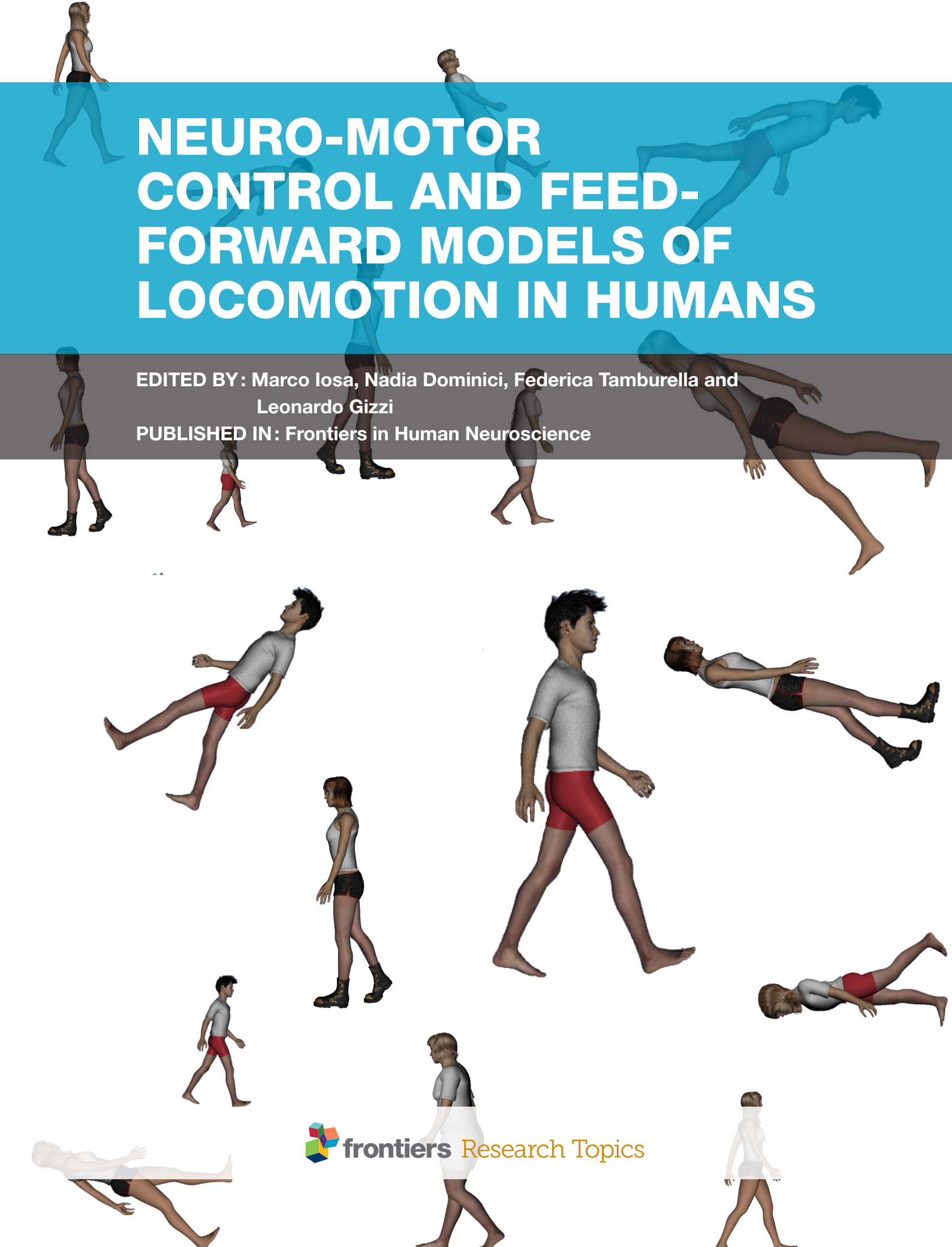
NEURO-MOTOR CONTROL AND FEED-FORWARD MODELS OF LOCOMOTION IN HUMANS

EDITED BY: Marco Iosa, Nadia Dominici, Federica Tamburella and Leonardo Gizzi

PUBLISHED IN: Frontiers in Human Neuroscience



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ISSN 1664-8714
ISBN 978-2-88919-614-2
DOI 10.3389/978-2-88919-614-2

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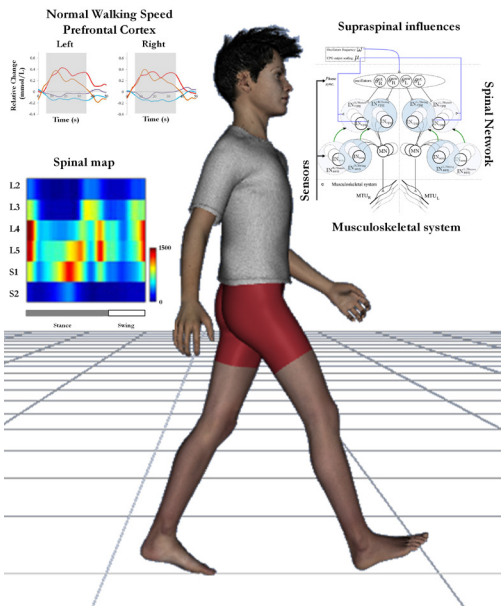
NEURO-MOTOR CONTROL AND FEED-FORWARD MODELS OF LOCOMOTION IN HUMANS

Topic Editors:
Marco Iosa, I.R.C.C.S. Fondazione Santa Lucia, Italy
Nadia Dominici, VU University Amsterdam, Netherlands
Federica Tamburella, I.R.C.C.S. Fondazione Santa Lucia, Italy
Leonardo Gizzi, Bernstein Center for Computational Neuroscience, Germany



A Walking Man. Above on the left, the relative change of oxyhemoglobin during walking and dual task walking (red and orange, respectively) and of deoxyhemoglobin during the same tasks (blue and purple, respectively) [Meester et al. 2014]. Below on the left, the spinal map of alpha motoneuron activity of the lumbosacral enlargement of a subject during normal walking [La Scaleia et al. 2014]. On the right, a schematic representation of the spinal network and supraspinal control [Dzeladini et al. 2014].

Locomotion involves many different muscles and the need of controlling several degrees of freedom. Despite the Central Nervous System can finely control the contraction of individual muscles, emerging evidences indicate that strategies for the reduction of the complexity of movement and for compensating the sensorimotor delays may be adopted.

Experimental evidences in animal and lately human model led to the concept of a central pattern generator (CPG) which suggests that circuitry within the distal part of CNS, i.e. spinal cord, can generate the basic locomotor patterns, even in the absence of sensory information. Different studies pointed out the role of CPG in the control of locomotion as well as others investigated the neuroplasticity of CPG allowing for gait recovery after spinal cord lesion. Literature was also focused on muscle synergies, i.e. the combination of (locomotor) functional modules, implemented in neuronal networks of the spinal cord, generating specific motor output by imposing a specific timing structure and appropriate weightings to muscle activations. Despite the great interest that this approach generated in the last years in the Scientific Community, large areas of investigations remain available for further improvement (e.g. the influence of afferent feedback and environmental constrains) for both experimental and simulated models.

However, also supraspinal structures are involved during locomotion, and it has been shown that they are responsible for initiating and modifying the features of this basic rhythm, for stabilising the upright walking, and for coordinating movements in a dynamic changing environment. Furthermore, specific damages into spinal and supraspinal structures result in specific alterations of human locomotion, as evident in subjects with brain injuries such as stroke, brain trauma, or people with cerebral palsy, in people with death of dopaminergic neurons in the substantia nigra due to Parkinson's disease, or in subjects with cerebellar dysfunctions, such as patients with ataxia.

The role of cerebellum during locomotion has been shown to be related to coordination and adaptation of movements. Cerebellum is the structure of CNS where are conceivably located the internal models, that are neural representations miming meaningful aspects of our body, such as input/output characteristics of sensorimotor system. Internal model control has been shown to be at the basis of motor strategies for compensating delays or lacks in sensorimotor feedbacks, and some aspects of locomotion need predictive internal control, especially for improving gait dynamic stability, for avoiding obstacles or when sensory feedback is altered or lacking. Furthermore, despite internal model concepts are widespread in neuroscience and neurocognitive science, neurorehabilitation paid far too little attention to the potential role of internal model control on gait recovery.

Many important scientists have contributed to this Research Topic with original studies, computational studies, and review articles focused on neural circuits and internal models involved in the control of human locomotion, aiming at understanding the role played in control of locomotion of different neural circuits located at brain, cerebellum, and spinal cord levels.

Citation: Iosa, M., Dominici, N., Tamburella, F., Gizzi, L., eds. (2015). Neuro-Motor Control and Feed-Forward Models of Locomotion in Humans. Lausanne: Frontiers Media.
doi: 10.3389/978-2-88919-614-2

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frontiers in Human Neuroscience

EDITORIAL
published: 02 June 2015
doi: 10.3389/fnhum.2015.00306

Editorial: Neuro-motor control and feed-forward models of locomotion in humans

Marco Iosa[1]*, *Leonardo Gizzi*[2], *Federica Tamburella*[3] and *Nadia Dominici*[4]

[1] Clinical Laboratory of Experimental Neurorehabilitation, Fondazione Santa Lucia I.R.C.C.S., Rome, Italy, [2] Department of Neurorehabilitation Engineering, Bernstein Focus Neurotechnology Göttingen - Bernstein Center for Computational Neuroscience, Göttingen, Germany, [3] SPInal REhabilitation Lab (SPIRE), Fondazione Santa Lucia I.R.C.C.S., Rome, Italy, [4] Faculty of Human Movement Sciences, MOVE Research Institute, VU University Amsterdam, Amsterdam, Netherlands

Keywords: gait, walking, central pattern generators (CPG), motor control, neurorehabilitation

> "He told me with amusement that when one is walking rapidly each step takes no more than half a second, and in that half second no fewer than 54 muscles are set in motion. I listened in awe. I at once directed my attention to my legs and tried to discover the infernal machine. I thought I had succeeded in finding it. I could not of course distinguish all its 54 parts, but I discovered something terrifically complicated which seemed to get out of order the instant I began thinking about it."

Well-depicted by Svevo in "Confessions of Zeno" (Svevo, 1923, 1989), the act of walking involves many different muscles and the necessity of controlling several degrees of freedom at once. This Research Topic has mainly been focused on the strategies adopted by the central nervous system for reducing the complexity of motor control and compensating for the sensorimotor delays. The studies published within this Research Topic addressed this issue at two levels of investigation, focusing on one side the neural circuitry, such as the so called central pattern generators in the spinal cord and the supraspinal structures, and on the other one on the cognitive processes involved during locomotion.

One of the paramount discoveries in locomotion is the existence of a central pattern generator (CPG), i.e., a neural circuitry within the spinal cord that can autonomously generate basic locomotor rhythmic patterns, even in the absence of brain connections and sensory information (Grillner, 1985). Although there is compelling evidence of existence of CPG in humans, a final proof is still lacking, also because CPGs have generally been investigated in reduced models including *in vitro* isolated preparations, genetically-engineered mice, spinal cord-transected animals, and virtual models. Guertin (2014) presented an extensive review of studies, concluding that the development of CPG-modulating clinical therapies is a necessary step for improving the locomotor function in patients with spinal cord injury. Similarly, the study of Dzeladini et al. (2014) enters in the debate between CPG and reflex-based human neuro-musculo-skeletal models, supplying a mixed model in which CPG are integrated in a reflex-based model. Their results highlighted potential advantages of CPGs as feed-forward components that can be interpreted as feedback predictors for stabilizing gait modulation. Further, their model perfectly replicated the harmonic structure of human gait (that has recently been found based on the so called golden ratio Iosa et al., 2013). Golden ratio is an irrational number at the basis of many biological and physical systems showing a omotetic harmonic structure, and the ratio between durations of stance and swing phases was found to coincide with the golden ratio (Iosa et al., 2013).

The hypothesis of Dzeladini could be supported by the results of two other studies published in this Research Topic. Awai and Curt (2014) reported a loss of intralimb coordination, especially related to the inability of modulating coordination when increasing speed from slow to comfortable, in patients with spinal cord injury. La Scaleia et al. (2014) found that coordination

OPEN ACCESS

Edited and reviewed by:
Hauke R. Heekeren, Freie Universität Berlin, Germany

***Correspondence:**
Marco Iosa, m.iosa@hsantalucia.it

Received: *27 January 2015*
Accepted: *12 May 2015*
Published: *02 June 2015*

Citation:
Iosa M, Gizzi L, Tamburella F and Dominici N (2015) Editorial: Neuro-motor control and feed-forward models of locomotion in humans. Front. Hum. Neurosci. 9:306. doi: 10.3389/fnhum.2015.00306

may be based on a discrete, temporal harmonic cyclic structure, along which, critical points delimiting burst components are shifted. In particular, despite the differences in the segmental level and intensity of the spinal activity, the motor-neurons' activation patterns exhibited two major bursts during different locomotor tasks: one around heel strike and the other around toe off, again in line with a schema of activations strictly related to the harmonic structure of gait. Practical guidelines on the methodological aspects for extracting neural control information in the guise of motor modules through electromyographic muscle activation patterns have been clearly depicted in the study of Oliveira et al. (2014).

Patients with complete sensory-motor lesions have a very limited chance of recovering the ability of walking and even if they recover the ability to ambulate, they are usually limited ambulators. The chances of walking recovery improve in less severe lesions and younger age. Motor and somatosensory evoked potentials can contribute toward diagnosing lesions of different neural structures and predicting the recovery of functional movements, as reported in the review by Scivoletto et al. (2014). The same group contributed also with an interesting study on the effects of enhancing somatosensory inputs through the application of kinesio-taping: spasticity can be reduced and gait ability improved in patients with spinal cord injury (Tamburella et al., 2014). These results are hence in line with the above reported importance of sensory feedback for modulating the rhythmic activity of motoneurons activations (La Scaleia et al., 2014). As described by Scivoletto et al. (2014), during the long and strenuous neurorehabilitation of patients with spinal cord injury, learning-dependent changes in CPG circuits can occur primarily through rhythmic peripheral influences imposed by the exercises. The gait training for these patients can also be improved through visual biofeedback, as reported by Schließmann et al. (2014), or robotic devices as reported in the studies of Del-Ama et al. (2014) and Sylos-Labini et al. (2014). In the former study the muscle examination of patients with spinal cord injury revealed improvements at knee and hip sagittal muscle functioning, the same joints as those found impaired in the study of Awai and Curt (2014). The latter reports the gait of subjects with spinal cord injury using a specifically developed wearable ambulatory exoskeleton (Sylos-Labini et al., 2014).

A comparison of the effects of robotic therapy against those obtained with sensory-based training in subjects with multiple sclerosis is reported in the study of Gandolfi et al. (2014). Multiple sclerosis is a chronic disease of the central nervous system characterized by a progressive decline in various neurological functions, with locomotion disturbances primary related to a reorganization of the postural control system and to deficits of central integration of sensory afferents. Outcomes resulted similar after the two different therapies, with a more pronounced improvement in gait function and balance, for robotic-aided and sensory integration based training, respectively.

The benefits obtained by gait training based on enhancing sensory feedback reported in the above studies suggest that human gait may involve a complex interplay between spinal and cortical circuits.

In fact, not only spinal, but also supraspinal structures are involved during locomotion. It has been shown that they are responsible for initiating (Jiang et al., 2015) and modifying the features of the gait basic rhythm, for stabilizing the upright walking, and for coordinating movements in a dynamic changing environment (Grasso et al., 2004). Furthermore, specific damages of supraspinal structures result in specific alterations of human locomotion, as evident in subjects with brain injuries such as stroke (Clark et al., 2010; Gizzi et al., 2011), brain trauma, or people with cerebral palsy (Iosa et al., 2012), in people with death of dopaminergic neurons in the *substantia nigra* due to Parkinson's disease, or in subjects with cerebellar dysfunctions, such as ataxia (Kirtley, 2006). The role of cerebellum during locomotion has been shown to be related to the coordination and adaptation of movements. Cerebellum is the structure of CNS where the internal models—neural representations miming meaningful aspects of our body, such as input/output characteristics of sensorimotor system—are conceivably developed (Wolpert et al., 1998). Internal model control has been shown to be at the basis of motor strategies for compensating delays or lack in sensorimotor feedback. Some aspects of locomotion require predictive internal control, especially for improving gait dynamic stability, avoiding obstacles, or when sensory feedback is altered or compromised. In their review focused on cerebellar contribution to feed-forward control of locomotion, Pisotta and Molinari (2014) hypothesized that sequence recognition is the mechanism by which the cerebellum facilitates the control of gait. Once again, the repetition of specific events during locomotion embedded into a predictable sequence seems to be a key-factor for facilitating locomotor control.

Sale et al. (2014) showed that, as for subjects with spinal cord injury, repetitive robotic gait training resulted effective also in a group of subjects with progressive supranuclear palsy, a rare neurodegenerative disease that causes the gradual deterioration and death of specific volumes of the brain (in particular midbrain, pallidum, thalamus, subthalamic nucleus, frontal lobes).

The role of cognitive functions during locomotion is still debated, with some authors (e.g., Ruchinskas et al., 2000) suggesting that locomotion is a largely automatized action, and others (Lamoth et al., 2011), who found that stability of gait is altered when cognition is impaired or during dual tasking in frail healthy people. The study of Meester et al. (2014), showed a greater activity in the prefrontal cortex, when a cognitive load was administered to the subject during walking. This adaptation, however, did not detrimentally affect the amplitude of soleus H-reflex or the spatiotemporal variables of gait. Analogously no correlation between walking speed and prefrontal cortex activity was found. On the other hand, the study of Begg et al. (2014) showed how other aspects of walking (e.g., increasing minimum toe clearance in subjects at risk of fall) can be improved through cognitive-motor training, such as a visual biofeedback.

The role of vision in gait is probably the most evident aspect needing the involvement of cortical areas during walking in the surrounding environment. Aprile et al. (2014) found that subjects with strabismus adopt different walking strategies to compensate

their deficits. They found that subjects with exotropia (an expanded visual field), showed larger step width than subjects with esotropia (a reduced visual field), suggesting a specific neurosensorial adaptation of gait with respect to abnormal binocular cooperation. These results are in line with the famous quote "Go where I'm looking, not look where I'm going" by Berthoz in his famous book "The brain's sense of movement," claiming the role of gaze-based feed-forward control involved in locomotion along a desired trajectory (Berthoz, 2000). The title of the study of Iosa and colleagues published in this Research Topic, "The brain's sense of walking…" (Iosa et al., 2014), is a clear tribute to Berthoz's work. In that study, as in the one from Fusco et al. (2014), the ability of imagining walking was under investigation. Motor imagery has been deeply investigated in literature, and it has been defined as a mental representation of an action without its physical execution. Fusco et al. (2014) pointed out three aspects about the intertwine between motor imagery and motor execution of gait actions: (1) they are correlated, but not always coincident; (2) agreement occurred only for some specific usual locomotor tasks (such as forward walking, but not for example for lateral walking); (3) motor execution resulted better simulated during dynamic motor imagery, than during static motor imagery, i.e., when a movement simulating the real one was performed. The study of Iosa et al. (2014) added that dynamic locomotor imagery is less formed in children with typical development and is impaired in children with cerebral palsy.

The abovementioned studies pointed out a number of neural structures involved in locomotion, which seem to paradoxically complicate, instead of simplifying, the management of all sensory and actuator systems necessary for the harmonious execution of human locomotion. However, the results published in this Research Topic appear to converge toward an intrinsic simplification of the problem: the involved neural systems seem to be responsive to the repetitive sequences of events occurring during gait, being facilitated in the generation, control, and prediction of walking by its intrinsic harmonic structure. Despite further studies being needed, neuroscience is giving important suggestions for a more effective neurorehabilitation, and for answering the question that arises when observing the elegant coordination and interplay of movement and balance, joints and muscles, senses and actuation, involved in human walking.

Funding

LG is supported by the EU Project "Integrative approach for the emergence of human like locomotion" (H2R; contract #600698).

References

Aprile, I., Ferrarin, M., Padua, L., Di Sipio, E., Simbolotti, C., Petroni, S., et al. (2014). Walking strategies in subjects with congenital or early onset strabismus. *Front. Hum. Neurosci.* 8:484. doi: 10.3389/fnhum.2014.00484

Awai, L., and Curt, A. (2014). Intralimb coordination as a sensitive indicator of motor-control impairment after spinal cord injury. *Front. Hum. Neurosci.* 8:148. doi: 10.3389/fnhum.2014.00148

Begg, R. K., Tirosh, O., Said, C. M., Sparrow, W. A., Steinberg, N., Levinger, P., et al. (2014). Gait training with real-time augmented toe-ground clearance information decreases tripping risk in older adults and a person with chronic stroke. *Front. Hum. Neurosci.* 8:243. doi: 10.3389/fnhum.2014.00243

Berthoz, A. (2000). *The Brain's Sense of Movement (Translated by G. Weiss)*. Boston, MA: Harvard University Press.

Clark, D. J., Ting, L. H., Zajac, F. E., Neptune, R. R., and Kautz, S. A. (2010). Merging of healthy motor modules predicts reduced locomotor performance and muscle coordination complexity post-stroke. *J. Neurophysiol.* 103, 844–857. doi: 10.1152/jn.00825.2009

Del-Ama, A. J., Gil-Agudo, A., Pons, J. L., and Moreno, J. C. (2014). Hybrid gait training with an overground robot for people with incomplete spinal cord injury: a pilot study. *Front. Hum. Neurosci.* 8:298. doi: 10.3389/fnhum.2014.00298

Dzeladini, F., van den Kieboom, J., and Ijspeert, A. (2014). The contribution of a central pattern generator in a reflex-based neuromuscular model. *Front. Hum. Neurosci.* 8:371. doi: 10.3389/fnhum.2014.00371

Fusco, A., Iosa, M., Gallotta, M. C., Paolucci, S., Baldari, C., and Guidetti, L. (2014). Different performances in static and dynamic imagery and real locomotion. An exploratory trial. *Front. Hum. Neurosci.* 8:760. doi: 10.3389/fnhum.2014.00760

Gandolfi, M., Geroin, C., Picelli, A., Munari, D., Waldner, A., Tamburin, S., et al. (2014). Robot-assisted vs. sensory integration training in treating gait and balance dysfunctions in patients with multiple sclerosis: a randomized controlled trial. *Front. Hum. Neurosci.* 8:318. doi: 10.3389/fnhum.2014.00318

Gizzi, L., Feldbæk Nielsen, J., Felici, F., and Farina, D. (2011). Impulses of activation but not motor modules are preserved in the locomotion of subacute stroke patients. *J. Neurophysiol.* 106, 202–210. doi: 10.1152/jn.00727.2010

Grasso, R., Ivanenko, Y. P., Zago, M., Molinari, M., Scivoletto, G., Castellano, V., et al. (2004). Distributed plasticity of locomotor pattern generators in spinal cord injured patients. *Brain* 127, 1019–1034. doi: 10.1093/brain/awh115

Grillner, S. (1985). Neurobiological bases of rhythmic motor acts in vertebrates. *Science* 228, 143–149. doi: 10.1126/science.3975635

Guertin, P. A. (2014). Preclinical evidence supporting the clinical development of central pattern generator-modulating therapies for chronic spinal cord-injured patients. *Front. Hum. Neurosci.* 8:272. doi: 10.3389/fnhum.2014.00272

Iosa, M., Fusco, A., Marchetti, F., Morone, G., Caltagirone, C., Paolucci, S., et al. (2013). The golden ratio of gait harmony: repetitive proportions of repetitive gait phases. *Biomed Res. Int.* 2013:918642. doi: 10.1155/2013/918642

Iosa, M., Marro, T., Paolucci, S., and Morelli, D. (2012). Stability and harmony of gait in children with cerebral palsy. *Res. Dev. Disabil.* 33, 129–135. doi: 10.1016/j.ridd.2011.08.031

Iosa, M., Zoccolillo, L., Montesi, M., Morelli, D., Paolucci, S., and Fusco, A. (2014). The brain's sense of walking: a study on the intertwine between locomotor imagery and internal locomotor models in healthy adults, typically developing children and children with cerebral palsy. *Front. Hum. Neurosci.* 8:859. doi: 10.3389/fnhum.2014.00859

Jiang, N., Gizzi, L., Mrachacz-Kersting, N., Dremstrup, K., and Farina, D. (2015). A brain-computer interface for single-trial detection of gait initiation from movement related cortical potentials. *Clin. Neurophysiol.* 126, 154–159. doi: 10.1016/j.clinph.2014.05.003

Kirtley, C. (2006). *Clinical Gait Analysis; Theory and Practice*. Philadelphia, PA: Elsevier.

Lamoth, C. J., van Deudekom, F. J., van Campen, J. P., Appels, B. A., de Vries, O. J., and Pijnappels, M. (2011). Gait stability and variability measures show effects of impaired cognition and dual tasking in frail people. *J. Neuroeng. Rehabil.* 8:2. doi: 10.1186/1743-0003-8-2

La Scaleia, V., Ivanenko, Y. P., Zelik, K. E., and Lacquaniti, F. (2014). Spinal motor outputs during step-to-step transitions of diverse human gaits. *Front. Hum. Neurosci.* 8:305. doi: 10.3389/fnhum.2014.00305

Meester, D., Al-Yahya, E., Dawes, H., Martin-Fagg, P., and Piñon, C. (2014). Associations between prefrontal cortex activation and H-reflex modulation during dual task gait. *Front. Hum. Neurosci.* 8:78. doi: 10.3389/fnhum.2014.00078

Oliveira, A. S., Gizzi, L., Farina, D., and Kersting, U. G. (2014). Motor modules of human locomotion: influence of EMG averaging, concatenation, and number of step cycles. *Front. Hum. Neurosci.* 8:335. doi: 10.3389/fnhum.2014.00335

Pisotta, I., and Molinari, M. (2014). Cerebellar contribution to feedforward control of locomotion. *Front. Hum. Neurosci.* 8:475. doi: 10.3389/fnhum.2014.00475

Ruchinskas, R. A., Singer, H. K., and Repetz, N. K. (2000). Cognitive status and ambulation in geriatric rehabilitation: walking without thinking? *Arch. Phys. Med. Rehabil.* 81, 1224–1228. doi: 10.1053/apmr.2000.6976

Sale, P., Stocchi, F., Galafate, D., De Pandis, M. F., Le Pera, D., Sova, I., et al. (2014). Effects of robot assisted gait training in progressive supranuclear palsy (PSP): a preliminary report. *Front. Hum. Neurosci.* 8:207. doi: 10.3389/fnhum.2014.00207

Schlieβmann, D., Schuld, C., Schneiders, M., Derlien, S., Glöckner, M., Gladow, T., et al. (2014). Feasibility of visual instrumented movement feedback therapy in individuals with motor incomplete spinal cord injury walking on a treadmill. *Front. Hum. Neurosci.* 8:416. doi: 10.3389/fnhum.2014.00416

Scivoletto, G., Tamburella, F., Laurenza, L., Torre, M., and Molinari, M. (2014). Who is going to walk? A review of the factors influencing walking recovery after spinal cord injury. *Front. Hum. Neurosci.* 8:141. doi: 10.3389/fnhum.2014.00141

Svevo, I. (1923). *La Coscienza di Zeno*, Trieste: Licinio Cappelli Editore.

Svevo, I. (1989). *Confessions of Zeno*. New York, NY: Vintage Books.

Sylos-Labini, F., La Scaleia, V., d'Avella, A., Pisotta, I., Tamburella, F., Scivoletto, G., et al. (2014). EMG patterns during assisted walking in the exoskeleton. *Front. Hum. Neurosci.* 8:423. doi: 10.3389/fnhum.2014.00423

Tamburella, F., Scivoletto, G., and Molinari, M. (2014). Somatosensory inputs by application of KinesioTaping: effects on spasticity, balance, and gait in chronic spinal cord injury. *Front. Hum. Neurosci.* 8:367. doi: 10.3389/fnhum.2014.00367

Wolpert, D. M., Miall, R. C., and Kawato, M. (1998). Internal models in the cerebellum. *Trends Cogn. Sci.* 2, 338–347. doi: 10.1016/S1364-6613(98)01221-2

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.

frontiers in
HUMAN NEUROSCIENCE

REVIEW ARTICLE
published: 30 May 2014
doi: 10.3389/fnhum.2014.00272

Preclinical evidence supporting the clinical development of central pattern generator-modulating therapies for chronic spinal cord-injured patients

Pierre A. Guertin [1,2] *

[1] *Department of Psychiatry and Neurosciences, Laval University, Quebec City, QC, Canada*
[2] *Spinal Cord Injury and Functional Recovery Laboratory, Laval University Medical Center (CHU de Quebec), Quebec City, QC, Canada*

Edited by:
Marco Iosa, Fondazione Santa Lucia, Italy

Reviewed by:
Auke Ijspeert, Ecole Polytechnique Federale de Lausanne (EPFL), Switzerland
Yuri P. Ivanenko, IRCCS Fondazione Santa Lucia, Italy

****Correspondence:***
Pierre A. Guertin, Spinal Cord Injury and Functional Recovery Laboratory, Laval University Medical Center (CHU de Quebec), 2705 Laurier Boulevard, Room RC-9800 (Neuroscience Unit), Quebec City, QC G1V 4G2, Canada
e-mail: Pierre.Guertin@crchul.ulaval.ca

Ambulation or walking is one of the main gaits of locomotion. In terrestrial animals, it may be defined as a series of rhythmic and bilaterally coordinated movement of the limbs which creates a forward movement of the body. This applies regardless of the number of limbs—from arthropods with six or more limbs to bipedal primates. These fundamental similarities among species may explain why comparable neural systems and cellular properties have been found, thus far, to control in similar ways locomotor rhythm generation in most animal models. The aim of this article is to provide a comprehensive review of the known structural and functional features associated with central nervous system (CNS) networks that are involved in the control of ambulation and other stereotyped motor patterns—specifically Central Pattern Generators (CPGs) that produce basic rhythmic patterned outputs for locomotion, micturition, ejaculation, and defecation. Although there is compelling evidence of their existence in humans, CPGs have been most studied in reduced models including *in vitro* isolated preparations, genetically-engineered mice and spinal cord-transected animals. Compared with other structures of the CNS, the spinal cord is generally considered as being well-preserved phylogenetically. As such, most animal models of spinal cord-injured (SCI) should be considered as valuable tools for the development of novel pharmacological strategies aimed at modulating spinal activity and restoring corresponding functions in chronic SCI patients.

Keywords: CPG, locomotion, SGE, ejaculation, LDC, defecation, SMC, micturition

INTRODUCTION

Locomotion is the act of self-propulsion by an animal (Hugues and Wiersma, 1960; Delcomyn, 1977; Kandel et al., 2000; Hopper and DiCaprio, 2004). Forms of terrestrial locomotion generally include walking, running, and hopping. In vertebrates, its control depends upon several neural systems that ensure propulsion, body orientation (equilibrium or postural control), and steering (goal-direction control) (Ivanenko et al., 2006). Certain areas of the brain have for role, through signals sent via descending neural pathways to the spinal cord, to trigger and modulate basic locomotor outputs generated spinally. The latter are organized essentially by a network localized in the lumbar segments of the spinal cord, generally referred to as the Central Pattern Generator (CPG) for locomotion. That network is responsible for much of the timing and pattern of the complex, rhythmic, coordinated muscle activities that underlie locomotion. In addition to this direct contribution from central systems, there are also different feedback and feedforward loops that use peripheral cues for proper adaptation of gait under different circumstances. Since most methods and experimental tools needed for studying these systems at the cellular level are rather invasive, it has remained difficult to study them in great details in humans. However, research in different animal models has revealed significant details about the organization and function of each constitutive element of these systems (Buschges et al., 2008). In most cases, great similarities have been found between neuronal circuits that generate rhythmic motor patterns among species. In fact, just examining similarities between various forms of locomotion among species already provide preliminary evidence of well-preserved spinal mechanisms throughout evolution. The neural system controlling locomotion is probably the most studied systems thus far among all spinal systems underlying comparable stereotyped motor behaviors such as micturition, ejaculation or defecation (Guertin and Steuer, 2009). Nonetheless, from compiling data gathered about these other systems more recently, it may be said that great similarities seem to exist among spinal systems that control these fundamental behaviors.

Abbreviations: CPG, Central Pattern Generator; SMC, Sacral or Spinal Micturition Center; LDC, Lumbosacral Defecation Center; SGE, Spinal Generator for Ejaculation; TX, Spinal Cord Transection; SCI, Spinal Cord Injury; NMDA, N-Methyl-D-aspartate; 5-HT, Serotonin; DA, Dopamine; CNS, Central Nervous System; EMG, Electromyogram.

DIFFERENT FORMS OF LOCOMOTION AMONG VERTEBRATE SPECIES

Although some differences exist between forward and backward locomotion in cats, mice humans, salamanders or lampreys, to name a few, most of the basic alternating patterns between flexor-like and extensor-like muscles are generally maintained even in invertebrate species (Székely et al., 1969; Pearson and Duysens, 1976; Kristan and Weeks, 1983; Robertson and Pearson, 1985; Pearson, 1993; Currie and Lee, 1997; Cheng et al., 1998; Currie, 1999; Matsumoto et al., 2007; Liu et al., 2012). For instance, lampreys and salamanders swim by contracting muscles on either side of the body in order to generate waves of flexion that travel the length of the body from nose to tail (rostrocaudal propagation of undulations), generally getting larger as they go along (Getting, 1977). Locomotion on land raises different complications, such as the effects of gravity. Nonetheless, walking—the most common gait in legged animals, shares comparable key features with swimming. Indeed, it is essentially composed of a series of rhythmic contractions between antagonist muscles of the main moving segments, the legs instead of the whole body. More specifically, it is the result of successive coordinated contractions between flexors and extensors of all limbs accompanied of bilateral alternation for bipeds and quadrupeds.

SEVERAL NEURAL NETWORKS INVOLVED

As mentioned earlier, it is now generally accepted that the basic motor patterns and rhythms underlying walking (i.e., the most extensively studied form of locomotion) are organized and generated in the spinal cord by the CPG (Grillner, 1981; Guertin, 2009a). However, many other networks in the CNS have key roles to play for succesfull locomotion to be achieved. For instance, supraspinal centers in the brainstem and forebrain are essential for initiating and controlling locomotor movements. One of them located in the cerebellum plays an important in sensorimotor control and in intra- and inter-limb coordination. The vermis and the intermediate region of the cerebellum receive information through the spinocerebellar pathways about the ongoing activities in the CPG and the somatosensory receptors. The information is conveyed to Purkinje neurons, transformed, returned to brainstem descending tract neurons also involved in the initiation of locomotion. Another important center is the mesencephalic locomotor region (MLR), first discovered in cats and later found in all vertebrate species tested to date (Orlovskii et al., 1966; Orlovski et al., 1966; Orlovsky and Shik, 1976; Jordan et al., 2008; Juvin et al., 2012). The reticulospinal tract that originates in the reticular formation has also been found to be essential for the production of locomotor activity evoked by brainstem stimulation in many different species, its activation is necessary for the initiation of locomotion in normal conditions (Grillner, 1981; Cheng and Magnuson, 2011; Le Ray et al., 2011). Along this idea, locomotor-initiating centers in the midbrain/pontine tegmentum and cerebellum converge in the reticular nucleus before descending to the spinal cord.

SPINAL CORD ORGANIZATION

It is impossible to provide an update on neural systems that control locomotion or other stereotyped motor behaviors, specifically those in the spinal cord, without reviewing first, fundamental details about spinal cord organization (Guertin, 2012). The spinal cord has often been considered as a simple relay between brain cells and effective organs (muscles, skin, etc.). However, it is increasingly recognized that the spinal cord is also a "command center" involved in the control and modulation of several functions (Kandel et al., 2000). With its one billion neurons (Kalat, 1998), it is definitely a significant structure of the CNS that can control both simple motor acts such as reflexes (e.g., monosynaptic excitatory, reciprocal inhibitory, withdrawal and crossed-extension reflexes) as well as more complex motor functions such as locomotion, bladder/bowel control, and sexual function (Guertin, 2012).

GROSS ANATOMY OF THE SPINAL CORD

The spinal cord constitutes the most caudally located structure of the CNS (Netter, 2006). Contained within the vertebral column, it extends from the medulla to the first lumbar vertebra (Patestas and Gartner, 2006). In humans, its long and thin elliptical structure varies in length between 43 and 45 cm comprising 31 segments—8 cervical, 12 thoracic, 5 lumbar, 5 sacral, and 1 coccygeal. Every segment is associated with a pair of nerves (and roots) on each side that comprises sensory nerve roots entering the spinal cord at each level, and motor roots emerging from the cord at each level. In rostral parts, the spinal nerves exit directly from (just above from C1 to C7 or just below from C8 and lower) the vertebra associated numerically with the corresponding spinal cord segment. However, in caudal parts of the spinal cord, the spinal nerves travel further down the column before exiting (also known as the cauda equine). In transverse sections, the spinal cord displays white and gray matter tissues. The white matter, located more peripherally, contains white matter tracts, ascending and descending myelinated fibers, carrying both sensory and motor inputs. The gray matter, more centrally located, is characterized by its butterfly-shape that contains unmyelinated cells and, specifically, simple as well as more complex spinal circuits. In the center, there is the central canal that contains cerebrospinal fluids formed mainly in the ventricular system (choroid plexus). Peripherally, the meninges are layers of tissue surrounding the spinal cord for its protection—the dura, the arachnoid and the pia. The latter is relatively thin and tightly associated with the surface of the spinal cord. The spinal cord is well-vascularized with blood vessels including the anterior spinal artery, the bilateral sulcal branches, the bilateral posterior spinal arteries, the pial arterial plexus as well as the anterior/posterior spinal veins, the anterior/posterior sulcal veins, and the pial venous plexus.

SPINAL CORD WHITE MATTER

Several tracts carry information between supraspinal (MLR, reticular formation, etc.) and spinal structures (e.g., motoneurons, simple reflexes, CPG, etc.). Ascending tracts convey sensory signals associated with the sense of touch, pressure, proprioception, and vibration via relatively large myelinated fibers (e.g., gracile fasciculus, cuneate fusciculus). They travel contralaterally (decussation) in the medulla prior to be redirected toward the thalamus and the sensory cortex. In contrast, the lateral or so-called anterolateral columns convey information about pain and

thermal sensation and decussate instead at the spinal cord level (close primary afferent entry) prior to ascend to the brain via a number of tracts (e.g., Lissauer's tract, dorsospinal tract, ventrospinal tract, spinocerebellar tract, spinothalamic tract, etc.).

The descending motor system is also divided in multiple tracts essentially composing the so-called pyramidal and extrapyramidal tracts. The corticospinal tracts, also called the pyramidal tracts, contain about one million axons on each side involved mainly in skilled movements. The extrapyramidal tracts are involved instead in the control of posture and locomotion (Bretzner and Drew, 2005; Barthélemy et al., 2011). The pyramidal tract originates from the cerebral cortex and from some brainstem motor nuclei. It constitutes the most direct descending motor pathway (e.g., some monosynaptic connections) between the motor cortex (Brodmann's areas 1, 2, 3, 4, and 6) and the final common motor pathway, namely the spinal motoneurons all segmental levels. About 80–90% of these corticospinal axons decussate contralaterally at the pyramid level in the medulla oblongata. From there, they form the lateral corticospinal tract that sends input to spinal motoneurons in the ventral horn. 10–20% descends instead ipsilaterally as the ventral corticospinal tract and decussates in the spinal cord prior to synapsing with spinal motoneurons. A few others form instead the corticobulbar tract that sends input to brainstem motoneurons for face, head and neck muscle control. Extrapyramidal tract neurons (rubrospinal, vestibulospinal, tectospinal and reticulospinal) originate from subcortical nuclei in the pons (reticular formation) and medulla oblongata where they send projections to spinal motoneurons of all segments. The rubrospinal tract receives inputs from the motor and premotor areas 4 and 6 prior to send projections contralaterally (decussation in the brain) to spinal motoneurons cervically. It is generally considered to facilitate flexion and inhibit extension in the upper extremities. The tectospinal tract sends axons in premotor lamina of the spinal cord (VI-VIII) cervically for neck and head motor control whereas the reticulospinal track sends projections to all levels of the spinal cord for autonomic control (e.g., cardiovascular and respiratory functions, blood pressure, micturition, defecation). The vestibulospinal tract sends projections laterally or ventrally on the ipsilateral side of the spinal cord to laminae VII and VIII for extensor control (English, 1985).

SIMPLE REFLEX PATHWAYS OF THE GRAY MATTER

As described above, white matter tracts are mainly "relays" sending signals up (sensory-related) and down (motor control) between supraspinal and spinal structures. In contrast, gray matter neurons in the spinal cord form reflex pathways and complex neuronal networks. Simple reflex arcs or pathways are normally associated with well-known classical reflexes (Matthews, 1972, 1991). By definition, a simple reflex, in normal condition, typically leads to a rapid, predictable, repeatable, stereotyped and involuntary motor reaction in response to the corresponding specific stimulus. Most reflexes are mediated locally, within one or two spinal segments, by relatively simple neural pathways in the spinal cord—involving generally either one (monosynaptic), two (disynaptic) or more (polysynaptic) synapses and corresponding neurons (i.e., interneurons and motoneurons). Simple reflexes may be of autonomic (related with inner organs, eyes, blood vessels, etc.) or somatic (related with skeletal muscle responses) origin. The latter has been more extensively studied mainly in the spinal cord-transected or decebrate cat models. Among the main somatic spinal reflexes, the Ia, Ib, II, and FRA (flexion reflex afferent) reflexes have been particularly well-characterized although their roles in the control of locomotion or other stereotyped motor behaviors remain incompletely understood (Henneman, 1974; Forssberg and Hirschfeld, 1988; Field-Fote et al., 2012). This said, activation of some of these reflexes via electrical stimulation of muscles, nerves or lumbar spinal cord area in spinal cord-injured (SCI) patients was shown to temporarely elicit or promote some waking movements with or without pharmacological aids (see recent work from Edgerton's or Gerasimenko's groups) (Edgerton et al., 2001).

MONOSYNAPTIC REFLEX

The so-called Ia monosynaptic reflex arc is a well-characterized somatic spinal reflex pathway. It is considered the simplest and fastest reflex of all, mediating primary afferent (Ia) inputs from muscle spindles typically activated by muscle stretch (e.g., following a tendon jerk, tendon tap or myotatic reflex). Ia inputs establish monosynaptic connections with homonymous alpha-motoneurons in the ventral horn gray matter. This excitatory reflex increases homonymous muscle contraction in response to muscle elongation while inhibiting antagonist muscle contraction via collateral branching. It is generally considered to play a role in tonus and postural adjustments which is important for terrestrial locomotion. Its level of activity is phase and task-dependent—i.e., larger response during standing compared with walking or running as well as during extension (stance phase) compared with flexion (swing phase) (Stein and Capaday, 1988). Findings in decerebrate and paralyzed cats provided clear evidence of its role also in reflexively-increased extensor activity during stance and in resetting rhythm or cycle probably via direct input upon CPG elements (Guertin et al., 1995; Angel et al., 1996, 2005). Hence, Ia inputs during locomotion may serve to compensate for an unsuspected increase of loading during ambulation as well as to increase tonus in extensor muscles during stance (Guertin et al., 1995; Pearson, 1995; Guertin, 1996).

Ib REFLEX PATHWAYS

Also known as the inverse myotatic reflex or autogenic inhibitory reflex pathway, the Ib reflex arc is associated with peripheral afferent inputs from Ib afferent fibers and Golgi tendon organs. It has been shown to inhibit homonymous and synergistic alpha-motoneurons at rest via a disynaptic arc (two synapses involving one inhibitory neuron called the Ib interneuron), originally believed to serve as a protective mechanism against excessive muscle contraction. This view has eventually changed when discovering that a wide range of muscle activity and load can alter Ib firing. It is also task-dependent since during locomotion, Ib inhibition is replaced by Ib excitation to homonymous and synergistic alpha-motoneurons at all joints of the lower extremities via a rhythmically active candidate excitatory interneuron located in the lumbar enlargement area (lamina VII) (Gossard et al., 1994; Angel et al., 1996, 2005). Its role during locomotion may be also

to enhance muscular contraction of extensors during the stance phase and to reset stepping to extension when activated during the swing phase (Guertin et al., 1995; Pearson, 1995).

FLEXION (WITHDRAWAL) REFLEX PATHWAYS

The FRA pathway is activated specifically by high-threshold afferent fibers (e.g., associated with cutaneous nociceptor A or C fibers, group II, III and IV muscle afferent fibers, etc.) (Knikou et al., 2009). Ipsilaterally, it involves at least two interneurons (three or more synapses) over several segments prior to synapse upon alpha-motoneurons ipsilaterally for extended flexor contraction and extended extensor inhibition. With weaker stimulus, only ipsilateral effects are found. However, with stronger stimulus, opposite contralateral effects may also be found (excitation of extensor muscles and inhibition of flexors)—the crossed-extension reflex. These reflex pathways play a role in reflex withdrawal of a limb (unilateral flexion) from a painful stimulus. Task-dependency has also been found since, during locomotion, a long-lasting burst of activity is unraveled ipsilaterally during pharmacologically-induced fictive locomotion (long-lasting FRA response) probably via CPG elements (specifically the flexor portion of the CPG) since FRA stimulation under experimental conditions was shown to reset the step cycle to flexion (Jankowska et al., 1967a,b; Perreault et al., 1995; Schomburg et al., 1998; Ollivier-Lanvin et al., 2011). Clinically, it is associated with the Babinski sign (i.e., tongue depressor-induced plantar extension) as an indication of neurological problems in adults (e.g., spinal pyramidal tracts-induced injury caused by trauma or tumor). The long-lasting FRA response as well as myoclonus can also be uncovered following FRA stimulation in patients with SCI.

CENTRAL PATTERN GENERATORS—EARLY EVIDENCE AND UNDERLYING CONCEPTS

To date, the best-characterized spinal network is undoubtedly the CPG for locomotion that directly controls the basic motor commands underlying ambulation (Guertin, 2009a; Guertin and Steuer, 2009). Seminal work from Flourens, Phillipson, Sherrington, and Graham Brown supported by subsequent evidence generated largely from the 1960s onwards showed that, across species, rhythmic and stereotyped motor behaviors including walking, flying, and swimming are controlled largely by a neuronal network generally referred to as the CPG for locomotion (Flourens, 1824; Freusberg, 1874; Sherrington, 1910; Graham Brown, 1911, 1914; Lhermitte, 1919; Grillner, 2006; Clarac and Pearlstein, 2007). Early observations from paraplegic dogs revealed the existence of locomotor-like movements that can occur spontaneously after a complete transection (TX) of the spinal cord. That was elicited specifically when dropping one of the limbs from a flexed position. Comparable observations by Philippson led him to conclude that the spinal cord could control locomotion using both central and reflex mechanisms. Sir Charles Sherrington's work in TX cats and dogs provided additional evidence that such spinal locomotor-like movements were the result of reflex actions from proprioceptors onto some spinal centers (Sherrington, 1910). However, it is Thomas Graham Brown, who described more directly the existence of a spinal neuronal network as main command center for locomotion (see also Stuart and Hultborn, 2008 for a thorough description of Sherrington and Graham Brown's original contributions) in anesthetized animals lying on one side when stepping movements in the hindlimbs were spontaneously expressed ("narcosis progression") after TX thoracically (Graham Brown, 1911, 1914; Stuart and Hultborn, 2008). More evidence of its existence cellularly (half-center now generally referred to as the CPG) occurred when intracellular recordings became possible in the 1960s. A Swedish group led by Anders Lundberg recorded interneurons located in the lumbar segments of the spinal cord (lamina VII), active following FRA stimulation. Since then, several theories have been proposed to tentatively describe the functional organization of that CPG—the Miller and Scott (1977) hypothesis involving Renshaw cells, the "ring" model to explain more complex locomotor patterns (e.g., backward vs. forward walking), the flexor burst generator with an assymetrical excitatory drive from a flexor burst generator as well as the unit burst generator model with symmetrically-organized burst generators (for each articulation or sets of muscles) even in absence of peripheral input. More recently, the "synergy model" from Bizzi's group and the bipartite model (or two-level CPGs) from McCrea's group were also proposed (Tresch et al., 1999; Bizzi et al., 2008; McCrea and Rybak, 2008).

CONCEPTUAL ORGANIZATION OF CPG FOR LOCOMOTION

HALF-CENTER MODEL

Sherrington used the term "half-center" to explain the spinal pathway for reciprocal inhibition. That expression was used again by Graham Brown in his model of spinal locomotor control based on his experiments showing that the basic pattern for stepping was generated entirely in the spinal cord even in absence of peripheral afferent contribution (due to narcosis) in spinal cord-transected cats, rabbits and guinea-pigs (Graham Brown, 1911, 1914). The animals, under general anesthesia, were lying on one side when stepping movements in the hindlimbs were spontaneously evoked ("narcosis progression") after a transection of the cord at the lower thoracic level. Since the level of anesthetic used was shown to abolish proprio- and extero-ceptive reflexes but not locomotor activity, Graham Brown proposed a "half-center" model made of two groups of spinal neurons reciprocally organized and mutually inhibiting each others that were capable of producing the basic rhythm and pattern for stepping. Activity in the first group of neurons (e.g., extensor half-center) would send motor commands to motoneurons (exciting extensors), and would inhibit simultaneously the reciprocal group of neurons (flexor half-center) preventing the excitation of antagonists (silencing flexors). After a period of "depression" (e.g., fatigue, adaptation, post-inhibitory rebound) of the extensor half-center, the flexor half-drive would predominate for a new phase of activity. Despite these findings, the general opinion of scientists between 1920 and 1960 remained that basic locomotor activity largely depends upon sensory input from the peripheral nervous system. However, it is Elzbieta Jankowska and Anders Lundberg who have provided in the 1960s using intracellular recording techniques the first direct evidence supporting the existence of Graham Brown's model

(Jankowska, 2008). They identified intracellularly interneurons located in the lumbar segments of the cord (specifically in the lamina VII) that are active following FRA stimulation. Specifically, one group of neurons was found to be activated by ipsilateral FRA, a second group by contralateral FRA (coFRA), and a third group by both ipsi- and contralateral stimulation. After injection of L-DOPA and nialamide in spinal cord-transected cats, FRA stimulation evoked a high frequency burst followed by a long-lasting self-sustained series of discharges. Some neurons did not even show any short latency effects during the stimulus train. These interneurons were found to be monosynaptically excited by ventro-lateral funiculus stimulation which contains descending fibers from the reticular formation. One of the most important features was the reciprocal organization between these groups of interneurons since coFRA stimulation abolished the long-latency discharges evoked by ipsilateral FRA and vice versa. Finally, it was proposed that Ia interneurons could participate in the production of the locomotor pattern by receiving strong excitatory input from FRA interneurons given their corresponding rhythmic activity in L-DOPA-treated cats (Jankowska et al., 1967a,b). However, in the 1970s, several neuroscientists began to provide evidence suggesting that, as it is, this half-center organization can not fully explain the complex patterns of muscle activation found during terrestrial locomotion (e.g., in quadrupeds) (Grillner, 1981).

MILLER AND SCOTT MODEL

Sharing similarities with the half-center model, the Miller and Scott hypothesis proposed that Renshaw cells rather than fatigue are responsible for the alternation between flexion and extension (Miller and Scott, 1977). Increasing activity in one pool of motoneurons (e.g., extensors) would be gradually inhibited by a corresponding increase of recurrent inhibition. Architecturally, this model takes into account known neuronal connections—it is constituted of a closed chain of neurons to which flexor and extensor motoneurons are connected in different parts. Renshaw cells and Ia inhibitory interneurons which are part of this chain of neurons are mainly responsible for reciprocal activation of the flexor and extensor motoneurons (Bergmans et al., 1969). Simultaneously, Renshaw cells would remove reciprocal inhibition of antagonists (recurrent facilitation) via their spindle Ia monosynaptic inhibitory input onto Ia interneurons allowing the flexor excitatory drive to take over for a new phase of activity (i.e., flexion). Interestingly, by varying the tonic input to the alpha motoneurons and Ia inbitory interneurons, coactivation of the flexors and extensors may be achieved. Although, the reciprocity between flexors and extensors is nicely explained by this model, the origin of the rhythmicity itself is rather unclear (Kriellaars, 1992; Kriellaars et al., 1994). Other criticisms came from results showing that Renshaw cell activity may be inhibited during locomotor activity although discrepancies were reported during fictive locomotion whereas both classes of neurons were reported not to be essential for the production of a basic locomotor pattern in motoneurons. Other evidence against the Miller and Scott model was provided by Jordan's group who showed that the basic locomotor pattern and Ia interneuron activity remain after i.v. injection of the nicotinic antagonist mecamylamine (MEC), which greatly reduces Renshaw cell activation (McCrea et al., 1980; reviewed in Guertin, 2009a).

RING MODEL

The highly conceptual "ring" model was one of the models subsequently proposed to explain the existence of complex locomotor patterns (e.g., taking into account differences between backward or forward walking, synchronization and phase coupling between activity of the ring and the cyclic afferent input, etc.) (reviewed in Guertin, 2009a). It is made of a closed chain of at least five groups of neurons (e.g., 2 pure extensors, 2 pure flexors and 1 bifunctional) that project to the motoneurons either directly or through specific interneurons. The sequence of these projections determines the order of activation of various muscles during the step cycle. At rest, a number of ring neurons are tonically inhibited by a certain group of spinal neurons. Activation of the monoaminergic descending system would result in the inhibition of the inhibitory neurons with consequent disinhibition of the ring neurons. Activity within the ring is based on a cyclically propagated inhibitory drive that would travel at different speeds from one group to the other depending on the excitability (e.g., modulated by afferent input) of the path ("ring") interconnecting them. The activity within the ring starts when the excitability level is raised (through disinhibition), so the neurons would discharge when not inhibited. A slow-propagated drive would activate neurons of a group for a longer period of time (e.g., pure extensor during the stance phase) whereas a fast-propagated one would activate a group of neurons for a brief moment in motoneurons to bifunctional muscles. However, this highly conceptual model has generally failed to convince most scientists in this field.

FLEXOR BURST GENERATOR MODEL

During those same years, other models such as the flexor burst generator were also proposed. Pearson and Duysens introduced this model for insects and cats (Pearson and Duysens, 1976; Duysens, 1977; Duysens et al., 2013). It consists of a rhythmic excitatory drive from the flexor burst generator to populations of flexor motoneurons. The burst generator would inhibit, via an inhibitory interneuron, the activity of extensors otherwise activated during the flexor silence by a tonic excitatory input. This asymmetrical model was abandoned later on in favor of a more symmetrical bipartite organization (i.e., in which both extensor and flexor portions are equally driven). This change in views is likely related to the subsequent description of a powerful feedback system associated with ankle extensor group I afferents that can reset rhythms and strongly excite most pools of hindlimb extensor motoneurons during locomotion. This said, recent data mainly from Brownstone's group provided evidence suggesting that an asymmetrical CPG organization should perhaps be re-considered. They proposed that a rhythm-generating layer, composed of a kernel of heterogeneous and electrotonically-coupled neurons, would project directly to the flexor half-center of the pattern formation layer (Brownstone and Wilson, 2008).

UNIT BURST GENERATOR MODEL

In the 70s and early 80s, the unit burst generator model contributed to the demonstration of a symmetrically-organized

generator in the spinal cord that can produce the basic pattern of motor commands for walking even in absence of peripheral input (Edgerton et al., 1976; Grillner, 1981). It was proposed essentially to explain that locomotion is not only a strictly alternating pattern of flexor and extensor activity (requiring all motoneurons to belong to one of these two groups) as proposed by the half-center model (initially by Graham Brown and subsequently by Lundberg et al.). Patterns of locomotor activity are often complex and may include some motoneuron pools that display activity during both the flexion and extension phases of the step cycle or that display differences in the onset and offset of activity in individual flexor and extensor pools. The persistence of such complex activity patterns following bilateral deafferentation of the hindlimbs in decerebrate cats, led Grillner and Zangger (1975) to conclude that the locomotor CPG does not simply generate an alternating activation of flexors and extensors but a more complex pattern that will sequentially start and terminate disctinctively, the activity in the appropriate muscles (Grillner and Zangger, 1974). Their idea was further developed in a proposal for a CPG architecture in which separate "modules" or unit burst generators would control subsets of motoneurons. First, Szekely et al. showed that the locomotor pattern (in the forelimbs) in freely moving newts was similar before and after a bilateral section of the dorsal roots (Székely et al., 1969). This was also shown in decerebrate cats leading then to the suggestion that a CPG could exist for each joint of each limb. Activity from these "units" would be tightly coupled during "normal" walking but individually controlled by supraspinal input to produce different types of motor patterns. This model emerged after analyzing more complex patterns of locomotor activity such as backward walking, climbing, etc. For instance, it was occasionally observed during fictive locomotion that one hindlimb motor nerve can display tonic activity while the others display a normal rhythmic pattern. Also, the activity of pluriarticular muscle nerves such as *semitendinosus* is sometimes in phase with extensors, or flexors, or both, which some authors found difficult to explain with a half-center type of model. Along this idea, Bizzi's group provided experimental and analytical results suggesting instead that sensory-dependent linear combinations of a small number of muscle synergies may generate diverse motor and locomotor patterns. Some of the cellular components of Grillner's CPG model were identified in the 80s using a simpler non-mammalian vertebrate nervous system preparation—the *in vitro* isolated lamprey preparation (Grillner and Wallén, 1985; Grillner, 2006). However, despite the attractiveness of this proposal, the unit burst generator model has not generally explained the existence of other complex patterns of motoneuronal activity such as those found during spontaneous deletions (see section below).

TWO-LEVEL-HALF-CENTER MODELS

Thus far, most models had failed to entirely explain the many patterns that can occur in the generally alternating activity of flexors and extensors during locomotion. In particular, unpredictable changes called "deletions" which refer to periods of silenced activity in some populations of motoneurons (e.g., extensors such as the *soleus*) accompanied of sustained or rhythmic activity in antagonist motoneurons (e.g., flexors such as the *tibialis anterior*) while post-deletion rhythm is generally maintained. This is essentially why attempts to explain these other types of changes have first been proposed based on bipartite CPG levels. Indeed, a detailed analysis of nerve and muscle activity during spontaneous walking in non-paralyzed cats or during fictive locomotor activity in paralyzed decorticated cats let Perret and Cabelguen to initially propose a bipartite or two-level (half-center-like) model that explains the complex biphasic activity in so-called bifunctional motoneurons (e.g., *semitendinosus*) (Perret and Cabelguen, 1980; McCrea and Rybak, 2008; reviewed in Guertin, 2009a). They proposed that not only one half-center but both half-centers (extensor and flexor ones) would send motor commands to bifunctional motoneurons. A variety of motoneuron patterns could be produced by modulating the half-centers output "en route" to these motoneurons. They also suggested that a rhythm generator would be functionally separated from a pattern generator since the rhythm and the amplitude of the locomotor drive potentials appear to be two distinct characteristics that can be independently and spontaneously changing. This paved the way to other studies from Kriellaars and Jordan who proposed a functional separation of pattern and amplitude (Kriellaars, 1992; Kriellaars et al., 1994). They showed that locomotor drive potentials monitored simultaneously (by dual intracellular recordings *in vivo*!) in pairs of motoneurons generally covary in amplitude in homonymous motoneurons while antagonist motoneurons inversely covary. The complex locomotor pattern in bifunctional motoneurons receiving input from both half-centers would be sculpted by controlling the amplitude of the flexor and extensor locomotor drive "en route" to these motoneurons. A similar separation of CPG function (rhythm vs. pattern and amplitude) was suggested in other studies to explain how sensory stimulation can also alter locomotor cycle timing without altering the level of motoneuron activity which has served in the 90s as basis to the elaboration of the most recently proposed multi-level models (2+ and 3 levels, see Rybak's work). Additional evidence from non-resetting deletions reported by McCrea et al. during fictive locomotion and scratch in the decerebrate cat strongly supported also this two-level CPG organization (Lafreniere-Roula and McCrea, 2005). Finally, mathematical models recently developed by McCrea and Rybak have constituted additional supports for the existence of multi-level CPG (half-center-like) organizations that can explain spontaneous deletions and other complex patterns of activity during locomotion (McCrea and Rybak, 2008; Knusel et al., 2013).

All and all, none of the above models have been refuted and yet, according to some, none of the conceptual models is capable to explain the wide variety and diversity of locomotor patterns that exist in real life.

CELLULAR CONSTITUENTS OF THE CPG FOR LOCOMOTION

Beyond these conceptual considerations about its organization, the CPG for locomotion has been characterized in part as a group of interneurons localized mainly in the lumbar area of the spinal cord (Giszter et al., 2007; Guertin, 2009a; Guertin and Steuer, 2009). With an *in vitro* isolated spinal cord preparation from neonatal rats, Kjaerulff et al. used sulforhodamine-101, an

activity-dependent marker/dye, to identify CPG neuron candidates in L1–L6 near the central canal as well as near the medial intermediate zone (Barajon et al., 1992; Kjaerulff et al., 1994). A comparable approach used by Cina and Hochman showed the existence of a restricted number of labeled cells (presumably CPG neuron candidates) more specifically in L1–L5 segmental areas (Hochman et al., 1994; Cina and Hochman, 2000). These findings are supported also by other studies that showed, using electrical stimulation or selective lesions, key rhythmogenic CPG elements specifically in L1 and L2 in mice (Nishimaru et al., 2000). This is also supported by findings from Dimitrijevic et al. (1998) in SCI patients following epidural stimulation near L1–L2 that triggered locomotor-like movements in the lower extremities (Dimitrijevic et al., 1998; Shapkova and Schomburg, 2001; Selionova et al., 2009). Observations in one patient with a complete spinal cord transectionmid-thoracically displaying spontaneous episodes of locomotor-like movements when lying in a bed could be interpreted as valuable evidence of a CPG in the lumbar cord of humans (Nadeau et al., 2010). Discrepancies may exist in other species regarding the exact localization of the CPG (e.g., in mid-lumbar segments in cats) (Langlet et al., 2005). In primitive vertebrate species such as lampreys, cellular components have been identified and even recorded from electrophysiologically nearly 30 years ago (LC cells, CC interneurons, etc.). Indeed, with its simpler neural system, it has been easier to investigate extensively neuronal activity from single spinal cells using the *in vitro* isolated spinal cord preparation from lampreys. The quest for dissecting further the CPGs has been supported also by findings obtained in parallel (or before) from non-mammalian and invertebrate species. In fact, studies on sea slugs, leeches, cockroaches, stick insects and crustacean locomotor (swimmeret) and motor (e.g., somatogastric system) pattern-generating networks have played a pivotal role in understanding further the cellular and network bases of rhythmic motor and locomotor patterns in both invertebrate and vertebrate species (Hugues and Wiersma, 1960; Pearson, 1993; Hopper and DiCaprio, 2004; Clarac and Pearlstein, 2007; Buschges et al., 2008; Harris-Warrick, 2011).

PHARMACOLOGICAL AND ELECTROPHYSIOLOGICAL PROPERTIES OF THE LOCOMOTOR CPG

Since the 1980s, pharmacological manipulations using a plethora of newly available receptor ligands (agonists and antagonists) have largely contributed to understand and further describe the detailed organization of the locomotor CPG (Rossignol et al., 2001; Harris-Warrick, 2011). Experiments in *in vitro* isolated rodents (rat, mouse and turtle preparations) as well as, subsequently, in *in vivo* TX rodents (adult rat and mouse models) led to substantial advances in cellular target identification (receptors and channels) associated with locomotor rhythm generation and modulation (Guertin and Hounsgaard, 1998a,b; Guertin, 2009b). Blood brain barrier (BBB) permeable ligands have served to pharmacologically "dissect" *in vivo* the contribution of specific receptors and channels to locomotor rhythm-generation (Guertin, 2008). Clear CPG-activating effects induced by specific drugs have also been found in various *in vitro* preparations from invertebrate and vertebrate species, although it is beyond the scope of this review to report on all of them (Grillner and Wallén, 1985; Sigvardt et al., 1985; Cazalets et al., 1990; Cowley and Schmidt, 1994; Kiehn and Kjaerulff, 1996; Schmidt et al., 1998; Schmidt and Jordan, 2000; Bonnot et al., 2002; Kiehn et al., 2008). As mentioned earlier, the noradrenergic system and specifically L-DOPA has been among the first systems to be associated with CPG activation in acutely TX cats and rabbits (Jankowska et al., 1967a,b; Viala and Buser, 1969; Grillner and Zangger, 1974). Clonidine, an alpha-2 adrenergic receptor agonist, administered during sensory stimulation (e.g., tail or sexual organ pinching) was also reported to enhance the effects of training and/or sensory stimulation (remains unclear) on locomotor rhythmogenesis in TX cats (Forssberg and Grillner, 1973; Barbeau and Rossignol, 1990, 1991; Pearson and Rossignol, 1991; Chau et al., 1998a,b; Rossignol et al., 2001). However, it has been difficult to determine site-specific actions (e.g., on motoneurons, CPG neurons or primary afferents) from results in many of these earlier studies that were not designed to specifically assess drug-induced CPG activation *per se* (i.e., given the use of additional stimuli including tail stimulation, sexual organ pinching, regular training or weight support assistance, see Lovely et al., 1986; Bélanger et al., 1996; Zhang et al., 2010). More recently, experiments conducted in our laboratory in a mouse model of paraplegia (a complete low-thoracic TX) with no assistance or additional stimuli (e.g., no training, no tail stimulation, no sexual organ pinching, and no weight-support assistance to avoid unspecific non-drug induced effects) have contributed to identify clearly a subset of transmembranal receptors involved in pharmacologically-elicited, CPG-mediated locomotor-like movements in the lower extremities (Guertin, 2008, 2009a,b). For instance, L-DOPA, serotonin (5-HT) or dopamine (DA) receptor ligands such as 8-OH-DPAT, buspirone (5-HT1A/7 agonists), quipazine (5-HT2A/2C agonist) or SKF-81297 (D1-like agonist) were found to trigger significant locomotor-like movements (i.e., rhythmic bilaterally alternating flexions and extensions involving one or several hindlimb joints) whereas others ligands such as 3-Trifluoromethylphenylpiperazine (TFMPP) (5-HT1B), m-CPP (5-HT2B/2C), SR57227A (5-HT3) or clonidine (adrenergic alpha-2 agonist) were shown to elicit mainly non-locomotor movements (i.e., non-bilaterally alternating movements, twitches, cramps, etc.) in TX mice (Guertin, 2004, 2005; Landry and Guertin, 2004; Guertin and Steuer, 2005; in rats, see Antri et al., 2005; Lapointe et al., 2008; Ung et al., 2008). Using selective antagonists and genetically-manipulated animals (e.g., 5-HT7KO mice), it has been clearly established that N-Methyl-D-aspartate (NMDA), 5-HT1, 5-HT7, 5-HT2A and D1 receptors were specifically involved in mediating such CPG-activating locomotor-like effects (Landry et al., 2006a; Lapointe et al., 2009). For instance, endogenous glutamate release and NMDA receptor activation were reported as critically important for quipazine-induced effects since a complete loss of induced movement was found in NMDA antagonist (MK-801)-treated animals previously pretreated with NMDA (Guertin, 2004). Regarding DA receptors, administration of D2, D3 or D4 agonists was found not to generate significant hindlimb locomotor-like movements whereas D1/D5 agonists such as SKF-81297 can potently elicit locomotor-like movements that

are lost in selective D1-like (D1/D5) antagonist-pretreated TX mice but no in D5 −/− paraplegic TX mice suggesting a specific contribution of the D1 subtype to CPG activation (Lapointe et al., 2009). All and all, pharmacological approaches in *in vivo*, untrained and non-assisted TX animals have contributed to identify a subset of CPG-activating compounds (and corresponding receptors confirmed with selective antagonists and knockout animal models) (Guertin, 2009a, 2012). However, none of these molecules were found to generate large amplitude weight bearing stepping movements *per se* in untrained, non-assisted and non-sensory stimulated TX animals suggesting that only partial CPG activating effects can be achieved using these ligands administered separately. Subsequent studies conducted in our laboratory have shown that only simultaneous activation of some of these candidate receptors can, using similar pharmacological approaches and animal models, induce full locomotor-inducing effects. Partial CPG-activating effects (i.e., associated with crawling rather than full weight bearing stepping) induced by some ligands, as mentioned above, were found indeed to turn into full CPG-activating effects (i.e., weight bearing stepping in non-assisted, untrained and non-stimulated paraplegic animals) by simultaneously administrating 8-OH-DPAT, quipazine, L-DOPA, or SKF-81297 (Guertin, 2009b; Guertin et al., 2010, 2011). Prior to these findings, numerous pharmacological studies aimed at improving locomotor function recovery after SCI have been also conducted in other *in vivo* models of SCI (e.g., in TX cats). However, it has remained difficult to consider those results as evidence of cellular targets associated with CPG activation or modulation given the used paradigm (experiments conducted during tail stimulation, sex organ pinching, body-weight supported, movement-assisted manually, etc.) (Forssberg and Grillner, 1973; Lovely et al., 1986; Barbeau and Rossignol, 1991; Bélanger et al., 1996). Some of our findings, reproduced subsequently in other laboratories, have thus been independently validated (Courtine et al., 2011; van den Brand et al., 2012).

This said, the extent to which the cellular network activated pharmacologically *in vivo* corresponds to already identified CPG neuron candidates (electrophysiologically or genetically) remains unclear (see section below). However, dual immunohistochemical experiments recently showed locomotor activity-labeled (c-fos) 5-HT1A-, 5-HT2A- or 5-HT7- positive neurons in the cat lumbar cord (specifically in laminae VII-VIII, Noga et al., 2009) suggesting that some of the locomotor activity-related receptors identified recently in *in vivo* models may indeed be located on CPG neurons (Hochman et al., 2012).

SPECIFIC IONIC CONDUCTANCES AND CHANNELS

This is also an aspect of the CPG that remains incompletely understood. It is generally accepted that one of the main cellular features expected to be found in CPG neuron candidates is its capacity to express endogenously rhythmic activity during fictive or real locomotion (Eken et al., 1989; MacLean et al., 1997; Brownstone and Wilson, 2008; Brocard et al., 2010). For several decades, it was also generally accepted that some neurons of the CPG were expected to express specifically autorhythmic or pacemaker-like properties given that locomotion is, by definition, a rhythmic motor behavior. There have been indeed several cell populations in the corresponding area of the CPG (i.e., lumbar segments in mammalian species) shown in the last few decades to be capable of expressing pacemaker-like properties in specific conditions such as in the presence of NMDA alone or combined, at lower dose, with serotonin and DA (e.g., bath-applied in the case of *in vitro* isolated spinal cord preparations) (Wang et al., 2006; Grob and Guertin, 2007). Plateau potential is another intrinsic property believed by some researchers to be associated with endogenous (tetrodotoxin-resistant) pacemaker-like activity (Eken et al., 1989). Transmembranal currents and channels underlying both of these properties (voltage oscillations, NMDA ionophore, L-type (CaV1.3) Ca^{2+} channel, persistent Na^{2+} current, I_h current, I_A current, I_{CAN}, I_{NaP} current, $I_{K(Ca)}$ current, post-inhibitory rebound have been relatively well-characterized in lamprey, turtle, tadpole, rodent and cat preparations with some species-dependent specificities (Grillner and Wallén, 1985; Reith and Sillar, 1998; Harris-Warrick, 2002, 2011; Grillner, 2006; Wang et al., 2006; Grob and Guertin, 2007). Nonetheless, the specific contribution of these properties (in motoneurons and some interneurons) to real life locomotion remains speculative and a source of debate.

GENETICALLY-IDENTIFIED NEURONS OF THE CPG FOR LOCOMOTION

In recent years, advances in genetics and transgenic murine models have largely contributed to the identification of specific CPG neuron candidates *per se* (Ginty et al., 1992; Lanuza et al., 2004; Goulding, 2009). Targets initially identified in the developing neural tube (i.e., 11 distinct populations of spinal neurons (dI1–dI6, V0–V3, VMN) based on the expression of transcription factors (e.g., Jessell's work) have largely contributed to these findings. Several populations of interneurons involved in locomotor activity were indeed characterized genetically (e.g., V0–V3). One of them is the population of V0 interneurons that was associated with left-right alternation since mice without V0 interneurons (lacking the transcriptional factor Dbx1) displayed bilateral synchrony rather than bilateral alternation during locomotion. Another population referred to as the V1 inhibitory interneurons (expressing the transcription factor Engrailed 1) was associated with high locomotor frequencies since slow rhythms were found in En1-DTA mice (Gosgnach et al., 2006). Other genetically-identified populations include the Chx10-expressing cells (V2a glutamatergic and V2b gabaergic interneurons) located in the intermediate zone of the gray matter in the lumbar spinal cord (Lundfald et al., 2007; Crone et al., 2008). These neurons were associated with frequency, amplitude and bilateral coordination since all of these parameters were affected in Chx10-DTA mice (lacking V2a interneurons). V3 interneurons (Sim-1 expressing cells) constitute another recently identified population shown to participate in the production of a robust and balanced rhythm during locomotion. Indeed, rhythmic activity was found to be partially disrupted in mice lacking V3 interneurons (Zhang et al., 2008). Genetically-engineered animals were also utilized to show that hopping instead of normal walking is displayed in mice lacking the ephA4 receptor-expressing lumbar interneurons. Finally, another population of CPG neuron candidate

called HB9 neurons was reported to provide neuronal excitation during locomotion. Although, no corresponding knockout model has been tested, compelling evidence suggests that the HB9 excitatory interneuron belongs to an asymmetrically-organized rhythm-generating network (Brownstone's and Jessell's work). Taken altogether, results from these exciting new studies in mice suggest that these genetically-characterized interneurons (V0–V3, EphA4, HB9) may constitute different cellular components of the CPG (Kullander et al., 2003; Wilson et al., 2005; Brownstone and Wilson, 2008). For instance, V0 interneurons form many cell types including commissural neurons which could establish inhibitory reciprocal connections between the two sides of the spinal cord. In turn, V1 interneurons may be associated with inhibitory interneurons such as the Ia inhibitory and Renshaw cells whereas HB9s may be excitatory interneurons constituting at least part of the rhythm generating layer. However, additional studies need to be conducted in order to fully characterize these promising new CPG neuron candidates (Butt et al., 2002; Wilson et al., 2007). It is important to mention also in most cases of gene deletions using these KO models, elimination of rhythmic motoneuron excitation or rhythmic inhibition is rarely found suggesting that several CPG secrets remain to be described.

CHARACTERISTICS OF OTHER CPGs

SPINAL GENERATOR FOR EJACULATION

A breakthrough finding in 2002 has drastically changed our view of the neurobiology of sexual function (Truitt and Coolen, 2002; Coolen et al., 2003). Indeed, Truitt and Coolen have shown that ejaculation critically depends upon a spinal generator or network of neurons referred to as the Spinal Generator for Ejaculation (SGE; Truitt and Coolen, 2002). Located in the lumbar segments L3 and L4, the SGE is defined as a circuit capable of producing self-sustained rhythmic output to pudendal motoneurons. The SGE was found to contain a key population of neurons (lumbar spino-thalamic neurons also called LSt cells) that (1) project to forebrain; (2) project to pudendal motoneurons (correspond to those located in the Onuf's nucleus in men); and (3) receive input from sexual organs via the pudendal and dorsal nerve of the penis. In fact, ejaculation is completely lost in animals undergoing LSt cell-lesioned procedures (using SSP-saporin). LSt cells are found in the vicinity of the central canal (lamina X and medial portion of lamina VII, and most of them specifically contain galanin, CCK, enkephalin and NK-1 receptors. Electrical stimulation of the pudendal or dorsal nerve of the penis nerves was shown to elicit ejaculatory responses in low-thoracic Tx rats. Comparable data were found in SCI men supporting the existence of LSt cells (or at least of a SGE) in humans as in other species. However, the type(s) of neurotransmitters involved and the subset of post-synaptic receptors associated with SGE activation and ejaculation remain largely unknown (Courtois et al., 2008, 2013).

This said, only a few families of compounds are known to modulate sexual function (either erection, ejaculation or both) (McKenna et al., 1991; Pomerantz et al., 1993; Vargas et al., 2004; Moreland and Makela, 2005; García-Bravo et al., 2006). For instance, Guttmann and Walsh showed (earlier case reports also exist) that the cholinesterase inhibitor prostigmine administered intrathecally (i.t.) could elicit spontaneous erection accompanied sometimes with ejaculation in SCI men (Guttman and Walsh, 1971). Midodrine (alpha-1 agonist) was also shown to induce either normal (anterograde—i.e., outside the urethral meatus) or abnormal (retrograde—i.e., back inside into the bladder) ejaculation in patients with SCI but, as with cholinesterase inhibitors, it was also found to increase blood pressure and autonomic dysreflexia (Jonas et al., 1979; Riley and Riley, 1982; Staerman et al., 2001; Blanchard-Dauphin et al., 2005; Courtois et al., 2008). In animal models, muscarine (i.p. or i.t.) was found to promote erection and sometimes ejaculation in sensory-stimulated Tx rats (i.e., penile sheath retraction known as the "penile reflex" model) (Durán et al., 2000). Alpha-1 agonists such as methoxamine (which, unlike midodrine, crosses the BBB) were reported to promote reflexively-induced ejaculation (either immediately after Tx or by urethral stimulation with fluid injection) in freely moving Tx rats. P-chloroamphetamine (amphetamine derivative that increases 5-HT release) was shown to elicit fictive ejaculation (Electromyogram (EMG) correlates) in anesthetized Tx rats) (Stafford et al., 2006). Supporting a role for subclasses of spinal 5-HT receptors, 5-HT receptor agonists such as m-CPP (5-HT2) or 8-OH-DPAT (5-HT1A) were also found to promote sometimes ejaculation in sensory-stimulated Tx (self-grooming) or non-Tx (copulating) rats (Camacho et al., 2007). However, mixed 5-HT-induced effects have also bee reported suggesting that serotonergic modulation may depend upon the site of action and route of delivery (inhibitory effects in brain vs. excitatory effects in spinal cord) (Yonezawa et al., 2008). Regarding DA agonists, none have been tested in Tx animals although D2-like agonists i.c.v. injected was reported to promote ejaculation in intact rats.

SPINAL MICTURITION CENTER

Micturition essentially depends, for the storage and periodic elimination of urine, on the coordinated activity of smooth and striated muscles in the several functional units of the lower urinary tract, namely the urinary bladder, the bladder neck, the urethra and the urethral sphincter (Mallory et al., 1991; de Groat et al., 1993; Andersson and Pehrson, 2003; Sugaya et al., 2005). The coordination between these organs is mediated by a complex neural control system that is located partly in the spinal cord (Schrøder, 1985; Birder and de Groat, 1993; Birder et al., 1999; Dolber et al., 2007). Central interneurons retrogradely labeled by injection of pseudorabies virus into the urinary bladder of the rat were found in regions receiving afferent input from the bladder (Nadelhaft and Vera, 1995; Sugaya et al., 1997). A comparable distribution was shown following injections of virus into the urethra (Vizzard's work) or the external urethral sphincter (EUS), indicating a prominent overlap of the interneuronal pathways controlling the various target organs of the lower urinary tract (Vizzard et al., 1995). In addition, spinal interneurons (located near dorsal commissure, superficial dorsal horn and sacral parasympathic nucleus) involved in processing afferent input from the lower urinary tract have been identified by c-fos expression following noxious or non-noxious stimulation of the bladder and urethra in rats (de Groat's work). Some of these interneurons send long

projections to the brain, whereas others make local connections in the spinal cord and participate in segmental spinal reflexes. These results provide strong evidence of a CPG for EUS control both in the thoracolumbar (T8-9 for storage of urine) and lumbosacral (L3-L4, L6-S1 for expulsion of urine) spinal cord. Various neurotransmitters have been associated with the control of the lower urinary tract including glutamate, tachykinins, pituitary-adenylate-cyclase-activating polypeptide, NO and ATP. Glutamate, acting on NMDA and non-NMDA receptors, seems to be critically involved in spinal and supraspinal reflex pathways that control the bladder and the EUS (Yoshiyama and de Groat, 2005). In contrast, GABA (γ-aminobutyric acid), glycine and enkephalins were reported to exert a tonic inhibitory control in the pontine micturition center (PMC) and regulate bladder capacity. Other neurotransmitters including DA, 5-HT, NA, and acetylcholine have either inhibitory or excitatory effects, depending on the type and location of activated receptors (de Groat et al., 1993; de Groat and Yoshimura, 2001; Chang et al., 2006; Dolber et al., 2007). For example, DA elicits inhibitory effects on micturition through D_1-like receptors and facilitatory effects through D_2-like receptors. Also, much like what has been shown with 5-HT1A/7 receptor agonists on CPG activation (Landry et al., 2006a), 5-HT1A/7 receptor agonist 8-OH-DPAT was found to induce rhythmic EUS relaxation during voiding in urethane-anesthetized chronic Tx rats. Other potential pharmacological treatments to facilitate micturition for instance after SCI; are (1) intravesical administration of drugs such as vanilloids which is aimed at desensitizing bladder afferents (Fowler et al., 1994); (2) injection of BoNT/A into the detrusor to temporarily block the pre-synaptic release of ACh from the parasympathetic innervation and produce a paralysis of the detrusor smooth muscle (Schurch et al., 2000); and (3) administration of NO donors such as isosorbide dinitrate to produce a significant reduction in striated sphincter pressure at rest and during dyssynergic contraction (Reitz et al., 2004).

SPINAL DEFECATION CENTER

Bowel control involves a complex interactions between the autonomic (sympathetic and parasympathetic), central (brain and spinal cord) and muscular systems (sphincters and smooth muscles) (Rostad, 1973). Experiments in spinal cord-transected rats (cervical level) have clearly shown the existence of a reflex defecation center (also known as the lumbosacral defecation center) located by retrograde labeling at the low lumbar and upper sacral level (i.e., L6-S1 in rats) (Shimizu et al., 2006). Evidence of increased defecation in humans caused by Lumbosacral Defecation Center (LDC)-mediated effects, induced by ghrelin receptor agonists, has been shown whereas direct evidence using intrathecal injections have been reported in animal models (Shimizu et al., 2006). Indeed, defecation induced pharmacologically was prevented by cutting the nerve pathways that connect the lumbosacral spinal cord and the colorectum, but not by severing the thoracic spinal cord (Ferens et al., 2011; Pustovit et al., 2014). Unfortunately, Ghrelin or its known agonists (e.g., ulimorelin) do not elicit defecation when administered orally and may increase blood pressure when delivered i.t.

CPGs IN HUMANS: EVIDENCE FROM DIRECT STIMULATION AND SPONTANEOUS PLASTICITY AND ROLE OF FEEDBACK AND FEEDFORWARD

Some people are reluctant to believe that spinal pattern generators or CPGs exist in humans given the lack of direct evidence. It is true that most data have been obtained from animal models, as described in this review. Experiments and tools used to explore CPG properties in animals are simply too invasive for comparable exploration in humans. But again, given that the spinal cord, among all structures of the CNS, is particularly well-preserved phylogenetically, it may be more reasonable to argue the opposite—are there evidence that humans do not have spinal CPGs given that all vertebrate species examined thus far stongly suggest the existence of comparable neural control systems for locomotion and other stereotyped motor behaviors?

The answer is "no", we do not have evidence against the existence of spinal CPGs in humans (Bussel et al., 1989, 1992). There is increasing evidence suggesting the existence of comparable spinal networks in primates including humans—at least for locomotion (Tan, 2006a,b, 2008). For instance, epidural or intraspinal stimulation near L1–L2 to successfully trigger locomotor-like movements in the lower extremities of completely SCI patients (Dimitrijevic et al., 1998; Holinski et al., 2011; Moshonkina et al., 2012). Given that electrical stimulation of CPG does not generally lead to full weght bearing stepping (e.g., often only to air-stepping or locomotor-like movements lying in bed, this suggests that only partial CPG activation may be achieved electrically. In turn, it has been possible to activate the locomotor CPG by administration i.t., of a variety of pharmacological agents to acutely spinalized marmoset monkeys (Callithrix jacchus) in the absence of phasic afferent input to the spinal cord (Fedirchuk et al., 1998). Spontaneous expression, within weeks or months post-SCI, of rhythmic movements (myoclonus or locomotor-like movements) in humans have also been reported (Pozos and Iaizzo, 1991; Bussel et al., 1992, 1988; Calancie et al., 1994; Chervin et al., 2003; Calancie, 2006; Consentino et al., 2006; Gerasimenko et al., 2008, 2010; Nadeau et al., 2010; Field-Fote et al., 2012; Gorodnichev et al., 2012; Rye and Trotti, 2012).

How could such spontaneous movements occur (Clemens et al., 2006; Harkema, 2008; Nadeau et al., 2010)? No clear answer exists in humans (Calancie et al., 1994; Byrnes et al., 2006). However, related spontaneous plasticity events have clearly been shown in animal models to be associated with some immediate early gene expression (IEG) in CPG-corresponding areas of the spinal cord. Changes in IEG expression levels post-SCI were associated also with a cascade of subsequent changes (TNF-α, NOS; C1qb, Galectin-3, and p22(phox), glycine receptors, GAD-67, 5-HT1A/7 receptors; NGF, BDNF, and NT-3) and spontaneous spinal locomotor-like activity (Chi et al., 1993; Yakovlev and Faden, 1994; Giroux et al., 1999; Tillakaratne et al., 2002; Gómez-Pinilla et al., 2004; Landry et al., 2006b; Lukacova et al., 2006; Peng et al., 2006; Schmitt et al., 2006; Giszter et al., 2007; Li et al., 2007; Ung et al., 2007).

It is clear though that despite direct stimulation electrically or pharmacologically, spinal-mediated stereotyped motor functions such as locomotion requires at least some feedforward and

feedback mechanisms for goal-oriented functional recovery. Interestingly, some feedback mechanisms may remain operational despite severe spinal cord lesions at the thoracic or cervical level. Indeed, it has clearly been shown in Tx cats, displaying some restored spinal-mediated locomotor activity on a motorized treadmill, that adaptation to different speeds remains possible (Edgerton's or Rossignol's work). However, to reach more complex levels of adaptation such as in climbing steps, feedforward mechanisms are probably required. Recent experiments in partially Tx rats showed that at least some descending fibers need to be intact or functional for animals, driven to walk by CPG-activating drug cocktails (such as those identified in Guertin's laboratory), for displaying complex patterns such as walking up stairs and changing directions during walking (SŁawinska et al., 2012; van den Brand et al., 2012). It is unclear which descending systems are most important for such key feedforward mechanisms, however, cells from the parietal cortex and cerebellar systems are key to visually-guided locomotion whereas brainstem circuits and corresponding descending tracts (e.g., vestibulo-spinal tracts) are known for significant roles in equilibrium and tonus (Gurfinkel and Shik, 1973; Aydogdu et al., 2011).

All in all, future drug treatments identified in animal models to modulate spinal network activity are thus likely to be associated with comparable efficacy data in humans given significant similarities in spinal cord structures and cellular properties from lampreys to primates. However, the role of feedback and feedforward mechanisms in patients with SCI that will be given CPG-modulating and reactivating-drug treatments remains to be explored.

FURTHER EVIDENCE FROM PHARMACOLOGICAL REACTIVATION OF THE CORRESPONDING SPINAL NETWORKS

As mentioned above and in different sections throughout this review, it is clear that different types of CPGs exist and a wide variety of responses may be triggered pharmacologically pending upon the classes and subclasses of transmembranal receptors activated or blocked by specific ligands or exogenous neurotransmitter administration (Johnston et al., 2003; Guertin and Steuer, 2009).

In my laboratory, an extended series of drug screening experiments has been conducted in recent years. Using an experimental model of complete SCI (a low-thoracic spinal cord transected mouse) that eases and accelerates the identification of CPG-activating compounds (i.e., given the completeness and high reproducibility of this exact type of injury), we identified families of ligands capable of inducing, within minutes post-administration (s.c., p.o. or i.p.), either episodes of locomotor movements in the hindlimbs, micturition, defecation or ejaculation. Given that spinal networks controlling these functions are located below injury level in our model and that brain-mediated effects through descending inputs are unlikely in Tx mice, then such well-coordinated and stereotyped motor behaviors must be induced, at least partly, by corresponding spinal network reactivation following administration of specific drug(s).

Reactivation of spinal catecholaminergic receptors using L-DOPA/carbidopa or DA receptor agonists and 5-HT1A receptors using buspirone or 8-OH-DPAT was found to trigger 45-min episodes of basic straightforward walking in chronic Tx mice (see Guertin, 2004 work since 2004). This patent-protected technologically, now referred to as Spinalon^{TM}, was shown to elicit comparable effects in other species (patent application WO 2006026850) whereas repeated administration was found to prevent muscular atrophy and anemia among other biological functions and systems. L-DOPA/carbidopa and buspirone were shown not to have drug-drug interactions *in vitro* and *in vivo*, providing evidence of safety in animals (Guertin, 2009b; Guertin et al., 2010, 2011). Clinical trials (Phase I/IIa) in paraplegic and tetraplegic patients that began recently (December 2013) may provide further evidence of safety in patients while possibly reporting the first evidence ever of efficacy in humans for a CPG-activating drug therapy.

Given that no sensory stimulation, no harness and no electrical stimulation are used for these tests in motor-complete SCI patients, if efficacy data in ASIA-As are to be found, this could be considered as clear evidence of a CPG in humans that remains functional and reactivable with drugs despite a SCI at higher (rostral) levels. Upon positive results, a Phase IIb and eventually a pivotal Phase III shall be undertaken to eventually meet criteria for new drug approval (NDA) in the U.S., Europe and Canada expected for the end of 2018.

Given the successes obtained with this approach, additional drug screening studies were conducted in our laboratory to identify also Sacral or Spinal Micturition Center (SMC), LDC and SGE activating compounds (Guertin, 2005, 2008, 2009b). We recently identified leads and provided proof-of-concept data *in vivo* clearly demonstrating that pharmacological reactivation of these other networks upon systemic drug delivery is possible at least in animal models of complete SCI.

A combination of transmembranal receptor agonists (that can not yet be revealed for intellectual property reasons) found to elicit superior effects was selected as our final candidate product suitable for full preclinical and clinical development. Now referred to as Micselon^{TM}, this drug combination was shown to elicit, within 15 to 30 min, episodes of micturition in SCI mice suffering of chronic urinary retention (patent application US 61/946,097). Again, the well-coordinated motor responses triggered by this therapy in completely SCI animals that induce in absence of sensory stimulation, providing strong evidence of SMC-activating effects. Lack of drug-drug interactions and evidence of efficacy in SCI patients remain to be shown prior to IND and NDA approval of this new experimental therapy against urinary retention.

A comparable technology was also found to restore some defecatory activities in SCI animals. Leads were identified using our technological platform adapted to assess defecation in paraplegic mice. Proof-of-concept data *in vivo* were obtained recently and Optimization phase studies with drug combinations revealed superior defecation-eliciting effects with a bi-therapy referred to as Transilon^{TM}. It was found to elicit within 30 min episodes of defecation in chronic SCI mice suffering of constipation (patent application US 61/946,113). Again, the well-coordinated motor

responses triggered by this therapy in animals in absence of sensory stimulation provide strong evidence of LDC-activating effects. Lack of drug-drug interactions and evidence of efficacy in SCI patients remain to be shown prior to regulatory approval of this new experimental therapy against chronic constipation (expected by 2020).

Finally, we found a 4th class of drug treatments capable of spinal network reactivation in completely SCI subjects. Lead compounds were identified although Optimization phase studies have not been completed yet. Nonetheless, we found compounds and specifically drug combinations (which specific identity can not be revealed yet for intellectual property reasons) that can elicit unconditionally (with or without erection) seminal emission alone or seminal emission accompanied of seminal expulsion (full ejaculatory motor pattern) in male Tx mice (patent application US 60/996,535). These results provided proof-of-concept evidence of efficacy (SGE activation) in *in vivo* animal models of SCI. Again, additional tests remain to be performed to show comparable efficacy in humans and thus to demonstrate the existence of SGE neurons remaining functional and reactivable pharmacologically in people with SCI suffering of anejaculation.

In conclusion, CPGs involved in the control of several stereotyped motor behaviors such as locomotion, micturition, defecation and ejaculation have clearly been shown in a wide variety of species (Vizzard et al., 1995; Dimitrijevic et al., 1998; Truitt and Coolen, 2002; Shimizu et al., 2006; Clarac and Pearlstein, 2007; Guertin, 2009a; Guertin and Steuer, 2009). The one underlying pattern and rhythm generation for locomotion is by the far the most extensively studied although cellular characterization remains incomplete. Other CPGs, although poorly characterized cellularly, have also been clearly shown in different species. Nonetheless, this has not prevented research from identifying drug treatments capable of reactivating these networks separately after SCI at higher (rostral) levels. In fact, several experimental drugs (drug combinations composed of known and rather safe molecules) are currently in development clinically or preclinically. These new pharmacological aids may provide both, further evidence of spinal CPGs in humans and, useful noninvasive approaches to temporarily restore some key biological functions in people with SCI or comparable neurological disorders (e.g., multiple sclerosis) (Holmes, 1915; Fung et al., 1990; Wainberg et al., 1990; Rémy-Néris et al., 1999; Edgerton et al., 2001; Guertin and Steuer, 2009; Thorpe et al., 2011; Dietz and Sinkjaer, 2012; Guertin, 2012; Harkema et al., 2012; Jones and Cavanna, 2012; Schürks and Bussfeld, 2013).

REFERENCES

Andersson, K. E., and Pehrson, R. (2003). CNS involvement in overactive bladder: pathophysiology and opportunities for pharmacological intervention. *Drugs* 63, 2595–2611. doi: 10.2165/00003495-200363230-00003

Angel, M. J., Guertin, P., Jiménez, I., and McCrea, D. A. (1996). Group I extensor afferents evoke disynaptic EPSPs in cat hindlimb extensor motorneurones during fictive locomotion. *J. Physiol.* 493, 851–861.

Angel, M. J., Jankowska, E., and McCrea, D. A. (2005). Candidate interneurones mediating group I disynaptic EPSPs in extensor motoneurones during fictive locomotion in the cat. *J. Physiol.* 563(Pt. 2), 597–610. doi: 10.1113/jphysiol.2004.076034

Antri, M., Barthe, J. Y., Mouffle, C., and Orsal, D. (2005). Long-lasting recovery of locomotor function in chronic spinal rat following chronic combined pharmacological stimulation of serotonergic receptors with 8-OHDPAT and quipazine. *Neurosci. Lett.* 384, 162–167. doi: 10.1016/j.neulet.2005.04.062

Aydogdu, I., Tanriverdi, Z., and Ertekin, C. (2011). Dysfunction of bulbar central pattern generator in ALS patients with dysphagia during sequential deglutition. *Clin. Neurophysiol.* 122, 1219–1228. doi: 10.1016/j.clinph.2010.11.002

Barajon, I., Gossard, J. P., and Hultborn, H. (1992). Induction of fos expression by activity in the spinal rhythm generator for scratching. *Brain Res.* 588, 168–172. doi: 10.1016/0006-8993(92)91359-m

Barbeau, H., and Rossignol, S. (1990). The effects of serotonergic drugs on the locomotor pattern and on cutaneous reflexes of the adult chronic spinal cat. *Brain Res.* 514, 55–67. doi: 10.1016/0006-8993(90)90435-e

Barbeau, H., and Rossignol, S. (1991). Initiation and modulation of the locomotor pattern in the adult chronic spinal cat by noradrenergic, serotonergic and dopaminergic drugs. *Brain Res.* 546, 250–260. doi: 10.1016/0006-8993(91)91489-n

Barthélemy, D., Grey, M. J., Nielsen, J. B., and Bouyer, L. (2011). Involvement of the corticospinal tract in the control of human gait. *Prog. Brain Res.* 192, 181–197. doi: 10.1016/b978-0-444-53355-5.00012-9

Bélanger, M., Drew, T., Provencher, J., and Rossignol, S. (1996). A comparison of treadmill locomotion in adult cats before and after spinal transection. *J. Neurophysiol.* 76, 471–491.

Bergmans, J., Burke, R., and Lundberg, A. (1969). Inhibition of transmission in the recurrent inhibitory pathway to motoneurones. *Brain Res.* 13, 600–602. doi: 10.1016/0006-8993(69)90270-4

Birder, L. A., and de Groat, W. C. (1993). Induction of c-fos expression in spinal neurons by nociceptive and nonnociceptive stimulation of LUT. *Am. J. Physiol.* 265, R326–R333.

Birder, L. A., Roppolo, J. R., Erickson, V. L., and de Groat, W. C. (1999). Increased c-fos expression in spinal lumbosacral projection neurons and preganglionic neurons after irritation of the lower urinary tract in the rat. *Brain Res.* 834, 55–65. doi: 10.1016/s0006-8993(99)01546-2

Bizzi, E., Cheung, V. C., d'Avella, A., Saltiel, P., and Tresch, M. (2008). Combining modules for movement. *Brain Res. Rev.* 57, 125–133. doi: 10.1016/j.brainresrev.2007.08.004

Blanchard-Dauphin, A., Rigot, J. M., and Thevenon, A. (2005). Treatment of ejaculation disorders by midodrine (Gutron) per os retrospective study of about 16 subjects. *Ann. Readapt. Med. Phys.* 48, 34–40. doi: 10.1016/j.annrmp.2004.09.004

Bonnot, A., Whelan, P. J., Mentis, G. Z., and O'Donovan, M. J. (2002). Locomotor-like activity generated by the neonatal mouse spinal cord. *Brain Res. Brain Res. Rev.* 40, 141–151. doi: 10.1016/s0165-0173(02)00197-2

Bretzner, F., and Drew, T. (2005). Contribution of the motor cortex to the structure and the timing of hindlimb locomotion in the cat: a microstimulation study. *J. Neurophysiol.* 94, 657–672. doi: 10.1152/jn.01245.2004

Brocard, F., Tazerart, S., and Vinay, L. (2010). Do pacemakers drive the central pattern generator for locomotion in mammals? *Neuroscientist* 16, 139–155. doi: 10.1177/1073858409346339

Brownstone, R. M., and Wilson, J. M. (2008). Strategies for delineating spinal locomotor-rhythm generating networks and the possible role of Hb9 interneurones in rhythmogenesis. *Brain Res. Rev.* 57, 64–76. doi: 10.1016/j.brainresrev.2007.06.025

Buschges, A., Akay, T., Gabriel, J. P., and Schmidt, J. (2008). Organizing network action for locomotion: insights from studying insect walking. *Brain Res. Rev.* 57, 162–171. doi: 10.1016/j.brainresrev.2007.06.028

Bussel, B., Roby-Brami, A., and Azouvi, P. (1992). "Organization of reflexes elicited by flexor reflex afferents in paraplegic man: evidence for a stepping generator," in *Muscle Afferents and Spinal Control of Movement*, eds L. Jami, E. Pierrot-Deseilligny and D. Zytnicki, 1st Edn. (Oxford, UK: Pergamon), 427–432.

Bussel, B., Roby-Brami, A., Yakovleff, A., and Bennis, N. (1989). Late flexion reflex in paraplegic patients. Evidence for a spinal stepping generator. *Brain Res. Bull.* 22, 53–56. doi: 10.1016/0361-9230(89)90127-5

Bussel, B. C., Roby-Brami, A., Yakovleff, A., and Bennis, N. (1988). "Evidences for the presence of a spinal stepping generator inpatients with a spinal cord section," in *Posture and Gait: Development, Adaptation and Modulation*, eds B. Amblard, A. Berthoz and F. Clarac (Amsterdam, the Netherlands: Elsevier), 273–278.

Butt, S. J., Harris-Warrick, R. M., and Kiehn, O. (2002). Firing properties of identified interneuron populations in the mammalian hindlimb central pattern generator. *J. Neurosci.* 22, 9961–9971.

Byrnes, K. R., Garay, J., Di Giovanni, S., De Biase, A., Knoblach, S. M., Hoffman, E. P., et al. (2006). Expression of two temporally distinct microglia-related gene clusters after spinal cord injury. *Glia* 53, 420–433. doi: 10.1002/glia.20295

Calancie, B. (2006). Spinal myoclonus after spinal cord injury. *J. Spinal Cord Med.* 29, 413–424.

Calancie, B., Needham-Shropshire, B., Jacobs, P., Willer, K., Zych, G., and Green, B. A. (1994). Involuntary stepping after chronic spinal cord injury. Evidence for a central rhythm generator for locomotion in man. *Brain* 117(Pt. 5), 1143–1159. doi: 10.1093/brain/117.5.1143

Camacho, F. J., Castro, M., Hernandez, V., and Paredes, R. G. (2007). Facilitation of ejaculation induced by 8-OH-DPAT does not produce conditioned place preference in male rats. *Behav. Neurosci.* 121, 579–585. doi: 10.1037/0735-7044.121.3.579

Cazalets, J. R., Grillner, P., Menard, I., Cremieux, J., and Clarac, F. (1990). Two types of motor rhythm induced by NMDA and amines in an in vitro spinal cord preparation of neonatal rat. *Neurosci. Lett.* 111, 116–121. doi: 10.1016/0304-3940(90)90354-c

Chang, H. Y., Cheng, C. L., Chen, J. J., and de Groat, W. C. (2006). Roles of glutamatergic and serotonergic mechanisms in reflex control of the external urethral sphincter in urethane-anesthetized female rats. *Am. J. Physiol. Regul. Integr. Comp. Physiol.* 291, R224–R234. doi: 10.1152/ajpregu.00780.2005

Chau, C., Barbeau, H., and Rossignol, S. (1998a). Effects of intrathecal alpha1- and alpha2-noradrenergic agonists and norepinephrine on locomotion in chronic spinal cats. *J. Neurophysiol.* 79, 2941–2963.

Chau, C., Barbeau, H., and Rossignol, S. (1998b). Early locomotor training with clonidine in spinal cats. *J. Neurophysiol.* 79, 392–409.

Cheng, J., Stein, R. B., Jovanocic, K., Yoshida, K., Bennett, D. J., and Han, Y. (1998). Identification, localization and modulation of neural networks for walking in mudpuppy (Necturus maculatus) spinal cord. *J. Neurosci.* 18, 4295–4305.

Cheng, J., and Magnuson, D. S. (2011). Initiation of segmental locomotor-like activities by stimulation of ventrolateral funiculus in the neonatal rat. *Exp. Brain Res.* 214, 151–161. doi: 10.1007/s00221-011-2816-7

Chervin, R. D., Consens, F. B., and Kutluay, E. (2003). Alternating leg muscle activation during sleep and arousals: a new sleep-related motor phenomenon? *Mov. Disord.* 18, 551–559. doi: 10.1002/mds.10397

Chi, S. I., Levine, J. D., and Basbaum, A. I. (1993). Peripheral and central contributions to the persistent expression of spinal cord fos-like immunoreactivity produced by sciatic nerve transection in the rat. *Brain Res.* 617, 225–237. doi: 10.1016/0006-8993(93)91090-f

Cina, C., and Hochman, S. (2000). Diffuse distribution of sulforhodamine-labeled neurons during serotonin-evoked locomotion in the neonatal rat thoracolumbar spinal cord. *J. Comp. Neurol.* 423, 590–602. doi: 10.1002/1096-9861(20000807)423:4<590::aid-cne5>3.0.co;2-l

Clarac, F., and Pearlstein, E. (2007). Invertebrate preparations and their contribution to neurobiology in the second half of the 20th century. *Brain Res. Rev.* 54, 113–161. doi: 10.1016/j.brainresrev.2006.12.007

Clemens, S., Rye, D., and Hochman, S. (2006). Restless legs syndrome: revisiting the dopamine hypothesis from the spinal cord perspective. *Neurology* 67, 125–130. doi: 10.1212/01.wnl.0000223316.53428.c9

Consentino, F. L., Iero, I., Lanuzza, B., Tripodi, M., and Ferri, R. (2006). The neurophysiology of the alternating leg muscle activation (ALMA) during sleep: study of one patient before and after treatment with pramipexole. *Sleep Med.* 7, 63–71. doi: 10.1016/j.sleep.2005.06.007

Coolen, L. M., Veening, J. G., Wells, A. B., and Shirpley, M. T. (2003). Afferent connections of the parvocellular subparafascicular thalamic nucleus in the rat: evidence for functional subdivisions. *J. Comp. Neurol.* 463, 132–156. doi: 10.1002/cne.10739

Courtine, G., van den Brand, R., and Musienko, P. (2011). Spinal cor injury: time to move. *Lancet* 377, 1896–1898. doi: 10.1016/s0140-6736(11)60711-3

Courtois, F., Carrier, S., Charvier, K., Guertin, P. A., and Journel, M. (2013). The control of male sexual responses. *Curr. Pharm. Des.* 19, 4341–4356. doi: 10.2174/13816128113199990333

Courtois, F. J., Charvier, K. F., Leriche, A., Vezina, J. G., Côté, M., and Bélanger, M. (2008). Blood pressure changes during sexual stimulation, ejaculation and midodrine treatment in men with spinal cord injury. *BJU Int.* 101, 331–337. doi: 10.1111/j.1464-410x.2007.07254.x

Cowley, K. C., and Schmidt, B. J. (1994). A comparison of motor patterns induced by N-methyl-D-aspartate, acetylcholine and serotonin in the in vitro neonatal rat spinal cord. *Neurosci. Lett.* 171, 147–150. doi: 10.1016/0304-3940(94)90626-2

Crone, S. A., Quinlan, K. A., Zagoraiou, L., Droho, S., Restrepo, C. E., Lundfald, L., et al. (2008). Genetic ablation of V2a ipsilateral interneurons disrupts left-right locomotor coordination in mammalian spinal cord. *Neuron* 60, 70–83. doi: 10.1016/j.neuron.2008.08.009

Currie, S. N. (1999). Fictive hindlimb motor patterns evoked by AMPA and NMDA in turtle spinal cord-hindlimb nerve preparations. *J. Physiol. Paris* 93, 199–211. doi: 10.1016/s0928-4257(99)80152-1

Currie, S. N., and Lee, S. (1997). Glycinergic inhibition contributes to the generation of rostral scratch motor patterns in the turtle spinal cord. *J. Neurosci.* 17, 3322–3333.

de Groat, W. C., and Yoshimura, N. (2001). Pharmacology of the lower urinary tract. *Ann. Rev. Pharmacol. Toxicol.* 41, 691–721. doi: 10.1146/annurev.pharmtox.41.1.691

de Groat, W. C., Booth, A. M., and Yoshimura, N. (1993). *Nervous Control of the Urogenital System (Autonomic Nervous System)* (Vol. 3), ed C. A. Maggi (London: Harwood Academic Publishers), 227–289.

Delcomyn, F. (1977). "Co-ordination of invertebrate locomotion," in *Mechanics and Energetics of Locomotion*, eds R. M. Alexander and G. Gold Spink (London: Chapman and Hall), 82–114.

Dietz, V., and Sinkjaer, T. (2012). Spasticity. *Handb. Clin. Neurol.* 109, 197–211. doi: 10.1016/B978-0-444-52137-8.00012-7

Dimitrijevic, M. R., Gerasimenko, Y., and Pinter, M. M. (1998). Evidence for a spinal central pattern generator in humans. *Ann. N Y Acad. Sci.* 860, 360–376. doi: 10.1111/j.1749-6632.1998.tb09062.x

Dolber, P. C., Gu, B., Zhang, X., Fraser, M. O., Thor, K. B., and Reiter, J. P. (2007). Activation of the external urethral sphincter central pattern generator by a 5-HT(1A) receptor agonist in rats with chronic spinal cord injury. *Am. J. Physiol. Regul. Integr. Comp. Physiol.* 292, R1699–R1706. doi: 10.1152/ajpregu.00142.2006

Durán, I., Gil, L., and Cueva-Rolon, R. (2000). Masculine copulatory behavior is facilitated by intrathecally administered muscarine. *Exp. Brain Res.* 134, 490–496. doi: 10.1007/s002210000488

Duysens, J., De Groote, F., and Jonkers, I. (2013). The flexion synergy, mother of all synergies and father of new models of gait. *Front. Comput. Neurosci.* 7:14. doi: 10.3389/fncom.2013.00014

Duysens, J. (1977). Reflex control locomotion as revealed by stimulation of cutaneous afferents in spontaneously walking premammillary cats. *J. Neurophysiol.* 40, 737–751.

Edgerton, V. R., Grillner, S., Sjöström, A., and Zangger, P. (1976). "Central generation of locomotion in vertebrates," in *Neural Control of Locomotion*, eds R. Herman, S. Grillner and P. Zangger (New York: Plenum Press), 439–464.

Edgerton, V. R., Leon, R. D., Harkema, S. J., Hodgson, J. A., London, N., Reinkensmeyer, D. J., et al. (2001). Retraining the injured spinal cord. *J. Physiol.* 533, 15–22. doi: 10.1111/j.1469-7793.2001.0015b.x

Eken, T., Hultborn, H., and Kiehn, O. (1989). Possible functions of transmitter-controlled plateau potentials in alpha motoneurones. *Prog. Brain Res.* 80, 257–267; discussion 239–242. doi: 10.1016/s0079-6123(08)62219-0

English, A. W. (1985). Interlimb coordination during stepping in the cat: the role of the dorsal spinocerebellar tract. *Exp. Neurol.* 87, 96–108. doi: 10.1016/0014-4886(85)90136-0

Fedirchuk, B., Nielsen, J., Petersen, N., and Hultborn, H. (1998). Pharmacologically evoked fictive motor patterns in the acutely spinalized marmoset monkey (Callithrix jacchus). *Exp. Brain Res.* 122, 351–361. doi: 10.1007/s002210050523

Ferens, D. M., Habgood, M. D., Saunders, N. R., Tan, Y. H., Brown, D. J., Brock, J. A., et al. (2011). Stimulation of defecation in spinal cord-injured rats by a centrally acting ghrelin receptor agonist. *Spinal Cord* 49, 1036–1041. doi: 10.1038/sc.2011.60

Field-Fote, E., Ness, L. L., and Ionno, M. (2012). Vibration elicits involuntary, step-like behavior in individuals with spinal cord injury. *Neurorehabil. Neural Repair* 26, 861–869. doi: 10.1177/1545968311433603

Flourens, M.-J.-P. (1824). *Recherches expérimentales sur les propriétés et les Fonctions du Système Nerveux, dans les Animaux Vertébrés, Experimental Studies on the Properties and Functions of the Nervous System in Vertebrate Animals.* Paris: Chez Crevot.

Forssberg, H., and Grillner, S. (1973). The locomotion of the acute spinal cat injected with clonidine i.v. *Brain Res.* 50, 184–186. doi: 10.1016/0006-8993(73)90606-9

Forssberg, H., and Hirschfeld, H. (1988). Phasic modulation of postural activation patterns during human walking. *Prog. Brain Res.* 76, 221–227. doi: 10.1016/s0079-6123(08)64508-2

Fowler, C. J., Beck, R. O., Gerrard, S., Betts, C. D., and Fowler, C. G. (1994). Intravesical capsaicin for treatment of detrusor hyperreflexia. *J. Neurol. Neurosurg. Psych.* 57, 169–173. doi: 10.1136/jnnp.57.2.169

Freusberg, A. (1874). Reflexbewegungen beim hunde. *Pflügers Arch* 9, 358–391. doi: 10.1007/bf01612347

Fung, J., Stewart, J. E., and Barbeau, H. (1990). The combined effects of clonidine and cyproheptadine with interactive training on the modulation of locomotion in spinal cord injured subjects. *J. Neurol. Sci.* 100, 85–93. doi: 10.1016/0022-510x(90)90017-h

García-Bravo, A. M., Suarez-Hernandez, D., Ruiz-Fernandez, M. A., Silva Gonzalez, O., Barbara-Bataller, E., and Méndez Suarez, J. L. (2006). Determination of changes in blood pressure during administration of Viagra in patients with spinal cord injury and erectile dysfunction. *Spinal Cord* 44, 301–308. doi: 10.1038/sj.sc.3101846

Gerasimenko, Y., Gorodnichev, R., Machueva, E., Pivovarova, E., Semyenov, D., Savochin, A., et al. (2010). Novel and direct access to the human locomotor spinal circuitry. *J. Neurosci.* 30, 3700–3708. doi: 10.1523/jneurosci.4751-09.2010

Gorodnichev, R. M., Pivovarova, E. A., Pukhov, A., Moiseev, S. A., Savokhin, A. A., and Moshonkina, T. R. (2012). [Transcutaneous electrical stimulation of the spinal cord: non-invasive tool for activation of locomotor circuitry in human]. *Fiziol. Cheloveka* 38, 46–56.

Gerasimenko, Y., Roy, R. R., and Edgerton, V. R. (2008). Epidural stimulation: comparison of the spinal circuits that generate and control locomotion in rats, cats and humans. *Exp. Neurol.* 209, 417–425. doi: 10.1016/j.expneurol.2007.07.015

Getting, P. A. (1977). Neuronal organization of escape swimming in Tritonia. *J. Comp. Physiol.* 121, 325–342. doi: 10.1007/bf00613012

Ginty, D. D., Bading, H., and Greenberg, M. E. (1992). Trans-synaptic regulation of gene expression. *Curr. Opin. Neurobiol.* 2, 312–316. doi: 10.1016/0959-4388(92)90121-z

Giroux, N., Rossignol, S., and Reader, T. A. (1999). Autoradiographic study of alpha1- and alpha2-noradrenergic and serotonin1A receptors in the spinal cord of normal and chronically transected cats. *J. Comp. Neurol.* 406, 402–414. doi: 10.1002/(sici)1096-9861(19990412)406:3<402::aid-cne8>3.3.co;2-6

Giszter, S., Patil, V., and Hart, C. (2007). Primitives, premotor drives and pattern generation: a combined computational and neuroethological perspective. *Prog. Brain Res.* 165, 323–346. doi: 10.1016/s0079-6123(06)65020-6

Gómez-Pinilla, F., Ying, Z., Roy, R. R., Hodgson, J., and Edgerton, V. R. (2004). Afferent input modulates neurotrophins and synaptic plasticity in the spinal cord. *J. Neurophysiol.* 92, 3423–3432. doi: 10.1152/jn.00432.2004

Gosgnach, S., Lanuza, G. M., Butt, S. J., Saueressig, H., Zhang, Y., Velasquez, T., et al. (2006). V1 spinal neurons regulate the speed of vertebrate locomotor outputs. *Nature* 440, 215–219. doi: 10.1038/nature04545

Gossard, J. P., Brownstone, R. M., Barajon, I., and Hultborn, H. (1994). Transmission in a locomotor related group Ib pathway from hindlimb extensor muscles in the cat. *Exp. Brain Res.* 98, 213–228. doi: 10.1007/bf00228410

Goulding, M. (2009). Circuits controlling vertebrate locomotion: moving in a new direction. *Nat. Rev. Neurosci.* 10, 507–518. doi: 10.1038/nrn2608

Graham Brown, T. (1911). The intrinsic factors in the act of progresson in the mammal. *Proc. R. Soc. Lond.* 84, 309–319. doi: 10.1098/rspb.1911.0077

Graham Brown, T. (1914). On the nature of the fundamental activity of the nervous centres; together with an analysis of the conditioning of rhythmic activity in progression and a theory of the evolution of function in the nervous system. *J. Physiol.* 48, 18–46.

Grillner, S. (1981). "Control of locomotion in bipeds, tetrapods and fish," in *Handbook of Physiology. The Nervous System II*, eds J. M. Brookhart and V. B. Mountcastle (Bethesda: Am. Physiol. Sco.), 1179–1236.

Grillner, S. (2006). Neuronal networks in motion from ion channels to behaviour. *An. R. Acad. Nac. Med. (Madr).* 123, 297–298.

Grillner, S., and Wallén, P. (1985). The ionic mechanisms underlying N-methyl-D-aspartate receptor-induced, tetrodotoxin-resistant membrane potential oscillations in lamprey neurons active during locomotion. *Neurosci. Lett.* 60, 289–294. doi: 10.1016/0304-3940(85)90592-0

Grillner, S., and Zangger, P. (1974). Locomotor movements generated by the deafferented spinal cord. *Acta Physiol. Scand.* 91, 38–39.

Grillner, S., and Zangger, P. (1975). How detailed is the central pattern generation for locomotion? *Brain Res.* 88, 367–371. doi: 10.1016/0006-8993(75)90401-1

Grob, M., and Guertin, P. A. (2007). [Role of Ca(2+) in the pacemaker-like property of spinal motoneurons]. *Med. Sci. (Paris)* 23, 64–66. doi: 10.1051/medsci/200723164

Guertin, P. (1996). "Central mediation of group 1 muscle afferent evoked adaptation of the locomotor step cycle in decerebrate cats". *Thesis*, (University of Manitoba).

Guertin, P., Angel, M. J., Perreault, M.-C., and McCrea, D. A. (1995). Ankle extensor group I afferents excite extensors throughout the hindlimb during fictive locomotion in the cat. *J. Physiol.* 487, 197–209.

Guertin, P. A. (2004). Role of NMDA receptor activation in serotonin agonist-induced air-stepping in paraplegic mice. *Spinal Cord* 42, 185–190. doi: 10.1038/sj.sc.3101580

Guertin, P. A. (2005). Semiquantitative assessment of hindlimb movement recovery without intervention in adult paraplegic mice. *Spinal Cord* 43, 162–166. doi: 10.1038/sj.sc.3101701

Guertin, P. A. (2008). A technological platform to optimize combinatorial treatment design and discovery for chronic spinal cord injury. *J. Neurosci. Res.* 86, 3039–3051. doi: 10.1002/jnr.21761

Guertin, P. A. (2009a). The mammalian central pattern generator for locomotion. *Brain Res. Rev.* 62, 45–56. doi: 10.1016/j.brainresrev.2009.08.002

Guertin, P. A. (2009b). Recovery of locomotor function with combinatory drug treatments designed to synergistically activate specific neuronal networks. *Curr. Med. Chem.* 16, 1366–1371. doi: 10.2174/092986709787846541

Guertin, P. A. (2012). "The spinal cord: functional organization, diseases and dysfunctions," in *Neuromethods: Animal Models in Spinal Cord Repair*, ed H. Aldskogius (New York, NY: Humana Press), 1–23.

Guertin, P. A., and Hounsgaard, J. (1998a). Chemical and electrical stimulation induce rhythmic motor activity in an in vitro preparation of the spinal cord from adult turtles. *Neurosci. Lett.* 245, 5–8. doi: 10.1016/s0304-3940(98)00164-5

Guertin, P. A., and Hounsgaard, J. (1998b). NMDA-Induced intrinsic voltage oscillations depend on L-type calcium channels in spinal motoneurons of adult turtles. *J. Neurophysiol.* 80, 3380–3382.

Guertin, P. A., and Steuer, I. (2005). Ionotropic 5-HT3 receptor agonist-induced motor responses in the hindlimbs of paraplegic mice. *J. Neurophysiol.* 94, 3397–3405. doi: 10.1152/jn.00587.2005

Guertin, P. A., and Steuer, I. (2009). Key central pattern generators of the spinal cord. *J. Neurosci. Res.* 87, 2399–2405. doi: 10.1002/jnr.22067

Guertin, P. A., Ung, R. V., and Rouleau, P. (2010). Oral administration of a tri-therapy for central pattern generator activation in paraplegic mice: proof-of-concept of efficacy. *Biotechnol. J.* 5, 421–426. doi: 10.1002/biot.200900278

Guertin, P. A., Ung, R. V., Rouleau, P., and Steuer, I. (2011). Effects on locomotion, muscle, bone and blood induced by a combination therapy eliciting weight-bearing stepping in nonassisted spinal cord-transected mice. *Neurorehabil. Neural Repair* 25, 234–242. doi: 10.1177/1545968310378753

Gurfinkel, V. S., and Shik, M. L. (1973). "The control of posture and locomotion," in *Motor Control*, eds A. A. Gydikov, N. T. Tankov and D. S. Kosarov (New York: Plenum Press), 217–234.

Guttman, L., and Walsh, J. J. (1971). Prostigmine assessment test of fertility in spinal man. *Paraplegia* 9, 39–51. doi: 10.1038/sc.1971.7

Harkema, S., Behrman, A., and Barbeau, H. (2012). Evidence-based therapy for recovery of function after spinal cord injury. *Handb. Clin. Neurol.* 109, 259–274. doi: 10.1016/B978-0-444-52137-8.00016-4

Harkema, S. J. (2008). Plasticity of interneuronal networks of the functionally isolated human spinal cord. *Brain Res. Rev.* 57, 255–264. doi: 10.1016/j.brainresrev.2007.07.012

Harris-Warrick, R. M. (2002). Voltage-sensitive ion channels in rhythmic motor systems. *Curr. Opin. Neurobiol.* 2, 646–651. doi: 10.1016/s0959-4388(02)00377-x

Harris-Warrick, R. M. (2011). Neuromodulation and flexibility in central pattern generator networks. *Curr. Opin. Neurobiol.* 21, 685–692. doi: 10.1016/j.conb.2011.05.011

Henneman, E. (1974). "Spinal reflexes and the control of movement," in *Medical Physiology* (Vol. 1), ed V. B. Mountcastle, 13th Edn. (Mosby, MO: St Louis), 651–667.

Hochman, S., Gozal, E. A., Hayes, H. B., Anderson, J. T., DeWeerth, S. P., and Chang, Y. H. (2012). Enabling techniques for in vitro studies on mammalian spinal locomotor mechanisms. *Front. Biosci. (Landmark Ed.)* 17, 2158–2180. doi: 10.2741/4043

Hochman, S., Jordan, L. M., and MacDonald, J. F. (1994). N-methyl-D-aspartate receptor-mediated voltage oscillations in neurons surrounding the central canal in slices of rat spinal cord. *J. Neurophysiol.* 72, 565–577.

Holinski, B. J., Mazurek, K. A., Everaert, D. G., Stein, R. B., and Mushahwar, V. K. (2011). Restoring stepping after spinal cord injury using intraspinal microstimulation and novel control strategies. *Conf. Proc. IEEE Eng. Med. Biol. Soc.* 2011, 5798–5801. doi: 10.1109/IEMBS.2011.6091435

Holmes, G. (1915). Spinal injuries of warfare. *Br. Med. J.* 2, 815–821. doi: 10.1136/bmj.2.2866.815

Hopper, S. L., and DiCaprio, R. A. (2004). Crustacean motor pattern generator networks. Invertebrate neural networks. *Neurosignals* 13, 50–69. doi: 10.1159/000076158

Hugues, G. M., and Wiersma, C. A. G. (1960). The coordination of swimmeret movements in the crayfish, procambarus clarkii (girard). *J. Exptl. Biol.* 39, 657–670.

Ivanenko, Y. P., Poppele, R. E., and Lacquantini, F. (2006). Motor control programs and walking. *Neuroscientist* 12, 339–348. doi: 10.1177/1073858406287987

Jankowska, E. (2008). Spinal interneuronal networks in the cat: elementary components. *Brain Res. Rev.* 57, 46–55. doi: 10.1016/j.brainresrev.2007.06.022

Jankowska, E., Jukes, M. G., Lund, S., and Lundberg, A. (1967a). The effect of DOPA on the spinal cord. 5. Reciprocal organization of pathways transmitting excitatory action to alpha motoneurones of flexors and extensors. *Acta Physiol. Scand.* 70, 369–388. doi: 10.1111/j.1748-1716.1967.tb03636.x

Jankowska, E., Jukes, M. G., Lund, S., and Lundberg, A. (1967b). The effect of DOPA on the spinal cord. 6. Half-centre organization of interneurones transmitting effects from the flexor reflex afferents. *Acta Physiol. Scand.* 70, 389–402. doi: 10.1111/j.1748-1716.1967.tb03637.x

Johnston, M. V., Mullaney, B., and Blue, M. E. (2003). Neurobiology of rett syndrome. *J. Child Neurol.* 18, 688–692. doi: 10.1177/08830738030180100501

Jonas, D., Linzbach, P., and Weber, W. (1979). The use of Midodrin in the treatment of ejaculation disorders following retroperitoneal lymphadenectomy. *Eur. Urol.* 5, 184–187.

Jones, R., and Cavanna, A. E. (2012). The neurobiology and treatment of restless legs syndrome. *Behav. Neurol.* 26, 283–292. doi: 10.1155/2013/585439

Jordan, L. M., Liu, J., Hedlund, P. B., Akay, T., and Pearson, K. G. (2008). Descending command systems for the initiation of locomotion in mammals. *Brain Res. Rev.* 57, 183–191. doi: 10.1016/j.brainresrev.2007.07.019

Juvin, L., Le Gal, J. P., Simmers, J., and Morin, D. (2012). Cervicolumbar coordination in mammalian quadrupedal locomotion: role of spinal thoracic circuitry and limb sensory inputs. *J. Neurosci.* 32, 953–965. doi: 10.1523/jneurosci.4640-11.2012

Kalat, J. W. (1998). *Biological Psychology.* 6th Edn. Pacific Grove, CA: Brooks/Cole Publishing Company.

Kandel, E. R., Schwartz, J. H., and Jessell, T. M. (2000). *Principles of Neural Science.* 4th Edn. New York: McGraw-Hill.

Kiehn, O., and Kjaerulff, O. (1996). Spatiotemporal characteristics of 5-HT and dopamine-induced rhythmic hindlimb activity in the in vitro neonatal rat. *J. Neurophysiol.* 75, 1472–1482.

Kiehn, O., Quinlan, K. A., Restrepo, C. E., Lundfald, L., Borgius, L., Talpalar, A. E., et al. (2008). Excitatory components of the mammalian locomotor CPG. *Brain Res. Rev.* 57, 56–63. doi: 10.1016/j.brainresrev.2007.07.002

Kjaerulff, O., Barajon, I., and Kiehn, O. (1994). Sulphorhodamine-labelled cells in the neonatal rat spinal cord following chemically induced locomotor activity in vitro. *J. Physiol.* 478, 265–273.

Knikou, M., Angeli, C. A., Ferreira, C. K., and Harkema, S. J. (2009). Flexion reflex modulation during stepping in human spinal cord injury. *Exp. Brain Res.* 196, 341–351. doi: 10.1007/s00221-009-1854-x

Knusel, J., Bicanski, A., Ryczko, D., Cabelguen, J. M., and Ljspeert, A. J. (2013). A salamder's flexible spinal network for locomotion, modeled at two levels of abstraction. *Integr. Comp. Biol.* 53, 269–282. doi: 10.1093/icb/ict067

Kriellaars, D. J., Brownstone, R. M., Noga, B. R., and Jordan, L. M. (1994). Mechanical entrainment of fictive locomotion in the decerebrate cat. *J. Neurophysiol.* 71, 2074–2086.

Kriellaars, D. J. (1992). *Generation and Peripheral Control of Locomotor Rhythm*. Ph.D. thesis. Manitoba: University of Manitoba.

Kristan, W. B. Jr., and Weeks, J. C. (1983). Neurons controlling the initiation, generation and modulation of leech swimming. *Symp. Soc. Exp. Biol.* 37, 243–260.

Kullander, K., Butt, S. J., Lebret, J. M., Lundfald, L., Restrepo, C. E., Rydstrom, A., et al. (2003). Role of EphA4 and EphrinB3 in local neuronal circuits that control walking. *Science* 299, 1889–1892. doi: 10.1126/science.1079641

Lafreniere-Roula, M., and McCrea, D. A. (2005). Deletions of rhythmic motoneuron activity during fictive locomotion and scratch provide clues to the organization of the mammalian central pattern generator. *J. Neurophysiol.* 94, 1120–1132. doi: 10.1152/jn.00216.2005

Landry, E. S., and Guertin, P. A. (2004). Differential effects of 5-HT1 and 5-HT2 receptor agonists on hindlimb movements in paraplegic mice. *Prog. Neuropsychopharmacol. Biol. Psychiatry* 28, 1053–1060. doi: 10.1016/j.pnpbp.2004.05.001

Landry, E. S., Lapointe, N. P., Rouillard, C., Levesque, D., Hedlund, P. B., and Guertin, P. A. (2006a). Contribution of spinal 5-HT1A and 5-HT7 receptors to locomotor-like movement induced by 8-OH-DPAT in spinal cord-transected mice. *Eur. J. Neurosci.* 24, 535–546. doi: 10.1111/j.1460-9568.2006.04917.x

Landry, E. S., Rouillard, C., Levesque, D., and Guertin, P. A. (2006b). Profile of immediate early gene expression in the lumbar spinal cord of low-thoracic paraplegic mice. *Behav. Neurosci.* 120, 1384–1388. doi: 10.1037/0735-7044.120.6.1384

Langlet, C., Leblond, H., and Rossignol, S. (2005). Mid-lumbar segments are needed for the expression of locomotion in chronic spinal cats. *J. Neurophysiol.* 93, 2474–2488. doi: 10.1152/jn.00909.2004

Lanuza, G. M., Gosgnach, S., Pierani, A., Jessell, T. M., and Goulding, M. (2004). Genetic identification of spinal interneurons that coordinate left-right locomotor activity necessary for walking movements. *Neuron* 42, 375–386. doi: 10.1016/s0896-6273(04)00249-1

Lapointe, N. P., Ung, R. V., Rouleau, P., and Guertin, P. A. (2008). Effects of spinal alpha(2)-adrenoceptor and I(1)-imidazoline receptor activation on hindlimb movement induction in spinal cord-injured mice. *J. Pharmacol. Exp. Ther.* 325, 994–1006. doi: 10.1124/jpet.107.134874

Lapointe, N. P., Rouleau, P., Ung, R. V., and Guertin, P. A. (2009). Specific role of D1 receptors in spinal network activation and rhythmic movement induction in vertebrates. *J. Physiol.* 587, 1499–1511. doi: 10.1113/jphysiol.2008.166314

Le Ray, D., Juvin, L., Ryczko, D., and Dubuc, R. (2011). Chapter 4—supraspinal control of locomotion: the mesencephalic locomotor region. *Prog. Brain Res.* 188, 51–70. doi: 10.1016/B978-0-444-53825-3.00009-7

Lhermitte, J. (1919). *La section totale de la Moelle Dorsale.* Bourges, France: Tardy Pigelet.

Li, X. L., Zhang, W., Zhou, X., Wang, X. Y., Zhang, H. T., Qin, D. X., et al. (2007). Temporal changes in the expression of some neurotrophins in spinal cord transected adult rats. *Neuropeptides* 41, 135–143. doi: 10.1016/j.npep.2007.02.001

Liu, Y. C., Bailey, I., and Hale, M. E. (2012). Alternative startle motor patterns and behaviors in the larval zebrafish (Danio rerio). *J. Comp. Physiol. A Neuroethol. Sens. Neural Behav. Physiol.* 198, 11–24. doi: 10.1007/s00359-011-0682-1

Lovely, R. G., Gregor, R. J., Roy, R. R., and Edgerton, V. R. (1986). Effects of training on the recovery of full-weight-bearing stepping in the adult spinal cat. *Exp. Neurol.* 92, 421–435. doi: 10.1016/0014-4886(86)90094-4

Lukacova, N., Kolesarova, M., Kucharova, K., Pavel, J., Kolesar, D., Radonak, J., et al. (2006). The effect of a spinal cord hemisection on changes in nitric oxide synthase pools in the site of injury and in regions located far away from the injured site. *Cell. Mol. Neurobiol.* 26, 1365–1383. doi: 10.1007/s10571-006-9092-2

Lundfald, L., Restrepo, C. E., Butt, S. J., Peng, C. Y., Droho, S., Endo, T., et al. (2007). Phenotype of V2-derived interneurons and their relationship to the axon guidance molecule EphA4 in the developing mouse spinal cord. *Eur. J. Neurosci.* 26, 2989–3002. doi: 10.1111/j.1460-9568.2007.05906.x

MacLean, J. N., Schmidt, B. J., and Hochman, S. (1997). NMDA receptor activation triggers voltage oscillations, plateau potentials and bursting in neonatal rat lumbar motoneurons in vitro. *Eur. J. Neurosci.* 9, 2702–2711. doi: 10.1111/j.1460-9568.1997.tb01699.x

Mallory, B. S., Roppolo, J. R., and de Groat, W. C. (1991). Pharmacological modulation of the pontine micturition center. *Brain Res.* 546, 310–320.

Matsumoto, N., Yoshida, M., and Uematsu, K. (2007). Effects of partial ablation of the cerebellum on sustained swimming in goldfish. *Brain Behav. Evol.* 70, 105–114. doi: 10.1159/000102972

Matthews, P. B. C. (1972). *Mammalian Muscle Receptors and Their Central Actions.* Baltimore, MD: Williams and Wilkins.

Matthews, P. B. C. (1991). The human stretch reflex and the motor cortex. *Trends Neurosci.* 14, 87–91. doi: 10.1016/0166-2236(91)90064-2

McCrea, D. A., and Rybak, I. A. (2008). Organization of mammalian locomotor rhythm and pattern generation. *Brain Res. Rev.* 57, 134–146. doi: 10.1016/j.brainresrev.2007.08.006

McCrea, D. A., Pratt, C. A., and Jordan, L. M. (1980). Renshaw cell activity and recurrent effects on motoneurons during fictive locomotion. *J. Neurophys.* 44, 475–488.

McKenna, K. E., Chung, S. K., and McVary, K. T. (1991). A model for the study of sexual function in anesthetized male and female rats. *Am. J. Physiol.* 261, 1276–1285.

Miller, S., and Scott, P. D. (1977). The spinal locomotor generator. *Exp. Brain Res.* 30, 387–403. doi: 10.1007/bf00237264

Moreland, A. J., and Makela, E. H. (2005). SSRIs in the treatment of premature ejaculation. *Ann. Pharmacother.* 39, 1296–1301.

Moshonkina, T. R., Makarovski, A. N., Bogacheva, I. N., Scherbakova, N. A., Savohin, A. A., and Gerasimenko, Y. P. (2012). Effects of spinal cord electrical stimulation in patients with vertebrospinal pathology. *Bull. Exp. Biol. Med.* 153, 16–20. doi: 10.1007/s10517-012-1632-9

Nadeau, S., Jacquemin, G., Fournier, C., Lamarre, Y., and Rossignol, S. (2010). Spontaneous motor rhythms of the back and legs in a patient with a complete spinal cord transection. *Neurorehabil. Neural Repair* 24, 377–383. doi: 10.1177/1545968309349945

Nadelhaft, I., and Vera, P. L. (1995). Central nervous system neurons infected by pseudorabies virus injected into the rat urinary bladder following unilateral transection of the pelvic nerve. *J. Comp. Neurol.* 359, 443–456. doi: 10.1002/cne.903590307

Netter, F. H. (2006). *Atlas of Human Anatomy.* Philadelphia, PA: Saunders/Elsevier.

Nishimaru, H., Takizawa, H., and Kudo, N. (2000). 5-Hydroxytryptamine-induced locomotor rhythm in the neonatal mouse spinal cord in vitro. *Neurosci. Lett.* 280, 187–190. doi: 10.1016/s0304-3940(00)00805-3

Noga, B. R., Johnson, D. M., Riesgo, M. I., and Pinzon, A. (2009). Locomotor-activated neurons of the cat. I. Serotonergic innervation and co-localization of 5-HT7, 5-HT2A, and 5-HT1A receptors in the thoraco-lumbar spinal cord. *J. Neurophysiol.* 102, 1560–1576. doi: 10.1152/jn.91179.2008

Ollivier-Lanvin, K., Krupka, A. J., AuYong, N., Miller, K., Prilutsky, B. I., and Lemay, M. A. (2011). Electrical stimulation of the sural cutaneous afferent nerve controls the amplitude and onset of the swing phase of locomotion in the spinal cat. *J. Neurophysiol.* 105, 2297–2308. doi: 10.1152/jn.00385.2010

Orlovski, G. N., Severin, F. V., and Shik, M. L. (1966). Effect of speed and load on coordination of movements during running of the dog. *Biophysics* 11, 414–417.

Orlovskii, G. N., Severin, F. V., and Shik, M. L. (1966). Locomotion induced by stimulation of the mesencephalon. *Dokl. Akad. Nauk. SSSR* 169, 1223–1226.

Orlovsky, G., and Shik, M. (1976). Control of locomotion: a neurophysiological analysis of the cat locomotor system. *Intern. Rev. Physiol. Ser. II* 10, 281–317.

Patestas, M. A., and Gartner, L. P. (2006). "Ascending sensory pathways," in *A Textbook of Neuroanatomy*, eds M. A. Patestas and L. P. Gartner (Etobicoke, ON: Wiley-Blackwell), 137–170.

Pearson, K., and Duysens, J. (1976). "Function of segmental reflexes in the control of stepping in cockroaches and cats," in *Neural Control of Locomotion*, eds R. Herman, S. Grillner, P. S. G. Stein and D. Stuart (New York: Plenum Press), 519–538.

Pearson, K. G. (1993). Common principles of motor control in vertebrates and invertebrates. *Annu. Rev. Neurosci.* 16, 265–297. doi: 10.1146/annurev.neuro.16.1.265

Pearson, K. G. (1995). Proprioceptive regulation of locomotion. *Curr. Opin. Neurobiol.* 5, 786–791. doi: 10.1016/0959-4388(95)80107-3

Pearson, K. G., and Rossignol, S. (1991). Fictive motor patterns in chronic spinal cats. *J. Neurophysiol.* 66, 1874–1887.

Peng, X. M., Zhou, Z. G., Glorioso, J. C., Fink, D. J., and Mata, M. (2006). Tumor necrosis factor-alpha contributes to below-level neuropathic pain after spinal cord injury. *Ann. Neurol.* 59, 843–851. doi: 10.1002/ana.20855

Perreault, M. C., Angel, M. J., Guertin, P., and McCrea, D. A. (1995). Effects of stimulation of hindlimb flexor group II afferents during fictive locomotion in the cat. *J. Physiol.* 487(Pt. 1), 211–220.

Perret, C., and Cabelguen, J. M. (1980). Main characteristics of the hindlimb locomotor cycle in the decorticate cat with special reference to bifunctional muscles. *Brain Res.* 187, 333–352. doi: 10.1016/0006-8993(80)90207-3

Pomerantz, S. M., Hepner, B. C., and Wertz, J. M. (1993). Serotonergic influences on male sexual behavior of rhesus monkeys: effects of serotonin agonists. *Psychopharmacology (Berl)* 111, 47–54. doi: 10.1007/bf02257406

Pozos, R. S., and Iaizzo, P. A. (1991). Shivering and pathological and physiological clonic oscillations of the human ankle. *J. Appl. Physiol. (1985)* 71, 1929–1932.

Pustovit, R. V., Callaghan, B., Kosari, S., Rivera, L. R., Thomas, H., Brock, J. A., et al. (2014). The mechanism of enhanced defecation caused by the ghrelin receptor agonist, ulimorelin. *Neurogastroenterol Motil.* 26, 264–271. doi: 10.1111/nmo.12259

Reith, C. A., and Sillar, K. T. (1998). A role for slow NMDA receptor-mediated, intrinsic neuronal oscillations in the control of fast fictive swimming in Xenopus laevis larvae. *Eur. J. Neurosci.* 10, 1329–1340. doi: 10.1046/j.1460-9568.1998.00144.x

Reitz, A., Knapp, P. A., Müntener, M., and Schurch, B. (2004). Oral nitric oxide donors: a new pharmacological approach to detrusor-sphincter dyssynergia in spinal cord injured patients? *Eur. Urol.* 45, 516–520. doi: 10.1016/j.eururo.2003.11.006

Rémy-Néris, O., Barbeau, H., Daniel, O., Boiteau, F., and Bussel, B. (1999). Effects of intrathecal clonidine injection on spinal reflexes and human locomotion in incomplete paraplegic subjects. *Exp. Brain Res.* 129, 433–440. doi: 10.1007/s002210050910

Riley, A. J., and Riley, E. J. (1982). Partial ejaculatory incompetence: the therapeutic effect of midodrine, an orally active selective alpha-adrenoceptor agonist. *Eur. Urol.* 8, p155–p160.

Robertson, R. M., and Pearson, K. G. (1985). Neural circuits in the flight system of the locust. *J. Neurophysiol.* 53, 110–128.

Rossignol, S., Giroux, N., Chau, C., Marcoux, J., Brustein, E., and Reader, T. A. (2001). Pharmacological aids to locomotor training after spinal injury in the cat. *J. Physiol.* 533(Pt. 1), 65–74. doi: 10.1111/j.1469-7793.2001.0065b.x

Rostad, H. (1973). Extrinsic and central nervous control of colonic motility. A survey of previous and new concepts based on experimental results. *J. Oslo City Hosp.* 23, 65–75.

Rye, D. B., and Trotti, L. M. (2012). Restless legs syndrome and periodic leg movements of sleep. *Neurol. Clin.* 30, 1137–1166. doi: 10.1016/j.ncl.2012.08.004

Schmidt, B. J., and Jordan, L. M. (2000). The role of serotonin in reflex modulation and locomotor rhythm production in the mammalian spinal cord. *Brain Res. Bull.* 53, 689–710. doi: 10.1016/s0361-9230(00)00402-0

Schmidt, B. J., Hochman, S., and MacLean, J. N. (1998). NMDA receptor-mediated oscillatory properties: potential role in rhythm generation in the mammalian spinal cord. *Ann. N Y Acad. Sci.* 860, 189–202. doi: 10.1111/j.1749-6632.1998.tb09049.x

Schmitt, C., Miranpuri, G. S., Dhodda, V. K., Isaacson, J., Vemuganti, R., and Resnick, D. K. (2006). Changes in spinal cord injury-induced gene expression in rat are strain-dependent. *Spine J.* 6, 113–119. doi: 10.1016/j.spinee.2005.05.379

Schomburg, E. D., Petersen, N., Barajon, I., and Hultborn, H. (1998). Flexor reflex afferents reset the step cycle during fictive locomotion in the cat. *Exp. Brain Res.* 122, 339–350. doi: 10.1007/s002210050522

Schrøder, H. D. (1985). Anatomical and pathoanatomical studies on the spinal efferent systems innervating pelvic structures. 1. Organization of spinal nuclei in animals. 2. The nucleus X-pelvic motor system in man. *J. Auton. Nerv. Syst.* 14, 23–48. doi: 10.1016/0165-1838(85)90123-7

Schurch, B., Stöhrer, M., Kramer, G., Schmid, D. M., Gaul, G., and Hauri, D. (2000). Botulinum-A toxin for treating detrusor hyperreflexia in spinal cord injured patients: a new alternative to anticholinergic drugs? Preliminary results. *J. Urol.* 164, 692–697. doi: 10.1016/S0022-5347(05)67283-7

Schürks, M., and Bussfeld, P. (2013). Multiple sclerosis and restless legs syndrome: a systematic review and meta-analysis. *Eur. J. Neurol.* 20, 605–615. doi: 10.1111/j.1468-1331.2012.03873.x

Selionova, V. A., Ivanenko, Y. P., Solopova, I. A., and Gurfinkel, V. S. (2009). Tonic central and sensory stimuli facilitate involuntary air-stepping in humans. *J. Neurophysiol.* 101, 2847–2858. doi: 10.1152/jn.90895.2008

Shapkova, E. Y., and Schomburg, E. D. (2001). Two types of motor modulation underlying human stepping by spinal cord electrical stimulation (SCES). *Acta Physiol. Pharmacol. Bulg.* 26, 155–157.

Sherrington, C. S. (1910). Flexion-reflex of the limb, crossed extension-reflex and reflex stepping and standing. *J. Physiol.* 40, 28–121.

Shimizu, Y., Chang, E. C., Shafton, A. D., Ferens, D. M., Sanger, G. J., Witherington, J., et al. (2006). Evidence that stimulation of ghrelin receptors in the spinal cord initiates propulsive activity in the colon of the rat. *J. Physiol.* 576(Pt. 1), 329–338. doi: 10.1113/jphysiol.2006.116160

Sigvardt, K. A., Grillner, S., Wallen, P., and Van Dongen, P. A. (1985). Activation of NMDA receptors elicits fictive locomotion and bistable membrane properties in the lamprey spinal cord. *Brain Res.* 336, 390–395. doi: 10.1016/0006-8993(85)90676-6

SŁawinska, U., Rossignol, S., Bennett, D. J., Schmidt, B. J., Frigon, A., Fouad, K., et al. (2012). Comment on "Restoring voluntary control of locomotion after paralyzing spinal cord injury". *Science* 338:328. doi: 10.1126/science.1226082

Staerman, F., Bryckaert, P. E., Youinou, Y., Colin, J., Brandt, B., and Lardennois, B. (2001). Pharmacologic stimulation of ejaculation with midodrine hydrochloride (Gutron) for medically assisted reproduction in spinal injury. *Prog. Urol.* 11, 1264–1268.

Stafford, S. A., Bowery, N. G., Tang, K., and Coote, J. H. (2006). Activation by p-chloroamphetamine of the spinal ejaculatory pattern generator in anaesthetized male rats. *Neuroscience* 140, 1031–1040. doi: 10.1016/j.neuroscience.2006.02.039

Stein, R. B., and Capaday, C. (1988). The modulation of human reflexes during functional motor tasks. *Trends Neurosci.* 11, 328–332. doi: 10.1016/0166-2236(88)90097-5

Stuart, D. G., and Hultborn, H. (2008). Thomas Graham Brown (1882–1965), Anders Lundberg (1920-) and the neural control of stepping. *Brain Res. Rev.* 59, 74–95. doi: 10.1016/j.brainresrev.2008.06.001

Sugaya, K., Nishijima, S., Miyazato, M., and Ogawa, Y. (2005). Central nervous control of micturition and urine storage. *J. Smooth Muscle Res.* 41, 117–132. doi: 10.1540/jsmr.41.117

Sugaya, K., Roppolo, J. R., Yoshimura, N., Card, J. P., and de Groat, W. C. (1997). The central neural pathways involved in micturition in the neonatal rat as revealed by the injection of pseudorabies virus into the urinary bladder. *Neurosci. Lett.* 223, 197–200. doi: 10.1016/s0304-3940(97)13433-4

Székely, G., Czéh, G., and Voros, G. (1969). The activity pattern of limb muscles in freely moving normal and deafferented newts. *Exp. Brain Res.* 9, 53–72. doi: 10.1007/bf00235451

Tan, U. (2006a). A new syndrome with quadrupedal gait, primitive speech, and severe mental retardation as a live model for human evolution. *Int. J. Neurosci.* 116, 361–369. doi: 10.1080/00207450500455330

Tan, U. (2006b). Evidence for "Uner Tan Syndrome" and the evolution of the human mind. *Int. J. Neurosci.* 116, 763–774. doi: 10.1080/00207450600588733

Tan, U. (2008). Uner Tan syndrome: review and report of four new cases. *Int. J. Neurosci.* 118, 211–225. doi: 10.1080/00207450701667808

Thorpe, A. J., Clair, A., Hochman, S., and Clemens, S. (2011). Possible sites of therapeutic action in restless legs syndrome: focus on dopamine and α2δ ligands. *Eur. Neurol.* 66, 18–29. doi: 10.1159/000328431

Tillakaratne, N. J., de Leon, R. D., Hoang, T. X., Roy, R. R., Edgerton, V. R., and Tobin, A. J. (2002). Use-dependent modulation of inhibitory capacity in the feline lumbar spinal cord. *J. Neurosci.* 22, 3130–3143.

Tresch, M. C., Saltiel, P., and Bizzi, E. (1999). The construction of movement by the spinal cord. *Nat. Neurosci.* 2, 162–167. doi: 10.1038/5721

Truitt, W. A., and Coolen, L. M. (2002). Identification of a potential ejaculation generator in the spinal cord. *Science* 297, 1566–1569. doi: 10.1126/science.1073885

Ung, R. V., Landry, E. S., Rouleau, P., Lapointe, N. P., Rouillard, C., and Guertin, P. A. (2008). Role of spinal 5-HT2 receptor subtypes in quipzine-induced hindlimb movements after a low-thoracic spinal cord transection. *Eur. J. Neurosci.* 28, 2231–2242. doi: 10.1111/j.1460-9568.2008.06508.x

Ung, R. V., Lapointe, N. P., Tremblay, C., Larouche, A., and Guertin, P. A. (2007). Spontaneous recovery of hindlimb movement in completely spinal cord transected mice: a comparison of assessment methods and conditions. *Spinal Cord* 45, 367–379.

van den Brand, R., Heutschi, J., Barraud, Q., DiGiovanna, J., Bartholdi, K., Huerlimann, M., et al. (2012). Restoring voluntary control of locomotion after paralyzing spinal cord injury. *Science* 336, 1182–1185. doi: 10.1126/science.1217416

Vargas, V. M., Torres, D., Corona, F., Vergara, M., Gomez, L. E., Delgado-Lezama, R., et al. (2004). Cholinergic facilitation of erection and ejaculation in spinal cord-transected rats. *Int. J. Impot Res.* 16, 86–90. doi: 10.1038/sj.ijir.3901169

Viala, D., and Buser, P. (1969). The effects of DOPA and 5-HTP on rhythmic efferent discharges in hind limb nerves in the rabbit. *Brain Res.* 12, 437–443. doi: 10.1016/0006-8993(69)90011-0

Vizzard, M. A., Erickson, V. L., Card, J. P., Roppolo, J. R., and de Groat, W. C. (1995). Transneuronal labeling of neurons in the adult rat brainstem and spinal cord after injection of pseudorabies virus into the urethra. *J. Comp. Neurol* 355, 629–640. doi: 10.1002/cne.903550411

Wainberg, M., Barbeau, H., and Gauthier, S. (1990). The effects of cyproheptadine on locomotion and on spasticity in patients with spinal cord injuries. *J. Neurol. Neurosurg. Psychiatry* 53, 754–763. doi: 10.1136/jnnp.53.9.754

Wang, D., Grillner, S., and Wallén, P. (2006). Effects of flufenamic acid on fictive locomotion, plateau potentials, calcium channels and NMDA receptors in the lamprey spinal cord. *Neuropharmacology* 51, 1038–1046. doi: 10.1016/j.neuropharm.2006.06.012

Wilson, J. M., Cowan, A. I., and Brownstone, R. M. (2007). Heterogeneous electronic coupling and synchronization of rhythmic bursting activity in mouse HB9 interneurons. *J. Neurophysiol.* 98, 2370–2381. doi: 10.1152/jn.00338.2007

Wilson, J. M., Hartley, R., Maxwell, D. J., Todd, A. J., Lieberam, I., Kaltschmidt, J. A., et al. (2005). Conditional rhythmicity of ventral spinal interneurons defined by expression of the Hb9 homeodomain protein. *J. Neurosci.* 25, 5710–5719. doi: 10.1523/jneurosci.0274-05.2005

Yakovlev, A. G., and Faden, A. I. (1994). Sequential expression of c-fos protooncogene, TNF-alpha and dynorphin genes in spinal cord following experimental traumatic injury. *Mol. Chem. Neuropathol.* 23, 179–190. doi: 10.1007/BF02815410

Yonezawa, A., Yoshizumi, M., Ebiko, M., Ise, S. N., Watanabe, C., Mizoguchi, H., et al. (2008). Ejaculatory response induced by a 5-HT2 receptor agonist m-CPP in rats: differential roles of 5-HT2 receptor subtypes. *Pharmacolo. Biochem. Behav.* 88, 367–373. doi: 10.1016/j.pbb.2007.09.009

Yoshiyama, M., and de Groat, W. C. (2005). Supraspinal and spinal α-amino-3-hydroxy-5-methylisoxazole-4-propionic acid and N-methyl-D-aspartate glutamatergic control of the micturition reflex in the urethane anesthetized rat. *Neuroscience* 132, 1017–1026. doi: 10.1016/j.neuroscience.2005.01.041

Zhang, S. X., Huang, F., Gates, M., White, J., and Holmberg, E. G. (2010). Tail nerve electrical stimulation induces body weight-supported stepping in rats with spinal cord injury. *J. Neurosci. Methods* 187, 183–189. doi: 10.1016/j.jneumeth.2010.01.008

Zhang, Y., Narayan, S., Geiman, E., Lanuza, G. M., Velasquez, T., Shanks, B., et al. (2008). V3 spinal neurons establish a robust and balanced locomotor rhythm during walking. *Neuron* 60, 84–96. doi: 10.1016/j.neuron.2008.09.027

Conflict of Interest Statement: Pierre A. Guertin is professor in neurosciences at Laval University as well as CEO of a biopharmaceutical company, Nordic Life Science Pipeline Inc.

Received: 20 December 2013; paper pending published: 07 February 2014; accepted: 11 April 2014; published online: 30 May 2014.

*Citation: Guertin PA (2014) Preclinical evidence supporting the clinical development of central pattern generator-modulating therapies for chronic spinal cord-injured patients. Front. Hum. Neurosci. **8**:272. doi: 10.3389/fnhum.2014.00272*

This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 26 June 2014
doi: 10.3389/fnhum.2014.00371

The contribution of a central pattern generator in a reflex-based neuromuscular model

Florin Dzeladini*, Jesse van den Kieboom and Auke Ijspeert

BioRob, School of Engineering, Institute of Bioengineering, École Polytechnique Fédérale de Lausanne, Lausanne, Switzerland

Edited by:
Federica Tamburella, IRCCS Fondazione Santa Lucia, Italy

Reviewed by:
Francesca Sylos Labini, IRCCS Santa Lucia Foundation, Italy
Marco Iosa, IRCCS Fondazione Santa Lucia, Italy

****Correspondence:***
Florin Dzeladini, BioRob, School of Engineering, Institute of Bioengineering École Polytechnique Fédérale de Lausanne, Station 14, INN 239 Lausanne, Vaud 1015, Switzerland
e-mail: florin.dzeladini@epfl.ch

Although the concept of central pattern generators (CPGs) controlling locomotion in vertebrates is widely accepted, the presence of specialized CPGs in human locomotion is still a matter of debate. An interesting numerical model developed in the 90s' demonstrated the important role CPGs could play in human locomotion, both in terms of stability against perturbations, and in terms of speed control. Recently, a reflex-based neuro-musculo-skeletal model has been proposed, showing a level of stability to perturbations similar to the previous model, without any CPG components. Although exhibiting striking similarities with human gaits, the lack of CPG makes the control of speed/step length in the model difficult. In this paper, we hypothesize that a CPG component will offer a meaningful way of controlling the locomotion speed. After introducing the CPG component in the reflex model, and taking advantage of the resulting properties, a simple model for gait modulation is presented. The results highlight the advantages of a CPG as feedforward component in terms of gait modulation.

Keywords: feedback, CPG, humans, locomotion, models, biological, movement, periodicity

1. INTRODUCTION

Central pattern generators (CPGs) are networks of neural cells that can generate coordinated rhythmic patterns in the absence of sensory feedbacks. The idea that CPG control locomotion in lower vertebrates has been widely accepted for several decades (Grillner and Wallen, 1985). Although many observations tend to favor the presence of such components in higher vertebrates (see MacKay-Lyons, 2002 for a review), the presence of specialized CPGs in human locomotion is still a matter of debate (Dimitrijevic et al., 1998). An interesting numerical model developed by Gentaro Taga in the 90s' demonstrated the role that CPGs could play in human locomotion. It was shown that walking and running could emerge from a rhythmic interaction (modeled by coupled oscillators, i.e., CPGs), between the central nervous system, the musculo-skeletal-system and the environment. The CPGs were modeled as a network of oscillators, coupled with the environment through joint angles and ground reaction forces (Taga, 1994). The intriguing robustness of the generated gaits against mechanical perturbations and changes in the environment was attributed to the use of CPGs and feedbacks, respectively, highlighting the important role of both components. However, more recently, a neuro-musculo-skeletal model (denoted FBL, for Feedback Based Locomotion) solely driven by reflex loops was proposed by Geyer and Herr (2010). The model showed a stability to perturbations similar to the previous model, without any CPG components, questioning the conclusions drawn by Taga et al. regarding the importance of CPGs to resist perturbations. Furthermore, the properties of the gaits produced by the FBL model were—in terms of muscles activity, joints angles and torques patterns—surprisingly close to those observed in humans. Yet, an important feature the reflex-driven neuro-musculo-skeletal system was unable to reproduce was the control of speed. Indeed, while in Taga's model, speed was controlled by a simple unique variable (the frequency of the oscillators), such a strategy is inapplicable in the reflex model. Although a preliminary speed control strategy has been proposed by Song and Geyer (2012), its complexity compared to the very simple descending signals, originating from the brain stem, able to control locomotion (found in lower vertebrates, such as the lamprey and the salamander, and even in cats) makes their relevance, from a biological point of view, questionable.

Given the striking properties of the reflex model, we wanted to study the possible benefits that a CPG would add to the model. We hypothesized that the reflex model would benefit from the presence of CPGs in terms of gait speed/step length control. The CPG component is derived from the feedback pathways, following an idea from Kuo (2002), where CPGs are viewed as feedback predictors. We use a variety of models combining CPG and feedbacks in different ways to study the relative importance of the different feedbacks/feedforward pathways. Finally, taking advantage of the properties of the CPG, a simple model for speed modulation is presented.

2. MATERIALS AND METHODS

In this section, we describe step-by-step how we generate the CPG-based extension of Geyer's FBL model, referred to as 3FBL (for FeedForward and Feedback Based Locomotion). We first present our implementation of the FBL model and detail its optimization. This model demonstrates that simple delayed feedback loops (i.e., delayed linear mapping between sensors state and muscles activities) combined with a simplified musculoskeletal

model (lower limb model of human based on anthropometric data, actuated by seven Hill muscle models per limb) is sufficient to generate walking at various frequencies and step lengths. Furthermore, when the objective function used for the optimization process includes a metabolic cost minimization criterion, the generated angles, torques and muscles activation are comparable to human walking data (replicating results found in Geyer and Herr, 2010 and Wang et al., 2012). Despite the interesting properties of the model, an important limitation is that, once a walking gait at a given speed and step length is obtained, the only way to modulate it is by tuning of the multiple feedback gains. For example, in Song and Geyer (2012), a speed controller has been derived based on feedback gains tuning. The proposed controller is able to switch between gaits of different speeds, but the strategy remains complex. In short, speed changes are obtained by switching between different sets of feedback gains; increasing speed is done by (1) switching to a set of gains that generate an acceleration, and (2) once the desired speed is reached, switching to a set of gains that generate a gait of the desired speed.

The gait modulation strategy we propose is based on evidence from lower vertebrates and quadrupeds suggesting that simple low dimensional descending signals are enough to modulate walking (speed changes and gait transitions) (Grillner and Wallen, 1985). Our strategy to introduce CPGs as a feedforward component is based on the assumption that CPGs can be viewed as feedback predictors. In other words, CPGs should be able to reproduce any feedback signals generated by a stable walking gait of the FBL model. Since the feedback signals can be of any shape, we do not want to make strong assumptions on the class of pattern. Therefore, we will use a special class of oscillators called "morphed non-linear phase oscillators," that have the ability to generate limit cycles of arbitrary shape (Ajallooeian et al., 2013). Note that we do not model individual neurons but rather use an abstract model of biological CPGs represented as a dynamical system exhibiting limit cycle behavior. This strategy is commonly used to test hypothesis on the role of biological CPGs (Ijspeert, 2008).

The CPGs will then be combined with feedback pathways using the strategy presented in Kuo (2002), offering an elegant and easy way to study the relative importance of the different feedback pathways. The proposed strategy will also permit to highlight the pathways that can be used as speed and step length modulators.

2.1. FBL DESCRIPTION

The pure feedback-based neuromuscular model of human locomotion (or FBL model) refers to a bio-inspired neuromuscular bipedal walking model developed by Geyer and Herr (2010) that we reimplemented and use as a starting point for our study. The following description is thus largely inspired by their work. Any differences with the original model will be explicitly stated.

In this study, all experiments are done using an implementation of the NMM library (a freely accessible C++ library that we developed to simulate neuromuscular models[1]) on the Webots robotic environment platform (Michel, 2004). This webots implementation[2] is based on an anthropometric model of human lower body (see Supplementary Figure 3, anthropometric data from Winter, 2009).

The FBL model uses feedback rules connecting different sources of sensory information (comprising muscle force and length feedbacks, ground reaction forces and joint angles) to Hill-type muscle models (details concerning the muscle model can be found in Geyer et al., 2003), which in turn generate effective joints torques. A state machine is used to switch between two sets of feedback rules: one to generate the stance phase control (mainly extensor muscles activity) and one to generate the swing phase control (mainly flexor muscles activity). Ground sensors placed under the feet are used to detect the state transition (takeoff and touchdown). The generation of the gait cycle is done through reflexes represented by a sequence of time delayed reactions (see **Figure 1**).

[1]The NMM library can be found online at https://bitbucket.org/efx/libnmm

[2]The Webots implementation of the NMM library can be found online at https://bitbucket.org/efx/sml

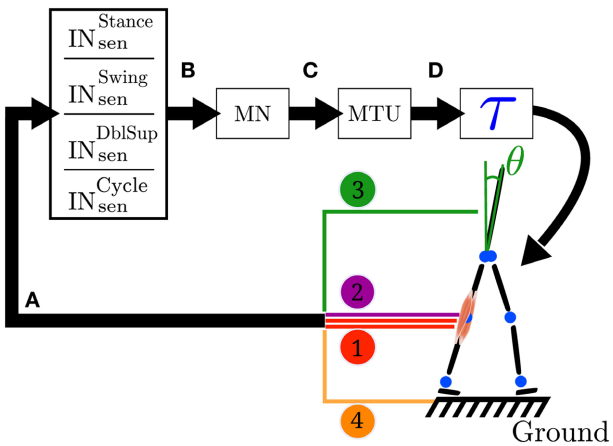


FIGURE 1 | Closed loop information flow of the FBL model. (A) Sensors signals stimulate (see Equation 1) a set of sensory interneurons (IN_{sen}). The sensors signals are represented by the colored line; 1 represents the muscle sensors, 2 represents the joint overextension/flexion prevention sensors, 3 represents the stability sensor generating a signal to maintain the trunk upright and 4 represents the ground sensors. There are four different types of sensory interneurons: IN_{sen}^{stance} which are active only during stance, IN_{sen}^{swing} only during swing, IN_{sen}^{dblsup}, only during the double support phase and IN_{sen}^{cycle} during the whole cycle. **(B)** Each IN_{sen} is connected to a unique motoneuron (MN). However a given MN receives inputs from several IN_{sen}. Connections between IN_{sen} and MN follow Equation 2. **(C)** In turn, each MN stimulates its corresponding muscle tendon unit (MTU). **(D)** Each MTU contributes to a torque (τ) on one or two joints, depending on whether it models a uni- or bi-articular muscle. Finally, the action of all the muscles on the body generates a movement, which induces a change in the sensors state and thereby closes the loop. Note that in the original model the link between sensors states and muscles activities is direct (i.e., no intermediary stage), while here the sensors to muscles mapping is separated in three more biologically relevant stages: sensory interneurons (IN_{sen}), motoneurons (MN) and muscle tendon units (MTU). Note that both the original and the FBL model are computationally equivalent.

While in the original model the link between sensors states and muscles activities was direct (i.e., no intermediate stage), in our work we separate the sensors to muscles mapping in three more biologically relevant stages (see **Figure 1** for details): sensory interneurons (IN_{sen}), motoneurons (MN) and muscle tendon units (MTU). The intermediate stages are added in order to prepare the extension of the model and makes no functional differences with the original model, as long as the overall delay between sensors and muscle activities is identical in both models. Stages A to C are implemented using the connection model defined in section 2.2.4. The sensors to torque mapping noted A to D (schematically represented in **Figure 1**) are presented below (see Supplementary Table 3 for a description of the different vector/matrices used):

A) Sensors to Interneurons
The activity of all interneurons can be written, in matrix form as:

$$X_{\text{in}_{\text{sen}}} = min\left\{1, max\left\{0, W\tilde{X}_{\text{sen}}\right\}\right\}^T \quad (1)$$

Where $X_{\text{in}_{\text{sen}}}$ is a vector of sensory interneurons activities, $\tilde{X}_{\text{sen}}$ is a vector of delayed sensors activities. W is the connection weights matrix linking the sensors and the interneurons. **Table 1** gives the list of the sensory interneurons present in a given limb.

B) Interneurons to Motoneurons
Given limbs state $s = (S_{\text{left}}, S_{\text{right}})$ (with $S_{\text{left}}, S_{\text{right}} \in S = \{\text{ST, STend, SW}\}$, where ST, SW and STend stand for stance, swing and double support finishing stance respectively) the activity of all the motoneurons can be written, in matrix form as:

$$X_{\text{mn}} = G^s X_{\text{in}_{\text{sen}}} + X^0_{\text{mn}} \quad (2)$$

Where: X_{mn} is the vector of motoneurons activities acting on limb L, $X_{\text{in}_{\text{sen}}}$ is a vector of sensory interneurons activities, in this case we assume no delay between interneurons and motoneurons (i.e., $\tilde{X}_{\text{in}} = X_{\text{in}}$). X^0_{mn} is a vector of basal motoneurons activities. G^s is a boolean matrix representing the connection state from interneurons to motoneurons given a limb state s. It ensures that the interneurons act on the motoneurons only when needed (i.e., stance feedback loops are active only during stance, swing feedback loops only during swing). For example if the interneuron $i = 18$ is connected to a motoneuron $j = 3$ and active only during left swing then $G^s(3, 18) = 1$ if $s = (\text{SW}, \cdot)$. Given a limb state s, the state of the considered limb S_{limb}, where limb can be either left or right is defined as a function of the level of the vertical ground reaction forces GRF^y_{limb} and the state of the contralateral limb S_{contra}. When $GRF^y_{\text{limb}} < 0.1$, the limb is considered in swing ($S_{\text{limb}} = \text{SW}$). If $GRF^y_{\text{limb}} \geqslant 0.1$ and S_{contra} switches from SW to ST then the current limb is in finishing stance ($S = \text{STend}$) otherwise the limb is in stance ($S_{\text{limb}} = \text{ST}$).

C) Motoneurons to muscle activities
A motoneuron acts on only one MTU, consequently the equation linking motoneurons to the MTUs stimulation is simply given by:

$$X_{\text{mtu}} = \tilde{X}_{\text{mn}} \quad (3)$$

Where: X_{mtu} is a vector of MTUs stimulation and $\tilde{X}_{\text{mn}}$ is a vector of delayed motoneurons activities. The MTU stimulation is constrained to the $[0.01, 1]$ interval. The lower bound of 0.01 is there to model the muscle tone (i.e., a minimal level of tension always produced by the motoneurons inervating a muscle). Its purpose is to permit quicker recruitment of muscles by maintaining a minimal non-zero level of tension. The MTU activation level A constrained to the $[0, 1]$ interval is linked to the MTU stimulation level by a first order differential equation modeling the excitation-contraction coupling:

Table 1 | List of the FBL sensory interneurons.

Sensory interneurons				
Abbreviation	**Type**	**From**	**To**	**ACTIVE_DURING**
GAS←GAS MFF, ST	1b	GAS	GAS	Stance
GLU←GLU MFF, SW	1b	GLU	GLU	Swing
HAM←HAM MFF, SW	1b	HAM	HAM	Swing
SOL←SOL MFF, ST	1b	SOL	SOL	Stance
TA←SOL MFF, ST	1b	SOL	TA	Stance (–)
VAS←VAS MFF, ST	1b	VAS	VAS	Stance
TA←TA MLF CY	1a	TA	TA	Cycle
HF←HAM MLF SW	1a	HAM	HF	Swing (–)
HF←HF MLF SW	1a	HF	HF	Swing (–)
HF←GSIF ST	3,4	iFoot,Trunk	HF	Stance
HAM←GSIF ST	3,4	iFoot,Trunk	HAM	Stance
GLU←GSIF ST	3,4	iFoot,Trunk	GLU	Stance
VAS←GCF STend	4	cFoot	VAS	Stance end (–)
HF←TLF SW	3	Trunk	HF	Swing
VAS←KNEE OPF	2	KNEE	VAS	Angle off (–)

The first column gives the abreviation of the interneuron. The abreviation indicates from which sensor the interneuron receives input from and to which MN it sends its output and is constructed as follow: MN←INsen_TYPE, ACTIVE_DURING. MN represents the motoneuron onto which the interneuron acts. If not specified, the motoneuron onto which the interneuron acts is on the same side as the sensors side (i.e., ipsilateral). INsen_TYPE represents the interneuron type. There are six different sensory interneurons; MFF (MTU force feedback), MLF (MTU length feedback), GSIF (ground and stability ipsilateral feedback), GCF (ground contralateral feedback), OPF (overextension prevention feedback), TLF (trunk lean feedback). ACTIVE_DURING indicates when the feedback is active; ST: feedback is active during stance, STend: feedback is active during double support finishing stance, SW: feedback is active during swing, CY: feedback is active during the whole cycle, AO: the feedback is active only when the angle of the corresponding joint goes beyond a certain limit, this is used only for the knee joint where the limit is fixed and set to 170°. The second column gives the type of the interneuron, as described in section 2.2.3. The third and fourth columns indicate the start and target of each feedback pathway. The last column specifies in which part of the cycle the feedback is active, the (–) sign refers to a inhibitory effect.

$$\frac{dA}{dt} = \tau_A\left(X_{\text{mtu}} - A\right),\ \tau_A = 100[s^{-1}] \quad (4)$$

D) Muscle activities to joint torques
The overall torque τ_j acting on joint j is given by :

$$\tau_j = \sum_{m \epsilon j} \tau_{m,j} + \tau_j^{lig}$$

Where τ_j^{lig} is the torque generated by the ligaments of joint j, $\tau_{m,j} = F_m \cdot r_m(\phi_j)$ is the torque generated by a MTU m on joint j, F_m is its force and r_m is the moment arm between MTU m and joint j (constant r_0 for hip joints and $r_0 cos(\phi - \phi_{\max})$ for knee and ankle joints, the r_0 and $\phi_{\max}$ values associated to each muscle-joint couples are given in **Table 2**).

2.2. FBL COMPONENTS

2.2.1. Ligament model

In animals, a ligament forms the joint that maintains two bones together. It also ensures that the angle formed by the bones stays within a given range. Its action is against the movement and engages only when the angle is beyond a certain limit, which depends on the joints (see Supplementary Table 5). Ligaments are modeled as non-linear spring damper acting as soft limit on the joints (Geyer and Herr, 2010). When the angle goes beyond the limit of the joint and the angular speed is not big enough to bring back the joint in its normal range a force is generated. The resulting torque τ_j^{lig} acting on joint j is modeled as :

$$\tau^{lig} = \begin{cases} k \cdot \Delta\phi \cdot \left(1 - \omega/\omega_{ref}\right) & \text{if } \Delta\phi > 0, \omega/\omega_{ref} > -1 \\ 0 & \text{else} \end{cases} \quad (5)$$

Where $k = 17.19[Nm/rad]$ is the spring damper stiffness, $\omega_{ref} = 1.74 \cdot 10^{-2}[rad/s]$ is the reference angular speed, used to normalize the joint angular speed, $\Delta\phi$ is the angle by which the joint limit is exceeded (i.e., difference between the actual angle and the limit angle, the axes are chosen so that $\Delta\phi > 0$ when the joint limit is passed) and $\omega[rad^{-1}]$ is the angular speed (the axes of rotation are chosen so that $\omega > 0$ when the angle is going toward the joint limit angle).

Note that this model of non-linear spring damper is also used in the model of H. Geyer to model the ground reaction forces to foot contacts. Here the contact of the robot with the ground are managed by the physical simulator of Webots.

Table 2 | List of the seven different muscles used in the FBL and derived models: GLU for gluteus, HF for hip flexor, VAS for vasilus, GAS for gastrocnemius, TA for tibialis, HAM for hamstring and SOL for soleus.

MTUs list and joints related parameters				
	Action	r_0[m]	ϕ_{max}[deg]	ϕ_{ref}[deg]
GLU	hip ext.	0.1	–	150
HF	hip flex.	0.1	–	180
VAS	knee ext.	0.06	165	125
SOL	ankle ext.	0.05	110	80
TA	ankle flex.	0.04	80	110
HAM	hip ext. knee flex.	0.08	–, 180	155, 180
GAS	ankle ext. knee flex.	0.05	110, 140	80, 165

The last two rows (HAM and GAS muscles) corresponds to bi-articular muscles (i.e., they span two joints), other rows are for uni-articular muscles. The second column shows the resulting action on the joint(s) onto which the muscle acts. The third column corresponds to the lever arm used for torque calculation. The fourth column gives the angle at which the action of the muscle on the joint is maximum (absent for the hip joint). The last column gives the reference angle of the muscle (i.e., the angle that corresponds to the muscle rest length).

2.2.2. Muscle model

The muscle model is based on the Hill model (Hill, 1938) and was developed by Geyer et al. (2003). A muscle is modeled together with its respective tendon (called muscle tendon unit, or MTU). An active, contractile element (CE) with two passive parallel elements (buffer elasticity BE and parallel elasticity PE) form the muscle, see Supplementary Figure 4. The active element represents the muscle active contractile element, while the two passive elements model the physical properties of the muscle fibers. The BE element prevents the muscle from collapsing, while the PE prevents the muscle length from going beyond a certain length. The tendon is modeled as a passive element in series with the muscle, called series elasticity (SE). The full mathematical formulation can be found in Geyer et al. (2003). The signal sent to the muscle by the motoneuron is related to the activity of the muscle with a first order differential equation accounting for neural delays, see section 2.2.4.

The force of a specific muscle j is linked to its activation level A_j by:

$$F_{\text{CE}} = F_{\max} \cdot f_l(l_{\text{CE}}) \cdot f_v(v_{\text{CE}}) \cdot A_j \quad (6)$$

Where : F_{CE} is the muscle force, $F_{\max}$ is the maximum force generated by the muscle, f_l and f_v respectively models the length-force and velocity-force relationship capturing main biological features of muscles, f_l and f_v equation can be found in Geyer et al. (2003). Given the muscle diagram depicted in **Figure 4** and applying Newton's third law of motion, we have that the net force generated by the muscle tendon unit (F_m) equals the force of the tendon F_{SE} :

$$F_m = F_{\text{SE}} = F_{\text{CE}} + F_{\text{PE}} - F_{\text{BE}} \quad (7)$$

The only unknown variables are the length and speed of the contractile element from which all muscle variables can be derived. Details on how v_{CE} is calculated can be found in Geyer et al. (2003). l_{CE} is then derived by integrating v_{CE}.

2.2.3. Sensors model

There are four different types of sensors (see **Figure 1**).

- Muscle sensors (type 1): there are two muscle sensors types. (1a) muscle length sensors, modeling the secondary muscle

spindles and (1b) muscle force sensors, modeling the Golgi tendons.

- Joint overextension/flexion prevention sensor (type 2): its intensity is proportional to the difference between the maximum tolerated angles and actual joint angle, and its direction is always against the movement. It is used to prevent knee joint overextension.
- Ground sensor (type 3): as in the original model (Geyer and Herr, 2010), there are two sensors under each foot that feel the reaction forces of the ground, located at the toe and heel position. In our case, the heel and toe sensors are provided by a Webots module called a TouchSensor that returns the cumulative force currently exerted on the sensor's body. Then, as in the original model, the value returned by the ground sensor is defined as being equal to the sum of the toe and heel sensors normalized by the total weight of the model.
- Stability sensor (type 4) measures the angle of the trunk in world coordinate and is used by stability feedback to bring the trunk toward a reference angle. These feedbacks are proportional-derivative control adapted to act on muscles and can be viewed as abstract models of descending pathways responsible for balance control originating from the cerebellum and the vestibular system.

2.2.4. Connection model

In the FBL, walking is generated by a sequence of time delayed reactions (or feedback loops) that connect sensory interneurons to muscles stimulation. The state of the output (y_j) is modeled as an affine transform of the sum of delayed weighted inputs ($\tilde{x}_i = x_i(t - T_{i,j})$):

$$y_j = f\left(W'\tilde{X}\right) = f\left(\sum_{i\,\varepsilon\,Input}\left(w_{j,i}\tilde{x}_{i,j}\right)\right)$$
$$= min\left\{1, max\left\{0, \sum_{i\,\varepsilon\,Input}\left(w_{j,i}\cdot x_i\left(t - T_{i,j}\right)\right) + x_j^0\right\}\right\} \quad (8)$$

Where the i-th index refers to input i and j-th index refers to the output j. Input-Output pairs are sensory neurons-sensory interneurons (stage A), sensory interneurons-motoneurons (stage B) and motoneurons to MTUs stimulation (stage C) shown on **Figure 1**. $\tilde{x}_{i,j}$ represent delayed input neuron activities meaning that a change in an input neuron will not affect the output neuron instantaneously but does so after a delay $T_{i,j}$ (modeling the fact that traveling speed of spikes depend on the properties of the nerve fiber). The delays are estimated assuming an average nerve fiber conductance of 80 m/s and estimated length between sensors and spinal cord. Note that the conductance of 80 m/s is the lower bound of extrafusal muscle fibers, golgi tendon organ and muscle spindle Ia conduction velocity (Siegel et al., 2006). We use three differents delays. A 2.5 ms delay to model the delay from hip muscles sensors and trunk stability sensors to their corresponding sensory interneuron and from the hip motoneurons to hip muscles. A 5 ms delay to model the delay from knee muscles sensors and knee joint angles sensors to their corresponding sensory interneurons and from the knee motoneurons to knee muscles and finally. A 10 ms delay for the ankle muscles sensors and ground sensors to their corresponding sensory interneuron and from the ankle motoneurons to ankle muscles. We assume no delay between sensory interneurons and motoneurons. $w_{j,i}$ is the connection weight from input x_i to output y_j and x_j^0 is the basal activity of the output (in vector format W is the vector of weights and $\tilde{X}$ is the vector of delayed input activity). The output is always constrained to the [0, 1] interval. For a neuron it can be viewed as its normalized firing frequency (1 meaning the neuron is firing at its maximum rate and 0 the neuron is not firing at all), for an MTU it can be viewed as a percentage of maximum muscle stimulation.

2.3. FBL SIMULATION ENVIRONMENT AND OPTIMIZATION

The model is implemented as described in Geyer et al. (2003) and Geyer and Herr (2010), i.e., 6° of freedom all constrained to the sagittal plane and 7 Hill type based muscles per limb. Simulations run with a time step of 1 ms. All differential equations are solved with a fourth order RungeKutta method, except for the muscle velocity which is integrated using the Euler method (as described in Geyer et al., 2003). In order to ensure convergence of the integration process, the integration time step of the muscle is reduced by a factor of 20 in comparison to the simulation time step.

Concerning the optimization, the open parameters of the system are the motoneurons basal activities (X^0_{mn} in Equation 2), the sensors parameters (trunk reference angle of the stability feedback, muscle length feedback offsets) and the feedback gains (non-zero values of matrix $W_{in,\mathrm{sen}}$ in Equation 1). The full model has 25 open parameters (the parameters and their associated ranges are given in Supplementary Table 1). In Geyer and Herr (2010), the parameters values were hand-tuned. When using those parameter values in our implementation, the produced gait shows a velocity of 1.1 [m/s]. The generated angles have a correlation with human data of 0.6, 0.7, and 0.9 for the HIP, KNEE, and ANKLE joint, respectively. The differences in produced gait between the original Geyer model and our implementation (for a given set of parameters) can be explained by the fact that we use a different simulation environment, bringing differences in the contact model and ground sensors. In almost all subsequent articles on FBL enhancement, optimization algorithms are used to set the parameters values. For example, in Song and Geyer (2012), the parameters were optimized to generate gaits of different speeds. The parameters were then analyzed in order to study the possibility to generate a speed controller through the direct modulation of reflex gains. The objective function used took into account the difference between target velocity and current velocity, a penalty term accounting for knee overextension and an energy expenditure term based on Bhargava et al. (2004).

In this article we also use optimization to instantiate parameters values of the FBL model. Since at least two criteria are always used (i.e., the minimization of energy and the penalty term accounting for knee overextension, and more as soon as one wants to optimize for an extra parameter, such as speed or step length), a good handling of multi-criteria evaluation is mandatory. We use a lexicographic ordering extension on top of the PSO

(Particle Swarm Optimization Kennedy and Eberhart, 1995) algorithm to handle multi-objectives fitness functions. Lexicographic ordering can be used only if the objectives can be written as constraints and ensures that the multi-objective optimization remains on the Pareto Front (Czyżżak and Jaszkiewicz, 1998; Li et al., 2008). Instead of using a unique multi-objective function (the usual average weighted sum or product of the multiple objectives can become difficult, due to the interaction between the different objectives), the different objectives are decoupled in single objective functions, that are sequentially optimized in corresponding stages. All except the last stage are constraint optimization. Each solution is evaluated according to one single objective function, following a sequential order. The solution is evaluated using the objective function of a given stage until the constraint of that stage is fulfilled. Therefore, each evaluated solution is defined by a tuple (s, v), where s is the stage reached and v is the fitness value obtained using the objective function of this stage. The solutions are then ranked according to their stages s and, within a stage, according to the value of the associated objective function v. In other words, assuming maximization, the following conditions hold:

- The stage are ordered so that a solution in a higher stage is always considered fitter.
- A solution can be in only one stage.
- Solutions in the same stage s_j are ordered using the fitness function f_j associated to that stage
- A solution is in stage s_i with $i > 0$, if all the constraints associated to stage $j < i$ are fulfilled but not the one of stage i.

Here we used 4 stages whose associated fitness functions and continuation criterion are given in Supplementary Table 4. The first stage optimizes for a walking gait that can cover at least a distance of d_{lim}. Since the model can generate gaits of various speed, we add a second stage to constrain the speed of the walking solution so as to facilitate further comparison between different obtained solutions. The third stage minimizes a penalty term accounting for knee overextension to favor human-like gaits. The fourth stage minimizes the metabolic energy expenditure. The model used for calculating the energy expenditure is based on a model of the energy consumption of a muscle as described in Bhargava et al. (2004) and as used in Wang et al. (2012).

Since we want to add a feedforward component to modulate the gait, the initial model should have the capacity to manage changes in acceleration, deceleration or step lengths, i.e., should be robust. However, optimizing for energy consumption on a flat ground will not favor the emergence of such gaits. In order to circumvent this issue and favor robust solutions, we optimize the feedback parameters on an environment with increasing and decreasing slope. The increasing/decreasing slope are modeled as simple trapezoidal structure (with max slope 5%). Furthermore, the length, slope and distance between trapezoidal structure are randomized (details concerning the environment can be found in Dzeladini, 2013). During the optimization process, each solution is evaluated on 5 different randomly generated environments, and only the worst fitness score is considered.

2.4. FBL EXTENSION: 3FBL

The extended model is a hybrid feedback and feedforward model, referred to as 3FBL. The CPG component (IN_{cpg}) generation is based on an idea from Kuo (2002), where feedforward signals produced by the CPGs are considered as feedback predictors. A direct way of combining such CPGs with feedbacks is to use a proportional term to control the relative importance of the CPG vs. the feedback it predicts, i.e., given the vector of CPG activities $X_{\text{in}_{\text{cpg}}}$, Equation 2 representing the motoneurons states becomes:

$$X_{\text{mn}} = G^s\left(\vec{\alpha} X_{\text{in}_{\text{sen}}} + (1 - \vec{\alpha}) X_{\text{in}_{\text{cpg}}}\right) + X^0_{\text{mn}} \tag{9}$$

Where: G^s, X_{mn}, X^0_{mn} and $X_{\text{in}_{\text{sen}}}$ are the same as in Equation 2. $X_{\text{in}_{\text{cpg}}}$ is the vector of feedforward interneurons activities. Note that here $X_{\text{in}_{\text{cpg}}}$ and $X_{\text{in}_{\text{sen}}}$ have the same dimension but all the components of $X_{\text{in}_{\text{cpg}}}$ referring to non-modeled sensory interneurons are set to 0. In the 3FBL models only the sensory interneurons related to muscles sensors are modeled with CPGs. Thereby, limiting the effective number of CPGs to 9 per limb. $\vec{\alpha}$ is a vector controlling the relative importance of sensory vs. CPG interneurons: a value of 0 in any of the α_i components will make the corresponding pathway exclusively feedforward-driven, whereas a value of 1 would make it solely feedback-driven (see **Figure 2**). Thus, when $\vec{\alpha} = \mathbf{1}$, the 3FBL becomes the FBL model. Conversely, when $\vec{\alpha} = \mathbf{0}$, the activity of all the sensory interneurons is ignored and the model becomes a purely feedforward-driven model.

Any IN_{cpg} is by definition a model of the underlying feedback pathway IN_{sen}. In this work we use two different abstract models of biological CPGs: a dynamical model $\text{IN}^{\text{osc}}_{\text{cpg}}$, generating periodic time varying signal and a constant model $\text{IN}^{\text{cst}}_{\text{cpg}}$, generating a constant signal (see section 2.5 for details). Both $\text{IN}^{\text{osc}}_{\text{cpg}}$ and $\text{IN}^{\text{cst}}_{\text{cpg}}$ can be viewed as a linear model of the underlying IN_{sen}. The

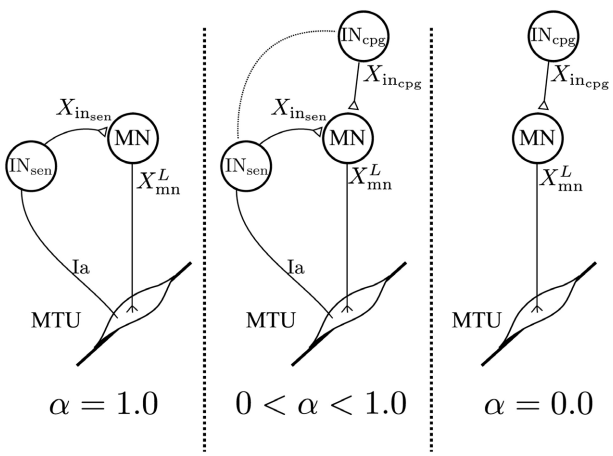


FIGURE 2 | Schematic representation of the spinal network for a specific feedback pathway. The value of α controls the proportion of feedback vs. feedforward. With $\alpha = 1.0$ the feedback pathway is solely feedback-driven. With $\alpha = 0.0$ the feedback pathway becomes a feedforward pathway. All values in-between create a feedback/feedforward pathway.

former is a model capturing the shape, timing and average activity while the latter only captures the average activity. Therefore, their combination with IN_{sen} can be viewed as a linearization of the underlying feedback pathways. Indeed, Equation 9 can be rewritten as:

$$X_{mn} = G^s\left(X_{in_{cpg}} + \vec{\alpha}\left(X_{in_{sen}} - X_{in_{cpg}}\right)\right) + X^0_{mn} \qquad (10)$$

This representation highlights the fact that, in the 3FBL model, the equation governing the activity of the motoneurons can be viewed as a linear feedforward term, plus a corrective term (i.e., the difference between the IN_{sen} and IN_{cpg} state). As expected, the effect of a IN^{osc}_{cpg}-IN_{sen} combination is different from the one of a IN^{cst}_{cpg}-IN_{sen} combination. On the one hand, increasing the proportion of IN^{cst}_{cpg} can be viewed as reducing the amplitude of the underlying IN_{sen}, without affecting its mean activity. In other words, the proportion of IN^{cst}_{cpg} vs. IN_{sen} controls the flatness of the IN_{sen}. On the other hand, combination of IN_{cpg} and IN_{sen} will neither significantly affect the shape, nor the average activity of the IN_{sen}, but will affect the timing.

2.5. 3FBL COMPONENTS

2.5.1. CPG-Constant model

In order to test whether a very simple model of feedback could already capture enough information to permit modulation, we decided to implement a CPG-Constant model, denoted IN^{cst}_{cpg}. IN^{cst}_{cpg} state, is a constant signal, whose value equals the average underlying IN_{sen} state. The average is calculated only on the part of the cycle where the feedback is active (e.g., for feedback active only during the stance, the average is calculated only during stance). This type of feedforward signal captures the average activity of the underlying feedback pathway. When combined with feedbacks (see section 2.4), the net effect is a flattening of the original feedback signal.

2.5.2. CPG-Oscillator model

In the oscillatory model, denoted IN^{osc}_{cpg}, each feedback predictor is modeled as a dynamical system reproducing the average shape and amplitude of the original feedback signal. In other words, CPGs can be viewed as a dynamical approximation of the sensory interneurons states $X_{in_{sen}}$ (see Equation 1). The dynamical system used for this purpose is a morphed oscillator (MO) (Ajallooeian et al., 2013). This oscillator is able to produce any shape, as long as this shape can be represented by a function that is both 1-periodic and derivable. The differential equation governing the oscillator is the following:

$$\dot{\theta} = \omega \qquad (11)$$

$$\dot{x} = \gamma\left(g(\theta) - x\right) + \frac{dg}{d\theta}\cdot\dot{\theta} + K \qquad (12)$$

Where $\dot{\theta}$ is the frequency of the oscillator, γ (here set to 100) controls the speed of convergence of the oscillator output x toward the shaping function $g(\theta)$, and $g(\theta)$ is the nominal function that shapes the output of the oscillator, this function is extracted from IN_{sen} states, see next paragraph.

2.5.2.1. Pattern generation. In order for the stability condition of the MO to be fulfilled, the pattern of the CPG must be represented by a first order differentiable 1-periodic function. Based on our hypothesis that CPGs can be viewed as feedback predictors, this function should reproduce the typical shape of the corresponding feedback pathway, for each cycle. The typical shape is derived as follow: (1) the sensory signals are recorded from a stable walking solution, (2) each sensory signal is split into cycles using the ipsilateral limb takeoff event (for feedback pathways active during swing), or the ipsilateral limb touchdown event (for all other feedback pathways), (3) each resulting sub signal is normalized in the temporal domain, in order to obtain a set of N repetitions of the sensory signal shape $p(\theta, i)$, $i = [1, \ldots, N]$, (4) the shaping function $g(\theta)$ is then derived using a third order spline interpolation of the mean signal.

$$g[\theta] = 1/N \sum_{i=1}^{N} p[\theta, i] \qquad (13)$$

2.5.2.2. CPG coupling with the environment. All oscillators have the same frequency ω initially set to an estimate of the FBL gait frequency from which the feedback patterns were extracted. In order to ensure that CPGs stay synchronized with the gait phases on which they should act, a coupling has to be defined. This coupling should ensure that:

1. IN_{cpg} will always start at the beginning of the gait phases during which it acts, at the touchdown/takeoff events of left limb for IN_{cpg} acting during left stance/left swing respectively, same holds for right limb. This event is called the synchronization event.
2. IN_{cpg} will never starts a new period before the gait phases on which it acts ends.

Consequently there should be four different oscillators driving the different IN_{cpg}, i.e., two for each limbs: one that uses touchdown as synchronization event (used by IN_{cpg} acting during stance or whole cycle) and an other one that uses takeoff as synchronization event (used by IN_{cpg} acting during swing), **Figure 2B** shows the organization of the spinal network. Each oscillator is coupled to the environment using the following frequency adaptation mechanisms implementing the two requested coupling properties:

1. If the oscillator is too slow compared to the walking frequency, the phase of the central clock is simply restarted and set to 0.0 at the synchronization event (see Supplementary Figure 2A).
2. If the oscillator is going too fast compared to the walking frequency, a slowing down mechanism takes action before the expected synchronization event (see Supplementary Figure 2B). It ensures that signals generated by the MOs will not start a new cycle before they should (e.g., for oscillators active during stance, before the limb touches the ground).

With both mechanisms turned on, the phase of oscillator i is defined as:

$$\dot{\theta}_i = \begin{cases} \omega & \text{if } t_i < p \cdot \frac{1}{w} \\ c(t_i) & \text{else} \end{cases} \quad (14)$$

$$\theta_i = 0 \quad \text{if } t_i > \frac{1}{\omega} \quad (15)$$

Where: θ_i is the phase of oscillator i, t_i is the time since the last synchronization event and p is the percentage of the phase at which the slowing down mechanism is turned on. $c(t)$ is a slowing down function that ensures that $\theta \leqslant 1.0, \forall\, t \in \mathbb{R}$ For the slowing down mechanism to enter in action after 90% of the period of the oscillator (i.e., $p = 0.9$), we can use the following function:

$$c(t_i) = 10\omega \cdot exp(-10\omega t_i - ln(10) + 9)$$

Details on how $c(t)$ is derived can be found in Dzeladini (2013).

2.5.3. Feedback sensitivity scale

For a feedback pathway i, the feedback sensitivity is noted $FDB_i^{sen} = 1 - \alpha_i$ and corresponds to the point at which the gait becomes unstable when (1) all other feedback pathways are kept as feedbacks (i.e., $\alpha_j = 1$ for all $j \neq i$) and (2) the feedback pathway i is combined with an IN_{cpg}^{osc}. A feedback sensitivity of 0 means that the feedback can be fully replaced by its cognate IN_{cpg}^{osc} predictor without destabilizing the stability of the generated gait.

2.6. 3FBL MODELS

In order to demonstrate the effect of feedback and CPG combinations, we created different models combining CPG and feedback components in different ways. Here we present only the 5 models exhibiting the most interesting properties in terms of speed modulation. The 5 models differ in their CPG-feedback combination vectors $\vec{\alpha}$ (see **Table 3** for details). Contrary to what might be expected, a 3FBL model with a IN_{cpg}^{osc}-IN_{sen} combination vectors of 0.5 for all muscle feedbacks pathways was not good in terms of speed modulation when considering global control variable acting on all CPGs. The first 4 models study the effect of a CPG addition on different group of muscles, namely the $3FBL_{ankle}^{osc}$, $3FBL_{hipA}^{osc}$, $3FBL_{hipB}^{osc}$, and $3FBL_{biArt}^{osc}$. The fifth model, referred to as $3FBL_{fdb}^{min}$, is a minimum feedback gait, designed to study the properties of gait with minimal feedback activity. That model was obtained as follows: IN_{cpg} are added starting from pathways acting on distal muscles. Pathways acting on distal muscles use CPG-CST models (IN_{cpg}^{cst}) and pathways acting on proximal muscles use CPG-OSC models (IN_{cpg}), using the lowest possible α (in the [0, 1] range). This methodology was chosen, with the aim of finding a gait with the minimal number of feedbacks. Note that other CPG-FDB combinations might be found using different methodologies. Using this methodology, the $3FBL_{fdb}^{min}$ gait generated stable walking, with a feedback activity corresponding to 35% of the IN_{sen} related to muscle feedbacks, and 45% of all the feedbacks (the feedback activity is defined as $\frac{\sum_i(\alpha_i)}{N}$, where N is the number of feedbacks).

2.7. 3FBL MODULATION : MODEL OF SUPRASPINAL INFLUENCES

We hypothesize that the use of a CPG component will facilitate speed control. Indeed, it is known that simple supraspinal signals are sufficient to modulate gait frequency in lower vertebrates and in mammals, as demonstrated by experiments on decerebrated cat walking on a treadmill, where speed changes and gait transitions can be elicited by

Table 3 | Description of the CPG-FDB combination map for the 5 different 3FBL models.

		$3FBL_{ankle}^{osc}$		$3FBL_{biArt}^{osc}$		$3FBL_{hipA}^{osc}$		$3FBL_{hipB}^{osc}$		$3FBL_{fdb}^{min}$	
		type	**1 − α**	**type**	**1 − α**	**type**	**1 − α**	**type**	**1 − α**	**type**	**1 − α**
ANKLE	SOL←SOL MFF, ST	Osc	0.5								
	TA←SOL MFF, ST									Cst	0.9
	TA←TA MLF CY									Cst	0.9
KNEE	GAS←GAS MFF, ST	Osc	O.5							Cst	0.9
	VAS←VAS MFF, ST										
	HAM←HAM MFF, SW			Osc	0.5	Osc	0.5	Osc	0.5	Osc	1.0
HIP	HF←HF MLF SW					Osc	0.5	Osc	0.5	Osc	1.0
	GLU←GLU MFF, SW					Osc	0.5			Osc	1.0
	HF←HAM MLF SW					Osc	0.5			Osc	0.0

Each row shows for a given feedback pathway, the type of CPG used (Osc stands for IN_{cpg}^{osc} and Cst for IN_{cpg}^{cst}) and the level of CPG-FDB (i.e., α) for the 5 different 3FBL models. The four first columns shows the most interesting 3FBL models in terms of speed control: $3FBL_{ankle}^{osc}$, with CPGs acting on distal extensor muscles, $3FBL_{hipA}^{osc}$ and $3FBL_{hipB}^{osc}$, with CPGs acting on HIP muscles and $3FBL_{biart}^{osc}$, with CPGs acting on the HAM bi-articular muscles. The last column shows the combination vector for $3FBL_{fdb}^{min}$ (i.e., the minimum feedback model). Note that the $3FBL_{fdb}^{min}$ also replaces the "VAS←GCF STend" and "HF←TLF SW" pathways by a CPG-CST predictor. Note that only the pathways related to muscle feedbacks are shown. Even though a full replacement of the "VAS←GCF STend" pathway by CPG-CST is possible without affecting the produced gait, the effect of a modulation produces no significant effect on the resulting gait (data not shown). This pathway is thus not used, except for the $3FBL_{fdb}^{min}$. The KNEE overextension prevention pathway ("VAS←KNEE OPF") and the pathways related to stability (i.e., "HF←GSIF ST," "HAM←GSIF ST," "GLU←GSIF ST" and "HF←TLF SW") are not used, as their role as feedback is evident. Moreover, even though a combination with CPG generates stable walking, walking becomes unstable even with very small modulation of the CPG parameters (data not shown).

varying the stimulation of the mesencephalic locomotor region. We model two different kinds of descending pathways (see **Figure 3**):

- Frequency : ω
  Controls the frequency of the CPG-OSC (ω value in Equation 14). This variable affects all oscillators as they share the same frequency.
- Activity modulation : μ
  Modulates the CPG activity of both CPG-OSC and CPG-CST. Effectively, the CPG output $X_{\text{in}_{\text{cpg}}}$ becomes $\mu \cdot X_{\text{in}_{\text{cpg}}}$, with $\mu > 0$ controlling the activity of the CPG.

3. RESULTS

The results are separated in three parts. In the first part, we compare the gait produced by the optimized FBL model with human

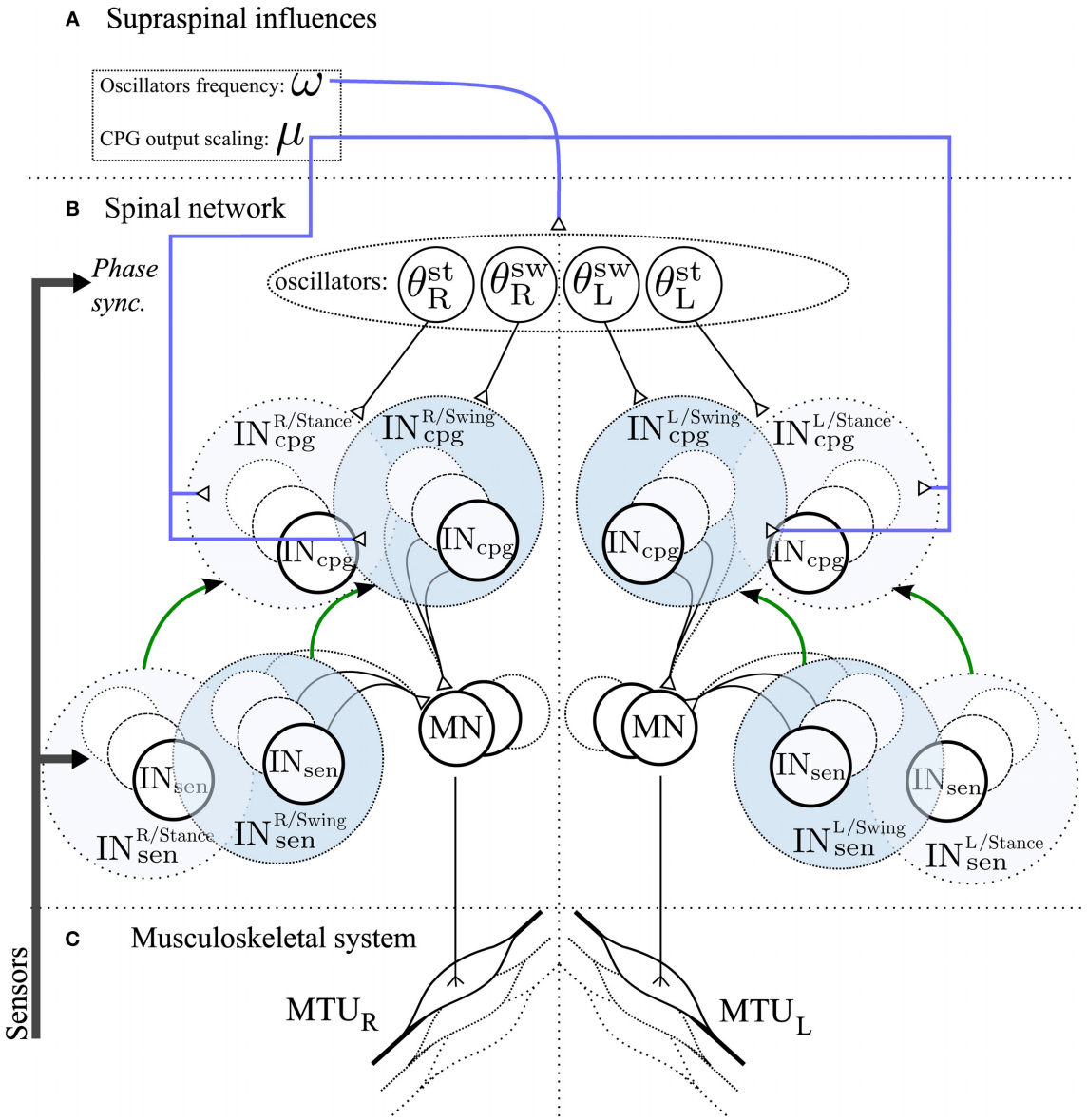


FIGURE 3 | Schematic representation of the spinal network and supraspinal control of the CPG network in the 3FBL model. The network is symmetric: left/right part of the figure corresponds to the part of the network acting on right/left limb muscles respectively. **(A)** Suprasinal influences: μ represents the activity modulation pathway and ω the frequency of the CPG network. All 4 oscillators share the same ω, but each CPG can have a different μ. If not stated otherwise, all $\text{IN}^{\text{osc}}_{\text{cpg}}$ and $\text{IN}^{\text{cst}}_{\text{cpg}}$ share the same amplitude modulation μ_{osc} and μ_{cst}, respectively. **(B)** Spinal network. Four oscillators, differing in their synchronization mechanism with the environment, drive the different IN_{cpg}. $\theta^{\text{st}}_{\text{R}}$,$\theta^{\text{sw}}_{\text{R}}$,$\theta^{\text{sw}}_{\text{L}}$ and $\theta^{\text{st}}_{\text{L}}$ are used by IN_{cpg} starting at right limb stance, right limb swing, left limb stance and left limb swing respectively. IN_{cpg} and IN_{sen} action on MN follows Equation 10. The green arrow between Sensory and CPG Interneurons pathway highlights the fact that each CPG pathway is a model of one sensory pathway. **(C)** Musculoskeletal system, there is one muscle corresponding to each individual motoneurons.

walking, in terms of metabolic cost, gait harmony and gait kinematics. In the second part, we present an analysis of the different feedback pathways of one specific solution of the FBL model. We analyse each feedback pathway separately and for each of them study the effect of a combination with their feedforward predictor. Finally, in the last part, we analyze the 3FBL models in terms of speed control.

3.1. FBL: FEEDBACKS BASED LOCOMOTION MODEL

In order to determine the ability of our optimization process to generate stable gait, we performed 10 runs of the same optimization process (as described in section 2.3) with different random initial condition. We observe that the optimization process always converges to a stable and symmetric walking solution, but to different solutions (local optima), hence leading to visually different gaits. **Figure 5F** gives a snapshot of the solution 1 during two cycles. Note that the presented results are, in terms of joint angles, joint torques and muscles activities, qualitatively similar to those presented in the paper describing the original model (Geyer and Herr, 2010).

3.1.1. Metabolic cost analysis

When comparing the cost of transport (CoT) between the 10 different solutions, we observed a value ranking from 2.2 to 3.5 $[\mathrm{Jm^{-1}kg^{-1}}]$ (CoT is defined as E/md, where E is the energy consumed during the run, m is the mass of the model, d is the traveled distance), see **Figure 4**. Five solutions show a CoT less than 25% higher than the net metabolic transport cost of $\sim$2.1 $[\mathrm{Jm^{-1}kg^{-1}}]$ found in human subjects of similar heights, weights and walking at the same speed (Weyand et al., 2010). This increase is comparable to the one found in Bhargava et al. (2004) and can be explained by the fact that, in our model, the upper body is modeled as a single rigid body, while the experimental values used for comparaison are for walking with arm swing. Indeed, it has been shown that, despite the fact that arm muscles consume energy to produce movement, they can still reduce the walking metabolic cost up to 12% (Collins et al., 2009). An other reason explaining the higher CoT could be the lack of feedbacks for stance preparation. Indeed, as most of the metabolic cost of walking comes from the stance phase, optimizing the properties of the limb joints before touchdown will affect the efficiency of walking, as shown in Donelan et al. (2002).

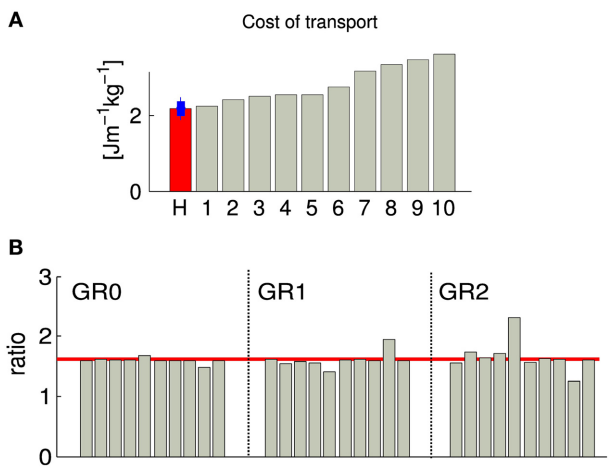


FIGURE 4 | Each gray bar corresponds to one solution of the same optimization process (optimizing for a stable gait walking at 1.3 m/s). (A) Normalized cost of transport. The red bar corresponds to the normalized cost of transport of human subject of the similar weight and walking speed as our obtained gait (data from Weyand et al., 2010), the blue bar shows the estimated standard deviation. **(B)** Duration proportion of the different gait phases. GR0 corresponds to the ratio between cycle duration and stance duration, GR1 corresponds to the ratio between stance duration and swing duration and GR2 corresponds to the ratio between swing duration and double stance support. The red line corresponds to the golden ratio $\phi = \frac{1+\sqrt{5}}{2}$. GR0, GR1, and GR2 are known to be statistically similar to the golden ratio in human walking at their preferred speed (Iosa et al., 2013).

3.1.2. Golden ratio analysis of gait harmony

As demonstrated in Iosa et al. (2013), the ratios between cycle/stance durations (noted GR0, commonly referred to as the duty factor), stance/swing durations (noted GR1), and swing/"double stance support" durations (noted GR2) is similar in healthy humans of different size, corpulence and age walking at preferred (self-chosen) speed, and satisfy the golden ratio ($\sigma = \frac{1+\sqrt{5}}{2}$). Note that the variability of GR1 is higher than GR0, and the variability of the GR2 is higher than GR1. We measured those three ratios in our 10 solutions, and observed that GR0 converges to σ in all cases, GR1 converges to values close to σ with higher variability and a bias to slightly smaller values, and GR2 is more variable, with a bias to values higher than σ. The bias observed in the cases of GR1 and GR2 indicates that there is a tendency to generate gaits with longer swing and shorter double stance support phases. This overestimation of the swing duration can be explained by the fact that our model does not have toes; the length of the foot being shorter, the legs tend to enter the swing phase earlier.

3.1.3. Gait analysis

We then compared the joint angles and torques trajectories of the 10 solutions, with human data (Winter, 2009). A correlation analysis revealed that all joints angles and torques are comparable to human data (see **Figures 5A,C**, if not stated otherwise, the solutions are ordered with increasing CoT). While the ANKLE torques show high correlation with humans, the HIP and KNEE torques correlations are substantially lower. This can be explained by the fact that, in our model, the HIP is completely fixed to the trunk. We thus do not model the characteristic pelvis movement observed in human walking. Regarding the joint angle correlations, we can see that the ANKLE angle correlation is not perfect. The low correlation can be explained by the differences in shape in late stance and early swing (see **Figure 5B**, right), which is due to the fact that the toe is not modeled. Indeed, the lack of toes will make the leg enters in swing earlier, thereby explaining both the reduced minimum angle and the earlier slope inversion (i.e., the swing/stance transition).

Another interesting difference between the model and human data can be noted at the ANKLE angle level during early stance. Indeed, while humans show an initial passive extension during early stance of about 1/10th of stance duration (black dotted

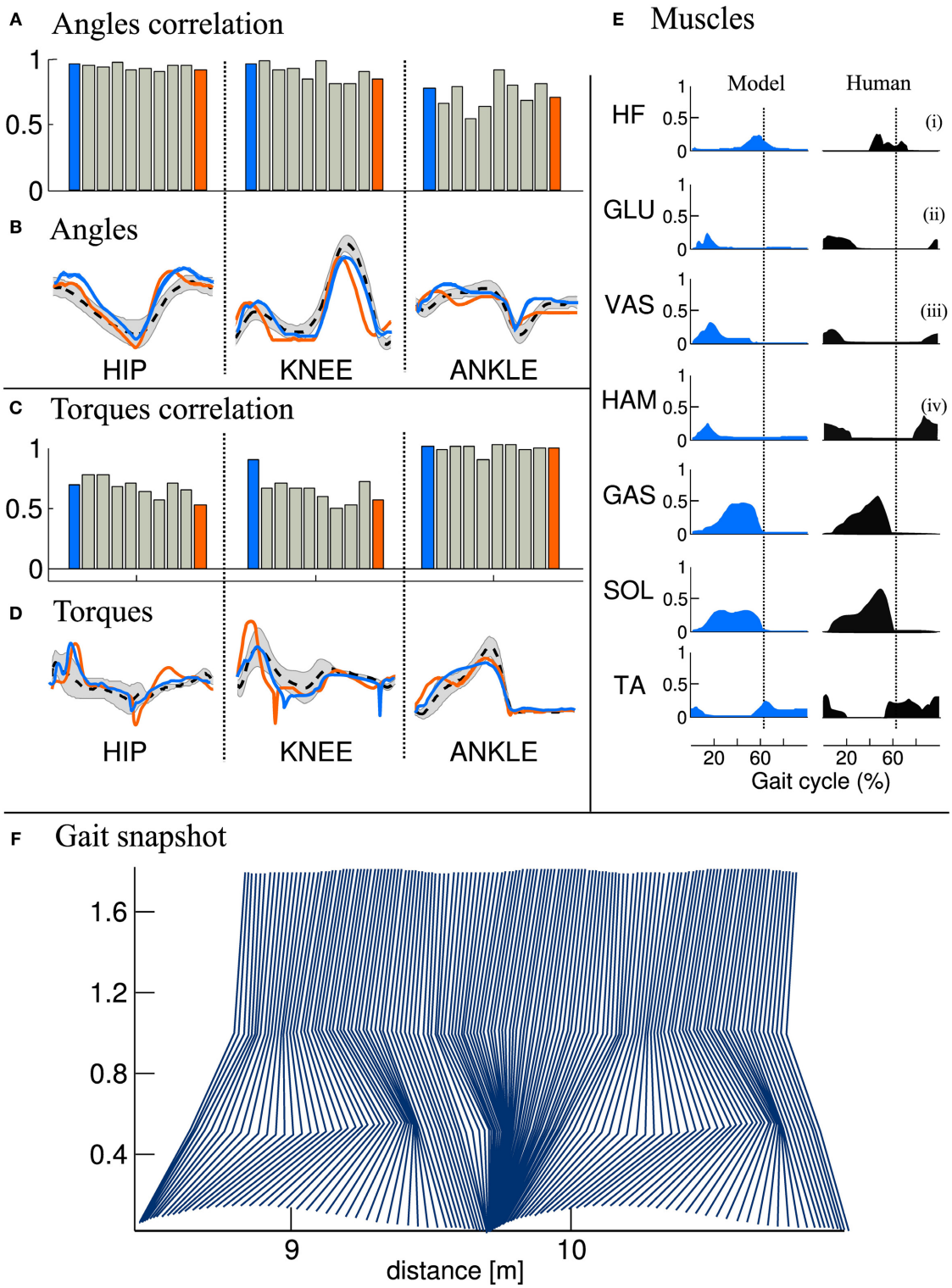


FIGURE 5 | Comparison of joints angle, joints torque and muscles activity extracted from the FBL models (10 optimization runs), with human data. Human joints angle and torque are taken from Winter (2009), muscles activities are adapted from Perry et al. (1992), as presented in Geyer and Herr (2010). **(A)** Joint angle correlation with human, **(B)** Average joint angle compared to human, **(C)** Joint torque correlation with human, **(D)** Average joint torque compared to human, **(E)** average muscles activity of solution 1 compared to human and **(F)** Gait snapshot of the solution 1 over two cycles. In **(A,C)**, the bar plots show the correlation with human for the different solutions and for the different joints. In **(B,D)** are shown typical human trajectories (black dotted line:mean, gray: standard deviation) and two

(Continued)

FIGURE 5 | Continued
mean trajectories from solution 1 and solution 10, blue and orange lines respectively. Each bar corresponds to one solution of the same optimization process (optimized for a stable gait walking at 1.3 m/s), the different solutions are ordered with increasing energy consumption (same as in **Figure 4**). The correlation were calculated on data extracted from 50 strides of steady state walking (sampling frequency of 1 Khz), spline interpolation was used to normalize the length of the vectors to 1000 points. The average of the normalized vector was then correlated with average human data. In **(E)** the subscripts show the compared muscles: (i) adductor longus, (ii) upper gluteus maximum, (iii) vastus lateralis, and (iv) semimembranosis. Note that the data was extracted from a model walking on a flat terrain without noise and external perturbations. Therefore, the standard deviation of the angles and torques trajectories and muscles activities is very small and thus not visible.

line in **Figure 5B** right), the model does not show this behavior. When looking carefully at the ANKLE angle pattern for solution 1 an initial passive extension is visible. However, this initial passive extension is very short and almost not visible in the figure (blue line in **Figure 5B** right, the ANKLE angle does not start at the same place due to a very fast and quick passive extension). The solution 10 (orange line in **Figure 5B**) does not show this behavior at all: the foot touches the ground horizontally. Several elements can explain this behavior, such as the lack of mechanism (e.g., feedback, CPG) for stance preparation, a shorter swing range (due to smaller HIP range or an under-extension of the knee) or the way the swing-stance transitions are designed, i.e., state machine with discrete transition.

When comparing muscles activities of solution 1 (see **Figure 5E**), we note that all the ANKLE muscles and HF muscle are close to human data. However, the GLU, VAS and HAM muscles do not show the typical activity observed during late swing in humans. This is in agreement with the conclusion drawn in the previous paragraph concerning the lack of a mechanism for stance preparation.

3.2. FEEDBACK PATHWAYS STUDY

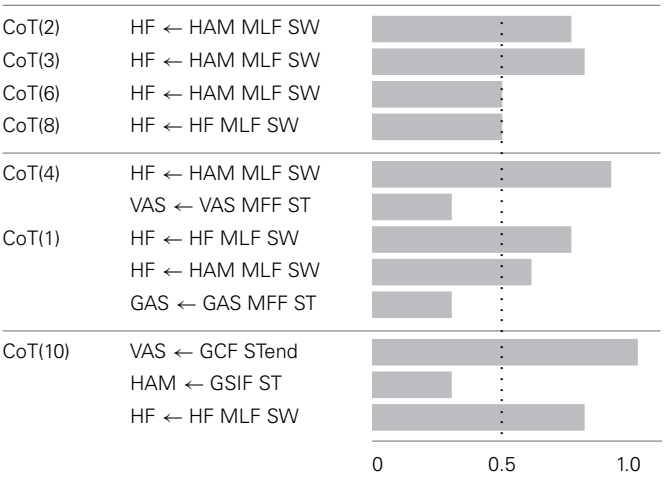
3.2.1. IN_{sen} signal analysis and prediction

Since the produced gaits are all symmetric and stable (i.e., close to perfectly periodic), the feedback signals should be very similar between cycles. Consequently, the quality of the feedback prediction should be very high (i.e., IN_{cpg}^{osc} should be very close to IN_{sen}). In order to study the quality of the prediction, we generated the IN_{cpg}^{osc} (as described in section 2.4) and ran them in a passive mode (no action on muscles, i.e., no link between IN_{cpg}^{osc} and MN). The Supplementary Figure 1 shows the actual IN_{sen} signals (dotted lines) and the reproduced signal (thick lines) over one step, for the worst gait (in terms of feedback prediction quality, i.e., similarity between IN_{sen} and IN_{cpg}^{osc}). We can see that the prediction is very close to the feedback signals; the lowest correlation between the original and the reproduced signals is of 0.98. Differences are noted as shifts and amplitude differences, and are due to small asymmetries in the gait. It is interesting to note that, even if those asymmetries are visible at the level of the feedbacks, their effects on the gait are very small. However, even small asymmetries between the IN_{sen} and their predictors (IN_{cpg}^{osc}) can create instabilities which makes their replacement difficult.

3.2.2. Feedbacks replacement

In order to study the possibility of replacing the feedbacks (IN_{sen}) by their full predictors (IN_{cpg}^{osc}), we ran a systematic search in which we increase $\beta = 1 - \alpha$ (i.e., the proportion of IN_{cpg}^{osc}) from 0 to 1.0 using the combination strategy presented in section 2.3.3. The systematic search is done for each feedback pathway i, where β_i is increased from 0 to 1 in steps of 0.1. All the others pathways are kept as feedbacks (i.e., $\beta_j = 0, j \neq i$).

Table 4 | Feedback sensitivity (see section 2.5.3) for the best 7 solutions (in terms of IN_{sen} replacement capacity, i.e., percentage of IN_{sen} that can not be replaced by a CPG-OSC model).



The first column shows the solution, ranked in term of cost of transport (CoT). The second column gives the name of the feedback pathway. The third column shows the feedback sensitivity (FDB^{sen}).

The Supplementary Table 2 shows, for each gait, the number of feedback pathways that could not be fully replaced (i.e., the feedback pathways that have a $FDB_i^{sen} \neq 0$. **Table 4** shows the feedback sensitivity of the 7 best gaits, in terms of the number of feedback pathways that can be replaced, i.e., in terms of feedback replacement capacity (see section 2.5.3 for details on the feedback sensitivity scale). It is interesting to note that feedback pathways acting on ANKLE muscles have a zero feedback sensitivity value which means that they can be fully replaced by a IN_{cpg}^{osc} model without loss of stability. The muscle length feedback pathway from HAM bi-articular muscle acting on the HF muscle always shows a high sensitivity (for gaits showing meaningful CoT), highlighting its importance for the stability of the gait. Even though feedback related to trunk stability (feedback type 4) are crucial to ensure stable walking and to enhance gait resistance to perturbations, they are not part of sensitive feedbacks. However, a gait with only one trunk stability feedback replaced is stable only in steady state walking; as soon as small perturbations (pushes and/or change in slope) are exerted on the model, the gait becomes unstable and falls.

Based on these results, we focus on gait 2, as it shows a good correlation with human data, a meaningful CoT and a low feedback sensitivity, for further analysis.

3.2.3. Feedbacks combination

Figure 6A shows the effect on the generated gait (in terms of CoT, stride length and speed) of an increase in the proportion of feedforward vs. feedback signal for one specific pathway while maintaining all the other pathways purely feedback driven (this was implemented by decreasing the feedback proportion by steps of 0.1 of one component of the $\vec{\alpha}$ vector at a time while keeping all other components at 1). **Figure 6A** Left and Right parts show the combination analysis of feedbacks with IN^{cst}_{cpg} and IN^{osc}_{cpg} respectively. As expected, the replacement of IN_{sen} by a constant model (i.e., IN^{cst}_{cpg}) has more effect on the gait characteristics, compared to the replacement of the IN_{sen} by an oscillatory model (i.e., IN^{osc}_{cpg}). This confirms that the latter captures more information from the IN_{sen} (i.e., the shape, timing and amplitude).

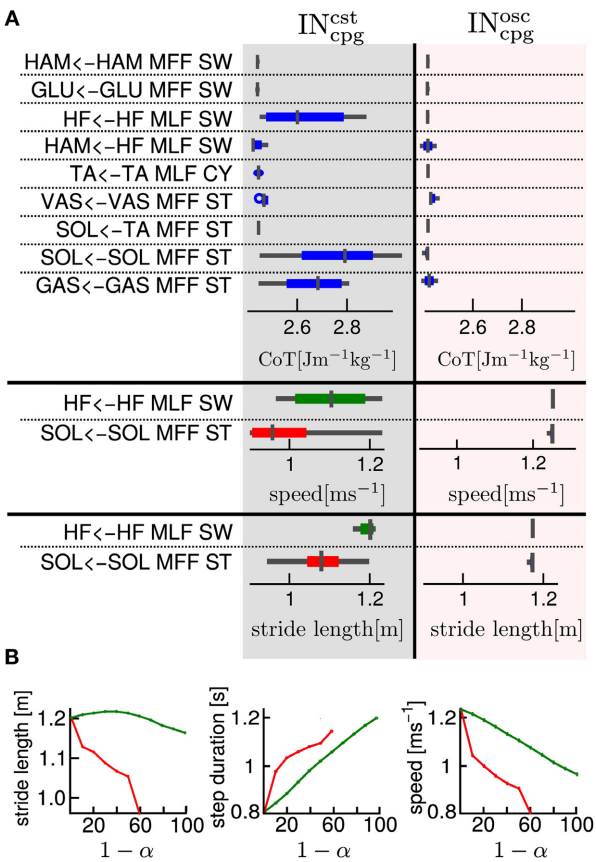


FIGURE 6 | (A) One by one feedback and feedforward combination effects on cost of transport, stride length and speed, for gait number 1. The first column gives the name of the feedback pathway considered. The second and third columns show for an IN_{sen}-IN^{cst}_{cpg} and an IN_{sen}-IN^{osc}_{cpg} respectively, a box plot of the variation of a measured variable when α varies from 1 to 0. In the first part of the table the considered variable is the cost of transport (CoT), in the second part, the speed and in the third part, the stride length. We show the speed and stride length box plot only for the two most interesting pathways in terms of feedback and feedforward combination effect on CoT. The box plot read as follow: the middle line is the median, the colored line represents 99% of the data assuming the data are normally distributed and the gray horizontal bar shows the range of the measured variable. A very thin box plot (no colored line visible) means that the variation of α had no effect on the considered variable, feedback pathway and IN_{cpg} model. As expected the IN_{sen}-IN_{cpg} combination for any α in the [0, 1] interval has very little effect on the CoT. **(B)** Relationship between IN^{cst}_{cpg} proportion and gait variables, for two selected feedbacks (red, "SOL←TA MFF, ST" and green, "GAS←GAS MFF, ST"). Left: relationship between stride length and $1-\alpha$ (i.e., the IN_{cpg} proportion), Middle: relationship between step duration and $1-\alpha$ and Right: relationship between speed and $1-\alpha$.

Despite the higher sensitivity of the IN^{cst}_{cpg}-IN_{sen} combination (i.e., percentage of IN_{sen} that could not be replaced by a constant model (IN^{cst}_{cpg}), several interesting effects of the IN^{cst}_{cpg}-IN_{sen} combination are noted, as shown in **Figure 6**. We observe that, for the "SOL←TA MFF, ST" and the "HF←HF MLF, SW" feedbacks, changes in α (i.e., proportion of IN^{cst}_{cpg} vs. IN_{sen}) produce large variations in speed and stride length. In the case of "SOL←TA MFF, ST", there is a linear relationship between the IN^{cst}_{cpg} proportion level and both the speed and the stride length. A decrease in stride length and speed is observed with the increase in IN^{cst}_{cpg} level, see **Figure 6B**.

3.3. 3FBL MODELS : FEEDFORWARD AND FEEDBACK BASED LOCOMOTION MODEL

In the previous section, we showed that all feedbacks can be combined with their CPG predictors, and that interesting properties, such as speed and step length variation, can be achieved, by playing with the CPG-FDB combination level when using CPG-CST predictors. While, in the previous section, feedback and CPG combinations were studied one pathway at the time, here we study effect of more complex combinations on 5 different 3FBL models exhibiting the most interesting property in terms of gait speed modulation (see section 2.6 for details).

3.3.1. $3FBL^{min}_{fdb}$: Minimal feedbacks gait

The $3FBL^{min}_{fdb}$ model is able to produce stable walking with a global feedback activity reduced from 100 to 45%. Its average speed on flat ground is 1.35[m/s] (3% increase compared to the underlying FBL model). When comparing the joint angles, torques and muscles activities between the two models, almost no differences can be observed at the HIP joint (see **Figure 7C**). However, differences are noted at the level of the ANKLE joint (see **Figure 7A**). Indeed, all muscles activities acting on the ANKLE joints show different muscle activation patterns than the corresponding FBL model. Interestingly, the differences observed in muscles activities do not produce important changes in the shape of the torque and angle patterns of the ANKLE joint. Nevertheless, the increase in extensor muscles activities produces a steeper increase in joint torque during stance. This increase in torques explains the observed increased ANKLE angle at takeoff. In turn, this increase in ANKLE angle also increases the duration of the stance phase, thereby explaining the observed shift of the KNEE pick angle in early swing (see **Figure 7B**).

The SOL muscle shows a different muscle activation pattern, while the "SOL←SOL MFF, ST" pathway, the only one acting on it, has not been replaced by a CPG (i.e., kept as pure feedback, $\alpha = 1$). Since the $3FBL^{min}_{fdb}$'s feedback / CPG combination map does not permit a combination of CPG-OSC with feedback for this specific pathway (even with $\alpha = 0.95$, i.e., pathway kept almost purely feedback), this change in activity is necessary to ensure a stable walking gait. This highlights

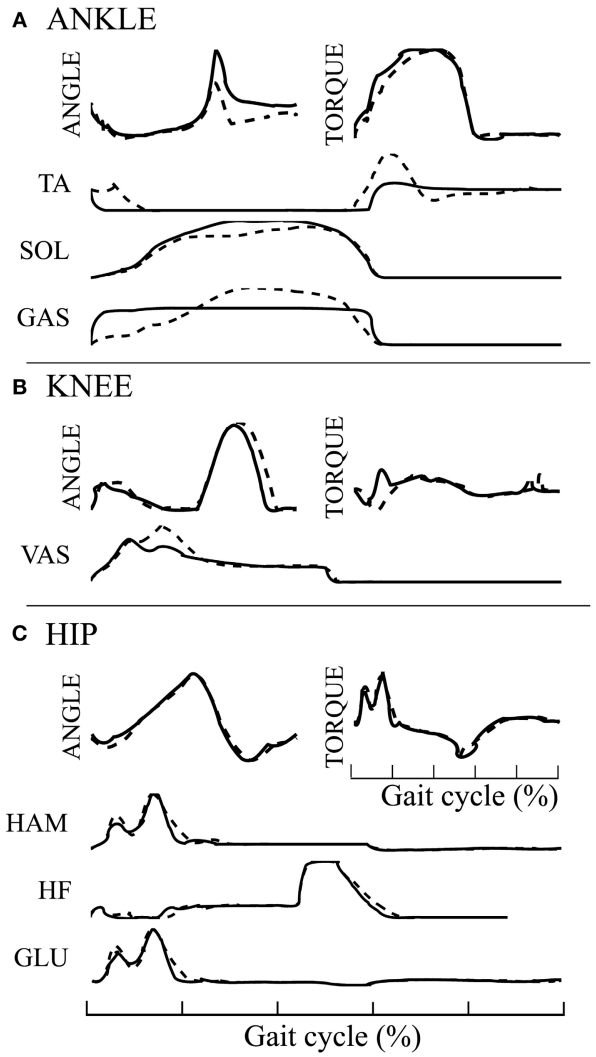


FIGURE 7 | Comparison of average joint angles, joint torques and muscles activation pattern between the $3FBL^{min}_{fdb}$ (black line) and the FBL models (dashed line) for solution 1. (A) ANKLE angle, torque and associated muscles activation level, **(B)** KNEE angle, torque and associated muscles activation level, and **(C)** HIP angle, torque and associated muscles activation level.

the important stabilizing role that muscle feedbacks play in locomotion.

It is important to note that, while in a stable walking regime reducing as much as possible the proportion of feedback signals for specific pathways does not significantly affect the generated gait, the replacement of feedbacks considerably reduce the gait robustness to perturbations. Indeed, recovery after 0.25[s] pushes is reduced from 40[N] to 28[N] compared to the original gait. This highlights the importance of feedback to adapt to perturbations.

Even though the $3FBL^{min}_{fdb}$ is valuable, as it shows that a large part of feedbacks can be removed from the FBL model, while a stable walking gait is still produced, it is not surprising that its modulation is almost impossible. Indeed, since a large part of the feedbacks are removed, even small modulations of CPG parameters render the gait unstable.

3.3.2. Systematic study of supraspinal signal modulation and their effects on gait

Using the model of supraspinal influences presented in section 2.7, we ran a systematic search on the effect of CPG amplitude and frequency modulation on the different 3FBL models presented in the previous section, using ω and μ_{osc} as parameters (the parameters are split into 11 values across a given range ([0.2, 2.5] for ω and [0.1, 4.0] for μ_{osc}).

The systematic search on the 4 chosen models acting on different group of muscles (see **Figure 8A**) indicates that all the models are stable in a large range of amplitudes and frequencies, except the $3FBL^{osc}_{hipA}$, that shows a more restricted region of stability. This can be due to the fact that the $3FBL^{osc}_{hipA}$ has more oscillators than the three other models. Note that the restricted region of stability does not imply a restricted range of speed. Indeed, small variations in ω (while μ_{osc} remains fixed) induces noticeable change in speed in this model; an increase in speed is observed with an increase in frequency. In other words, changing the frequency of the $3FBL^{osc}_{hipA}$ is sufficient to entrain the whole musculoskeletal system. Interestingly, this model—which is the only model with a high number of CPGs acting on proximal muscles—is the only one that shows an increase in speed when increasing the CPG network frequency. This suggest that CPGs acting on proximal muscles are required to produce a frequency-driven entrainment of the system.

Interestingly, the $3FBL^{osc}_{hipB}$—which has only two CPGs acting on proximal muscles, compared to four in the case of the $3FBL^{osc}_{hipA}$—shows almost no change in speed when the frequency ω is modulated (while μ_{osc} is fixed). Possibly, the frequency modulation of only two CPGs at the HIP level is not sufficient to produce a frequency-driven entrainment of the system. However, increasing μ_{osc} leads to a significant decrease in gait velocity. This decrease in speed with increasing amplitude is likely an effect of the "HF←HF MLF, SW," as this effect is not observed in the $3FBL^{osc}_{biArt}$, which differs from the $3FBL^{osc}_{hipB}$ model only by the absence of a CPG component for this feedback pathway. Indeed, the "HF←HF MLF, SW" is a negative feedback, and thus increasing the amplitude of its associated CPG (i.e., μ_{osc}) will reduce the activity of the HF muscle, reducing the HIP flexion velocity and hence increasing the duration of the swing, which in turn decreases the gait speed (as the stride length does not change significantly).

Surprisingly, as little as one oscillator is sufficient to allow significant changes in speed (shown by the $3FBL^{osc}_{biArt}$, see **Figure 8B**). The changes in speed are mainly induced by a modulation of the amplitude μ_{osc}, but with an opposite effect compared to the $3FBL^{osc}_{hipB}$ (i.e., an increase in μ_{osc} leads to an increase in the gait velocity). However, since this effect is accompanied with a shortening of the stride length, this model is unlikely to be relevant; indeed, in humans an increase in speed is usually concomitant to an increase in stride length (Murray et al., 1966).

Note that small changes in speed are still possible with a modulation of the frequency ω, both in the case of the $3FBL^{osc}_{biArt}$

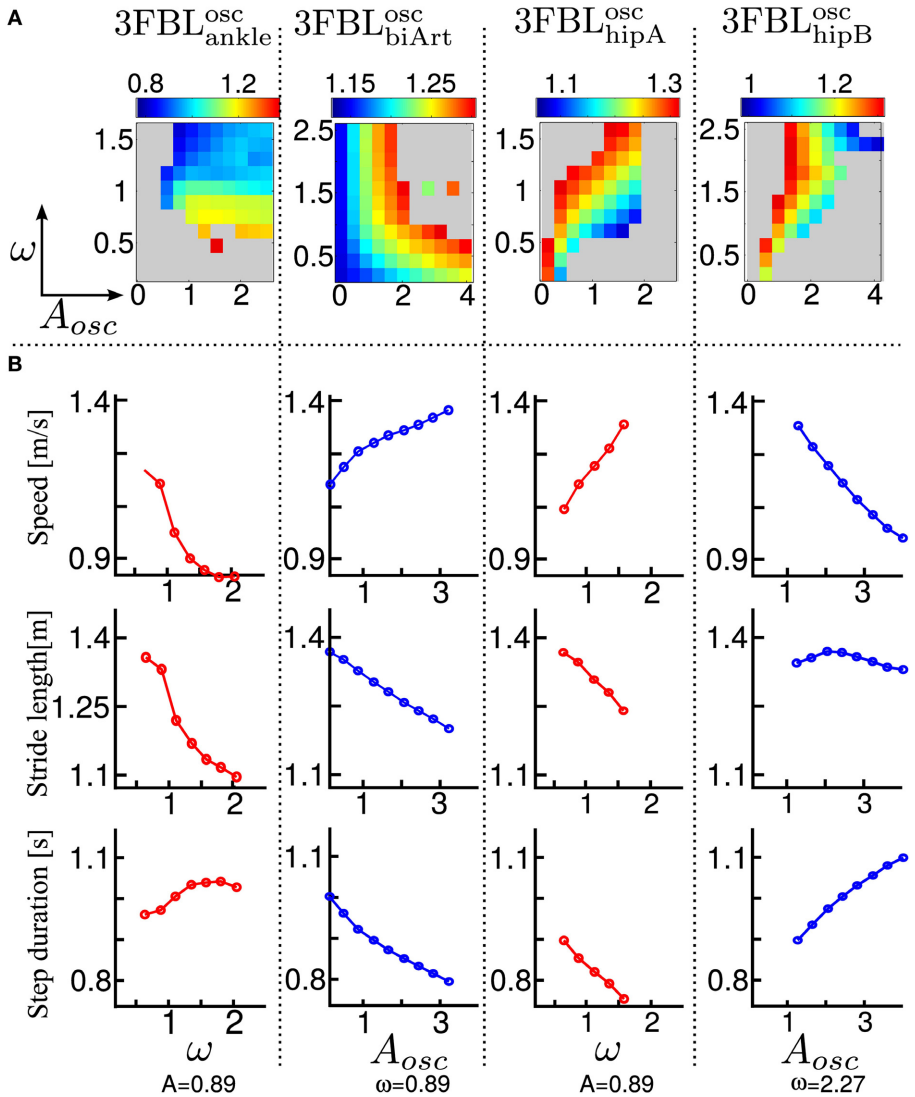


FIGURE 8 | Systematic search study of CPG parameters (supraspinal influences) for the different 3FBL models. The systematic search is done for two parameters: ω, the frequency of the CPG network and μ_{osc}, the CPG-OSC amplitude modulation. Each column corresponds to a given 3FBL model (name at the top, see **Table 3**). **(A)** Heat map of the systematic search. The color indicates the speed of the gait for a given (μ_{osc},ω) pair (gray color means that the gaits was unstable or asymmetric). **(B)** Highest variation in speed possible while maintaining one of the parameters constant (based on the heat map). A red/blue line means that (μ_{osc}/ω) is kept constant, respectively. The value of the constant parameter is indicated at the bottom. The first row shows the speed, the second the stride length, and the third the step duration. Note that the 3FBL$^{min}_{fdb}$ is not shown as its modulation is almost not possible.

and 3FBL$^{osc}_{hipB}$, but to a lesser extent than the 3FBL$^{osc}_{hipA}$. This is expected, as a lower number of CPG—acting on proximal muscles—will have a lower frequency-driven entrainment capacity.

Concerning the pathways acting on distal muscles (i.e., the 3FBL$^{osc}_{ankle}$ model), large changes in speed and step length are observed. However, contrary to what might be expected, an increase in frequency produces a decrease in speed. This is an artifact only possible because of the synchronization mechanism used to ensure the lock-in of the CPG with the mechanical system (see section 2.5.2.2). This effect is thus mainly related to a change in the duration of the burst of the feedforward signal (induced by the change in frequency), rather than to an entrainment between the two systems (i.e., CPG and musculoskeletal system). In other words, the observed gait modulations are due to a modulation of the shape of the signal (change in amplitude and/or duration).

Importantly, increases in speed induced by supraspinal influences on the different 3FBL models do not have the same effect on the gait characteristics (i.e., stride length and step duration). Modulation of the 3FBL$^{osc}_{hipA}$ or 3FBL$^{osc}_{hipB}$ parameters induce very little change in stride length ($< 5\%$). This is explained by the fact those CPGs are active only during swing and modulate the swing speed, but do not impact the swing length (and hence the stride length). Conversely, an increase in speed in the 3FBL$^{osc}_{ankle}$ induces

a significant increase in stride length, as increasing the propulsive force will increase the swing length and thereby the stride length. As previously mentioned, the opposite effect is observed for the $3FBL^{osc}_{biArt}$ (i.e., a decrease in stride length).

In real humans, it is known that, up to a certain point, increases in speed are usually accomplished by a decrease in step duration (i.e., increase in frequency), as well as by an increase in stride length (Murray et al., 1966). As expected, the 4 models exhibit a decrease in step duration with the increase in speed. Interestingly, only a modulation occurring on distal muscles also shows an increase in stride length, suggesting the propulsive force modulation as a means of velocity control.

Results suggest 2 ways of controlling speeds: (1) frequency modulation of CPGs acting on proximal muscles, (2) modulation of burst duration, amplitude and timing of CPGs acting on distal muscles.

4. DISCUSSION

4.1. FBL

The analysis of gaits generated by the optimized FBL model (see section 3.1 for details) highlighted several similarities to healthy humans. Moreover, some solutions of different runs from the same optimization process showed ANKLE kinematics similarities to children suffering from cerebral palsy, highlighting the role that the FBL model could play in terms of modeling locomotion diseases. Children with cerebral palsy show a typical ANKLE flexion (instead of extension) in the early stance, followed by a double bump, visible at both the angle and torque level (Iosa et al., 2010). This is conceivably linked to a reduced hip range of motion, a weakness of tibialis anterior and/or a hypertone of gastrocnemious. Suprisingly some of the solutions described in section 3.1, such as solution 10 (orange line in **Figures 5B,D** right), show both features observed in children with cerebral palsy, i.e., ANKLE flexion in early stance and the double bump visible in both the torque and the angle. Furthermore, solution 10 shows a smaller HIP range of motion compared to solution 1. Finally, the tibilias anterior was found less active at the beginning of gait cycle compared to human physiological gait, as reported for children with cerebral palsy. Conversely, the double bump noted in the model seemed not to be related to an increased muscular activity of gastrocnemious. These interesting similarities, as well as the potential role of the model in disease/injury modeling should be further investigated.

4.2. FBL EXTENSION

Our approach—to use a dynamical system model of CPGs playing the role of feedback predictors—offers an easy and intuitive way of studying the relative importance of the different feedback pathways, and allowed us to highlight several aspects regarding speed control.

4.2.1. *CPG modulations on both proximal and distal muscles allow speed control*

Mixing a constant predictor (CPG-CST) and feedbacks for as little as one pathway already enables speed and step length control. Increasing the level of CPG-CST for one specific pathway results in a flattening of the original feedback signal. Flattening the "SOL←SOL MFF, ST" feedback (i.e., the SOL positive muscle force feedback, active during stance) induces a clear decrease in both the gait speed and stride length, while flattening the "HF←HF MLF, SW" feedback (i.e., the HF negative muscle length feedback, active during swing) induces a clear decrease in the gait speed, but has little effect on the stride length (see **Figure 6B**). Those two observations confirm the intuition that speed changes would arise differently, depending on whether the control is applied during stance or swing. While speed control arising from stance control would more likely use extensor distal muscles, a speed control arising from swing control would more likely use proximal muscles. On the one hand, to be effective, a control acting during the stance should affect the propulsive force, which is mainly controlled by extensor muscles acting on the ankle joint (i.e., SOL and GAS muscles). It is thus not surprising that a modulation of feedback pathways acting on ankle extensor muscles during the stance affects the speed of locomotion (see **Figure 6A**). The effect on stride length is understood as the result of the modulation of the propulsive force: decreasing the propulsive force will decrease the swing length and thereby decrease the stride length. On the other hand, for the control acting during the swing at the level of the HIP flexors, the decrease in speed is not accompanied with any clear reduction in stride length (see **Figure 6B** green), meaning that it is the speed of the swing, but not its amplitude that induced the change in speed.

Similarly, the 3FBL models with CPG components acting on different groups of muscles confirm that speed control can arise from distal muscles extensors during the stance phase, and proximal muscles during the swing phase. We show that changes in speed, induced by a modulation of feedforward signals acting at the level of the ankle muscles, is unlikely due to a modulation of the frequency of the CPG network (see section 3.3.2), but rather induced by changes in burst duration and timing. Conversely, the results from a control acting during the swing at the level of proximal muscles shows that they could, indeed, be due to a modulation of the frequency of a CPG network.

When the CPG activity is modulated, the rest of the system (i.e., the remaining feedbacks) should adapt to the new conditions. Therefore, it is the combined effects of both CPGs and feedbacks that changes the gait properties (such as speed, step length, step duration). It has already been demonstrated that feedbacks acting at the level of the ankle produce such speed-adaptive behaviors (Markowitz et al., 2011). Here we show that this is true regardless of whether the control is applied at the level of proximal or distal muscles.

The proposed spinal architecture was able to generate speed transition ranging from 0.75 to 1.35 [m/s]. While this can seem relatively small compared to the controller proposed in Song and Geyer (2012), in which speed transition ranging from 0.8 to 1.6 [m/s] were obtained, the strategy proposed in this article has the advantage that changes in speed can be obtained without changing the reflex parameters. Furthermore, as the proportion of feedbacks vs. CPGs (i.e., α vector) of the 3FBL models were hand tuned, larger range of speed could be obtained through optimization. Finally, co-optimizing the feedback and feedforward components could also increase the range of speed. Indeed, as already stated, the 3FBL can be viewed as a system made

of two components: a feedforward component and a corrective term, accounting for the differences between the feedback and the feedforward pathways (see section 3.2.3). In this context, the FBL model is a 3FBL model where the feedforward component is zero: the feedback parameters of the FBL are thus optimized for a model without any feedforward component. In this regard, since the 3FBL models were designed on top of an existing FBL model, the feedback parameters are not optimized to work with a non-zero feedforward component. This could also explain the low robustness of the $3FBL_{fdb}^{min}$ model. Furthermore, in a biological point of view, it is obvious that the feedforward components should evolve together with the feedback components. Consequently, in the future, we will investigate the co-evolution of the feedforward and feedback components.

4.2.2. Stable locomotion is produced even with a significant decrease in feedback activity

The $3FBL_{fdb}^{min}$ model shows that stable locomotion can be produced despite a significant decrease in feedback activity. Indeed, stable walking is produced even with a 65% percent reduction in muscle feedback activity. As expected, this large decrease in feedback activity reduces the robustness of the gait to external perturbations (pushes and slope variation), and also considerably reduces the possibilities to control the gait (change in speed/stride length are not possible). This shows that some pathways are more important than others regarding their role as gait stabilizer which can be beneficial to both perturbation resistance and control of the gait.

4.2.3. Exploiting the low dimensional organization of feedback pathways

Interestingly, in all the optimized FBL gaits, all the feedback pathways can be represented with as little as 4 signals found by non-negative matrix factorization (98% correlations between the original signals and the reconstructed one, data not shown). Since motoneurons are a simple linear combination of feedback pathways, the same conclusions are valid when analyzing the motoneurons signals. This low dimensional representation is also found in humans EMG patterns (Clark et al., 2009; Dominici et al., 2011), where only 4 signals, the so-called "motor-primitives," are necessary to faithfully represent the EMG patterns of adult human walking. It would thus be interesting to exploit this low dimensional structure when modeling the feedforward components. In other words, we could model the CPGs as a set of motor-primitives that can be combined together to generate the different motoneurons states. Therefore, instead of viewing the CPG as a feedback predictor, we would view it as a motoneuron predictor. Based on the presented results, our new hypothesis is that the modulation of the timing, amplitude and duration of the motor-primitive will offer a better control of the gait, in terms of speed, stride length, gait transition and adaptation to increasing/decreasing slope.

5. CONCLUSION

In this work, we presented a method to introduce CPGs as feedforward components in a feedback based model of human walking. The proposed strategy is based on the idea that, in a feedback driven system, the feedforward component can be viewed as a feedback predictor. We implemented the feedback predictors using morph oscillators as abstract models of biological CPGs. Thanks to the intrinsic robustness inherited from the feedback pathways, the modulation of CPGs network's frequency and amplitudes were possible, over a broad range, without affecting the overall walking stability. Furthermore, the modulation of the CPGs network's parameters allowed smooth and stable speed changes in a range of 0.6 [m/s]. Preliminary results shows that the same strategy can be used to adapt to larges increase in slope (up to 30%) and to broader speed range (up to 0.8 [m/s]) suggesting that the idea of using feedback predictor as gait modulator can be extended to a large range of applications, highlighting the role biological CPGs could play on top of a reflex based rhythmic movement.

ACKNOWLEDGMENTS

The work presented here was performed in the SYMBITRON project which is supported by EU research program FP7, FET-Proactive initiative "Symbiotic human-machine interaction" (ICT-2013-10) under project contract #611626 and EU research program FP7, FET-Proactive initiative "Whole-body Adaptive Locomotion and Manipulation" (ICT-2013-10) under project contract #611832. The SYMBITRON project is coordinated by University of Twente and the WALKMAN project by the Istituto Italiano di Tecnologia (IIT). We thanks Steeve Berger for its contribution to the implementation of the FBL model. We thanks Yuri Ivanenko, Andrea d'Avella and Nadine Ait Bouziad for helpful discussions.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: http://www.frontiersin.org/journal/10.3389/fnhum.2014.00371/abstract

REFERENCES

Ajallooeian, M., van den Kieboom, J., Mukovskiy, A., Giese, M. A., and Ijspeert, A. J. (2013). A general family of morphed nonlinear phase oscillators with arbitrary limit cycle shape. *Physica D* 263, 41–56. doi: 10.1016/j.physd.2013.07.016

Bhargava, L. J., Pandy, M. G., and Anderson, F. C. (2004). A phenomenological model for estimating metabolic energy consumption in muscle contraction. *J. Biomech.* 37, 81–88. doi: 10.1016/S0021-9290(03)00239-2

Clark, D. J., Ting, L. H., Zajac, F. E., Neptune, R. R., and Kautz, S. A. (2009). Merging of healthy motor modules predicts reduced locomotor performance and muscle coordination complexity post-stroke. *J. Neurophysiol.* 103, 844–857. doi: 10.1152/jn.00825.2009

Collins, S. H., Adamczyk, P. G., and Kuo, A. D. (2009). Dynamic arm swinging in human walking. *Proc. R. Soc. B Biol. Sci.* 276, 3679–3688. doi: 10.1098/rspb.2009.0664

Czyżżak, P., and Jaszkiewicz, A. (1998). Pareto simulated annealing—a metaheuristic technique for multiple-objective combinatorial optimization. *J. Multi-Crit. Decis. Anal.* 7, 34–47. doi: 10.1002/(SICI)1099-1360(199801)7:1<34::AID-MCDA161>3.0.CO;2-6

Dimitrijevic, M. R., Gerasimenko, Y., and Pinter, M. M. (1998). Evidence for a spinal central pattern generator in humansa. *Ann. N.Y. Acad. Sci.* 860, 360–376. doi: 10.1111/j.1749-6632.1998.tb09062.x

Dominici, N., Ivanenko, Y. P., Cappellini, G., d'Avella, A., Mondi, V., Cicchese, M., et al. (2011). Locomotor primitives in newborn babies and their development. *Science* 334, 997–999.

Donelan, J. M., Kram, R., and Kuo, A. D. (2002). Simultaneous positive and negative external mechanical work in human walking. *J. Biomech.* 35, 117–124. doi: 10.1016/S0021-9290(01)00169-5

Dzeladini, F. (2013). *Implementation of a Human Feedback-based Locomotion and its Control by means of a Feedforward Component inspired by Central Pattern Generators*. Master Thesis, EPFL, Lausanne.

Geyer, H., and Herr, H. (2010). A muscle-reflex model that encodes principles of legged mechanics produces human walking dynamics and muscle activities. *IEEE Trans. Neural Syst. Rehabil. Eng.* 18, 263–273. doi: 10.1109/TNSRE.2010.2047592

Geyer, H., Seyfarth, A., and Blickhan, R. (2003). Positive force feedback in bouncing gaits? *Proc. R. Soc. B Biol. Sci.* 270, 2173–2183. doi: 10.1098/rspb.2003.2454

Grillner, S., and Wallen, P. (1985). Central pattern generators for locomotion, with special reference to vertebrates. *Ann. Rev. Neurosci.* 8, 233–261. doi: 10.1146/annurev.ne.08.030185.001313

Hill, A. V. (1938). The heat of shortening and the dynamic constants of muscle. *Proc. R. Soc. B Biol. Sci.* 126, 136–195. doi: 10.1098/rspb.1938.0050

Iosa, M., Fusco, A., Marchetti, F., Morone, G., Caltagirone, C., Paolucci, S., et al. (2013). The golden ratio of gait harmony: repetitive proportions of repetitive gait phases. *BioMed. Res. Int.* 2013, 1–7. doi: 10.1155/2013/918642

Iosa, M., Morelli, D., Nanni, M. V., Veredice, C., Marro, T., Medici, A., et al. (2010). Functional taping: a promising technique for children with cerebral palsy. *Dev. Med. Child Neurol.* 52, 587–589. doi: 10.1111/j.1469-8749.2009.03539.x

Ijspeert, A. J. (2008). Central pattern generators for locomotion control in animals and robots: a review. *Neural Netw.* 21, 642–653. doi: 10.1016/j.neunet.2008.03.014

Kennedy, J., and Eberhart, R. (1995). "Particle swarm optimization," in *Proceedings of IEEE International Conference on Neural Networks*, Vol. 4 (Perth, WA), 1942–1948. doi: 10.1109/ICNN.1995.488968

Kuo, A. D. (2002). The relative roles of feedforward and feedback in the control of rhythmic movements. *Motor Control* 6, 129–145.

Li, B.-B., Wang, L., and Liu, B. (2008). An effective PSO-Based hybrid algorithm for multiobjective permutation flow shop scheduling. *IEEE Trans. Syst. Man Cybern. A Syst. Hum.* 38, 818–831. doi: 10.1109/TSMCA.2008.923086

MacKay-Lyons, M. (2002). Central pattern generation of locomotion: a review of the evidence. *Phys. Ther.* 82, 69–83.

Markowitz, J., Krishnaswamy, P., Eilenberg, M. F., Endo, K., Barnhart, C., and Herr, H. (2011). Speed adaptation in a powered transtibial prosthesis controlled with a neuromuscular model. *Philos. Trans. R. Soc. B Biol. Sci.* 366, 1621–1631. doi: 10.1098/rstb.2010.0347

Michel, O. (2004). Webots: professional mobile robot simulation. *J. Adv. Robot. Syst.* 1, 39–42.

Murray, M. P., Kory, R. C., Clarkson, B. H., and Sepic, S. (1966). Comparison of free and fast speed walking patterns of normal men. *Am. J. Phys. Med. Rehabil.* 45, 8–24. doi: 10.1097/00002060-196602000-00002

Perry, J., k, S. T., and Davids, J. R. (1992). Gait analysis: normal and pathological function. *J. Pediatr. Orthop.* 12, 815. doi: 10.1097/01241398-199211000-00023

Siegel, A., Sapru, H. N., and Siegel, H. (2006). *Essential Neuroscience*. Philadelphia, PA: Lippincott Williams and Wilkins.

Song, S., and Geyer, H. (2012). "Regulating speed and generating large speed transitions in a neuromuscular human walking model," in *IEEE International Conference on Robotics and Automation* (St Paul, MN).

Taga, G. (1994). Emergence of bipedal locomotion through entrainment among the neuro-musculo-skeletal system and the environment. *Physica D* 75, 190–208. doi: 10.1016/0167-2789(94)90283-6

Wang, J. M., Hamner, S. R., Delp, S. L., and Koltun, V. (2012). Optimizing locomotion controllers using biologically-based actuators and objectives. *ACM Trans. Graph.* 31, 1–11. doi: 10.1145/2185520.2185521

Weyand, P. G., Smith, B. R., Puyau, M. R., and Butte, N. F. (2010). The mass-specific energy cost of human walking is set by stature. *J. Exp. Biol.* 213(Pt 23), 3972–3979. doi: 10.1242/jeb.048199

Winter, D. A. (2009). *Biomechanics and Motor Control of Human Movement*. Hoboken, NJ: John Wiley and Sons, Inc. doi: 10.1002/9780470549148

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.

Received: 28 February 2014; accepted: 14 May 2014; published online: 26 June 2014.
*Citation: Dzeladini F, van den Kieboom J and Ijspeert A (2014) The contribution of a central pattern generator in a reflex-based neuromuscular model. Front. Hum. Neurosci. **8**:371. doi: 10.3389/fnhum.2014.00371*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 17 March 2014
doi: 10.3389/fnhum.2014.00148

Intralimb coordination as a sensitive indicator of motor-control impairment after spinal cord injury

Lea Awai* and Armin Curt

Spinal Cord Injury Center, Balgrist University Hospital, University of Zurich, Zurich, Switzerland

Edited by:
Marco Iosa, Istituto di Ricovero e Cura a Carattere Scientifico – Fondazione Santa Lucia, Italy

Reviewed by:
Giorgio Scivoletto, Istituto di Ricovero e Cura a Carattere Scientifico – Fondazione Santa Lucia, Italy
Randy D. Trumbower, Emory University, USA

****Correspondence:***
Lea Awai, Spinal Cord Injury Center, Balgrist University Hospital, University of Zurich, Forchstrasse 340, CH-8008 Zurich, Switzerland
e-mail: lawai@paralab.balgrist.ch

Background: Recovery of walking function after neurotrauma, e.g., after spinal cord injury, is routinely captured using standardized walking outcome measures of time and distance. However, these measures do not provide information on possible underlying mechanisms of recovery, nor do they tell anything about the quality of gait. Subjects with an incomplete spinal cord injury are a very heterogeneous group of people with a wide range of functional impairments. A stratification of these subjects would allow increasing sensitivity for hypothesis testing and a more targeted treatment strategy.

Methods: The gait of incomplete spinal cord injured subjects was compared to healthy control subjects by analyzing kinematic data obtained by a 3-D motion capture system. Hip–knee angle-angle plots (cyclograms) informed on the qualitative aspect of gait and the intralimb coordination. Features of the cyclogram, e.g., shape of the cyclogram, cycle-to-cycle consistency and its modulation due to changes in walking speed were discerned and used to stratify spinal cord injured subjects.

Results: Spinal cord injured subjects were unable to modulate their cyclogram configuration when increasing speed from slow to preferred. Their gait quality remained clearly aberrant and showed even higher deviations from normal when walking at preferred speed. Qualitative categorization of spinal cord injured subjects based on their intralimb coordination was complemented by quantitative measures of cyclogram shape comparison.

Discussion: Spinal cord injured subjects showed distinct distortions of intralimb coordination as well as limited modulation to changes in walking speed. The specific changes of the cyclograms revealed complementary insight in the disturbance of lower-limb control in addition to measures of time and distance and may be a useful tool for patient categorization and stratification prior to clinical trial inclusion.

Keywords: spinal cord injury, human, gait, intralimb, coordination, categorization, cyclogram

INTRODUCTION

The most obvious impairment after spinal cord injury (SCI) is the complete or partial loss of lower-limb motor function, clinically assessed as decreased walking speed and alterations in time–distance measures (e.g., step length, step frequency, double-, and single-limb support phase; Krawetz and Nance, 1996; Pepin et al., 2003). These parameters are often used to monitor recovery and to capture locomotor capacity, but they lack the ability to unveil underlying neurological mechanisms (Dobkin et al., 2006; Tamburella et al., 2013). However, the quality of walking, especially the ability to coordinate lower-limb segments and joints, represents aspects of gait that complement the information about walking capacity and additionally provides insights into underlying mechanisms. A clinical inventory for gait quality assessment, relying on a subjective evaluation of SCI walking by a trained therapist has been developed (Field-Fote et al., 2001). Yet, it is not easy to quantify and scale gait quality and even compare it across different neuromotor disorders (Robinson and Smidt, 1981). The latter may be one of the reasons for the limited amount of published studies performing quantitative kinematic analysis of gait quality. Previous studies in healthy people have shown that, irrespective of walking speed, lower-limb segments are controlled interdependently as quantified by the co-variation of the elevation angles (Borghese et al., 1996; Grasso et al., 1998; Lacquaniti et al., 1999). This constraint in multi-segmental coordination suggests an underlying rule of motor control ensuring secure upright locomotion. The controlled behavior of multiple segments subserves the ultimate goal of controlling limb endpoints, which is believed to be controlled on different levels within very restricted boundaries. At the cost of aberrant muscle-activity pattern, the endpoint is controlled in a way that its trajectory shows very little variability in healthy individuals and gets closer to normal during training in iSCI subjects (Winter, 1992; Ivanenko et al., 2003). Compared to elevation angles, joint angles seem to be more variable and less reproducible (Borghese et al., 1996; Grasso et al., 1998). Lower-limb kinematics (i.e., hip-, knee-, and ankle-angle profiles) have been assessed in SCI (Abel et al., 2002; Gil-Agudo et al., 2011) and reveal aberrant and heterogeneous behaviors. But the simple description of joint angles during a gait cycle does not inform on intersegmental coordination. The intralimb

coordination, measured as the simultaneous coordination of hip- and knee-angles, was found to be distorted in incomplete SCI (iSCI) and to be less amendable after locomotor training (Field-Fote and Tepavac, 2002; Nooijen et al., 2009). However, the degree of distortion has not been quantified and the measure of intralimb coordination has not been further analyzed. Also, the speed modulation of the intralimb coordination has not been considered, which might reveal information on underlying deficits. The aim of the present study was to characterize intralimb coordination of iSCI subjects qualitatively as well as quantitatively by means of hip–knee cyclograms. These measures may reveal different patterns and categories of walking impairment in iSCI subjects and may uncover mechanisms of lower-limb motor control. The latter findings may be applied to improve the stratification of patients for tailored (i.e., specific to the impairment) interventions to improve walking outcomes in acute as well as chronic iSCI.

MATERIALS AND METHODS

SUBJECTS

Incomplete SCI inclusion criteria: a diagnosed iSCI; age: 18 years or older; iSCI subjects need to be able to at least stand and walk without the assistance of another person (no manual leg movement by therapist). Exclusion criteria: Subjects suffering from neurological disorders other than SCI; gait impairments not caused by SCI. Healthy subjects with no neurological disorders or gait impairments served as the control group. The study was approved by the ethics committee of the Canton of Zurich, Switzerland. Participants gave their written informed consent. 19 iSCI subjects (4 female, 15 male; age: 50.0 ± 15.9 years; height: 172.6 ± 7.7 cm; weight: 75.8 ± 13.2 kg; **Table 1**) and 19 healthy control subjects (10 female, 9 male; age: 40.7 ± 13.7 years; height: 173.2 ± 9.3 cm; 68.9 ± 13.0 kg) were included in this study.

MATERIALS

Kinematic data was recorded using eight infrared cameras (T10, Vicon motion systems Ltd., Oxford, UK) at 200 Hz and two synchronized digital high-speed video cameras (pilot series, Basler AG, Ahrensburg, D) at 100 Hz. 16 reflective markers (16 mm diameter) were placed on bony landmarks according to the Vicon Plug-in Gait lower-body model. During treadmill (TM) walking pressure sensors underneath the TM belt recorded the force distribution of the footsoles at 120 Hz (Zebris FDM-T, zebris Medical GmbH, Isny im Allgäu, D). Kinematic data was recorded and post-processed using the Vicon Nexus Software (1.7.1). Trajectories were smoothed and gaps interpolated using Woltring's cross-validatory quintic-spline routine with a mean squared error (MSE) of 10 mm^2. Continuous data from ∼20 consecutive gait cycles was cut into individual gait cycles and time-normalized using linear interpolation. Data from the pressure sensors underneath the TM belt was recorded by the same PC using the Zebris WinFDM-T software (02.01.01). Recordings were synchronized via a ±5 V trigger signal.

PROTOCOL

Subjects walked barefooted both overground (OG) and on a TM, where walking speed could be controlled and varied. iSCI subjects were allowed to use assistive devices for OG walking or to hold handrails when walking on the TM. Subjects were first asked to walk OG to assess their preferred walking speed. On the TM, iSCI subjects were then asked to walk at a slow speed (0.5 km/h ≈ 0.14 m/s) and at preferred OG walking speed.

OUTCOME VARIABLES

Approximately twenty gait cycles were analyzed per walking speed. The hip and knee angles were time-normalized to one gait cycle (500 samples) using linear interpolation. The intralimb coordination was illustrated by hip–knee cyclograms whose vertical and horizontal expansion corresponded to the maximal hip- and knee-range of motion [ROM (°)] during walking, respectively. The within-subject cycle-to-cycle consistency of these cyclograms was quantified using the angular component of coefficient of correspondence (ACC; Field-Fote and Tepavac, 2002). Inter-subject (within-group) variability of the cyclogram was assessed after translation of the centroids of cyclograms to the origin. The inter-subject variability was calculated as the cumulative ellipse area with half axes (a and b, see Eq. 1) corresponding to the between-subject standard deviation of hip- and knee-angles, respectively, for 20 equal bins of time-normalized cyclograms:

$$\mathrm{Var_n} = \sum_{i=1}^{20} \Pi * a_{n,i} b_{n,i} \tag{1}$$

Index *n* refers to subject number, *i* describe the bin number. The shape difference between two cyclograms was quantified as the square root of the sum of squared distances (SSD) after uniform scaling and translation of the cyclogram centroids to the origin:

$$\mathrm{SSD_{j,k}} = \sqrt{\sum_i \left(\alpha_{j,i} - \alpha_{k,i}\right)^2 + \left(\beta_{j,i} - \beta_{k,i}\right)^2} \tag{2}$$

Equation 2 compares cyclogram *j* to cyclogram *k* where α and β correspond to the scaled and transformed hip- and knee-angles, respectively, at sample point *i*. Within-group SSD was calculated as the average SSD of every pairing of subjects within a group [mean of n*(n-1)/2 SSDs]. The between group SSD was calculated by comparing the mean cyclograms of two groups. Data analysis was performed using custom-written Matlab scripts (Matlab R2013a; The MathWorks Inc., Natick, MA, USA). A categorization based on the presence or absence of specific characteristics of the cyclogram shape was performed with the goal of stratifying patients into different groups of impairment. Within each group, iSCI subjects were additionally ranked according to the severity of alterations, resulting in a continuous ranking of iSCI subjects based on their cyclogram (lower ranking meaning worse cyclogram).

STATISTICAL ANALYSIS

Normality of data distribution was tested using histograms and QQ plots. Non-normally distributed ACC values were compared using the non-parametric Kruskal–Wallis test and, if revealing significance, the following *post hoc* tests were applied: independent data was tested using a Mann–Whitney *U* test and a Wilcoxon signed rank test was employed for paired data samples. The significance level was corrected for using Bonferroni

Table 1 | Descriptive data of patients.

ID	Age (years)	Sex	Cause of SCI	Level of SCI	Disease progress	LEMS	Assistive device
01	73	Male	Traumatic	C5	Chronic	24.5	Wheeler
02	24	Male	Traumatic	C3	Chronic	24.5	–
08	60	Female	Spinal ischemia	T5	122 d	23.0	Wheeler
09	48	Female	Traumatic	T7	Chronic	15.0	One stick
10	39	Female	Spinal myelitis	C, T	46 d	19.0	Wheeler
12	65	Male	Hematoma	C6	Chronic	24.0	–
13	23	Male	Traumatic	C7	1 year	9.5	Wheeler
15	78	Male	Diverse	C3/4	50 d	22.5	Wheeler
16	60	Male	Spinal canalstenosis	T9/10	76 d	25.0	–
17	55	Male	Cervicalmyelopathy	C5	Chronic	19.5	–
18	63	Female	Epiduralabscess	T4	Chronic	23.0	Crutches
19	43	Male	Traumatic	C2	Chronic	24.0	Two sticks
20	61	Male	Disc prolapse	C2	Chronic	25.0	–
21	40	Male	Traumatic	C5	Chronic	22.5	–
22	64	Male	Spondylitis, abscess	T4	Chronic	23.5	–
23	32	Male	Traumatic	T11	75 d	8.0	Crutches
24	36	Male	Traumatic	L4/5	78 d	23.5	–
25	41	Male	Traumatic	C7	1 year	24.0	–
26	48	Female	Disc prolapse	T10	15 d	24.0	Two sticks

LEMS = mean value of left and right lower extremity motor score (max = 25).

correction resulting in an α-value of $0.05/4 = 0.0125$. In order to assess the relation between the qualitative cyclogram ranking and objective measures (i.e., walking speed, hip-, and knee-ROM) non-parametric Spearman correlation coefficient ρ (rho) was calculated. Matlab R2013a was used for all statistical analyses.

RESULTS

Incomplete SCI subjects exhibited a mean preferred walking speed of 0.57 ± 0.32 m/s while control subjects preferably walked at 1.16 ± 0.15 m/s.

PATIENT CATEGORIZATION

The cyclograms of healthy control subjects at preferred speed exhibited typical characteristics that could be found in all subjects. These features are listed in the box on the left of **Figure 1A**. iSCI subjects showed one or more alterations of certain properties of the cyclogram and depending on the degree of deviation were classified into four groups of impairment (**Figure 1B**). The specific alterations of cyclogram characteristics are described in the boxes on the left. Accordingly, four iSCI subjects ended up in group 1 (unpredictable gait pattern), six iSCI subjects qualified for group 2 (crouched gait pattern), five iSCI subjects were classified into group 3 (resilient gait pattern), and finally four iSCI subjects exhibited a regular gait pattern (group 4). Correlation analysis between the cyclogram ranking and preferred OG speed, hip-ROM, and knee-ROM revealed a high correlation between preferred speed and cyclogram ranking (Spearman's $\rho = 0.751$; **Figure 2B**) while both hip- and knee-ROM were relatively indifferent to the quality of the cyclogram (Spearman's $\rho = 0.267$ and 0.330, respectively; **Figure 2C**).

INTRALIMB COORDINATION

The Kruskal–Wallis test revealed significant group differences in non-normally distributed ACC (χ^2: 59.59, $p < 0.001$). At the slow walking speed (0.5 km/h) the cyclograms of healthy subjects were similarly distorted and irregular as iSCI subject's cyclograms (**Figure 3A**). In line with this, the *post hoc* test revealed that the ACC did not differ between the two groups at the slow speed (Mann–Whitney U test: $p = 0.840$; **Figure 2A**). In contrast, control subjects normalized their cyclogram configuration to a very uniform shape between subjects when walking at preferred speed (**Figure 3A**). Likewise, the ACC was higher at preferred speed compared to the slow speed (Wilcoxon signed rank test: $p < 0.001$). iSCI subjects were also able to increase their ACC from slow to preferred speed (Wilcoxon signed rank test: $p = 0.005$), but their value was still significantly lower compared to healthy subjects (Mann-Whitney U test: $p < 0.001$). In contrast to control subjects, iSCI subject's cyclograms did not converge toward normal and were still very heteromorphic. The within-group cyclogram variability during a gait cycle was similar in healthy control subjects and in iSCI subjects at the slow walking speed (**Figure 3B**). The cumulative variability for 20 equal time bins was 3238.6 mm^2 in controls and 3149.9 mm^2 among iSCI subjects. However, at preferred walking speed, control subjects showed a remarkably

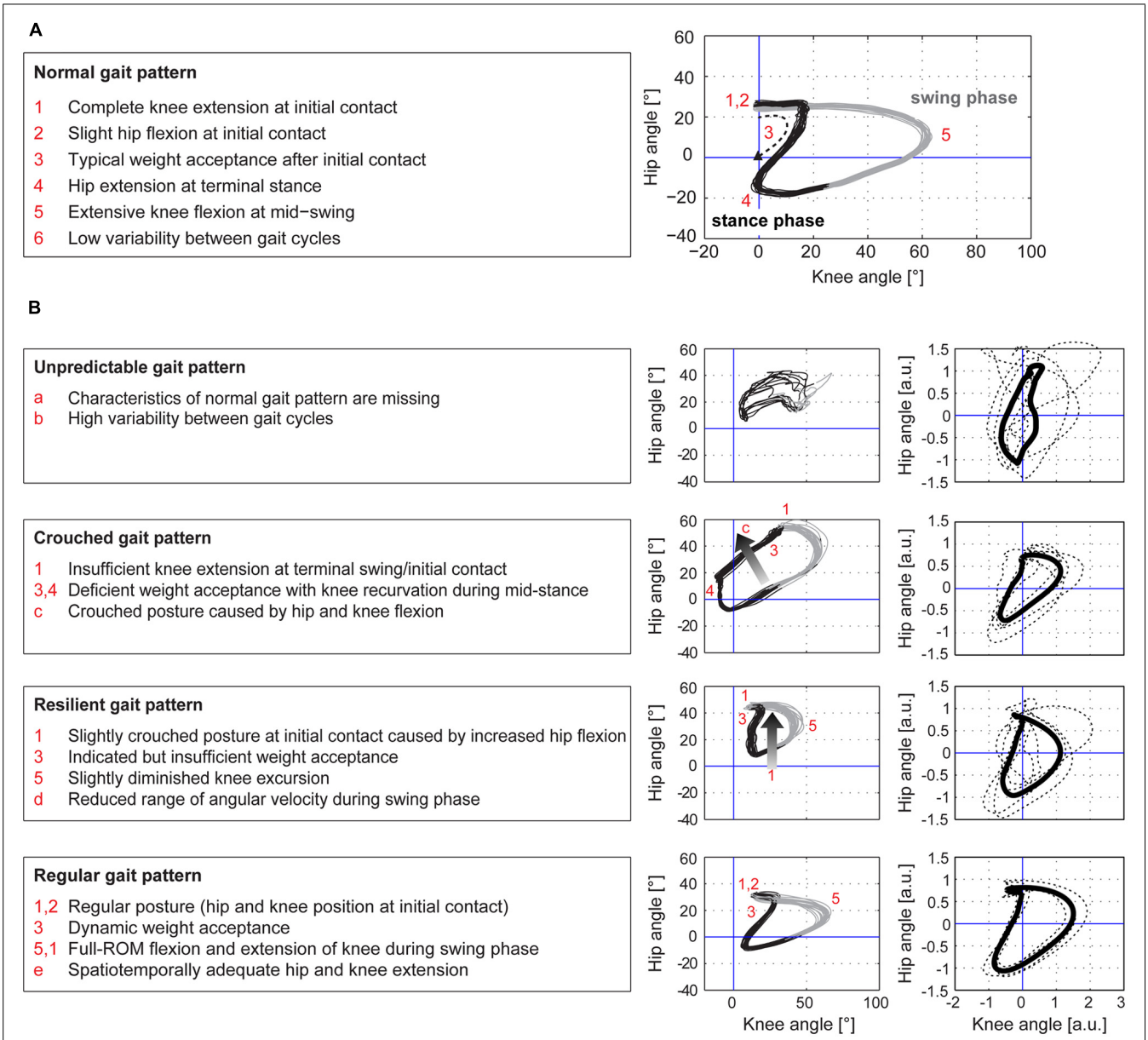


FIGURE 1 | Hip-knee cyclograms of healthy controls and iSCI subjects. The hip–knee cyclogram of a healthy control subject with its typical characteristics is depicted in panel **(A)**. iSCI subjects were classified into four groups of impairment, group 1 (unpredictable gait pattern) being the most affected group while group 4 (regular gait pattern) showed normal cyclograms **(B)**. The criteria for each group are listed in the boxes on the left. The cyclograms in the left column show one representative example per group, the right column depicts the individual cyclograms of each iSCI subject within a group (dashed lines) and the group mean cyclogram (solid bold line) after translation of the cyclogram's centroid to origin (zero) and uniform scaling.

lower variability than iSCI subjects. The cumulative variability at preferred speed for 20 time bins among control subjects was 650.9 mm^2 while it was even higher at preferred speed compared to the slow speed in iSCI subjects (4152.0 mm^2). The SSD, representing the amount of shape difference after uniform scaling and translation (**Table 2**), was smaller within the healthy control group (SSD = 6.44) compared to iSCI subjects (SSD = 32.96) at preferred speed, but showed no difference at the slow speed (healthy: SSD = 14.00, iSCI: SSD = 11.74; **Figure 4**). The between-group shape difference was identical at the slow speed (SSD = 6.49) as at preferred speed (SSD = 6.46). **Table 2** also shows the values of cyclogram shape comparison between each of the four groups of iSCI subjects compared to the control group. At preferred speed, the difference to normal shape according to the SSD coincided with the qualitative categorization of the cyclogram configuration.

DISCUSSION

The kinematic analysis of walking pattern in human iSCI allows for categorization of lower-limb control during walking while these

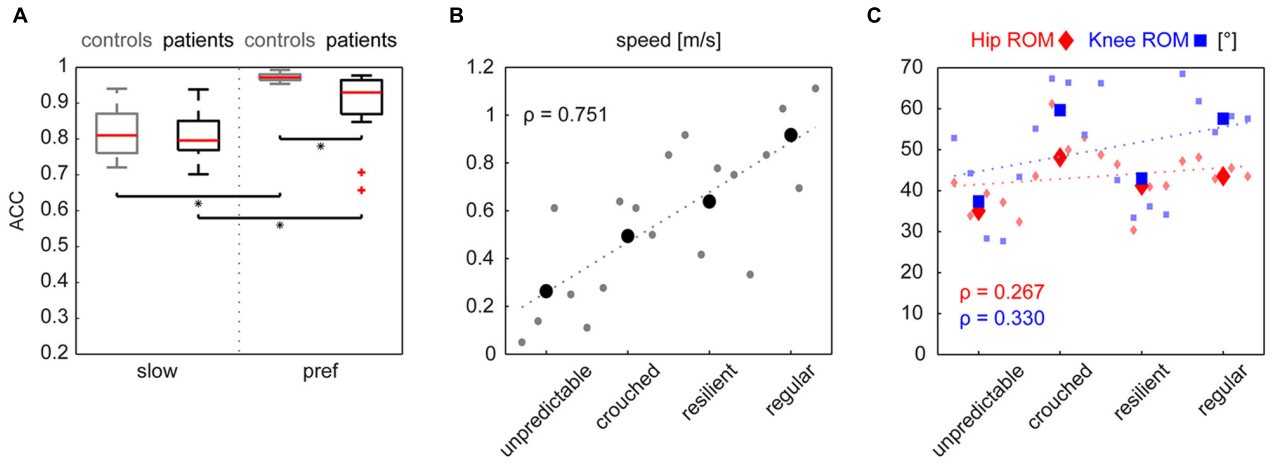


FIGURE 2 | (A) The cycle-to-cycle shape consistency within a subject is quantified as the angular component of coefficient of correspondence (ACC). This value can attain values between 0 (no congruency between cycles) and 1 (complete shape congruency from cycle to cycle). Both groups increased their cyclogram consistency when changing from a slow to preferred walking speed. iSCI subjects only showed lower values at preferred walking speed. Statistical significance ($p < 0.0125$) is indicated by an asterisk. Panel **B** shows the correlation indicated by Spearman's ρ of the cyclogram ranking to preferred OG walking speed and to hip- and knee-ROMs **(C)**. Little dots represent single values of individual subjects while the larger symbols represent the mean value per group.

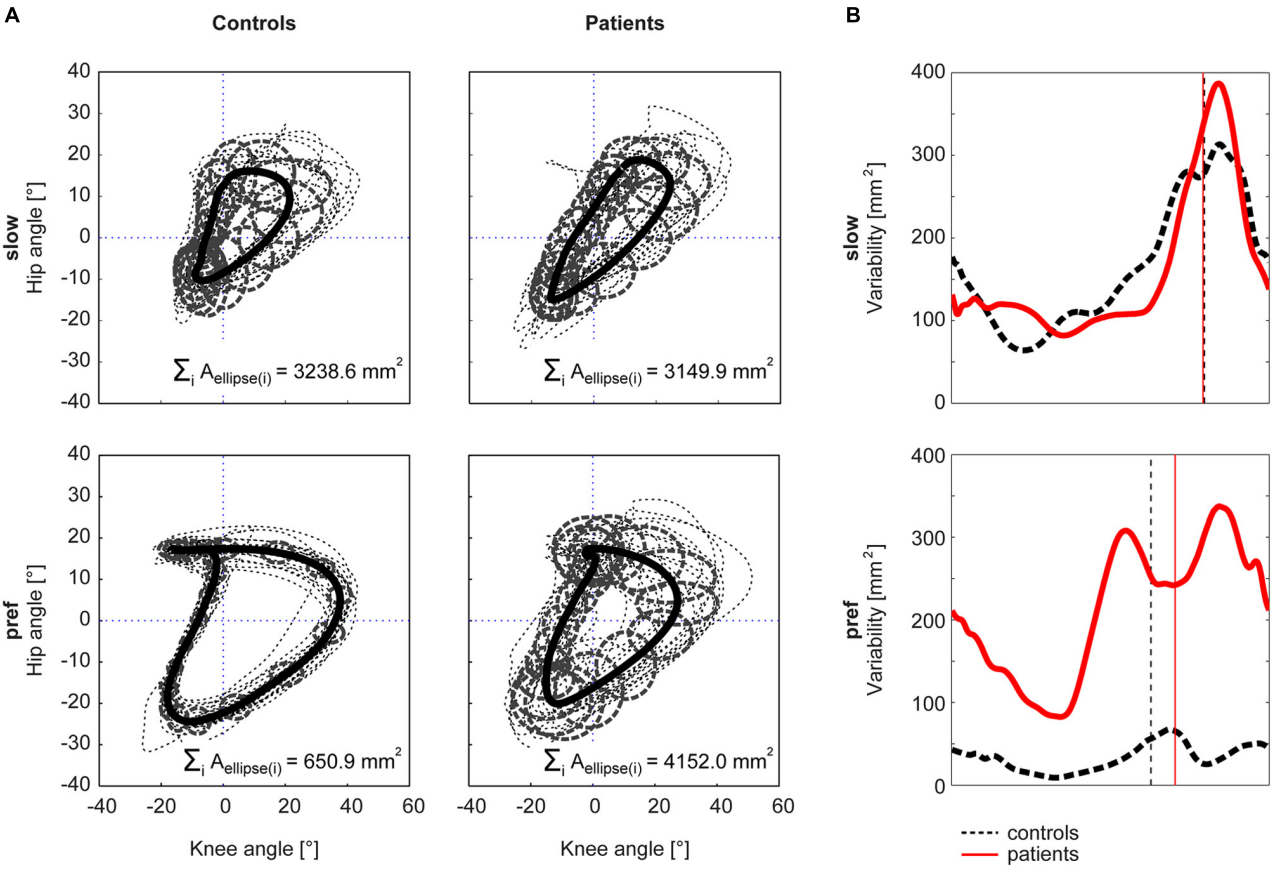


FIGURE 3 | Within-group cyclogram variability. The variability of the cyclogram between subjects within a group was quantified by the cumulated elliptic area for 20 bins per gait cycle **(A)**. The half axes of the ellipse correspond to the between-subject standard deviation of the hip- and knee-angles, respectively. **(B)** shows the course of the variability at every time point of a gait cycle. The control group is represented by a black dashed line, iSCI group is depicted in red. The vertical lines mark the time point of toe-off.

Table 2 | Cyclogram-shape differences.

Group comparison	SSD	
	Slow	Preferred
Within control group	14.00	6.44
Within patients	11.74	32.96
Between controls and patients	6.49	6.46
Between group 1 and controls	9.02	13.64
Between group 2 and controls	7.20	8.86
Between group 3 and controls	9.77	5.46
Between group 4 and controls	4.22	2.73

SSD = square root of sum of squared distances. The within-group SSD was calculated by comparing the cyclograms of subjects each by each and then calculating the mean value. The between-groups SSD was calculated by comparing the mean cyclogram per group.

qualitative readouts of gait performance are complementary to measures of walking distance and speed. While the latter measures are able to quantify the capacity of walking the qualitative scoring provides additional information regarding intralimb motor control. The consideration of these combined outcomes will be meaningful to better target treatment interventions and improve the stratification of most suitable patients for clinical trials.

MODULATION OF INTRALIMB COORDINATION

Visual inspection and quantitative analysis of the cyclograms at a slow walking speed (i.e., 0.5 km/h) revealed that healthy control subjects exhibited similarly variable intralimb coordination as iSCI subjects. Speed increments in the two groups to preferred walking speed, however, induced distinct adaptations in cyclogram configuration. While effectively all control subjects gained a very regular and inter-subjectively uniform cyclogram at their preferred walking speed, iSCI subjects maintained considerable shape variability between subjects and their cyclograms remained different from the typical shape found in healthy subjects. The visually observed lack of cyclogram modulation in iSCI subjects was also made evident by the quantification of shape differences represented by the SSD. According to this value, the inter-subject shape difference within the iSCI group was bigger at preferred speed compared to the slow speed. The lack of shape assimilation is contrary to healthy subjects and might suggest that patient's impairments become more pronounced when walking at preferred speed. In iSCI subjects with most severe shape deformations (group 1 with the highest cyclogram impairments and the lowest walking speed) the increase in speed even induced an increase in shape deformity. This increase in shape deformity did not occur randomly as the reproducibility of this aberrant shape (i.e., ACC) increased at preferred walking speed, indicating that iSCI subjects have learned and consolidated an aberrant walking pattern in a rather abnormal manner.

NEURAL MECHANISMS

The lack of cyclogram modulation when increasing slow walking speed to preferred speed, at which locomotion is supposed to be more economic (Cotes and Meade, 1960) may be associated with a limited access to supraspinal control. We deliberately refer to the term "supraspinal", including corticospinal as well as other descending motor tracts (e.g., rubrospinal or reticulospinal). Several aspects of locomotion, e.g., reciprocal activation of lower-limb muscles, appear to be primarily controlled at a spinal level and depend less on supraspinal efforts as revealed in preclinical studies in animals (Barbeau and Rossignol, 1987; Duysens and Van de Crommert, 1998). In humans, however, the correct spatiotemporal coordination of the lower-limb muscles and joints during locomotion (complex postural control and segmental interplay) seems to be more dependent on intact and sufficient supraspinal control (Kuhn, 1950). Interestingly, in the present study, an individual iSCI subject was not able to normalize his or her cyclogram when increasing the walking speed from slow to preferred speed despite increasing supraspinal neural drive (Bachmann et al., 2013). However, those iSCI subjects with a higher preferred OG walking speed, probably reflecting greater access to supraspinal speed-driving centers, showed a

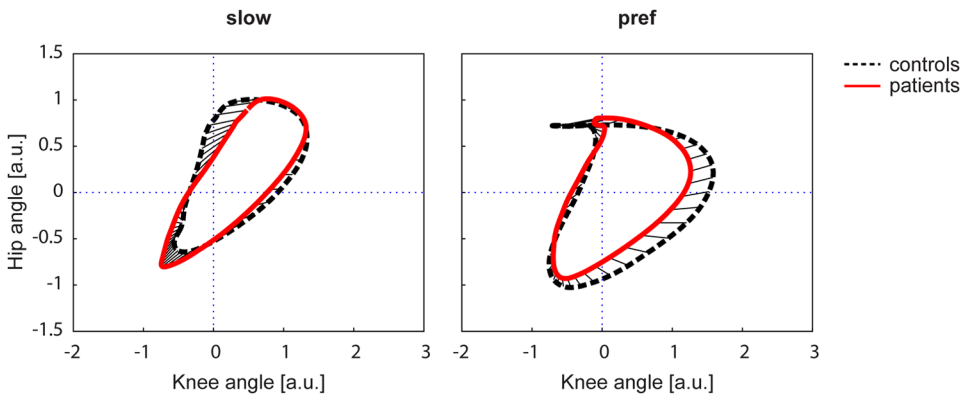


FIGURE 4 | The square root of the sum of squared distances (SSD) calculated the difference in shape of two figures. The cyclograms are shown after translation of the centroid to origin (zero) and uniform scaling. The control group is represented by a black dashed line, iSCI group is depicted in red. The straight black lines indicate the deviation of the two figures. a.u. = arbitrary units.

closer-to-normal cyclogram. The distinct input on walking control can also be found in Parkinson's subjects, where the relation between step length and cadence with changes in speed is distorted [contrary to iSCI (Pepin et al., 2003)] while the hip–knee cyclogram retains a normal but small scaled shape (Morris et al., 1998, 2005). The distortion of cyclograms in iSCI subjects may not only originate from motor deficits, but may also be a consequence of impaired sensory feedback. Motor behavior is known to be affected in disorders associated with severe sensory impairment (Courtemanche et al., 1996; Manor and Li, 2009). Studies with deafferented subjects particularly revealed that the interjoint coordination was distorted while performing an upper-limb task (Sainburg et al., 1993). Even though skilled upper-limb movements depend more strongly on voluntary control compared to lower-limb movements (Nakajima et al., 2000; MacLellan et al., 2013), a deterioration of peripheral sensory feedback is sufficient to disrupt complex interjoint limb movements (Sainburg et al., 1993).

IMPLICATIONS FOR TREATMENT AND CLINICAL TRIALS

The most widely used categorization scheme for SCI is the American Spinal Injury Association (ASIA) Impairment Scale (AIS). This scale is of rather low resolution and does not well distinguish patients upon walking abilities. In the present iSCI-subject sample the majority was classified as AIS D but ranged from AIS C/D to even AIS A (a subject with lesion level below T11). The categorization of iSCI subjects based on the cyclogram can complement the ASIA assessments and may serve several goals. From a therapist's point of view treatment interventions aiming at improving locomotion may become better tailored to the patient's walking impairment. Depending on the aim of training studies (e.g., improvement of quantitative measures and/or gait quality) different outcome measures should be chosen. Interventional studies may reveal that walking speed is improved while the gait quality remains impaired, or vice versa. Clinical trials investigating novel interventions (e.g., effect of drugs or cell transplantation) may have harmful or desired beneficial effects. By deploying the most sensitive assessment battery, differential effects may be discerned, and mechanisms of action may be recognized.

LIMITATIONS

A sample size of 19 iSCI subjects is too low to be able to state whether the categorization based on the cyclogram quality is sufficient to stratify every iSCI subject. The categorization should be tested in a larger cohort, from which one could retrieve further quantitative measures that may serve as threshold values for future guidelines.

CONCLUSION

In clinical routines walking ability is captured by walking speed and distance covered, but the quality of walking is frequently ignored or reported according to subjective rater evaluation. Yet, the present study shows that the intralimb coordination represented by the hip–knee cyclogram reveals reliable and sensitive information on walking capacity after iSCI and may be used to stratify patients prior to clinical trial inclusion. Further, the cyclogram with its quantifiable parameters may provide additional insights into gait control across different neuromotor disorders beyond those assessed by time-distance measures.

ACKNOWLEDGMENTS

We thank B. Huber for assistance with subject recruitment and kinematic data acquisition. Funding: this work was supported by the European Commission's Seventh Framework Program [CP-IP 258654, NEUWalk]; the Wolf Foundation, Switzerland; the Clinical Research Priority Program CRPP Neurorehab UZH.

REFERENCES

Abel, R., Schablowski, M., Rupp, R., and Gerner, H. J. (2002). Gait analysis on the treadmill – monitoring exercise in the treatment of paraplegia. *Spinal Cord* 40, 17–22. doi: 10.1038/sj.sc.3101239

Bachmann, L. C., Matis, A., Lindau, N. T., Felder, P., Gullo, M., and Schwab, M. E. (2013). Deep brain stimulation of the midbrain locomotor region improves paretic hindlimb function after spinal cord injury in rats. *Sci. Transl. Med.* 5, 208ra146. doi: 10.1126/scitranslmed.3005972

Barbeau, H., and Rossignol, S. (1987). Recovery of locomotion after chronic spinalization in the adult cat. *Brain Res.* 412, 84–95. doi: 10.1016/0006-8993(87)91442-9

Borghese, N. A., Bianchi, L., and Lacquaniti, F. (1996). Kinematic determinants of human locomotion. *J. Physiol.* 494 (Pt 3), 863–879.

Cotes, J. E., and Meade, F. (1960). The energy expenditure and mechanical energy demand in walking. *Ergonomics* 3, 97–119. doi: 10.1080/00140136008930473

Courtemanche, R., Teasdale, N., Boucher, P., Fleury, M., Lajoie, Y., and Bard, C. (1996). Gait problems in diabetic neuropathic patients. *Arch. Phys. Med. Rehabil.* 77, 849–855. doi: 10.1016/S0003-9993(96)90269-5

Dobkin, B., Apple, D., Barbeau, H., Basso, M., Behrman, A., Deforge, D., et al. (2006). Weight-supported treadmill vs over-ground training for walking after acute incomplete SCI. *Neurology* 66, 484–493. doi: 10.1212/01.wnl.0000202600.72018.39

Duysens, J., and Van de Crommert, H. W. (1998). Neural control of locomotion; the central pattern generator from cats to humans. *Gait Posture* 7, 131–141. doi: 10.1016/S0966-6362(97)00042-8

Field-Fote, E. C., Fluet, G. G., Schafer, S. D., Schneider, E. M., Smith, R., Downey, P. A., et al. (2001). The spinal cord injury functional ambulation inventory (SCI-FAI). *J. Rehabil. Med.* 33, 177–181. doi: 10.1080/16501970175030064 5

Field-Fote, E. C., and Tepavac, D. (2002). Improved intralimb coordination in people with incomplete spinal cord injury following training with body weight support and electrical stimulation. *Phys. Ther.* 82, 707–715.

Gil-Agudo, A., Perez-Nombela, S., Forner-Cordero, A., Perez-Rizo, E., Crespo-Ruiz, B., and Del Ama-Espinosa, A. (2011). Gait kinematic analysis in patients with a mild form of central cord syndrome. *J. Neuroeng. Rehabil.* 8, 7. doi: 10.1186/1743-0003-8-7

Grasso, R., Bianchi, L., and Lacquaniti, F. (1998). Motor patterns for human gait: backward versus forward locomotion. *J. Neurophysiol.* 80, 1868–1885.

Ivanenko, Y. P., Grasso, R., Zago, M., Molinari, M., Scivoletto, G., Castellano, V., et al. (2003). Temporal components of the motor patterns expressed by the human spinal cord reflect foot kinematics. *J. Neurophysiol.* 90, 3555–3565. doi: 10.1152/jn.00223.2003

Krawetz, P., and Nance, P. (1996). Gait analysis of spinal cord injured subjects: effects of injury level and spasticity. *Arch. Phys. Med. Rehabil.* 77, 635–638. doi: 10.1016/S0003-9993(96)90000-3

Kuhn, R. A. (1950). Functional capacity of the isolated human spinal cord. *Brain.* 73, 1–51. doi: 10.1093/brain/73.1.1

Lacquaniti, F., Grasso, R., and Zago, M. (1999). Motor patterns in walking. *News Physiol. Sci.* 14, 168–174.

MacLellan, M. J., Ivanenko, Y. P., Catavitello, G., La Scaleia, V., and Lacquaniti, F. (2013). Coupling of upper and lower limb pattern generators during human crawling at different arm/leg speed combinations. *Exp. Brain Res.* 225, 217–225. doi: 10.1007/s00221-012-3364-3365

Manor, B., and Li, L. (2009). Characteristics of functional gait among people with and without peripheral neuropathy. *Gait Posture* 30, 253–256. doi: 10.1016/j.gaitpost.2009.04.011

Morris, M., Iansek, R., Matyas, T., and Summers, J. (1998). Abnormalities in the stride length-cadence relation in parkinsonian gait. *Mov. Disord.* 13, 61–69. doi: 10.1002/mds.870130115

Morris, M., Iansek, R., Mcginley, J., Matyas, T., and Huxham, F. (2005). Three-dimensional gait biomechanics in Parkinson's disease: evidence for a centrally mediated amplitude regulation disorder. *Mov. Disord.* 20, 40–50. doi: 10.1002/mds.20278

Nakajima, K., Maier, M. A., Kirkwood, P. A., and Lemon, R. N. (2000). Striking differences in transmission of corticospinal excitation to upper limb motoneurons in two primate species. *J. Neurophysiol.* 84, 698–709.

Nooijen, C. F., Ter Hoeve, N., and Field-Fote, E. C. (2009). Gait quality is improved by locomotor training in individuals with SCI regardless of training approach. *J. Neuroeng. Rehabil.* 6, 36. doi: 10.1186/1743-0003-6-36

Pepin, A., Norman, K. E., and Barbeau, H. (2003). Treadmill walking in incomplete spinal-cord-injured subjects: 1. Adaptation to changes in speed. *Spinal Cord* 41, 257–270. doi: 10.1038/sj.sc.3101452

Robinson, J. L., and Smidt, G. L. (1981). Quantitative gait evaluation in the clinic. *Phys. Ther.* 61, 351–353.

Sainburg, R. L., Poizner, H., and Ghez, C. (1993). Loss of proprioception produces deficits in interjoint coordination. *J. Neurophysiol.* 70, 2136–2147.

Tamburella, F., Scivoletto, G., and Molinari, M. (2013). Balance training improves static stability and gait in chronic incomplete spinal cord injury subjects: a pilot study. *Eur. J. Phys. Rehabil. Med.* 49, 353–364.

Winter, D. A. (1992). Foot trajectory in human gait: a precise and multifactorial motor control task. *Phys. Ther.* 72, 45–53; discussion 54–46.

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.

Received: 15 January 2014; paper pending published: 04 February 2014; accepted: 27 February 2014; published online: 17 March 2014.

Citation: Awai L and Curt A (2014) Intralimb coordination as a sensitive indicator of motor-control impairment after spinal cord injury. Front. Hum. Neurosci. ***8****:148. doi: 10.3389/fnhum.2014.00148*

This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 15 May 2014
doi: 10.3389/fnhum.2014.00305

Spinal motor outputs during step-to-step transitions of diverse human gaits

Valentina La Scaleia[1,2,3], Yuri P. Ivanenko[3], Karl E. Zelik[3] and Francesco Lacquaniti[1,2,3]*

[1] *Department of Systems Medicine, University of Rome Tor Vergata, Rome, Italy*
[2] *Centre of Space Bio-medicine, University of Rome Tor Vergata, Rome, Italy*
[3] *Laboratory of Neuromotor Physiology, Santa Lucia Foundation, Rome, Italy*

Edited by:
Leonardo Gizzi, Pain Clinic Center for Anesthesiology, Emergency and Intensive Care Medicine University Hospital Göttingen, Germany

Reviewed by:
Massimo Sartori, Medical University Göttingen, Germany
Anderson Souza Oliveira, Aalborg University, Denmark

****Correspondence:***
Francesco Lacquaniti, Department of Systems Medicine, University of Rome Tor Vergata, IRCCS Santa Lucia Foundation, Via Ardeatina 306, 00178 Roma, Italy
e-mail: lacquaniti@med.uniroma2.it

Aspects of human motor control can be inferred from the coordination of muscles during movement. For instance, by combining multimuscle electromyographic (EMG) recordings with human neuroanatomy, it is possible to estimate alpha-motoneuron (MN) pool activations along the spinal cord. It has previously been shown that the spinal motor output fluctuates with the body's center-of-mass motion, with bursts of activity around foot-strike and foot lift-off during walking. However, it is not known whether these MN bursts are generalizable to other ambulation tasks, nor is it clear if the spatial locus of the activity (along the rostrocaudal axis of the spinal cord) is fixed or variable. Here we sought to address these questions by investigating the spatiotemporal characteristics of the spinal motor output during various tasks: walking forward, backward, tiptoe and uphill. We reconstructed spinal maps from 26 leg muscle EMGs, including some intrinsic foot muscles. We discovered that the various walking tasks shared qualitative similarities in their temporal spinal activation profiles, exhibiting peaks around foot-strike and foot-lift. However, we also observed differences in the segmental level and intensity of spinal activations, particularly following foot-strike. For example, forward level-ground walking exhibited a mean motor output roughly 2 times lower than the other gaits. Finally, we found that the reconstruction of the spinal motor output from multimuscle EMG recordings was relatively insensitive to the subset of muscles analyzed. In summary, our results suggested temporal similarities, but spatial differences in the segmental spinal motor outputs during the step-to-step transitions of disparate walking behaviors.

Keywords: EMG, motoneurons, spinal mapping, tiptoe, uphill, backward, walking

INTRODUCTION

Muscle activity during human locomotion is coordinated by tens of thousands of alpha-motoneurons (MNs), organized along the spinal cord (Romanes, 1951; Sharrard, 1964; Tomlinson and Irving, 1977). Bio-imaging techniques are being developed to increase our understanding of this spinal neural function, but generally these techniques (e.g., functional magnetic resonance imaging) remain difficult or impossible to use for studying the spinal cord during walking (Harel and Strittmatter, 2008; Stroman et al., 2014). Furthermore, most available techniques do not distinguish between activation of sensory neurons and that of MNs in the spinal cord. However, mapping muscle activations onto the rostrocaudal location of MN-pools in the human spinal cord provides a compact representation of the total motor output (Yakovenko et al., 2002; Ivanenko et al., 2006; O'Donovan et al., 2008; Monaco et al., 2010; Warp et al., 2012). This mapping also provides a complementary perspective to conventional approaches to understanding neural control, which often rely on detailed analyses of individual muscle activity and inter-muscular coordination (e.g., D'Avella and Bizzi, 2005; Ting, 2007; Giszter et al., 2010; D'Avella et al., 2013).

In previous studies, the spinal mapping method was used to investigate development and aging (Monaco et al., 2010; Ivanenko et al., 2013b), as well as the relationship between the spatiotemporal organization of the spinal motor output and the biomechanics of human locomotion (Ivanenko et al., 2008; Cappellini et al., 2010; MacLellan et al., 2012). In particular, Cappellini et al. (2010) found that, during both forward and backward walking on level ground, the spatial activity of the spinal cord fluctuated with the center-of-body-mass (COM) motion, with bursts of activity around touchdown and foot lift-off. However, it is not known whether these bursts of activity around touchdown and toe-off are generalizable to other gaits nor is it clear if the spatial location of the activity (along the rostrocaudal axis of the spinal cord) is fixed or variable for different gaits. A better understanding of

Abbreviations: AbdDM, abductor digiti minimi; AddL, adductor longus; AddM, adductor magnus; BFL, biceps femoris, long head; BFS, biceps femoris, short head; BW, backward walking; EDB, extensor digitorum brevis; EHB, extensor hallucis brevis; EHL, extensor hallucis longus; EMG, electromyography; ES, erector spinae (at L2 level); FDB, flexor digitorum brevis; FDHL, flexor digitorum/hallucis longus; FW, forward walking; Gmax, gluteus maximus; Gmed, gluteus medius; Ilio, iliopsoas; LG, gastrocnemius lateralis; MC, maximum contraction; MG, gastrocnemius medialis; MN, motoneuron; PerL, peroneus longus; PerS, peroneus brevis; RF, rectus femoris; Sart, sartorius; SD, standard deviation; Semit, semitendinosus; Sol, soleus; TA, tibialis anterior; TFL, tensor fasciae latae; Vlat, vastus lateralis; Vmed, vastus medialis; GT, greater trochanter; LE, lateral femur epicondyle; LM, lateral malleolus; HE, heel; 5MP, fifth metatarsophalangeal joint.

spinal motor outputs during different locomotion modes may provide further insights into adaptability and modularity of neural control (Lacquaniti et al., 2013; Bagnall and McLean, 2014), interspecies comparison (Carlson-Kuhta et al., 1998; Yakovenko et al., 2002), and may thus also have important clinical implications (Grasso et al., 2004; Scivoletto et al., 2007; Coscia et al., 2011; Oetgen and Peden, 2012; Hoogkamer et al., 2014).

Thus, the purpose of this study was to investigate these questions about motor output during several different locomotor tasks: forward, backward and digitigrade (tiptoe) walking on level ground, and walking on an inclined surface. These tasks may also be relevant to clinical, rehabilitation or sport applications. For instance, toe walking is observed in patients with various neurologic and developmental abnormalities (Oetgen and Peden, 2012), backward locomotion is used increasingly in sports and rehabilitation (Hoogkamer et al., 2014) and uphill walking may be appropriate exercise for obese individuals at risk for musculoskeletal pathology or pain (Haight et al., 2014). We used the recordings from 26 leg muscles (including intrinsic foot muscles that have not typically been considered) to reconstruct spinal motor outputs with specific interest in identifying common and idiosyncratic features across locomotor gaits.

MATERIALS AND METHODS

EXPERIMENTAL PROTOCOL

We recorded surface electromyograms (EMGs) and foot motion for 8 subjects (4 males, 4 females, 25.6 ± 2.6 years old, 1.78 ± 0.11 m, 76 ± 16 kg) during 4 ambulation tasks: walking forward, backward, tiptoe and uphill (20% inclined grade), all at 4 km/hr. These tasks were selected to represent biomechanically distinct walking gaits that were cyclic (for EMG analysis purposes) and could be performed at fixed speed on a treadmill. The treadmill speed was selected because it was sufficiently fast to distinguish myoelectric activity from the baseline noise (at slower speeds some muscle EMGs were small and therefore difficult to quantify), but also slow enough that most subjects could perform all the tasks. However, two out of the eight subjects were not able to walk backward at 4 km/hr. Each walking trial lasted 40 s and was performed barefoot on a standard treadmill. Prior to data collection, the subjects were trained on each task, allowing them time to acclimate to the various walking conditions, and all subjects gave informed consent prior to participation. The protocol was approved by the Ethics Committee of the Santa Lucia Institute.

We also collected several additional trials to help identify maximum contraction (MC) magnitude for each muscle EMG. Before collecting the walking data, we asked subjects to perform a set of quasi-static maneuvers against manual resistance. These included: flexing/extending/abducting the toes, plantarflexing/dorsiflexing/inverting/everting the ankle, flexing/extending the knee, flexing/extending/abducting/adducting the hip and flexing the back. Each exercise was performed for 5 s, during which subjects were instructed to perform maximal contractions. Two additional forward walking trials (3 and 5 km/hr) were also recorded and used to help determine MC values.

DATA COLLECTION

We recorded kinematic data bilaterally at 100 Hz using a Vicon-612 system (Vicon, Oxford, UK) with nine cameras. Infrared reflective markers (diameter 14 mm) were place bilaterally over the following landmarks: greater trochanter (GT), lateral femural epicondyle (LE), lateral malleolus (LM), heel (HE), and fifth metatarsophalangeal joint (5MP).

We recorded EMG activity by means of surface electrodes from 26 muscles simultaneously on the right side of each subject. These included one muscle from the lower back (erector spinae (ES) at L2 level), two muscles from the buttocks [gluteus maximus (Gmax) and gluteus medius (Gmed)], 11 muscles from the thigh [iliopsoas (Ilio), tensor fasciae latae (TFL), sartorius (Sart), adductor magnus (AddM), adductor longus (AddL), vastus medialis (Vmed), vastus lateralis (Vlat), rectus femoris (RF), biceps femoris long head (BFL), biceps femoris short head (BFS), semitendinosus (Semit)], six muscles from the shank [tibialis anterior (TA), peroneus longus (PerL), peroneus brevis (PerS), medial gastrocnemius (MG), lateral gastrocnemius (LG), soleus (Sol)], and six muscles from the foot [extensor hallucis longus (EHL), flexor digitorum/hallucis longus (FDHL), extensor hallucis brevis (EHB), extensor digitorum brevis (EDB), abductor digit minimi (AbdDM), flexor digitorum brevis (FDB)]. The activations of flexor digitorum longus and flexor hallucis longus were indistinguishable in our surface EMG recordings, due to close proximity of the muscles, and thus are reported together. We placed EMG electrodes based on suggestions from SENIAM (seniam.org), the European project on surface EMG. To this end, we located the muscle bellies by means of palpation and oriented the electrodes along the main direction of the fibers (Winter, 1991; Kendall et al., 2005). The placement of EMG electrode for muscles in the foot and shank segments is illustrated in **Figure 1** for convenience since some of the foot muscles are less commonly recorded in literature.

All EMGs were recorded at 4000 Hz using a Delsys Trigno Wireless System (Boston, MA), except the flexor digitorum brevis which was recorded using a synchronized Delsys Bagnoli System (at 1000 Hz). Due to the recording site of flexor digitorum brevis (on the plantar surface of the foot), the lower-profile Bagnoli electrode was needed. Some electrodes became partially or fully detached during testing, and signals were thus not usable. These EMGs were removed on a subject-specific basis. On average we analyzed 23.4 ± 1.7 muscle EMG from each subject.

DATA PROCESSING

The beginning of the gait cycle (foot-strike) was defined based on kinematic events. We used vertical height of the right HE marker for forward walking and limb elevation angle (based on maximum GT-5MP virtual segment displacement) for backward, tiptoe and inclined walking. Similarly, for stance to swing transition (foot-lift) we used limb elevation angle (based on minimum GT-5MP virtual segment displacement) for forward, tiptoe and inclined walking, and minimum vertical height of the HE marker for backward walking. The usage of these criteria was based on the different kinematic endpoint (foot) behaviors for the various gaits (Ivanenko et al., 2007). While the differences in definition of gait cycle initiation may have introduced minor time shifts between

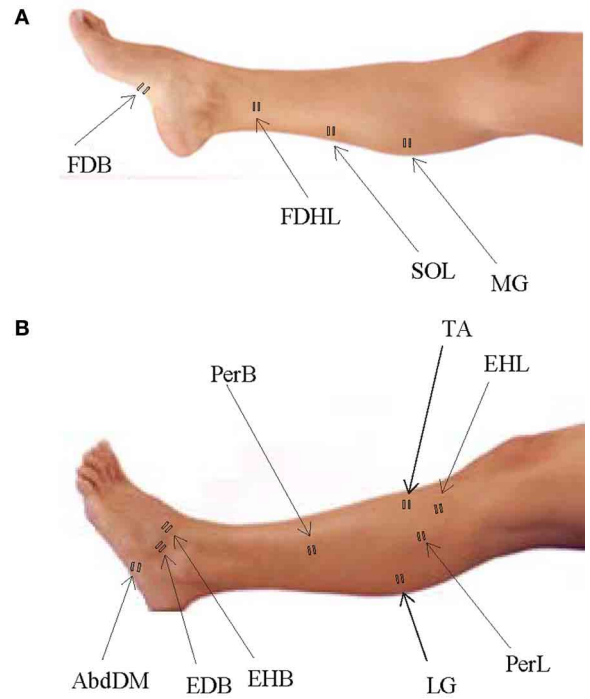


FIGURE 1 | Sites of EMG electrode placement for muscles in the foot and shank segments. (A) Plantar surface of the foot and medial aspect of the leg with sites of electrode placement. **(B)** Dorsal surface of the foot and lateral aspect of the leg with sites of electrode placement.

tasks, all muscle EMGs for a single task shift together in time and so for each individual gait this did not impact the fidelity of spinal map reconstruction. General gait parameters [cycle duration, anterior-posterior foot (5MP) excursion] and joint (ankle, knee and hip) angular range of motion were calculated to characterize the kinematics of gaits studied (**Figure 2**).

We processed EMG data using standard filtering and rectifying methods. We applied a 30 Hz high-pass filter, then rectified the EMG signals and applied a 10 Hz low-pass filter (all filters, zero-lag 4th order Butterworth). To reduce residual baseline noise, which appear as offsets in the EMG envelopes, we subtracted the minimum signal from each EMG. This assumes that at some point during walking each muscle is effectively "off" (not actively contracting). Some subjects exhibited artifacts in the foot muscles, generally linked to foot-strike and foot-lift events. In order to remove these artifacts, high-pass filtering of these muscles was performed using a 150 Hz cut-off frequency (rather than 30 Hz). A prior study on cut-off frequency (Potvin and Brown, 2004) and informal tests on locomotor EMGs (Zelik et al., 2014) confirmed that this artifact-removal filter had minimal effect on the shape of the muscle activation pattern. For illustrative purposes, the EMGs filtered at higher cut-off frequency were then rescaled to match peak amplitude of the 30 Hz filtered signal (**Figures 3**, **4**). However, this rescaling procedure did not affect the calculation of the motor output since EMGs were eventually normalized to their MC amplitudes before mapping to the spinal cord (detailed below).

We divided EMGs into gait cycles based on foot kinematics, then interpolated each stride to 200 time points, and finally averaged across gait cycles (individually for each subject and task). This yielded an ($m \times 200$) EMG matrix for each task, where m equaled the number of muscles analyzed. Inter-subject mean (and standard deviation) values for EMG were then computed from these subject-specific data. In addition to calculating the ensemble-averaged EMGs (across strides and subjects), we also present some EMG waveforms of individual strides in order to examine inter-stride variability in the spinal motor output.

EMG NORMALIZATION

We normalized EMGs by the MC magnitude across all trials. Normalization was performed to account for the differences in μV magnitudes recorded between muscles. We defined MC magnitude as the muscle's maximum EMG signal from either dynamic (walking) or quasi-static trials (during which subjects were instructed to perform maximal contractions against manual resistance, see *Experimental Protocol*). Thus, all EMGs were considered on a scale from 0 to 1, where 0 indicates that a muscle is inactive and 1 represents maximum muscle activation. Across all quasi-static trials (EMGs were low-pass filtered as described previously), we looked at a sliding 1-s window (by incrementally shifting each time step) and computed the average EMG during each. The highest average EMG found during any 1-s window was defined as the maximum quasi-static activation magnitude. Similarly, maximum dynamic activation magnitude was defined for each muscle as the peak stride-averaged EMG across all walking tasks. The normalization constant for each muscle was then defined as the larger of the quasi-static and dynamic activation magnitudes.

We note that normalization to muscle physiological cross sectional area (PCSA) was not used in this study for reconstructing the segmental spinal outputs, which has occasionally been done in the past (e.g., MacLellan et al., 2012; Ivanenko et al., 2013b). This is because the number of motor units for each muscle is not related to PCSA in a simple way (e.g., number of motor units does not scale proportionally with size of muscle; Feinstein et al., 1955; Christensen, 1959; McComas et al., 1997; McComas, 1998).

MOTOR OUTPUT CALCULATIONS

To characterize the spinal motor output, EMG-activity was mapped onto the estimated rostrocaudal location of MN-pools in the human spinal cord from L2 and S2 segments. Because this method has been thoroughly documented in previous papers (Ivanenko et al., 2006, 2013b; MacLellan et al., 2012), we describe it only briefly here. The maps were constructed by adding up the contributions of each muscle to the total activity at each spinal segment, using the myotomal charts of Kendall et al. (2005) to link muscles to their spinal innervation levels (see **Figure S1**). The motor output pattern of each spinal segment S_j was estimated by the following equation:

$$S_j = \frac{\sum_{i=1}^{m_j}\left(\frac{k_{ji}}{n_i} \times EMG_i\right)}{\sum_{i=1}^{m_j}\left(\frac{k_{ji}}{n_i}\right)} \times MN_j \qquad (1)$$

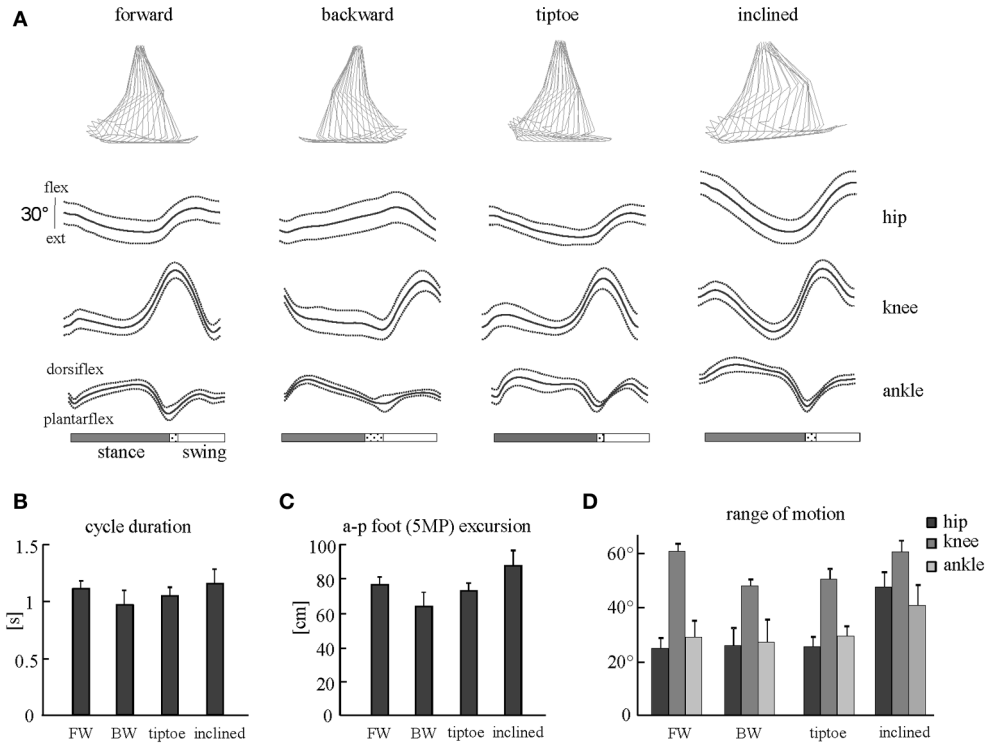


FIGURE 2 | Kinematic patterns during forward, backward, tiptoe and uphill (20% inclined) walking at 4 km/hr. (A) Ensemble averages (±SD) of hip, knee, and ankle joint angles of the right leg. Hip and knee angles increase in flexion, ankle angle in dorsi-flexion. The dotted region between stance and swing phases depicts inter-subject standard deviation (SD), and is centered at average foot-lift. *On the top*: stick diagrams for a single stride in one representative subject. **(B)** Cycle duration (mean +SD) for different gaits. **(C)** Anterior-posterior foot (5MP marker) excursion. **(D)** Peak-to-peak amplitudes (+SD) of angular motion.

where EMG_i represents the normalized, subject-specific envelope of muscle activity, k_{ji} is a weighting coefficient for the i-th muscle (to signify if the j-th spinal level is a major, $k_{ji} = 1$, or minor, $k_{ji} = 0.5$, MN source, see **Figure S1**), m_j is the number of muscles innervated by the j-th spinal segment, and n_i is the total number of spinal levels that innervate the i-th muscle, again accounting for major and minor sources (for instance, for the soleus muscle, $n_i = 1 + 1 + 0.5 = 2.5$, see **Figure S1**). Thus, the fractional part of Equation 1 can range in value from 0 (inactive) to 1 (maximum activation of that spinal segment). To account for size differences in MN pools at each spinal level, this fractional activity value was then multiplied by the segment-specific number of MNs (MN_j). This MN pool size normalization primarily affects the boundary segments L2 and S2, which contain 2–3 times fewer MNs than the other segments (**Table S1**, Tomlinson and Irving, 1977). We note that Equation 1 is slightly modified with respect to our previous studies (Grasso et al., 2004; Ivanenko et al., 2006, 2013b) in order to better account for the different number of muscles that innervate each spinal segment and the heterogeneity in the MN pools along the lumbosacral enlargement. Thus, our updated calculation yields spinal motor output in units of number of (active) MNs.

The primary assumptions implicit in this analysis are that (1) the rectified EMG provides an indirect measure of the net firing rate of MNs for each muscle (Yakovenko et al., 2002), and (2) the set of recorded muscles is representative of the total motor output from each spinal segment. The first assumption seems reasonable given that mean EMG has been found to increase linearly with the net motor unit firing rate (Hoffer et al., 1987; Day and Hulliger, 2001). However, a limitation is that this method does not account for confounds due to other physiological properties, such as the effects of muscle length or velocity on the EMG signals. To test the second assumption, we compared the activation maps obtained from all 26 recorded muscles with those obtained from reduced subsets of muscles (detailed in *Muscle subset analysis* section below).

To obtain the averaged (across subjects) spinal maps, we calculated the spinal motor output for each subject based on stride-averaged EMGs, and then we averaged it across subjects. We computed two summary metrics to describe the spinal maps: *mean segmental output* and *mean temporal output*. For each condition, we averaged the motor output patterns over the entire gait cycle to find the subject-specific *mean segmental output* and then averaged it across subjects to obtain mean ± SD. Similarly, we averaged the motor output across the spinal segments L2 to S2 to find the *mean temporal output* across the gait cycle. From this mean temporal output waveform, we found the maximum peak in the first half of the stance phase and defined it as activation *burst 1*, and the peak in the second half of the

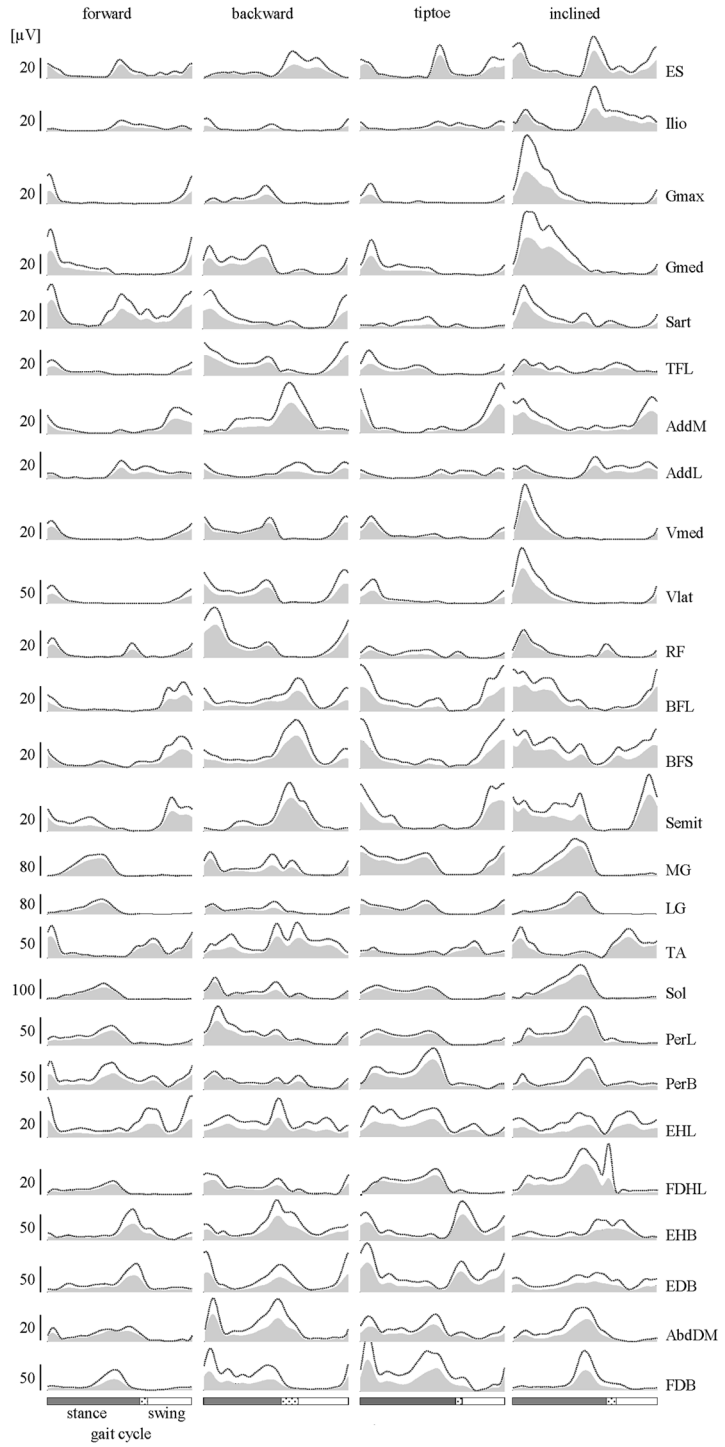


FIGURE 3 | EMGs. Subject-averaged patterns of muscle activity during forward, backward, tiptoe and uphill (20% inclined) walking at 4 km/hr. Mean EMGs and inter-subject standard deviations are plotted across a normalized gait cycle from foot-strike to ipsilateral foot-strike. The dotted region between stance and swing phases depicts inter-subject standard deviation (SD), and is centered at average foot-lift.

gait cycle as *burst 2*. To characterize the total intensity of the spinal output for each task, we computed for each subject the *mean motor output* by averaging across both spatial segments and gait cycle, and then we averaged it across subjects. In addition to creating subject-specific spinal maps from stride-averaged EMG envelopes, we also computed maps for individual strides and compared them with those obtain from ensemble-averaged strides.

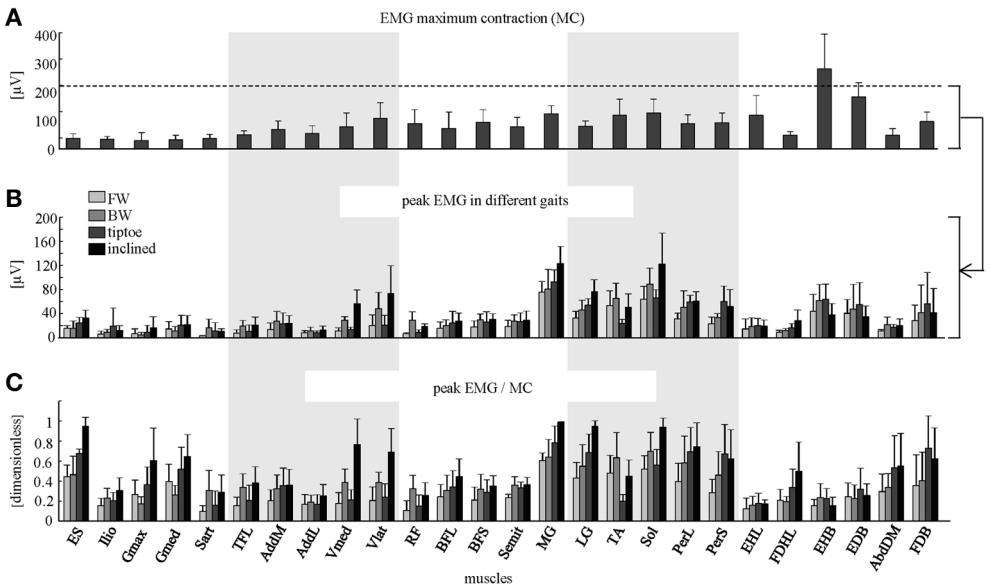


FIGURE 4 | Peak EMG magnitudes. (A) Maximum contraction (MC) values (+SD) are shown (based on peak EMGs from dynamic and quasi-static trials; see Methods for full details). Peak EMG amplitudes (+SD) are depicted in μV **(B)** and as a percentage of MC **(C)** for forward, backward, tiptoe and inclined walking at 4 km/hr.

MUSCLE SUBSET ANALYSIS

Practical considerations limit the number of muscles from which we could record. Thus, there is the potential issue of how the specific selection of the muscles affects the resulting spatiotemporal maps of MN activity. To evaluate the sensitivity of the spinal maps approach we compared the motor outputs obtained from analyzing all 26 muscles with those obtained from subsets of these muscles. Subsets were chosen as follows: (1) the 20 non-foot muscles (TA, Sol, MG, LG, RF, Vmed, Vlat, AddL, AddM, ES, TFL, PerL, PerB, BFL, BFS, Semit, Sart, Ilio, Gmax, and Gmed) and (2) 12 commonly recorded muscles (TA, Sol, MG, LG, RF, Vmed, Vlat, ES, TFL, BFL, Semit, and Gmax). For forward walking we also made 26 additional comparisons by correlating maps from each unique set of 25 muscles (i.e., by systematically eliminating each individual muscle) with the map constructed from all muscles. The correlation coefficient (r) was calculated for each subject and condition. Averaged correlation coefficients were then reported for each comparison.

STATISTICS

To compare activation waveforms we computed linear correlations (*r*-values). For instance, to compare segmental activations of individual subjects with those of averaged maps, correlation coefficient was computed for each subject and each segment, and then they were averaged first across subjects for each segment and then across segments. Similarly, to compare the maps obtained by different sets of muscles, correlation coefficient was computed for each segment, and then the data for all segments were averaged. Since correlation coefficients have non-normal distributions, their mean estimates were computed based on the normally distributed, Z-transformed values.

Repeated measures (RM) ANOVA was used to evaluate differences in the kinematics and the *mean motor output* across different gaits, and *post-hoc* Tukey's HSD test was used to determine statistical significance. Since only six out of the eight subjects were able to walk backward at 4 km/hr their missing data for this condition for the ANOVA were replaced by the unweighted mean value estimated from all other subjects. Reported results are considered significant for $p < 0.05$.

RESULTS

KINEMATICS

General gait parameters and ensemble-averaged joint angular movements are reported in **Figure 2**. We observed that cycle duration and foot excursion were slightly but significantly lower for backward walking than for forward and inclined walking ($p < 0.006$, **Figures 2B,C**). These two parameters were also larger for inclined walking relative to tip-toe walking ($p < 0.03$). The range of hip and ankle angular motion was significantly larger during inclined walking than for the other tasks ($p < 0.001$, **Figure 2D**). The peak-to-peak amplitude of the knee joint oscillations was significantly smaller for backward and tip-toe walking than for forward and inclined walking ($p < 0.0002$, **Figure 2D**).

EMG

Lower-limb EMGs (**Figure 3**) were qualitatively consistent with those reported elsewhere in the literature for forward (Winter, 1991; Ivanenko et al., 2006), backward (Thorstensson, 1986; Grasso et al., 1998; Ivanenko et al., 2008), tiptoe (Perry et al., 2003; Romkes and Brunner, 2007) and inclined walking (Lange et al., 1996; Franz and Kram, 2012). In this study we extended the number of recorded muscles relative to our previous studies (Ivanenko et al., 2006, 2013b). In particular, we included intrinsic foot

muscles, which demonstrated their own unique activation patterns with bursts principally around the stance to swing transition of gait (**Figure 3**).

Averaged EMG waveforms for the deeply located and interconnected muscles during forward walking were consistent with those reported in the literature. The deep hip flexors (Ilio) demonstrated the major peak of activity around lift-off (Rab, 1994; Andersson et al., 1997; Ivanenko et al., 2008). EMG recordings of AddL and AddM showed main bursts at foot lift-off and during swing, respectively (Winter, 1991). The activity of BFL and BFS (at the end of swing and beginning of stance) was similar to that reported by University of California Berkeley (1953). Intramuscular recordings of foot muscle activity (Gersten et al., 1956; Mann and Inman, 1964) showed a good correspondence with our data (**Figure 3**). Specifically, EHL activity showed two peaks around foot lift-off and heel strike, respectively, while the FDHL showed activity beginning in early stance and continuing until the foot lift-off (Gersten et al., 1956). The EDB and AbdDM became active ∼20% of the cycle and the FDB at 40% of the cycle, remaining active until just before foot lift-off (Mann and Inman, 1964).

The amplitude of EMG signals (in μV) varied considerably across muscles, both during walking and in terms of MC (**Figures 4A,B**). We found that normalizing to MC tended to increase the relative activation magnitude of proximal muscles (e.g., ES, Ilio, Gmax, Gmed, TFL, AddL, Sart, Vmed) and some intrinsic foot muscles (e.g., FDHL, AddDM) and thus their contribution to the spinal maps (**Figure 4C**).

AVERAGE SPINAL MAPS

We observed task-specific spinal motor outputs for each walking condition (**Figure 5**), although with qualitative similarities in temporal profile. In particular, two prominent periods of activity were observed in the mean temporal output of each task (**Figure 5**, bottom): the first following foot-strike (∼5–10% of the gait cycle) and the second preceding foot-lift (∼40–55%). However the timing of the second burst relative to foot-lift varied considerably between tasks, occurring later in tiptoe than forward walking, for example (**Figure 5**). In contrast to the qualitative temporal similarities, we found substantial differences in the spatial localization and intensity of spinal activation for each gait (**Figure 5**). In particular, we found that the mean motor output (spinal activation averaged across the entire gait cycle and all spinal segments) was significantly lower for forward walking than for the other tasks ($p < 0.01$, **Figure 6A**). We also found that the loci of mean segmental outputs shifted somewhat as a function of gait (**Figure 5**, see plots to the right of each spinal map). For instance, forward and tiptoe walking exhibited principle activations in L5 and S1, whereas backward and inclined walking showed a more distributed output with roughly similar intensities from L3 to S1. These differences in spatial level of spinal activation were even more evident during the major "spots" of activity (identified as burst 1 and 2 from the mean temporal output). During burst 1 (after foot-strike, **Figure 6B**) peak motor output was at the spinal level L3 for inclined walking, L4 for forward walking, L5 for tiptoe walking and S1 for backwards walking (although this gait exhibited a relatively constant intensity from L3-S1). Differences were less evident for burst 2 (**Figure 6C**), when most gaits exhibited peak motor outputs from spinal segments L5 and S1.

SUBJECT-SPECIFIC SPINAL MAPS

The major features observed in the average spinal maps were also present in subject-specific maps. In particular, 6 out of the 8 tested subjects exhibited bimodal (two peaked) motor output profiles for all gaits (**Figure 7**). The remaining 2 subjects also showed the bimodal temporal profile for most gaits except for forward walking (s8 subject, **Figure 7A**) and backward walking

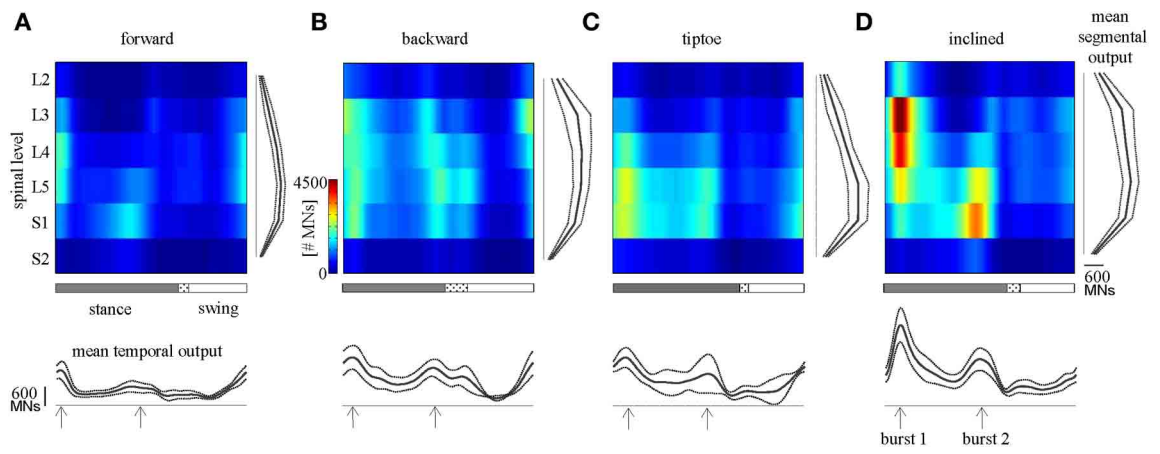


FIGURE 5 | Spinal maps. Depicted here are estimates of averaged (across subjects) spatiotemporal spinal motor outputs computed from EMGs for **(A)** forward, **(B)** backward, **(C)** tiptoe, and **(D)** uphill (20% inclined) walking, all at 4 km/hr. Motor output (reported in units of number of MNs) is plotted as a function of gait cycle and spinal segment level. Waveforms plotted below the maps correspond to the mean temporal output pattern averaged first across all 6 segments and then across subjects (mean ± SD, $n = 8$ subjects). Note the tendency for peaks to occur around early and late stance (labeled as burst 1 and 2). Curves to the right of maps represent the mean segmental output averaged first across the entire gait cycle and then across subjects. In the gait cycle, the dotted region between stance and swing phases depicts inter-subject standard deviation (centered at average foot-lift).

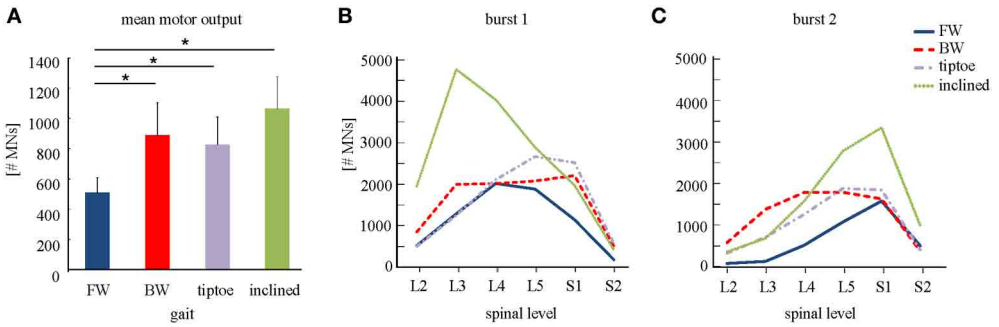


FIGURE 6 | Spinal motor output. (A) Depicted are mean (+SD) motor outputs (averaged across both gait cycle and spinal levels). Asterisks denote significant differences between conditions. Segment-specific magnitudes of motor output are also shown for **(B)** burst 1 of spinal activity (occurring after foot-strike; see **Figure 5**), and **(C)** burst 2 (occurring around foot-lift).

(s5 subject, **Figure 7B**). In these cases, mean temporal output was found to have an additional peak at the beginning of the swing phase. Individual subjects also exhibited small differences in the segmental level of spinal activation, particularly during load acceptance following foot-strike (**Figures 5**, **7**). Nevertheless we found a strong correlation between subject-specific and average maps (0.85 ± 0.13 depicted in **Figure 5**), consistent with previously published findings (Ivanenko et al., 2006).

EMG profiles exhibit stride-to-stride variability related to dynamic stability and walking speed maintenance (Hausdorff, 2007; Kang and Dingwell, 2009). Examples of the spinal maps of individual strides are illustrated in **Figure 8**. Despite individual variations in the segmental level of spinal activation, the major features depicted in the stride-averaged maps (**Figure 7**) are representative of the general trends in individual strides (**Figure 8**). We found the mean correlation coefficient between segmental output waveforms of individual and ensemble-averaged strides was 0.90 ± 0.04 (average from **Table 1**).

SENSITIVITY TO THE NUMBER OF MUSCLES ANALYZED

We found that the spinal maps were relatively insensitive to the subset of muscles analyzed. Spinal maps computed from 20 and 12 muscle subsets were strongly correlated with the maps computed from the full set of 26 recorded muscles, with average correlation coefficients between 0.98–0.99 and 0.91–0.96, for each task (**Table 2**). The motor outputs evaluated at each individual spinal segment were also found to be in good agreement, with r values (always greater than 0.9 using 20 muscles, and generally greater than 0.85 using 12 muscles). The only exception was that, with 12 muscles, the L5 segment correlation dropped to 0.74–0.90. Forward walking maps obtained by excluding a single recorded muscle were also highly correlated with those obtained from the full set of 26 muscles ($r = 0.99 \pm 0.01$).

DISCUSSION

The overall behavior of the body and limbs during walking is determined by the interplay of neural and mechanical factors. Here we observed that spinal motor outputs corresponded to the major phases of biomechanical force production during diverse walking tasks (Winter, 1991; Perry et al., 2003; DeVita et al., 2007; Franz and Kram, 2012). Specifically, the elevated MN outputs during the gait cycle produce muscle contractions during the step-to-step transition, in which both limbs act to redirect the body's velocity in a way that is thought to improve walking economy (Donelan et al., 2002). However, during the step-to-step transition, we observed differences in the loci of the segmental spinal activity across gaits (**Figures 5**, **6**). This suggests that even if similar biomechanical functions are performed by the limbs (i.e., redirection of the body during the transition), it may be accomplished differently, through a gait-specific coordination of muscles. Thus, high-level features of locomotion may be flexibly encoded by neural circuits to generate muscle activation patterns based on gait-specific constraints and feedback.

Various neural control strategies have been proposed for transforming such task-level goals to muscle-level execution, for example using a hierarchical, modular architecture under feedback control (Ting et al., 2012). The pulse-like features of the spinal motor output observed in this study may be consistent with "drive-pulse" rhythmic elements or neural primitives, which have been hypothesized to underlie the spinal circuitry of animals (Giszter et al., 2007). Although the precise neuronal substrates remain largely unknown (but see Hart and Giszter, 2010), it is believed that a crucial role is played by central pattern generators (Grillner, 1981). Specifically, it has been proposed that motor activation patterns may emerge from a multi-layered organization of the spinal neural networks with two functionally distinct levels, one for rhythm generation and the other for muscle pattern generation (McCrea and Rybak, 2008). In this study we found that the spatial loci of MN pool activations depends greatly on the walking task (**Figures 5**, **6**), indicating that "drive pulse" rhythmic elements may be significantly modulated by task-specific sensory feedback. Since muscle activation timing was linked to major force production events around foot touchdown and foot-lift, it suggests that pre-programmed motoneuronal drive may be principally mediated by afferent force and kinematic-related feedback (Duysens et al., 1998; Nielsen and Sinkjaer, 2002; Pearson, 2004). There is also supporting evidence from a previous study on cats that neuromotor coordination may be modulated by critical points that correspond to key biomechanical events (Saltiel and Rossignol, 2004).

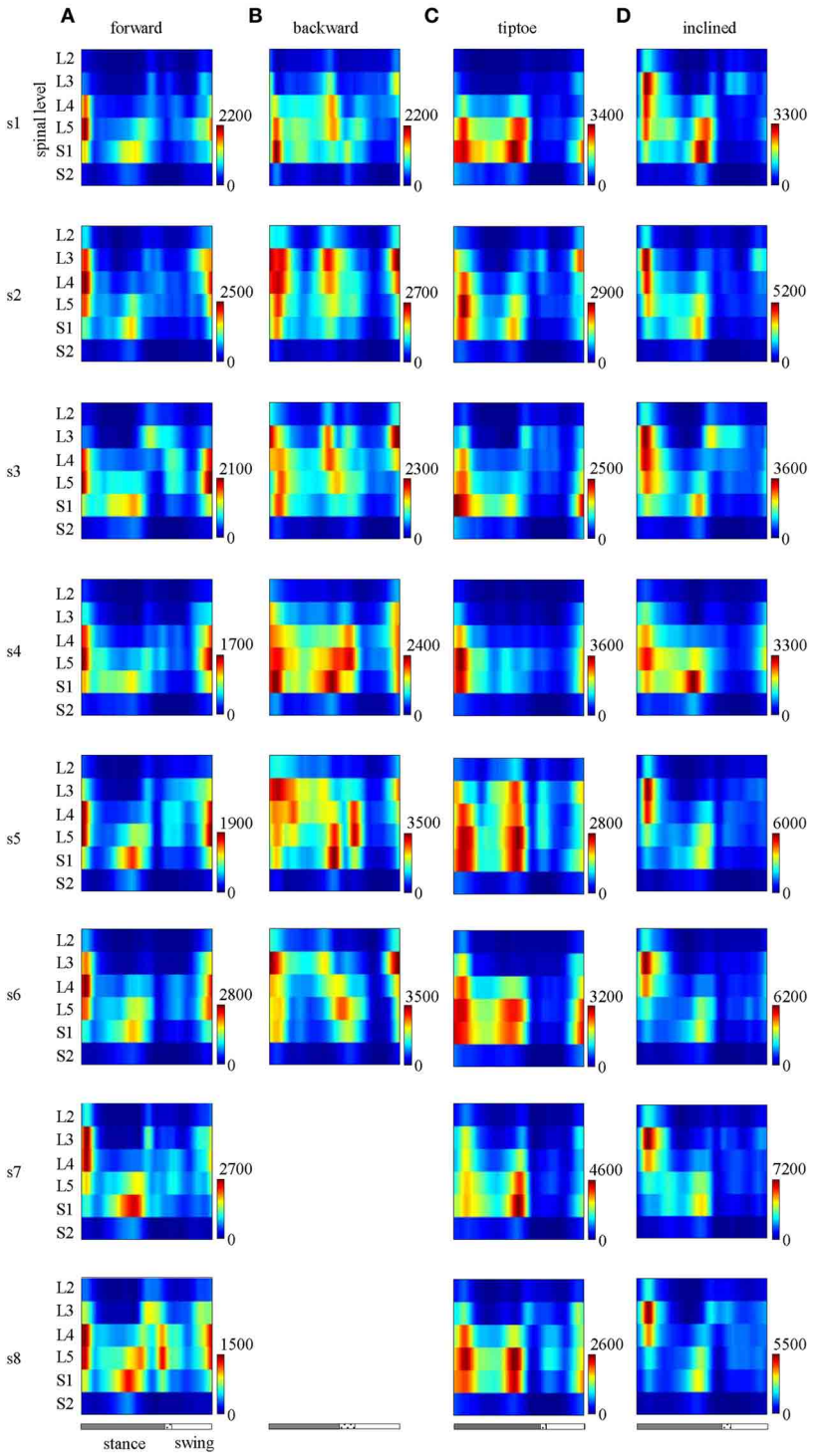


FIGURE 7 | Spinal maps of MN activity of the lumbosacral enlargement in all subjects for all gaits (A–D). Note similar temporal features (main peaks around foot-strike and foot-lift) of the segmental output between individual and averaged (**Figure 5**) spinal maps.

It is worth noting that we observed similarities in the spinal activation maps across subjects (**Figure 7**) and strides (**Figure 8**, **Table 1**), specifically in terms of temporal activation peaks around foot-strike and foot-lift. Thus the spinal mapping methodology seem to provide a robust and repeatable means to reconstruct MN pool activity. Meanwhile, previous literature has demonstrated that the spinal maps do vary for individuals with neuromotor impairments (Grasso et al., 2004; Coscia et al., 2011; Ivanenko

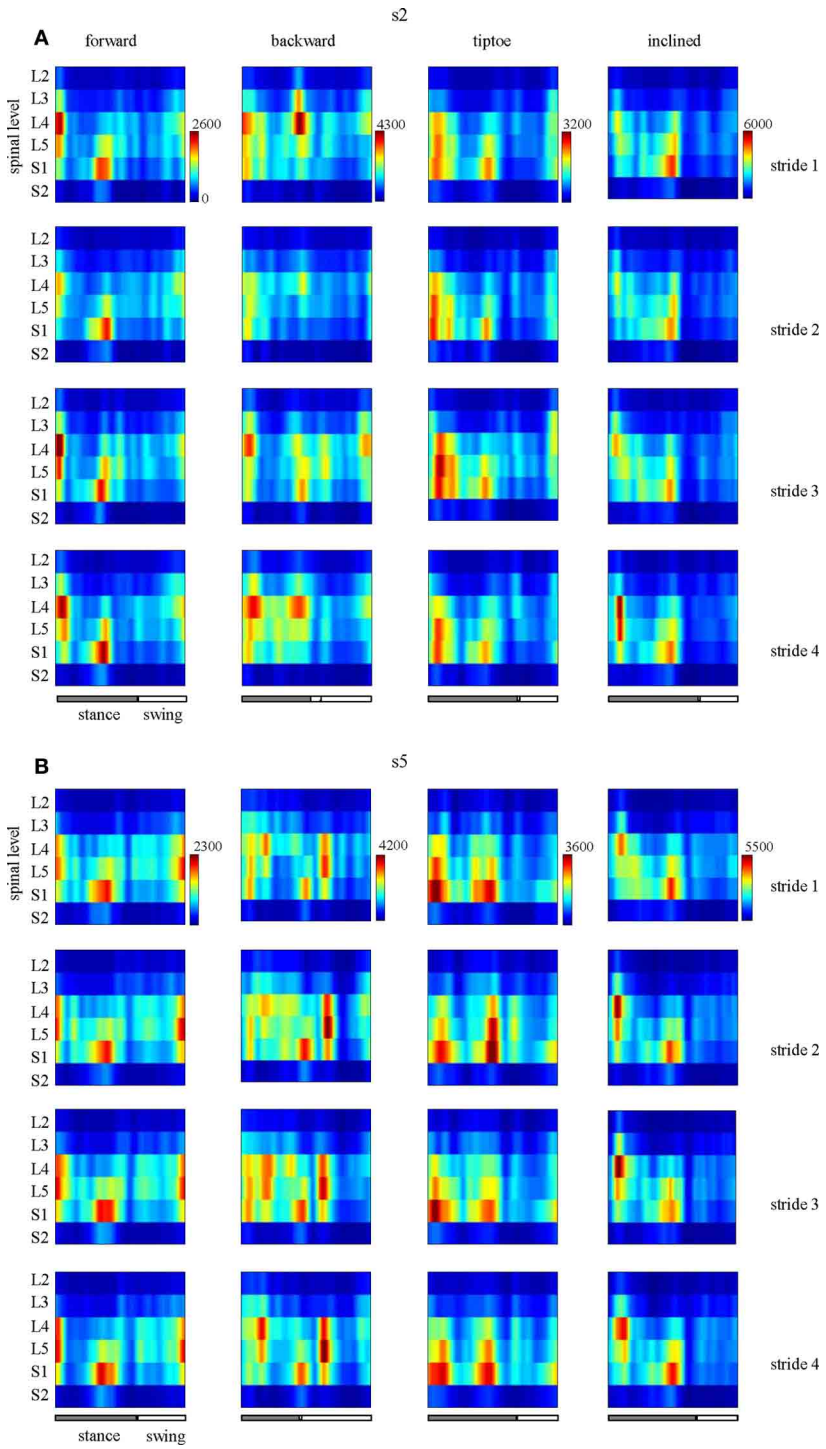


FIGURE 8 | Examples of spatiotemporal maps of MN activity of the lumbosacral enlargement in two subjects [s2 and s5, (A,B), respectively] for all gaits. For each individual, four individual strides are shown. Note similar spatiotemporal features (activity around foot-strike and foot-lift) of the segmental output between individual and averaged (**Figure 7**) strides for these subjects.

et al., 2013a) and throughout the aging process (Monaco et al., 2010) and during childhood development (Ivanenko et al., 2013b). Taken together, the robustness of the methodology and the population-specific activations suggest that spinal mapping approach may be useful for assessing or differentiating gait performance in clinical populations.

It is also interesting to compare spinal maps between plantigrade and digitgrade gaits. Human adults typically walk with a

characteristic heel-to-toe progression (plantigrade gait), whereas many animals walk only on their toes (digitgrade gait). In this study we observed roughly a doubling of the intensity of spinal motor output during tiptoe walking (**Figure 6A**), which is known to incur increased energetic costs compared to plantigrade gait (Cunningham et al., 2010). This increase in motor output was due, in part, to differences in the spinal activity after foot-strike, which was both increased in magnitude and spatially shifted toward more distal segments (L5/S1). The spinal maps for human tiptoe walking were, however, qualitatively different from maps constructed from digitigrade feline locomotion (Yakovenko et al., 2002). In cats, the primary MN activation during walking occurs during midstance and with roughly constant intensity, likely the result of neuromechanical differences associated with their flexed limb posture and quadrupedal gait. This comparison also highlights the potential utility of spinal maps for studying interspecies motor control.

There are several limitations to the spinal mapping approach, many of which have been previously documented (Cappellini et al., 2010). Briefly, the reconstruction and interpretation of spinal maps assume anatomical similarity of motor pools across individuals and that rectified EMG provides an indirect measure of the net MN firing rate (Yakovenko et al., 2002). Another potential concern is related to EMG cross-talk, which is always a potential issue with surface EMG recordings: in particular for deep muscles like Ilio that have a relativity small superficial region for recording and for smaller foot muscles (e.g., flexor digitorum longus) that are in close proximity to larger calf muscles. In the previous study (Ivanenko et al., 2013b), the cross-talk issue was addressed by modeling the potential effect of different levels of cross-talk in the EMG profiles. The spinal segmental output was reconstructed by adding up incrementally the magnitude of cross-talk from adjacent muscles (from 10 to 100%). While the intensity and the width of the main loci of activation could be affected by adding cross-talk, this procedure did not give rise to the appearance of new loci of activation or significant time shifts in the spinal maps. Given the similar spinal mapping methodology in this study, we do not expect that the similarities in spinal maps reconstructions (based on different set of muscles EMGs) were due to cross-talk. Furthermore, the spinal maps during walking have been shown to be similar when reconstructed from EMGs obtained using surface and intramuscular electrodes (Ivanenko et al., 2006). Consistent maps have also been produced in different studies (Ivanenko et al., 2008; Cappellini et al., 2010; Monaco et al., 2010; Coscia et al., 2011; MacLellan et al., 2012). We only tested four walking tasks (**Figure 5**), but other gaits may show additional (e.g., skipping) or temporally shifted (e.g., running) spots of activity specific for force production in those gaits (Ivanenko et al., 2008). Finally our analysis was also based on a limited set of muscles. However, we found the spinal maps to be relatively robust and insensitive to the subset of muscles analyzed (see Results, **Table 2**), presumably because the lumbosacral enlargement innervates numerous muscles and each muscle is innervated by several segments.

In summary, we found that the MN activation patterns exhibited two major bursts during diverse walking tasks, one around foot-strike and the other around foot-lift, but with differences in the segmental level and intensity of the spinal activity. We also found further evidence that spinal MN mapping provides a robust method for estimating spinal motor output, which is relatively insensitive to the subset of EMGs analyzed. We also suggest that spinal motor mapping can be used to assess the recruitment of

Table 1 | Inter-stride variability in the segmental spinal output for different gaits.

	FW	BW	Tiptoe	Inclined
L2	0.90 ± 0.05	0.85 ± 0.02	0.86 ± 0.06	0.94 ± 0.04
L3	0.90 ± 0.05	0.83 ± 0.02	0.87 ± 0.06	0.94 ± 0.04
L4	0.93 ± 0.01	0.79 ± 0.03	0.89 ± 0.02	0.93 ± 0.03
L5	0.87 ± 0.05	0.72 ± 0.04	0.89 ± 0.01	0.88 ± 0.04
S1	0.91 ± 0.02	0.84 ± 0.03	0.93 ± 0.01	0.92 ± 0.01
S2	0.94 ± 0.02	0.85 ± 0.06	0.93 ± 0.01	0.92 ± 0.02

For each subject, correlation coefficients between segmental motor outputs (based on 26 recorded muscles) of individual and ensemble-averaged strides were obtained and averaged across strides. Then, for each spinal segment, these correlation coefficients were averaged across subjects and reported in this table (mean ± SD).

Table 2 | Sensitivity of spinal maps to the muscle subset analyzed.

	20 muscles				12 muscles			
	FW	BW	Tiptoe	Inclined	FW	BW	Tiptoe	Inclined
L2	1.00	1.00	1.00	1.00	0.94	0.97	0.91	0.98
L3	1.00	1.00	1.00	1.00	0.94	0.96	0.86	0.98
L4	0.99	0.99	0.99	0.99	0.92	0.93	0.91	0.94
L5	0.92	0.97	0.99	0.95	0.74	0.90	0.83	0.74
S1	0.96	0.99	0.99	0.99	0.92	0.98	0.95	0.98
S2	0.99	0.99	0.98	1.00	0.99	0.99	0.99	1.00
Total	0.98 ± 0.03	0.99 ± 0.01	0.99 ± 0.01	0.99 ± 0.02	0.91 ± 0.08	0.96 ± 0.03	0.91 ± 0.05	0.94 ± 0.09

Correlation coefficients are reported for comparisons between motor outputs based on all 26 recorded muscles and those obtained from subsets of 20 or 12 muscles. For each gait, correlations are reported for individual spinal segments and for the total spinal map (mean ± SD).

specific motor pools when using epidural electrical stimulation or corticospinal neuroprostheses for restoring locomotor functions (Capogrosso et al., 2013; Borton et al., 2014).

ACKNOWLEDGMENTS

The financial support from the Italian Health Ministry, Italian University Ministry (PRIN project), Italian Space Agency (CRUSOE and COREA grants), EU FP7-ICT program (MINDWALKER grant #247959 and AMARSi grant #248311) and Whitaker International Program is gratefully acknowledged.

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: http://www.frontiersin.org/journal/10.3389/fnhum.2014.00305/abstract

Here we report the anatomical data used to reconstruct the segmental motor output during human locomotion (see Methods). These data include segmental muscle innervation in humans (**Figure S1**) and the number of MNs in each segment of the human spinal cord (**Table S1**).

Figure S1 | Spinal segment muscle innervation. This graphic is adapted from Kendall et al. (2005), which compiled segmental innervation charts for muscles by integrating the anatomical and clinical data of several different sources. The blue color in the chart below denotes an innervation agreed upon by five or more sources, and the light blue color denotes agreement of three to four sources. In our spinal map analysis, we used the weighting coefficient (k_{ji}) to define the major (1) and minor (0.5) innervation segments for each muscle.

Table S1 | Number of motoneurons (MNs) in each segment of the human spinal cord (adapted from Tomlinson and Irving, 1977).

REFERENCES

Andersson, E., Nilsson, J., and Thorstensson, A. (1997). Intramuscular EMG from the hip flexor muscles during human locomotion. *Acta Physiol. Scand.* 161, 361–370. doi: 10.1046/j.1365-201X.1997.00225.x

Bagnall, M. W., and McLean, D. L. (2014). Modular organization of axial microcircuits in zebrafish. *Science* 343, 197–200. doi: 10.1126/science.1245629

Borton, D., Bonizzato, M., Beauparlant, J., Digiovanna, J., Moraud, E. M., Wenger, N., et al. (2014). Corticospinal neuroprostheses to restore locomotion after spinal cord injury. *Neurosci. Res.* 78, 21–29. doi: 10.1016/j.neures.2013.10.001

Capogrosso, M., Wenger, N., Raspopovic, S., Musienko, P., Beauparlant, J., Bassi Luciani, L., et al. (2013). A computational model for epidural electrical stimulation of spinal sensorimotor circuits. *J. Neurosci.* 33, 19326–19340. doi: 10.1523/JNEUROSCI.1688-13.2013

Cappellini, G., Ivanenko, Y. P., Dominici, N., Poppele, R. E., and Lacquaniti, F. (2010). Migration of motor pool activity in the spinal cord reflects body mechanics in human locomotion. *J. Neurophysiol.* 104, 3064–3073. doi: 10.1152/jn.00318.2010

Carlson-Kuhta, P., Trank, T. V., and Smith, J. L. (1998). Forms of forward quadrupedal locomotion. II. A comparison of posture, hindlimb kinematics, and motor patterns for upslope and level walking. *J. Neurophysiol.* 79, 1687–1701.

Christensen, E. (1959). Topography of terminal motor innervation in striated muscles from stillborn infants. *Am. J. Phys. Med.* 38, 65–78. doi: 10.1097/00002060-195904000-00005

Coscia, M., Monaco, V., Capogrosso, M., Chisari, C., and Micera, S. (2011). Computational aspects of MN activity estimation: a case study with post-stroke subjects. *IEEE Int. Conf. Rehabil. Robot. Proc.* 2011, 5975405. doi: 10.1109/ICORR.2011.5975405

Cunningham, C. B., Schilling, N., Anders, C., and Carrier, D. R. (2010). The influence of foot posture on the cost of transport in humans. *J. Exp. Biol.* 213, 790–797. doi: 10.1242/jeb.038984

D'Avella, A., and Bizzi, E. (2005). Shared and specific muscle synergies in natural motor behaviors. *Proc. Natl. Acad. Sci. U.S.A.* 102, 3076–3081. doi: 10.1073/pnas.0500199102

D'Avella, A., Cesqui, B., and Lacquaniti, F. (2013). "Identifying muscle synergies from EMG decomposition: approaches, evidence, and potential application to neurorehabilitation," in *Converging Clinical and Engineering Research on Neurorehabilitation*, eds J. L. Pons, D. Torricelli, and M. Pajaro (Berlin; Heidelberg: Springer Berlin Heidelberg), 1243–1247.

Day, S. J., and Hulliger, M. (2001). Experimental simulation of cat electromyogram: evidence for algebraic summation of motor-unit action-potential trains. *J. Neurophysiol.* 86, 2144–2158.

DeVita, P., Helseth, J., and Hortobagyi, T. (2007). Muscles do more positive than negative work in human locomotion. *J. Exp. Biol.* 210, 3361–3373. doi: 10.1242/jeb.003970

Donelan, J. M., Kram, R., and Kuo, A. D. (2002). Mechanical work for step-to-step transitions is a major determinant of the metabolic cost of human walking. *J. Exp. Biol.* 205, 3717–3727.

Duysens, J., van Wezel, B. M., van de Crommert, H. W., Faist, M., and Kooloos, J. G. (1998). The role of afferent feedback in the control of hamstrings activity during human gait. *Eur. J. Morphol.* 36, 293–299. doi: 10.1076/ejom.36.4.0293

Feinstein, B., Lindegard, B., Nyman, E., and Wohlfart, G. (1955). Morphologic studies of motor units in normal human muscles. *Acta Anat. (Basel)* 23, 127–142. doi: 10.1159/000140989

Franz, J. R., and Kram, R. (2012). The effects of grade and speed on leg muscle activations during walking. *Gait Posture* 35, 143–147. doi: 10.1016/j.gaitpost.2011.08.025

Gersten, J. W., Mastellone, A. F., and Sheffield, F. J. (1956). Electromyographic study of the muscles of the foot in normal walking. *Am. J. Phys. Med.* 35, 223–236.

Giszter, S. F., Hart, C. B., and Silfies, S. P. (2010). Spinal cord modularity: evolution, development, and optimization and the possible relevance to low back pain in man. *Exp. Brain Res.* 200, 283–306. doi: 10.1007/s00221-009-2016-x

Giszter, S., Patil, V., and Hart, C. (2007). Primitives, premotor drives, and pattern generation: a combined computational and neuroethological perspective. *Prog. Brain Res.* 165, 323–346. doi: 10.1016/S0079-6123(06)65020-6

Grasso, R., Bianchi, L., and Lacquaniti, F. (1998). Motor patterns for human gait: backward versus forward locomotion. *J. Neurophysiol.* 80, 1868–1885.

Grasso, R., Ivanenko, Y. P., Zago, M., Molinari, M., Scivoletto, G., Castellano, V., et al. (2004). Distributed plasticity of locomotor pattern generators in spinal cord injured patients. *Brain* 127, 1019–1034. doi: 10.1093/brain/awh115

Grillner, S. (1981). "Control of locomotion in bipeds, tetrapods and fish," in *Handbook of Physiology: Section 1: The Nervous System, volume II, Part.1 Motor Control*, eds V. B. Brooks, J. M. Brookhart, and V. B. Mountcastle (Bethesda, MD: American Physiological Society), 1179–1236.

Haight, D. J., Lerner, Z. F., Board, W. J., and Browning, R. C. (2014). A comparison of slow, uphill and fast, level walking on lower extremity biomechanics and tibiofemoral joint loading in obese and nonobese adults. *J. Orthop. Res.* 32, 324–330. doi: 10.1002/jor.22497

Harel, N. Y., and Strittmatter, S. M. (2008). Functional MRI and other non-invasive imaging technologies: providing visual biomarkers for spinal cord structure and function after injury. *Exp. Neurol.* 211, 324–328. doi: 10.1016/j.expneurol.2008.02.017

Hart, C. B., and Giszter, S. F. (2010). A neural basis for motor primitives in the spinal cord. *J. Neurosci.* 30, 1322–1336. doi: 10.1523/JNEUROSCI.5894-08.2010

Hausdorff, J. M. (2007). Gait dynamics, fractals and falls: finding meaning in the stride-to-stride fluctuations of human walking. *Hum. Mov. Sci.* 26, 555–589. doi: 10.1016/j.humov.2007.05.003

Hoffer, J. A., Sugano, N., Loeb, G. E., Marks, W. B., O'Donovan, M. J., and Pratt, C. A. (1987). Cat hindlimb motoneurons during locomotion. II. Normal activity patterns. *J. Neurophysiol.* 57, 530–553.

Hoogkamer, W., Meyns, P., and Duysens, J. (2014). Steps forward in understanding backward gait: from basic circuits to rehabilitation. *Exerc. Sport Sci. Rev.* 42, 23–29. doi: 10.1249/JES.0000000000000000

Ivanenko, Y. P., Cappellini, G., Dominici, N., Poppele, R. E., and Lacquaniti, F. (2007). Modular control of limb movements during human locomotion. *J. Neurosci.* 27, 11149–11161. doi: 10.1523/JNEUROSCI.2644-07.2007

Ivanenko, Y. P., Cappellini, G., Poppele, R. E., and Lacquaniti, F. (2008). Spatiotemporal organization of alpha-motoneuron activity in the human spinal cord during different gaits and gait transitions. *Eur. J. Neurosci.* 27, 3351–3368. doi: 10.1111/j.1460-9568.2008.06289.x

Ivanenko, Y. P., Cappellini, G., Solopova, I. A., Grishin, A. A., Maclellan, M. J., Poppele, R. E., et al. (2013a). Plasticity and modular control of locomotor patterns in neurological disorders with motor deficits. *Front. Comput. Neurosci.* 7:123. doi: 10.3389/fncom.2013.00123

Ivanenko, Y. P., Dominici, N., Cappellini, G., Di Paolo, A., Giannini, C., Poppele, R. E., et al. (2013b). Changes in the spinal segmental motor output for stepping during development from infant to adult. *J. Neurosci.* 33, 3025–3036. doi: 10.1523/JNEUROSCI.2722-12.2013

Ivanenko, Y. P., Poppele, R. E., and Lacquaniti, F. (2006). Spinal cord maps of spatiotemporal alpha-motoneuron activation in humans walking at different speeds. *J. Neurophysiol.* 95, 602–618. doi: 10.1152/jn.00767.2005

Kang, H. G., and Dingwell, J. B. (2009). Dynamics and stability of muscle activations during walking in healthy young and older adults. *J. Biomech.* 42, 2231–2237. doi: 10.1016/j.jbiomech.2009.06.038

Kendall, F., McCreary, E., Provance, P., Rodgers, M., and Romani, W. (2005). *Muscles. Testing and Function with Posture and Pain*. Baltimore, MD: Lippincott Williams and Wilkins.

Lacquaniti, F., Ivanenko, Y. P., d'Avella, A., Zelik, K. E., and Zago, M. (2013). Evolutionary and developmental modules. *Front. Comput. Neurosci.* 7:61. doi: 10.3389/fncom.2013.00061

Lange, G. W., Hintermeister, R. A., Schlegel, T., Dillman, C. J., and Steadman, J. R. (1996). Electromyographic and kinematic analysis of graded treadmill walking and the implications for knee rehabilitation. *J. Orthop. Sports Phys. Ther.* 23, 294–301. doi: 10.2519/jospt.1996.23.5.294

MacLellan, M. J., Ivanenko, Y. P., Cappellini, G., Sylos Labini, F., and Lacquaniti, F. (2012). Features of hand-foot crawling behavior in human adults. *J. Neurophysiol.* 107, 114–125. doi: 10.1152/jn.00693.2011

Mann, R., and Inman, V. T. (1964). Phasic activity of intrinsic muscles of the foot. *J. Bone Joint Surg. Am.* 46, 469–481.

McComas, A. J. (1998). 1998 ISEK Congress Keynote Lecture: motor units: how many, how large, what kind? International Society of Electrophysiology and Kinesiology. *J. Electromyogr. Kinesiol.* 8, 391–402. doi: 10.1016/S1050-6411(98)00020-0

McComas, A. J., Quartly, C., and Griggs, R. C. (1997). Early and late losses of motor units after poliomyelitis. *Brain J. Neurol.* 120(Pt 8), 1415–1421. doi: 10.1093/brain/120.8.1415

McCrea, D. A., and Rybak, I. A. (2008). Organization of mammalian locomotor rhythm and pattern generation. *Brain Res. Rev.* 57, 134–146. doi: 10.1016/j.brainresrev.2007.08.006

Monaco, V., Ghionzoli, A., and Micera, S. (2010). Age-related modifications of muscle synergies and spinal cord activity during locomotion. *J. Neurophysiol.* 104, 2092–2102. doi: 10.1152/jn.00525.2009

Nielsen, J. B., and Sinkjaer, T. (2002). Afferent feedback in the control of human gait. *J. Electromyogr. Kinesiol.* 12, 213–217. doi: 10.1016/S1050-6411(02)00023-8

O'Donovan, M. J., Bonnot, A., Mentis, G. Z., Arai, Y., Chub, N., Shneider, N. A., et al. (2008). Imaging the spatiotemporal organization of neural activity in the developing spinal cord. *Dev. Neurobiol.* 68, 788–803. doi: 10.1002/dneu.20620

Oetgen, M. E., and Peden, S. (2012). Idiopathic toe walking. *J. Am. Acad. Orthop. Surg.* 20, 292–300. doi: 10.5435/JAAOS-20-05-292

Pearson, K. G. (2004). Generating the walking gait: role of sensory feedback. *Prog. Brain Res.* 143, 123–129. doi: 10.1016/S0079-6123(03)43012-4

Perry, J., Burnfield, J. M., Gronley, J. K., and Mulroy, S. J. (2003). Toe walking: muscular demands at the ankle and knee. *Arch. Phys. Med. Rehabil.* 84, 7–16. doi: 10.1053/apmr.2003.50057

Potvin, J. R., and Brown, S. H. M. (2004). Less is more: high pass filtering, to remove up to 99% of the surface EMG signal power, improves EMG-based biceps brachii muscle force estimates. *J. Electromyogr. Kinesiol.* 14, 389–399. doi: 10.1016/j.jelekin.2003.10.005

Rab, G. T. (1994). "Muscle," in *Human Walking, 2nd Edn.*, eds J. Rose and J. G. Gamble (Baltimore, MD: Willaims and Wilkins), 101–122.

Romanes, G. J. (1951). The motor cell columns of the lumbo-sacral spinal cord of the cat. *J. Comp. Neurol.* 94, 313–363. doi: 10.1002/cne.900940209

Romkes, J., and Brunner, R. (2007). An electromyographic analysis of obligatory (hemiplegic cerebral palsy) and voluntary (normal) unilateral toe-walking. *Gait Posture* 26, 577–586. doi: 10.1016/j.gaitpost.2006.12.010

Saltiel, P., and Rossignol, S. (2004). Critical points in the forelimb fictive locomotor cycle and motor coordination: evidence from the effects of tonic proprioceptive perturbations in the cat. *J. Neurophysiol.* 92, 1329–1341. doi: 10.1152/jn.00563.2003

Scivoletto, G., Ivanenko, Y., Morganti, B., Grasso, R., Zago, M., Lacquaniti, F., et al. (2007). Plasticity of spinal centers in spinal cord injury patients: new concepts for gait evaluation and training. *Neurorehabil. Neural Repair* 21, 358–365. doi: 10.1177/1545968306295561

Sharrard, W. J. (1964). The segmental innervation of the lower limb muscles in man. *Ann. R. Coll. Surg. Engl.* 35, 106–122.

Stroman, P. W., Wheeler-Kingshott, C., Bacon, M., Schwab, J. M., Bosma, R., Brooks, J., et al. (2014). The current state-of-the-art of spinal cord imaging: methods. *Neuroimage* 84, 1070–1081. doi: 10.1016/j.neuroimage.2013.04.124

Thorstensson, A. (1986). How is the normal locomotor program modified to produce backward walking? *Exp. Brain Res.* 61, 664–668. doi: 10.1007/BF00237595

Ting, L., Chvatal, S., Safavynia, S., and McKay, J. (2012). Review and perspective: neuromechanical considerations for predicting muscle activation patterns for movement. *Int. J. Numer. Methods Biomed. Eng.* 28, 1003–1014. doi: 10.1002/cnm.2485

Ting, L. H. (2007). Dimensional reduction in sensorimotor systems: a framework for understanding muscle coordination of posture. *Prog. Brain Res.* 165, 299–321. doi: 10.1016/S0079-6123(06)65019-X

Tomlinson, B. E., and Irving, D. (1977). The numbers of limb motor neurons in the human lumbosacral cord throughout life. *J. Neurol. Sci.* 34, 213–219. doi: 10.1016/0022-510X(77)90069-7

University of California, Berkeley. (1953). *The Pattern of Muscular Activity in the Lower Extremity During Walking.* Prosthetic research project, Series II, Issue 25. *Report Presented to the Advisory Committee on Artificial Limbs, National Research Council* (Los Angeles, CA).

Warp, E., Agarwal, G., Wyart, C., Friedmann, D., Oldfield, C. S., Conner, A., et al. (2012). Emergence of patterned activity in the developing zebrafish spinal cord. *Curr. Biol.* 22, 93–102. doi: 10.1016/j.cub.2011.12.002

Winter, D. A. (1991). *The Biomechanics and Motor Control of Human Gait: Normal, Elderly and Pathological.* Waterloo, ON: Waterloo Biomechanics.

Yakovenko, S., Mushahwar, V., VanderHorst, V., Holstege, G., and Prochazka, A. (2002). Spatiotemporal activation of lumbosacral motoneurons in the locomotor step cycle. *J. Neurophysiol.* 87, 1542–1553.

Zelik, K. E., La Scaleia, V., Ivanenko, Y. P., and Lacquaniti, F. (2014). Can modular strategies simplify neural control of multidirectional human locomotion? *J. Neurophysiol.* 111, 1686–1702. doi: 10.1152/jn.00776.2013

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.

Received: 29 October 2013; accepted: 25 April 2014; published online: 15 May 2014.
Citation: La Scaleia V, Ivanenko YP, Zelik KE and Lacquaniti F (2014) Spinal motor outputs during step-to-step transitions of diverse human gaits. Front. Hum. Neurosci. ***8****:305. doi: 10.3389/fnhum.2014.00305*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 23 May 2014
doi: 10.3389/fnhum.2014.00335

Motor modules of human locomotion: influence of EMG averaging, concatenation, and number of step cycles

Anderson S. Oliveira[1]*, Leonardo Gizzi[2], Dario Farina[3] and Uwe G. Kersting[1]

[1] *Department of Health Science and Technology, Center for Sensory-Motor Interaction, Aalborg University, Aalborg, Denmark*
[2] *Pain Clinic Center for Anesthesiology, Emergency and Intensive Care Medicine, University Hospital Göttingen, Göttingen, Germany*
[3] *Department of Neurorehabilitation Engineering, Bernstein Focus Neurotechnology Göttingen, Bernstein Center for Computational Neuroscience, University Medical Center Göttingen, Georg-August University, Göttingen, Germany*

Edited by:
Nadia Dominici, EPFL, Switzerland

Reviewed by:
Jinsook Roh, Rehabilitation Institute of Chicago, USA
David J. Clark, Malcom Randall VA Medical Center, USA

***Correspondence:**
Anderson S. Oliveira, Department of Health Science and Technology, Center for Sensory-Motor Interaction, Aalborg University, Fredrik Bajers Vej 7 D-3, DK-9220 Aalborg, Denmark
e-mail: oliveira_dkbr@hotmail.com

Locomotion can be investigated by factorization of electromyographic (EMG) signals, e.g., with non-negative matrix factorization (NMF). This approach is a convenient concise representation of muscle activities as distributed in motor modules, activated in specific gait phases. For applying NMF, the EMG signals are analyzed either as single trials, or as averaged EMG, or as concatenated EMG (data structure). The aim of this study is to investigate the influence of the data structure on the extracted motor modules. Twelve healthy men walked at their preferred speed on a treadmill while surface EMG signals were recorded for 60 s from 10 lower limb muscles. Motor modules representing relative weightings of synergistic muscle activations were extracted by NMF from 40 step cycles separately (EMG_{SNG}), from averaging 2, 3, 5, 10, 20, and 40 consecutive cycles (EMG_{AVR}), and from the concatenation of the same sets of consecutive cycles (EMG_{CNC}). Five motor modules were sufficient to reconstruct the original EMG datasets (reconstruction quality >90%), regardless of the type of data structure used. However, EMG_{CNC} was associated with a slightly reduced reconstruction quality with respect to EMG_{AVR}. Most motor modules were similar when extracted from different data structures (similarity >0.85). However, the quality of the reconstructed 40-step EMG_{CNC} datasets when using the muscle weightings from EMG_{AVR} was low (reconstruction quality ~40%). On the other hand, the use of weightings from EMG_{CNC} for reconstructing this long period of locomotion provided higher quality, especially using 20 concatenated steps (reconstruction quality ~80%). Although EMG_{SNG} and EMG_{AVR} showed a higher reconstruction quality for short signal intervals, these data structures did not account for step-to-step variability. The results of this study provide practical guidelines on the methodological aspects of synergistic muscle activation extraction from EMG during locomotion.

Keywords: locomotion, variability, EMG, muscle synergies, motor modules, neural control

INTRODUCTION

Surface electromyography (EMG) represents indirectly the neural inputs from many sources (supraspinal, reflex activities, somatosensory information) to the muscles and has therefore been widely used to define neural strategies to perform motor tasks (Lacquaniti et al., 2012). An increasing number of investigations have been applying factorization analyses on multi-muscle surface EMG signals in order to extract basic motor patterns or modules (also called muscle synergies) that concisely represent the neural strategies for recruiting muscles during locomotor tasks (Ivanenko et al., 2005; Cappellini et al., 2006; Lacquaniti et al., 2012; Oliveira et al., 2013a). These investigations reported a low-dimensional model for representing the neural control of muscles during human locomotion, which is characterized by activation signals that define the instants of recruitment of specific motor modules related to biomechanical sub-tasks (Lacquaniti et al., 2012).

Human locomotion is a largely automatized motor behavior, therefore the step-to-step variability of the main neural inputs to the muscles is limited. Studies applying factorization analysis focusing on human locomotion usually report a low-dimensional set of four to six motor modules to represent neural inputs to the muscles (Ivanenko et al., 2004; Lacquaniti et al., 2012). Differences in the number of motor modules needed for an accurate description (i.e., dimensionality) may be related to a variable number of muscles included in the EMG dataset and different low-pass filtering among studies (Hug et al., 2012; Steele et al., 2013). In addition, there is a wide range of number of step cycles used for extracting representative motor modules; for example, some studies used 4–12 cycles (Monaco et al., 2010), others 10–25 cycles (Merkle et al., 1998; Ivanenko et al., 2004, 2005; Gizzi et al., 2011; Oliveira et al., 2013a; Sartori et al., 2013b), and in some cases up to 30 cycles (Clark et al., 2010). The number of step cycles used for the estimation of the synergistic activation is relevant for applications to biofeedback and rehabilitation technologies, when the extracted motor modules are used either for feedback purposes or as a basis for controlling the interaction with robotic devices. For example, because the

motor modules provides a concise representation of relative muscle activations, their online estimation can be used by a training supervisor to focus the attention of the patient on those muscles that are abnormally activated during a certain phase of the step cycle.

Previous investigations extracting motor modules from single trials reported reconstruction quality over 90% (Ivanenko et al., 2004, 2005), whereas analyses in which consecutive step cycles were concatenated and analyzed together resulted in a lower reconstruction quality (Gizzi et al., 2011; Oliveira et al., 2012, 2013a,b). Reduced reconstruction quality in concatenated analyses may be an effect of natural step-to-step variability contained in surface EMG signals, which may be crucial for specific kinematic adjustments during locomotion. Recently, de Rugy et al. (2013) have raised concerns about the use of factorization analysis because they noticed that even small reconstruction errors in muscle activity could correspond to relatively important changes in force production. Therefore, although EMG factorization analysis is a promising tool for locomotion rehabilitation and robotic control (Gizzi et al., 2012; Moreno et al., 2013; Sartori et al., 2013a), there are many aspects that still remain unclear for an optimal and consistent application of such methodology.

In this study we explored the differences in the extracted motor modules when varying the EMG data structure by comparing the factorization results when using single step EMG (EMG_{SNG}), averaged EMG (EMG_{AVR}), and concatenated EMG (EMG_{CNC}). Applications in neurotechnologies for rehabilitation, e.g., biofeedback, would benefit from the analysis on the shortest time interval (single cycles) that would allow adaptive/reactive responses. However, single cycle factorization would present fast variations on a cycle basis. These variations may be relevant in some applications, e.g., in patients with high intrinsic step-to-step variability, but not in others. The hypothesis of the study was that the use of different data structures (single trials, averaging or concatenating EMG signals) to identify motor modules influences the extracted dimensionality and/or the modules. The results obtained are of practical relevance when using EMG factorization for the study of human locomotion.

METHODS

SUBJECTS

Twelve healthy men (age: 28 ± 4 years; body mass: 80.8 ± 8 kg; body height: 178 ± 4 cm) volunteered for the experiment. One subject was left-dominant and all others were right-dominant. Exclusion criteria included history of knee or ankle ligament injury, current lower-extremity injury, recent (within 6 months) low back injury, or vestibular dysfunction. All subjects provided written informed consent before participation and the procedures were approved by the ethical committee of Northern Jutland (N-20130015).

EXPERIMENTAL SETUP

In a single session subjects were initially asked to perform familiarization to the treadmill (Woodway Pro, Foster Court Waukesha, USA) by walking for 5 min. Subsequently, preferred walking speed was determined following previous literature (Choi and Bastian, 2007) and after a 2-min rest period, subjects walked at the selected speed for 5 min during which surface EMG and walking cadence were recorded from the last 60 s (see **Figure 1A** for illustration).

DATA COLLECTION

EMG signals were recorded in bipolar derivations with pairs of Ag/AgCl electrodes (Ambu Neuroline 720 01-K/12; Ambu, Ballerup, Denmark) with 22 mm of center-to-center spacing. Prior to electrode placement the skin was shaved and lightly abraded. A reference electrode was placed on the right tibia. The EMG signals were recorded from a portable EMG amplifier (Biovision EMG-Amp, Germany) stored in a backpack together with a mini-computer. The EMG signals were sampled at 2000 Hz (12 bits per sample), band-pass filtered (second-order, zero lag Butterworth, bandwidth 10–500 Hz). The EMG signals were recorded from the following muscles of the right side (dominant side for 11 out of 12 subjects) according to Barbero et al. (2011): tibialis anterior (TA), soleus (SO), gastrocnemius lateralis (GL), gastrocnemius medialis (GM), vastus lateralis (VL), vastus medialis (VM), rectus femoris (RF), biceps femoris (BF), semitendinosus (ST), and gluteus maximus (GX). A uniaxial accelerometer was placed on the right tibia, which measured the vertical acceleration synchronized to the EMG measurements.

DATA ANALYSIS

Accelerometry

Data from tibia vertical acceleration were low-pass filtered (60 Hz) and step cycles were determined following previously reported methods (Kersting, 2011). Individual step cycles were time-normalized to 200 data points for one step cycle.

Surface EMG

The segmentation for EMG factorization was defined from the accelerometer data, from which step cycles were determined. After segmentation, the surface EMG signals from the 10 muscles were band-pass filtered (20–500 Hz), full-wave rectified, low-pass filtered (10 Hz) and time-normalized in order to obtain 200 data points for one step cycle.

Motor modules extraction

For each subject, non-negative matrix factorization (NMF, Lee and Seung, 2001) was applied in order to process the EMG_{SNG} extraction and identify motor modules and activation signals from the 40 consecutive step cycles separately. Subsequently, the vectors representing muscle weightings and activation signals were averaged for each subject which could be compared to motor modules from the other two processing methods. In addition, NMF was applied in EMG datasets containing two, three, five, 10, 20, and 40 consecutive step cycles in two processing modalities. The first (EMG_{AVR}) consisted of averaging the different number of step cycles for subsequent extraction of motor modules. The second method (EMG_{CNC}) consisted of the concatenation of a given amount of step cycles for subsequent extraction of motor modules. In this case, all variability from sequential step cycles is accounted for during NMF analysis, which may reduce reconstruction quality for longer datasets including

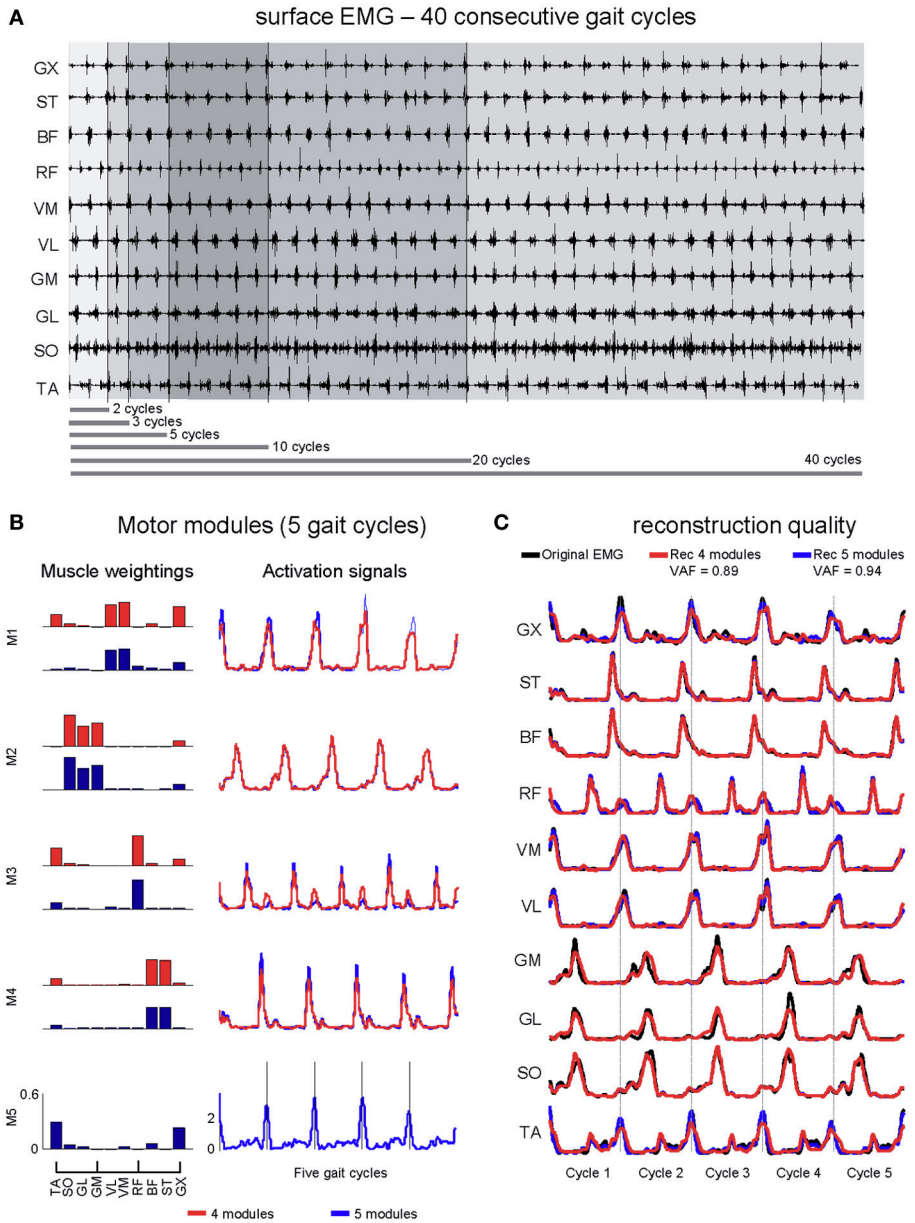


FIGURE 1 | Illustrative sample of surface EMG signals (A) from 40 consecutive walking step cycles. (B) Non-negative matrix factorization provides similar muscle weightings by using 4 and 5 motor modules (similarity >0.9 for the first four motor modules), however differences in the activation signals can be qualitatively observed, especially for M1 and M3. The reconstruction of the original EMG dataset is shown in **(C)**. The original EMG (*black lines*) from some muscles (i.e., VL, VM, BF, ST) is well reconstructed by using both 4 modules (Rec 4 modules, *red lines*) or five modules (Rec 5 modules, *blue lines*). On the other hand, muscles such as TA, RF, and GX exhibited better reconstruction only by using 5 modules.

a greater number of consecutive cycles (see **Figures 1B,C** for illustration). For all three EMG processing methods, individual surface EMG channels were normalized by the peak activation, so that all channels were ranging from 0 to 1 in amplitude.

Motor module model

The EMG signals $X(k)$ recorded from M muscles were factorized as

$$X(k) \approx X_r(k) = S \cdot P(k) \quad (1)$$

where $X_r(k)$ is the muscle activity vector reconstructed by the factorization, S is a scalar matrix (synergy matrix or motor module matrix), and $P(k)$ are the activation signals, of dimension $N < M$. In Equation (1), the EMG $X(k)$ are obtained by linear transformation of the activation signals $P(k)$ with gain factors s_{mn} (the entries of the synergy matrix, Lee and Seung, 2001).

Dimensionality

After extracting the motor modules, the estimated muscular activation pattern was compared with the experimental pattern by

means of the variability accounted for (VAF) value, defined as the variation that can be explained by the model: VAF = 1− SSE/SST, where SSE (sum of squared errors) is the unexplained variation and SST (total sum of squares) is the pooled variation of the data. The reconstruction quality was analyzed by plotting the VAF as a function of the number of modules, and the minimum acceptable number of modules was identified as the point in which this curve pronouncedly changes its slope (d'Avella et al., 2003; Muceli et al., 2010), and additionally, the number of modules must also successfully reconstruct at least 90% of the original EMG content. In addition, we reconstructed EMG signals from the three processing methods and number of steps in two different ways: (a) the combination of extracted muscle weightings with activation signals obtained from randomly generated matrix (i.e., activation signals free to vary) and (b) the combination of extracted activation signals with muscle weightings obtained from randomly generated matrix (i.e., muscle weightings free to vary). The latter analysis provided the quality of EMG reconstruction (i.e., VAF) that can be achieved by using random variability, which was hypothesized to be lower than the VAF obtained by reconstructing EMG signals with the factorization obtained by NMF. The muscle weightings and activation signals free to vary were obtained by iterating 1000 times the NMF update rules (Lee and Seung, 1999), only for muscle weightings or activation signals, respectively.

Similarities

The muscle weightings and activation signals from two sets were compared by computing the similarity between the best matched pairs, as described in d'Avella et al. (2003). Similarities were then calculated by computing the scalar product between pairs of vectors (motor modules or activation signals), normalized by the product of the norms of each column (d'Avella et al., 2003; Muceli et al., 2010), which prioritizes the comparison between the shapes of vectors rather than amplitude. Similarity can vary from 0 (no curve shape matching) to 1 (perfect curve shape matching) and previous investigations have used values above 0.8 to define if a pair of vectors is similar (Gizzi et al., 2011; Oliveira et al., 2013b). Intra-subject similarity analyses were conducted for individual motor modules and individual activation signals between the different numbers of step cycles for each given EMG processing method. In addition, intra-subject similarities between methods were calculated for each sequence of step cycles.

EMG reconstructed from different muscle weightings

Additionally to similarity analysis, we fixed the muscle weightings extracted from 2, 3, 5, 10, and 20 steps of the first half of the recording, and used such weightings for reconstructing another sequence of 2, 3, 5, 10, and 20 concatenated steps from the second half, as well as the whole sequence of 40 cycles. This analysis reflected the situation in which the motor modules are computed from only an initial portion of the recording and then used to explain the remaining part of the recording. This procedure was conducted by using muscle weightings from EMG_{CNC} and from EMG_{AVR}. For instance, we reconstructed the concatenated EMG from 40 step cycles by using its original activation signals combined to the muscle weightings from shorter concatenation periods (2, 3, 5, 10, and 20 cycles). By doing so we aimed at directly testing the reconstruction performance of motor modules extracted from different concatenation lengths as a measure of their representativeness for a longer signal interval.

Statistical analysis

The degrees of similarity between individual motor modules and between individual activation signals were compared by a One-Way ANOVA. The significance level was set to $p < 0.05$. A Two-Way ANOVA was used in order to verify the effects of EMG processing method (EMG_{AVR} vs. EMG_{CNC}) and number of step cycles (two, three, five, 10, 20, and 40) on the reconstruction quality (VAF). In addition, Student *t*-tests were used to investigate differences between intra-subject similarities among the EMG processing methods. All statistical procedures were conducted using SPSS 18.0 (SPSS, Inc., Chicago, IL, USA).

RESULTS

DIMENSIONALITY

The analysis of dimensionality from EMG_{SNG} revealed that four motor modules were required to reconstruct unilateral muscular activations with an overall reconstruction quality of 88% (VAF = 88 ± 3%, average across all muscles, **Figure 2A**). For three out of 12 subjects the VAF was higher than 90% for all muscles by reconstructing the EMG from four motor modules. However, for most of subjects, muscles such as TA, RF, and GX showed poorer reconstruction quality than the average (∼80%). By using five motor modules the overall reconstruction quality increased to 93 ± 2%, and all muscles could reach an average reconstruction quality >90% (see **Figures 1B,C** for illustration).

The calculated VAF by combining randomly generated muscle weightings and the extracted activation signals to reconstruct the original EMG datasets was 38 ± 8, 36 ± 11, and 32 ± 9% for EMG_{SNG}, EMG_{AVR}, and EMG_{CNC} respectively. Similarly, the results from calculating the VAF by combining randomly generated activation signals and the extracted muscle weightings to reconstruct the original EMG datasets was 39 ± 10%, 33 ± 12%, and 37 ± 10% for EMG_{SNG}, EMG_{AVR}, and EMG_{CNC} respectively. Both simulations showed a very poor reconstruction quality in comparison to the extracted motor modules, which suggests that the extracted motor modules provide meaningful information that random variability cannot reproduce.

AVERAGING vs. CONCATENATING EMG SIGNALS

By fixing the number of modules to five we compared the results from EMG_{AVR} and EMG_{CNC}. The Two-Way ANOVA revealed no EMG processing vs. number of step cycles interaction, however there was a significant effect of the EMG processing method ($p < 0.001$, $F = 90.5$). The reconstruction accuracy was approximately 94% (VAF = 0.94 ± 0.02, **Figure 2B**) for motor modules from EMG_{AVR}, and slightly lower (∼90%) when using EMG_{CNC} (VAF = 0.90 ± 0.03). In both cases, the number of step cycles used for the calculation did not significantly influence the estimates.

MOTOR MODULES FROM TREADMILL WALKING

Four out of the five motor modules could be assigned to biomechanical subtasks of walking (**Figure 3**). Module 1 (M1) consists of the activation of knee extensors and GX (see muscle weightings

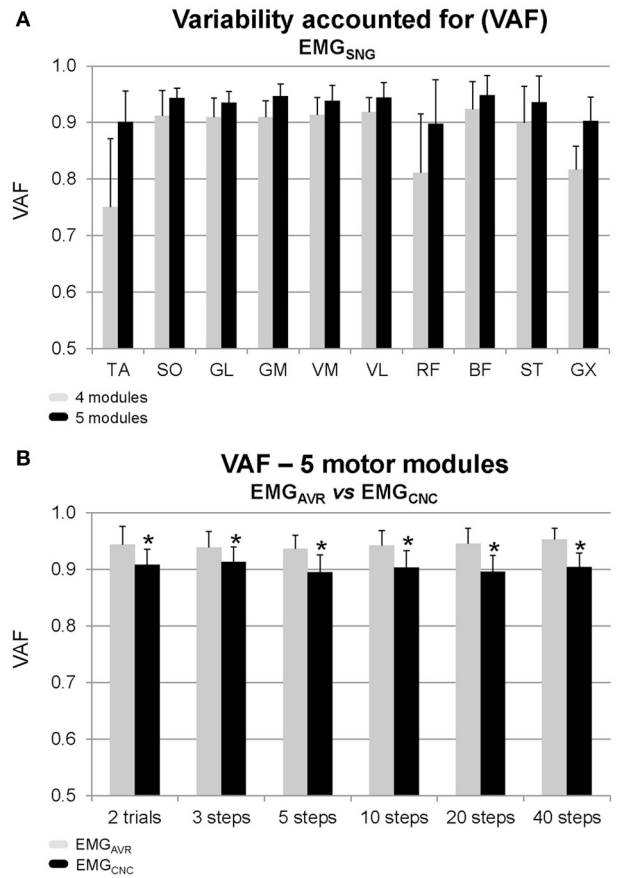


FIGURE 2 | Mean ± *SD* of the variation accounted for (VAF) from the factorization analysis of individual step cycles (A) by considering to reconstruct the original EMG dataset using four motor modules (*gray bars*) and five motor modules (*black bars*) from single step cycles (EMG$_{SNG}$). (B) The VAF from the reconstruction of original EMG datasets by using five motor modules from averaged EMG (EMG$_{AVR}$, *gray bars*) and from concatenated (EMG$_{CNC}$, *black bars*) in different amounts of step cycles. *Denotes significant difference in relation to EMG$_{AVR}$ ($p < 0.05$).

in **Figure 3A**) at the beginning of the stance period (see activation signals in **Figure 3B**). Module 2 (M2) relates to forward propulsion, in which the plantarflexors are predominantly recruited. Module 3 (M3) relates to the leg swing, in which TA and RF are recruited throughout the swing phase, and Module 4 (M4) is related to the recruitment of the hamstring muscles (ST, BF) prior to the subsequent initial contact. The fifth module (M5) involves the recruitment of ankle joint muscles as well as RF and GX, with no clear burst-like activity throughout the step cycle. The recruitment of this motor module is predominant at initial contact, transition from stance to swing phase and prior to subsequent initial contact.

INTRA-SUBJECT SIMILARITIES

High similarities considering all ranges of step cycles (>0.8) were found for individual muscle weightings and individual activation signals of all motor modules except M5, regardless the used EMG processing method for motor modules extraction (**Table 1**). However, EMG$_{SNG}$ exhibited reduced intra-subject similarity for individual muscle weightings in comparison to EMG$_{AVR}$ and EMG$_{CNC}$ for most of the modules (ANOVA One-Way, $p < 0.05$). In addition, EMG$_{SNG}$ also exhibited reduced intra-subject similarity for individual activation signals in comparison to EMG$_{AVR}$ for all motor modules (t-Student test, $p < 0.05$).

SIMILARITY AMONG EMG PROCESSING METHODS

Intra-subject similarity among methods (**Figure 4**) was high between EMG$_{SNG}$ vs. EMG$_{AVR}$ (similarity = 0.95 ± 0.09 considering all ranges of cycles numbers and the five motor modules), as well as between EMG$_{SNG}$ vs. EMG$_{CNC}$ (similarity = 0.94 ± 0.10). A 1-way ANOVA test for each motor module did not reveal any statistical differences ($p > 0.05$). Similarity between EMG$_{AVR}$ vs. EMG$_{CNC}$ was slightly reduced (0.92 ± 0.16), especially for M5 (0.80 ± 0.15). For the motor module related to leg swing (M3) similarity between EMG$_{SNG}$ vs. EMG$_{AVR}$ (0.96 ± 0.01) was slightly higher than the similarity between EMG$_{SNG}$ vs. EMG$_{CNC}$ (0.93 ± 0.01) and EMG$_{AVR}$ vs. EMG$_{CNC}$ (0.90 ± 0.01).

RECONSTRUCTED EMG FROM DIFFERENT CONCATENATIONS OF STEP CYCLES

The reconstruction of original EMG using muscle weightings from EMG$_{CNC}$ revealed that the lower the number of concatenated step cycles, the lower is the quality of reconstruction for longer concatenation periods (**Figure 5A**). On the other hand, the use of weightings from EMG$_{AVR}$ did not provide a similar reconstruction quality (**Figure 5B**). In a more detailed analysis concerning the reconstruction of 40 steps (**Figure 5C**), it was observed that the use of muscle weightings from EMG$_{CNC}$ provided higher reconstruction quality in comparison to EMG$_{AVR}$. Moreover, the highest reconstruction quality was achieved by using the concatenation of 20 steps (VAF = 0.8 ± 0.04, **Figure 5C**). In addition, the overall quality of reconstruction by using a given number of step cycles to reconstruct the EMG datasets of the different number of steps is shown in **Figure 5D**. It was also observed that EMG$_{CNC}$ provided higher reconstruction quality in comparison to EMG$_{AVR}$, and especially for EMG$_{CNC}$ the shorter the concatenation period, the poorer is the quality of reconstruction.

DISCUSSION

We studied the influence of the data structure (e.g., number of step cycles) and their processing (averaging/concatenation) on the EMG factorization analysis during locomotion. The results indicated that the number of step cycles and their pre-processing did not impact the estimated dimensionality and had a relatively small effect on the extracted motor modules, as intra-subject similarities demonstrated that these motor modules were predominantly similar regardless of the number of analyzed step cycles. However, further analyses applying muscle weightings from EMG$_{AVR}$ and shorter EMG$_{CNC}$ intervals to reconstruct longer locomotion intervals (e.g., 40 step cycles) demonstrated poor reconstruction quality, while optimal reconstructions were found by using at least 20 steps.

As expected, the extraction of motor modules from individual step cycles revealed a certain degree of step-to-step variability

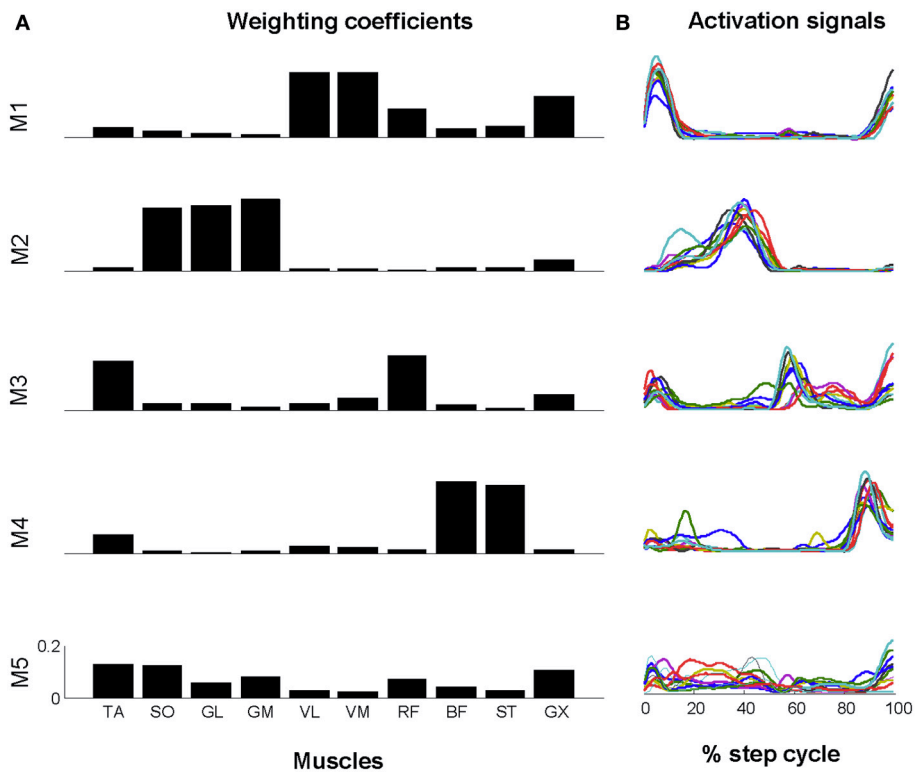


FIGURE 3 | Motor modules extracted from EMG_{AVR} of all subjects concatenated. Muscle weightings were averaged for illustration **(A)** and each activation signal for single subjects (each subject represented by a color) is shown in **(B)**. The first four activation signals exhibited well-defined bursts of activity at specific timing, however M5 did not presented clear pattern of activation throughout the step cycle, as well as between subjects.

Table 1 | Mean ± *SD* intra-subject similarities for each motor module (M1–M5) extracted by using surface EMG from single step cycles (EMG_{SNG}), averaged EMG (EMG_{AVR}), and concatenated EMG (EMG_{CNC}).

	M1	M2	M3	M4	M5
MUSCLE WEIGHTINGS					
EMG_{SNG}	0.89 ± 0.10*	0.89 ± 0.11*	0.86 ± 0.13	0.85 ± 0.03*	0.58 ± 0.14*
EMG_{AVR}	0.97 ± 0.02	0.97 ± 0.02	0.92 ± 0.12	0.98 ± 0.01	0.78 ± 0.18
EMG_{CNC}	0.94 ± 0.06	0.96 ± 0.02	0.86 ± 0.20	0.97 ± 0.02	0.78 ± 0.19
ACTIVATION SIGNALS					
EMG_{SNG}	0.92 ± 0.08*	0.93 ± 0.03*	0.82 ± 0.10*	0.92 ± 0.06*	0.60 ± 0.10*
EMG_{AVR}	0.99 ± 0.01	0.97 ± 0.02	0.91 ± 0.12	0.96 ± 0.05	0.79 ± 0.17

*For each subject similarity was computed across all ranges of step cycles, therefore it was not possible to compute similarities for the concatenated activation signals. *Denotes significant difference in relation to the other EMG processing methods.*

in the results. Because of this variability, when muscle weightings extracted from EMG_{AVR} or from EMG_{CNC} with small number of step cycles were fixed for reconstructing the original EMG data from different concatenation periods, the reconstruction quality was generally poor. These results suggest that, although different EMG processing methods can reveal predominantly similar vectors for weightings and timing properties, the details of muscle activities and their variability are better extracted by concatenating at least 20 step cycles. This number of step cycles, when concatenated, allowed to capture most of the step-by-step variability generated during longer periods of locomotion.

The reconstruction of original EMG datasets by using four motor modules was not high for TA, RF, and GX in most of the tested subjects. However, the four extracted modules were still consistent when five modules were extracted, and were comparable to those reported in previous literature (Ivanenko et al.,

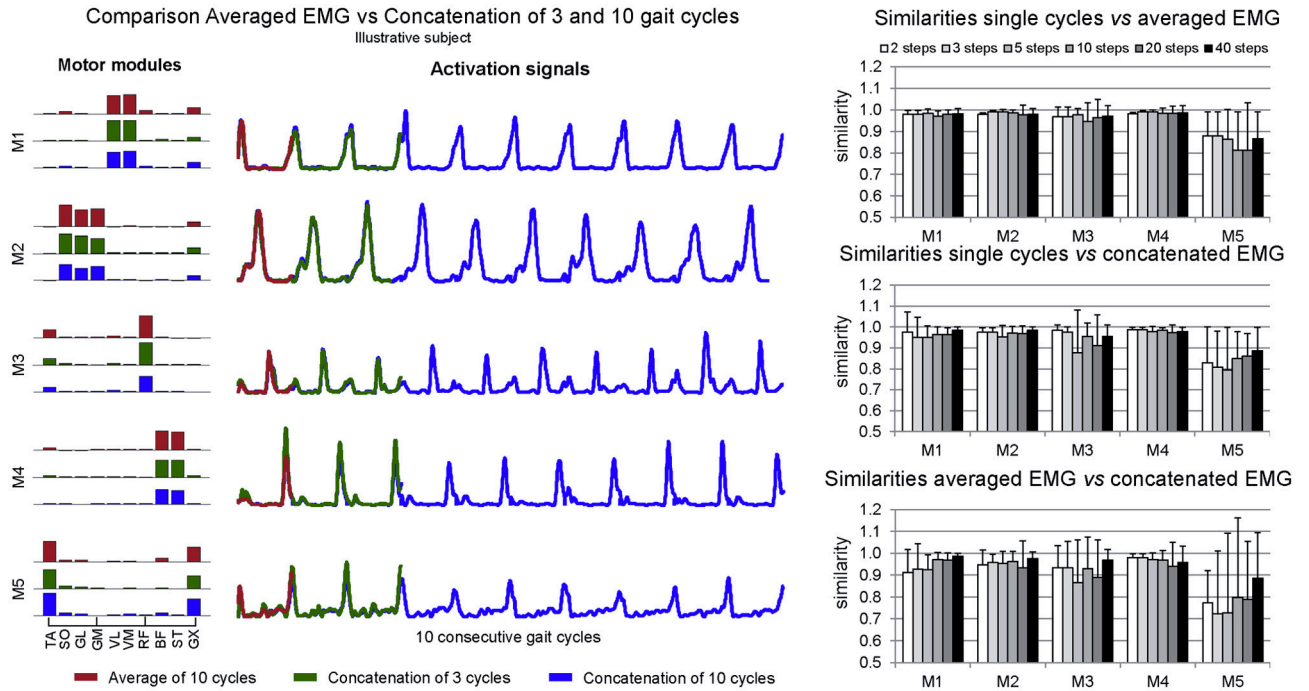


FIGURE 4 | Illustrative comparison of averaged EMG and concatenated EMG from a single subject (*left*). On the right, mean ± *SD* similarities between muscle weightings extracted from single trials vs. averaged EMG (*top*), from single trials vs. concatenated EMG (*middle*) and from averaged EMG vs. single trials (*bottom*), for each analyzed number of step cycles (from three to 40 step cycles).

2004; Clark et al., 2010; Oliveira et al., 2012). In our study, a fifth motor module that was not directly associated to a biomechanical subtask, was required to complement the activation of ankle joint muscles, and of the RF and GX muscles at the transition instances of the step cycle (swing-to-stance and stance-to-swing). This type of motor modules with relatively small biomechanical relevance has been previously reported by Monaco et al. (2010) who defined it as systematic information with robust inter-subject muscle groups, especially at high cadences. Ivanenko et al. (2004, 2005) also described less relevant motor patterns that could be dropped from the analysis because of their lack of significance. In the present study, the fifth motor module could only capture a marginal portion of the EMG variance and may not be necessary to understand the main features of the global EMG data. However, the consistently lower reconstruction quality for the same muscles when using four modules may indicate that additional temporal adjustments in muscle recruitment might be needed in order to produce optimal limb kinematics. The exact source of such activity can only be speculated, involving sensorial/afferent inputs to the muscles (Rossignol et al., 2006) and/or direct modulation from cortical neurons (Petersen et al., 2012). Gwin et al. (2011) have shown increased spectral power in the alpha and beta bands of cortical activity during step transitions, the predominant periods in which the described fifth module was recruited. Therefore, these additional components should not be disregarded while extracting motor modules if the purpose of the experiment requires high-quality EMG reconstruction.

The high similarity observed across EMG processing methods, including different numbers of step cycles, may initially suggest that single steps can be representative of all variability contained in longer locomotion periods. Therefore, we also analyzed the intra-subject variability that motor modules exhibited with the different EMG processing methods. This analysis showed a reduced similarity among the extracted motor modules from EMG_{SNG} in comparison to the other methods, suggesting that individual motor modules from EMG_{SNG} do not contain sufficient EMG variability for representing the EMG step pattern. When using the EMG_{AVR} or EMG_{CNC} datasets, the variability of the entire recording was included, either by averaging or by factorizing the whole signal interval, which explains the higher intra-subject similarity for these EMG processing methods. Although these results from longer ambulation periods were superior than those extracted from single trials, we also used a cross-validation procedure for verifying if the weightings could be shared between concatenation periods while generating successful reconstruction (Oliveira et al., 2013b). The use of muscle weightings from the concatenation of less than 10 step cycles reconstructed the original EMG from 40 step cycles by less than 70% whereas, when using 20 cycles, the VAF raised to 80% on average. This result suggests that a rather long locomotion period is preferable to optimally represent the modular organization of human locomotion and its variability over time. Interestingly, the use of weightings from EMG_{AVR} did not reach the same reconstruction quality as those from EMG_{CNC}, even though most of the weightings from these two conditions presented similarities

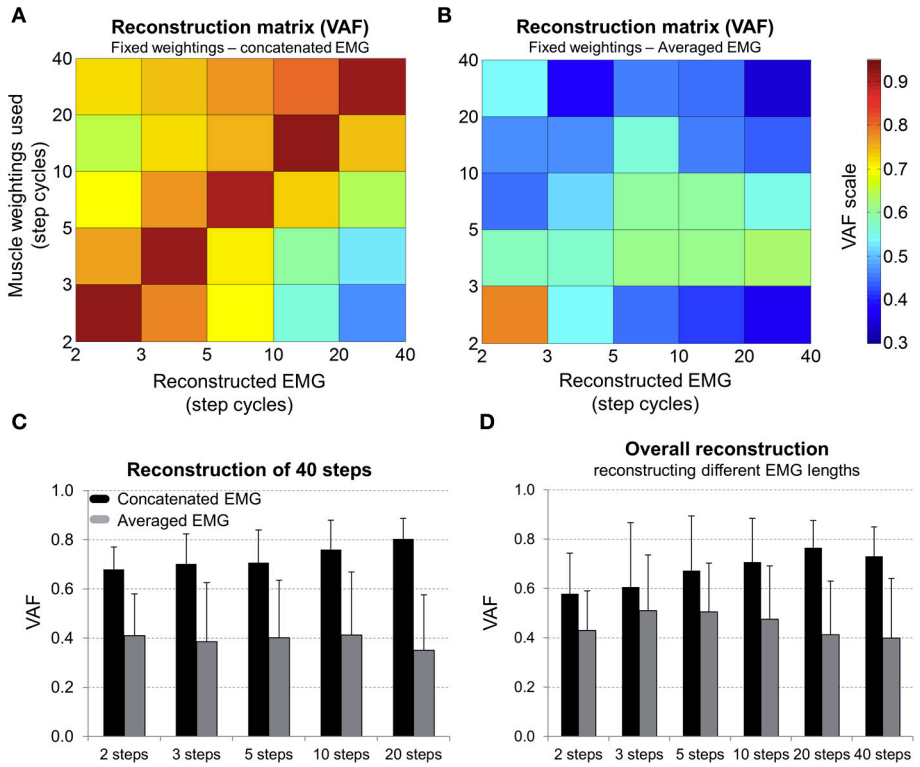


FIGURE 5 | (A) Colormap representing the variability accounted for (VAF) the reconstruction of surface EMG weightings by combining fixed muscle weightings from a given number of step cycles to the activation signals of a given number of step cycles. **(B)** Mean (SD) VAF of reconstructing EMG from 40 concatenated step cycles by using muscle weightings from the concatenation of 2–20 steps. **(C)** Mean (SD) VAF of reconstructing an entire 40 step cycles EMG dataset by using muscle weightings from the concatenation (*black bars*) and from the average of 2–20 continuous step cycles (*gray bars*). **(D)** Mean (SD) VAF displaying overall reconstruction quality achieved by using muscle weightings from the concatenation (*black bars*) and average (*gray bars*) of specific number of step cycles (from 2 to 40) to reconstruct all other step ranges.

above 90%. This observation may indicate the limitation of this similarity measure. Therefore, the comparison of results from factorization analysis of different tasks may not be exclusively based on similarity indexes, and the use of a cross-validation method such as fixing the muscle weightings in combination with timing properties of the signal to be reconstructed may be more valuable.

In the present investigation we recorded 10 lower limb muscles directly involved in locomotor mechanics. Previous investigations have recorded the EMG activity from up to 32 muscles and found five principal components that modulate muscle recruitment (Ivanenko et al., 2005), while other studies containing fewer muscles reported four motor modules (McGowan et al., 2010; Monaco et al., 2010; Gizzi et al., 2011). Our results are therefore in agreement with these previous reports and we speculate that the addition of other muscles such as hip extensors, adductors, and abductors may lead to an increased dimensionality. However the outcome of the methodological comparisons may be preserved if the results are extracted from locomotion at constant speed. The use of treadmill walking in this investigation provided an ideal model of locomotion in a controlled environment and at a fixed speed. The lack of kinematic measurements is a limitation of this investigation, however there is an extensive body of literature describing walking kinematics and its variability, and its relationship to EMG variability (Winter and Yack, 1987; Ivanenko et al., 2002; Kang and Dingwell, 2006). Indeed, despite the considerable EMG variability during locomotion, lower limb kinematics appear less variable (Winter and Yack, 1987; Ivanenko et al., 2002; Kang and Dingwell, 2006) due to inertial and damping properties of body segments that smoothen individual muscle force fluctuations (Kang and Dingwell, 2006, 2009). This observation supports the conclusion that muscle recruitment can be essentially modulated to control the overall limb kinematics (Ivanenko et al., 2002). Another limitation of this study is the subject sample of only healthy subjects. The results obtained may not be entirely applicable for clinical cases in which there is more variability in kinematics and muscle recruitment (Clark et al., 2010; Gizzi et al., 2011).

In summary, the present investigation showed that the dimensionality of motor modules was not influenced by the number of step cycles used for EMG factorization. We also noted that the dimensionality must be accurately defined such that the reconstruction of all the involved muscles reaches acceptable levels. In this experiment, four motor modules could account for most of the EMG variability and could be assigned to biomechanical subtasks, but for optimal muscle reconstruction a fifth motor module was required. In addition, although muscle weightings from the

factorization of different numbers of step cycles and processing methods are predominantly similar, the use of muscle weightings from the factorization of a sufficient number of concatenated step cycles can better represent locomotion over longer periods.

AUTHOR CONTRIBUTIONS

Anderson S. Oliveira, Leonardo Gizzi, Uwe G. Kersting, and Dario Farina designed the experiment. Anderson S. Oliveira and Uwe G. Kersting performed the experiments. Anderson S. Oliveira, Leonardo Gizzi, and Dario Farina analyzed and interpreted the data. Anderson S. Oliveira, Leonardo Gizzi, Uwe G. Kersting, and Dario Farina drafted the manuscript and all authors approved the final version. Experiments were performed at Aalborg University.

ACKNOWLEDGMENT

This project was partly sponsored by the EU Project "Integrative approach for the emergence of human like locomotion" (H2R; contract #600698).

REFERENCES

Barbero, M., Merletti, R., and Rainoldi, A. (2011). *Atlas of Muscle Innervation Zones*. New York, NY: Springer.

Cappellini, G., Ivanenko, Y. P., Poppele, R. E., and Lacquaniti, F. (2006). Motor patterns in human walking and running. *J. Neurophysiol.* 95, 3426–3437. doi: 10.1152/jn.00081.2006

Choi, J. T., and Bastian, A. J. (2007). Adaptation reveals independent control networks for human walking. *Nat. Neurosci.* 10, 1055–1062. doi: 10.1038/nn1930

Clark, D. J., Ting, L. H., Zajac, F. E., Neptune, R. R., and Kautz, S. A. (2010). Merging of healthy motor modules predicts reduced locomotor performance and muscle coordination complexity post-stroke. *J. Neurophysiol.* 103, 844–857. doi: 10.1152/jn.00825.2009

d'Avella, A., Saltiel, P., and Bizzi, E. (2003). Combinations of muscle synergies in the construction of a natural motor behavior. *Nat. Neurosci.* 6, 300–308. doi: 10.1038/nn1010

de Rugy, A., Loeb, G. E., and Carroll, T. J. (2013). Are muscle synergies useful for neural control? *Front. Comput. Neurosci.* 7:19. doi: 10.3389/fncom.2013.00019

Gizzi, L., Nielsen, J. F., Felici, F., Ivanenko, Y. P., and Farina, D. (2011). Impulses of activation but not motor modules are preserved in the locomotion of subacute stroke patients. *J. Neurophysiol.* 106, 202–210. doi: 10.1152/jn.00727.2010

Gizzi, L., Nielsen, J. F., Felici, F., Moreno, J. C., Pons, J. L., and Farina, D. (2012). Motor modules in robot-aided walking. *J. Neuroeng. Rehabil.* 9, 76. doi: 10.1186/1743-0003-9-76

Gwin, J. T., Gramann, K., Makeig, S., and Ferris, D. P. (2011). Electrocortical activity is coupled to gait cycle phase during treadmill walking. *Neuroimage* 54, 1289–1296. doi: 10.1016/j.neuroimage.2010.08.066

Hug, F., Turpin, N. A., Dorel, S., and Guével, A. (2012). Smoothing of electromyographic signals can influence the number of extracted muscle synergies. *Clin. Neurophysiol.* 123, 1895–1896. doi: 10.1016/j.clinph.2012.01.015

Ivanenko, Y., Grasso, R., Macellari, V., and Lacquaniti, F. (2002). Control of foot trajectory in human locomotion: role of ground contact forces in simulated reduced gravity. *J. Neurophysiol.* 87, 3070–3089. doi: 10.11512/jn.00815.2001

Ivanenko, Y. P., Cappellini, G., Dominici, N., Poppele, R. E., and Lacquaniti, F. (2005). Coordination of locomotion with voluntary movements in humans. *J. Neurosci.* 25, 7238–7253. doi: 10.1523/JNEUROSCI.1327-05.2005

Ivanenko, Y. P., Poppele, R. E., and Lacquaniti, F. (2004). Five basic muscle activation patterns account for muscle activity during human locomotion. *J. Physiol.* 556, 267–282. doi: 10.1113/jphysiol.2003.057174

Kang, H. G., and Dingwell, J. B. (2006). A direct comparison of local dynamic stability during unperturbed standing and walking. *Exp. Brain Res.* 172, 35–48. doi: 10.1007/s00221-005-0224-6

Kang, H. G., and Dingwell, J. B. (2009). Dynamics and stability of muscle activations during walking in healthy young and older adults. *J. Biomech.* 42, 2231–2237. doi: 10.1016/j.jbiomech.2009.06.038

Kersting, U. G. (2011). Regulation of impact forces during treadmill running. *Footwear Sci.* 3, 59–68. doi: 10.1080/19424280.2011.552074

Lacquaniti, F., Ivanenko, Y. P., and Zago, M. (2012). Patterned control of human locomotion. *J. Physiol.* 590, 2189–2199. doi: 10.1113/jphysiol.2011.215137

Lee, D. D., and Seung, H. S. (1999). Learning the parts of objects by non-negative matrix factorization. *Nature* 401, 788–791. doi: 10.1038/44565

Lee, D. D., and Seung, H. S. (2001). Algorithms for non-negative matrix factorization. *Adv. Neural Inf. Process. Syst.* 13, 556–562.

McGowan, C. P., Neptune, R. R., Clark, D. J., and Kautz, S. A. (2010). Modular control of human walking: adaptations to altered mechanical demands. *J. Biomech.* 43, 412–419. doi: 10.1016/j.jbiomech.2009.10.009

Merkle, L., Layne, C., Bloomberg, J., and Zhang, J. (1998). Using factor analysis to identify neuromuscular synergies during treadmill walking. *J. Neurosci. Methods* 82, 207–214. doi: 10.1016/S0165-0270(98)00054-5

Monaco, V., Ghionzoli, A., and Micera, S. (2010). Age-related modifications of muscle synergies and spinal cord activity during locomotion. *J. Neurophysiol.* 104, 2092–2102. doi: 10.1152/jn.00525.2009

Moreno, J. C., Barroso, F., Farina, D., Gizzi, L., Santos, C., Molinari, M., et al. (2013). Effects of robotic guidance on the coordination of locomotion. *J. Neuroeng. Rehabil.* 10:79. doi: 10.1186/1743-0003-10-79

Muceli, S., Boye, A. T., d'Avella, A., and Farina, D. (2010). Identifying representative synergy matrices for describing muscular activation patterns during multidirectional reaching in the horizontal plane. *J. Neurophysiol.* 103, 1532–1542. doi: 10.1152/jn.00559.2009

Oliveira, A. S., Gizzi, L., Kersting, U. G., and Farina, D. (2012). Modular organization of balance control following perturbations during walking. *J. Neurophysiol.* 108, 1895–1906. doi: 10.1152/jn.00217.2012

Oliveira, A. S., Silva, P. B., Lund, M. E., Gizzi, L., Farina, D., and Kersting, U. G. (2013b). Effects of perturbations to balance on neuromechanics of fast changes in direction during locomotion. *PLoS ONE* 8:e59029. doi: 10.1371/journal.pone.0059029

Oliveira, A. S., Silva, P. B., Lund, M. E., Kersting, U. G., and Farina, D. (2013a). Fast changes in direction during human locomotion are executed by impulsive activation of motor modules. *Neuroscience* 228, 283–293. doi: 10.1016/j.neuroscience.2012.10.027

Petersen, T. H., Willerslev-Olsen, M., Conway, B. A., and Nielsen, J. B. (2012). The motor cortex drives the muscles during walking in human subjects. *J. Physiol.* 590, 2443–2452. doi: 10.1113/jphysiol.2012.227397

Rossignol, S., Dubuc, R., and Gossard, J. P. (2006). Dynamic sensorimotor interactions in locomotion. *Physiol. Rev.* 86, 89–154. doi: 10.1152/physrev.00028.2005

Sartori, M., Gizzi, L., and Farina, D. (2013a). "Musculoskeletal modeling of human locomotion based on low-dimensional impulsive activation signals: perspectives for neurotechnologies," in *Converging Clinical and Engineering Research on Neurorehabilitation,* (New York, NY: Springer), 1239–1242. doi: 10.1007/978-3-642-34546-3_206

Sartori, M., Gizzi, L., Lloyd, D. G., and Farina, D. (2013b). A musculoskeletal model of human locomotion driven by a low dimensional set of impulsive excitation primitives. *Front. Comput. Neurosci.* 7:79. doi: 10.3389/fncom.2013.00079

Steele, K. M., Tresch, M. C., and Perreault, E. J. (2013). The number and choice of muscles impact the results of muscle synergy analyses. *Front. Comput. Neurosci.* 7:105. doi: 10.3389/fncom.2013.00105

Winter, D., and Yack, H. (1987). EMG profiles during normal human walking: stride-to-stride and inter-subject variability. *Electroencephalogr. Clin. Neurophysiol.* 67, 402–411. doi: 10.1016/0013-4694(87)90003-4

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.

Received: 13 December 2013; accepted: 03 May 2014; published online: 23 May 2014.
*Citation: Oliveira AS, Gizzi L, Farina D and Kersting UG (2014) Motor modules of human locomotion: influence of EMG averaging, concatenation, and number of step cycles. Front. Hum. Neurosci. **8**:335. doi: 10.3389/fnhum.2014.00335*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

REVIEW ARTICLE
published: 13 March 2014
doi: 10.3389/fnhum.2014.00141

Who is going to walk? A review of the factors influencing walking recovery after spinal cord injury

Giorgio Scivoletto[1,2]*, *Federica Tamburella*[1,2], *Letizia Laurenza*[1], *Monica Torre*[1] and *Marco Molinari*[1,2]

[1] Spinal Cord Unit, IRCCS Fondazione S. Lucia, Rome, Italy
[2] Clinical and Research Movement Analysis Lab, Fondazione S. Lucia, Rome, Italy

Edited by:
Nadia Dominici, Swiss Federal Institute of Technology in Lausanne, Switzerland

Reviewed by:
John Francis Ditunno, Thomas Jefferson University, USA
Armin E. P. Curt, University Hospital Balgrist, Switzerland
Kristin Musselman, University of Saskatchewan, Canada

***Correspondence:**
Giorgio Scivoletto, Spinal Cord Unit, IRCCS Fondazione S. Lucia, Via Ardeatina 306, 00179 Rome, Italy
e-mail: g.scivoletto@hsantalucia.it

The recovery of walking function is considered of extreme relevance both by patients and physicians. Consequently, in the recent years, recovery of locomotion become a major objective of new pharmacological and rehabilitative interventions. In the last decade, several pharmacological treatment and rehabilitative approaches have been initiated to enhance locomotion capacity of SCI patients. Basic science advances in regeneration of the central nervous system hold promise of further neurological and functional recovery to be studied in clinical trials. Therefore, a precise knowledge of the natural course of walking recovery after SCI and of the factors affecting the prognosis for recovery has become mandatory. In the present work we reviewed the prognostic factors for walking recovery, with particular attention paid to the clinical ones (neurological examination at admission, age, etiology gender, time course of recovery). The prognostic value of some instrumental examinations has also been reviewed. Based on these factors we suggest that a reliable prognosis for walking recovery is possible. Instrumental examinations, in particular evoked potentials could be useful to improve the prognosis.

Keywords: spinal cord injury, walking recovery, prognostic factors

INTRODUCTION

Walking recovery is one of the main goals of patients after SCI: walking is rated at first place (together with bladder and bowel function) at least by patients with incomplete lesions (Ditunno et al., 2008a). Furthermore, an epidemiological study shows an increase of the number of patients with incomplete lesions (e.g., with chances of walking recovery) (Pagliacci et al., 2003). Therefore, the recovery of ambulation has become the target of several pharmacological and rehabilitative approaches (Wernig and Muller, 1992; Domingo et al., 2012) and a precise evaluation of the natural recovery of walking and of the prognostic factors influencing this function has become mandatory (Steeves et al., 2007).

In the present work we reviewed the effect of several clinical and demographic features on the prognosis for walking recovery. Furthermore, because one of the main problems of the acute phase of SCI is the lack of reliable examinations, we considered the prognostic value of neurophysiological and neuroimaging examinations.

Finally, the effect of early pharmacological and surgical interventions on walking recovery will be examined.

MATERIALS AND METHODS

A systematic search was performed of all papers as well as websites mentioning spinal cord injury and walking The literature search was conducted without time limits to identify papers that explicitly mentioned the walking capacity in patients with SCI. Databases included PubMed, Ovid MEDLINE, CINAHL, PsychINFO, Cochrane Central Register of Controlled Trials and Scopus, which includes Embase citations. All study designs, including case reports, were included, with no restrictions on the ages of participants. Non-English articles and animal studies were excluded. The following search terms were used: prognosis prediction, SCI, paraplegia/tetraplegia/quadriplegia, ambulation/gait and walking/walking capacity. In addition, other databases, such as Google and a hand search of Spinal Cord yielded other citations not identified by the above strategy.

Two authors (Giorgio Scivoletto and Federica Tamburella) independently identified and classified the papers through a review of the abstracts, texts, and references and circulated them to the authors' panel.

CLINICAL EXAMINATION

The most relevant prognostic factor for functional recovery in SCI patients is the neurological status at the moment of the first examination. The physical examination of these patients has been standardized by the American Spinal Injury Association in the International Standards for Neurological Classification of Spinal Cord Injury (ISNCSCI) (American Spinal Injury Association, 2000). Based on this examination it is possible to establish the neurological level of injury, as well as the severity of the lesion (impairment). Components also include a rectal examination for voluntary anal contraction and anal sensation (**Figures 1**, **2**). Patients are considered to have a complete lesion (AIS impairment A), according to the ASIA Impairment Scale (AIS), in the absence of sensory or motor function at the lowest sacral segments. Incomplete lesions are defined when sensation and/or motor function are preserved below the neurologic level of injury, and in particular in the lowest sacral segments (anal sensation, including deep anal pressure and voluntary external anal sphincter contraction) (**Figure 2**).

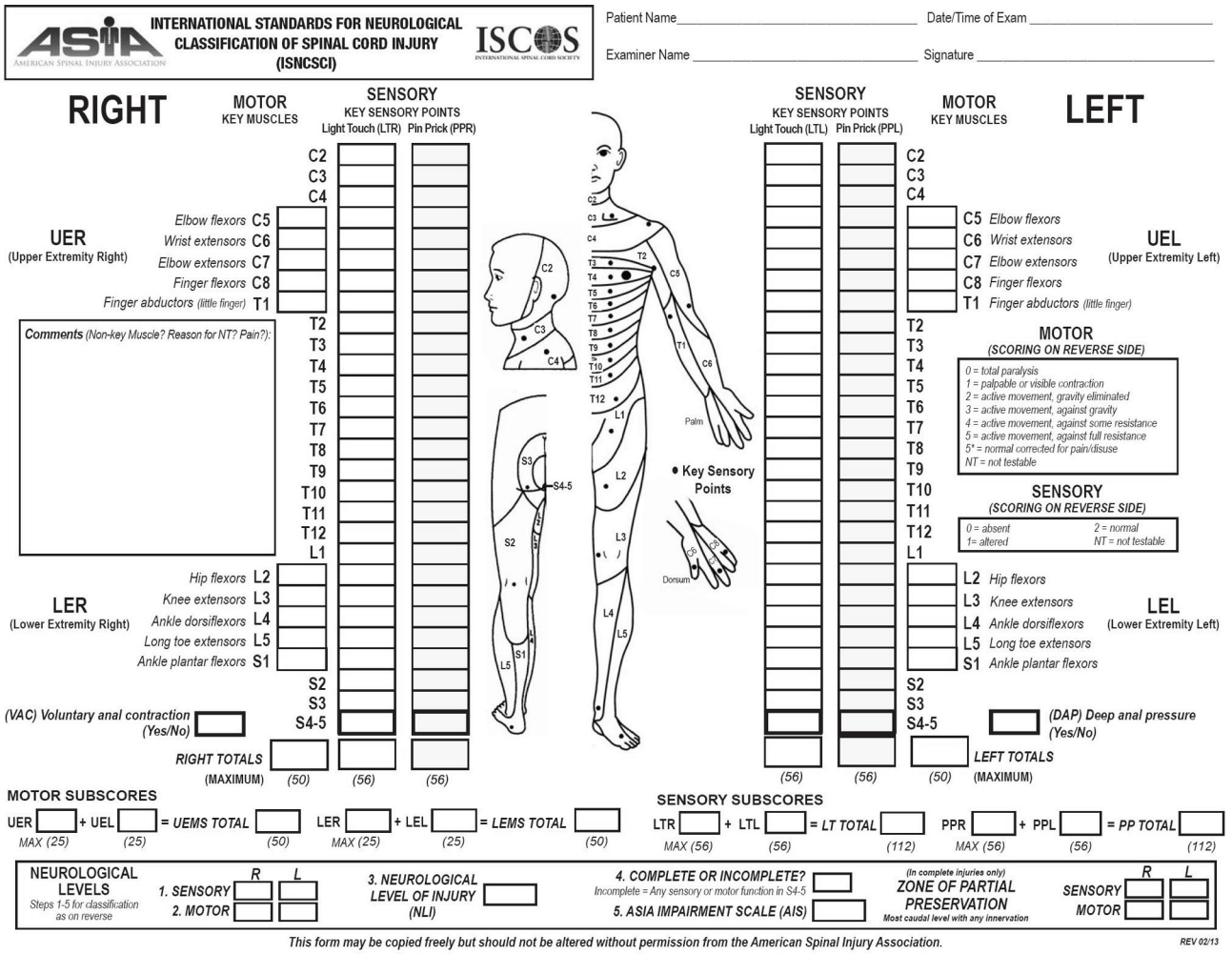


FIGURE 1 | Scoring sheet for the International Standards for Neurological Classification of Spinal Cord Injury. American Spinal Injury Association: International Standards for Neurological Classification of Spinal Cord Injury, revised 2013; Atlanta, GA. Reprinted 2013.

This examination should usually be performed at 72 h after the lesion because this timing seems to have a more accurate prognostic value than earlier assessment (Herbison et al., 1991).

AIS GRADE CONVERSION AND WALKING RECOVERY

For the aim of this review we would define walking recovery as the regained ability to walk independently in the community, with or without the use of devices and braces. This is also defined "functional walking" and has been described by several authors (Hussey and Stauffer, 1973) as the capacity to walk reasonable distances both in and out of home unassisted by another person.

For a long time AIS grade conversion has been considered the basis to predict the possibility of achieving functional walking. However, a recent article by van Middendorp et al. (2009) questioned the relationship between AIS grade conversion and ability to walk as we will show below.

Patients with AIS impairment A (motor and sensory complete lesion) at their first examination have very few chances of neurological recovery below the lesion. When the examination is performed at 72 h post-injury, 80% of the initial AIS A patients remain as AIS A, with about 10% converting to AIS B (i.e., some sensory function) and about 10% converting to AIS C (with some motor recovery below the lesion) (Burns et al., 2012). However, if the first examination is performed later, the percentage of improvement decreases dramatically to 2.5% (Scivoletto et al., 2004a) (**Table 1**). Accordingly, the possibility of patients with AIS impairment A of achieving functional walking is very limited too. Furthermore, also between the patients who converted to an incomplete lesion only 14% recovered some walking function (van Middendorp et al., 2009). The AIS A patients who achieve some walking function usually are low thoracic or lumbar levels (T12-L3) and need braces and devices to walk (Ditunno et al., 2008b; **Table 2**). Finally, these patients are usually limited ambulators, with slow average velocities and great energy expenditure (Vaccaro et al., 1997).

AIS grade B patients (those with motor complete, sensory incomplete lesion at 72 h examination) usually show some motor recovery and they can convert to AIS C or even AIS D grade. However, the overall recovery of ambulation is considered to be about 33% (Katoh and el Masry, 1995; van Middendorp et al.,

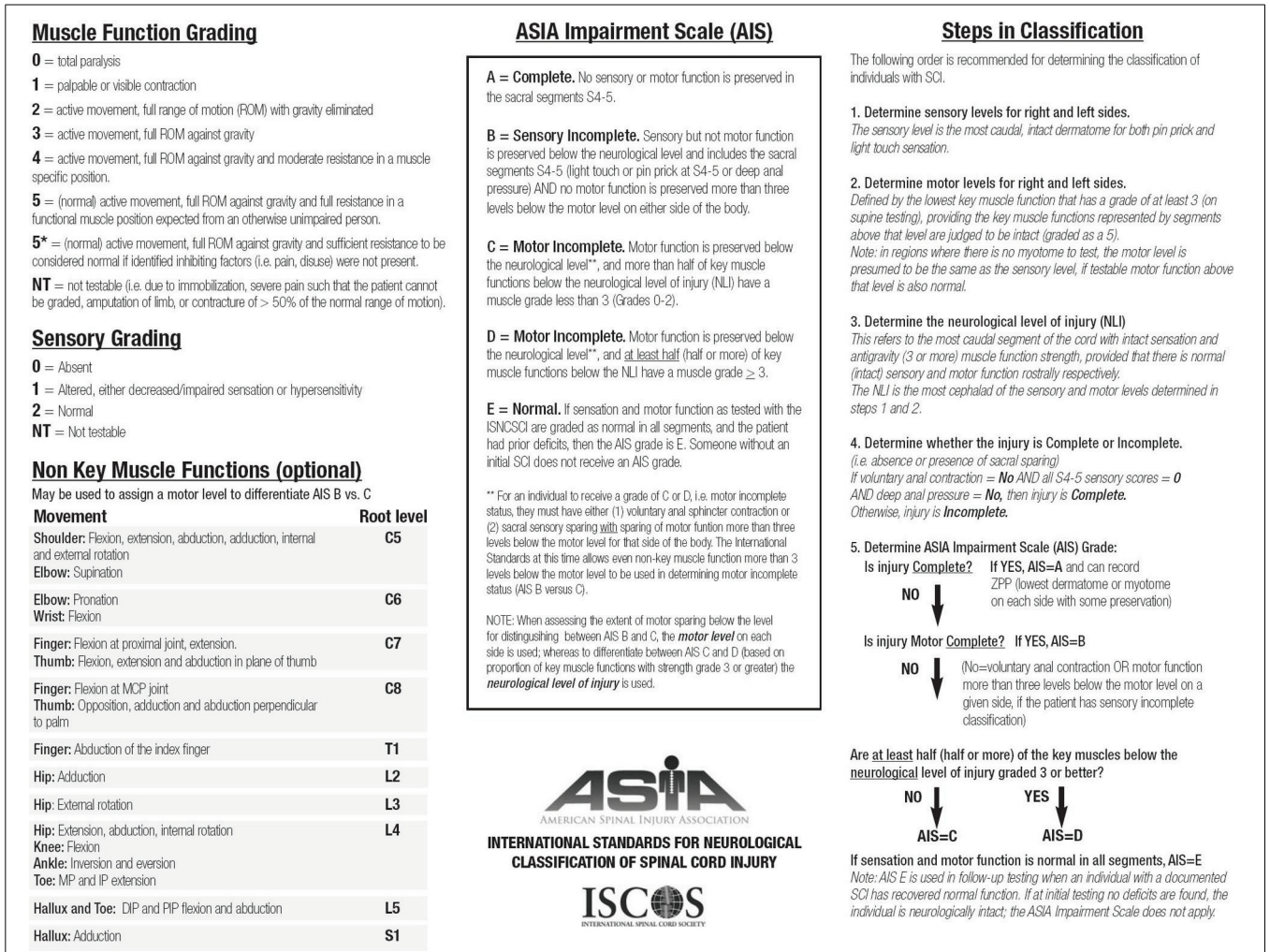


FIGURE 2 | Scoring sheet for the International Standards for Neurological Classification of Spinal Cord Injury. American Spinal Injury Association: International Standards for Neurological Classification of Spinal Cord Injury, revised 2013; Atlanta, GA. Reprinted 2013.

Table 1 | Prediction of recovery according to AIS impairment scale.

AIS grade at admission	A	B	C	D
First examination at 72 h[10]	**One-year follow-up AIS grade**			
A	84%	8%	5%	3%
B	10%	30%	29%	31%
C	2%	2%	25%	67%
D	2%	1%	2%	85%
First examination at 30 days[11]	**One-year follow-up AIS grade**			
A	95%	0	2,5%	2,5%
B	0	53%	21%	26%
C	1%	0	45%	54%
D	2%	0	0	96%

2009). The percentage of walking recovery may vary depending on the modality of the sensation spared at the lowest sacral segments. Several studies reported a relationship between pinprick preservation and recovery in AIS B patients. AIS grade B patients with pinprick preservation have a better walking recovery than those with light touch only (Foo et al., 1981; Crozier et al., 1991; Waters et al., 1994a; Katoh and el Masry, 1995; Oleson et al., 2005) (**Table 2**). This finding has an anatomical basis at the spinal cord level. The preservation of pinprick perception together with light touch one indicates less extensive damage to the spino-thalamic tracts and posterior column. Therefore, in these cases, there is a high likelihood of some sparing of the motor pathways conveyed by the nearby cortico-spinal tracts (Oleson et al., 2005).

Motor incomplete (AIS C) patients have a better prognosis for walking recovery than sensory incomplete ones. The overall rate of recovery is about 75% (Maynard et al., 1979; Crozier et al., 1992; Waters et al., 1994b; van Middendorp et al., 2009). This percentage includes both the patients who converted to AIS D and those who remained AIS C but achieve at least some walking function (van Middendorp et al., 2009); these patients probably have low thoracic or lumbar lesions and walk with braces and devices. Several factors may influence the chance of walking recovery in these patients: lower extremity strength, motor recovery timing, age and upper extremity strength for tetraplegic patients are the

Table 2 | Prediction of functional walking according to AIS impairment and other features.

AIS/lesion level at admission	Functional walking/ authors (references)
AIS A/cervical lesion	0% (Waters et al., 1994a,b) 0% (Ditunno et al., 2008b)
AIS A/thoracic and lumbar lesions	5% (Waters et al., 1994a,b) 8.5% (Ditunno et al., 2008b)
AIS at admission and sensation	**% recovery of community ambulation at 1 year post-injury/authors (references)**
AIS B (only light touch preservation)	0% (Waters et al., 1994a,b) 11% (Crozier et al., 1991) 33% (Waters et al., 1994a,b)
AIS B (light touch + pin prick preservation)	89% (Crozier et al., 1991) 66% (Foo et al., 1981) 75% (Katoh and el Masry, 1995)
AIS at admission and age	**% recovery of community ambulation at 1 year post-injury/authors (references)**
AIS C < 50 years	91% (Burns et al., 1997) 71% (Scivoletto et al., 2003)
AIS C > 50 years	42% (Burns et al., 1997) 25% (Scivoletto et al., 2003)
AIS D < 50 years	100% (Burns et al., 1997) 100% (Scivoletto et al., 2003)
AIS D > 50 years	100% (Burns et al., 1997) 80% (Scivoletto et al., 2003)

most important ones (Crozier et al., 1992; Waters et al., 1994b). In AIS C patients age seems to be a strong prognostic factor for walking recovery. Age represents a clear negative prognostic factor for walking recovery: AIS C subjects younger than 50 years have a chance of achieving functional walking of 80–90%, but this percentage dramatically decreases to 30–40% in older patients (**Table 2**) (Perot and Vera, 1982; Foo, 1986; Burns et al., 1997; Scivoletto et al., 2003). Different hypotheses have been offered to explain the negative effect of age. The functional potential for a given neurological deficit is lower at older age; this may be considered reasonable since functional abilities generally decline as people's age increases. In normal ageing "reserve (peak) capacity" (or "vitality") (DiGiovanna, 2000) seems to peak at around 30 years of age, and then gradually declines until death. Disease processes, including SCI and its complications, are considered to accelerate this process of decline. Jakob et al. (2009) offers another possible explanation. In his study he found that age is not correlated with neurologic recovery, but is correlated with a worse functional outcome in terms of independence in daily life activities and walking function. He therefore suggested that the neurological recovery is not directly related to the functional outcome and that elderly patients have difficulties in translating neurological recovery into positive functional changes.

Finally, AIS D patients at admission have very good ambulation prognosis at 1 year post-injury (Burns et al., 1997; Scivoletto et al., 2003). All patients, regardless of age, who initially were classified as AIS D (within 72 h) were able to walk at the time of discharge from inpatient rehabilitation (Burns et al., 1997; van Middendorp et al., 2009).

OTHER CLINICAL FACTORS

In addition to AIS grade, several other factors evaluated at 72 h after the lesion have been considered in the prognosis of walking recovery and are examined below.

REFLEXES

In the very early examination of SCI patients the presence/absence of the delayed plantar response (DPR) must be assessed. DPR is characterized by a delayed response to an unusually strong stimulus to the sole of the foot (Weinstein et al., 1997). The onset of this response following the stimulus could be 500 ms or a full second following the initiation of the stimulus (Weinstein et al., 1997). The DPR shows a reciprocal relationship with the Babinski sign and it is particularly relevant because it allows the prognosis during the spinal shock phase (Ko et al., 1999). The DPR is a negative prognostic indicator as it is more often present and lasts longer (more than 1 day) in SCI patients who do not recover any voluntary movement (Weinstein et al., 1997; Ko et al., 1999).

SYNDROMES

Based on the distribution of sensory and motor loss, the ISNCSCI allow to identify several incomplete spinal cord syndromes with different prognostic values.

The *central cord syndrome (CCS)* is mostly seen following cervical lesion. It represents about 9% of the total SCIs and 44% of the clinical syndromes (Mckinley et al., 2007) and is characterized by a greater involvement of the upper extremities than the lower extremities. The CCS is a clinical picture that recognizes several causes (with and without bone injury) and several different mechanisms (including direct injury of the spinal cord or vascular injuries) (Mckinley et al., 2007) that primarily affects the center of the spinal cord and generally has a favorable prognosis as to independence in daily life activities and bladder and bowel function recovery (Newey et al., 2000; Dvorak et al., 2005; Aito et al., 2006). Because of the lesser involvement of the lower extremities, CCS is considered to have a good prognosis for walking recovery too (Merriam et al., 1986; Penrod et al., 1990; Roth et al., 1990; Burns et al., 1997; Aito et al., 2006). The percentage of patients who recover walking varies from 40 to 97%, but is strongly influenced by age. Several studies confirm that younger patients (less than 50 years old) have twice the chance of achieving independent walking than older ones (Foo, 1986; Merriam et al., 1986; Penrod et al., 1990; Roth et al., 1990; Burns et al., 1997; Newey et al., 2000; Dvorak et al., 2005; Aito et al., 2006).

The *Brown-Séquard syndrome (BSS)* is characterized by ipsilateral hemiplegia and contralateral hemianalgesia due to spinal hemisection (Brown-Sequard, 1868). It accounts for 2–4% of all traumatic SCIs and 17% of the clinical syndromes (Mckinley et al., 2007). The pure form of BSS is rarely seen and the Brown-Séquard Plus Syndrome (relative ipsilateral hemiplegia with a relative contralateral hemianalgesia) is much more frequent (Roth et al., 1991). BSS is more frequent at cervical level and is usually associated with stab-wound injuries (Gentleman and Harrington,

1984). BSS is characterized by a good functional prognosis. About 75% of patients achieve independent walking at discharge from rehabilitation (Stahlman and Hanley, 1992). In this framework an important predictor for walking recovery is the distribution of the impairment: if the upper limb is weaker than the lower limb, then patients are more likely to ambulate at discharge (Kirshblum and O'Connor, 1998).

The *anterior cord syndrome* is due to a lesion that involves the anterior two thirds of the spinal cord and preserves the posterior columns (Maynard et al., 1997), and account for 1% of all the SCIs and 5% of the clinical syndromes (Mckinley et al., 2007). It may derive from a retropulsed disc or bone fragments (Bauer and Errico, 1991), direct injury to the anterior spinal cord, or with lesions of the anterior spinal artery that provides the blood supply to that tract of spinal cord (Cheshire et al., 1996). Lesions of the anterior spinal artery may result from diseases of the aorta, cardiac or aortic surgery, embolism, polyarteritis nodosa, or angioplasty (Cheshire et al., 1996). Anterior cord syndrome is characterized by a variable loss of motor as well as pinprick sensation with a relative preservation of light touch, proprioception, and deep-pressure sensation. Due to the massive involvement of the anterior and lateral spinal cord with inclusion of the corticospinal tracts, only 10–20% of the patients with an anterior cord syndrome have the chance to recover muscle function, and even in those with some recovery, usually motor strength is low and coordination is lacking; consequently these patients have low walking recovery chances (Bohlman, 1979).

Etiology of the lesion

Most of the literature on SCI is focused on the rehabilitation of traumatic patients, despite the relevant incidence of non-traumatic lesions, considered to account for a percentage of the total SCIs varying from 30 to 80% (Buchan et al., 1972; Celani et al., 2001; Citterio et al., 2004). Patients with non-traumatic lesions differ from their traumatic counterparts for several prognostic factors. They are usually older, with a more even distribution of genders and a higher frequency of incomplete lesions. Therefore, a direct comparison of these two populations is difficult (Scivoletto et al., 2011). However, when the confounding effect of these factors is eliminated by means of statistics, patients with non-traumatic spinal cord lesions can achieve comparable rates of functional gains as their traumatic spinal cord injury counterparts (Mckinley et al., 2000; McKinley et al., 2001; Mckinley et al., 2002). With regard to walking function, recently a number of articles compared the recovery of ambulation in traumatic and non-traumatic SCIs and found that the two populations achieve comparable walking capacity with an overall percentage of patients varying from 35 (Scivoletto et al., 2011) to 49% (Marinho et al., 2012).

GENDER

There are only few studies on gender related differences in neurological and functional outcomes after inpatient rehabilitation of SCI (Greenwald et al., 2001; Scivoletto et al., 2004b; Sipski et al., 2004). Two of them (Greenwald et al., 2001; Scivoletto et al., 2004b) found no significant differences between the two genders with regard to daily life independence, motor efficiency, American Spinal Injury Association motor scores (Greenwald et al., 2001) and walking function (Scivoletto et al., 2004b). However, Sipski et al. (2004) found gender-related differences in daily life independence, but did not specifically focus on walking recovery. Women with SCI may have more natural neurologic recovery than men, but, for a given level and degree of neurologic injury, men tend to do better functionally than women at time of discharge from rehabilitation (Sipski et al., 2004).

FORMULAS AND ALGORITHMS

In the last three decades several attempts have been made to link one or more of the above mentioned factors (and of the results of instrumental examinations discussed below) to the prognosis for walking recovery.

Waters et al. (1994b) examined the relationship between lower extremity strength at first examination in incomplete paraplegics and walking recovery: all patients with an initial (1-month) lower extremity motor score of $\geq$10 points ambulated in 1 year. Seventy percent of patients with an initial motor score between 1 and 9 ambulated at 1 year. Furthermore, all patients with an initial hip flexor or knee extensor Grade $\geq$2 ambulated in the community at 1 year.

The same author examined the odds of walking recovery in incomplete tetraplegics and found that, although the relationship between initial lower extremity motor score and walking holds true for tetraplegics, these patients have less chance to achieve ambulation (Waters et al., 1994a): 63% of the patients with an initial lower extremity motor score of $\geq$10 points ambulated by 1 year, vs. 21% of those with an initial motor score between 1 and 9 (Waters et al., 1994a). In addition, Waters stressed the relationship between upper extremities strength and ambulation recovery in tetraplegics: patients who are community or household ambulators have significant higher motor scores. The author linked this datum to the importance of upper extremities strength for devices use during walking (Waters et al., 1994a).

Crozier et al. (1992) focused on the timing of recovery of lower extremity motor strength and concluded that early recovery of quadriceps strength is an excellent prognostic factor for ambulation. All patients with an initial quadriceps strength of at least Grade 2/5 who attained a grade of $\geq$3/5 in at least one quadriceps by 2 months post-injury achieved functional ambulation (ability to walk independently in the community, with or without the use of devices and braces) at follow-up. However, only 25% of those who did not recover quadriceps strength of 3/5 within 2 months were able to walk at follow-up.

More recently, Zörner et al. (2010) developed an algorithm based on outcome predictors and aimed at identifying subgroups of patients in the sub-acute phase who could achieve functional walking. For patients with incomplete paraplegia, lower extremity motor scores, pinprick scores and age were the best predictors for walking recovery. For patients with incomplete tetraplegia the more reliable predictors were the lower extremity motor scores, the tibial SSEP score and the AIS grade.

In 2011 van Middendorp et al. (2011) produced a simple clinical prediction rule based on the combination of age ($<$65 vs. $\geq$65 years), motor scores of the quadriceps femoris (L3), gastrocsoleus (S1) muscles, and light touch sensation of dermatomes

L3 and S1. This rule showed an excellent discrimination capacity in recognizing patients who achieved independent ambulation (ability to walk independently, with or without braces and orthoses for <10 m) at follow-up from those who were dependent walkers or non-walkers.

INSTRUMENTAL EXAMINATION

SOMATOSENSORY EVOKED POTENTIALS (SSEPs) (Table 3)

SSEPs are used for clinical diagnosis in patients with neurologic disease, and many studies have been performed to determine the value of SSEPs in the prediction of walking recovery in SCI patients (Young and Dexter, 1979; Kaplan and Rosen, 1981; Young, 1985; Foo, 1986; Ziganow, 1986; Katz et al., 1991; Aalfs et al., 1993; Jacobs et al., 1995; Curt and Dietz, 1997).

Most of these studies conclude that early SSEPs can predict motor improvement and ambulation outcome in SCI patients. However, SSEPs do not seem to offer additional prognostic accuracy if compared to clinical examination according to the ISNCSCI for both complete and incomplete patients (Young and Dexter, 1979; Kaplan and Rosen, 1981; Perot and Vera, 1982; Chabot et al., 1985; Katz et al., 1991; Aalfs et al., 1993; Curt and Dietz, 1997).

When a reliable clinical examination, together with the ISNCSCI is impossible (patients unresponsive, for example because sedated or under the effect of alcohol or drugs, or uncooperative, for example because of pain) then SSEPs are helpful to determine if they have SCI (Curt and Dietz, 1997). In addition, SSEPs may be helpful to differentiate between SCI and hysteric paraplegia, a differential diagnosis that may be very difficult (Kaplan et al., 1985).

Motor Evoked Potentials (MEPs) (Table 3)

Transcranial magnetic stimulation allows an examination of the conductivity of the motor tracts following cortical or spinal lesions in humans. According to a study of Curt, MEPs can contribute toward diagnosing lesions of different neurologic structures within the spinal cord and in predicting the recovery of functional movements (Curt et al., 1998). The study shows that MEPs recordings are sensitive to indicate motor tract lesions in approximately 90% of SCI patients and predictive for the recovery of upper and lower limb motor function. In this sense they are of similar prognostic value to clinical examination in the prediction of functional recovery. MEPs can be used in combination with the ASIA protocol to follow the recovery of clinical motor functions in relation to that of descending motor tracts for impulse transmission. In Curt's study, MEPs were highly predictive of ambulatory capacity. All patients with elicitable MEPs at initial examination recovered a muscle strength of 3/5 or more of the respective muscles. Not surprisingly, MEPs recordings in SCI patients are more sensitive than SSEPs recordings for revealing the involvement of motor tract fibers and are at least as sensitive as the ASIA protocol in predicting the resulting functional deficit. Similarly to SSEPs, the use of MEP recordings is mostly appropriate in patients who are uncooperative (approximately 15% of patients with acute SCI) (Bozzo et al., 2011).

Table 3 | Prognostic value of SSEPs and MEPs.

	Six months walking capacity			
	Normal (%)	Functional (%)	Therapeutic (%)	No walking function (%)
LOWER LIMBS SSEPS AND AMBULATION (Curt and Dietz, 1997)				
Intial SSEP evaluation				
Normal	83	17	0	0
Present, altered	10	60	10	20
Absent	0	7	13	80
LOWER LIMBS MEP AND AMBULATION (Curt et al., 1998)				
Intial MEP evaluation				
Normal	100	0	0	0
Absent	11	0	78	

Magnetic resonance imaging (Table 4)

Before the advent of MRI, there were no imaging methods to assess the severity of traumatic SCI. MRI provides a rapid non-invasive means of evaluating the condition of spinal cord parenchyma and depicting the injured spinal cord and accurately showing the extent of macroscopic damage (Yamashita et al., 1991). It should be noted, however, that to the best of our knowledge, no study examined the relationship between MRI aspect and walking recovery, but only with neurologic recovery (AIS grade conversion) that is only partially related to walking (see above).

For prognostic purposes the T2 sagittal images seem to be the most useful ones, while T1 and axial images do not correlate with the prognosis (Bozzo et al., 2011). A damaged spinal cord exhibits a variable amount of intramedullary hemorrhage and edema. Both the presence of these two features and the amount of parenchyma that is affected by hemorrhage and edema are directly related to the degree of initial neurologic deficit and to the prognosis (Bondurant et al., 1990; Flanders et al., 1990). Based on these aspects, Bondurant and associates (Bondurant et al., 1990) proposed a classification which consider four different MRI patterns: Pattern 1 shows a normal MRI signal in the cord; pattern 2 represents single-level edema; pattern 3 is multi-level edema; and pattern 4 is mixed hemorrhage and edema.

Most studies showed that patients with spinal cord hemorrhage will have decreased motor power, lower motor recovery rates, and fewer muscles with useful function, 1 year after injury in comparison with subjects with small, non-hemorrhagic lesions (Bondurant et al., 1990; Flanders et al., 1990, 1996; Yamashita et al., 1991; Schaefer et al., 1992; Marciello et al., 1993; Sato et al., 1994; Ramón et al., 1997); hemorrhage on initial MRI (within 15 days from the lesion) is associated with a complete injury in almost 100% of the patients (Ramón et al., 1997). If no hemorrhage is seen on initial MRI, patients will have an incomplete lesion and have a significantly better prognosis for motor recovery in the upper and lower extremities, as well as improvement in their Frankel and/or ASIA impairment scale classification (Schaefer et al., 1992).

It is unclear whether the size of the hemorrhage is a prognostic feature. Some authors (Flanders et al., 1990; Schaefer et al., 1992;

Table 4 | MRI and lesion severity.

Authors	Results
PRESENCE OF HEMORRHAGE AT INITIAL EXAMINATION	
Marciello et al., 1993	Hemorrage = low upper extremity and no lower extremity recovery
Flanders et al., 1990	Hemorrage = decreased motor power, lower motor recovery rate, and fewer muscles with useful function
Ramón et al., 1997	Hemorrage = complete injury
SIZE OF HEMORRHAGE	
Boldin et al., 2006; Flanders et al., 1990; Schaefer et al., 1992	Small hemorrhage = higher recovery rates
Bondurant et al., 1990; Flanders et al., 1996	No relationship between hemorrhage size and recovery
PRESENCE OF EDEMA	
Flanders et al., 1996	Edema = prognosis of recovery to functional levels (D/E)
Ramón et al., 1997	Edema = association with incomplete syndromes
SIZE OF EDEMA	
Flanders et al., 1990; Flanders et al., 1996; Ramón et al., 1997	Degree of edema is inversely proportional to initial impairment and future recovery
Boldin et al., 2006; Flanders et al., 1990	Multiple levels involvement = poorer prognosis and greater chance of complete lesions
Flanders et al., 1996	Involvement of only one to three segments = improved prognosis

Boldin et al., 2006) have shown that small hemorrhages may offer higher recovery rates; others showed no difference based on the size of the hemorrhage (Bondurant et al., 1990; Flanders et al., 1996).

With regard to spinal cord edema, this MRI finding seems to have a good prognostic value. In incomplete SCIs, the finding of edema in MRI is associated with a good prognosis of neurological recovery (Flanders et al., 1996). Furthermore, the incomplete syndromes, such as the Brown-Sèquard syndrome, seem to be associated with the edema pattern (Ramón et al., 1997). However, if the edema involves multiple levels, it tends to be associated with a poorer prognosis and a greater chance of having a complete lesion (Flanders et al., 1996; Boldin et al., 2006). If the cord edema is limited to one to three segments only, then the lesion is usually milder in nature, with an improved prognosis (Bauer and Errico, 1991).

Based on the classification of Bondurant et al. (1990), Bozzo et al. (2011) reviewed the data of several articles (Schaefer et al., 1992; Shimada and Tokioka, 1999; Andreoli et al., 2005) and found a correlation with the AIS conversion of patients. As already reported hemorrhage is the more severe MRI aspect, with about 95% of patients remaining with the same AIS grade of admission examination. Patients with diffuse edema also showed a poor improvement, as only 28% of them showed an improvement of AIS grade. Conversely, patients with single level edema pattern showed a good neurological outcome as 90% of them improved for a mean of 1.9 AIS grades.

Other positive correlations have been described: greater degree of cord compression, greater degree of canal compromise, and the severity of soft tissue injuries seem to be all associated with poorer neurological outcomes (Flanders et al., 1996; Selden et al., 1999; Dai and Jia, 2000; Miyanji et al., 2007; Song et al., 2008).

TREATMENT

In the last decade several interventions aiming at reducing the spinal cord damage (neuroprotection) have been proposed (Becker and McDonald, 2012). However, these interventions are still at an experimental level (Becker and McDonald, 2012). Therefore, in the following paragraphs we will focus only on the use and efficacy of high dose methylprednisolone (which, although questioned, is still the most widely used pharmacological treatment in the acute phase of SCI) and of early surgical intervention. It should be noticed that in both cases, studies referred to neurological improvement rather than to walking recovery. Therefore, data on the efficacy of these treatments on ambulation are not available.

METHYLPREDNISOLONE

The administration of high-dose methylprednisolone (MP) to patients with spinal cord injuries has been reported in the National Acute Spinal Cord Injury Studies (NASCIS, NASCIS-II, and NASCIS-III) (Bracken et al., 1984, 1990, 1997). Since then, the use of MP increased and became a standard of care for acute traumatic SCIs (Hurlbert, 2001). It has been hypothesized that MP attenuates the inflammatory cascade and lessens lipid peroxidation, thus decreasing secondary Spinal Cord damage (Delamarter et al., 1995). In the NASCIS studies, the 24 and 48 h administration of high dose MP produced an important neurologic recovery (AIS grade improvement) paralleled by a functional amelioration (Bracken et al., 1997). However, several recent revisions of NASCIS protocols and other randomized trials questioned the efficacy of steroids administration to achieve a neurologic improvement (Hurlbert, 2001; Matsumoto et al., 2001; Suberviola et al., 2008; Bydon et al., 2013). Furthermore, the 48-h–infusion of MP seems to be associated with an increased risk of pneumonia, sepsis, gastrointestinal bleeding, and steroid myopathy (Pointillart et al., 2000; Quian et al., 2004).

Based on these evidences, both the Consortium for Spinal Cord Medicine clinical practice guidelines (Consortium for Spinal Cord Medicine, 2007) and the neurosurgical guidelines (2002) consider the use of high-dose MP to be a treatment option rather than a standard.

SURGERY TRIALS

The undisputed benefits of surgical treatment for unstable vertebral injuries include decreased hospital stay, fewer sequelae from prolonged immobilization, and more rapid admission to the rehabilitation system (Raineteau and Schwab, 2001).

Despite these evidence, the timing of decompression of the neural elements, and, in particular, the efficacy of early decompression (within 24 h) in improving neurologic recovery is still a matter of debate (Fehlings and Tator, 1999; Fehlings and Perrin, 2005). A meta-analysis of studies of early decompression from 1966 through 2000 (La Rosa et al., 2004), showed that surgery performed within 24 h produced a significant improvement in neurological recovery compared with late surgery, but concluded that the evidence was not strong and that early surgery could be considered only as a practice option.

Starting from this framework, a recent prospective multicentric study (Fehlings et al., 2012) demonstrated that the odds of achieving a 2 AIS grade improvement is 2.8 times higher in patients undergoing early surgical decompression (within 24 h). However, a recent meta-analysis (van Middendorp et al., 2013) reported a lack of statistical robustness of the articles examined, therefore the relationship between early surgery and better neurological outcome is still to be demonstrated.

DISCUSSION

This review demonstrates that the chance of walking recovery after a SCI can be accurately predicted on the base of demographic data and clinical examination. Patients with complete sensory-motor lesions have very limited possibility of achieving walking function at follow up, and also if they are able to ambulate they usually are "limited ambulators." The chances of walking recovery improve in less severe lesions, as demonstrated by AIS B and C subjects. AIS B patients can recover walking especially if their clinical picture shows a less severe involvement of the spinal cord (light touch and pinprick conservation = some sparing of the spino-thalamic and posterior columns tracts = higher possibility of cortico-spinal tracts preservation). Finally, subjects with AIS C lesions are bound to walk, especially the younger ones. This prognosis for walking may be sustained and empowered by instrumental examinations that help to assess the severity of the lesion and, in some cases (SSEPs and MEPs) are directly correlated with walking function.

The need to predict outcome based on expected neurological recovery and associated functional recovery has been emphasized as essential for health care planning (Ditunno, 1999) and this need is partially unmet.

During the first few days after SCI, definitive management strategies are formulated, which often include aggressive surgical decompression of the spinal cord (Wilson et al., 2012). This is also the time of greatest anguish for an injured patient and their family as they face significant prognostic uncertainty. A precise knowledge of the prognosis makes it possible to answer questions regarding function that patients usually ask after spinal cord injury: "Will I walk again?" and "What will I be able to do?" Furthermore, in countries with health care systems based on insurance, rehabilitation professionals have to justify and fight for appropriate services; furthermore they have to know how to allocate resources. Therefore, predicting recovery has become a rehabilitative imperative (Ditunno, 1999).

Finally, better knowledge of the course and prognosis of recovery after SCI and an understanding of the underlying mechanisms would help in the development of strategies and treatments to enhance neurological recovery. The number of interventions, therapies, and devices that have been developed and proposed to improve functional outcomes after SCI is enormous; several of these proposal will undergo clinical trials in the near future. Some early stage SCI clinical trials have recently been started and some experimental therapies have been introduced into clinical practice without a clinical trial being completed. Prognostic data are essential to evaluate the efficacy of new drugs and therapies (for example to distinguish between the natural recovery and the effect of treatments) and to project the clinical trials (for example to calculate the number of patients needed to obtain statistical power) (Fawcett et al., 2006).

LIMITATIONS

This article has several limitations due to the nature of the works examined. Some of them are based on small sample sizes and the definition of walking function and of follow up time points vary across the studies. Furthermore, these articles mainly represent the experience from USA and, in part, Europe. Therefore, they do not reflect the whole world standards of care. As SCI management may differ in different geographical areas, the rates of recovery of walking could vary to. Finally, the distribution in time of the works examined is not regular. Although the study of the prognostic factors is still a matter of interest, most of the articles related to clinical factors date back to the 80 s and 90 s. Some prognostic factors may change over time as SCI management evolves. Based on these limitations the results of these studies could not be necessarily generalizable However, the factors that we examined here are still considered the base of the prognosis of SCI outcome (Burns et al., 2012).

FUNDING

Supported in part by grant RC12G of the Italian Ministry of Health and grant P133 of the International Foundation for Research in Paraplegia to Giorgio Scivoletto.

REFERENCES

(2002). Management of acute central cervical spinal cord injuries. *Neurosurgery* 50(3 Suppl.), S166–S172.

Aalfs, C. M., Koelman, J. H., Meyjes, F. E., and de Visser, B. W. O. (1993). Posterior tibial and sural nerve somatosensory evoked potentials: a study in spastic paraparesis in spinal cord lesions. *Electroencephalogr. Clin. Neurophysiol.* 89, 437–441. doi: 10.1016/0168-5597(93)90118-9

Aito, S., D'Andrea, M., Werhagen, L., Farsetti, L., Cappelli, S., Bandini, B., et al. (2006). Neurological and functional outcome in traumatic central cord syndrome. *Spinal Cord* 45, 292–297. doi: 10.1038/sj.sc.3101944

American Spinal Injury Association. (2000). *International standards for neurological classifications of spinal cord Injury (revised)*. Chicago, IL: American Spinal Injury Association.

Andreoli, C., Colaiacomo, M. C., Rojas Beccaglia, M., Di Biasi, C., Casciani, E., and Gualdi, G. (2005). MRI in the acute phase of spinal cord traumatic lesions: Relationship between MRI findings and neurological outcome. *Radiol. Med.* 110, 636–645.

Bauer, R. D., and Errico, T. J. (1991). "Cervical spine injuries," in *Spinal Trauma*, eds T. J. Errico, R. D. Bauer, and T. Waugh (Philadelphia, PA: JB Lippincott), 71–121.

Becker, D., and McDonald, J. W. 3rd. (2012). Approaches to repairing the damaged spinal cord: overview. *Handb. Clin. Neurol.* 109, 445–461. doi: 10.1016/B978-0-444-52137-8.00028-0

Bohlman, H. H. (1979). Acute fractures and dislocations of the cervical spine. An analysis of three hundred hospitalized patients and review of the literature. *J. Bone Joint Surg. Am.* 61, 1119–1142.

Boldin, C., Raith, J., Fankhauser, F., Haunschmid, C., Schwantzer, G., and Schweighofer, F. (2006). Predicting neurologic recovery in cervical spinal cord injury with postoperative MR imaging. *Spine* 31, 554–559. doi: 10.1097/01.brs.0000201274.59427.a4

Bondurant, F. J., Cotler, H. B., Kulkarni, M. V., McArdle, C. B., and Harris, J. H. (1990). Acute spinal cord injury: a study using physical examination and magnetic resonance imaging. *Spine* 15, 161–168. doi: 10.1097/00007632-199003000-00002

Bozzo, A., Marcoux, J., Radhakrishna, M., Pelletier, J., and Goulet, B. (2011). The role of magnetic resonance imaging in the management of acute spinal cord injury. *J. Neurotrauma* 28, 1401–1411. doi: 10.1089/neu.2009.1236

Bracken, M. B., Collins, W. F., Freeman, D. F., Shepard, M. J., Wagner, F. W., Silten, R. M., et al. (1984). Efficacy of methylprednisolone in acute spinal cord injury. *JAMA* 251, 45–52. doi: 10.1001/jama.1984.03340250025015

Bracken, M. B., Shepard, M. J., Collins, W. F., Holford, T. R., Young, W., Baskin, D. S., et al. (1990). A randomized, controlled trial of methylprednisolone or naloxone in the treatment of acute spinal-cord injury. Results of the second national acute spinal cord injury study. *N. Engl. J. Med.* 322, 1405–1411. doi: 10.1056/NEJM199005173222001

Bracken, M. B., Shepard, M. J., Holford, T. R., Leo-Summers, L., Aldrich, E. F., Fazl, M., et al. (1997). Administration of methylprednisolone for 24 or 48 hours or tirilazad mesylate for 48 hours in the treatment of acute spinal cord injury. Results of the third national acute spinal cord injury randomized controlled trial. national acute spinal cord injury study. *JAMA* 277, 1597–1604. doi: 10.1001/jama.1997.03540440031029

Brown-Sequard, C. E. (1868). Lectures on the physiology and pathology of the central nervous system and the treatment of organic nervous affections. *Lancet* 2, 593–595, 659–662, 755–757, 821–823. doi: 10.1016/S0140-6736(02)52465-X

Buchan, A. C., Fulford, G. E., Jellineck, E., Kerr, W. G., Newsam, J. E., and Stark, G. D. (1972). A preliminary survey of the incidence and etiology of spinal paralysis. *Paraplegia* 10, 23–28. doi: 10.1038/sc.1972.5

Burns, A. S., Marino, R. J., Flanders, A. E., and Flett H. (2012). Clinical diagnosis and prognosis following spinal cord injury. *Handb. Clin. Neurol.* 109, 47–62. doi: 10.1016/B978-0-444-52137-8.00003-6

Burns, S. P., Golding, D. G., Rolle, W. A. Jr., Graziani, V., and Ditunno, J. F. (1997). Recovery of ambulation in motor incomplete tetraplegia. *Arch. Phys. Med. Rehabil.* 78, 1169–1172. doi: 10.1016/S0003-9993(97)90326-9

Bydon, M., Lin, J., Macki, M., Gokaslan, Z. L., and Bydon, A. (2013). The current role of steroids in acute spinal cord injury. *World Neurosurg.* doi: 10.1016/j.wneu.2013.02.062. [Epub ahead of print].

Celani, M. G., Spizzichino, L., Ricci, S., Zampolini, M., and Franceschini, M. (2001). Spinal cord injury in Italy: a multicenter retrospective study. *Arch. Phys. Med. Rehab.* 82, 589–596. doi: 10.1053/apmr.2001.21948

Chabot, R., York, D. H., Watts, C., and Waugh, W. A. (1985). Somatosensory evoked potentials evaluated in normal subjects in spinal cord injured patients. *J. Neurosurg.* 63, 544–551. doi: 10.3171/jns.1985.63.4.0544

Cheshire, W. E., Santos, C. C., Massey, E. W., and Howard, J. E. (1996). Spinal cord infarction: etiology and outcome. *Neurology* 47, 321–330. doi: 10.1212/WNL.47.2.321

Citterio, A., Franceschini, M., Spizzichino, L., Reggio, A., Rossi, B., and Stampacchia G. (2004). Nontraumatic spinal cord injury: an Italian survey. *Arch. Phys. Med. Rehabil.* 85, 1483–1487. doi: 10.1016/j.apmr.2003.09.028

Consortium for Spinal Cord Medicine. (2007). *Early Acute Management in Adults with Spinal Cord Injury: a Clinical Practice Guideline for Health-Care Providers*. Washington, DC: Paralyzed Veterans of America.

Crozier, K. S., Cheng, L. L., Graziani, V., Zorn, G., Herbison, G., and Ditunno, J. F. Jr. (1992). Spinal cord injury: prognosis for ambulation based on quadriceps recovery. *Paraplegia* 30, 762–767. doi: 10.1038/sc.1992.147

Crozier, K. S., Graziani, V., Ditunno, J. F. Jr., and Herbison, G. J. (1991). Spinal cord injury: prognosis for ambulation based on sensory examination in patients who are initially motor complete. *Arch. Phys. Med. Rehabil.* 72, 119–121.

Curt, A., and Dietz V. (1997). Ambulatory capacity in spinal cord injury: significance of somatosensory evoked potentials and ASIA protocols in predicting outcome. *Arch. Phys. Med. Rehabil.* 78, 39–43. doi: 10.1016/S0003-9993(97)90007-1

Curt, A., Keck, M. E., and Dietz, V. (1998). Functional outcome following spinal cord injury: significance of motor-evoked potentials and ASIA scores. *Arch. Phys. Med. Rehabil.* 79, 81–86. doi: 10.1016/S0003-9993(98)90213-1

Dai, L., and Jia, L. (2000). Central cord injury complicating acute cervical disc herniation in trauma. *Spine* 25, 331–335. doi: 10.1097/00007632-200002010-00012

Delamarter, R. B., Sherman, J., and Carr, J. B. (1995). Pathophysiology of spinal cord injury: recovery after immediate and delayed compression. *J. Bone Joint Surg. Am.* 77, 1042–1049.

DiGiovanna, A. G. (2000). *Human Ageing: Biological Perspectives, 2nd Edn.* New York, NY: McGraw Hill Companies.

Ditunno, J. F. (1999). The John Stanley Coulter Lecture. Predicting recovery after spinal cord injury: a rehabilitation imperative. *Arch. Phys. Med. Rehabil.* 80, 361–364.

Ditunno, J. F., Scivoletto, G., Patrick, M., Biering-Sorensen, F., Abel, R., and Marino, R. (2008b). Validation of the walking index for spinal cord injury in a US and European clinical population. *Spinal Cord* 46, 181–188. doi: 10.1038/sj.sc.3102071

Ditunno, P. L., Patrick, M., Stineman, M., and Ditunno, J. F. (2008a). Who wants to walk? Preferences for recovery after SCI: a longitudinal and cross-sectional study. *Spinal Cord.* 46, 500–506. doi: 10.1038/sj.sc.3102172

Domingo, A., Al-Yahya, A. A., Asiri, Y., Eng, J. J., Tania Lam, T., and Spinal Cord Injury Rehabilitation Evidence Research Team. (2012). A systematic review of the effects of pharmacological agents on walking function in people with spinal cord injury. *J. Neurotrauma* 29, 865–879. doi: 10.1089/neu.2011.2052

Dvorak, M. F., Fisher, C. G., Hoekema, J., Boyd, M., Noonan, V., Wing, P. C., et al. (2005). Factors Predicting motor recovery and functional outcome after traumatic central cord syndrome a long-term follow-up. *Spine* 30, 2303–2311. doi: 10.1097/01.brs.0000182304.35949.11

Fawcett, J. W., Curt, A., Steeves, J. D., Coleman, W. P., Tuszynski, M. H., Lammertse, D., et al. (2006). Guidelines for the conduct of clinical trials for spinal cord injury as developed by the ICCP panel: spontaneous recovery after spinal cord injury and statistical power needed for therapeutic clinical trials. *Spinal Cord* 45, 190–205. doi: 10.1038/sj.sc.3102007

Fehlings, M. G., and Perrin, R. G. (2005). The role and timing of early decompression for cervical spine injury: update with a review of the recent clinical evidence. *Injury* 36(Suppl. 2), B13–B26. doi: 10.1016/j.injury.2005.06.011

Fehlings, M. G., and Tator, C. H. (1999). An evidence-based review of decompressive surgery in acute spinal cord injury: rationale, indications, and timing based on experimental and clinical studies. *J. Neurosurg. Spine* 91, 1–11. doi: 10.3171/spi.1999.91.1.0001

Fehlings, M. G., Vaccaro, A., Wilson, J. R., Singh, A. W., Cadotte, D., Harrop, J. S., et al. (2012). Early versus delayed decompression for traumatic cervical spinal cord injury: results of the Surgical Timing in Acute Spinal Cord Injury Study (STASCIS). *PLoS ONE* 7:e32037. doi: 10.1371/journal.pone.0032037

Flanders, A. E., Schaefer, D. M., Doan, H. T., Mishkin, M. M., Gonzalez, C. F., Northrup, B. E. (1990). Acute cervical spine trauma: correlation of MR imaging findings with degree of neurologic deficit. *Radiology* 177, 25–33.

Flanders, A. E., Spettell, C. M., Tartaglino, L. M., Friedman, D. P., and Herbison, G. J. (1996). Forecasting motor recovery after cervical spinal cord injury: value of MR imaging. *Radiology* 201, 649–655.

Foo, D. (1986). Spinal cord injury in forty-four patients with cervical spondylosis. *Paraplegia* 24, 301–306. doi: 10.1038/sc.1986.42

Foo, D., Subrahmanyan, T. S., and Rossier, A. B. (1981). Post-traumatic acute anterior spinal cord syndrome. *Paraplegia*. 19, 201–205. doi: 10.1038/sc.1981.42

Gentleman, D., and Harrington, M. (1984). Penetrating injury of the spinal cord. *Injury* 16, 7–8. doi: 10.1016/0020-1383(84)90101-3

Greenwald, B. D., Seel, R. T., Cifu, D. X., and Shah, A. N. (2001). Gender-related differences in acute rehabilitation lengths of stay, charges, and functional outcomes for a matched sample with spinal cord injury: a multicenter investigation. *Arch. Phys. Med. Rehabil.* 82, 1181–1187. doi: 10.1053/apmr.2001.24891

Herbison, G. J., Zerby, S. A., Cohen, M. E., Marino, R. J., and Ditunno, J. E. (1991). Motor power difference within the first two weeks post-SCI in cervical spinal cord quadriplegic subjects. *J. Neurotrauma* 9, 373–380. doi: 10.1089/neu.1992.9.373

Hurlbert, R. J. (2001). The role of steroids in acute spinal cord injury. An evidence-based analysis. *Spine* 26, S39–S46. doi: 10.1097/00007632-200112151-00009

Hussey, R. W., and Stauffer, E. S. (1973). Spinal cord injury: requirements for ambulation. *Arch. Phys. Med. Rehabil.* 54, 544–547.

Jacobs, S. R., Yeaney, N. K., Herbison, G. J., and Ditunno, J. F. Jr. (1995). Future ambulation prognosis as predicted by somatosensory evoked potentials in motor complete and incomplete quadriplegia. *Arch. Phys. Med. Rehabil.* 76, 635–641. doi: 10.1016/S0003-9993(95)80632-6

Jakob, W., Wirz, M., van Hedel, H. J., Dietz, V., and EM-SCI Study Group. (2009). Difficulty of elderly SCI subjects to translate motor recovery—"body function"–into daily living activities. *J. Neurotrauma* 26, 2037–2044. doi: 10.1089/neu.2008.0824

Kaplan, B. J., Friedman, W. A., and Gavenstein, D. (1985). Somatosensory evoked potential in hysterical paraplegia. *Surg. Neurol.* 23, 502–506. doi: 10.1016/0090-3019(85)90246-0

Kaplan, P. E., and Rosen, J. S. (1981). Somatosensory evoked potentials in spinal cord injured patients. *Paraplegia* 19, 118–122. doi: 10.1038/sc.1981.26

Katoh, S., and el Masry, W. S. (1995). Motor recovery of patients presenting with motor paralysis and sensory sparing following cervical spinal cord injuries. *Paraplegia* 33, 506–509.

Katz, R. T., Tolkeikis, R. J., and Knuth, A. E. (1991). Somatosensory-evoked and dermatomal-evoked potentials are not clinically useful in the prognostication of acute spinal cord injury. *Spine* 16, 730–735. doi: 10.1097/00007632-199107000-00007

Kirshblum, S. C., and O'Connor, K. C. (1998). Predicting neurologic recovery in traumatic cervical spinal cord injury. *Arch. Phys. Med. Rehabil.* 79, 1456–1466.

Ko, H-Y., Ditunno, J. F., Graziani, V., and Little, J. W. (1999). The pattern of reflex recovery during spinal shock. *Spinal Cord* 37, 402–409. doi: 10.1038/sj.sc.3100840

La Rosa, G., Conti, A., Cardali, S., Cacciola, F., and Tomasello F. (2004). Does early decompression improve neurological outcome of spinal cord injured patients? Appraisal of the literature using a meta-analytical approach. *Spinal Cord* 42, 503–512. doi: 10.1038/sj.sc.3101627

Marciello, M., Flanders, A. E., Herbison, G. J., Schaefer, D. M., Friedman, D. P., and Lane, J. I. (1993). Magnetic resonance imaging related to neurologic outcome in cervical spinal cord injury. *Arch. Phys. Med. Rehabil.* 74, 940–946.

Marinho, A. R., Flett, H. M., Craven, C., Ottensmeyer, C. A., Parsons, D., and Verrier, M. C. (2012). Walking-related outcomes for individuals with traumatic and non-traumatic spinal cord injury inform physical therapy practice. *J. Spinal Cord Med.* 35, 371–381. doi: 10.1179/2045772312Y.0000000038

Matsumoto, T., Tamaki, T., Kawakami, M., Yoshida, M., Ando, M., and Yamada, H. (2001). Early complications of high-dose methylprednisolone sodium succinate treatment in the follow-up of acute cervical spinal cord injury. *Spine* 26, 426–430. doi: 10.1097/00007632-200102150-00020

Maynard, F. M., Glen, G. R., Fountain, S., Wilmot, C., and Hamilton R. (1979). Neurological prognosis after traumatic quadriplegia. *J. Neurosurg.* 50, 611–616. doi: 10.3171/jns.1979.50.5.0611

Maynard, F. M. Jr., Bracken, M. B., Creasey, G., Ditunno, J. F. Jr., Donovan, W. H., Ducker, T. B., et al. (1997). International standards for neurological and functional classification of spinal cord injury patients (revised). *Spinal Cord* 35, 266–274. doi: 10.1038/sj.sc.3100432

McKinley, W., Santos, K., Meade, M., and Brooke, K. (2007). Incidence and outcomes of spinal cord injury clinical syndromes. *J. Spinal Cord Med.* 30, 215–224.

McKinley, W. O., Huang, M. E., and Tewksbury, M. A. (2000). Neoplastic vs. traumatic spinal cord injury: an inpatient rehabilitation comparison. *Am. J. Phys. Med. Rehabil.* 79, 138–144. doi: 10.1097/00002060-200003000-00005

McKinley, W. O., Seel, R. T., Gadi, R. K., and Tewksbury, M. A. (2001). Nontraumatic vs. traumatic spinal cord injury. *Am. J. Phys. Med. Rehab*. 80, 693–699. doi: 10.1097/00002060-200109000-00010

Mckinley, W. O., Tewksbury, M. A., and Mujteba, N. M. (2002). Spinal stenosis vs traumatic spinal cord injury: a rehabilitation outcome comparison. *J. Spinal Cord Med*. 25, 28–32.

Merriam, W. E., Taylor, T. K. F., Ruff, S. J., and McPhail, M. J. (1986). A reappraisal of acute traumatic central cord syndrome. *J. Bone Joint Surg*. 68B, 708–713.

Miyanji, F., Furlan, J. C., Aarabi, B., Arnold, P. M., and Fehlings, M. G. (2007). Acute cervical traumatic spinal cord injury: MR imaging findings correlated with neurologic outcome—prospective study with 100 consecutive patients. *Radiology* 243, 820–827. doi: 10.1148/radiol.2433060583

Newey, M. L., Sen, P. K., and Fraser, R. D. (2000). The long-term outcome after central cord syndrome: a study of the natural history. *J. Bone Joint Surg. Br.* 82, 851–855. doi: 10.1302/0301-620X.82B6.9866

Oleson, C. V., Burns, A. S., Ditunno, J. F., Geisler, F. H., and Coleman, W. P. (2005). Prognostic value of pinprick preservation in motor complete, sensory incomplete spinal cord injury. *Arch. Phys. Med. Rehabil.* 86, 988–992. doi: 10.1016/j.apmr.2004.09.031

Pagliacci, M. C., Celani, M. G., Spizzichino, L., Zampolini, M., Aito, S., Citterio, A., et al. (2003).Gruppo Italiano Studio Epidemiologico Mielolesioni (GISEM) Group. spinal cord lesion management in Italy: a 2-year survey. *Spinal Cord* 41, 620–628. doi: 10.1038/sj.sc.3101521

Penrod, L. E., Hegde, S. K., and Ditunno, J. E. (1990). Age effect on prognosis for functional recovery in acute, traumatic central cord syndrome. *Arch. Phys. Med. Rehabil.* 71, 963–968.

Perot, P. L., and Vera, C. L. (1982). Scalp-recorded somatosensory evoked potentials to stimulation of nerves in the lower extremities and evaluation of patients with spinal cord trauma. *Ann. N.Y. Acad. Sci.* 388, 359–368. doi: 10.1111/j.1749-6632.1982.tb50802.x

Pointillart, V., Petitjean, M. E., Wiart, L., Vital, J. M., Lassie, P., Thicoipe, M., et al. (2000). Pharmacological therapy of spinal cord injury during the acute phase. *Spinal Cord* 38, 71–76. doi: 10.1038/sj.sc.3100962

Quian, T., Guo, X., Levi, A. D., Vanni, S., Shebert, R. T., and Sipski, M. L. (2004). High-dose methylprednisolone may cause myopathy in acute spinal cord injury patients. *Spinal Cord* 43, 199–203. doi: 10.1038/sj.sc.3101681

Raineteau, O., and Schwab, M. E. (2001). Plasticity of motor systems after incomplete spinal cord injury. *Nat. Rev. Neurosci.* 2, 263–273. doi: 10.1038/35067570

Ramón, S., Domínguez, R., Ramírez, L., Paraira, M., Olona, M., Castelló, T., et al. (1997). Clinical and magnetic resonance imaging correlation in acute spinal cord injury. *Spinal Cord* 35, 664–673. doi: 10.1038/sj.sc.3100490

Roth, E. J., Lawler, M. H., and Yarkony, G. M. (1990). Traumatic central cord syndrome: clinical features and functional outcomes. *Arch. Phys. Med. Rehabil.* 71, 18–23.

Roth, E. J., Park, T., Pang, T., Yarkony, G. M., and Lee, M. Y. (1991). Traumatic cervical Brown-Sequard and Brown-Sequard plus syndromes: the spectrum of presentations and outcomes. *Paraplegia* 29, 582–589. doi: 10.1038/sc.1991.86

Sato, T., Kokubun, S., Rijal, K. P., Ojima, T., Moriai, N., Hashimoto, M., et al. (1994). Prognosis of cervical spinal cord injury in correlation with magnetic resonance imaging. *Paraplegia* 32, 81–85. doi: 10.1038/sc.1994.14

Schaefer, D. M., Flanders, A. E., Osterholm, J. L., and Northrup, B. E. (1992). Prognostic significance of magnetic resonance imaging in the acute phase of cervical spine injury. *J. Neurosurg.* 76, 218–223. doi: 10.3171/jns.1992.76.2.0218

Scivoletto, G., Farchi, S., Laurenza, L., and Molinari, M. (2011). Traumatic and non-traumatic spinal cord lesions: an Italian comparison of neurological and functional outcomes. *Spinal Cord* 49, 391–396. doi: 10.1038/sc.2010.85

Scivoletto, G., Morganti, B., Ditunno, P., Ditunno, J. F., and Molinari, M. (2003). Effects on age on spinal cord lesion patients' rehabilitation. *Spinal Cord* 41, 457–464. doi: 10.1038/sj.sc.3101489

Scivoletto, G., Morganti, B., and Molinari, M. (2004a). Neurologic recovery of spinal cord injury patients in Italy. *Arch. Phys. Med. Rehabil*. 85, 485–489. doi: 10.1016/S0003-9993(03)00766-4

Scivoletto G., Morganti B., and Molinari, M. (2004b). Sex-related differences of rehabilitation outcomes of spinal cord lesion patients. *Clin. Rehabil.* 18, 709–713. doi: 10.1191/0269215504cr749oa

Selden, N. R., Quint, D. J., Patel, N., D'Arcy, H. S., and Papadopoulos, S. M. (1999). Emergency magnetic resonance imaging of cervical spinal cord injuries: clinical correlation and prognosis. *Neurosurgery* 44, 785–792.

Shimada, K., and Tokioka, T. (1999). Sequential MR studies of cervical cord injury: correlation with neurological damage and clinical outcome. *Spinal Cord* 37, 410–415. doi: 10.1038/sj.sc.3100858

Sipski, M. L., Jackson, A. B., Gómez-Marín, O., Estores, I., and Stein, A. (2004). Effects of gender on neurologic and functional recovery after spinal cord injury. *Arch. Phys. Med. Rehabil.* 85, 1826–1836. doi: 10.1016/j.apmr.2004.04.031

Song, K. J., Kim, G. H., and Lee, K. B. (2008). The efficacy of the modified classification system of soft tissue injury in extension injury of the lower cervical spine. *Spine* 33, E488–E493. doi: 10.1097/BRS.0b013e31817b6191

Stahlman, G. C., and Hanley, E. N. (1992). "Surgical management of spinal injuries," in *Skeletal Trauma*, eds B. D. Browner, J. B. Jupiter, A. M. Levine, and P. G. Trafton (Philadelphia: WB Saunders), 837–860.

Steeves, J. D., Lammertse, D., Curt, A., Fawcett, J. W., Tuszynski, M. H., Ditunno, J. F., et al. (2007). International campaign for cures of spinal cord injury paralysis. guidelines for the conduct of clinical trials for spinal cord injury (SCI) as

developed by the ICCP panel: clinical trial outcome measures. *Spinal Cord* 45, 190–205. doi: 10.1038/sj.sc.3102008

Suberviola, B., Gonzalez-Castro, A., Llorca, J., Ortiz-Melon, F., and Minambres, E. (2008). Early complications of high-dose methylprednisolone in acute spinal cord injury patients. *Injury* 39, 748–752. doi: 10.1016/j.injury.2007.12.005

Vaccaro, A. R., Daugherty, R. J., Sheehan, T. P., Dante, S. J., Cotler, J. M., Balderston, R. A., et al. (1997). Neurologic outcome of early versus late surgery for cervical spinal cord injury. *Spine* 22, 2609–2613. doi: 10.1097/00007632-199711150-00006

van Middendorp, J. J., Hosman, A. J., and Doi, S. A. (2013). The effects of the timing of spinal surgery after traumatic spinal cord injury: a systematic review and meta-analysis. *J. Neurotrauma* 30, 1781–1794. doi: 10.1089/neu.2013.2932

van Middendorp, J. J., Hosman, A. J., Donders, A. R., Pouw, M. H., Ditunno, J. F. Jr., Curt, A., et al. (2011). A clinical prediction rule for ambulation outcomes after traumatic spinal cord injury: a longitudinal cohort study. *Lancet* 377, 1004–1010. doi: 10.1016/S0140-6736(10)62276-3

van Middendorp, J. J., Hosman, A. J., Pouw, M. H., EM-SCI Study Group, and Van de Meent, H. (2009). ASIA impairment scale conversion in traumatic SCI: is it related with the ability to walk? A descriptive comparison with functional ambulation outcome measures in 273 patients. *Spinal Cord* 47, 555–560. doi: 10.1038/sc.2008.162

Waters, R. L., Adkins, R. H., Yakura, J. S., and Sie, I. (1994a). Motor and sensory recovery following incomplete tetraplegia. *Arch. Phys. Med. Rehabil.* 75, 306–311. doi: 10.1016/0003-9993(94)90034-5

Waters, R. L., Adkins, R. H., Yakura, J. S., and Sie, I. (1994b). Motor and sensory recovery following incomplete paraplegia. *Arch. Phys. Med. Rehabil.* 75, 67–72. doi: 10.1016/0003-9993(94)90034-5

Weinstein, D. E., Ko, H. Y., Graziani, V., and Ditunno, J. F. Jr. (1997). Prognostic significance of the delayed plantar reflex following spinal cord injury. *Spinal Cord Med*. 20, 207–211.

Wernig, A., and Muller, S. (1992). Laufband locomotion with body weight support improved walking in persons with severe spinal cord injuries. *Paraplegia* 30, 229–238. doi: 10.1038/sc.1992.61

Wilson, J. R., Grossman, R. G., Frankowski, R. F., Kiss, A., Davis, A. M., Kulkarni, A. V., et al. (2012). A clinical prediction model for long-term functional outcome after traumatic spinal cord injury based on acute clinical and imaging factors. *J. Neurotrauma* 29, 2263–2271. doi: 10.1089/neu.2012.2417

Yamashita, Y., Takahashi, M., Matsuno, Y., Kojima, R., Sakamoto, Y., Oguni, T., et al. (1991). Acute spinal cord injury: magnetic resonance imaging correlated with myelopathy. *Br. J. Radiol.* 64, 201–209. doi: 10.1259/0007-1285-64-759-201

Young, J. S., and Dexter, W. R. (1979). Neurological recovery distal to the zone of injury in 172 cases of closed, traumatic spinal cord injury. *Paraplegia* 16, 39–49. doi: 10.1038/sc.1978.6

Young, W. (1985). "Somatosensory evoked potentials (SEPs) in spinal cord injury," in *Spinal Cord Monitoring*, eds J. Schranml and S. J. Jones (Berlin: Springer-Verlag), 127–142.

Ziganow, S. (1986). Neurometric evaluation of the cortical somatosensory evoked potential in acute incomplete spinal cord injuries. *Electroencephalogr. Clin. Neurophysiol.* 65, 86–93. doi: 10.1016/0168-5597(86)90040-7

Zörner, B., Blanckenhorn, W. U., Dietz, V., EM-SCI Study Group, and Curt, A. (2010). Clinical algorithm for improved prediction of ambulation and patient stratification after incomplete spinal cord injury. *J. Neurotrauma* 27, 241–252. doi: 10.1089/neu.2009.0901

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.

Received: 04 December 2013; accepted: 26 February 2014; published online: 13 March 2014.

Citation: Scivoletto G, Tamburella F, Laurenza L, Torre M and Molinari M (2014) Who is going to walk? A review of the factors influencing walking recovery after spinal cord injury. Front. Hum. Neurosci. ***8****:141. doi: 10.3389/fnhum.2014.00141*

This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 30 May 2014
doi: 10.3389/fnhum.2014.00367

Somatosensory inputs by application of KinesioTaping: effects on spasticity, balance, and gait in chronic spinal cord injury

Federica Tamburella*, Giorgio Scivoletto and Marco Molinari

Spinal Cord Unit, Clinical Movement Analysis and Research Laboratory, IRCCS Santa Lucia Foundation, Rome, Italy

Edited by:
Nadia Dominici, Swiss Federal Institute of Technology (EPFL), Switzerland

Reviewed by:
Antonio Oliviero, Hospital Nacional de Paraplejicos, Spain
Johanna Jonsdottir, Foundation Don Gnocchi, Italy

****Correspondence:***
Federica Tamburella, Spinal Cord Unit, Clinical Movement Analysis and Research Laboratory, IRCCS Santa Lucia Foundation, Via Ardeatina 306 - 00179 Rome, Italy
e-mail: f.tamburella@hsantalucia.it

Introduction: Leg paralysis, spasticity, reduced interlimb coordination, and impaired balance are the chief limitations to overground ambulation in subjects with incomplete spinal cord injury (SCI). In recent years, the application of KinesioTaping (KT) has been proposed to enhance sensory inputs, decreasing spasticity by proprioception feedback and relieving abnormal muscle tension. Because no studies have examined KT-based techniques in SCI subjects, our goal was to analyze the effects of ankle joint KT on spasticity, balance, and gait.

Materials and Methods: A randomized crossover case control design was used to compare the effects of KT and conventional nonelastic silk tape (ST) in 11 chronic SCI subjects**,** AIS level D, with soleus/gastrocnemius (S/G) muscle spasticity and balance and gait impairments. Treatment: 48 h of treatment with KT or ST was followed by 48 h with the other technique after 1 week. A single Y-strip of Cure© tape (KT) and ST was to the S and G muscles with 0% stretch. Before and 48 h after of application of KT and ST, clinical data on the range of motion (ROM), spasticity, clonus, pain, balance, and gait were collected. Stabilometric platform assessment of center of pressure (COP) movements; bidimensional gait analysis; and recording of electromyographic (EMG) activity of the S, G, and tibialis anterior and extensor hallucis lungus muscles were also performed.

Results: Only KT had significant effects on spasticity ($p < 0.05$), clonus ($p < 0.001$) and COP movements ($p < 0.05$), kinematic gait parameters ($p < 0.001$), and EMG activity ($p < 0.001$). Comparison between ST and KT improvements pointed out significant differences as concerns ROM ($p < 0.001$), spasticity ($p < 0.001$), clonus ($p < 0.001$), pain ($p < 0.001$), COP parameters ($p < 0.05$), and most kinematic gait data ($p < 0.05$).

Discussion: Short-term application of KT reduces spasticity and pain and improves balance and gait in chronic SCI subjects. Although these data are promising, they require confirmation in a larger cohort of patients.

Keywords: spinal cord injury, KinesioTaping, balance, gait, Electromyography

INTRODUCTION

In designing effective gait rehabilitation programs after spinal cord injury (SCI), knowledge of the neuronal mechanisms that mediate and, in particular, influence the afferent feedbacks in the function of the damaged spinal cord is paramount (Hubli and Dietz, 2013). Locomotion requires continuous modulation of spinal central pattern generator (CPG) circuits to adapt to the everchanging environment. Feedback from a variety of sources, such as visual, vestibular, somatosensory, and proprioceptive circuits, must be interpreted and integrated into CPG activity to generate locomotion that is effective under all conditions (Hubli and Dietz, 2013). In this complex framework, sensory feedback and context-specific gait requirements interact in affecting muscle synergies (Horak and Nashner, 1986).

In recent years, increasing cutaneous stimuli through neuromuscular KinesioTaping (KT) has been proposed to enhance somatosensory inputs (Halseth, 2004). Alexander et al. reported decreased H-reflex amplitude after KT of the trapezius, suggesting that it influences muscle tone (Alexander et al., 2003). This KT-dependent H-reflex decline indicates that it is inhibitory and adjusts muscle activity through proprioception feedback (Lin et al., 2011). KT has been used in neurological pathologies (Kilbreath et al., 2006; Karadag-Saygi et al., 2010; Cortesi et al., 2011), including stroke and multiple sclerosis, and various orthopedic disorders (Alexander et al., 2003; Halseth, 2004; Thelen et al., 2008; Lin et al., 2011), generally improving muscle tone, range of motion, center of pressure balance parameters, and pain symptoms.

No study has addressed the use of KT in subjects with SCI. Major gait impairments in incomplete SCI are caused by ankle spasticity (Scivoletto et al., 2008; Arazpour et al., 2013) and decreased balance (Scivoletto et al., 2008; Tamburella et al., 2013a), both of which are positively affected by KT in neurological (Cortesi et al., 2011) and nonneurological disorders

(Alexander et al., 2003; Halseth, 2004; Lin et al., 2011). Thus, we examined KT treatment in controlling ankle muscle tone in subjects with incomplete SCI, determining its effects on spasticity, balance, and gait by clinical and instrument-based evaluations.

MATERIALS AND METHODS

STUDY DESIGN—POPULATION

A randomized crossover case control design was used to compare the effects of KT and conventional nonelastic silk tape (ST) on ankle muscles in subjects with chronic incomplete SCI. Patient selection was based on the clinical assessment, per the American Spinal Injury Association (ASIA) standards for neurological status, and on the degree of ankle spasticity, per the modified Ashworth scale (MAS). The inclusion criteria were chronic SCI lesion (i. e., at least 12 months post-injury), AIS level D, and MAS higher than 2 bilaterally in the soleus/gastrocnemius muscles. The exclusion criteria were the presence of other neurological or orthopedic impairments, participation in other studies, and pharmacological treatment for spasticity in the previous 4 weeks. This study was approved by the local ethics committee.

From January 1, 2013 to April 30, 2013, 33 consecutive patients who were admitted to the Spinal Cord Rehabilitation outpatient service of Santa Lucia Foundation were examined by an experience neurologist (Giorgio Scivoletto), of whom 11 subjects met the inclusion criteria. The demographics and clinical features of the SCI subjects are reported in **Table 1**.

INTERVENTION: KT AND ST TREATMENT

After enrollment, SCI subjects were randomized into 2 treatment groups. Group A ($n = 6$) underwent 48 h of KT, followed by 48 h of ST 1 week later. Group B ($n = 5$) received 48 h of ST treatment, followed by 48 h of KT treatment after 1 week (**Figure 1**). All subjects underwent clinical and instrumental evaluations before (T0) and immediately after treatment ($T_{48\,h}$). Electromyography (EMG) was performed only in Group B before, during, and after KT treatment. A certified KT practitioner (Federica Tamburella) administered all taping procedures. Clinical and instrumental outcomes were measured at T0 and $T_{48\,h}$ after removal of the KT by a different researcher (L.M.) who was blinded to the treatment.

KT and ST were applied bilaterally to the plantar-flexor ankle muscles, soleus (S), and gastrocnemius (G), per Luque-Suarez et al. (Luque-Suarez et al., 2014). Standard 5-cm single-strip nonelastic silk tape and Cure© tape were used for the ST and KT, respectively. Y-strip tapes were applied to the S and G muscles with the subject in a prone position, the with knee extended and the ankle in 90° passive dorsiflexion. Both tapes were applied directly to the skin using a decompressive muscle technique, with 0% stretch, from the calcaneus to the medial and lateral femoral condyles (**Figure 2**). To maximize their adhesion, the tape strips were warmed by rubbing them in the hands several times on the application zone (Luque-Suarez et al., 2014).

SETUP AND EVALUATION OF OUTCOMES

All assessments were performed by the same examiner at the same time each day before application of the tape (T_0) and after 48 h ($T_{48\,h}$) of KT or ST treatment. Neurological status was assessed using the American Spinal Injury Association (ASIA) and ASIA Impairment Scale (AIS) (American Spinal Injury Association, 2000). AIS levels A and B indicate complete motor lesions, and AIS levels C and D reflect incomplete motor lesions. Active and passive range of motion (ROM) was measured using a standard manual goniometer (Fong et al., 2011).

The Modified Ashworth Scale (MAS) (Gregson et al., 1999) was used to evaluate ankle spasticity. Spasms, clonus, and pain were scored using the Penn modified Spasm Frequency Scale (PSFS) (Penn, 1988), Spinal Cord Assessment Tool for Spastic Reflexes subscale for clonus assessment (SCATS) (Benz et al., 2005), and Global Pain Scale (GPS) (Wewers and Lowe, 1990), respectively. Balance and gait were assessed using the Berg Balance Scale (BBS) (Lemay and Nadeau, 2010), Walking Index for Spinal Cord Injury (WISCI) (Ditunno and Dittuno, 2001), 10-meter walk test (10WT) (Rossier and Wade, 2001), 6-min walking test (6MWT) (Poole-Wilson, 2000), and timed up and go test (TUG) (Podsiadlo and Richardson, 1991). Walking time tests were performed using a self-selected walking device, if needed (Patrick et al., 2011) and scored using the WISCI, as reported in **Table 1**. All subjects we subjected to instrument-based balance and gait analyses as detailed below.

Table 1 | Patients' clinical and epidemiological data.

Patients	Sex	Age	Weight(Kg)	Height (cm)	Etiology	Lesion level	Years since SCI	MAS	WISCI level
PT1	M	34	85	1.82	Traumatic	C6	4	3	13
PT2	M	69	75	1.65	Traumatic	C6	8	2	18
PT3	F	35	60	1.76	Nontraumatic (Degenerative)	T9	4	5	19
PT4	M	51	74	1.73	Nontraumatic (Vascular)	C6	3	2	18
PT5	F	41	60	1.64	Nontraumatic (Vascular)	T6	4	2	19
PT6	M	52	80	1.78	Traumatic	C6	3	2	20
PT7	F	77	67	1.66	Nontraumatic (Vascular)	T10	7	2	19
PT8	M	58	66	1.73	Nontraumatic (Tumoral)	T11	2	4	19
PT9	F	41	55	1.7	Nontraumatic (Tumoral)	T7	10	4	20
PT10	M	72	81	1.64	Nontraumatic (Degenerative)	C7	6	3	13
PT11	F	38	64	1.6	Traumatic	T8	12	3	20
Mean		52	70	170			5.72	2.9	18
SD		16	10	0.07			3.19	1.04	2.57

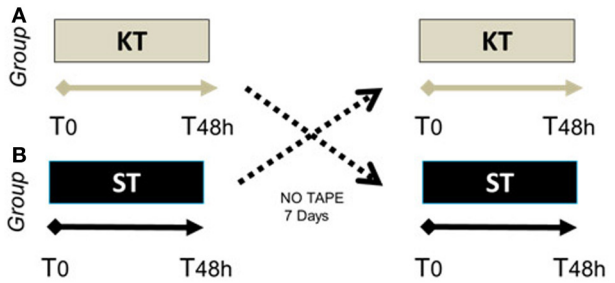


FIGURE 1 | Randomized crossover case control study schema. Group **(A)** ($n = 6$), SCI patients who underwent 48 h of KT, followed by 48 h of ST 1 week later. Group **(B)** ($n = 5$), SCI patients who received 48 h of ST treatment, followed by 48 h of KT 1 week later.

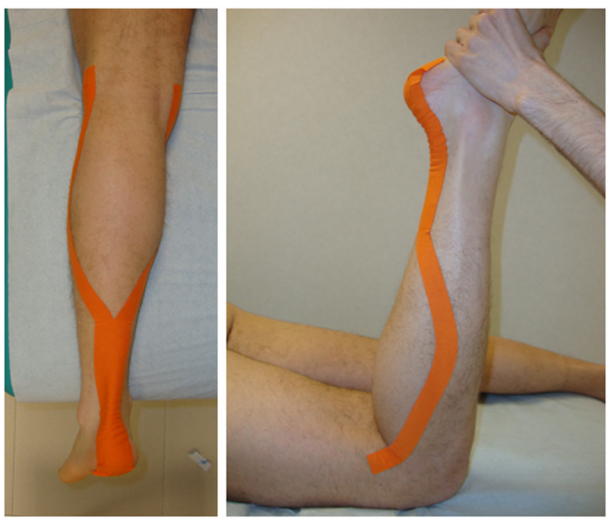


FIGURE 2 | Application of KT and ST tape to soleus and gastrocnemius ankle muscles.

The visual analog scale (VAS) was administered at $T_{48\,h}$ to assess perception of reductions in spasticity. Patients were asked to quantify the reduction in spasticity due to the tape, on a scale from 0 (no reduction in spasticity) to 10 (maximum reduction in spasticity). Electromyography (EMG) analyses were performed only for KT-treated subjects in Group B.

EVALUATION OF BALANCE

Stabilometric parameters were analyzed using a 320 × 75-cm (length × width) static force platform (Platform BPM 120, Physical Support Italia, Italy). The signals were amplified and acquired using dedicated software (Physical Gait Software Vv. 2.66, Physical Support Italia, Italy). Static stability was assessed per Tamburella et al. for chronic SCI subjects (Tamburella et al., 2013a). Patients stood barefoot in a natural and relaxed position with their arms by their sides, without shoes and with both heels lined up, under 2 sensory conditions: eyes open (OE) facing forward to a target 1.5 m away and eyes closed (CE). For each condition, the recording time was set to 51.2 s. and measurements were recorded 3 times and averaged.

We considered the following quantitative COP parameters:

- Length indicators: path length (L, mm), mean (V, L divided by trial duration), anteroposterior velocity (V_{AP}, mm/s) laterolateral velocity (V_{LL}, mm/s), and mean position of planar laterolateral COP.
- Surface indicators: area of the ellipse encompassing 90% of COP samples (A, cm^2) and length of its semiaxes (X and Y, cm).

EVALUATION OF GAIT

Locomotion kinematic gait data were recorded and analyzed using the bidimensional KineView Motion System ® (Kineview, Hafnarfjordur, Iceland) per the protocol for chronic SCI subjects in Tamburella et al. (2013a,b), based on 3 strides at a self-determined velocity. Spatial movements of the lower extremity segments were monitored, based on the position of passive markers that were placed per the Helen Hayes biomechanical model (Kadaba et al., 1990). Kinematic data were reconstructed offline using Matlab (Mathworks, Inc., version 7.1, Natick, Massachusetts, USA).

The following kinematic data were considered: speed (m/s), cadence (N° step/min), stride length (STRIDE: mean of right and left stride in m), stance phase (STANCE: mean of right and left stance phase expressed as the percentage of gait cycle), and double-time support phase (DTS: mean of right and left double-time support phase, expressed as the percentage of gait cycle). STRIDE was defined as the event between 2 successive instances of foot-ground contact. STANCE was defined as the event from foot-ground contact to liftoff, and DTS was the time during which both feet were in contact with the ground (Huxham et al., 2006; Tamburella et al., 2013a,b). Foot-ground contact was determined manually from video recordings (Tamburella et al., 2013a,b). All gait variables were averaged from the kinematic data of the 3 trials.

EMG ASSESSMENT

For Group B patients, surface EMGs of tibialis anterior (TA), extensor hallucis longus (EHL), S, and G muscle activity were analyzed. Recordings were made before (T_0), 5 minutes after KT was applied (T_1), and after the KT was removed ($T_{48\,h}$). EMG data were acquired through 4 wireless EMG sensors, 1 for each muscle, affixed per SENIAM recommendations (Oliveira et al., 2012) using EMG Delsys. EMG data were processed using EMG Works Analysis (Delsys, Boston, USA) using a pass-band filter between 10 and 450 Hz, and successively a 50-Hz notch filter. Root mean square (RMS) values, with a window of 0.250 and an overlap of 0.0625, were obtained from the filtered data. Data on each muscle were then imported into Matlab (Mathworks, Inc., version 7.1, Natick, Massachusetts, USA) to analyze muscle coactivation by calculating the coactivation index (CI) (Kellis et al., 2003).

$$CI = \frac{\int EMG(S+G)}{\int EMG[(TA+EHL)+(S+G)]} \cdot 100$$

CI is a relative measure of antagonist (S and G) contribution to total activation (S and G + TA and EHL) during the dorsiflexion

task (Kellis et al., 2003). Thus, an increase in CI reflects a rise in co-contraction. CI ranged from 0 to 100%, with 100% indicating full muscle coactivation, defined as coactivation (i. e., simultaneous activity) of all ankle muscles. EMG data were recorded while patients were asked to perform maximal voluntary contraction (MVC) during 5 dorsiflexion active movements lying down with knees flexed and extended. Data were averaged across the 5 active tasks.

STATISTICAL ANALYSIS

No participant withdrew from the trial, and all outcome measures were obtained for all SCI subjects. Descriptive statistics were generated for all variables. Prior to the statistical comparisons, normal distribution of the data was confirmed by Kolmogorov-Smirnov test.

Treatment effects were analyzed by grouping the KT and ST data on Group A and B subjects. Paired *t*-test was used to compare the effects of treatment, evaluated as T0 vs. $T_{48\,h}$, for each KT or SK treatment groups. At T0 and $T_{48\,h}$, KT and ST were compared by independent *t*-test and Mann-Whitney *U*-test for ordinal and nonordinal variables, respectively.

For each clinical and instrument-based parameter, the percentage of improvement due to KT and ST was calculated as follows:

Percentage of improvement $= [(T_{48\,h}$ data-T_0 data)/$T_0]$*100. Treatment effects on percentage of improvement data were analyzed by independent *t*-test or Mann-Whitney *U*-test when appropriate.

CI data on KT-treated Group B patients were analyzed by repeated measures ANOVA, with time (T0 vs. T_1 vs. $T_{48\,h}$) as the main within-group factor, followed by Bonferroni *post-hoc* test when the ANOVA results reached significance.

Statistical significance was considered at $p < 0.05$ ($^{*}p < 0.05$, $^{**}p < 0.005$, $^{***}p < 0.001$). All statistical tests were performed using the Statistical Package for the Social Sciences Software (SPSS), version 12.0 (Chicago, IL).

RESULTS

No clinical or instrument-based assessment differed significantly between Groups A and B at T_0(**Table 2**) ($p > 0.05$).

CLINICAL ASSESSMENT

The clinical assessment results are shown in **Table 2**. As expected, almost no changes were observed between T_0 and $T_{48\,h}$in the ST group. Conversely, vs. T_0, KT treatment at $T_{48\,h}$ significantly improved passive ($p < 0.005$) and active ROM ($p < 0.001$), SCATS score with the knees flexed and extended ($p < 0.001$), PSFS ($p < 0.001$), BBS ($p < 0.001$), and 6MWT ($p < 0.001$). Compared with ST, KT had significant treatment effects $T_{48\,h}$ on SCATS with the knees flexed and extended ($p < 0.05$) and on MAS ($p < 0.05$).

Based on percentage of improvement values, we noted significant treatment effects on active and passive dorsiflexion ROM ($p < 0.001$), pathological reflex with the knees flexed ($p < 0.005$) and extended ($p < 0.001$), PSFS ($p < 0.001$), GPN ($p < 0.001$), BBS, and 6MWT ($p < 0.001$).

With regard to perception of spasticity, VAS score was 7.9 ± 1.2 after KT and 2.5 ± 1.3 after ST ($p < 0.05$).

EVALUATION OF BALANCE

Between T0 and $T_{48\,h,}$ KT significantly improved L, C, V_{LL}, and V_{AP} under the OE and CE conditions ($p < 0.05$). Compared with ST, at $T_{48\,h}$, KT had significant treatment effects on L, V, V_{LL} ($p < 0.05$), and V_{AP} ($p < 0.001$) under the OE condition and on L and V under the CE condition ($p < 0.05$). Based on

Table 2 | Clinical and instrumental assessment.

Clinical data	KT Mean (*SD*)		ST Mean (*SD*)		*P*		
					KT vs. ST		
	T_0	$T_{48\,h}$	T_0	$T_{48\,h}$	T_0	$T_{48\,h}$	Percentage of improvement
Passive ROM (°)	88.64 (11.63)	78.64** (10.88)	85.27 (13.07)	85.27 (13.07)	n.s.	n.s.	0.001***
Active ROM (°)	90.18 (13.47)	80.45*** (12.41)	87.27 (14.55)	87 (14.21)	n.s.	n.s.	0.001***
SCATS (flexed knee)	2.18 (0.82)	1.55*** (0.82)	2.09 (0.91)	2.09 (0.91)	n.s.	0.008*	0.002**
SCATS (extended knee)	2.18 (0.82)	1.09*** (0.54)	2.09 (0.91)	2.09 (0.91)	n.s.	0.008*	0.001***
MAS	3.82 (1.17)	1.82 (0.75)	3.6 (0.84)	2.45 (0.93)	n.s.	0.05*	0.001***
PSFS	2.73 (1.35)	1*** (1.26)	2.09 (1.51)	2.09 (1.51)	n.s.	n.s.	0.001***
GPS	2.45 (3.3)	1 (2.49)	2 (3.26)	2 (3.26)	n.s.	n.s.	0.04*
BBS	39.64 (7.7)	42.82*** (7.15)	40.36 (7.68)	40.55 (8)	n.s.	n.s.	0.001***
WISCI	18 (2.57)	18 (2.57)	18 (2.57)	18 (2.57)	n.s.	n.s.	n.s.
6 MWT (m)	231.65 (106.479)	259.63*** (116.13)	253.63 (125.16)	251.25 (125.02)	n.s.	n.s.	0.001***
10 MWT (s)	24.62 (17.07)	19.94 (14.28)	22.97 (18.11)	21.94 (17.32)	n.s.	n.s.	n.s.
TUG (s)	25.69 (17.50)	20.45 (13.30)	22.11 (14.34)	21.27 (13.92)	n.s.	n.s.	n.s.

*The KT, $T_{48\,h}$ column lists the statistical comparison of T0 vs. $T_{48\,h}$ data. Comparisons between KT and SHAM data at T0 and $T_{48\,h}$ and percentage of improvement between KT and ST groups are reported in the last columns of the table (*p < 0.05, **p < 0.005, ***p < 0.001). Gray cells indicate significant p-values. n.s., not significant; for clinical scales abbreviations, see main text.*

percentage of improvement values, significantly treatment effects were pointed out for L, V, and V_{AP} ($p < 0.05$) under the OE and CE conditions and enhanced A, Y, and V_{LL} under the CE condition (A and V_{LL}: $p < 0.05$; Y and V_{AP} $p < 0.005$) (**Table 3**).

EVALUATION OF GAIT

The effects of KT on STRIDE, STANCE, and DTS at $T_{48\,h}$ vs. T0 were significant ($p < 0.001$). Further STRIDE ($p < 0.001$), STANCE, and DTS ($p < 0.005$) improved with KT at $T_{48\,h}$ compared with ST. Comparison of percentage improvement values demonstrated significant treatment effects for all kinematic parameters (speed, cadence, and DTS: $p < 0.05$; STRIDE and STANCE: $p < 0.001$) (**Table 4**).

ASSESSMENT OF EMG CI

CI, as assessed with the knees flexed or extended, declined significantly immediately after application of KT ($p < 0.001 - F[19.046]$). After 48 h of treatment, this effect was maintained only with the knees flexed ($p < 0.001 - F[0.820]$) (**Figure 3**).

To determine the most notable effects of KT, the results were divided into primary and secondary outcome measures, as reported in **Table 5**. MAS, BBS, CoP V, 6MWT, STRIDE, and STANCE were identified as the most important outcomes, and the remaining data were considered secondary outcomes and used to evaluate additional effects of the intervention.

DISCUSSION

In this study, we examined the effects of KT treatment in chronic incomplete SCI subjects compared with nonelastic ST on functional relevant aspects of the post-SCI condition—, i. e., ankle muscle spasticity, balance, and gait. By MAS and analysis of functional balance and gait, 48 h of KT treatment improved all primary outcomes: MAS, BBS, V CoP, 6MWT, STRIDE, and STANCE (**Figure 4**), indicating better functional status after KT, with reduced spasticity and improved balance and gait. In general no adverse events were observed, and subjects reported no discomfort during KT treatment.

KT is used to enhance sensory inputs, decreasing spasticity through proprioception feedback and relieving abnormal muscle tension, in healthy athletic subjects (Thelen et al., 2008; Lin et al., 2011). Few studies have examined KT in neurological lesions, such as multiple sclerosis (Cortesi et al., 2011) and stroke (Kilbreath et al., 2006; Karadag-Saygi et al., 2010). In multiple sclerosis, Cortesi et al. observed positive effects of KT of the ankle on COP balance parameters, suggesting that ankle taping helps stabilize body posture immediately (Cortesi et al., 2011). In stroke patients, KT of the gluteus muscles increases hip extension during gait, suggesting that muscle activation improves through cutaneous stimuli (Kilbreath et al., 2006), whereas no positive effects were obtained by combining ankle KT and botulinum toxin to reduce plantar flexor spasticity (Karadag-Saygi et al., 2010). No data are available on the effects of KT in SCI subjects.

The crossover paradigm that we used allowed us to blind subjects to the treatment allocation and limit the risk of compliance in analyzing the effects of KT vs. ST (Mills et al., 2009). To prevent overflow effects of KT, an interval of 7 days separated application of the tapes (Chen et al., 2013; de Hoyo et al., 2013).

Although significant differences were observed in passive/active ROM, clonus, spasms, BBS, 6MWT, and most COP

Table 3 | Balance assessment.

CoP parameters		KT Mean (*SD*)		ST Mean (*SD*)		*P*		
						KT vs. ST		
		T_0	$T_{48\,h}$	T_0	$T_{48\,h}$	T_0	$T_{48\,h}$	Percentage of improvement
Open eyes	A (cm^2)	2.53 (3.10)	2.72 (5.81)	2.33 (3.16)	2.67 (3.14)	n.s.	n.s.	n.s.
	X (cm)	1.32 (0.98)	1.26 (1.15)	1.37 (0.97)	1.26 (0.97)	n.s.	n.s.	n.s.
	Y (cm)	1.33 (0.89)	1.22 (0.79)	1.27 (0.71)	1.4 (0.85)	n.s.	n.s.	n.s.
	L (mm)	193.68 (104.44)	157.07* (76.78)	183.47 (95.57)	183.19 (107.36)	n.s.	0.01*	0.01*
	V (mm/s)	3.78 (2.04)	3.08* (1.49)	(3.64) (1.92)	3.66 (2.1)	n.s.	0.01*	0.02*
	V_{LL} (mm/s)	2.79 (1.48)	2.26* (1.1)	2.68 (1.39)	2.59 (1.45)	n.s.	0.02*	n.s.
	V_{AP}(mm/s)	2.05 (1.19)	1.65* (0.79)	2 (1.15)	2.08 (1.29)	n.s.	0.001***	0.01*
Closed eyes	A (cm^2)	5.00 (6.12)	4.19 (5.74)	3.28 (4.36)	4.53 (6)	n.s.	n.s.	0.04*
	X (cm)	1.69 (1.60)	1.52 (1.23)	1.60 (1.1)	1.52 (1.3)	n.s.	n.s.	n.s.
	Y (cm)	1.94 (1.30)	1.63 (1.18)	1.46 (0.97)	1.83 (1.36)	n.s.	n.s.	0.002**
	L (mm)	283.37 (188.56)	246.69* (165.09)	244.33 (143.46)	268.13 (199.79)	n.s.	0.03*	0.02*
	V (mm/s)	5.53 (3.68)	4.82* (3.22)	4.77 (2.80)	5.21 (3.92)	n.s.	0.03*	0.02*
	V_{LL} (mm/s)	3.84 (2.47)	3.29* (2.18)	3.36 (1.88)	3.51 (2.48)	n.s.	n.s.	0.05*
	V_{AP} (mm/s)	3.30 (2.42)	2.89* (2.05)	2.79 (1.84)	3.21 (2.6)	n.s.	n.s.	0.007*

The KT, $T_{48\,h}$ column lists the statistical comparison of T0 vs. $T_{48\,h}$ data. Comparisons between KT and SHAM data at T0 and $T_{48\,h}$ and percentage of improvement between KT and ST groups are reported in the last columns of the table ($p < 0.05$, **$p < 0.005$, ***$p < 0.001$). Gray cells indicate significant p-values. n.s., not significant; for abbreviation of COP parameters, see main text.*

Table 4 | Gait assessment.

Kinematic gait data	KT Mean (*SD*)		ST Mean (*SD*)		*P*		
					KT vs. ST		
	T_0	$T_{48\,h}$	T_0	$T_{48\,h}$	T_0	$T_{48\,h}$	Percentage of improvement
Speed (m/s)	0.54 (0.2)	0.56 (0.24)	0.51 (0.18)	0.50 (0.19)	n.s.	n.s.	0.02*
Cadence (steps/min)	69.67 (22.61)	71.09 (24.80)	66.47 (20.52)	65.79 (20.55)	n.s.	n.s.	0.04*
STRIDE (m)	1.04 (0.15)	1.15*** (0.19)	1 (0.13)	0.97 (0.13)	n.s.	0.001***	0.001***
STANCE (%)	72.42 (24.39)	64.99*** (21.63)	69.94 (23.81)	65.49 (23.66)	n.s.	0.005**	0.001***
DTS (%)	27.44 (11.05)	24.46*** (9.20)	26.66 (10.93)	24.64 (10.52)	n.s.	0.005**	0.05*

The KT, $T_{48\,h}$ column lists the statistical comparison of T0 vs. $T_{48\,h}$ data. Comparisons between KT and SHAM data at T0 and $T_{48\,h}$ and percentage of improvement between KT and ST groups are reported in the last columns of the table ($p < 0.05$, **$p < 0.005$, ***$p < 0.001$). Gray cells indicate significant p-values. n.s., not significant; DTS, double-time support phase.*

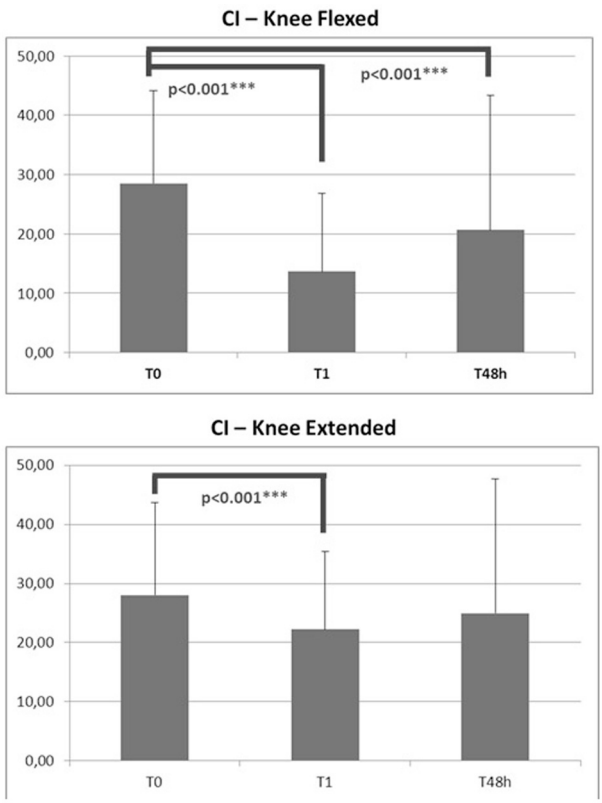


FIGURE 3 | CI index at T_0, T_1, and $T_{48\,h}$ for EXP group. ***$p < 0.001$.

parameters and kinematic gait data after 48 h of KT, almost no changes were observed after 48 h of ST treatment, as expected, due to the chronic condition. Treatment effects were analyzed by comparing improvements after 48 h of KT and ST treatments. KT reduced MAS and improved active and passive ankle ROM, which was paralleled by a decrease in spasticity-associated symptoms, clonus, and pain. Control of ankle spasticity is paramount in improving balance and gait in SCI subjects (Arazpour et al., 2013). KT had significant therapeutic effects on balance and gait, both of which improved with regard to the clinical scales and stabilometric and kinematic data. After KT treatment, there was better control of balance, as confirmed by the decline in V and L COP parameters (Cortesi et al., 2011; Tamburella et al., 2013a), as well as an improvement in kinematic gait parameters. Decreases in STANCE and DTS reflect improved dynamic postural stability (Tamburella et al., 2013b), which has been suggested to specifically improve gait in subjects with chronic motor incomplete SCI (Tamburella et al., 2013a).

To determine the possible mechanism of these improvements, EMG data were collected in Group B patients before, during, and after KT treatment. CI has been proposed as an index of spasticity in stroke subjects (Hu et al., 2007) and is indicative of fatigue-induced decreases in muscular co-contraction in healthy athletic subjects (Missenard et al., 2008). In this study, we used the CI to evaluate EMG activity of agonist vs. antagonist ankle muscles. A high degree of CI reflects excessive antagonistic muscle contractions during dynamic activities compared with agonist muscle activity, impairing function and increasing the metabolic cost of performing the task (Knarr et al., 2012). Our EMG data demonstrated a significant reduction in CI with KT, suggesting improved motor outcome (Hu et al., 2009) and confirming our clinical data on spasticity.

Notably, CI improved immediately after application of KT—an effect that was maintained, although slightly reduced, after 48 h. The lack of significance of the CI data at 48 h with the knee extended confirm the high variability of spasticity measurements in this posture compared with the knee flexed (**Figure 3**). The significant reduction in CI due to KT might be explained by 2 reasons: the increase in EMG activation of the TA and EHL and the decreased co-contraction phase of the antagonist S and G muscles. Considering the findings of Alexander et al. (2003), in which amplitude of the H-reflex decreased after KT of the trapezius in healthy subjects, it is conceivable that KT also adjusts muscle activity by inhibiting proprioception feedback (Lin et al., 2011) also in SCI subjects.

To improve outcomes and methods of applying the tape, it is necessary to understand the mechanism that leads to better upright balance and gait. The effects of KT were clinically significant immediately after its application, implying that the

Table 5 | Primary and secondary outcome measures.

			KT Mean (*SD*)		ST Mean (*SD*)		*P:* KT vs. ST		
			T_0	$T_{48\,h}$	T_0	$T_{48\,h}$	T_0	$T_{48\,h}$	% of improvement
Primary Outcomes	MAS		3.82 (1.17)	1.82 (0.75)	3.6 (0.84)	2.45 (0.93)	n.s.	0.05*	0.001***
	BBS		39.64 (7.7)	42.82*** (7.15)	40.36 (7.68)	40.55 (8)	n.s.	n.s.	0.001***
	V (mm/s)	OE	3.78 (2.04)	3.08* (1.49)	(3.64) (1.92)	3.66 (2.1)	n.s.	0.01*	0.02*
		CE	5.53 (3.68)	4.82* (3.22)	4.77 (2.80)	5.21 (3.92)	n.s.	0.03*	0.02*
	6 MWT (m)		231.65 (106.479)	259.63*** (116.13)	253.63 (125.16)	251.25 (125.02)	n.s.	n.s.	0.001***
	Stride (m)		1.04 (0.15)	1.15*** (0.19)	1 (0.13)	0.97 (0.13)	n.s.	0.001***	0.001***
	Stance (%)		72.42 (24.39)	64.99*** (21.63)	69.94 (23.81)	65.49 (23.66)	n.s.	0.005**	0.001***
Secondary Outcomes	Clinical Data		Passive/Active ROM, SCATS, PSFS, GPS, WISCI, 10MWT, TUG (for details, see **Table 2**)						
	Instrumental Data		BALANCE: A, X, Y, L, V_{LL}, V_{AP} for both visual conditions: OE and CE (for details, see **Table 3**)						
			GAIT: Speed, Cadence, DTS (for details see **Table 4**)						

Clinical and instrumental data on spasticity, balance, and gait have been divided into primary and secondary outcome measures. The KT, $T_{48\,h}$ column lists the statistical comparison of T0 vs. $T_{48\,h}$ data. Comparisons between KT and SHAM data at T0 and $T_{48\,h}$ and percentage of improvement between KT and ST groups are reported in the last columns of the table ($p < 0.05$, **$p < 0.005$, ***$p < 0.001$). Gray cells indicate significant p-values. n.s., not significant; for abbreviations, see main text.*

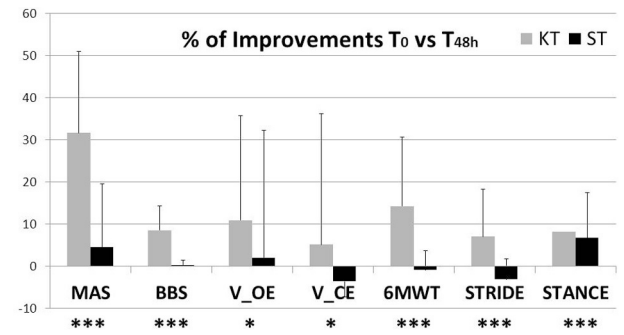


FIGURE 4 | Percentage of improvement due to 48-h application of KT or ST. *$p < 0.05$, ***$p < 0.001$.

changes were not due to long-term learning, as reported in multiple sclerosis (Cortesi et al., 2011). In addition to the secondary effects of spasticity changes, the alterations in the balance control system might be explained by changes in skin receptor inputs due to application of KT (Morasso and Schieppati, 1999). The mechanical effects of applying tape to the skin might increase skin receptor output, stimulating supraspinal centers and thus enhancing kinesthetic and joint position sense (Simoneau et al., 1997; Halseth, 2004) and improving balance.

In analyzing the effects of KT on gait, sensory components cannot be dismissed. Applying pressure to and stretching the skin with KT can stimulate cutaneous mechanoreceptors and enhance signal information of joint movement or joint position (Hsu et al., 2009) (Riemann and Lephart, 2002). The importance of sensory inputs in influencing the activity of gait central pattern generators (CPGs) is highlighted (Grillner, 2006). In SCI patients, changes in CPG circuits are well documented (Grillner, 2006; Roy et al., 2012) and are learning-dependent, primarily through rhythmic peripheral influences that imposed by the exercise—for instance, during robotic gait training (Curt et al., 2008; van Hedel and Dietz, 2010). The importance and effectiveness of sensory input in modulating stepping in SCI has been demonstrated in a wide range of experiments (Edgerton and Roy, 2009); for example, modulation of sensory information influences spinal circuit reorganization to be effective from milliseconds to months (Roy et al., 2012). Thus, sensory modulation through KT might not only influence spasticity but also intervene in longlasting reorganization of spinal gait circuits. In this theoretical framework, the influences of KT on gait merit studies not only in subjects with SCI but in all neurological gait pathologies.

Subjectively, the VAS results indicate a significant reduction in perception of spasticity after KT treatment and that spasticity is negatively associated with quality of life after SCI (Westerkam et al., 2011).

The significance of the sensory effects of KT must also be considered in analyzing its effects on pain. In our study, despite the short-term treatment, GPS declined significantly after KT and but not with ST. Treatment significance was present when comparing KT and ST GPS improvements. Although this study did not aim to evaluate the effects of KT on pain, our results are consistent with data in chronic low back pain patients (Paoloni et al., 2011) and merit dedicated studies, possibly with longer application times of KT.

In conclusion, KT is a valid technique to reduce spasticity and related symptoms in the short term and improve balance and gait in chronic incomplete SCI subjects. Further studies are needed to determine its long-lasting effects.

LIMITATIONS

The sample size of SCI subjects ($n = 11$) was small, which might have limited the statistical relevance of the study. Nevertheless, the statistical differences were large, rendering the statistical error that was caused by sample size negligible. Further, as suggested by Friston (2012), significant results that are based on a small sample

indicate a greater treatment effect than equivalent results in a large sample.

A follow-up study with a longer KT application is necessary to confirm these preliminary data, and a theory on the neurophysiological effects of taping would facilitate the generation of experimental hypotheses.

AUTHOR CONTRIBUTIONS

Federica Tamburella: Substantial contributions to the conception and design of the work; the acquisition, analysis, and interpretation of data; drafting of the manuscript and revising it with regard to intellectual content. Giorgio Scivoletto: Substantial contributions to the conception and design of the work, the interpretation of data, and revision of the work with regard to intellectual content. Marco Molinari: Substantial contributions to revision of the work with regard to intellectual content and final approval of the version to be published.

ACKNOWLEDGMENTS

This work was supported by the Italian Ministry of Health (RC08G) and the European Commission in the Seventh Framework Program ICT-2013- 611626 SYMBITRON. Thanks to Luca Muzzioli for data acquisition. The manuscript was reviewed for English language by "Blue Pencil" Science.

REFERENCES

Alexander, C. M., Stynes, S., Thomas, A., Lewis, J., and Harrison, P. J. (2003). Does tape facilitate or inhibit the lower fibres of trapezius? *Man. Ther.* 8, 37–41. doi: 10.1054/math.2002.0485

American Spinal Injury Association. (2000). *International Standard for Neurological Classification of Spinal Cord Injury (rev)*. Chicago: American Spinal Injury Association, 1–23.

Arazpour, M., Bani, M. A., Hutchins, S. W., Curran, S., and Javanshir, M. A. (2013). The influence of ankle joint mobility when using an orthosis on stability in patients with spinal cord injury: a pilot study. *Spinal Cord* 51, 750–754. doi: 10.1038/sc.2013.78

Benz, E. N., Hornby, T. G., Bode, R. K., Scheidt, R. A., and Schmit, B. D. (2005). A physiologically based clinical measure for spastic reflexes in spinal cord injury. *Arch. Phys. Med. Rehabil.* 86, 52–59. doi: 10.1016/j.apmr.2004.01.033

Chen, C. H., Huang, T. S., Chai, H. M., Jan, M. H., and Lin, J. J. (2013). Two stretching treatments for the hamstrings: proprioceptive neuromuscular facilitation versus kinesio taping. *J. Sport Rehabil.* 22, 59–66.

Cortesi, M., Cattaneo, D., and Jonsdottir, J. (2011). Effect of kinesio taping on standing balance in subjects with multiple sclerosis: a pilot study\m{1}. *NeuroRehabilitation* 28, 365–372. doi: 10.3233/NRE-2011-0665

Curt, A., van Hedel, H. J., Klaus, D., and Dietz, V. (2008). Recovery from a spinal cord injury: significance of compensation, neural plasticity, and repair. *J. Neurotrauma* 25, 677–685. doi: 10.1089/neu.2007.0468

de Hoyo, M., Alvarez-Mesa, A., Sanudo, B., Carrasco, L., and Dominguez, S. (2013). Immediate effect of kinesio taping on muscle response in young elite soccer players. *J. Sport Rehabil.* 22, 53–58.

Ditunno, P. L., Dittuno, J. F. Jr. (2001). Walking index for spinal cord injury (WISCI II): scale revision. *Spinal Cord* 39, 654–656. doi: 10.1038/sj.sc.3101223

Edgerton, V. R., and Roy, R. R. (2009). Robotic training and spinal cord plasticity. *Brain Res. Bull.* 78, 4–12. doi: 10.1016/j.brainresbull.2008.09.018

Fong, C. M., Blackburn, J. T., Norcross, M. F., McGrath, M., and Padua, D. A. (2011). Ankle-dorsiflexion range of motion and landing biomechanics. *J. Athl. Train.* 46, 5–10. doi: 10.4085/1062-6050-46.1.5

Friston, K. (2012). Ten ironic rules for non-statistical reviewers. *Neuroimage* 61, 1300–1310. doi: 10.1016/j.neuroimage.2012.04.018

Gregson, J. M., Leathley, M., Moore, A. P., Sharma, A. K., Smith, T. L., and Watkins, C. L. (1999). Reliability of the Tone Assessment Scale and the modified Ashworth scale as clinical tools for assessing poststroke spasticity. *Arch. Phys. Med. Rehabil.* 80, 1013–1016. doi: 10.1016/S0003-9993(99)90053-9

Grillner, S. (2006). Biological pattern generation: the cellular and computational logic of networks in motion. *Neuron* 52, 751–766. doi: 10.1016/j.neuron.2006.11.008

Halseth, T. (2004). WMJDML. The effects of kinesioTM taping on proprioception at the ankle. *J. Sports Sci. Med.* 3, 1–7.

Horak, F. B., and Nashner, L. M. (1986). Central programming of postural movements: adaptation to altered support-surface configurations. *J. Neurophysiol.* 55, 1369–1381.

Hsu, Y. H., Chen, W. Y., Lin, H. C., Wang, W. T., and Shih, Y. F. (2009). The effects of taping on scapular kinematics and muscle performance in baseball players with shoulder impingement syndrome. *J. Electromyogr. Kinesiol.* 19, 1092–1099. doi: 10.1016/j.jelekin.2008.11.003

Hu, X., Tong, K. Y., Song, R., Tsang, V. S., Leung, P. O., and Li, L. (2007). Variation of muscle coactivation patterns in chronic stroke during robot-assisted elbow training. *Arch. Phys. Med. Rehabil.* 88, 1022–1029. doi: 10.1016/j.apmr.2007.05.006

Hu, X. L., Tong, K. Y., Song, R., Zheng, X. J., Lui, K. H., Leung, W. W., et al. (2009). Quantitative evaluation of motor functional recovery process in chronic stroke patients during robot-assisted wrist training. *J. Electromyogr. Kinesiol.* 19, 639–650. doi: 10.1016/j.jelekin.2008.04.002

Hubli, M., and Dietz, V. (2013). The physiological basis of neurorehabilitation–locomotor training after spinal cord injury. *J. Neuroeng. Rehabil.* 10:5. doi: 10.1186/1743-0003-10-5

Huxham, F., Gong, J., Baker, R., Morris, M., and Iansek, R. (2006). Defining spatial parameters for non-linear walking. *Gait Posture* 23, 159–163. doi: 10.1016/j.gaitpost.2005.01.001

Kadaba, M. P., Ramakrishnan, H. K., and Wootten, M. E. (1990). Measurement of lower extremity kinematics during level walking. *J. Orthop. Res.* 8, 383–392. doi: 10.1002/jor.1100080310

Karadag-Saygi, E., Cubukcu-Aydoseli, K., Kablan, N., and Ofluoglu, D. (2010). The role of KinesioTaping combined with botulinum toxin to reduce plantar flexors spasticity after stroke. *Top. Stroke Rehabil.* 17, 318–322. doi: 10.1310/tsr1704-318

Kellis, E., Arabatzi, F., and Papadopoulos, C. (2003). Muscle co-activation around the knee in drop jumping using the co-contraction index. *J. Electromyogr. Kinesiol.* 13, 229–238. doi: 10.1016/S1050-6411(03)00020-8

Kilbreath, S. L., Perkins, S., Crosbie, J., and McConnell, J. (2006). Gluteal taping improves hip extension during stance phase of walking following stroke. *Aust. J. Physiother.* 52, 53–56. doi: 10.1016/S0004-9514(06)70062-9

Knarr, B. A., Zeni, J. A. Jr., and Higginson, J. S. (2012). Comparison of electromyography and joint moment as indicators of co-contraction. *J. Electromyogr. Kinesiol.* 22, 607–611. doi: 10.1016/j.jelekin.2012.02.001

Lemay, J. F., and Nadeau, S. (2010). Standing balance assessment in ASIA D paraplegic and tetraplegic participants: concurrent validity of the Berg Balance Scale. *Spinal Cord* 48, 245–250. doi: 10.1038/sc.2009.119

Lin, J. J., Hung, C. J., and Yang, P. L. (2011). The effects of scapular taping on electromyographic muscle activity and proprioception feedback in healthy shoulders. *J. Orthop. Res.* 29, 53–57. doi: 10.1002/jor.21146

Luque-Suarez, A., Gijon-Nogueron, G., Baron-Lopez, F. J., Labajos-Manzanares, M. T., Hush, J., and Hancock, M. J. (2014). Effects of KinesioTaping on foot posture in participants with pronated foot: a quasi-randomised, double-blind study. *Physiotherapy* 100, 36–40. doi: 10.1016/j.physio.2013.04.005

Mills, E. J., Chan, A. W., Wu, P., Vail, A., Guyatt, G. H., and Altman, D. G. (2009). Design, analysis, and presentation of crossover trials. *Trials* 10:27. doi: 10.1186/1745-6215-10-27

Missenard, O., Mottet, D., and Perrey, S. (2008). The role of cocontraction in the impairment of movement accuracy with fatigue. *Exp. Brain Res.* 185, 151–156. doi: 10.1007/s00221-007-1264-x

Morasso, P. G., and Schieppati, M. (1999). Can muscle stiffness alone stabilize upright standing? *J. Neurophysiol.* 82, 1622–1626.

Oliveira, A. S., Gizzi, L., Kersting, U. G., and Farina, D. (2012). Modular organization of balance control following perturbations during walking. *J. Neurophysiol.* 108, 1895–1906. doi: 10.1152/jn.00217.2012

Paoloni, M., Bernetti, A., Fratocchi, G., Mangone, M., Parrinello, L., Del Pilar, C. M., et al. (2011). Kinesio Taping applied to lumbar muscles influences clinical and electromyographic characteristics in chronic low back pain patients. *Eur. J. Phys. Rehabil. Med.* 47, 237–244.

Patrick, M., Ditunno, P., Ditunno, J. F., Marino, R. J., Scivoletto, G., Lam, T., et al. (2011). Consumer preference in ranking walking function utilizing the

walking index for spinal cord injury II. *Spinal Cord* 49, 1164–1172. doi: 10.1038/sc.2011.77

Penn, R. D. (1988). Intrathecal baclofen for severe spasticity. *Ann. N. Y. Acad. Sci.* 531, 157–166. doi: 10.1111/j.1749-6632.1988.tb31822.x

Podsiadlo, D., and Richardson, S. (1991). The timed "Up and Go": a test of basic functional mobility for frail elderly persons. *J. Am. Geriatr. Soc.* 39, 142–148.

Poole-Wilson, P. A. (2000). The 6-minute walk. A simple test with clinical application. *Eur. Heart J.* 21, 507–508. doi: 10.1053/euhj.1999.1970

Riemann, B. L., and Lephart, S. M. (2002). The sensorimotor system, Part II: the role of proprioception in motor control and functional joint stability. *J. Athl. Train.* 37, 80–84.

Rossier, P., and Wade, D. T. (2001). Validity and reliability comparison of 4 mobility measures in patients presenting with neurologic impairment. *Arch. Phys. Med. Rehabil.* 82, 9–13. doi: 10.1053/apmr.2001.9396

Roy, R. R., Harkema, S. J., and Edgerton, V. R. (2012). Basic concepts of activity-based interventions for improved recovery of motor function after spinal cord injury. *Arch. Phys. Med. Rehabil.* 93, 1487–1497. doi: 10.1016/j.apmr.2012.04.034

Scivoletto, G., Romanelli, A., Mariotti, A., Marinucci, D., Tamburella, F., Mammone, A., et al. (2008). Clinical factors that affect walking level and performance in chronic spinal cord lesion patients. *Spine* 33, 259–264. doi: 10.1097/BRS.0b013e3181626ab0

Simoneau, G. G., Degner, R. M., Kramper, C. A., and Kittleson, K. H. (1997). Changes in ankle joint proprioception resulting from strips of athletic tape applied over the skin. *J. Athl. Train.* 32, 141–147.

Tamburella, F., Scivoletto, G., Cosentino, E., and Molinari, M. (2013b). Walking in water and on land after an incomplete spinal cord injury. *Am. J. Phys. Med. Rehabil.* 92(10 Suppl. 1), e4–e15. doi: 10.1097/PHM.0b013e3182a1e6c3

Tamburella, F., Scivoletto, G., and Molinari, M. (2013a). Balance training improves static stability and gait in chronic incomplete spinal cord injury subjects: a pilot study. *Eur. J. Phys. Rehabil. Med.* 49, 353–364.

Thelen, M. D., Dauber, J. A., and Stoneman, P. D. (2008). The clinical efficacy of kinesio tape for shoulder pain: a randomized, double-blinded, clinical trial. *J. Orthop. Sports Phys. Ther.* 38, 389–395. doi: 10.2519/jospt.2008.2791

van Hedel, H. J., and Dietz, V. (2010). Rehabilitation of locomotion after spinal cord injury. *Restor. Neurol. Neurosci.* 28, 123–134. doi: 10.3233/RNN-2010-0508

Westerkam, D., Saunders, L. L., and Krause, J. S. (2011). Association of spasticity and life satisfaction after spinal cord injury. *Spinal Cord* 49, 990–994. doi: 10.1038/sc.2011.49

Wewers, M. E., and Lowe, N. K. (1990). A critical review of visual analogue scales in the measurement of clinical phenomena. *Res. Nurs. Health* 13, 227–236. doi: 10.1002/nur.4770130405

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.

Received: 09 January 2014; accepted: 13 May 2014; published online: 30 May 2014.
Citation: Tamburella F, Scivoletto G and Molinari M (2014) Somatosensory inputs by application of KinesioTaping: effects on spasticity, balance, and gait in chronic spinal cord injury. Front. Hum. Neurosci. ***8****:367. doi: 10.3389/fnhum.2014.00367*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 12 June 2014
doi: 10.3389/fnhum.2014.00416

Feasibility of visual instrumented movement feedback therapy in individuals with motor incomplete spinal cord injury walking on a treadmill

Daniel Schließmann[1†], Christian Schuld[1†], Matthias Schneiders[1], Steffen Derlien[2], Maria Glöckner[2], Till Gladow[3], Norbert Weidner[1] and Rüdiger Rupp[1]*

[1] *Experimental Neurorehabilitation, Spinal Cord Injury Center, Heidelberg University Hospital, Heidelberg, Germany*
[2] *Institut für Physiotherapie, University Hospital Jena, Jena, Germany*
[3] *HASOMED GmbH, Magdeburg, Germany*

Edited by:
Federica Tamburella, IRCCS Fondazione Santa Lucia, Italy

Reviewed by:
Giorgio Scivoletto, IRCCS Fondazione Santa Lucia, Italy
Juan C. Moreno, Spanish National Research Council, Spain

****Correspondence:***
Rüdiger Rupp, Experimental Neurorehabilitation, Spinal Cord Injury Center, Heidelberg University Hospital, Schlierbacher Landstrasse 200a, Heidelberg 69118, Germany
e-mail: ruediger.rupp@med.uni-heidelberg.de

† These authors have contributed equally to this work.

Background: Incomplete spinal cord injury (iSCI) leads to motor and sensory deficits. Even in ambulatory persons with good motor function an impaired proprioception may result in an insecure gait. Limited internal afferent feedback (FB) can be compensated by provision of external FB by therapists or technical systems. Progress in computational power of motion analysis systems allows for implementation of instrumented real-time FB. The aim of this study was to test if individuals with iSCI can normalize their gait kinematics during FB and more importantly maintain an improvement after therapy.

Methods: Individuals with chronic iSCI had to complete 6 days (1 day per week) of treadmill-based FB training with a 2 weeks pause after 3 days of training. Each day consists of an initial gait analysis followed by 2 blocks with FB/no-FB. During FB the deviation of the mean knee angle during swing from a speed matched reference (norm distance, ND) is visualized as a number. The task consists of lowering the ND, which was updated after every stride. Prior to the tests in patients the in-house developed FB implementation was tested in healthy subjects with an artificial movement task.

Results: Four of five study participants benefited from FB in the short and medium term. Decrease of mean ND was highest during the first 3 sessions (from 3.93 ± 1.54 to 2.18 ± 1.04). After the pause mean ND stayed in the same range than before. In the last 3 sessions the mean ND decreased slower (2.40 ± 1.18 to 2.20 ± 0.90). Direct influences of FB ranged from 60 to 15% of reduction in mean ND compared to initial gait analysis and from 20 to 1% compared to no-FB sessions.

Conclusions: Instrumented kinematic real-time FB may serve as an effective adjunct to established gait therapies in normalizing the gait pattern after incomplete spinal cord injury. Further studies with larger patient groups need to prove long term learning and the successful transfer of newly acquired skills to activities of daily living.

Keywords: spinal cord injury, visual realtime feedback, proprioception, gait rehabilitation, treadmill, motion analysis, motor learning

INTRODUCTION

A spinal cord injury (SCI) leads to persistent sensorimotor impairments in the lower (paraplegia) and upper (tetraplegia) extremities (McDonald and Sadowsky, 2002) resulting in limitations of mobility and participation in society. Over the last decade a clear trend toward a higher proportion of cases with motor incomplete SCI (iSCI) can be seen worldwide. Currently about 60% of all new spinal cord injuries are incomplete (National Spinal Cord Injury Statistical Center, 2013). Individuals with initially preserved motor functions below the level of lesion have a good prognosis to become ambulatory after 3–6 month (Katoh and el Masry, 1995; Burns et al., 1997). However, even in those subjects who reach a sufficient level of ambulation, sensory and in particular proprioceptive impairments or spasticity may be present and limiting walking function. Restrictions in deep sensibility lead to an altered or absent intrinsic afferent feedback to the central nervous system. It is known that a loss of afferent feedback severely affects motor control particularly within a changing environment (Riemann and Lephart, 2002; Schmidt and Lee, 2011). Therefore, one characteristic of neurological gait disorders is a non-physiological and instable gait pattern, often accompanied by the fear to fall (Sanes et al., 1985; Dietz, 2002). Under normal conditions the control of the walking pattern requires little attention. However, adaptation of the walking pattern to changing environments is mainly controlled by cerebellar and cerebral motor areas (Morton and Bastian, 2006). These

conscious processes preferentially access the spatial and not the temporal walking pattern (Malone and Bastian, 2010).

If intrinsic feedback is impaired, extrinsic feedback (FB) provided by human therapists or technical systems may compensate for the loss of sensory function and may help to restore a physiological walking pattern. It was shown that patients with cerebral palsy are able to normalize their pathological gait pattern, if deviations from a normal walking pattern are fed back while walking in a locomotion robot on a treadmill (Labruyere et al., 2013; Meyer-Heim and van Hedel, 2013). Within a training program following motor learning principles FB is an effective tool to boost motor learning (Krakauer, 2006). It can be provided using haptic, visual, auditory modalities either alone or in combinations, because all have dedicated strengths and weaknesses (Sigrist et al., 2011, 2013). Besides modality, FB frequency and timing are important factors for performance and learning efficacy (Park et al., 2000; Maslovat et al., 2009; Sigrist et al., 2013). High-frequency or concurrent FB seems to be beneficial for inexperienced learners and for those confronted with complex tasks (Wulf et al., 1998; Wulf and Shea, 2002), whereas for simple motor tasks lower FB frequencies seem to be more beneficial (Maslovat et al., 2009).

While principles of motor learning and FB are well investigated in sport sciences and commonly applied in optimizing sportsmen performance (Wulf et al., 2002; Anderson et al., 2005; Schmidt and Lee, 2011; Sigrist et al., 2011; Keogh and Hume, 2012; Thow et al., 2012), the integration of FB methods in gait rehabilitation programs of iSCI patients has just recently started (Banz et al., 2008; Duschau-Wicke et al., 2010; Schuck et al., 2012; Govil and Noohu, 2013). Most studies related to FB during walking are based on the Lokomat, which is a motor-driven gait orthosis for robotic assistance during treadmill training (Colombo et al., 2001). Beside its actuator components the device can be extended to use sensor data for quantification of hip and knee joint angles and torques for FB (Duschau-Wicke et al., 2010; Schuck et al., 2012). Due to the small number of sensors important gait parameters e.g., kinematics of the feet cannot be assessed easily. Additionally, information about the natural walking pattern of a user cannot be obtained with the locomotion robot due to its mechanical constraints allowing only movements in the sagittal plane.

Another issue is the implementation of the feedback. In a recent study focusing on the feasibility of patient-cooperative robotic gait training FB was provided in form of a moving reference avatar, which is overlaid by the real kinematic parameter of the user (Schuck et al., 2012). Although, the specific contribution of the FB on the improvement of overground walking were not assessed and systematic investigations are missing, based on clinical experience it can be assumed that this rather unspecific FB may be less efficient compared to joint or segment specific FB. This statement is underlined by the results of a recently published randomized controlled study, which showed positive effects of FB of the electromyogram derived from the M. gluteus maximus on the walking function of individuals with iSCI (Govil and Noohu, 2013). However, in already ambulatory individuals it might be more important to achieve a more physiological walking pattern to avoid long-term complications instead of a higher walking velocity. If a more physiological walking pattern is in the focus of the therapy, a kinematic gait analysis is indispensable.

So far all studies investigating the effect of FB on the walking pattern of individuals with iSCI have shown that (1) motivation rises resulting in a more active participation of the participants during the training sessions (Krakauer, 2006), that (2) they are able to alter their gait pattern during FB and that (3) positive effects on the gait capabilities after the FB training can be maintained in the short-term. However, retention and potential learning effects during no-FB sessions after a longer pause period are mainly unknown. Hence to date, it is not entirely clear if subjects with iSCI and the associated sensorimotor impairments are able to influence their pathological gait pattern during FB application. Most importantly, it is still an open question, if iSCI subjects have an improved long-term outcome i.e., that the application of FB has the characteristics of a therapy. The following study represents a feasibility study aiming at improving the gait pattern of ambulatory chronic iSCI individuals with predominantly sensory impairments by application of a novel motion analysis based real-time FB system. The technical implementation of the feedback modality had to be tested with motor unimpaired subjects in a pilot study before applying it to individuals with iSCI.

MATERIALS AND METHODS

MOTION ANALYSIS AND FEEDBACK SYSTEM

For the intended study a dedicated FB system featuring real-time motion analysis (Motion Analysis Inc., Santa Rosa, CA, USA) consisting of 8 Hawk cameras to capture a volume of approximately 1.5 × 1.4 × 2 m on a custom-made treadmill (Rupp et al., 1998) and a proprietary FB software (**Figure 1**) was developed. A standard Helen Hayes marker set (Kadaba et al., 1990) consisting of 20 retroflective markers (Ø 10 mm) together with the dedicated biomechanical model of the lower extremities was used to calculate angles of the hip, knee and ankle joints online with a sampling frequency of 200 Hz. Gait events were estimated algorithmically by detecting local extremes in the trajectories of heel and toe markers by a validated proprietary implementation optimized for low detection latencies (Schablowski-Trautmann, 2005).

The commercial software of the motion analysis system (Cortex) streams marker trajectories, analog data and the model based joint angles with a low latency of less than 2 frames via multicast into the local area network. The low-level module of the proprietary FB software written in C++ using Cortex SDK 4.1.6 (Motion Analysis Inc., Santa Rosa, CA, USA) and the QT toolkit 4.8.4 (Digia Plc, Helsinki, Finland) receives the multicast stream, performs basic signal processing tasks e.g., low latency gait event detection, time distance parameter calculation and gait cycle normalization. The high-level module based on the MATLAB 2010a engine (The MathWorks, Inc., Natick, MA, USA) computes parameters for use as FB values e.g., deviations of the current step from a physiological kinematic walking pattern (see below) or deviations of the focal step from an artificial movement task, like the one used for feasibility testing with healthy individuals (see Pilot study with non-impaired individuals). By making extensive use of high-level MATLAB functions FB parameters can be visualized by a projector on a canvas in 3.5 m distance. FB

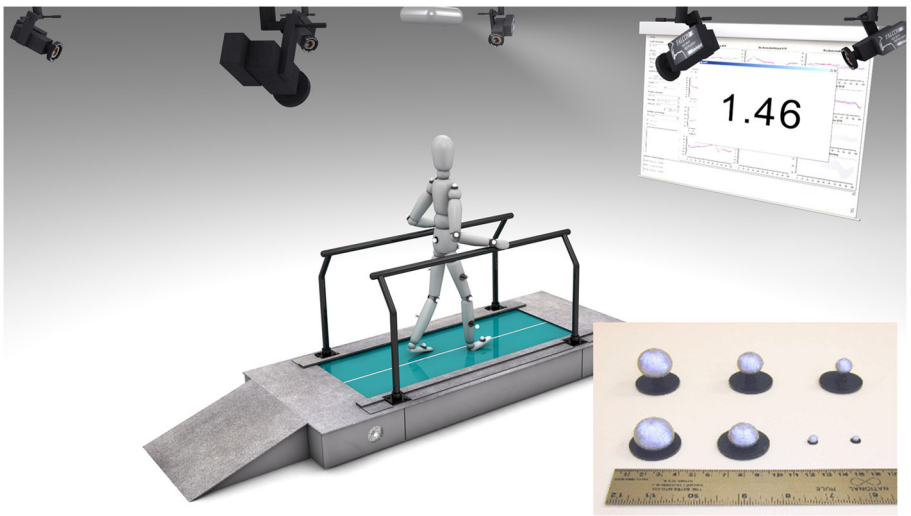


FIGURE 1 | Components of the feedback system used in this study. Infrared motion capture system using reflective markers associated with a customized treadmill. Knee joint angles are calculated in real-time during every step and compared to reference values. The calculated norm distance value is fed back to the participant using a projector and canvas.

was provided with a latency of <150 ms after the pre-defined gait event, which is in this case the heel strike.

For implementation of the FB, joint angles were normalized to 100% gait cycle. A scalable norm distance (ND) measure (Wolf et al., 2006) was used as FB parameter in the iSCI cohort. In general, ND reflects the deviations from a physiological gait pattern. It is defined as the difference between the actual, pathological joint angle x_p and the mean norm angle $\overline{x_n}$ of a gait velocity dependent cohort of non-impaired subjects (Schablowski-Trautmann, 2005) weighted by the standard deviation of the norm angle σ_n at point k within a step normalized (0 – 100 percent gait cycle) joint angle:

$$ND_p[k] = \left| \frac{x_p[k] - \overline{x_n}[k]}{\sigma_n[k]} \right| \quad (1)$$

The ND has in contrast to other methods like averaging the inherent advantage of including a-priori knowledge of normal walking when used as a FB parameter. ND values between 0 and 1 indicate physiological gait patterns, whereas values >>1 indicate pathological ones (Schablowski-Trautmann et al., 2006).

PILOT STUDY WITH NON-IMPAIRED INDIVIDUALS

In a case series motor unimpaired individuals walking on a treadmill received visual FB about their mean knee flexion angle (0° flexion angle means fully extend knee) during swing phase. Treadmill speed was set at 0.8 m/s for all participants. Each participant first started with an initial reference measurement of 120 s to assess the normal walking pattern without FB. This reference measurement was followed by four sessions each lasting for 120 s, in which the subject had to perform different artificial movement tasks with the support of visual FB (**Figure 2A**). During each session the individuals had to adjust their flexion angle of one knee during swing phase to a predefined percentage

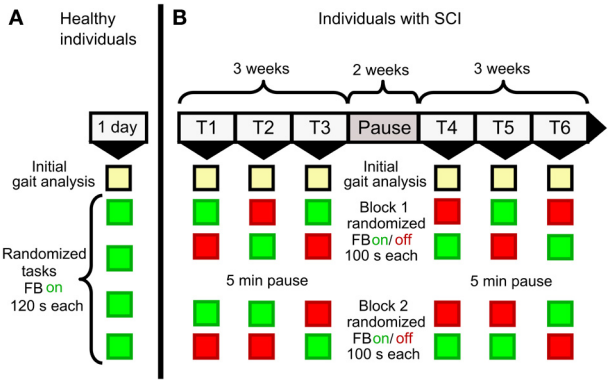


FIGURE 2 | Study protocol for (A) non-impaired and (B) iSCI individuals. Non-impaired individuals completed a reference measurement (initial gait analysis) followed by 4 sessions with artificial movement tasks (+20, +40, −20, −40% of previously recorded mean knee angles during normal walking) in randomized order. Individuals with iSCI completed 6 weeks (1 training day per week, two blocks of feedback/ no-feedback sessions per day of training) of FB training with a pause after the third week. Those blocks were randomized, except for the first block of the first day of training, which started with a FB session. Equivalent to the condition in the non-impaired participants each session started with an initial gait analysis.

(+20, −20, +40, −40%) of the previously determined reference angle, which is the mean knee angle during swing phase averaged over all detected strides on one side. The order of movement tasks, as well as the body side to which FB was applied, was randomized. The definition of the artificial movement tasks on the basis of each individual's own reference angle was made to assure that tasks were equally challenging for each participant.

Visual FB consisted of a line diagram (**Figure 3**) where the mean knee flexion angle during swing phase is displayed on the

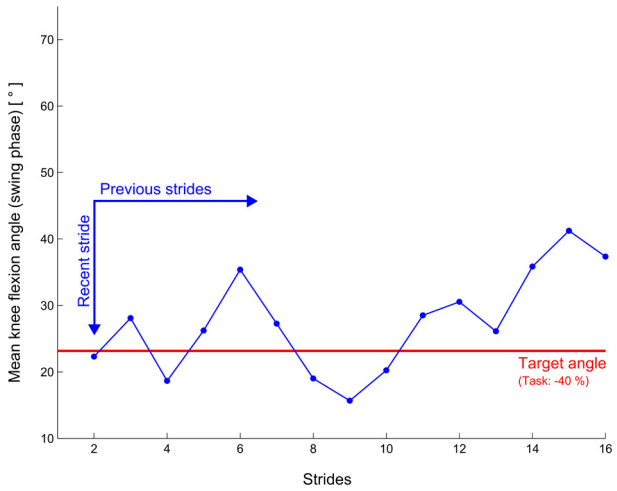


FIGURE 3 | Illustration of the feedback display of the pilot study with non-impaired subjects. The blue graph is shifted to the right on every stride.

y-axis and the latest 16 FB values on the x-axis. The diagram was updated after every stride of the respective side. A red horizontal line indicated the optimum of the movement task i.e., the desired mean knee flexion angle during swing phase. After receiving a brief verbal introduction on relevant gait events, the movement tasks and FB implementation, the participants were instructed to adjust their knee flexion in order to match the FB value as good as possible to the red line. The different movement tasks were presented consecutively with about 10 s pause in between to let the operator switch to the next task. Participants were asked to walk normally during this transition period.

A movement task was considered successfully performed, if the stepwise mean group performance accuracy as measured by the accumulated standard error dropped below 1°.

FEASIBILITY STUDY WITH iSCI INDIVIDUALS

For the feasibility study, individuals with chronic iSCI (date of injury >12 months) and predominantly sensory deficits in the lower extremities were included. Additionally, basic walking function i.e., walking with a walker or with less support by walking aids, was preserved (Walking Index for Spinal Cord Injury II (Marino et al., 2010) (WISCI II) ≥13). Patients were recruited by screening of the medical records of the SCI Center of the Heidelberg University Hospital. Subjects with stiff-knee gait pattern were selected by observational gait analysis. A stiff-knee gait is characterized by an insufficient flexion of the knee during swing phase. Hand and arm function had to be preserved to a degree sufficient to hold on to parallel bars during treadmill walking.

Each individual completed 2 episodes of 3 weeks of FB training on the treadmill with 1 training day per week and a 2 weeks pause after the first episode (**Figure 2B**) resulting in 6 training days in total. Every training day started with an initial gait analysis to assess the individual's current gait kinematics followed by 2 therapy blocks each consisting of one FB/no-FB session (100 s each) in randomized order, except for the very first block on the first training day, which always started with a FB session. To follow the idea of distributed practice (Krakauer, 2006) therapy blocks included no-FB sessions to avoid full dependence on FB but rather promote medium and long-term motor learning. Visual FB was given after every stride as an absolute number of the ND (see equation 1) of the knee angle during swing phase of the most affected leg. The number was updated on every heel strike of this leg. Font size was set to 180 points to ensure legibility for every participant and the number was displayed with two fractional digits (**Figure 1**). In contrast to the pilot study with non-impaired individuals, only numerical and no graphical representation of ND was presented. Prior to FB sessions, individuals were instructed to actively lower this number by altering their gait pattern. The operator suggested movement strategies such as "Perform faster thigh movements during hip flexion" or "Actively increase your knee flexion by trying to lift the heel up closer to the buttock." Additionally, hints for altering cadence and foot placement were given that might help an individual to achieve a more physiological gait pattern. The verbal instructions were the same for FB and no-FB sessions, where individuals were asked to focus on the movement task. During walking sessions the operator reduced communication to a minimum in order to avoid any distraction of the subject from the movement task. For each initial gait analysis, individuals were asked to walk like in daily life, without concentration on their walking pattern. In this case, no additional verbal information was provided. At study inclusion, treadmill speed was adjusted to the individual comfortable walking speed to minimize mental and physical fatigue during the training session. The self-selected walking speed of each individual was kept constant during all sessions. Keeping the treadmill speed constant is a prerequisite for comparing the NDs between sessions due to the large influence of walking speed on gait kinematics (Schablowski-Trautmann et al., 2006).

For standardized assessment of the SCI related sensorimotor deficits motor and sensory (pin prick discrimination, light touch appreciation) examinations were performed at study onset according to the International Standards for Neurological Classification of Spinal Cord Injury (ISNCSCI) published by the American Spinal Injury Association (ASIA) (Kirshblum et al., 2011). A high-quality ISNCSCI assessment was assured by trained assessors (Schuld et al., 2013) and computational scaling, scoring and classification (Schuld et al., 2012) including the ASIA Impairment Scale. The preserved degree of proprioception in the knee joint was assessed prior to the first training session by a vibratory sensation testing at the epicondylus medialis femoris of the leg selected for FB using a tuning fork (64 Hz testing frequency, scale 0–8, minimal to maximal sensitivity).

Confirmatory statistics were not considered due to the small sample size and the feasibility character of the study. Therefore, only descriptive statistics were performed. The main goal of the study was to test the feasibility of this completely new therapy approach in iSCI and to determine effect sizes for future clinical trials. Descriptive statistics and visualization were performed using R 3.0.0 (R Core Team, 2012) and MATLAB 2010a. This feasibility study has been approved by the institutional review board. All non-impaired and impaired study participants gave written informed consent.

RESULTS

PILOT STUDY

Nine motor unimpaired individuals (age: 31.4 ± 5.3 years; 2 females, 7 males, **Table 1**) completed the FB training. All participants managed to perform every movement task within the training duration of 120 s. **Figure 4** depicts the mean performance curves per task. The number of strides varied within participants and tasks. Accordingly, the ensemble average was only calculated for the least common number of strides per person and session, which was 83 for this study.

The −40% task was successfully performed after 23 strides (red line in **Figure 4A**) and the mean error during the FB trial was 1.87 ± 3.88°. During the movement task of −20% the desired knee flexion angle was reached after 10 strides (**Figure 4C**) with a mean error of 0.58 ± 1.85°. The +20% task was successfully fulfilled after 7 strides with a mean error of −0.30 ± 1.85° (**Figure 4E**) and +40% after 15 strides with a mean error of −0.92 ± 3.17° (**Figure 4G**) respectively. Movement tasks with positive algebraic signs which needed an adjustment of the mean knee angle during swing phase toward larger values yielded slightly smaller outcome values both in movement task approximation and mean errors compared to their negative counterparts. Reflecting the magnitude of the before mentioned average mean errors of the gait tasks, gait cycle curves were below (**Figures 4B,D**) and slightly above (**Figures 4F,H**) the mean normal gait cycle during swing phase.

FEASIBILITY STUDY

Five chronic (23.6 ± 8.6 months after injury) individuals (age: 55.2 ± 8.5 year, 3 female, 2 male) with iSCI (4 ASIA Impairment Scale (AIS) D, 1 AIS B) successfully completed all 6 days of training (self-selected gait speed 0.5 ± 0.1) (**Table 2**). Individuals with iSCI showed a strong decrease in mean ND of the initial gait analysis without FB from 3.93 ± 1.54 on the first day of training to 2.20 ± 0.90 on the sixth day of training. The strongest decrease of 1.75 (3.93 ± 1.54 to 2.18 ± 1.04) is observed over the course of the first three sessions (**Figure 5**). After a 2 week pause, on the fourth day of training mean ND remains on the same level compared to the third day (2.18 ± 1.04 and 2.40 ± 1.18, respectively) before the pause. Compared to the first half of the total days of training a smaller decrease of 0.21 (2.40 ± 1.18 to 2.20 ± 0.90) occurred over the second half. Considerable direct benefits of FB occurred in four individuals with a significant decrease of the mean ND of all FB sessions (ranging from 60 to 15%) relative to all initial gait analyses. Direct benefits of FB sessions relative to no-FB sessions ranged from 20 to 1% (**Figure 6A**). Individual 10401 showed a slight decrease in performance (−6 and −5%, respectively). Four individuals reduced their ND already in the first FB session (**Figure 6B**). The decrease in ND observed in the first FB sessions ranged from 73 to 21% relative to the first initial gait analysis, and −24% for individual 10401. Decreases in ND of the first FB session relative to the first no-FB session ranged from 49 to 0.03%. Individuals 10401 and 10586 showed increases in ND (−7.5 and −17%, respectively).

Vibration sensitivity in the knee joints was measured for all individuals, however due to hyperaesthesia, individual 10,586 did not yield any reliable data for this assessment. Individuals with a high sensitivity to vibration in the knee joint showed a high immediate benefit from FB training—both related to initial and no-FB trails—, whereas in patients with lower or no sensitivity lower direct and immediate benefits from FB were observed (**Figures 6A,B**). This correlation can also be seen at a medium-term learning level, displayed as the individual difference in ND between the first initial gait analysis and the first gait analysis after a 2 weeks pause (**Figure 6C**). Individuals with a high immediate benefit from FB also showed high levels of retention after the 2 weeks pause (**Figures 6B,C**).

To visualize the relation between ND and kinematics and the effect of FB on the knee angle trajectory, short (within session) and medium-term benefits are exemplarily described for individual 10447 (**Figure 7**). The stiff-knee gait with decreased knee flexion during swing phase has been confirmed by the initial gait analysis (**Figure 7A**) with a high mean ND value of 5.85. During the FB trial, swing phase kinematics approached the norm trajectory and ND values decreased to 1.36 (**Figure 7B**). The initial gait analysis of the third training session showed an intermediate ND value of 3.66 and a knee flexion angle with an intermediate distance to the norm curve (**Figure 7C**).

Table 1 | Data of non-impaired individuals participating in the pilot study.

ID	Sex	FB side	Age (years)	Mean swing knee flexion angle [°]
10569	m	R	35	48.01
10672	f	L	23	38.06
10671	f	R	28	32.28
10440	m	L	38	36.34
10144	m	L	38	46.45
10635	m	R	34	43.46
10588	m	R	26	40.74
10567	m	R	31	41.80
10620	m	L	30	48.66
		Mean	31.44	41.76
		SD	5.25	5.54

Mean knee flexion angle during swing phase as recorded in the reference measurement episode.

DISCUSSION

FB IN NON-IMPAIRED SUBJECTS

Our results show that the implemented visual FB setup of frequent terminal FB using a novel treadmill based real-time FB system is effective in supporting non-impaired individuals to alter their normal gait patterns. The artificial movement task of precisely altering the knee flexion angle during swing phase could be mastered with support of FB. In our setup the participants successfully managed to adapt their walking pattern to the movement task within a few steps, which represents a very short adaptation time. In another study qualitative real-time video feedback was used to support study participants in improving their gait symmetry on a split-belt treadmill (Malone and Bastian, 2010). The study results show that it took healthy

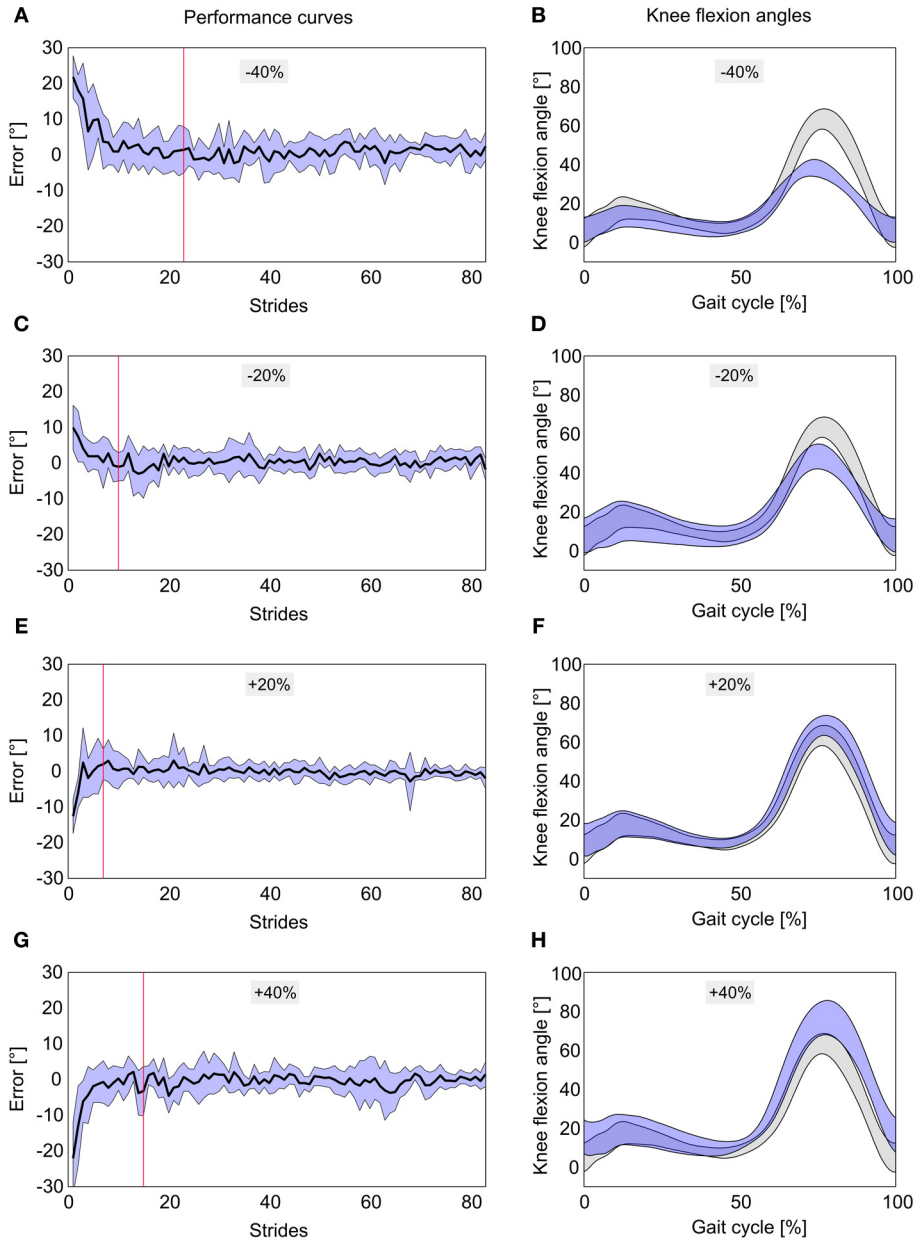


FIGURE 4 | Performance curves and mean knee flexion angles to the movement tasks −40% (A,B), −20% (C,D), +20% (E,F), and +40% (G,H) of normal knee flexion angles. Performance curves display 83 steps during feedback training averaged over 9 non-impaired individuals ± *SD* (blue area). Red vertical lines mark the point where the SE of the mean error is within ±1°. Mean knee flexion angles are displayed as blue curves averaged over 9 non-impaired individuals. Gray curves represent mean knee flexion angles ± *SD* of all participants during normal walking (initial gait analysis).

subject approximately 250 strides to compensate for the gait deviations induced by the different belt speeds. Our findings either lead to the conclusions that the tasks were very easy, or that the implemented FB paradigm was effective, or both. Most volunteers judged the task as easy and the visual display as comprehensive. However, lacking any retention test we only assessed performance but not motor learning. Interestingly, tasks requiring less knee flexion during swing phase i.e., −20 and −40%, appeared to be slightly more difficult than the ones requiring more knee flexion. One possible explanation could be that increased knee flexion is commonly used for obstacle stepping in daily life and thus inherent to each participant's movement repertoire.

In conclusion, this experiment shows the technical feasibility of the FB system and confirms our hypothesis that the modulation of knee angles during swing phase is an easy task for healthy individuals, which is a mandatory prerequisite for providing this type of FB for iSCI subjects.

Table 2 | Characteristics of individuals with SCI included in the feasibility study.

ID	Sex	AIS	NLI	Age	MAI	LEMS (max. 50)	PP (max. 112)	LT (max. 112)	VIB (max. 8)	WISCI II (max. 20)	S [m/s]
10447	m	D	Th8	42	22	47	73	88	6	16	0.6
10521	f	B	L3	65	25	47	96	102	6	13	0.6
10522	f	D	C6	59	23	48	109	108	1	20	0.46
10401	f	D	Th6	54	36	43	107	91	0	16	0.42
10586	m	D	Th10	56	12	48	93	107	NA	16	0.4
			Mean	55.2	23.6	46.6	95.6	99.2	3.25	16.2	0.5
			SD	8.5	8.6	2.1	14.4	9.2	3.2	2.5	0.1

Age, sex, months after injury (MAI), data according to the International Standards for Neurological Classification of Spinal Cord Injury (ISNCSCI): Asia Impairment Scale (AIS), Neurological Level of Injury (NLI), Lower Extremity Motor Score (LEMS), Pin Prick score (PP), Light Touch score (LT), and the Walking Index for Spinal Cord Injury II (WISCI II). Knee joint vibration sensation was measured (VIB) as an estimation for proprioception. Treadmill speed (S) was set to the physical capability of each individual.

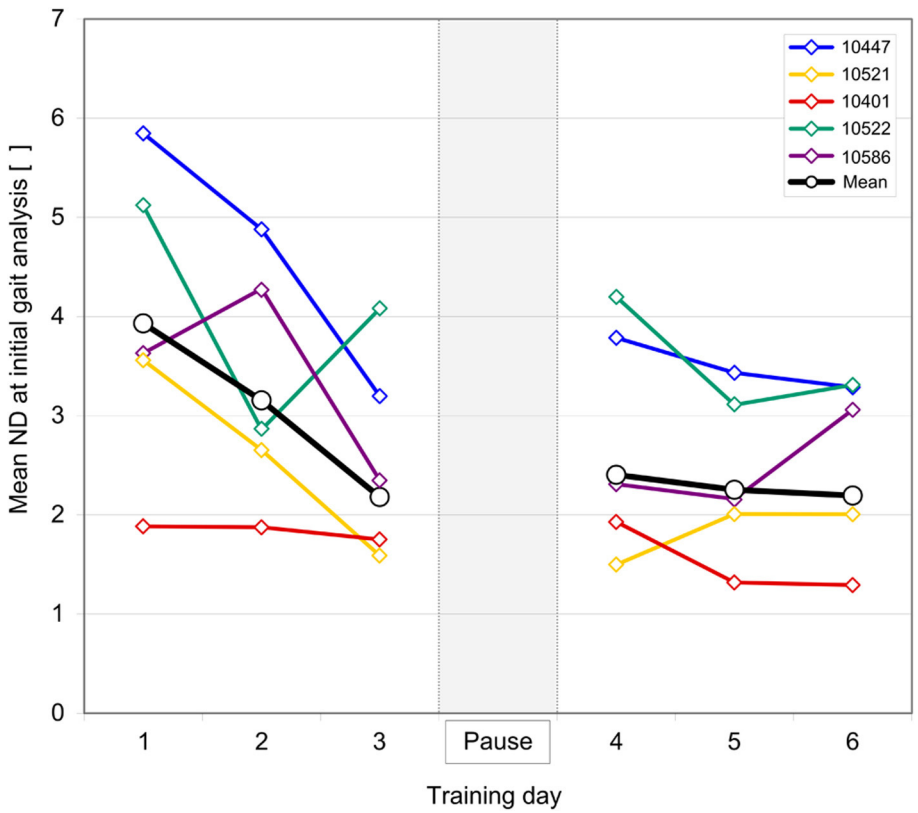


FIGURE 5 | Learning curves of individuals with iSCI over 6 days of training with a 2 weeks pause in between. Norm distance (ND) values were derived from initial gait analysis (without feedback) preceding every feedback session. Brown lines indicate mean ND over all individuals with iSCI.

FB IN INDIVIDUALS WITH iSCI

This study is to our knowledge the first 3 dimensional lower-body kinematic approach for a real-time FB therapy in individuals with iSCI. Our results showed that 4 out of 5 chronic iSCI individuals with stiff-knee gait and predominant sensory impairments are capable to adapt their pathological gait pattern toward more physiological knee kinematics during the application of visual FB. More importantly, the carry-over effects seen after 2 weeks of pause confirm that at least medium-term motor learning occurred. Study participants continued their therapy program during the study. Therefore, the improvements occurred during the study cannot be attributed to changes in the regular training intensity, which qualifies the application of real-time FB as a therapy in individuals with iSCI.

Beyond the expected benefits of FB in the motor learning context, it can be assumed that discussing the results of the initial gait analysis with the patients supported participants in consciously altering their otherwise unconsciously imprinted pathologic motor programs. This is indicated by the strong immediate effects between initial gait analysis and first FB session

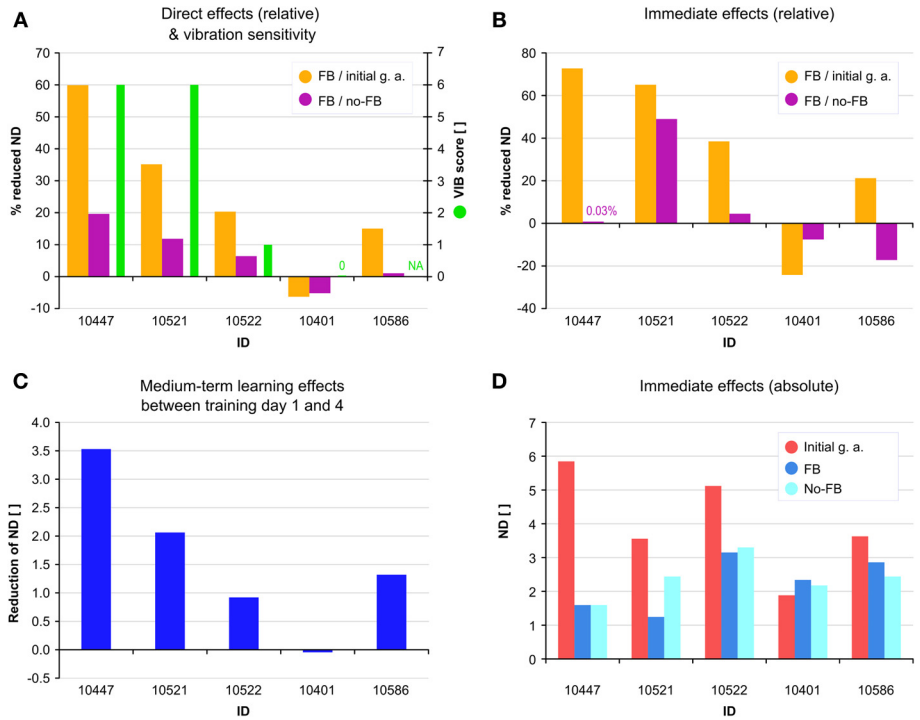


FIGURE 6 | (A) Direct effects of feedback FB as reduced norm distance (ND) during all FB sessions relative to all initial gait analyses (orange) and relative to all no-FB sessions (purple). Vibration sensitivity (VIB) score is displayed as green bars. **(B)** Immediate effects of FB during the first FB session relative to the first initial gait analysis (orange) and relative to the first no-FB session (purple). **(C)** Medium-term learning effects as reduction of ND between day 1 and 4 of training. **(D)** Immediate effects of FB displayed as absolute values in ND during the first initial gait analysis (red), first FB session (blue) and first no-FB session (bright blue).

on the first training day. The extent and direction of this immediate change (**Figures 6B,D**) seems to reflect the size of the medium-term (**Figure 6C**) and the direct effects (**Figure 6A**). Contrary, when FB was switched off during the first no-FB trial (**Figure 6B**), patients showed very diverse short-term carry-over effects ranging from unchanged performance over considerable performance loss up to performance gains (individual 10,586). Of course, these observations are made on an individual level during the very first application of the FB system and may therefore not be representative for the progress of each individual over the course of the study. Similar to healthy subjects individuals with iSCI can adapt their kinematic pattern to the given movement task within the first FB session. As the amount of individual improvement within the first FB session seems to be associated with direct- and medium-term effects, the improvement during the initial session may identify therapy responders and serve as a marker for the final therapy outcome. In addition to the previously discussed conscious altering of movement patterns, some form of learning could also have occurred on the spinal level (Wolpaw and Carp, 1990; Thompson and Wolpaw, 2014). Due to the fact that already in the first session, which lasts for 200 s, positive effects of FB training occurred, we suggest that alterations in conscious supraspinal motor control most contribute to the early changes in motor behavior and task performance. On the medium- and long-term, adaptations on spinal level might contribute to the manifestation of the more physiological movement pattern.

The individuals with iSCI differed substantially in vibration sensing in the knee joint and in initial performance regarding ND of their treated leg. Interestingly, we observed that patients with low vibration sensitivity benefited less from FB directly, as well as in the medium-term. This observation does not support the assumption that FB may totally compensate for sensory impairments. For the adaptation of the kinematic pattern during FB sessions and for the formation or alteration of a motor program afferent, proprioceptive information is needed (Nielsen and Sinkjaer, 2002; Schmidt and Wrisberg, 2008). Our study participants only received FB in the form of knowledge of results through a rather abstract number after the stride. According to the specificity of learning hypothesis, the most optimal afferent information is integrated into the developing motor program (Proteau, 1992). Considering this, the external FB provided in our specific setup may not be optimal to totally compensate the disturbed internal, proprioceptive feedback. To the contrary, due to its abstract nature, the FB might have provided only moderate guidance (Maslovat et al., 2009; Sigrist et al., 2013), but was intended to promote retention, as individuals were intended to make better use of their preserved proprioception. Our therapy regime represents a motor learning and restorative approach rather than compensatory strategy and therefore does not impose maximal guidance. However, considering the very low sample size, such interpretations must be treated very carefully.

Open questions for future studies include the detection of the right target group for this kind of FB training. Which degree of

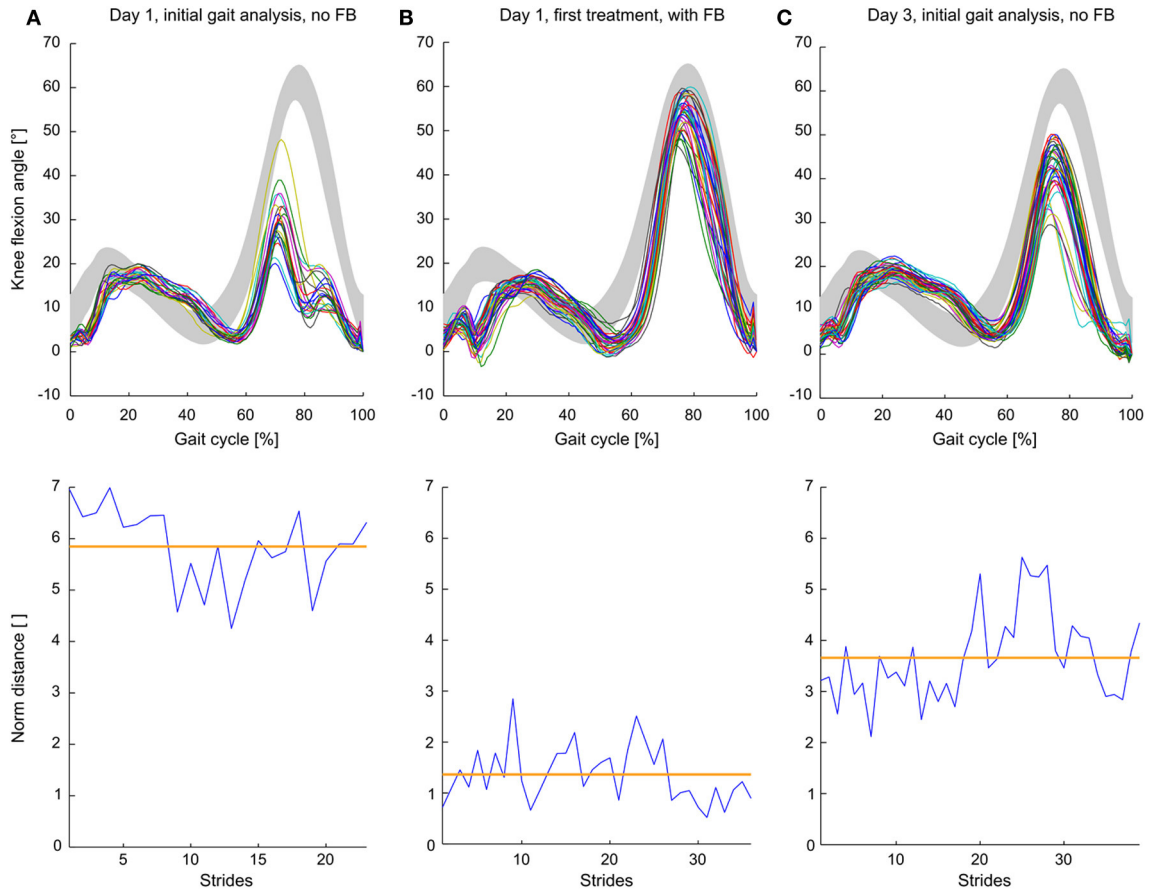


FIGURE 7 | Step normalized knee angle curves and norm distance values of a single individual with iSCI during (A) initial gait analysis (no feedback) on day 1, (B) first feedback trial during the first day of training and (C) initial gait analysis (no feedback) on the third day. Gray curves depict mean knee flexion angles ± *SD* of non-impaired subjects. Orange lines show mean norm distance values.

motor capabilities, proprioception and range of motion are necessary and which degree of spasticity can be tolerated to still achieve a positive therapy effect? Our study participants had an almost unimpaired motor function (LEMS near 50), were able to walk for at least a few minutes and had some spared proprioception at least in the most affected joints. According to our study results, it may be concluded that real-time FB can be effectively used as a therapy in ambulating iSCI patients with predominantly sensory problems.

The effects of FB on gait rehabilitation have been rarely investigated in the iSCI population. Mainly robotic devices have been used (Banz et al., 2008; Schuck et al., 2012) for that purpose. Obvious advantages include prolonged therapy sessions and the potential to include non-ambulating individuals, but the fixed trajectories in the sagittal plane prevent an assessment of natural gait kinematics. With optical motion tracking patients' unrestricted walking pattern can be assessed. This provides patients the freedom of variability e.g., to experiment and solve the movement task in various ways.

On the other hand this freedom in task execution carries some inherent risks, because the movement task can be successfully fulfilled with different strategies. Although the reduction of ND indicates a positive change in the knee kinematics, it does not necessarily indicate a convergence to a more physiological walking pattern of all joints of the lower extremities. As the kinematics of only one joint was in the focus of the FB therapy, the rest of joints could have slinked away from their physiological movement patterns. Therefore, in future studies FB training should be implemented in such a way that the kinematics of the most affected joint should be normalized with the constraint, that kinematics of other ipsilateral and contralateral joints, are kept in a physiological range. Those studies should include proper retention tests directly after end of training and also several months later. FB displays should be more motivating, following a multimodal approach (Sigrist et al., 2013). In analogy to the assist-as-needed principle recently applied in locomotion robots (Emken et al., 2007), FB therapy should accommodate to the patients' progress and needs. This can be done by adjusting FB complexity and frequency and herewith find a compromise between challenging and supporting the patient.

Treadmill walking differs from walking over ground due to the constant velocity, the inability to walk curves, a different optical flow (Brennan et al., 2012) and hand rails for balance and/or body weight support. Additionally, spatial and temporal

gait parameters acquired on the treadmill may be transferred differentially to over ground walking (Yen et al., 2012). Therefore, it needs to be investigated, if improvements of our FB training on the treadmill can be transferred to over ground walking. Even though it has been shown that removing vision during treadmill adaptation could improve overground transfer of the new walking pattern (Torres-Oviedo and Bastian, 2010), the ultimate goal would be to train in everyday life situations. For this purpose, mobile gait analysis systems are promising tools that provide the possibility for measurement of joint angles and time-distance parameters. In an ongoing project called "RehaGait" (BMWi, German Federal Ministry of Economic Affairs and Energy, Project number KF2906702KJ2) the commercially available gait analysis system RehaWatch® (HASOMED GmbH, Magdeburg, Germany) based on inertial sensors is currently redesigned to provide FB for different gait affecting conditions (Parkinson's disease, stroke, cerebral palsy, iSCI). Moreover, a portable FB system has enormous spatial and economical advantages compared with a stationary motion-analysis based system, and can address a wide range of patients and institutions. FB in mobile systems can be provided through Augmented Reality glasses, auditory or haptic feedback. This introduces a dual-task in the training, which might degrade the time needed for adaptation, but enhances retention of the newly acquired skills (Torres-Oviedo et al., 2011).

Considering the interesting correlation of the capacity to respond to FB training and vibratory perception, reliable proprioception assessments are desired beyond the discrimination capacity of the tuning fork, which is frequently used in clinical practice as diagnostic tools for polyneuropathy in diabetes (Pourhamidi et al., 2014).

CONCLUSION

We show that instrumented real-time movement feedback based on kinematic variables is a promising technique to evoke short- and medium-term changes in individuals with incomplete spinal cord injury and prominent sensory deficits. It can be assumed that FB supported participants in consciously altering their otherwise unconsciously imprinted pathologic motor programs. Further studies including more patients and other gait pathologies are needed to reveal the underlying physiological mechanisms for this observation and to identify the most effective FB parameters, training strategies and characteristics of potential responders. Particularly, the role of proprioception in motor learning of individuals with iSCI needs to be further investigated.

ACKNOWLEDGMENTS

We would like to thank all participants of the study for their patience and motivation. We thank M. Lürssen for the creation of the three dimensional graphical model of our motion analysis laboratory (depicted in **Figure 1**). We also acknowledge financial support by Ruprecht-Karls-University Heidelberg within the funding program "Open Access Publishing."

REFERENCES

Anderson, R., Harrison, A., and Lyons, G. M. (2005). Accelerometry-based feedback–can it improve movement consistency and performance in rowing? *Sports Biomech.* 4, 179–195. doi: 10.1080/14763140508522862

Banz, R., Bolliger, M., Colombo, G., Dietz, V., and Lunenburger, L. (2008). Computerized visual feedback: an adjunct to robotic-assisted gait training. *Phys. Ther.* 88, 1135–1145. doi: 10.2522/ptj.20070203

Brennan, A. A., Bakdash, J. Z., and Proffitt, D. R. (2012). Treadmill experience mediates the perceptual-motor aftereffect of treadmill walking. *Exp. Brain Res.* 216, 527–534. doi: 10.1007/s00221-011-2956-9

Burns, S. P., Golding, D. G., Rolle, W. A. Jr., Graziani, V., and Ditunno, J. F. Jr. (1997). Recovery of ambulation in motor-incomplete tetraplegia. *Arch. Phys. Med. Rehabil.* 78, 1169–1172. doi: 10.1016/S0003-9993(97)90326-9

Colombo, G., Wirz, M., and Dietz, V. (2001). Driven gait orthosis for improvement of locomotor training in paraplegic patients. *Spinal Cord* 39, 252–255. doi: 10.1038/sj.sc.3101154

Dietz, V. (2002). Proprioception and locomotor disorders. *Nat. Rev. Neurosci.* 3, 781–790. doi: 10.1038/nrn939

Duschau-Wicke, A., Caprez, A., and Riener, R. (2010). Patient-cooperative control increases active participation of individuals with SCI during robot-aided gait training. *J. Neuroeng. Rehabil.* 7, 43. doi: 10.1186/1743-0003-7-43

Emken, J. L., Benitez, R., and Reinkensmeyer, D. J. (2007). Human-robot cooperative movement training: learning a novel sensory motor transformation during walking with robotic assistance-as-needed. *J. Neuroeng. Rehabil.* 4, 8. doi: 10.1186/1743-0003-4-8

Govil, K., and Noohu, M. M. (2013). Effect of EMG biofeedback training of gluteus maximus muscle on gait parameters in incomplete spinal cord injury. *NeuroRehabilitation* 33, 147–152. doi: 10.3233/NRE-130939

Kadaba, M. P., Ramakrishnan, H. K., and Wootten, M. E. (1990). Measurement of lower extremity kinematics during level walking. *J. Orthop. Res.* 8, 383–392. doi: 10.1002/jor.1100080310

Katoh, S., and el Masry, W. S. (1995). Motor recovery of patients presenting with motor paralysis and sensory sparing following cervical spinal cord injuries. *Paraplegia* 33, 506–509. doi: 10.1038/sc.1995.110

Keogh, J. W., and Hume, P. A. (2012). Evidence for biomechanics and motor learning research improving golf performance. *Sports Biomech.* 11, 288–309. doi: 10.1080/14763141.2012.671354

Kirshblum, S. C., Waring, W., Biering-Sorensen, F., Burns, S. P., Johansen, M., Schmidt-Read, M., et al. (2011). Reference for the 2011 revision of the International Standards for Neurological Classification of Spinal Cord Injury. *J. Spinal Cord Med.* 34, 547–554. doi: 10.1179/107902611X13186000420242

Krakauer, J. W. (2006). Motor learning: its relevance to stroke recovery and neurorehabilitation. *Curr. Opin. Neurol.* 19, 84–90. doi: 10.1097/01.wco.0000200544.29915.cc

Labruyere, R., Gerber, C. N., Birrer-Brutsch, K., Meyer-Heim, A., and van Hedel, H. J. (2013). Requirements for and impact of a serious game for neuropediatric robot-assisted gait training. *Res. Dev. Disabil.* 34, 3906–3915. doi: 10.1016/j.ridd.2013.07.031

Malone, L. A., and Bastian, A. J. (2010). Thinking about walking: effects of conscious correction versus distraction on locomotor adaptation. *J. Neurophysiol.* 103, 1954–1962. doi: 10.1152/jn.00832.2009

Marino, R. J., Scivoletto, G., Patrick, M., Tamburella, F., Read, M. S., Burns, A. S., et al. (2010). Walking index for spinal cord injury version 2 (WISCI-II) with repeatability of the 10-m walk time: inter- and intrarater reliabilities. *Am. J. Phys. Med. Rehabil.* 89, 7–15. doi: 10.1097/PHM.0b013e3181c560eb

Maslovat, D., Brunke, K. M., Chua, R., and Franks, I. M. (2009). Feedback effects on learning a novel bimanual coordination pattern: support for the guidance hypothesis. *J. Mot. Behav.* 41, 45–54. doi: 10.1080/00222895.2009.10125923

McDonald, J. W., and Sadowsky, C. (2002). Spinal-cord injury. *Lancet* 359, 417–425. doi: 10.1016/S0140-6736(02)07603-1

Meyer-Heim, A., and van Hedel, H. J. (2013). Robot-assisted and computer-enhanced therapies for children with cerebral palsy: current state and clinical implementation. *Semin. Pediatr. Neurol.* 20, 139–145. doi: 10.1016/j.spen.2013.06.006

Morton, S. M., and Bastian, A. J. (2006). Cerebellar contributions to locomotor adaptations during splitbelt treadmill walking. *J. Neurosci.* 26, 9107–9116. doi: 10.1523/JNEUROSCI.2622-06.2006

National Spinal Cord Injury Statistical Center. (2013). Spinal cord injury facts and figures at a glance. *J. Spinal Cord Med.* 36, 1–2. doi: 10.1179/1079026813Z.000000000136

Nielsen, J. B., and Sinkjaer, T. (2002). Afferent feedback in the control of human gait. *J. Electromyogr. Kinesiol.* 12, 213–217. doi: 10.1016/S1050-6411(02)00023-8

Park, J. H., Shea, C. H., and Wright, D. L. (2000). Reduced-frequency concurrent and terminal feedback: a test of the guidance hypothesis. *J. Mot. Behav.* 32, 287–296. doi: 10.1080/00222890009601379

Pourhamidi, K., Dahlin, L. B., Englund, E., and Rolandsson, O. (2014). Evaluation of clinical tools and their diagnostic use in distal symmetric polyneuropathy. *Prim. Care Diabetes* 8, 77–84. doi: 10.1016/j.pcd.2013.04.004

Proteau, L. (1992). "On the specificity of learning and the role of visual information for movement control," in *Vision and Motor Control*, Vol. 85, eds L. Proteau and D. Elliott (Oxford: North-Holland), 67–103. doi: 10.1016/S0166-4115(08)62011-7

R Core Team. (2012). *R: A Language and Environment for Statistical Computing*. Vienna: R Foundation for Statistical Computing.

Riemann, B. L., and Lephart, S. M. (2002). The sensorimotor system, Part II: the role of proprioception in motor control and functional joint stability. *J. Athl. Train.* 37, 80–84.

Rupp, R., Schablowski, M., and Gerner, H. J. (1998). [Development and evaluation of a diagnostic treadmill for three-dimensional imaging of gait dynamics]. *Biomed. Tech.* 43(Suppl.), 192–193. doi: 10.1515/bmte.1998.43.s1.192

Sanes, J. N., Mauritz, K. H., Dalakas, M. C., and Evarts, E. V. (1985). Motor control in humans with large-fiber sensory neuropathy. *Hum. Neurobiol.* 4, 101–114.

Schablowski-Trautmann, M. (2005). *Konzept zur Analyse der Lokomotion auf dem Laufband bei imkompletter Querschnittlähmung mit Verfahren der nichtlinearen Dynamik*. Karlsruhe: KIT Scientific Publishing, Universität Karlsruhe.

Schablowski-Trautmann, M., Kogel, M., Rupp, R., Mikut, R., and Gerner, H. J. (2006). From diagnostics to therapy–conceptual basis for real-time movement feedback in rehabilitation medicine. *Biomed. Tech.* 51, 299–304. doi: 10.1515/BMT.2006.061

Schmidt, R. A., and Lee, T. D. (2011). *Motor Control and Learning: A Behavioral Emphasis*, 5 Edn. Champaign, IL: Human Kinetics.

Schmidt, R. A., and Wrisberg, C. A. (2008). *Motor Learning and Performance, 3rd Edn.* Champaign, IL: Human Kinetics.

Schuck, A., Labruyere, R., Vallery, H., Riener, R., and Duschau-Wicke, A. (2012). Feasibility and effects of patient-cooperative robot-aided gait training applied in a 4-week pilot trial. *J. Neuroeng. Rehabil.* 9, 31. doi: 10.1186/1743-0003-9-31

Schuld, C., Wiese, J., Franz, S., Putz, C., Stierle, I., Smoor, I., et al. (2013). Effect of formal training in scaling, scoring and classification of the International Standards for Neurological Classification of Spinal Cord Injury. *Spinal Cord* 51, 282–288. doi: 10.1038/sc.2012.149

Schuld, C., Wiese, J., Hug, A., Putz, C., Hedel, H. J., Spiess, M. R., et al. (2012). Computer implementation of the international standards for neurological classification of spinal cord injury for consistent and efficient derivation of its subscores including handling of data from not testable segments. *J. Neurotrauma* 29, 453–461. doi: 10.1089/neu.2011.2085

Sigrist, R., Rauter, G., Riener, R., and Wolf, P. (2013). Augmented visual, auditory, haptic, and multimodal feedback in motor learning: a review. *Psychon. Bull. Rev.* 20, 21–53. doi: 10.3758/s13423-012-0333-8

Sigrist, R., Schellenberg, J., Rauter, G., Broggi, S., Riener, R., and Wolf, P. (2011). Visual and auditory augmented concurrent feedback in a complex motor task. *Presence* 20, 15–32. doi: 10.1162/pres_a_00032

Thompson, A. K., and Wolpaw, J. R. (2014). Operant conditioning of spinal reflexes: from basic science to clinical therapy. *Front. Integr. Neurosci.* 8:25. doi: 10.3389/fnint.2014.00025

Thow, J. L., Naemi, R., and Sanders, R. H. (2012). Comparison of modes of feedback on glide performance in swimming. *J. Sports Sci.* 30, 43–52. doi: 10.1080/02640414.2011.624537

Torres-Oviedo, G., and Bastian, A. J. (2010). Seeing is believing: effects of visual contextual cues on learning and transfer of locomotor adaptation. *J. Neurosci.* 30, 17015–17022. doi: 10.1523/JNEUROSCI.4205-10.2010

Torres-Oviedo, G., Vasudevan, E., Malone, L., and Bastian, A. J. (2011). Locomotor adaptation. *Prog. Brain Res.* 191, 65–74. doi: 10.1016/B978-0-444-53752-2.00013-8

Wolf, S., Loose, T., Schablowski, M., Doderlein, L., Rupp, R., Gerner, H. J., et al. (2006). Automated feature assessment in instrumented gait analysis. *Gait Posture* 23, 331–338. doi: 10.1016/j.gaitpost.2005.04.004

Wolpaw, J. R., and Carp, J. S. (1990). Memory traces in spinal cord. *Trends Neurosci.* 13, 137–142. doi: 10.1016/0166-2236(90)90005-U

Wulf, G., McConnel, N., Gartner, M., and Schwarz, A. (2002). Enhancing the learning of sport skills through external-focus feedback. *J. Mot. Behav.* 34, 171–182. doi: 10.1080/00222890209601939

Wulf, G., and Shea, C. H. (2002). Principles derived from the study of simple skills do not generalize to complex skill learning. *Psychon. Bull. Rev.* 9, 185–211. doi: 10.3758/BF03196276

Wulf, G., Shea, C. H., and Matschiner, S. (1998). Frequent feedback enhances complex motor skill learning. *J. Mot. Behav.* 30, 180–192. doi: 10.1080/00222899809601335

Yen, S. C., Schmit, B. D., Landry, J. M., Roth, H., and Wu, M. (2012). Locomotor adaptation to resistance during treadmill training transfers to overground walking in human SCI. *Exp. Brain Res.* 216, 473–482. doi: 10.1007/s00221-011-2950-2

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.

Received: 29 January 2014; accepted: 23 May 2014; published online: 12 June 2014.
*Citation: Schließmann D, Schuld C, Schneiders M, Derlien S, Glöckner M, Gladow T, Weidner N and Rupp R (2014) Feasibility of visual instrumented movement feedback therapy in individuals with motor incomplete spinal cord injury walking on a treadmill. Front. Hum. Neurosci. **8**:416. doi: 10.3389/fnhum.2014.00416*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

CLINICAL CASE STUDY ARTICLE
published: 13 May 2014
doi: 10.3389/fnhum.2014.00298

Hybrid gait training with an overground robot for people with incomplete spinal cord injury: a pilot study

Antonio J. del-Ama[1], Ángel Gil-Agudo[1], José L. Pons[2] and Juan C. Moreno[2]*

[1] *Biomechanics and Technical Aids Unit, National Hospital for Spinal Cord Injury, Toledo, Spain*
[2] *Bioengineering Group, Spanish National Research Council, Madrid, Spain*

Edited by:
Nadia Dominici, Swiss Federal Institute of Technology Lausanne, Switzerland

Reviewed by:
Yuri P. Ivanenko, Istituto di Ricovero e Cura a Carattere Scientifico Fondazione Santa Lucia, Italy
Bouri Mohamed, Swiss Federal Institute of Technology Lausanne, Switzerland

***Correspondence:**
Juan C. Moreno, Bioengineering Group, Spanish National Research Council, Carretera Campo Real 0,200, 28500 Madrid, Spain
e-mail: jc.moreno@csic.es

Locomotor training has proved to provide beneficial effect in terms of mobility in incomplete paraplegic patients. Neuroprosthetic technology can contribute to increase the efficacy of a training paradigm in the promotion of a locomotor pattern. Robotic exoskeletons can be used to manage the unavoidable loss of performance of artificially driven muscles. Hybrid exoskeletons blend complementary robotic and neuro-prosthetic technologies. The aim of this pilot study was to determine the effects of hybrid gait training in three case studies with persons with incomplete spinal cord injury (iSCI) in terms of locomotion performance during assisted gait, patient-robot adaptations, impact on ambulation and assessment of lower limb muscle strength and spasticity. Participants with iSCI received interventions with a hybrid bilateral exoskeleton for 4 days. Assessment of gait function revealed that patients improved the 6 min and 10 m walking tests after the intervention, and further improvements were observed 1 week after the intervention. Muscle examination revealed improvements in knee and hip sagittal muscle balance scores and decreased score in ankle extensor balance. It is concluded that improvements in biomechanical function of the knee joint after the tested overground hybrid gait trainer are coherent with improvements in gait performance.

Keywords: lower extremity, gait, spinal cord injury, hybrid exoskeleton, muscular electrical stimulation, motor recovery

INTRODUCTION

Locomotor training has proved to provide beneficial effect in terms of mobility in incomplete paraplegic patients. Improvement of locomotor activity occurs independently of the spontaneous recovery of the spinal cord function. With training functional movements under close physiological conditions, sensory inputs and central neuronal circuits become activated, with important spinal cord regeneration effects. Such training paradigm of stepping that relies on the entrainment of control of movement by driving the limbs through trajectories can be implemented with wearable actuated exoskeletons.

A further training approach is the use of EMS to activate paralyzed muscles for inducing gait. The importance of the EMS-induced gait training approach emerges from the demonstrated benefits it provides to the patient, mainly related to muscle strength and cardiorespiratory fitness (Creasey et al., 2004; Nightingale et al., 2007; Graupe et al., 2008; Lam et al., 2010). Besides, EMS can contribute to the stimulation of sensory input from the muscle that may be beneficial in case of damage. Nevertheless, it is not so effective yet in gait recovery (Thrasher et al., 2006), due to muscle fatigue, rapidly induced by EMS, leading to interruptions in training. The combination of a robotic exoskeleton with an EMS system results in a Hybrid Exoskeleton (del-Ama et al., 2012c). Hybrid exoskeletons posses several advantages for implementing novel alternatives of gait training. Firstly, hybrid exoskeletons takes advantage of the fine control of joint trajectories and the ability of delivering power that can compensate the poor quality of EMS-induced joint movement (Goldfarb and Durfee, 1996). On the other hand, muscle power generated by EMS can reduce the energy demand of the exoskeleton, thereby requiring less powerful joint actuators, which would result in a lighter system. Secondly, hybrid exoskeletons may promote more effective neural plasticity than other standard practices like treadmill training, because of the intensive, community-based gait practice involved. This gait practice occurs during daily training, and thus, increased user participation is promoted during walking training.

Designing a control strategy that adequately manages muscle fatigue is crucial to the development of a successful hybrid exoskeleton that can provide longer periods of training. Muscle fatigue caused by EMS is critical in cases of muscle atrophy, which are typically found in the SCI population. Effective closed-loop control of EMS has been proposed to manage muscle performance based on the kinetics of human–robot interaction (Stauffer

Abbreviations: 6mWT, 6 minutes walking test; 10mWT, 10 meters walking test; AAN, assist-as-needed; BP, blood pressure; CO, cooperative-only condition; EMS, electrical muscle stimulation; FSR, force sensing resistor; FTI, force time integral; HC, hybrid-stiff condition; HGO, hip guidance orthosis; HKAFO, hip knee ankle foot orthosis; HP, hybrid-cooperative condition; HR, heart rate; ILC, iterative learning controller; iSCI, incomplete spinal cord injury; KAFO, knee ankle foot orthosis; MFE, muscle fatigue estimator; MMT, manual muscle test; NP, neuroprosthetic; NTTI, normalized torque-time integral; PWM, pulse width modulation; QUEST, Quebec user evaluation of satisfaction with assistive technology; RMS, root mean square; VAS, visual analog scale; WISCI II, walking index for spinal cord injury version 2.

et al., 2009; del-Ama et al., 2012d). Besides, closed-loop control of joint movement would be required to counteract the effect of muscle fatigue (Goldfarb et al., 2003). An extensive review of control approaches and management of muscle fatigue of hybrid exoskeletons is reported in (del-Ama et al., 2012b).

While various wearable exoskeletons were successful in achieving gait in subjects with spinal cord injuries (Dollar and Herr, 2007, 2008; Hesse et al., 2010), this has generally been proposed as a functional substitution. An automatic treatment combining the AAN and cooperative-control principles, using state-of-the art hybrid technologies, could produce feasible systems in which the robot handles efficient delivery of EMS-induced torque. Besides, AAN control strategies of hybrid NP and robotic systems must work in parallel with the human system. These systems are likely to achieve sustained training sessions with EMS if incorporating techniques to control the appearance of muscle fatigue (del-Ama et al., 2012a).

Kinesis is a hybrid robotic device that has been developed for overground gait training in incomplete spinal cord injuries. The objective of this study is to evaluate the performance of Kinesis and the hybrid-cooperative walking therapy within the target population. In a previous study we conducted the evaluation of the effect of hybrid control of walking in a group of healthy subjects, demonstrating the feasibility of the ambulatory hybrid exoskeleton to effectively balance robotic and EMS during walking in intact humans (del-Ama et al., 2013). Based on that, we propose the current pilot study for validation of the hybrid-cooperative control approach. A comprehensive protocol was for evaluation of the impact on the walking function of the patient.

The design of this pilot clinical evaluation comprises pre–post assessment of the intervention effect in walking function of the patients. In particular, the design follows two objectives: (1) to assess the direct effects on walking of gait with Kinesis, focusing on control aspects and Kinesis-patient mutual adaptations; (2) to assess the impact of training with Kinesis on the ambulation of a sample of incomplete spinal cord injured subjects. We report the effects of hybrid gait training in three cases of persons with iSCI in terms of locomotion performance during assisted gait with and without EMS, patient-robot adaptations, impact on ambulation and assessment of lower limb muscle strength and spasticity.

MATERIALS AND METHODS

PARTICIPANTS

Participants with incomplete SCI ($n = 3$) were enrolled in the study to test the hybrid bilateral exoskeleton. The inclusion criteria considered patients whose lesion was categorized as conus medullaris, (injuries that affect the spinal levels among L1 and L2). The prognosis of functional recovery of walking is that these patients can walk short distances but depending on the wheelchair for community ambulation. Therefore, a successful overground hybrid walking therapy may provide benefits to this population. The functional characteristics of this lesion in relation to the walking function are: (a) preserved hip flexion ability, (b) partial ability to generate voluntary knee extension, (c) paralysis of ankle joint, and (d) presence of mild to severe spasticity.

Subject 1 is a male of 35 years old, 65 kg and 1.8 m height. He had a SCI L5 Asia impairment scale (AIS) D resulting in mild walking impairment. A secondary consequence of the lesion was impaired balance during left leg stance. Subject 2 is a male of 43 years old, 75 kg and 1.77 m height. He had a traumatic lesion, resulting in a SCI L4 AIS grade D. The patient had a passive limitation on the articular range of both knees, which caused to adapt the kinematic pattern of the left leg to meet these limits. The patient walked with fixed left knee joint during stance (at maximum extension) and a compensatory kinematic pattern for the right knee during the swing phase. Subject 3 is a male of 40 years old, 70 kg and 1.8 m height. He suffered an accident resulting in a SCI at L1 level, AIS A, with partial motor preservation at L3 level, and partial sensitive preservation at L4 level. This lesion represents the most impaired functional condition that met the inclusion criteria. The use of parallel bars was selected for the walking experimental conditions involving patient 3, given his functional status.

HYBRID GAIT TRAINER

The Kinesis system is a bilateral wearable knee-ankle-foot orthosis, equipped with active actuators at the knee hinges and a passive elastic actuator at the ankle. FSRs are employed for monitoring floor contact and custom force sensors are available to measure human–robot interaction torques. Kinesis has a PC-controlled stimulator that delivers biphasic current-controlled rectangular pulses to knee extensor (rectus femoris and vastus lateralis) and flexor (semitendinosus and biceps femoris) muscles through surface electrodes. The controller of Kinesis is comprised by the EMS controller and the robotic controller, both managed within a high-level controller is in charge of a cooperative behavior controlling exoskeleton actuators, EMS of knee joint muscles, and management of muscle fatigue. The EMS controller is comprised by a iterative learning control algorithm for knee flexor muscles, which is only active during swing phases of gait, and a PID controller for knee extensor muscles, which is active during all the gait phases. The control task of the dual EMS controller is to minimize the interaction leg-exoskeleton forces. The robotic joints features an admittance controller, which allows to regultate the assistance of the robotic joints during walking through modulation of the stiffness of a force field imposed aroung the knee joint trajectory. Such trajectory was extracted from a normative database available in our laboratory.

The goal of the AAN controller is to reduce robotic assistance during overground ambulatory walking. To this extent, the controller is comprised by a two-state finite machine. The first state (learning state) allows the ILC to "learn" the stimulation patterns for the flexor muscles while the leg are driven by the exoskeleton. Once the algorithm has converged, the AAN controller steps into the monitoring state, where the stimulation patterns obtained in the learning state are held constant, and the assistance of the exoskeleton is modulated (increased or decrased) regarding the muscle power obtained from the EMS. This is estimated through observing the maximum angle achieved during the swing phase. If the angle exceeds 60°, the robotic assistance can be reduced, and *vice versa*.

Further details on the implementation of Kinesis EMS-robot cooperative control can be found in (del-Ama et al., 2013).

SAFETY MEASURES

Safety measures implemented in the hybrid gait trainer included mechanical stops in the physiological limits of motion placed in ankle and knee robotic joints. In addition to this, the admittance controller of the knee joint was programmed with a software limit at maximum and minimum positions. In case of exceeding these limits, the state machine locked the motor shaft, and moved it to a default safe position. A third software safety measure consisted on the limitation of the maximum output torque required to the motor. An equivalent safe strategy was implemented in the EMS controller to set output limits for pulse width and amplitude modulation. Finally, a mechanical safety button was enabled to physically disconnect the energy supply of the entire hardware system. A risk analysis was conducted to verify the adequate actuation and response of the safety measures before actually moving to the experimental evaluation.

PROCEDURES

The established protocol was comprised by 2 weeks (**Figure 1**). During the first week (intervention week) several experiments related with hybrid walking training were conducted.

The first session consisted of a examination of response to EMS (S in **Figure 1**). The objectives for this session were to quantify the muscular response to the stimulation and also to get the patient used to the stimulation. Within this session, both flexor and extensor knee muscle groups of both legs were stimulated for 15 min. At the following session (T in **Figure 1**), the Kinesis device was introduced to the patients and learning exercises were carried out (**Figure 2**). The objective of this session was to train the patient to use the hybrid gait trainer and getting acquainted with the walking technique: bend to the side to lift the heel prior to initiate a step and then pressing the button. The Kinesis hybrid gait trainer was adjusted to the patient anthropometry within this session. The total walking time in this session did not exceed from 10 min. The remaining three sessions of the intervention were the actual hybrid walking experiments. As explained below, different configurations of the AAN controller were investigated in separate days (HC, CO, and HP in **Figure 1**, defined below).

During the second week (no intervention week) no intervention was administered. The objective of this design was to assess patient walking function before and after, and 1 week after the intervention with Kinesis (and associated experiments). For that purpose, assessment of gait function was performed prior-, post-, and 1 week after the hybrid walking intervention. The walking function assessment, denoted as EI, EII, and EIII in **Figure 1**, took place in separated days. The protocol was equal for all evaluation sessions, and was comprised by a MMT (Office, 1943) score of the sagittal plane, spasticity assessment by the Ashworth and PENN scales (Ashworth, 1964), and a 6mWT, where the time to cover the first 10 m was registered (10mWT). Patients used assistive devices for the walking conditions if regularly used to walk, and all conditions were assisted by a physiotherapist.

As introduced above, three configurations of the AAN controller have been included in the study (HC, CO, and HP in **Figure 1**). It is recalled here that Kinesis hybrid-cooperative controller is designed to modulate stimulation and robotic assistance during walking, which corresponds to the HC control configuration (hereinafter only configuration). In order to provide a better understanding of the performance of the hybrid system during overground gait training, two additional walking experimental conditions where designed (CO and HP in **Figure 1**), this is, to separately test the performance of the NP and the robotic components. The following codes are used to identify the walking experimental conditions:

- HC: hybrid-cooperative. During this condition EMS and robotic assistance are modulated during walking. The implementation of the controller used in this condition and its validation with healthy users presented elsewhere (del-Ama et al., 2013).
- CO: cooperative-only. During this condition the ILC was disabled, thus Kinesis can only bring adaptable robotic assistance to the patient during the swing phase stimulation of extensor muscles was not disabled to provide support during the stance phase.
- HP: hybrid-stiff. During this condition, the robotic assistance of Kinesis was held constant while the EMS controller operates similarly to HC configuration (learning and monitoring states enabled).

Each condition was tested in separated sessions to avoid fatigue-related effects. The conditions were tested under this sequence for each subject: HC-CO-HP. Measuring of BP and HR immediately prior and after the walking test were included for monitoring the physiological impact.

DATA ANALYSIS

In order to evaluate the direct effects of the intervention we analyzed the gait function and the biomechanical performance during the HC conditions (results during the CO and HP conditions are not presented in the current study). Assessment of gait function was performed gathering data from the 6mWT and 10mWT, MMT for the lower limbs and Ashworth spasticity scale. Results were group averaged and compared across assessment points. Mean

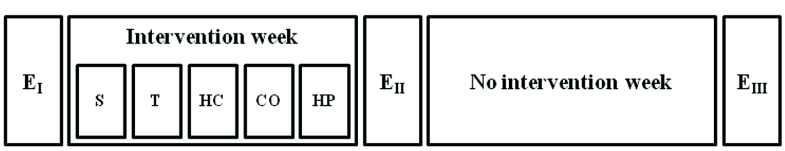


FIGURE 1 | Protocol for evaluation of hybrid gait training with Kinesis overground robot in iSCI. Examination sessions and walking conditions are identified as follows. Walking function evaluation session: E; examination of response to EMS: S; training session: T; hybrid-cooperative condition: HC; cooperative-only condition: CO; hybrid-stiff condition: HP.

FIGURE 2 | Experimental setup for data collection and testing hybrid gait training with Kinesis overground robot.

and standard deviation for, 10mWT and 6mWT were obtained. Kinesis performance was assessed in terms of actual knee angle, torque interaction, stimulator control output and torque field stiffness.

The normalized average stimulation output for knee extensor and flexor muscles (acronym NILC for flexor muscles) were calculated during the swing and stance phases respectively. This normalized average was calculated integrating the stimulator output during the walking phase (for swing and stance separately), and dividing the result by the maximum stimulation output theoretically achievable, which corresponds to a 450 μs saturated output for the entire walking phase. This normalized stimulation output gives a representative value ∈ [0, 1] where 0 means no stimulation during the entire phase, and 1 means a constant, saturated stimulation output of 450 μs for the entire walking phase.

A VAS was used to assess user fatigue, pain, and comfort. The VAS consists of a 10 centimeters rectangle that is extensively used to assess perceived pain in clinical settings (DeVine et al., 2011). With this scale, the user rates the pain perception placing a mark inside the rectangle, rating from no pain at all at the left edge of the rectangle, to intolerable pain at the right edge of the rectangle. Measuring the distance from the left edge to the mark gives a value ∈ [0, 10] that represents the user perceived pain, or fatigue and comfort in this protocol.

RESULTS

OVERGROUND ROBOTIC-GUIDED WALKING WITH NEURO-PROSTHETIC CONTROL (HC)

Overground walking performance was evaluated for the HC condition, with representative data from a single participant (Subject 1) with iSCI shown in **Figure 3**. Time-evolution of knee patterned and actual kinematics, interaction forces and the ILC stimulation output for the flexor muscles for both legs during walking with the HC, are represented. For this subject, knee kinematics of both legs were successfully controlled, and there were no dangerous situations reported during the experiment. Stimulation of left leg flexor muscles during swing (**Figures 3A,B**) was significantly lower than for the right leg (**Figures 3C,D**). A knee hyper-extension pattern at the right leg was obtained during the stance phase, maintained until prior swing phase (see actual knee joint angle, **Figures 3C,D**) and this hyper-extension was also evident in the measured interaction forces. On the contrary, extension (positive) interaction forces were observed during the swing phase of the right leg. Cycle-related performance during the HC experiment was analized for each subject, with data from participants with iSCI shown in **Figure 4**. The average normalized intensity of quadriceps EMS for the stance phases of the walking experiment were also analyzed for each subject, shown in **Figure 5**.

In Subject 1, differences in the functional status of both legs were observed (**Figures 4A,B**), coherent with the demand of EMS intensity and the stiffness of the exoskeleton. For the left leg, EMS intensity (represented in black) was lower when compared to the right leg muscles (maximum reached: 40% for the left, 97% for the right), and the first learning period required more steps. Interestingly, after the first muscle fatigue detection of the left leg (cycle 20), the second learning period led to a lower stimulation intensity, while the NTTI[1] further decreased during the second monitoring period (cycles 25–29). This decrease could not be attributed to an effect of EMS, because the pulse duration was significantly low to produce muscle contraction (10% of NILC). NTTI was increased in the last five cycles of the walking trial, due to the augmentation of muscle fatigue. There was a marked reduction in the interaction force for both legs, up to a 40% of the initial TTI. The EMS was considerably high for the right leg, above the 80% of the maximum achievable stimulation, for most of the trial. Decay on muscle performance was detected at cycle 10, probably due to the fatigue produced by the high intensity of EMS that resulted in the first iteration period (**Figure 4B**, cycles 1–5). The second iteration required a longer period (cycles 11–18), achieving a similar reduction in TTI. An increase on TTI was observed for the left leg for the last five cycles of the trial. The stiffness of the left knee joint was progressively reduced during the monitoring periods, while the stiffness of the right knee joint could not be reduced, although the stimulation was high and a reduction in NTTI was achieved.

In the case of Subject 2, the performance in the cycle domain during the HC condition is presented in **Figures 4C,D**. In particular, the obtained EMS patterns are summarized. NILC for both legs achieved the 70% of the maximum achievable stimulation intensity during the swing phase. After the first learning period of the EMS controlling the right leg, a decrease on NTTI is observed along

[1] Normalized time integral of the interaction torque during the swing phase.

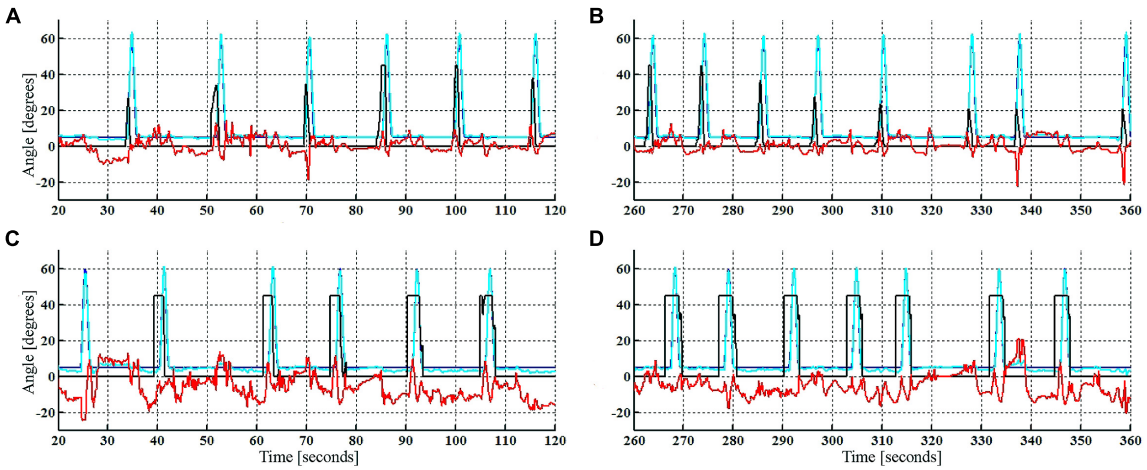


FIGURE 3 | Reference (blue) and actual (cyan) knee joint sagittal angle, interaction forces (red, [N·m/deg]), and ILC stimulation output (black, [μs]) during HC walking condition. First steps of left leg **(A)**, last steps of left leg **(B)**, first steps of right leg **(C)**, last steps of right leg **(D)**.

with an increase on joint stiffness. Muscle fatigue was detected in cycle 27 which activated a second monitoring state. A more moderate increase was required EMS intensity for the left leg during the first learning period, reaching similar equivalent right leg EMS intensity, and related to the observed decrease on NTTI. After the learning period, the stiffness was slightly reduced.

In the case of Subject 3, the performance in the cycle domain during HC walking condition is presented in **Figures 4E,F**. Left knee joint interaction forces were of low magnitude during the stance phase. The intensity of EMS was lower if compared to the right leg (85% of the maximum achievable intensity, black line) after the learning prior. The stiffness decreased with a minor decay during the monitoring period. Stimulation of right knee flexor muscles exhibited a saturated pattern from the first cycles the swing phase, as was observed in the cycle domain, where the NILC for the right leg (after the first learning period) reached approximately 100% of the maximum achievable EMS (**Figure 4F**). The NTTI shown a decrease (red line) after cycle 10. Stiffness increased during the monitoring period (magenta line), as expected.

Physiological effort data (**Figure 6** and **Table 2**) shows that the HC condition resulted in an increase in the systolic and diastolic BP after the experiment. Subjective perception of fatigue, pain and comfort were rated under the 50% of the scale for across all conditions (**Figure 6** and **Table 2**).

IMPACT ON WALKING FUNCTION

Evaluations of gait functionality were conducted prior, after, and 1 week after the intervention week (**Figure 1**). The results are presented in terms of relative changes on the outcome measures. These walking tests were conducted with diverse types of external support (patients 1 and 2 without external aids, patient 3 used a walker). **Table 1** shows the relative changes on the time needed to reach 10 m, and the distance covered in 6 min. The data revealed significant improvements after week I–II with less time required to complete 10 m and covered more distance in 6 min. One week after intervention, the patients further improved on gait function (II–III), but overall this differential improvement was smaller than post intervention I–II. The improvement of the intervention I–II was sustained and augmented at the final evaluation (I–III).

Muscle examination and grading strength (MMT) was performed after the interventions and revealed significant improvements on muscles controlling hip and knee joints. Data from the group of patients are provided in **Figure 7** and **Table 2** with MMT scores for hip, knee and ankle joints in the sagittal plane. In particular, results show that highest increments on hip MMT score were found for the I–II intervention week, sustained until the I–III intervention week (**Figure 7A** and **Table 2**). The increase on hip MMT can be explained to the effort required to move the extra weight of the exoskeleton, whilst no assistance was provided at hip joint level. A greater impact was observed on the muscle strength of flexor muscles than on extensor muscles. It can hypothesized that the effort to extend the hip be lower as result of the passive hip extension produced as consequence of contralateral leg flexion combined with trunk forward lean. Data also revealed increments in knee flexor and extensor muscle groups (intervention week I–II). In particular, higher improvement in knee extensor muscles was observed in session week II–III, and on the contrary, the flexor muscles shown a relative decrement on MMT for this intervention. When the knee was passively driven by the exoskeleton, no increments on knee muscular MMT would be obtained. For the knee flexor muscles, decreased MMT score was observed during session week II–III, which may be explained by a significantly lower demand during standard rehabilitation exercises in comparison with the hybrid walking condition. As a result, the increase on the flexor MMT score was not sustained in week II–III.

For the ankle flexor muscles (**Figure 7C**), increased MMT score was observed after I–II and II–III weeks, which was sustained after 1 week (week I–III). The increased scores were higher for the no-intervention week (II–III vs. I–II). Ankle extensor muscles revealed a decrease after the I–II intervention week, but

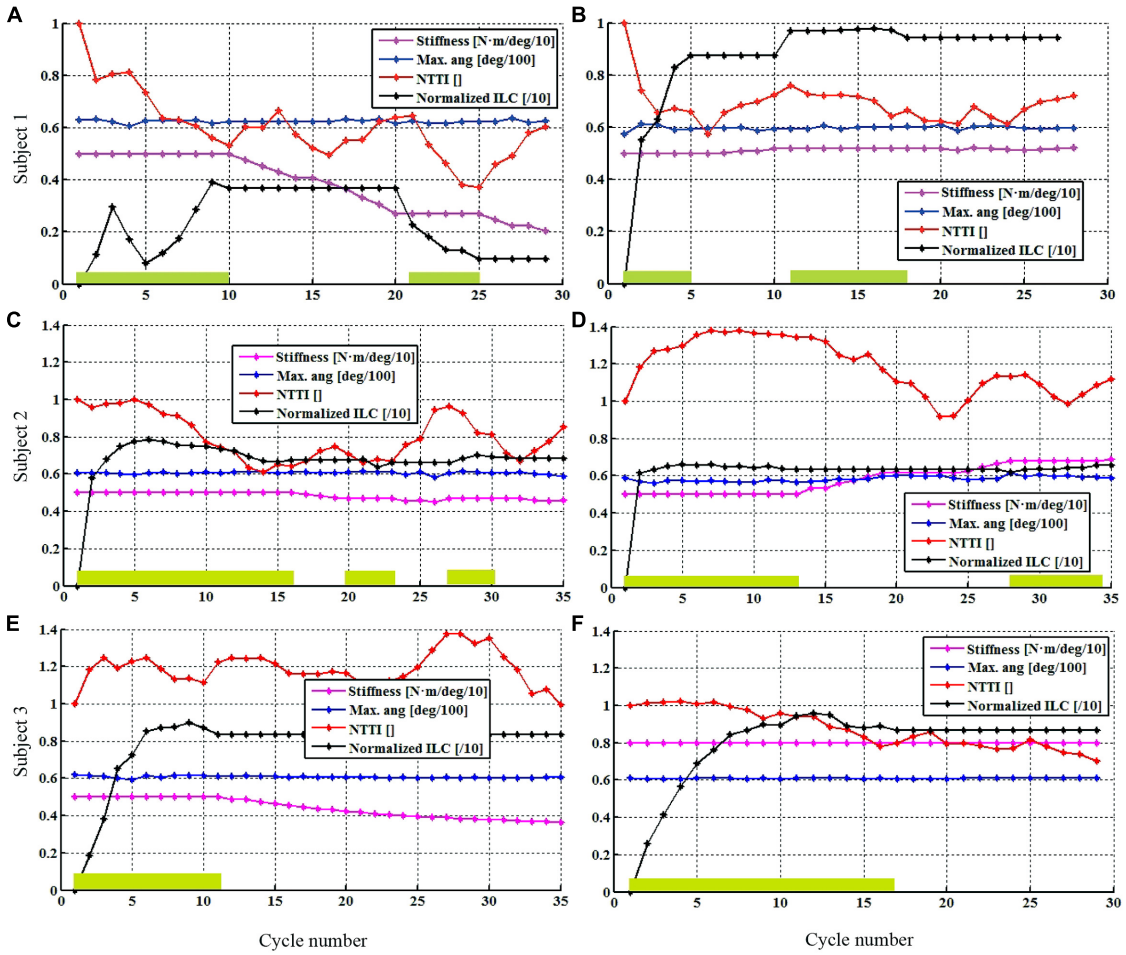


FIGURE 4 | Single-subject performance in the cycle domain during the HC condition. Controller stiffness (magenta line), NTTI (red), maximum angle achieved during flexion (blue line), normalized torque-time integral (red line), and NILC (black line) of left **(A,C,E)** and right **(B,D,F)** legs. Green boxes indicate when EMS learning state was active within the cycle. Controller stiffness, maximum angle and normalized stimulation curves are scaled for visualization purposes.

were increased after the II–III non-intervention week (**Table 2**). A hypothesis for this finding is that the spring-driven ankle actuator of the exoskeleton, which supported the foot during the swing phase, could have reduced the ankle extensor muscle ability. This phenomenon has been described previously in experiments with healthy subjects walking with a pneumatic KAFO-type exoskeleton (Kao et al., 2010). Nevertheless, other variables not related to the study may have influence on these results. Finally, a single-subject difference in articular knee joint range after the intervention session I–II with an increase on the knee joint range of movement in patient 2 (left knee ROM: 10° extension, 110° flexion; right knee: 5° extension, 120° flexion).

Analysis of spasticity measures after hybrid gait training revealed marked differences in the ASHWORTH and PENN scales for measuring spasticity (**Figure 8**) for all subjects. Average relative increments in Ashworth scale was -0.2 ± 0.4 and Penn spasm frequency scale was -0.4 ± 0.5 after the intervention week.

DISCUSSION

The purpose of this study was the assessment of the effects of the overground gait therapy by means of overground exoskeletons with neuro-prosthetic control. This analysis could be useful to provide the clinicians with objective data for interpretation of the appropriateness of this methodology as part of the treatment for the patient population. As such, evaluation of biomechanical and functional variables during and after hybrid gait training in these conditions provides insight into the response of iSCI patients to such treatment. Only few previous studies including NP technology and wearable exoskeletons provided preliminary insight into the potential outcome of such overground training paradigm in the promotion of a locomotor pattern (Esquenazi et al., 2012; Zeilig et al., 2012).

EFFECTS IN GAIT PERFORMANCE

Evaluation of the participants' response revealed that tested hybrid walking intervention as a therapy was tolerated by the patients;

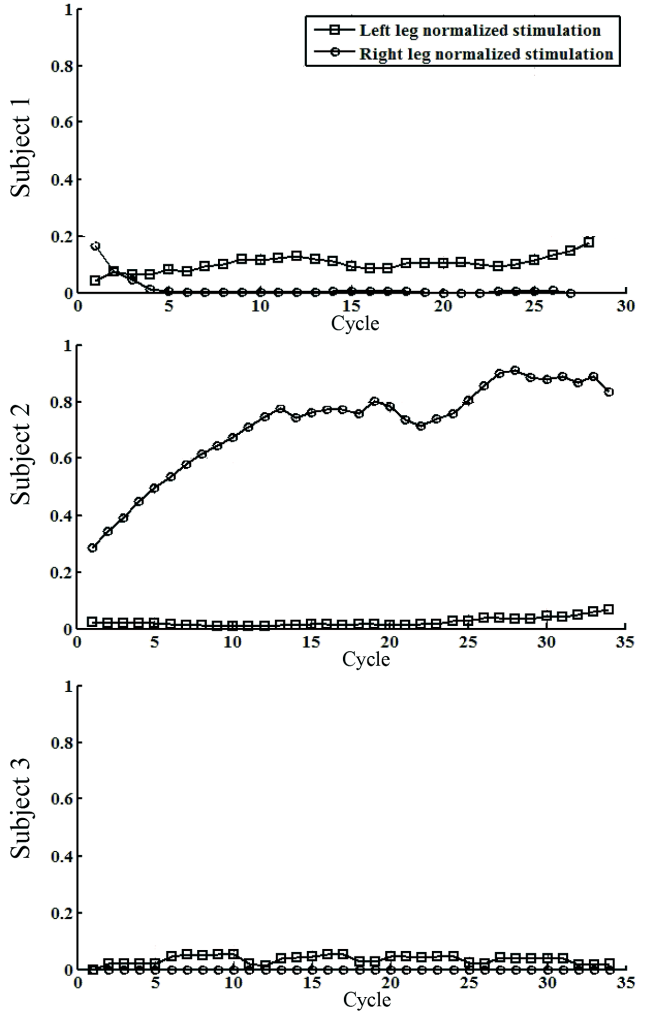


FIGURE 5 | Single-subject performance of quadriceps EMS during the walking experiment. The data represents the averaged stimulation for the knee extensor muscles during the stance phases of walking. Cycle means number of step.

no adverse effects were produced and the physical demand was tolerable. The participants were able to complete 6 min of walking with the system after 1 day of practice. The improvement in the 6mWT, the 10mWT, lower limb MMT and spasticity indexes demonstrate that walking function of the patients was improved after the intervention with the hybrid trainer. The hybrid control system adapted to the residual function of the participants during the task, modulating EMS and robotic assistance as needed. The observation of perceived pain, fatigue and comfort scores for the HC configuration revealed that training with Kinesis led to values on average were below 0.5. This provides an innovative indication of the positive tolerance of the participants to the proposed hybrid walking therapy. Our previous review of the literature did not trow explicit results of this evaluation but some specific claims on the lack of pain perception (Goldfarb et al., 2003), and improved feeling of safety have been reported (Popovic et al., 1989). In comparison with the reported results on perception of robotic-assisted walking (Esquenazi et al., 2012; Zeilig et al., 2012), our results are equivalent in terms of pain but with lower scores in relation to fatigue.

HUMAN–ROBOT INTERACTION DEMANDS FOR HYBRID GAIT TRAINING

From the perspective of assisted gait training, shortening the preliminary training period that is required to start a robot-aided treatment is important for the key components of the recovery after neurological damage, such as active engagement, motivation and patient-cooperative control. It is to be noted that the participants in this study were able to use the system as an overground robot-aided treatment after 1 day of practice (session T in **Figure 1**). This is significantly lower to requirements reported with the Rewalk, 25 days (average) needed by the patients to achieve autonomous use of Rewalk for an equivalent period of time (5–10 min; Esquenazi et al., 2012; Zeilig et al., 2012). This is important from a clinical perspective, although clear differences between Kinesis and Rewalk systems, the evaluation protocols and patient conditions should be considered when comparing requirements across devices. Rewalk controller demands patients to learn a strategy to trigger gait initiation and stopping by tilting the trunk. With this function, patients are required to learn a strategy to drive the Rewalk, maintaining balance with the help of walking crutches. With the Kinesis system used in the present study, the patient triggers each step when feeling table and ready to keep stepping, and also, the walker aid during the training condition provides a more stable support than crutches. These key differences can be reflected when comparing the results for 10mWT and 6mWT. On the one hand, Kinesis hybrid gait training led in average to double time to walk 10 m, and half distance covered in 6 min than Rewalk. On the other hand, Kinesis sequential manual trigger of stepping allowed the patients to efficiently drive the after a single day training session. This is important to bring the hybrid therapy early to the patient, avoiding long training periods as reported for gait neuroprosthetics (Thrasher et al., 2006). Human–robot interfaces to drive walking control with robotic exoskeletons for training require more research and development to more naturally operate the device without increasing the required training time an maintaining safety (Quintero et al., 2012).

ASSIST-AS-NEEDED HYBRID GAIT TRAINING

The extent of cooperative assistance provided by the hybrid robot is a key factor that may contribute to observed differences between treatment groups. Evaluations of the control-cooperative and AAN features of the hybrid gait trainer reveal, in our opinion, significant advances in the state-of-the art of control approaches for hybrid exoskeletons. First, the proposed hybrid controller allowed adapting the artificial muscular stimulation patterns to the neuromuscular status and performance of the patient. The hybrid controller showed to efficiently deliver stimulation as-needed to knee flexor and extensor muscles as a function of gait phase and voluntary contribution. Accordingly, customized stimulation patterns are delivered for each muscle of each leg to stably drive the knee joints. Across individuals, the dynamically obtained EMS patterns varied muscle activation from no

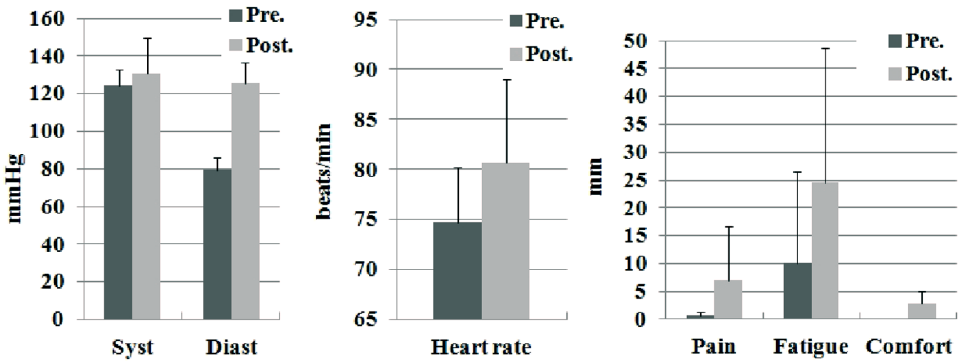


FIGURE 6 | Pre and post condition measures of physiological effort and subjective perceptions. The reported data are average and standard deviation for systolic BP, diastolic BP, heart rate, pain, fatigue, and comfort.

stimulation (e.g., Subject 1) to maximum stimulation (e.g., Subject 3) to specific functional requirements during stance or swing phases. Finally, these results supports close-loop muscle stimulation from a functional standpoint. However, the closed-loop control of muscle stimulation tested in the current study does not optimally manage the relationship between intensity of the EMS and force production. The results show that for a number of gait cycles EMS within the hybrid controller was not able to produce a significant muscular contraction for any normalized stimulation intensity below the 20%. Moreover, the opposite situation was also noticed in a number of trials, characterized by a saturated output of the EMS controller that could not produced a muscle contraction (e.g., Subject 3, leg knee flexor muscles). These effects constitute inherent limitations of the hybrid control system, which is not able to map the non-linear relationship between muscle stimulation and force production. Such feature in future controllers of hybrid gait exoskeletons would be important to provide a criteria for more efficient modulation of EMS regarding the range of muscle activation of targeted muscles.

Muscle fatigue estimation in hybrid exoskeletons has been addressed by off-line methods, such as the isometric recruitment curve (Goldfarb et al., 2003). The muscle performance monitor within the hybrid gait controller relies on leg-exoskeleton physical interaction as feedback for modulation control output. This scheme is expected to contribute to delays the appearance of muscle fatigue. However, the current results indicate that voluntary leg movements influence the physical interaction, with an effect on the accuracy of muscle fatigue estimation. Two mechanisms can explain the effects on the physical interaction. First, the varying volitional contribution to joint force provided by the subject. In this case, patient slacking would lead to false detection of muscle fatigue. Second, the method for online normalization of the FTI values that could lead to low accuracy in FTI estimation. This suggests that more robust methods to uncouple patient contribution to the movement from EMS performance and robot assistance are needed to improve muscle fatigue management and therefore optimize patient volitional contribution.

The impact of the hybrid walking therapy on patient gait abilities was evaluated. Greater improvements in gait function, sagittal MMT score, and spasticity, for the intervention week I–II was concluded with the observation week II–III. There are some limitations to this study. First, the sample size was low, and with heterogeneous lesions, therefore we recommend caution when generalizing the study results. Second, we tested walking conditions to reveal effects of the hybrid approach in a non-randomized testing order of configurations due to practical considerations. As a result, this may led to uncontrolled learning

Table 1 | Pre–post condition results of 10mWT an 6mWT tests during the experimental conditions.

	I–II	II–III	I–III
10mWT [sec]	−13.6 ± 28.2	−16.2 ± 33.5	−29.8 ± 61.7
6mWT [m]	44.2 ± 59.3	17.8 ± 21.4	62.0 ± 79.6

The reported data are average and standard deviation.

Table 2 | Summary of evaluation results: 10mWT and 6mWT tests and subjective scales.

	HC
10mWT [sec]	3.3 ± 1.6
6mWT [m]	17.0 ± 20.2
Systolic BP [mmHg]	6.7 ± 20.8
Diastolic BP [mmHg]	7.3 ± 15.0
Hear rate [beat/min]	6.0 ± 14.6
Pain [cm]	6.3 ± 17.3
Comfort [cm]	2.9 ± 2.1
Fatigue [cm]	14.7 ± 50.5

Data provided for fatigue, pain, BP and HR are pre–post relative increment. The reported data are average and standard deviation.

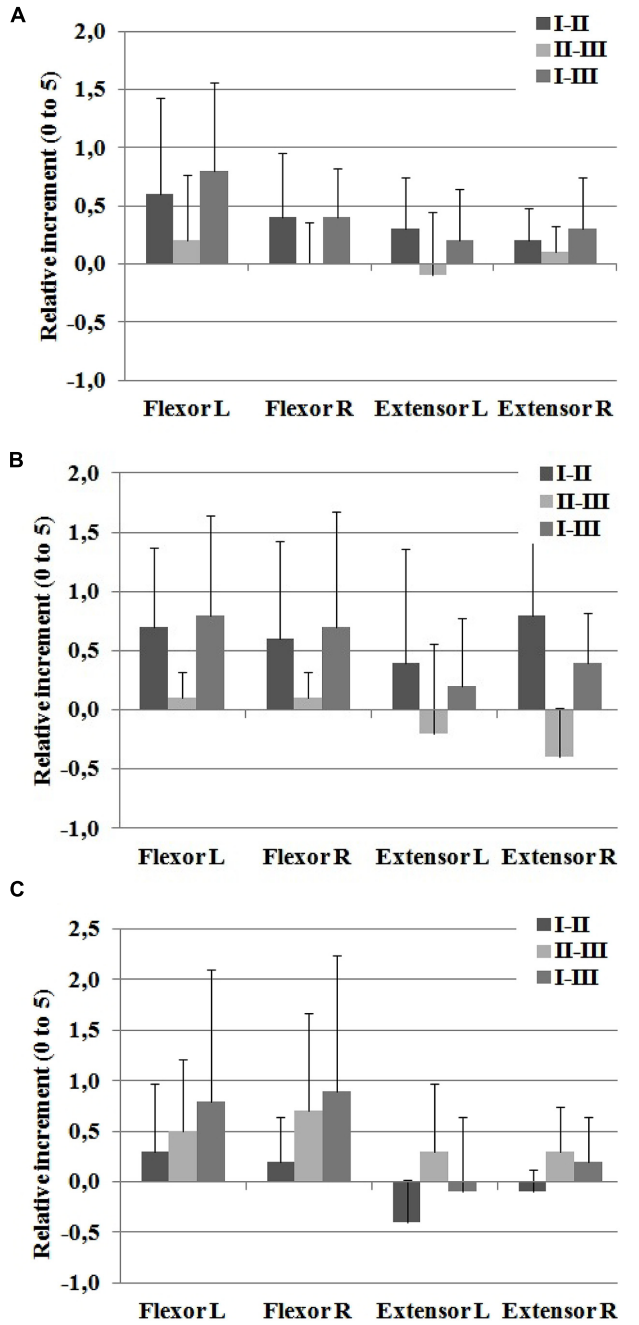


FIGURE 7 | Sagittal manual muscle testing (MMT) score for (A) hip, (B) knee, and (C) ankle joints. The reported data are average and standard deviation of the relative changes between the examination sessions I–II, II–III, and I–III.

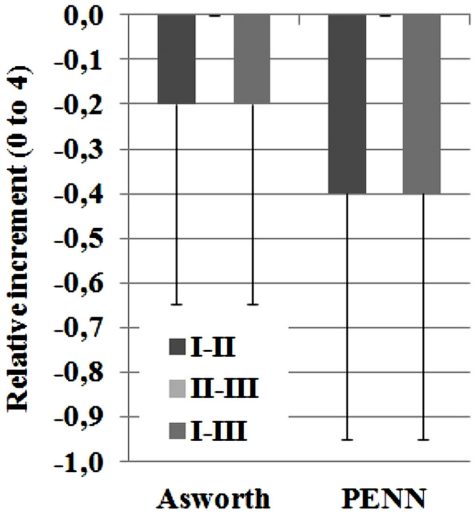


FIGURE 8 | Results for spasticity ASHWORTH and PENN scales. Score can range from 0 to 4. The reported data are median and range.

effects. A third limitation is that we did not test the effect of intensity and long-term retention of hybrid gait training. However, these results encourage the design of further studies that would allow knowing whether iSCI can retain the functional gains over longer time periods as a function of treatment dose.

In summary, participants tolerated the hybrid gait intervention delivered by Kinesis. The patients were able to complete 6 min of walking with the system after 1 day of practice. Mutual adaptations were observed between the patient and the system that were assessed through the analysis of the physical interaction. The hybrid-cooperative control was able to compensate bilateral pathologic walking patterns by autonomously increasing the stimulation of the knee joint muscles and increasing the displayed stiffness of the robotic actuators. The improvement in the 6mWT, the 10mWT, lower limb MMT, and spasticity indexes demonstrate this training paradigm, termed hybrid gait training, to improve short-term locomotor performance that is consistent with promoted recovery of neuromuscular control for gait. The results of the current pilot study provide proof-of-concept for the feasibility of combining neuroprosthesis and actuated exoskeletons as tools for robotic training of gait function in motor incomplete SCI patients, guaranteeing and motivating further research in the field. Further work is required to optimize several open questions of hybrid gait control, such as assessment of volitional muscle contribution during EMS and optimization of muscle fatigue detection and quantification. Furthermore, testing the hybrid strategies on multiple joints will be interesting to cope with requirements of wider patient populations. Also, research is required to elucidate the changes in neuromuscular activity of targeted lower limb muscles during and after the intervention and to elucidate the mechanisms that may explain a recovery of neural control. The reduced number of subjects studied and the small amount of training time (6 min for the walking experiments T, HC, CO, and HP) limits extrapolating the results of the hybrid walking therapy to the incomplete SCI population. Besides, the heterogeneity of the functional status of the patients can be also regarded as a limitation in the study design. While this is true for elucidating the effects of the hybrid training, the results showed the ability of Kinesis system and AAN controller to adapt to such different functional conditions. In conclusion, future clinical trials are required to establish the long-term therapeutic benefits of overground hybrid training in restoring gait function

in a wide and more representative population of incomplete SCI subjects.

ACKNOWLEDGMENT

This project is funded by the Spanish Consolider-Ingenio Programme, project "HYPER" (contract number CSD2009-00067).

REFERENCES

Ashworth, B. (1964). Preliminary trial of carisoprodol in multiple sclerosis. *Practitioner* 192, 540–542.

Creasey, G. H., Ho, C. H., Triolo, R. J., Gater, D. R., DiMarco, A. F., Bogie, K. M., et al. (2004). Clinical applications of electrical stimulation after spinal cord injury. *J. Spinal Cord Med.* 27, 365–375.

del-Ama, A. J., Bravo-esteban, E., Moreno, J. C., Gómez-soriano, J., Koutsou, A. D., Gil-agudo, Á., et al. (2012a). "Knee muscle fatigue estimation during isometric artificially elicited contractions in incomplete spinal cord injured subjects," in *Proceedings of the 2012 International Conference on Neurorehabilitation (ICNR2012): Converging Clinical and Engineering Research on Neurorehabilitation* (Berlin: Springer), 329–333.

del-Ama, A. J., Koutsou, A. D., and Moreno, J. C. (2012b). Review of hybrid exoskeletons to restore gait following spinal cord injury. *J. Rehabil. Res. Dev.* 49, 497–514. doi: 10.1682/JRRD.2011.03.0043

del-Ama, A. J., Koutsou, A. D., Moreno, J. C., De-los-Reyes, A., Gil-Agudo, A., and Pons, J. L. (2012c). Review of hybrid exoskeletons to restore gait following spinal cord injury. *J. Rehabil. Res. Dev.* 49, 497–514. doi: 10.1682/JRRD.2011.03.0043

del-Ama, A. J., Moreno, J. C., Gil-Agudo, A., De-los-Reyes, A., and Pons, J. L. (2012d). Online assessment of human-robot interaction for hybrid control of walking. *Sensors* 12, 215–225. doi: 10.3390/s120100215

del-Ama, A. J., Gil-agudo, Á., Pons, J. L., and Moreno, J. C. (2013). Hybrid FES-robot cooperative control of ambulatory gait rehabilitation exoskeleton. *J. Neuroeng. Rehabil.* 11, 27. doi: 10.1186/1743-0003-11-27

DeVine, J., Norvell, D. C., Ecker, E., and Fourney, D. R. (2011). Evaluating the correlation and responsiveness of patient-reported pain with function and quality-of-life outcomes after spine surgery. *Spine (Phila Pa 1976).* 36, S69–S74. doi: 10.1097/BRS.0b013e31822ef6de

Dollar, A. M., and Herr, H. (2007). Active orthoses for the lower-limbs: challenges and state of the art. *IEEE Int. Conf. Rehabil. Robot.* 1, 968–977.

Dollar, A. M., and Herr, H. (2008). Lower extremity exoskeletons and active orthoses: challenges and state-of-the-art. *IEEE Trans. Robot.* 24, 144–158. doi: 10.1109/TRO.2008.915453

Esquenazi, A., Talaty, M., Packel, A., and Saulino, M. (2012). The ReWalk powered exoskeleton to restore ambulatory function to individuals with thoracic-level motor-complete spinal cord injury. *Am. J. Phys. Med. Rehabil.* 91, 911–921. doi: 10.1097/PHM.0b013e318269d9a3

Goldfarb, M., and Durfee, W. K. (1996). Design of a controlled-brake orthosis for FES-aided gait. *IEEE Trans. Rehabil. Eng.* 4, 13–24. doi: 10.1109/86.486053

Goldfarb, M., Korkowski, K., Harrold, B., and Durfee, W. (2003). Preliminary evaluation of a controlled-brake orthosis for FES-aided gait. *IEEE Trans. Neural Syst. Rehabil. Eng.* 11, 241–248. doi: 10.1109/TNSRE.2003.816873

Graupe, D., Cerrel-Bazo, H., Kern, H., and Carraro, U. (2008). Walking performance, medical outcomes and patient training in FES of innervated muscles for ambulation by thoracic-level complete paraplegics. *Neurol. Res.* 30, 123–130. doi: 10.1179/174313208X281136

Hesse, S., Waldner, A., and Tomelleri, C. (2010). Innovative gait robot for the repetitive practice of floor walking and stair climbing up and down in stroke patients. *J. Neuroeng. Rehabil.* 7:30. doi: 10.1186/1743-0003-7-30

Kao, P.-C., Lewis, C. L., and Ferris, D. P. (2010). Short-term locomotor adaptation to a robotic ankle exoskeleton does not alter soleus Hoffmann reflex amplitude. *J. Neuroeng. Rehabil.* 7:33. doi: 10.1186/1743-0003-7-33

Lam, T., Wolfe, D., Eng, J. J., and Domingo, A. (2010). *Lower Limb Rehabilitation Following Spinal Cord Injury*, Version 4, Vol. 3. Vancouver, BC: Spinal Cord Injury Rehabilitation Evidence.

Nightingale, E. J., Raymond, J., Middleton, J. W., Crosbie, J., and Davis, G. M. (2007). Benefits of FES gait in a spinal cord injured population. *Spinal Cord* 45, 646–657. doi: 10.1038/sj.sc.3102101

Office, H. M. S. (ed.). (1943). *Aids to the Investigation of Peripheral Nerve Injuries.* London: Medical Research Council (Great Britain), Nerve injuries Committee.

Popovic, D., Tomovic, R., and Schwirtlich, L. (1989). Hybrid assistive system-the motor neuroprosthesis. *IEEE Trans. Biomed. Eng.* 36, 729–737. doi: 10.1109/10.32105

Quintero, H. A., Farris, R. J., Ha, K., and Goldfarb, M. (2012). Preliminary assessment of the efficacy of supplementing knee extension capability in a lower limb exoskeleton with FES. *Conf. IEEE Eng. Med. Biol. Soc.* 2012, 3360–3363. doi: 10.1109/EMBC.2012.6346685

Stauffer, Y., Allemand, Y., Bouri, M., Fournier, J., Clavel, R., Metrailler, P., et al. (2009). The WalkTrainer – a new generation of walking reeducation device combining orthoses and muscle stimulation. *IEEE Trans. Neural Syst. Rehabil. Eng.* 17, 38–45. doi: 10.1109/TNSRE.2008.2008288

Thrasher, T. A., Flett, H. M., and Popovic, M. R. (2006). Gait training regimen for incomplete spinal cord injury using functional electrical stimulation. *Spinal Cord* 44, 357–361. doi: 10.1038/sj.sc.3101864

Zeilig, G., Weingarden, H., Zwecker, M., Dudkiewicz, I., Bloch, A., and Esquenazi, A. (2012). Safety and tolerance of the ReWalk™ exoskeleton suit for ambulation by people with complete spinal cord injury: a pilot study. *J. Spinal Cord Med.* 35, 96–101. doi: 10.1179/2045772312Y.0000000003

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.

Received: 15 December 2013; accepted: 23 April 2014; published online: 13 May 2014.
Citation: del-Ama AJ, Gil-Agudo Á, Pons JL and Moreno JC (2014) Hybrid gait training with an overground robot for people with incomplete spinal cord injury: a pilot study. Front. Hum. Neurosci. ***8****:298. doi: 10.3389/fnhum.2014.00298*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 16 June 2014
doi: 10.3389/fnhum.2014.00423

EMG patterns during assisted walking in the exoskeleton

Francesca Sylos-Labini[1,2]*, Valentina La Scaleia[1,2], Andrea d'Avella[1], Iolanda Pisotta[3], Federica Tamburella[3], Giorgio Scivoletto[3], Marco Molinari[3], Shiqian Wang[4], Letian Wang[5], Edwin van Asseldonk[5], Herman van der Kooij[4,5], Thomas Hoellinger[6], Guy Cheron[6], Freygardur Thorsteinsson[7], Michel Ilzkovitz[8], Jeremi Gancet[8], Ralf Hauffe[9], Frank Zanov[9], Francesco Lacquaniti[1,2,10] and Yuri P. Ivanenko[1]

[1] *Laboratory of Neuromotor Physiology, Santa Lucia Foundation, Rome, Italy*
[2] *Centre of Space Bio-medicine, University of Rome Tor Vergata, Rome, Italy*
[3] *Spinal Cord Rehab Unit and CaRMA Lab, Santa Lucia Foundation, Rome, Italy*
[4] *Biomechanical Engineering, Delft University of Technology, Delft, Netherlands*
[5] *Biomechanical Engineering, University of Twente, Enschede, Netherlands*
[6] *Laboratory of Neurophysiology and Movement Biomechanics, Université Libre de Bruxelles, Brussels, Belgium*
[7] *OSSUR, Reykjavík, Iceland*
[8] *Space Applications Services N.V./S.A., Zaventem, Belgium*
[9] *ANT Neuro, Berlin, Germany*
[10] *Department of Systems Medicine, University of Rome Tor Vergata, Rome, Italy*

Edited by:
Marco Iosa, Fondazione Santa Lucia, Italy

Reviewed by:
Juan C. Moreno, Spanish National Research Council, Spain
Stefano Masiero, University of Padua, Italy

****Correspondence:***
Francesca Sylos-Labini, Laboratory of Neuromotor Physiology, IRCCS Fondazione Santa Lucia, 306 via Ardeatina, 00179 Rome, Italy
e-mail: f.syloslabini@hsantalucia.it

Neuroprosthetic technology and robotic exoskeletons are being developed to facilitate stepping, reduce muscle efforts, and promote motor recovery. Nevertheless, the guidance forces of an exoskeleton may influence the sensory inputs, sensorimotor interactions and resulting muscle activity patterns during stepping. The aim of this study was to report the muscle activation patterns in a sample of intact and injured subjects while walking with a robotic exoskeleton and, in particular, to quantify the level of muscle activity during assisted gait. We recorded electromyographic (EMG) activity of different leg and arm muscles during overground walking in an exoskeleton in six healthy individuals and four spinal cord injury (SCI) participants. In SCI patients, EMG activity of the upper limb muscles was augmented while activation of leg muscles was typically small. Contrary to our expectations, however, in neurologically intact subjects, EMG activity of leg muscles was similar or even larger during exoskeleton-assisted walking compared to normal overground walking. In addition, significant variations in the EMG waveforms were found across different walking conditions. The most variable pattern was observed in the hamstring muscles. Overall, the results are consistent with a non-linear reorganization of the locomotor output when using the robotic stepping devices. The findings may contribute to our understanding of human-machine interactions and adaptation of locomotor activity patterns.

Keywords: robotic exoskeleton, assisted gait, EMG patterns, spinal cord injury, neuroprosthetic technology

INTRODUCTION

Exoskeleton robotic devices are now often used in the rehabilitation practice to assist physical therapy of individuals with neurological disorders (Sale et al., 2012; Moreno et al., 2013). To provide patients with some degree of locomotion capability, passive (unpowered) orthoses are often prescribed (Hsu et al., 2008). However, passive devices have many limitations, including the high energy expenditure and low utilization by individuals with severe walking impairments (Wang et al., submitted). Active (powered) exoskeletons and new control implementations are extensively developed in recent years to provide new possibilities for severely paralyzed patients to walk (Fitzsimmons et al., 2009; Swinnen et al., 2010; Cheron et al., 2012; del-Ama et al., 2012; Roy et al., 2012; Sale et al., 2012; Wang et al., submitted). Many of these devices include some form of body weight support and adjustable levels of robotic guidance forces.

Investigating locomotor responses in individuals after neurological lesions, as well as in healthy subjects, when using the robotic devices, is fundamental to the development of improved rehabilitation strategies and to explore the mechanisms involved in improving locomotor function (Ivanenko et al., 2013). Even in neurologically intact subjects, the use of external devices for stepping can affect motor patterns (Hidler and Wall, 2005; Lam et al., 2008; Van Asseldonk et al., 2008; Moreno et al., 2013), modify the "locomotor body scheme" and result in distortions in the body and space representation (Ivanenko et al., 2011). There is still a lack of knowledge on the effect of robotic gait assistance on the locomotor function and its recovery in injured humans due to the complex nature of the control of locomotion, compensatory strategies, and plasticity of neuronal networks.

Several studies emphasized the importance of minimizing passive guidance and stabilization provided during gait rehabilitation

(Israel et al., 2006), establishing baseline patterns (Hidler and Wall, 2005) and reduction of metabolic cost of ambulant exoskeletons (Malcolm et al., 2013). Different artificial control schemes can induce different locomotor patterns. Here we used a control strategy of the exoskeleton consisting in weight shift to the stance side to trigger a step and to provide predefined reference joint trajectories (Wang et al., 2013). This exoskeleton assisted both posture (knee stabilization during stance, weight shift, lateral stabilization) and leg movements. The main purpose of this study was to report the muscle activation patterns in a sample of intact and injured subjects while walking with a robotic exoskeleton and, in particular, to quantify the level of muscle activity during assisted gait. It can be argued that robotic-guided walking should reduce leg muscle activity in healthy subjects to a lower level and might affect the motor output in patients as well. To verify this hypothesis, we investigated the adaptation of muscle activation patterns in neurologically intact human adults and spinal cord injury (SCI) patients using a recently developed exoskeleton (called MINDWALKER, https://www.mindwalker-project.eu).

METHODS

PARTICIPANTS

Six healthy volunteers (age range between 21 and 36 years, five males and one female, mean height 1.72 ± 0.09 m [mean ±*SD* (standard deviation)], weight 69 ± 12 kg) participated in this study. We also tested the MINDWALKER exoskeleton on SCI subjects (**Table 1**). Patient inclusion criteria were the following: age 18–45 years, traumatic/non-traumatic SCI, at least 5 month after injury with stable neurological score, complete lesion (AIS A, B at the time of inclusion) from below T7, inability to ambulate over ground without at least moderate assistance, Mini-Mental State Examination score >26. Exclusion criteria were: presence of transmissible diseases, such as (but not limited to) hepatitis, human immunodeficiency virus or Creutzfeldt-Jacob disease, symptomatic orthostatic hypotension or 30-mmHg drop when upright, subjects with spine-stabilizing devices for whom their treating surgeon contraindicates gait, contraindications for lower extremities weight bearing (pelvic or leg fracture, chronic joint pain), untreatable chronic pain, untreatable spasticity (Ashworth scale score >3), severe reduction in lower limb joint's range of motion, pressure sore stage 2 or higher, skin injuries or problems such as blisters, burns, wounds from operation, or other superficial wounds at the scalp, debilitating disease prior to SCI that causes exercise intolerance and limits mobility-related self-care and instrumental activities of daily living, premorbid major depression or psychosis, suicide attempt caused the SCI, unlikely to complete the intervention or return for follow-up, participation in another research. The studies conformed to the Declaration of Helsinki, and informed consent was obtained from all participants according to the procedures of the Ethics Committee of the Santa Lucia Foundation.

BRIEF DESCRIPTION OF MINDWALKER EXOSKELETON

The detailed description of the exoskeleton and its control is provided elsewhere (Wang et al., 2013; Wang et al., submitted). Briefly, this exoskeleton is aimed at providing a research prototype that can empower lower limb disabled people (especially SCI patients) to walk on level ground (**Figure 1A**). Based on human anatomy and joint range of motion (RoM), the desired degrees of freedom (five at each leg) and joint RoM for the exoskeleton are specified to allow sitting, standing, and walking. In each leg, three degrees of freedom (DoFs: hip ab/adduction, hip flexion/extension and knee flexion/extension) are powered by series elastic actuators and two DoFs (hip endo/exo rotation and ankle dorsi/plantar flexion), are passively sprung with certain stiffness (800 and 180 Nm/rad, respectively). The exoskeleton weighs 28 kg excluding batteries and it bears its own weight by transferring the weight via its footplates to the ground. The exoskeleton can be attached to the wearer at five main locations: footplate, shank, thigh, pelvis, and torso (**Figure 1A**).

A finite-state machine based controller was implemented for providing gait assistance in both sagittal and frontal planes and the swing phase initiation was triggered using trunk motion (Wang et al., 2013). For example, leaning to the left and forward triggers a right step: when the estimated center of mass (CoM) falls into a predefined region, the controller detected the intention of the subject and initiates assisted weight shift to left. Then the state transits automatically to right swing. Weight shift is initiated by the subject and completed by the exoskeleton. This control strategy is relatively simple, as well as it takes advantage of natural lateral trunk oscillations that always accompany normal walking (Cappellini et al., 2010).

In this study, two control modes of the exoskeleton were used, namely, "EXO assisted" and "EXO-unassisted." In the EXO-unassisted mode, healthy subjects wore the exoskeleton with all motorized joints in torque control mode, in which the references were 0 torque. In this mode, the exoskeleton joints were moved by the human. As the controller bandwidth is limited (Wang et al., 2013) the actual exerted torques by the exoskeleton will not be zero and therefore we quantified the actual torques

Table 1 | Subject characteristics.

Patient	Age, year	Gender	Weight, kg	Height, m	Lesion level	ASIA	Aethiology	Lesion time, months
p1	19	M	50	1.80	T12-L1	B	Trauma	5
p2	21	M	67	1.78	T7	A	Trauma	26
p3	22	M	70	1.80	T11-T12	A	Trauma	36
p4	43	M	78	1.74	T9-T10	A	Trauma	49

Lesion level indicates the clinical neurological level, lesion time the time interval between lesion diagnosis and data recording.

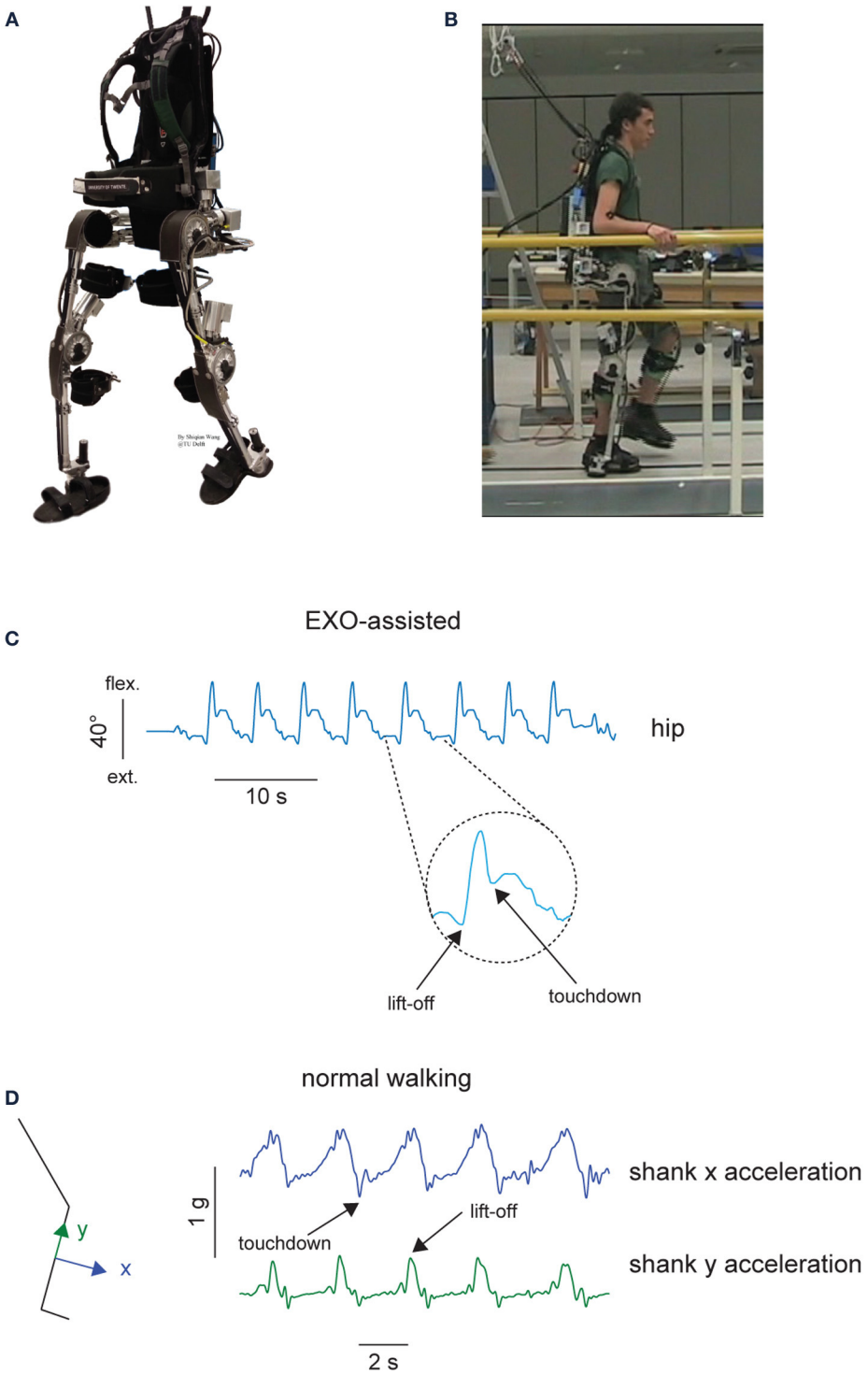


FIGURE 1 | Experimental setup. (A) MINDWALKER exoskeleton. Each leg has five degrees of freedom. Shank and thigh segments have telescopic tubular structure to accommodate different subject statues. The exoskeleton is attached to the wearer at five main locations: footplate, shank, thigh, pelvis, and torso. Footplates are made of carbon fiber and have braces to host human feet. Shank braces are used to support most of the weight of the user in standing and walking while thigh braces are added to loosely constrain the upper leg and support the wearer during standing up. Pelvis and backpack braces are used to attach the upper body to the wearer. **(B)** A healthy subject during walking in the exoskeleton. **(C)** Definition of touchdown and lift-off events from the hip joint angle during walking in the exoskeleton. **(D)** Definition of touchdown and lift-off events from the shank inertial sensor accelerations during normal walking.

measured by exoskeleton sensors (see Results and **Figure 2B**). In the EXO-assisted mode, all exoskeleton joints were following predefined joint angles provided with variable joint impedances, the walking trajectories during the swing phase (reference joint angles) were defined based on walking patterns of a healthy subject walking in the MINDWALKER exoskeleton in the EXO-unassisted mode (Wang et al., 2013). Hip and knee flexion angles were slightly increased during swing to ensure sufficient foot clearance. Since the reference trajectories for the swing phases were predefined, the swing phase durations were similar across conditions.

EXPERIMENT DESCRIPTION

Four experimental conditions in healthy individuals were recorded in the same experimental session: EXO-assisted, EXO-unassisted, NM slow, NM self-selected. "NM slow" referred to normal slow walking without the exoskeleton and "NM self-selected" normal walking at self-selected speed without the

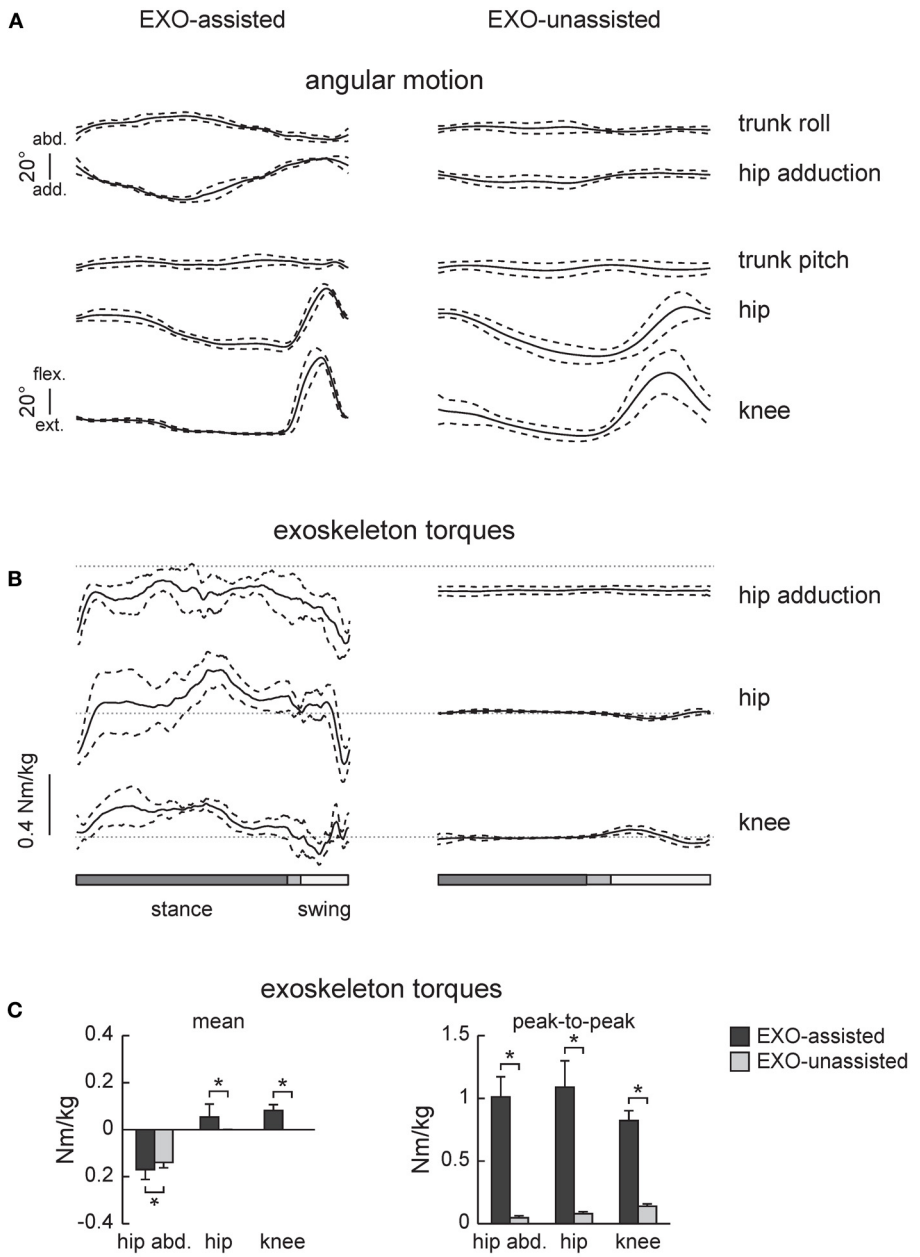


FIGURE 2 | Joint angles and exoskeleton torques recorded in healthy subjects during walking in the exoskeleton (EXO-assisted and EXO-unassisted). (A) Ensemble-averaged (mean ±*SD*, $n = 6$) joint angular movements. **(B)** Ensemble-averaged joint torques recorded in three powered actuators of the exoskeleton (knee and hip flexion/extension and hip ab/adduction). Note little torques in the hip and knee joints in the zero-torque mode (right panel) due to the absence of assistance. **(C)** Mean torques and peak-to-peak oscillations of torques. Asterisks denote significant differences.

exoskeleton. In the first two conditions, the participants were asked to walk along a 8-m walkway and were allowed to place the abducted arms on horizontal handrails located at the side of the walkway (**Figure 1B**), to provide stability/assistance if needed. A safety harness worn by the wearer was attached to an overhead suspension system moving along with the wearer, which only came into action when the subject fell. Typically, we collected the data from 2–4 trials while walking in the exoskeleton following a short period of training (1–2 trials). On average, 8–15 strides were recorded and analyzed in each experimental condition. The total duration of the experimental session was about 1–2 h. In the EXO-assisted condition, the subjects were instructed to move their CoM forward and toward a side to trigger a contralateral step (for example leaning to the left and forward would trigger a right step). In the EXO-unassisted condition, the subjects were told to just walk at their preferred pace bearing the exoskeleton to reach the end of the walkway.

In the latter two conditions, healthy subjects walked without the exoskeleton along a 8-m walkway at slow and self-selected speeds. Gait initiation and gait termination strides were excluded from the analysis. About 10 strides were analyzed in each subject in each condition. The high speeds of normal walking were not recorded because walking in the exoskeleton was rather slow (see Results).

One experimental condition in SCI participants was recorded, that is the EXO-assisted condition. A similar protocol as used in healthy individuals (control) was employed, for comparison. In participants with complete lesions, familiarization with MINDWALKER usage was more difficult and required several days of exoskeleton training (2 or 3 times/week) for a total of session ranging between 5 and 8. SCI participants achieved the control of balance holding the handrails located at the side of the walkway. All subjects presented high motivation since the first trial and throughout testing and the comparison between the first and last sessions for the whole group of patients present only minimal changes in the mean walking speed. No clinical changes were observed, between first and last trials, in the clinical scales (the detailed description of behavioral assessment and the physiological cost index are provided elsewhere, Pisotta et al., submitted), indicating that MINDWALKER usage does not affect the functional neurological status, consistent with a limited effectiveness of robot-assisted gait training in severely paralyzed individuals (Swinnen et al., 2010; Roy et al., 2012; Sale et al., 2012). Here we analyzed the stepping pattern in the last session, after familiarization with MINDWALKER usage.

DATA RECORDING, PROCESSING, AND GAIT EVENT DETECTION

In the exoskeleton walking conditions (EXO-assisted and EXO-unassisted), joint angles and torques at aforementioned powered DoFs were recorded by the MINDWALKER exoskeleton at 1000 Hz and downsampled at 100 Hz to be used with the muscle activity recordings (Wang et al., 2013). Gait cycle events (touchdown and lift-off) were defined based on the kinematic data of the hip flexion-extension angle: touchdown as the first local minimum following the maximum and lift-off as the first local minimum preceding the maximum (**Figure 1C**). These kinematic criteria were verified by comparison with the events detected by inertial signals from the sensor placed on the TA muscle using a similar method as during normal walking (Jasiewicz et al., 2006). In general, the difference between the time events measured from kinematics (**Figure 1C**) and inertial sensors was less than 4%. We divided the recorded kinematic and kinetic data into gait cycles (touchdown as the beginning of the gait cycle), then interpolated each stride to 200 time points, and finally averaged across gait cycles (individually for each subject). Joint torques were normalized to the total body weight (subject + exoskeleton) prior to averaging across subjects. It is worth noting that these are not the net joint torques of the subjects but the resulting torques exerted by the exoskeleton to move the subject's limbs and whole body.

Electromyographic (EMG) activity was recorded by means of surface electrodes from 11 muscles simultaneously on the right side of each subject. These included vastus medialis (VM), rectus femoris (RF), biceps femoris long head (BF), semitendinosus (ST), tibialis anterior (TA), medial gastrocnemius (MG), and soleus (Sol), anterior deltoid (DELTa), posterior deltoid (DELTp), flexor carpi ulnaris (FCU), extensor carpi ulnaris (ECU). We placed EMG electrodes based on suggestions from SENIAM (seniam.org), the European project on surface EMG, and by palpating to locate the muscle bellies and orienting the electrodes along the main direction of the fibers (Winter, 1991; Kendall et al., 2005). All EMGs were recorded at 2000 Hz using a Delsys Trigno Wireless System (Boston, MA).

The EMG sensors of the Delsys Trigno Wireless System also contained 3D accelerometers, and we recorded and filtered (5 Hz low-pass zero-lag 4th order Butterworth) these inertial signals from the sensor placed on the TA muscle in order to define the gait cycle during walking without the exoskeleton (NM slow and NM self-selected walking): based on the method of Jasiewicz et al. (2006), touchdown was identified by minima in the shank x acceleration while lift-off was identified by maxima in the shank y acceleration (**Figure 1D**).

EMG DATA ANALYSIS

EMG data were processed using standard filtering and rectifying methods. We applied a 60 Hz high-pass filter, then rectified the EMG signals and applied a 10 Hz low-pass filter (all filters, zero-lag 4th order Butterworth). EMG data were time-interpolated over a time base with 200 points for individual gait cycles ($i = 1 \div 200$) and averaged.

In addition to computing the ensemble-averaged EMG waveforms (Winter, 1991; Perry, 1992), we calculated for each muscle and each subject the mean and maximum EMG activity and the center-of-activity (CoA) throughout the gait cycle. The CoA during the gait cycle was calculated using circular statistics ("circ_mean.m" function in the CircStat Matlab toolbox, Berens, 2009) and plotted in polar coordinates (polar direction denoted the phase of the gait cycle—with angle θ that varies from 0 to 360° corresponding to 0 and 100% cycle, respectively—and radius denoted the mean EMG activity of the muscle). The CoA of the EMG waveform was calculated as the angle of the vector (first trigonometric moment) which points to the CoM of that circular distribution using the following formulas:

$$A = \sum_{t=1}^{200} (\cos\theta_t \times EMG_t) \quad (1)$$

$$B = \sum_{t=1}^{200} (\sin\theta_t \times EMG_t) \quad (2)$$

$$CoA = \tan^{-1}(B/A) \quad (3)$$

The CoA has been used previously to characterize the overall temporal shifts of EMG or motoneuron activity (Yakovenko et al., 2002; Ivanenko et al., 2006; Sylos-Labini et al., 2011) and was chosen because it was impractical to reliably identify a single peak of activity in the majority of muscles. It can be helpful to understand if the distribution of muscular activity remains unaltered across different conditions.

STATISTICS

Descriptive statistics included means ± standard deviation (SD) of the mean. The mean torque and peak-to-peak torque amplitudes were computed and compared across conditions. A repeated measure (RM) ANOVA was used to evaluate the effect of condition (on all parameters except for CoA) in healthy individuals. *Post-hoc* tests and multiple comparisons analysis were performed by means of the Bonferroni test. Circular statistics on directional data (Batschelet, 1981) were used to characterize the mean CoA for each muscle (see preceding text) and its variability across strides (angular SD). The Watson-William test was used for circular data (CoA) to evaluate the effect of condition in healthy individuals. Unpaired t-test was used to test differences in the exoskeleton torques and mean EMGs between controls and SCI patients. Statistics on Pearson's correlation coefficients was performed on the normally distributed, Z-transformed values. Reported results are considered significant for $p < 0.05$.

RESULTS

Results were presented and briefly discussed in the following manner to make clear comparison: first, comparisons on kinematic and kinetic data were made between EXO-assisted and EXO-unassisted walking conditions in healthy individuals; second, for the same two walking conditions, EMG data in lower limbs were presented; third, EMG data in EXO-assisted and NM slow walking were given; and finally, for the EXO-assisted walking condition, comparisons were made between healthy and SCI subjects based on the kinematic/kinetic and EMG measurements.

EXO-ASSISTED vs. EXO-UNASSISTED: KINEMATICS AND KINETICS IN HEALTHY SUBJECTS

In general, as shown in **Figure 3A**, EXO-unassisted walking was faster than EXO-assisted walking (mean cycle duration was 3.2 ± 0.8 s vs. 6.1 ± 0.8 s). The swing phase durations were similar and the difference were caused by the dead time in stance, since in EXO-assisted walking, the subjects needed to move their trunk and trigger the swing step by step.

Figures 2A,B illustrates ensemble-averaged angular movements and joint torques in healthy subjects in these two walking conditions. The amplitude of the knee and hip joint angular movements in the sagittal plane were similar (**Figure 2A**), however, hip abduction was larger during assisted walking since lateral trunk movements were necessary to trigger the swing phase.

EXO-assisted walking in the exoskeleton requires relatively large torques in the hip and knee joints (**Figure 2B**). For instance, peak-to-peak amplitudes in the knee and hip joints (normalized to the wearer-EXO's weight) were about 1 Nm/kg (**Figure 2C**). Nevertheless, the torques that the exoskeleton applied to the subject (in the sagittal plane) were compatible to those exerted by subjects during normal overground walking (Winter, 1991). Note though that these torques were exerted by the exoskeleton (in order to move the subject's limbs and body) rather than by the subjects themselves. As expected, during unassisted walking these torques were very small (**Figure 2B**, right panel), which was dictated by the closed-loop torque control performance of the exoskeleton.

EXO-ASSISTED vs. EXO-UNASSISTED: EMG PATTERNS IN HEALTHY SUBJECTS

Figure 3B illustrates ensemble-averaged EMG patterns of leg muscles in control subjects in different walking conditions. During EXO-assisted walking the exoskeleton provided all necessary torques to support the body and move the legs forward while during unassisted walking the subjects moved their and exoskeleton's legs together. Accordingly, it was not surprising that during unassisted walking the amplitude of EMG activity was typically larger than that during assisted walking (**Figure 3B**, left two panels). Specifically, the mean activity was significantly larger for the RF, VM, TA, MG, and Sol muscles while it was comparable for BF and smaller for ST (**Figure 4A**). It is also worth noting that EMG waveforms differed for BF and ST: in particular, there was no activity in these muscles at the beginning of the stance phase during not-assisted walking (**Figure 3B**). The correlation analysis confirmed similarities in the EMG waveforms for RF, VM, MG, and Sol muscles and differences for BF, ST, and TA muscles between these two conditions (**Table 2**).

EXO-ASSISTED vs. NM SLOW WALKING: EMG PATTERNS IN HEALTHY SUBJECTS

During normal walking, muscle activity typically increases with increasing walking speed (Ivanenko et al., 2006). Therefore, comparisons of normal and pathological gait are typically performed at similar walking speeds. Assisted walking in the exoskeleton (EXO assisted) was relatively slow compared to normal walking (**Figure 3A**). The swing phase duration was also longer for the EXO assisted condition (**Figure 3A**).

Since the movements of the limbs were performed by the exoskeleton, one would expect substantially lower muscle activity during assisted walking. Interestingly, contrary to our expectations, assisted walking in the exoskeleton was not accompanied by reduced EMG activity. In fact, the activity of most muscles (RF, VM, TA, MG, Sol) did not change significantly, while the activity of BF and ST even increased during assisted walking despite the slower walking speed in this condition (**Figure 4A**).

EMG waveforms of some leg muscles also differed between assisted gait and normal walking (**Figure 3B**). For instance, RF and VM activity contained additional bursts during the swing

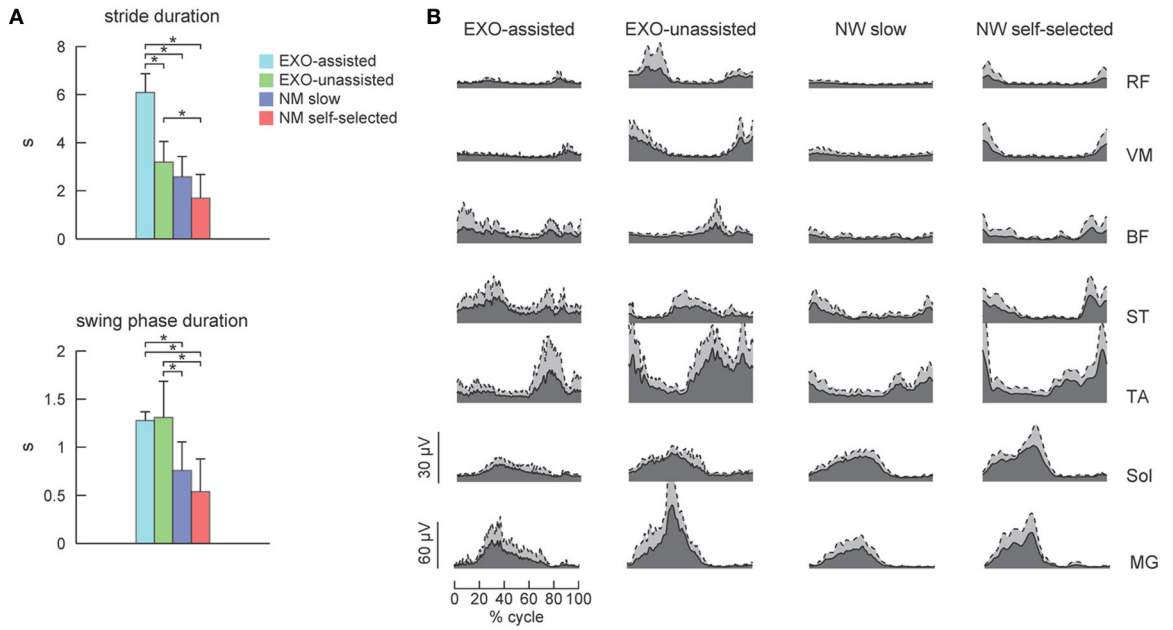


FIGURE 3 | EMG patterns in healthy subjects during walking in the exoskeleton and during normal overground walking. (A) Stride and swing durations (mean +*SD*, $n = 6$) for each experimental condition. **(B)** Time course of ensemble-averaged EMG patterns (dark area, gray area corresponds to *SD*). Asterisks denote significant differences across conditions.

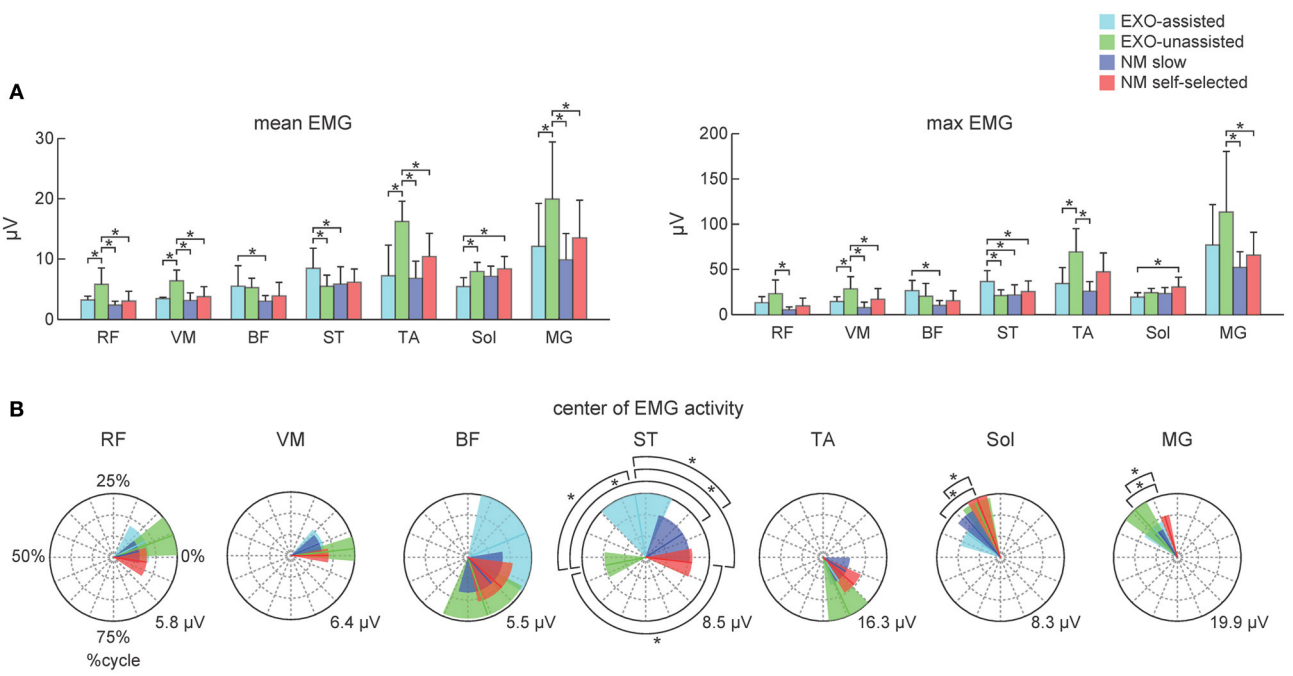


FIGURE 4 | Characteristics of EMG activity during assisted and normal walking in control subjects. (A) Mean and maximum EMG activities (left and right panels, respectively) for each muscle (mean +*SD*). **(B)** Polar plots of the center of EMG activity. Polar direction denotes the relative time over the gait cycle (time progresses clockwise), radius of the vector denotes the mean EMG activity of the muscle and the width of the sector denotes angular *SD* (across subjects). Polar grid with circles was also shown to ease comparisons (the number in the right corner of each plot corresponds to the value of the external circle). Asterisks denote significant differences across conditions.

Table 2 | Pearson correlation coefficients (mean ±*SD*, *n* = 6) between EMG waveforms for different conditions in control subjects.

Muscle	Condition	EXO-assisted	EXO-unassisted	NW slow	NW self-selected
RF	EXO-assisted	–	0.32 ± 0.29*	0.10 ± 0.21	0.07 ± 0.10
	EXO-unassisted	0.32 ± 0.29*	–	0.58 ± 0.52*	0.35 ± 0.27*
	NW slow	0.10 ± 0.21	0.58 ± 0.52*	–	0.32 ± 0.13*
	NW self-selected	0.07 ± 0.10	0.35 ± 0.27*	0.32 ± 0.13*	–
VM	EXO-assisted	–	0.70 ± 0.35*	0.41 ± 0.26*	0.29 ± 0.13*
	EXO-unassisted	0.70 ± 0.35*	–	0.61 ± 0.28*	0.56 ± 0.20*
	NW slow	0.41 ± 0.26*	0.61 ± 0.28*	–	0.50 ± 0.24*
	NW self-selected	0.29 ± 0.13*	0.56 ± 0.20*	0.50 ± 0.24*	–
BF	EXO-assisted	–	0.01 ± 0.34	−0.11 ± 0.29	0.25 ± 0.41
	EXO-unassisted	0.01 ± 0.34	–	0.26 ± 0.34	0.47 ± 0.51
	NW slow	−0.11 ± 0.29	0.26 ± 0.34	–	0.29 ± 0.09*
	NW self-selected	0.25 ± 0.41	0.47 ± 0.51	0.29 ± 0.09*	–
ST	EXO-assisted	–	−0.07 ± 0.33	0.20 ± 0.36	0.17 ± 0.17*
	EXO-unassisted	−0.07 ± 0.33	–	−0.24 ± 0.13*	−0.34 ± 0.10*
	NW slow	0.20 ± 0.36	−0.24 ± 0.13*	–	0.38 ± 0.13*
	NW self-selected	0.17 ± 0.17*	−0.34 ± 0.10*	0.38 ± 0.13*	–
TA	EXO-assisted	–	0.35 ± 0.49	0.14 ± 0.21	0.22 ± 0.07*
	EXO-unassisted	0.35 ± 0.49	–	0.33 ± 0.28*	0.26 ± 0.30
	NW slow	0.14 ± 0.21	0.33 ± 0.28*	–	0.72 ± 0.21*
	NW self-selected	0.22 ± 0.07*	0.26 ± 0.30	0.72 ± 0.21*	–
Sol	EXO-assisted	–	0.65 ± 0.34*	0.60 ± 0.30*	0.73 ± 0.34*
	EXO-unassisted	0.65 ± 0.34*	–	0.74 ± 0.14*	0.78 ± 0.25*
	NW slow	0.60 ± 0.30*	0.74 ± 0.14*	–	0.74 ± 0.14*
	NW self-selected	0.73 ± 0.34*	0.78 ± 0.25*	0.74 ± 0.14*	–
MG	EXO-assisted	–	0.67 ± 0.37*	0.65 ± 0.25*	0.74 ± 0.37*
	EXO-unassisted	0.67 ± 0.37*	–	0.72 ± 0.24*	0.87 ± 0.38*
	NW slow	0.65 ± 0.25*	0.72 ± 0.24*	–	0.79 ± 0.25*
	NW self-selected	0.74 ± 0.37*	0.87 ± 0.38*	0.79 ± 0.25*	–

Asterisks denote correlation coefficients significantly different from zero, t-test.

phase (**Figure 3B**), BF and ST muscles were activated in mid-stance and early swing during assisted walking (**Figure 3B**) and the center of activity of the ST muscle differed significantly between normal and assisted walking (**Figure 4B**). The correlation analysis showed low correlations for most muscles: only VM, Sol, and MG muscles demonstrated significant correlations between these two conditions (**Table 2**).

SCI vs. HEALTHY SUBJECTS IN EXO-ASSISTED CONDITION: KINEMATICS, KINETICS, AND EMG PATTERNS

SCI patients walked slightly slower than the control subjects (**Figure 5A**, on average, the cycle duration was 6.1 ± 0.8 s in control subjects and 7.6 ± 1.1 s in SCI patients), though the swing phase duration (**Figure 5A** right panel) and the angular movements (**Figure 5B** left panel) were similar. The amplitude (peak-to-peak) of the exoskeleton torques was also similar (only the knee torque was larger, **Figure 5C**), suggesting that the exoskeleton provided the main forces for stepping in both control and SCI subjects.

Despite similarities in the kinematics and dynamics of movements, EMG patterns differed in SCI patients. Overall, they used more upper limb muscles for stepping (DELTp and ECU **Figure 5E**) though there was also variability in using the arms muscles between subjects (compare, for instance, the control and the SCI subject in **Figure 5D**). EMG activity in the lower limb muscles was typically minute if any in SCI patients, though one SCI patient demonstrated consistent activity in the BF, ST, RF, and MG muscles during the swing phase and beginning of stance (**Figure 5D** right panel).

DISCUSSION

We investigated the effect of walking with an exoskeleton on the muscular activation patterns in healthy subjects and SCI patients. Strikingly, despite exoskeleton assistance in both posture and leg movements, the overall muscle activity level in healthy subjects was not reduced at all, as one would expect, further supporting the importance of sensory input and suitability of using robotic exoskeletons for entraining lumbosacral locomotor circuitry. The

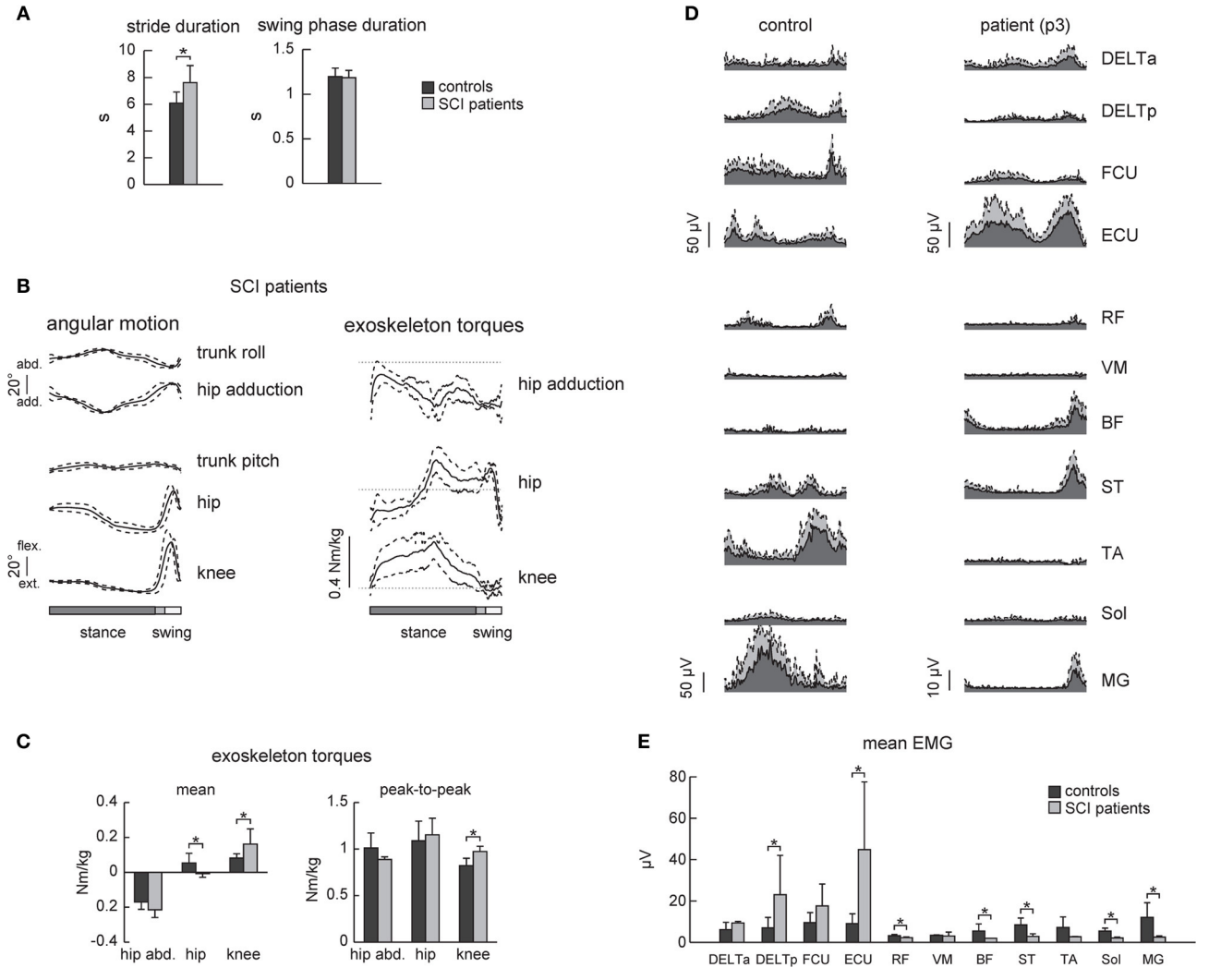


FIGURE 5 | EMG activity in healthy subjects and SCI patients during walking in the exoskeleton ("EXO-assisted" mode). (A) Joint angles and exoskeleton torques recorded in SCI patients during walking in the exoskeleton. The same format as in **Figure 2**. **(B)** Mean torques and peak-to-peak oscillations of torques in control subjects and patients. **(C)** An example of EMG activity in the upper and lower limb muscles in a healthy subject (left) and SCI patient (right, p3 **Table 1**) during walking in the exoskeleton. Note EMG activity in the upper limb muscles in both subjects. Note also some EMG activity in the ST, BF, and MG muscles in the patient despite neurologically complete paraplegia. **(D)** Mean (+ *SD*) EMG activity of the upper and lower limb muscles. Asterisks denote significant differences. **(E)** Mean EMG activities for each muscle (mean + *SD*).

results also showed a non-linear reorganization of EMG patterns under different walking conditions (**Figures 3–5**, **Table 2**). Below we discuss the findings in the context of adaptability of locomotor patterns and human-machine interactions.

EMG PATTERNS IN HEALTHY SUBJECTS

To assess similarities in the EMG waveforms across conditions, we used both the correlation analysis and calculated the center of EMG activity in the gait cycle. Both invariant features and significant variations in the EMG waveforms were observed across different walking conditions (**Figure 4**, **Table 2**).

For instance, the correlation analysis revealed that in EXO-assisted and normal slow walking Sol and MG muscles demonstrated significant correlations (**Table 2**), which could be explained by the fact that the exoskeleton ankles were not powered and in both conditions human ankles were actively contributing to locomotion and to antigravity calf muscle activity during foot loading in the stance phase (Nielsen and Sinkjaer, 2002). The most variable pattern was generally observed in the hamstring muscles (BF and ST). This can be explained by the important contribution of stretch reflexes in this muscle in the context of a "passive" contribution (Duysens et al., 1998), but it can also be interpreted in terms of the more proximal muscles being less dependent on sensory feedback than the distal ones (in the context of "active" contribution from central sources). Another explanation can be related to the fact that the hamstring muscle (in particular, the semimembranosus and semitendinosus muscles) is specifically involved in the locking of the erect posture

by producing a tonic activation against the action of gravity (Cheron et al., 1997). Indeed, an anticipated inhibition of the hamstring activity worked in conjunction with the phasic activation of the TA and the action of gravity (Cheron et al., 1997). In the present "EXO-assisted" situation, such inhibitory modulation related to normal graviception can be disturbed by the presence of these artificial forces. The context-specific function of the hamstring muscle was also reported in other experimental conditions (Ivanenko et al., 2000; Sylos-Labini et al., 2014).

The amplitude of EMG activity varied across conditions. Walking in EXO-unassisted mode in healthy individuals was accompanied by the augmented motor output (**Figures 3**, **4**), likely due to additional inertia and weight of the exoskeleton. Strikingly, however, walking in the EXO-assisted mode was not accompanied by the reduction of leg muscle EMG activity despite limb movement assistance. This can be explained in part by the important contribution of afferent feedback to the pre-programmed motoneuronal drive (Nielsen and Sinkjaer, 2002), different biomechanical demands and the "active" nature of stepping in the exoskeleton (the subject was not fully "relaxed," needed to maintain the upper trunk posture and provide small lateral trunk displacements to trigger step transitions) even though the limb movements were guided by the exoskeleton. Another possible cause could be the intermittent contact between the exoskeleton and the subject. The brace connections were not tight and had slag (for comforts and to prevent overloading human joints since minor misalignments could not be avoided.). It could be that the subject was ambulating on his own and the exoskeleton was acting as disturbances to the subject due to the intermittent contacts.

Adaptive non-linear changes in both amplitude and temporal envelope have been reported in other walking conditions as well (Hidler and Wall, 2005; Israel et al., 2006; Lam et al., 2008; Van Asseldonk et al., 2008; Moreno et al., 2013). For instance, with body weight unloading (Ivanenko et al., 2002), most muscles (e.g., gluteus maximus and distal leg extensors) decrease their activity, while other muscles demonstrate a "paradoxical" increment of activation (e.g., quadriceps) or considerable changes in the activation waveforms (hamstring muscles). Even the amplitude of EMG activity of "anatomical" synergists may diverge remarkably: lateral and medial gastrocnemius muscles at different walking speeds (Huang and Ferris, 2012), soleus and gastrocnemius muscles at different levels of limb loading (McGowan et al., 2010). In addition, muscle activity patterns are shaped by the direction of progression (e.g., forward vs. backward, Grasso et al., 1998, or walking along a curved path, Courtine et al., 2006). In particular, such studies suggest that a comparison of normal and pathological gait should be preferably performed in the same stepping conditions.

Taken together, the data support the idea of plasticity and distributed networks for controlling human locomotion (Scivoletto et al., 2007; Ivanenko et al., 2013). Tens of muscles participate in the control of limb and body movements during locomotion, and redundancy in the neuromuscular system is an essential element of gait adaptability (Winter, 1989; Cai et al., 2006; Noble and Prentice, 2006; Molinari, 2009; Duysens et al., 2013; Ivanenko et al., 2013). Due to muscle redundancy, various neuromotor strategies may exist to compensate for decreased muscle strength and pathology (Grasso et al., 2004; Goldberg and Neptune, 2007; Huang and Ferris, 2012; Gordon et al., 2013).

EMG PATTERNS IN SCI PATIENTS

Flexibility and adaptability of locomotor patterns are evident from monitoring and analyzing the spatiotemporal spinal segmental output after SCI (Grasso et al., 2004; Scivoletto et al., 2007). For instance, in motor incomplete paraplegics who recovered independent control of their limbs, an additional activation burst, related to abnormal activation of the quadriceps muscle, is often present in the lumbosacral enlargement (Ivanenko et al., 2013). Patients can be trained to step with body weight support unassisted, but they use activity patterns in individual muscles that were often different from healthy individuals (Grasso et al., 2004).

In this study we used the reference patterns based on pre-recorded trajectories from unimpaired volunteer walking in the device while it is operated in a transparent mode. Other approaches may be based on patient specific patterns by recording the gait trajectory while the patient walks with manual assistance (Aoyagi et al., 2007), but this may be done only in individuals with less severe paresis of the lower limbs. Further investigations are needed regarding the possible effect that the selected reference gait pattern may have on the findings and also regarding possible solutions for reference gait pattern customization for SCI.

Patients with severe SCI disorders frequently show EMG patterns different from those of healthy individuals suggesting that human spinal cord can interpret differently loading- or velocity-dependent sensory input during stepping (Beres-Jones and Harkema, 2004). Complete paraplegics also use more their arms and largely rely on proximal and axial muscles to assist the leg movements and balance control (**Figures 5D,E**, see also Grasso et al., 2004). During assisted walking in the exoskeleton, complete paraplegics typically showed little if any leg muscle activity (**Figure 5E**). Only one patient (p3, **Table 1**) demonstrated consistent activity in the BF, ST, RF, and MG muscles during swing and beginning of stance (**Figure 5D** right panel), suggesting the contribution of stretch- or loading-related afferent inputs to muscle activity (Maegele et al., 2002; Beres-Jones and Harkema, 2004; Grasso et al., 2004). Nevertheless, this reflex-related activity might be beneficial for potential gait rehabilitation since there is a relationship between facilitation of segmental reflexes and the ability to recover gait (Dietz et al., 2009; Thompson and Wolpaw, 2014). Thus, in addition to gait assistive aspects of exoskeleton robotic devices in severely paralyzed individuals, the proposed approach may also be beneficial for gait rehabilitation. We did not test in this study the effect of robot-assisted gait training in persons with SCI. Longer sessions would be required to evaluate the adequate learning paradigm, likely in combination with other central pattern generator-modulating therapies (Roy et al., 2012; Guertin, 2014) and biofeedback that might help the patients to adapt their movement patterns and to improve their motivation (Lünenburger et al., 2007).

CONCLUSIONS

Overall, the results are consistent with a non-linear reorganization of the locomotor output when using the robotic stepping devices. The findings may contribute to our understanding of human-machine interactions and adaptation of locomotor activity patterns. Locomotor movements can be accommodated to various external conditions, and some of the suggestions in this article may possibly be revised as empirical data on the sensorimotor interactions when walking with different types of exoskeletons accumulate. The effect of learning and adaptation is also an interesting avenue of future research. Such investigations may have important implications related to the construction of gait rehabilitation technology.

ACKNOWLEDGMENT

The financial support of the European Union FP7-ICT program (MINDWALKER grant #247959) and of the Italian Space Agency (COREA grant) are gratefully acknowledged.

REFERENCES

Aoyagi, D., Ichinose, W. E., Harkema, S. J., Reinkensmeyer, D. J., and Bobrow, J. E. (2007). A robot and control algorithm that can synchronously assist in naturalistic motion during body-weight-supported gait training following neurologic injury. *IEEE Trans. Neural Syst. Rehabil. Eng.* 15, 387–400. doi: 10.1109/TNSRE.2007.903922

Batschelet, E. (1981). *Circular Statistics in Biology*. New York, NY: Academic Press.

Berens, P. (2009). CircStat: a MATLAB toolbox for circular statistics. *J. Stat. Softw.* 31, 1–21.

Beres-Jones, J. A., and Harkema, S. J. (2004). The human spinal cord interprets velocity-dependent afferent input during stepping. *Brain* 127, 2232–2246. doi: 10.1093/brain/awh252

Cai, L. L., Courtine, G., Fong, A. J., Burdick, J. W., Roy, R. R., and Edgerton, V. R. (2006). Plasticity of functional connectivity in the adult spinal cord. *Philos. Trans. R. Soc. Lond. B Biol. Sci.* 361, 1635–1646. doi: 10.1098/rstb.2006.1884

Cappellini, G., Ivanenko, Y. P., Dominici, N., Poppele, R. E., and Lacquaniti, F. (2010). Motor patterns during walking on a slippery walkway. *J. Neurophysiol.* 103, 746–760. doi: 10.1152/jn.00499.2009

Cheron, G., Bengoetxea, A., Pozzo, T., Bourgeois, M., and Draye, J. P. (1997). Evidence of a preprogrammed deactivation of the hamstring muscles for triggering rapid changes of posture in humans. *Electroencephalogr. Clin. Neurophysiol.* 105, 58–71. doi: 10.1016/S0924-980X(96)96544-3

Cheron, G., Duvinage, M., De Saedeleer, C., Castermans, T., Bengoetxea, A., Petieau, M., et al. (2012). From spinal central pattern generators to cortical network: integrated BCI for walking rehabilitation. *Neural Plast.* 2012:375148. doi: 10.1155/2012/375148

Courtine, G., Papaxanthis, C., and Schieppati, M. (2006). Coordinated modulation of locomotor muscle synergies constructs straight-ahead and curvilinear walking in humans. *Exp. Brain Res.* 170, 320–335. doi: 10.1007/s00221-005-0215-7

del-Ama, A. J., Moreno, J. C., Gil-Agudo, A., de-los-Reyes, A., and Pons, J. L. (2012). Online assessment of human-robot interaction for hybrid control of walking. *Sensors* 12, 215–225. doi: 10.3390/s120100215

Dietz, V., Grillner, S., Trepp, A., Hubli, M., and Bolliger, M. (2009). Changes in spinal reflex and locomotor activity after a complete spinal cord injury: a common mechanism? *Brain* 132, 2196–2205. doi: 10.1093/brain/awp124

Duysens, J., De Groote, F., and Jonkers, I. (2013). The flexion synergy, mother of all synergies and father of new models of gait. *Front. Comput. Neurosci.* 7:14. doi: 10.3389/fncom.2013.00014

Duysens, J., van Wezel, B. M., van de Crommert, H. W., Faist, M., and Kooloos, J. G. (1998). The role of afferent feedback in the control of hamstrings activity during human gait. *Eur. J. Morphol.* 36, 293–299. doi: 10.1076/ejom.36.4.0293

Fitzsimmons, N. A., Lebedev, M. A., Peikon, I. D., and Nicolelis, M. A. L. (2009). Extracting kinematic parameters for monkey bipedal walking from cortical neuronal ensemble activity. *Front. Integr. Neurosci.* 3:3. doi: 10.3389/neuro.07.003.2009

Goldberg, E. J., and Neptune, R. R. (2007). Compensatory strategies during normal walking in response to muscle weakness and increased hip joint stiffness. *Gait Posture* 25, 360–367. doi: 10.1016/j.gaitpost.2006.04.009

Gordon, K. E., Kinnaird, C. R., and Ferris, D. P. (2013). Locomotor adaptation to a soleus EMG-controlled antagonistic exoskeleton. *J. Neurophysiol.* 109, 1804–1814. doi: 10.1152/jn.01128.2011

Grasso, R., Bianchi, L., and Lacquaniti, F. (1998). Motor patterns for human gait: backward versus forward locomotion. *J. Neurophysiol.* 80, 1868–1885.

Grasso, R., Ivanenko, Y. P., Zago, M., Molinari, M., Scivoletto, G., Castellano, V., et al. (2004). Distributed plasticity of locomotor pattern generators in spinal cord injured patients. *Brain* 127, 1019–1034. doi: 10.1093/brain/awh115

Guertin, P. A. (2014). Preclinical evidence supporting the clinical development of central pattern generator-modulating therapies for chronic spinal cord-injured patients. *Front. Hum. Neurosci.* 8:272. doi: 10.3389/fnhum.2014.00272

Hidler, J. M., and Wall, A. E. (2005). Alterations in muscle activation patterns during robotic-assisted walking. *Clin. Biomech.* 20, 184–193. doi: 10.1016/j.clinbiomech.2004.09.016

Hsu, J. D., Michael, J., and Fisk, J. (2008). *AAOS Atlas of Orthoses and Assistive Devices*. Philadelphia, PA: MOSBY Elsevier.

Huang, S., and Ferris, D. P. (2012). Muscle activation patterns during walking from transtibial amputees recorded within the residual limb-prosthetic interface. *J. Neuroeng. Rehabil.* 9:55. doi: 10.1186/1743-0003-9-55

Israel, J. F., Campbell, D. D., Kahn, J. H., and Hornby, T. G. (2006). Metabolic costs and muscle activity patterns during robotic- and therapist-assisted treadmill walking in individuals with incomplete spinal cord injury. *Phys. Ther.* 86, 1466–1478. doi: 10.2522/ptj.20050266

Ivanenko, Y. P., Cappellini, G., Solopova, I. A., Grishin, A. A., Maclellan, M. J., Poppele, R. E., et al. (2013). Plasticity and modular control of locomotor patterns in neurological disorders with motor deficits. *Front. Comput. Neurosci.* 7:123. doi: 10.3389/fncom.2013.00123

Ivanenko, Y. P., Dominici, N., Daprati, E., Nico, D., Cappellini, G., and Lacquaniti, F. (2011). Locomotor body scheme. *Hum. Mov. Sci.* 30, 341–351. doi: 10.1016/j.humov.2010.04.001

Ivanenko, Y. P., Grasso, R., and Lacquaniti, F. (2000). Influence of leg muscle vibration on human walking. *J. Neurophysiol.* 84, 1737–1747.

Ivanenko, Y. P., Grasso, R., Macellari, V., and Lacquaniti, F. (2002). Control of foot trajectory in human locomotion: role of ground contact forces in simulated reduced gravity. *J. Neurophysiol.* 87, 3070–3089.

Ivanenko, Y. P., Poppele, R. E., and Lacquaniti, F. (2006). Spinal cord maps of spatiotemporal alpha-motoneuron activation in humans walking at different Speeds. *J. Neurophysiol.* 95, 602–618. doi: 10.1152/jn.00767.2005

Jasiewicz, J. M., Allum, J. H. J., Middleton, J. W., Barriskill, A., Condie, P., Purcell, B., et al. (2006). Gait event detection using linear accelerometers or angular velocity transducers in able-bodied and spinal-cord injured individuals. *Gait Posture* 24, 502–509. doi: 10.1016/j.gaitpost.2005.12.017

Kendall, F. P., McCreary, E. K., Provance, P. G., Rodgers, M. M., and Romani, W. A. (2005). *Muscles: Testing and Function, with Posture and Pain*. 5th Edn. Baltimore, MD: Lippincott Williams & Wilkins.

Lam, T., Wirz, M., Lünenburger, L., and Dietz, V. (2008). Swing phase resistance enhances flexor muscle activity during treadmill locomotion in incomplete spinal cord injury. *Neurorehabil. Neural Repair* 22, 438–446. doi: 10.1177/1545968308315595

Lünenburger, L., Colombo, G., and Riener, R. (2007). Biofeedback for robotic gait rehabilitation. *J. Neuroeng. Rehabil.* 4:1. doi: 10.1186/1743-0003-4-1

Maegele, M., Müller, S., Wernig, A., Edgerton, V. R., and Harkema, S. J. (2002). Recruitment of spinal motor pools during voluntary movements versus stepping after human spinal cord injury. *J. Neurotrauma* 19, 1217–1229. doi: 10.1089/08977150260338010

Malcolm, P., Derave, W., Galle, S., and De Clercq, D. (2013). A simple exoskeleton that assists plantarflexion can reduce the metabolic cost of human walking. *PLoS ONE* 8:e56137. doi: 10.1371/journal.pone.0056137

McGowan, C. P., Neptune, R. R., Clark, D. J., and Kautz, S. A. (2010). Modular control of human walking: adaptations to altered mechanical demands. *J. Biomech.* 43, 412–419. doi: 10.1016/j.jbiomech.2009.10.009

Molinari, M. (2009). Plasticity properties of CPG circuits in humans: impact on gait recovery. *Brain Res. Bull.* 78, 22–25. doi: 10.1016/j.brainresbull.2008.02.030

Moreno, J. C., Barroso, F., Farina, D., Gizzi, L., Santos, C., Molinari, M., et al. (2013). Effects of robotic guidance on the coordination of locomotion. *J. Neuroeng. Rehabil.* 10:79. doi: 10.1186/1743-0003-10-79

Nielsen, J. B., and Sinkjaer, T. (2002). Afferent feedback in the control of human gait. *J. Electromyogr. Kinesiol.* 12, 213–217. doi: 10.1016/S1050-6411(02)00023-8

Noble, J. W., and Prentice, S. D. (2006). Adaptation to unilateral change in lower limb mechanical properties during human walking. *Exp. Brain Res.* 169, 482–495. doi: 10.1007/s00221-005-0162-3

Perry, J. (1992). *Gait Analysis: Normal and Pathological Function*. Thorofare, NJ: SLACK Incorporated.

Roy, R. R., Harkema, S. J., and Edgerton, V. R. (2012). Basic concepts of activity-based interventions for improved recovery of motor function after spinal cord injury. *Arch. Phys. Med. Rehabil.* 93, 1487–1497. doi: 10.1016/j.apmr.2012.04.034

Sale, P., Franceschini, M., Waldner, A., and Hesse, S. (2012). Use of the robot assisted gait therapy in rehabilitation of patients with stroke and spinal cord injury. *Eur. J. Phys. Rehabil. Med.* 48, 111–121.

Scivoletto, G., Ivanenko, Y., Morganti, B., Grasso, R., Zago, M., Lacquaniti, F., et al. (2007). Plasticity of spinal centers in spinal cord injury patients: new concepts for gait evaluation and training. *Neurorehabil. Neural Repair* 21, 358–365. doi: 10.1177/1545968306295561

Swinnen, E., Duerinck, S., Baeyens, J.-P., Meeusen, R., and Kerckhofs, E. (2010). Effectiveness of robot-assisted gait training in persons with spinal cord injury: a systematic review. *J. Rehabil. Med.* 42, 520–526. doi: 10.2340/16501977-0538

Sylos-Labini, F., Ivanenko, Y. P., Cappellini, G., Gravano, S., and Lacquaniti, F. (2011). Smooth changes in the EMG patterns during gait transitions under body weight unloading. *J. Neurophysiol.* 106, 1525–1536. doi: 10.1152/jn.00160.2011

Sylos-Labini, F., Ivanenko, Y. P., Maclellan, M. J., Cappellini, G., Poppele, R. E., and Lacquaniti, F. (2014). Locomotor-like leg movements evoked by rhythmic arm movements in humans. *PLoS ONE* 9:e90775. doi: 10.1371/journal.pone.0090775

Thompson, A. K., and Wolpaw, J. R. (2014). Operant conditioning of spinal reflexes: from basic science to clinical therapy. *Front. Integr. Neurosci.* 8:25. doi: 10.3389/fnint.2014.00025

Van Asseldonk, E. H. F., Veneman, J. F., Ekkelenkamp, R., Buurke, J. H., Van der Helm, F. C. T., and van der Kooij, H. (2008). The effects on kinematics and muscle activity of walking in a robotic gait trainer during zero-force control. *IEEE Trans. Neural Syst. Rehabil. Eng.* 16, 360–370. doi: 10.1109/TNSRE.2008.925074

Wang, L., Wang, S., van Asseldonk, E. H. F., and van der Kooij, H. (2013). "Actively controlled lateral gait assistance in a lower limb exoskeleton," in *2013 IEEE/RSJ International Conference on Intelligent Robots and Systems (IROS)* (Tokyo), 965–970.

Winter, D. A. (1989). Biomechanics of normal and pathological gait: implications for understanding human locomotor control. *J. Mot. Behav.* 21, 337–355. doi: 10.1080/00222895.1989.10735488

Winter, D. A. (1991). *The Biomechanics and Motor Control of Human Gait: Normal, Elderly and Pathological*. Waterloo, ON: University of Waterloo Press.

Yakovenko, S., Mushahwar, V., VanderHorst, V., Holstege, G., and Prochazka, A. (2002). Spatiotemporal activation of lumbosacral motoneurons in the locomotor step cycle. *J. Neurophysiol.* 87, 1542–1553.

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.

Received: 22 January 2014; accepted: 27 May 2014; published online: 16 June 2014.
*Citation: Sylos-Labini F, La Scaleia V, d'Avella A, Pisotta I, Tamburella F, Scivoletto G, Molinari M, Wang S, Wang L, van Asseldonk E, van der Kooij H, Hoellinger T, Cheron G, Thorsteinsson F, Ilzkovitz M, Gancet J, Hauffe R, Zanov F, Lacquaniti F and Ivanenko YP (2014) EMG patterns during assisted walking in the exoskeleton. Front. Hum. Neurosci. **8**:423. doi: 10.3389/fnhum.2014.00423*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 22 May 2014
doi: 10.3389/fnhum.2014.00318

Robot-assisted vs. sensory integration training in treating gait and balance dysfunctions in patients with multiple sclerosis: a randomized controlled trial

Marialuisa Gandolfi[1], Christian Geroin[1], Alessandro Picelli[1], Daniele Munari[1], Andreas Waldner[2], Stefano Tamburin[3], Fabio Marchioretto[4] and Nicola Smania[1,5]*

[1] *Department of Neurological and Movement Sciences, Neuromotor and Cognitive Rehabilitation Research Center, University of Verona, Verona, Italy*
[2] *Department of Neurological Rehabilitation, Private Hospital Villa Melitta, Bolzano, Italy*
[3] *Neurology Section, Department of Neurological and Movement Sciences, University of Verona, Verona, Italy*
[4] *Neurological Unit, Sacro Cuore–Don Calabria Hospital, Verona, Italy*
[5] *Neurological Rehabilitation Unit, Azienda Ospedaliera Universitaria Integrata, Verona, Italy*

Edited by:
Marco Iosa, Fondazione Santa Lucia, Italy

Reviewed by:
Federica Tamburella, Fondazione Santa Lucia, Italy
Alessandra Pompa, Fondazione Santa Lucia, Italy

Correspondence:
Nicola Smania, Department of Neurological and Movement Sciences, Neuromotor and Cognitive Rehabilitation Research Center (CRRNC), University of Verona, P.le L.A. Scuro 10, 37134, Verona, Italy
e-mail: nicola.smania@univr.it

Background: Extensive research on both healthy subjects and patients with central nervous damage has elucidated a crucial role of postural adjustment reactions and central sensory integration processes in generating and "shaping" locomotor function, respectively. Whether robotic-assisted gait devices might improve these functions in Multiple sclerosis (MS) patients is not fully investigated in literature.

Purpose: The aim of this study was to compare the effectiveness of end-effector robot-assisted gait training (RAGT) and sensory integration balance training (SIBT) in improving walking and balance performance in patients with MS.

Methods: Twenty-two patients with MS (EDSS: 1.5–6.5) were randomly assigned to two groups. The RAGT group ($n = 12$) underwent end-effector system training. The SIBT group ($n = 10$) underwent specific balance exercises. Each patient received twelve 50-min treatment sessions (2 days/week). A blinded rater evaluated patients before and after treatment as well as 1 month post treatment. Primary outcomes were walking speed and Berg Balance Scale. Secondary outcomes were the Activities-specific Balance Confidence Scale, Sensory Organization Balance Test, Stabilometric Assessment, Fatigue Severity Scale, cadence, step length, single and double support time, Multiple Sclerosis Quality of Life-54.

Results: Between groups comparisons showed no significant differences on primary and secondary outcome measures over time. Within group comparisons showed significant improvements in both groups on the Berg Balance Scale ($P = 0.001$). Changes approaching significance were found on gait speed ($P = 0.07$) only in the RAGT group. Significant changes in balance task-related domains during standing and walking conditions were found in the SIBT group.

Conclusion: Balance disorders in patients with MS may be ameliorated by RAGT and by SIBT.

Keywords: sensory feedback, proprioception, postural balance, motor skills disorders, physiological adaptations

INTRODUCTION

Multiple Sclerosis (MS) is a chronic disease of the central nervous system characterized by a progressive decline in various neurologic functions such as vision, sensation, coordination and balance, muscle strength and tone (Nelson et al., 1995; Speers et al., 2002). All these impairments might contribute to walking disturbances, which represent a hallmark presentation of MS (Larocca, 2011). Longitudinal studies showed that up to 80% of patients with MS necessitate an assistive device for walking with disease progression (Weinshenker et al., 1989; Confavreux et al., 2000). This condition affects participation outcomes such as quality of life, daily living activities and work (Larocca, 2011).

Data from studies on healthy subjects showed that gait involves a complex interplay between cortical and spinal circuits. A full review of neural correlates of walking control is beyond this perspective. Nevertheless, the overall evidence that locomotion control relies on the integrity of feedback and feed forward mechanisms of movement control (including postural adjustments) (Dietz et al., 2002; Nielsen and Sinkjaer, 2002; Pearson, 2004) has been well established.

Although a wide range of movement control dysfunctions might contribute to gait impairment in people with MS, balance disorders are thought to contribute to most of MS walking-related disabilities. Indeed, they negatively influence gait performance by

reducing gait velocity, shortening steps length, increasing double support time and decreasing single support and swing times (Cameron and Lord, 2010). It is worth noting that most of MS patients with walking disturbances report having balance problems even when they have minimal or no clinically assessable impairments (Cameron et al., 2008; Larocca, 2011; Zackowski et al., 2013).

Several studies investigating locomotion disturbances in patients with MS showed that both MS-specific reorganization of the posture control system (Corradini et al., 1997) and deficits of central integration of sensory afferents are involved as primary mechanisms. The former consists of having very delayed onset of both compensatory (CPAs) (Cameron et al., 2008; Huisinga et al., 2014) and anticipatory postural adjustment (APAs) (Krishnan et al., 2012a,b) while standing and walking. The latter consists of the inability of the central nervous system to use different sensory input (mainly vestibular, somatosensory, and visual) in order to create a system of coordinates on which the body's postural control is based (Smania et al., 2008). Central integration deficits are a rather underestimated issue in MS people even though they affect postural adjustment reactions and then balance during upright posture and gait (Huisinga et al., 2014).

As a whole, these evidence support that specific treatments aimed at improving the efficiency of postural reactions can improve gait quality and might potentially contribute to an improvement in activity, community participation, and quality of life in people with MS (Cameron et al., 2008).

Rehabilitation studies have shown that different approaches may be useful in treating balance disturbances stemming from neurological dysfunctions. Conventional balance rehabilitation strategies proved to be effective in both stroke and Parkinson disease (Smania et al., 2008, 2010). Recently, new strategies for balance rehabilitation have been put forward. On one hand, preliminary data on the effects of balance exercises stressing the processing (and/or central integration) of specific sensory afferents (i.e., somatosensory, visual, and vestibular) led to improvement of gait and balance performance in patients with stroke and spinal cord injury (Tamburella et al., 2013). On the other hand, robot-assisted training of gait in patients with Parkinson's disease has shown not only to improve gait but also balance parameters. The effects of both these approaches have been, at least in part, related to the role of somatosensory inputs in balance control. Indeed, somatic sensation (in particular proprioception) is very important for the efficiency of both feedback and feed forward control of gait and posture (Riemann and Lephart, 2002).

The aim of this study was to compare the effectiveness of end-effector robot-assisted gait training (RAGT) and sensory integration balance training (SIBT) in improving walking and balance performance in patients with MS. The hypothesis was that both types of training might promote central neural processes involved in feedback and feed forward control of gait and balance. The rationale behind the study is twofold. First, it would further explore the potential field of application of new technological devices, which are increasingly being used in clinical practice even though their mechanisms of action are still unknown. Second, it would be relevant to find new approaches which allow training patients in a safe and efficient manner even when neurological condition is severe.

MATERIALS AND METHODS

TRIAL DESIGN

A single blind RCT comparing the effects between the experimental (RAGT) and control group (SIBT) on walking and balance disorders was performed (allocation ratio 1:1). The examiner was blinded to group assignment.

PARTICIPANTS

Outpatients with relapsing remitting or secondary progressive MS (Polman et al., 2011) were enrolled in the Neurological Rehabilitation Unit of the Department of Neurological and Movement Sciences, University Hospital, Italy. Inclusion criteria were: age 30–60 years, Expanded Disability Status Score (EDSS) between $1.5 \geq x \leq 6.5$ (Kurtzke, 1983), Mini Mental State Examination score ≥ 24 (Folstein et al., 1975), ability to maintain standing position without aids for at least 1 min and ability to walk independently for at least 15 m, absence of concomitant neurological or orthopedic conditions that may interfere with ambulation. Exclusion criteria were: any type of rehabilitation intervention in the month prior to recruitment, MS relapse during the three months prior to recruitment, pharmacological therapy not well defined and/or changed during the study, presence of paroxysmal vertigo, lower limb botulinum toxin injections within the previous 12 weeks. All patients were informed regarding the experimental nature of the study. Written informed consent was given by patients. The research was performed in accordance with the Helsinki Declaration. The ethical approval was obtained from the ethics committee of the Azienda Ospedaliera Universitaria Integrata (Verona-Italy) (Prog.CE 1893). Patients enrolled in this study are a subgroup of a clinical trial registered at the http://clinicaltrials.gov (NCT01564511).

INTERVENTIONS

Prior to the start of the study, authors designed RAGT and SIBT protocols and instructed two treating physiotherapists, one for the RAGT group and the other for the SIBT group. Treatment procedures consisted of 12 individual sessions of 50 min, twice weekly (Monday and Wednesday or Tuesday and Friday) for six consecutive weeks. Both treatments were tailored to suit each patient's ability and task complexity was progressively increased as the patient improved. Patients were not allowed to receive other physiotherapy during the study, but were given no other activity restrictions. Training procedures were administered in the morning around 10 AM, to ensure that fatigue did not influence the patient's performance. Participants were allowed to wear their usual footwear and orthoses.

RAGT training

The RAGT group was treated by means of the electromechanical Gait Trainer GT1 (Reha-Stim, Berlin, Germany) (Hesse et al., 1999). During RAGT, individuals were secured in a harness with their feet on footplates, while movements of the center of mass were controlled in a phase-dependent manner by ropes attached to the harness. Patients received a 40 min of RAGT followed by

10 passive lower limb joint mobilizations and stretching exercises. The overall duration of RAGT therapy, including the time getting in and out was 40 min while the net RAGT lasted 30 min. Each training session consisted of two 15-min sessions, separated by a 5-min rest if required by the patient. In the first session we trained patients at 20% of supported body-weight and 1.3 km/h of speed; in the second session at 10% of supported body-weight and 1.6 km/h of speed (Picelli et al., 2012). The use of body weight support permits patients to walk more symmetrically with higher velocities resulting in a facilitation of the lower limb muscles and in a more effective gait (Hesse, 2008). In our view, the rationale for supporting body weight was to increase safety and compliance with the RAGT. Patients were instructed to "help" the GT1 gait-like movement during training. Patients unable to maintain the pace were excluded. The step-length was evaluated with the GAITRite system (CIR Systems, Havertown, PA) and individually defined (Givon et al., 2009).

SIBT training

The SIBT patients underwent a specific training program aimed at improving the ability to integrate multisensory inputs during balance responses (Nichols, 1997). Each session consisted of exercises fitting to three different levels of difficulty and repeated under three different sensory conditions (free vision, wearing a mask and wearing an helmet) (Smania et al., 2008). Level I included tasks that induced external destabilizations of the center-of-body mass (CoP) while standing on a stable and comfortable surface (i.e., the physiotherapist shifted the pelvis in the frontal and sagittal direction asking the patient to actively maintain balance standing on the floor). These tasks mainly involved feedback postural control. Level II included exercises of self-destabilization of the CoP. The patient performed voluntary motor actions in both static and dynamic conditions while standing on a stable and comfortable surface (i.e., performing a single-step simulation, shifting his/her weight from one foot to the other in the frontal direction standing on the floor). These tasks mainly involved feed-forward postural control. Level III consisted of exercises of external destabilization and exercises of self-destabilization of the CoP while standing on different types of compliant surfaces (i.e., increasing weight shifting and decreasing the amplitude of the base of support while standing on foam support bases of different consistency). Three different foam sections were used according to the patient's abilities (1.5, 3.5, and 8 cm). These tasks required continuous feedback and feed forward postural adjustments. During each treatment session, a total of 10 exercises (3 from level I, 3 from level II, 4 from level III) were repeated several times (2–5 times) within a 5-min period (Smania et al., 2008).

OUTCOMES

An examiner, who was blinded to the patients' group allocation, performed all evaluations. Primary and secondary outcomes were measured before (T0), after treatment (T1) and at 1-month follow-up (T2). Patients were examined around 10 AM in the morning to reduce the effect of fatigue frequently reported later in the day.

Primary outcomes measures

Gait speed (cm/s). It was assessed by the GAITRite system (Gold version 3.2b; CIR System Inc, Havertown, PA, USA) (Menz et al., 2004). Patients walk along a mat with integrated sensors 4 times in a self-selected speed. Patients were allowed to use orthoses but not other walking aids (i.e., cane).

Berg balance scale (BBS). A 14-item validated scale used to evaluate both static and dynamic balance disorders after rehabilitative interventions in individuals with MS (Range of score: 0–4 points per task; higher = better performance) (Cattaneo et al., 2006).

Secondary outcomes measures

Activities-specific balance confidence scale (ABC). A validated and reliable interview that evaluates the patient's perceived level of balance confidence during 16 daily living activities such as walking, bending, standing and reaching (Range of score: 0–100 points per activity; higher = more confident) (Powell and Myers, 1995).

Sensory organization balance test (SOT). A validated balance test to evaluate central integration deficit of sensory inputs in patients with neurologic impairment. The patient stands barefoot with arms alongside the body and feet in a heel-to-toe position and maintains standing balance under 6 different sensory conditions according to the original protocol. The sensory conditions are: (1) stable surface eyes open, closed, and dome condition; and (2) compliant surface eyes open, closed and dome condition. A stopwatch records the amount of time a patient maintains erect standing without activating any postural reaction. Five 30-s trials are carried out for each condition (Range of score: 0–150 s; higher = better performance) (Shumway-Cook and Horak, 1986).

Stabilometric assessment (SA). A widely used instrument to evaluate balance disorders in patients with neurological impairment. Patients are evaluated in the standing position on an electronic monoaxial platform (Technobody© platform)[1]. The feet position on the platform is standardized for all patients using a V-shaped frame. The subjects while standing placed the medial borders of the feet alongside the frame. The malleolus are aligned to vertical bars. The distance between two malleolus is 3 cm and the medial borders of the feet were extra-rotated 12° with regard to the anterior-posterior axis. Patients are evaluated while standing without upper limbs support. An operator stands behind them in order to prevent the risk of falling (Cattaneo and Jonsdottir, 2009).

The patient is tested in two consecutive conditions (eyes-open and eyes-closed) each lasting 30-s according to Cattaneo and Jonsdottir's protocol (Cattaneo and Jonsdottir, 2009). Main stabilometric parameters evaluated are sway area and length of CoP trajectory.

Fatigue severity scale (FSS). A 9-item self-reported questionnaire which assesses the perceived level of fatigue on a 7-point scale (Range of score: 1–7; higher = worse performance) (Krupp et al., 1989).

[1]Website: http://www.tecnobody.it.

Gait analysis

The following spatiotemporal gait parameters were evaluated by means of GAITRite System: cadence (step/min), step length (cm), single support time (% of cycle) and double support time (% of cycle) (Menz et al., 2004).

Multiple sclerosis quality of Life-54 (MSQOL-54). A 54-item validated structured self-report questionnaire that evaluates both generic and MS-specific domains of health-related QoL with 12 subscales. Two summary scores, physical health (PHC) and mental health (MHC) are reported (Range of score: 0–100; higher = better performance) (Solari et al., 1999).

Randomization procedure

After screening, an independent blinded collaborator who was not involved in the treatment or care of patients, randomly assigned eligible patients to the RAGT or SIBT according to a simple software-generated randomization scheme (Dallal, 2007).

STATISTICAL ANALYSIS

The Mann-Whitney test was used for testing differences between groups at baseline. The Friedman's ANOVA was used to analyse within-group changes in performance overtime, whilst the Wilcoxon signed rank tests to compare within-group changes from baseline/post-treatment and baseline/follow-up measures. The Mann-Whitney test was used for between-group comparisons. For this purpose, we computed the differences (Δ) between post- and pre-treatment performance and between follow-up and pre-treatment performance for all outcome measures. We set the alpha level for significance at 0.05, however, to adjust for multiple comparisons we used a Bonferroni correction (alpha level = 0.025). Descriptive analysis was used to evaluate the effect size measures between the 2 independent groups (Cohen's *d* calculation) (Cohen, 1988). All statistical analysis was carried out using the SPSS for Macintosh statistical package, version 16.0.

RESULTS

PARTICIPANTS

Thirty-two patients were evaluated for eligibility between September 2011 and November 2013. Five patients were excluded because they did not meet the inclusion criteria and 1 declined to participate. Thus, 26 patients were randomly allocated to the RAGT ($n = 14$) or SIBT ($n = 12$).

Two patients in the RAGT and 2 in the SIBT did not complete the allocated intervention due to difficulty in transportation or medical complications to treatment sessions (**Figure 1**). Therefore, 12 patients in the RAGT and 10 in the SIBT completed the study (**Table 1**).

Multiple separate independent-sample Mann-Whitney tests showed that there was no significant difference between groups as to age, EDSS, disease duration, and all baseline clinical measures at T0 (**Table 1**).

Primary outcome measures

Between groups comparisons showed no significant changes on primary outcome measures over time (**Table 2**).

In the RAGT group we found within-group changes (Friedman' ANOVA) approaching significance on gait speed ($P = 0.07$) and significant changes on BBS ($P = 0.001$) over time (**Figure 2**). In the SIBT group overall significant changes were found only on BBS ($P = 0.001$). Pairwise comparisons are reported in **Table 3A** and **Figure 2**.

Secondary outcome measures

Between groups comparisons showed significant differences on performance at SOT compliant surface-dome condition ($P = 0.048$) in favor of the SIBT training at follow-up (**Table 3A** and **Figure 2**). No significant differences between groups in all secondary outcome measures were found over time (**Table 2**).

In the RAGT group within-group significant changes (Friedman' ANOVA) on ABC ($P = 0.017$), on SOT stable surface eyes-closed condition ($P = 0.04$) and step length ($P = 0.017$) were found over time. Changes approaching significance were found on SOT stable surface dome condition ($P = 0.06$).

In the SIBT group within-group significant changes (Friedman' ANOVA) on ABC ($P = 0.001$), on SOT stable surface eyes-open condition ($P = 0.013$), on SOT stable surface eyes-closed condition ($P = 0.027$), on SOT compliant surface dome condition ($P = 0.003$), sway area eyes-closed ($P = 0.04$), step length ($p = 0.002$), double support ($P < 0.001$) and MSQOL-PH ($P = 0.032$) were found over time. Changes approaching significance were found on SOT compliant surface eyes-closed ($P = 0.06$) and on FSS ($P = 0.052$). Pairwise comparisons are reported in **Tables 3A**,**B** and **Figure 2**.

DISCUSSION

Results showed that RAGT and SIBT might improve step length, postural stability and the level of balance confidence perceived while performing daily activities in patients with MS. These training effects may be maintained for at least 1 month post-treatment.

So far many approaches have been proposed to improve walking and balance in people with MS (Armutlu et al., 2001; Schuhfried et al., 2005; Newman et al., 2007; Benedetti et al., 2009; Broekmans et al., 2010; Cakt et al., 2010; Hebert et al., 2011; Swinnen et al., 2012; Nilsagård et al., 2013). However, only few RCT studies (Beer et al., 2008; Lo and Triche, 2008; Schwartz et al., 2012; Vaney et al., 2012; Straudi et al., 2013) evaluated whether RAGT may be superior to conventional walking training in terms of gait performance. Furthermore, one study of them evaluated treatment effects on mobility assessed by Time Up and Go Test (Straudi et al., 2013) and only one on balance impairment by means BBS (Schwartz et al., 2012). Beer et al. (2008) found a moderate to large effect size, although not significant, for walking speed, distance and knee-extensor strength favoring RAGT. In the present study both groups' outcomes values returned to baseline at six months follow-up (Beer et al., 2008). Lo et al. (Lo and Triche, 2008) reported in their crossover study no differences in gait outcomes between treatment groups after 6 sessions of training. Vaney et al. (2012) reported that the over ground walking group improved gait speed insignificantly more than the RAGT. Straudi (Straudi et al., 2013) and colleagues showed walking endurance, as well as spatio-temporal gait parameters improvements after RAGT. In the present study within-group analysis showed no significant effects on the TUG test (Straudi et al., 2013). Finally, Schwartz et al. (2012) revealed

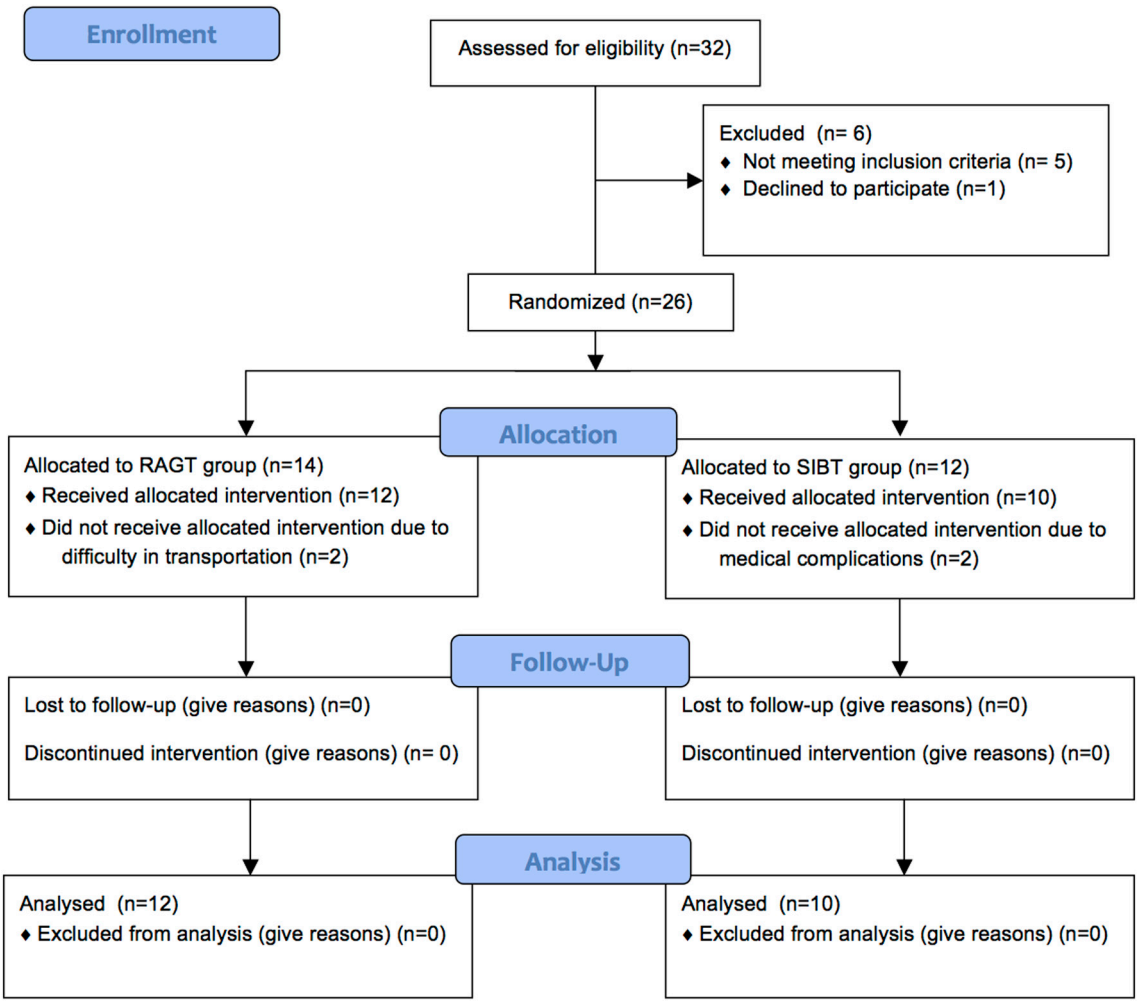


FIGURE 1 | Flow diagram of the study.

beneficial effects in term of gait, mobility, and balance comparable to conventional walking treatment. In the present study both RAGT and conventional walking training groups showed significant improvement on TUG test without any difference between groups (Schwartz et al., 2012). The conventional walking exercise appeared to have better long-term influence on postural control compared to RAGT in this study.

This is the first pilot study that evaluates the effects of an end-effector RAGT compared to a SIBT in walking and balance performance in patients with MS.

It is worthy to note that in all previous studies an exoskeleton device (Lokomat) was used as RAGT, and the control group consisted of over ground walking training. Thus, differences with our study were twofold. On one hand, the type of device used as RAGT was an end-effector device (Gang Trainer GT1) and on the other hand the type of treatment used as "control" condition was specific SIBT.

The Gang Trainer GT1 (Hesse et al., 1999) is an end-effector system, on which the harness-secured patients were positioned on 2 footplates, whose movements simulated stance and swing in a highly physiological manner. The body weight could be partially relieved, and ropes attached to the patient controlled the vertical and lateral movements of the center of mass in a phase-dependent manner (Hesse et al., 1999). Results showed that the GT1 training might promote changes on gait speed approaching significance. This might be attributed to the limited sample size and/or to low intensity of training procedures (30 min of RAGT, twice a week). Nevertheless, it is important to note that treatment procedures for RAGT in people with MS are still undefined in terms of intensity and variability of exercise.

Interestingly, significant changes in the GT1 group were found also on postural stability. This can be considered as one of the most important findings in our study because the majority of the existing literature on RAGT in MS patients does not evaluate this outcome. The issue of balance recovery is very relevant in MS rehabilitation studies (Cameron and Lord, 2010).

Walking can be seen as a repeated sequence of balance challenges (Cameron and Lord, 2010) and changes in gait observed in people with MS are largely the result of changes in postural control (Cameron and Lord, 2010). Overall evidence on

Table 1 | Baseline demographic and clinical features of the patients.

	RAGT group (n = 12) Mean (*SD*)	SIBT group (n = 10) Mean (*SD*)	*P value* (*Z*)
Age (years)	50.83 (8.42)	50.1 (6.29)	0.640 (−468)
Range	38–63	42–60	
Sex (Male/Female)	5/7	1/9	
EDSS	3.96 (0.75)	4,35 (0.67)	0.101 (−1.640)
Range	3–5.5	3.5–5.5	
Disease duration (years)	13.5 (7.60)	14.9 (8.68)	0.731 (−0.344)
Range	5–34	5–27	
PRIMARY OUTCOME MEASURE			
Gait speed (cm/s)	79.42 (21.14)	81.31 (16.81)	0.895 (−0.132)
BBS (0–56)	47.17 (5.27)	46.50 (6.69)	0.921 (−0.100)
SECONDARY OUTCOME MEASURES			
ABC scale (0–100)	59.68 (11.31)	61.90 (7.06)	0.226 (−1.210)
SOT S. surface (0–150)			
EO	118.73 (39.52)	114.56 (38.66)	0.691 (−0.398)
EC	63.37 (28.12)	52.23 (24.65)	0.210 (−1.253)
Dome	62.75 (40.07)	57.89 (31.97)	0.895 (−0.132)
SOT C. surface (0–150)			
EO	96.48 (37.76)	110.43 (23.27)	0.322 (−0.990)
EC	50.59 (32.95)	39.06 (16.82)	0.598 (−0.528)
Dome	58.52 (45.85)	30.96 (15.80)	0.176 (−1.352)
Stabilometric assessment EO condition			
Sway area (mm^2)	83.48 (83.53)	128.54 (84.03)	0.121 (−1.550)
Length CoP (mm)	499.66 (499.0)	504.60 (408.06)	0.644 (−0.462)
Stabilometric assessment EC condition			
Sway area (mm^2)	250.48 (261.99)	509.22 (342.79)	0.065 (−1.846)
Length CoP (mm)	951.66 (1045.29)	1157.90 (951.54)	0.429 (−0.791)
Gait analysis			
Cadence (step/min)	117.13 (28.22)	106.32 (37.49)	0.138 (−1.481)
SL (cm)	56.88 (19.57)	58.62 (25.99)	0.843 (−0.198)
SS time (% of cycle)	33.85 (4.23)	40.84 (16.49)	0.235 (−1.187)
DS time (% of cycle)	31.30 (5.48)	39.54 (14.35)	0.187 (−1.319)
FSS (1–7)	4.40 (1.386)	4.03 (2.25)	0.598 (−0.528)
MSQOL-54 PHC (0–100)	64.17 (6.53)	59.59 (10.67)	0.288 (−1.061)
MSQOL-54 MHC (0–100)	59.01 (21.69)	59.51 (20.70)	1.000 (0.000)

*SD, standard deviation; P value (Z), p value and corresponding t.statistic; EDSS, expanded disability status scale; BBS, berg balance scale (higher score = better performance); ABC scale, activities balance confidence scale (higher score = better performance); SOT, sensory organization balance test; EO, eyes-open condition; EC, eyes-closed condition; S, stable; C, compliant; Length CoP, length of CoP trajectory; FSS, fatigue severity scale (higher score = worse performance); SL, step length; SS, single support time; DS, double support time; MSQOL-54 PHC, multiple sclerosis quality of Life-54 physical health (higher score = better performance); MSQOL-54 MHC, multiple sclerosis quality of Life-54 mental health (higher score = better performance); *Statistically significant. p value significant if <0.05.*

Table 2 | Descriptive and inferential statistics for clinical outcome measures.

Outcome variables	Mann-Whitney test between-group differences	
	T1 vs. T0 *P* value (Effect size)	T2 vs. T0 *P* value (Effect size)
PRIMARY OUTCOME MEASURE		
Gait speed (cm/s)	0.644 (0.10)	0.895 (0.02)
BBS (0–56)	0.547 (0.13)	0.091 (0.28)
SECONDARY OUTCOME MEASURES		
ABC scale (0–100)	0.741 (0.09)	0.692 (0.08)
SOT—S. surface (0–150) EO	0.197 (−0.34)	0.075 (−0.37)
EC	0.947 (−0.13)	0.210 (−0.27)
Dome	0.187 (0.29)	0.553 (0.15)
SOT—C. surface (0–150) EO	0.843 (−0.05)	0.553 (0.01)
EC	0.644 (−0.12)	0.291 (−0.30)
Dome	0.129 (−0.31)	0.048 (−0.32)*
SA EO condition		
Sway area (mm^2)	0.817 (−0.20)	0.553 (−0.10)
Length CoP (mm)	0.468 (−0.05)	0.895 (−0.10)
SA EC condition		
Sway area (mm^2)	0.210 (0.20)	0.598 (0.10)
Length CoP (mm)	0.767 (0.20)	0.895 (−0.03)
Gait analysis		
Cadence (step/min)	0.322 (−0.22)	0.339 (−0.25)
SL (cm)	0.235 (−0.11)	0.644 (0.02)
SS time (% of cycle)	0.742 (0.15)	0.974 (−0.15)
DS time (% of cycle)	0.065 (0.41)	0.166 (0.15)
FSS (1–7)	0.276 (0.18)	0.391 (0.08)
MSQOL-54 PHC (0–100)	0.261 (−0.27)	0.869 (−0.06)
MSQOL-54 MHC (0–100)	0.667 (−0.15)	0.235 (0.07)

*Before, pre-treatment; After, post-treatment; SD, standard deviation; P Value, p value; BBS, berg balance scale (higher score = better performance); ABC scale, activities balance confidence scale (higher score = better performance); SOT, sensory organization balance test; EO, eyes-open condition; EC, eyes-closed condition; S, stable; C, compliant; SA, stabilometric assessment; Length CoP, length of CoP trajectory; FSS, fatigue severity scale (higher score = worse performance); SL, step length; SS, SINGLE support time; DS, double support time; MSQOL-54 PHC, multiple sclerosis quality of Life-54 physical health (higher score = better performance); MSQOL-54 MHC, multiple sclerosis quality of Life-54 mental health; *Statistically significant; p value significant if <0.05.*

healthy subjects showed that gait is the result of intricate dynamic interactions between a central program, so-called central pattern generator (CPG), and feedback mechanisms. The central program is based on a genetically determined spinal circuitry that allows generating basic gait functions such as starting, stopping, and steer locomotion (Rossignol et al., 2006). In contrast, feedback mechanisms activated by afferents inputs resulting from skin, muscles and special senses (vision, vestibular, and auditory) dynamically shape gait pattern to the environments necessities (Rossignol et al., 2006). To deal with these environments

Table 3A | Descriptive and inferential statistics for clinical outcome measures.

Outcome variables	Before Mean (SD)		After mean (*SD*)		1-month FU		Comparisons Wilcoxon signed ranks test		Comparisons Wilcoxon signed ranks test	
	RAGT	SIBT	RAGT	SIBT	RAGT	SIBT	RAGT		SIBT	
							T1 vs. T0 *P* value (95% CI)	T2 vs. T0 *P* value (95% CI)	T1 vs. T0 *P* value (95% CI)	T2 vs. T0 *P* value (95% CI)
PRIMARY OUTCOME MEASURE										
Gait speed (cm/s)	79.42 (21.14)	81.31 (16.81)	86.49 (23.05)	85.11 (12.96)	86.04 (21.67)	87.44 (13.93)	0.117 (−1.67; 15.81)	0.050 (−2.17; 15.41)	0.515 (−8.51; 16.10)	0.333 (−6.63; 18.87)
BBS (0−56)	47.17 (5.27)	46.50 (6.69)	52.58 (2.64)	50.70 (5.74)	53.33 (2.06)	50.00 (5.46)	0.007 (1.95; 8.88)*	0.002 (3.07; 9.25)*	0.007 (1.38; 7.01)*	0.018 (0.38; 6.61)*
SECONDARY OUTCOME MEASURES										
ABC scale (0−100)	59.68 (11.31)	61.90 (7.06)	70.79 (11.04)	72.14 (10.24)	69.09 (10.38)	70.63 (9.72)	0.010 (2.81; 19.41)*	0.023 (1.88; 16.94)*	0.008 (3.94; 14.48)*	0.012 (3.67; 12.04)*
SOT (0−150)—S. surface EO	118.73 (39.52)	114.56 (38.66)	125.76 (34.80)	143.89 (10.88)	123.22 (35.09)	137.61 (24.38)	0.86 (−2.82; 16.88)	0.386 (−5.38; 14.37)	0.015 (0.55; 58.09)*	0.021 (2.49; 43.59)*
EC	63.37 (28.12)	52.23 (24.65)	71.53 (30.50)	65.82 (20.40)	73.83 (31.19)	79.13 (37.07)	0.136 (−7.20; 23.52)	0.209 (−10.08; 30.97)	0.047 (2.01; 25.15)	0.009 (7.70; 46.09)*
Dome	62.75 (40.07)	57.89 (31.97)	77.15 (38.17)	59.36 (27.36)	76.77 (36.18)	65.83 (40.28)	0.022 (1.27; 27.51)*	0.059 (0.30; 27.72)	0.878 (−14.54; 17.47)	0.241 (−5.58; 21.46)
SOT (0−150)—C. surface EO	96.48 (37.76)	110.43 (23.27)	100.92 (38.83)	116.56 (18.98)	95.73 (37.12)	109.46 (36.75)	0.285 (−7.10; 15.99)	0.790 (−11.97; 10.46)	0.260 (−4.13; 16.37)	0.445 (−24.38; 22.44)
EC	50.59 (32.95)	39.06 (16.82)	63.03 (35.75)	58.86 (18.41)	56.70 (34.49)	64.90 (35.38)	0.272 (−12.55; 37.43)	0.272 (−15.14; 27.36)	0.013 (6.82; 32.77)*	0.028 (5.10; 46.58)
Dome	58.52 (45.85)	30.96 (15.80)	63.33 (37.54)	51.33 (27.06)	56.23 (37.13)	48.87 (33.81)	0.583 (−14.19; 23.81)	0.695 (−25.61; 21.02)	0.007 (8.30; 32.43)*	0.009 (3.24; 32.59)*

*Before, pre-treatment; After, post-treatment; SD, standard deviation; P Value (Z), p value; BBS, berg balance scale (higher score = better performance); ABC scale, activities balance confidence scale (higher score = better performance); SOT, sensory organization balance test; EO, eyes-open condition; EC, eyes-closed condition; S, stable; C, compliant; SA, stabilometric assessment; Length CoP, length of CoP trajectory; FSS, fatigue severity scale (higher score = worse performance); SL, step length; SS, single support time; DS, double support time; MSQOL-54 PHC, multiple sclerosis quality of Life-54 physical health (higher score = better performance); MSQOL-54 MHC, multiple sclerosis quality of Life-54 mental health; *Statistically significant; for post-hoc analysis p value significant if <0.025 for Bonferroni correction for primary outcome measures.*

Table 3B | Descriptive and inferential statistics for clinical outcome measures.

Outcome variables	Before mean (SD)		After mean (SD)		1-month FU		Comparisons Wilcoxon signed ranks test		Comparisons Wilcoxon signed ranks test	
	RAGT	SIBT	RAGT	SIBT	RAGT	SIBT	RAGT		SIBT	
							T1 vs. T0 P value (95% CI)	T2 vs. T0 P value (95% CI)	T1 vs. T0 P value (95% CI)	T2 vs. T0 P value (95% CI)
SECONDARY OUTCOME MEASURES										
SA EO condition										
Sway area (mm^2)	83.48 (83.53)	128.54 (84.03)	64.55 (48.67)	141.44 (124.10)	120.40 (108.76)	165.21 (133.04)	0.182 (−50.33; 12.50)	0.594 (−26.66; 60.37)	0.799 (−55.69; 81.49)	0.683 (−54.53; 127.87)
Length CoP (mm)	499.66 (499.01)	504.60 (408.06)	398.16 (354.27)	422.70 (369.64)	369.08 (342.85)	413.77 (336.63)	0.055 (−227.26; 24.26)	0.182 (−441.53; 118.84)	0.241 (−234.92; 71.12)	0.169 (−245.09; 63.44)
SA EC condition										
Sway area (mm^2)	250.48 (261.99)	509.22 (342.79)	243.32 (230.54)	407.14 (311.95)	266.43 (216.58)	480.28 (330.37)	0.657 (−136.77; 122.44)	0.530 (−103.80; 135.69)	0.169 (−282.46; 78.30)	0.959 (−200.36; 142.48)
Length CoP (mm)	951.67 (1045.29)	1157.9 (951.54)	935.17 (1146.48)	1035.4 (993.74)	797.00 (736.14)	1154.78 (1053.15)	0.480 (−135.59; 102.59)	0.424 (−451.72; 142.38)	0.285 (−352.55; 107.55)	0.953 (−541.29; 304.09)
Gait analysis										
Cadence (step/min)	117.13 (28.22)	106.32 (37.49)	114.45 (24.75)	107.29 (42.01)	114.53 (33.78)	111.07 (40.04)	0.959 (−35.39; 11.40)	0.959 (−37.47; 13.61)	0.859 (−12.43; 14.17)	0.374 (−8.46; 17.02)
SL (cm)	56.88 (19.57)	58.62 (25.99)	62.39 (22.04)	65.95 (28.44)	59.49 (20.06)	60.99 (27.02)	0.084 (−0.32; 11.34)	0.028 (−0.00; 5.23)	0.005 (2.09; 12.56)*	0.508 (−3.80; 8.52)
SS time (% of cycle)	33.85 (4.23)	40.84 (16.49)	34.20 (4.39)	39.44 (16.84)	33.48 (4.93)	38.01 (10.17)	0.814 (−1.79; 2.48)	0.799 (−14.07; 2.15)	0.799 (−6.65; 3.85)	0.508 (−8.13; 2.46)
DS time (% of cycle)	31.30 (5.48)	39.54 (14.35)	29.79 (4.96)	31.40 (12.61)	28.49 (5.60)	28.03 (5.88)	0.084 (−3.42;.40)	0.074 (−16.38; 1.26)	0.005 (−15.25; −1.01)*	0.005 (−20.27; −2.73)*
FSS (1–7)	4.40 (1.38)	4.03 (2.25)	3.96 (1.17)	3.02 (1.50)	4.13 (1.81)	3.12 (1.84)	0.530 (−1.60;.73)	0.789 (−2.07;.84)	0.036 (−1.97; −0.05)	0.059 (−1.89;.07)
MSQOL−54 PHC (0–100)	64.17 (6.53)	59.59 (10.67)	60.84 (9.01)	61.34 (8.16)	60.79 (5.85)	57.30 (9.60)	0.507 (−8.04; 2.51)	0.169 (−7.20; 1.57)	0.214 (−3.60; 6.74)	0.214 (−5.75; 1.63)
MSQOL−54 MHC (0–100)	59.01 (21.69)	59.51 (20.7)	61.11 (19.58)	65.24 (15.34)	63.82 (15.01)	62.10 (18.38)	0.574 (−3.78; 7.28)	0.093 (−1.79; 9.79)	0.314 (−4.55; 14.87)	0.953 (−7.21; 11.88)

*Before, pre-treatment; After, post-treatment; SD, standard deviation; P Value (Z), p value; BBS, berg balance scale (higher score = better performance); ABC scale, activities balance confidence scale (higher score = better performance); SOT, sensory organization balance test; EO, eyes-open condition; EC, eyes-closed condition; S, stable; C, compliant; SA, stabilometric assessment; Length CoP, length of CoP trajectory; FSS, fatigue severity scale (higher score = worse performance); SL, step length; SS, single support time; DS, double support time; MSQOL-54 PHC, multiple sclerosis quality of Life-54 physical health (higher score = better performance); MSQOL-54 MHC, multiple sclerosis quality of Life-54 mental health; *Statistically significant; post-hoc analysis p value significant if <0.025 for Bonferroni correction for primary outcome measures.*

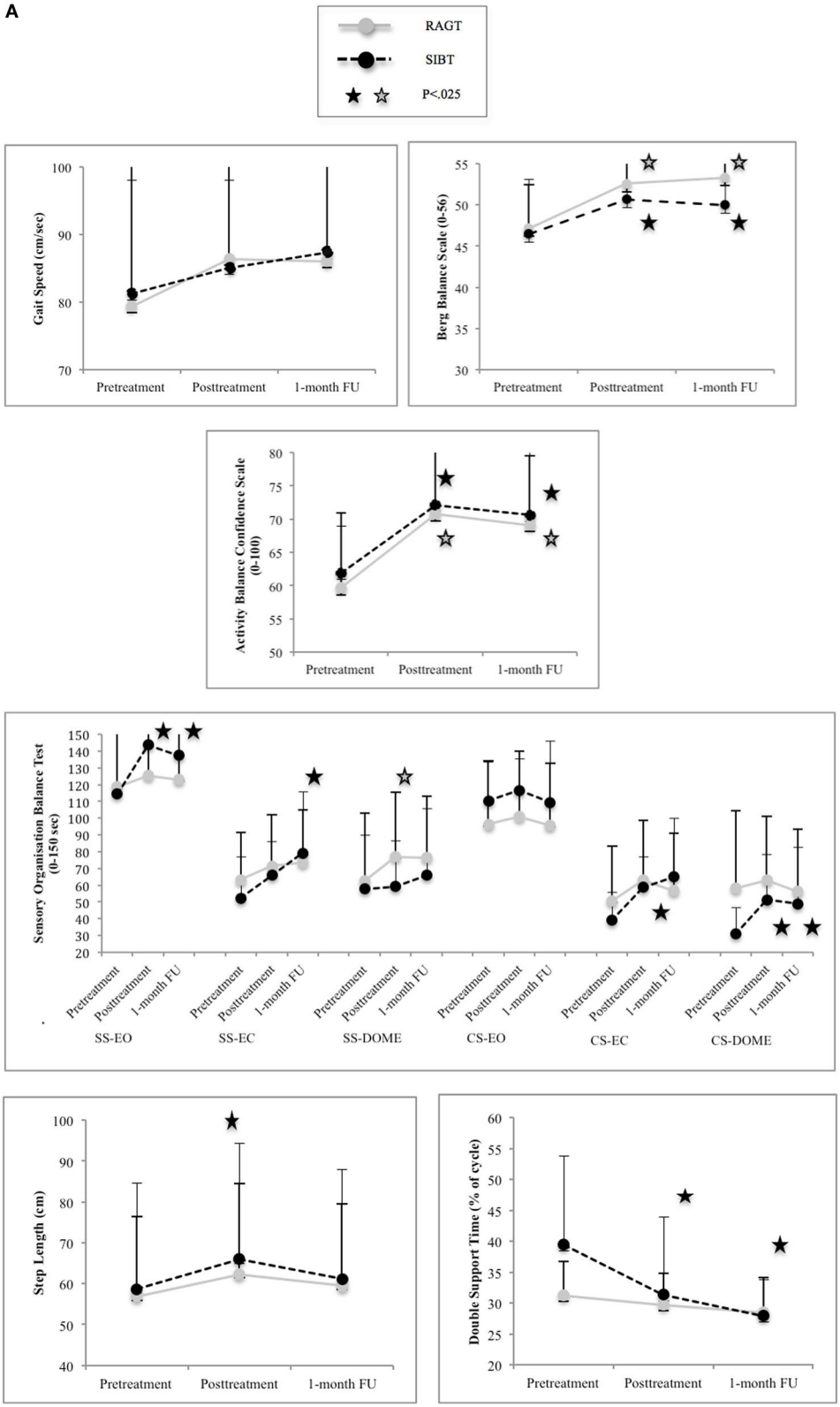


FIGURE 2 | Continued

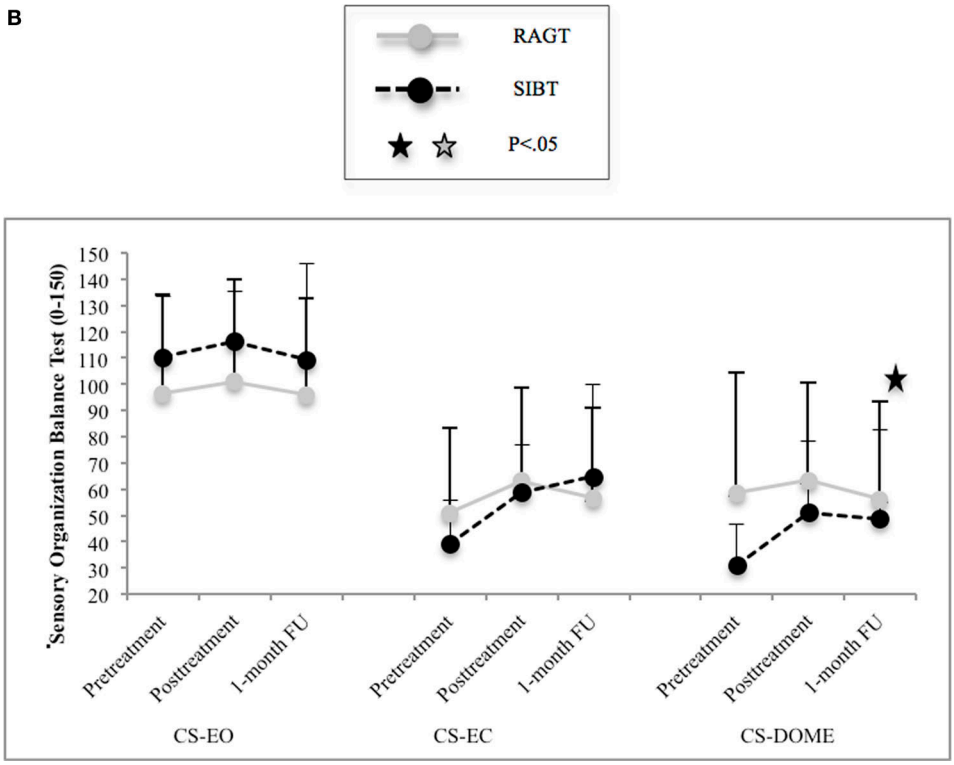


FIGURE 2 | (A) Within group analysis: mean performance and standard errors at primary and statistical significant secondary outcome measures. Abbreviations: SS, stable surface; CS, complaint surface; EO, eyes-open condition; Dome, Dome condition; FU, follow-up. **(B)** Between group comparison: mean performance and standard errors at secondary organization balance test (SOT) (only statistical significant value). Abbreviations: CS, complaint surface; EO, eyes-open condition; EC, eyes-closed condition; Dome, Dome condition; FU, follow-up.

necessities the central nervous system employs compensatory postural adjustments or so-called feedback mechanisms (CPAs), and/or feed-forward or anticipatory postural adjustments (APAs) (Massion, 1992). Both mechanisms appear to be affected in MS patients while standing (Cameron et al., 2008) and walking (Huisinga et al., 2014).

Currently, interventions that specifically address proprioceptive and/or central processing deficits are likely to be particularly effective in MS patients. Proprioceptive information, in fact, plays a crucial role with respect to the knowledge on external environment (i.e., body position knowledge, sensorimotor control of functional joint stability and feedback postural adjustments) and in motor control during internally generated motor commands (internal model). The concept behind the study is that the task-specific balance training should improve gait performance and vice versa because postural control is essential for walking.

Our findings cannot be fully discussed with those by Straudi (Straudi et al., 2013) and Schwartz et al. (2012) owing to differences about patients EDSS score range, sample size, duration of treatment and treatment types used as both conventional and experimental treatment. In our study, balance outcome measures included also a subjective measure of confidence in performing various ambulatory activities (ABC) as well as to objective measure designed to assess static balance and fall risk (BBS). Data showed that RAGT and SIBT have a significant both post-treatment and long-term effect (1 months). Additionally, our findings suggest that these improvement may generalize in patients while performing daily activities.

A possible explanation of the balance improvements is that GT1 approach act as a form of "destabilization training." For the first time in literature we might introduce the concept of "task specific balance training" by end-effector RAGT.

This training might play a role for reinforcing the neuronal circuits that contribute to postural control. In particular, RAGT represents an external force that could interfere with the abnormal experience of balance and gait. An end-effector system may represent a more suitable device for this purpose. It enables wheelchair-bound subjects to practice a gait-like movement with minimal assistance. The harness-secured patients are positioned on 2 footplates, whose movements simulate stance and swing in a highly physiological manner. In this context the patient has a reduced number of constraints acting at different lower limb levels. A reduced number of constraints and more freedom during exercise, especially for pelvic movements, may be an optimal environment for learning. Similar results with an end-effector system were found in patients affected by Parkinson's disease (Picelli et al., 2012, 2013). Lastly, we cannot exclude that muscle strength improvements could contribute to this effect.

This is a significant result, given that MS patients suffer from balance disorders very early during the disease even when gait

disorders are minimal. From a clinical perspective having another rehabilitation strategy for these high disabling disorders is very relevant. Nevertheless future studies on larger sample and involving patients stratified by EDSS would allow us to better understand which approach (robot assisted balance training or SIBT) and for which patients would be more useful to improve balance task related domains and/or gait related domains. MS patients with different degrees of disability require different needs in terms of treatment's type, intensity, and frequency.

As to SIBT effects, significant improvements on BBS and ABC paralleled significant effects in sensory-motor integration ability and dynamic balance performance. Indeed, patients in the SIBT group showed improvements during SOT conditions (stable surface-opened eyes and closed eyes) and in most difficult performance (compliant surface-closed eyes and dome conditions). In healthy subjects balance control is a complex process involving the reception and the integration of visual, sensorimotor, and vestibular sensory inputs, which allows the planning and execution of the movements required to maintain balance during upright posture and gait (Merfeld et al., 1999). The ability of the central nervous system to process these different types of sensory information leads to the establishment of a system of coordinates on which the body's postural control is based (Merfeld et al., 1999). For instance, in the static standing position healthy adults use sensorimotor information, which originates from pressure receptors, joint receptors, and muscle proprioceptors, to build the main reference coordinates for balance (Maurer et al., 2006). When sensorimotor information is inadequate, visual and vestibular systems become involved to maintain balance. This central integration of sensory inputs allows potential sensory conflicts generated by inadequate afferent information to be overcome. The capability to analyse, compare, and select the pertinent sensory information is very important in order to prevent falling (Cattaneo and Jonsdottir, 2009). It has been showed that imbalance in MS may not only be related to a primary sensory deficit but to a disturbed integration of the available sensory information (Smedal et al., 2006). Moreover, studies of postural responses indicate that imbalance in people with MS is unlike imbalance from cerebellar disorders (Cameron and Lord, 2010).

Several studies evaluated the efficacy of rehabilitation for improving balance in people with MS (Armutlu et al., 2001; Smedal et al., 2006; Nilsagård et al., 2013). Evidence supports that the interventions most likely to be effective are those related to sensory facilitation and central integration deficits (Cattaneo et al., 2007). This type of intervention has been proposed in few studies in order to ameliorate specifically the postural control in elderly (Hu and Woollacott, 1994), in stroke patients (Smania et al., 2008; Bayouk et al., 2006) and in Parkinson disease patients (Smania et al., 2010). So far, only two RCT studies have addressed this issue suggesting promising effects in MS patients (Cattaneo et al., 2007; Elwishy, 2012). Abeer et al. enrolled fifty patients (EDSS $\leq$ 4.5) randomized to receive balance rehabilitation just for motor strategies or sensorimotor balance rehabilitation program that aimed at improving motor and sensory strategies. Each treatment lasted 50 min/session, 3 times/week for 8 weeks. Before and after treatment balance was assessed clinically for standing balance by BBS and instrumentally for somatosensory and neuromuscular control aspects by Biodex Stability system. Data showed significant differences in all outcome measures between the two groups in favor of the sensorimotor training (Elwishy, 2012). Cattaneo et al. (Cattaneo et al., 2007) pointed out that such sensorimotor balance training might improve also dynamic balance aspects assessed by the Dynamic Gait Index. Although differences between our work and the study by Cattaneo et al. and by Elwishy (Elwishy, 2012) were the wide variation of EDSS score, sample size, the duration of treatment and outcome measures used, our findings further support that specific balance training may induce positive effects in improving central integration of the sensory input. That is, patients underwent SIBT may have improved the ability to integrate somatosensory and vestibular inputs, becoming less reliant on visual input, as well as use the most appropriate sensory strategies to control their posture and prevent falls (Shumway-Cook and Horak, 1986; Cattaneo et al., 2007). The execution of exercises with the use of surfaces and vision manipulation aimed at challenging postural control could improve the somatosensory integration processes during dynamic tasks. This type of intervention has been proposed in few studies in order to ameliorate specifically the postural control in elderly (Hu and Woollacott, 1994), in stroke patients (Bayouk et al., 2006; Smania et al., 2008), in Parkinson disease patients (Smania et al., 2010) and in MS patients (Cattaneo et al., 2007; Elwishy, 2012).

An important finding that required discussion was the no statistically significant differences between the end-effector RAGT and the SIBT in primary outcome measures. Moreover, for improvements in secondary outcome measures, between-group differences were in favor of the SIBT for one SOT condition (compliant surface-dome). According to our hypothesis, these results support the assumption that this form of RAGT, which practices a gait-like movement with minimal assistance, allows patients to train postural and gait control. Many advantages that further support the use of end-effector RAGT may be acknowledged: the patient may be trained safely owing to body harness, the complexity of the tasks might be improved over time by changing the amount of body weight support and, finally, it does not necessarily require a one-by-one physical therapist assistance. Further, recent work has demonstrated that end-effector RAGT enables patients repetitive practice of stair climbing, which is considered a more demanding balance task than gait (Hesse et al., 2010). Stair climbing requires a high level of postural control and walking ability. It is important to consider that end-effector RAGT may allow us to develop specific programs combining gait training with sensory integration exercises in order to further improve walking. For instance by using an end-effector system (Hesse et al., 2010) with body weight support might be an optimal strategy for implementing a true sensorimotor training for people affected by MS. There is growing interest in developing new technological approaches for these disturbances.

The point to use end-effector RAGT and SIBT in people with MS is that overall evidence supports that these patients have CPAs and APAs as well as sensory integration deficits leading to "internal representation" of motor and sensory signals impairments.

People with MS have a strong delay in CPAs onset in terms of magnitude and velocity (Cameron et al., 2008; Huisinga et al.,

2014). This has been attributed mainly to a reduced velocity of signals propagation in somatosensory and motor pathway. Recent findings point toward the somatosensory afferent inputs as primary causative role of impaired CPAs postural adjustment (Huisinga et al., 2014). To compensate the prolonged postural responses, and then to prevent falls, people with MS often implement a predictive strategy (APAs). Recent findings showed that APAs are impaired in people with MS in terms of delayed onset of muscles activation, reduced magnitude and less directional specific activation of muscles (Krishnan et al., 2012a,b). These alterations lead to an impaired dynamic shifting of CoP during gait initiation (Remelius et al., 2008) and to a reduction of the excursion of the center of mass in the sagittal and transversal plane (Remelius et al., 2008).

Another important mechanism involved in gait encompasses afferent input. Widespread research revealed afferent inputs are involved in motor output shaping during walking (Dietz et al., 2002; Nielsen and Sinkjaer, 2002; Pearson, 2004). It has been demonstrated that human action execution (i.e., to make a cup of coffee) required three main phases: motor planning, motor execution, and movement control. Feed-forward and feedback mechanisms are thought to be involved mainly in the last phase (movement control) in order to perform efficient goal-directed movements (Frey et al., 2011). These mechanisms are involved in forming the so-called "internal model," which is an "internal representation" of motor and sensory signals related to a specific motor execution. It has been demonstrated that an "internal model" exists also for lower limb (Emken and Reinkensmeyer, 2005; Lam et al., 2006). It allows walking to face postural perturbation and/or novel dynamic environments. Lesions at the central nervous system due to MS can be viewed as generating a novel dynamic environment that must be learned in order to walk effectively. Experiments ranging from animals (Lou and Bloedel, 1988; Hodgson et al., 1994) to humans (Lam et al., 2003; Pang et al., 2003; Emken and Reinkensmeyer, 2005; Lam et al., 2006) showed lasting modifications in response to continuous disturbance of walking followed by aftereffects after removal the disturbance. These aftereffects that follow a period of training under specific walking condition may suggest the possible formation or adaptation of the motor output ("internal model" shaping).

Limitations of the present study are the small sample size, the clinical heterogeneity of patients according to EDSS score, the lack of patients' stratification by neurological severity, the lack of a follow-up assessment at 3 or more months after training and the lack of assessment of CPAs and APAs with electromyography. Future studies should determine frequency, duration, and other important aspects of RAGT parameters, such as speed, and need for body weight support. Finally, postural destabilizations and sensory strategies might add a substantial value to ongoing therapy.

TIPS FOR FUTURE RESEARCH

Treatment of gait and balance dysfunction in people with MS has developed significantly in recent years. Studies demonstrated the potential effect of various interventions for improving walking and balance disorders, with benefits reported also by the patients. However, there are cloudy hypotheses that are driving this research area. To speed up the progress in this field of research, several crucial points should taken into account when planning future studies (Zackowski et al., 2013):

(1) To perform randomized controlled trial on larger sample in order to evaluate SIBT and end-effector RAGT effectiveness in MS patients. In our opinion, the RAGT devised for this purpose should be an end-effector one in order to improve gait and balance too.
(2) To evaluate the effects of treatments combining SIBT and end-effector RAGT.
(3) To develop new technological software that may include for instance exercises with sensory augmentations for the impaired proprioception and/or with the use of surfaces and vision manipulation aimed at improving somatosensory integration processes (Elwishy, 2012), sub sensory mechanical noise or vibration applied to the sole of the foot, haptic learning (without vision) and functional electrical stimulation cycling to coordinate further leg muscles during walking.
(4) To couple visual information and feedback in order to improve the awareness of disturbed walking and to engage actively the patients' participation and motivation during training.
(5) To amplify the patient's movement errors (Emken and Reinkensmeyer, 2005) aimed at inducing postural adaptations and favorably adjust leg movement trajectories. Unexpected perturbation during swing phase of walking might be useful (Emken and Reinkensmeyer, 2005).

CONCLUSIONS

This is the first study comparing the effects on walking and balances between the end-effector RAGT and SIBT. These preliminary results suggest that the end-effector RAGT training may act as task-specific balance training in order to promote central neural processes involved in gait and balance control.

ACKNOWLEDGMENTS

This work was supported by funding from the Italian Multiple Sclerosis Foundation grant (FISM 2009/R/27).

AUTHOR CONTRIBUTIONS

All authors gave a substantial contribution to the design of the work and acquisition, analysis, interpretation of data; Marialuisa Gandolfi conceived of the study, carried out the studies, data acquisition, analysis and interpretation, drafted the manuscript. Christian Geroin and Alessandro Picelli carried out the studies, data acquisition, analysis and interpretation, drafted the manuscript and performed the statistical analysis. Daniele Munari carried out the studies, data acquisition and interpretation, drafted the manuscript. Nicola Smania, Andreas Waldner, Stefano Tamburin, and Fabio Marchioretto conceived of the study, participated in its design and coordination, and assisted in drafting the manuscript. All authors read and approved the final manuscript. All authors declare 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 are appropriately investigated and resolved.

REFERENCES

Armutlu, K., Karabudak, R., and Nurlu, G. (2001). Physiotherapy approaches in the treatment of ataxic multiple sclerosis: a pilot study. *Neurorehabil. Neural Repair* 15, 203–211. doi: 10.1177/154596830101500308

Bayouk, J. F., Boucher, J. P., and Leroux, A. (2006). Balance training following stroke: effects of task-oriented exercises with and without altered sensory input. *Int. J. Rehabil. Res.* 29, 51–59. doi: 10.1097/01.mrr.0000192100.67425.84

Beer, S., Aschbacher, B., Manoglou, D., Gamper, E., Kool, J., and Kesselring, J. (2008). Robot-assisted gait training in multiple sclerosis: a pilot randomized trial. *Mult. Scler.* 14, 231–236. doi: 10.1177/1352458507082358

Benedetti, M. G., Gasparroni, V., Stecchi, S., Zilioli, R., Straudi, S., and Piperno, R. (2009). Treadmill exercise in early multiple sclerosis: a case series study. *Eur. J. Phys. Rehabil. Med.* 45, 53–59.

Broekmans, T., Roelants, M., Alders, G., Feys, P., Thijs, H., and Eijnde, B. O. (2010). Exploring the effects of a 20-week whole-body vibration training programme on leg muscle performance and function in persons with multiple sclerosis. *J. Rehabil. Med.* 42, 866–872. doi: 10.2340/16501977-0609

Cakt, B. D., Nacir, B., Genç, H., Saraçoğlu, M., Karagöz, A., Erdem, H. R., et al. (2010). Cycling progressive resistance training for people with multiple sclerosis: a randomized controlled study. *Am. J. Phys. Med. Rehabil.* 89, 446–457. doi: 10.1097/PHM.0b013e3181d3e71f

Cameron, M. H., Horak, F. B., Herndon, R. R., and Bourdette, D. (2008). Imbalance in multiple sclerosis: a result of slowed spinal somatosensory conduction. *Somatosens. Mot. Res.* 25, 113–122. doi: 10.1080/08990220802131127

Cameron, M. H., and Lord, S. (2010). Postural control in multiple sclerosis: implications for fall prevention. *Curr. Neurol. Neurosci. Rep.* 10, 407–412. doi: 10.1007/s11910-010-0128-0

Cattaneo, D., and Jonsdottir, J. (2009). Sensory impairments in quiet standing in subjects with multiple sclerosis. *Mult. Scler.* 15, 59–67. doi: 10.1177/1352458508096874

Cattaneo, D., Jonsdottir, J., Zocchi, M., and Regola, A. (2007). Effects of balance exercises on people with multiple sclerosis: a pilot study. *Clin. Rehabil.* 21, 771–781. doi: 10.1177/0269215507077602

Cattaneo, D., Regola, A., and Meotti, M. (2006). Validity of six balance disorders scales in persons with multiple sclerosis. *Disabil. Rehabil.* 28, 789–795. doi: 10.1080/09638280500404289

Cohen, J. (1988). *Statistical Power Analysis for the Behavioral Sciences*. 2nd ed. Hillsdale, NJ: Lawrence Erlbaum.

Confavreux, C., Vukusic, S., Moreau, T., and Adeleine, P. (2000). Relapses and progression of disability in multiple sclerosis. *N. Engl. J. Med.* 343, 1430–1438. doi: 10.1056/NEJM200011163432001

Corradini, M. L., Fioretti, S., Leo, T., and Piperno, R. (1997). Early recognition of postural disorders in multiple sclerosis through movement analysis: a modeling study. *IEEE Trans. Biomed. Eng.* 44, 1029–1038. doi: 10.1109/10.641330

Dallal, G. (2007). *Random Generator*. Available online at: www.randomization.com

Dietz, V., Müller, R., and Colombo, G. (2002). Locomotor activity in spinal man: significance of afferent input from joint and load receptors. *Brain* 125, 2626–2634. doi: 10.1093/brain/awf273

Elwishy, A. B. A. (2012). Effect of sensorimotor integration balance program in patients with Multiple Sclerosis: a single blinded randomized controlled study. *Med. J. Cairo Univ.* 80, 85–93.

Emken, J. L., and Reinkensmeyer, D. J. (2005). Robot-enhanced motor learning: accelerating internal model formation during locomotion by transient dynamic amplification. *IEEE Trans. Neural Syst. Rehabil. Eng.* 13, 33–39. doi: 10.1109/TNSRE.2004.843173

Folstein, M. F., Folstein, S. E., and McHugh, P. R. (1975). "Mini-mental state." A practical method for grading the cognitive state of patients for the clinician. *J. Psychiatr. Res.* 12, 189–198. doi: 10.1016/0022-3956(75)90026-6

Frey, S. H., Fogassi, L., Grafton, S., Picard, N., Rothwell, J. C., Schweighofer, N., et al. (2011). Neurological principles and rehabilitation of action disorders: computation, anatomy, and physiology (CAP) model. *Neurorehabil. Neural Repair* 25, 6S–20S. doi: 10.1177/1545968311410940

Givon, U., Zeilig, G., and Achiron, A. (2009). Gait analysis in multiple sclerosis: characterization of temporal-spatial parameters using GAITRite functional ambulation system. *Gait Posture* 29, 138–142. doi: 10.1016/j.gaitpost.2008.07.011

Hebert, J. R., Corboy, J. R., Manago, M. M., and Schenkman, M. (2011). Effects of vestibular rehabilitation on multiple sclerosis-related fatigue and upright postural control: a randomized controlled trial. *Phys. Ther.* 91, 1166–1183. doi: 10.2522/ptj.20100399

Hesse, S. (2008). Treadmill training with partial body weight support after stroke: a review. *NeuroRehabilitation* 23, 55–65.

Hesse, S., Sarkodie-Gyan, T., and Uhlenbrock, D. (1999). Development of an advanced mechanised gait trainer, controlling movement of the centre of mass, for restoring gait in non-ambulant subjects. *Biomed. Tech. (Berl).* 44, 194–201. doi: 10.1515/bmte.1999.44.7-8.194

Hesse, S., Waldner, A., and Tomelleri, C. (2010). Innovative gait robot for the repetitive practice of floor walking and stair climbing up and down in stroke patients. *J. Neuroeng. Rehabil.* 7, 30. doi: 10.1186/1743-0003-7-30

Hodgson, J. A., Roy, R. R., de Leon, R., Dobkin, B., and Edgerton, V. R. (1994). Can the mammalian lumbar spinal cord learn a motor task? *Med. Sci. Sports Exerc.* 26, 1491–1497. doi: 10.1249/00005768-199412000-00013

Hu, M. H., and Woollacott, M. H. (1994). Multisensory training of standing balance in older adults: I. Postural stability and one-leg stance balance. *J. Gerontol.* 49, 52–61. doi: 10.1093/geronj/49.2.M52

Huisinga, J. M., St. George, R. J., Spain, R., Overs, S., and Horak, F. B. (2014). Postural response latencies are related to balance control during standing and walking in patients with multiple sclerosis. *Arch. Phys. Med. Rehabil.* 17. doi: 10.1016/j.apmr.2014.01.004

Krishnan, V., Kanekar, N., and Aruin, A. S. (2012a). Feedforward postural control in individuals with multiple sclerosis during load release. *Gait Posture* 36, 225–230. doi: 10.1016/j.gaitpost.2012.02.022

Krishnan, V., Kanekar, N., and Aruin, A. S. (2012b). Anticipatory postural adjustments in individuals with multiple sclerosis. *Neurosci. Lett.* 506, 256–260. doi 10.1016/j.neulet.2011.11.018

Krupp, L. B., LaRocca, N. G., Muir-Nash, J., and Steinberg, A. D. (1989). The fatigue severity scale. Application to patients with multiple sclerosis and systemic lupus erythematosus. *Arch. Neurol.* 46, 1121–1123. doi: 10.1001/archneur.1989.00520460115022

Kurtzke, J. F. (1983). Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). *Neurology* 33, 1444–1452. doi: 10.1212/WNL.33.11.1444

Lam, T., Anderschitz, M., and Dietz, V. (2006). Contribution of feedback and feedforward strategies to locomotor adaptations. *J. Neurophysiol.* 95, 766–773. doi: 10.1152/jn.00473.2005

Lam, T., Wolstenholme, C., and Yang, J. F. (2003). How do infants adapt to loading of the limb during the swing phase of stepping? *J. Neurophysiol.* 89, 1920–1928. doi: 10.1152/jn.01030.2002

Larocca, N. G. (2011). Impact of walking impairment in multiple sclerosis: perspectives of patients and care partners. *Patient* 4, 189–201. doi: 10.2165/11591150-000000000-00000

Lo, A. C., and Triche, E. W. (2008). Improving gait in multiple sclerosis using robot-assisted, body weight supported treadmill training. *Neurorehabil. Neural Repair* 22, 661–671. doi: 10.1177/1545968308318473

Lou, J. S., and Bloedel, J. R. (1988). A new conditioning paradigm: conditioned limb movements in locomoting decerebrate ferrets. *Neurosci. Lett.* 84, 185–190. doi: 10.1016/0304-3940(88)90405-3

Massion, J. (1992). Movement, posture and equilibrium: interaction and coordination. *Prog. Neurobiol.* 38, 35–56. doi: 10.1016/0301-0082(92)90034-C

Maurer, C., Mergner, T., and Peterka, R. J. (2006). Multisensory control of human upright stance. *Exp. Brain Res.* 171, 231–250. doi: 10.1007/s00221-005-0256-y

Menz, H. B., Latt, M. D., Tiedemann, A., Mun San Kwan, M., and Lord, S. R. (2004). Reliability of the GAITRite walkway system for the quantification of temporo-spatial parameters of gait in young and older people. *Gait Posture* 20, 20–25. doi: 10.1016/S0966-6362(03)00068-7

Merfeld, D. M., Zupan, L., and Peterka, R. J. (1999). Human use internal models to estimate gravity and linear acceleration. *Nature* 98, 615–618. doi: 10.1038/19303

Nelson, S. R., Di Fabio, R. P., and Anderson, J. H. (1995). Vestibular and sensory interaction deficits assessed by dynamic platform posturography in patients with multiple sclerosis. *Ann. Otol. Rhinol. Laryngol.* 104, 62–68.

Newman, M. A., Dawes, H., van den Berg, M., Wade, D. T., Burridge, J., and Izadi, H. (2007). Can aerobic treadmill training reduce the effort of walking and fatigue in people with multiple sclerosis: a pilot study. *Mult. Scler.* 13, 113–119. doi: 10.1177/1352458506071169

Nichols, D. S. (1997). Balance retraining after stroke using force platform biofeedback. *Phys. Ther.* 77, 553–558.

Nielsen, J. B., and Sinkjaer, T. (2002). Afferent feedback in the control of human gait. *J. Electromyogr. Kinesiol.* 12, 213–217. doi: 10.1016/S1050-6411(02)00023-8

Nilsagård, Y. E., Forsberg, A. S., and von Koch, L. (2013). Balance exercise for persons with multiple sclerosis using Wii games: a randomised, controlled multi-centre study. *Mult. Scler.* 19, 209–216. doi: 10.1177/1352458512450088

Pang, M. Y., Lam, T., and Yang, J. F. (2003). Infants adapt their stepping to repeated trip-inducing stimuli. *J. Neurophysiol.* 90, 2731–2740. doi: 10.1152/jn.00407.2003

Pearson, K. G. (2004). Generating the walking gait: role of sensory feedback. *Prog. Brain Res.* 143, 123–129.

Picelli, A., Melotti, C., Origano, F., Neri, R., Waldner, A., and Smania, N. (2013). Robot-assisted gait training versus equal intensity treadmill training in patients with mild to moderate Parkinson's disease: a randomized controlled trial. *Parkinsonism Relat. Disord.* 19, 605–610. doi: 10.1016/j.parkreldis.2013.02.010

Picelli, A., Melotti, C., Origano, F., Waldner, A., Gimigliano, R., and Smania, N. (2012). Does robotic gait training improve balance in Parkinson's disease? A randomized controlled trial. *Parkinsonism Relat. Disord.* 18, 990–993. doi: 10.1016/j.parkreldis.2012.05.010

Polman, C. H., Reingold, S. C., Banwell, B., Clanet, M., Cohen, J. A., Filippi, M., et al. (2011). Diagnostic criteria for multiple sclerosis: 2010 revisions to the McDonald criteria. *Ann. Neurol.* 69, 292–302. doi: 10.1002/ana.22366

Powell, L. E., and Myers, A. M. (1995). The activities-specific balance confidence (ABC) scale. *J. Gerontol. A Biol. Sci. Med. Sci.* 50, M28–M34. doi: 10.1093/gerona/50A.1.M28

Remelius, J. G., Hamill, J., Kent-Braun, J., and Van Emmerik, R. E. (2008). Gait initiation in multiple sclerosis. *Motor Control* 12, 93–108.

Riemann, B. L., and Lephart, S. M. (2002). The sensorimotor system, Part II: the role of proprioception in motor control and functional joint stability. *J. Athl. Train.* 37, 80–84.

Rossignol, S., Dubuc, R., and Gossard, J. P. (2006). Dynamic sensorimotor interactions in locomotion. *Physiol. Rev.* 86, 89–154. doi: 10.1152/physrev.00028.2005

Schuhfried, O., Mittermaier, C., Jovanovic, T., Pieber, K., and Paternostro-Sluga, T. (2005). Effects of whole-body vibration in patients with multiple sclerosis: a pilot study. *Clin. Rehabil.* 19, 834–842. doi: 10.1191/0269215505cr919oa

Schwartz, I., Sajin, A., Moreh, E., Fisher, I., Neeb, M., Forest, A., et al. (2012). Robot-assisted gait training in multiple sclerosis patients: a randomized trial. *Mult. Scler.* 18, 881–890. doi: 10.1177/1352458511431075

Shumway-Cook, A., and Horak, F. B. (1986). Assessing the influence of sensory interaction of balance. Suggestion from the field. *Phys. Ther.* 66, 1548–1550.

Smania, N., Corato, E., Tinazzi, M., Stanzani, C., Fiaschi, A., Girardi, P., et al. (2010). Effect of balance training on postural instability in patients with idiopathic Parkinson's disease. *Neurorehabil. Neural Repair* 24, 826–834. doi: 10.1177/1545968310376057

Smania, N., Picelli, A., Gandolfi, M., Fiaschi, A., and Tinazzi, M. (2008). Rehabilitation of sensorimotor integration deficits in balance impairment of patients with stroke hemiparesis: a before/after pilot study. *Neurol. Sci.* 29, 313–319. doi: 10.1007/s10072-008-0988-0

Smedal, T., Lygren, H., Myhr, K. M., Moe-Nilssen, R., Gjelsvik, B., Gjelsvik, O., et al. (2006). Balance and gait improved in patients with MS after physiotherapy based on the Bobath concept. *Physiother. Res. Int.* 11, 104–116. doi: 10.1002/pri.327

Solari, A., Filippini, G., Mendozzi, L., Ghezzi, A., Cifani, S., Barbieri, E., et al. (1999). Validation of Italian multiple sclerosis quality of life 54 questionnaire. *J. Neurol. Neurosurg. Psychiatr.* 67, 158–162. doi: 10.1136/jnnp.67.2.158

Speers, R. A., Kuo, A. D., and Horak, F. B. (2002). Contributions of altered sensation and feedback responses to changes in coordination of postural control due to aging. *Gait Posture* 16, 20–30. doi: 10.1016/S0966-6362(02)00003-6

Straudi, S., Benedetti, M. G., Venturini, E., Manca, M., Foti, C., and Basaglia, N. (2013). Does robot-assisted gait training ameliorate gait abnormalities in multiple sclerosis? A pilot randomized-control trial. *NeuroRehabilitation* 33, 555–563. doi: 10.3233/NRE-130990

Swinnen, E., Beckwée, D., Pinte, D., Meeusen, R., Baeyens, J. P., and Kerckhofs, E. (2012). Treadmill training in multiple sclerosis: can body weight support or robot assistance provide added value? A systematic review. *Mult. Scler. Int.* 2012, 240274. doi: 10.1155/2012/240274

Tamburella, F., Scivoletto, G., and Molinari, M. (2013). Balance training improves static stability and gait in chronic incomplete spinal cord injury subjects: a pilot study. *Eur. J. Phys. Rehabil. Med.* 49, 353–364.

Vaney, C., Gattlen, B., Lugon-Moulin, V., Meichtry, A., Hausammann, R., Foinant, D., et al. (2012). Robotic-assisted step training (lokomat) not superior to equal intensity of over-ground rehabilitation in patients with multiple sclerosis. *Neurorehabil. Neural Repair*. 26, 212–221. doi: 10.1177/1545968311425923

Weinshenker, B. G., Bass, B., Rice, G. P., Noseworthy, J., Carriere, W., Baskerville, J., et al. (1989). The natural history of multiple sclerosis: a geographically based study. I. Clinical course and disability. *Brain* 112, 133–146. doi: 10.1093/brain/112.1.133

Zackowski, K. M., Cameron, M., and Wagner, J. M. (2013). 2nd International Symposium on Gait and Balance in Multiple Sclerosis: interventions for gait and balance in MS. *Disabil. Rehabil*. doi: 10.3109/09638288.2013.833306. [Epud ahead of print].

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.

Received: 17 March 2014; accepted: 29 April 2014; published online: 22 May 2014.
Citation: Gandolfi M, Geroin C, Picelli A, Munari D, Waldner A, Tamburin S, Marchioretto F and Smania N (2014) Robot-assisted vs. sensory integration training in treating gait and balance dysfunctions in patients with multiple sclerosis: a randomized controlled trial. Front. Hum. Neurosci. ***8****:318. doi: 10.3389/fnhum.2014.00318*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

REVIEW ARTICLE
published: 25 June 2014
doi: 10.3389/fnhum.2014.00475

Cerebellar contribution to feedforward control of locomotion

Iolanda Pisotta and Marco Molinari*

Neurological Rehabilitation Department A and CaRMA Lab, I.R.C.C.S. Fondazione Santa Lucia Rome, Rome, Italy

Edited by:
Nadia Dominici, VU University Amsterdam, Netherlands

Reviewed by:
Masao Ito, RIKEN, Japan
Richard E. Poppele, University of Minnesota, USA

****Correspondence:***
Marco Molinari, Neurological Rehabilitation Department A and CaRMA Lab, I.R.C.C.S. Fondazione Santa Lucia Rome, Via Ardeatina 306, 00179 Rome, Italy
e-mail: m.molinari@hsantalucia.it

The cerebellum is an important contributor to feedforward control mechanisms of the central nervous system, and sequencing—the process that allows spatial and temporal relationships between events to be recognized—has been implicated as the fundamental cerebellar mode of operation. By adopting such a mode and because cerebellar activity patterns are sensitive to a variety of sensorimotor-related tasks, the cerebellum is believed to support motor and cognitive functions that are encoded in the frontal and parietal lobes of the cerebral cortex. In this model, the cerebellum is hypothesized to make predictions about the consequences of a motor or cognitive command that originates from the cortex to prepare the entire system to cope with ongoing changes. In this framework, cerebellar predictive mechanisms for locomotion are addressed, focusing on sensorial and motoric sequencing. The hypothesis that sequence recognition is the mechanism by which the cerebellum functions in gait control is presented and discussed.

Keywords: locomotion, corticocerebellar loops, feedforward control, movement prediction, sequencing hypothesis

INTRODUCTION

Based on animals and humans studies, much has been learned about how the cerebellum coordinates normal movement and how it contributes to motor adaptation and motor learning.

Cerebellar damage does not cause a loss of movement; instead, it effects clear and consistent abnormalities in movement, including lack of coordination, increased variability, tremor, and poor accuracy. Notably, cerebellar damage induces greater impairments to movements that require predictive control versus those that require reactive control. As demonstrated by Morton and Bastian (2006) using an elegant task that was based on splitbelt treadmill walking, cerebellar damage impairs the ability to adapt to predictable but not sudden unpredictable changes. Recently developed functional theories on predictive control of the cerebellum explain the effects of cerebellar damage on eye and limb movements and on walking (Koziol et al., 2014).

In motor control theories, the term "predictive" refers to the feedforward component of a movement that is planned in advance and is unchanged by online peripheral feedback. Predictive control is typically assessed at the earliest stage of the movement, during which corrections that are based on peripheral feedback are not possible. This type of control is used to make any online corrections that might be necessary as a movement unfolds, and it requires that the conditions of later movement stages be known in advance. Although the hypothesis that the cerebellum is important for predictive control is not new, claims of its importance in locomotive control are relatively recent (Morton and Bastian, 2006). Further, current data on the significance of cerebellar predictive control in areas outside of the motor domain and for supporting learning and functional recovery have piqued the interest of neuroscientists in the hopes of better understanding cerebellar control mechanisms.

In this framework, we will discuss cerebellar predictive mechanisms for locomotion, focusing on the type of information that is processed—sensorial or motoric—and on sequence recognition as the mechanisms for understanding cerebellar function in making predictions.

CEREBELLUM AND FEEDFORWARD CONTROL

The cerebellum has an exquisitely simple cellular organization, which has been well known since the beginning of the last century, based on the work of Ramon y Cajal (see Sotelo, 2008). Since then, scientists have been intrigued by its function. Nevertheless, after more than a century of dedicated studies, there is no consensus on how the cerebellum operates. Among the various theories that exist, the hypothesis that the cerebellum mediates predictive motor control in locomotion is gaining momentum (Bastian, 2006).

The forward model of motor control postulates that to make a motor-to-somatosensory prediction, the cerebellum receives an efference copy of a motor command from the primary motor cortex. This information allows the cerebellum to make a prediction with regard to the sensory consequences of such motor commands, allowing the musculoskeletal system to prepare to successfully execute a movement. During movement, predicted sensations are then compared with the actual incoming sensations. If there is a positive match, the pattern is maintained for the next movement. The lack of a match is associated with an alert signal that is sent back to the motor cortical and subcortical areas, which activates feedback movement corrections and calibration of the forward model (Shadmehr et al., 2010).

This process allows the corticocerebellar circuit to act as somatic event detectors that respond, particularly to unexpected stimuli (Restuccia et al., 2007). Various studies on the internal forward model have demonstrated that the cerebellum generates motor-to-somatosensory predictions (Izawa et al., 2012; Popa et al., 2012; Knolle et al., 2013).

Thus, consensus is building that the cerebellum is more involved in learning to associate motor commands with novel sensory consequences–i.e., the forward model—than in learning to correlate sensory goals with novel motor commands (the inverse model).

SEQUENCING AND PREDICTION

In 1997, Braitenberg, Heck, and Sultan proposed sequence detection and generation as the basic operational mode of the cerebellum in the motor domain (Braitenberg et al., 1997). Since then, the sequencing properties of cerebellar processing have been studied extensively, and sequencing has been reported to be the more frequently impaired function in a large cohort of subjects with cerebellar damage (Tedesco et al., 2011).

Few years ago, we proposed to consider sequencing the basic mechanism that allows cerebellar prediction in all functional domains (Molinari et al., 2008). Traditionally, sequencing has not been recognized as a discrete cognitive function, and it can be defined as a supramodal function, the relationships with other functions of which, such as working memory and timing, remain unknown. Acquiring and acting on a serial order of events is a fundamental ability that effects sequencing structure knowledge. To recognize that stimuli are presented in a particular order, sensory information that pertains to a stimulus must be kept active in a working memory system and compared with subsequent stimuli. Like many instrumental abilities, sequence knowledge can be acquired incidentally through experience (implicit learning) or intentionally through explicit effort (declarative learning). A schematic of cerebellar sequence mechanisms for prediction is shown in **Figure 1**.

Whenever feedforward control is needed, the cerebellum intervenes by identifying predictable patterns of motor or cognitive command sequences and linking them with learned sensory or cognitive consequences. This process allows anticipatory responses to be generated in all relevant cerebellar domains.

Cerebellar input has a facilitating effect on the contralateral cerebral cortex, and chronic cerebellar damage (Di Lazzaro et al., 1994) and cerebellar conditioning transcranial magnetic stimulation (TMS; Grimaldi et al., 2014) reduce the excitability of the contralateral motor cortex. Furthermore, the cerebellar influence on the cerebral cortex is not limited to motor areas, and the nature and functional significance of the overall cerebellar influence over the cerebral cortex is the subject of much debate (Dalal et al., 2013). At least for the motor domain, it is widely accepted that cerebellar input conveys information for sensory motor integration.

TMS experiments in rats support this model (Ben Taib et al., 2005; Oulad Ben Taib and Manto, 2008). In rats, as in humans, the enhancement of excitability in the contralateral motor cortex after sustained somatosensory stimulation is cerebellum-dependent (Kaelin-Lang et al., 2002; Luft et al., 2002). Cerebellum-dependent neurophysiological changes are not limited to the motor cortex, and cerebellar output might also directly affect the somatosensory cortex. The parietal cortex projects to the cerebellum through the pontine nuclei in a topographically organized manner (Schmahmann and Pandya, 1989), and it receives the cerebellar return loop through the thalamus (Giannetti and Molinari, 2002; Allen et al., 2005; Clower et al., 2005). Cerebellar influences on the parietal somatosensory cortex have been demonstrated in cats (Kolodziejak et al., 2000) and in patients with unilateral cerebellar lesions (Restuccia et al., 2001). In subjects with unilateral cerebellar damage, late N24 and P24 components of the somatosensory-evoked potentials decline significantly in the contralateral somatosensory cortex (Restuccia et al., 2001). A magnetoencephalography (MEG) study that compared expected and unexpected sensory stimuli in evoking cerebellar and cortical responses has linked the somatosensonsory evoked potentials findings with the prediction and sequencing theories of cerebellar function. A regular train of somatosensory stimuli induces evoked potentials in the contralateral somatosensory cortex and ipsilateral cerebellum. If the stimulus is omitted at random, while no activity is recorded in the somatosensory cortex, cerebellar activity is markedly enhanced (Tesche and Karhu, 2000). This response after an unpredictable omission in a predictable sequence has been interpreted as

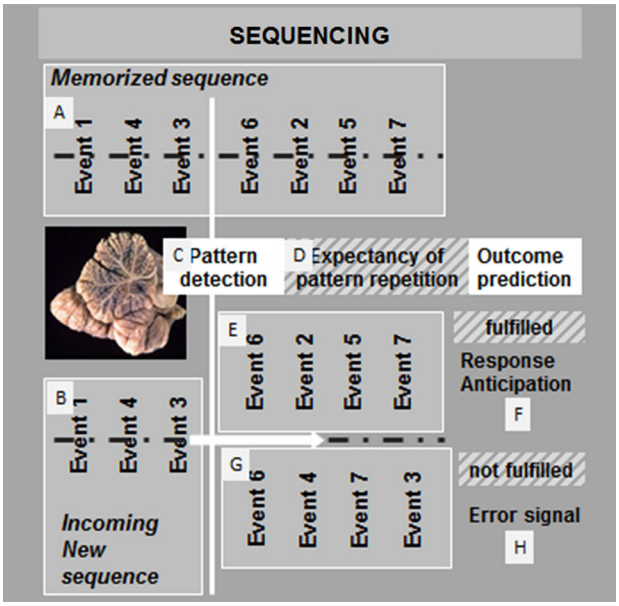


FIGURE 1 | Proposed mechanism of cerebellar sequencing for prediction. Incoming events are continuously monitored in the cerebellar circuits. Relations between events are compared in the cerebellar corticonuclear microcomplex (Ito, 2005) and stored in a working memory buffer **(A)**. Through the same mechanisms, sequences of new incoming events are compared with previously stored event sequences **(B)**. If a match is recognized **(C)**, then an expectancy of repetition is generated **(D)**. The cerebellar monitoring of the flow of events continues, and as long as the prediction is maintained **(E)**, response anticipation is conveyed, and feedforward control can function smoothly **(F)**. If prediction fails **(G)**, then an error signal is activated by the cerebellar output system **(H)**, and feedforward control is interrupted or corrected.

proof of the ability of the cerebellum to code expectancy (Ivry, 2000).

Clinical evidence of cerebellar function in coding expectancy has been confirmed by 2 groups, both of which used mismatch negativity protocols. Restuccia et al. (2007) used a somatosensory mismatch negativity (S-MMN) protocol in patients with unilateral cerebellar lesions. S-MMN is believed to be generated by differences between current and prior inputs, supported by an automatic change-detection cortical process. This process is blocked or impaired if the cerebellar input is absent. Subjects who are affected by unilateral hemispheric cerebellar stroke do not develop S-MMN responses in the contralateral cortex. Similar findings have been observed in an MMN auditory paradigm (Moberget et al., 2008), focusing on timing expectancy. Both studies have demonstrated that the cerebellum is part of the MMN circuit and that it is critical for generating expectancy and making sensory predictions.

These findings complement the theories on cerebellar function in the prediction of sensory events (Nixon, 2003) and the long-standing hypothesis that the cerebellum acts as a comparator (Ito, 2005). In this theoretical framework, it is conceivable that through a comparison of time and space characteristics of actual and preceding stimuli, predictable event sequences can be recognized and stored. Thus, sequencing in the sensorimotor domain is evident, but is this also true when cognitive functions are considered?

Behavioral or script sequencing can be defined as the process that allows spatial and temporal relations to be recognized correctly among behaviorally relevant actions (Sirigu et al., 1998). Script sequencing is altered in subjects with cerebellar damage who are tested in an ad hoc card sorting task (Leggio et al., 2011) and is interpreted as a prediction deficit in the cognitive/behavioral domain. Card sequencing tasks require visual or verbal material to be examined to understand spatial, temporal, and/or semantic relationships and correctly reconstruct the strings in logical sequences.

The test in Leggio et al. (2008) consisted of 11 sets of cards, each comprising 6 cartoon-like drawings, including sentences (to examine verbal factors), behavioral figures (for behavioral factors), and abstract figures (for spatial factors), that were to be ordered in a logical sequence by patients. The influence of the lesion was analyzed by grouping patients by lesion type (focal or atrophic) and lesion side (right or left). Patients with cerebellar damage developed cognitive sequencing impairments, and lesion side and characteristics of the material that were to be sequenced correlated. Specifically, patients with left lesions performed poorly only on script sequences that were based on pictorial material, and patients with right lesions encountered difficulties with script sequences that required verbal elaboration. The presence of right/left and pictorial/verbal differences is consistent with the hypothesis that cerebrocerebellar interactions are organized in segregated corticocerebellar loops, in which specificity is related to the characteristics of the information that is processed—not to the mode of function (Leggio et al., 2008).

CEREBELLAR GAIT

Locomotion can be considered a purposeful, goal-directed behavior that is initiated by signals that arise from volitional processing in the cerebral cortex or emotional processing in the limbic system and sustained by basic locomotor motor patterns that are generated by spinal interneuronal networks—i.e., the central pattern generator (CPG) circuits. Locomotor control mechanisms are complex and, in addition to the CPGs, involve various subcortical and cortical control areas (for review, see Takakusaki, 2013). In this network, the cerebellum is considered dispensable for steady-state locomotion but crucial for avoiding obstacles and adapting to novel conditions.

Cerebellar gait ataxia is characterized by staggering, irregular stepping, veering, and excessive high lifting of the feet above the ground. This clinical condition has been linked to the inability to control relative movements between leg joints during locomotion. Starting from clinical observation, the coordination of multijoint activity through the scaling of movement variables has been considered the core of cerebellar motor function (Topka et al., 1998). Of the movement variables that are cerebellum-dependent, the timing of muscle activity, especially of antagonist muscles, has long been favored (Frysinger et al., 1984).

In 2001, Earhart and Bastian questioned cerebellar function in the timing or scaling of individual joint movements during gait by asking subjects with cerebellar damage to step on a surface that was inclined at various angles while walking. Healthy subjects mastered the task by using several temporal strategies, with systematic shifts in the timing of muscle activity and peak joint angles, based on the changes in inclination. Notably, cerebellar subjects were able to produce appropriate timing shifts at most joints, demonstrating preserved selection of the basic timing of motor patterns. Conversely, the presence of abnormal relative joint movements and the decomposition of movement implicated the cerebellum in adjusting the relative movement of multiple joints, especially to accommodate external constraints (Earhart and Bastian, 2001). Collectively, animal studies and clinical evidence have demonstrated cerebellar function in adaptive gait control, effecting constant recalibration of walking patterns to navigate various terrains and environments smoothly.

Cerebellar adaptation is not based on sensory feedback information. Subjects with cerebellar damage are impaired in locomotor tasks that require prediction, whereas they have good control when reactive control is needed (Morton and Bastian, 2006). As discussed, one possible mechanism of sustaining prediction is sequencing, which can intervene at various levels of locomotor control. Thus, similar to what has been observed in sMMN paradigms (Restuccia et al., 2007), fixed sequences of sensory information, funneled by spinocerebellar fibers during locomotion (Jankowska et al., 2011), have been hypothesized to be recognized by the cerebellum, effecting correct prediction of the neuromuscular requirements of the subsequent step. Alterations in the predicted sequence will enhance the cerebellar output system, allowing cortical and brainstem locomotor regions to adapt.

Conversely, subjects with cerebellar atrophies are not only impaired in managing obstacles and adapting to novel environment, they develop ataxic gait in well-learned environments and on smooth surfaces (Mari et al., 2014), implicating cerebellar

processing in controlling steady-state locomotion. This clinical profile also exists in subjects with cerebellar stroke; nevertheless, gait ataxia is generally mild, from which patients recover well (Bultmann et al., 2014). Pascual-Leone and colleagues (Farzan et al., 2013) recently reported that 21 days of cerebellar TMS reduces gait ataxia in patients who are affected by idiopathic late-onset cerebellar atrophy. This finding suggests that low-frequency TMS reduces the inhibitory control of the cerebellar cortex over the dentate nucleus, favoring the function of dentate nucleus output.

To this end, we would like to advance an alternative hypothesis. Considering the established function role of motor learning and adaptation in allowing forward control strategies to be generated and the lack of cerebellar influence on reactive adjustments and well-learned automatic movements, it is conceivable that cerebellar TMS inhibits the cerebellar output, allowing motor circuits to act in the absence of cerebellar influences. Thus, the experimental condition that was proposed by Pasqual-Leone and colleagues could approximate a cerebellar focal lesion—i.e., after a stroke. Both conditions are associated with better locomotion than in the presence of cerebellar atrophy.

It follows that cerebellar gait ataxia due to cerebellar atrophy might be the result of erroneous cerebellar predictions. Altered cerebellar processing will insert virtual errors into the forward control models, inducing continuous correction of the ongoing motor command. If this hypothesis is true, the inhibition of cerebellar processing—e.g., by TMS or tDCS—would improve gait in subjects with cerebellar ataxia but will have little or no effect on subjects with ataxia due to focal cerebellar damage. Conversely, focal damage to the cerebellum is associated with balance and gait problems, primarily in the acute/subacute phase followed by efficient spontaneous functional recovery (Bultmann et al., 2014). This evidence suggests that the motor system recovers more efficiently from the absence of cerebellar processing than from alterations to it. This view is supported by recent literature on cerebellar stimulation (Farzan et al., 2013; Ferrucci et al., 2013; Grimaldi et al., 2014), suggesting that transcranial cerebellar stimulation is a feasible neurorehabilitation intervention that can be used to treat gait ataxia (Block and Celnik, 2012).

CONCLUSION

The importance of feedforward control in motor control and the significance of cerebellar processing in this function are well established. Nevertheless, in locomotion control, cerebellar function is neglected, and studies have focused primarily on spinal and cortical locomotion control mechanisms. Current evidence implicates forward models as more important locomotion control mechanisms, but the relative importance of forward and inverse models to locomotion remains unknown. Recent reports indicates that cerebellar processing intervenes in locomotion by providing advance information on subsequent step events, suggesting how such motor prediction can be obtained per the sequencing hypothesis of cerebellar function. In nearly all cerebellar functional domains—from motor to cognition—cerebellar symptoms can be attributed to impairments in recognizing repeated sequenced patterns. Only recognition of a previously experienced pattern allows a prediction to be made and thus effective feed-forward control to be instigated. This theory of cerebellar function has implications for the symptomatic treatment of gait disturbances, and preliminary results on magnetic and electrical modulation of cerebellar function are guiding the development of an effective treatment for ataxic gait.

REFERENCES

Allen, G., McColl, R., Barnard, H., Ringe, W. K., Fleckenstein, J., and Cullum, C. M. (2005). Magnetic resonance imaging of cerebellar-prefrontal and cerebellar-parietal functional connectivity. *Neuroimage* 28, 39–48. doi: 10.1016/j.neuroimage.2005.06.013

Bastian, A. J. (2006). Learning to predict the future: the cerebellum adapts feedforward movement control. *Curr. Opin. Neurobiol.* 16, 645–649. doi: 10.1016/j.conb.2006.08.016

Ben Taib, N. O., Manto, M., Pandolfo, M., and Brotchi, J. (2005). Hemicerebellectomy blocks the enhancement of cortical motor output associated with repetitive somatosensory stimulation in the rat. *J. Physiol.* 567(Pt. 1), 293–300. doi: 10.1113/jphysiol.2005.088229

Block, H. J., and Celnik, P. (2012). Can cerebellar transcranial direct current stimulation become a valuable neurorehabilitation intervention? *Expert Rev. Neurother.* 12, 1275–1277. doi: 10.1586/ern.12.121

Braitenberg, V., Heck, D., and Sultan, F. (1997). The detection and generation of sequences as a key to cerebellar function: experiments and theory. *Behav. Brain Sci.* 20, 229–245; discussion 245–277. doi: 10.1017/s0140525x9721143x

Bultmann, U., Pierscianek, D., Gizewski, E. R., Schoch, B., Fritsche, N., Timmann, D., et al. (2014). Functional recovery and rehabilitation of postural impairment and gait ataxia in patients with acute cerebellar stroke. *Gait Posture* 39, 563–569. doi: 10.1016/j.gaitpost.2013.09.011

Clower, D. M., Dum, R. P., and Strick, P. L. (2005). Basal ganglia and cerebellar inputs to 'AIP'. *Cereb. Cortex* 15, 913–920. doi: 10.1093/cercor/bhh190

Dalal, S. S., Osipova, D., Bertrand, O., and Jerbi, K. (2013). Oscillatory activity of the human cerebellum: the intracranial electrocerebellogram revisited. *Neurosci. Biobehav. Rev.* 37, 585–593. doi: 10.1016/j.neubiorev.2013.02.006

Di Lazzaro, V., Restuccia, D., Molinari, M., Leggio, M. G., Nardone, R., Fogli, D., et al. (1994). Excitability of the motor cortex to magnetic stimulation in patients with cerebellar lesions. *J. Neurol. Neurosurg. Psychiatry* 57, 108–110. doi: 10.1136/jnnp.57.1.108

Earhart, G. M., and Bastian, A. J. (2001). Selection and coordination of human locomotor forms following cerebellar damage. *J. Neurophysiol.* 85, 759–769.

Farzan, F., Wu, Y., Manor, B., Anastasio, E., Lough, M., Novak, V., et al. (2013). Cerebellar TMS in treatment of a patient with cerebellar ataxia: evidence from clinical, biomechanics and neurophysiological assessments. *Cerebellum* 12, 707–712. doi: 10.1007/s12311-013-0485-8

Ferrucci, R., Brunoni, A., Parazzini, M., Vergari, M., Rossi, E., Fumagalli, M., et al. (2013). Modulating human procedural learning by cerebellar transcranial direct current stimulation. *Cerebellum* 12, 485–492. doi: 10.1007/s12311-012-0436-9

Frysinger, R. C., Bourbonnais, D., Kalaska, J. F., and Smith, A. M. (1984). Cerebellar cortical activity during antagonist cocontraction and reciprocal inhibition of forearm muscles. *J. Neurophysiol.* 51, 32–49.

Giannetti, S., and Molinari, M. (2002). Cerebellar input to the posterior parietal cortex in the rat. *Brain Res. Bull.* 58, 481–489. doi: 10.1016/s0361-9230(02)00815-8

Grimaldi, G., Argyropoulos, G. P., Boehringer, A., Celnik, P., Edwards, M. J., Ferrucci, R., et al. (2014). Non-invasive cerebellar stimulation—a consensus paper. *Cerebellum* 13, 121–138. doi: 10.1007/s12311-013-0514-7

Ito, M. (2005). Bases and implications of learning in the cerebellum—adaptive control and internal model mechanism. *Prog. Brain Res.* 148, 95–109. doi: 10.1016/s0079-6123(04)48009-1

Ivry, R. (2000). Exploring the role of the cerebellum in sensory anticipation and timing: commentary on Tesche and Karhu. *Hum. Brain Mapp.* 9, 115–118. doi: 10.1002/(sici)1097-0193(200003)9:3<115::aid-hbm1>3.3.co;2-x

Izawa, J., Criscimagna-Hemminger, S. E., and Shadmehr, R. (2012). Cerebellar contributions to reach adaptation and learning sensory consequences of action. *J. Neurosci.* 32, 4230–4239. doi: 10.1523/JNEUROSCI.6353-11.2012

Jankowska, E., Nilsson, E., and Hammar, I. (2011). Processing information related to centrally initiated locomotor and voluntary movements by feline spinocerebellar neurones. *J. Physiol.* 589, 5709–5725. doi: 10.1113/jphysiol.2011.213678

Kaelin-Lang, A., Luft, A. R., Sawaki, L., Burstein, A. H., Sohn, Y. H., and Cohen, L. G. (2002). Modulation of human corticomotor excitability by somatosensory input. *J. Physiol.* 540(Pt. 2), 623–633. doi: 10.1113/jphysiol.2001.012801

Knolle, F., Schrager, E., and Kotz, S. A. (2013). Cerebellar contribution to the prediction of self-initiated sounds. *Cortex* 49, 2449–2461. doi: 10.1016/j.cortex.2012.12.012

Kolodziejak, A., Dziduszko, J., Niechaj, A., and Tarnecki, R. (2000). Influence of acute cerebellar lesions on somatosensory evoked potentials (SEPs) in cats. *J. Physiol. Pharmacol.* 51, 41–55.

Koziol, L., Budding, D., Andreasen, N., D'Arrigo, S., Bulgheroni, S., Imamizu, H., et al. (2014). Consensus paper: the cerebellum's role in movement and cognition. *Cerebellum* 13, 151–177. doi: 10.1007/s12311-013-0511-x

Leggio, M. G., Chiricozzi, F. R., Clausi, S., Tedesco, A. M., and Molinari, M. (2011). The neuropsychological profile of cerebellar damage: the sequencing hypothesis. *Cortex* 47, 137–144. doi: 10.1016/j.cortex.2009.08.011

Leggio, M. G., Tedesco, A. M., Chiricozzi, F. R., Clausi, S., Orsini, A., and Molinari, M. (2008). Cognitive sequencing impairment in patients with focal or atrophic cerebellar damage. *Brain* 131, 1332–1343. doi: 10.1093/brain/awn040

Luft, A. R., Kaelin-Lang, A., Hauser, T. K., Buitrago, M. M., Thakor, N. V., Hanley, D. F., et al. (2002). Modulation of rodent cortical motor excitability by somatosensory input. *Exp. Brain Res.* 142, 562–569. doi: 10.1007/s00221-001-0952-1

Mari, S., Serrao, M., Casali, C., Conte, C., Martino, G., Ranavolo, A., et al. (2014). Lower limb antagonist muscle co-activation and its relationship with gait parameters in cerebellar ataxia. *Cerebellum* 13, 226–236. doi: 10.1007/s12311-013-0533-4

Moberget, T., Karns, C. M., Deouell, L. Y., Lindgren, M., Knight, R. T., and Ivry, R. B. (2008). Detecting violations of sensory expectancies following cerebellar degeneration: a mismatch negativity study. *Neuropsychologia* 46, 2569–2579. doi: 10.1016/j.neuropsychologia.2008.03.016

Molinari, M., Chiricozzi, F., Clausi, S., Tedesco, A., De Lisa, M., and Leggio, M. (2008). Cerebellum and detection of sequences, from perception to cognition. *Cerebellum* 7, 611–615. doi: 10.1007/s12311-008-0060-x

Morton, S. M., and Bastian, A. J. (2006). Cerebellar contributions to locomotor adaptations during splitbelt treadmill walking. *J. Neurosci.* 26, 9107–9116. doi: 10.1523/jneurosci.2622-06.2006

Nixon, P. D. (2003). The role of the cerebellum in preparing responses to predictable sensory events. *Cerebellum* 2, 114–122. doi: 10.1080/14734220309410

Oulad Ben Taib, N., and Manto, M. (2008). Reinstating the ability of the motor cortex to modulate cutaneomuscular reflexes in hemicerebellectomized rats. *Brain Res.* 1204, 59–68. doi: 10.1016/j.brainres.2008.02.014

Popa, L. S., Hewitt, A. L., and Ebner, T. J. (2012). Predictive and feedback performance errors are signaled in the simple spike discharge of individual purkinje cells. *J. Neurosci.* 32, 15345–15358. doi: 10.1523/JNEUROSCI.2151-12.2012

Restuccia, D., Della, M. G., Valeriani, M., Leggio, M. G., and Molinari, M. (2007). Cerebellar damage impairs detection of somatosensory input changes. A somatosensory mismatch-negativity study. *Brain* 130(Pt. 1), 276–287. doi: 10.1093/brain/awl236

Restuccia, D., Valeriani, M., Barba, C., Le Pera, D., Capecci, M., Filippini, V., et al. (2001). Functional changes of the primary somatosensory cortex in patients with unilateral cerebellar lesions. *Brain* 124(Pt. 4), 757–768. doi: 10.1093/brain/124.4.757

Schmahmann, J. D., and Pandya, D. N. (1989). Anatomical investigation of projections to the basis pontis from posterior parietal association cortices in rhesus monkey. *J. Comp. Neurol.* 289, 53–73. doi: 10.1002/cne.902890105

Shadmehr, R., Smith, M. A., and Krakauer, J. W. (2010). Error correction, sensory prediction and adaptation in motor control. *Annu. Rev. Neurosci.* 33, 89–108. doi: 10.1146/annurev-neuro-060909-153135

Sirigu, A., Cohen, L., Zalla, T., Pradat-Diehl, P., Van Eeckhout, P., Grafman, J., et al. (1998). Distinct frontal regions for processing sentence syntax and story grammar. *Cortex* 34, 771–778. doi: 10.1016/s0010-9452(08)70780-9

Sotelo, C. (2008). Viewing the cerebellum through the eyes of Ramón Y Cajal. *Cerebellum* 7, 517–522. doi: 10.1007/s12311-008-0078-0

Takakusaki, K. (2013). Neurophysiology of gait: from the spinal cord to the frontal lobe. *Mov. Disord.* 28, 1483–1491. doi: 10.1002/mds.25669

Tedesco, A. M., Chiricozzi, F. R., Clausi, S., Lupo, M., Molinari, M., and Leggio, M. G. (2011). The cerebellar cognitive profile. *Brain* 134, 3672–3678. doi: 10.1093/brain/awr266

Tesche, C. D., and Karhu, J. J. (2000). Anticipatory cerebellar responses during somatosensory omission in man. *Hum. Brain Mapp.* 9, 119–142. doi: 10.1002/(sici)1097-0193(200003)9:3<119::aid-hbm2>3.3.co;2-i

Topka, H., Konczak, J., Schneider, K., Boose, A., and Dichgans, J. (1998). Multijoint arm movements in cerebellar ataxia. *Exp. Brain Res.* 119, 493–503. doi: 10.1007/s002210050365

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.

Received: 26 February 2014; accepted: 10 June 2014; published online: 25 June 2014.
*Citation: Pisotta I and Molinari M (2014) Cerebellar contribution to feedforward control of locomotion. Front. Hum. Neurosci. **8**:475. doi: 10.3389/fnhum.2014.00475*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 17 April 2014
doi: 10.3389/fnhum.2014.00207

Effects of robot assisted gait training in progressive supranuclear palsy (PSP): a preliminary report

Patrizio Sale[1]*, Fabrizio Stocchi[1], Daniele Galafate[1], Maria Francesca De Pandis[2], Domenica Le Pera[1], Ivan Sova[1], Manuela Galli[1,3], Calogero Foti[4] and Marco Franceschini[1]

[1] *Department of Neurorehabilitation, IRCCS San Raffaele Pisana, Rome, Italy*
[2] *San Raffaele Cassino, Cassino, Italy*
[3] *Dipartimento di Elettronica, Informazione e Bioingegneria-Politecnico di Milano, Milano, Italy*
[4] *Physical Rehabilitation Medicine Chair, Clinical Sciences and Translational Medicine DPT, Tor Vergata University, Roma, Italy*

Edited by:
Marco Iosa, Fondazione Santa Lucia, Italy

Reviewed by:
Leonardo Gizzi, University Hospital Göttingen, Germany
Giovanni Morone, IRCCS Santa Lucia Foundation, Italy

***Correspondence:**
Patrizio Sale, Department of Neurorehabilitation, IRCCS San Raffaele Pisana, Via della Pisana 235, 00163, Rome, Italy
e-mail: patrizio.sale@gmail.com

Background and Purpose: Progressive supranuclear palsy (PSP) is a rare neurodegenerative disease clinically characterized by prominent axial extrapyramidal motor symptoms with frequent falls. Over the last years the introduction of robotic technologies to recover lower limb function has been greatly employed in the rehabilitative practice. This observational trial is aimed at investigating the changes in the main spatiotemporal following end-effector robot training in people with PSP.

Method: Pilot observational trial.

Participants: Five cognitively intact participants with PSP and gait disorders.

Interventions: Patients were submitted to a rehabilitative program of robot-assisted walking sessions for 45 min, 5 times a week for 4 weeks.

Main outcome measures: The spatiotemporal parameters at the beginning (T0) and at the end of treatment (T1) were recorded by a gait analysis laboratory.

Results: Robot training was feasible, acceptable and safe and all participants completed the prescribed training sessions. All patients showed an improvement in the gait spatiotemporal index (Mean velocity, Cadence, Step length, and Step width) (T0 vs. T1).

Conclusions: Robot training is a feasible and safe form of rehabilitation for cognitively intact people with PSP. The lack of side effects and the positive results in the gait parameter index in all patients support the recommendation to extend the trials of this treatment. Further investigation regarding the effectiveness of robot training in time is necessary.

Trial registration: ClinicalTrials.gov NCT01668407.

Keywords: progressive supranuclear palsy, PSP, gait analysis, lower limb, robot

INTRODUCTION

Progressive supranuclear palsy (PSP) is a rare neurodegenerative disease that causes the gradual deterioration and death of specific volumes of the brain (Richardson et al., 1963; Steele et al., 1964). Ludolph and colleagues showed that the neuropathologic features of PSP include marked midbrain atrophy and atrophy of the pallidum, thalamus, subthalamic nucleus, and frontal lobes (Ludolph et al., 2009). Clinical criteria for the diagnosis of PSP are: a gradually progressive disorder, an onset at the age of 40 years or later, a presence of vertical supranuclear palsy, a slowing of vertical saccades and a postural instability with falls in the first year of disease onset. In particular, the first symptoms in two-thirds of the cases are: loss of balance, lunging forward when mobilizing, fast walking, bumping into objects or people, and falls (Lubarsky and Juncos, 2008). Other common early symptoms are changes in personality, general slowing of movement and visual symptoms. Postural instability and gait impairment are the most important disorders in the early phases of the disease. The PSP subject shows a short, shuffling stepped gait, gait freezing, lurching, unsteady gait or spontaneous falls (Lubarsky and Juncos, 2008). In particular, subjects with PSP have decreased step length, step velocity, and a significantly slower ability to break a fall from the center of gravity than controls (Welter et al., 2007). Although some disorders are similar to those found in Parkinsonism (rigidity, bradykinesia, postural instability), PSP differs in several ways. The PSP subject does not have a forward flexed posture like the Parkinson Disease (PD) subject and falls in PSP tend to be backwards (Boeve, 2007). Pharmacotherapy with carbidopa/levodopa and with dopamine agonists typically is ineffective in managing the disorders (Rampello et al., 2005). Non-pharmacologic therapies such as physical therapy and occupational therapy are potentially useful, with the main goals being the maintenance of functional ambulation and the reduction of falls and associated injuries (Steffen et al., 2007). Gait recovery in all patients with impairments of the central nervous system (CNS) is an integral part of rehabilitation and often influences

the possibility of a patient returning home or to work (Schmidt et al., 2007). Neurologic motor rehabilitation is directed toward the re-learning of motor skills. In particular, the recovery of walking is a crucial aspect of rehabilitation, improving the quality of life and patient's independence. For non-ambulatory neurological patients, mechanically assisted walking with body weight support (BWS) has been suggested as a strategy to facilitate walking (Richards et al., 1993; Hesse et al., 2001) because it provides the opportunity to complete more practice of the whole task than would be possible by assisting overground walking. In the last years, robot-aided walking is considered a promising tool for gait rehabilitation (Colombo et al., 2000) in various neurological disease (Beer et al., 2008; Lo and Triche, 2008; Hesse et al., 2010). The robotic devices have been developed to relieve physical therapists from the strenuous and not ergonomic burden of manual BWS (Volpe et al., 2001; Winchester and Querry, 2006; Steffen et al., 2007). Furthermore, the use of robotic devices is currently advised to prevent the risk of falls and to improve gait velocity with the total safety of the patient (Morone et al., 2014). Moreover, robot devices can be used to give inpatients an intensive program (in terms of many repetitions) of complex gait cycles. The robotic machines, offering practice up to 1000 steps per session, used either an exoskeleton or an end-effector approach. Currently, a robotic task-specific repetitive approach, i.e., numerous practices of complex gait cycles, is regarded as the most promising to restore motor function after neurological disease (Dobkin, 2004; Dobkin and Duncan, 2012). Evidence-based approaches of rehabilitation in PSP are lacking and currently the majority of research on these subjects consists of case reports involving only a small number of patients. Until now, only few studies have been carried out on physiotherapy intervention in the PSP gait and balance disorder (Izzo et al., 1986; Sosner et al., 1993; Suteerawattananon et al., 2002; Steffen et al., 2014). Usually the subject with PSP needs rehabilitative training to improve balance and gait problems and to prevent the frequent falls. The rehabilitation programs for patients with PSP generally include limb-coordination activities, tilt-board balancing, gait training, strength training with progressive resistive exercises and isokinetic exercises and stretching of the neck muscles (Miyai et al., 2002). Even if several studies have demonstrated the efficacy of workout, including treadmill training, in subjects with PD (Zampieri and Di Fabio, 2006; Semprini et al., 2009; Mehrholz et al., 2010), so far little evidence has been found on the effects of the treadmill training therapy on tauopathies or other parkinsonian disorders (Suteerawattananon et al., 2002; Steffen et al., 2007) and no study has been conducted on the employment of robot assisted treatment in PSP subjects. This study was aimed at investigating the effects on improvement of the walking function by a change in spatiotemporal parameters using end-effector robotic rehabilitation locomotion training in patients with PSP.

MATERIALS AND METHODS

This study was a pilot observational trial. The subject had been on stable doses of medication for at least 4 weeks prior to study onset and showed an endurance sufficient to keep an upright position, assisted or unassisted, for at least 15 min. A preliminary medical examination included a physical and neurological test besides a gait analysis. The inclusion criteria for all groups were: (a) diagnosis of idiopathic PSP according to the UK Brain Bank criteria, without any other significant neurological or orthopaedic disorder; (b) age between 18 and 90 years old; (c) ability to walk, unassisted or with little assistance, for at least 25 feet. The following exclusion criteria were identified: (d) inability to understand instructions required by the study (Informed Consent Test of Comprehension); (e) primarily wheelchair bound; (f) chronic and ongoing alcohol or drug abuse, active depression, anxiety or psychosis that might interfere with the use of the equipment or testing.

INSTRUMENTAL ASSESSMENTS

Trained professionals, who were not involved in the research treatment and blind to patients' treatment, performed all instrumental and clinical assessments. All outcome assessments were collected in the ON phase 1 h and half after oral assumption of the usual dose of medication. The 3D-Gait analysis (3D-GA) was conducted using the following equipment: a 12-camera optoelectronic system with passive markers (ELITE2002, BTS, Italy) to measure the kinematics of the movement; 2 force platforms (Kistler, CH) to obtain the kinetic data of the movement (i.e., ground reaction forces); 2 TV camera Video systems (BTS, Italy) synchronized with the optoelectronic and force platform systems for video recording. To evaluate the kinematics of each body segment, markers were positioned as described by Davis and colleagues (Sale et al., 2012, 2013a). Subjects were asked to walk barefoot, at their own natural pace (self-selected and comfortable speed), along a 10-meter walkway where the two force platforms were placed. At least seven trials were collected for each subject in order to ensure data consistency. All graphs obtained from GA were normalized as a % of the gait cycle and kinetic data were normalized for individual body weights. In order to quantify the gait pattern of participants involved in this study, a specific software (Smartanalyser, BTS, Italy) enabled the calculation of some indices (time/distance parameters, joint angles values in specific gait cycle instant, peak values in ankle power graph) starting from those data.

PRIMARY OUTCOMES

A primary outcome was the change in spatiotemporal parameter. In particular we recorded: the gait velocity assessed by mean velocity (m/s), which measured the rate of change of position, recorded in meters per second; the cadence (step/min) that measured the number of steps taken in a given period of time, which was then converted into the number of steps taken per minute; the step length (mm) that measured the average distance (mm) between two successive placements of the same foot; the stride length (mm) that measured the average distance (mm) between two successive placements of the same foot, the step width (mm) that measured the medio-lateral distance between the two feet during double support; the stance time (stride %) that measured the duration of the stance phase; the swing time (stride %) that measured the duration of the swing phase and the double support (stride %) that measured the duration of double support.

ROBOT THERAPY

Each subject was asked to perform 20 sessions (5 days a week for 4 weeks) of robot assisted gait training, using the commercially

available end effector system machines G-EO system device (Reha Technology AG; Olten, Switzerland). The trajectories of the footplates and the vertical and horizontal movements of the center of mass were fully programmable, enabling wheelchair-bound subjects not only the repetitive practice of simulated floor walking but also up and down stair climbing. One therapist, who has experience in machine-supported gait rehabilitation, assisted the patients with putting on the harness while sitting in their wheelchair, getting onto the G-EO System in the wheelchair using a ramp from the rear, fixing the feet on the plates, hoisting the patient, attaching the lateral ropes, and setting the therapy parameters memorized by the G-EO System computer. The footplates had 3 DoF each, allowing to control the length and the height of the steps and the foot plate angles. The maximum step length corresponded to 550 mm, the maximum achievable height of the steps was 400 mm and the maximum angles were ±90°. The maximum speed of the foot plates was 2.3 km/h. The exercise included a robot-assisted walking therapy, at variable speeds, for a maximum of 45 min, with a partial BWS. All participants started with 30–40% of BWS and an initial speed of 1.5 km/h; speed was then increased to a range of 2.2–2.5 km/h maximum and initial BWS was decreased. Two further DoF controlled the BWS system and the lateral displacement of the hip. The graphic user interface (GUI) showed during the training the actual trajectory, so that the therapists were able to control and to correct it. Changes during the training were made for step length, step height, the terminal stance and the initial contact inclination angles of the feet, the vertical and the lateral excursions of the CoM, and for the relative position of the suspension point with respect to the footplates. The computer saved the trajectory settings. The adaptive control was applied only to both of the 3 DoF intended to control the legs. The remaining 2 DoF for the control of the center of mass were excluded from being master in the adaptive mode. In particular, the patients must apply the necessary force for moving the footplates along the selected trajectory. The necessary force for moving the footplates along the selected trajectory settings was set by a force level slider in the software of the robot. The computer put the footplates and the mechanics of the G-EO Systems to virtual zero friction and once the intended force level on the lower limb was reached, there could be amplification to the movement, according to the value of the amplification level selected in the robot by therapist. The amplification level provided for additional acceleration while executing the movement of the footplates along the selected trajectory. By adding acceleration to the necessary force for moving, the footplates lowered. This provided a smooth and continuous movement of the footplates. After 45 min the session was stopped. During each session, the patients practiced 5–30 min of simulated floor walking followed by 5 to 10 min of repetitive simulated stair climbing up and down. The patient practiced a minimum of 300 steps on the simulated floor and climbed a minimum of 50 steps on the simulated stair during each session. Breaks were optional, but uninterrupted training intervals of at least 5 min for simulated floor walking and 3 min of simulated stair climbing were required. Heart rate and blood pressure were monitored at the beginning and at the end of each session. Subjects who did not retrieve sessions and interrupted the treatment for more than 3 consecutive days were excluded from the study.

STATISTICAL ANALYSIS

All the previously defined parameters were computed for each participant. Mean values and standard deviations of all indexes were calculated for each group. The Kolomogorov–Smirnov tests were used to verify if the parameters were normally distributed. As this was not the case, we used Wilcoxon's tests in order to detect significant changes between data at baseline (T0) and endpoint (T1). The T0 and T1 data of all patients and CG were compared with Mann-Whitney U tests. Statistical significance was set at $p < 0.05$. The Mann-Whitney test was used to compare median scores between groups.

ETHICAL ASPECTS

This study was performed in accordance with the Declaration of Helsinki and was approved by the ethics committees of IRCCS San Raffaele Pisana. Informed consent was obtained from all subjects enrolled in this study.

RESULTS

We screened 15 patients, 5 of whom satisfied the inclusion criteria. No dropouts were recorded during the treatment and all subjects fulfilled the protocol (compliant subjects: $N = 5$). The distribution of the study subjects ($N = 5$) by age, gender, and main clinical and demographical characteristics are shown in **Table 1**. The median age was 67.80 ± 11.71 years. Participant demographics are presented in **Table 1**. **Table 2** summarizes the observed mean ± SD for all tests (T0 vs. T1), as measured on the compliant subjects at T0 ($N = 5$), T1 ($N = 5$) (**Table 2**). Gait velocity (T0 0.54 ± 0.173 m/s and T1 0.69 ± 0.150 m/s) and cadence (T0 83.00 ± 9.618 and T1 93.60 ± 15.437) improved respectively by 15 and 23.8%. Participants also demonstrated an improvement of 11% in step length left (T0 421.00 ± 98.831 mm and T1 466.40 ± 105.749 mm) and of 35% in step length right (T0 363.20 ± 94.767 mm and T1 429.80 ± 67.570 mm) and a decrease of 9% of Step width (T0 166.60 ± 24.460 mm and T1 153.60 ± 43.678 mm) (**Table 2**). Due to a small analyzed sample size no statistical significance was found in all the analyzed parameters. No significant changes were found in SpO2 (T0 97.5 ± 1.6 T1 97 ± 4.1, and in the HR (T0 82.3 ± 13.1 T1 86.9 ± 16) after the trainings in all patients. The patients workload and statisfaction

Table 1 | Patients' clinic and demographic data at baseline.

Experimental group *(n = 5)*		
	n	***Mean ± SD***
Dropout	0	
Compliants	5	
Male	3	
Female	2	
Age		67.8 ± 11.7
Height		161.2 ± 3.56
Weight		72.00 ± 10.58
Years of disease		3.6 ± 1.85
Psp rating scale		32.00 ± 9.24
PSP staging scale		2.4 ± 0.5
Walking handicap scale		3.2 ± 1.48

Legend n, number; SD, standard deviation

Table 2 | Results of gait analysis spatiotemporal parameters of all single patients and mean at T0 and T1.

	Sex	Età	Altezza	Peso	T0 Mean velocity (m/s)	T1 Mean velocity (m/s)	T0 Cadence (step/min)	T1 Cadence (step/min)	T0 Step length (mm) DX	T1 Step length (mm) DX	T0 Step length (mm) SX	T1 Step length (mm) SX	T0 Step width (mm)	T1 Step width (mm)
P1	Female	52	158	70	0.53	0.68	91	113	349	384	353	344	173	165
P2	Female	68	165	68	0.49	0.62	70	72	328	471	492	562	156	143
P3	Male	78	158	70	0.43	0.57	77	86	349	410	336	400	205	220
P4	Male	80	160	62	0.42	0.63	84	97	268	359	365	436	159	101
P5	Female	61	165	90	0.84	0.95	93	100	522	525	559	590	140	139
Mean		67.80	161.20	72.00	0.54	0.69	83.00	93.60	363.20	429.80	421.00	466.40	166.60	153.60
SD		11.71	3.56	10.58	0.173	0.150	9.618	15.437	94.767	67.570	98.831	105.749	24.460	43.678

	Sex	T0 Stance time (% stride) DX	T1 Stance time (% stride) DX	T0 Stance time (% stride) SX	T1 Stance time (% stride) SX	T0 Double supp. (% stride) DX	T1 Double supp. (% stride) DX	TO Double supp. (% stride) SX	T1 Double supp. (% stride) SX
P1	Female	62%	63%	62%	63%	14%	14%	12%	11%
P2	Female	60%	63%	71%	62%	19%	17%	11%	8%
P3	Male	62%	65%	58%	61%	14%	15%	10%	9%
P4	Male	64%	60%	68%	57%	15%	8%	14%	8%
P5	Female	60%	59%	59%	57%	13%	11%	6%	6%
Mean		62%	62%	64%	60%	15%	13%	11%	8%
SD		0.017	0.024	0.057	0.028	0.023	0.035	0.030	0.018

assessed by a Visual Analogical Scale showed a tendency toward positive feelings regarding the training process. Moreover regarding the fatigue the patients reported that during and at the end of the training training there was no excessive fatigue. The patients did clearly feel safe and comfortable with the robot at the end of the training.

DISCUSSION

Many authors have shown the efficacy of robot-assisted gait training on improving the walking function in several neurological diagnoses (Wirz et al., 2005; Morone et al., 2012; Schwartz et al., 2012; Sale et al., 2013a) but the process aimed at restoring this function in patients with a neurological pathology is challenged by the complexity and variability of these disorders (Sale et al., 2012). As far as we know, this is the first study that examines the effects of end effector robot-assisted training in individuals with PSP. This study shows that twenty 45-min sessions of robot-assisted training is a treatment that could stimulate and enhance the beneficial effects of motor training on gait recovery in patients with PSP. This trial protocol is easy, reproducible and safe and it allows the training of PSP subjects with moderate to severe lower limb impairment. As demonstrated, in gait training to walk repetitively in a natural manner similar to the over-ground gait and with the correct proprioceptive and exteroceptive feedback is of the most critical importance (Sale et al., 2012). In particular our adaptive training robotic protocol where the patient interacts with the robot can overcome all the limitations about the repetitively (and repeatability) of the movement with respect to the human-human interaction. Until now, several studies have demonstrated that robotic gait training can slow the clinical progression of gait disease in PD patients, but so far, no other evidence has been found in the PSP population. In particular, robot-assisted training may improve postural instability in patients with PD (Picelli et al., 2012a) or may develop aspects of walking ability (Picelli et al., 2012b). In our recent paper we demonstrated that the use of the end-effector lower limb robotic device in PD patients increases a short period lower limb motor recovery in idiopathic PD patients, improving above all the gait velocity, the step length and the stride length (Sale et al., 2013b). To date, there are not many studies on the gait rehabilitation in patients with PSP. Recently Steffen and colleagues published an interesting case report on the use of the treadmill in gait recovery in one patient with PSP, with a follow-up of 10 years. In particular, the author showed how a patient with PSP participated consistently in a regular group exercise program for 10 years and how he reduced fall frequency, maintained balance and endurance and retained community ambulation using a walker (Steffen et al., 2014). Falls and the associated trauma are one of the main causes of morbidity and mortality in these disorders and the lower limb training could effectively prevent it; falls are also the leading cause of unintentional injury and hospitalization in people aged 65 years and older (Dellinger and Stevens, 2006; Bridenbaugh and Kressig, 2011). Different methods of gait rehabilitation have been used so far in neurological lower limb physiotherapy to prevent the falls, such as manually assisted over-ground training and manually assisted treadmill training or robotic training with or without the BWS (Sale et al., 2012). The advantage of these electromechanical devices, compared with treadmill training with partial BWS in PD and Stroke patients, may be the reduced effort required by therapists, as they no longer need to set the paretic limbs or assist trunk movements (Hesse et al., 2003). However, the effort in the development of robotic gait training is not limited to the improvement of working conditions of physiotherapists, but rather in providing the patient with an engaging, challenging and effective rehabilitation tool. Our choice to use the robotic device in PSP gait recovery has many bases. Our training shows in all patients an improvement in each spatio-temporal parameter, in particular cadence, step length, stride length, velocity, and reduction of step width. These are the parameters mostly connected to the risk of falls. In particular, several studies have identified changes in certain spatial and temporal gait parameters as independent predictors of the fall risk. Tailored and colleagues showed in a sample of older people that there were significant main effects of gait condition and of faller status for mean value measures (velocity, stride length, double support time, and stride width) and for variability measures (swing time variability and stride length variability); the examination of individual gait parameters indicated that the multiple fallers walked more slowly, had a shorter stride length, spent more time in double support, had a wider support width and showed more variability in stride length and swing time (Taylor et al., 2013). Maki observed in his study that increased stride-to-stride variability in stride length, stride speed and double support time as well as increased stride width were predictive of falls in the ensuing 6 months for residents of a senior living facility (Maki, 1997). Our data, compared with the results of Tailored and Maki, suggest that the robot therapy, with the intention to recover the leg movements, could increase and help these patients to prevent falls and other trauma. Several conditions should however be considered when interpreting these data. Most importantly, these are simply case descriptions. Changes in these 5 individuals could have been due to a variety of factors. Possibly the improvements could represent progresses in test performance. Or perhaps the observed changes could be due to usual day-to-day, but it should be noted that the trends in the repeated measures were fairly consistent and showed improvement in all patients. A long period follow-up is required to confirm our hypothesis. Furthermore, it is unknown to what extent these findings will generalize in other individuals in the early and middle stages of PSP. These 5 individuals were in the early to middle stages of PSP and highly motivated, which may have contributed to their reported adherence, even after completing the supervised part of the program. Our experience and various examined articles showed that robot-assisted gait therapy provides versatile control approaches as a framework in the design of optimal rehabilitation interventions and experimental motor control studies, but the high cost of robot devices raises the question of efficiency in comparison with other training strategies.

CONCLUSION

The focus on gait recovery represents one of the most innovative features of this study and makes this research useful in clinical practice. Spatial-temporal gait analysis can detect discrete gait disorders, which are not perceptible to the naked eye, and several

gait changes have been identified as fall predictors. Early detection allows early intervention. The positive results on improvement in spatiotemporal parameter of the PSP subject by the Robot Therapy, the lack of side effects strongly recommend extending the use of a Robot Therapy in the recovery of gait performance. This rehabilitation training could provide new opportunities in PSP rehabilitation thanks to a sensorimotor approach aimed at functional recovery.

AUTHOR CONTRIBUTIONS

All authors contributed to the design of the study, to draft and review the manuscript. Patrizio Sale, Calogero Foti, Daniele Galafate, Domenica Le Pera contributed to data analysis. Patrizio Sale, Marco Franceschini, Maria Francesca De Pandis and Fabrizio Stocchi contributed in the supervision of the research study. All authors read and approved the final manuscript.

REFERENCES

Beer, S., Aschbacher, B., Manoglou, D., Gamper, E., Kool, J., and Kesselring, E. J. (2008). Robot-assisted gait training in multiple sclerosis: a pilot randomized trial. *Mult. Scler.* 14, 231–236. doi: 10.1177/1352458507082358

Boeve, B. F. (2007). Links between frontotemporal lobar degeneration, corticobasal degeneration, progressive supranuclear palsy, and amyotrophic lateral sclerosis. *Alzheimer Dis. Assoc. Disord.* 21, S31–S38. doi: 10.1097/WAD.0b013e31815bf454

Bridenbaugh, S. A., and Kressig, R. W. (2011). Laboratory review: the role of gait analysis in seniors' mobility and fall prevention. *Gerontology* 57, 256–264. doi: 10.1159/000322194

Colombo, G., Joerg, M., Schreier, R., and Dietz, V. (2000). Treadmill training of paraplegic patients using a robotic orthosis. *J. Rehabil. Res. Dev.* 37, 693–700.

Dellinger, A. M., and Stevens, J. A. (2006). The injury problem among older adults: mortality, morbidity and costs. *J. Safety Res.* 37, 519–522. doi: 10.1016/j.jsr.2006.10.001

Dobkin, B. H. (2004). Strategies for stroke rehabilitation. *Lancet Neurol.* 3, 528–536. doi: 10.1016/S1474-4422(04)00851-8

Dobkin, B. H., and Duncan, P. W. (2012). Should body weight-supported treadmill training and robotic-assistive steppers for locomotor training trot back to the starting gate? *Neurorehabil. Neural Repair* 26, 308–317. doi: 10.1177/1545968312439687

Hesse, S., Schmidt, H., Werner, C., and Bardeleben, A. (2003). Upper and lower extremity robotic devices for rehabilitation and for studying motor control. *Curr. Opin. Neurol.* 16, 705–710. doi: 10.1097/00019052-200312000-00010

Hesse, S., Waldner, A., and Tomelleri, C. (2010). Innovative gait robot for the repetitive practice of floor walking and stair climbing up and down in stroke patients. *J. Neuroeng. Rehabil.* 28, 7–30. doi: 10.1186/1743-0003-7-30

Hesse, S., Werner, C., Bardeleben, A., and Barbeau, H. (2001). Body weight-supported treadmill training after stroke. *Curr. Atheroscler. Rep.* 3, 287–294. doi: 10.1007/s11883-001-0021-z

Izzo, K., DiLorenzo, P., and Roth, A. (1986). Rehabilitation in progressive supranuclear palsy: case report. *Arch. Phys. Med. Rehabil.*67, 473–476.

Lo, A., and Triche, E. (2008). Improving gait in multiple sclerosis using robot-assisted, body weight supported treadmill training. *Neurorehabil. Neural Repair* 22, 661–671. doi: 10.1177/1545968308318473

Lubarsky, M., and Juncos, J. L. (2008). Progressive supranuclear palsy: a current review. *Neurologist* 14, 79–88. doi: 10.1097/NRL.0b013e31815cffc9

Ludolph, A. C., Kassubek, J., Landwehrmeyer, B. G., Mandelkow, E., Mandelkow, E. M., Burn, D. J., et al. (2009). Tauopathies with parkinsonism: clinical spectrum, neuropathologic basis, biological markers, and treatment options. *Eur. J. Neurol.* 16, 297–309. doi: 10.1111/j.1468-1331.2008.02513.x

Maki, B. E. (1997). Gait changes in older adults: predictors of falls or indicators of fear. *J. Am. Geriatr. Soc.* 45, s313–320.

Mehrholz, J., Friis, R., Kugler, J., Twork, S., Storch, A., and Pohl, M. (2010). Treadmill training for patients with Parkinson's disease. *Cochrane Database Syst. Rev.* CD007830. doi: 10.1002/14651858.CD007830

Miyai, I., Fujimoto, Y., Yamamoto, H., Ueda, Y., Saito, T., Nozaki, S., et al. (2002). Long-term effect of body weight-supported treadmill training in Parkinson's disease: a randomized controlled trial. *Arch. Phys. Med Rehabil.* 83, 1370–1373. doi: 10.1053/apmr.2002.34603

Morone, G., Iosa, M., Bragoni, M., De Angelis, D., Venturiero, V., Coiro, P., et al. (2012). Who may have durable benefit from robotic gait training?: a 2-year follow-up randomized controlled trial in patients with subacute stroke. *Stroke* 43, 1140–1142. doi: 10.1161/STROKEAHA.111.638148

Morone, G., Iosa, M., Pratesi, L., and Paolucci, S. (2014). Can overestimation of walking ability increase the risk of falls in people in the subacute stage after stroke on their return home? *Gait Posture* 39, 965–970. doi: 10.1016/j.gaitpost.2013.12.022

Picelli, A., Melotti, C., Origano, F., Waldner, A., Fiaschi, A., Santilli, V., et al. (2012b). Robot-assisted gait training in patients with Parkinson disease: a randomized controlled trial. *Neurorehabil. Neural Repair* 26, 353–361. doi: 10.1177/1545968311424417

Picelli, A., Melotti, C., Origano, F., Waldner, A., Gimigliano, R., and Smania, N. (2012a). Does robotic gait training improve balance in Parkinson's disease? A randomized controlled trial. *Parkinsonism Relat. Disord.* 18, 990–993. doi: 10.1016/j.parkreldis.2012.05.010

Rampello, L., Buttá, V., Raffaele, R., Vecchio, I., Battaglia, G., Cormaci, G., et al. (2005). Progressive supranuclear palsy: a systematic review. *Neurobiol Dis.* 20, 179–186. doi: 10.1016/j.nbd.2005.03.013

Richards, C. L., Malouin, F., Wood-Dauphinee, S., Williams, J. I., Bouchard, J. P., and Brunet, D. (1993). Task-specific physical therapy for optimization of gait recovery in acute stroke patients. *Arch. Phys. Med. Rehabil.* 74, 612–620. doi: 10.1016/0003-9993(93)90159-8

Richardson, J. C., Steele, J., and Olszewski, J. (1963). Supranuclear ophthalmoplegia, pseudobulbar palsy, nuchal dystonia and dementia. A clinical report on eight cases of 'heterogeneous system degeneration.' *Trans. Am. Neurol. Assoc.* 88, 25–29.

Sale, P., De Pandis, M. F., Stocchi, F., Domenica, L. P., Sova, I., Cimolin, V., et al. (2013b). Robot-assisted walking training for individuals with Parkinson's disease: a pilot randomized controlled trial. *BMC Neurol.* 13:50. doi: 10.1186/1471-2377-13-50

Sale, P., De Pandis, M. F., Vimercati, S. L., Sova, I., Foti, C., Tenore, N., et al. (2013a). The relation between Parkinson's disease and ageing. Comparison of the gait patterns of young Parkinson's disease subjects with healthy elderly subjects. *Eur. J. Phys. Rehabil. Med.* 49, 161–167.

Sale, P., Franceschini, M., Waldner, A., and Hesse, S. (2012). Use of the robot assisted gait therapy in rehabilitation of patients with stroke and spinal cord injury. *Eur. J. Phys. Rehabil. Med.* 48, 111–121.

Schmidt, H., Werner, C., Bernhardt, R., Hesse, S., and Krüger, J. (2007). Gait rehabilitation machines based on programmable footplates. *J. Neuroeng. Rehabil.* 4, 2. doi: 10.1186/1743-0003-4-2

Schwartz, I., Sajin, A., Moreh, E., Fisher, I., Neeb, M., Forest, A., et al. (2012). Robot-assisted gait training in multiple sclerosis patients: a randomized trial. *Mult. Scler.* 18, 881–890. doi: 10.1177/1352458511431075

Semprini, R., Sale, P., Foti, C., Fini, M., and Franceschini, M. (2009). Gait impairment in neurological disorders: a new technological approach. *Funct. Neurol.* 24, 179–183.

Sosner, J., Wall, G., and Sznajder, J. (1993). Progressive supranuclear palsy: clinical presentation and rehabilitation of two patients. *Arch. Phys. Med. Rehabil.*74, 537–539. doi: 10.1016/0003-9993(93)90120-Y

Steele, J. C., Richardson, J. C., and Olszewski, J. (1964). Progressive Supranuclear Palsy. A heterogeneous degeneration involving the brain stem, basal ganglia and cerebellum with vertical gaze and pseudobulbar palsy, nuchal dystonia and dementia. *Arch Neurol.* 10, 333–359. doi: 10.1001/archneur.1964.00460160003001

Steffen, T. M., Boeve, B. F., Mollinger-Riemann, L. A., and Petersen, C. M. (2007). Long-term locomotor training for gait and balance in a patient with mixed progressive supranuclear palsy and corticobasal degeneration. *Phys. Ther.* 87, 1078–1087. doi: 10.2522/ptj.20060166

Steffen, T. M., Boeve, B. F., Petersen, C. M., Dvorak, L., and Kantarci, K. K. (2014). Long-term exercise training for an individual with mixed corticobasal degeneration and progressive supranuclear palsy features: 10-year follow-up. *Phys. Ther.* 94, 289–296. doi: 10.2522/ptj.20130052

Suteerawattananon, M., MacNeill, B., and Protas, E. J. (2002). Supported treadmill training for gait and balance in a patient with progressive supranuclear palsy. *Phys. Ther*. 82, 485–495.

Taylor, M. E., Delbaere, K., Mikolaizak, A. S., Lord, S. R., and Close, J. C. (2013). Gait parameter risk factors for falls under simple and dual task conditions in cognitively impaired older people. *Gait Posture* 37, 126–130. doi: 10.1016/j.gaitpost.2012.06.024

Volpe, B. T., Krebs, H. I., and Hogan, N. (2001). Is robot-aided sensorimotor training in stroke rehabilitation a realistic option? *Curr. Opin. Neurol.*14, 745–752. doi: 10.1097/00019052-200112000-00011

Welter, M. L., Do, M. C., Chastan, N., Torny, F., Bloch, F., du Montcel, S. T., et al. (2007). Control of vertical components of gait during initiation of walking in normal adults and patients with progressive supranuclear palsy. *Gait Posture* 26, 393–399. doi: 10.1016/j.gaitpost.2006.10.005

Winchester, P., and Querry, R. (2006). Robotic orthoses for body weight-supported treadmill training. *Phys. Med. Rehabil. Clin. N. Am.* 17, 159–172. doi: 10.1016/j.pmr.2005.10.008

Wirz, M., Zemon, D. H., Rupp, R., Scheel, A., Colombo, G., Dietz, V., et al. (2005). Effectiveness of automated locomotor training in patients with chronic incomplete spinal cord injury: a multicenter trial. *Arch. Phys. Med. Rehabil.* 86, 672–680. doi: 10.1016/j.apmr.2004.08.004

Zampieri, C., and Di Fabio, R. P. (2006). Progressive supranuclear palsy: disease profile and rehabilitation strategies. *Phys. Ther.* 86, 870–880.

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.

Received: 15 December 2013; paper pending published: 21 January 2014; accepted: 24 March 2014; published online: 17 April 2014.

Citation: Sale P, Stocchi F, Galafate D, De Pandis MF, Le Pera D, Sova I, Galli M, Foti C and Franceschini M (2014) Effects of robot assisted gait training in progressive supranuclear palsy (PSP): a preliminary report. Front. Hum. Neurosci. ***8****:207. doi: 10.3389/fnhum.2014.00207*

This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 18 February 2014
doi: 10.3389/fnhum.2014.00078

Associations between prefrontal cortex activation and H-reflex modulation during dual task gait

***Daan Meester*[1]*, *Emad Al-Yahya*[1,2], *Helen Dawes*[1], *Penny Martin-Fagg*[1] and *Carmen Piñon*[1]**

[1] *Movement Science Group, Department of Sport and Health Sciences, Faculty of Health and Life Sciences, Oxford Brookes University, Headington, Oxford, UK*
[2] *Department of Physiotherapy, Faculty of Rehabilitation, The University of Jordan, Amman, Jordan*

Edited by:
Marco Iosa, Fondazione Santa Lucia, Italy

Reviewed by:
Floriana Pichiorri, Fondazione Santa Lucia–Istituto Di Ricovero E Cura A Carattere Scientifico, Italy
Erik B. Simonsen, University of Copenhagen, Denmark

***Correspondence:**
Daan Meester, Movement Science Group, Department of Sport and Health Sciences, Faculty of Health and Life Sciences, Oxford Brookes University, Headington, Oxford OX3 0BP, UK
e-mail: dmeester@brookes.ac.uk

Walking, although a largely automatic process, is controlled by the cortex and the spinal cord with corrective reflexes modulated through integration of neural signals from central and peripheral inputs at supraspinal level throughout the gait cycle. In this study we used an additional cognitive task to interfere with the automatic processing during walking in order to explore the neural mechanisms involved in healthy young adults. Participants were asked to walk on a treadmill at two speeds, both with and without additional cognitive load. We evaluated the impact of speed and cognitive load by analyzing activity of the prefrontal cortex (PFC) using functional Near-Infrared Spectroscopy (fNIRS) alongside spinal cord reflex activity measured by soleus H-reflex amplitude and gait changes obtained by using an inertial measuring unit. Repeated measures ANOVA revealed that fNIRS Oxy-Hb concentrations significantly increased in the PFC with dual task (walking while performing a cognitive task) compared to a single task (walking only; $p < 0.05$). PFC activity was unaffected by increases of walking speed. H-reflex amplitude and gait variables did not change in response to either dual task or increases in walking speed. When walking under additional cognitive load participants adapted by using greater activity in the PFC, but this adaptation did not detrimentally affect H-reflex amplitude or gait variables. Our findings suggest that in a healthy young population central mechanisms (PFC) are activated in response to cognitive loads but that H-reflex activity and gait performance can successfully be maintained. This study provides insights into the mechanisms behind healthy individuals safely performing dual task walking.

Keywords: gait, dual task, fNIRS, H-reflex, motor control, prefrontal cortex

INTRODUCTION

Walking is a largely automatic process although it is controlled by the cortex, brain stem and spinal cord, and modulated through integration of neural signals from central and peripheral inputs at spinal and supraspinal level (Nielsen, 2003; Yang and Gorassini, 2006). Activation of cortical motor networks, including the motor, premotor, and prefrontal cortex (PFC) has been observed during walking (Fukuyama et al., 1997; Hanakawa et al., 1999). However, whilst it has been reported that cognitive tasks interfere with walking performance (Suzuki et al., 2004; Al-Yahya et al., 2011), the underlying mechanism of how cortical interference affects gait and mobility has not yet been described. Walking has been shown to be facilitated by selective moderation of central drive as a result of inhibitory activity by intracortical neurones which suppress motoneuronal activation (Petersen et al., 2001). This effect is apparent in the strong modulation of the soleus H-reflex throughout the gait cycle whereby the H-reflex decreases or is absent during the swing phase of gait, facilitating ankle dorsiflexion, and increases approaching heel contact and stance phases, thus assisting weight bearing (Yang and Gorassini, 2006; Makihara et al., 2012). The H-reflex is considered to provide valuable information on the involvement of the corticospinal tract in the control of peripheral reflexes and movement during walking (Knikou, 2008a,b). There is further evidence of phase-dependent soleus H-reflex modulation, observed during walking in patients with spinal cord injuries (Knikou et al., 2009), which supports the contribution from sensory afferents in walking control. Exploring gait parameters alongside H-reflex and cortical mechanisms may offer an insight into the mechanisms involved in gait control.

In this study we explored the impact of an additional cognitive task, which placed demands on the PFC (McCulloch, 2007), on walking at self-selected and fast walking speeds (Suzuki et al., 2004; Suzuki et al., 2008; Al-Yahya et al., 2009). We set out to investigate PFC activation and any consequential effects on the soleus H-reflex alongside gait performance. To date, a reduced H-reflex amplitude, indicating a depressed spinal excitability to improve stability, has been observed when performing an additional cognitive load during standing (Weaver et al., 2012); but the effect of cognitive load on neural mechanisms during walking has not been explored.

Furthermore, walking speed associated changes have been demonstrated in both central and peripheral mechanisms, where both the activity of the PFC (Suzuki et al., 2004), and H-reflex amplitude were shown to increase with higher walking speed (Simonsen et al., 2012).

We hypothesized that additional cognitive load would increase PFC activity and through projections from the PFC reduce the

H-reflex amplitude during normal walking speed. We further expected that increasing both speed and the cognitive load would provoke a further increased activity in the PFC and reduce speed related changes in the H-reflex amplitude (Petersen et al., 2009), with greater changes in the PFC associated with a reduced H-reflex during stance and greater alterations in gait parameters. As such, our study sets out to explore the mechanism behind healthy individuals safely performing dual task walking.

MATERIALS AND METHODS

Seventeen healthy subjects (7 men; 10 women), 15 right handed and 2 left handed, participated in this study. Mean age was 27.8 ± 6.3 with age range 22–44 years; mean height and weight were 1.75 ± .11 m and 69.1 ± 15.2 kg, respectively. All subjects gave written informed consent according to the Declaration of Helsinki before the start of the experiments and this study was approved by the University Research Ethics Committee. Subjects walked on a treadmill while concurrently performing a cognitive task at a normal and faster walking speed. H-reflexes were elicited in the right soleus and measures of fNIRS were performed on the PFC.

STUDY DESIGN

Standard methodology, utilizing several practice trials was used to familiarize participants with the treadmill and varying speeds (Woodway ELG 75, Germany) and thus determine preferred walking speed close to normal over ground walking speed (Voloshin, 2000). A faster walking speed was determined by increasing the normal walking speed by 20% (Voloshin, 2000).

The treadmill was programmed for five repetitions of walking and dual task walking alternated with rest periods in which the treadmill was stationary. Both walking and walking with distraction were performed in blocks of 30 s, and rest periods varied from 20 to 40 s. The rest periods had a varying length to prevent subjects anticipating the start of the next block. Subjects performed five repetitions of walking and walking with distraction at each of the two speeds. For the cognitive task, subjects were asked to count backward in steps of seven from a number presented by the investigator.

fNIRS IMAGING

A continuous wave (782 nm, 859 nm) fNIRS instrument (Oxymon, Artinis Medical Systems, The Netherlands) was used to measure PFC activation. Two identical plastic holders consisting of four optodes each (two sources, two detectors) in a 4-channel arrangement with an inter optode separation of 30 mm were placed on each participant's forehead using a custom-built spring-loaded array optode holder covering the area linking Fp1, F3, and F7 and the area linking Fp2, F4, and F8 according to the international 10–20 EEG electrode system, which corresponds to the left and the right PFC, respectively (Leff et al., 2008). To monitor hemodynamic responses, blood pressure, and heart rate were measured at baseline and at the end of the program.

H-REFLEXES

H-reflexes were elicited in the right soleus muscle (SOL) during single and dual task blocks. A constant current high voltage stimulator (Digitimer Ltd. DS7A, UK) was used to elicit H-reflexes and M-waves. H-reflex recruitment curves were obtained while subjects were standing. H_{max} and M_{max} were measured to determine the intensity which elicited 20–25% of M_{max} (Simonsen and Dyhre-Poulsen, 1999; Phadke et al., 2010). A footswitch (Odstock Medical Ltd, UK) under the subject's right heel provided data to time the stimulation within the gait cycle. The footswitch was used to trigger the stimulator to elicit a H-reflex during mid-stance (30% of gait cycle; Hughes and Jacobs, 1979). To prevent depression of the H-reflex and subject anticipation of the reflex, stimulations were given every four, five, or six heel strikes; corresponding to an inter-stimulus-time (ISI) of 4–5 s, which is known to be long enough to measure consecutive H-reflexes (Knikou and Taglianetti, 2006; Jeon et al., 2007).

EMG RECORDING AND NERVE STIMULATION

Based on earlier research (Capaday and Stein, 1986, 1987; Simonsen and Dyhre-Poulsen, 1999), the right SOL was selected for EMG recordings. Ag–AgCl electrodes (55 mm diameter) were placed on the muscle belly and as a stimulating electrode on the tibial nerve (Konrad, 2005). The cathode was placed in the popliteal fossa with the anode at a distance of 2 cm medial to the cathode. Researchers located the nerve using small moveable electrodes, before positioning the actual stimulation electrodes, which were secured with Velcro tape to prevent slippage during locomotion. EMG leads were attached to the leg and upper body to reduce movement artifacts and prevent subjects from tripping.

STEP TIME

Step time was measured using an inertial measuring unit (Philips, Eindhoven, The Netherlands) comprising a tri-axial accelerometer, gyroscope, and magnetometer placed on the center of mass (Esser et al., 2012). Post-processing and analysis was performed in a pre-written program in LabVIEW2010 (National Instruments, Austin, TX, USA). Step time was taken as the gait variable of interest with the time interval between trough-to-trough center of mass excursions during one gait cycle (Esser et al., 2009).

DATA PROCESSING

Raw fNIRS signals were collected at a sample rate of 10 Hz. Deoxy-Hb and Oxy-Hb concentrations were calculated (Oxysoft 2.1.6), filtered with a low pass filter set at.67 Hz (Labview 6.1) and visually inspected for motion artifacts, missing signals, and noisy signals. Blocks with missing signals or artifacts were excluded from analysis. A moving average filter with a width of 4 s was used to smooth the signal. Block averages of the 5 task + rest repetitions were calculated and the middle 10 s of each task and rest periods used for statistical analyses. To offset low spatial resolution of fNIRS, and provide a better indication of general measured activity in the PFC, the four channels on both the left PFC and the right PFC were averaged.

Signal software was used for data acquisition and analysis (CED Signal 3.09, UK). EMG signals were pre amplified 1000 times and high passed filtered at 30 Hz (NL844; Digitimer). Consequently signals were low pass filtered at 200 Hz (NL135; Digitimer) before H-reflexes were sampled at 1000 Hz (Tokuno et al., 2007). Peak-to-peak amplitude of the H-reflex measured during walking was

normalized by expressing the walking H-reflex as a percentage of the standing H-reflex elicited at the same intensity. Variability of normalized H-reflexes was determined using the standard deviation.

STATISTICS

Descriptive statistics were performed on demographic and gait control parameters. Paired *t*-tests were used to examine differences in hemoglobin concentrations during task and rest blocks. The effects of task and speed on brain measures, H-reflex amplitudes and step times were examined using repeated measures ANOVA models. To investigate relationships between central and peripheral mechanisms, changes in Oxy-Hb concentrations, H-reflex amplitude variability, and step time variability were explored through Pearson correlations. For all statistical tests, alpha level was set at 0.05 a priori, and SPSS Bonferroni adjusted *p*-values are quoted.

RESULTS

DESCRIPTIVES

Individuals' average self-selected normal walking speed was 1.22 ± SD 0.24 m/s, range 0.7–1.5 m, and faster walking speed was 1.48 ± 0.26 m/s, range 1.0–1.7 m/s. Blood pressure and heart rate were stable with a mean blood pressure of 117 ± 10.6/75 ± 6.7 mmHg, range 98/63 mmHg to 136/89 mmHg and a mean heart rate of 75 ± 12.3 bpm, range 60–109 bpm. Blood pressure and heart rate did not significantly ($p > 0.05$) change from baseline to normal and faster walking speed. Cognitive task score was not significantly ($p > 0.05$) different between speeds. Average answer rate was 10.3 ± 3.8 answers during normal walking and 10.5 ± 3.8 during faster walking with, respectively, mean error rates of 0.4 ± 0.4 and 0.4 ± 0.3.

NIRS IMAGING

Average Oxy-Hb and Deoxy-Hb concentrations are summarized in **Figures 1** and **2**. Repeated measures ANOVA results are shown in **Table 1**. For single and dual task blocks at normal and faster walking speed, relative Oxy-Hb concentrations were significantly ($p < 0.05$) higher during the task compared to the average rest block followed after each task in both hemispheres. Deoxy-Hb changes were significantly (0.011) lower during dual task blocks compared to rest in the right PFC when walking at a faster walking speed.

In the right cortex Oxy-Hb concentrations increased significantly with dual task ($F = 4.632$; $p = 0.049$) from 0.23 ± 0.1 mmol/l to 0.34 ± 0.1 mmol/l at normal speed and from 0.21 ± 0.1 to 0.51 ± 0.1 at faster speed. In the left cortex, a trend was shown toward significant increases ($F = 3.535$; $p = 0.080$) of Oxy-Hb concentrations with dual task, with increases from 0.23 ± 0.1 mmol/l to 0.38 ± 0.1 mmol/l and 0.22 ± 0.1 mmol/l to 0.46 ± 0.1 mmol/l for normal and faster walking speed, respectively. Deoxy-Hb concentrations were not significantly affected by task or speed. Increases and decreases in Oxy-Hb and Deoxy-Hb were not significantly different between speeds. No significant interactions were found between task and speed for either Oxy-Hb and Deoxy-Hb concentrations.

H-REFLEX AND STEP TIME

Averages and variability of H-reflex amplitudes and step times are described in **Table 2** and **Figure 3**. Changes of mean normalized H-reflex, step time, and variability of H-reflex and step time were not significantly ($p > 0.05$) different between tasks and walking speeds. Furthermore repeated measures ANOVAs did not show interactions between task and speed for both parameters (see **Table 3**).

CORRELATIONS BETWEEN CENTRAL AND PERIPHERAL MEASURES

No significant relationships were found between PFC activity, H-reflex, and step times. Changes in Oxy-Hb concentrations did not correlate with H-reflex variability and step time variability. Changes in Oxy-Hb concentrations in the left cortex due to dual

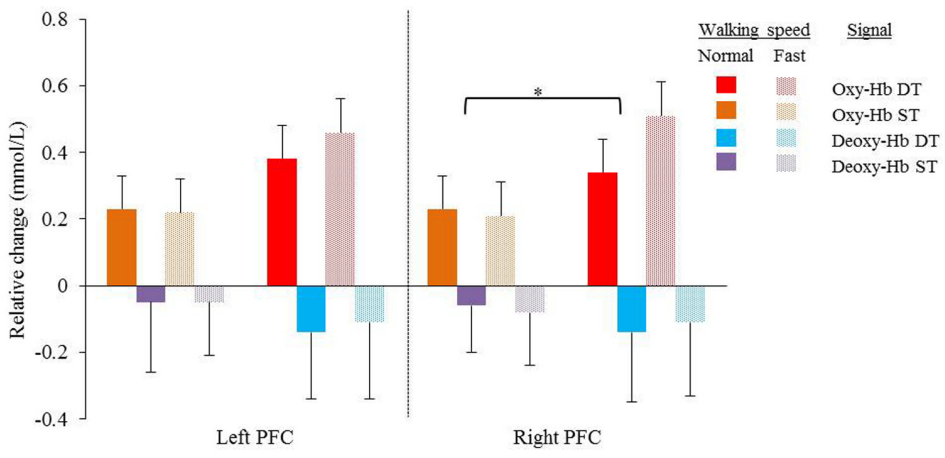


FIGURE 1 | Mean relative changes and standard deviations in Oxy-Hb (red and orange) and Deoxy-Hb (purple and blue) during normal and fast (dotted bars) walking in the left and right cortex. Results of single task (orange and purple) and dual task walking (red and blue) are presented. PFC = prefrontal cortex, Oxy-Hb = oxy hemoglobin, Deoxy-Hb = deoxy hemoglobin, ST = single task, DT = dual task. Significant higher Oxy-Hb concentration change during dual task walking compared to single task walking in the right cortex (*$p = 0.049$).

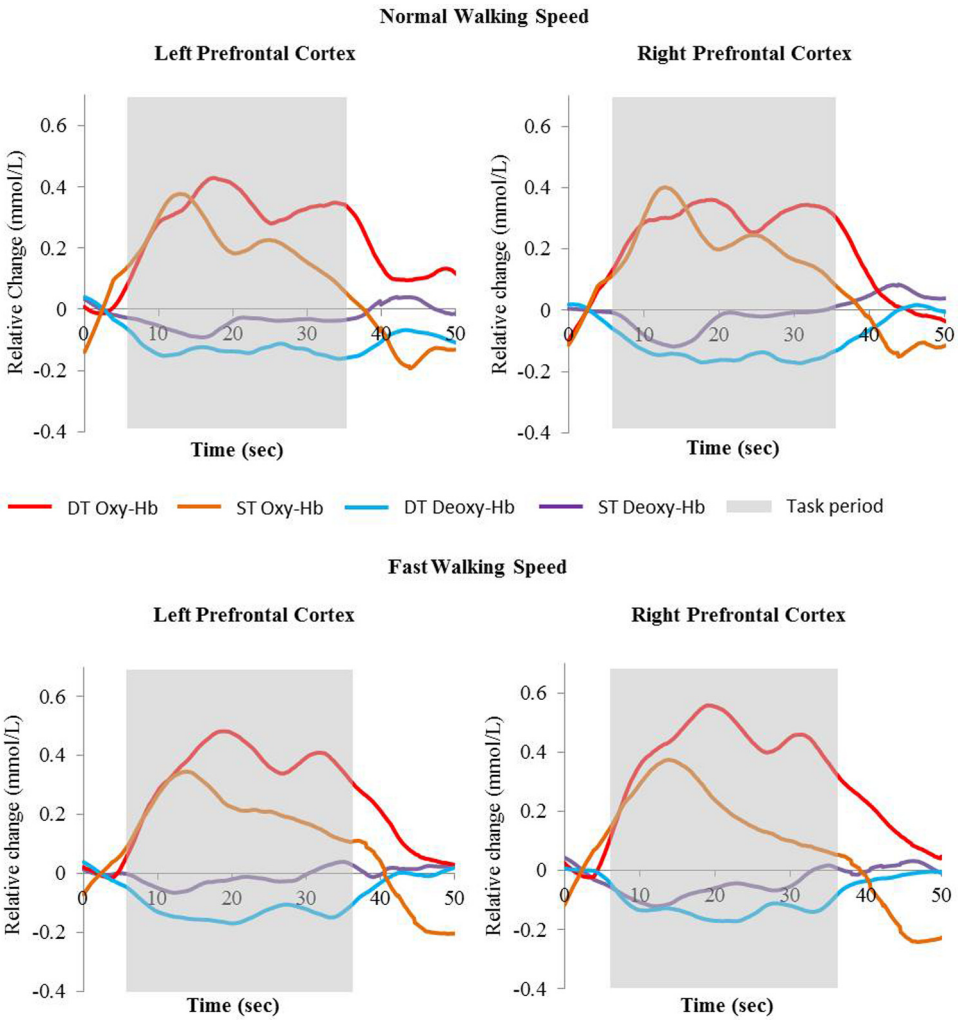


FIGURE 2 | Relative change of Oxy-Hb (red and orange) and Deoxy-Hb (blue and purple) concentrations during single task walking and dual task walking at normal and fast walking speed. Task period are indicated in gray. Oxy-Hb = oxyhemoglobin, Deoxy-Hb = deoxyhemoglobin, ST = single task, DT = dual task.

Table 1 | Repeated measures showing the effect of task and speed on Oxy hemoglobin and Deoxy hemoglobin concentrations in the left and right prefrontal cortex.

Summary statistics of ANOVA for Oxy and Deoxy hemoglobin concentrations

	Left PFC hemisphere				Right PFC hemisphere			
	Oxy-Hb		Deoxy-Hb		Oxy-Hb		Deoxy-Hb	
Effect	***F***	***Sig.***	***F***	***Sig.***	***F***	***Sig.***	***F***	***Sig.***
Task	3.535	0.080	3.396	0.085	4.632	**0.049***	2.107	0.169
Speed	0.213	0.651	0.188	0.736	1.776	0.204	0.045	0.835
Task*Speed	0.471	0.503	0.076	0.786	2.425	0.142	1.231	0.286

*Task = single and dual task walking, speed = normal and faster walking speed, PFC = prefrontal cortex, Oxy-Hb = oxy hemoglobin, Deoxy-Hb = deoxy hemoglobin. Significant higher Oxy-Hb concentration change during dual task walking compared to single task walking in the right prefrontal cortex (*p = 0.049).*

Table 2 | Averages + standard deviations of H-reflex amplitude, variability, step times, and step time variability.

H-reflex and step time averages and variability				
	Normal walking speed		Fast walking speed	
	Single task	Dual task	Single task	Dual task
H-reflex (%)	103.7 ± 24.4	105.9 ± 25.5	109.0 ± 26.9	106.7 ± 33.2
H-reflex variability (%)	14.2 ± 7.8	18.1 ± 10.3	16.9 ± 12.5	17.6 ± 11.3
Step time (ms)	528.4 ± 41.3	532.4 ± 46.1	524.3 ± 39.8	517.6 ± 38.3
Step time variability (ms)	105.0 ± 134.1	124.6 ± 139.4	63.4 ± 81.3	54.1 ± 48.5

Variability of the normalized H-reflex and step time was measured using the standard deviation.

Table 3 | Repeated measures showing the effect of task and speed on normalized H-reflex, H-reflex variability, step time, and step time variability.

Summary statistics of ANOVA for H-reflex amplitudes and step times								
	H-reflex		H-reflex variability		Step time		Step time variability	
Effect	***F***	***Sig.***	***F***	***Sig.***	***F***	***Sig.***	***F***	***Sig.***
Task	0.001	0.973	2.266	0.153	0.966	0.341	1.436	0.251
Speed	0.868	0.366	0.376	0.549	0.108	0.746	0.205	0.658
Task*Speed	0.951	0.345	2.255	0.154	3.339	0.088	3.387	0.087

Variability of the normalized H-reflex and step time was measured using the standard deviation.

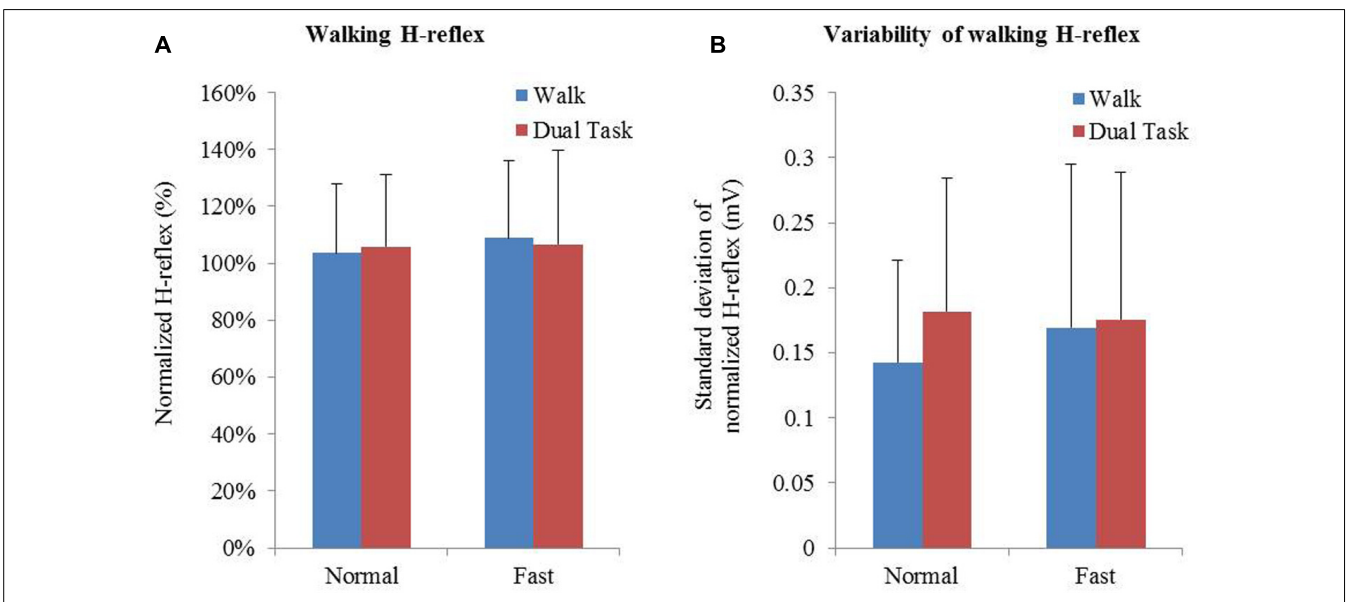


FIGURE 3 | (A) Means and standard deviation of normalized H-reflex. **(B)** Mean variability of H-reflex+standard deviation. Variability of the normalized H-reflex and step time was measured using the standard deviation.

task and changes in step time showed the highest correlation of 0.420 close toward a trend; $p = 0.11$. Error rates of cognitive task performance were not correlated with significantly higher or lower concentrations of Oxy-Hb or Deoxy-Hb or changes due to single and dual task. Moreover changes in H-reflex variability did not correlate with step time variability.

DISCUSSION AND CONCLUSION

We found healthy young adults responded to additional cognitive loading during treadmill walking with increased PFC activation, but unlike individuals after stroke or the elderly, this activation was not associated with altered gait parameters (Al-Yahya, 2011). Further, there was no change in the amplitude of the H-reflex during stance in either fast or dual task walking conditions. It was hypothesized that the H-reflex amplitude would reduce during the stance phase of walking when participants were simultaneously performing a cognitive task. However, whilst we observed no change in amplitude of the reflex there was a trend toward increase in H-reflex amplitude variability under both fast and dual task walking conditions. In earlier studies Capaday and Stein (1986, 1987) found decreases in H-reflex amplitudes from standing to walking and with increasing walking speed throughout the gait cycle, whereas Simonsen and Dyhre-Poulsen (1999), Simonsen et al. (2012) showed increases in H-reflex amplitudes from walking to running. In agreement with these inconsistent results this study confirmed that there is no clear direction in which the H-reflex amplitude is altered, but that an increased walking task difficulty by speed or dual task may increase the variability of the H-reflex amplitude. The absence of changes in gait parameters between different walking conditions indicates that young healthy individuals are able to cope with additional cognitive loads and changes in speed during walking. Therefore it is proposed that the observed increases in PFC activity allowed individuals to perform additional tasks simultaneously, without affecting cortical output onto the measured peripheral reflexes and thus gait control. When exploring correlations between dual task changes in PFC activation and step time, the highest Pearson r^2 found was 0.420 which was not significant ($p = 0.11$). This indicates that in a healthy young population central mechanisms are activated in response to cognitive loads but that reflex activity and gait performance can successfully be maintained.

Our findings are important as they set out a non-pathological response of reflex control alongside central adaptations to cognitive load in healthy young adults at both self-selected and fast walking speeds. Previous studies (Capaday and Stein, 1986, 1987; Chalmers and Knutzen, 2000; Ferris et al., 2001) have found both decreases and increases in H-reflex amplitude with increases in walking speed (Simonsen and Dyhre-Poulsen, 1999; Schneider et al., 2000; Simonsen et al., 2012) which suggest that central control mechanisms are involved in H-reflex pathways during activities like walking. The stance phase of gait is important for stability and propulsion during gait and thus of importance to understand mechanisms affecting balance. Our study found a very stable H-reflex during the stance phase of walking within younger subjects. The swing phase of the gait cycle shows a different response which could now be explored in dual task walking conditions. Our findings suggest that central control, measured with prefrontal activation changes, occurred in response to altered walking demands but that these did not affect peripheral reflexes, as measured by the H-reflex through supra-spinal cortical outputs directly controlling motor neuron excitability. Increased impact of cognitive load has been shown during backward walking (Kurz et al., 2012) and during dual tasking in older adults (Seidler et al., 2011). The increase in associated gait decrements in the older populations, particularly those with neurological damage, suggests an age-related shift from automatic to attentional control of movement as walking ability declines (Seidler et al., 2011). Investigation of the impact on both PFC and H-reflex in the older population may elucidate the mechanism behind this behavioral response.

In our younger population, no significant changes in PFC activity were found with increased speed, suggesting there might be differences in control mechanisms of faster speeds, or greater capacity for adaptation in younger population. Importantly peripheral changes were not related to cortical changes. This supports the hypothesis that in healthy individuals there is adequate central capacity to cope with subtle changes in walking and that any peripheral changes may be minimal and separately mediated.

The methodologies used in this study do have some limitations. fNIRS is a developing modality with great opportunities (Belda-Lois et al., 2011), but it also has a poor spatial resolution, low depth penetration and is variable with regards to signal quality between individuals (Toronov et al., 2007; Seraglia et al., 2011). Furthermore due to practical reasons we only measured the PFC, and were limited due to patient comfort in our testing time thus limiting our ability to test the H-reflex throughout the gait cycle and from exploring other motor networks (Suzuki et al., 2004; Kurz et al., 2012; Karim et al., 2013) which may provide further insight into gait and balance control. Our study may be underpowered and prone to type II error. The use of the H-reflex in order to explore walking has inherent practical challenges of using the appropriate intensity of stimulus, timing of the stimulus (Simonsen et al., 2013), protocol (Mynark, 2005), and control of the amount of body weight (Hwang et al., 2011); however, we used techniques with established reliability (Simonsen and Dyhre-Poulsen, 2011; König et al., 2013). Nevertheless the H-reflex has been shown to be effective in exploring the normal response to postural threat, and perhaps by measuring the reflex in mid stance changes at heel strike or other areas of the gait cycle were missed (Krauss and Misiaszek, 2007). Although gait control has not been explored before, differences in the H-reflex during upright stance have been found between the elderly and young in balance responses (Baudry et al., 2010; Baudry and Duchateau, 2012). Considering the high variability in H-reflex and fNIRS between participants, the measures used, although normalized, may not have been sensitive enough to pick up correlations between central and peripheral mechanisms. However, it is important to replicate this research in elderly individuals and neurological populations to explore relationships between the mechanisms in those populations.

We used a treadmill for our study, which is not reflective of an overground walking and normal walking control, since individuals

were unable to respond to simultaneous cognitive demand by slowing down (Al-Yahya et al., 2009). Although previous studies using treadmill testing have shown changes in gait parameters, the method may lack some ecological validity for understanding gait control for community mobility. Our population selected an average walking speed of 1.2 ms^{-1} which is lower than the average walking speed for this age group (Bohannon and Williams Andrews, 2011) resulting in fast walking speeds, set at 120% of normal walking speed, which were more reflective of a normal walking pace.

Our results have shown that cognitive load does increase activity in the PFC but this is not associated with a change in H-reflex modulation during stance or gait parameters. Gait control mechanisms under speed and dual task conditions now need to be explored in older adults, and people prone to falls or poor balance and mobility.

ACKNOWLEDGMENTS

We would like to thank members of the Movement Science Group of Oxford Brookes University for their input in setting up this research. We would also like to thank the participants who took part. Furthermore we would like to acknowledge the funding support from the Elizabeth Casson Trust for Prof. Helen Dawes and the Stroke Association for Mr. Daan Meester.

REFERENCES

Al-Yahya, E. (2011). *Neural Correlates of Cognitive Motor Interference While Walking*. Ph.D. Thesis, Oxford Brookes University, Oxford.

Al-Yahya, E., Dawes, H., Collett, J., Howells, K., Izadi, H., Wade, D. T., et al. (2009). Gait adaptations to simultaneous cognitive and mechanical constraints. *Exp. Brain Res.* 199, 39–48. doi: 10.1007/s00221-009-1968-1

Al-Yahya, E., Dawes, H., Smith, L., Dennis, A., Howells, K., and Cockburn, J. (2011). Cognitive motor interference while walking: a systematic review and meta-analysis. *Neurosci. Biobehav. Rev.* 35, 715–728. doi: 10.1016/j.neubiorev.2010.08.008

Baudry, S., and Duchateau, J. (2012). Age-related influence of vision and proprioception on Ia presynaptic inhibition in soleus muscle during upright stance. *J. Physiol.* 590, 5541–5554. doi: 10.1113/jphysiol.2012.228932.

Baudry, S., Maerz, A. H., and Enoka, R. M. (2010). Presynaptic modulation of Ia afferents in young and old adults when performing force and position control. *J. Neurophysiol.* 103, 623–631. doi: 10.1152/jn.00839.2009

Belda-Lois, J.-M., Mena-Del Horno, S., Bermejo-Bosch, I., Moreno, J. C., Pons, J. L., Farina, D., et al. (2011). Rehabilitation of gait after stroke: a review towards a top-down approach. *J. Neuroeng. Rehabil.* 8, 66. doi: 10.1186/1743-0003-8-66.

Bohannon, R. W., and Williams Andrews, A. (2011). Normal walking speed: a descriptive meta-analysis. *Physiotherapy* 97, 182–189. doi: 10.1016/j.physio.2010.12.004.

Capaday, B. Y. C., and Stein, R. B. (1986). Amplitude modulation of the soleus H-reflex in the human during walking and standing. *J. Neurosci.* 6, 1308–1313.

Capaday, B. Y. C., and Stein, R. B. (1987). Difference in the amplitude of the human soleus h reflex during walking and running. *J. Physiol.* 392, 513–522.

Chalmers, G. R., and Knutzen, K. M. (2000). Soleus Hoffmann-reflex modulation during walking in healthy elderly and young adults. *J. Gerontol. A Biol. Sci. Med. Sci.* 55, B570–B579. doi: 10.1093/gerona/55.12.B570

Esser, P., Dawes, H., Collett, J., Feltham, M. G., and Howells, K. (2012). Validity and inter-rater reliability of inertial gait measurements in Parkinson's disease: a pilot study. *J. Neurosci. Methods* 205, 177–181. doi: 10.1016/j.jneumeth.2012.01.005

Esser, P., Dawes, H., Collett, J., and Howells, K. (2009). IMU: inertial sensing of vertical CoM movement. *J. Biomech.* 42, 1578–1581. doi: 10.1016/j.jbiomech.2009.03.049

Ferris, D. P., Aagaard, P., Simonsen, E. B., Farley, C. T., and Dyhre-Poulsen, P. (2001). Soleus H-reflex gain in humans walking and running under simulated reduced gravity. *J. Physiol.* 530, 167–180. doi: 10.1111/j.1469-7793.2001.0167m.x

Fukuyama, H., Ouchi, Y., Matsuzaki, S., Nagahama, Y., Yamauchi, H., Ogawa, M., et al. (1997). Brain functional activity during gait in normal subjects: a SPECT study. *Neurosci. Lett.* 228, 183–186. doi: 10.1016/S0304-3940(97)00381-9

Hanakawa, T., Katsumi, Y., Fukuyama, H., Honda, M., Hayashi, T., Kimura, J., et al. (1999). Mechanisms underlying gait disturbance in Parkinson's disease: a single photon emission computed tomography study. *Brain* 122(Pt 7), 1271–1282. doi: 10.1093/brain/122.7.1271

Hughes, J., and Jacobs, N. (1979). Normal human locomotion. *Prosthet. Orthot. Int.* 3, 4–12. doi: 10.3109/03093647909164693

Hwang, S., Jeon, H. -S., Kwon, O. -Y., and Yi, C. -H. (2011). The effects of body weight on the soleus H-reflex modulation during standing. *J. Electromyogr. Kinesiol.* 21, 445–449. doi: 10.1016/j.jelekin.2010.11.002

Jeon, H.-S., Kukulka, C. G., Brunt, D., Behrman, A. L., and Thompson, F. J. (2007). Soleus H-reflex modulation and paired reflex depression from prone to standing and from standing to walking. *Int. J. Neurosci.* 117, 1661–1675. doi: 10.1080/00207450601067158

Karim, H., Schmidt, B., Dart, D., Nancy, B., and Huppert, T. (2013). Functional near-infrared spectroscopy (fnirs) of brain function during active balancing using a video game system. *Gait Posture* 35, 367–372. doi: 10.1016/j.gaitpost.2011.10.007

Knikou, M. (2008a). The H-reflex as a probe: pathways and pitfalls. *J. Neurosci. Methods* 171, 1–12. doi: 10.1016/j.jneumeth.2008.02.012

Knikou, M. (2008b). The H-reflex as a probe: pathways and pitfalls. *J. Neurosci. Methods* 171, 1–12. doi: 10.1016/j.jneumeth.2008.02.012

Knikou, M., Angeli, C. A., Ferreira, C. K., and Harkema, S. J. (2009). Soleus H-reflex modulation during body weight support treadmill walking in spinal cord intact and injured subjects. *Exp. Brain Res.* 193, 397–407. doi: 10.1007/s00221-008-1636-x

Knikou, M., and Taglianetti, C. (2006). On the methods employed to record and measure the human soleus H-reflex. *Somatosen. Mot. Res.* 23, 55–62. doi: 10.1080/08990220600702715

König, N., Reschke, A., Wolter, M., Müller, S., Mayer, F., and Baur, H. (2013). Plantar pressure trigger for reliable nerve stimulus application during dynamic H-reflex measurements. *Gait Posture* 37, 637–639. doi: 10.1016/j.gaitpost.2012.09.021

Konrad, P. (2005). *The ABC of EMG*. Scottsdale: Noraxon U.S.A. Inc., 1–60.

Krauss, E. M., and Misiaszek, J. E. (2007). Phase-specific modulation of the soleus H-reflex as a function of threat to stability during walking. *Exp. Brain Res.* 181, 665–672. doi: 10.1007/s00221-007-0962-8

Kurz, M. J., Wilson, T. W., and Arpin, D. J. (2012). Stride-time variability and sensorimotor cortical activation during walking. *Neuroimage* 59, 1602–1607. doi: 10.1016/j.neuroimage.2011.08.084

Leff, D. R., Elwell, C. E., Orihuela-Espina, F., Atallah, L., Delpy, D. T., Darzi, A. W., et al. (2008). Changes in prefrontal cortical behaviour depend upon familiarity on a bimanual co-ordination task: an fNIRS study. *Neuroimage* 39, 805–813. doi: 10.1016/j.neuroimage.2007.09.032

Makihara, Y., Segal, R. L., Wolpaw, J. R., and Thompson, A. K. (2012). H-reflex Modulation in the human medial and lateral gastrocnemii during standing and walking. *Muscle Nerve* 45, 116–125. doi: 10.1002/mus.22265.H-reflex

McCulloch, K. (2007). Attention and dual-task conditions: physical therapy implications for individuals with acquired brain injury. *J. Neurol. Phys. Ther.* 31, 104–118. doi: 10.1097/NPT.0b013e31814a6493

Mynark, R. G. (2005). Reliability of the soleus H-reflex from supine to standing in young and elderly. *Clin. Neurophysiol.* 116, 1400–1404. doi: 10.1016/j.clinph.2005.02.001

Nielsen, J. B. (2003). How we walk: central control of muscle activity during human walking. *Neuroscientist* 9, 195–204.

Petersen, N. T., Butler, J. E., Marchand-Pauvert, V., Fisher, R., Ledebt, A., Pyndt, H. S., et al. (2001). Suppression of EMG activity by transcranial magnetic stimulation in human subjects during walking. *J. Physiol.* 537, 651–656. doi: 10.1111/j.1469-7793.2001.00651.x

Petersen, T., Rosenberg, K., Petersen, N., and Nielsen, J. (2009). Cortical involvement in anticipatory postural reactions in man. *Exp. Brain Res.* 193, 161–171. doi: 10.1007/s00221-008-1603-6

Phadke, C. P., Klimstra, M., Zehr, E. P., Thompson, F. J., and Behrman, A. L. (2010). Soleus h-reflex modulation during stance phase of walking with altered arm swing patterns. *Motor Control* 14, 116–125.

Schneider, C., Lavoie, B. A., and Capaday, C. (2000). On the origin of the soleus H-reflex modulation pattern during human walking and its task-dependent differences. *J. Neurophysiol.* 83, 2881–2890.

Seidler, R. D., Bernard, J. A., Burutolu, T. B., Fling, B. W., Gordon, M. T., Gwin, J. T., et al. (2011). Motor control and aging: links to age-related brain structural, functional, and biochemical effects. *Neurosci. Biobehav. Rev.* 34, 721–733. doi: 10.1016/j.neubiorev.2009.10.005

Seraglia, B., Gamberini, L., Priftis, K., Scatturin, P., Martinelli, M., and Cutini, S. (2011). An exploratory fNIRS study with immersive virtual reality: a new method for technical implementation. *Front. Hum. Neurosci.* 5:176. doi: 10.3389/fnhum.2011.00176

Simonsen, E. B., Alkjær, T., and Raffalt, P. C. (2012). Reflex response and control of the human soleus and gastrocnemius muscles during walking and running at increasing velocity. *Exp. Brain Res.* 219, 163–174. doi: 10.1007/s00221-012-3075-y

Simonsen, E. B., Alkjær, T., and Raffalt, P. C. (2013). Influence of stimulus intensity on the soleus H-reflex amplitude and modulation during locomotion. *J. Electromyogr. Kinesiol.* 23, 438–442. doi: 10.1016/j.jelekin.2012.10.019

Simonsen, E. B., and Dyhre-Poulsen, P. (1999). Amplitude of the human soleus H reflex during walking and running. *J. Physiol.* 515 (Pt 3), 929–939. doi: 10.1111/j.1469-7793.1999.929ab.x

Simonsen, E. B., and Dyhre-Poulsen, P. (2011). Test-retest reliability of the soleus H-reflex excitability measured during human walking. *Hum. Mov. Sci.* 30, 333–340. doi: 10.1016/j.humov.2010.02.009

Suzuki, M., Miyai, I., Ono, T., and Kubota, K. (2008). Activities in the frontal cortex and gait performance are modulated by preparation. An fNIRS study. *Neuroimage* 39, 600–607. doi: 10.1016/j.neuroimage.2007.08.044

Suzuki, M., Miyai, I., Ono, T., Oda, I., Konishi, I., Kochiyama, T., et al. (2004). Prefrontal and premotor cortices are involved in adapting walking and running speed on the treadmill: an optical imaging study. *Neuroimage* 23, 1020–1026. doi: 10.1016/j.neuroimage.2004.07.002

Tokuno, C. D., Carpenter, M. G., Thorstensson, A., Garland, S. J., and Cresswell, A. G. (2007). Control of the triceps surae during the postural sway of quiet standing. *Acta Physiol. (Oxf.)* 191, 229–236. doi: 10.1111/j.1748-1716.2007.01727.x

Toronov, V. Y., Zhang, X., and Webb, A. G. (2007). A spatial and temporal comparison of hemodynamic signals measured using optical and functional magnetic resonance imaging during activation in the human primary visual cortex. *Neuroimage* 34, 1136–1148. doi: 10.1016/j.neuroimage.2006.08.048

Voloshin, A. (2000). The influence of walking speed on dynamic loading on the human musculoskeletal system. *Med. Sci. Sports Exerc.* 32, 1156–1159. doi: 10.1097/00005768-200006000-00019

Weaver, T. B., Janzen, M. R., Adkin, A. L., and Tokuno, C. D. (2012). Changes in spinal excitability during dual task performance. *J. Mot. Behav.* 44, 289–294. doi: 10.1080/00222895.2012.702142

Yang, J. F., and Gorassini, M. (2006). Spinal and brain control of human walking: implications for retraining of walking. *Neuroscientist* 12, 379–389. doi: 10.1177/1073858406292151

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.

Received: 20 Nov 2013; accepted: 31 Jan 2014; published online: 18 February 2014.
Citation: Meester D, Al-Yahya E, Dawes H, Martin-Fagg P and Piñon C (2014) Associations between prefrontal cortex activation and H-reflex modulation during dual task gait. Front. Hum. Neurosci. ***8****:78. doi: 10.3389/fnhum.2014.00078*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 08 May 2014
doi: 10.3389/fnhum.2014.00243

Gait training with real-time augmented toe-ground clearance information decreases tripping risk in older adults and a person with chronic stroke

Rezaul K. Begg[1]*, Oren Tirosh[1], Catherine M. Said[2,3], W. A. Sparrow[1], Nili Steinberg[1,4], Pazit Levinger[1] and Mary P. Galea[5]

[1] *Gait and Balance Research Group, College of Sport and Exercise Science, Institute of Sport, Exercise and Active Living, Victoria University, Melbourne, VIC, Australia*
[2] *Physiotherapy Department, Austin Health, Melbourne, VIC, Australia*
[3] *Physiotherapy, The University of Melbourne, Melbourne, VIC, Australia*
[4] *Wingate College of Physical Education and Sport Sciences, Wingate Institute, Netanya, Israel*
[5] *Department of Medicine (Royal Melbourne Hospital), The University of Melbourne, Parkville, Melbourne, VIC, Australia*

Edited by:
Marco Iosa, Fondazione Santa Lucia, Italy

Reviewed by:
Federica Tamburella, I.R.C.C.S. Fondazione Santa Lucia, Italy
Roberta Annicchiarico, Fondazione Santa Lucia, Italy

****Correspondence:***
Rezaul K. Begg, Gait and Balance Research Group, Institute of Sport, Exercise and Active Living, Victoria University, PO Box 14428, Melbourne, VIC 8001, Australia
e-mail: rezaul.begg@vu.edu.au

Falls risk increases with ageing but is substantially higher in people with stroke. Tripping-related balance loss is the primary cause of falls, and Minimum Toe Clearance (MTC) during walking is closely linked to tripping risk. The aim of this study was to determine whether real-time augmented information of toe-ground clearance at MTC can increase toe clearance, and reduce tripping risk. Nine healthy older adults (76 ± 9 years) and one 71 year old female stroke patient participated. Vertical toe displacement was displayed in real-time such that participants could adjust their toe clearance during treadmill walking. Participants undertook a session of unconstrained walking (no-feedback baseline) and, in a subsequent Feedback condition, were asked to modify their swing phase trajectory to match a "target" increased MTC. Tripping probability (PT) pre- and post-training was calculated by modeling MTC distributions. Older adults showed significantly higher mean MTC for the post-training retention session (27.7 ± 3.79 mm) compared to the normal walking trial (14.1 ± 8.3 mm). The PT on a 1 cm obstacle for the older adults reduced from 1 in 578 strides to 1 in 105,988 strides. With gait training the stroke patient increased MTC and reduced variability (baseline 16 ± 12 mm, post-training 24 ± 8 mm) which reduced obstacle contact probability from 1 in 3 strides in baseline to 1 in 161 strides post-training. The findings confirm that concurrent visual feedback of a lower limb kinematic gait parameter is effective in changing foot trajectory control and reducing tripping probability in older adults. There is potential for further investigation of augmented feedback training across a range of gait-impaired populations, such as stroke.

Keywords: gait, augmented feedback, toe-clearance, tripping, stroke

INTRODUCTION

The World Health Organization reports that falls injuries are the second leading cause of unintentional death after road accidents and are the major precursor to death in the elderly (WHO, 2002). Falls risk is considerably increased with ageing (Gillespie et al., 2003) but is substantially higher in stroke patients (AIHW, 2008; Batchelor et al., 2012). It is estimated that approximately 73% of stroke patients fall at least once in the first 6-months following discharge from inpatient rehabilitation (Forster and Young, 1995). The primary cause of falls-related injury is tripping (Cohen et al., 2003) due to unanticipated foot contact with ground-based objects, sufficient to irretrievably destabilize the individual (Nagano et al., 2011). Tripping is a hazard in the everyday environment either when walking over easily anticipated significant obstacles or lower but more frequently occurring surface irregularities, that may not always be accommodated by changes to swing limb trajectory. Goldie et al. (2000) found that in the homes of 22 stroke patients recently discharged from hospital, 2.5 to 4.4 obstacles were encountered for every 10 m of ambulation. Obstacles have been directly implicated in 10% of falls following stroke (Forster and Young, 1995). Foot trajectory during the swing phase of the gait cycle must not only maintain progression in the direction of travel, reflected in step length, but also incorporate a vertical displacement component sufficient to accommodate changes in support surface elevation.

Minimum Toe Clearance (MTC) is a critical event close to mid-swing in the walking gait cycle, when foot-ground clearance is minimal (1–2 cm). Low toe clearance at MTC has, therefore, been investigated as a predictor of tripping risk (Begg et al., 2007; Best and Begg, 2008). One gait adaptation to minimize tripping risk is to increase ground clearance at MTC while reducing variability. Recent work in our laboratory demonstrated that augmented vertical displacement information provided by projecting the real-time toe trajectory onto a screen (**Figure 1**), enabled

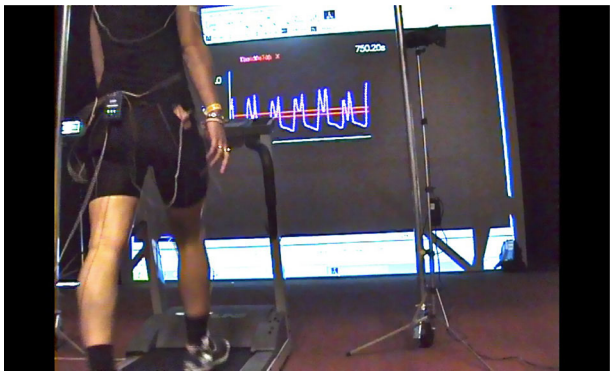


FIGURE 1 | Preferred-speed treadmill walking with visual augmented information of minimum toe clearance (MTC) provided using a real-time projection of the right foot sagittal trajectory. The MTC target band established in a baseline trial is shown as two parallel lines and MTC is the low point between the two trajectory peaks.

young participants to both increase MTC and decrease variability (Tirosh et al., 2013). This gait-specific movement information (i.e., ground clearance at MTC) represents a general class of information provided by an external source, which supplements the performer's intrinsic, task-specific sensory information. The effectiveness of such "augmented information" for changing movement-related characteristics is well known in motor behavior research and the proposal that augmented information of kinematic variables could be used to optimize motor performance has also been long-established (Newell et al., 1983). A further consideration in designing the present experiment was that motor learning is operationally defined not only by the "relative permanence" of performance in a later retention condition (e.g., Salmoni et al., 1984; Sparrow and Summers, 1992) but also evidence of transfer of training from one action, to another similar but unpracticed movement. In this paper we measured gait cycle characteristics and toe height at MTC in both limbs, i.e., both the trained and untrained limb, to determine any evidence of transfer to the untrained foot's movement characteristics.

It is increasingly recognized in gait research that adaptive locomotor control is dependent on complex interactions between the lower limbs, as reflected in stride phase variables that are unequal or "asymmetrical" (Nagano et al., 2013). A further motivation for examining both limbs simultaneously was that with both ageing and gait pathology there is evidence of compensatory adaptations that protect the walker from balance loss, reflected in "functional" or adaptive changes to gait cycle variables. With ageing it has been suggested that the dominant limb adopts a primary role of forward progression while the non-dominant limb serves to secure gait stability (e.g., Sadeghi et al., 2000). It was of interest to identify changes to gait cycle parameters with training consistent with functional adaptations that would secure balance while also facilitating progression.

Visual augmented information has been demonstrated to improve balance and gait across a range of clinical populations such as children with cerebral palsy and people with multiple sclerosis (Basmajian, 1981; Seeger et al., 1981; Baram and Miller, 2006; Boonstra et al., 2008; Langhorne et al., 2011). Augmented feedback has also been adapted to gait therapy in adults with hemiplegia (Batavia et al., 2001). A review by Van Vliet and Wulf (2006) of extrinsic feedback and motor learning after stroke concluded that biofeedback improved longer term performance compared to no-feedback conditions. A recent randomized control experiment with stroke patients assessed the effectiveness of visual feedback for balance training (Rao et al., 2012). These researchers used foot-ground reaction forces from a force plate as biofeedback and showed larger gains in balance test scores and functional independence measures for the experimental group compared to no-feedback controls. This recent study provides evidence of accelerated positive effects of feedback training for gait retraining and the restoration of walking ability after stroke. Providing feedback of toe vertical displacement at MTC during walking may facilitate treadmill-based gait training in both healthy older individuals and older adults who have sustained neurological deficits due to stroke.

The research questions addressed here were first, whether, relative to an initial baseline condition, healthy older participants could increase toe height at MTC with augmented information using the real-time visual presentation procedure devised by our research group (e.g., Begg et al., 2012; Tirosh et al., 2013). The second question to be addressed was the feasibility of translating these procedures to neurological rehabilitation by applying similar augmented training procedures to an older individual who had sustained a stroke. Evidence of the feedback's effectiveness would be demonstrated first by improved performance in a post-training retention trial when augmented information was withdrawn and, second, evidence of greater vertical toe displacement at MTC in the untrained limb (i.e., transfer of training).

MATERIALS AND METHODS

PARTICIPANTS

Nine healthy community living older adults (aged 76 ± 9 years) and one 71 year old female who had sustained a stroke participated in the experiment. The stroke patient was 21 years post left sided hemorrhage, which resulted in right sided hemiplegia. She walked with a single point stick (gait speed 0.37 m/s). She had a Step Test score of 6 with the affected limb supporting and 0 with the unaffected limb supporting. She was unable to perform a Timed Chair Stand Test and had a Stroke Rehabilitation Assessment of Movement Lower Limb sub score of 13/20. She had no spasticity in gastrocnemius or soleus. All participants gave their written informed consent using procedures mandated and approved by the Victoria University Human Research Ethics Committee. In addition, the protocol for the stroke participant was approved by the Austin Hospital Human Research Ethics Committee.

DATA ACQUISITION

All participants walked on a motor driven treadmill at preferred speed. Healthy older individuals performed three conditions within a single session; (i) 10 min walking (Baseline); (ii) 20 min walking with augmented MTC information (Feedback); (iii) 10 min walking with no feedback (Retention). The stroke patient completed training and testing over nine sessions, as

follows: (i) 5 min walking (Baseline); (ii) eight sessions over 4 weeks with augmented MTC information (Feedback) which included four sessions of continuous feedback for 5 min and four sessions faded feedback (4 min feedback and 2 min no feedback); (iii) 5 min walking with no feedback (Retention), immediately post-training.

In Feedback the right toe trajectory was projected in real-time on a screen in front of the participants with instructions to elevate the right toe when walking (i.e., increase ground clearance at MTC) by maintaining vertical displacement within an upper and lower bound (bandwidth) superimposed on the trajectory. The right foot was the stroke patient's affected limb. The displayed target bounds were calculated from the Baseline trajectory data; the lower bound was 1.5 × mean toe height at MTC in Baseline and the upper bound was the lower bound mean + 3 × standard deviations of toe height at MTC in Baseline. In Retention all participants were asked to reproduce the Feedback toe height at MTC but no trajectory display was provided.

Three-dimensional lower limb position data were sampled at 100 Hz using two Optotrak Certus (Northern Digital Inc.) motion analysis camera units positioned on either side of the treadmill. Optotrak infrared emitting diodes (IREDS) were placed on the distal extremity of the 5th metatarsal head (toe) and the heel. Participants wore their own comfortable shoes to which heel and toe IRED markers were attached, representing the anatomical location. A static data sample was obtained to provide reference coordinates. All participants wore a safety harness.

DATA ANALYSIS

The Optotrak system provided x-y-z spatial coordinates of each marker with the x-axis parallel to the walking direction (anterior–posterior), the z-axis vertical, i.e., perpendicular to the treadmill belt and the y-axis lateral and perpendicular to the x and z axes (medio-lateral). The swing phase events right toe off (RTO) and right heel strike (RHS) were identified by applying gait event algorithms using the heel and toe velocity and acceleration (Zeni et al., 2008). Toe height at MTC was computed as the toe vertical local minimum between the first maximum following toe-off and the second maximum of vertical displacement (Nagano et al., 2011). MTC toe height descriptive statistics (mean, standard deviation, skewness, and kurtosis) for both feet were computed. Obstacle contact frequency was calculated as the probability of tripping (PT) over a 1 cm high obstacle using MTC distribution modeling devised by Best and Begg (2008) and then compared pre- and post-training to assess the effect of training on toe height at MTC.

To determine condition and foot effects on toe clearance at MTC for the older group the gait variables were entered into two One-Way repeated measures Multivariate Analysis of Variance (MANOVA) procedure, with condition (Baseline, Retention) and foot (Left, Right) the within-subject factors. ANOVA F-ratios with probability <0.05 were accepted as statistically significant. All statistical analyses were undertaken using *Statistica* (StatSoft Inc.).

RESULTS

Table 1 presents stride-cycle data for the older group and the stroke patient at baseline and in the retention trial following gait training with augmented toe-height information. ANOVA results indicated that for the older participants none of the walking cycle variables in **Table 1** changed significantly from baseline to retention. In interpreting these results it is, however, worthwhile to note that these variables are influenced by walking velocity which was held constant across the two conditions (Older Group = 0.72 m/s; stroke patient = 0.28 m/s). Lower walking speed in the stroke patient was reflected in short steps of long duration relative to the older group. The stroke patient's hemiplegia was most clearly reflected in a right step (the affected limb) that was 10 cm longer than the left step. In addition, the stroke patient's right stance duration (as a percentage of stride time) was shorter (73%) than for the left foot (80%). The stroke patient's step width was 2–4 cm greater than the older group.

As illustrated in **Table 2** the older group showed similar toe height at MTC for the two feet in baseline. Following training, as expected, higher mean right toe vertical displacement at MTC was seen in Retention (35.5 mm) compared to baseline (14.4 mm), with the ANOVA results confirming that the difference between these two means reflected a significant condition effect [$F_{(1, 32)} = 6.96$, $p < 0.05$]. For the stroke patient, toe-ground clearance in the affected right limb was 13 mm lower than for the left toe in Baseline, while in Retention the unaffected and non-treated left toe cleared the ground at MTC by 10 mm *more* than the feedback influenced right toe. This result confirmed that the trained foot toe height improved with feedback training. It is of further interest to note in **Table 2** that the stroke patient's *unaffected* left toe height in Baseline was almost twice that of the older sample, while the affected right toe clearance

Table 1 | Gait cycle parameters for a group of older individuals and one stroke participant with an affected right limb.

	Older group				Stroke participant			
	Baseline		Retention		Baseline		Retention	
	Left foot	Right foot	Left foot	Right foot	Left foot	Right foot	Left foot	Right foot
Stride length (m)	0.87	0.87	0.91	0.91	0.61	0.61	0.68	0.68
Stride duration (s)	1.21	1.21	1.25	1.25	2.21	2.19	2.46	2.44
Step length (m)	0.45	0.42	0.48	0.43	0.26	0.36	0.30	0.39
Step duration (s)	0.61	0.60	0.64	0.61	1.10	1.14	1.25	1.20
Step width (m)	0.21	0.21	0.20	0.20	0.23	0.23	0.23	0.24
Stance duration (%)	69	70	69	69	80	73	81	74

(16 mm) was comparable to the older participants' toe-ground clearances.

While there was no condition effect on toe height variability in older participants, augmented information training of the right foot may have increased right toe-height variability relative to baseline. In contrast the stroke patient's right toe standard deviation *decreased* from 13.0 mm in Baseline to 8.8 mm in Retention. In addition to the toe height mean and standard deviation, the skewness and kurtosis characteristics of the toe-height distribution in **Table 2** may have affected the predicted frequency of contact (for a 1 cm obstacle). The results show that all but one of the older subjects had positively skewed ($S > 0$; right skew) MTC and one other older person demonstrated negative kurtosis ($K < 0$) in the Baseline condition. Subject E3 had an exceptionally high negative skew ($S = -8.9$) to the right foot MTC that caused the overall group mean skew to be less than zero in Baseline. This subject's skew, however, became positive in Retention ($S = 0.57$). Subject E1 had negative Kurtosis ($K = -4.4$) which after training became positive ($K = 3$). Most interesting is the increase in predicted step frequency and associated reduction in probability of tripping (PT) from baseline to retention for all participants on the trained limb. The PT on a 1 cm high obstacle for the older adults reduced on average from 1 in every 578 strides to 1 in $>10^5$ strides. With gait training the stroke patient's estimated tripping probability over a 1 cm obstacle at MTC was 1 in 3 strides at baseline and 1 in 161 strides immediately post-training.

Qualitative changes in the toe swing phase trajectory are illustrated in **Figures 2**, **3**. **Figure 2** shows the immediate effect of augmented information on the stroke patient's toe-trajectory control by comparing the Baseline and Retention toe-ground clearance time-series over multiple steps. It is noteworthy that MTC biofeedback tended to increase toe-ground clearance throughout the swing phase, not only at the targeted MTC event. Training effects on the stroke patient's lower limb trajectory control are highlighted further in the typical one gait cycle (Heel Contact to Heel Contact) plot in **Figure 3**. With training the plot more closely resembles that of the control older adult at the top of the figure with increased toe elevation and a more clearly defined MTC event.

DISCUSSION

Previous research has identified MTC at approximately mid-swing as a gait variable for predicting foot-contact with surface obstacles (e.g., Begg and Best, 2002). The aim of this experiment was to determine the efficacy of treadmill-based gait training

Table 2 | Toe height characteristics at the Minimum Toe Clearance (MTC) swing-phase event.

	Older group				Stroke participant			
	Baseline		Retention		Baseline		Retention	
	Left foot	Right foot	Left foot	Right foot	Left foot	Right foot	Left foot	Right foot
Toe height (mm)	15.3	14.4	19.5*	35.5*	29.1	16.6	34.9	24.9
Toe height *SD* (mm)	3.6	5.4	4.0	8.3	5.4	13.0	5.3	8.8
Skewness	1.20	−0.36	1.04	0.73	0.44	2.88	0.60	2.08
Kurtosis	1.66	1.38	1.58	1.60	0.29	11.27	−0.13	8.35
Contact frequency	2482	641	269	$>10^5$	$>10^5$	3	$>10^5$	161

SD is toe height standard deviation. Contact Frequency is the predicted frequency of toe contact with a 1 cm high obstruction at MTC, i.e., 641 represents predicted contact once in every 641 strides. Higher frequency represents lower tripping probability.

$^*p < 0.05$, Significant difference between Baseline vs. Retention conditions.

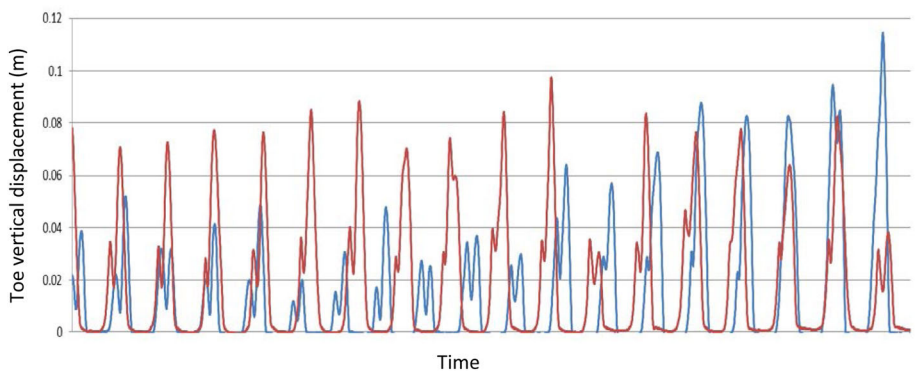


FIGURE 2 | A sample of the real-time vertical toe-ground displacement for the Stroke Participant during preferred speed treadmill gait training, Blue—Pre-training; Red—Post-training (Retention).

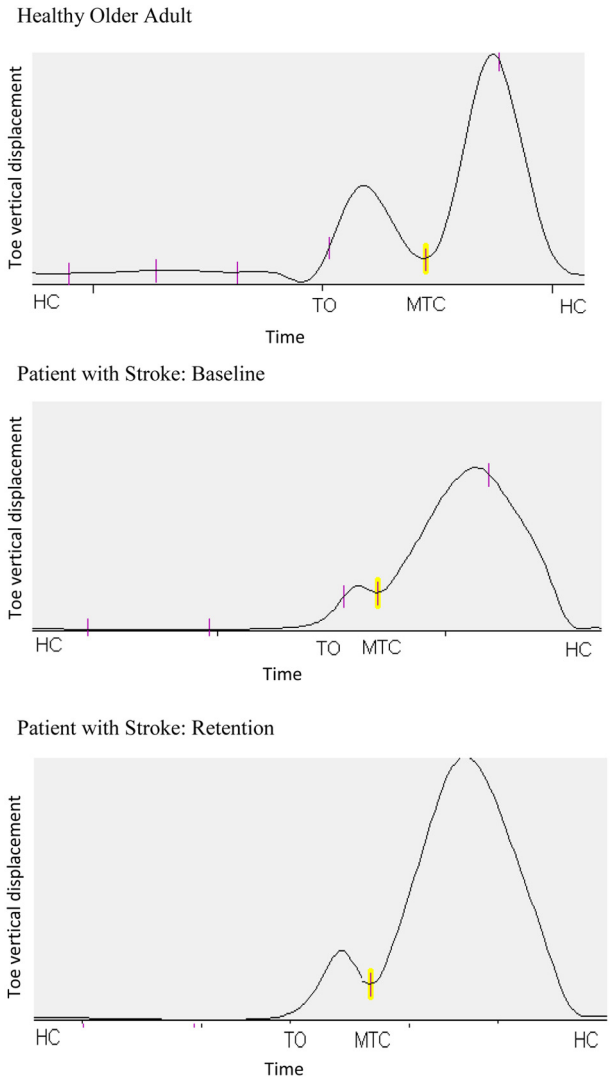


FIGURE 3 | Typical toe vertical displacement for one Healthy Older Adult and the Stroke Participant in pre-training Baseline and in Retention following training. The plots are one complete time-normalized stride cycle, defined by consecutive heel contact (HC) events. The swing-phase is from the first HC to Toe-Off (TO) and MTC is the Minimum Toe Clearance event at approximately mid-swing.

with concurrent augmented information to increase toe height at MTC in older adults and reduce the predicted frequency of toe-ground contact. Consistent with the review by Barrett et al. (2010) Baseline MTC was in the range 10–20 mm for the older group and the stroke patient's *affected* limb MTC (16 mm) was comparable to the older adult data. Despite the stroke patient's significant gait impairment, as reflected in spatial and temporal gait cycle variables, it is interesting that high toe-clearance was maintained in both feet, perhaps a precautionary strategy but, toe-clearance was considerably greater in the unaffected limb.

When provided with augmented MTC information both the older group and the stroke patient maintained an elevated MTC in retention. Toe height variability in the stroke patient's affected limb reduced with feedback training while toe height at MTC increased. Increased toe height is associated with safer progression but in contrast to the stroke patient who decreased her right toe clearance variability from baseline to retention the older group increased variability in the right limb following training. As a consequence of increased Retention variability in the older group and variability *declining* with training in the stroke participant's treated right foot, toe-ground variability in retention was approximately equivalent. Positive skew indicates a distribution biased to the right (higher MTC), consistent with one of three strategies employed by the elderly to minimize tripping (Begg et al., 2007). The two other strategies to reduce tripping risk are either to increase MTC central tendency or reduce MTC variability (Begg et al., 2007; Best and Begg, 2008), the former observed in the present results following training.

Overall the above toe-ground clearance height results confirm that concurrent ("real-time") visual information of lower limb kinematic gait parameters is effective in increasing anticipatory control of mid-swing toe height in both healthy older adults and the older adult with stroke. All participants reduced tripping probability in retention as reflected in an increased number of strides that would contact a 1 cm obstacle assuming no anticipatory corrections to toe-trajectory. The use of training using feedback to improve anticipatory ("feedforward") control has been studied only to a limited degree (e.g., Tsao and Hodges, 2007). The mechanism underlying the improvement in MTC is yet to be elucidated, however it is possible that the augmented feedback of MTC plus task-specific training enhanced the internal model of gait to improve foot trajectory (Kawato, 1999). The results also revealed that the stroke patient's *un*affected left toe also increased height at MTC in Retention relative to baseline (29.1 and 34.9 mm, respectively). Changes to the kinematic characteristics of the untrained limb imply transfer of training, which could be very important in unilaterally affected populations. If transfer could be confirmed in future work with gait-impaired populations it would raise the possibility of training a patient's unaffected limb to induce positive effects on the injured or neurologically impaired contralateral limb.

The finding of longer stance duration (80% of stride time) in the stroke patient's non-affected limb (compared to 73% in the affected, right limb) and a spatially shorter left step suggest a stability-related adaptation, similar to that previously reported in older adults (Nagano et al., 2013). Step length of older adults' dominant limb was asymmetrically larger suggesting "functional asymmetry." Older adults increased the proportion of step time in double support and reduced step length on the treadmill indicating adaptation that may preserve their balance (Nagano et al., 2013). Previous literature also reported asymmetry between the affected and non-affected limb's spatial-temporal parameters following stroke (e.g., Allen et al., 2011). Further evidence of the stroke patient employing gait adaptations to maintain stability is a greater step width (1–4 cm) than the older group.

Results are only available from one participant with stroke, which limits the generalizability of these results. However, the results are encouraging and support the feasibility of utilizing augmented feedback-based gait training to retrain walking following stroke. Future randomized controlled trials with

appropriate sample size and power are required to demonstrate the effectiveness of this technique in people with stroke.

To complement biomechanical information concerning augmented information effects on the lower-limb's "end-point control," reflected in toe height at MTC, it is also important to determine how hip, knee, and ankle joint angles are modulated to control MTC.

ACKNOWLEDGMENTS

The authors would like thank Rob van der Straaten and Dr. Lisa James for assistance with the data collection. The probability of tripping software was developed by Dr. Russell Best. The work was supported in part by the Australian Government's Collaborative Research Networks (CRN) program.

REFERENCES

AIHW. (2008). *Australian Institute of Health and Welfare: Australia's Health.* Canberra: Australian Government.

Allen, J. L., Kautz, S. A., and Neptune, R. R. (2011). Step length asymmetry is representative of compensatory mechanisms used in post-stroke hemiparetic walking. *Gait Posture* 33, 538–543. doi: 10.1016/j.gaitpost.2011.01.004

Baram, Y., and Miller, A. (2006). Virtual reality cues for improvement of gait in patients with multiple sclerosis. *Neurology* 66, 178–181. doi: 10.1212/01.wnl.0000194255.82542.6b

Barrett, R. S., Mills, P. M., and Begg, R. K. (2010). A systematic review of the effect of ageing and falls history on minimum foot clearance characteristics during level walking. *Gait Posture* 32, 429–435. doi: 10.1016/j.gaitpost.2010.07.010

Basmajian, J. V. (1981). Biofeedback in rehabilitation: a review of principles and practices. *Arch. Phys. Med. Rehabil.* 62, 469–475.

Batavia, M., Gianutsos, J. G., Vaccaro, A., and Gold, J. T. (2001). A do-it-yourself membrane-activated auditory feedback device for weight bearing and gait training: a case report. *Arch. Phys. Med. Rehabil.* 82, 541–545. doi: 10.1053/apmr.2001.21931

Batchelor, F. A., Mackintosh, S. M., Said, C. M., and Hill, K. D. (2012). Falls after stroke. *Int. J. Stroke* 7, 482–490. doi: 10.1111/j.1747-4949.2012.00796.x

Begg, R. K., and Best, R. J. (2002). "Estimating the probability of foot-ground contact in young and older individuals during walking," in *Proceedings of the Fourth Australasian Biomechanics Conference*, eds T. M. Bach, D. Orr, R. Baker, and W. A. Sparrow (Melbourne: La Trobe University).

Begg, R. K., Best, R. J., Taylor, S., and Dell'Oro, L. (2007). Minimum foot clearance during walking: strategies for the minimization of trip-related falls. *Gait Posture* 25, 191–198. doi: 10.1016/j.gaitpost.2006.03.008

Begg, R. K., Tirosh, O., Straaten, R. V. D., and Sparrow, W. A. (2012). "Real-time biofeedback of gait parameters using infrared position sensors," in *Refereed Conference Paper* (4 pages)—*Sixth International Conference on Sensing Technology—ICST 2012* (Kolkata: IEEE Explore).

Best, R. J., and Begg, R. K. (2008). A method for calculating the probability of tripping while walking. *J. Biomech.* 41, 1147–1151. doi: 10.1016/j.jbiomech.2007.11.023

Boonstra, T. A., van der Kooij, H., Munneke, M., and Bloem, B. R. (2008). Gait disorders and balance disturbances in Parkinson's disease: clinical update and pathophysiology. *Curr. Opin. Neurol.* 21, 461–471. doi: 10.1097/WCO.0b013e328305bdaf

Cohen, L., Miller, T., Sheppard, M. A., Gordon, E., Gantz, T., and Atnafou, R. (2003). Bridging the gap: bringing together intentional and unintentional injury prevention efforts to improve health and well being. *J. Safety Res.* 34, 473–483. doi: 10.1016/j.jsr.2003.03.005

Forster, A., and Young, J. (1995). Incidence and consequences of falls due to stroke: a systematic enquiry. *Br. Med. J.* 311, 83–86. doi: 10.1136/bmj.311.6997.83

Gillespie, L. D., Gillespie, W. J., Robertson, M. C., Lamb, S. E., Cumming, R. G., and Rowe, B. H. (2003). Interventions for preventing falls in elderly people. *Cochrane Database Syst. Rev*. 4, CD000340.

Goldie, P., Kay, D., and Patla, A. (2000). "An analysis of the home environment encountered by stroke patients," in *Sixth International Physiotherapy Congress of the Australian Physiotherapy Association* (Canberra: Australian Physiotherapy Association) (ACT Branch), 10–11.

Kawato, M. (1999). Internal models for motor control and trajectory planning. *Curr. Opin. Neurobiol.* 9, 718–727. doi: 10.1016/S0959-4388(99)00028-8

Langhorne, P., Bernhardt, J., and Kwakkel, G. (2011). Stroke rehabilitation. *Lancet* 377, 1693–1702. doi: 10.1016/S0140-6736(11)60325-5

Nagano, H., Begg, R. K., Sparrow, W. A., and Taylor, S. (2011). Ageing and limb dominance effects on foot-ground clearance during treadmill and overground walking. *Clin. Biomech.* 26, 962–968. doi: 10.1016/j.clinbiomech.2011.05.013

Nagano, H., Begg, R. K., Sparrow, W. A., and Taylor, S. (2013). A comparison of treadmill and overground walking effects on step cycle asymmetry in young and older individuals. *J. Appl. Biomech.* 29, 188–193.

Newell, K. M., Quinn, J. T., Sparrow, W. A., and Walter, C. B. (1983). Information feedback for learning a rapid arm movement. *Hum. Mov. Sci.* 2, 255–269.

Rao, N., Zielke, D., Keller, S., Burns, M., Sharma, A., Krieger, R., et al. (2012). Pregait balance rehabilitation in acute stroke patients. *Int. J. Rehabil. Res.* 36, 112–117. doi: 10.1097/MRR.0b013e328359a2fa

Sadeghi, H., Prince, F., Sadeghi, S., and Labelle, H. (2000). Principal component analysis of the power developed in the flexion/extension muscles of the hip in able-bodied gait. *Med. Eng. Phys.* 22, 703–710. doi: 10.1016/S1350-4533(01)00010-8

Salmoni, A. W., Schmidt, R. A., and Walter, C. B. (1984). Knowledge of results and motor learning: a review and critical reappraisal. *Psychol. Bull.* 95, 355–386.

Seeger, B. R., Caudrey, D. J., and Scholes, J. R. (1981). Biofeedback therapy to achieve symmetrical gait in hemiplegic cerebral palsied children. *Arch. Phys. Med. Rehabil.* 62, 364–368.

Sparrow, W. A., and Summers, J. J. (1992). Performance on trials without knowledge of results (KR) in reduced relative frequency presentations of KR. *J. Mot. Behav*. 24, 197–209. doi: 10.1080/00222895.1992.9941615

Tirosh, O., Cambell, A., Begg, R. K., and Sparrow, W. A. (2013). Biofeedback training effects on minimum toe clearance variability during treadmill walking. *Ann. Biomed. Eng*. 41, 1661–1669. doi: 10.1007/s10439-012-0673-6

Tsao, H., and Hodges, P. W. (2007). Immediate changes in feedforward postural adjustments following voluntary motor training. *Exp. Brain Res*. 181, 537–546. doi: 10.1007/s00221-007-0950-z

Van Vliet, P. M., and Wulf, G. (2006). Extrinsic feedback for motor learning after stroke: what is the evidence? *Disabil. Rehabil.* 28, 831–840. doi: 10.1080/09638280500534937

WHO. (2002). *The Injury Chart Book: A Graphical Overview of the Global Burden of Injuries*. Geneva: Department of Injuries and Violence Prevention, World Health Organization.

Zeni, J. A. Jr., Richards, J. G., and Higginson, J. S. (2008). Two simple methods for determining gait events during treadmill and overground walking using kinematic data. *Gait Posture* 27, 710–714. doi: 10.1016/j.gaitpost.2007.07.007

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.

Received: 14 December 2013; accepted: 02 April 2014; published online: 08 May 2014.
*Citation: Begg RK, Tirosh O, Said CM, Sparrow WA, Steinberg N, Levinger P and Galea MP (2014) Gait training with real-time augmented toe-ground clearance information decreases tripping risk in older adults and a person with chronic stroke. Front. Hum. Neurosci. **8**:243. doi: 10.3389/fnhum.2014.00243*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 10 July 2014
doi: 10.3389/fnhum.2014.00484

Walking strategies in subjects with congenital or early onset strabismus

Irene Aprile[1]*, Maurizio Ferrarin[2], Luca Padua[1,3], Enrica Di Sipio[1], Chiara Simbolotti[1], Sergio Petroni[4], Costanza Tredici[5] and Anna Dickmann[5]

[1] *SM Provvidenza Movement Laboratory, Don Carlo Gnocchi Foundation, IRCCS, Milan, Rome, Italy*
[2] *Biomedical Technology Department, Don Carlo Gnocchi Foundation, IRCCS, Milan, Italy*
[3] *Neuroscience Department of Catholic University, Rome, Italy*
[4] *Ophthalmology Department, Bambino Gesù Children's Hospital, Rome, Italy*
[5] *Department of Surgical Sciences of Head and Neck, Institute of Ophtalmology, Catholic University, Rome, Italy*

Edited by:
Marco Iosa, Fondazione Santa Lucia, Italy

Reviewed by:
Giovanni Morone, IRCCS Santa Lucia Foundation, Italy
Elena Bergamini, Università degli Studi di Roma Foro Italico, Italy

****Correspondence:***
Irene Aprile, SM Provvidenza Movement Laboratory, Don Carlo Gnocchi Foundation, 401 Casal Del Marmo Street, 00166 Rome, Italy
e-mail: iaprile@dongnocchi.it

Introduction: In congenital strabismus, sensory adaptations occur hampering the correct development of normal binocular vision. The aim of this study is to investigate if patients with congenital or early onset exotropic or esotropic strabismus adopt different walking strategies with respect to healthy subjects. Our hypothesis is that the abnormal binocular cooperation, occurring in patients with exotropic or esotropic strabismus, could influence neurosensorial adaptation of the gait pattern.

Materials and Methods: Twenty-five patients were enrolled: 19 with esotropic (ESO) and 6 with exotropic strabismus (EXO). All patients underwent an ophthalmological and orthoptic evaluation. Biomechanical data were collected using a stereophotogrammetric system and a force platform. Twenty-seven age-matched healthy subjects (HS) were used as controls.

Results: The comparison between patients with ESO and patients with EXO strabismus showed that the maximal power at the knee and at the ankle was lower in EXO group ($p < 0.01$ and $p < 0.05$, respectively). The step width was statistically different between ESO and EXO groups ($p < 0.01$), lower in patients with ESO and higher in patients with EXO strabismus when compared with HS (though not statistically significant). The deviation angle values showed a relationship with the step width (at the near fixation $p < 0.05$) and with the maximal power at the knee and at the ankle (at the far fixation for the knee $p < 0.001$ and for the ankle $p < 0.05$; at the near fixation for the knee $p < 0.05$): in the patients with EXO the increased angle deviation is related to larger step width and to lower power at the knee and at the ankle. In the patients with ESO strabismus this relationship is less robust.

Discussion: Patients with EXO and ESO strabismus adopt different strategies to compensate their walking difficulties, and these strategies are likely due to an expanded binocular visual field in patients with EXO and to a reduced visual field in patients with ESO strabismus.

Keywords: gait analysis, strabismus, binocular visual field, walking strategy

INTRODUCTION

Strabismus is a condition characterized by a misalignment of the visual axes which leads to anomalies of the sensory and motor balance of both eyes. Depending on the age at onset, strabismus may determine different consequences: if it is congenital or starts in the first months after birth (that is during the period of maximal cortical plasticity) sensory adaptations (suppression, anomalous retinal correspondence) occur hampering the correct development of normal binocular vision and stereopsis (normal binocular cooperation). When strabismus occurs in the period in which cortical plasticity is no longer possible, sensory adaptations do not take place and patients will experience diplopia and confusion.

Strabismus may be classified according to various clinical aspects, i.e., age at onset, comitant or non-comitant (if the amount of deviation is the same in all gaze positions or not), alternating (if the patient can freely fix with each eye), or monocular (if the same eye is always the fixing one). When strabismus is always present, it is called heterotropia, if it is latent it is called heterophoria.

Direction of deviation is a common manner to describe strabismus: if the direction of the deviated eye is toward the temple it is an exodeviation, whereas, if the direction of deviation is toward

the nose it is an esodeviation (**Figure 1**). As a rule, the amount of the deviation (generally measured in Prismatic Diopters - PD) is negative for exodeviation and positive for esodeviation.

Walking is a complex task for the peripheral and central nervous system, which has to generate and control dynamic instability in order to produce the propulsive force needed for forward progression. Walking effectively through a complex environment requires successful integration of both sensory and motor functions. Visual function abnormalities may pose significant challenges to an individual in terms of such integration.

The visual system provides information not only on target distance and presence of obstacles, but also on maintaining balance during walking (Hallemans et al., 2010) adjusting trajectories when an obstacle appears or the target is shifted (Reynolds and Day, 2005).

Vision controls dynamic stability providing information about the environment (objects near and far) thus helping regulate walking both at local (step by step basis) and global (route planning) levels. So, individuals with vision abnormalities may take longer to walk safely through the environment.

A study investigated the effects of optic flow field alterations on spatio-temporal gait parameters and on joint kinematics (Konczak, 1994), and others have studied gait in patients with visual deprivation (Hallemans and Aerts, 2009; Hallemans et al., 2009a,b). In one study a significant influence of reversible visual deprivation on gait parameters has been observed in HS (Iosa et al., 2012).

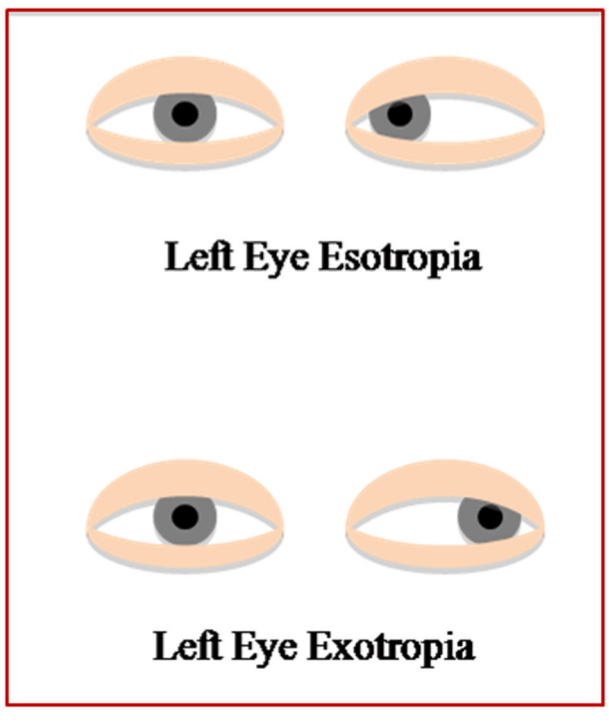


FIGURE 1 | Description of the eye direction, deviated toward the temple in patients with EXO and deviated toward the nose in patients with ESO.

No studies on gait strategies of subjects with abnormal sensory and motor eyes cooperation, at birth or started in the first months after birth (as in congenital/early onset strabismus), have been reported. Subjects with congenital or early onset strabismus have an abnormal binocular vision since the time of maximal cortical plasticity, and they could develop different walking adaptive strategies with respect to those observed after reversible visual deprivation in HS.

Several authors (Odenrick et al., 1984; Matsuo et al., 2006, 2010; Matheron et al., 2007; Legrand et al., 2011; Gaertner et al., 2013a; Lions et al., 2013; Przekoracka-Krawczyk et al., 2014) found that subjects with congenital or early onset strabismus have a significantly lower static and dynamic balance than HS. However, a binocular visual stimulation plays an important role in postural control of patients with strabismus (Gaertner et al., 2013b).

Our hypothesis is that the abnormal binocular cooperation, occurring in patients with congenital or early onset exotropic or esotropic strabismus, could influence neurosensorial adaptations of the gait pattern.

The aim of this study was to investigate if patients with exotropic or esotropic congenital or early onset strabismus adopt different walking strategies with respect to HS.

MATERIALS AND METHODS

POPULATION

A sample of patients with strabismus and a group of age-matched HS were analyzed. Twenty-five patients with strabismus were enrolled in this study: 13 males and 12 females, range 5–50 years. In particular, 6 patients with exotropic strabismus (EXO) of mean age 17.33 years (*SD*: 17.31), and 19 patients with esotropic strabismus (ESO) of mean age 13.16 years (*SD*: 6.64). Inclusion criteria were congenital or early onset strabismus (within 1 year of age) and age $\geq$ 5 years. All patients were able to walk independently without assistance or walking aids. Exclusion criteria included strabismus acquired after 1 year of age, lack of cooperation for age or mental retardation, presence of systemic or neurological diseases as well as of orthopedic or postural problems. We excluded patients with cognitive impairment, cardiologic diseases that made walking risky, or other diseases liable to cause motor gait impairment (i.e., radiculopathy, bone fracture, etc.). The HS group consisted of 27 subjects (14 females and 13 males, mean age 17.93 years, range 5–46).

All the participants gave their informed consent prior to participating in the study, which complied with the Helsinki Declaration.

OPHTHALMOLOGICAL AND ORTHOPTIC EVALUATION

Both patients and HS underwent a complete ophthalmological and orthoptic evaluation. Subjects with strabismus were divided, according to the direction of deviation, into two groups: patients with esotropic and patients with exotropic strabismus (**Table 1**). In no patient a vertical or cyclotorsional component was associated with the horizontal deviation. The amount of strabismus was always measured by means of Prismatic Cover test, both at far fixation (6 m) and at near fixation (0.40 m), considering that difference in the amount of deviation may depend on the

Table 1 | Anthropometric and ophthalmological/orthoptic evaluation of patients with strabismus.

PATIENT	SEX	AGE (yy)	Mass (Kg)	Stature (m)	Visual acuity RE	Visual acuity LE	Deviation angle (PD) at far fixation	Deviation angle (PD) at near fixation
IMAL	M	5	18	1.10	6/6	6/7.5	XT50	XT′45
PESI	M	5	17	1.10	6/7.5	6/6	XT14	XT′20
MOAD	F	6	21	1.20	6/6	6/7.5	ET10	ET′18
GAFR	M	7	32	1.30	6/12	6/7.5	ET25	ET′30
FASO	F	8	24	1.25	6 / 9.5	6/9.5	ET12	ET′35
INDE	F	7	19	1.21	6/15	6/9.5	ET30	ET′35
CAFI	M	7	24	1.23	6/7.5	6/6	ET35	ET′40
VEAR	M	8	35	1.38	6/7.5	6/9.5	ET6	ET′10
CHVA	F	8	34	1.30	6/6	6/9.5	ET20	ET′18
ALMA	M	8	30	1.32	6/6	6/12	ET75	ET′70
QUTO	M	9	27	1.30	6/7.5	6/6	ET30	ET′35
BOVI	F	10	33	1.40	6/6	6/6	XT2	XT′8
TAIL	F	11	35	1.44	6/9.5	6 / 7.5	ET40	ET′75
DILE	M	11	55	1.55	6/6	6/6	XT8	XT′16
CAMA	F	12	30	1.42	6/6	6/9.5	ET25	ET′35
DESI	F	13	47	1.58	6/9.5	6/12	ET18	ET′20
VARO	M	15	65	1.70	6/6	6/7.5	ET16	ET′20
CHAD	M	16	73	1.80	6/6	6/6	ET35	ET′30
SULA	F	19	40	1.58	6/9.5	6/7.5	ET14	ET′8
FEAN	M	23	80	1.76	6/6	6/7.5	XT16	XT′40
COAR	F	23	49	1.63	6/12	6/7.5	ET50	ET′60
PAIL	F	24	72	1.81	6/6	6/6	ET4	ET′8
PADA	M	24	75	1.80	6/7.5	6/6	ET12	ET′8
NALA	F	24	55	1.53	6/6	6/6	ET20	ET′14
CAFR	M	50	76	1.74	6/6	6/6	XT35	XT′35

RE, right eye; LE, left eye; PD, Prismatic Diopters; ET, esotropia at far fixation; ET′, esotropia at near fixation; XT, exotropia at far fixation; XT′, exotropia at near fixation.

distance of fixation. Due to the presence of a congenital or early onset strabismus a normal binocular vision was absent in all patients.

In the HS group all subjects were emmetropic or with a best corrected visual acuity of 6/6 and showed a normal binocular vision and stereopsis $\geq 60''$ at the orthoptic evaluation.

Gait analysis

The gait analysis was performed using the Smart D500 stereophotogrammetric systems (BTS Bioengineering, Milan, Italy). The system consists of eight infrared cameras (sampling rate of 200 Hz) to acquire movement of reflective spherical markers placed on anatomical landmarks. Kinetic data were acquired at 1 kHz using a single 600 × 400 mm piezoelectric force platform (Kistler, Winterthur, Switzerland). The patients were equipped with 22 retro-reflective markers, according to Davis protocol (Davis et al., 1991). The marker-set is composed of 18 markers directly applied to the skin and 4 wands placed at 1/3 the length of the body segment. In particular, it places a wand on the femur and on the leg, so that the plane containing the three points is parallel to the frontal plane. For each subject anthropometric measures were collected; to increase the reliability of the biomechanical measures, markers placement and anthropometric measures were performed by the same operator (Winter, 1979).

Before formal measurements were started, practice sessions were performed to let participants become familiar with the procedure. Both patients and HS were asked to walk barefoot straight ahead along a level surface of approximately 6 m at their self-selected speed. Ten linear walking trials were acquired for each subject. To avoid fatigue, groups of 5 trials were separated by 1 min rest.

Three-dimensional (3D) kinematics, dynamics were considered if complete data of at least one stride (indifferently right or left) were available, otherwise the trial was discarded. Trials, which presented evident artifacts due to technical problems (missing detection of some markers or improper foot-strike on platform), were excluded.

Data analysis

Three-dimensional marker trajectories were tracked using a frame-by-frame tracking system (Smart Tracker-BTS, Milan, Italy). Data were processed using 3D reconstruction software (SMARTAnalyzer, BTS, Milan, Italy) and MATLAB software (MATLAB 7.4.0, MathWorks, Natick, MA, USA).

The following parameters were considered:

Spatio-temporal parameters. Gait cycle duration was defined as the interval between two consecutive heel contacts of the same

foot. To reduce the possible variability due to the age-range of the sample, we have normalized all height-dependent gait parameters (i.e., step length, step width and walking speed) by subject height. The following parameters were calculated: cycle time [s], stance [%], swing [%] and double support phase duration [%], step length [%H], step width [%H], cadence [step/min], walking speed [%H/s]. All dynamic parameters were normalized to body weight.

Kinematic parameters. The lower limb joint kinematics in the sagittal plane was considered and the hip, knee and ankle joint range of motion (ROM) [°] were calculated.

Kinetic parameters. The maximal power and maximal moment in early and late stance at the hip and knee were calculated; for the ankle the maximal power and maximal moment in late stance were considered. Early stance is defined as the percentage of the gait cycle ranging from 0 to 30, and late stance is defined as the percentage of the gait cycle ranging from 30 to 60.

Statistical analysis

The statistical analysis was performed using the STATSOFT (Tulsa, OK, USA) package. All data were tested for normality with the Shapiro–Wilk test. Since the variables were not normally distributed, the Mann-Whitney test was used for all the investigated variables (spatio-temporal, kinematic and kinetic data) to determine differences between the following group pairs: patients with strabismus vs. HS, patients with ESO vs. patients with EXO strabismus, patients with ESO strabismus vs. HS, patients with EXO strabismus vs. HS. Moreover, the Spearman's rank correlation coefficient test was used to evaluate the correlations between deviation angle values at far and near fixation and spatio-temporal, kinematic and kinetic data, was used. The significance level was set for all parameters at $p < 0.05$.

RESULTS

COMPARISON BETWEEN SUBJECTS WITH STRABISMUS AND HEALTHY SUBJECTS

In **Table 2** mean, standard deviation, and *p*-value of age, anthropometric measures and gait analysis results are reported. No significant differences were found in age, stature, and mass between patients and HS. Kinetic results showed a significantly lower maximal moment in patients than in HS at the hip ($p < 0.001$ in early stance, and $p < 0.0001$ in late stance), at the knee ($p < 0.0001$ in early and late stance) and at the ankle ($p < 0.05$). Moreover a significantly lower range of motion of the knee was found in patients than in HS ($p < 0.01$).

Table 2 | Spatio-temporal, kinematic, and kinetic parameters in patients and in HS.

Variable	Patients (25 cases)	Healthy Subjects (27 cases)	*p*-value
	Mean (*SD*)	Mean (*SD*)	
Age [yy]	14.04 (9.98)	17.93 (12.20)	NS
ANTHROPOMETRIC			
Mass [Kg]	42.64 (20.57)	46.85 (20.25)	NS
Stature [m]	1.46 (0.23)	1.48 (0.25)	NS
TIME DISTANCE			
Cycle Time [s]	1.04 (0.13)	1.01 (0.15)	NS
Stance [%]	59.32 (1.65)	58.87 (1.75)	NS
Swing [%]	40.96 (1.66)	41.13 (1.80)	NS
DBS [%]	8.71 (1.32)	8.53 (1.72)	NS
Step length [%H]	34.50 (2.60)	34.54 (2.09)	NS
Step width [%H]	9.49 (1.91)	9.52 (1.63)	NS
Cadence [step/min]	117.78 (15.47)	122.01 (20.27)	NS
Walking speed [%H/s]	73.38 (12.63)	75.64 (14.97)	NS
KINEMATIC			
Hip SAG ROM [°]	46.10 (5.19)	46.11 (3.51)	NS
Knee SAG ROM [°]	57.59 (4.78)	60.71 (3.93)	<0.01
Ankle SAG ROM [°]	28.43 (5.33)	30.11 (5.52)	NS
KINETIC			
Hip max moment (early stance) [N*m/Kg]	0.70 (0.10)	0.79 (0.06)	<0.001
Hip max moment (late stance) [N*m/Kg]	0.19 (0.10)	0.06 (0.05)	<0.0001
Knee max moment (early stance) [N*m/Kg]	0.30 (0.09)	0.41 (0.06)	<0.0001
Knee max moment (late stance) [N*m/Kg]	0.10 (0.04)	0.13 (0.03)	<0.05
Ankle max moment [N*m/Kg]	1.30 (0.17)	1.46 (0.08)	<0.0001
Hip max power (early stance) [W/Kg]	1.19 (0.37)	1.29 (0.21)	NS
Hip max power (late stance) [W/Kg]	0.69 (0.21)	0.59 (0.15)	NS
Knee max power (early stance) [W/Kg]	0.46 (0.16)	0.49 (0.13)	NS
Knee max power (late stance) [W/Kg]	0.43 (0.15)	0.46 (0.18)	NS
Ankle max power [W/Kg]	3.07 (0.69)	3.08 (0.56)	NS

COMPARISON BETWEEN ESOTROPIC AND EXOTROPIC SUBJECTS AND HEALTHY SUBJECTS

In **Table 3**, age, anthropometric and gait analysis data (mean, *SD* and *p*-values) in ESO, EXO and HS are reported. Furthermore, the comparison between each groups of patients and HS (ESO vs. HS, EXO vs. HS) and the comparison between the two groups of patients (ESO vs. EXO) are shown. No differences in age, stature and mass were found among the three groups.

Regarding spatio-temporal results, **Figure 2** describes as the step length progressively reduced in the three groups (HS, ESO, and EXO). The step width was smaller in patients with ESO and wider in patients with EXO when compared with HS, respectively (**Figure 3**). A significant difference in the step width was observed when patients with ESO and patients with EXO were compared ($p < 0.01$, see **Table 3**).

Regarding kinematic results patients with ESO and patients with EXO had a significantly reduced ROM of the knee than

Table 3 | Comparison between groups: patients with esotropic strabismus (ESO) vs. Healthy Subjects (HS); patients with exotropic strabismus (EXO) vs. Healthy Subjects (HS).

Variable	Patients with ESO	*p*-value	Patients with EXO	*p*-value	*p*-value
	Mean (*SD*)	(ESO vs. HS)	Mean (*SD*)	(EXO vs. HS)	(ESO vs. EXO)
Age [yy]	13.00 (6.69)	NS	17.33 (17.31)	NS	NS
ANTHROPOMETRIC					
Mass [Kg]	41.42 (18.43)	NS	46.50 (28.03)	NS	NS
Stature [m]	1.46 (0.21)	NS	1.44 (0.30)	NS	NS
TIME DISTANCE					
Cycle Time [s]	1.04 (0.13)	NS	1.04 (0.14)	NS	NS
Stance [%]	59.28 (1.63)	NS	59.44 (1.89)	NS	NS
Swing [%]	41.09 (1.62)	NS	40.56 (1.89)	NS	NS
DBS [%]	8.66 (1.19)	NS	8.86 (1.79)	NS	NS
Step Length [%H]	35.11 (2.36)	NS	32.58 (2.55)	NS	NS
Step Width [%H]	8.86 (1.16)	NS	11.51 (2.52)	NS	<0.01
Cadence [step/min]	117.86 (15.45)	NS	117.53 (17.00)	NS	NS
Walking speed [%H/s]	74.68 (11.73)	NS	69.27 (15.60)	NS	NS
KINEMATIC					
Hip SAG ROM [°]	47.23 (5.09)	NS	42.68 (4.11)	NS	NS
Knee SAG ROM [°]	58.59 (4.13)	<0.05	54.40 (5.70)	<0.05	NS
Ankle SAG ROM [°]	29.63 (5.12)	NS	24.64 (4.42)	NS	NS
KINETIC					
Hip max moment (early stance) [N*m/Kg]	0.70 (0.08)	<0.0001	0.71 (0.17)	NS	NS
Hip max moment (late stance) [N*m/Kg]	0.15 (0.09)	<0.0001	0.29 (0.06)	<0.0001	<0.01
Knee max moment (early stance) [N*m/Kg]	0.31 (0.09)	<0.001	0.24 (0.06)	<0.0001	NS
Knee max moment (late stance) [N*m/Kg]	0.10 (0.03)	NS	0.09 (0.05)	NS	NS
Ankle max moment [N*m/Kg]	1.33 (0.13)	<0.0001	1.21 (0.25)	<0.01	NS
Hip max power (early stance) [W/Kg]	1.19 (0.36)	NS	1.21 (0.44)	NS	NS
Hip max power (late stance) [W/Kg]	0.70 (0.24)	NS	0.64 (0.09)	NS	NS
Knee max power (early stance) [W/Kg]	0.51 (0.15)	NS	0.39 (0.07)	<0.001	<0.01
Knee max power (late stance) [W/Kg]	0.43 (0.16)	NS	0.41 (0.08)	NS	NS
Ankle max power [W/Kg]	3.25 (0.59)	NS	2.49 (0.69)	NS	<0.05

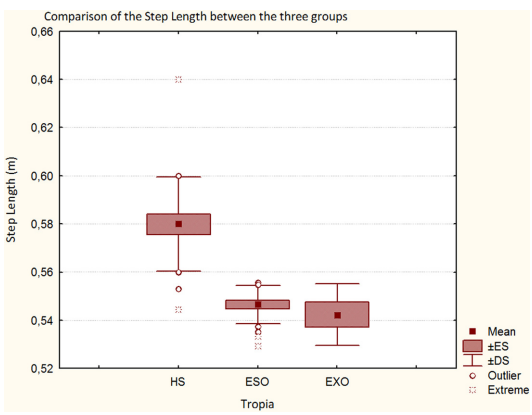


FIGURE 2 | Step length in HS, in patients with ESO and in patients with EXO.

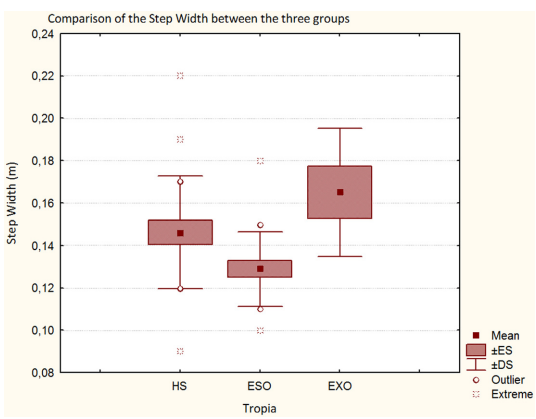


FIGURE 3 | Step width in HS, in patients with ESO and in patients with EXO.

HS ($p < 0.05$). A reduction (though not statistically significant) of the hip ROM was found between patients with EXO and HS and of the hip and ankle ROM comparing ESO and EXO groups.

Regarding kinetic results, comparing patients with ESO and HS significant differences in hip moment (in early and late stance, $p < 0.001$), in knee moment (in early stance, $p < 0.001$), and in ankle moment were found ($p < 0.0001$), (see **Table 3**).

Comparing patients with EXO and HS, significant differences in hip moment (in late stance $p < 0.0001$), in knee moment (in early stance $p < 0.0001$), in ankle moment ($p < 0.01$) were found. Moreover a significant reduced maximal power at the knee (in early stance) was found between patients with EXO and HS ($p < 0.001$). Finally, comparing patients with ESO and patients with EXO, significant differences in hip moment (in late stance, $p < 0.01$), and mainly in the maximal power at the knee (in early stance, $p < 0.01$) and at the ankle ($p < 0.05$) were found. The maximal power at the knee and at the ankle is lower in patients with EXO than patients with ESO. Moreover, the maximal power at the ankle is similar between HS and patients with ESO but strongly reduced in the patients with EXO (**Figure 4**).

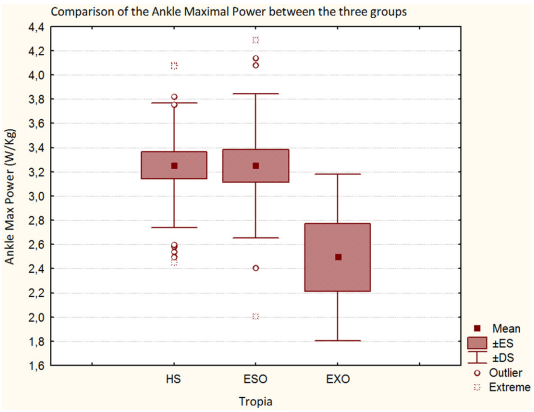


FIGURE 4 | Maximal Power at the ankle in HS, in patients with ESO and in patients with EXO.

RELATIONSHIP BETWEEN ORTHOPTIC AND GAIT PARAMETERS

The deviation angle values at far fixation showed a statistically significant relationship with the maximal power at the knee ($p < 0.001$) and at the ankle ($p < 0.05$): in patients with more negative values (expression of the severity of the motor misalignment in subjects with exotropic strabismus) a lower power at the knee and at the ankle was observed (**Figures 5A,B**). The deviation angle values at near fixation showed a significant correlation with the maximal power at the knee ($p < 0.05$): in patients with more negative values a lower power at the knee (**Figure 5C**) was observed. Regarding the maximal power at the ankle, a similar relationship was observed but without statistical significance (**Figure 5D**).

Furthermore an interesting significant relation was observed between the step width and angle values at near fixation ($p < 0.05$): patients with EXO with more negative values had a larger

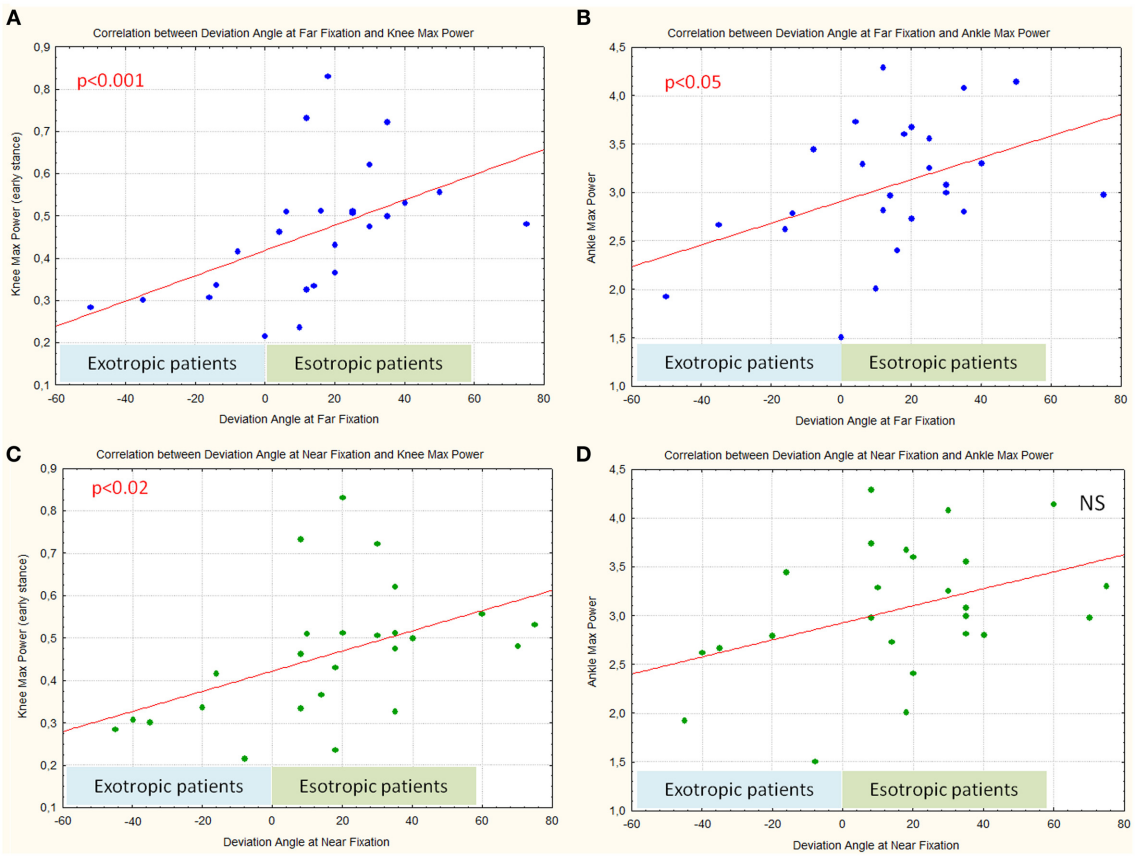


FIGURE 5 | Correlation between deviation angle values at far (A,B) and near fixation (C,D) and Maximal Power at the knee and at the ankle.

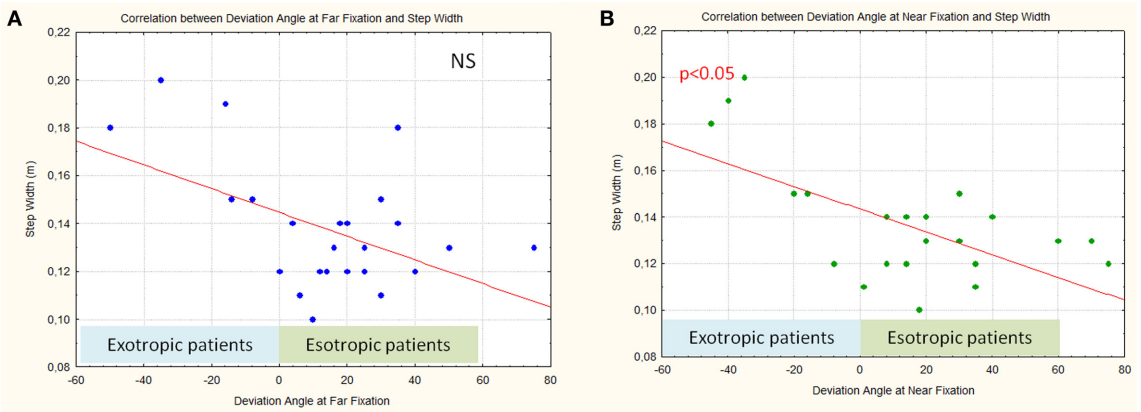


FIGURE 6 | Correlation between deviation angle values at far fixation (A) and near fixation (B) and step width.

step width (**Figure 6A**), moving toward more positive values; patients with ESO had a decreased step width. A similar relationship, between the step width and angle values at far fixation was observed (though not statistically significant).

It is interesting to underline that an accurate analysis of **Figures 5**, **6** shows that, considering only patients with ESO, the correlation between deviation angle and gait parameters (step width and power at the knee and ankle) seems less robust. In fact, when patients with EXO and patients with ESO were separately considered we found that patients with EXO maintain a significant relationship between step width and power at knee with deviation angle ($p < 0.05$), while patients with ESO maintain a significant relationship only between step width and deviation angle ($p < 0.05$).

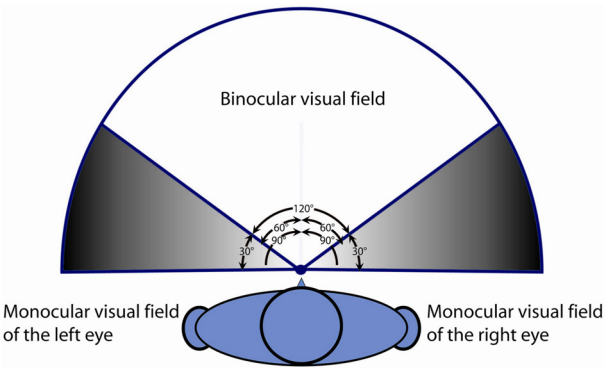


FIGURE 7 | Description of the normal binocular visual field and monocular visual field of the right and left eye.

DISCUSSION

The results of our study show the presence of different walking strategies in patients with ESO and EXO strabismus. In particular we observed significant differences in step width between the two groups. It is important to note that the larger step width is generally considered as a feature characterizing walking instability, while a smaller step width is recognized as an expression of good stability (Schrager et al., 2008). Regarding this parameter, patients with ESO and EXO showed different behavior when compared to HS: patients with ESO had a smaller step width than HS while patients with EXO had a larger step width than HS.

A possible explanation might rely on the characteristics of Binocular Visual Field (BVF) in patients with strabismus. The integrity of binocular visual field is recognized as an important factor influencing walking (Graci et al., 2009, 2010). In normal subjects the visual fields of both eyes widely overlap in the central part of each hemi-field (binocular visual field), as only a small part (the "temporal crescent" emerging from the more medial part of the nasal retina) is seen monocularly (**Figure 7**).

In patients with strabismus, despite a normality of visual fields when monocularly tested, BVF shows some anomalies concerning its extent (also depending on the direction of deviation eso/exo) when compared to HS. In esotropia BVF is more restricted and, in many cases, can show an expansion after strabismus surgery (Wortham and Greenwald, 1989; Kushner, 1994; Rosenbaum, 1999). Conversely, patients with EXO may exhibit an expanded visual field (von Noorden and Campos, 2002), in some way similar to the vision experienced by animals characterized by lateralization of the eyes (in this condition binocular overlap of visual field is absent or extremely limited). Especially in subjects with EXO, who can easily alternate eye fixation, the brain receives images at each moment from one eye or the other, suppressing the visual field originated by the non-fixing eye, and this could negatively influence the gait. Probably this behavior in patients with EXO could explain the larger step width and the worse walking performance with less power at the knee and ankle than patients with ESO.

Our results show a significant correlation between step width and near deviation angle value. In patients with EXO the step width proportionally increased with the angle of deviation. This is in agreement with the fact that modifications in perception of BVF seem to strongly depend on the amount of ocular misalignment. A similar correlation trend was observed between step width and the amount of deviation at far fixation (though without statistical significance). The small number of patients with

EXO enrolled in the study could explain this result. The presence of a reduced BVF in patients with ESO and an expanded one in patients with EXO might lead to a gait adjustment based on modifications of step width. The direct correlation between the amount of strabismus and step width could support the hypothesis of an adaptation strategy used to compensate the BVF anomalies.

Kinetic data showed that in patients with EXO power at the ankle, and above all at the knee joints, proportionally reduces when the angle of deviation increases. This supports the hypothesis of a decrease of gait performance in EXO patients proportional to the angle of deviation, which induce a more cautious gait characterized by enlarged step width and reduced maximum power at knee and ankle joint level.

It is interesting to note that when patients with ESO and patients with EXO were separately considered, the correlation between step width and deviation angle was maintained in both groups, the correlation between knee power and deviation angle was lost in patients with ESO. To confirm this finding, a larger sample of patients is needed.

The results of our study show that subjects with EXO and ESO adopt different strategies to compensate their walking difficulties and these different strategies are probably due to an expanded visual field in patients with EXO and in a reduced visual field in patients with ESO.

Further studies should also be addressed to which other aspects of the binocular vision disorders can influence the different walking strategies that patients with ESO and EXO adopt. Moreover, a short and long follow-up could be useful to verify the natural evolution of these different strategies in the two groups of patients.

Our results about walking strategies of patients with congenital or early onset ESO and EXO, suggest the application of integrated rehabilitation therapies focused not only on gait training but also on visual field training.

ACKNOWLEDGMENTS

We would like to thank Daniele Coraci and Cristiano Pecchioli for their help in making figures and Chiara Briani for her help in the linguistic revision of the paper.

REFERENCES

Davis, R. B. 3rd., Ounpuu, S., Tyburski, D., and Gage, J. R. (1991). A gait data collection and reduction technique. *Hum. Mov. Sci.* 10, 575–587. doi: 10.1016/0167-9457(91)90046-Z

Gaertner, C., Creux, C., Espinasse-Berrod, M. A., Orssaud, C., Dufier, J. L., and Kapoula, Z. (2013a). Postural control in nonamblyopic children with early-onset strabismus. *Invest. Ophthalmol. Vis. Sci.* 54, 529–536. doi: 10.1167/iovs.12-10586

Gaertner, C., Creux, C., Espinasse-Berrod, M. A., Orssaud, C., Dufier, J. L., and Kapoula, Z. (2013b). Benefit of bi-ocular visual stimulation for postural control in children with strabismus. *PLoS ONE* 8:e60341. doi: 10.1371/journal.pone.0060341

Graci, V., Elliott, D. B., and Buckley, J. G. (2009). Peripheral visual cues affect minimum-foot-clearance during overground locomotion. *Gait Posture* 30, 370–374. doi: 10.1016/j.gaitpost.2009.06.011

Graci, V., Elliott, D. B., and Buckley, J. G. (2010). Utility of peripheral visual cues in planning and controlling adaptive gait. *Optom. Vis. Sci.* 87, 21–27. doi: 10.1097/OPX.0b013e3181c1d547

Hallemans, A., and Aerts, P. (2009). Effects of visual deprivation on intra-limb coordination during walking in children and adults. *Exp. Brain Res.* 198, 95–106. doi: 10.1007/s00221-009-1937-8

Hallemans, A., Beccu, S., Van Loock, K., Ortibus, E., Truijen, S., and Aerts, P. (2009a). Visual deprivation leads to gait adaptations that are age- and context-specific: I. Step-time parameters. *Gait Posture* 30, 55–59. doi: 10.1016/j.gaitpost.2009.02.018

Hallemans, A., Beccu, S., Van Loock, K., Ortibus, E., Truijen, S., and Aerts, P. (2009b). Visual deprivation leads to gait adaptations that are age- and context-specific: II. Kinematic parameters. *Gait Posture* 30, 307–311. doi: 10.1016/j.gaitpost.2009.05.017

Hallemans, A., Ortibus, E., Meire, F., and Aerts, P. (2010). Low vision affects dynamic stability of gait. *Gait Posture* 32, 547–551. doi: 10.1016/j.gaitpost.2010.07.018

Iosa, M., Fusco, A., Morone, G., and Paolucci, S. (2012). Effects of visual deprivation on gait dynamic stability. *ScientificWorldJournal* 2012:974560. doi: 10.1100/2012/974560

Konczak, J. (1994). Effects of optic flow on the kinematics of human gait: a comparison of young and older adults. *J. Mot. Behav.* 26, 225–236. doi: 10.1080/00222895.1994.9941678

Kushner, B. J. (1994). Binocular field expansion in adults after surgery for esotropia. *Arch. Ophthalmol.* 112, 639–643. doi: 10.1001/archopht.1994.01090170083027

Legrand, A., Quoc, E. B., Vacher, S. W., Ribot, J., Lebas, N., Milleret, C., et al. (2011). Postural control in children with strabismus: effect of eye surgery. *Neurosci. Lett.* 501, 96–101. doi: 10.1016/j.neulet.2011.06.056

Lions, C., Bui-Quoc, E., and Bucci, M. P. (2013). Postural control in strabismic children versus non strabismic age-matched children. *Graefes Arch. Clin. Exp. Ophthalmol.* 251, 2219–2225. doi: 10.1007/s00417-013-2372-x

Matheron, E., Le, T. T., Yang, Q., and Kapoula, Z. (2007). Effects of a two-diopter vertical prism on posture. *Neurosci. Lett.* 423, 236–240. doi: 10.1016/j.neulet.2007.07.016

Matsuo, T., Narita, A., Senda, M., Hasebe, S., and Ohtsuki, H. (2006). Body sway increases immediately after strabismus surgery. *Acta Med. Okayama* 60, 13–24.

Matsuo, T., Yabuki, A., Hasebe, K., Shira, Y. H., Imai, S., and Ohtsuki, H. (2010). Postural stability changes during the prism adaptation test in patients with intermittent and constant exotropia. *Invest. Ophthalmol. Vis. Sci.* 51, 6341–6347. doi: 10.1167/iovs.10-5840

Odenrick, P., Sandstedt, P., and Lennerstrand, G. (1984). Postural sway and gait of children with convergent strabismus. *Dev. Med. Child Neurol.* 26, 495–499. doi: 10.1111/j.1469-8749.1984.tb04477.x

Przekoracka-Krawczyk, A., Nawrot, P., Czaiska, M., and Michalak, K. P. (2014). Impaired body balance control in adults with strabismus. *Vision Res.* 98, 35–45. doi: 10.1016/j.visres.2014.03.008

Reynolds, R. F., and Day, B. L. (2005). Visual guidance of the human foot during a step. *J. Physiol.* 569, 677–684. doi: 10.1113/jphysiol.2005.095869

Rosenbaum, A. L. (1999). The goal of adult strabismus surgery is not cosmetic. *Arch. Ophthalmol.* 117:250. doi: 10.1001/archopht.117.2.250

Schrager, M. A., Kelly, V. E., Price, R., Ferrucci, L., and Shumway-Cook, A. (2008). The effects of age on medio-lateral stability during normal and narrow base walking. *Gait Posture* 28, 466–471. doi: 10.1016/j.gaitpost.2008.02.009

von Noorden, G. K., and Campos, E. (2002). *Binocular Vision and Ocular Motility: Theory and Management of Strabismus*. St. Louis, MO: Mosby.

Winter, D. A. (1979). *Biomechanics of Human Movement*. New York, NY: Wiley.

Wortham, E., and Greenwald, M. J. (1989). Expanded Binocular peripheral visual fields following surgery for esotropia. *J. Pediatr. Ophthalmol. Strabismus* 26, 109–112.

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.

Received: 27 February 2014; accepted: 16 June 2014; published online: 10 July 2014.
Citation: Aprile I, Ferrarin M, Padua L, Di Sipio E, Simbolotti C, Petroni S, Tredici C and Dickmann A (2014) Walking strategies in subjects with congenital or early onset strabismus. Front. Hum. Neurosci. ***8****:484. doi: 10.3389/fnhum.2014.00484*
This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 02 October 2014
doi: 10.3389/fnhum.2014.00760

Different performances in static and dynamic imagery and real locomotion. An exploratory trial

Augusto Fusco[1,2]*, Marco Iosa[2], Maria Chiara Gallotta[1], Stefano Paolucci[2], Carlo Baldari[1†] and Laura Guidetti[1†]

[1] Department of Movement, Human and Health Sciences, University of Rome Foro Italico, Rome, Italy
[2] Clinical Laboratory of Experimental Neurorehabilitation, IRCCS Fondazione Santa Lucia, Rome, Italy

Edited by:
Leonardo Gizzi, University Hospital Göttingen, Germany

Reviewed by:
Natalie Mrachacz-Kersting, Aalborg University, Denmark
Aidan Moran, University College Dublin, Ireland

***Correspondence:**
Augusto Fusco, Department of Movement, Human and Health Sciences, University of Rome Foro Italico, Piazza L. De Bosis 15, 00135 Rome, Italy
e-mail: a.fusco@hsantalucia.it; augusto.fusco@uniroma4.it

[†] These authors have contributed equally to this work.

Motor imagery (MI) is a mental representation of an action without its physical execution. Recently, the simultaneous movement of the body has been added to the mental simulation. This refers to dynamic motor imagery (dMI). This study was aimed at analyzing the temporal features for static and dMI in different locomotor conditions (natural walking, NW, light running, LR, lateral walking, LW, backward walking, BW), and whether these performances were more related to all the given conditions or present only in walking. We have been also evaluated the steps performed in the dMI in comparison with the ones performed by real locomotion. 20 healthy participants (29.3 ± 5.1 years old) were asked to move towards a visualized target located at 10 mt. In dMI, no significant temporal differences respect the actual locomotion were found for all the given tasks (NW: $p = 0.058$, LR: $p = 0.636$, BW: $p = 0.096$; LW: $p = 0{,}487$). Significant temporal differences between static imagery and actual movements were found for LR ($p < 0.001$) and LW ($p < 0.001$), due to an underestimation of time needed to achieve the target in imagined locomotion. Significant differences in terms of number of steps among tasks were found for LW ($p < 0.001$) and BW ($p = 0.036$), whereas neither in NW ($p = 0.124$) nor LR ($p = 0.391$) between dMI and real locomotion. Our results confirmed that motor imagery is a task-dependent process, with walking being temporally closer than other locomotor conditions. Moreover, the time records of dMI are nearer to the ones of actual locomotion respect than the ones of static motor imagery.

Keywords: walking, dynamic motor imagery, human locomotion, healthy, mental representation, chronometry

INTRODUCTION

Motor imagery (MI) is defined as a mental rehearsal of an action without its actual performance (Decety, 1996). Evidences have been provided that MI has positive effects on the performance of motor skills, likely by developing of a better implicit understanding about spatial and kinesthetic features required for completing the task (Driskell et al., 1994; Callow et al., 2013). Thereafter, MI has attracted increasing interest of researchers in sport science, psychology, cognitive neuroscience, and, finally, for promoting recovery after a neurological damage in medical sciences worldwide (Müller et al., 2007; Schuster et al., 2011).

Functional imaging studies have shown as motor imagery and motor preparation and execution partially share the same brain networks, as parietal cortex, cortical motor areas, basal ganglia and cerebellum, supporting the hypothesis of functional equivalence (Jeannerod, 1994; Holmes and Collins, 2001; Guillot et al., 2008; Macuga and Frey, 2012).

Many conditions have been pointed out for their role in influencing the MI. For example, MI has been recognized to be closer to real movement when performed with a body posture consistent with the one needed for executing the required action. Conversely, when body posture was not coherent with the imagined movement, the characteristics of similarity were found reduced (Vargas et al., 2004; Fourkas et al., 2006; Saimpont et al., 2012). In addition, the environment and the context in which the MI is performed may improve the mental rehearsal by facilitating the formation of more vivid and precise mental images (Guillot and Collet, 2005; Callow et al., 2006). Recently, it has been hypothesized the existence of an egocentric internal model representing the features of own body and its interaction with the external environment, identified as self-centered mental imagery (Land, 2014). This is a cerebral region deputed to form our conscious percept of a stable world, providing information required by the motor system.

With the introduction of theories related to the Motor Cognition, the same concept of MI is partially changed, revising it as a part of a continuum between preparation and execution of an action where the intentional movement is related to a command following imagined actions (Jeannerod, 2006; MacIntyre and Moran, 2010). Under a pragmatic perspective, the above may explain why MI could impact positively on movements and vice-versa.

Most of the studies on MI have been focused on arm and hand movements. Locomotion received less attention, probably

because it involves simulated full-body movements and the concurrent use of environmental information (Kunz et al., 2009). Studies have showed that imagined and physically executed actions share common features. Participants increased the time spent in imagining the walk towards a target located at different distances (Courtine et al., 2004; Plumert et al., 2005; Kunz et al., 2009), consistently with the Fitts' model on human movement (Fitts, 1954). Further, when participants were asked to walk blindfolded towards a target, they had to combine the motor acts with the imagination (Mittelstaedt and Mittelstaedt, 2001). When MI tasks were assigned adding some constraints, such as the requirement of walking at slower/faster speeds (Bredin et al., 2005) or with shorter/longer steps (Mittelstaedt and Mittelstaedt, 2001) or walking on stilts (Dominici et al., 2009), participants' motor performances were negatively affected, likely due to the difficulties in imagining an action out of its standard execution.

Recently, the simultaneous coupling of imagination and movement has been introduced in MI trials, referring to it as "dynamic motor imagery" (dMI; MacIntyre and Moran, 2010). This definition concerns about a specific sequence in the MI processes which are associated to movements miming in part those mentally represented, with the same specific features of the action in relation to temporal or spatial invariance (Guillot et al., 2013). The dMI is conceptually different from MI, which is a condition that occurs in the absence of any overt or potential movement (Guillot and Collet, 2010). However, despite its theoretical definition, trials have evidenced that, during MI, a subliminal muscular activity was possible, suggesting by it that the motor control is not completely inhibited (Guillot et al., 2007; Lebon et al., 2008). Consequently, it has been suggested that a motor output is possible and may be included within MI (Morris et al., 2005).

In some preliminary studies where imagery and movement were associated, Callow et al. (2006) reported as high-level junior skiers who moved the body from side to side, simulating the actions during a skiing competition, experienced more vivid images and increased their confidence in the performance of athletic movements. Vergeer and Roberts (2006) reported the improvement of stretching exercises in terms of flexibility in participants with more vivid imagery of their exercises. Finally, a recent study has shown that the technical performance can be improved in active high jumpers (Guillot et al., 2013). These studies have led some authors to conclude that applying a dynamic support to the imagination could result in improvements of the performance (Smith et al., 2007). Nevertheless, at the current stage data are still poor.

The first aim of this study was to evaluate if dynamic MI is equivalent or superior to MI in representing temporal features of real execution for different types of locomotion. These different locomotor conditions were normal walking, light running, lateral walking and backward walking. Because MI performances have shown to be related to the usual physical practice (Aglioti et al., 2008), we have also hypothesized that both MI and dMI could be closer to real performances during normal walking than into the other tested locomotor conditions.

MATERIALS AND METHODS

PARTICIPANTS AND PROTOCOL

Twenty healthy volunteers were enrolled in this study (8 males; 12 females; mean age: 29.3 ± 5.1 years). They were asked to stand on a line marked on the floor in front of another line taped on the ground, at a distance of at 10 mt, unknown by the participants.

They were asked to imagine achieving the target in one of four possible randomized conditions: normal walking (W), light running (LR), lateral walking (LW), and backward walking (BW). Imagery could be accompanied by stepping in place (dynamic motor imagery, dMI) or not (static motor imagery, sMI) the required type of locomotion.

After having performed these tasks for all requested conditions, individuals were asked to really perform the task, going towards the target according to the randomizations sequence. The randomized sequence of locomotor types was repeated for each one of the three tasks, sMI, dMI, real performance (RP), performed in this order.

To avoid some possible learning and/or cognitive influencing effects, we tested naïve subjects to the requested tasks. No verbal information were given to the participants about the target distance and their performances, as similarly conducted in previous studies (Bredin et al., 2005; Iosa et al., 2012a). The experimental trial was conducted in the same indoor environment, to avoid possible influence of different settings (Lappin et al., 2006; Iosa et al., 2012a).

The study was conducted in accordance with the Declaration of Helsinki about experiments on human subjects. This study was approved by the local ethical committee of our institution (Research Rehabilitation Hospital) where all tests were carried out. Signed informed consent was obtained from each participant.

MEASUREMENT SETTINGS

The main measures were related to temporal features among static imagery, dynamic imagery and actual performance for all the analyzed forms of locomotion. The response time data was the main outcome measure for all behavioral experiments. Consistently with previous studies, we compared the duration of the performance between real and imagined motor acts (Collet et al., 2011). In the assessment of MI, the chronometric tests have widely proved to be a reliable technique both in healthy subjects and in patients (Malouin et al., 2008). Execution time during MI is close to that of actual execution (Guillot and Collet, 2005). We do not use self-report inventories to measure MI performances in order to avoid the risk to involve subject's conscious awareness, potentially altering the data (Collet et al., 2011).

During sMI, time was measured using a chronometer by a professional sports chronograph digital timer stopwatch (JUNSD®). Participants were instructed to use the chronograph by themselves to mark the beginning and the end of each trial, as already performed in previous study (Lebon et al., 2012). During dMI, measures were taken with accelerometers. They work by means of a wearable inertial sensor device (FreeSense, Sensorize s.r.l., Rome; sampling frequency = 100 Hz), located inside an elastic belt on an area of their back corresponding to the L2-L3 spinous processes, close to the body center of mass.

Accelerometric inertial sensor, a suitable simple quantitative technique, was used to objectively assess the dynamic gait stability of subjects during walking and to estimate spatiotemporal parameters in many previous studies both in healthy subjects and in patient with several diseases (Iosa et al., 2012b,c,d, 2013). This device is lightweight (93 g) and contains a triaxial accelerometer to measure accelerations along the three body axes (antero-posterior, AP; latero-lateral, LL; and cranio-caudal, CC) and three gyroscopes to measure angular velocities around the above axes. We used it to measure movement time and for estimating the number of performed steps (corresponding to the negative peak of antero-posterior acceleration) in place or along the pathway in dMI and RP. The acceleration data were recorded during consecutive steps and these signals were analyzed after the subtraction of their mean values and after low-pass filtering at 20 Hz and were summarized in parameters for each body axis, as commonly used in previous studies (Iosa et al., 2012d,e).

STATISTICAL ANALYSIS

Means and standard deviations were computed for all the investigated parameters (demographic data of subjects, spatio-temporal results obtained by tests). Repeated measure analysis of variance using as within subject factor the task (sMI, dMI, RP) were performed using time as dependent variable and number of steps performed by subjects for each one of the four locomotion condition (normal walking, light running, backward walking and lateral walking). *Post-hoc* analyses were performed when needed, correcting the level of significance in accordance to Bonferroni correction ($p < 0.025$); for all the other analyses this level was set at 0.05. Pearson's correlation coefficient (R) was computed for evaluating the relationship between the mean values computed into the four different locomotor types between sMI and RP, dMI and RP, and between sMI and RP.

RESULTS

The mean time spent by participants during the three tasks (sMI, dMI, RP) in the four locomotor conditions (light running, normal walking, backward walking and lateral walking) is shown in **Figure 1**.

Repeated measure analysis of variance showed that significant differences were found for all conditions among tasks, except for backward walking (see **Table 1**). *Post-hoc* analyses revealed that: (i) time was not different between dMI and RP for any of the locomotor condition; (ii) time was different between sMI and RP for light running and lateral walking; and (iii) time was different between sMI and dMI for light running, normal and lateral walking.

The temporal correlation between simulated dMI and RP resulted statistically significant ($R = 0.972$, $p = 0.028$, as evident in **Figure 1**). The correlation between sMI and RP was lower and not statistically significant ($R = 0.890$, $p = 0.110$). As shown in **Figure 1**, it was mainly due to an underestimation of time needed to achieve the target by lateral walking during static motor imagery.

Number of steps could be analyzed only in dMI and RP. Repeated measure analysis of variance showed significant differences in lateral walking ($F = 22.733$, $p < 0.001$) and in backward walking ($F = 5.087$, $p = 0.036$), whereas neither in normal walking ($F = 2.609$, $p = 0.124$) nor in running ($F = 0.770$, $p = 0.391$) significant differences were observed in terms of performed steps between dMI and real performances. As clearly shown in **Figure 2**, there was a significant correlation in terms of number of steps between dMI and RP ($R = 0.991$, $p = 0.009$).

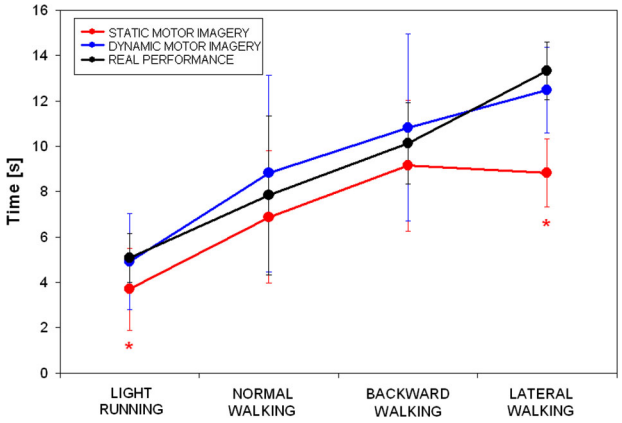


FIGURE 1 | Mean and standard deviations of time spent during the three tasks (static motor imagery in red, dynamic motor imagery in blue and actual locomotion in black) in the four locomotor conditions (* $p < 0.025$ in the *post hoc* showed in Table 1).

DISCUSSION

The first aim of this study was to investigate whether dMI was superior to static motor imagery into imagining the achievement of a target placed at given distance (10 mt) in different locomotor conditions. Our results clearly supported this hypothesis, with similar temporal performances between dMI and real performances. Conversely, the time spent during sMI, resulted significantly underestimated with respect to the one really needed in light running and lateral walking. Individuals showed a good capacity to mentally estimate the needed time only for normal and backward walking.

We had hypothesized that normal walking could be more easily performed than other types of locomotion, according to idea that usual physical practice could enhance MI (Aglioti et al., 2008), probably for a greater neural overlap (Guillot et al., 2013). This hypothesis was supported by our results. Surprisingly, a temporal similarity between sMI and RP was obtained also during backward walking. Despite some differences, the kinematics of forward and backward human locomotion are quite similar (Viviani et al., 2011), and it could explain our results.

For the other locomotor conditions (light running and lateral walking), significant differences were found between sMI and RP. This result suggests that motor imagery could be a process dependent on the motor act to imagine. At the same time, this dependency was not significant when imagery is coupled with external movements miming in part those mentally represented.

Table 1 | Results of repeated measure analysis of variance and relevant *post-hoc* Analyses on time spent by subjects in the three tasks: static motor imagery (sMI), dynamic motor imagery (dMI) and real performance (RP) (* $p < 0.025$).

Time	Analysis of variance		*Post-hoc*		
Locomotor task	***F***	***P***	**sMI vs. RP**	**dMI vs. RP**	**sMI vs. dMI**
Light running	16.011	**<0.001**	**<0.001***	0.636	**<0.001***
Normal walking	9.433	**<0.001**	0.041	0.058	**<0.001***
Backward walking	**2.494**	0.096	—	—	—
Lateral walking	11.392	**<0.001**	**<0.001***	0.487	**<0.001***

For dMI, we have also analyzed the number of simulated steps. They resulted significantly different from those really performed to achieve the target only during backward and lateral walking. Especially, for this last condition, the number of steps was roughly halved in respect of actual one. Hence, lateral walking resulted the task more difficult to be imagined. Differently from the other locomotor conditions, the movement of one leg never pass over the other leg position during actual lateral walking. Likely, participants did not take into account this peculiarity when they had been asked to imagine to perform this task.

Our results confirm that the temporal features of dMI are similar to those of RP even for complex movements (Olsson et al., 2008). At the same time, this was also applicable for spatial features (performed steps) during normal walking and light running. The result could be due to the fact that dMI, more than sMI, involves brain networks with a more vivid imagination of movement with a spatial updating and re-calibration of the distance perception, that are critical factors in locomotor performance, especially for less complex movements (or, in our case, for more usual movements) (Guillot et al., 2013).

Our results could be also explained on the basis of the theory of internal models. Actually, with regards to the latter, it has been hypothesized the existence of a specific internal model related to walking, called locomotor body schema (Dominici et al., 2009). Spino-cerebellar neuronal networks could encode information of limb length, combining them with information of limb kinematics, for computing a predictive measure of step length and hence of walked distance (Ivanenko et al., 2011).

During MI, the body schema could provide the needed information about limb length, independently if in static or dynamic condition, however, MI could benefit of simulation of limb kinematics, for the proprioceptive and sensory inputs, together with possible information related to the efferent motor commands. Hence, the processing of this information may differ significantly in static and dynamic imagery due to a diversity of the available information, resulting in a better temporal (and for usual locomotion also spatial) correlation between dMI and RP. This locomotor body schema should be taken into account also perceived distance and spatial updating, in accordance with the walking imagery (Loomis et al., 1992; Kunz et al., 2009). However, it should be noted that Guillot et al. have reported that dMI was effective also for upper limb movements: mimicking the gestures of the upper limbs, the athletes were able to improve their kinesthetic ability (Guillot et al., 2013). Our results on dMI support this idea and confirm the functional

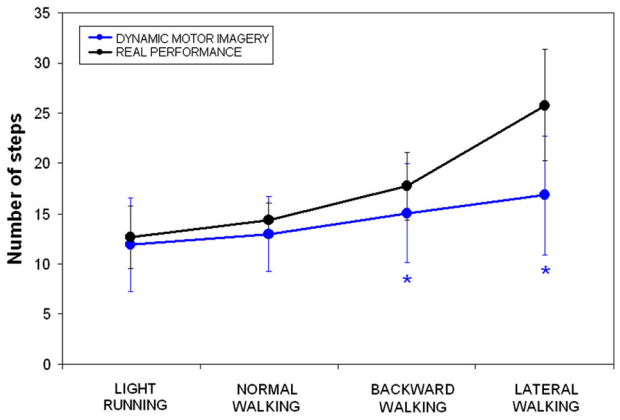


FIGURE 2 | Mean and standard deviations of number of steps during dynamic imagery (blue) and actual (black) locomotion in the four conditions (* $p < 0.025$ in the *post-hoc* analyses).

equivalence between action and imagination (Jeannerod, 1994; Decety, 1996).

Dynamic motor imagery showed temporal similarity with real performances independently by the locomotor conditions, but from a spatial point of view this similarity was limited to walking and running. In consideration of our results, this locomotor body schema seemed to be motor act-dependent and more effective for these two locomotor acts. There are two possible explanations for these results: backward and lateral walking are uncommon in normal daily-living and/or the direction of body progression did not coincide with the front of the body. Lateral and backward walking are more common during such type of sports (for example, basketball, soccer, volley) and future studies could investigate if athletes have better performances than non-athletes in these other types of locomotion. Globally, MI has been found to be more effective in athletes (Rushall and Lippman, 1998; Bredin et al., 2005; Callow et al., 2013).

Limitations of this study were probably due to some uncontrolled conditions. For example, lateral walking was explained to the participants as a lateral movement without leg crossing. Despite of it, and despite of the fact that they correctly performed the test as asked during RP, they be-halved the number of steps into the imagery tasks, such as they imagined both the legs as doing propulsive forward movements. Then, we did not administered a questionnaire about the vividness of motor imagery, as

in previous study (Guillot et al., 2013). Therefore, future studies should focus on the possible differences between simple and complex movements, both for healthy and patients, analyzing the different motor impairments, also using appropriate measures (Beauchet et al., 2010). Differently from previous researches, our study has the worth to have investigated the MI in target-directed different locomotor conditions, possible in the real-life situations and important in such sports.

In conclusion, our study approach was innovative, focusing on the concepts of motor imagery in locomotion. Our results confirmed that motor imagery is a task-dependent process also for the human locomotion. In fact, imagined and executed walking are more temporally closer respect than other locomotor conditions, showing a functional equivalence. Moreover, the time records of dMI are nearer to the ones of actual locomotion respect than the ones of static motor imagery, revealing a potential important role of dMI for improving performances.

ACKNOWLEDGMENTS

This research was supported by funds from the Department of Health Sciences of University of Rome Foro Italico (Cod. RIC042013).

REFERENCES

Aglioti, S. M., Cesari, P., Romani, M., and Urgesi, C. (2008). Action anticipation and motor resonance in elite basketball players. *Nat. Neurosci.* 11, 1109–1116. doi: 10.1038/nn.2182

Beauchet, O., Annweiler, C., Assal, F., Bridenbaugh, S., Herrmann, F. R., Kressig, R. W., et al. (2010). Imagined timed up & go test: a new tool to assess higher-level gait and balance disorders in older adults? *J. Neurol. Sci.* 294, 102–106. doi: 10.1016/j.jns.2010.03.021

Bredin, J., Kerlirzin, Y., and Israël, I. (2005). Path integration: is there a difference between athletes and non-athletes? *Exp. Brain Res.* 167, 670–674. doi: 10.1007/s00221-005-0251-3

Callow, N., Roberts, R., and Fawkes, J. Z. (2006). Effects of dynamic and static imagery on vividness of imagery, skiing performance and confidence. *J. Im. Res. Sport Phys. Act.* 1, 1–13. doi: 10.2202/1932-0191.1001

Callow, N., Roberts, R., Hardy, L., Jiang, D., and Edwards, M. G. (2013). Performance improvements from imagery: evidence that internal visual imagery is superior to external visual imagery for slalom performance. *Front. Hum. Neurosci.* 7:697. doi: 10.3389/fnhum.2013.00697

Collet, C., Guillot, A., Lebon, F., MacIntyre, T., and Moran, A. (2011). Measuring motor imagery: combining psychometric, qualitative, chronometric and psychophysiological techniques. *Exerc. Sport Sci. Rev.* 39, 89–92. doi: 10.1097/jes.0b013e31820ac5e0

Courtine, G., Papaxanthis, C., Gentili, R., and Pozzo, T. (2004). Gait dependent motor memory facilitation in covert movement execution. *Brain Res. Cogn. Brain Res.* 22, 67–75. doi: 10.1016/j.cogbrainres.2004.07.008

Decety, J. (1996). The neurophysiological basis of motor imagery. *Behav. Brain Res.* 77, 45–52. doi: 10.1016/0166-4328(95)00225-1

Dominici, N., Daprati, E., Nico, D., Cappellini, G., Ivanenko, Y. P., and Lacquaniti, F. (2009). Changes in the limb kinematics and walking-distance estimation after shank elongation: evidence for a locomotor body schema? *J. Neurophysiol.* 101, 1419–1429. doi: 10.1152/jn.91165.2008

Driskell, J., Copper, C., and Moran, A. (1994). Does mental practice enhance performance? A meta-analysis. *J. Appl. Psychol.* 79, 481–492. doi: 10.1037/0021-9010.79.4.481

Fitts, P. M. (1954). The information capacity of the human motor system in controlling the amplitude of movement. *J. Exp. Psych.* 47, 381–391. doi: 10.1037/h0055392

Fourkas, A. D., Ionta, S., and Aglioti, S. M. (2006). Influence of imagined posture and imagery modality on corticospinal excitability. *Behav. Brain Res.* 168, 190–196. doi: 10.1016/j.bbr.2005.10.015

Guillot, A., and Collet, C. (2005). Duration of mentally simulated movement: a review. *J. Mot. Behav.* 37, 10–20. doi: 10.3200/jmbr.37.1.10-20

Guillot, A., and Collet, C. (2010). *The Neurophysiological Foundations of Mental and Motor Imagery.* Oxford: Oxford University Press.

Guillot, A., Collet, C., Nguyen, V. A., Malouin, F., Richards, C., and Doyon, J. (2008). Functional neuroanatomical networks associated with expertise in motor imagery ability. *Neuroimage* 41, 471–1483. doi: 10.1016/j.neuroimage.2008.03.042

Guillot, A., Lebon, F., Rouffet, D., Champely, S., Doyon, J., and Collet, C. (2007). Muscular responses during motor imagery as a function of muscle contraction types. *Int. J. Psychophysiol.* 66, 18–27. doi: 10.1016/j.ijpsycho.2007.05.009

Guillot, A., Moschberger, K., and Collet, C. (2013). Coupling movement with imagery as a new perspective for motor imagery practice. *Behav. Brain Funct.* 9:8. doi: 10.1186/1744-9081-9-8

Holmes, P. S., and Collins, D. J. (2001). The PETTLEP approach to motor imagery: a functional equivalence model for sport psychologists. *J. Appl. Sport Psychol.* 13, 60–83. doi: 10.1080/104132001753155958

Iosa, M., Fusco, A., Morone, G., and Paolucci, S. (2012a). Walking there: environmental influence on walking-distance estimation. *Behav. Brain Res.* 226, 124–132. doi: 10.1016/j.bbr.2011.09.007

Iosa, M., Fusco, A., Morone, G., and Paolucci, S. (2012b). Effects of visual deprivation on gait dynamic stability. *ScientificWorldJournal* 2012:974560. doi: 10.1100/2012/974560

Iosa, M., Fusco, A., Morone, G., Pratesi, L., Coiro, P., Venturiero, V., et al. (2012c). Assessment of upper-body dynamic stability during walking in patients with subacute stroke. *J. Rehabil. Res. Dev.* 49, 439–450. doi: 10.1682/jrrd.2011.03.0057

Iosa, M., Marro, T., Paolucci, S., and Morelli, D. (2012e). Stability and harmony of gait in children with cerebral palsy. *Res. Dev. Disabil.* 33, 129–135. doi: 10.1016/j.ridd.2011.08.031

Iosa, M., Morelli, D., Marro, T., Paolucci, S., and Fusco, A. (2013). Ability and stability of running and walking in children with cerebral palsy. *Neuropediatrics* 44, 147–154. doi: 10.1055/s-0033-1336016

Iosa, M., Morone, G., Fusco, A., Pratesi, L., Bragoni, M., Coiro, P., et al. (2012d). Effects of walking endurance reduction on gait stability in patients with stroke. *Stroke Res. Treat.* 2012:810415. doi: 10.1155/2012/810415

Ivanenko, Y. P., Dominici, N., Daprati, E., Nico, D., Cappellini, G., and Lacquaniti, F. (2011). Locomotor body scheme. *Hum. Mov. Sci.* 30, 341–351. doi: 10.1016/j.humov.2010.04.001

Jeannerod, M. (1994). The representing brain: neural correlates of motor intention and imagery. *Behav. Brain Sci.* 17, 187–245. doi: 10.1017/s0140525x00034026

Jeannerod, M. (2006). *Motor Cognition*. New York: Oxford University Press.

Kunz, B. R., Creem-Regehr, S. H., and Thompson, W. B. (2009). Evidence for motor simulation in imagined locomotion. *J. Exp. Psychol. Hum. Percept. Perform.* 35, 1458–1471. doi: 10.1037/a0015786

Land, M. F. (2014). Do we have an internal model of the outside world? *Philos. Trans. R. Soc. Lond. B Biol. Sci.* 369:20130045. doi: 10.1098/rstb.2013.0045

Lappin, J. S., Shelton, A. L., and Rieser, J. J. (2006). Environmental context influences visually perceived distance. *Percept. Psychophys.* 68, 571–581. doi: 10.3758/bf03208759

Lebon, F., Byblow, W. D., Collet, C., Guillot, A., and Stinear, C. M. (2012). The modulation of motor cortex excitability during motor imagery depends on imagery quality. *Eur. J. Neurosci.* 35, 323–331. doi: 10.1111/j.1460-9568.2011.07938.x

Lebon, F., Rouffet, D., Collet, C., and Guillot, A. (2008). Modulation of EMG power spectrum frequency during motor imagery. *Neurosci. Lett.* 435, 181–185. doi: 10.1016/j.neulet.2008.02.033

Loomis, J. M., Da Silva, J. A., Fujita, N., and Fukusima, S. S. (1992). Visual space perception and visually directed action. *J. Exp. Psychol. Hum. Percept. Perform.* 18, 906–921. doi: 10.1037//0096-1523.18.4.906

MacIntyre, T., and Moran, A. (2010). "Meta-imagery processes among elite sports performers," in *The Neurophysiological Foundations of Mental and Motor Imagery*, eds C. Collet and A. Guillot (New York: Oxford University Press), 227–244.

Macuga, K. L., and Frey, S. H. (2012). Neural representations involved in observed, imagined and imitated actions are dissociable and hierarchically organized. *Neuroimage* 59, 2798–2807. doi: 10.1016/j.neuroimage.2011.09.083

Malouin, F., Richards, C., Durand, A., and Doyon, J. (2008). Reliability of mental chronometry for assessing motor imagery ability after stroke. *Arch. Phys. Med. Rehab.* 89, 311–319. doi: 10.1016/j.apmr.2007.11.006

Mittelstaedt, M. L., and Mittelstaedt, H. (2001). Idiothetic navigation in humans: estimation of path length. *Exp. Brain Res.* 139, 318–332. doi: 10.1007/s002210100735

Morris, T., Spittle, M., and Watt, A. P. (2005). *Imagery in Sport.* Champaign, IL: Human Kinetics.

Müller, K., Bütefisch, C. M., Seitz, R. J., and Hömberg, V. (2007). Mental practice improves hand function after hemiparetic stroke. *Restor. Neurol. Neurosci.* 25, 501–511.

Olsson, C. J., Jonsson, B., and Nyberg, L. (2008). Internal imagery training in active high jumpers. *Scand. J. Psychol.* 49, 133–140. doi: 10.1111/j.1467-9450.2008.00625.x

Plumert, J. M., Kearney, J. K., Cremer, J. F., and Recker, K. (2005). Distance perception in real and virtual environments. *ACM Trans. Appl. Perc.* 2, 216–233. doi: 10.1145/1077399.1077402

Rushall, B. S., and Lippman, L. G. (1998). The role of imagery in physical performance. *Int. J. Sport Psychol.* 29, 57–72.

Saimpont, A., Malouin, F., Tousignant, B., and Jackson, P. L. (2012). The influence of body configuration on motor imagery of walking in younger and older adults. *Neuroscience* 222, 49–57. doi: 10.1016/j.neuroscience.2012.06.066

Schuster, C., Hilfiker, R., Amft, O., Scheidhauer, A., Andrews, B., Butler, J. A., et al. (2011). Best practice for motor imagery: a systematic literature review on motor imagery training elements in five different disciplines. *BMC Med.* 9:75. doi: 10.1186/1741-7015-9-75

Smith, D., Wright, C., Allsopp, A., and Westhead, H. (2007). It's all in the mind: PETTLEP-based imagery and sports performance. *J. Appl. Sport Psychol.* 19, 80–92. doi: 10.1080/10413200600944132

Vargas, C. D., Olivier, E., Craighero, L., Fadiga, L., Duhamel, J. R., and Sirigu, A. (2004). The influence of hand posture on corticospinal excitability during motor imagery: a transcranial magnetic stimulation study. *Cereb. Cortex* 14, 1200–1206. doi: 10.1093/cercor/bhh080

Vergeer, E., and Roberts, J. (2006). Movement and stretching imagery during flexibility training. *J. Sports Sci.* 24, 197–208. doi: 10.1080/02640410500131811

Viviani, P., Figliozzi, F., Campione, G. C., and Lacquaniti, F. (2011). Detecting temporal reversals in human locomotion. *Exp. Brain Res.* 214, 93–103. doi: 10.1007/s00221-011-2809-6

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.

Received: 28 February 2014; accepted: 09 September 2014; published online: 02 October 2014.

Citation: Fusco A, Iosa M, Gallotta MC, Paolucci S, Baldari C and Guidetti L (2014) Different performances in static and dynamic imagery and real locomotion. An exploratory trial. Front. Hum. Neurosci. ***8****:760. doi: 10.3389/fnhum.2014.00760*

This article was submitted to the journal Frontiers in Human Neuroscience.

frontiers in
HUMAN NEUROSCIENCE

ORIGINAL RESEARCH ARTICLE
published: 27 October 2014
doi: 10.3389/fnhum.2014.00859

The brain's sense of walking: a study on the intertwine between locomotor imagery and internal locomotor models in healthy adults, typically developing children and children with cerebral palsy

***Marco Iosa*[1]*, *Loredana Zoccolillo*[2], *Michela Montesi*[1,3], *Daniela Morelli*[2], *Stefano Paolucci*[1] *and Augusto Fusco*[1]**

[1] *Clinical Laboratory of Experimental Neurorehabilitation, IRCCS Fondazione Santa Lucia, Rome, Italy*
[2] *Department of Children Neurorehabilitation, IRCCS Fondazione Santa Lucia, Rome, Italy*
[3] *School of Physiotherapy, University of Rome Tor Vergata, IRCCS Fondazione Santa Lucia, Rome, Italy*

Edited by:
Nadia Dominici, VU University Amsterdam, Netherlands

Reviewed by:
Charalambos Papaxanthis, Université de Bourgogne, France
Nadia Dominici, VU University Amsterdam, Netherlands

***Correspondence:**
Marco Iosa, Clinical Laboratory of Experimental Neurorehabilitation, IRCCS Fondazione Santa Lucia, Via Ardeatina 306, 00179, Rome, Italy
e-mail: m.iosa@hsantalucia.it

Motor imagery and internal motor models have been deeply investigated in literature. It is well known that the development of motor imagery occurs during adolescence and it is limited in people affected by cerebral palsy. However, the roles of motor imagery and internal models in locomotion as well as their intertwine received poor attention. In this study we compared the performances of healthy adults (n = 8, 28.1 ± 5.1 years old), children with typical development (n = 8, 8.1 ± 3.8 years old) and children with cerebral palsy (CCP) (n = 12, 7.5 ± 2.9 years old), measured by an optoelectronic system and a trunk-mounted wireless inertial magnetic unit, during three different tasks. Subjects were asked to achieve a target located at 2 or 3 m in front of them simulating their walking by stepping in place, or actually walking blindfolded or normally walking with open eyes. Adults performed a not significantly different number of steps (p = 0.761) spending not significantly different time between tasks (p = 0.156). Children with typical development showed task-dependent differences both in terms of number of steps (p = 0.046) and movement time (p = 0.002). However, their performance in simulated and blindfolded walking (BW) were strictly correlated (R = 0.871 for steps, R = 0.673 for time). Further, their error in BW was in mean only of −2.2% of distance. Also CCP showed significant differences in number of steps (p = 0.022) and time ($p < 0.001$), but neither their number of steps nor their movement time recorded during simulated walking (SW) were found correlated with those of blindfolded and normal walking (NW). Adults used a unique strategy among different tasks. Children with typical development seemed to be less reliable on their motor predictions, using a task-dependent strategy probably more reliable on sensorial feedback. CCP showed less efficient performances, especially in SW, suggesting an altered locomotor imagery.

Keywords: gait, motor imagery, internal model, locomotion, locomotor body schema, cerebral palsy

INTRODUCTION

"Go where I'm looking, not look where I'm going", with this expression Alain Berthoz in his book "The Brain's sense of movement" claimed the role of gaze-based feed-forward control involved in locomotion along a desired trajectory (Berthoz, 2000). In fact, gaze turns towards the desired trajectory in advance of the feet, suggesting that subjects follow an internal model of the predicted trajectory. When visual feedback is not available, such as during walking in a dark environment, control of locomotion should rely even more on motor predictions (Iosa et al., 2012a).

Two neural mechanisms underlie the mental representation of an action (Ito, 2008; Wolpert and Flanagan, 2010): motor imagery (Beisteiner et al., 1995) and internal motor models (Kawato, 1999). Motor imagery has been defined as a voluntary dynamic state during which a subject mentally simulates a given action (Decety, 1996). Similar neural structures are involved in motor imagery and movement execution: parietal cortex, cortical motor areas, basal ganglia and cerebellum (Decety, 1996; Jeannerod, 2001). Many studies reported that the time imagined as needed for executing an action is strictly related to that really needed to complete that action. Motor imagery is developed during childhood and it reaches an asymptote during adolescence (Smits-Engelsman and Wilson, 2013). This development has been found altered in children with cerebral palsy (CCP; Mutsaarts et al., 2006, 2007).

An internal model is an implicit cerebellar neural mechanism (Wolpert et al., 1998), that can mimic the input/output characteristics, or their inverse, of real structures of our body

(such as sensori-motor apparatus (Kawato, 1999)) or of the external environment (such as ecological invariants (McIntyre et al., 2001; Zago et al., 2004)). Internal models are conceivably located into the cerebellum and allow for anticipating consequences of an action given the actual state and a-priori information (feedforward internal models) or for computing the commands needed to obtain a desired trajectory (inverse internal models) (Imamizu et al., 2000).

It has been suggested that motor imagery can be regarded as the conscious experience of internal models (Jeannerod, 2001). On the other hand, internal sensori-motor models could be useful both during motor performances in predicting the motion of body segments as well as during motor imagery (Gentili et al., 2004). The development of motor imagery during childhood was hypothesized to be related to the fact that with age, children become less reliant on feedback and more attuned to feedforward control of movements, capturing aspects of motor prediction and involving a-priori information in their motor behaviors (Smits-Engelsman and Wilson, 2013).

Many studies were focused on the mental representation of walking, mainly based on two different approaches: those in which subjects were explicitly asked to imagine to walk and mental chronometry paradigm was used to verify if imagination time is related to real movement time (Decety, 1996; Bakker et al., 2007), and those in which subjects were asked to perform an action without the support of feedback, such as blindfolded walking (BW) towards a target the position of which was previously memorized (Dominici et al., 2009; Iosa et al., 2012a). This last ability implies the use of an internal model, sometimes called locomotor body schema for computing the needed number of steps suitable to achieve the target in absence of visual support (Dominici et al., 2009; Ivanenko et al., 2011). Despite locomotor imagery and locomotor internal model are conceivably intertwined (Jeannerod, 2001; Gentili et al., 2004), subjects' performances during walking imagery task and walking distance estimation have been rarely compared (Decety et al., 1989).

Many studies reported an alteration of motor imagery in CCP (Mutsaarts et al., 2006, 2007; Crajé et al., 2010), despite a recent study questioned it for locomotor imagery (Spruijt et al., 2013). In children with developmental coordination disorder their problems in generating a mental representation of an intended action have been hypothesized to be associated with internal modeling deficit (Gabbard and Bobbio, 2011). No studies investigated if the alterations in motor imagery in CCP have been due to (or associated with) alterations in their locomotor internal model, evaluated by means of a walking distance estimation test performed without visual support.

According with the scenario depicted above, we tested locomotor imagery and locomotor internal model in three populations of subjects hypothesizing that their intertwine could be: (1) strict in healthy adults, (2) not completely formed yet in typically developing children (TDC); and (3) altered in CCP.

MATERIAL AND METHODS

PARTICIPANTS

Three groups of subjects were enrolled in this study: healthy adult group (HAG: 28.1 ± 5.1 years, age range: 23–37 years; 8 subjects: 3 males and 5 females), children clinically defined as typically developing by their pediatrician (TDC: 8.1 ± 3.8 years, age range: 4–14 years; 8 subjects: 4 males and 4 females) and a group formed by (CCP: 7.5 ± 2.9 years, 12 subjects: 7 males and 5 females). The two groups of children were age-matched ($p = 0.785$, u-test). CCP were affected by hemi- or diparesis, had a walking ability classified as level I in the Gross Motor Function Classification System (Palisano et al., 1997), and were able to understand the given instructions (IQ >= 49 assessed by a psychologist using the revised version of Wechsler Intelligence Scale for Children). Their age range was 4–12 years; their mean IQ was 85 ± 20 (range: 53–110). Four subjects had left hemiparesis, five right hemiparesis, three subjects had diparesis.

Local ethics committee approved the study procedures, designed in accordance to the Declaration of Helsinki on human experimentation and the signed informed consent of adult subjects or parents/legal guardian of children were obtained.

TASKS

Because mental representation by itself is difficult to be measured, all previous studies used paradigms in which outcome measures were hypothesized to be representative of these internal representation, such as measuring response time in Hand Laterality Judgment Task to assess motor imagery (Boonstra et al., 2012). In our study, subjects performed the three tasks described in details below: simulated walking (SW) to assess locomotor imagery, BW to assess locomotor internal model, and normal walking (NW).

Subjects were asked to stand on a strip of tape fixed on the ground (starting line) and to image or to actually walk towards one of two possible targets formed by two other strips placed on the ground at 2 and 3 m from the starting-line and parallel to that. All subjects performed the tasks wearing their common shoes. For each one of the following tasks, both distances (2 and 3 m) were tested in a randomized order. Because it has been demonstrated a practice effect occurring in repeated measurements of motor imagery tasks, implying a progressive improvement of the performances over trial repetition (Philbeck et al., 2008; Boonstra et al., 2012), only one trial was performed for each distance. So, instead of testing two or more trials for the same distance, we preferred to test two slightly different distances for avoiding the phenomenon of learning and/or progressive recalibration, similarly to what done in some previous studies (Smith et al., 2010; Iosa et al., 2012a). Only two distances were tested for avoiding the possible reduction of compliance in children. Target distances were set at 2 and 3 m because it has been shown that even healthy adults significantly undershot the target in indoor environments for distance longer than 3 m, probably because adopting a conservative strategy leaded by the fear to hit a wall (Iosa et al., 2012a).

In the first task SW, subjects were asked to image to walk towards the target and at the same time to simulate walking by means of stepping in place. No instructions were given about looking or not the target during imagination, so subjects could freely decide where to look or to close their eyes. No information was given about the fact that we recorded their number of steps

and movement time, neither these parameters were mentioned to subjects.

In the second task BW subjects were asked to walk towards the target after being blindfolded. During these trials, subjects were reassured that an experimenter can promptly advise them if they were going to hit a wall, but none of them needed his intervention. According to previous study (Iosa et al., 2012a), to avoid some possible learning effect of the first trial on the second one, no verbal feedback was given to the participants about their performances and they had been guided back to the starting-line still blindfolded by the experimenter.

In the last task NW, subjects were asked to stand on the starting-line and then to achieve by NW the target line formed by tape on the ground with their eyes opened at their self-selected comfortable speed. This test was performed in order to measure the normal self-selected spatio-temporal gait parameters under visual control.

MEASURES

All the tests were performed within a rectangle (length: 6 m, width: 2.5 m) formed by optoelectronic bars placed on the ground in our laboratory (Optogait® with inertial unit gyko, Microgate, Italy; sampling frequency = 100 Hz). Half of the electronic bars contained an infrared light emitter each 1.04 cm and the other half a receiver at the same distance. This optoelectronic system was used for measuring the number of performed steps and their spatio-temporal related parameters. In the blindfolded task, analogously to previous studies (Iosa et al., 2012a,b), the experimenter also measured the distance between the target and the middle point of the two malleoli of the subject with a graduated tape, increasing the resolution from 1 cm to 1 mm. During all the above tasks, participants wore an elastic belt containing a wireless triaxial accelerometer (inertial unit gyko, sampling frequency = 100 Hz) located on the back in correspondence of L2–L3 spinous processes, close to the subject's center of mass, and providing acceleration signals along the three body axis.

The outcome measures for all the three tasks were: number of performed steps (measured by optoelectronic system) and movement time (measured by accelerometer). SW performance was hypothesized to be informative on motor imagery, BW performance on locomotor internal model, and NW was used as reference condition. The accelerometer allowed for identifying start and stop of subject's movements (as done in previous studies (Iosa et al., 2012a,c,d)) and their time difference represented the movement time, i.e., the time spent by subjects during simulated and actual walking. For BW also the error (as well as its absolute value independent by under- or over-shooting the target) was measured as the difference between walked distance and real target distance and it was expressed in percentage of real target distance.

STATISTICAL ANALYSIS

Mean and standard deviation were computed for summarizing the outcome measures. Because of the small sample sizes and because not all the data sets resulted normally distributed, non parametric statistics was used for analyzing data. Friedman's analysis was performed to assess the effect of task (3 levels: simulated, blindfolded, and NW) within groups. For HCG and TDC the number of data included into that analysis was 16 (8 subjects for each group per two distances), whereas for CCP it was 24 (12 subjects for 2 distances). *Post-hoc* comparisons were performed using Wilcoxon signed rank test.

The correlation between the values of each parameter recorded in two different tasks was computed on the above data sets using Spearman's coefficient (R). Percentage differences with respect to NW were computed as the difference between values recorded in SW or BW and that of NW, divided by NW-value and multiplied per 100. Percentage walking error in blindfolded task was computed as distance from target divided by target distance and multiplied per 100. Comparisons among groups were performed using Kruskal-Wallis analysis, and Mann-Whitney u-test was applied for comparing two independent samples, such as in between group *post-hoc* analyses.

The threshold for statistically significant difference was set at 0.05 for all the analyses, but for *post-hoc* tests for which Bonferroni correction was applied.

RESULTS

Figure 1 shows mean and standard deviation of number of steps and movement time for the three groups in the three tasks for the two tested distances. **Table 1** shows the results of Friedman's analyses and relevant *post-hoc* comparisons.

HEALTHY ADULT GROUP (HAG)

In healthy adults, neither the number of steps nor the movement time significantly differed between tasks for healthy adults (**Table 1**). The percentage differences with respect to NW were lower for number of steps than for movement time in SW (4% vs. 26%, respectively, $p = 0.011$) and slightly in BW (1% vs. 10%, $p = 0.100$).

Significant correlations were also found between the recorded values of each parameter in different tasks (**Table 2**). The only exception was the correlation of movement time values between SW and BW, that only approached the significant threshold ($p = 0.089$).

Some of adult subjects undershot the target during BW, resulting in a negative mean spatial error ($-13 \pm 11\%$, in percentage of the distance). The corresponding mean absolute error was $13 \pm 11\%$.

TYPICALLY DEVELOPING CHILDREN (TDG)

As observable in **Figure 1**, for children with typical development, the performances of simulated and BW were similar each other, but quite different from that of NW. In fact, both number of performed steps and especially movement time significantly differed between tasks for these children (**Table 1**). It was due to a significantly higher number of steps and longer time spent during both blindfolded and simulated conditions with respect to NW.

The percentage differences with respect to NW were higher than those observed for healthy adults, but remained lower for number of steps than for movement time (SW: 60% vs. 112%, $p = 0.017$; BW: 49% vs. 97%, $p = 0.010$, respectively).

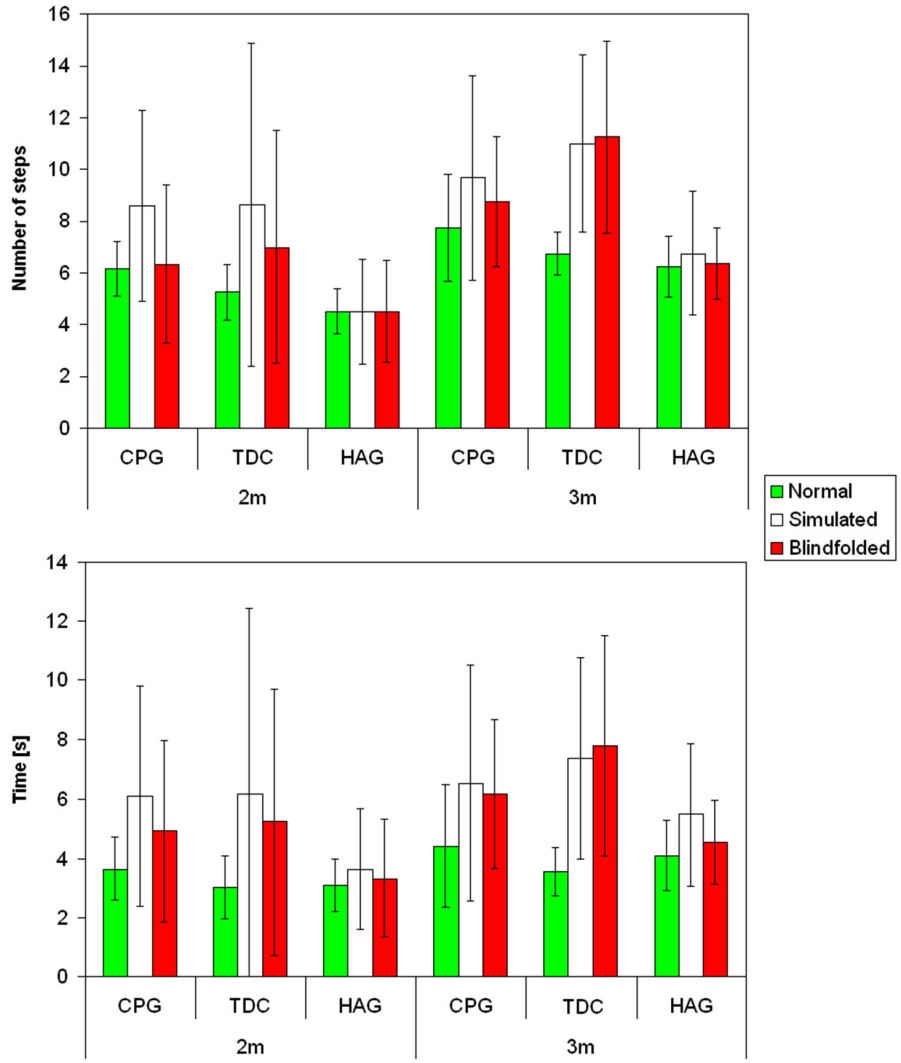


FIGURE 1 | Mean and standard deviations for the number of steps (above) and movement time (below) for children with cerebral palsy (CCP), typically developing children (TDC) and healthy children group (HCG) during normal (green), simulated (white) and blindfolded (red) walking for the two tested distances.

Analogously, significant correlations were found between tasks for the number of steps, whereas the movement time resulted significantly correlated only between SW and BW (**Table 2**).

During BW, the error of children was in mean close to zero: $-2.2 \pm 8.9\%$, with an absolute error of $12.3 \pm 7.3\%$, similar to that found for HAG.

Table 1 | Within group comparisons of number of steps and movement time in healthy adults (HAG), typically developing children (TDC) and children with cerebral palsy (CCP).

Parameter	Group	Friedman's analysis		*Post-hoc* analyses Wilcoxon Signed Rank Test (*p*-value)		
		χ^2	p	SW vs. NW	BW vs. NW	SW vs. BW
Number of steps	HAG	0.545	0.761	—	—	—
	TDC	0.614	**0.046**	**0.010**	**0.006**	0.475
	CCP	7.624	**0.022**	**0.015**	0.175	0.068
Movement time	HAG	3.714	0.156	—	—	—
	TDC	12.133	**0.002**	**0.004**	**<0.001**	0.798
	CCP	24.343	**<0.001**	**<0.001**	**<0.001**	0.170

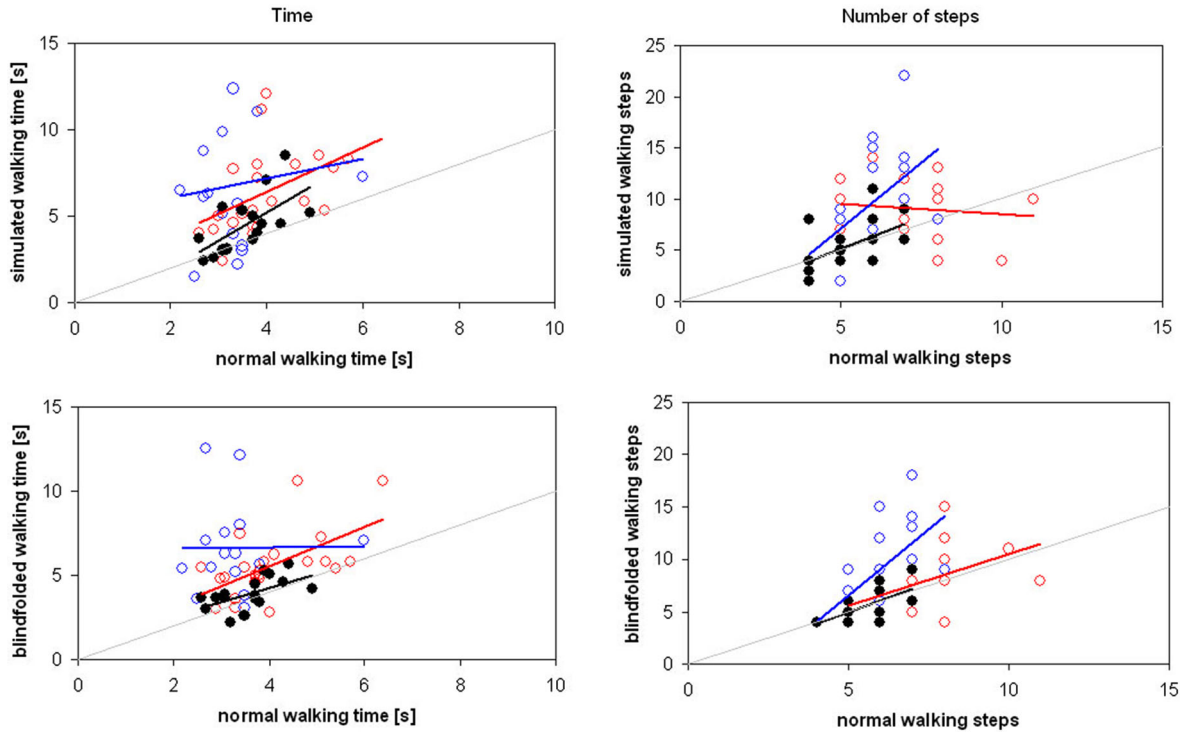


FIGURE 2 | Time (on the left) and number of steps (on the right) spent for covering the distances of 2 and 3 m in simulated (above) and blindfolded (below) walking by adults (black filled circles), children with typical development (blue empty circles) and children with cerebral palsy (red empty circles). Regression lines are reported in the same color of relevant data, whereas the gray line is the theoretical line of equality.

Table 2 | Spearman's correlation coefficient (*R*) between two conditions for time and number of steps.

	Movement time Number of steps	Simulated walking	Blindfolded walking	Normal walking
	Simulated	—	0.439	**0.616***
HAG	Blindfolded	**0.743****	—	**0.629****
	Normal	**0.547***	**0.738****	—
	Simulated	—	**0.673****	0.059
TDC	Blindfolded	**0.871****	—	−0.012
	Normal	**0.504***	**0.682****	—
	Simulated	—	0.344	**0.690****
CCP	Blindfolded	0.034	—	**0.572****
	Normal	−0.070	**0.669****	—

*$p <= 0.05$, **$p <= 0.001$.

CHILDREN WITH CEREBRAL PALSY (CCP)

Figure 1 shows that, the performances of CCP were partially similar to those of children with typical development, but some peculiar differences. Similarly to them (and differently from healthy adults), number of steps and movement time resulted both dependent on tasks (**Table 1**). *Post-hoc* analyses revealed that, especially for number of steps, it was mainly due to differences between simulated and NW. Similarly to HAG and TDC, the percentage differences of number of steps with respect to NW were lower than the related differences of movement time (SW: 37% vs. 47%, $p = 0.458$; BW: 8% vs. 39%, $p < 0.001$).

Differently from children with typical development, the number of steps of CCP during SW was not significantly correlated neither with that of NW nor with that of BW (**Table 2**, **Figure 2**). This result was independent by the target distance (**Figure 3**). It was partially due to six out of the 12 children with CP, who did not imagine that more steps were needed for covering 3 m with respect to 2 m during SW. None adult and just one child with TD showed a similar behavior. Because during BW, this rate was reduced to 2 out of 12 children with CP, the correlation between the number of steps performed in BW and SW was not statistically significant. Moreover, this correlation was completely absent in these six children ($R = -0.114$, $p = 0.724$). If these subjects were excluded from the analysis, the correlation slightly improved, but not significantly ($R = 0.310$, $p = 0.326$). Neither age (7.0 ± 3.2 vs. 8.0 ± 2.6, $p = 0.375$) nor IQ (93.2 ± 20.3 vs. 80.3 ± 16.2, $p = 0.093$) resulted significantly lower in these children with respect to the other six children. Neither the number of steps really performed by these two subgroups of children with CP resulted statistically different (for 2 m: $p = 0.699$, for 3 m: $p = 0.280$). Of these six children, three had left hemiparesis, two right hemiparesis, and one diplegia (the other group was formed by three children with right hemiparesis, two with diplegia and a child with left hemiparesis).

The correlation between number of steps in SW and BW remained not statistically significant even when re-evaluated

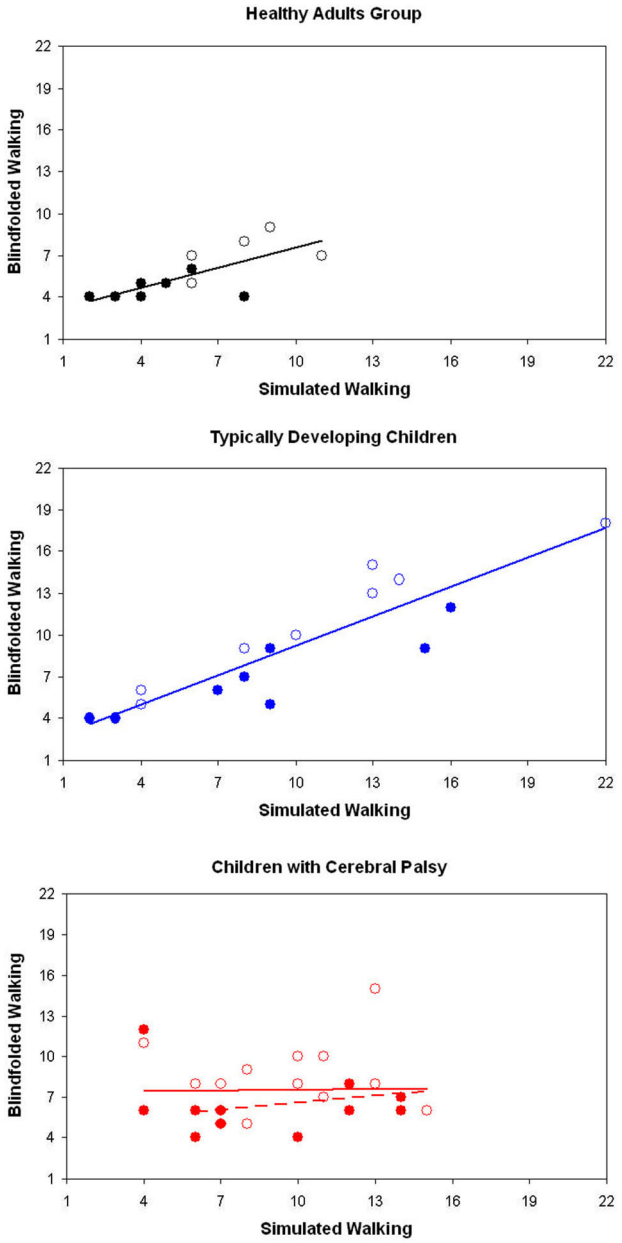


FIGURE 3 | Number of steps recorded during simulated and blindfolded walking for healthy adults (above), typically developing children (in the middle) and children with cerebral palsy (CCP), for the distance of 2 m (filled markers) and 3 m (empty markers). Regression lines are reported in the same color of all the relevant data.

using as covariate IQ-score, age or number of steps needing during NW (IQ: $R = 0.047$; age: $R = 0.136$, steps: $R = 0.095$, respectively, $p > 0.5$ for all of them).

The mean error performed during BW was $-15.5 \pm 21.2\%$ for CCP, with an absolute error of $19.6 \pm 17.3\%$. The mean error was significantly different in the three groups of subjects ($\chi^2 = 9.281$, $p = 0.010$), and *post-hoc* analyses revealed that this difference was significant between CCP and TDC ($p = 0.008$), but not between CCP and HAG ($p = 0.658$).

DISCUSSION

KEY FINDINGS

The aim of this study was to investigate the intertwine between locomotor imagery and internal locomotor model in three groups of subjects. Our results confirmed the hypothesis of a strict intertwine in healthy adults, who showed similar performances in all the three tasks. As shown in **Figure 3**, their performances were correlated also between simulated and BW.

We also hypothesized that this intertwined was not completely formed in TDC. We found that their performance was task-dependent, despite low errors, suggesting an adaptable motor behavior. Anyway, their performances between simulated and BW were strictly correlated. Finally, we hypothesized that the intertwine between locomotor imagery and locomotor internal model could be loss in CCP. Our results confirmed it, suggesting a more marked deficit in motor imagery than in internal model.

The performances in terms of movement time were found even more statistically different between tasks for the two groups of children, and with larger differences with respect to NW in all the three groups. It suggested that the parameter leading the motor strategies was the number of steps. Healthy adults performed in simulating and blindfolded tasks about the same number of steps performed during NW, showing a good ability in estimating the actual number of steps needed for traveling a given distance. It is in accordance with the suggestion that subjects encoded a seen distance in terms of "action units" more than in terms of meters: for adults involved in our study these action units seemed to be the steps needed for covering that distance (Berthoz, 2000). These results can also be read in conjunction with those of Dominici et al. (2009) about blindfolded healthy subjects walking on stilts who undershot the target. Authors explained these results suggesting that subjects had probably planned a reduced number of "expected" longer strides with their lengthened legs, but in real, their actual step length was not longer for a reduction of hip sagittal range of motion, resulting into a reduced traveled distance. This behavior implied that subjects did not take into account proprioceptive signals related to the reduction of hip flexion/extension range or otolith signals related to head forward movement, but based their strategy only on the prediction of the needed steps. It is conceivable that also in our study healthy adults estimated the number of needed steps independently by the task.

Children with typical development showed a different strategy, adapting their number of steps in a task-dependent manner. Despite the differences, the correlation between simulated and BW was statistically significant for both number of steps and movement time. The correlation was statistically significant also for number of steps in simulated and BW with respect to NW, but not for movement time.

These results are in accordance with those reporting that correlation between real and imagined movement times is poor under 8 years, increases during adolescence, and is robust only in adults (Skoura et al., 2009; Smits-Engelsman and Wilson, 2013). Despite it, TDC showed good performances in achieving the target during BW. They adapted their walking in absence of visual feedback performing a higher number of steps and spending more time to complete the trial, probably also taking into account their sensorial feedback. Children could be less self-confident of

adults in their internal representations and rely on sensorial feedbacks for selecting their locomotor strategy (Smits-Engelsman and Wilson, 2013). The reason for which children are less reliant in prediction of movements could be related to the fact that they had not yet a standardized comfortable step length. In fact, they are still developing, implying a progressive increase of step length, that is not only proportional to anthropometric growth (Sutherland et al., 1980), but also related to changes in thigh, shank and foot kinematics (Ivanenko et al., 2004). Also brain development should be taken into account. It has also been shown that the relationship between motor imagery and motor skill becomes stronger with age (Caeyenberghs et al., 2009), achieving an asymptote after adolescence (Smits-Engelsman and Wilson, 2013). Hence, motor imagery development has been suggested to follow brain development and to reflect the unfolding of internal modeling processes in healthy subjects (Caeyenberghs et al., 2009).

However, we found differences between imaging (simulating) walking and predicting the effect of their (blindfolded) walking (i.e., the achievement or not of the target) in CCP. It conceivably suggests that motor imagery ability and its development can only captures some aspects of the implicit processes involved into motor prediction (Smits-Engelsman and Wilson, 2013), and this cognitive capture could be difficult for children with CP. In fact, in CCP, neither number of steps nor movement time resulted significantly correlated between simulated and blindfolded performances. It was mainly due to altered locomotor imagery. In fact, differently from other subjects and differently from real walking, half of children with CP did not imagine more steps needed for covering the longer distance with respect to the shorter one. These subgroup of children were neither younger nor with lower IQ than the other children with CP. Anyway, also in the other children the correlation between simulated and blindfolded performances was poor.

During BW, their mean errors remained quite higher than those performed by age-matched TDC. BW could be a difficult task for children with CP also for their reduced upright gait stability (Iosa et al., 2012d, 2014). However, both number of steps and movement time were found correlated between blindfolded and NW. These results suggested that their motor imagery could be more markedly impaired than their locomotor internal model.

There are some factors that could have affected the performance of CCP during SW, mainly their motor and their cognitive impairments. Further, age could influence both cognitive and motor performances. However, correlation between simulated and BW remained not statistically significant even when locomotor functioning, IQ or age were introduced as covariates. It could be due to the reduced sample size, as reported into the next session about the limit of our study. Howsoever, it was evident that the performance of children with CP was more altered during simulated than during BW. It could be possible that these children needed to have sensorial feedback to improve their performances. Further, their internal locomotor model, being located into cerebellum, could be less impaired than their motor imagery, involving brain areas potentially affected by cerebral palsy.

LIMITS

The main limit of our study was the reduced size of enrolled samples. Despite in line with previous studies (Mittelstaedt and Mittelstaedt, 2001; Stevens, 2005), and sufficient to highlight statistically significant differences, the reduced sample size did not allow us for dividing subjects in homogenous subgroups, for example for dividing children with CP in relationship to the damaged brain areas. Further studies should clarify how the effects of damages in specific areas could impair motor imagination and locomotor predictions.

Then, SW tasks in this study can not be considered as a pure protocol of motor imagery, because it involved stepping in place for eliciting information about motor imagery. Further, subjects involved in this study were asked to perform a "spatial" task towards a target, without any kind of time constraints: it could have limited the role played by time estimations in subjects' performances. The fact that the distances were the same for all the subjects could have increased the task difficulty for CCP because they needed a higher number of steps for covering the given distances. However, their performance in imaging to walk towards a target located at 2 m from them remained poorer than that of TDC for the distance of 3 m. Finally, for avoiding learning effects, subjects performed each trial just one time, not allowing for averaging performances on a wide amount of trials.

THEORY AND FUTURE RESEARCHES

It is not so common for a healthy adult to walk without visual feedback, despite it sometimes happens, especially during night in his/her own house. This ability implies the construction of a mental map of the surrounding environment, but it is also related to the ability of imaging ourselves moving on that map (Palermo et al., 2008). What is the unit of measure of that neural map? Even if adult subjects have poor performance in judging a distance in terms of meters (Iosa et al., 2012a), our results showed that subjects were able to estimate the correct number of steps needed to achieve a target. So, it suggests the hypothesis of an inverse internal model in which a standard step length is encoded as action unit for estimating distances in terms of number of steps needed to cover them.

Children with typical development showed a task-dependent strategy: their more adaptable motor behavior seemed to be less reliant on a-priori internal predictions. However also their performances resulted accurate and it could be due to a proper exploitation of sensorial afferent feedback. How can sensorial feedback be transformed into information about traveled distance? Even if including sensorial feedback this processing needs an internal model. The first possibility is that subjects can use an internal model exploiting their proprioceptive signals of lower limb joint angles and a-priori information about their lower limb length (internalized into their body schema) (Mittelstaedt and Mittelstaedt, 2001; Dominici et al., 2009). Alternatively, traveled distance could be estimated by double-integration of the otolith signals, i.e., by an internal model doubly integrating head accelerations. This last hypothesis is supported by accurate performances of subjects even during blindfolded passive target-directed translations (Israel et al., 1997). It has also been suggested that inertial and proprioceptive information

could be combined and optimized in a task-dependent weighted averaging (Mittelstaedt and Mittelstaedt, 2001). Our results can not clarify which one of these models, or if a combination of them was used by TDC and further studies are needed.

In accordance with literature (Mutsaarts et al., 2006, 2007), CCP showed poor capacity of imagining their motor actions. Further, the errors performed by these children in BW were systematic and brought them to undershoot the target. Many explanations are possible and need further studies to be deeply investigated: a reduced upright gait stability that may lead them to use a more conservative strategy (Iosa et al., 2012a), an altered proprioception (Riquelme and Montoya, 2010), an altered body image (Hammar et al., 2009), or an altered internal locomotor model. This last hypothesis opens a scenario that needs further studies and related to the action-observation processing in children with an altered development. They usually observe healthy subjects, for example their typically developing age-matched school mates, walking with longer steps, and it could contribute to form an altered locomotor body schema, overestimating their walking ability. It has been shown that the subliminal activations of motor pathways during observation of others' actions, mediated by the mirror neuron system, were different between possible and impossible actions, even if these actions have an identical intention (Borroni et al., 2011). What happens when a subject for whom an action is impossible observes another subject performing that action? However, it is noteworthy that even if the performances of CCP during BW showed an error resulting in undershooting the target, the number of steps was found correlated with NW. It did not happen for simulating walking, suggesting the possibility that motor imagery could be more affected than internal locomotor models in these children. Further studies are needed to investigate this scenario, especially because new action-observation rehabilitative protocols have recently been proposed for these children.

Although only a small part of human motor activity is reflected at the conscious level, motor and sensory components of action are deeply intertwined, suggesting inherent linkage between perception and action in the system of internal representation (Jeannerod, 2001; Rizzolatti and Sinigaglia, 2007; Ivanenko et al., 2011). Our results supported the hypothesis of the development of a fascinating link between the internal models probably stored in the cerebellum and their conscious cerebral counterpart forming a sense of walking into the brain.

REFERENCES

Bakker, M., de Lange, F. P., Stevens, J. A., Toni, I., and Bloem, B. R. (2007). Motor imagery of gait: a quantitative approach. *Exp. Brain Res.* 179, 497–504. doi: 10.1007/s00221-006-0807-x

Beisteiner, R., Höllinger, P., Lindinger, G., Lang, W., and Berthoz, A. (1995). Mental representations of movements. Brain potentials associated with imagination of hand movements. *Electroencephalogr. Clin. Neurophysiol.* 96, 183–193. doi: 10.1016/0168-5597(94)00226-5

Berthoz, A. (2000). *The Brain's Sense of Movement* (translated by G. Weiss). Boston: Harvard University Press.

Boonstra, A. M., de Vries, S. J., Veenstra, E., Tepper, M., Feenstra, W., and Otten, E. (2012). Using the hand laterality judgement task to assess motor imagery: a study of practice effects in repeated measurements. *Int. J. Rehabil. Res.* 35, 278–280. doi: 10.1097/MRR.0b013e328355dd1e

Borroni, P., Gorini, A., Riva, G., Bouchard, S., and Cerri, G. (2011). Mirroring avatars: dissociation of action and intention in human motor resonance. *Eur. J. Neurosci.* 34, 662–669. doi: 10.1111/j.1460-9568.2011.07779.x

Caeyenberghs, K., Tsoupas, J., Wilson, P. H., and Smits-Engelsman, B. C. (2009). Motor imagery development in primary school children. *Dev. Neuropsychol.* 34, 103–121. doi: 10.1080/87565640802499183

Crajé, C., Aarts, P., Nijhuis-van der Sanden, M., and Steenbergen, B. (2010). Action planning in typically and atypically developing children (unilateral cerebral palsy). *Res. Dev. Disabil.* 31, 1039–1046. doi: 10.1016/j.ridd.2010.04.007

Decety, J. (1996). The neurophysiological basis of motor imagery. *Behav. Brain Res.* 77, 45–52. doi: 10.1016/0166-4328(95)00225-1

Decety, J., Jeannerod, M., and Prablanc, C. (1989). The timing of mentally represented actions. *Behav. Brain Res.* 34, 35–42. doi: 10.1016/s0166-4328(89)80088-9

Dominici, N., Daprati, E., Nico, D., Cappellini, G., Ivanenko, Y. P., and Lacquaniti, F. (2009). Changes in the limb kinematics and walking-distance estimation after shank elongation: evidence for a locomotor body schema? *J. Neurophysiol.* 101, 1419–1429. doi: 10.1152/jn.91165.2008

Gabbard, C., and Bobbio, T. (2011). The inability to mentally represent action may be associated with performance deficits in children with developmental coordination disorder. *Int. J. Neurosi.* 121, 113–120. doi: 10.3109/00207454.2010.535936

Gentili, R., Cahouet, V., Ballay, Y., and Papaxanthis, C. (2004). Inertial properties of the arm are accurately predicted during motor imagery. *Behav. Brain Res.* 155, 231–239. doi: 10.1016/j.bbr.2004.04.027

Hammar, G. R., Ozolins, A., Idvall, E., and Rudebeck, C. E. (2009). Body image in adolescents with cerebral palsy. *J. Child Health Care* 13, 19–29. doi: 10.1177/1367493508098378

Imamizu, H., Miyauchi, S., Tamada, T., Sasaki, Y., Takino, R., Pütz, B., et al. (2000). Human cerebellar activity reflecting an acquired internal model of a new tool. *Nature* 403, 192–195. doi: 10.1038/35003194

Iosa, M., Fusco, A., Morone, G., and Paolucci, S. (2012a). Walking there: environmental influence on walking-distance estimation. *Behav. Brain Res.* 226, 124–132. doi: 10.1016/j.bbr.2011.09.007

Iosa, M., Fusco, A., Morone, G., and Paolucci, S. (2012b). Effects of visual deprivation on gait dynamic stability. *ScientificWorldJournal* 2012:974560. doi: 10.1100/2012/974560

Iosa, M., Fusco, A., Morone, G., and Paolucci, S. (2014). Development and decline of upright gait stability. *Front. Aging Neurosci.* 6:14. doi: 10.3389/fnagi.2014.00014

Iosa, M., Fusco, A., Morone, G., Pratesi, L., Coiro, P., Venturiero, V., et al. (2012c). Assessment of upper-body dynamic stability during walking in patients with subacute stroke. *J. Rehabil. Res. Dev.* 49, 439–450. doi: 10.1682/JRRD.2011.03.0057

Iosa, M., Marro, T., Paolucci, S., and Morelli, D. (2012d). Stability and harmony of gait in children with cerebral palsy. *Res. Dev. Disabil.* 33, 129–135. doi: 10.1016/j.ridd.2011.08.031

Israel, I., Grasso, R., Georges-Francois, P., Tsuzuku, T., and Berthoz, A. (1997). Spatial memory and path integration studied by selfdriven passive linear displacement I. Basic properties. *J. Neurophysiol.* 77, 3180–3192.

Ito, M. (2008). Control of mental activities by internal models in the cerebellum. *Nat. Rev. Neurosci.* 9, 304–313. doi: 10.1038/nrn2332

Ivanenko, Y. P., Dominici, N., Cappellini, G., Dan, B., Cheron, G., and Lacquaniti, F. (2004). Development of pendulum mechanism and kinematic coordination from the first unsupported steps in toddlers. *J. Exp. Biol.* 207, 3797–3810. doi: 10.1242/jeb.01214

Ivanenko, Y. P., Dominici, N., Daprati, E., Nico, D., Cappellini, G., and Lacquaniti, F. (2011). Locomotor body scheme. *Hum. Mov. Sci.* 30, 341–351. doi: 10.1016/j.humov.2010.04.001

Jeannerod, M. (2001). Neural simulation of action: a unifying mechanism for motor cognition. *Neuroimage* 14, S103–S109. doi: 10.1006/nimg.2001.0832

Kawato, M. (1999). Internal models for motor control and trajectory planning. *Curr. Opin. Neurobiol.* 9, 718–727. doi: 10.1016/s0959-4388(99)00028-8

McIntyre, J., Zago, M., Berthoz, A., and Lacquaniti, F. (2001). Does the brain model Newton's laws? *Nat. Neurosci.* 4, 693–694. doi: 10.1038/89477

Mittelstaedt, M. L., and Mittelstaedt, H. (2001). Idiothetic navigation in humans: estimation of path length. *Exp. Brain Res.* 139, 318–332. doi: 10.1007/s002210100735

Mutsaarts, M., Steenbergen, B., and Bekkering, H. (2006). Anticipatory planning deficits and task context effects in hemiparetic cerebral palsy. *Exp. Brain Res.* 172, 151–162. doi: 10.1007/s00221-005-0327-0

Mutsaarts, M., Steenbergen, B., and Bekkering, H. (2007). Impaired motor imagery in right hemiparetic cerebral palsy. *Neuropsychologia* 45, 853–859. doi: 10.1016/j.neuropsychologia.2006.08.020

Palermo, L., Iaria, G., and Guariglia, C. (2008). Mental imagery skills and topographical orientation in humans: a correlation study. *Behav. Brain Res.* 192, 248–253. doi: 10.1016/j.bbr.2008.04.014

Palisano, R., Rosenbaum, P., Walter, S., Russell, D., Wood, E., and Galuppi, B. (1997). Development and reliability of a system to classify gross motor function in children with cerebral palsy. *Dev. Med. Child Neurol.* 39, 214–223. doi: 10.1111/j.1469-8749.1997.tb07414.x

Philbeck, J. W., Woods, A. J., Arthur, J., and Todd, J. (2008). Progressive locomotor recalibration during blind walking. *Percept. Psychophys.* 70, 1459–1470. doi: 10.3758/pp.70.8.1459

Riquelme, I., and Montoya, P. (2010). Developmental changes in somatosensory processing in cerebral palsy and healthy individuals. *Clin. Neurophysiol.* 121, 1314–1320. doi: 10.1016/j.clinph.2010.03.010

Rizzolatti, G., and Sinigaglia, C. (2007). Mirror neurons and motor intentionality. *Funct. Neurol.* 22, 205–210.

Skoura, X., Vinter, A., and Papaxanthis, C. (2009). Mentally simulated motor actions in children. *Dev. Neuropsychol.* 34, 356–367. doi: 10.1080/87565640902801874

Smith, A. D., Howard, C. J., Alcock, N., and Cater, K. (2010). Going the distance: spatial scale of athletic experience affects the accuracy of path integration. *Exp. Brain Res.* 206, 93–98. doi: 10.1007/s00221-010-2398-9

Smits-Engelsman, B. C., and Wilson, P. H. (2013). Age-related changes in motor imagery from early childhood to adulthood: probing the internal representation of speed-accuracy trade-offs. *Hum. Mov. Sci.* 32, 1151–1162. doi: 10.1016/j.humov.2012.06.006

Spruijt, S., Jouen, F., Molina, M., Kudlinski, C., Guilbert, J., and Steenbergen, B. (2013). Assessment of motor imagery in cerebral palsy via mental chronometry: the case of walking. *Res. Dev. Disabil.* 34, 4154–4160. doi: 10.1016/j.ridd.2013.08.044

Stevens, J. A. (2005). Interference effects demonstrate distinct roles for visual and motor imagery during the mental representation of human action. *Cognition* 95, 329–350. doi: 10.1016/j.cognition.2004.02.008

Sutherland, D. H., Olshen, R., Cooper, L., and Woo, S. L. (1980). The development of mature gait. *J. Bone Joint Surg. Am.* 62, 336–353.

Wolpert, D. M., and Flanagan, J. R. (2010). Motor learning. *Curr. Biol.* 20, R467–R472. doi: 10.1016/j.cub.2010.04.035

Wolpert, D. M., Miall, R. C., and Kawato, M. (1998). Internal models in the cerebellum. *Trends Cogn. Sci.* 2, 338–347. doi: 10.1016/s1364-6613(98)01221-2

Zago, M., Bosco, G., Maffei, V., Iosa, M., Ivanenko, Y. P., and Lacquaniti, F. (2004). Internal models of target motion: expected dynamics overrides measured kinematics in timing manual interceptions. *J. Neurophysiol.* 91, 1620–1634. doi: 10.1152/jn.00862.2003

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.

Received: 26 February 2014; accepted: 06 October 2014; published online: 27 October 2014.

Citation: Iosa M, Zoccolillo L, Montesi M, Morelli D, Paolucci S and Fusco A (2014) The brain's sense of walking: a study on the intertwine between locomotor imagery and internal locomotor models in healthy adults, typically developing children and children with cerebral palsy. Front. Hum. Neurosci. ***8****:859. doi: 10.3389/fnhum.2014.00859*

This article was submitted to the journal Frontiers in Human Neuroscience.