{rec} during the HTTs were not affected by HA (**Table 4**). A two-way ANOVA showed no significant main effect of condition ($p = 0.36$) or condition-time interaction ($p = 0.65$) for T_{rec} measured each minute of HTTs (**Figure 5**). There was an average reduction in T_{rec} at rest, although this was not significant ($p = 0.07$; **Table 4**). AUC for T_{rec} during HTTs (calculated from minute averages) was not significantly different between HTTs ($p = 0.39$). Mean ($p = 0.26$) and peak ($p = 0.13$) skin temperatures (T_{sk}) during HTTs were not affected by HA (**Table 4** and **Figure 5**). Mean HR ($p = 0.48$; **Figure 5**) and mean ($p = 0.45$) and peak ($p = 0.38$) percentage of age-estimated HR maximum (%HR_{max}) during HTTs was not different between HTTs (**Table 4**).

There was a significant change in number of active sweat glands immediately following HTTs (main effect: $p = 0.01$,

TABLE 2 | Performance, physiological, and psycho-physical responses during heat acclimation sessions averaged across Days 1–4 and Days 5–9.

	Days 1–4	Days 5–9
Mean power output (W)	108 ± 17	117 ± 21*
Mean $T_{\text{rec}75}$ (°C)	38.4 ± 0.2	38.2 ± 0.2*
Resting T_{rec} (°C)	37.3 ± 0.4	37.2 ± 0.2
Peak T_{rec} (°C)	38.6 ± 0.2	38.5 ± 0.2*
ΔT_{rec} (°C)	1.4 ± 0.4	1.2 ± 0.3
Peak T_{sk} (°C)	35.5 ± 0.9	35.0 ± 1.0*
Mean HR (beats · minute ⁻¹)	150 ± 6	145 ± 6*
Mean %HR _{max} (%)	78 ± 3	75 ± 4*
SR _{BSA} (g · h ⁻¹ · m ⁻²)	694 ± 105	705 ± 143
Sweat loss (%BM)	2.9 ± 0.5	2.9 ± 0.6

Data are presented as mean ± SD. T_{rec} , rectal temperature; $T_{\text{rec}75}$, mean rectal temperatures recorded during the final 75-min of the session; ΔT_{rec} , maximal change in rectal temperature during exercise from rest; HR, heart rate; %HR_{max}, percentage of age-estimated maximum heart rate; SR_{BSA}, estimated sweat rate relative to body surface area; %BM, percentage of body mass. *Significantly different from Days 1–4 ($p < 0.05$).

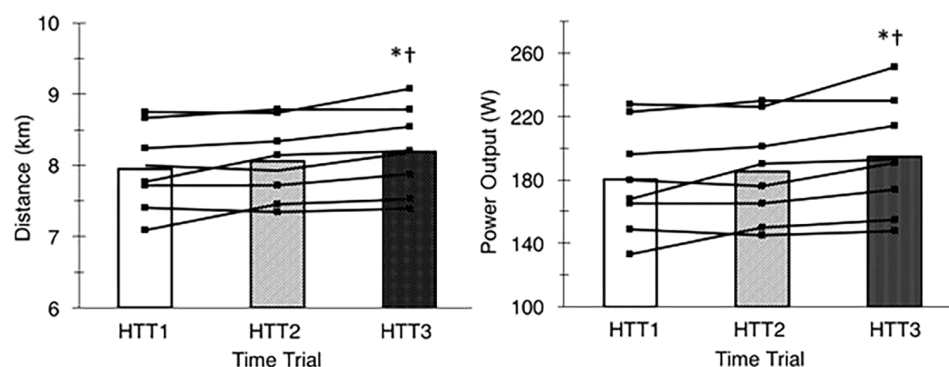


FIGURE 3 | Distance cycled (**left**) and mean power output (**right**) during time trials in hot conditions (35°C, 30% RH) performed pre-acclimation (HTT1), following 4-days heat acclimation (HTT2), and following 9-days heat acclimation (HTT3). Bars represent mean value and black lines represent individual participant data.

*Significant increase from HTT1 ($p < 0.05$); †Significant increase from HTT2 ($p < 0.05$).

TABLE 3 | Performance measures during time trials in the heat (35°C, 30% RH).

	HTT1	HTT2	HTT3
Distance (km)	7.96 ± 0.58	8.06 ± 0.55	8.20 ± 0.59*†
Mean power (W)	180 ± 34	185 ± 32	195 ± 36*†
Mean speed (km · h ⁻¹)	31.9 ± 2.3	32.3 ± 2.2	32.8 ± 2.4*†

Data are presented as mean ± SD. HTT1, time trial pre-acclimation; HTT2, time trial following 4-days heat acclimation; HTT3, time trial following 9-days heat acclimation. *Significant increase from HTT1; †Significant increase from HTT2, ($p < 0.05$).

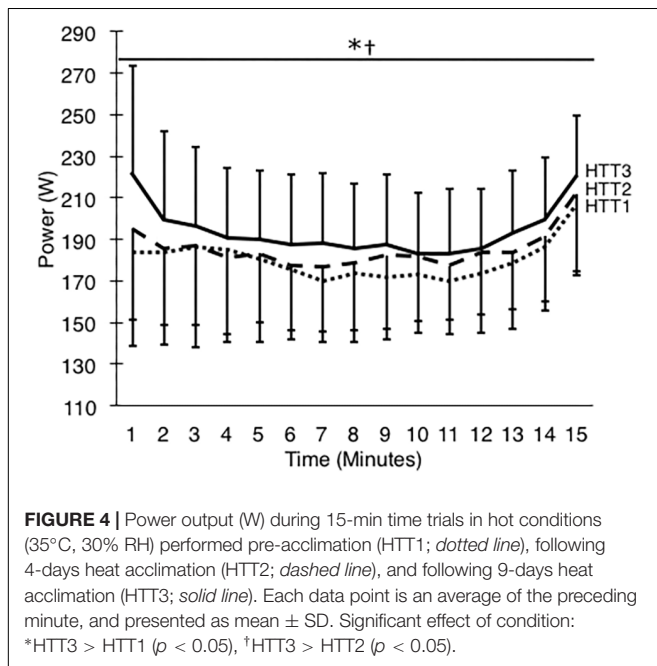


FIGURE 4 | Power output (W) during 15-min time trials in hot conditions (35°C, 30% RH) performed pre-acclimation (HTT1; dotted line), following 4-days heat acclimation (HTT2; dashed line), and following 9-days heat acclimation (HTT3; solid line). Each data point is an average of the preceding minute, and presented as mean ± SD. Significant effect of condition: *HTT3 > HTT1 ($p < 0.05$), †HTT3 > HTT2 ($p < 0.05$).

$\eta_p^2 = 0.64$). *Post hoc* analysis indicated that number of active sweat glands increased by 33% (+17 active sweat glands per cm², [+3, +30]; $p = 0.02$) from HTT1 to HTT3, and by 22% (+12 active sweat glands per cm², [+6, +17]; $p < 0.01$) from HTT2 to HTT3 (Table 4). There was no difference in number of active sweat glands from HTT1 to HTT2 (+5 active sweat glands per cm², [-7, +17]; $p = 0.62$). An example of sweat gland activity recorded following HTTs is depicted in Figure 6.

There was no significant difference in SR_{BSA} during HTTs and including the 75 min of HA that followed (main effect: $p = 0.08$). There were no differences in blood lactate at rest (immediately preceding HTTs; $p = 0.34$) or immediately following HTTs ($p = 0.41$; Table 4). Ratings of thermal sensation taken in the environmental chamber immediately before exercise were significantly different between the HTTs (main effect: $p = 0.02$; Table 4), and on average, corresponded to “Warm” (HTT1), “Warm” (HTT2) and “Slightly Warm” (HTT3). *Post hoc* pairwise comparisons indicated that differences were between HTT1 and HTT3 ($p = 0.03$). There were no significant differences between HTT1 and HTT2 ($p = 0.10$) or between HTT2 and HTT3 ($p = 0.08$). Ratings of thermal comfort taken in the environmental chamber immediately before exercise were not significantly different between HTTs (average ratings corresponded to ratings

TABLE 4 | Physiological and psychophysical measures recorded during time trials in hot conditions.

	HTT1	HTT2	HTT3
Thermoregulatory			
Resting T_{rec} (°C)	37.2 ± 0.4	37.2 ± 0.3	37.0 ± 0.4
Mean T_{rec} (°C)	37.7 ± 0.2	37.7 ± 0.3	37.6 ± 0.3
Peak T_{rec} (°C)	38.1 ± 0.3	38.1 ± 0.3	38.1 ± 0.4
ΔT_{rec} (°C)	0.9 ± 0.5	0.9 ± 0.3	1.0 ± 0.6
Mean T_{sk} (°C)	34.6 ± 0.6	34.1 ± 0.7	34.5 ± 0.9
Peak T_{sk} (°C)	35.0 ± 0.5	34.5 ± 0.5	35.0 ± 0.5
Cardiovascular			
Mean HR (beats · minute ⁻¹)	168 ± 14	165 ± 9	168 ± 12
Mean %HR _{max} (%)	87 ± 7	86 ± 4	87 ± 6
Peak %HR _{max} (%)	94 ± 5	93 ± 5	95 ± 4
Sudomotor response			
Active sweat glands per cm ²	58 ± 23	63 ± 24	75 ± 25*†
Blood lactate			
Pre-test (mmol · L ⁻¹)	1.2 ± 0.7	1.0 ± 0.4	1.0 ± 0.5
Post-test (mmol · L ⁻¹)	11.1 ± 4.1	11.3 ± 1.7	12.8 ± 2.7
Thermal comfort (10-point scale)			
Pre-test	3 ± 2	3 ± 1	2 ± 1
Post-test	5 ± 1	5 ± 2	5 ± 2
Thermal sensation (13-point scale)			
Pre-test	9 ± 1	9 ± 1	8 ± 0*
Post-test	10 ± 1	10 ± 1	10 ± 1

Data are presented as mean ± SD. HTT1, pre-acclimation time trial; HTT2, time trial following 4-days heat acclimation; HTT3, time trial following 9-days heat acclimation. T_{rec} , rectal temperature; ΔT_{rec} , change in rectal temperature during time trial; T_{sk} , weighted mean skin temperature; HR, heart rate; %HR_{max}, percentage of age-estimated maximum heart rate. *Significant difference from HTT1, †significant difference from HTT2, ($p < 0.05$).

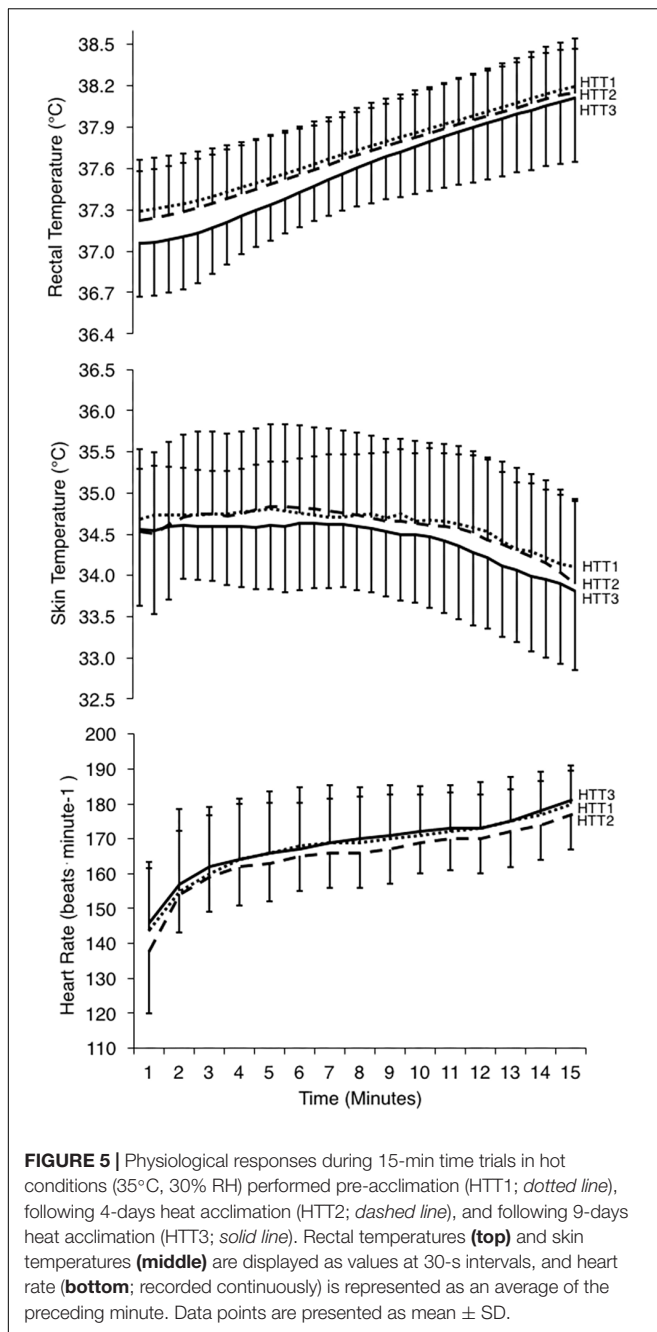
between “Comfortable” and “Slightly Uncomfortable”; $p = 0.39$; Table 4). Ratings of thermal sensation and thermal comfort taken at the end of HTTs were not significantly different between HTTs (“Hot” [$p = 0.25$] and “Uncomfortable” [$p = 0.53$], respectively; Table 4).

DISCUSSION

This study was designed to determine whether STHA (4-days) is sufficient to improve self-paced endurance performance in hot conditions in females, as has been observed in males, or whether a longer heat acclimation stimulus (i.e., 9-days) is required. In this study’s female cohort, STHA did not significantly improve time-trial performance in the heat; however, 9-days HA did. These results were consistent with the study hypothesis, which predicted that STHA would be insufficient to improve self-paced performance in females, and that a longer heat acclimation stimulus would be required to induce the physiological adaptations needed for performance improvements in the heat.

Self-Paced Endurance Performance

Following STHA, female participants showed no significant performance improvements in distance cycled, mean power



output, or speed during HTT2, as compared to HTT1. This is in direct contrast to a number of studies in male cohorts, where males have shown meaningful physiological adaptations and improved endurance performance in the heat following STHA (Garrett et al., 2009, 2012; Chen et al., 2013; Racinais et al., 2015; Guy et al., 2016; James et al., 2016; Willmott et al., 2016; Wingfield et al., 2016). Thus, it appears that STHA using 90 min of daily exercise heat stress is insufficient to improve endurance performance in females, reflecting the lack of physiological adaptation to heat acclimation previously demonstrated in females following STHA (Mee et al., 2015). The current study's

performance results following STHA differ from those observed by Sunderland et al. (2008), who reported a 33% improvement in distance run during a repeated shuttle run performance test (Loughborough Intermittent Shuttle Test) following STHA (4-days) in a female cohort. Of note, time to exhaustion is the main outcome measure of the Loughborough Intermittent Shuttle Test. This outcome is influenced by technique (i.e., ability to change direction and accelerate; Mendez-Villanueva and Buchheit, 2013), making it less reliable and subject to greater variation than the self-paced performance trial used in the current study (Hickey et al., 1992; Gosens et al., 2015; Borg et al., 2018). Furthermore, the behavioral regulation of performance possible in a self-paced time trial is not available in a time to exhaustion protocol (Schlader et al., 2011). Indeed, the lower pre-exercise thermal sensation reported by participants after 9-days HA may be an indication of perceptual changes contributing to behavioral regulation (i.e., pacing). Thus, the self-paced performance test used in the current study is a more reliable and holistic assessment of performance than a time to exhaustion test. Despite efforts in the current study to create an "intense" heat stimulus by combining isothermic heat acclimation, HIIT, and permissive dehydration, it still appears that females require either a longer daily heat exposure (Mee et al., 2018), or a greater number of heat exposures (as observed in the current study and by Mee et al., 2015) to improve exercise performance in the heat.

This is the first study to quantify improvements in self-paced time trial performance following a longer (i.e., 9-days) heat acclimation stimulus in a female cohort. The $\sim 8\%$ mean improvement in mean power output in HTT3 as compared to HTT1 is comparable to performance improvements observed in male, or mostly male cohorts following similar heat acclimation protocols. Keiser et al. (2015) showed that male participants experienced a $\sim 10\%$ improvement in power output during a 30-min self-paced time trial following 10-days heat acclimation (daily bouts: 90-min cycling at 50% $\text{VO}_{2\text{max}}$ in 38°C, 30%RH). Lorenzo et al. (2010) also found that participants (10 males and 2 females) had an 8% mean improvement in power output during their 1-h self-paced time trial following 10-days heat acclimation (daily bouts: 90-min cycling at 50% $\text{VO}_{2\text{max}}$ in 40°C, 30%RH). In the current study, improvements in mean power output coincided with improvements in mean cycling speed and distance covered from HTT1 to HTT3. These data demonstrate that 9-, but not 4-days heat acclimation, improves endurance performance outcomes in females.

Physiological Measures

Participants exhibited reduced markers of physiological strain (i.e., T_{rec} , T_{sk} and HR) during days 5–9 of HA, as compared to days 1–4. These physiological changes occurred in spite of an increased mean power output during days 5–9 of HA. Although these data indicate a reduction in the desired stimulus across the heat acclimation protocol, it also indicates that the greatest heat stimulus was administered during STHA. Furthermore, the improved performance in HTT3 as compared to the previous HTTs indicates that this reduced stimulus during days 5–9 was still effective in producing HA-related performance improvements. Also, this HA protocol produced

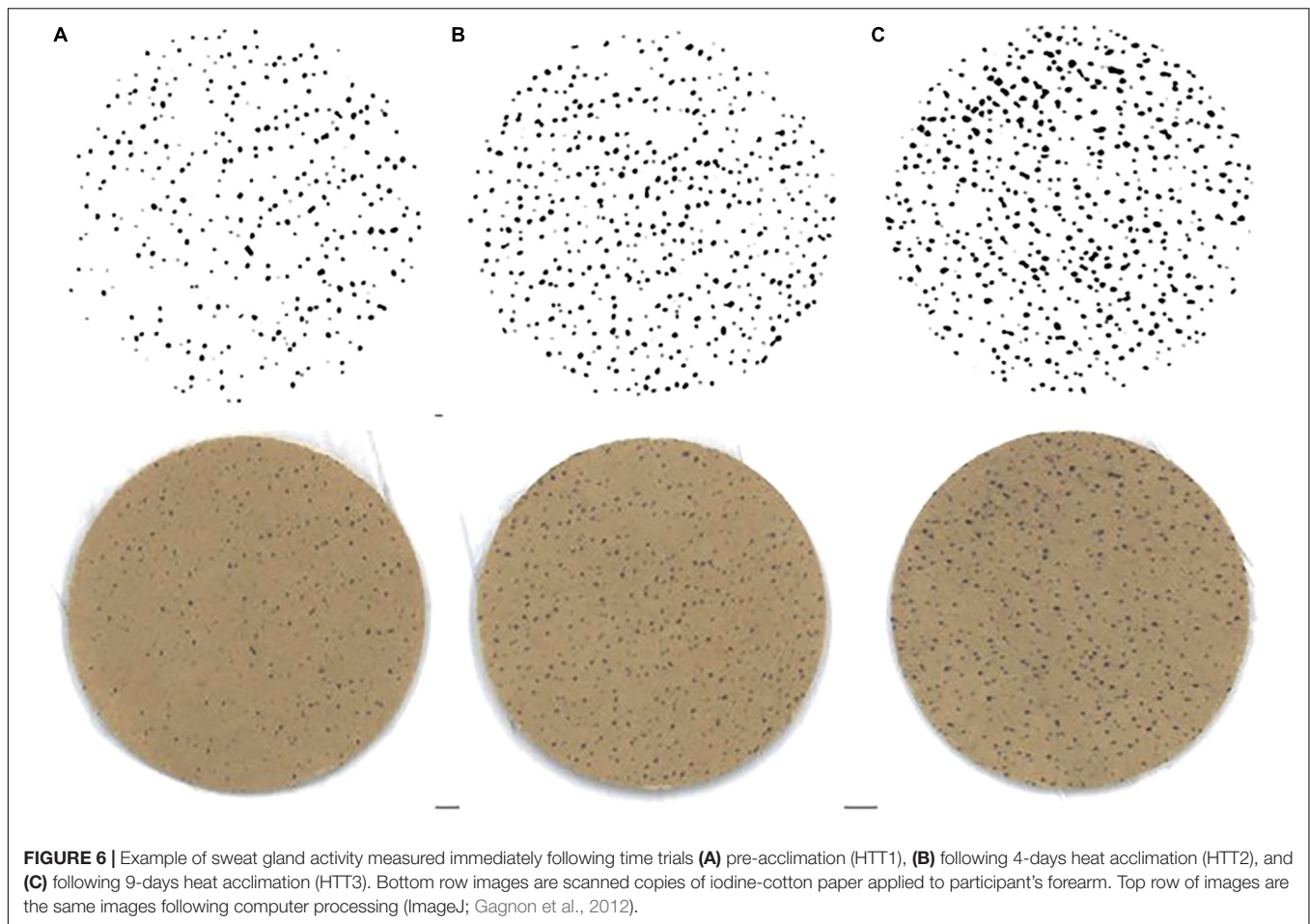


FIGURE 6 | Example of sweat gland activity measured immediately following time trials (A) pre-acclimation (HTT1), (B) following 4-days heat acclimation (HTT2), and (C) following 9-days heat acclimation (HTT3). Bottom row images are scanned copies of iodine-cotton paper applied to participant's forearm. Top row of images are the same images following computer processing (ImageJ; Gagnon et al., 2012).

a sufficient dehydration stimulus, as the $\sim 3\%$ body mass loss achieved across HA days 1–4 and 5–9 in addition to permissive dehydration presumably exceeded the osmotic threshold required for compensatory fluid regulatory responses (i.e., 2% body mass loss; Cheuvront and Kenefick, 2014). However, as we did not measure changes in plasma volume, it is unknown whether participants experienced the fluid regulatory responses typically associated with heat acclimation.

There was a trend for a lower T_{rec} at rest before HTT3, which appeared to influence T_{rec} during the initial minutes of HTT3 (albeit not significantly). Menstrual cycle phase and associated changes in female sex hormones influence resting T_{rec} (Inoue et al., 2005) and the overall thermoregulatory set point range (Charkoudian and Stachenfeld, 2016). This may have contributed to the non-significant change in resting T_{rec} observed in the current study. By the end of each HTT, T_{rec} reached similar values ($\sim 38.1^\circ\text{C}$). This is perhaps unsurprising as a previous study has shown that heat acclimation does not change the maximal T_{rec} reached ($40.1\text{--}40.2^\circ\text{C}$) during a 43.4-km time trial in the heat, despite a lower T_{rec} for the first 80% of the post-acclimation time trial (Racinais et al., 2015).

In the current study, there was an observed increase of active sweat glands at the end of HTT3 (Table 4). This contrasts findings in male cohorts, where sweat

gland activation did not increase following 8–10-days heat acclimation (Inoue et al., 1999; Lee et al., 2010; Poirier et al., 2016). In the current study, the number of active sweat glands (75 ± 25 per cm^2) at the end of HTT3 were lower than values previously reported in acclimated males ($\sim 96\text{--}108$ per cm^2 ; Inoue et al., 1999; Lee et al., 2010; Poirier et al., 2016) and unacclimated females (~ 93 per cm^2 ; Knip, 1969). Therefore, changes observed following a 15-min HTT may not indicate improved maximal sweat gland activation *per se*, but rather earlier activation of the sweat glands. Although there is large intra-subject coefficient variation associated with this measure, the 33 and 22% mean improvements following HTT3 in comparison to HTT1 and HTT2, respectively, surpass the $\sim 11\%$ coefficient of variation reported by Gagnon et al. (2012).

Perspectives

These results contribute to the limited research that informs the expected performance outcomes of heat acclimation for female athletes. The results of this study indicate that while heat acclimation can be an effective training component in preparation for competition in the heat, female athletes may require up to 9 days of 90-min heat acclimation sessions before

experiencing performance improvements. However, there will be individual variation in how athletes (male or female) respond to heat acclimation (Racinais et al., 2012). In the current study, three participants' performance deteriorated in HTT2 as compared to HTT1, whereas four participants showed improvements and one participant showed no change. Thus, some female athletes may achieve meaningful performance benefits after 4-days heat acclimation, while others could require longer than 9-days. A heat acclimation protocol lasting longer than 9-days has yet to be initiated in a female cohort, which would be hypothesized to further stabilize adaptations and improve performance (Racinais et al., 2015). It is also unclear how different phases of the menstrual cycle/contraception may affect heat adaptation during acclimation. Future research is also needed to clarify the impact of mixed-intensity heat acclimation on longer performance tests in both male and female athletes.

Considerations

Despite the absence of a control group, it is unlikely that performance improvements in HTT3 were due to learning or training effects. After preliminary testing and familiarizations, HTT1 was the fourth time that participants would have completed the 15-min time trial, minimizing learning effects. Furthermore, performance improvements in the current study are similar to previous studies (Lorenzo et al., 2010; Keiser et al., 2015), where control groups showed no improvements.

It is possible that the high-intensity heat acclimation protocol used in the current study may have caused a general fatigue that impaired performance during HTT2 and HTT3 (Schmit et al., 2018; Reeve et al., 2019). However, this is a negative bias as fatigue-related performance impacts would presumably have been greatest at HTT3. A further consideration is that heat acclimation adaptations are specific to the type/intensity of exercise employed (Wingfield et al., 2016). Therefore, the 15-min of HIIT undertaken at the beginning of each HA session may have facilitated specific adaptations. Whether this type of mixed-intensity heat acclimation (15-min HIIT + 75-min isothermic HA) would be equally or more effective than steady-state isothermic heat acclimation protocols typically reported in the literature remains unknown.

This study did not control for menstrual cycle. Recent data has shown that performance under heat stress is not affected by menstrual cycle or oral contraceptive pill (OCP) use in trained female athletes (Lei et al., 2017, 2018), nor does menstrual cycle affect whole-body heat loss (Notley et al., 2018). Eumenorrheic participants and OCP users did not cross over phases between HTT1 and HTT2. Participants were counterbalanced in their phases in HTT3, with both eumenorrheic participants being in opposite phases and both OCP users being in opposite phases (i.e., pill-taking, or non-pill-taking). None of the other four participants [contraceptive implant or copper intrauterine device (IUD)] were menstruating during the protocol, mitigating concerns of premenstrual symptoms that could affect performance (Giacomoni et al., 2000). Despite this, variable hormonal states may have affected the degree of relative heat stimulus administered when targeting an absolute core temperature of 38.5°C during heat acclimation sessions.

Finally, it should be noted that measures of sweat gland activity were taken from sites on the forearm and are not a precise indication of whole-body sweat gland adaptations given the regional heterogeneity of sweat gland activity. While an increased sweat gland activity may imply a better use of body surface area to dissipate heat, sweat gland activation is not directly proportional to local sweat output of the area (Poirier et al., 2016). In future, measures of local sweat output should be combined with measures of sweat gland activation to fully understand sex differences in peripheral sudomotor adaptations.

CONCLUSION

This study was the first to document performance outcomes during self-paced time trials in a female cohort following STHA (4-days) and 9-days high-intensity, isothermic HA. In the current study, females did not show an improvement in self-paced endurance performance following STHA. This differs from the well-documented performance improvements previously observed in male cohorts following STHA. However, following 9-days HA, females achieved meaningful improvements in self-paced endurance performance. These improvements included an ~8% increase in mean power output, a ~3% increase in distance cycled, and a ~3% increase in speed when performing a 15-min self-paced time trial in hot conditions (HTT). These data offer a reference for the changes which female athletes can expect when undergoing heat acclimation with the aim of improving self-paced endurance exercise performance in hot conditions, and provides further evidence that STHA may be insufficient for female athletes.

ETHICS STATEMENT

This study was carried out in accordance with the recommendations of the University of Birmingham Ethics Committee with written informed consent from all subjects.

AUTHOR CONTRIBUTIONS

NK, SL, and RL contributed to the conception and design of the study, acquisition, analysis, and interpretation of the data, drafted, revised, and approved the final version of the manuscript.

FUNDING

The study was funded by the University of Birmingham.

ACKNOWLEDGMENTS

The authors would like to thank all the participants for their time and effort in the completion of this study.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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The Effects of Endurance Exercise in Hypoxia on Acid-Base Balance, Potassium Kinetics, and Exogenous Glucose Oxidation

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Purpose: To investigate the carbohydrate metabolism, acid-base balance, and potassium kinetics in response to exercise in moderate hypoxia among endurance athletes.

Methods: Nine trained endurance athletes [maximal oxygen uptake ($\text{VO}_{2\text{max}}$): 62.5 ± 1.2 mL/kg/min] completed two different trials on different days: either exercise in moderate hypoxia [fraction of inspired oxygen (FiO_2) = 14.5%, HYPO] or exercise in normoxia (FiO_2 = 20.9%, NOR). They performed a high-intensity interval-type endurance exercise consisting of 10×3 min runs at 90% of $\text{VO}_{2\text{max}}$ with 60 s of running (active rest) at 50% of $\text{VO}_{2\text{max}}$ between sets in hypoxia (HYPO) or normoxia (NOR). Venous blood samples were obtained before exercise and during the post-exercise. The subjects consumed ^{13}C -labeled glucose immediately before exercise, and we collected expired gas samples during exercise to determine the ^{13}C -excretion (calculated as $^{13}\text{CO}_2/^{12}\text{CO}_2$).

Results: The running velocities were significantly lower in HYPO (15.0 ± 0.2 km/h) than in NOR (16.4 ± 0.3 km/h, $P < 0.0001$). Despite the lower running velocity, we found a significantly greater exercise-induced blood lactate elevation in HYPO compared with in NOR ($P = 0.002$). The bicarbonate ion concentration ($P = 0.002$) and blood pH ($P = 0.002$) were significantly lower in HYPO than in NOR. There were no significant differences between the two trials regarding the exercise-induced blood potassium elevation ($P = 0.87$) or ^{13}C -excretion (HYPO, 0.21 ± 0.02 mmol/39 min; NOR, 0.14 ± 0.03 mmol/39 min; $P = 0.10$).

Conclusion: Endurance exercise in moderate hypoxia elicited a decline in blood pH. However, it did not augment the exercise-induced blood K^+ elevation or exogenous glucose oxidation (^{13}C -excretion) compared with the equivalent exercise in normoxia among endurance athletes. The findings suggest that endurance exercise in moderate hypoxia causes greater metabolic stress and similar exercise-induced elevation of blood K^+ and exogenous glucose oxidation compared with the same exercise in normoxia, despite lower mechanical stress (i.e., lower running velocity).

Keywords: hypoxia, endurance exercise, carbohydrate metabolism, acid-base balance, K^+

OPEN ACCESS

Edited by:

Matthew J. Barnes,
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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 09 January 2019

Accepted: 09 April 2019

Published: 16 May 2019

Citation:

Sumi D, Kasai N, Ito H and
Goto K (2019) The Effects
of Endurance Exercise in Hypoxia on
Acid-Base Balance, Potassium
Kinetics, and Exogenous Glucose
Oxidation. *Front. Physiol.* 10:504.
doi: 10.3389/fphys.2019.00504

INTRODUCTION

Carbohydrate metabolism during endurance exercise in hypoxia has been evaluated using several parameters, including blood glucose, lactate response, and the respiratory exchange ratio (RER) (Friedmann et al., 2004; Katayama et al., 2010; Morishima et al., 2014). However, carbohydrate oxidation using the RER may be overestimated during exercise in hypoxia due to the lower oxygen uptake and hyperventilation (Ogawa et al., 2007; Ofner et al., 2014; Sharma et al., 2019). An evaluation of carbohydrate oxidation using a stable isotope (^{13}C) is an alternative procedure to overcome this problem (Harvey et al., 2007; Blondin et al., 2010; Smith et al., 2010; Tremblay et al., 2010).

The energy supply via the glycolytic system is enhanced during endurance exercise in hypoxia (Buchheit et al., 2012; Sumi et al., 2018a). During exercise, lactate and hydrogen ions (H^+) are produced in working muscle via the augmented energy supply from the glycolytic system and are subsequently released into the blood circulation by Na^+/H^+ exchanger isoform 1 and monocarboxylate transporters (Juel, 1997, 2006), which elicits metabolic acidosis (lower muscle pH). Furthermore, the augmented metabolic stress stimulates the opening of muscle ATP-sensitive K^+ (K_{ATP}) channels, which subsequently increases the K^+ efflux from working muscle into the extracellular fluid (Davies, 1990; Nielsen et al., 2002). Therefore, endurance exercise in hypoxia with greater intramuscular and blood acidification may promote the accumulation of blood K^+ compared with exercise in normoxia. In our previous study (Sumi et al., 2018b), we evaluated the effect of endurance exercise in hypoxia on acid-base balance and K^+ responses among middle-long distance runners. However, in this study, we used cycling exercise although middle-long distance runners were recruited. Therefore, the subjects performed unaccustomed exercise modality compared with their daily exercise training (running), which may be associated with unexpected results from the hypothesis. The lower muscle pH and accumulation of K^+ in extracellular fluid during exercise are considered factors limiting sustained power output during endurance exercise (Trivedi and Danforth, 1966; Sahlin et al., 1981; Fitts, 1994; Sejersted and Sjøgaard, 2000; Bangsbo and Juel, 2006; Allen et al., 2008), whereas these factors may augment training adaptations (Mohr et al., 2007; Iaia et al., 2008; Bangsbo et al., 2009). For instance, Mohr et al. (2007) compared the effects of two different intense training regimens on skeletal muscle ion transport proteins and exercise performance. Sprint endurance training (30 s runs), with greater disturbance of muscle ion homeostasis (higher blood lactate and K^+ concentrations) during the training sessions, caused greater adaptation of Na^+/H^+ exchanger isoform 1 and the Na^+/K^+ -ATPase $\alpha 1$ isoform. These adaptations also led to lower venous H^+ and K^+ concentrations during intensive running exercise and further improved exercise performance compared with a normal endurance training group. Because the training adaptations arise from repetition of the acute physiological responses in each training session, determination of the exercise-induced acid-base balance and K^+ responses would greatly improve our understanding of the mechanism underlying the enhanced endurance exercise capacity after several weeks of

endurance training in hypoxia (Dufour et al., 2006; Czuba et al., 2011, 2013, 2017).

Therefore, the present study evaluated the acid-base balance, K^+ kinetics, and exogenous glucose oxidation in response to endurance (running) exercise in moderate hypoxia among endurance runners. We hypothesized that endurance exercise in moderate hypoxia would facilitate the exercise-induced decrease in blood pH, elevate the K^+ concentration, and promote exogenous glucose oxidation compared with the same relative intensity of exercise in normoxia.

MATERIALS AND METHODS

Subjects

Nine endurance athletes (middle-long distance runners who belonged to the same truck and filed club in university) participated in the study. Exclusion criteria were (1) an unhealthy person (all subjects were required to pass the medical check conducted in university every year), (2) no experience in hypoxic training at least 3 months before the present experiment. Their means and standard errors (SE) for age, height, and body mass were 20.7 ± 0.9 years, 172.5 ± 2.2 cm, and 61.6 ± 2.8 kg, respectively. All athletes were born and living at sea level, and they maintained specific middle-long distance running training 5 days per week (approximately 70 km per week). They gave written informed consent after being informed of the purpose and risks associated with the experiment. This study was approved by the Ethics Committee for Human Experiments at Ritsumeikan University, Japan.

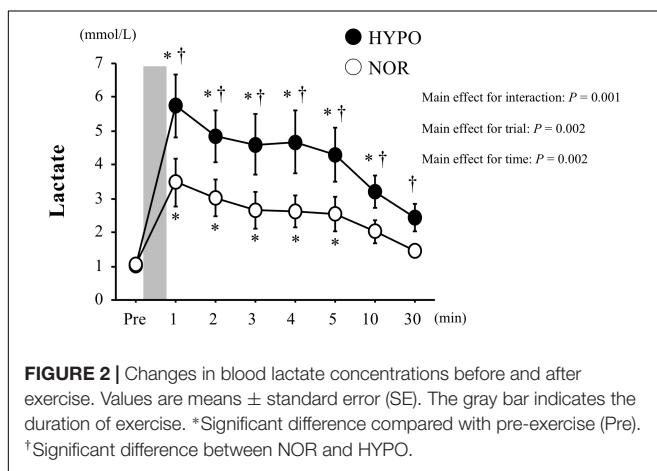
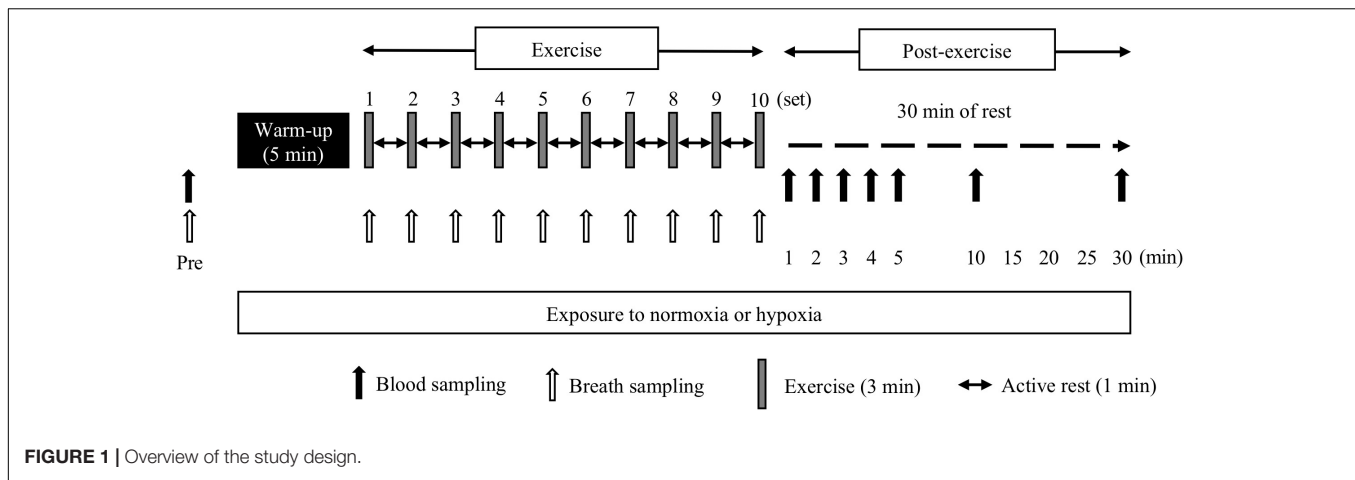
Experimental Design

The subjects visited the laboratory four times during the study. During the first and second visits, maximal oxygen uptake ($\text{VO}_{2\text{max}}$) tests were completed using a treadmill (Valiant; Lode, Groningen, the Netherlands) in both normoxia [inspired oxygen fraction (FiO_2) = 20.9%] and normobaric hypoxia (FiO_2 = 14.5%, equivalent to a simulated altitude of 3000 m). Each test was separated by 3 days and randomized.

During the third and fourth occasions, the subjects performed two experimental trials in either hypoxia (FiO_2 = 14.5%, HYPO) or normoxia (FiO_2 = 20.9%, NOR) on different days. We used a cross-over design and each trial was separated by 1 week. The two trials were started from the same time of the day, and the order of the two trials was randomized. As shown in **Figure 1**, all subjects completed high-intensity interval running on a treadmill at the same exercise intensity relative to $\text{VO}_{2\text{max}}$ evaluated in hypoxia and normoxia. After completing the exercise, the subjects rested for 30 min under the respective condition (i.e., either normoxia or hypoxia). Changes in blood variables and the $^{13}\text{CO}_2/^{12}\text{CO}_2$ ratio in the expired gas were monitored during and after the exercise to clarify the effects of exercise in hypoxia on the acid-base balance and potassium and exogenous glucose oxidation kinetics.

Exercise Protocols

All exercise sessions in HYPO and NOR were conducted using a treadmill (Elevation series E95Ta; Life Fitness, Tokyo, Japan).



Both trials were conducted in an environmentally controlled chamber. The hypoxic chamber used in this study was the whole-room type, and the hypoxic condition was established by nitrogen insufflation. The subjects ran for 5 min at 60% of $\text{VO}_{2\text{max}}$ (warm-up exercise) from 15 min after entering the hypoxic chamber. From 5 min after the warm-up exercise, the subjects started an interval-type endurance exercise [10×3 min run at 90% of $\text{VO}_{2\text{max}}$ separated by a 1 min run (active rest) at 50% of $\text{VO}_{2\text{max}}$] in hypoxia or normoxia. This exercise regimen was designed to mimic the actual training regimen of well-trained endurance athletes (Niess et al., 2003; Sumi et al., 2018a,b). Each trial was separated by 1 week. The two trials started at the same time of the day, and the order of the two trials was randomized. To avoid psychological influences, the subjects were not informed about whether the trial was conducted in hypoxia or normoxia. Room temperature and humidity were set at 22°C and 50%, respectively.

Maximal Oxygen Uptake in Normoxia or Hypoxia (Preliminary Measurement)

The initial running velocity was set at 12 km/h, and the running velocity was increased by 2 km/h every 2 min until it reached

16 km/h. Once the running velocity reached 16 km/h, it was increased by 0.6 km/h every minute until volitional exhaustion (Sumi et al., 2018a). During the test, expired gases were collected and analyzed using an automatic gas analyzer (AE300S; Minato Medical Science, Tokyo, Japan). The collected data were averaged every 30 s. Heart rate (HR) was measured continuously during the test using a wireless HR monitor (Accurex Plus; Polar Electro Oy, Kempele, Finland). The $\text{VO}_{2\text{max}}$ tests were performed twice in either normoxia or hypoxia, and the order of the two $\text{VO}_{2\text{max}}$ tests was randomized.

Blood Variables

Following an overnight fast, the subjects visited the laboratory at 8:00 and rested before the first blood collection. A polyethylene catheter was inserted into an antecubital vein after a 20 min rest, and a baseline blood sample was obtained. Blood samples were collected 1, 2, 3, 4, 5, 10, and 30 min after the participants completed the exercise. All blood samples for determinations of blood gases and electrolytes were collected using a 2.5 mL syringe containing heparin. A 10 mL syringe was used to obtain serum and plasma samples. Serum and plasma samples were obtained after 10 min of centrifugation at 4°C (3000 rpm), and the samples were stored at -80°C until analysis.

The blood glucose, lactate, serum insulin, and ketone body concentrations as well as the blood-gas variables of the blood samples were measured. The blood-gas parameters, including the hydrogen ion concentration (pH), oxygen partial pressure (pO_2), carbon dioxide partial pressure (pCO_2), bicarbonate ion (HCO_3^-), and base excess (BE); the potassium (K^+), sodium (Na^+), and hemoglobin (Hb) concentrations; and the hematocrit (Hct) levels were measured using an automatic blood-gas analyzer (OPTI CCA TS, Sysmex, Hyogo, Japan). The exercise-induced plasma volume shift (%) was calculated using the Dill and Costill (1974) equation as follows:

$$\Delta PV(\%) = 100 \times [(Hb_{\text{pre}}/Hb_{\text{post}}) \times (100 - Hct_{\text{post}}) / (100 - Hct_{\text{pre}}) - 1],$$

where Hct is in % and Hb is in g/dL.

These analyses were completed within 15 min after blood collection, and the samples were put on ice until analysis. The blood glucose and lactate concentrations were measured using a glucose analyzer (Free style; Nipro, Osaka, Japan) and a lactate analyzer (Lactate Pro; ARKRAY, Kyoto, Japan) immediately after blood collection. The serum insulin and ketone body concentrations were measured at a clinical laboratory (SRL, Tokyo, Japan). The intra-assay coefficients of variability were 3.2 and 2.9% for the serum insulin and ketone body concentrations, respectively.

Exogenous Glucose Oxidation Kinetics

Immediately before beginning the exercise, the subjects consumed 500 mg of ^{13}C -glucose (D-Glucose- U - $^{13}\text{C}_6$, ^{13}C : 99 atom%; Chlorella Industry, Tokyo, Japan) dissolved in 100 mL of purified water. In the ^{13}C -glucose, the carbon atoms at all six positions in each glucose molecule were labeled with ^{13}C . Before consuming the ^{13}C -glucose, a baseline breath sample was collected using a 1.3 L sampling bag (Otsuka Pharmaceutical, Tokyo, Japan). Ten breath samples were collected until the end of the exercise in each set. The $^{13}\text{CO}_2/^{12}\text{CO}_2$ ratio in the sample bag was evaluated using an infrared spectrometer (POC one; Otsuka Pharmaceutical). The $^{13}\text{CO}_2/^{12}\text{CO}_2$ ratio was expressed as the absolute increase between samplings during exercise and at baseline.

The measured $^{13}\text{CO}_2$ and $^{12}\text{CO}_2$ abundance ratio was converted into the actual amount of excreted ^{13}C and then converted using the formula to evaluate the ^{13}C kinetics. The ^{13}C excretion per unit time was calculated using the following equation (Schneider et al., 1978; Tanaka et al., 2013):

$$^{13}\text{C} - \text{excretion} = (\Delta\%^{13}\text{C}/100) \times 300 \times \text{BSA}.$$

Body surface area (BSA) can be estimated using the formula proposed by Du Bois and Du Bois (1916):

$$\text{BSA} = (W^{0.425} \times H^{0.725}) \times 0.007184,$$

where W is the body weight measured in kilograms, and H is the body height measured in centimeters.

Cardiorespiratory Variables and RPE

The oxygen uptake (VO_2), carbon dioxide output (VCO_2), RER, and expired minute ventilation (VE) were determined breath by breath in repetitions (reps) 5 and 10 of the interval exercise, and the average values of the respiratory variables during the final 1 min of each rep were calculated. Percutaneous oxygen saturation (SpO_2) was measured at reps 5 and 10 during the interval exercise using a finger pulse oximeter (Smart Pulse; Fukuda Denshi, Tokyo, Japan) placed on the tip of the right forefinger. HR was recorded every 5 s during exercise; the average values were calculated during the final 1 min of each 3 min rep. The subjects indicated their rating of perceived exertion for respiratory strain (RPE-R) and leg muscle strain (RPE-L) at the end of each exercise rep using a 10-point scale to measure perceived exertion (Wilson and Jones, 1991).

Energy Expenditure and Substrate Oxidation

The energy expenditure (EE) during exercise was calculated using the Weir (1949) equation, where VO_2 and VCO_2 are expressed as L/min. The values of VO_2 and VCO_2 were the averages during the final 1 min in rep 10 during the interval exercise:

$$\text{Energy expenditure (kcal/min)} = 3.9 \times \text{VO}_2 + 1.1 \times \text{VCO}_2.$$

The rates of carbohydrate and fat oxidation were calculated using the following equations (Peronnet and Massicote, 1991; Manetta et al., 2002), where VO_2 and VCO_2 are expressed as L/min. VO_2 and VCO_2 values were the averages of the last 1 min of rep 10 during the interval exercise:

$$\text{Carbohydrate (g/min)} = 4.585 \times \text{VCO}_2 - 3.226 \times \text{VO}_2$$

$$\text{Fat (g/min)} = 1.695 \times \text{VO}_2 - 1.701 \times \text{VCO}_2.$$

Statistical Analyses

Data are expressed as means \pm SE. Two-way analysis of variance (ANOVA) with repeated measures was used to test the interaction (trial \times time) and main effects (trial, time). When the ANOVA revealed a significant interaction or main effect, the Tukey–Kramer test was performed as a *post hoc* analysis to identify differences. The area under the curve (AUC) for ^{13}C -excretion was compared between the two trials using a paired *t*-test. For all tests, $P < 0.05$ were considered to indicate statistical significance.

RESULTS

$\text{VO}_{2\text{max}}$ and Running Velocity

$\text{VO}_{2\text{max}}$ was significantly lower in HYPO (43.6 ± 1.4 mL/kg/min) than in NOR (62.5 ± 1.2 mL/kg/min, $P < 0.0001$). The maximal running velocity during $\text{VO}_{2\text{max}}$ test was significantly lower in HYPO (17.4 ± 0.1 km/h) than in NOR (19.6 ± 0.3 km/h, $P < 0.0001$). Consequently, the running velocity during each 3 min of the interval exercise (90% of $\text{VO}_{2\text{max}}$) was significantly lower in HYPO (15.0 ± 0.2 km/h) than in NOR (16.4 ± 0.3 km/h, $P < 0.0001$).

Metabolites, Blood Gas, and Plasma Volume Kinetics

The blood lactate concentration was significantly increased after exercise in both trials (main effect for time, $P = 0.002$). Moreover, it was significantly higher in HYPO than in NOR after exercise (interaction, $P = 0.001$; main effect for trial, $P = 0.002$, **Figure 2**).

The blood glucose concentrations increased significantly with exercise in both trials (main effect for time, $P < 0.0001$). However, there was no significant difference between the two trials at any time point. The serum insulin and ketone body concentrations did not change significantly over time in either trial. Moreover, no significant difference was observed between the two trials. In HYPO, the blood pO_2 decreased significantly

after exercise (main effect for time, $P = 0.001$), whereas the blood pO_2 increased significantly after exercise in NOR (main effect for time, $P = 0.001$). Consequently, the blood pO_2 remained significantly lower in HYPO after exercise compared with in NOR (interaction, $P < 0.0001$; main effect for trial, $P < 0.0001$). The blood pCO_2 decreased significantly after exercise in both trials (main effect for time, $P < 0.0001$). The exercise-induced reduction in blood pCO_2 was significantly greater in HYPO after exercise (interaction, $P = 0.475$; main effect for trial, $P < 0.0001$). The plasma volume decreased significantly after exercise in both trials. However, there was no significant difference between the two trials at any time point (Table 2).

Acid-Base Balance

After exercise, the blood pH was significantly lower in HYPO than in NOR (interaction, $P = 0.002$). The HCO_3^- concentration decreased significantly after exercise in both trials (main effect for time, $P < 0.0001$). However, HYPO showed significantly lower HCO_3^- concentrations after exercise (interaction, $P = 0.434$; main effect for trial, $P = 0.002$). The BE decreased significantly after exercise in both trials (main effect for time, $P < 0.0001$). The exercise-induced reduction in BE was significantly greater in HYPO after exercise (main effect for trial, $P = 0.018$, Figure 3).

Blood K^+

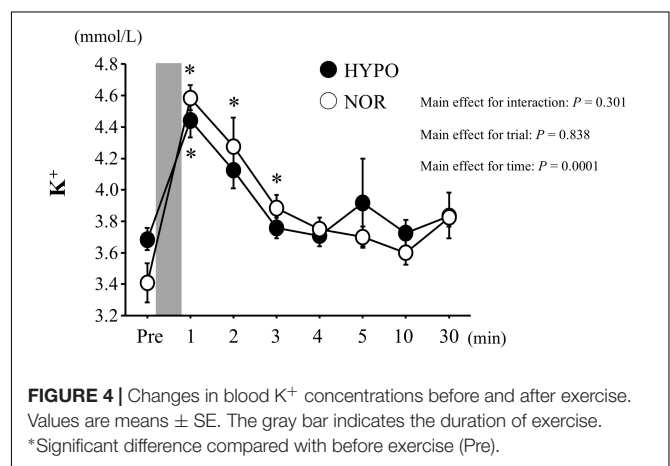
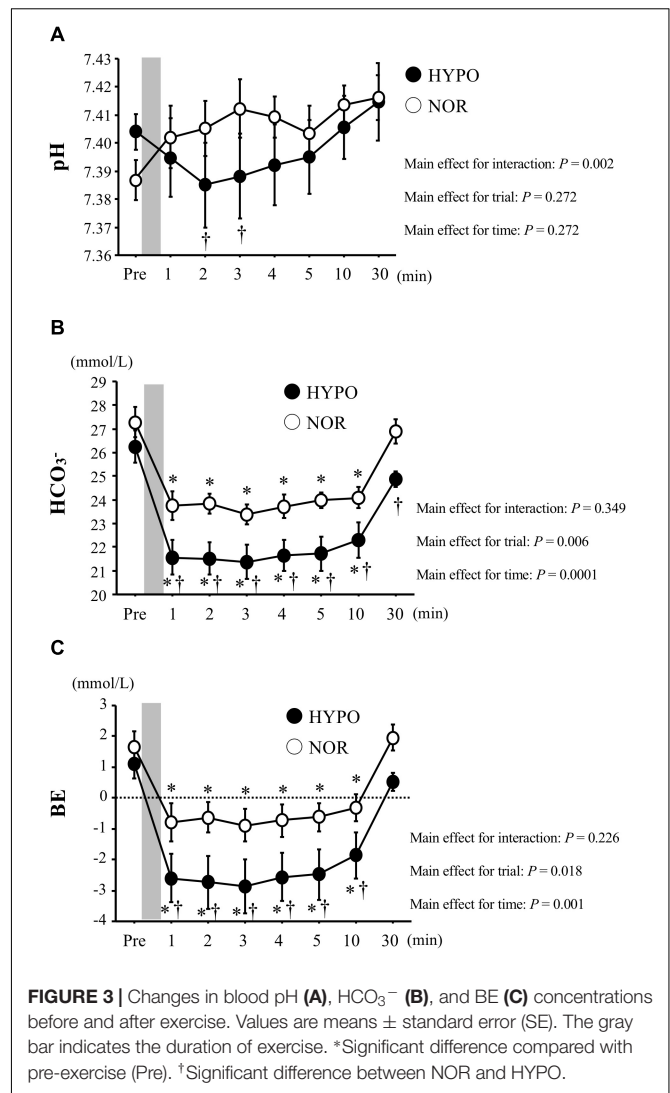
The blood K^+ concentrations increased significantly after exercise in both trials (main effect for time, $P < 0.0001$). However, the exercise-induced elevations of blood K^+ did not differ significantly between the two trials (Figure 4).

Exogenous Glucose Oxidation Kinetics During Exercise

The ^{13}C -excretion calculated by $^{13}CO_2/^{12}CO_2$ increased during exercise in both trials (main effect for time, $P < 0.0001$), whereas there was no significant difference between HYPO and NOR (interaction, $P = 0.146$; main effect for trial, $P = 0.09$). The AUC for the ^{13}C -excretion (during exercise) did not differ significantly between HYPO and NOR ($P = 0.09$, Figure 5).

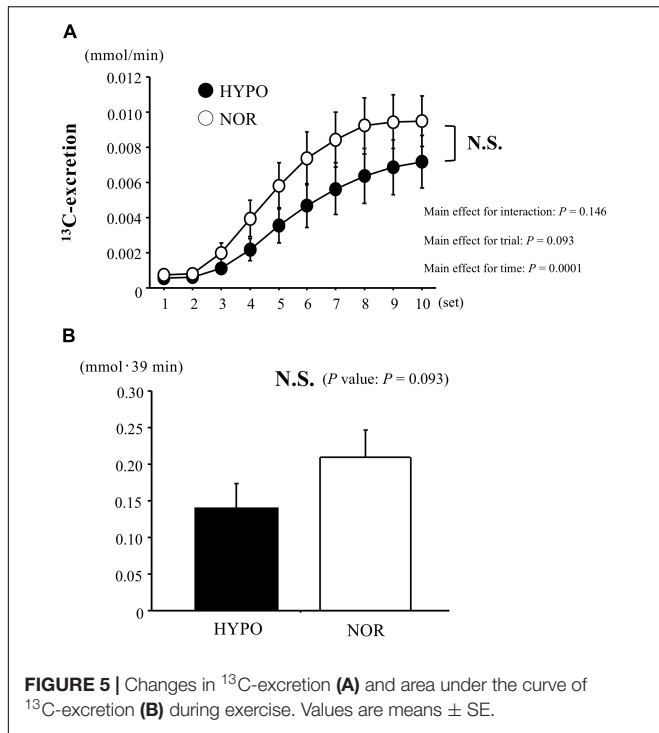
Cardiorespiratory Variables, Substrate Oxidation, and Energy Expenditure During Exercise

Table 1 shows the cardiorespiratory variables during exercise in each trial. VO_2 , VCO_2 , and SpO_2 remained significantly lower in HYPO than in NOR throughout the exercise (main effect for trial, $P < 0.0001$). By contrast, HYPO showed a significantly higher RER during exercise compared with NOR (main effect for trial, $P < 0.0001$). During exercise, the VE and HR did not differ significantly between HYPO and NOR. The CHO oxidation during exercise was significantly higher in HYPO than in NOR (main effect for trial, $P = 0.03$). By contrast, HYPO showed a significantly lower fat oxidation (main effect for trial, $P = 0.002$) and EE (main effect for trial, $P < 0.0001$) during exercise compared with NOR.



RPE

RPE-R and RPE-L did not differ significantly between HYPO and NOR during exercise.



DISCUSSION

As we hypothesized, the exercise-induced acidification of blood (i.e., lower blood pH and higher blood lactate) was significantly greater in HYPO than in NOR, although the running velocity was significantly lower in HYPO. By contrast, there was no significant difference in the exercise-induced elevation of blood K^+ concentrations or ^{13}C -excretion between HYPO and NOR.

The exercise-induced blood lactate elevation was more profound in HYPO than in NOR. Moreover, HYPO caused significantly higher RER and CHO oxidation (evaluated by VO_2 and VCO_2) during exercise compared with NOR. These results indicate that carbohydrate metabolism during endurance exercise was augmented in hypoxia compared with in normoxia, which was consistent with previous findings (Katayama et al., 2010;

TABLE 1 | Cardiorespiratory variables, substrate oxidation and energy expenditure during exercise.

	NOR	HYPO	p-value
SpO_2 (%)	95.0 ± 0.3	78.3 ± 1.1	$P < 0.0001$
VO_2 (mL/kg/min)	52.1 ± 0.9	41.1 ± 1.6	$P < 0.0001$
VCO_2 (mL/kg/min)	47.9 ± 1.2	42.1 ± 1.2	$P = 0.0003$
RER	0.92 ± 0.02	1.03 ± 0.02	$P = 0.004$
VE (L/min)	100 ± 6.0	105 ± 3.8	$P = 0.315$
HR (beats/min)	177 ± 2.4	176 ± 3.2	$P = 0.545$
CHO oxidation (g/min)	3.1 ± 0.3	3.7 ± 0.6	$P = 0.044$
Fat oxidation (g/min)	0.4 ± 0.1	0.1 ± 0.0	$P = 0.002$
EE (kcal/min)	15.7 ± 0.7	12.6 ± 0.5	$P < 0.0001$

Values are mean \pm SE.

TABLE 2 | Blood variables and plasma volume shift before exercise and during post-exercise.

	Pre	1	2	3	4	5	10	30 (min)
Glucose (mg/dL)	NOR 78 \pm 1.5	94 \pm 6.6*	104 \pm 6.1*	103 \pm 6.0*	99 \pm 4.3*	100 \pm 6.3*	93 \pm 6.0	78 \pm 4.1
	HYPO 77 \pm 2.2	101 \pm 4.0*	106 \pm 4.7*	102 \pm 6.0*	107 \pm 5.0*	101 \pm 4.8*	90 \pm 4.8	77 \pm 2.6
Insulin ($\mu\text{LU/mL}$)	NOR 2.64 \pm 0.6	1.69 \pm 0.4	—	—	—	—	—	3.40 \pm 1.2
	HYPO 2.55 \pm 0.6	1.93 \pm 0.4	—	—	—	—	—	2.67 \pm 0.6
Ketone body ($\mu\text{mol/L}$)	NOR 127.7 \pm 48.0	96.2 \pm 17.3	—	—	—	—	—	166.1 \pm 58.7
	HYPO 88.7 \pm 25.8	85.8 \pm 6.2	—	—	—	—	—	76.7 \pm 14.3
pO ₂ (mmHg)	NOR 49 \pm 5.0	73 \pm 3.2*	85 \pm 1.6*	90 \pm 1.8*	90 \pm 2.4*	87 \pm 3.2*	77 \pm 4.3*	55 \pm 5.5
	HYPO 61 \pm 4.2	47 \pm 1.4†	56 \pm 1.3†	58 \pm 1.1†	58 \pm 1.4†	57 \pm 1.7†	51 \pm 2.0†	45 \pm 3.6†
pCO ₂ (mmHg)	NOR 47 \pm 1.4	39 \pm 1.1*	39 \pm 0.5*	38 \pm 0.3*	39 \pm 0.5*	40 \pm 0.5*	39 \pm 0.7*	43 \pm 1.2
	HYPO 44 \pm 1.7	36 \pm 1.0†	36 \pm 0.7†	36 \pm 0.5†	36 \pm 0.5†	36 \pm 0.6†	36 \pm 0.9†	40 \pm 1.5
ΔPV (%)	NOR 0.0 \pm 0.0	-12.7 \pm 2.9*	-12.5 \pm 3.3*	-7.0 \pm 6.3*	-11.3 \pm 2.9*	-9.8 \pm 3.6*	-4.3 \pm 2.3*	-4.4 \pm 6.5
	HYPO 0.0 \pm 0.0	-15.0 \pm 2.9*	-15.9 \pm 2.7*	-13.9 \pm 2.2*	-14.8 \pm 2.3*	-12.8 \pm 2.4*	-9.1 \pm 2.0*	-5.6 \pm 2.2*

Values are means \pm SE. *Significant difference vs. Pre. †Significant difference vs. Normoxia. PV, plasma volume.

Buchheit et al., 2012; Sumi et al., 2018a). The facilitated carbohydrate metabolism during endurance exercise under our HYPO condition may be explained by the increased ATP produced via the glycolytic system to compensate for the hypoxia-induced decline in ATP production via the aerobic system (Friedmann et al., 2007; Ogawa et al., 2007).

Many studies have evaluated carbohydrate metabolism during endurance exercise in hypoxia using traditional procedures, including blood glucose, lactate responses, and RER (Friedmann et al., 2004; Katayama et al., 2010; Morishima et al., 2014). Among these, the determination of RER during submaximal endurance exercise is most often used to monitor the substrate oxidation pattern, whereas carbohydrate oxidation may be overestimated by RER during endurance exercise in hypoxia due to the lower VO_2 and hyperventilation (Ogawa et al., 2007; Ofner et al., 2014; Sharma et al., 2019). By contrast, we evaluated carbohydrate oxidation using a stable isotope (^{13}C) as an alternative procedure to overcome this limitation. The consumed ^{13}C -labeled glucose is oxidized mainly in working muscles during endurance exercise and is subsequently excreted in the expired gas as $^{13}\text{CO}_2$. Therefore, the $^{13}\text{CO}_2/^{12}\text{CO}_2$ ratio during endurance exercise reflects the amount of exogenous glucose oxidation (Harvey et al., 2007; Blondin et al., 2010; Smith et al., 2010; Tremblay et al., 2010). The ingestion of ^{13}C -labeled glucose, specifically the exogenous glucose oxidation pattern in the tissues (e.g., skeletal muscle and liver), can be used to evaluate carbohydrate oxidation during endurance exercise in hypoxia. Moreover, the circulating blood during endurance exercise is predominantly distributed to working muscle; therefore, the augmented ^{13}C -excretion during endurance exercise mainly reflects glucose oxidation in working muscle. Furthermore, we determined the serum ketone body concentrations (an indication of liver metabolism). However, there was no significant difference in the serum ketone body concentration over time, suggesting that energy metabolism in the liver (with a subsequent increase in serum ketone body concentration) was not augmented during the exercise. As previous studies revealed that endurance exercise in hypoxia promotes carbohydrate metabolism compared with exercise in normoxia (Katayama et al., 2010; Buchheit et al., 2012; Sumi et al., 2018a), we initially hypothesized that HYPO would elicit exogenous glucose oxidation during endurance exercise (i.e., increased ^{13}C -excretion in HYPO compared with in NOR). However, no significant difference was observed in ^{13}C -excretion between HYPO and NOR. Several factors are involved in exogenous glucose oxidation during exercise, but the EE during exercise strongly affects the amount of carbohydrate oxidation (Gautier et al., 1996). In our study, the EE during the 10×3 min run was significantly lower in HYPO than in NOR due to the lower running velocity. Therefore, future research should include comparisons involving the same EE in hypoxia and normoxia. Additionally, gastric emptying and the intestinal absorption of glucose also affect the exogenous glucose oxidation kinetics after oral consumption of labeled glucose. Unfortunately, the effects of exercise in hypoxia on gastric emptying and intestinal absorption remain unclear, although no study has reported that hypoxia altered gastric emptying or intestinal absorption.

Notably, the blood lactate elevation and CHO oxidation were significantly greater in HYPO, whereas ^{13}C -excretion during endurance exercise did not differ significantly between the two trials. Because ^{13}C -excretion during endurance exercise indicates exogenous glucose oxidation, the greater blood lactate elevation and CHO oxidation in HYPO may reflect facilitated muscle glycogen utilization (augmented endogenous glycogen utilization) during the exercise. The exercise in hypoxia promoted exercise-induced adrenaline elevation and sympathetic nerve activation (Wadley et al., 2006; Mazzeo, 2008; Katayama et al., 2010), and these promote muscle glycogenolysis during exercise (Watt et al., 2001; Wadley et al., 2006). Further research to address muscle glycogen utilization during endurance exercise in hypoxia would greatly aid our understanding of the physiological factors behind the augmented exercise-induced blood lactate elevation in hypoxia.

There was no significant difference in blood K^+ levels between the two trials. Our initial hypothesis was that HYPO would augment the exercise-induced blood K^+ elevation compared with NOR. Because lower pH increases the opening of K_{ATP} channels, it subsequently promotes K^+ efflux from working muscle into the bloodstream (Davies, 1990; Nielsen et al., 2002). The accumulation of extracellular K^+ has two possible explanations: (1) enhanced release of K^+ from the working muscle and (2) decreased K^+ re-uptake during exercise. During endurance exercise, the release of K^+ commonly exceeds the K^+ re-uptake, which consequently leads to the accumulation of K^+ in the interstitium and blood. Unfortunately, the effects of exercise in hypoxia on the muscle Na^+-K^+ -pump (K^+ re-uptake) remain unclear. Street et al. (2005) determined the effects of sodium citrate ingestion on exercise-induced acidosis and potassium accumulation. The sodium citrate-ingestion group (CIT) had significantly higher blood and interstitial pH after exercise compared with a placebo water-ingestion group (PLA). Furthermore, CIT caused significantly lower exercise-induced interstitial K^+ elevation compared with PLA (CIT: 8.0 mM, PLA: 11.0 mM). However, there was no significant difference in blood K^+ concentrations between CIT and PLA (CIT, 4.1 mM; PLA, 4.2 mM). Therefore, due to the lack of data on the interstitial K^+ response, caution is necessary when interpreting the K^+ response in our study.

In our previous study (Sumi et al., 2018b), we investigated the effect of endurance exercise (cycling exercise) in hypoxia on acid-base balance and K^+ responses. Consequently, endurance exercise in hypoxia caused higher blood pH and lower K^+ concentrations compared with the same exercise in normoxia, which was not consistent with outcomes in the present study in spite of similar study design. However, the difference in absolute workload between hypoxia and normoxia was greater in the above previous study (approximately 20% lower in hypoxia) compared with the present study (approximately 10% lower in hypoxia). Furthermore, different exercise modality (cycling exercise in the previous study vs. running exercise in the present study) may explain different outcomes between the two studies.

In conclusion, high-intensity interval running in moderate hypoxia elicited decreased blood pH and elevated blood lactate

despite the lower running velocity. However, it did not affect the exercise-induced blood K^+ elevation or exogenous glucose oxidation. Therefore, our findings suggest that, despite the lower mechanical stress (lower running velocity), endurance exercise in moderate hypoxia causes higher metabolic stress and similar exercise-induced elevations of blood K^+ and exogenous glucose oxidation compared with the same exercise in normoxia. The training in hypoxia may be an efficient training strategy to elicit training adaptations (e.g., improved CHO metabolism and muscle buffer capacity) with reduced risk of injury in lower legs.

AUTHOR CONTRIBUTIONS

DS and KG contributed to the study design, data collection, analysis, and manuscript writing. NK contributed to the data

collection, analysis. HI contributed to the study design, data collection, and analysis. All authors read and approved the final manuscript.

FUNDING

This study was supported by Grant-in-Aid for Scientific Research from Japan Society for the Promotion of Science.

ACKNOWLEDGMENTS

We would like to thank all of the participants who participated in the study.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Mild Hypobaric Hypoxia Enhances Post-exercise Vascular Responses in Young Male Runners

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OPEN ACCESS

Edited by:

Toby Mündel,
Massey University, New Zealand

Reviewed by:

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Gregoire P. Millet,
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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 04 December 2018

Accepted: 17 April 2019

Published: 24 May 2019

Citation:

Saito Y, Nakamura M, Eguchi K
and Otsuki T (2019) Mild Hypobaric
Hypoxia Enhances Post-exercise
Vascular Responses in Young Male
Runners. *Front. Physiol.* 10:546.
doi: 10.3389/fphys.2019.00546

It has been reported that sustained post-exercise vasodilation may be linked to exercise-induced angiogenesis. The present study aimed to evaluate whether mild hypobaric hypoxia enhances the post-exercise reduction in systemic vascular resistance in young male runners. Seven male intercollegiate runners (aged 19–21 years) performed maximal incremental treadmill running under conditions of hypobaric hypoxia (corresponding to 2,200 m above sea level, hereinafter referred to as HH) and normobaric normoxia (corresponding to sea level, hereinafter referred to as NN). A third exercise test was performed under NN conditions, consisting of submaximal exercise with the same absolute exercise volume as was achieved during HH (submaximal exercise under NN conditions, hereinafter referred to as NNsubmax). Blood pressure and cardiac output (CO) were measured before and at 15, 30, and 60 (p60) minutes after exercise. Compared with NN, exercise time was shorter in HH and NNsubmax conditions ($p < 0.05$). Systolic blood pressure and mean blood pressure (MBP) were lower after exercise in HH conditions ($p < 0.05$). No condition-related differences were found in CO. Total peripheral resistance (TPR, defined as the ratio of MBP to CO) was significantly lower after exercise compared to baseline for all conditions ($p < 0.05$). However, the decrease in TPR was maintained longer after exercise in HH compared with NN and NNsubmax conditions ($p < 0.05$). At p60, TPR was lower than baseline for HH conditions ($p < 0.05$), whereas after exercise in NN, and NNsubmax conditions, TPR recovered to baseline by p60. Decreases in systemic vascular resistance after exercise were maintained longer under mild HH conditions compared with NN despite the lower exercise volume of the former.

Keywords: athletes, maximal exercise, mild hypobaric hypoxia, vascular resistance, post-exercise vasodilation

INTRODUCTION

During recovery from exercise, dynamic changes in neural, and local factors induce immediate hyperemia (0–20 min after exercise) and sustained vasodilation (20–120 min after exercise) (Laughlin et al., 2012). Halliwill et al. (2013) proposed that sustained vasodilation after exercise may contribute to the growth and remodeling of microvasculature. Increased capillary supply improves the delivery of oxygenated blood to working muscles, and thus increases the arteriovenous

oxygen differences by increasing the diffusion area within tissues. These changes lead to increased oxygen uptake and consequent improved performance in endurance exercise (Brodal et al., 1977). Therefore, establishing a method which can prolong or enhance sustained post-exercise vasodilation may be beneficial for endurance athletes who require maximal aerobic capacity.

Altitude training is a popular method used by competitive athletes, because hematological mechanisms (changes in red blood cell mass), and non-hematological mechanisms (such as angiogenesis, glucose transport, glycolysis, and PH regulation) of adaptation to hypoxia may enhance competitive performance (Gore et al., 2007; Girard et al., 2017). Additionally, combining exposure to hypoxia with exercise has been reported to improve O₂ transport and/or metabolism within muscles (Lundby et al., 2009), suggesting that exercise under such conditions may induce angiogenesis. If angiogenesis is in fact induced by post-exercise vasodilation, then exercise-induced vasodilation might be enhanced at higher altitudes. Indeed, inspiration of hypoxic gas has been shown to increase vascular conductance during submaximal exercise via the endothelial function-related pathway, as described by Casey et al. (2010), and enhance flow-mediated dilation (FMD) after submaximal exercise (Katayama et al., 2013) in young sedentary males. However, hemodynamics after intense (maximal and supramaximal) endurance exercise under conditions of hypobaric hypoxia have not been explored. It has been reported that maximal exercise under normoxic conditions causes a transient increase in reactive hyperemia (Schroeder et al., 2019), and that high-intensity interval training under hypoxic conditions improves maximal oxygen uptake and performance during repeated-sprint exercise (Brocherie et al., 2017). Studies on endurance athletes could have clinical implications for other athletes because the effects of exercise and altitude may differ between trained and sedentary individuals.

The accompanying negative effects of altitude training must be considered along with the training benefits. The incidence and severity of acute mountain sickness symptoms such as headaches, dizziness, fatigue, and restless sleep depend on the altitude reached and rate of ascent (Hackett et al., 1976). The hypoxic conditions in the studies mentioned above (Casey et al., 2010; Katayama et al., 2013) were relatively severe, at <80% arterial oxygen saturation (SpO₂); in terms of safety, studies in a milder hypoxic environment are important. To the best of our knowledge, there have been no investigations into the effects of exercise under mild hypoxic conditions on sustained post-exercise vasodilation.

We hypothesized that mild hypobaric hypoxia can enhance sustained post-exercise vasodilation in endurance athletes. To test this hypothesis, we evaluated the effects of artificial altitude conditions equivalent to 2,200 m above sea level on the systemic vascular resistance after maximal endurance exercise in young male runners. We chose 2,200 m because the arterial O₂ saturation reduced at this altitude may be sufficient to stimulate vasodilation after exercise, as indicated by a previous study which reported that 8 weeks of aerobic training under similar hypobaric hypoxic conditions improved FMD, and arterial stiffness in postmenopausal women (Nishiwaki et al., 2011).

MATERIALS AND METHODS

Subjects

Seven male intercollegiate long-distance runners were recruited for this study [age (mean \pm standard deviation) 19.9 ± 0.9 years; height, 173.0 ± 4.3 cm; weight, 57.8 ± 6.2 kg]. None of the subjects had cardiovascular disease, including hypertension.

A power calculation was performed to calculate adequate sample size for repeated measures two-way analysis of variance (ANOVA) analysis of post-exercise changes in systolic blood pressure (SBP). We used the G*Power 3 program (Faul et al., 2007). The sample size of this study ($n = 7$) was adequate to detect an interaction at 80% power with an α value of 5% when effect size was assumed as the medium (0.25) (Cohen, 1992).

All subjects gave written informed consent after verbal explanation of this study and the anticipated risks, in accordance with the Declaration of Helsinki. The study and all its procedures were carried out in accordance with the recommendations of, and approved by, the Ethics Committee of the Japan Institute of Sports Sciences.

Procedure

Trials were conducted under conditions of normobaric normoxia (760 mmHg, corresponding to sea level, hereinafter denoted NN) and hypobaric hypoxia (568 mmHg, corresponding to 2,200 m above sea level, hereinafter denoted HH) in a hypobaric chamber. In our pilot study, SpO₂ was approximately 10% lower in HH compared with NN conditions, and HH conditions resulted in decreased exercise time. Thus, as the third condition, submaximal exercise was performed with the same absolute workload as in HH conditions, under NN conditions (hereinafter denoted NNsubmax, 760 mmHg). This aimed to compare the effects of exercise of equivalent workload in hypoxic vs. normoxic conditions. Subjects underwent NN and HH trials in a randomized order. The NNsubmax trial was conducted after the HH trial, or after both the HH and NN trials.

Subjects visited the chamber four times over a period of 3–16 days. During the first visit, we explained the study in detail and the subjects became accustomed to using the experimental instruments. Data collection was carried out during the other three visits. Changes in cardiovascular status for the three different conditions were measured during a 60-min recovery period after acute exercise. Subjects fasted for 12 h prior to each test and were instructed to avoid strenuous activity and caffeine for 24 h prior to each test.

Exercise

Exercise Mode

The exercise test consisted of running on a treadmill (Biomill, 4 Assist, Tokyo, Japan). Subjects warmed up on the treadmill for 10 min while heart rate (HR) was monitored with an electrocardiographic signal transmitter (ZS-910P, Nihon Kohden, Tokyo, Japan). Running speed was self-paced and therefore not defined. Next, the maximal incremental running test was performed. The test began with the treadmill speed set at 286 m/min and the speed was increased by 14–20 m/min every

3 min in the first four stages. Starting at the fifth stage, the speed was increased by 11 m/min every 3 min. In the NN and HH trials, the test continued until subjects became exhausted. For the NNsubmax trials, the trial ended when subjects reached the time that was attained in the previous HH trial.

The exercise test was discontinued if three or more of the following five criteria were met: (1) a rating of perceived exertion greater than 17 on the Borg Scale, (2) a respiratory exchange ratio greater than 1.1, (3) no increase in HR with increasing running speed, (4) plateaued oxygen uptake ($\dot{V}O_2$) (increase of 150 mL or less) with increasing running speed, and (5) subject request for discontinuation due to fatigue.

Exercise-Related Measurements

Ventilatory parameters and the oxygen (O_2) and carbon dioxide (CO_2) concentrations of expired air were analyzed breath-by-breath during the running test using an open-circuit spirometry gas analysis system (AS300, Minato Medical Science, Osaka, Japan). The system was calibrated prior to measurements under NN and HH conditions with a calibration gas of known O_2 and CO_2 concentration and constant volume.

Mean minute ventilation ($\dot{V}E$), $\dot{V}O_2$, CO_2 production ($\dot{V}CO_2$), and HR were calculated for every 30 s of exercise. The mean values of $\dot{V}E$, $\dot{V}O_2$, and $\dot{V}CO_2$ during exercise were defined as $\dot{V}E_{mean}$, $\dot{V}O_{2mean}$, and $\dot{V}CO_{2mean}$, respectively. Peak $\dot{V}O_2$ and HR were defined as $\dot{V}O_{2peak}$ and HR_{peak} , respectively.

Post-exercise

Rest and Recovery

The air temperature in the chamber was maintained between 23 and 25°C. Baseline measurements of cardiovascular parameters were taken after the subjects had rested for at least 15 min in the supine position. Five minutes after the completion of the exercise, the subjects were instructed to rest in the supine position for 55 min (i.e., to reach 60 min after exercise), and the cardiovascular status was measured at 15 (p15), 30 (p30), and 60 (p60) minutes after cessation of exercise. Subjects were allowed to drink water *ad libitum* during the experimental period.

Cardiovascular Measurements

Oxygen saturation

Measurements of SpO_2 were performed using a pulse oximeter (OLV-3100, Nihon Kohden, Tokyo, Japan) placed on the tip of the left index finger before exercise.

Arterial blood pressure and heart rate

Diastolic blood pressure (DBP), SBP, and HR were each measured three times at the upper right arm using the oscillometric method (Jentow, Nihon Colin, Aichi, Japan). The mean values of the three measurements of each parameter were used for analysis. Mean blood pressure (MBP) was estimated using the following formula: $MBP = 2/3(DBP) + 1/3(SBP)$.

Cardiac function

Echocardiography was performed (SSD-6500, Aloka Company, Tokyo, Japan) according to the recommendations of the American Institute of Ultrasound in Medicine (Sahn et al., 1978). First, B-mode long axis views of the left ventricle from the left

parasternal region were obtained with a 1.88-MHz sector probe. Next, M-mode measurements were made over 10 continuous heartbeats. Echocardiographic video images were converted from analog to digital information (Mini Converter, Blackmagic Design, Fremont, CA, United States), exported, and stored in a general-purpose computer. The images were analyzed offline using image-processing software (ImageJ, National Institute of Health, Bethesda, MD, United States). Left ventricular end-diastolic diameter and left ventricular end-systolic diameter were measured, and the mean values over three heartbeats calculated. End-diastolic volume and end-systolic volume were calculated using the method of Teichholz et al. (1976). Stroke volume was obtained by subtracting left ventricular end-systolic volume from left ventricular end-diastolic volume. CO was calculated as the product of HR and stroke volume. Total peripheral resistance (TPR) was calculated using the following formula: $TPR = MBP/CO$. The between-day coefficient of variation for stroke volume was calculated to be 6.8% in the present study.

Flow-mediated dilation

Measurements of FMD were taken at baseline and p60, after cardiovascular parameters were measured. Subjects were placed in the supine position with the right arm in 90 degrees of abduction. The length between the acromion and olecranon was measured, and the midpoint was marked to identify the location for probe placement. Ultrasonographic images were obtained and analyzed in the same manner as echocardiographic images. A longitudinal image of the brachial artery and a Doppler image of brachial blood flow were recorded simultaneously in duplex mode using a 5-MHz linear probe. The insonation angle was set at <65 degrees to the direction of blood flow (Picot and Embree, 1994). The sample volume gate was adjusted to cover the width of the vessel. Images were recorded for 1 min at rest. Next, a rapid cuff inflator (E-20, D. E. Hokanson, Bellevue, WA, United States) on the right forearm was inflated to 250 mmHg for 5 min to occlude blood flow in the forearm. After reperfusion, there was a recovery period of 2 min. Vessel diameter and blood flow velocity were recorded continuously from 1 min before ischemia to 2 min after reperfusion.

Vessel diameter was measured every 10 s. Three measurements were made for each image and the mean value was calculated. The mean blood velocity (MBV) of one cardiac cycle was measured at the same time as diameter measurements were taken. The percent increase in vessel diameter from baseline to maximum vessel diameter after reperfusion was defined as %FMD. Shear rate was calculated using the following formula: $\text{shear rate} = 8 \times \text{vessel diameter}/\text{MBV}$ (Parker et al., 2009). The shear rate area under the curve was calculated as the sum total of shear rates every 10 s from the onset of cuff deflation to the point of maximum vessel diameter. We excluded one subject from the analysis due to technical problems during the NNsubmax trial. The between-day coefficient of variation for vessel diameter was found to be 0.8% in the present study.

Statistical Analysis

Data are presented as means \pm standard deviation. The significance level was set at 0.05. We used SPSS version

21 (IBM Co., Armonk, NY, United States) for all statistical analyses. One-way (condition) ANOVA was used for saturation and exercise parameters. Two-way (condition \times time) repeated-measures ANOVA was used to analyze time-dependent changes in cardiovascular parameters. For *post hoc* analysis, the Bonferroni method was used where significant values were found.

RESULTS

Oxygen Saturation

We confirmed that SpO₂ in HH conditions was $93.0 \pm 1.4\%$, which was lower than that in NN ($99.1 \pm 0.7\%$) or NNsubmax conditions ($99.0 \pm 0.6\%$) ($p < 0.05$).

Exercise Parameters

Recorded exercise parameters are shown in **Table 1**. Exercise time, an index of performance, was shorter in HH and NNsubmax conditions compared with NN ($p < 0.05$). There were no differences in $\dot{V}CO_{2\text{mean}}$, $\dot{V}O_{2\text{mean}}$, $\dot{V}O_{2\text{peak}}$, and HRpeak between the conditions, although the difference in $\dot{V}O_{2\text{peak}}$ per kg body weight was close to statistical significance ($p = 0.07$). However, $\dot{V}E_{\text{mean}}$ was higher in HH than in the other two conditions ($p < 0.05$).

Post-exercise Hemodynamics

Time course measurements for blood pressures are shown in **Figures 1A–C**. Both SBP and MBP were lower at p30 and p60 than baseline in HH conditions ($p < 0.05$). Regarding DBP, no difference was observed between the trials. The condition-related differences in HR, as assessed

TABLE 1 | Exercise parameters.

	NN	HH	NNsubmax	<i>p</i> -value
Exercise time, min	13.3 ± 2.6	$10.2 \pm 1.3^*$	$10.2 \pm 1.3^*$	$p < 0.05$
$\dot{V}E_{\text{mean}}$, L/min	99 ± 9	$119 \pm 13^{*\dagger}$	88 ± 8	$p < 0.05$
$\dot{V}CO_{2\text{mean}}$, L/min	3.06 ± 0.29	3.15 ± 0.26	2.88 ± 0.30	NS
$\dot{V}O_{2\text{mean}}$, L/min	3.28 ± 0.35	3.14 ± 0.32	3.19 ± 0.33	NS
$\dot{V}O_{2\text{peak}}$, L/min	3.52 ± 0.37	3.27 ± 0.31	3.42 ± 0.32	NS
$\dot{V}O_{2\text{peak}}$, mL/min/kg	61.0 ± 3.7	56.2 ± 4.5	58.8 ± 2.8	$p = 0.07$
HRpeak, beats/min	190 ± 12	182 ± 5	180 ± 11	NS

Data are presented as means \pm standard deviation. Key: * $p < 0.05$, compared with NN; $^{\dagger}p < 0.05$, compared with NNsubmax. NN, maximal exercise under normobaric normoxic conditions; HH, maximal exercise under hypobaric hypoxic conditions; NNsubmax, submaximal exercise under normobaric normoxic conditions; NS, non-significant; $\dot{V}E_{\text{mean}}$, mean minute ventilation during exercise; $\dot{V}CO_{2\text{mean}}$, mean carbon dioxide production during exercise; $\dot{V}O_{2\text{mean}}$, mean oxygen uptake during exercise; $\dot{V}O_{2\text{peak}}$, peak oxygen uptake; HRpeak, peak heart rate.

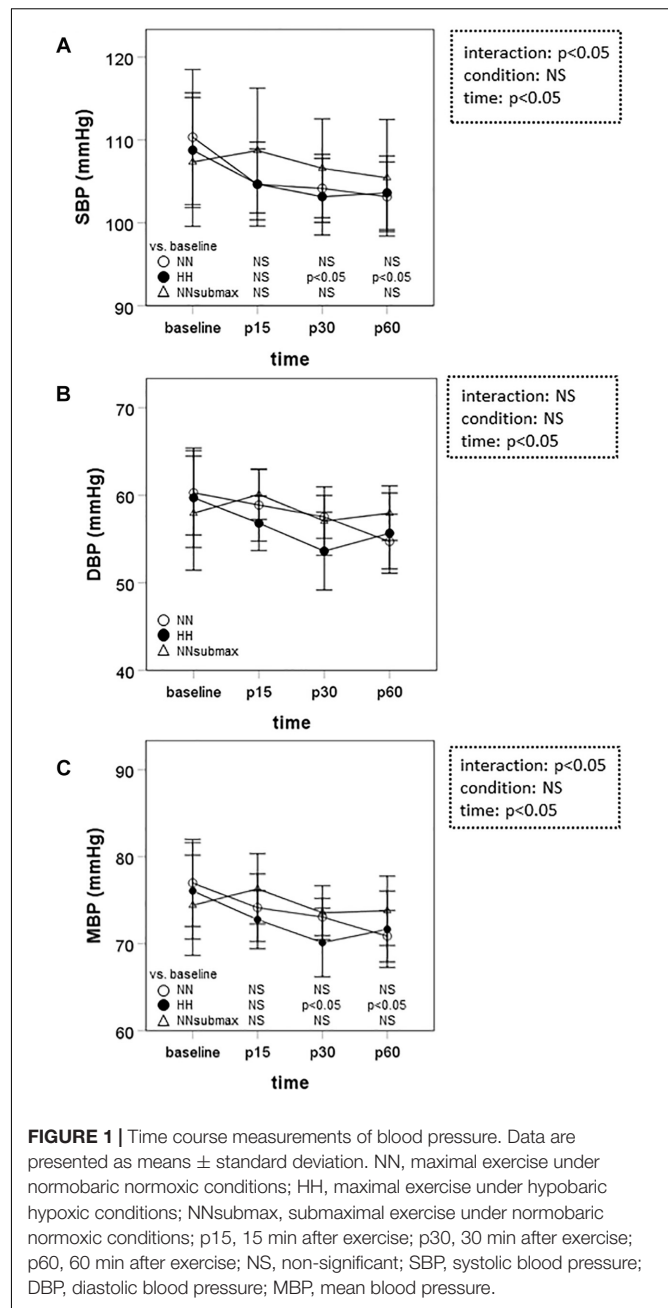


FIGURE 1 | Time course measurements of blood pressure. Data are presented as means \pm standard deviation. NN, maximal exercise under normobaric normoxic conditions; HH, maximal exercise under hypobaric hypoxic conditions; NNsubmax, submaximal exercise under normobaric normoxic conditions; p15, 15 min after exercise; p30, 30 min after exercise; p60, 60 min after exercise; NS, non-significant; SBP, systolic blood pressure; DBP, diastolic blood pressure; MBP, mean blood pressure.

by ANOVA, were not statistically significant between HH and NNsubmax ($p = 0.06$) (**Table 2**). Neither stroke volume (**Table 2**) nor CO (**Figure 2**) showed any significant differences between the conditions.

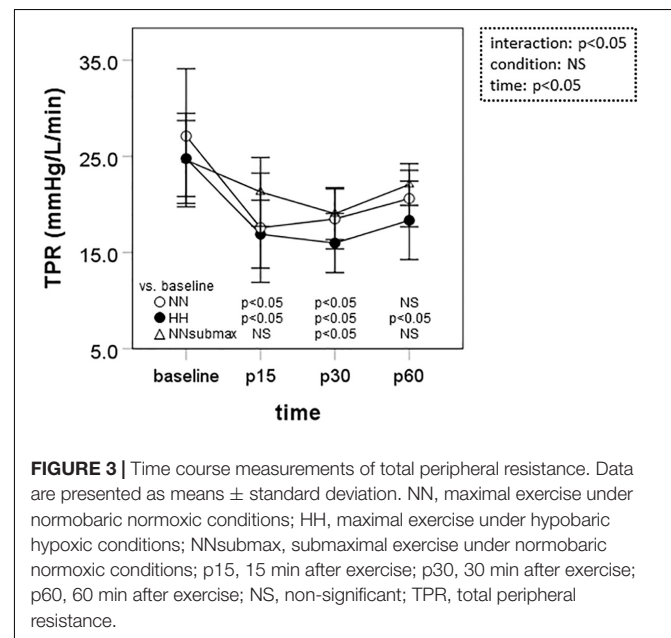
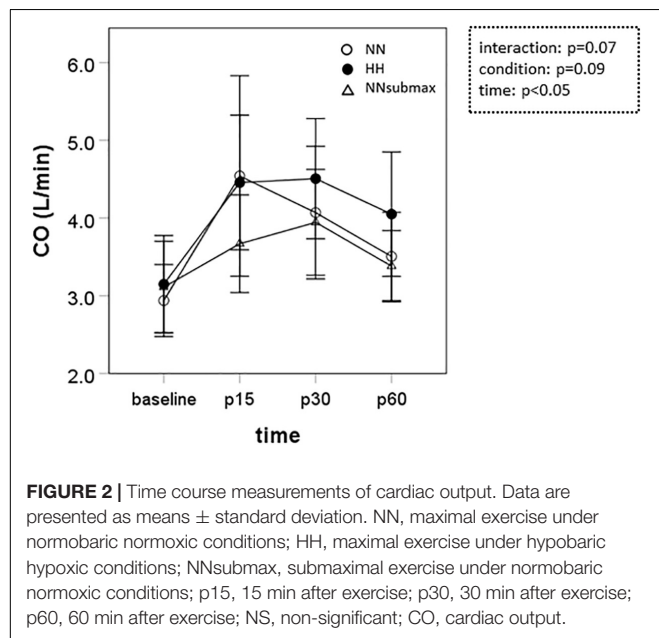
Time course measurements of TPR are shown in **Figure 3**. Under HH conditions, TPR was below baseline until the end of the measurement period ($p < 0.05$). In NN and NNsubmax conditions, TPR was only lower than baseline until p30 ($p < 0.05$).

The FMD parameters are shown in **Table 3**. The pre-occlusion diameter at p60 was higher in HH compared with NNsubmax conditions ($p < 0.05$). There were no other significant differences

TABLE 2 | Values and results of analysis of variance of cardiac function before and after exercise.

	Condition	Baseline	p15	p30	p60	p-value
HR, beats/min	NN	49 ± 7	73 ± 13	64 ± 14	58 ± 9	Interaction: NS
	HH	50 ± 9	75 ± 9	67 ± 10	60 ± 11	Condition: $p < 0.05$
	NNsubmax	48 ± 10	67 ± 11	60 ± 11	55 ± 8	Time: $p < 0.05$
SV, mL	NN	60 ± 12	62 ± 13	64 ± 7	61 ± 9	Interaction: $p = 0.05$
	HH	64 ± 11	59 ± 12	67 ± 11	68 ± 11	Condition: NS
	NNsubmax	65 ± 13	55 ± 8	66 ± 6	62 ± 6	Time: $p < 0.05$

Data are presented as means ± standard deviation. NN, maximal exercise under normobaric normoxic conditions; HH, maximal exercise under hypobaric hypoxic conditions; NNsubmax, submaximal exercise under normobaric normoxic conditions; p15, 15 min after exercise; p30, 30 min after exercise; p60, 60 min after exercise; NS, non-significant; HR, heart rate; SV, stroke volume.



between baseline and p60 under any of the studied conditions except for the peak diameter ($p < 0.05$).

DISCUSSION

We hypothesized that mild hypobaric hypoxia enhances sustained post-exercise vasodilation in endurance athletes. This is the first study to investigate the effects of mild hypoxia on vasodilation after intense endurance exercise. The differences in SBP, MBP, and TPR responses to exercise under the different conditions of this study suggest that the reduction in systemic vascular resistance after intensive endurance exercise persists longer after exercise in mild HH conditions despite the lower exercise volume.

Systemic vascular resistance is controlled by neural and local factors (Pescatello et al., 2004). Sympathetic activity is a neural factor that increases systemic vascular resistance. In general, sympathetic activity increases with short-term exposure to hypoxia via chemical receptors (Smith and Muentert, 2000). In the conditions of the present study, however,

hypoxia-induced sympathetic vasoconstriction may have been overcome by compensatory vasodilation. With regards to local factors, hypoxia augments release of the endothelium-derived vasodilators adenosine (Leuenberger et al., 1999), prostaglandin (Messina et al., 1992), and nitric oxide (NO) (Casey et al., 2010). Hypoxia-induced expression of endothelial NO synthase mRNA (Shaul et al., 1995; Le Cras et al., 1998; Xiao et al., 2001) may be implicated in hypoxia-induced vascular responses because receptors for hypoxia-inducible factor, a transcriptional regulator, are present in the endothelial NO synthase promoter area (Coulet et al., 2003). However, brachial artery FMD, which represents an index of NO-mediated vasodilation, was not affected by hypoxia in the present study. Katayama et al. (2016) reported that reactive hyperemia in response to exercise was not enhanced in the inactive limb of subjects breathing a hypoxic gas mixture (12.0% fraction of inspired oxygen, FiO_2) compared to a normoxic gas mixture (21.0% FiO_2), although it was enhanced in the active limb (Katayama et al., 2013). We measured FMD in the upper arm, which was active, although secondary to the lower limbs. Effects of hypoxia on post-exercise FMD might have been identified if FMD had been measured

TABLE 3 | Values and results of analysis of variance of flow-mediated dilation.

	Condition	Baseline	p60	p-value
Brachial artery pre-occlusion diameter, mm	NN (n = 7)	3.8 ± 0.2	3.9 ± 0.3	Interaction: $p < 0.05$
	HH (n = 7)	3.9 ± 0.2	3.9 ± 0.3	Condition: NS
	NNsubmax (n = 6)	3.9 ± 0.2	3.8 ± 0.3*	Time: NS
Brachial artery peak diameter, mm	NN (n = 7)	4.1 ± 0.2	4.1 ± 0.2	Interaction: NS
	HH (n = 7)	4.1 ± 0.3	4.2 ± 0.2	Condition: NS
	NNsubmax (n = 6)	4.0 ± 0.3	4.1 ± 0.3	Time: $p < 0.05$
FMD, %	NN (n = 7)	5.6 ± 3.5	4.0 ± 4.5	Interaction: NS
	HH (n = 7)	5.7 ± 2.2	8.6 ± 4.2	Condition: NS
	NNsubmax (n = 6)	4.0 ± 2.0	7.5 ± 1.2	Time: NS
SRauc, AU	NN (n = 7)	588 ± 139	597 ± 147	Interaction: NS
	HH (n = 7)	498 ± 158	616 ± 229	Condition: NS
	NNsubmax (n = 6)	548 ± 221	464 ± 79	Time: NS
FMD/SRauc	NN (n = 7)	0.011 ± 0.008	0.006 ± 0.007	Interaction: NS
	HH (n = 7)	0.012 ± 0.005	0.016 ± 0.009	Condition: NS
	NNsubmax (n = 6)	0.014 ± 0.021	0.017 ± 0.004	Time: NS

Data are presented as means ± standard deviation. Key: * $p < 0.05$, compared with HH. NN, maximal exercise under normobaric normoxic conditions; HH, maximal exercise under hypobaric hypoxic conditions; NNsubmax, submaximal exercise under normobaric normoxic conditions; p60, 60 min after exercise; NS, non-significant; FMD, flow-mediated dilation; SRauc, shear rate area under the curve; AU, arbitrary unit. We excluded one subject from the analysis due to technical problems during the NNsubmax trial.

in the arteries of the leg. On the other hand, it is possible that local factors other than NO contribute to the impact of hypoxia on TPR. A previous study showed that sustained post-exercise vasodilation is dependent on activation of the histamine H1 and H2 receptors (McCord and Halliwill, 2006). However, it remains unclear whether the histamine response is modified by hypoxia. Further studies are needed to understand the details of the mechanisms underlying the prolonged post-exercise decrease in systemic vascular resistance that was observed in HH conditions in this study.

Most of the subjects of previous studies on hypoxic vasodilation were sedentary young individuals (Casey et al., 2010; Katayama et al., 2013). In contrast, we showed that competitive runners exhibited sustained post-exercise decreases in systemic vascular resistance after exercise in mild HH conditions. The expected angiogenesis of the microvasculature, which may be related to post-exercise sustained vasodilation, is more important in such endurance athletes due to their requirement for increased aerobic capacity compared with sedentary individuals. In addition, HH conditions may be more beneficial than conditions of normobaric hypoxia for endurance-trained athletes who compete in high-speed running, because the reduced air density modifies the air resistance, and facilitates high-speed movements (Ward-Smith, 1984). However, normobaric hypoxia may have other benefits for endurance athletes (Millet et al., 2012).

Our findings could contribute to the development of a new training method for athletes. Furthermore, elderly or hypertensive subjects who need to improve their vascular function may benefit from hypoxic exercise. The combination of hypoxic stimuli and exercise improves several parameters of vascular function, including reduced blood pressure, which are pertinent for the reduction of cardiovascular risks (Millet et al., 2016). Therefore, we propose that altitude exercise has possible clinical applications to various populations, although

further studies are needed for this potential to be realized. First, although we have demonstrated that the post-exercise reduction in SBP, MBP, and TPR persist for longer after exercise in HH conditions, we did not observe the recovery of baseline for these parameters. Future studies of post-exercise hemodynamics with increased observation periods could provide important insight. Second, the effects of hypoxia on vasculature depend on the hypoxic level, as indicated by the finding that mortality from stroke decreases with increasing altitude of the place of residence (12% per 1,000 m) (Faeh et al., 2009). The effects of low- and high-intensity hypoxia may differ with regards to the mechanisms involved in post-exercise vasodilation. Identifying the optimal altitude for enhancement of post-exercise vasodilation would be highly significant for athletes and clinicians.

The present study had some limitations. First, we examined only acute-vessel responses to a single bout of endurance exercise. A long-term intervention study is needed to ascertain whether the sustained vasodilation after exercise under conditions of hypoxia contributes to growth and remodeling of the microvasculature. Second, since we studied young male collegiate runners, we cannot generalize the present findings to the entire population. Third, the sample size was relatively small, and we found that some differences in measurements did not reach statistically significant level.

CONCLUSION

Our findings suggest that reductions in systemic vascular resistance induced by endurance exercise might persist longer under mild HH conditions (equivalent to 2,200 m above sea level), even when the absolute exercise volume is reduced, and compared with NN conditions (equivalent to sea level). Mild HH

and post-exercise hemodynamics seem to have additive effects in young runners when compared with NN conditions.

ETHICS STATEMENT

This study was carried out in accordance with the recommendations of Ethical Guidelines for Medical and Health Research Involving Human Subjects, the Ministry of Education, Culture, Sports, Science and Technology of Japan with written informed consent from all subjects. All subjects gave written informed consent in accordance with the Declaration of Helsinki. The protocol was approved by the Ethics Committee of the Japan Institute of Sports Sciences.

AUTHOR CONTRIBUTIONS

YS contributed to the entire process of the work including the design of the study, acquisition, analysis, interpretation of the

data, and writing of the manuscript. MN and KE contributed to the acquisition and interpretation of the data. TO contributed to the interpretation of data and the critical repeated revisions of the manuscript. All authors contributed to manuscript revision and have read and approved the submitted version.

FUNDING

This study was funded by a Grant-in-Aid for Scientific Research No. 23700855 from the Japan Society for the Promotion of Science.

ACKNOWLEDGMENTS

We thank Prof. Seiji Kushibe of Josai University, his track and field club, and the medical staff of the Japan Institute of Sports Sciences. We are especially grateful to Dr. Ryuichi Ajisaka of the University of Tsukuba for informative advice on the manuscript.

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- Conflict of Interest Statement:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Conventional and Alternative Strategies to Cope With the Subtropical Climate of Tokyo 2020: Impacts on Psychological Factors of Performance

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OPEN ACCESS

Edited by:

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Specialty section:

This article was submitted to
Movement Science and Sport
Psychology,
a section of the journal
Frontiers in Psychology

Received: 04 December 2018

Accepted: 15 May 2019

Published: 04 June 2019

Citation:

Coudeville GR, Sinnaph S,
Robin N, Collado A and Hue O (2019)
Conventional and Alternative
Strategies to Cope With
the Subtropical Climate of Tokyo
2020: Impacts on Psychological
Factors of Performance.
Front. Psychol. 10:1279.
doi: 10.3389/fpsyg.2019.01279

The thermal discomfort caused by a hot or hot-wet climate can have negative effects on human performance. The 2020 Summer Olympic and Paralympic Games will take place in Tokyo's hot and humid summer period, possibly exposing athletes to severe environmental stressors. In addition to technical, tactical, physical and nutritional preparation, Olympians and Paralympians need an optimal psychological state to turn in their best performances, especially in terms of emotional control, concentration and motivation. Yet, the tropical climate can have many negative effects on these factors. Better understanding of the negative effects of this climate and the strategies to manage them might be crucial for competitors, coaches and their teams in Japan. At the psychological level, cooling interventions before, during and/or immediately after exercise were mainly studied on perceptual responses. However, the effects of these interventions on other psychological components such as cognitive abilities or psychological states and the use of psychological techniques have been little explored, especially in hot-wet climate. Thus, this article proposes to take stock of the knowledge on the conventional and alternative strategies that help athletes to psychologically cope with the subtropical climate of Tokyo.

Keywords: cognitive abilities, hot-wet climate, heat stress, cooling, thermal comfort, mental technique

INTRODUCTION

Competing at the Olympic/Paralympic Games requires high level physical, technical, tactical, nutritional, and mental preparation. Athletes have to manage event stressors like anxiety (Nicholls and Levy, 2016) and competitive stressors like preparation, expectations, and opponents (Fletcher and Sarkar, 2012). Tokyo, lying in a humid subtropical zone, will add another important stressor: the hot and wet climate in the summer season. The Köppen Climate Classification subtype for this climate is warm temperature, fully humid and hot summer (i.e., subtropical climate). The hottest month of the year is August with an average temperature of 27.4°C (81.3°F) corresponding to the month of the Games. The average temperatures in Tokyo have risen by three degrees Celsius in the last one hundred years. Two degrees of these three are attributed to the urban heat island (UHI) effect and one to global warming. UHI is a kind of heat accumulation phenomenon in which temperatures in urban areas are markedly higher than those in surrounding areas

(Matsumoto et al., 2017). One of the causes of this effect is asphalt concrete (i.e., the most common pavement surfacing materials) exposed to solar irradiation (Mohajerani et al., 2017). During the competitions, athletes from certain disciplines will be directly exposed (e.g., triathlon, road cycling, marathon, 20 and 50 km race walk). The particularities of Tokyo 2020 are required to mobilize all strategies to succeed in achieving sports performance in high thermal stresses without risk for health. Associated with intense physical activity, if not regulated, these high thermal stresses may provoke hyperthermia, which disrupts brain function (e.g., cerebral blood flow reduction, brain temperature increase, and cerebral oxygenation compromising), which in turn alters the cognitive abilities (Bain et al., 2015). Therefore, it is important to determine strategies that allow better regulation (e.g., cooling intervention) and better adaptation to this hyperthermia (e.g., acclimatization, mental preparation) without risk to health. In addition to the heat-related physiological problems that affect athletes' health, notably heat stroke (Brotherhood, 2008), the thermal environment can have negative consequences on performance (Hue, 2011). During prolonged aerobic exercise, this stressor is known to negatively impact both physiological (e.g., Maughan et al., 2012) and psychological performances (e.g., Vasmatazidis et al., 2002). The literature proposes cooling strategies to deal with these deleterious effects of heat (Bongers et al., 2017). Regarding the psychological components, most studies are interested in the perceptual responses of heat stress with or without cooling intervention. However, the effects of these interventions on other psychological components such as cognitive abilities (e.g., reaction time, attention, executive function) or psychological states (i.e., mood, emotional, expectation, and motivational effects), and the use of psychological interventions, are little known, specifically in the tropical climate (i.e., TropC). This brief review deals with the available strategies to cope with the impact of hot and hot-wet climates on perceptual responses and cognitive abilities. We first focus on the effects of conventional strategies (i.e., training in the thermal conditions and cooling interventions). We then focus on alternative or complementary strategies (i.e., menthol ingestion techniques and mental techniques). We also aimed to study the impact of these different strategies on other psychological components that have not been examined so far (e.g., self-confidence, motivation, flow states). In addition to the theoretical interest, this article provides information to coaches on how best to prepare their athletes for the climate conditions of Tokyo 2020 but also for the upcoming events (e.g., 2022 FIFA World Cup Qatar). To establish this mini-review, citations from *Pubmed* and *Sciencedirect* were identified from the earliest record until April 2019 using the following search terms: hot climate, hot-wet climate, strategies, sport, performance, and psychology. Included studies required using adult participants (≥ 18 years) and ambient temperatures ($\geq 35^{\circ}\text{C}$ and $\leq 40\%$ rH for studies conducted in hot climates and around 31°C and $\geq 50\%$ rH for studies conducted in TropC). All studies without any link or involvement in competitive physical activities were excluded.

IMPACTS OF CONVENTIONAL STRATEGIES TO DEAL WITH THERMAL STRESS

The main usual strategies to limit the negative effects of heat or TropC are training in the thermal stress and cooling interventions (for a review, see Bongers et al., 2017). For greater clarity, we distinguished the studies conducted in hot climate from those conducted in TropC and distinguished the factors related to perceptual responses from those related to cognitive abilities.

Training in Hot and Tropical Climate In Hot Climate

Thermal adaptation encompasses physiological factors, behavioral adjustments, and psychological factors (Marialena and Koen, 2003). According to Pryor et al. (2019), heat acclimation in a hot environment is induced by repeated exercise-heat exposures that result in temporary physiological adaptations. This reduces thermal load and cardiovascular strain, and improves heat dissipation mechanisms during exercise. In addition, Taylor (2014) reviewed that the adaptations previously evoked can improve aerobic performance, enhance exercise-heat tolerance, and reduce the risk of exertional heat illness. Likewise, a series of studies investigated whether training in the same thermal conditions as the competition would enable athletes or soldiers to better adapt to heat stress (see Heathcote et al., 2018, for a review). Several studies showed an improved thermal comfort after acclimation (Sunderland et al., 2008; Costa et al., 2014). Recently, Malgoire et al. (2018) compared the effects of a 15-day aerobic training program on soldiers in a hot-dry versus a temperate environment. They showed that the physiological modifications (e.g., rectal temperature) were mostly the same in the two environments but that thermal discomfort and the rating of perceived exertion (RPE) were much lower in the heat-training group than in the control group. Equivalent results were obtained for thermal comfort, sensation and perceived exertion in athletes prior to the Marathon des Sables following short-term heat acclimation (Willmott et al., 2017). Being accustomed to withstanding high training loads and high environmental stresses, athletes perceive the difficulty of an exercise as less onerous than the objective evaluation of it, especially compared to less well-accustomed athletes.

Several studies have shown the positive effects of thermal adaptation on cognitive abilities. Radakovic et al. (2007) examined the effects of exertional heat stress and acclimation status on the physiological and cognitive abilities of soldiers. The participants performed an exertional heat stress test in a cool environment, a hot environment to which they were not acclimatized, or a hot environment after undergoing 10 days of passive or active acclimation. The results showed that participants in the acclimatized group did not experience any adverse effects of heat stress, unlike the group of non-acclimatized participants who experienced a slight decrease in attention. Although different, these populations of athletes and soldiers may

have commonalities in coping with a stressful environment (e.g., rigor, resilience, determination).

In Tropical Climate

The literature on the impact of acclimation to TropC on psychological factors is far sparser. Schmit et al. (2017), testing the impact of an 8-day training camp, with or without a cooling vest, on 13 triathletes who performed two 20-km cycling time-trials in TropC (35°C and 50% rH), found positive effects on thermal comfort. Then, it can be assumed that acclimation to the environment reduces the effects of heat stress and thus avoids the risk of cognitive disturbances. Thus, training in TropC may be one of the most important strategies to sensitize the delegations to Tokyo 2020. In this regard, Racinais et al. (2015) recommended repeated exercise-heat exposures over 1–2 weeks, as well as incentives for hydration during the athletes' effort.

Cooling Interventions

Cooling interventions, such as cold-water immersion/ingestion or cooling garments, have been developed to prevent the physiological and psychological consequences of heat and TropC (for a review, see Jones et al., 2012; Bongers et al., 2017).

In Hot Climate

Ruddock et al. (2017) reviewed the effectiveness of cooling strategies during continuous exercise in heat. The results were from studies in a hot climate with lower relative humidity, but they are nonetheless interesting and indicate that cooling decreases the RPE and thermal perception during fixed-intensity exercise, which would improve endurance performance. Several studies have also found that head cooling decreases the RPE and thermal discomfort during hyperthermic exercise (e.g., Armada-da-Silva et al., 2004; Mündel et al., 2007). For example, Simmons et al. (2008) tested nine physically active, non-heat-acclimated volunteers. Participants performed two exercises (i.e., 12-min constant-load cycling tests at 70% VO_2max) separated by a 90-min period of passive heating in two conditions: with or without head and face cooled. They showed that head cooling during passive heating reduced RPE and improved thermal comfort during subsequent exercise in the heat.

While cooling interventions in the heat appear to have positive effects on the RPE and thermal comfort, the benefits to cognitive abilities are less clear-cut. Their effectiveness seems limited and dependent on the timing of implementation (Schmit et al., 2017) and the type of task (Shibasaki et al., 2017). Along this line, Gaoua et al. (2011) assessed whether attention and memory task performance would be positively affected by cold pack application to the head during passive heating compared to passive heating without cold packs and a control situation. Head cooling had a more positive effect on working memory capacity, and rapid visual processing was no longer negatively impacted, which was the case for passive heating without the cold packs. However, head cooling showed no beneficial effect on pattern recognition memory. In another experiment with three conditions (hot, 50°C vs. hot, 50°C with cold packs vs. control, 20°C), Racinais et al. (2008) investigated whether cooling

(i.e., cold packs to the head) would limit the alterations in motor training and cognitive function induced by the passive hyperthermia. They showed that cooling preserved memory capacity but not visual memory.

In Tropical Climate

Arngrimsson et al. (2004) showed that wearing a cooling vest during warm-up in TropC reduces thermal discomfort at the start of a 5-km run race compared to control condition, but this decrease fades after 3.2-km of running. That said, according to the authors, reducing perceived thermal discomfort and cardiovascular strain at the beginning of the race seems to play a role in improving performance in TropC (i.e., 32°C and 50% rH). Moreover, Cleary et al. (2014) evaluated the effectiveness of intermittent superficial cooling (i.e., 5-min of wearing a cooling vest) in 10 college students over 60-min of intense American football training in TropC. Thermal sensation was significantly decreased by this technique but not thirst, RPE, nor heat illness symptoms.

Regarding the impact of cooling strategies on cognitive abilities, Lee et al. (2014) evaluated the efficacy of neck cooling on cognitive task (i.e., symbol digit matching, search and memory, digit span, choice reaction time and psychomotor vigilance test), following exercise-induced hyperthermia. They showed that the neck-cooling collar seemed to enhance performance only in the high-complexity tasks (i.e., search and memory test). Ando et al. (2015) also examined the influence of neck-cooling interventions (with a wet towel and fanning) on cognitive functions during an exercise in TropC (35°C and 70% rH). They particularly focused on working memory and executive function in eight participants at rest and after cycling 10-min in both conditions (i.e., TropC vs. TropC+cooling intervention) but found that the cooling intervention in TropC did not effectively attenuate the impairment of either. Bandelow et al. (2010) used another cooling method to examine the cognitive effects of exercising (i.e., complex visuo-motor, fine motor speed, visual/auditory and visuo-spatial working memories) in TropC in a series of three matches between two soccer teams. The environmental conditions were about 34°C and 63% rH. The players sat under a canopy in 25°C temperature for 15-min before the match and for 10-min during the half-time break. Of the four cognitive functions assessed, only complex visuo-motor speed was improved.

Thus, some cooling interventions may not be efficient enough (e.g., Shibasaki et al., 2017) or may even be counterproductive. Indeed, internal cooling by ingesting large volumes of cold water or ice during exercise might cause gastrointestinal discomfort, allergy or cold-stimulus headache in some individuals (Stevens et al., 2013). Given the disadvantages of certain conventional interventions and the complexity of implementing them during sports competitions, other less invasive and easier to implement techniques should be considered to avoid the inconvenience of overly restrictive or embarrassing cooling interventions and yet maintain the psychological benefits.

IMPACTS OF ALTERNATIVE STRATEGIES TO DEAL WITH THERMAL STRESS

In addition to the conventional interventions to deal with heat stress, other techniques can counter heat and TropC. Thus, these techniques may be used as alternatives or complements.

Menthol Ingestion Techniques to Create a Cooling Sensation

A series of studies has dealt with subjective body cooling through menthol. Although menthol does not lower the core or skin temperature of athletes (Barwood et al., 2015), it stimulates the cold receptors (Cheung, 2010) and induces a cool feeling (Mündel and Jones, 2010), which modifies thermal perceptions. Menthol-induced cold perception has been shown to increase exercise intensity and performance in hot climate (see Cheung, 2010; Mündel and Jones, 2010) and in TropC (e.g., Riera et al., 2014). For example, Stevens et al. (2016) examined the impacts of ice-slurry ingestion and menthol mouth rinse (25 mL of an L-menthol solution at 22°C at a concentration of 0.01% during 5 s) prior to an endurance running performance in the heat. Although ice-slurry reduced the core temperature, it neither decreased the thermal sensation during exercise nor improved the performance on a 5-km run. In contrast, the menthol mouth rinse improved the thermal sensation during exercise and the running performance. However, care is needed when using this technique. Indeed, a false thermal afferent signal, as when the actual temperature of the skin differs from the cold perception, can increase the risk of developing heat-related illnesses (see Valente et al., 2015), the brain being fooled by the menthol-induced cold signal. Moreover, a high menthol concentration could alter thermal perception to an extent that a hot deep body temperature could be ignored. This could lead to even more serious health risks such as heat stroke or hyperthermia, and subsequently to generated cerebral blood flow reduction, brain temperature increase or/and cerebral oxygenation compromising (Bain et al., 2015).

Although these results suggest that the modified perceptive signal is stronger than the physiological signal, it should be possible to find a technique that retains this perceptual advantage without the risk of a side effect on health. Other techniques, such as mental techniques, might avoid the downside of a menthol intervention while maintaining its psychological benefits for motivation and performance. These can be used to manage thermal stress and create a cold feeling that could help athletes to psychologically deal with the thermal environment.

Mental Techniques to Manage Thermal Stress

The techniques of mental preparation are useful to manage stress during competition, but it might be worthwhile to examine the classic mental preparation techniques to determine which ones would be most appropriate for coping with climate stressors. Many variables have been explored, including athletes' resilience and adaptation (Fletcher and Sarkar, 2012). The question remaining: Which strategy is best for coping

with climatic factors? In the field of sport and exercise, few studies have investigated psychological techniques in relation to ambient temperature. Barwood et al. (2008) tested whether a training package of four psychological skills (i.e., goal setting, arousal regulation, mental imagery, positive self-talk) would increase the distance covered during three maximal-effort runs of 90-min in the heat (30°C and 40% rH). They showed that the package lowered the temptation to reduce exercise intensity during the maximal-effort runs while increasing the distance covered by 8% in the last 90-min run. However, it was not possible to determine which of the skills had the beneficial influence. Wallace et al. (2016), therefore, used only one psychological skill (i.e., motivational self-talk) and showed that a 2-week intervention significantly improved endurance capacity and executive function in the heat. Similarly, it might also be interesting to examine how conventional mental interventions could be specifically adapted to coping with TropC. From this viewpoint and even though it has not been shown thus far, mindfulness might be an appropriate strategy. This technique has three components (*awareness* of current thoughts, emotions and bodily sensations; *acceptance*, which is a non-judgmental attitude toward one's current thoughts, emotions, and bodily sensations; and *commitment* to goal-relevant attention focus and behavior) (Gardner and Moore, 2007; Thienot et al., 2014). Although the study did not take place in a particular climate condition, Haase et al. (2015) showed that a 7-week mindfulness intervention (2 full days+6 sessions of 90-min/week) changed the way high-level athletes treated interoceptive information and increased their ability to regulate the anxiety related to unpleasant feelings. This technique seems a more promising way to mentally prepare athletes for the Olympic and Paralympic Games in TropC.

Mental Techniques to Create a Cold Feeling

Few studies in the health field have examined the effect of mental techniques such as hypnosis on thermal factors. Langlade et al. (2002) investigated the influence of hypnotic suggestion and showed that heat detection and heat-pain thresholds were increased. Younus et al. (2003) studied the effect of hypnosis (4 × 1-h/week) on hot flashes in ten healthy volunteers and four breast cancer patients and showed that the frequency, duration, and severity of the hot flashes were significantly reduced. Elkins et al. (2013) obtained equivalent results. However, another question is whether this technique is also effective for coping with the discomfort of a hot and wet environment. For this reason, it would be interesting to examine whether an intervention in TropC consisting of hypnotic cold suggestion would have specific effects on psychological markers (i.e., thermal comfort, thermal sensation, affect) and the motivation to perform exercise compared with a control situation (in TropC with a neutral intervention). Finally, the scientific literature has not yet studied the effects of TropC and cooling strategies on other psychological factors like motivation, self-confidence or flow states. It would therefore

TABLE 1 | Practical implications for dealing with thermal stress.

1	Consider acclimatizing athletes by conducting training sessions for the Olympic preparation period in a tropical environment (training base camp in Japan or a region with a TropC, like the French West Indies). By becoming psychologically accustomed to the difficult conditions, the athletes will experience less stress (heat stress) and disturbance once in Japan. This will also allow for experimentation and selection of pre-cooling methods for all types of outdoor exercise (not only aerobic exercise). To facilitate acclimatization and not just jet lag and chronobiology issues, plan to arrive in Japan as soon as possible before the start of competitions. Given the very special conditions of the Tokyo City (i.e., UHI), consider setting up and training at the nearest competition venues.
2	Anticipate the physical preparation before acclimatization in Tokyo. Schedule physical training in the warmest hours of the country of origin, in warm rooms and with windproof clothing. Once you are in TropC in Japan, keep the same physical goals before setting higher goals. Then, increase the goals more gradually than if you were in a temperate climate. You can begin to get used to gradually by scheduling a physical training at the coolest hours of the day (early in the morning and evening), then after 1 week scheduling training hours on the same time slots of the competitions. Recommendations for nutrition, hydration and sleep, important in a temperate climate, are even more numerous in TropC (e.g., drink fresh water regularly before, during and after exercise, nap and sleep in cool rooms to facilitate better recovery).
3	Consider all conventional cooling techniques that do not seem to have an adverse effect on psychological factors. Consider a set of pre-, per-, post-, and combined cooling interventions in agreement with each athlete. For example, drinking cold water at the preferential temperature, cold wet towels on the head, and a cold jacket before and after warm-up and during timeout of the competition. Check the rules of the sport to guard against any appeal from the organizers or opponents concerning the use of a cooling strategy (e.g., cold packs, collar, cooling vest) during competition.
4	With each athlete, try to find the most effective and enjoyable technique or combination of techniques. Although this has not been demonstrated, it is possible that there is a strong inter-individual difference in the acceptability and effectiveness of the same technique across athletes. Some techniques may, for example, be psychologically beneficial for some athletes, neutral for others, and disturbing for others.
5	While it is important to support and encourage the athlete in their efforts, it is even more important to do so in TropC where physical goals are more difficult to achieve. With each athlete, select from among all the mental preparation techniques those that seem most adapted to cope with TropC (e.g., the mindfulness program). Include the techniques selected for the Olympic or Paralympic Games within the athletes' performance routines so that they can obtain the benefits (i.e., better thermal comfort, reduced perception of exercise difficulty, increased motivation) without suffering undesirable effects (i.e., concentration decrement, distraction, discomfort).
6	Use performance routines (including selected cooling and mental interventions) at each training session prior to the Games to allow athletes to obtain and maintain a flow state for longer times during competition.

be interesting to examine the effects on these psychological factors that are directly involved in performance. Likewise, no studies have compared high performance athletes with

mid-level athletes in terms of their ability to cope with heat stress. Thus, it would be interesting to examine whether psychological components such as motivation, resilience or achievement goals allow high performance athletes to feel perceptions (e.g., thermal comfort, RPE) more bearable than mid-level athletes.

CONCLUSION

Although the conventional strategies are of interest for managing physiological responses during long-duration exercise (see Bongers et al., 2017), they have more relative effects on psychological factors, particularly in TropC. In addition to the disadvantages of implementing pre-cooling (cold-water immersion, cooling garments, cold packs applied to the head) or per-cooling (cold-water, ice-slurry, and/or menthol ingestion), there are also the psychological costs. Indeed, if the physiological benefits (central temperature decrement) are less than the psychological cost (increase in cognitive disturbance), the strategy in real performance situations will prove counterproductive particularly in highly demanding cognitive tasks.

The results concerning the efficacy of these techniques on cognitive abilities depend of the intervention used and the factor investigated. It seems very important to match the expected benefits of a particular technique (conventional or alternative) with the type of performance. Although all psychological factors are important in high-level practice, psychological needs differ across sports. On the one hand, precision sports (e.g., 50-m pistol, archery), team sports (e.g., football, basketball) and duel sports (e.g., tennis, fencing) will primarily require cognitive resources (e.g., attention, decision-making) through subjective intervention (e.g., cold suggestion), and the use of objective strategies (e.g., a cooling vest during timeouts) could be beneficial if they do not disturb the athlete. On the other hand, aerobic sports (e.g., the marathon, 50-km race walk) will mainly require motivational resources through training adaptation in TropC, the use of objective strategies (e.g., cooling equipment before the race), and the use of mental techniques (e.g., mindfulness intervention) should be beneficial.

Last, a parameter that deserves more study is inter-individual variability. Some high-level athletes may be much more supportive of certain strategies, whereas, others prefer one or a combination of different strategies. Not all athletes are inclined to drink crushed ice or take an ice bath before a race. Cooling techniques are not necessarily adapted to all sports modalities and all athletes. We need to focus on the strategy (not the method), which will depend on the severity of the environment and the athletes' perceptions. As perceptions differ from one athlete to another, the psychological feelings are essential. This aspect is enhanced by the fact that the majority of the studies presented in this review are with weak samples and with mid-level athletes as there are few studies of high-level athletes related to the climate. For this reason, we encourage delegations to have their

athletes train in TropC a few weeks before the Tokyo Games (i.e., acclimation training during preparation camp) and to arrive on site early enough to have the time to test strategies and find the most effective one in terms of perceptions (e.g., thermal comfort, RPE), cognitive abilities (e.g., reaction time, attention) and feelings and emotions (e.g., self-confidence, motivation or flow states). With this goal in mind, we also encourage delegations to consider easy-to-use alternative strategies to preserve the psychological benefits in TropC without constraining the athletes to cooling interventions that are potentially deleterious for their performance (see **Table 1**).

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AUTHOR CONTRIBUTIONS

All authors contributed to the manuscript redaction, from the plan conception to the review of literature to the corrections.

FUNDING

Some of the results obtained from the laboratory experimentations have been partially funded by PO-Feder 2014–2020, “ACTE-APPLI”, n°215-FED-213.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Wearing a Cooling Vest During Half-Time Improves Intermittent Exercise in the Heat

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OPEN ACCESS

Edited by:

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Kobe University, Japan

Reviewed by:

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 02 November 2018

Accepted: 21 May 2019

Published: 07 June 2019

Citation:

Chaen Y, Onitsuka S and
Hasegawa H (2019) Wearing
a Cooling Vest During Half-Time
Improves Intermittent Exercise
in the Heat. *Front. Physiol.* 10:711.
doi: 10.3389/fphys.2019.00711

Endurance and intermittent exercise performance are impaired by high ambient temperatures. Various countermeasures are considered to prevent the decline in exercise performance in the heat, convenient, and practical cooling strategies attracts attention. The purpose of this study was to investigate the effect of wearing a new type of cooling vest which cooled torso and neck during half-time (HT) on intermittent exercise performance that imitated intermittent athletic games. All measurements on the experiments were carried out with the bicycle ergometer. Eight male soccer players performed a familiarization session and two experimental trials of a 2 × 30 min intermittent cycling exercise protocol, which consisted of a 5 s maximal power pedaling (body weight × 0.075 kp) every minutes separated by 25 s unloaded pedaling (80 rpm) and rest (30 s) in the heat (33.0°C; 50% relative humidity). The two trials included cooling-vest condition (VEST) and control condition (CON), and the difference is with or without wearing cooling vest imposed for 15 min at HT. Mean and peak power output, rectal (Tre) and skin temperature (neck, upper back, chest, right upper arm, and thigh), heart rate (HR), deep thigh temperature, rating of perceived exertion (RPE), and thermal comfort (TC) and thermal sensation (TS) were measured. Mean power output at 2nd half was significantly greater ($p < 0.05$) in VEST (3rd trial: 589 ± 58 W, 4th trial: 584 ± 58 W) than in CON (3rd trial: 561 ± 53 W, 4th trial: 561 ± 53 W). HR were significantly lower in VEST during HT and higher in VEST at the last maximal pedaling ($p < 0.05$). At the end of HT, neck skin temperature and mean skin temperature were significantly lower in VEST ($32.04 \pm 1.47^\circ\text{C}$, $33.76 \pm 1.08^\circ\text{C}$, respectively) than in CON ($36.69 \pm 0.78^\circ\text{C}$, $36.14 \pm 0.67^\circ\text{C}$, respectively) ($p < 0.05$). During 2nd half, TS, TC, and RPE were significantly lower in VEST than in CON ($p < 0.05$). There was no significant difference in Tre and deep thigh temperature throughout each conditions. These results indicate that wearing a new type of cooling vest during HT significantly improves intermittent exercise performance in the heat with decreased neck and mean skin temperature and improved subjective responses.

Keywords: intermittent exercise, cooling vest, skin temperature, heat, thermal comfort

INTRODUCTION

In many team sports, athletes frequently perform in the heat, and are required to sustain exercise performance. Compared to the temperate environment, the core and skin temperatures increase in the heat, resulting in increased cardiovascular and metabolic strain and thermal perceptual load, as well as decreased exercise performance. Given that Tokyo 2020 Olympic and Paralympic games

will be held in extremely hot and humid conditions ($=33^{\circ}\text{C}$, 70–80% relative humidity), athletes should begin to prepare for this. Therefore, various countermeasures are considered to prevent the decline in exercise performance in the heat, requiring convenient and practical cooling strategies. Although it is well established that cooling interventions, such as water immersion or cooling with a large fan, are effective in normalizing the body temperature, these methods are not practical in the actual sports field (Quod et al., 2006). Additionally, the use of cooling vests is popular in many field team-sports due to their practicality and ease of use. Thus, wearing a cooling vest, which can be used conveniently, is one of the methods to prevent the decline in exercise performance in the heat (Randall et al., 2015).

However, details of the mechanism underlying the decline in exercise performance in the heat have not been clarified yet, but it is known that the central nervous system is involved (Nybo and Nielsen, 2001). Signals sensed from the peripheral thermoreceptors, such as in the skin, are sent to the hypothalamus, which is the center of temperature regulation (Tucker and Noakes, 2009). It has been suggested that the hypothalamus that senses high skin temperature or high thermal perception selectively regulates exercise intensity to complete an exercise task within a range not exceeding the critical limiting temperature. Therefore, wearing a cooling vest has a beneficial effect in reducing skin temperature and thermal perception and in relieving thermal strain sent from the peripheral thermoreceptors to the hypothalamus; thus, the selective decrease in exercise intensity is suppressed and endurance exercise performance is improved (Faulkner et al., 2015; Schmit et al., 2017). Previous studies that investigated improvement in exercise performance by wearing a cooling vest adopted an endurance exercise protocol such as self-paced trials (Arngrimsson et al., 2004; Faulkner et al., 2015; Schmit et al., 2017). However, intermittent sporting activities, such as soccer, rugby, and basketball, are also impaired when the ambient temperature is elevated (Duffield and Marino, 2007; Castle et al., 2006). As with endurance exercise, intermittent performance deteriorates in the heat (Drust et al., 2005). Furthermore, it has been suggested that intermittent exercise in the heat increases the thermal and metabolic strain further compared to endurance exercise (Ekblom et al., 1971; Mora-Rodriguez et al., 2008). Success in intermittent sports is greatly linked to the ability to perform repeated bouts of high intensity sprint exercises (Tyler et al., 2015). In a previous study, the potential cause of the decline in sprint performance observed in team sports activities in the heat was considered to be a decline in the function of the central nervous system caused by an increase in core, cerebral, and skin temperatures (Girard et al., 2015). Therefore, the physiological mechanism of declining intermittent exercise performance in the heat is similar to that of declining endurance exercise performance, and the selective decrease in exercise intensity by the motor cortex via thermal information from hypothalamus may also be caused by intermittent exercise performance. Therefore, wearing a cooling vest can suppress the selective decrease in exercise intensity and improve intermittent exercise in the heat.

Intermittent athletic games, such as soccer and rugby, have short rest periods between the exercise bouts and some cooling interventions are possible (Arngrimsson et al., 2004); countermeasures of heat are required for practical and convenient cooling interventions. In previous studies of pre-cooling methods, lower body cooling using ice packs improved subsequent sprint performance (Castle et al., 2006). However, it is also suggested that sufficient re-warming is required after cooling, since high-intensity exercise performance declined with decreasing the temperature of working muscles (Sleivert et al., 2001). It is difficult to do sufficiently re-warming in a short time such as during a half-time (HT); thus, it may be inappropriate to cool the working muscles directly during HT. Therefore, it is considered that wearing a cooling vest, which decreases skin temperatures without decreasing working muscle temperatures, can be effective because it can continue cooling even during HT. The purpose of this study was to investigate the effect of wearing a cooling vest, which can cool the torso and neck regions during HT, on intermittent exercise performance, imitating intermittent athletic games. We hypothesized that wearing a cooling vest which cooled the torso and neck regions decreases skin temperature and improves subjective sensation, and subsequent intermittent exercise performance is improved in the heat.

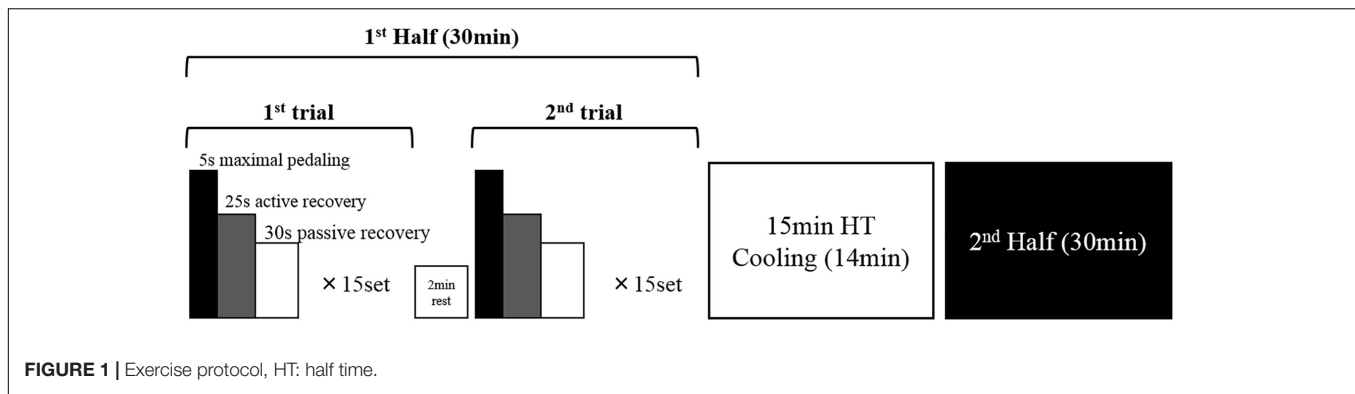
MATERIALS AND METHODS

Participants

Eight non-heat-acclimated male soccer players (age: 21 ± 1.6 years, height: 174 ± 5 cm, mass: 64 ± 4 kg) volunteered for this study. They abstained from alcohol and caffeine consumption 24 h before the experiment, and they ingested same food and drink (Calorie Mate and Energen; Otsuka Pharmaceutical Co., Ltd., Japan) 2 h before the testing. All trials took place during the winter season with mean temperatures ranging from 10°C to 19°C to avoid the influence of natural heat acclimatization. The study procedures were approved by the Ethics in Human Research Committee of Hiroshima University, and all participants signed an informed consent form before the start of the study.

Experimental Design

Participants completed one familiarization session before completing two experimental sessions in a randomized cross-over design. In one experimental session, participants wore the cooling vest (VEST) at the HT, and for the other session, they wore the cooling vest without ice packs (CON) at the HT. In the familiarization trial, they performed with the same measurement and protocol as the CON. Each session was separated by at least 4 days and completed at the same time of day to control for the effect of circadian rhythm on body temperature. All sessions took place in a climate environmental chamber in hot ambient conditions (33.0°C , 50% relative humidity). During all sessions, participants were allowed to consume water (33°C) up to 750 ml during the 2-min rest and HT, and all participants drank all the water.



Exercise Protocol

Given that the measuring equipment was attached in all participants in the heat, they stayed in the heat for 30 min before the start of the exercise. All sessions were completed with the use of a cycling ergometer (POWERMAX-V2; COMBI, Japan). Participants completed warm-up exercises consisting of 7 min of cycling (body mass \times 0.01 kp) and 3 sets of maximal pedaling (body mass \times 0.075 kp) at 2 min before the exercise protocol. Participants completed a laboratory-based intermittent exercise protocol designed to replicate the demands of actual intermittent athletic games. The protocol consisted of two 30-min halves separated by a 15-min HT, with the half consisting of two trials separated by a 2-min rest period. One set consisted of a 5-s maximal pedaling (body mass \times 0.075 kp), 25-s active recovery (no load, 80 rpm), and 30-s passive recovery. One trial consisted of 15 sets. Participants performed a total of four trials (Figure 1).

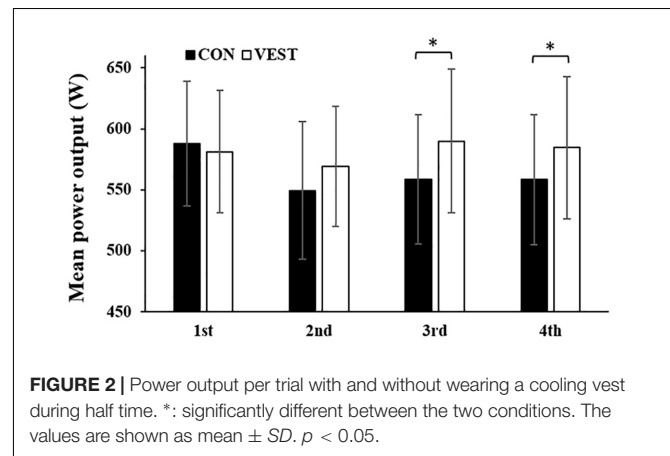
Cooling Intervention

The cooling vest (Mizuno Co., Ltd., Japan), which can cool the neck, upper body, back, and side, was used during HT. Immediately after the 2nd trial ended, participants took off their underwear and training shirt, wiped their sweat, changed into a new training shirt, and wore the cooling vest. Given 1 min was required to change clothes, the actual cooling time was 14 min. In the VEST, ice packs (approximately -1°C) of gel material frozen in the freezer beforehand were put inside the pockets of the cooling vest. Since it was kept in the freezer until immediately before cooling, the cooling effect remained for the duration of 14 min. On the other hand, in the CON, cooling vests with similar weight as the used in the VEST (1.9 kg) were used.

MEASUREMENTS

Performance Index

We measured the mean and maximal power outputs as the intermittent exercise performance. The mean power output was an average of 5-s maximal pedaling, and the maximal power output was calculated by the following equation:



Maximal power output =

body mass \times 0.075 kp \times maximum rotation speed.

Physiological Index

In this study, rectal (T_{re}), skin, and deep thigh temperatures, heart rate (HR), blood lactate, dehydration rate, and urine specific gravity were measured. Participants self-inserted a rectal probe approximately 10 cm past the anal sphincter. Skin temperatures were measured by attaching a thermistor probe on the neck, upper back, chest, upper arm, and thigh with active flex (BAND-AID; Johnson & Johnson, Japan) and insulation material seal (temperature insulation pad; Nihon Kohden Corporation, Japan) to accurately measure skin temperature. The mean skin temperature (T_{sk}) was calculated using the formula developed by Roberts et al. (1977):

$$T_{sk} = (0.43 \times T_{chest}) + (0.25 \times T_{upper arm}) + (0.32 \times T_{thigh}).$$

Participants also wore a HR monitor (model RS400; Polar Electro Oy, Kempe, Finland) that was attached before entering the environmental chamber. Deep thigh temperature measured by the deep body temperature monitor (CM-210, Terumo Co., Ltd., Japan) which detects the tissue temperature 5–10 mm below the skin surface using the zero heat flow method

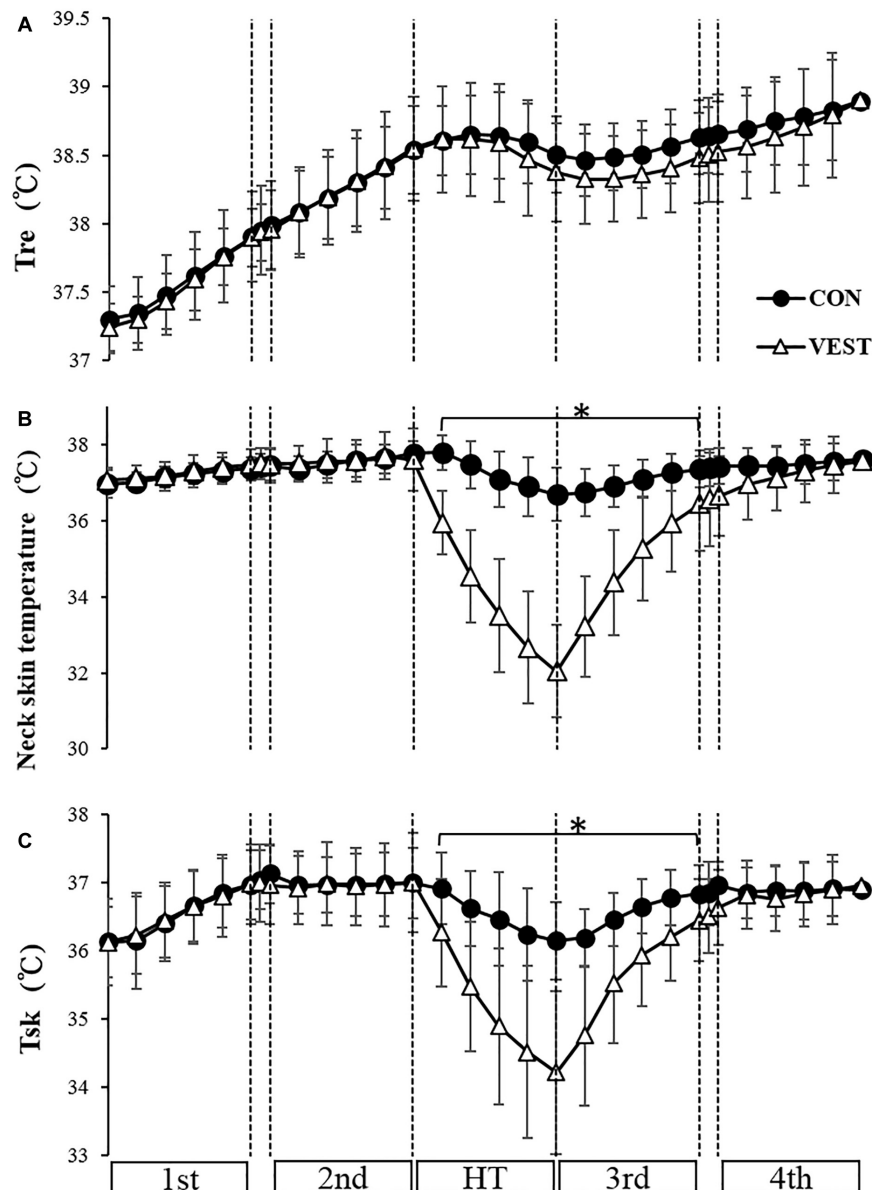


FIGURE 3 | Change in rectal temperature (A), neck skin temperature (B) and mean skin temperature (C) during exercise protocol. *: significantly different between the two conditions. The values are shown as mean \pm SD. $p < 0.05$.

(Yamakage and Namiki, 2003). This monitor measures skin surface temperature beneath a thermal insulating pad containing a heater, which equilibrates the skin temperature with the deep tissue temperature when heat flow from the skin is maintained at zero. The consistency between muscle temperature measured using a needle thermocouple and the zero heat flow method was evaluated previously (Togawa et al., 1976; Wakabayashi et al., 2018). Tre, skin, and deep thigh temperature, HR were measured every 3 min during the experiment. Blood lactate was taken from the fingertip before starting the exercise and at the end of the trials and analyzed using an automated blood lactate analyzer (Lactate Pro; Arkrey, Japan). The body weight was measured using a scale

(UC-300, A&D Co., Ltd., Japan) with the participants naked. The urine specific gravity was measured using a digital urine specific gravity refractometer (UG-D; Atago Co., Ltd., Japan) before and after the experiment. Changes in nude body mass were used to estimate gross sweat loss adjusted for fluid intake.

Perceptual Index

Upper body thermal sensation and thermal comfort (TSupper, TCupper), neck thermal sensation and thermal comfort (TSneck, TCneck), and whole body thermal sensation and thermal comfort (TS, TC) were taken every 3 min during the experiment. TS and TC were rated using a 13-point scale that ranged from -6

(very cold) to 6 (very hot) and -6 (very uncomfortable) to 6 (very comfortable) (Olesen and Brager, 2004). Rating of perceived exertion (RPE) (Borg, 1973) was taken every 3 sprints in all trials.

Statistical Analyses

All statistical calculations were performed using SPSS version 25.0. Mean power output, maximal power output, T_{re} , HR, T_{sk} , all skin temperatures, deep thigh temperature, blood lactate, all TS and TC, and RPE were analyzed using two-way repeated measures ANOVA (conditions \times time). Dehydration rate and urine specific gravity were analyzed using one-way ANOVA (conditions). Where significant effects were identified, *post hoc* pairwise comparisons with Bonferroni correction were conducted. Where an interaction effect was observed, a paired samples *t*-test was conducted with Bonferroni correction applied. All data were checked for normal distribution by Kolmogorov–Smirnov test, and the violation of sphericity prior to analysis and Greenhouse-Geisser epsilon correction were used to adjust the degrees of freedom. The accepted level of significance for all analyses was $p < 0.05$. All data were presented as mean \pm SD.

RESULTS

Performance

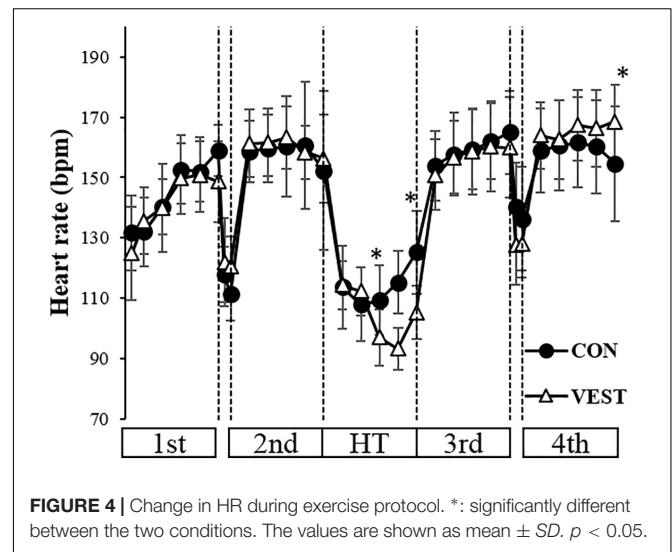
Mean power output in the 2nd half was significantly greater ($p < 0.05$; **Figure 2**) in VEST (3rd trial: 589 ± 58 W, 4th trial: 584 ± 58 W) than in CON (3rd trial: 561 ± 53 W, 4th trial: 561 ± 53 W). Seven out of the eight participants had improved mean power output in the 3rd trial in VEST than in CON, and all eight participants had improved mean power output in the 4th trial in VEST than in CON. Maximal power output was not significantly different between the two conditions.

Body Temperature

Baseline values of all body temperatures were taken at the start of the 1st trial. T_{re} at the end of HT was 0.13°C lower in VEST ($38.3 \pm 0.4^\circ\text{C}$) than in CON ($38.5 \pm 0.3^\circ\text{C}$); however, there were no significant differences between the two conditions (**Figure 3A**). Neck skin temperature and T_{sk} from the start of HT to the end of the 3rd trial was significantly lower in VEST than in CON ($p < 0.05$; **Figures 3B,C**). Owing to the failure of the measuring equipment, upper back skin temperature was analyzed in 6 participants. Chest and upper back skin temperature from the start of HT to the end of the 3rd trial was significantly lower in VEST than in CON ($p < 0.05$, end of HT, CON: $36.45 \pm 1.1^\circ\text{C}$, VEST: $31.13 \pm 1.93^\circ\text{C}$, CON: $36.8 \pm 0.72^\circ\text{C}$, VEST: $32.36 \pm 1.69^\circ\text{C}$). There were no significant differences between the two conditions in upper arm and thigh skin temperatures. There were no significant differences between the two conditions in deep thigh temperature (at the end of HT, CON: $37.2 \pm 0.5^\circ\text{C}$, VEST: $37.5 \pm 0.3^\circ\text{C}$).

Heart Rate and Blood Lactate

Heart Rate was significantly lower in VEST during HT and higher in VEST at the last maximal pedaling than in CON



($p < 0.05$; **Figure 4**). Blood lactate at the end of warming up was 3.0 ± 1.2 mmol/L in CON and 3.3 ± 1.2 mmol/L in VEST, whereas at the end of the 4th trial, it was 5.3 ± 1.3 mmol/L in CON and 5.3 ± 1.8 mmol/L in VEST. Blood lactate did not differ between the two conditions.

Body Fluid Balance

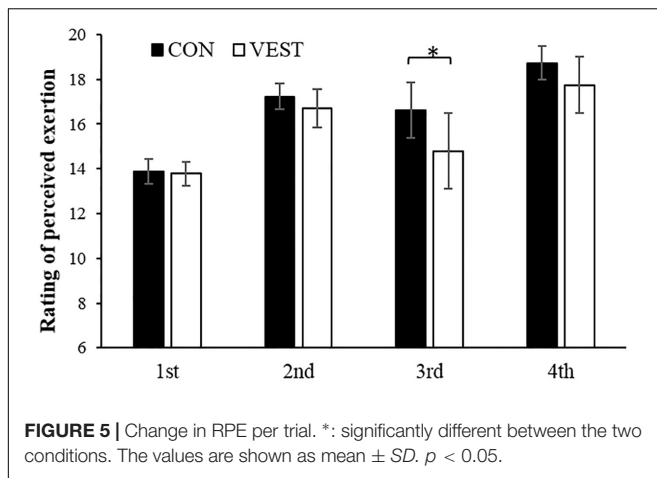
The rate of sweat loss was $1.8 \pm 0.9\%$ in CON and $1.5 \pm 0.7\%$ in VEST, and there were no significant differences between the two conditions. The urine specific gravity was 1.020 ± 0.006 in CON and 1.020 ± 0.007 in VEST before the exercise, whereas it was 1.023 ± 0.002 in CON and 1.023 ± 0.002 in VEST after the exercise. There were no significant differences in the urine specific gravity between the two conditions.

Perceptual Index

All perceptual data (TS, TC, and RPE) changed over the time during the intermittent exercise. TS was significantly lower in the 2nd half in VEST (**Table 1**). All TCs were significantly higher in the 2nd half in VEST (**Table 1**). Changes in RPE were not significantly different between the two conditions; however, the mean RPE per trial were not significantly lower in VEST ($p < 0.05$; **Figure 5**).

DISCUSSION

In this study, we investigated the effect of wearing a cooling vest, which cooled the torso and neck regions during HT, on intermittent exercise performance that imitated intermittent athletic games in the heat. We hypothesized that wearing a cooling vest decreases skin temperature and improves subjective sensation, and compared with no-cooling condition, subsequent intermittent exercise performance is improved in the heat. Our study suggests that wearing the cooling vest that covered the torso and neck during HT significantly improves intermittent exercise



performance in the heat with decreased neck and mean skin temperature as well as improved subjective responses.

As reported in previous studies, wearing a cooling vest has a beneficial effect on skin temperatures, resulting in improved exercise performance. Therefore, in this study, it is considered that decreasing skin temperature by wearing the cooling vest improved performance. The mean skin temperature decreased by approximately 2.8°C through cooling, and it was significantly lower than of the CON until the end of the 3rd trial. Although ambient temperature or cooling time is different, the studies of Faulkner et al. (2015) and Schmit et al. (2017) indicated that the mean skin temperature decreased by approximately 3°C and 1.8°C, respectively, by wearing the cooling vest, and this reduction was maintained for approximately 15 min. Thus, this study showed that the mean skin temperature decreased as much as those in previous studies, which showed that wearing the cooling vest improves endurance exercise performance in the heat. Although the peripheral skin blood flow increases due to heat dissipation and the amount of blood supply to the skeletal muscle decreases (Bell et al., 1983; Gonzalez-Alonso et al., 1999), the reduction in skin temperature decreases

the peripheral skin blood flow, and it potentially reduces cardiovascular strain and increases blood supply to the skeletal muscle (Sleivert et al., 2001; Gonzalez-Alonso and Calbet, 2003). Thus, in this study, wearing the cooling vest decreased skin temperature, which increases blood supply to the skeletal muscle; hence, participants were able to demonstrate higher power output even in the heat.

The decrease in neck skin temperature also positively influenced intermittent exercise performance. The neck skin temperature decreased by approximately 5°C by wearing the cooling vest, and it was significantly lower than that of the CON from HT to the end of the 3rd trial. The cooling vest used in this study was a new type of cooling vest, which can cool the neck, unlike the conventional one. Tyler et al. (2010) reported the improvement of endurance performance by cooling the neck during exercise without decreases in skin temperature, core temperature, and HR. In addition, it was reported that continuous neck cooling also has a beneficial effect on intermittent exercise performance (Sunderland et al., 2015). These improvements of performance are due to the fact that the neck is adjacent to the temperature-regulating centers and high sensory thermoreceptor (Shvartz, 1976; Gordon et al., 1990; Cotter and Taylor, 2005). Furthermore, it is possible that neck cooling decreased the cerebral temperature and affected mitigation of central fatigue. In the heat, it has been reported that central fatigue is caused by excessive increases in body temperature (Nybo and Nielsen, 2001), resulting in the decline of exercise performance (Hasegawa and Cheung, 2013). Ansley et al. (2008) improved the TS and submaximal exercise performance by cooling the face, and reported that cooling of the blood flow into the head decreases the cerebral temperature, which was considered as one of the factors that improve exercise performance. Furthermore, it is suggested that the neck cooling affects the arterial blood and a subsequent reduction in cerebral temperature (Caputa et al., 1986; Tyler and Sunderland, 2011). From the abovementioned studies, it is speculated that reduction of cerebral temperature and mitigation of central fatigue improve exercise performance, however, the current study did not measure cerebral temperature

TABLE 1 | Thermal sensation (TS) and thermal comfort (TC) during exercise protocol.

		Baseline	Post 1st	Post 2nd	Post HT	Post 3rd	Post 4th
TS (whole)	CON	2.3 ± 0.9	3.8 ± 0.6	5.8 ± 0.3	2.8 ± 1.0*	5.1 ± 0.9*	6 ± 0.0*
	VEST	2.3 ± 1.1	3.6 ± 0.7	5.3 ± 0.5	-2.2 ± 1.4	3.8 ± 1.1	5.5 ± 0.5
TS (neck)	CON	2.5 ± 1.0	3.8 ± 0.6	5.8 ± 0.3	2.8 ± 1.0*	5.1 ± 0.9*	6 ± 0.0*
	VEST	2.3 ± 1.1	3.6 ± 0.7	5.3 ± 0.5	-3.6 ± 0.1	3.8 ± 1.1	5.5 ± 0.5
TS (upper)	CON	2.3 ± 0.9	3.8 ± 0.6	5.8 ± 0.3	2.8 ± 1.0*	5.1 ± 0.9*	6 ± 0.0*
	VEST	2.3 ± 0.9	3.6 ± 0.7	5.3 ± 0.5	-2.8 ± 0.8	3.8 ± 1.1	5.5 ± 0.5
TC (whole)	CON	-2.1 ± 0.6	-3.5 ± 0.5	-5.3 ± 0.9	-3.1 ± 1.2*	-4.8 ± 0.9*	-6 ± 0.0*
	VEST	-2 ± 1.1	-3.5 ± 0.5	-5.1 ± 0.8	2.6 ± 1.8	-3.7 ± 1.1	-5.5 ± 0.5
TC (neck)	CON	-2.1 ± 0.6	-3.5 ± 0.5	-5.3 ± 0.9	-3 ± 0.8*	-4.8 ± 0.9*	-6 ± 0.0*
	VEST	-2 ± 1.1	-3.5 ± 0.5	-5.1 ± 0.8	2.6 ± 1.8	-3.7 ± 1.1	-5.5 ± 0.5
TC (upper)	CON	-2.1 ± 0.6	-3.5 ± 0.5	-5.3 ± 0.9	-3.1 ± 0.9*	-4.8 ± 0.9*	-6 ± 0.0*
	VEST	-1.9 ± 0.9	-3.5 ± 0.5	-5.1 ± 0.8	2.6 ± 1.8	-3.7 ± 1.1	-5.5 ± 0.5

*: significantly different between the two conditions. The values are shown as mean ± SD. $p < 0.05$.

and cerebral blood flow. Thus, it is necessary to investigate these measurements in the future.

In this study, TS and TC were classified into parts and measured; all parts of TS and TC were significantly lower in VEST ($p < 0.05$). Participants felt the coolest at the end of HT in all parts of TS (respectively, whole: -2.2 ± 1.4 , neck: -3.6 ± 1.0 , upper: -2.8 ± 0.8), and participants felt that the neck was the coolest. Given that increases in skin temperature during exercise worsen thermal perception and TC (Flouris and Schlader, 2015), in the present study, it is considered that these improvements in thermal perception are due to the decrease in skin temperature (Schlader et al., 2011). The finding that the participants felt that the neck was the coolest, supports the assertion of a previous study reporting that thermoreceptive senses possibly differ depending on the body parts and region of the neck (Cotter and Taylor, 2005; Nakamura et al., 2013; Filingeri et al., 2014). However, in a previous study on neck cooling, TS of the neck significantly improved by neck cooling, but TS of the whole body was not significantly improved (Tyler and Sunderland, 2011). Thus, it was suggested that not only neck cooling but also upper body cooling, such as the chest or upper back, may be necessary for cooling the whole body and a cooling vest is useful for this. In addition, in the 3rd trial, the mean value of RPE was significantly lower in VEST. The difference in RPE is considered to be due to the improvement of TS and TC, and mean power outputs in the 3rd trial showed a high value despite the low RPE, indicating the usefulness of the cooling vest which covered the torso and neck. It has been suggested that sending thermal information to the hypothalamus indirectly affects reduction of power output (Tucker and Noakes, 2009). Therefore, mixed cooling of the torso and neck using the new type of cooling vest beneficially affects the subjective sensation, and the selective lower exercise intensity by the motor cortex was suppressed because of alleviation of thermal strain, which is sent from the peripheral thermoreceptors to the hypothalamus, thereby resulting in high performance as demonstrated by the participants.

In this study, intermittent exercise performance in the heat was improved without changes in T_{re} by improving the skin temperature, TS, and TC with the use of the cooling vest that cooled the neck, compared to that in CON. Tyler et al. (2010) reported that if an athlete is under sufficient thermal strain, time-trial performance in the heat can only be significantly enhanced by reducing neck temperature and TS, without significantly altering the physiological or peripheral neuroendocrinological responses to the exercise bout. However, improving performance due to reduction of skin temperature and TS without changes in T_{re} may be accompanied by a risk of heat illness. The neck region is an area of high alliesthesia thermosensitivity and also is an area that can be cooled effectively (Tyler and Sunderland, 2011). Cooling the surface of the neck allowed the athletes to tolerate higher core temperature and HR (Tyler and Sunderland, 2011). However, it should be noted that the perceived level of thermal strain is dampened by cooling intervention, and the possibility of excessive core temperature increases due to high performance compared to no cooling. On the other hand, in this study, it was noteworthy that even though participants demonstrated high power output in VEST, T_{re} at the end of exercise was similar

between VEST and CON. From these results, it was suggested that improving intermittent exercise performance by wearing the cooling vest does not cause excessive body temperature increases and does not increase the risk of athlete's heat illness, and this method allowed athletes to play safely at the actual competition site.

Intermittent athletic games, such as soccer and rugby, have short rest periods between the exercise bouts; therefore, various countermeasures, such as convenient and practical cooling strategies, are considered to prevent the decline in exercise performance in the heat. In this study, the exercise protocol imitated intermittent exercise games, and wearing the cooling vest during HT improved intermittent exercise performance. In the cooling time of previous studies on cooling vests, Duffield and Marino (2007) applied the cooling vest for 15 min before exercise and 10 min during HT, whereas Castle et al. (2006) applied the cooling vest for 20 min before exercise. Compared to these previous studies, sprint performance improved in this study despite the short cooling time, because the cooling vest was able to cool the neck. Furthermore, it was important to note that the degree of lowering deep thigh temperature of the working muscle was similar between VEST and CON, suggesting that the cooling vest had no negative effects on subsequent intermittent exercise. The previous study reported that application of ice packs on the thigh for 45 min had a negative effect on subsequent full-pedaling performance (Sleivert et al., 2001). Thus, it is suggested that direct cooling on the working muscle may influence the subsequent high-intensity exercise performance and sufficient re-warming is required. That is, direct cooling on the working muscle in a short time, such as HT, may not be suitable, and this study showed that the cooling vest is the appropriate cooling strategy during short resting periods. Therefore, the application of the new type of cooling vest, which cooled the neck in a relatively short time, improved subsequent intermittent exercise performance without lowering the performance of working muscle. This is considered to be the attractive cooling strategy for athletes involved in intermittent sporting activities.

In previous studies on cooling vests, wearing the cooling vest improved exercise performance without improvement of physiological index, such as core temperature or HR (Faulkner et al., 2015; Schmit et al., 2017). In contrast, in this study, HR was significantly lower in VEST during HT and higher in VEST at the last maximal pedaling. The reason for this improvement is that the cooling vest used in this study was able to cool the neck. By wearing the cooling vest, the peripheral region, such as the skin, is cooled down and the necessity of heat dissipation is alleviated. As a result, it can be predicted that the HR decreased because the blood flow in the center increased and the cardiovascular strain was alleviated. However, in the previous study showing improved intermittent performance by cooling the neck, HR was not significantly decreased only by neck cooling (Tyler and Sunderland, 2011). Therefore, it is considered that mixed cooling of the torso and neck is necessary for the improvement of physiological indexes other than skin temperature; hence, our hypothesis that cooling vests covers torso and neck are useful for intermittent exercise performance.

LIMITATION

The limitation of this study was the reproducibility of intermittent athletic game. First, participants spent HT in the heat, but, in an actual intermittent game, the player spends the rest period in the locker room in which room temperature is cooled by air conditioning. In this study, we wanted to investigate the effects of the cooling vest itself, thus participants spent HT in the heat. However, in the future, it is necessary to set a more practical rest period. Second, there was an issue associated with exercise intensity. Özgünen et al. (2010) investigated the influence of the heat on physiological index and performance during actual soccer games. They reported that the peak core temperature was observed at the end of the 1st half. However, in this study, the peak core temperature was observed later in the 2nd half, which differed from the result of the actual competition. These varying results are considered to be caused by the configuration of the exercise intensity. In this protocol, since the exercise intensity was controlled except for the 5-s maximal pedaling, the exercise task of all active recovery had the same exercise intensity. However, in the actual competition, a decline in performance was also observed in other parameters such as jogging in the 2nd half compared to the 1st half (Özgünen et al., 2010). Therefore, in this study, it is considered that the active recovery had the same intensity in the 1st half and the 2nd half, and it resulted in the same value of T_{re} between the two conditions. In future research, exercise intensity other than maximal pedaling should not be controlled, and evaluation of exercise performance will also be necessary. In addition, we could not measure skin blood flow, cerebral temperature, muscle blood flow, and blood pressure in this study. In future research, to

clarify the mechanism of circulatory dynamics, it is necessary to measure these indexes.

CONCLUSION

By wearing the cooling vest which cools the neck and upper torso regions during HT that imitates intermittent athletic competitions, such as soccer, in the heat, physiological indicators, such as skin temperature and HR, and subjective sensation, such as TS or TC, were significantly improved, and subsequent intermittent exercise performance improved. In addition, as the improvement in performance was observed in a relatively short cooling time, the cooling vest is more practical to use than the cooling interventions requiring large-scale facilities such as a cold water immersion, suggesting that wearing the cooling vest is effective at the actual competition site.

AUTHOR CONTRIBUTIONS

YC designed the study with assistance from HH. All authors completed the data collection, data analysis, and manuscript preparation.

ACKNOWLEDGMENTS

We express our gratitude to all participants in this study. We would like to thank Editage (www.editage.jp) for English language editing.

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Interventions to Minimize Jet Lag After Westward and Eastward Flight

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Keywords: time zone, adaptation, sleep, circadian, light, melatonin, exercise, athlete

INTRODUCTION

Air travel across several time zones, i.e., transmeridian flight, causes negative effects—some of which occur during flight and some of which occur in the days after flight. Anecdotally, these effects are often referred to collectively as jet lag, but they are actually two separate phenomena—travel fatigue and jet lag—each with their own causes and consequences (Waterhouse et al., 2004). Travel fatigue refers to a collection of symptoms that occur during or immediately after long flights. These symptoms include fatigue, disorientation, and headache (Waterhouse et al., 2004)—primarily caused by the sleep loss, dehydration, hypoxia, and discomfort associated with being in an aircraft with confined space, recline-restricted seats, low air pressure, low humidity, etc., for 8–14 h (Brown et al., 2001; Roach et al., 2018). In contrast, jet lag refers to a collection of symptoms that occur in the days after flight across three or more time zones. These symptoms include headache, irritability, daytime sleepiness, difficulty sleeping at night, poor mental and physical performance, and poor gastrointestinal function (Waterhouse et al., 2004)—primarily caused by the mismatch between the circadian system, or internal body clock, which is synchronized to time cues in the departure time zone, and the desired timing of sleep and wake, which are typically synchronized to time cues in the destination time zone.

In August 2020, the Olympic Games will be held in Tokyo, Japan. Athletes will travel from all over the world to compete in the Games, and many will have to travel across several time zones. For example, athletes traveling to Japan from North America and Western Europe will face time zone changes of 8–11 h west and 6–8 h east, respectively. Some athletes will travel to Japan, or nearby countries, weeks before their events, while others will arrive in Japan in the days prior to competition. In either case, athletes will want to adjust to the new time zone as quickly as possible so that they can prepare well and/or compete at the highest level.

The purpose of this manuscript is to discuss the causes and consequences of jet lag and to provide examples of how to use judiciously timed light exposure/avoidance and/or exogenous melatonin ingestion to adapt the circadian system to a new time zone after transmeridian flight. These guides could be applied by athletes competing in the Tokyo 2020 Olympic Games, but they could also be applied by athletes traveling to other countries for training or competition, or by non-athletes traveling for business or pleasure.

JET LAG IS CAUSED BY THE DESYNCHRONY BETWEEN THE CIRCADIAN SYSTEM AND LOCAL TIME CUES

The term “circadian rhythms” refers to rhythms with a period of approximately 24 h (Halberg, 1959). Many physiological and psychological variables in humans have been shown to alter rhythmically with a ~24 h period, including core body temperature (Dijk et al., 1992; Zhou et al., 2011a, 2017); cortisol (Scheer et al., 2009), blood pressure (Scheer et al., 2010), heart rate (Scheer et al., 2010), hunger (Sargent et al., 2016), cognitive performance (Dijk et al., 1992; Darwent et al., 2010; Matthews et al., 2010), strength (Reilly et al., 1997; Sargent et al., 2010), balance

OPEN ACCESS

Edited by:

Till Roenneberg,
Ludwig Maximilian University of
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Reviewed by:

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Specialty section:

This article was submitted to
Chronobiology,
a section of the journal
Frontiers in Physiology

Received: 09 April 2019

Accepted: 09 July 2019

Published: 31 July 2019

Citation:

Roach GD and Sargent C (2019)
Interventions to Minimize Jet Lag After
Westward and Eastward Flight.
Front. Physiol. 10:927.
doi: 10.3389/fphys.2019.00927

(Sargent et al., 2012b), flexibility (Reilly et al., 1997), dexterity (Matthews et al., 2012a,b), subjective alertness (Dijk et al., 1992; Zhou et al., 2012; Kosmadopoulos et al., 2014), subjective fatigue (Ferguson et al., 2012), subjective sleepiness (Kosmadopoulos et al., 2017), and objective sleepiness (Lavie, 1986; Dijk and Czeisler, 1995; Paech et al., 2010, 2012; Sargent et al., 2012a).

In humans, circadian rhythms in physiological and psychological variables are endogenously generated by a central circadian pacemaker—located within the suprachiasmatic nucleus of the hypothalamus—with a period of approximately 24.2 h (Czeisler et al., 1999; Zhou et al., 2011b). These rhythms are entrained to the period of a 24 h day by environmental signals or “zeitgebers,” meaning time-givers (Aschoff et al., 1971). Sunlight is the most powerful zeitgeber for humans (Wever et al., 1983; Czeisler et al., 1986), but non-photic stimuli such as social contact, eating, and physical activity, may also play a role (Mistlberger and Skene, 2004). Most peripheral cell types, including those in the major organ systems—heart, lungs, liver, pancreas—also contain their own circadian oscillators, which are kept in coherent phase relationships by the suprachiasmatic nucleus (Yamada and Forger, 2010; Bass, 2012; Schibler et al., 2015).

The circadian system cannot immediately entrain to the timing of zeitgebers in a new time zone (Wever, 1980), so after long-haul flights to the west or east, the circadian system is initially aligned with the timing of zeitgebers at the point of departure rather than zeitgebers at the new location (Winget et al., 1984). A period of desynchrony follows while the circadian system is entrained to the timing of zeitgebers in the new time zone—and it is this period of desynchrony that gives rise to the symptoms of jet lag.

THE TIMING OF THE HUMAN CIRCADIAN SYSTEM CAN BE RESET BY LIGHT, MELATONIN, AND EXERCISE

The timing of the human sleep/wake and circadian systems are related such that the production of endogenous melatonin begins ~2 h before habitual bedtime (Burgess et al., 2003b; Burgess and Eastman, 2005), the daily minimum of the core body temperature rhythm (CBTmin), which coincides with the daily low-point of the circadian cycle, occurs ~7 h after melatonin onset (Cagnacci et al., 1996; Brown et al., 1997; Eastman et al., 2000), and the daily peak of the core body temperature rhythm (CBTmax), which coincides with the daily high-point of the circadian cycle, occurs ~12 after CBTmin (Dijk et al., 1992). Therefore, a person who normally sleeps from 23:00 to 07:00 will have melatonin onset at ~21:00, CBTmin at ~04:00, and CBTmax at ~16:00 (**Figure 1**). Maximal sleepiness, and poorest mental/physical performance, occur in the 2–3 h either side of CBTmin, and maximal alertness, and greatest mental/physical performance, occur in the 2–3 h either side of CBTmax (Dijk et al., 1992). The protocols required to directly assess the timing of melatonin onset and CBTmin are invasive, time-consuming, and costly, so these variables are typically estimated based on the habitual timing of sleep and wake. However, work is currently being conducted to develop

biomarkers of circadian phase based on the analysis of white blood cells from a single sample (Ueda et al., 2004; Wittenbrink et al., 2018).

Immediately after westward flight, the circadian system will be running ahead of the local time zone. For example, after a flight from London to Los Angeles (8 h west), when the body clock is ready for bed at 23:00 London time, it will only be 15:00 in Los Angeles. To adjust to the new time zone, the circadian system has to delay, or shift backward, or move later. Conversely, immediately after eastward flight, the circadian system will be running behind the local time zone. For example, after a flight from Los Angeles to London (8 h east), when it is time to get up at 07:00 in London, it is only 23:00 in Los Angeles, so the body clock will be ready for bed. To adjust to the new time zone, the circadian system has to advance, or shift forward, or move earlier.

Delays and advances in the timing of the circadian system can be facilitated by appropriately timed light exposure, melatonin ingestion, and/or exercise (**Figure 1**). The direction and size of the shift in the timing of the circadian system in response to these stimuli depend on the time of day, or more correctly, the circadian phase, that the stimuli occur. These effects are described by phase response curves (PRCs):

- (A) Phase response curve to light. Light exposure in the ~12 h prior to CBTmin shifts the circadian system backward/late, or causes a phase delay; light exposure in the ~12 h after CBTmin shifts the circadian system forward/earlier, or causes a phase advance; and the largest shifts occur when light exposure occurs in the 3–6 h either side of CBTmin (Czeisler et al., 1989; Khalsa et al., 2003). Furthermore, the degree to which the timing of the circadian system can be shifted by exposure to light depends the duration of the exposure (Rimmer et al., 2000), the intensity of the light (Boivin et al., 1996), and the wavelength of the light (Wright and Lack, 2001; Rüger et al., 2013). The largest phase shifts occur when the duration of exposure is longer, when the intensity of light is higher, and when the wavelength of light is shorter (i.e., blue).
- (B) Phase response curves to melatonin. Melatonin ingested in the late afternoon or early evening, a few hours prior to the onset of endogenous melatonin production and several hours prior to CBTmin, shifts the circadian system forward/earlier, or causes a phase advance (Burgess et al., 2008, 2010). Conversely, melatonin ingested in the morning, a few hours after habitual get-up time and several hours after CBTmin, shifts the circadian system backward/late, or causes a phase delay (Burgess et al., 2008, 2010). Separate PRCs have been created for exogenous melatonin at a “physiological” dose of 0.5 mg and at a “pharmacological” dose of 3.0 mg. For a 0.5 mg dose, it is estimated that maximum advances occur for ingestion ~10.5 h before CBTmin, or ~5.5 h before habitual bedtime, and maximum delays occur for ingestion ~6.5 h after CBTmin, or ~3.5 h after habitual get-up time (Burgess et al., 2010). For a 3.0 mg dose, it is estimated that maximum advances occur for ingestion ~11.5 h before CBTmin, or ~6.5 h before habitual bedtime, and maximum delays occur for ingestion ~4 h

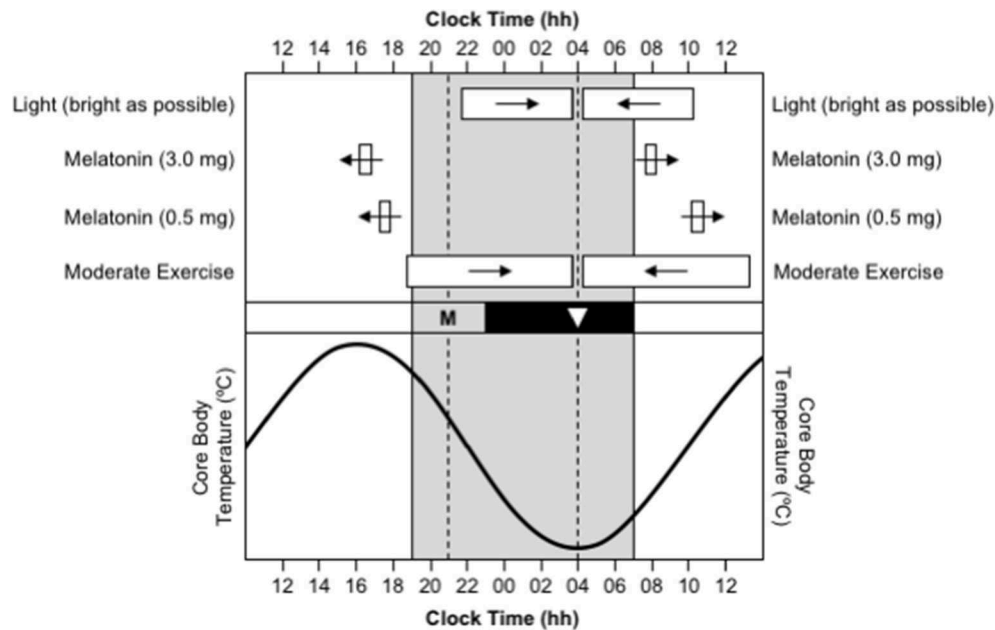


FIGURE 1 | Use of appropriately timed stimuli to shift the timing of the human circadian system. In all sections, the gray background represents night-time. In the bottom section, the sinusoid-shaped line indicates the daily rhythm of core body temperature. In the middle section, the black bar represents habitual bedtime, “M” represents the evening onset of endogenous melatonin production, and the inverted triangle represents the daily minimum of core body temperature. In the top section, the white bars indicate the optimal timing of light exposure, exogenous melatonin ingestion, and moderate-intensity exercise, required to facilitate delays (right-pointing arrows) and advances (left-pointing arrows) in the circadian system. This figure was inspired by a similar figure presented by Waterhouse et al. (2007).

after CBTmin, or ~ 1 h after habitual get-up time (Burgess et al., 2008). The two doses of exogenous melatonin produce phase shifts of a similar size, but the 3.0 mg dose produces more reliable phase shifts than the 0.5 mg dose (Burgess et al., 2010). Goodness of fit data were not provided for either PRC, so the apparent precision of the estimated timing of melatonin ingestion for maximum advances and delays should be read with caution.

- (C) Phase response curve to exercise. There is some evidence that exercise has phase-shifting properties (Eastman et al., 1995; Buxton et al., 2003), and a phase response curve to a 1 h bout of moderate-intensity exercise has been published recently (Youngstedt et al., 2019). The PRC indicates that exercise in the ~ 9 h prior to CBTmin shifts the circadian system backward/later, or causes a phase delay, and exercise in the ~ 9 h after CBTmin shifts the circadian system forward/earlier, or causes a phase advance. However, the protocol to obtain the data to construct this PRC was conducted with moderate-intensity light at 50 lux, instead of low-intensity light at <10 – 15 lux, so a PRC for the effects of exercise independent of light is still to be established.

This manuscript provides examples of how to use light and/or melatonin to shift the timing of the circadian system so that jet lag can be overcome as quickly as possible (Figures 4, 5). Light and melatonin can be used independently, but their phase-shifting effects are additive—particularly for phase advances—so using them together should produce a greater effect than either one on

its own (Wirz-Justice et al., 2004; Revell et al., 2006; Burke et al., 2013). In contrast, there is no evidence that the effects of light and exercise are additive, so exercise is not included in the adaptation guides. However, the appropriate times to conduct exercise for its phase-shifting properties coincide with the appropriate times for light exposure.

THE EXPERIENCE OF JET LAG DEPENDS ON THE DIRECTION OF TRAVEL

The most obvious consequences of jet lag are poor night-time sleep, excessive daytime sleepiness, and poor mental and physical performance (Waterhouse et al., 2004). However, the experience of jet lag greatly depends on the direction of travel. Consider the difference in the manifestation of jet lag between westward and eastward flights over 8 time zones—as occurs with travel between Western Europe (UTC+0 h) and the USA’s west coast (UTC-8 h). Immediately after flying 8 h west, say from London to Los Angeles, the circadian system is still entrained to the timing of zeitgebers in London, so the daily low-point of the circadian cycle occurs at 04:00 London time, which is 20:00 in Los Angeles, and the daily high-point of the circadian cycle occurs at 16:00 London time, which is 08:00 in Los Angeles (Figure 2A). Consequently, at least initially in Los Angeles, a person will feel sleepy in the evening and they will have difficulty staying asleep until their normal wake time in the morning. If they are an athlete, they will also have difficulty training or competing at their highest

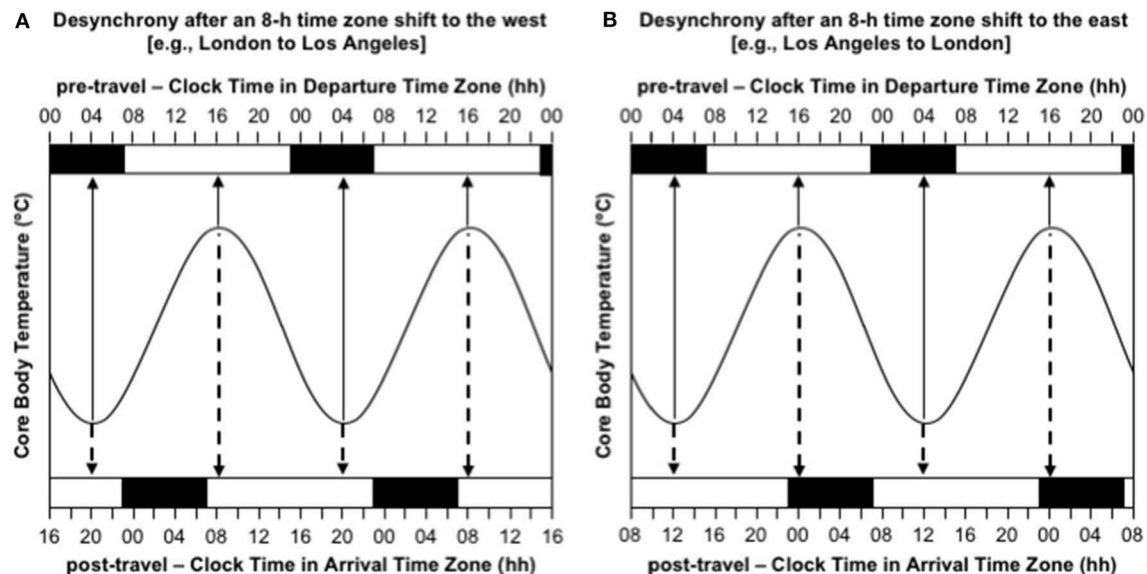


FIGURE 2 | Desynchrony between the circadian system and the desired timing of sleep/wake immediately after time zone shifts to the west (**A**) and east (**B**). In both panels, the sinusoid-shaped line represents core body temperature; the solid arrows indicate the timing of the daily minimum and maximum of core body temperature in the departure time zone; the dashed arrows indicate the timing of the daily minimum and maximum of core body temperature in the arrival time zone; the black bars at the top axis represent night-time sleep in the departure time zone; the black bars at the bottom axis represent night-time sleep in the arrival time zone; time is double-potted with 2×24 h periods. (**A**) Desynchrony after 8-h time zone shift to the west (e.g., London to Los Angeles). (**B**) Desynchrony after an 8-h time zone shift to the east (e.g., Los Angeles to London).

level in the evening. In contrast, immediately after flying 8 h east, say from Los Angeles to London, the circadian system is still entrained to the timing of zeitgebers in Los Angeles, so the daily low-point of the circadian cycle occurs at 04:00 Los Angeles time, which is midday in London, and the daily high-point of the circadian cycle occurs at 16:00 Los Angeles time, which is midnight in London (**Figure 2B**). Consequently, at least initially in London, a person will feel sleepy in the late morning and early afternoon and they will have difficulty falling asleep at their normal bedtime in the evening. If they are an athlete, they will also have difficulty training and/or competing at their highest level in the late morning and early afternoon.

SUNLIGHT CAN EITHER HELP OR HINDER ADAPTATION TO A NEW TIME ZONE

From anecdotal reports, it seems that a common perception among laypersons, is that maximizing exposure to sunlight in a new time zone is an effective strategy for overcoming jet lag. In general, this approach will work quite well after westward travel, but it may actually be counterproductive after eastward travel. To overcome jet lag, the circadian system must adjust so that it becomes aligned with the desired timing of sleep and wake in the new time zone. The most effective way to adjust the circadian system, or shift the timing of the body clock, is with exposure to light. However, light exposure, *per se*, does not cause the circadian system to align with the new time zone. Rather, light exposure shifts the timing of the body clock either backward

(delay) or forward (advance), as required after westward and eastward travel, respectively.

Consider the difference in the effectiveness of indiscriminately maximizing exposure to sunlight after westward and eastward travel over 8 time zones. Immediately after flying 8 h west, say from London to Los Angeles, the circadian system is still entrained to the timing of zeitgebers in London, so CBTmin occurs at 04:00 London time, which is 20:00 in Los Angeles. To adapt to the new time zone in Los Angeles, the circadian system must delay by 8 h, so that CBTmin occurs at the normal time of 04:00 instead of 20:00 (**Figure 3A**). If a person indiscriminately seeks sunlight during the daytime, they will be exposed to sunlight before CBTmin at 20:00, which will provide a delay signal, and they will be exposed to little or no sunlight after CBTmin, because the sun sets earlier than 20:00 in Los Angeles at most times of year, so they will not receive an advance signal (**Figure 3A**). In this case, maximizing exposure to sunlight would aid the desired phase delay. In contrast, immediately after flying 8 h east, say from Los Angeles to London, the circadian system is still entrained to the timing of zeitgebers in Los Angeles, so CBTmin occurs at 04:00 Los Angeles time, which is midday in London. To adapt to the new time zone in London, the circadian system must advance by 8 h, so that CBTmin occurs at the normal time of 04:00 instead of midday (**Figure 3B**). If a person indiscriminately seeks out sunlight during the daytime, they will be exposed to sunlight before CBTmin at midday, which will provide a delay signal, and they will be exposed to sunlight after CBTmin at midday, which will provide an advance signal (**Figure 3B**). In this case, maximizing exposure to sunlight would

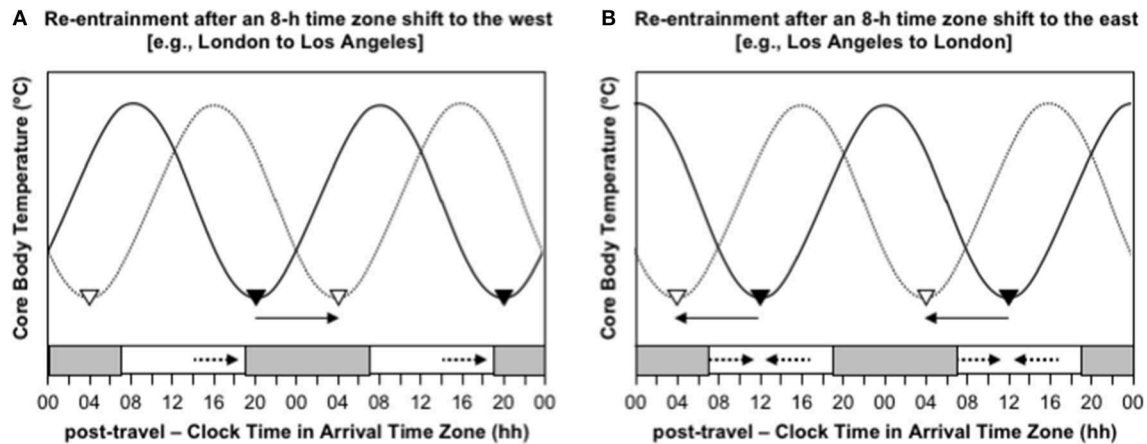


FIGURE 3 | Re-entrainment of the circadian system to local time cues after time zone shifts to the west **(A)** and east **(B)**. In both panels, the solid sinusoid-shaped line with black inverted triangles represent core body temperature and the timing of its daily minimum immediately after flight; the dashed sinusoid-shaped line with white inverted triangles represent core body temperature and the timing of its daily minimum after re-entrainment to the arrival time zone; the gray bars at the bottom axis represent night-time in the arrival time zone; the solid arrows represent the direction of the phase shift required for adaptation to the new time zone; the dashed arrows represent the direction of the phase shift signal provided by light exposure at that time of day; time is double-plotted with 2×24 h periods. **(A)** Re-entrainment after an 8-h time zone shift to the west (e.g., London to Los Angeles). **(B)** Re-entrainment after an 8-h time zone shift to the east (e.g., Los Angeles to London).

provide contradictory signals, which would inhibit adaptation to the new time zone. A better approach would be to avoid sunlight before midday and maximize exposure to sunlight in the 3–6 h after midday—that would limit the delay signal and maximize the desired advance signal.

ENDOGENOUS MELATONIN HAS HYPNOTIC AND CHRONOBIOTIC PROPERTIES

Exogenous melatonin is both a chronobiotic and an hypnotic, i.e., it can shift the timing of the circadian system (Arendt and Skene, 2005), but it can also make it easier to fall asleep and/or stay asleep (van den Heuvel et al., 2005; Zhdanova, 2005). The hypnotic effects of melatonin depend on the time of day, or more correctly, the circadian phase, that the melatonin is ingested (Wyatt et al., 2006). For exogenous melatonin, doses of ≤ 0.5 mg and 1–5 mg are typically considered to be physiological and pharmacological, respectively. If ingested when endogenous melatonin is high, i.e., during body clock night-time, neither a physiological 0.3 mg dose, nor a pharmacological 5.0 mg dose, of exogenous melatonin increase sleepiness. In contrast, if ingested when endogenous melatonin is low, i.e., during body clock daytime, both doses of exogenous melatonin increase sleepiness to a similar extent. Therefore, if melatonin is used as a chronobiotic at either physiological or pharmacological doses, its potential hypnotic effects should also be considered. This manuscript provides advice on how to exploit the chronobiotic effects of a pharmacological 3.0 mg dose, rather than a physiological 0.5 mg dose, because the higher dose produces more reliable phase shifts than the lower dose (Burgess et al., 2010).

SCHEDULES FOR SHIFTING THE TIMING OF THE CIRCADIAN SYSTEM AFTER TRANSMERIDIAN AIR TRAVEL

After long-haul flights across multiple time zones, the circadian system is initially aligned with the timing of zeitgebers at the point of departure rather than zeitgebers at the new location. To overcome jet lag, the timing of the circadian system must shift so that it becomes aligned with the new time zone. Adaptation guides have previously been presented for relatively large time zone shifts of 7–9 h (Eastman and Burgess, 2009; Revell and Eastman, 2012). The following sub-sections provide examples of how to use light and/or melatonin to shift the timing of the circadian system so that jet lag can be overcome as quickly as possible after rapid time zone changes of 3, 6, 9, and 12 h to the west and east.

The adaptation guides are based on three major assumptions:

- Assumption 1—A person who normally sleeps from 23:00 to 07:00 will have the daily minimum of their core body temperature rhythm (CBTmin) at $\sim 04:00$. For people with earlier or later bedtimes, the timing of light and melatonin should be adjusted accordingly. For example, a person who normally sleeps from 22:00 to 06:00 will have CBTmin at $\sim 03:00$ instead of $\sim 04:00$, so the timing of light and melatonin should be 1 h earlier than in the examples, and a person who normally sleeps from midnight to 08:00 will have CBTmin at $\sim 05:00$ instead of $\sim 04:00$, so the timing of light and melatonin should be 1 h later than in the examples.
- Assumption 2—A person wants their main daily sleep period to occur at the same local time in the arrival time zone as in their normal time zone (i.e., 23:00 to 07:00 in the

examples provided). In some cases, this may be difficult to achieve in practice, particularly on the first few days after travel. For example, after a shift of 9 time zones west (**Figure 4C**), a person may be sleepy in the evening, so they may wish to go to bed earlier than usual (e.g., 21:00–05:00 instead of 23:00–07:00). Similarly, after a shift of 9 time zones east (**Figure 5C**), a person may not be sleepy in the evening, so they may wish to go to bed later than usual (e.g., 01:00–09:00 instead of 23:00–07:00). In situations where the timing of the main sleep period differs from the timing in the relevant guide, this should not interfere with adaptation provided that light exposure/avoidance and/or melatonin ingestion still occur at the appropriate time.

- Assumption 3—Arrival in the new time zone occurs at 13:00 local time. In certain cases where arrival is earlier or later, it may be necessary for the adaptation schedule to be advanced or delayed by a day, respectively. For example, for a 21:00 arrival after a 9 h westward time zone change, the day 0 schedule should be delayed to occur on day 1 because arrival would occur after the critical time for light exposure, so adaptation has to begin one day “late” (**Figure 4C**). Conversely, for a 06:00 arrival after a 6 h eastward time zone change, the day 1 schedule should be advanced to occur on day 0 because arrival would occur prior to the critical time for light exposure, so adaptation can begin one day “early” (**Figure 5B**).

When a schedule requires light exposure during the daytime, it is best to be outside in sunlight without sunglasses. When a schedule requires light exposure after sunset, bright indoor light, or a light box, or light-emitting glasses should be used. When a schedule requires light avoidance, it is best to be inside with lights off or as dim as possible—it may even be appropriate to have a nap (limited to 1 h so as not to interfere with night-time sleep). When a schedule requires light avoidance during the daytime and being outside is unavoidable, wrap-around sunglasses with minimal light transmission should be worn.

Complete adaptation after transmeridian flight is achieved when the circadian system shifts sufficiently such that CBTmin in the arrival time zone occurs at the same time as it occurred in the departure time zone (i.e., 04:00, assuming habitual bed time of 23:00–07:00). However, this can take several days, so from a practical point of view, it is most important that the circadian system shifts sufficiently such that CBTmin occurs during the main night-time sleep period in the new time zone—a state of partial adaptation (Eastman and Burgess, 2009). Once CBTmin—the daily low-point of the circadian cycle—occurs during the night-time, sleep should be longer and of better quality, daytime sleepiness should be reduced, and mental and physical performance should be higher, i.e., the symptoms of jet lag should be greatly reduced. This distinction between complete and partial adaptation has previously been applied to shift workers switching from day work to night work (Lee et al., 2006).

NB. If melatonin is used by an athlete, a responsible party must ensure that it is not a prohibited substance under the relevant

drug code and that it is sourced from a reputable supplier (to ensure purity and dose accuracy).

Adaptation schedules after westward flight:

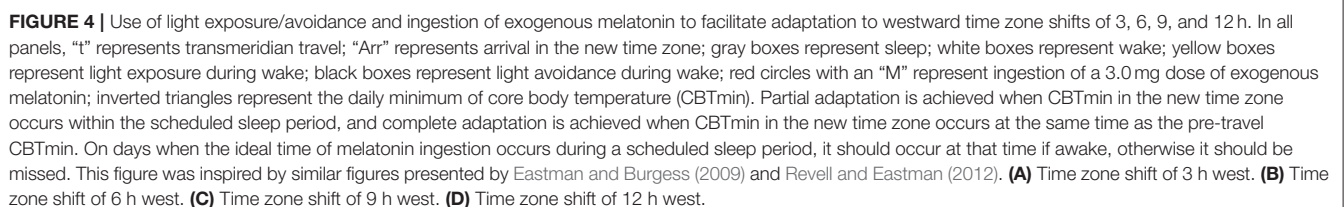
- Time zone change of 3 h west (e.g., from Wellington, New Zealand, to Tokyo, Japan). Immediately after flight, CBTmin will occur at 01:00 local time, instead of 04:00, and CBTmax will occur at 13:00 local time, instead of 16:00. To adapt to the new time zone, the circadian system has to delay by 3 h—partial and complete adaptation should be achieved on days 1 and 4, respectively (**Figure 4A**).
- Time zone change of 6 h west (e.g., from Anchorage, USA, to Tokyo, Japan). Immediately after flight, CBTmin will occur at 22:00 local time, instead of 04:00, and CBTmax will occur at 10:00 local time, instead of 16:00. To adapt to the new time zone, the circadian system has to delay by 6 h—partial and complete adaptation should be achieved on days 2 and 6, respectively (**Figure 4B**).
- Time zone change of 9 h west (e.g., from Minneapolis, USA, to Tokyo, Japan). Immediately after flight, CBTmin will occur at 19:00 local time, instead of 04:00, and CBTmax will occur at 07:00 local time, instead of 16:00. To adapt to the new time zone, the circadian system has to delay by 9 h—partial and complete adaptation should be achieved on days 3 and 7, respectively (**Figure 4C**).

Adaptation schedules after eastward flight:

- Time zone change of 3 h east (e.g., from Dhaka, Bangladesh, to Tokyo, Japan). Immediately after flight, CBTmin will occur at 07:00 local time, instead of 04:00, and CBTmax will occur at 19:00 local time, instead of 16:00. To adapt to the new time zone, the circadian system has to advance by 3 h—partial and complete adaptation should be achieved on days 1 and 4, respectively (**Figure 5A**).
- Time zone change of 6 h east (e.g., from Doha, Qatar, to Tokyo, Japan). Immediately after flight, CBTmin will occur at 10:00 local time, instead of 04:00, and CBTmax will occur at 22:00 local time, instead of 16:00. To adapt to the new time zone, the circadian system has to advance by 6 h—partial and complete adaptation should be achieved on days 3 and 6, respectively (**Figure 5B**).
- Time zone change of 9 h east (e.g., from London, United Kingdom, to Tokyo, Japan). Immediately after flight, CBTmin will occur at 13:00 local time, instead of 04:00, and CBTmax will occur at 01:00 local time, instead of 16:00. To adapt to the new time zone, the circadian system has to advance by 9 h—partial and complete adaptation should be achieved on days 5 and 8, respectively (**Figure 5C**).

Adaptation schedules after a time zone change of 12 h east/west (e.g., from Buenos Aires, Argentina, to Tokyo, Japan):

- Immediately after flight, CBTmin will occur at 16:00 local time, instead of 04:00, and CBTmax will occur at 04:00 local time, instead of 16:00. To adapt to the new time zone, the circadian system could either delay or advance by 12 h. However, given that the human circadian system has a natural period of ~24.2 h such that it has a greater propensity to delay than to



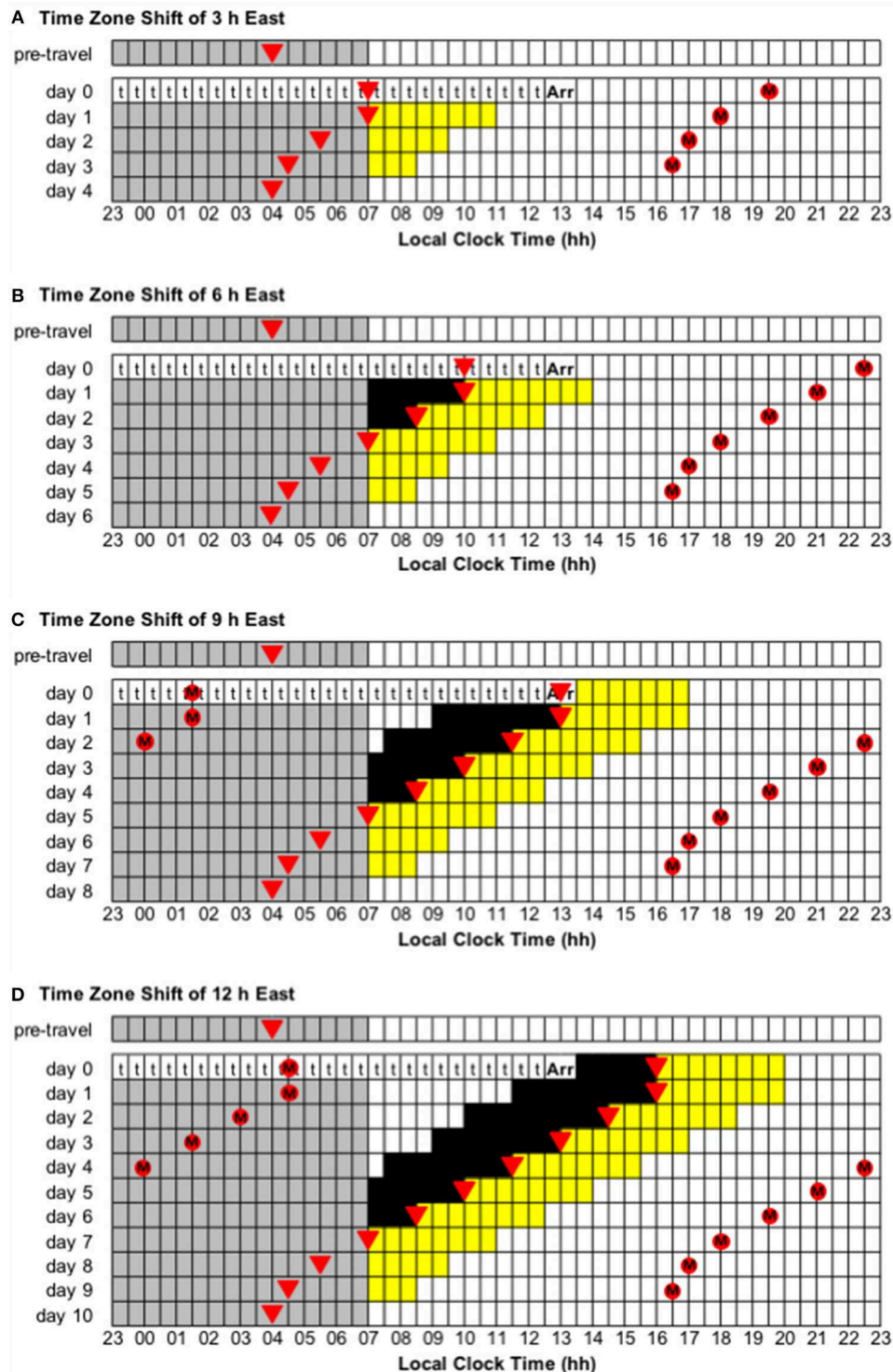


FIGURE 5 | Use of light exposure/avoidance and ingestion of exogenous melatonin to facilitate adaptation to eastward time zone shifts of 3, 6, 9, and 12 h. In all panels, “t” represents transmeridian travel; “Arr” represents arrival in the new time zone; gray boxes represent sleep; white boxes represent wake; yellow boxes represent light exposure during wake; black boxes represent light avoidance during wake; red circles with an “M” represent ingestion of a 3.0 mg dose of exogenous melatonin; inverted triangles represent the daily minimum of core body temperature (CBTmin). Partial adaptation is achieved when CBTmin in the new time zone occurs within the scheduled sleep period, and complete adaptation is achieved when CBTmin in the new time zone occurs at the same time as the pre-travel CBTmin. On days when the ideal time of melatonin ingestion occurs during a scheduled sleep period, it should occur at that time if awake, otherwise it should be missed. This figure was inspired by similar figures presented by Eastman and Burgess (2009) and Revell and Eastman (2012). **(A)** Time zone shift of 3 h east. **(B)** Time zone shift of 6 h east. **(C)** Time zone shift of 9 h east. **(D)** Time zone shift of 12 h east.

advance (Czeisler et al., 1999; Zhou et al., 2011b), it is more common to adapt by delay. Using a delay schedule, partial and complete adaptation should be achieved on days 6 and 10, respectively (Figure 4D). Using an advance schedule, partial and complete adaptation should be achieved on days 7 and 10, respectively (Figure 5D).

EVIDENCE OF EFFICACY FOR JET LAG INTERVENTIONS BASED ON LIGHT AND/OR MELATONIN

This manuscript contains recommendations regarding the use of judiciously timed light exposure/avoidance and ingestion of exogenous melatonin to minimize jet lag by facilitating adaptation of the circadian system to a new time zone. These recommendations are primarily based on information contained in phase response curves, which describe the effects of light and melatonin on the timing of the circadian system (Czeisler et al., 1989; Khalsa et al., 2003; Burgess et al., 2008, 2010). Computer-based simulations with experimentally-validated mathematical models have demonstrated that schedules of light exposure and avoidance could be used to increase the rate of adaptation after a rapid shift in the timing of the light-dark cycle (Serkh and Forger, 2014). Laboratory-based trials have established that both light and melatonin, when administered alone, can shift the timing of the circadian system (Deacon and Arendt, 1995; Middleton et al., 1997; Burgess et al., 2003a; Smith and Eastman, 2009), and when used in combination, the phase-shifting effects of light and melatonin are additive (Revell et al., 2006; Paul et al., 2011). Only a few field studies have been conducted to examine the efficacy of light-based interventions for the treatment of jet lag—and their results are equivocal (Boulos et al., 2002; Lahti et al., 2007; Thompson et al., 2012). In contrast, a meta-analysis of 10 field studies examining the efficacy of melatonin-based interventions indicates that they are effective at shifting the body clock and at reducing subjective ratings of jet lag (Herxheimer and Petrie, 2002). To date, the efficacy of combined light/melatonin interventions has not been assessed in field-based settings, so this is a critical next step for the advancement of knowledge in this field.

COULD ADAPTATION BEGIN BEFORE/DURING TRANSMERIDIAN TRAVEL?

Specific guides have not been provided here, but it is possible to begin shifting the circadian system in the desired direction before and/or during transmeridian flight (see Eastman and Burgess, 2009; Revell and Eastman, 2012). The potential advantage of pre-shifting is that it should reduce the amount of time required to adapt to local time cues in the arrival time zone, such that the symptoms of jet lag are less pronounced and/or occur over fewer days. Conversely, the potential disadvantages of pre-shifting are that it could interfere with sleep and be socially disruptive prior

to travel and it could make it more difficult to estimate the timing of the daily minimum in core body temperature (CBT_{min}), and thus the appropriate times for light exposure/avoidance and/or melatonin ingestion, upon arrival in the new time zone.

To delay the circadian system in the 3–4 days prior to westward travel, gradually move bedtime and get-up time later (i.e., 30–60 min per day), maximize evening light exposure, minimize morning light exposure, and take 3.0 mg of melatonin 1 h after rising from bed. To advance the circadian system in the 3–4 days prior to eastward travel, gradually move bedtime and get-up time earlier (i.e., 30–60 min per day), minimize evening light exposure, maximize morning light exposure, and take 3.0 mg of melatonin 6.5 h before bed.

To shift the circadian system during travel, rather than setting a watch and attempting to align sleep and wake with the arrival time zone, light exposure/avoidance and melatonin ingestion should be timed according to the departure time zone. To delay the circadian system during westward travel, maximize light exposure in the ~3 h before CBT_{min}, avoid light in the ~3 h after CBT_{min} (by sleeping if possible), and take 3.0 mg of melatonin 4 h after CBT_{min}. To advance the circadian system during eastward travel, take 3.0 mg of melatonin 11.5 h before CBT_{min}, avoid light in the ~3 h prior to CBT_{min} (by sleeping if possible), and maximize light exposure in the ~3 h after CBT_{min}.

CONCLUSIONS

Long-haul flight over several time zones causes both travel fatigue and jet lag. The most obvious consequences of jet lag are poor sleep at night, excessive sleepiness during the day, and poor mental and physical performance. These consequences occur because the human circadian system cannot immediately adapt to time cues in a new time zone. This manuscript has presented recommendations on how to minimize jet lag using judiciously timed light exposure/avoidance and ingestion of exogenous melatonin to facilitate adaptation of the circadian system to a new time zone. These recommendations are based on the latest information regarding the effects of light and melatonin on the human circadian system. There are potential barriers to the practical implementation of these recommendations, so it will be critical to assess their efficacy in natural settings, preferably using experimental designs with randomization to treatment and control groups.

AUTHOR CONTRIBUTIONS

GR and CS contributed to conception of the manuscript. GR wrote the first draft of the manuscript. GR and CS contributed to revision of the manuscript. GR and CS read and approved the submitted version of the manuscript.

FUNDING

The authors are grateful for funding to support this manuscript from the Australian Research Council's Discovery Project scheme (DP160104909).

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Conflict of Interest Statement: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Modification and Applicability of Questionnaires to Assess the Recovery-Stress State Among Adolescent and Child Athletes

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OPEN ACCESS

Edited by:

Matthew J. Barnes,
Massey University, New Zealand

Reviewed by:

Will Hopkins,
Victoria University, Australia
Nicola McCulloch,
Northumbria University,
United Kingdom

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Specialty section:

This article was submitted to
Exercise Physiology,
a section of the journal
Frontiers in Physiology

Received: 03 November 2018

Accepted: 31 October 2019

Published: 20 November 2019

Citation:

Kölling S, Ferrauti A, Meyer T,
Pfeiffer M and Kellmann M (2019)
Modification and Applicability
of Questionnaires to Assess
the Recovery-Stress State Among
Adolescent and Child Athletes.
Front. Physiol. 10:1414.
doi: 10.3389/fphys.2019.01414

Despite the general consensus regarding the implementation of self-report measures in the training monitoring, there is a lack of research about their applicability and comprehensibility among developing athletes. However, this target group needs special considerations to manage the increasing training demands while maintaining health and performance. This study deals with challenges of applying recovery-stress questionnaires which were validated with adult populations among developing athletes and presents a possible approach to enhance their applicability. In two phases, the Acute Recovery and Stress Scale (ARSS), a 32-adjective list covering eight scales, and the 8-item derived version, the Short Recovery and Stress Scale (SRSS) were answered by 1052 athletes between 10 and 16 years. Phase 1 included 302 14- to 16-year-old athletes who used the original questionnaires with the additional option to mark “I don’t understand,” while modified versions with additional explanations (phase 2) were applied to 438 adolescents (14.7 ± 0.6 years) and 312 child athletes (11.8 ± 1.1 years). Data of the original validation sample ($n = 442$) were reanalyzed to examine measurement invariance between adults and adolescents. The results showed comparable psychometric properties to the validation sample (e.g., $r_{it} > 0.30$) and acceptable fit indices via confirmatory factor analyses (CFA), although more difficulties and limitations were present within the younger groups (e.g., Cronbach’s α between 0.50 and 0.87), especially among 10- and 11-year-olds. The original as well as the modified SRSS, on the other hand, indicated good applicability (Cronbach’s α between 0.72 and 0.80). Multigroup CFA revealed measurement invariance of the original ARSS among adults and adolescents and of the modified ARSS among adolescents and children. Overall, the present study confirmed the assumption that questionnaires designed by and for adults cannot be directly transferred to younger athletes. The peculiarities and differences in the cognitive and affective development of each age

group need to be considered. Future research needs to identify a cut-off age to start the proper use of psychometric tools, especially for state-oriented assessments for routine application in training monitoring. Further modifications and long-term investigations are necessary to implement psychometric monitoring in high-performance environments within youth sport.

Keywords: training, monitoring, psychometrics, development, sports

INTRODUCTION

Despite the goal of the International Olympic Committee to develop healthy, capable, and resilient young athletes (Bergeron et al., 2015), training demands on developing athletes are high in order to achieve the elite level. In addition to their sport, these athletes are facing a double burden due to school and social commitments and other non-sport stressors. Life event stress, as an example, was shown to predict injury occurrence among junior soccer players (Johnson and Ivarsson, 2011). In general, there is consensus about the necessity to manage an adequate balance between stress and recovery (Kellmann and Beckmann, 2018; Kellmann et al., 2018), which is supported by the systematic review of Drew and Finch (2016) who are indicating an emerging moderate evidence for the relationship between training load and the risk of injury and illness. Therefore, effective management of training and competition, such as periodization or the length of mid-season and off-season breaks, plays an essential role in the maintenance of performance and injury prevention (Jones et al., 2017). For instance, Phibbs et al. (2018) recently analyzed the weekly match and training loads of adolescent rugby union players during 14 weeks. They found a large within-player variability that represented the inconsistent match scheduling which, furthermore, exposed the players to an increased risk of injury. According to a recent systematic review of longitudinal studies investigating the association between training load with injury and illness, it is not only the magnitude of external training load but also the increase of the intensity of external load (e.g., speed, weights) as well as the internal load (e.g., perceived exertion, heart rate) which result in an augmented stress and injury risk (Jones et al., 2017). Excessive training overload combined with inadequate recovery may lead to non-functional overreaching (NFOR) and can develop into the overtraining syndrome (OTS) which is characterized by symptoms of fatigue, performance decline, and mood disturbances (Meeusen et al., 2013).

DiFiori et al. (2014) raise the concern of overuse injury and burnout resulting from an increased pressure to begin with high-intensity training and the emphasis on competitive success already in youth sport. In their position statement, they point out the lack of research on the incidence and prevalence of overuse injuries in children and adolescents. Nevertheless, there is some evidence supporting the relevance and need for special attention to develop prevention programmes. A survey among 11- to 18-year-old English athletes ($N = 376$) revealed that approximately one third has experienced a state of NFOR or OTS (Matos et al., 2011). Similar rates were found in adolescent swimmers ($N = 231$) across Greece, Japan, Sweden, and the

United States, with 34.6% of the total sample and a range from 20.5 to 45.1% between countries (Raglin et al., 2000). Bergeron et al. (2015) emphasize that there is still a lack of evidence-based injury prevention strategies in sports with a high risk of injury, such as rugby, field hockey, soccer, volleyball, running, lacrosse, gymnastics, martial arts, tennis, and wrestling.

One important approach is monitoring the athlete's training response and recovery-stress state to ensure the readiness to perform as well as to sustain the athlete's health and well-being and prevent injuries in the long-term (Murray, 2017; Kellmann et al., 2018). This is further important in terms of effective talent development and preserving the limited talent pool (Murray, 2017). Especially among adolescent athletes, it seems important to take into account their individual perception and assessment of the training load, as Brink et al. (2014) have shown that under-17 and under-19 soccer players perceived the training as harder than it was intended to be by the coach. Even though coaches showed an altered rating of observed exertion to align with the athletes' responses after training sessions, small to moderate differences were still found in a study of youth hockey, netball, rugby, and soccer players (Scantlebury et al., 2018). Despite the documentation of the training load and measuring the internal load via physiological responses (e.g., heart rate, creatine kinase), self-report measures are a vital source of information (Kellmann, 2000; Bourdon et al., 2017; Scantlebury et al., 2017). As the manifestation of the OTS is a process over a period of time, psychological changes and mood disturbances have been identified as successful indicators (Steinacker et al., 1999; Meeusen et al., 2013). According to a systematic review, acute and chronic training loads were better reflected by subjective measures indicating an impaired well-being following acute increases of training as well as chronic training and improvements after acute decreases in training load (Saw et al., 2016). Considering the implementation of psychometric monitoring tools, Saw et al. (2017) highlight the importance of established questionnaires which fulfill the quality criteria in terms of a theoretical basis, reliability, and validity. While there is a number of instruments available (for an overview see Nässi et al., 2017b), their applicability among adolescents or even children needs to be considered critically and should not be applied before thorough pretesting (Borgers et al., 2000). While it seems that, with the help of parents, children at the age of five may already be able to provide reliable and valid replies to their health-related quality of life (Varni et al., 2007), Williams et al. (1994) point out that young people may have difficulties applying the *Rating of Perceived Exertion* scale (Borg, 1998), as it demands comprehension and translation of the verbal expressions and the range of numbers to their presumably

rudimentary concept of exercise and the accompanied sensations. Therefore, the *Children's Effort Rating Table* has been developed for 6- to 9-year old children (Williams et al., 1994). Another modification has been reported by Yelling et al. (2002) who have illustrated the verbal and numerical rating scale with pictorial images of exertion. However, the recovery-stress continuum is multi-dimensional and cannot be simplified by assessing only the exertion or the absence thereof (Kellmann, 2010; Heidari et al., 2018). While it is recommended to capture different aspects of recovery and stress (e.g., mood, emotional well-being), it is doubtful whether existing questionnaires which were developed and validated among adults can be transferred to be used on younger athletes. In general, there are two requirements that need to be fulfilled before implementing self-report measures in this context, i.e., the cognitive development to read and understand the items and the children's level of self-perception to differentiate their current psychophysiological state and its representation on rating scales. Borgers et al. (2000) differentiate between reading ability, which involves the vocabulary in general and its decoding, and language ability which involves reading comprehension.

An eligible tool for training monitoring is the Acute Recovery and Stress Scale (ARSS) and its shortened version, the Short Recovery and Stress Scale (SRSS, Kellmann et al., 2016) which are established instruments to assess multiple facets of recovery and stress states (i.e., physical, mental, emotional, and overall dimensions). These were developed to support every-day and long-term training monitoring by showing sensitivity to change in an economical way (Hitzschke et al., 2017). Several studies indicate their sport-specific applicability as well as validity in different training settings (Kölling et al., 2015; Collette et al., 2018; Pelka et al., 2018). However, their application for athletes younger than 16 years has not been examined yet. As the 32 items of the ARSS assess the current recovery-stress state on the basis of single adjectives, the understanding of them by children and adolescents needs to be investigated. A particularity of the SRSS is its derivation of the ARSS's scales. While four items are comprised into one of the ARSS's scales, these eight scales are assessed as single items in the SRSS and represent a somewhat broader construct of the recovery-stress dimensions. The corresponding adjectives (ARSS items) serve as descriptors below each SRSS item to support their meaning. However, it needs to be verified whether additional explanations are needed among younger athletes. The present study aims at pointing out likely challenges of application and demonstrating possible approaches to modify and adapt existing tools for younger athletes.

MATERIALS AND METHODS

Participants

Overall, 1052 athletes (75.6% male) participated in the different phases of the study. The majority (83.9%) was engaged in team sports such as soccer and handball, while 15.8% belonged to individual sports. **Table 1** provides an overview of participants' characteristics in each of the phases. The group of phase 1

consisted of 302 athletes between 14 and 16 years. Most of the data was collected in several selection-focused training camps. During a nationwide selection course of the handball association, 239 players of that age group were recruited. Additionally, 17 athletes were part of an under-15 and 21 athletes of an under-16 soccer team. In order to retain the anonymity of the athletes and to prevent distorted responses, the questionnaires were answered without individual demographic information. In phase 2, participants were divided into the group of adolescents between 14 and 16 years ($n = 438$) and child athletes between 10 and 13 years ($n = 312$). The athletes and their parents were informed about the purpose of the study and informed consent was attained by athletes as well as parents prior to the data collection. Ethical approval was obtained by the local ethic committee.

Procedure

The study consisted of two evaluation phases which were conducted successively (**Table 1**). In phase 1, the ARSS was applied among adolescents with the option to mark "*I don't understand*" beside the original rating scale, while the SRSS remained in its original form. Following initial feedback based on the answers and the most common ratings, four items were identified and modified with additional adjectives to test them in phase 2 among another group of adolescents and child athletes. As a second alteration, the SRSS was also modified with a sentence for each item to describe the different domains of recovery and stress. In each phase of data collection, the questionnaires were answered in a paper version. As the questionnaires were distributed among cooperating sports clubs, the researchers were not present during the process of completing them. The athletes were instructed by the persons who handed out the scales. As the psychometric parameters of the study will be compared with statistics of the original (e.g., dispersion measures, correlation coefficients, Cronbach's alpha, fit indices), the characteristics of the validation sample which were presented in the manual serve as reference values (**Table 1**).

INSTRUMENTS

The ARSS is a 32-item adjective list (e.g., "*rested*," "*tired*") that is rated from 0 ("*does not apply at all*") to 6 ("*fully applies*") (Kellmann et al., 2016; Kellmann and Kölling, 2019). Eight scales are then generated by summarizing four items which cover the *Recovery* dimension (*Physical Performance Capability*, *Mental Performance Capability*, *Emotional Balance*, *Overall Recovery*) and the *Stress* dimension (*Muscular Stress*, *Lack of Activation*, *Negative Emotional State*, *Overall Stress*). As depicted in **Table 2**, the original ARSS showed satisfactory discriminatory power of the items ($r_{it} = 0.51$ to 0.82) and, as shown in **Table 3**, good scale homogeneity ($\alpha = 0.76$ to 0.90) for the validation sample ($N = 574$, 21 ± 6.8 years). The factorial structure of the original was further supported via confirmatory factor analysis (Kellmann et al., 2016).

The SRSS is a derivation of the ARSS using the eight scales as items which are rated on the scale from 0 to 6

TABLE 1 | Overview of the studies, participants' characteristics and response patterns.

	Reference Sample ^a	Phase 1	Phase 2	Phase 2
Age group	Adults (≥ 16 years)	Adolescents (14–16 years)	Adolescents (14–16 years)	Children (10–13 years)
Questionnaires	Original ARSS, Original SRSS	Original ARSS + “I don’t understand,” Original SRSS	Modified ARSS + “I don’t understand,” Modified SRSS	Modified ARSS + “I don’t understand,” Modified SRSS
N (male, female)	574 (279, 293)	302 (183, 119)	438 (383, 55)	312 (232, 79)
Age ($M \pm SD$)	21.0 \pm 6.8	14–16	14.7 \pm 0.6	11.8 \pm 1.1
Complete item responses (n [%])		202 (66.9%)	263 (60.0%)	118 (37.8%)
Percentage of item non-responses		0.8%	1.1%	1.5%
Percentage of “I don’t understand” responses		0.8%	0.7%	4.5%

ARSS, Acute Recovery and Stress Scale; SRSS, Short Recovery and Stress Scale; ^a, reference values as published in the German manual (Kellmann et al., 2016, p. 27).

(Kellmann et al., 2016; Kellmann and Kölling, 2019). The four related adjectives are listed as descriptors of the items to provide examples of each construct. The *Short Recovery Scale* (Physical Performance Capability, Mental Performance Capability, Emotional Balance, and Overall Recovery) and the *Short Stress Scale* (Muscular Stress, Lack of Activation, Negative Emotional State, and Overall Stress) revealed acceptable discriminatory power ($r_{it} = 0.37$ to 0.66) as well as satisfactory scale homogeneity with $\alpha = 0.70$ and 0.76 , respectively, in the validation sample (Tables 4, 5, respectively).

In phase 1 of the study, each item of the ARSS could also be answered with the option “I don’t understand” next to the Likert-type rating scale, while the original SRSS was used.

In phase 2, one ARSS item each of four scales (i.e., *Emotional Balance*, *Muscular Stress*, *Negative Emotional State*, and *Overall Stress*) was modified with additional descriptions. These were added in brackets behind each item [e.g., “depressed (e.g., feeling down)”. Additionally, “I don’t understand” (next to the rating scale) could be ticked as well. For the SRSS, a sentence was added to each item (e.g., *Physical Performance Capability*: “I am full of energy and feel ready for training/competition”).

Statistical Analyses

In this publication, three statistical approaches were examined. The first step was a descriptive analysis using SPSS 25 to compare means and standard deviations separated by the different groups in each phase (i.e., adolescents and children). For single items, discriminatory power was assessed via corrected item-total correlations (r_{it}). Cronbach’s α was determined to analyze internal consistency of the scales. In addition, response patterns of each group were analyzed and the “I don’t understand” responses are displayed divided into the single age subgroups. Due to the missing demographic information in phase 1, the frequency of these responses (in percentage) is presented only for the participants of phase 2. Spearman correlation coefficients (r_s) were calculated to examine the relationship between the ARSS scales and the corresponding SRSS items. The descriptive values which were reported in the manual serve as benchmark for the present study.

The second approach was to perform confirmatory factor analyses (CFA) and, as a third approach, to examine

measurement invariance of the ARSS using R (Lavaan package version 0.6-3 by Rosseel, 2012; semTools package version 0.5-1 by Jorgensen et al., 2018). For this purpose, parts of the original data set of the validation sample was reanalyzed and fit indices were compared with the adolescent sample of phase 1. Only data of participants above 16 years were used from the validation sample to avoid an overlap of that age category. This reduced the sample size to $n = 442$ among the adults. Separate CFAs were performed among children and adolescents of the current data collection, as a modified questionnaire was used in phase 2. For the default model, inferential and descriptive fit statistics and the critical thresholds were selected [i.e., χ^2 with df and p -values, comparative fit index (CFI > 0.90), root mean square error of approximation (RMSEA < 0.08) \pm 90% confidence interval [90%-CI], standardized root mean residual (SRMR < 0.10)] as commonly reported in the literature (Hu and Bentler, 1999; Beauducel and Wittmann, 2005). Robust maximum likelihood estimators were applied to account for non-normal multivariate distribution. To examine measurement invariance across groups, i.e., if the recovery and stress models are comparable between the samples, multigroup CFA was conducted (Cheung and Rensvold, 2002). In a first step, the least restrictive model was estimated to analyze the same associations of items and factors, and the same number of factors (i.e., configural invariance). For the second model, all factor loadings were constrained to be invariant across groups to analyze metric invariance (i.e., weak measurement invariance). A third model tested whether the observed indicators show equal intercepts when regressed on the latent factors (i.e., scalar/strong invariance). Change of the fit indices were evaluated based on recommendations by Cheung and Rensvold (2002) for CFI (i.e., $\Delta CFI \leq -0.01$) and by Chen (2007) for changes of RMSEA (i.e., $\Delta RMSEA < 0.015$) and SRMR (i.e., $\Delta SRMR < 0.01$), whereas χ^2 -Difference test was not performed as both references do not recommend it and as the test provided by the semTools package is not applicable to the robust estimation method.

Due to the exploratory nature of the study, statistical analyses were performed only with those participants who provided complete responses. As a consequence, the sample sizes were reduced considerably for all groups (i.e., adolescents phase 1: $n = 202$, adolescents phase 2: $n = 263$, children phase 2: $n = 118$).

TABLE 2 | Means, standard deviations and item-total correlations of the ARSS.

		Reference Values ^a : Adults (<i>N</i> = 574)			Phase 1: Adolescents (<i>n</i> = 202)			Phase 2: Adolescents (<i>n</i> = 263)			Phase 2: Children (<i>n</i> = 118)		
		M	SD	<i>r</i> _{it}	M	SD	<i>r</i> _{it}	M	SD	<i>r</i> _{it}	M	SD	<i>r</i> _{it}
Recovery Dimension	Physical Performance Capability												
	Item 1	3.4	1.4	0.77	4.4	1.1	0.68	4.1	1.3	0.72	4.4	1.4	0.53
	Item 2	4.0	1.4	0.71	4.8	1.1	0.59	4.7	1.2	0.70	5.1	1.1	0.47
	Item 3	3.3	1.5	0.79	4.2	1.3	0.69	4.1	1.4	0.75	4.6	1.3	0.69
	Item 4	3.3	1.5	0.82	4.3	1.3	0.75	4.2	1.5	0.72	4.7	1.3	0.70
	Mental Performance Capability												
	Item 1	4.0	1.3	0.59	4.9	1.06	0.63	4.6	1.2	0.73	4.6	1.3	0.57
	Item 2	4.2	1.3	0.67	5.0	1.13	0.52	4.6	1.3	0.66	4.9	1.2	0.43
	Item 3	3.9	1.3	0.74	4.7	1.08	0.62	4.7	1.2	0.67	4.8	1.2	0.59
	Item 4	3.5	1.4	0.68	4.6	1.12	0.69	4.5	1.2	0.59	4.6	1.4	0.50
	Emotional Balance												
	Item 1	4.0	1.5	0.55	4.4	1.1	0.36	4.6	1.3	0.53	5.1	1.1	0.40
	Item 2	3.5	1.4	0.51	4.2	1.4	0.31	4.1	1.4	0.55	4.2	1.4	0.18
	Item 3	4.2	1.4	0.60	5.3	1.0	0.46	4.9	1.2	0.60	5.2	1.0	0.34
	Item 4	3.7	1.4	0.58	4.6	1.2	0.38	4.3	1.3	0.51	4.5	1.3	0.31
	Overall Recovery												
	Item 1	3.4	1.4	0.70	4.0	1.2	0.66	3.8	1.4	0.66	4.2	1.5	0.57
	Item 2	3.0	1.5	0.72	3.8	1.4	0.64	3.6	1.6	0.66	3.8	1.7	0.61
	Item 3	2.9	1.5	0.65	4.0	1.4	0.52	3.5	1.6	0.66	4.0	1.8	0.43
	Item 4	3.0	1.5	0.70	4.0	1.4	0.71	3.7	1.5	0.66	4.2	1.7	0.63

(Continued)

TABLE 2 | Continued

		Reference Values ^a : Adults (N = 574)			Phase 1: Adolescents (n = 202)			Phase 2: Adolescents (n = 263)			Phase 2: Children (n = 118)		
		M	SD	r _{it}	M	SD	r _{it}	M	SD	r _{it}	M	SD	r _{it}
Stress	Muscular Stress												
Dimension	Item 1	2.3	1.6	0.74	1.3	1.3	0.67	1.8	1.5	0.68	1.3	1.7	0.65
	Item 2	2.6	1.7	0.77	1.5	1.4	0.68	1.9	1.6	0.73	1.1	1.4	0.67
	<i>Item 3</i>	1.8	1.6	0.75	1.1	1.3	0.66	1.9	1.7	0.64	1.3	1.8	0.64
	Item 4	2.5	1.8	0.66	1.3	1.4	0.67	1.8	1.6	0.60	1.5	1.7	0.53
	Lack of Activation												
	Item 1	1.6	1.6	0.70	0.5	0.9	0.39	0.8	1.3	0.66	0.6	1.2	0.57
	Item 2	1.6	1.6	0.74	0.7	1.1	0.56	1.1	1.4	0.66	0.7	1.4	0.53
	Item 3	1.6	1.6	0.71	0.3	0.8	0.52	0.7	1.2	0.70	0.5	1.2	0.63
	Item 4	2.0	1.6	0.65	0.9	1.1	0.55	1.2	1.4	0.53	0.6	1.2	0.46
	Negative Emotional State												
	<i>Item 1</i>	1.8	1.7	0.59	0.7	1.1	0.34	1.0	1.4	0.53	0.7	1.3	0.51
	Item 2	2.2	1.7	0.56	0.9	1.1	0.53	1.4	1.4	0.55	1.1	1.3	0.54
	Item 3	1.7	1.6	0.66	0.5	0.9	0.61	0.9	1.3	0.69	0.8	1.3	0.54
	Item 4	2.1	1.7	0.61	1.0	1.4	0.51	1.6	1.6	0.45	1.5	1.7	0.36
	Overall Stress												
	<i>Item 1</i>	2.8	1.7	0.71	1.6	1.4	0.58	2.0	1.6	0.67	1.6	1.8	0.58
	Item 2	2.1	1.7	0.76	1.3	1.3	0.64	1.7	1.5	0.76	1.4	1.6	0.71
	Item 3	2.0	1.6	0.70	1.0	1.2	0.66	1.6	1.5	0.70	1.0	1.4	0.60
	Item 4	2.5	1.8	0.76	1.2	1.3	0.64	1.8	1.7	0.75	1.1	1.5	0.66

ARSS, Acute Recovery and Stress Scale; ^a, reference values as published in the German manual (Kellmann et al., 2016, p. 41); items in italic font indicate modification in phase 2.

TABLE 3 | Values of the internal consistency (Cronbach's α) of the ARSS scales across the groups.

		Reference Values ^a : Adults (N = 574)	Phase 1: Adolescents (n = 202)	Phase 2: Adolescents (n = 263)	Phase 2: Children (n = 118)
Recovery Dimension	Physical Performance Capability	0.90	0.84	0.87	0.78
	Mental Performance Capability	0.84	0.80	0.83	0.73
	<i>Emotional Balance</i>	0.76	0.59	0.75	0.50
	Overall Recovery	0.85	0.81	0.83	0.76
Stress Dimension	<i>Muscular Stress</i>	0.87	0.84	0.83	0.80
	Lack of Activation	0.86	0.71	0.81	0.75
	<i>Negative Emotional State</i>	0.79	0.70	0.75	0.69
	Overall Stress	0.88	0.81	0.87	0.81

ARSS, Acute Recovery and Stress Scale; ^a, reference values as published in the German manual (Kellmann et al., 2016, p. 41); scales in italic font contained one modified item in phase 2.

RESULTS

Response rates are depicted in **Table 1**. The children group provided the majority of missing data with 37.5% rating the ARSS items completely, while 4.5% of missing values were attributable to the “*I don't understand*” rating. Up to two thirds (phase 1) and more than half (phase 2) of the adolescent groups returned fully completed ARSS ratings, respectively. Less than 1% of missing data was accounted for by “*I don't understand*” answers. **Figure 1** shows the percentages of items from the *Recovery* dimension which the participants of phase 2 answered with “*I don't understand*.” **Figure 2** displays the percentages for the *Stress* dimension. Within the *Recovery* dimension, there was no item that was not understood by more than 20% of each age group, with the exception of one item in the scale *Mental Performance Capability* which the 10- (21.8%) and 11-year-olds (22.6%) did not understand. Within the *Stress* dimension, over 30% of the 10- and 27.4% of the 11-year-olds marked the same items of *Muscular Stress* and *Lack of Activation* as difficult to understand.

Table 2 shows means, standard deviations, and item-total correlations for the three groups of the study compared to the original data of the validation sample as reported in the manual (Kellmann et al., 2016). On the descriptive level, all of the *Recovery* scores were higher than the original data. Among the *Stress* dimension, scores of each group were apparently lower than in the validation sample. Values were rarely >2 . The standard deviations, on the other hand, appeared somewhat similar across the different groups. Item-total correlations ranged within comparable degrees between the groups. In the children group of phase 2, discriminatory power was rather weak (i.e., $r_{it} = 0.18$) for just one of the items that had been modified with an explanation. The remaining coefficients reached values above 0.30 across the different groups. **Table 3** compares the Cronbach's α values of the three groups with the original data of the validation sample. As these analyses were performed with complete responses, participants who marked “*I don't understand*” were not included. The validation sample of the original population presented the highest values throughout the scales, while the lowest values were found among the child athletes. *Emotional Balance*, in particular, revealed poor internal

consistency ($\alpha = 0.50$), while the remaining scales showed acceptable ranges of Cronbach's α . Among adolescents, however, increased values can be identified when comparing phase 1 and phase 2, where the scale contained one modified item (i.e., $\alpha = 0.59$ vs. $\alpha = 0.75$). Improved values were also identified for *Negative Emotional State* (i.e., $\alpha = 0.70$ vs. $\alpha = 0.75$) and for *Overall Stress* (i.e., $\alpha = 0.81$ vs. $\alpha = 0.87$).

Table 4 provides an overview of the SRSS's means, standard deviations and item-total correlations for the three groups and the validation sample. While means of the *Short Recovery Scale* appeared to be similar across the different samples, the validation sample presented higher scores among the *Short Stress Scale* compared to the study groups. Item-total correlations were above 0.30 across all groups. A comparison of Cronbach's α values of the *Short Recovery Scale* and the *Short Stress Scale* can be found in **Table 5**. For all groups, the *Short Recovery Scale* showed higher internal consistency than the validation sample, while Cronbach's α of the *Short Stress Scale* was higher in the validation sample compared to phase 1 adolescents and phase 2 children.

Spearman correlations between the ARSS scales and the corresponding SRSS items are shown in **Table 6**. Compared to the validation sample, similar or higher relationships within the *Recovery* dimension were identified across the three study groups. Within the *Stress* dimension, correlation coefficients were higher in the validation sample, whereas phase 2 adolescents revealed the highest correlation among *Overall Stress* of all groups. Strong correlations (i.e., $r_s \geq 0.70$) appeared only within the validation sample (*Lack of Activation*, *Negative Emotional State*) and within adolescents in phase 2 (*Physical Performance Capability*, *Overall Recovery*, *Overall Stress*).

The results of the CFA and the Multigroup CFA between adults and adolescents (phase 1) with the original ARSS are depicted in **Table 7**. Both groups revealed decent fit indices in the *Recovery* dimension. In addition, all of the fit indices were within the recommended thresholds in the three conditions of invariance analysis. The CFI did not change when comparing models of configural and metric invariance, while the change of the remaining fit indices did not exceed the suggested cut-off values. Regarding the *Stress* dimension, the initial model was acceptable despite the RMSEA values among adults ($\chi^2 = 400.80$,

TABLE 4 | Means, standard deviations and item-total correlations of the SRSS.

	Reference Values ^a : Adults (N = 574)				Phase 1: Adolescents (n = 199)				Phase 2: Adolescents (n = 261)				Phase 2: Children (n = 115)			
	M	SD	r _{it}		M	SD	r _{it}		M	SD	r _{it}		M	SD	r _{it}	
Recovery Dimension	PPC	4.2	1.5	0.62	4.4	1.1	0.64		4.4	1.2	0.63		4.9	1.1	0.46	
	MPC	4.4	1.3	0.51	4.7	1.0	0.54		4.7	1.1	0.60		4.9	1.0	0.61	
	EB	4.3	1.5	0.37	4.7	1.0	0.47		4.6	1.1	0.48		5.0	1.0	0.42	
	OR	3.7	1.6	0.53	4.1	1.3	0.61		3.9	1.4	0.63		4.4	1.3	0.60	
Stress Dimension	MS	3.1	1.8	0.49	1.6	1.4	0.58		2.3	1.6	0.61		1.6	1.8	0.67	
	LA	2.4	1.9	0.58	0.5	0.9	0.47		1.1	1.4	0.62		0.9	1.4	0.43	
	NES	2.4	1.9	0.48	0.9	1.3	0.39		1.2	1.4	0.55		0.9	1.2	0.45	
	OS	2.9	1.8	0.66	1.4	1.3	0.63		1.9	1.6	0.70		1.5	1.6	0.58	

SRSS, Short Recovery and Stress Scale; PPC, Physical Performance Capability; MPC, Mental Performance Capability; EB, Emotional Balance; OR, Overall Recovery; MS, Muscular Stress; LA, Lack of Activation; NES, Negative Emotional State; OS, Overall Stress; ^a, reference values as published in the German manual (Kellmann et al., 2016, p. 54).

$df = 98$, $p < 0.001$, CFI = 0.914, SRMR = 0.071, RMSEA = 0.091 [90%-CI = 0.082,0.101]). Model fit was slightly improved following modifications (i.e., covariation of measurement errors within *Lack of Activation*) which were then applied to the model of the adolescents who showed a better fit than the adults (Table 7). The analyses of measurement invariance showed good fits, despite the borderline RMSEA's upper limit of the 90%-CI in each step. The change of the fit indices was within the recommended thresholds, while the CFI increased by 0.001 in the model of metric invariance.

The results of the CFA and Multigroup CFA among both groups of phase 2 are displayed in Table 8. The initial *Recovery* model fit was acceptable despite the RMSEA values for the adolescents ($\chi^2 = 221.69$, $df = 98$, $p < 0.001$, CFI = 0.926, SRMR = 0.054, RMSEA = 0.077 [90%-CI = 0.063,0.090]), while it was overall somewhat poor for the children ($\chi^2 = 165.54$, $df = 98$, $p < 0.001$, CFI = 0.862, SRMR = 0.082, RMSEA = 0.085 [90%-CI = 0.062,0.106]). Table 8 shows the fit indices of the final model. Measurement invariance was found in each step, while the upper limit of the 90%-CI of RMSEA slightly exceeded the recommended threshold in each of the models. The modified *Stress* model of the adult sample was applied to both groups of phase 2. While the fit indices were just within an acceptable range for the adolescents ($\chi^2 = 178.98$, $df = 94$, $p < 0.001$, CFI = 0.947, SRMR = 0.058, RMSEA = 0.069 [90%-CI = 0.053,0.084]), it was considerably poorer among the children ($\chi^2 = 191.48$, $df = 94$, $p < 0.001$, CFI = 0.850, SRMR = 0.082, RMSEA = 0.106 [90%-CI = 0.084,0.127]). A second modification through covariance relationships within *Muscular Stress* led only to marginal improvements of the model in both groups (see Table 8). Nevertheless, measurement invariance was found with acceptable fit indices and changes of fit, despite the RMSEA's upper limit in each model.

DISCUSSION

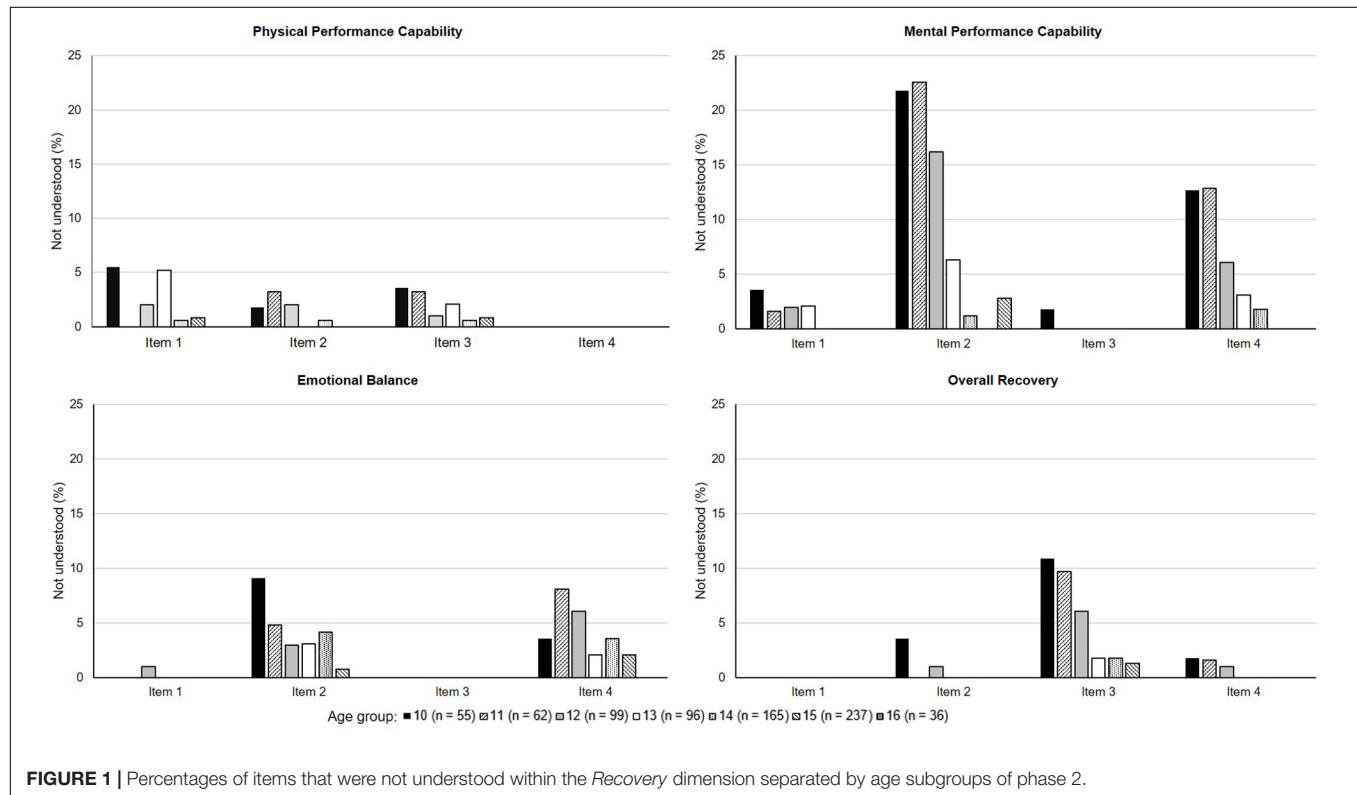
In the light of early specialization and intensified training among developing athletes, monitoring training load and the recovery-stress state has gained significance in youth sport as part of effective training management and health prevention. As it is questionable whether self-report measures which were developed for and with adults can be applied among younger athletes, it was the aim of the present study to examine psychometric properties of two established questionnaires in their original form as well as with initial modifications to approach the level of comprehension.

Overall, the results confirm that the understanding of the items is difficult among younger athletes. Although issues other than the lack of comprehensibility may be responsible for missing data, the majority of the children did not return complete ARSS ratings and most of the missing values were due to the “*I don't understand*” option. Specifically, the age group of 10- and 11-year-olds was identified to most frequently mark items as “*I don't understand*” across the dimensions of *Recovery* and *Stress*, with at least one item of *Mental Performance Capability*, *Muscular Stress*, *Lack of Activation*, and *Negative Emotional State*. The

TABLE 5 | Values of the internal consistency (Cronbach's α) of the *Short Recovery Scale* and the *Short Stress Scale* across the groups.

	Reference Values ^a : Adults (<i>N</i> = 574)	Phase 1: Adolescents (<i>n</i> = 199)	Phase 2: Adolescents (<i>n</i> = 261)	Phase 2: Children (<i>n</i> = 115)
Short Recovery Scale	0.70	0.76	0.78	0.73
Short Stress Scale	0.76	0.72	0.80	0.73

^a, reference values as published in the German manual (Kellmann et al., 2016, p. 54).

**FIGURE 1** | Percentages of items that were not understood within the *Recovery* dimension separated by age subgroups of phase 2.

descriptive statistics of the items served as another indicator of limited applicability as recovery items were consistently rated higher and stress items lower by the participants of the study groups compared to the validation sample. One reason could be that the younger athletes have either not yet developed the awareness and interpretation of their psychophysiological state or they have difficulties in expressing their current perception of recovery and stress in numerical graduations. This may explain the low internal consistency of the ARSS scale *Emotional Balance* and the low item-total correlation of item 2 (which corresponds to “feeling down”) among the children group in phase 2, although a description of that item was provided. Another explanation may be the number of response options. Borgers et al. (2004) found out that offering more than six options appeared to cause a decrease in scale reliability for children between 8 and 16 years.

Interestingly, modifying single items of the ARSS seemed to contribute to a better understanding among the adolescents, as improved Cronbach's α values were found comparing phase 1 to phase 2. In general, it is recommended that the instructions and questions of a questionnaire should be simple with clear and unambiguous wording. This is especially important when working with children between 8 and 11 years

(Borgers et al., 2000). As the ARSS only presents a list of adjectives, which may partly have ambiguous meanings, limited applicability seems to be induced among the children group and response bias may be an issue. Borgers et al. (2003) argue that children younger than 10 years might not be able to answer questionnaires reliably, which is expressed in their difficulties to apply the response options. Moreover, it seems that adolescents around the age of 11 may provide consistent answers which improves with age and may be stabilized around the age of 14 (Borgers et al., 2000).

The descriptive item statistics of the SRSS were comparable across the study groups, although the stress ratings were lower than in the validation sample. While the original SRSS revealed acceptable internal consistency among adolescents, which was quite similar to the validation sample, the modified SRSS indicated even higher values for adolescents as well as children. It has to be noted that the missing option to mark “I don't understand” is a limiting factor of the study design, and the issue of response bias cannot be ruled out. Nevertheless, the results suggest that the SRSS might be applicable for athletes from the age of 10 onward. The correlational patterns of the ARSS and the SRSS across the study groups imply that both

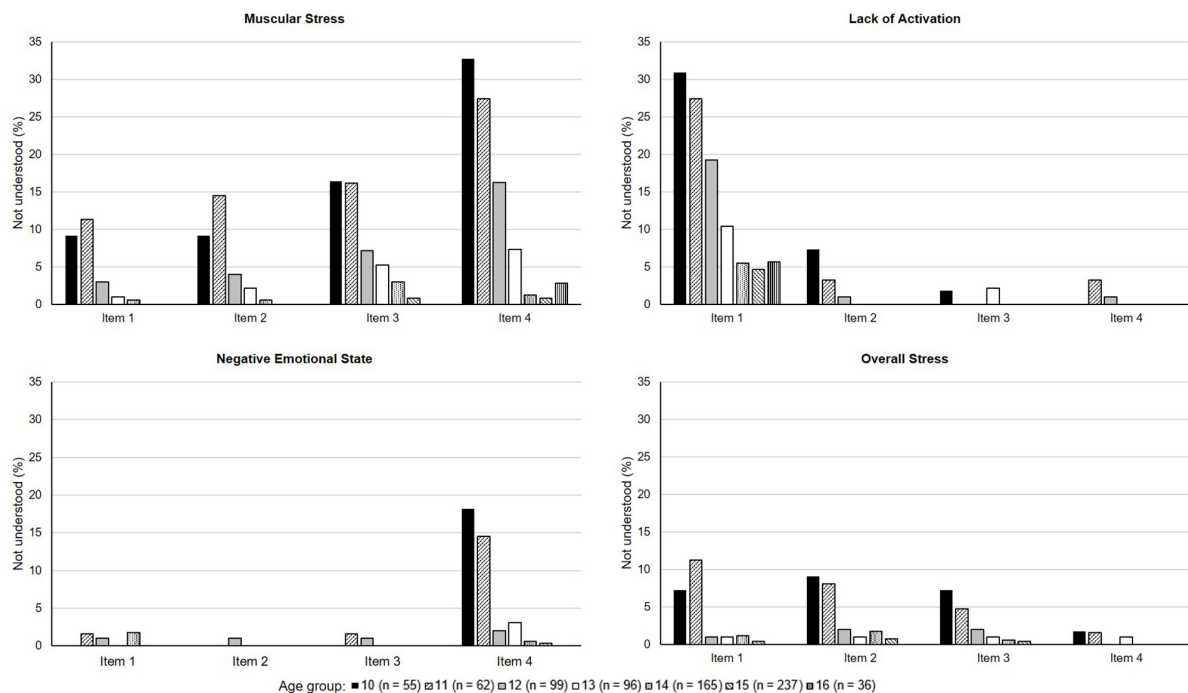


FIGURE 2 | Percentages of items that were not understood within the *Stress* dimension separated by age subgroups of phase 2.

TABLE 6 | Spearman correlations between the ARSS scales and corresponding SRSS items across the groups.

	Reference Values ^a : Adults (N = 574)	Phase 1: Adolescents (n = 199)	Phase 2: Adolescents (n = 261)	Phase 2: Children (n = 115)
Physical Performance Capability	0.62	0.66	0.76	0.58
Mental Performance Capability	0.49	0.58	0.65	0.63
Emotional Balance	0.46	0.56	0.60	0.45
Overall Recovery	0.64	0.66	0.72	0.65
Muscular Stress	0.69	0.65	0.68	0.55
Lack of Activation	0.74	0.46	0.63	0.52
Negative Emotional State	0.70	0.56	0.62	0.52
Overall Stress	0.67	0.65	0.72	0.63

ARSS, *Acute Recovery and Stress Scale*; SRSS, *Short Recovery and Stress Scale*; ^a, reference values as published in the German manual (Kellmann et al., 2016, p. 63); all correlations are significant on the level $p < 0.001$.

assess the recovery-stress state, but they can be considered as independent questionnaires, as the coefficients did not reveal perfect correlations. This finding was also present across different data collections with the original tools (Kellmann et al., 2016; Nässi et al., 2017a; Kellmann and Kölling, 2019).

Multigroup CFA was performed to examine if the ARSS is measuring the same construct across groups. As a first step, the models need to show a decent model fit in each group separately (Cheung and Rensvold, 2002). This was found for the *Recovery* model in every group. Considering the rather borderline values of the RMSEA's 90%-CI across groups, the *Stress* construct might be critically discussed. Especially among the children, the model seems to fit somewhat poorly to the data. However, the descriptive rather than normative nature of the fit indices and

their cut-offs has to be pointed out, so that there is actually no consensus definition of an ideal fit (Worthington and Whittaker, 2006). At the level of configural invariance, the models of the adults and adolescents of phase 1 as well as those of phase 2 were combined. In both group comparisons, the model fit indicates that the basic factor structure can be considered equal among the groups. Thus, the original items of the ARSS seem to assess the same pattern of *Recovery* and *Stress* of participants between 14 and 16 years as of adults. The same conclusion can be drawn for the modified ARSS. Weak measurement invariance can be assumed when the factor loadings are equivalent between groups. The model fit did not decrease out of the recommended range in either condition (i.e., original ARSS, modified ARSS) nor in the dimensions (i.e., *Recovery*, *Stress*). Even the third

TABLE 7 | Multigroup confirmatory factor analysis with the adult sample ($n = 442$) and adolescents of phase 1 ($n = 199$).

	Model	χ^2	df	p	CFI	SRMR	RMSEA	90% CI	Δ CFI	Δ SRMR	Δ RMSEA
Recovery Dimension	Adults	253.37	98	<0.001	0.944	0.047	0.067	0.057 0.077	—/—	—/—	—/—
	Adolescents (Phase 1)	152.57	98	<0.001	0.950	0.054	0.058	0.039 0.075	—/—	—/—	—/—
	Configural Invariance	408.58	196	<0.001	0.950	0.047	0.064	0.056 0.073	—/—	—/—	—/—
	Metric Invariance	421.97	208	<0.001	0.950	0.049	0.062	0.054 0.071	0.000	0.002	−0.002
	Scalar Invariance	473.28	220	<0.001	0.941	0.056	0.066	0.058 0.074	−0.009	0.007	0.004
Stress Dimension	Adults	321.82	94	<0.001	0.935	0.063	0.081	0.072 0.091	—/—	—/—	—/—
	Adolescents (Phase 1)	164.19	94	<0.001	0.930	0.063	0.070	0.051 0.087	—/—	—/—	—/—
	Configural Invariance	480.52	188	<0.001	0.934	0.059	0.078	0.069 0.086	—/—	—/—	—/—
	Metric Invariance	473.33	200	<0.001	0.935	0.065	0.075	0.066 0.084	0.001	0.006	−0.003
	Scalar Invariance	525.59	212	<0.001	0.925	0.073	0.078	0.070 0.086	−0.010	0.008	0.003

CFI, Comparative Fit Index; SRMR, Standardized Root Mean Residual; RMSEA, Root Mean Error of Approximation; CI, Confidence Interval; specifications of the final Stress model are identical between groups.

TABLE 8 | Multigroup confirmatory factor analysis among phase 2 participants with adolescents ($n = 261$) and children ($n = 115$).

	Model	χ^2	df	p	CFI	SRMR	RMSEA	90% CI	Δ CFI	Δ SRMR	Δ RMSEA
Recovery Dimension	Adolescents (Phase 2)	198.53	94	<0.001	0.938	0.051	0.072	0.058 0.086	—/—	—/—	—/—
	Children	158.71	94	<0.001	0.870	0.078	0.084	0.061 0.106	—/—	—/—	—/—
	Configural Invariance	357.78	188	<0.001	0.923	0.056	0.076	0.064 0.088	—/—	—/—	—/—
	Metric Invariance	368.82	200	<0.001	0.923	0.061	0.073	0.061 0.085	0.000	0.005	−0.003
	Scalar Invariance	387.42	212	<0.001	0.921	0.062	0.072	0.061 0.083	−0.002	0.001	−0.001
Stress Dimension	Adolescents (Phase 2)	166.44	91	<0.001	0.952	0.055	0.066	0.050 0.082	—/—	—/—	—/—
	Children	174.90	91	<0.001	0.870	0.082	0.100	0.078 0.123	—/—	—/—	—/—
	Configural Invariance	335.23	182	<0.001	0.931	0.060	0.077	0.064 0.090	—/—	—/—	—/—
	Metric Invariance	351.77	194	<0.001	0.930	0.066	0.076	0.063 0.088	−0.001	0.006	−0.001
	Scalar Invariance	371.50	206	<0.001	0.928	0.067	0.075	0.062 0.087	−0.002	0.001	−0.001

CFI, Comparative Fit Index; SRMR, Standardized Root Mean Residual; RMSEA, Root Mean Error of Approximation; CI, Confidence Interval; specifications of the final Stress model are identical between groups.

model seems to provide acceptable fit which indicates strong measurement invariance that would allow for the comparison of the latent mean between groups. Nevertheless, in the present study, data were collected in a range of naturalistic situations which could not be controlled. As the underlying construct of acute recovery and stress represent a state that is assumed to change over the course of time (and in response to stress or recovery stimuli), the within-individual stability of the construct needs to be analyzed over time.

Considering the results and initial implications, coaches and practitioners need to appreciate that the period of adolescence is critical for the maturation of neurobiological processes, among others, which may contribute to cognitive and affective behavior (Yurgelun-Todd, 2007). Moreover, Blakemore and Choudhury (2006) point out the sensitivity of the brain to experiential input in terms of executive function and social cognition due to the synaptic reorganization. The developing brain as well as behavioral and cognitive systems mature along different timetables which causes heightened vulnerability in adolescents (Steinberg, 2005). In terms of cognitive efficiency in response to emotionally related stimuli, McGivern et al. (2002) found a decrement at the onset of puberty. This may support the rather poor statistics of the emotionally related scales in the

present study. While it may be possible in surveys to use standardized questionnaires that are similar to those for adults among the age group of 11 to 15–16 years (Borgers et al., 2000), precautions should be considered. As the present study revealed, it is important to test the questionnaires among the target populations and provide modifications to enhance reliable responses. In some cases, it may be sufficient to explain the questionnaire when handed out for the first time and to be available for further questions. Otherwise, items or scales that have been known as being problematic should rather not be interpreted and analyzed at all.

Limitations and Future Directions

Some limitations of the study, especially regarding phase 1, need to be commented on. As the anonymity of the athletes in phase 1 was the priority, valuable information could not be assessed and the analyses were limited to the overall group level. Moreover, pre- post measurements to examine improvements of understanding within the individuals were not possible. On the other hand, the high performance level of the phase 1 group was an advantage, as the participants were familiar with training and exercise which may facilitate their general understanding of the topic of the questionnaires.

Furthermore, it was the aim to explore the psychometric properties among those who provided complete responses which caused a considerable reduction of the sample sizes. Appropriate statistical measures, such as multiple imputation, may be considered in future analyses to adjust for missing item scores. Although it may be of minor relevance at the level of the items' understanding, team sport athletes were somewhat overrepresented. Therefore, the present results should be considered as preliminary investigation in this area. Moreover, it seems worthwhile to analyze the psychometric properties for each age group to identify possible cut-offs which differentiate between the applicability of the original and the need for modified versions. Therefore, larger sample sizes should be recruited in future studies. This may further allow for separate gender analyses, since female athletes were underrepresented in this study. As the participants gave their responses at different times and various settings (e.g., in a training camp, before or after an intensive training), the sensitivity to change needs to be investigated systematically once the modifications are completed.

In the present study, a top-down approach was chosen to evaluate the recovery-stress model that was established for adults among the younger clientele. As suggested by Ravens-Sieberer and Bullinger (1998), a mixture of top-down and bottom-up methods is preferable. With the help of bottom-up tactics, the children's concepts of recovery and stress and perceptions of their psychophysiological response to training as well as relevant recovery and stress dimensions may be considered.

ETHICS STATEMENT

This study was carried out in accordance with the recommendations of the ethical committee of the Faculty

of Psychology at the Ruhr University Bochum with written informed consent from all subjects. All subjects gave written informed consent in accordance with the Declaration of Helsinki. The protocol was approved by the ethical committee of the Faculty of Psychology at the Ruhr University Bochum (application number 308).

AUTHOR CONTRIBUTIONS

SK planned and designed the study, conducted measurements, analyzed the data, and prepared the manuscript. AF, TM, and MP edited the manuscript. MK planned and designed the study and edited the manuscript. All authors read and approved the submitted version.

FUNDING

The current study was funded by the German Federal Institute of Sport Science. The research was realized in the project "REGman—Optimization of Training and Competition: Management of Regeneration in Elite Sports" (Grant Number IIA1-081901/12-20). We acknowledge support by the DFG Open Access Publication Funds of the Ruhr University Bochum.

ACKNOWLEDGMENTS

The authors would like to thank all people who assisted in the recruitment and data collection. Special thanks goes to the participants for providing valuable information.

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Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Serving Patterns of Women's Badminton Medalists in the Rio 2016 Olympic Games

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OPEN ACCESS

Edited by:

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Specialty section:

This article was submitted to
Movement Science and Sport
Psychology,
a section of the journal
Frontiers in Psychology

Received: 23 August 2019

Accepted: 20 January 2020

Published: 05 February 2020

Citation:

Gómez-Ruano M-Á, Cid A,
Rivas F and Ruiz L-M (2020) Serving
Patterns of Women's Badminton
Medalists in the Rio 2016 Olympic
Games. *Front. Psychol.* 11:136.
doi: 10.3389/fpsyg.2020.00136

The aim of the present study was to describe and identify the serving performance profiles of medalists during an elite women's badminton tournament taking notational and temporal variables into account. The sample was composed of the 14 matches ($n = 1,052$ rallies) played by the three medalists during the 2016 women's singles Olympic Games badminton event (Rio, Brazil). The independent variable studied was serving player (medalist/opponent); while the dependent variables were related to notational analysis: serve type, set, and point won by the server/receiver; and the time-related variables: number of strokes per rally, rally time, rest time, and frequency of strokes. The main results showed that: (i) temporal parameters were similar for total match duration but shorter for rally time, and longer for rest time and with more strokes per rally than found in previous research; (ii) the serve effectiveness showed neutral values when analyzing serving by all the players, medalists, and opponents (around 50%); (iii) the two-step cluster analysis identified how successful players used the serve when playing short rallies with backhand short and flick serves (cluster 1), and forehand long serves (cluster 2); and during long rallies with the use of the backhand short serve, forehand short serve and forehand long serve (cluster 3). On the other hand, medalists and their opponents used forehand long serves during set 1 with durations of 8.80 s (cluster 5); and the opponents showed an independent performance using the forehand short serve during sets 1 and 2 (cluster 4); and (iv) the classification tree analysis (Exhaustive CHAID) identified the importance of different serving patterns with the gold medal player using more backhand and forehand flick serves, and the main use of backhand short serves during sets 1 and 2 in all the tournament stages. The bronze medalist used more forehand long serves during all sets, and the silver medalist showed a mixed performance of serves using the forehand short serve, the backhand short serve and the forehand long serve. The current findings may help coaches and players to manage different serving and playing patterns during training and matches according to the serve and rally requirements.

Keywords: racket sports, performance indicators, female, notational analysis, elite player performance

INTRODUCTION

Badminton is a sport characterized by a combination of speed, endurance, and power displayed during high intensity and short-duration actions that have short rest intervals between points (Laffaye et al., 2015). In particular, it is complex and dynamic and the player tries to produce quick responses (i.e., decision-making) disrupting his/her opponent's actions, and then, win the point (Chow et al., 2014). In fact, the demands placed on a badminton player are focused on tactical, technical, and temporal adaptations to the dynamics of each context (e.g., behaviors and tactics when serving or receiving, set intervals before and after point 11, playing long or short rallies, the different importance of sets 1, 2 and 3, etc.) during the match (Abián et al., 2014; Laffaye et al., 2015). Under the current badminton regulations (i.e., scoring system) the players play more aggressively using different tactics and a higher frequency of strokes during longer matches (e.g., faster game play with more points to be played, greater variations of rally time and rest time, and more unpredictability during the intervals and sets) (Laffaye et al., 2015).

Based on this rationale, the available research on badminton (Cabello et al., 2004; Abián et al., 2014; Laffaye et al., 2015) has focused its attention on notational analysis and the temporal structure during elite competitions. On the one hand, notational analysis has been widely used to investigate the individual's performance in badminton and racket sports providing relevant information about players' technical and tactical behaviors during matches and rallies such as type of serve, serve effectiveness, point outcome, number of strokes used, type of shots, effectiveness when serving or receiving, etc. (Lees, 2003; Abdullahi and Coetzee, 2017). On the other hand, the temporal structure of this sport has complemented the notational analysis with relevant information about the game/match duration, rally time, rest time, density of play, number of strokes per rally or time between strokes (Phomsoupha and Laffaye, 2015; Laffaye et al., 2015). This information is extremely important due to its high applicability to real contexts when training and playing matches, setting the appropriate loads or task constraints according to the requirements of competition (Chiminazzo et al., 2018).

Specifically, these performance analyses in elite badminton have extensively studied differences according to the sex of the players, the stage or phase of competition (e.g., group or knockout stages), the final outcome of the match (i.e., winning and losing), or the players quality/strength such as the best or worst players (Barreira et al., 2016; Chiminazzo et al., 2018). However, the analysis of how successful players (i.e., medalists) perform and score points when serving at the elite level in badminton is still inconclusive. This approach has been largely studied from the perspective of developing sporting talent, analyzing the athlete (e.g., anthropometric and physiological factors, genetics, birthdate, motivation, or psychological skills), the environment (e.g., birthplace, parents, family, or coaches support), the importance of practice or training (e.g., early specialization or the volume of training), and other potential factors (e.g., injuries, recovery, or socio-economic status) of medalists (Sarkar et al., 2015; Rees et al., 2016). These characteristics showed by successful athletes reflect a determined focus when training

and competing (i.e., mastering key technical, tactical, and psychological skills) with a direct impact on their performance (Starkes and Ericsson, 2003). Despite this approach of scientific research, specific performance analysis (i.e., technical, tactical, or temporal) of medalists has been developed in individual sports such as running or swimming events (Hollings et al., 2014; Mytton et al., 2015) with concluding remarks of performance features during competition that characterize their success (e.g., better performances for medalists during the last part of running or swimming races or better adaptation to different paces according to race contexts). In racket sports successful players use different effective serving and playing patterns that make it possible to defeat their opponents during rallies and matches (Chiminazzo et al., 2018).

In particular, the serve in badminton is the first stroke of the point and plays a key tactical role as it is not affected by any previous action by the opponent. The serve is thus one of the most used strokes in badminton (Abdullahi and Coetzee, 2017; Chiminazzo et al., 2018) that needs to be under the full control of the server in order to potentially gain any spatial and temporal (e.g., short and long serves) advantage over the receiver during the consecutive strokes played in each point (Pearce, 2002; Alcock and Cable, 2009). However, according to Bialik (2016) the serve does not represent an advantage in women's badminton singles where only 55% of the points were won when serving. The serve can then be considered as a way to start to play the point but not a key stroke to win direct points. Therefore, the analysis of actions performed by successful players monitoring serve type, serve effectiveness and playing patterns during the rallies may reflect their individual performance features that lead to success. Thus, the specific study of key performance indicators in elite badminton may define the characteristics of successful players when serving, and then reflect the performance profiles during their matches according to some key notational (i.e., type of serve or serve effectiveness) and temporal (i.e., number of strokes, rally time, rest time, or frequency of strokes) variables. Therefore, the aim of the present study was to describe and identify the serving performance profiles of successful players (medalists) during an elite women's badminton tournament taking notational and temporal variables into account. It was hypothesized that successful players use different serving and playing patterns that imply quicker and more difficult technical-tactical actions to score points during the matches.

MATERIALS AND METHODS

Sample

The sample was composed of 14 matches (Group stage, Quarter-final, Semi-finals, and Final matches) played by the three medalists (Gold, Silver, and Bronze) from the 2016 women's singles Olympic Games badminton event (Rio, Brazil). Only one match was excluded from the sample (Bronze medal match) due to the fact that one player was injured and did not play the match. The final sample included the analysis of 1,052 rallies played by the three medalists. All matches were publicly available on TV and the data was used with the approval of the Universidad

Politécnica de Madrid Ethics Committee and in accordance with the European Data Protection Law.

Procedure

The analyses were carried out using an observation tool in a video analysis program (Dartfish, Friburgo, Switzerland). Four trained observers (graduates in Sports Sciences with 10 years' experience as badminton coaches) collected the variables with good and very good inter and intra-rater reliability values (Kappa: >0.81 ; correlation coefficient $r > 0.86$; ICC: > 0.85 , and standard error of measurement: < 0.46) (Altman, 1991; Hopkins, 2000).

The independent variable studied was the serving player (medalist/opponent); while the dependent variables were related to *notational analysis*: serve type (forehand short serve, forehand long serve, forehand flick, backhand short serve, and backhand flick), set (1st, 2nd, or 3rd), and point won by the server or the receiver; and the *temporal structure* variables: number of strokes per rally, rally time (time in s of the rally duration between the serve and the end of the point), rest time (time in s between the end of the point and the serve action of the next immediate point), and frequency of strokes (the time in s between opposing players' strokes).

Statistical Analysis

Firstly, descriptive analyses (median and lower/upper quartiles) were run for temporal parameters (total match and set duration, number of strokes per rally, rally time, rest time, and frequency of strokes) during all matches and each set (1st, 2nd, and 3rd) in order to show the measures of centrality of time-related demands during the championship.

Secondly, the crosstabs commands were used to study the relationships (Pearson's Chi-square test) between the point won when serving or receiving and the type of serve used by the server (medalist or opponent). Fisher's exact test was applied when the Expected Frequency Distribution was lower than 5 or the count of cases in one cell was lower or equal to 5 (Field, 2013). In order to estimate Effect sizes (ES) Cramer's V test was used considering the following range values: 0.10 = small effect, 0.30 = medium effect, and 0.50 = large effect (Volker, 2006).

Thirdly, in order to analyze the variables that best explain the players' performance when serving, the sample was grouped into different clusters that described the specificities of rallies played by the medalists and their opponents during the tournament. Then, a two-step cluster analysis was run considering the variables: type of serve, set, serving player, if the point was won by the server or the receiver, and temporal parameters (rally time, rest time, number of strokes, and frequency of strokes). The clustering technique automatically (log-likelihood distance measure) determined the best number of clusters (types of rallies played) using the Schwartz's Bayesian Information Criterion (BIC). The model obtained was good with a Silhouette measure value of 0.5. Additionally, the clusters were differentiated using the Kruskal-Wallis H non-parametric test for numerical variables (temporal parameters: rally time, rest time, number of strokes, and frequency of strokes). The *post hoc* pairwise comparisons (Dunn's test with the Bonferroni's correction) were run to identify differences among clusters. The crosstabs

command (Pearson's Chi-square test) was used to differentiate the categorical variables (type of serve, set, medalist condition, and point won) among clusters.

Lastly, the Exhaustive CHAID (Chi-squared automatic interaction detection) classification tree analysis was used to determine the differences between the performance playing patterns of the three medalists according to the temporal (rally time, rest time, number of strokes, and frequency of strokes) and notational (type of serve, set, set interval, round, and point won by the server, or receiver) variables. This model made it possible to split the medalists' sample according to nodes (sub-groups) based on the impact of the medalist (gold-, silver-, or bronze-medal) condition. The algorithm used considers a nominal dependent variable and nominal and numerical independent variables. The Chi-square test identifies the relationships between independent variables, and then finds the best predictors (temporal and notational variables) that most influence the dependent variable (Schnell et al., 2013). The algorithm used completes three steps on each node of the root (merging, splitting, and stopping) in order to find the predictors that exert the most influence on the dependent variable. The exhaustive CHAID assesses all splitting possibilities for each independent variable, and the merging step improves the searching procedure to find (and merge) those similar pairs until only a single pair remains. The model provides a graphical presentation of the final tree (hierarchical tree, see **Figure 1**) where the impact of each independent variable makes it possible to split the root node (node 0) into branches with n descendent nodes. The tree continues descending with each branch that assesses the remaining significant independent variables (improving the search of splitting nodes). The terminal nodes are established when no further split can be made (Schnell et al., 2013).

The statistical specifications considered in this model were: (i) $p < 0.05$; (ii) Pearson's Chi-square test was used to check relationships among independent variables; (iii) the maximum number of iterations was 100; (iv) the minimum change in expected cell frequencies was 0.001; (v) the Bonferroni adjustment was used; and (vi) a maximum of three levels were considered in the tree model. Lastly, the risk of misclassification was estimated as a measure of model reliability (Schnell et al., 2013). All statistical analyses were performed using the statistical software IBM SPSS statistics for Windows, version 22.0 (IBM, Corp., Armonk, NY, United States).

RESULTS

Table 1 shows the descriptive results of temporal variables (median, lower, and upper quartile) during the matches played by medalists during the Tournament.

The distribution of type of serve for points won by the server or receiver is presented in **Table 2** (percentage and case numbers). The results showed that 49.6% of the points were won by the server and the type of serve was not significantly ($p > 0.05$) associated to winning the point when serving. The analysis splitting by medalist and opponents (see **Table 2**) showed a significant relationship for medalists between type of serve

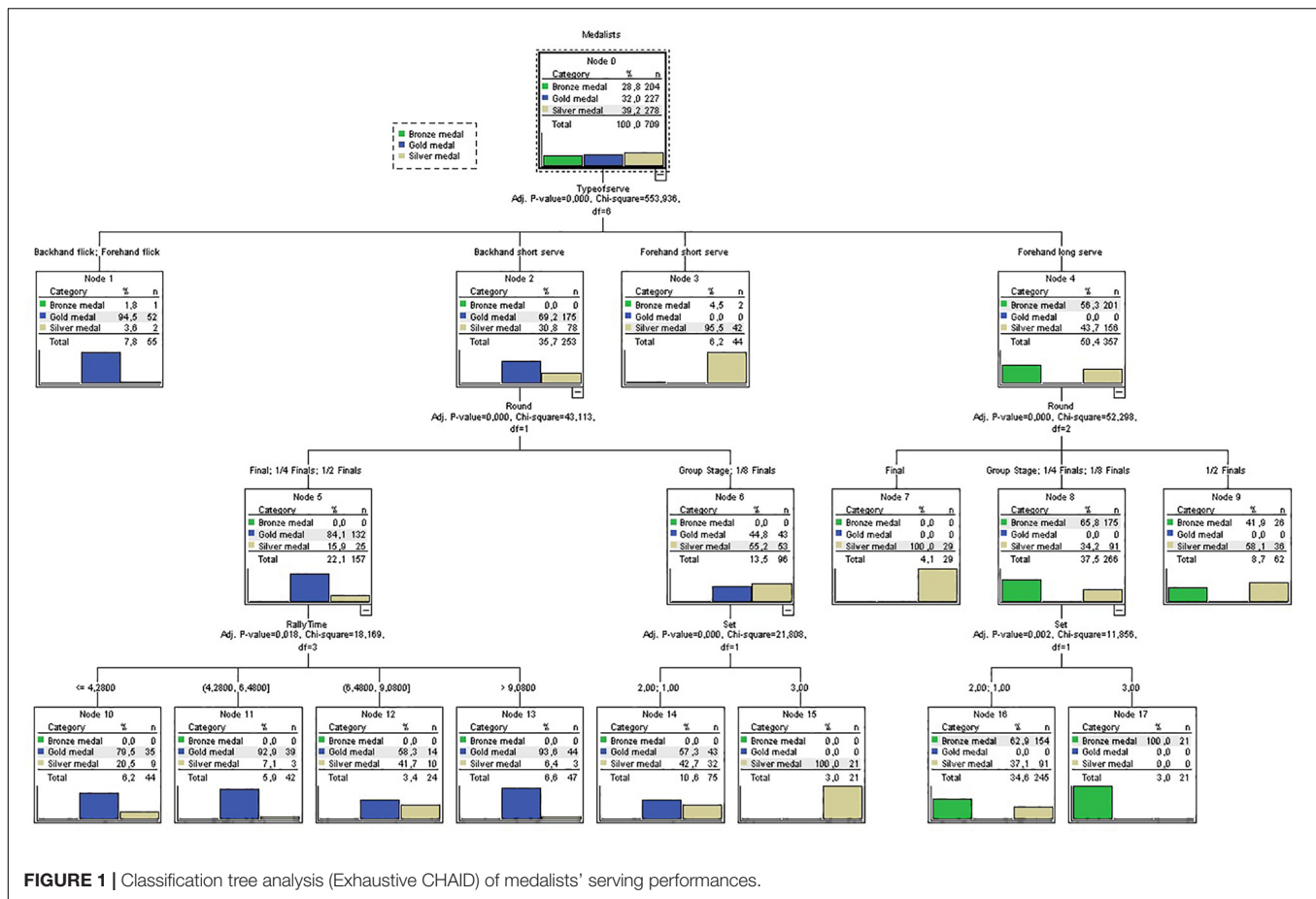


FIGURE 1 | Classification tree analysis (Exhaustive CHAID) of medalists' serving performances.

and points won when they serve with the backhand flick and backhand short serve (AR = 4.3 and 5.1, respectively). In addition, the results for opponents (see Table 2) showed significant relationships between type of serve and winning the point serving when using the forehand flick and forehand short serve (AR = 3.0 and 8.2, respectively).

The clustering technique (two-step cluster, see Table 3) identified five different clusters (rallies) according to the notational (type of serve, set, serving player, point won by the server, or receiver) and temporal (rally time, rest time, number of strokes, and frequency of strokes) variables. The most frequent rally was cluster 3 (27.3%, played mainly by the medalists using the backhand short serve, and forehand long and short serves, during sets 2 and 3, rally duration of 12.3 s, frequency of strokes of 0.98, and greater effectiveness for the server: 56.9%), cluster 4 (22.3%, played by the opponents using the forehand flick and forehand short serve, and backhand short serve, during sets 1 and 2, with rally times of 6.63 s, frequency of strokes of 0.92, and neutral effectiveness for the server: 51.0%), cluster 5 (21.5%, played by both players using the forehand long serve, during set 1, with rally times of 8.80 s, frequency of strokes of 1.10, and 49.0% of effectiveness for the server), cluster 1 (15.2%, played by the medalists using the backhand short serve, backhand flick and forehand short serve, during sets 1 and 2, rally times of 6.27 s, frequencies of strokes of 0.92, and 49.5% of effectiveness

for the server), and cluster 2 (13.7%, played mainly by medalists using all serves (except the backhand flick), during set 2, with rally durations of 7.87 s, frequencies of strokes of 1.11, and lower effectiveness for the server: 44.7%).

Significant differences were identified among clusters for type of serve, serving player, and set ($p < 0.05$; see Table 4). No significant ($p > 0.05$) relationships were identified among clusters for point won by the server of the receiver (Table 3). Additionally, the time-related variables showed significant differences among clusters for rally time, rest time, frequency of strokes, and number of strokes (all $p < 0.01$). The pairwise comparisons showed clear differences among rallies (clusters) with cluster 3 as the longest rally and clusters 1 and 4 as the shortest and quickest ones (see Table 4).

The classification tree model was run to identify specific playing performances of medalists when monitoring for temporal (number of strokes, rally time, rest time, and frequency of strokes) and notational (tournament round, set, interval, type of service, and outcome) variables in the statistical analysis. The results showed only four significant variables (type of serve, round, set, and rally time) when classifying medalists' performance (three-stage tree). The following factors led to 17 nodes (12 final nodes) of contrasting groups classifying medalists mainly by type of serve (level 1), round (level 2) and rally time and set (level 3). Figure 1 shows the categories for predictor variable (medalists:

TABLE 1 | Descriptive results (median, lower, and upper quartile) for match and set temporal parameters during the matches studied.

	Median	Quartile	
		Lower	Upper
Match duration (min)	41.8	38.3	53.5
Set 1 duration (min)	22.1	17.7	24.8
Set 2 duration (min)	20.7	17.9	23.3
Set 3 duration (min)	41.8	38.3	53.5
Rally time (s)	7.87	4.74	12.6
Rest time (s)	22.1	17.1	29.6
Strokes per rally (n)	8.0	5.0	13.0
Frequency (s)	1.01	0.90	1.12
Set 1			
Rally time (s)	7.78	4.93	12.6
Rest time (s)	21.7	17.0	29.7
Strokes per rally (n)	7.50	5.0	13.0
Frequency (s)	1.02	0.91	1.14
Set 2			
Rally time (s)	7.83	4.37	12.5
Rest time (s)	21.6	16.6	29.3
Strokes per rally (n)	8.00	4.0	13.0
Frequency (s)	1.01	0.90	1.12
Set 3			
Rally time (s)	8.34	5.07	15.0
Rest time (s)	26.0	20.5	37.1
Strokes per rally (n)	9.0	5.0	15.0
Frequency (s)	0.97	0.88	1.07

gold-, silver-, and bronze-medal) and also the 17 nodes defined by the classification tree model.

Level 1 (root node) is split by the type of serve showing the gold medalist using more backhand flicks and forehand flicks (node 1: 94.5%; $n = 52$) and backhand short serves (node 2: 69.2%; $n = 175$). The silver medalist used more forehand short serves (node 3: 95.5%; $n = 42$) and the bronze medalist used more forehand long serves (node 4: 56.4%; $n = 201$). Level 2 showed the importance of the round for the backhand short serve (from node 2) where there was a greater use of this serve during the final, semi-finals, and quarter-finals by the gold medalist (node 5: 84.1%; $n = 132$) and during the group stage and round of 16 for the silver medalist (node 6: 55.2%; $n = 53$). In addition, level 2 showed the importance of the round for the forehand long serve (from node 4) where the silver medalist used this serve more often during the final and semi-finals (nodes 7: 100%; $n = 29$; and node 9: 58.1%; $n = 36$, respectively) and the bronze medalist during the group stage, quarter-final, and round of 16 (node 8: 65.8%; $n = 175$).

Level 3 showed the importance of rally time when using the backhand short serve during the final, semi-finals and quarter-finals by the gold medalist (from node 5), the set when using the backhand short serve during group stage and round of 16 (from node 6), and the set when using the forehand long serve during the group stage, quarter-final and round of 16 (from node 8). On the one hand, the importance of rally time showed greater use

of the backhand short serve by the gold medalist during rallies with time durations ranged between 4.28 and 6.48 s (node 11: 92.9%; $n = 39$) and longer than 9.08 s (node 13: 93.6%; $n = 44$). In addition, the use of the backhand short serve was greater by the gold medalist during sets 1 and 2 of the group stage, quarter-final and round of 16 (node 14: 57.3%; $n = 43$), and greater by the silver medalist during set 3 (node 15: 100%; $n = 21$). On the other hand, the significant effect of the set for the bronze medalist showed more actions when using forehand long serves during the group stage, quarter-final, and round of 16 matches in set 3 (node 17: 100%; $n = 21$) and sets 1 and 2 (node 16: 62.9%; $n = 154$) than the silver medalist. The classification tree model explained 74.8% of total variance after cross-validation analysis.

DISCUSSION

The aim of the current study was to describe and identify the serving performance profiles of rallies played by successful players (medalists) when taking notational and temporal variables into account during the women's badminton Olympic Games (Rio, 2016). As was argued successful players (medalists) performed differently when playing rallies using a wider range of serve types than their opponents with a different impact on their points and match behaviors as identified in the two-step cluster and decision tree analyses (Starkes and Ericsson, 2003; Rees et al., 2016). These main findings may reflect a better technical and tactical preparation to serve and play the point managing fatigue, next point preparation or stress/pressure during the set/match (Taylor et al., 2008; Barreira et al., 2016; Chiminazzo et al., 2018).

Temporal Analysis

The results of temporal parameters of matches played by medalists during the tournament showed similar total match duration to previous research that studied international badminton tournaments (Cabello et al., 2004; Torres-Luque et al., 2019). However, the rally time of current matches showed shorter durations (7.87 compared with values of 9–10 s) than previous studies. This finding reflects the fact that during the tournament successful players showed the same total time duration but played rallies at a higher intensity (8 strokes per rally and frequency of strokes of 1.01) and with longer rest time periods. This general trend is in agreement with Phomsoupha and Laffaye (2015) and Chiminazzo et al. (2018) who described a new temporal structure of elite badminton with high-intensity and short-duration intermittent actions that require longer rest periods. Along these lines, successful players may reflect a better adaptation to playing quick actions due to a better mastery of technical, tactical, and psychological abilities with a direct impact on match behaviors (Starkes and Ericsson, 2003). Additionally, medalists played sets 1 and 2 with similar time duration but shorter rally time, more strokes per rally, shorter frequency of strokes, and longer rest time periods than presented in the available research (Torres-Luque et al., 2019). Specifically, these results suggest a better use by medalists of stoppages, end of points and breaks for managing pressure, fatigue, and recovery than their opponents (Taylor et al., 2008). However, during

TABLE 2 | Frequency distribution of type of serve and point won by the server or receiver for all players, medalists, and opponents (Crosstab Command: Pearson's Chi-square, degrees of freedom, significance, and effect size).

All players	Point won						χ ²	df	p	ES
	Server			Receiver						
	N	%	AR	N	%	AR				
Backhand flick	29	5.6	0.8	24	4.5	−0.8	1.284	4	0.86	0.04
Backhand short serve	149	28.5	0.6	143	27.0	−0.6				
Forehand flick	7	1.3	−0.2	8	1.5	0.2				
Forehand long serve	257	49.2	−0.9	276	52.1	0.9				
Forehand short serve	80	15.3	0.2	79	14.9	−0.2				
Total	522	49.6		530	50.4					
Medalists serving										
Backhand flick	29	8.1	4.3	1	0.4	−4.3	93.19†	4	<0.001*	0.38
Backhand short serve	131	36.7	5.1	43	17.6	−5.1				
Forehand flick	0	0.0	−3.2	7	2.9	3.2				
Forehand long serve	178	49.9	−1.1	133	54.3	1.1				
Forehand short serve	19	5.3	−7.0	61	24.9	7.0				
Total	350	49.4		359	50.6					
Opponents serving										
Backhand flick	0	0.0	−3.7	23	8.1	3.7	105.83†	4	<0.001*	0.48
Backhand short serve	18	10.9	−5.6	100	35.1	5.6				
Forehand flick	7	4.2	3.0	1	0.4	−3.0				
Forehand long serve	79	47.9	−0.5	143	50.0	0.5				
Forehand short serve	61	37.0	8.2	18	6.3	−8.2				
Total	172	50.1		171	49.9					

**p* < 0.05; †Fisher's exact test was used as the Expected Frequency Distribution was lower than 5. AR, adjusted residuals.

set 3 medalists recorded a longer set duration, and rest time, but shorter rally time than in a previous study that analyzed the whole competition (Torres-Luque et al., 2019). This result may reflect the fact that medalists usually play the decisive set 3 only during the eliminatory phase where the highest level of performance between players generates an open outcome. Thus, successful players used stoppages, end of points and breaks for managing pressure, fatigue, and recovery during these critical moments of the match (Taylor et al., 2008).

On the other hand, the specific sample of the Olympic Games and the analysis of only medalists' matches may have an impact on the current identified trends of temporal structure as was argued in the available research (Abdullahi and Coetzee, 2017). These results show that the time structure in elite badminton (Olympic Games) is a critical highly trained issue for players trying to perform at the highest level.

Type of Serve and Effectiveness

The results of serve effectiveness showed neutral values when analyzing all the players, medalists and opponents serving (49.6, 49.4, and 50.1%). The current results are in agreement with Bialik (2016) who identified that the serve is not an advantage in badminton. Thus, serving can be considered as a way to start the point that should potentially gain some spatial and temporal (e.g., short, flick, or long serves) advantage over the receiver during the consecutive strokes played in each point (Pearce, 2002; Alcock and Cable, 2009). However, the results showed significant

relationships of points won serving for medalists and opponents. Specifically, medalists won more points serving via the backhand flick and backhand short serve; while opponents won more points serving using the forehand flick and forehand short serve. In particular, as the serve is not affected by any previous action of the opponent, the server should manage the most effective serve during each context of badminton matches (Abdullahi and Coetzee, 2017; Chiminazzo et al., 2018). Thus, opponents start the point with less risky serves (e.g., forehand ones) than medalists (Yadav et al., 2007).

Two-Step Cluster Analysis

Successful actions in badminton require forcing the opponent to perform under spatial and temporal conditions (e.g., close to the net, moving from corner to corner, or corner-net-corner sequences) and then, generating open spaces to win the rally (Chow et al., 2014). Despite this general tactical approach, players have to serve trying to gain some advantage (spatial) during the next strokes to counteract the opponent's behaviors (Bialik, 2016). Therefore, due to the complex nature of badminton and the neutral serve effectiveness (i.e., ranging from 46 to 56%) the use of different type of serves during matches may allow successful players to adapt to the different scenarios that they have to deal with. In particular, the results of the two-step cluster analysis showed how successful players used the serve when playing short rallies with the backhand short and flick serves (cluster 1: 6.27 s, during sets 1 and 2 and 49.5% of serve

TABLE 3 | Results of rally types (clusters, % and n) identified by the two-step cluster analysis based on type of serve, serving player, set, point won by the server or receiver, rally time, rest time, frequency and number of strokes (I = predictor's importance; and BIC = Schwartz's Bayesian Information Criterion; Q1 = lower quartile; Q3 = upper quartile).

	Cluster 1			Cluster 2			Cluster 3			Cluster 4			Cluster 5		
Variables	15.2% (n = 160)			13.7% (n = 144)			27.3% (n = 287)			22.3% (n = 235)			21.5% (n = 226)		
Type of serve I = 1.0	%			%			%			%			%		
Backhand flick	17.6			0.0			1.4			0.0			0.0		
Backhand short serve	68.2			3.7			34.7			25.5			0.0		
Forehand flick	0.3			0.9			0.0			8.3			0.0		
Forehand long serve	0.0			94.5			49.3			0.0			100		
Forehand short serve	13.8			0.9			14.6			66.2			0.0		
Serving player I = 0.69															
Medalist	100			75.1			72.9			0.0			59.1		
Opponent	0.0			24.9			27.1			100			40.9		
Set I = 0.42															
Set 1	51.2			8.8			1.4			37.2			100		
Set 2	48.0			90.3			27.1			62.8			0.0		
Set 3	0.0			0.9			71.5			0.0			0.0		
Point won I = 0.38															
Server	49.5			44.7			56.9			51.0			49.0		
Receiver	50.5			55.3			43.1			49.0			51.0		
Temporal variables	Median (Q1/Q3)			Median (Q1/Q3)			Median (Q1/Q3)			Median (Q1/Q3)			Median (Q1/Q3)		
Rest time I = 0.12	22.2	17.1	28.3	19.4	15.3	29.9	27.5	21.8	37.6	22.5	18.7	28.4	20.5	16.2	27.2
Frequency I = 0.11	0.92	0.85	1.02	1.11	1.00	1.30	0.98	0.91	1.07	0.92	0.81	0.99	1.10	1.01	1.21
Rally time I = 0.11	6.27	3.83	9.73	7.87	4.98	11.2	12.3	6.45	21.6	6.63	3.83	12.4	8.80	6.02	13.5
Strokes I = 0.06	7.00	4.00	10.0	7.00	4.00	11.5	13.0	7.00	21.0	8.00	4.00	14.0	8.00	5.00	13.0
BIC	10326.95			9296.13			8472.99			7911.768			7456.60		

effectiveness), and forehand long serves (cluster 2: 7.87 s, during set 2 and 44.7% of serve effectiveness); and during longer rallies with the use of the backhand short serve, forehand short serve and forehand long serve (cluster 3: 12.3 s, during sets 2 and 3, and 56.9% of effectiveness). These results reinforce the idea that successful players are better prepared technically to execute a wide variety of serves according to each specific context (Taylor et al., 2008). In particular, the analysis of quick rallies such as clusters 1 and 2 reflected a lower serving effectiveness (49.5 and 44.7%, respectively) during sets 1 and 2. However, the results of cluster 3 may point to the better tactical and mental preparation to be more successful (56.9% of serve effectiveness) during long rallies played mainly during sets 2 and 3. This performance is related to successful players in elite badminton that can adapt the intensity required during the rallies using a variety of tactical patterns that lead to more successful actions as the match goes on (Barreira et al., 2016; Chiminazzo et al., 2018). In addition, these actions (cluster 3) lead to long rest times (ranging from 21 to 37 s) for medalists to manage fatigue and pressure before serving the next point.

On the other hand, medalists and their opponents played the rallies in a similar way using forehand long serves during set 1 with durations of 8.80 s and 49.0% of effectiveness (cluster 5). In particular, after the introduction of the new scoring system the forehand long serve is the most used serve to start the point and

TABLE 4 | Statistical differences among clusters in the categorical and numerical variables analyzed.

Categorical variables	χ^2	p	ES (ESI)
Type of serve	1319.62†	<0.001	0.54 (Large effect)
Serving player	455.381	<0.001	0.66 (Large effect)
Set	1169.68	<0.001	0.75 (Large effect)
Point won	5.346	0.252	0.07 (Small effect)
Numerical variables	χ^2	p	Post hoc
Rest time	53.605	<0.001	1 vs. 2-3; 2 vs. 4-5; 3 vs. 5
Frequency	258.550	<0.001	2 vs. 1-3-4-5; 5 vs. 1-3-4
Rally time	97.400	<0.001	3 vs. 1-2-4-5; 5 vs. 1-2-4
Strokes	71.933	<0.001	1 vs. 5; 2 vs. 5; 3 vs. 1-2-4-5

†Fisher's exact test was used as the Expected Frequency Distribution was lower than 5; ESI = effect size interpretation.

to generate cross-court shots forcing the opponent to run and return the shuttlecock (Alcock and Cable, 2009). Therefore, this result may suggest that at the beginning of the match both players perform with this serve trying to disrupt the opponent's strategies as a way to induce fatigue and constant adaptation to each stroke (Chow et al., 2014).

Lastly, the opponents showed an independent performance using the forehand short serve during sets 1 and 2, with short rally

times (6.63 s) and neutral effectiveness (51%). As was identified in previous studies (Laffaye et al., 2015; Chiminazzo et al., 2018) the performance required at the highest level may lead competitors to play below or beyond the competition requirements, and then opponents may be forced to play taking less risks when serving via short serves. Additionally, when medalists are receiving, they showed better strategical preparation against forehand serves (probably more predictable serves for them) and then performed anticipatory actions forcing the opponent to play quick rallies that involve 3 to 5 strokes (Yadav et al., 2007; Chiminazzo et al., 2018).

Classification Tree Analysis

The results of the classification tree analysis identified the importance of different serving patterns for each medalist based on some key variables (type of serve, round, rally time, and set) to classify their performances from a multivariate (integrated) approach. Particularly, the gold medalist was characterized by the use of more backhand and forehand flick serves, and the dominant use of the backhand short serve during sets 1 and 2 in all the tournament stages. On the contrary, the bronze medalist showed more forehand long serves during all sets. Lastly, the silver medalist showed a mixed performance of serves using the forehand short serve, the backhand short serve during set 3 of the group stage and round of 16, and the forehand long serve during the final and semi-finals. These findings reflect the importance of identifying individual playing patterns (profiles) that help players to be aware of serving strategies and performance areas they need to monitor according to the opponent (O'Donoghue, 2013). In particular, the silver medalist is a versatile player than manages a wider range of serve types than the gold and bronze medalists; and the bronze medalist mainly used forehand long serves. Therefore, these individual performance profiles based on multiple performance indicators make it possible to describe the idiosyncratic playing patterns of each player from technical, tactical, and strategical approaches (O'Donoghue, 2013). Thus, the analysis of successful players (medalists) is needed in order to update and manage the current player's performance and its evolution tournament to tournament in accordance with the serving and playing patterns identified (Menescardi et al., 2019).

The present study has some limitations that need to be acknowledged and addressed in future research. On the one hand, it analyzed the matches played by medalists but the opponents playing patterns were neither identified nor monitored during the multivariate analysis (classification and decision tree). As was identified, the type of rallies and playing patterns should be studied in depth in order to analyze the different players' profiles in elite men and women's badminton from a long-term perspective (i.e., individual players and by country/continent). On the other hand, further studies should consider larger datasets (Super Series, European and World Championships) to test

the importance of winning/losing and successful/unsuccessful conditions. Additionally, some variables can be included to analyze the ending action (type of technique), the zones of the court or the sequence of actions in each rally due to the importance of dynamic analysis in racket sports.

The findings of the current study have some practical implications that can be implemented during training and competitions. The information obtained about the temporal-related demands during matches played by successful players can be used to simulate these competitive scenarios during high-intensity sets/matches. Specifically, identified trends obtained with the two-step cluster analysis can help coaches and players to monitor different serving and playing patterns during training and matches according to the serve and rally requirements. Lastly, the analysis of each medalist's performance profile makes it possible to better describe and identify the serving strategies during tournaments. This individual approach would allow anticipating how to take advantage (i.e., most used strokes and tactics) of successful players according to serving strategies and playing patterns.

DATA AVAILABILITY STATEMENT

The datasets generated for this study are available on request to the corresponding author.

ETHICS STATEMENT

Written informed consent was not required, as the data used was publicly available on championship TV.

AUTHOR CONTRIBUTIONS

All co-authors equally contributed to the manuscript. M-ÁG-R designed, wrote, and supervised the draft of the article. AC collected the data and described the main variables and factors to use for. FR collected the data, discussed the design, and analyzed the article. L-MR contributed to the introduction, rationale and discussion of main findings, and supervised the final draft of the document.

FUNDING

The present study was supported by the Spanish Ministry of Economy and competitiveness with the project "Estudio de los complejos de juego y los perfiles de rendimiento en bádminton de élite COMPLEXBAD" (DEP2015- 67231-R).

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Conflict of Interest: FR was employed by company Spanish Badminton Federation.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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