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RECEIVED 15 March 2023

ACCEPTED 13 June 2023

PUBLISHED 28 July 2023

CITATION

Batista Taborda PA, Valente TS,
de Lima Carvalho MV, da Silva MVGB
and Paranhos da Costa MJR (2023)
Estimation of genetic parameters
for milking temperament in
Holstein-Gyr cows.
Front. Anim. Sci. 4:1187273.
doi: 10.3389/fanim.2023.1187273

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Estimation of genetic parameters for milking temperament in Holstein-Gyr cows

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Introduction: Dairy cattle with poor temperament can cause several inconveniences during milking, leading to labor difficulties, increasing the risk of accidents with animals and workers, and compromising milk yield and quality. This study aimed to estimate variance components and genetic parameters for milking temperament and its genetic correlations with milk yield in crossbred Holstein-Gyr cattle.

Methods: Data were collected at three commercial farms, resulting in 5,904 records from 1,212 primiparous and multiparous lactating cows. Milking temperament (MT), measured as the milking temperament of each cow, was assessed during pre-milking udder preparation (RP) and when fitting the milking cluster (RF) by ascribing scores from 1 (cow stands quietly) to 8 (the cow is very agitated, with vigorous movements and frequent kicking). The number of steps and kicks were also recorded during pre-milking udder preparation (S_{RP} and K_{RP} , respectively) and when fitting the milking cluster (S_{RF} and K_{RF} , respectively). Milk yield (MY) was obtained from each farm database. In two of them, MY was recorded during the monthly milk control (that could or could not coincide with the date when the milking temperament assessments were carried out) and in the remaining farm, MY was recorded on the same day that the milking temperament assessments were made. Genetic parameters were estimated using the THRGibbs1f90 program applying a threshold model, which included 89 contemporary groups as fixed effects, animal age at the assessment day and the number of days in milking as covariates, and direct additive genetic and residual effects as random effects.

Results and discussions: The heritability estimates were $MT = 0.14 \pm 0.03$ (for both, M_{RP} and M_{RF}), $MY = 0.11 \pm 0.08$, $S_{RP} = 0.05 \pm 0.03$, $K_{RP} = 0.14 \pm 0.05$, $S_{RF} = 0.10 \pm 0.05$, and $K_{RF} = 0.32 \pm 0.16$. The repeatability estimates were 0.38 ± 0.05 , 0.42 ± 0.02 , and 0.84 ± 0.006 for MT_{RP} , MT_{RF} , and MY, respectively; and 0.38 ± 0.02 , 0.30 ± 0.07 , 0.52 ± 0.02 , and 0.46 ± 0.15 for S_{RP} , K_{RP} , S_{RF} , and K_{RF} .

respectively. The estimates of most genetic correlation coefficients between $MT_{RP}-MT_{RF}$ were all strong and positive ($MT_{RR}-MT_{RF}= 0.63 \pm 0.10$, $MT_{RP}-S_{RP}= 0.65 \pm 0.12$, $MT_{RP}-K_{RP}= 0.56 \pm 0.16$, $MT_{RF}-S_{RF}= 0.77 \pm 0.06$, and $MT_{RF}-K_{RF}= 0.56 \pm 0.34$) except for MY ($MT_{RP}-MY= 0.26 \pm 0.26$ and $MT_{RF}-MY= 0.21 \pm 0.23$). Despite the low magnitude of MT heritability, it can be included as a selection trait in the breeding program of Holsteins-Gyr cattle, although its genetic progress will be seen only in the long term. Due to the low accuracy of the genetic correlation estimates between MT and MY and the high range of the 95% posterior density interval, it cannot be affirmed by this study that the selection of a milking temperament trait will infer on milk yield. More data is therefore needed per cow and more cows need to be observed and measured to increase the reliability of the estimation of these correlations to be able to accurately interpret the results.

KEYWORDS

dairy cattle, genetic correlation, Girolando, heritability, reactivity, workability

1 Introduction

Individual variability has been observed in the behavior of dairy cattle in response to a stressor or environmental challenges, leading to considerable impacts on performance, reproduction, health, and animal welfare (Sutherland et al., 2012; Haskell et al., 2014; Friedrich et al., 2015; Hedlund and Lövlie, 2015; Marçal-Pedroza et al., 2021). Previous studies have suggested that calmer cows during milking facilitate handling procedures and have higher production rates and milking speed (Wickhman, 1979; Lawstuen et al., 1988; Cue et al., 1996; Samoré et al., 2010; Sewalem et al., 2011; Hedlund and Lövlie, 2015) in comparison to nervous cows. Breuer et al. (2000), working with Holstein cows, reported that special attention is required for animals showing a higher level of body and leg movements and kicks during milking, which inevitably leads to difficulties and increased labor time when carrying out the handling procedures. These variables have been used to characterize the level of stress during milking, and, consequently, are expected to accompany the inhibition of milk ejection and decreased milk yield (Breuer et al., 2000; Haskell et al., 2014). More recently, Marçal-Pedroza et al. (2021) reported that dairy cows' temperament is also related to metabolic efficiency and enteric CH₄ emissions, directly affecting the sustainability of this system.

As reviewed by Haskell et al. (2014) and Chang et al. (2020), milking temperament is low to moderately heritable and genetically correlated with milk production, workability, health, and reproductive traits. Low heritability estimates (0.07) for the milking temperament of Holstein cows were reported by Pryce et al. (2000) and Hiendleder et al. (2003) when applying a score from 1 (defined by the authors as "nervous/aggressive") to 9 ("quiet/docile"). Sewalem et al. (2011), working with records of 1,940,092 Holstein cows and applying a score ranging from 1 ("very nervous") to 5 ("very calm"), reported a heritability of 0.13. Similarly, Cue

et al. (1996) reported heritability of 0.14 and 0.17 for the milking temperament of Holstein and Jersey cows, respectively, and a higher estimate (0.33) for Ayrshire cows. These authors used a scoring system that ranged from 1 ("vicious") to 9 ("placid"). With a similar scoring system, ranging from 1 ("acceptable") to 5 ("undesirable"), Visscher and Goddard (1995) estimated a heritability of 0.22 for Holstein (14,596 records) and 0.25 for Jersey (4,695 records) cows.

It is important to highlight that most of the estimations of variance and covariance components and genetic parameters for milking temperament have been carried out assessing *Bos taurus* cattle breeds, such as Holstein cows (Visscher and Goddard, 1995; Cue et al., 1996; Pryce et al., 2000; Hiendleder et al., 2003; Sewalem et al., 2011; Pires et al., 2013; Stephansen et al., 2018). Indeed, little has been done for *Bos indicus* breeds, such as Girolando or Gyr cows, regardless of the importance of using local breeds to improve profitability while reducing health and welfare issues. Thus, it is important to develop additional studies addressing *Bos indicus* breeds and their crosses.

The introduction of the milking temperament trait as a selection index in dairy production is a tool to select calmer animals and in the long term achieve a genetic change in the herd for this characteristic (Haskell et al., 2014; Chang et al., 2020). If we focus on genetic-environment interaction, the *Bos indicus* breeds are more adapted to tropical conditions, but their temperament is a concern for dairy producers since the cows are usually more reactive to the milking procedures and present a higher fear of human approach and less productivity (Fordyce et al., 1982; Paranhos da Costa et al., 2015). Crossing *Bos indicus* with *Bos taurus* animals is one strategy to address this problem, and in Brazil, the greater part (80%) of milk production is provided by crossing Holstein (*Bos Taurus*) and Gyr cattle (*Bos indicus*) (Ferreira et al., 2002; Madalena et al., 2012). To the best of our knowledge, there is no information available in the literature regarding the estimation of the genetic parameters for milking temperament for Girolando cattle. For this

reason, the present study contributes a novel approach for Brazilian dairy producers and other dairy systems that use *Bos taurus* and *Bos indicus* crossbreed cattle. Thus, this study aimed to estimate the genetic and phenotypic parameters of milking temperament, as well as its genetic correlation with milk yield in crossbred Holstein-Gyr (HG) cattle raised in Brazil.

2 Materials and methods

The Committee of the Ethical Use of Animals of the Faculty of Agricultural and Veterinary Sciences, São Paulo State University (UNESP), Jaboticabal, SP, Brazil, approved this study (Protocol n. 005215/18).

2.1 Animals and housing conditions

The study was conducted at three commercial dairy farms associated with the Girolando Breeders Association (GIROLANDO) from April 2018 to May 2019, resulting in 5,904 records from 1,212 lactating Holstein-Gyr cows, daughters of 155 sires and 663 dams. The Girolando Breeders Association provided the pedigree data containing 19,531 sires and 349,222 dams.

On two farms (Farms 1 and 2), the cows were housed in a free stall housing environment, and on the remaining farm (Farm 3), they were kept in pastures. The cows were milked twice daily (in the mornings and afternoons). In Farms 2 and 3, they were milked in herringbone parlors with automatic cluster removal systems, while in Farm 1, they were milked in a rotary/carrousel parlor. On all farms, the cows are separated from the calves following calving before 24 hours and integrated within the milking herds. The replacement of animals in the dairy herd is from the calves themselves born on the farms.

All dairy cows evaluated in the present study were born between 2009 and 2017. Most of them were $\frac{3}{4}$ Holstein-Gyr (632, ~52%), followed by F1 Holstein-Gyr (513, ~42%), and only 67 (~6%) represented other Holstein-Gyr crosses. Around sixty-six per cent (803) cows were primiparous and 409 (~34%) were multiparous. Of the primiparous cows, 452 (~56%) were $\frac{3}{4}$ Holstein-Gyr, 314 (~39%) were $\frac{1}{2}$ Holstein-Gyr and 37 (~5%) were other Holstein-Gyr crosses; of the multiparous cows, 196 (~48%) were $\frac{1}{2}$ Holstein-Gyr, 176 (~43%) were $\frac{3}{4}$ Holstein-Gyr, and 37 (~9%) were other Holstein-Gyr crosses.

Cow birth seasons were classified as rainy (September to February) and dry (March to August). The average age at first calving was 32 ± 14 months (ranging from 21.8 to 61.3 months). Lactation days were calculated as the number of days in lactation that the cow was at the time of the milking temperament measurement, ranging from 9 to 305 days, with 408 cows at the beginning of lactation (9 to 100 days), 595 at the middle of lactation (100 to 200 days) and 209 cows at the end of lactation (200 to 305 days), with an average milk yield of 20.5 ± 6.3 L/day (ranging from 3.0 to 59.4 L/day).

After pre-milking udder preparation and before the commencement of the milking process, 298 (~25%) cows received

an application of exogenous oxytocin and 914 (~75%) did not. Of the cows that received an injection of exogenous oxytocin, 159 (~53%) were primiparous and 139 (~47%) were multiparous; of those cows that did not receive an application of exogenous oxytocin, 644 (~70%) were primiparous and 270 (~30%) were multiparous.

2.2 Milking temperament assessment

From each farm, phenotypic data of milking temperament was collected as milking temperament scores during three consecutive days for three consecutive months, totaling nine measuring events for each cow during the first milking of the day. However, not all animals were available to be recorded nine times for reasons out of our control, such as health issues or other treatment, resulting in an unequal number of available measurements per cow. Of the total data collected, most animals (300 cows, ~25%) were evaluated only three times, 188 cows (~16%) had six measurement events, and 85 cows (~7%) had nine measurement events. Detailed information about this is shown in Table 1.

Milking temperament was scored during pre-milking udder preparation (MT_{RP}) and when fitting the milking cluster (MT_{RF}) by assigning one of the scores described in Table 2. The number of steps and kicks were also recorded during pre-milking udder preparation (S_{RP} and K_{RP} , respectively) and when fitting the milking cluster (S_{RF} and K_{RF} , respectively).

Milk yield (MY) was obtained from the farms' database. In two of them, MYs were recorded during the monthly milk recording records (that could or could not coincide with the days on which the milking temperament assessments were carried out), and in the remaining farm, MYs were recorded on the same days that milking temperament assessments were made. These differences concerning the recording of MY may have contributed to errors in the genetic correlation estimates of MY with MT_{RP} and MT_{RF} .

TABLE 1 The number of cows and respective percentages according to the number of records of milking temperament measurement events.

Number of records of milking temperament measurement events	Number of cows	%
1	52	4
2	145	12
3	300	25
4	68	6
5	134	11
6	188	16
7	97	8
8	143	12
9	85	7
Total	1212	100

TABLE 2 Description of the milking temperament scores used to assess Holstein-Gyr cows' milking temperament during pre-milking udder preparation and when fitting the milking cluster.

Scores	Descriptions
1	Cow stands quietly.
2	Cow remains still, but it can arch its back and stretch its legs.
3	Cow gently moves only one hind leg.
4	Cow gently moves both back legs, backing them up in an alternate way.
5	Cow shows occasional vigorous hind legs movements.
6	Cow shows vigorous hind and front legs movements.
7	Cow kicks.
8	Cow is very agitated, showing vigorous movements and frequent kicking, making the milking procedure impossible without tying her hind legs.

Adapted from Paranhos da Costa and Broom (2001).

2.3 Statistical analyses

In total, 89 contemporary groups (CG) were categorized by farm, year and season of cows' birth, and genetic group (including mainly 3/4 Holstein-Gyr, 1/2 Holstein-Gyr cows, and other groups, including 1/4, 3/8, and 5/8 Holstein-Gyr cows). The THRGIBBS1F90 software (Miszta et al., 2015) was used to estimate the (co)variance components and genetic parameters by implementing a Bayesian inference using the Gibbs sampling algorithm. A multi-trait analysis was performed to estimate the variance components, heritability, and repeatability of milking temperament scores, and the number of steps and kicks during pre-milking preparation (MT_{RP} , S_{RP} and K_{RP} , respectively) and when fitting the milking clusters (MT_{RF} , S_{RF} and K_{RF} , respectively). Genetic and phenotypic correlations of MY with MT_{RP} and MT_{RF} were also estimated. Since MT_{RP} and MT_{RF} were categorical variables, the Bayesian threshold was the most appropriate method for conducting genetic analyses, which assumes that the number of levels is related to an underlying continuous scale containing fixed and random effects (Van Tassell et al., 1998). For MT_{RP} data, scores from 1 to 7 were considered (score 8 was eliminated due to only having a few recorded instances, which were therefore included in score 7), while for MT_{RF} data, all scores were considered (from 1 to 8). The number of steps and kicks during pre-milking udder preparation (S_{RP} and K_{RP}) and when fitting the milking cluster (S_{RF} and K_{RF}) and MY were considered continuous variables.

The animal model used included direct additive genetic and residual effects as random effects and CG as a fixed effect; the animal age at the time of milking temperament scoring (with linear and quadratic regressions), and the number of days in milk (linear regression) were included as covariates for all traits. The matrix presentation of the general model used is as follows:

$$y = XQ + Za + Wpe + e$$

where: y is the vector of observations; β is the vector of fixed effects; a is the vector of the direct additive genetic effect of the animal; pe is the vector related to permanent environment random

effects of the animal (each daily milking temperament measurement considered as repeated measurements on the cow); X , Z , and W are known incidence matrices relating β , a , and pe to y ; and e is the vector of residuals.

It was assumed that $E[y] = X\beta$; $\text{Var}(a) = A\otimes G$; $\text{Var}(pe) = I\otimes PE$; $\text{Var}(e) = I\otimes R$, where A is the relationship matrix among all animals in the pedigree file containing 19,531 sires and 349,222 dams, \otimes is the direct product, G is the (co)variance matrix of direct additive genetic effects, PE is the (co)variance matrix of permanent environmental effects, I is the identity matrix, and R is the (co)variance matrix of residual effects.

The vectors β , a , and pe are location parameters from the conditional distribution. A uniform distribution of β was assumed a priori, which reflects a vague prior knowledge about this vector. For the (co)variance matrices of random effects, inverted Wishart distributions were defined as prior distributions. Thus, the distribution of y given the parameters of location and scale was assumed (Van Tassell and Van Vleck, 1996):

$$y|\beta, a, R \sim \text{MVN}[X\beta + Za + Wpe, \text{INR}]$$

For analysis, chains of 1,200,000 iterations were generated, with samplings every 20 cycles. The first 300,000 iterations were discarded as fixed burn-in. Thus, 45,000 samples were used for parameter estimations.

Data convergence was checked through the criteria proposed by Geweke (1992) and Heidelberger and Welch (1983) using the R software, with the Bayesian Output Analysis (BOA) package in R 4.1.0 software (The R Development Core Team).

After obtaining the correctly converged variances, heritability (h^2) and repeatability (R) for milking temperament, the number of steps (SRP , S_{RF}) and kicks (KRP , K_{RF}), and phenotypic (r_{P1P2}) and genetic (r_{A1A2}) correlations between milking temperament and milk yield during pre-milking udder preparation and when fitting the milking cluster were estimated as:

$$h^2 = \frac{\sigma^2 a}{\sigma^2 a + \sigma^2 pe + \sigma^2 e} \quad R = \frac{(\sigma^2 a + \sigma^2 pe)}{(\sigma^2 a + \sigma^2 pe + \sigma^2 e)}$$

$$r_{P1P2} = \left(\frac{C(P1, P2)}{\sigma_{P1} \sigma_{P2}} \right) \quad r_{A1A2} = \left(\frac{Cov(A1, A2)}{\sigma_{A1} \sigma_{A2}} \right)$$

where: $\sigma^2 a$ is additive genetic variance; $\sigma^2 pe$ is permanent environmental variance (due to repeated measurements of milking temperament records per cow); σ^2 is residual variance; $Cov(P1, P2)$ is phenotype co(variance) between two traits; $Cov(A1, A2)$ is genetic co(variance) between two traits; σ_{P1} and σ_{P2} are phenotypes standard deviation of traits 1 and 2; and σ_{A1} and σ_{A2} are genetic standard deviations of traits 1 and 2.

3 Results and discussion

For all the phenotypic data collected, cows presented higher temperament scores during pre-milking udder preparation (MT_{RP} : 4.33 ± 1.43) compared to when fitting the milking cluster (MT_{RF} : 2.74 ± 1.47), with mode values of 5 and 1, respectively. In the same

way, during udder preparation, there was a higher number of steps (S_{RP} : 5.08 ± 3.69 , ranging from 0 to 38) and kicks (K_{RP} : 0.11 ± 0.63 , ranging from 0 to 16) when compared to when fitting the milking cluster (S_{RF} : 1.61 ± 2.05 , ranging from 0 to 42, and K_{RF} : 0.01 ± 0.23 , ranging from 0 to 10). The high SD and CV (%) in the number of kicks during pre-milking udder preparation (K_{RP}), and number of steps and kicks when fitting the milking cluster (S_{RF} and K_{RF} , respectively) indicate important individual differences in the way that cows react to these handling procedures (Table 3).

According to the convergence criteria applied in this study for all trait analyses, the number of remaining Markov chains (45,000) was adequate for obtaining the convergence of all parameters estimated. Table 4 shows the posterior means of additive genetic, permanent environment, and residual variances, and heritability and repeatability obtained for milking temperament-related traits and milk yield.

The posterior means of heritability for milking temperament during pre-milking udder preparation (MT_{RP}) and when fitting the milking cluster (MT_{RF}) were 0.14 ± 0.03 . These results are in line with the values reported in the literature, which are like those estimated by Wickham (1979); Lawstuen et al. (1988); Cue et al. (1996), and Sewalem et al. (2011) for Holstein cows (h^2 ranging from 0.11 to 0.14). However, the heritability estimated in the present study was lower than that found by O'Bleness et al. (1960); Dickson et al. (1970), and Visscher and Goddard (1995) for Holstein cows (0.40, 0.47, and 0.22, respectively). The estimated mean heritability for the number of steps

(0.05 ± 0.03) and kicks (0.14 ± 0.05) was estimated during pre-milking udder preparation, and the estimations for the number of steps and kicks when fitting the milking clusters were 0.10 ± 0.05 and 0.32 ± 0.16 , respectively. It should be noted that the only literature currently available for discussion regarding the estimation of genetic parameters for milking temperament is based entirely on *Bos taurus* dairy cattle herds, while in this study results from *Bos taurus* x *Bos indicus* dairy crosses are presented.

The MY heritability in our study was of lower magnitude (0.11 ± 0.08) than that obtained in other studies with Holstein, Gyr, and Brown Swiss breeds (0.20, 0.22, and 0.24; Rennó et al., 2003; Lagrotta et al., 2010; Campos et al., 2015, respectively), as well as than the estimate reported by the national breeding program for Girolando cattle ($h^2_{MY} = 0.29$) (da Silva et al., 2020).

The repeatability estimates of this study were moderate for milking temperament and the number of steps and kicks during pre-milking udder preparation and when fitting the milking cluster, ranging from 0.30 to 0.52 (Table 5). Similar results were reported by Erf et al. (1992); Kramer et al. (2013), and Wethal and Heringstad (2019). These authors estimated values ranging from 0.32 to 0.56 in Holstein, Brown Swiss, and Norwegian Red cattle herds, respectively.

A strong, positive, and favorable genetic correlation (0.63 ± 0.10) was observed between MT_{RP} and MT_{RF} (Table 5). In the same way, genetic correlations between MT_{RP} - S_{RP} (0.65 ± 0.12), MT_{RP} - K_{RP} (0.56 ± 0.16), MR_{RF} - S_{RF} (0.77 ± 0.06), and MT_{RF} - K_{RF} ($0.56 \pm$

TABLE 3 Means, standard deviations (SD), mode, minimum (Min), and maximum (Max) values of the coefficients of variation (CV, %) for milking temperament, and number of steps and kicks during pre-milking udder preparation (MT_{RP} , S_{RP} , and K_{RP} , respectively), and milking temperament, and the number of steps and kicks when fitting the milking cluster (MT_{RF} , S_{RF} , and K_{RF} , respectively) and milk yield (MY) in the dataset of Holstein-Gyr cross cattle.

Traits	N	Mean \pm SD	Mode	Min	Max	CV (%)
MT_{RP}	5,904	4.33 ± 1.43	5	1	7	32.94
SRP	5,904	5.08 ± 3.69	-	0	38	72.74
KRP	5,904	0.11 ± 0.63	-	0	16	546.87
MT_{RF}	5,904	2.74 ± 1.47	1	1	8	53.84
SRF	5,904	1.61 ± 2.05	-	0	42	126.84
KRF	5,904	0.01 ± 0.23	-	0	10	1747.82
MY	5,904	20.47 ± 6.32	-	3.00	49.00	30.87

TABLE 4 Descriptive statistics of posterior density (95% highest posterior density intervals, HPD) of variance components, heritability (h^2) and repeatability (R) estimates for milking temperament, number of steps and kicks, and milk yield of Holstein-Gyr cross cattle.

Traits	σ^2_a	σ^2_{pe}	σ^2_e	$h^2 \pm SD$	HPD	$R \pm SD$	HPD
MT_{RP}	0.15 ± 0.18	0.28 ± 0.41	0.80 ± 1.24	0.14 ± 0.03	0.08-0.21	0.38 ± 0.05	0.28-0.47
SRP	0.58 ± 0.33	3.98 ± 0.43	7.53 ± 0.31	0.05 ± 0.03	0.01-0.10	0.38 ± 0.02	0.34-0.41
KRP	0.42 ± 0.18	0.47 ± 0.23	2.13 ± 0.22	0.14 ± 0.05	0.05-0.24	0.30 ± 0.07	0.15-0.44
MT_{RF}	0.10 ± 0.02	0.19 ± 0.03	0.40 ± 0.05	0.14 ± 0.03	0.08-0.20	0.42 ± 0.02	0.38-0.46
SRF	1.75 ± 0.80	7.05 ± 0.92	7.95 ± 0.80	0.10 ± 0.05	0.03-0.19	0.52 ± 0.02	0.48-0.57
KRF	1.46 ± 1.00	0.58 ± 0.40	2.41 ± 1.03	0.32 ± 0.16	0.04-0.61	0.46 ± 0.15	0.17-0.73

σ^2_a , genetic additive variance; σ^2_{pe} , permanent environmental variance; σ^2_e , residual variance; SD, standard deviation; MT_{RP} , milking temperament during pre-milking udder preparation; MT_{RF} , milking temperament when fitting the milking cluster; S_{RP} , steps during pre-milking udder preparation; S_{RF} , steps when fitting the milking cluster; K_{RP} , kicks during pre-milking udder preparation; K_{RF} , kicks when fitting the milking cluster; MY, milk yield.

TABLE 5 Posterior estimates of genetic (above diagonal) and phenotypic (below diagonal) correlations (mean \pm standard deviation) and the highest posterior density interval containing 95% of the observations (inside brackets) between milking temperament, number of steps and kicks, and milk yield traits of Holstein-Gyr cross cattle.

Traits	MT _{RP}	S _{RP}	K _{RP}	MT _{RF}	S _{RF}	K _{RF}	MY
MT _{RP}	–	0.65 \pm 0.12 (0.44 – 0.89)	0.56 \pm 0.16 (0.25 – 0.88)	0.63 \pm 0.10 (0.44 – 0.82)	0.57 \pm 0.18 (0.24–0.90)	0.34 \pm 0.32 (-0.25 – 0.91)	0.26 \pm 0.26 (-0.14 – 0.86)
SRP	0.76 \pm 0.05 (0.66 – 0.85)	–	0.69 \pm 0.28 (-0.03 – 0.94)	0.57 \pm 0.14 (0.30 – 0.84)	0.65 \pm 0.17 (0.31–0.99)	0.56 \pm 0.24 (0.1 – 0.94)	0.55 \pm 0.28 (-0.006 – 0.96)
KRP	0.75 \pm 0.12 (0.51 – 0.96)	0.69 \pm 0.16 (0.38 – 0.93)	–	0.44 \pm 0.17 (0.12– 0.77)	0.51 \pm 0.34 (-0.22 - 0.94)	0.46 \pm 0.35 (-0.23 - 1)	-0.07 \pm 0.28 (-0.60 – 0.52)
MT _{RF}	0.42 \pm 0.09 (0.25 – 0.59)	0.38 \pm 0.07 (0.23 – 0.52)	0.64 \pm 0.12 (0.39 – 0.87)	–	0.77 \pm 0.06 (0.64 – 0.87)	0.56 \pm 0.34 (-0.34 – 0.88)	0.21 \pm 0.23 (-0.19 – 0.72)
SRF	0.33 \pm 0.09 (0.16 – 0.50)	0.39 \pm 0.26 (-0.11 – 0.90)	0.33 \pm 0.34 (-0.34 – 0.99)	0.88 \pm 0.02 (0.84 - 0.92)	–	0.66 \pm 0.49 (-0.65 – 0.99)	0.43 \pm 0.26 (-0.80 – 0.87)
KRF	0.36 \pm 0.34 (-0.20 – 0.98)	-0.25 \pm 0.40 (-1 – 0.54)	0.30 \pm 0.40 (-0.49 – 0.99)	0.44 \pm 0.35 (-0.22 – 0.93)	0.35 \pm 0.41 (-0.44 – 0.97)	–	0.64 \pm 0.54 (-0.45 – 0.99)
MY	-0.09 \pm 0.08 (-0.25 – 0.06)	-0.07 \pm 0.25 (-0.49 – 0.48)	-0.03 \pm 0.30 (-0.62 – -0.55)	-0.02 \pm 0.07 (-0.16 – 0.13)	-0.16 \pm 0.10 (-0.39 - 0.04)	0.11 \pm 0.22 (-0.39 - 0.53)	–

MT_{RP}, milking temperament during pre-milking udder preparation; MT_{RF}, milking temperament when fitting the milking cluster; S_{RP}, steps during pre-milking udder preparation; S_{RF}, steps when fitting the milking cluster; K_{RP}, kicks during pre-milking udder preparation; K_{RF}, kicks when fitting the milking cluster; MY, milk yield.

0.34) were also high and positive. Thus, only one of these traits can be used to assess a Holstein-Gyr cow's temperament during milking to implement a breeding program that includes milking temperament-related traits. It can be inferred in this study that the most reactive cows measured through milking temperament scores showed a greater expression of steps and kicks, and, in the inverse, cows with a lower milking temperament score expressed fewer steps and kicks. It is advisable to implement the counting of the number of steps during the different moments in the milking process; it is easy to measure and does not need any score for its measurement. These results confirm what was suggested by Breuer et al. (2000) when it was recommended that the number of steps and kicks should be counted as an alternative to measuring milking temperament in dairy herds.

The phenotypic and genetic correlation estimates between milking temperament, number of steps and kicks, and milk yield cannot allow the orientation and degree of the phenotypic and genetic correlations to be inferred, since the estimated value of the standard deviations and the highest posterior density interval containing 95% have a very high range, including the zero; therefore, the values of the correlations estimated can be negative, zero, or positive. Consequently, it cannot be affirmed by this study that the selection of a milking temperament trait will infer on milk yield. More data is therefore needed per cow, and more cows need to be measured to increase the reliability of the estimation of these correlations to be able to accurately interpret the results.

4 Conclusions

Although the heritability estimated for milking temperament and the number of steps and kicks during pre-milking udder

preparation and when fitting the milking cluster reached low magnitude, there is a possibility that if the selection is made through this trait, long-term genetic progress can be seen. Thus, the estimations of heritability and repeatability for milking temperament justify the inclusion of this trait as a selection criteria trait for the Holstein-Gyr cross in Brazil.

This study confirms that milking temperament during pre-milking udder preparation has a positive and high genetic correlation with milking temperament when fitting the milking cluster. Furthermore, a positive genetic correlation also exists between milking temperament and counting the steps and kicks during pre-milking udder preparation and when fitting the milking cluster. Animals with high milking temperaments are known to express more steps and kicks during the milking process making handling difficult. Counting steps during milking is an appropriate measurement for including milking temperament in selection indexes for the Holstein-Gyr cross, because it is easy and inexpensive to measure, and it can be used to assess milking temperament objectively.

More records are needed to estimate the genetic and phenotypic correlations between milking temperament and milk yield more accurately since they could not be affirmed in this study due to the high standard errors of the estimates, as well as the high range of the 95% posterior density interval.

Data availability statement

The original contributions presented in the study are included in the article/supplementary materials. Further inquiries can be directed to the corresponding author/s.

Ethics statement

The animal study was reviewed and approved by Committee of the Ethical Use of Animals of the Faculty of Agricultural and Veterinary Sciences, São Paulo State University (UNESP), Jaboticabal, SP, Brazil (Protocol n. 005215/18). Written informed consent was obtained from the owners for the participation of their animals in this study.

Author contributions

MdC and TV contributed to the conception and design of the study. PB organized the database, performed the statistical analysis, and wrote the first draft of the manuscript. TV and MdS performed the statistical analysis. MC organized the database. All authors contributed to the article and approved the submitted version.

Funding

This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001, which had no role in the study design, data collection and analysis, the decision to publish, or preparation of the manuscript.

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Acknowledgments

The study was part of the doctoral thesis of the first author (Paula A. Batista Taborda), prepared for the Graduate Program in Genetics and Animal Breeding at São Paulo State University (UNESP), Faculty of Agricultural and Veterinary Sciences, Jaboticabal, SP, Brazil. Special appreciation is expressed to the owners or managers of the farms Santa Luzia (Mauricio Silveira Coelho), Calciolândia (Jordane Silva and Ronaldo Lazzarini Santiago), and Boa Fé (Jônadan Ma) and their staff for their support and making it possible to carry out this research.

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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