

## Synchronous stimulation and monitoring of soleus H reflex during robotic body weight-supported ambulation in subjects with spinal cord injury

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**Abstract**—We evaluated the accuracy of a novel method for recording the soleus H reflex at specific points in the gait cycle during robotic locomotor training in subjects with spinal cord injury (SCI). Hip goniometric information from the Lokomat system defined midstance and midswing points within the gait cycle. Soleus H reflex stimulation was synchronized to these points during robotic-assisted ambulation at 1.8 and 2.5 km/h. Motor stimulus intensity was monitored and adjusted in real time. Analysis of 50 H reflex cycles during each speed and gait phase showed that stimulation accuracy was within 0.5° of the defined hip joint position and that >85% of the H reflex cycles met the +/-10% M wave criterion that was established during quiet standing. This method allows increased consistency of afferent information into the segmental spinal and supraspinal circuitry and, thus, evaluation of H reflex characteristics during robotic ambulation in subjects with SCI.

**Key words:** body weight-supported treadmill training, gait training, H reflex, locomotor training, motor control, muscle afferents, reflex activity, rehabilitation, robotic-aided training, spinal cord injury.

### INTRODUCTION

The soleus H reflex has been used as a tool for assessing monosynaptic reflex excitability in humans during both rest and voluntary activity [1]. The amplitude of the soleus H reflex is modulated in a task-dependent

manner, such as standing versus walking [2–5]. A profound phase-dependent modulation, including modulation associated with ambulation speed, also occurs during walking and running [2,5–8]. In addition to reflex modulation coupled to muscle activation, isolated joint angle-dependent modulation of H reflex activity has been demonstrated at the ankle [9–10] and the hip [11] independent of motor neuronal excitation. The differences in the H reflex modulation in these tasks is evidence that the changes seen during the gait cycle are not simply due to the  $\alpha$ -motor neuron excitation level, as indicated by electromyography (EMG), but also may be modulated by supraspinal, homonymous, and heteronymous afferent inputs and interneuronal activity, as well as by intrinsic

**Abbreviations:** ANOVA = analysis of variance, ASIA = American Spinal Injury Association, BWSTT = body weight-supported treadmill training, DGO = driven gait orthosis, EMG = electromyography,  $H_{\max}$  = maximal H reflex amplitude,  $M_{H\max}$  = M wave amplitude at  $H_{\max}$ ,  $M_{\max}$  = maximal M wave amplitude, SCI = spinal cord injury, SD = standard deviation.

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properties of the motor neuron. To evaluate H reflex modulation during the gait cycle over different conditions or time, a methodology that addresses lower-limb joint position, loading, and phase-dependent muscle activation patterns is preferred.

The most common method described in previous investigations of timing stimulation of the soleus H reflex during gait has been the use of a foot switch marker that identifies the initial contact of the stance phase or the step cycle EMG patterns of the soleus and tibialis anterior muscles. This method involves the conduction of pseudo-random stimulations of the H reflex across the gait cycle, with post hoc division of the gait cycle into 8 to 20 phases. Custom computer algorithms assign the acquired reflex cycles to the appropriate phase of gait on the basis of timing latencies or EMG activity for signal-averaging of 8 to 10 reflex cycles at each point in the gait cycle [1,4–5,12]. Although this method has been effective, analysis of H reflex modulation across varying speeds would require recalculation of gait phases on the basis of changing latencies with changes in ambulation speed. Additionally, dividing the gait cycle into a number of phases (8 to 10) that include a range of joint positions could cause increased variance of the H reflex across the phase as a result of variations in muscle activation and joint position at the hip and ankle.

The H reflex is also a commonly used clinical tool for assessing reflex excitability after spinal cord injury (SCI) [13–17]. H reflex responses after SCI have been shown to be different than normal spinal cord physiology at rest and during stepping [1,18–20]. Body weight-supported treadmill training (BWSTT) has been increasingly applied as a clinical tool for rehabilitation of standing and walking in patients with SCI [21–25]. Recent developments in robotic devices for BWSTT have provided researchers and clinicians the unique ability to monitor joint position, torques, and subject performance with increased accuracy,

precision, and subject tolerance. The high repeatability and control of gait kinematics with robotic locomotor systems allow for improved control of many of the factors that modulate H reflex excitability. Robotic locomotor training systems should allow accurate synchronization of H reflex stimulation to defined points in the gait cycle. This methodology should provide more consistent and controlled afferent information from muscle activation, joint position, and loading than the routinely used random stimulation cycles. Robotic BWSTT may also allow better control of or reduced step-to-step variability than manually assisted BWSTT. Data collected in this manner may provide new insights into afferent and central regulation of human motor control during the natural motor task of walking, especially in subjects with SCI who are unable to step on their own and have been difficult to study during ambulation.

We conducted this study to develop and evaluate a methodology that would result in more precise and accurate stimulation of the soleus H reflex synchronously with specific points in the gait cycle during robotic BWSTT in subjects with and without SCI.

## METHODS

### Subjects

The local committees for the protection of human subjects at the Dallas Department of Veterans Affairs Medical Center and The University of Texas Southwestern Medical Center approved this investigation. A total of 26 subjects (17 male, 9 female) volunteered to participate, including 4 subjects without SCI and 22 subjects with SCI who had varying degrees of injury completeness and functional ability. **Table 1** summarizes the subject demographics. All subjects provided written consent.

**Table 1.**

Demographics of participants in H reflex methodology study. American Spinal Injury Association (ASIA) spinal cord injury (SCI) classifications of A and B are described as motor complete, ASIA C and D as motor incomplete.

Classification	Sex		No.	Age (yr) (mean ± SD)	Time Since Injury (mo) (mean ± SD)
	Male	Female			
ASIA A	7	1	8	34.4 ± 5.2	60.8 ± 40.2
ASIA B	2	2	4	35.5 ± 12.2	51.9 ± 36.6
ASIA C	2	4	6	28.2 ± 10.0	44.8 ± 39.6
ASIA D	3	1	4	44.0 ± 11.7	26.4 ± 15.2
Non-SCI	3	1	4	35.3 ± 3.5	—

SD = standard deviation.

## H Reflex Instrumentation

To record the soleus H reflex EMG, we placed subjects in a prone position and placed Ag-Cl surface electrodes (Blue Sensor, Ambu; Ballerup, Denmark) over the soleus muscle. We prepared the electrode sites by shaving the skin and mildly abrading it with prep-paper and alcohol to reduce skin impedance to less than 5 k $\Omega$ . The recording electrode was placed over the distal third of the soleus muscle just below the insertion of the gastrocnemius muscle onto the Achilles tendon in order to selectively record from the soleus. The reference electrode was placed over the Achilles tendon approximately 6 cm above the calcaneus. A reference ground electrode was placed over the fibular head.

The recording electrode signal was amplified at a fixed gain of 500 by a bioamplifier (Biopac EMG 100C, Biopac Systems Inc; Santa Barbara, California) and was bandpass-filtered between 1 Hz and 5 kHz. The signal was sent to a 16 bit analog-to-digital converter (Biopac MP 150, Biopac Systems Inc; Santa Barbara, California) and sampled at 2 KHz on a Pentium personal computer. The computer-based data acquisition system (AcqKnowledge, Biopac Systems Inc; Santa Barbara, California) collected, monitored, and stored the signal on hard disk for post hoc analysis.

The soleus H reflex was elicited by stimulation of the tibial nerve in the popliteal fossa. A 2 in.-diameter polymer anode was placed anteriorly just above the patella. We used a handheld electrode to locate the optimum site for nerve stimulation distal to the popliteal fossa. The criterion for the optimum site was the motor point that yielded the largest M wave amplitude during low-intensity stimulation. The handheld electrode was replaced with a 1 in. polymer cathode (Empi; St. Paul, Minnesota) that was placed on the skin at the optimum stimulation site. The electrodes were secured with adhesive tape so that the stimulating electrodes constantly contacted the underlying skin during all locomotor tasks. The cathode placement distal to the crease of the popliteal fossa helped avoid electrode movement relative to the nerve during the experiment and ambulation. The nerve stimulus was a 1 ms monophasic square pulse delivered by a constant current stimulator (Digitimer DS7A, Digitimer Limited; Hertfordshire, United Kingdom).

## Robotic Instrumentation

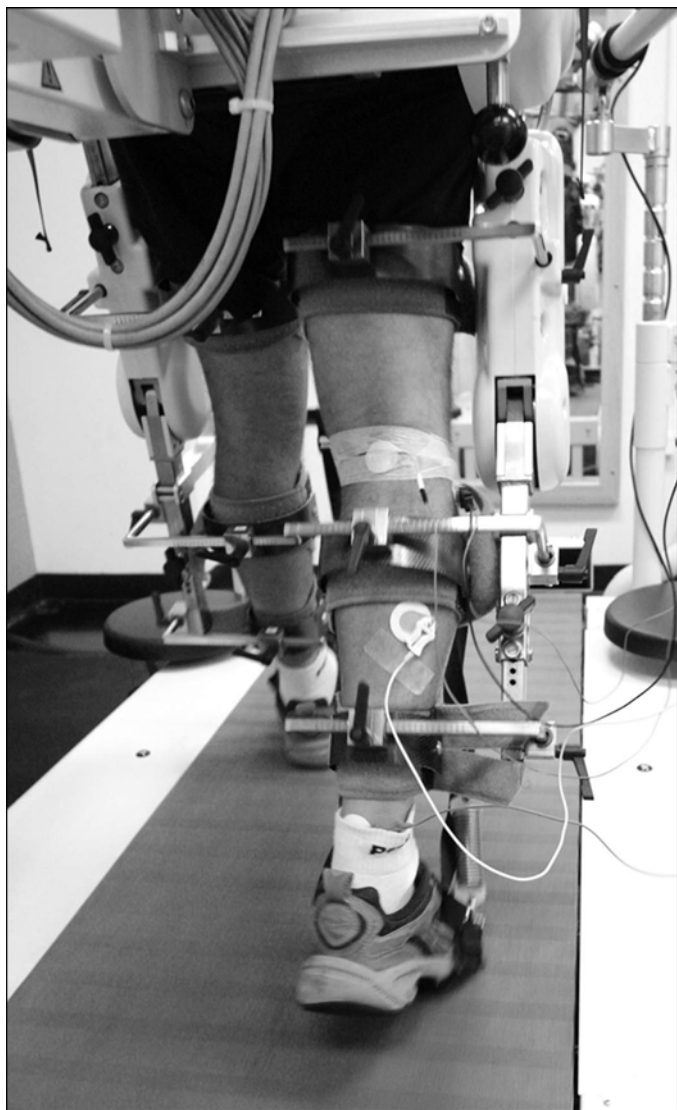
Once subjects were instrumented for H reflex acquisition, they were transferred into the Lokomat<sup>®</sup> (Hocoma

AG; Volketswil, Switzerland) robotic gait orthosis. The Lokomat driven gait orthosis (DGO) and Lokolift (Hocoma AG; Volketswil, Switzerland) dynamic unweighting system assist subjects during standing and walking. Subjects were fitted with a weight-supporting harness, and the Lokolift body weight-support system helped them stand on the treadmill. We set body-weight support at 40 percent of the subject's weight to ensure that a consistent protocol was used with the subjects from each American Spinal Injury Association (ASIA) classification and with the non-SCI subjects. This protocol included the provision of an adequately safe environment for the subjects with SCI, as well as sufficient support for them to ambulate with robotic assistance for a minimum of 30 minutes. The rigid-framed DGO was secured and aligned to the subject with cloth cuffs that attached around the thigh and shank of the lower leg. The foot and ankle were controlled by attachment of the spring-loaded straps on the lower arm of the DGO to the subject's forefoot. Pelvic straps connected the DGO to the weight-supporting harness. Although the Lokomat uses a rigid frame structure that is aligned with the subject's hip and knee and helps stabilize the pelvis, the limb is secured with cloth straps that cannot guarantee that the actual joint position will be accurately aligned with the Lokomat joint axis during ambulation. During locomotion, the subject's gait pattern was assisted by direct-drive linear actuators aligned bilaterally at the hip and knee and computer-controlled to generate a symmetrical gait pattern synchronized to the speed of the underlying treadmill (**Figure 1**). The Lokomat computer interface allowed the investigator to adjust parameters of step length, hip, and knee range of motion to approximate normal kinematics for each subject. Colombo et al. published a more detailed description of the Lokomat device [26–27].

The Lokomat DGO included a computer interface card that allowed goniometric position and direct current motor force information from the hip and knee joints to be output in real time during locomotor tasks. This information was integrated with the external data acquisition equipment to collect and monitor subjects' hip joint information within the Lokomat DGO and dynamically synchronize the H reflex stimulation and response during the gait cycle according to the defined criterion.

## H Reflex Protocol

We analyzed H reflex responses by calculating the peak-to-peak amplitude of the evoked motor response



**Figure 1.**

H reflex instrumentation with subject walking in Lokomat<sup>®</sup> (Hocoma AG; Volketswil, Switzerland). H reflex stimulation and recording electrodes are positioned before subject is placed in Lokomat.

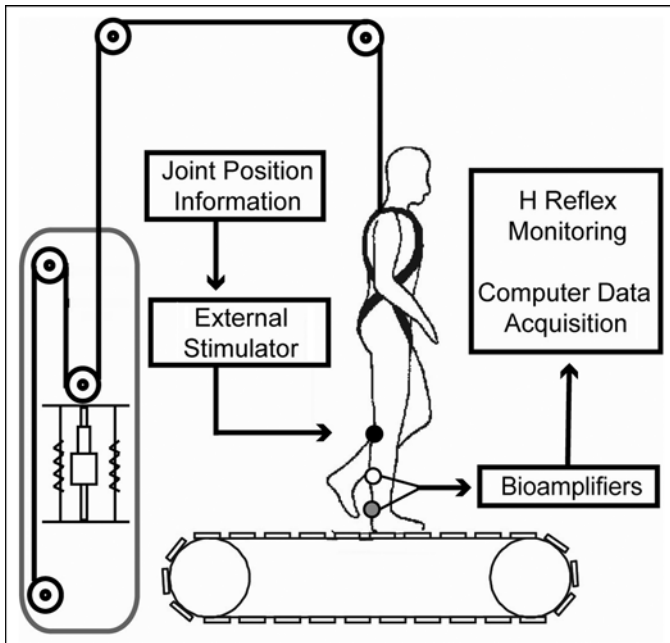
recorded from the soleus muscle. Data acquisition was triggered by each stimulation onset. Software-controlled graphical displays allowed for a 100 ms time-amplitude window representing the current H reflex stimulus-response, as well as a graphical time-amplitude display of the serial stimulus-response curves. Before testing the H reflex under synchronized robotic locomotor conditions, we recorded the H reflex and M wave recruitment characteristics with the subject in the prone position. We used a manually triggered ramping protocol to define the stimulus-response relationship. The stimulus intensity

was gradually increased from a level below the H reflex or motor (M wave) threshold to an intensity eliciting the maximal M wave amplitude ( $M_{\max}$ ). Specific identified variables were the maximal H reflex amplitude ( $H_{\max}$ ), the M wave amplitude at  $H_{\max}$  ( $M_{H_{\max}}$ ), and the  $M_{\max}$ . After placing the subject in the Lokomat, we repeated the stimulus ramping protocol with the subject in a quiet standing position with 40 percent body-weight unloading. The stimulus-response output obtained in quiet standing was then used to standardize the stimulus intensity for all locomotor tasks. The  $M_{H_{\max}}$  during quiet standing was identified as the desired independent variable to control during locomotor tasks. The M wave amplitude is the response of the  $\alpha$ -motor fibers to direct stimulation. Maintaining the same proportion of activated  $\alpha$ -motor fibers is widely assumed to consistently activate Ia afferents, allowing valid evaluation of H-reflex characteristics, particularly  $H_{\max}$  across different tasks [2,28].

### Synchronized Ambulation Protocol

Once placed in the Lokomat system, subjects remained at 40 percent body-weight support and ambulated at both 1.8 and 2.5 km/h. Hip and knee joint positions were sampled at 500 Hz from the Lokomat goniometric output (Biopac MP150). A hardware digital output channel was set so that the Lokomat hip position information triggered the external stimulator output at specifically defined points in the gait cycle for soleus H reflex acquisition (**Figure 2**).

Initially, the midstance position of the gait cycle was defined and selected as the criterion for synchronized stimulation of the H reflex. Midstance (20% into the gait cycle from initial contact) was selected as a period of single-limb support, and afferent input through the lower limb was similar to standing with the ankle in approximately 5° dorsiflexion. Midstance was defined in the software algorithm as 0° hip position from the Lokomat goniometric output. We added a midswing protocol (75% into the gait cycle from initial contact) as a second trial after confirming the stability of the midstance-synchronized acquisition protocol and collected data on 15 of the 22 subjects (5 ASIA A, 2 ASIA B, 2 ASIA C, 3 ASIA D, and 3 non-SCI). Midswing was selected as a point of limb unloading and defined in the software algorithm as the point of maximal hip flexion ( $30^\circ \pm 2^\circ$ ) recorded from the Lokomat goniometric output (**Figure 3**). The corresponding position of the knee during the defined midstance and midswing phases of the Lokomat's programmed kinematic path



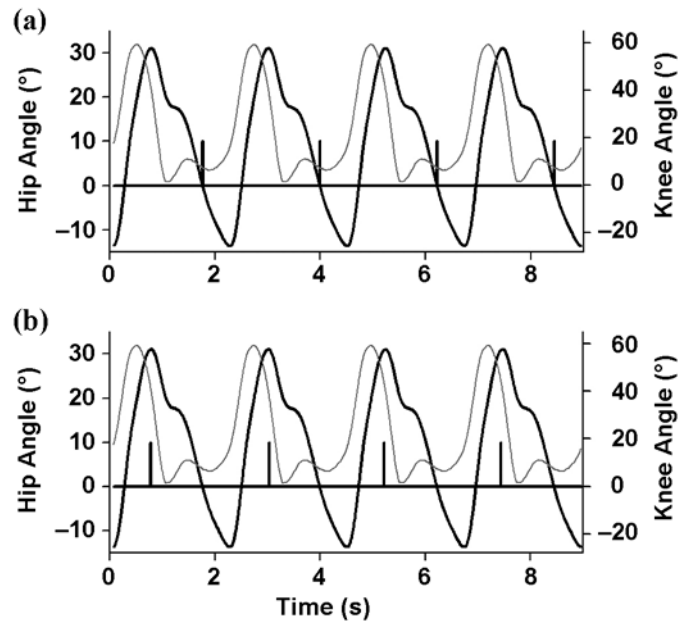
**Figure 2.**

Specific points of gait cycle are defined and used to control stimulation of soleus H reflex. Motor response is monitored for necessary adjustment of stimulus intensity.

were  $7^\circ \pm 1^\circ$  and  $43^\circ \pm 2^\circ$  of knee flexion, respectively (**Figure 3**). The software algorithm was set to control the external stimulator at these defined points. Trial 1 was H reflex stimulation synchronized to midstance at both the 1.8 and 2.5 km/h ambulation speeds. Stimulation triggering frequency was approximately 0.5 to 1 Hz, depending on speed and cadence, but user control of the stimulator output to the subject allowed for interruption of the synchronized trigger to prevent postactivation depression of the stimulus.

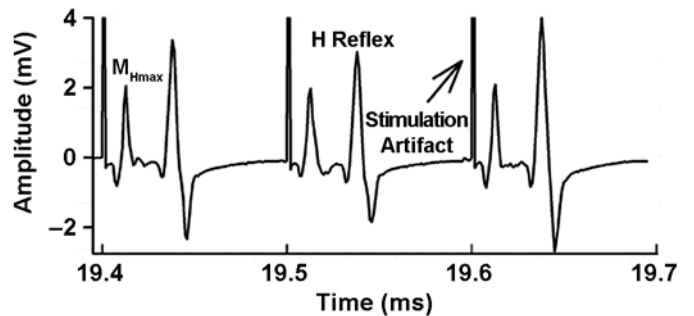
During the locomotor tasks at 1.8 and 2.5 km/h, the  $M_{Hmax}$  was monitored in real time through the software time-amplitude graphical displays. Deviations greater than  $\pm 10$  percent of the  $M_{Hmax}$  standardized in the quiet-standing condition for more than two sequential steps resulted in adjustment of the stimulus intensity to restore the appropriate  $M_{Hmax}$  value. The criterion of  $\pm 10$  percent of  $M_{Hmax}$  was initially defined as the acceptable M wave variability that would minimally affect H reflex amplitude response variability.

The series of H reflex events were recorded for 100 ms event windows triggered by the onset of the stimulation artifact (**Figure 4**). A total of 50 synchronized



**Figure 3.**

Output of Lokomat<sup>®</sup> (Hocoma AG; Volketswil, Switzerland) goniometric position of hip (black line) and knee (gray line) during ambulation. Software control of external stimulator was set to predefined points in gait cycle: (a) midstance, defined as  $0^\circ$  of hip flexion, and (b) midswing, defined as point of maximal hip flexion. Synchronized stimulator control pulse is shown.



**Figure 4.**

Graphical output of sequential H reflex stimulation cycles. H reflex data acquisition was triggered by stimulus artifact and collected for 100 ms, allowing viewing and analysis of serial H reflex cycles without acquisition of interstimulus information. Post hoc analysis defined each 100 ms H reflex event. Events that met set criterion for M wave amplitude were signal-averaged for measurement of H reflex amplitude.

H reflex cycles were collected at each walking speed to evaluate M wave variability. Ambulation speed was increased from 1.8 to 2.5 km/h without software control adjustments because stimulation control was synchronized

to real-time hip joint position with each step cycle. In addition to the steady state ambulation H reflex cycles,  $M_{\max}$  data were also acquired at each ambulation speed to confirm the stability of the maximum motor neuron stimulation amplitude.

For Trial 2, the software stimulator control window was adjusted so that the defined midswing parameters became the stimulus triggers. The synchronized stimulation protocol at the two walking speeds and at  $M_{\max}$  were repeated. Stimulation intensity was modified as needed to maintain the  $M_{H_{\max}}$  measured in quiet standing and used during the midstance trial. After the two walking trials, we remeasured skin impedance to identify any changes that may have affected electrical signal amplitudes. Post hoc analysis included stringent selection of the H reflex cycles meeting the  $\pm 10$  percent of  $M_{H_{\max}}$  criterion from the quiet standing and ambulation trials. All acceptable cycles were signal-averaged with software event selection and signal-processing algorithms (DataPac, Run Technologies; Mission Viejo, California).

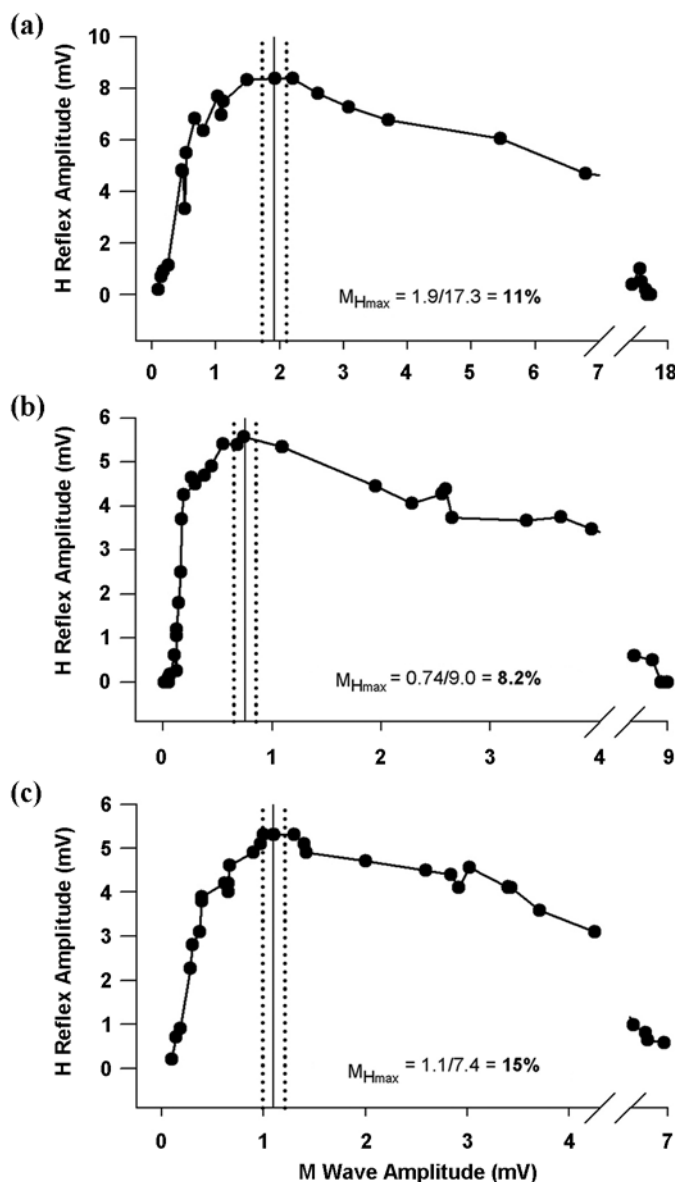
### Data Analysis

Data are presented as mean  $\pm$  standard deviation (SD) for central tendency and variance. Statistical comparisons of subjects within ASIA classifications and non-SCI subjects for standing and walking or midstance and midswing phases were analyzed with paired *t*-tests that evaluated differences in the measured variables (Excel, Microsoft Corporation; Redmond, Washington). Two-way analysis of variance (ANOVA) with Bonferroni analysis compared differences between subjects within ASIA classifications and non-SCI subjects for midswing and midstance variables (SPSS, SPSS Inc; Chicago, Illinois). The level of significance was set at  $p < 0.05$  for all analyses.

## RESULTS

An important construct of this methodology was to maintain the same proportion of  $\alpha$ -motor neuron activation during sequential step cycles by monitoring the  $M_{H_{\max}}$  during synchronized stimulations. The effect of the predefined threshold criterion of  $\pm 10$  percent of  $M_{H_{\max}}$  on H reflex amplitude variability was analyzed with initial data collection in motor complete, motor incomplete, and non-SCI subjects. The typical response to the increasing stimulus intensity ramp was a higher sensitivity of H wave amplitude to increasing M wave

amplitude on the ascending portion of the H-M sensitivity curve, with an attenuated sensitivity to increasing M wave amplitude at  $H_{\max}$  and across the descending portion of the curve to  $M_{\max}$  (Figure 5). This finding was



**Figure 5.** M wave to H reflex response sensitivity graphs. Data represent tibial-nerve stimulation ramping protocol from no M wave to maximal M wave amplitude ( $M_{\max}$ ). Presented are typical responses of subjects (a) without spinal cord injury (SCI), (b) with American Spinal Injury Association (ASIA) A (motor complete) SCI, and (c) with ASIA C (motor incomplete) SCI. Solid vertical line represents measured M wave that elicited maximal H reflex ( $M_{H_{\max}}$ ). Dotted vertical line represents  $\pm 10\%$  of  $M_{H_{\max}}$ .  $M_{H_{\max}}$  percentage of  $M_{\max}$  is calculated.

consistent among the subjects. Identification of  $M_{Hmax}$  and the  $\pm 10$  percent criterion lines provided evidence that maintaining this range of motor nerve activation minimally affected H wave variability within this range. This criterion was then used to adjust stimulus intensity as needed during data acquisition and for post hoc acceptance for H reflex cycles for all subjects and trials.

The  $M_{max}$  varied for each individual subject but was stable for any subject across conditions. The percentage of  $M_{max}$  where  $M_{Hmax}$  occurred also varied with each subject (**Figure 5**). In some subjects,  $H_{max}$  occurred before the onset of an M wave (0% of  $M_{max}$ ), while in other subjects,  $H_{max}$  occurred at  $>20$  percent of  $M_{max}$ . Grouped data by SCI classification for this percentage of  $M_{max}$  in standing and walking are presented in **Table 2**. Paired *t*-test statistical analysis showed no significant difference in the  $M_{Hmax}$  percentage of  $M_{max}$  between standing and walking for subjects within ASIA classifications and for non-SCI subjects ( $p > 0.05$ ).

The described methodology resulted in precise synchronicity of the soleus H reflex stimulation with the defined midstance and midswing phases of the gait cycle. The internal latency of the system from hip joint position trigger to stimulation output to the subject was measured as  $16 \pm 4$  ms. During BWSTT at varying speeds, the stimulation onset was highly accurate and repeatable. Direct measurement of 20 sequential step cycles at both 1.8 and 2.5 km/h in 10 subjects during midstance resulted in a pooled stimulation output to subjects at  $0.3^\circ \pm 0.2^\circ$  of Lokomat hip extension position. Once the stimulus intensity was adjusted during a testing condition to obtain  $M_{Hmax}$ , then intratrial adjustment within  $\pm 3$  mA of current maintained the  $M_{Hmax}$  criterion in each of the prone,

standing, and ambulation trials. During the midswing trials, the position of the knee was flexed approximately  $30^\circ$  more than during midstance. This alteration in electrode distance required an increase of 3 to 5 mA in stimulation intensity compared with the midstance level. Once established, the stimulation criterion in midswing was also maintained within  $\pm 3$  mA

M wave amplitudes during serial H reflex stimuli indicated minimal variability during a specific walking task. During post hoc analysis, 1,150 step cycles at each ambulation speed were evaluated in midstance and 350 step cycles in midswing. The  $M_{Hmax}$  variability resulted in acceptance of  $87.5 \pm 12.6$  percent and  $84.7 \pm 12.7$  percent of midstance H reflex complexes of pooled data at 1.8 and 2.5 km/h, respectively. During midswing,  $88.8 \pm 1.4$  percent and  $90.6 \pm 1.8$  percent of the acquired H reflex cycles met the  $M_{Hmax}$  criterion of pooled data at 1.8 and 2.5 km/h, respectively. Paired *t*-test results indicated no difference between H reflex acceptance rates between ambulation speeds for a given gait phase ( $p > 0.05$ ). Acceptance rates for stance and swing phases by SCI classification are presented in **Table 3**. The ANOVA between subjects within the ASIA classifications and the non-SCI subjects indicated no significant differences between H reflex cycle acceptance rates during each gait phase.

The total ambulation time required for each trial of 50 steady state cycles at each speed was only 4 to 5 minutes, even in subjects with SCI. The swing trial was conducted on a separate occasion for seven of the subjects. For the remaining eight subjects, it was completed in sequence after the stance phase trial on the same testing day and was established as the standard protocol. The

**Table 2.**

$M_{Hmax}$  percentage of  $M_{max}$ . Standing and walking data (mean  $\pm$  standard deviation) shown grouped by spinal cord injury (SCI) classification. No significant difference was found between standing and walking percentages ( $p > 0.05$ ).

Classification (No. of Subjects)	Standing	Walking
ASIA A (8)	$15.6 \pm 7.7$	$15.6 \pm 9.1$
ASIA B (4)	$8.5 \pm 9.1$	$10.0 \pm 10.1$
ASIA C (6)	$8.4 \pm 4.9$	$8.4 \pm 4.8$
ASIA D (4)	$11.5 \pm 7.8$	$11.8 \pm 9.4$
Non-SCI (4)	$14.3 \pm 4.4$	$13.9 \pm 7.2$

ASIA = American Spinal Injury Association,  $M_{Hmax}$  = M wave amplitude at maximal H reflex amplitude,  $M_{max}$  = maximal M wave amplitude.

**Table 3.**

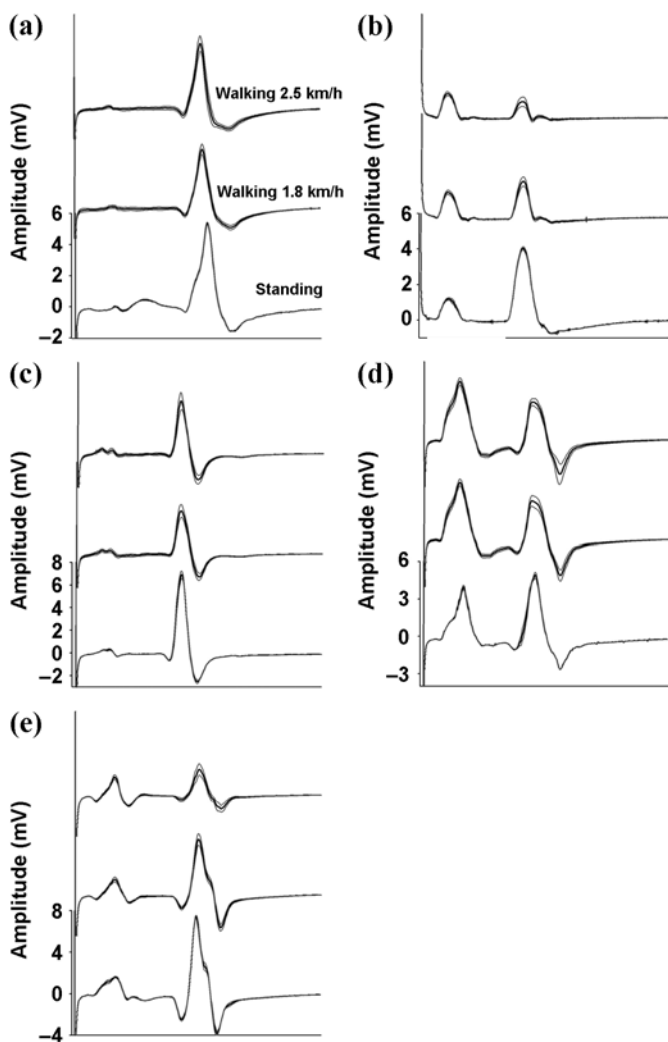
Acceptance rates (mean  $\pm$  standard deviation %) of H reflex cycles meeting  $M_{Hmax}$  criterion. Data are pooled for 1.8 and 2.5 km/h ambulation speeds ( $n = 22$  for stance, 15 for swing). No significant differences were found between spinal cord injury (SCI) classification groups for each gait phase ( $p > 0.05$ ).

Classification	Stance	Swing
ASIA A	$83.6 \pm 14.2$	$86.1 \pm 4.2$
ASIA B	$82.5 \pm 9.9$	$88.7 \pm 5.6$
ASIA C	$86.0 \pm 10.6$	$84.9 \pm 4.1$
ASIA D	$89.2 \pm 14.6$	$89.5 \pm 8.1$
Non-SCI	$88.9 \pm 12.4$	$90.1 \pm 4.2$
All Subjects	$86.1 \pm 12.6$	$88.1 \pm 4.9$

ASIA = American Spinal Injury Association,  $M_{Hmax}$  = M wave amplitude at maximal H reflex amplitude.

total ambulation time for collection of swing and stance phase data was within 30 minutes for all subjects.

Ultimately, the  $M_{H_{max}}$  criterion for monitoring stimulation intensity and post hoc H reflex cycle acceptance resulted in signal-averaged data. Examples of signal-averaged data during the midstance trial for a typical subject in each ASIA classification and for non-SCI subjects are shown in **Figure 6**. The  $M_{H_{max}}$  defined in the standing condition was maintained across ambulation at 1.8 and



**Figure 6.** Signal-averaged H reflex cycles. Presented are typical subjects (a) with American Spinal Injury Association (ASIA) A (motor complete) spinal cord injury (SCI), (b) with ASIA B (motor complete) SCI, (c) with ASIA C (motor incomplete) SCI, (d) with ASIA D (motor incomplete) SCI, and (e) without SCI during standing (3–5 cycles) and ambulation (approximately 35–40 cycles) at 1.8 and 2.5 km/h during midstance-synchronized stimulation.

2.5 km/h during the midstance and midswing trials (only midstance phase is shown). The signal-averaged graphs indicated that variability of the M wave within the  $\pm 10$  percent criterion resulted in low H wave variability during standing (three to five stimulation cycles) and increased variability in H wave amplitudes during walking. The M wave variance was controlled in all trials, thus providing evidence that the variance in H reflex amplitudes was attributable to the integration of spinal and supraspinal inputs on H reflex modulation and not to methodology limits. The variability of the differences in  $M_{H_{max}}$  in relation to  $H_{max}$  that occurs on an individual subject basis is also evident in **Figure 6**.

## DISCUSSION

This study developed and evaluated a methodology that could integrate the recent advances in robotic technology for assisted ambulation in subjects with SCI with the acquisition of the soleus H reflex. The goal was to increase the precision and accuracy of acquiring the soleus H reflex at specific points in the gait cycle during robotic BWSTT in subjects with and without SCI. The main criteria for evaluating the methodology were (1) its accuracy in stimulating the H reflex at a predefined point in the gait cycle, (2) its ability to change ambulation speed while maintaining synchronized H reflex stimulation, (3) its ability to monitor M wave amplitude in real time at the defined  $M_{H_{max}}$  in order to maintain equal stimulus intensity during ambulation and across the different protocol phases, and (4) the variability of the  $M_{H_{max}}$  during steady state ambulation speed to evaluate the number of step cycles needed to obtain sufficient H reflex cycles meeting M wave criterion while minimizing the ambulation time for subjects with SCI.

With the accuracy of the goniometric output from the Lokomat coupled to the control of the external stimulator, the soleus H reflex was acquired with less than  $1^\circ$  variability in the Lokomat hip joint position on sequential gait cycles. Although hip joint position controlled stimulation, knee joint position within the Lokomat was also consistent and therefore reduced the variability of afferent information from the knee on the H reflex. One must consider that, along with the Lokomat reporting mechanical joint positions with high accuracy, actual hip and knee joint positions are likely to be slightly different because of the motion freedom of the limb held in place by the



cloth cuffs.\* Although this difference may occur, the repeatability of the gait pattern should allow better control of stimulation guidance than was previously possible during free walking or with manual BWSTT.

The majority of the studies investigating the soleus H reflex during ambulation used random or incremental H reflex stimulation across the gait cycle and then used a post hoc division of H reflex cycles into 8 to 20 bins that fell within a specified range of the gait cycle. For calculation of the H reflex amplitude of each bin, 8 to 10 reflex cycles were averaged [1,3–6,12]. Although this method may be time efficient for acquiring H reflex cycles across the entire gait cycle, it results in division of the normalized gait cycle (100%) into bins in which the hip and knee joints would be positioned at random points covering 5 to 12 percent of the full step cycle but signal-averaged together for a single data point. Variation in the joint position of the knee, ankle, and specifically the hip, may alter afferent input from joint loading and muscle activation across a single bin. Afferent input from step to step may also vary within a given bin. Given that H reflex amplitudes have been shown to be modulated by both phase-dependent muscle activation levels during ambulation [2,5–8] and by ankle [9–10] and hip joint positions [11,29], precisely stimulating sequential H reflex cycles during the gait cycle may reduce variability in modulation factors that affect H reflex output. Robotic-controlled BWSTT provides highly repeatable gait characteristics that would be difficult to control during therapist-assisted BWSTT.

In addition to the high precision of gait-synchronized H reflex stimulation during robotic-controlled BWSTT, ambulation speed with this method could be adjusted as desired or needed without loss of stimulation precision or the need for instrumentation adjustments. This reduced the time required to complete the protocol of 50 H reflex cycles at two different ambulation speeds with body-weight support. Collection of only 10 to 12 H reflex cycles for signal-averaging at a specific gait phase, as is commonly reported, could be completed within a short time and at several points within the gait cycle within 5 minutes of ambulation. During investigations of H reflex modulation in subjects with SCI, completing the protocol in a minimal amount of time is a significant benefit because of subject tolerance and fatigue factors. However, even with the assistance of robotics, fatigue would still remain a

factor. Therefore, rapid acquisition of the required H reflex cycles would augment the ability to minimize subject fatigue and changes in motor activation. In order to approximate a synchronized stimulation to a specific point in the gait cycle, methods that use latency from a set trigger such as initial contact would require tedious measurements to adjust latency parameters. These measurements would include measurements and calculations for each change in ambulation speed and for each individual subject's cadence for any given speed. Although changes in body-weight support were not measured in this investigation, future use of this methodology should allow adjustments with confidence in simulation synchronization.

Maintaining the same proportion of activated  $\alpha$ -motor fibers, measured by the M-wave, is widely assumed to demonstrate a constant level of Ia afferent activation and allow valid evaluation of H reflex characteristics, particularly  $H_{\max}$ , across tasks [2,28]. Considerable variability exists in how previous investigations have selected the H reflex stimulation intensity criteria. Methods include selecting a constant percentage of the  $M_{\max}$  to be maintained across conditions. Investigators have used intensities ranging from 10 to 30 percent [7,30–33], multiple stimulation intensities with post hoc analysis to match M wave cycles [2,6], or have not reported. Acceptable M wave variance for these studies was between  $\pm 3$  to 5 percent of selected intensity. If a single stimulus is selected based on  $M_{\max}$  intensity across subjects, data from this investigation would suggest that the position of the stimulus intensity on the M-H recruitment curve would vary between subjects. This variability may complicate the interpretation of group results because individual data may be collected at different portions of the curve. If the selected stimulus intensity is on the ascending limb of the M-H recruitment curve, small changes in M wave amplitude may alter H reflex responses. The methodology presented here selected M wave intensity that corresponded with the individual's maximal H reflex response in quiet standing and with the acceptance tolerance of the criterion  $\pm 10$  percent. The  $H_{\max}$  portion of the recruitment curve has been shown to be an area of attenuated change in H wave amplitude with M wave change [2]. Our data supported that the criterion of  $\pm 10$  percent  $M_{H_{\max}}$  resulted in minimal H reflex variability. The chosen stimulus intensity  $M_{H_{\max}}$  in quiet standing was used across all trials. The  $M_{H_{\max}}$  was monitored in real time and required minimal or no adjustment from the initial quiet standing value across experimental conditions.

\*Joe Hidler, personal communication and unpublished data, June 2006.

Comparing H reflex acceptance rates is difficult because previous investigations have not stated the total number of H reflex cycles accepted compared with the number of cycles collected or the M wave and H wave variability. The synchronization methodology resulted in 85 percent or greater of the 50 H reflex cycles meeting the rigid post hoc criterion of  $M_{Hmax} \pm 10$  percent. H reflex acceptance rates did not differ between ambulation speeds or between subjects with or without SCI. The minimal variability of the  $M_{Hmax}$  may result partly from the consistency of joint position and afferent input previously discussed with this methodology. This high level of stability will allow us to reduce the number of H reflex cycles collected and further reduce the necessary data collection time.

We chose midstance and midswing to allow evaluation of synchronized H reflex acquisition at two different positions within the gait cycle, with very different joint and muscle activation patterns. Midstance was defined to maximally load the hip and knee and approximate a neutral ankle position, thus maximizing afferent information from the kinetic chain. This position would be similar to the loading characteristics during standing, which has been shown to have increased H reflex amplitudes [2,4], and would allow a comparison of H reflex modulation in a dynamic ambulation activity and static standing with very similar positioning. Midswing was defined at the point of maximal hip flexion. Previous studies have indicated that passive hip flexion of 20° to 30° attenuates the H reflex amplitude compared with hip extension [11,29]. Therefore, the midswing position would allow the investigation of dynamic modulation of similar hip positioning. The Lokomat uses a spring mechanism placed under the forefoot to control the ankle position for foot clearance. During the stance phase, the strap-spring mechanism that controls the ankle minimally affects the loading characteristics in the Lokomat. During swing, however, the spring force of the ankle mechanism maintains the ankle in neutral to slight dorsiflexion for safety and does not require active use of the tibialis anterior, even in subjects without SCI. For this investigation, the stability of  $M_{Hmax}$  and  $M_{max}$  was consistent in both midstance and midswing. Although actual limb position and the robotic-limb joint axis at the hip, knee, and ankle during gait will differ, the differences may not be clinically significant and robotic support provides a level of repeatability that would be difficult to impossible to control manually.

The main limitation to this methodology is that it was specifically developed with the specialized Lokomat

robotic device that offered dynamic goniometric output. Robotic devices continue to develop, and the Lokomat device is currently in 10 Department of Veterans Affairs facilities and more than 100 are in service worldwide, with the potential for increased clinical and research use in the future. All the hardware and software used with the Lokomat are commercially available and require no proprietary equipment or programming abilities. The crossover applications that would allow use of this methodology with other research measures or electronic goniometers for overground, synchronized H reflex stimulation are expected to be developed.

## CONCLUSIONS

Commercially available hardware and software can obtain a highly repeatable soleus H reflex response synchronously with any specified position in the gait cycle during robotic BWSTT in subjects with all ASIA levels of SCI. Synchronized, kinematic-controlled stimulation increased the consistency of hip and knee positions and may provide increased control of the muscle length and muscle force afferent information presented to the segmental spinal and supraspinal circuitry that generate and modulate the H reflex. Visual and software analysis of M wave amplitude during acquisition required minimal intratrial stimulus-intensity adjustment and resulted in a high percentage of acceptable reflex cycles with no differences between subjects with different ASIA SCI classifications and non-SCI subjects. Although 50 H reflex cycles were collected during ambulation, the stability of the data suggests that a reduced number of H reflex cycles could be used without affecting data quality. The growing development of robotic technologies is creating new tools for the clinical rehabilitation and scientific investigation of mechanisms of neural injury and SCI repair. This methodology may be used to investigate various research interests and provide new insights into spinal cord rehabilitation.

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## REFERENCES

1. Capaday C. Neurophysiological methods for studies of the motor system in freely moving human subjects. *J Neurosci Methods*. 1997;74(2):201–18. [PMID: 9219889]
2. Capaday C, Stein RB. Amplitude modulation of the soleus H-reflex in the human during walking and standing. *J Neurosci*. 1986;6(5):1308–13. [PMID: 3711981]
3. Lavoie BA, Devanne H, Capaday C. Differential control of reciprocal inhibition during walking versus postural and voluntary motor tasks in humans. *J Neurophysiol*. 1997;78(1):429–38. [PMID: 9242291]
4. Schneider C, Lavoie BA, Capaday C. On the origin of the soleus H-reflex modulation pattern during human walking and its task-dependent differences. *J Neurophysiol*. 2000;83(5):2881–90. [PMID: 10805685]
5. Simonsen EB, Dyhre-Poulsen P. Amplitude of the human soleus H reflex during walking and running. *J Physiol*. 1999;515(Pt 3):929–39. [PMID: 10066917]
6. Capaday C, Stein RB. Difference in the amplitude of the human soleus H reflex during walking and running. *J Physiol*. 1987;392:513–22. [PMID: 3446790]
7. Ferris DP, Aagaard P, Simonsen EB, Farley CT, Dyhre-Poulsen P. Soleus H-reflex gain in humans walking and running under simulated reduced gravity. *J Physiol*. 2001;530(Pt 1):167–80. [PMID: 11136869]
8. Sinkjaer T, Andersen JB, Larsen B. Soleus stretch reflex modulation during gait in humans. *J Neurophysiol*. 1996;76(2):1112–20. [PMID: 8871224]
9. Hwang IS. Assessment of soleus motoneuronal excitability using the joint angle dependent H reflex in humans. *J Electromyogr Kinesiol*. 2002;12(5):361–66. [PMID: 12223168]
10. Pinniger GJ, Nordlund M, Steele JR, Cresswell AG. H-reflex modulation during passive lengthening and shortening of the human triceps surae. *J Physiol*. 2001;534(Pt 3):913–23. [PMID: 11483720]
11. Knikou M, Rymer WZ. Hip angle induced modulation of H reflex amplitude, latency and duration in spinal cord injured humans. *Clin Neurophysiol*. 2002;113(11):1698–1708. [PMID: 12417222]
12. Ethier C, Imbeault MA, Ung V, Capaday C. On the soleus H-reflex modulation pattern during walking. *Exp Brain Res*. 2003;151(3):420–25. [PMID: 12827331]
13. Calancie B, Alexeeva N, Broton JG, Molano MR. Interlimb reflex activity after spinal cord injury in man: Strengthening response patterns are consistent with ongoing synaptic plasticity. *Clin Neurophysiol*. 2005;116(1):75–86. [PMID: 15589186]
14. Calancie B, Broton JG, Klose KJ, Traad M, Difini J, Ayyar DR. Evidence that alterations in presynaptic inhibition contribute to segmental hypo- and hyperexcitability after spinal cord injury in man. *Electroencephalogr Clin Neurophysiol*. 1993;89(3):177–86. [PMID: 7686850]
15. Little JW, Halar EM. H-reflex changes following spinal cord injury. *Arch Phys Med Rehabil*. 1985;66(1):19–22. [PMID: 3966862]
16. Muller R, Dietz V. Neuronal function in chronic spinal cord injury: Divergence between locomotor and flexion- and H-reflex activity. *Clin Neurophysiol*. 2006;117(7):1499–1507. [PMID: 16690351]
17. Schindler-Ivens SM, Shields RK. Soleus H-reflex recruitment is not altered in persons with chronic spinal cord injury. *Arch Phys Med Rehabil*. 2004;85(5):840–47. [PMID: 15129411]
18. Andersen JB, Sinkjaer T. The stretch reflex and H-reflex of the human soleus muscle during walking. *Motor Control*. 1999;3(2):151–57. [PMID: 10198147]
19. Faist M, Mazevet D, Dietz V, Pierrot-Deseilligny E. A quantitative assessment of presynaptic inhibition of Ia afferents in spastics. Differences in hemiplegics and paraplegics. *Brain*. 1994;117(Pt 6):1449–55. [PMID: 7820579]
20. Fung J, Barbeau H. Effects of conditioning cutaneomuscular stimulation on the soleus H-reflex in normal and spastic paretic subjects during walking and standing. *J Neurophysiol*. 1994;72(5):2090–2104. [PMID: 7884446]
21. Behrman AL, Lawless-Dixon AR, Davis SB, Bowden MG, Nair P, Phadke C, Hannold EM, Plummer P, Harkema SJ. Locomotor training progression and outcomes after incomplete spinal cord injury. *Phys Ther*. 2005;85(12):1356–71. [PMID: 16305274]
22. Dietz V, Harkema SJ. Locomotor activity in spinal cord-injured persons. *J Appl Physiol*. 2004;96(5):1954–60. [PMID: 15075315]
23. Protas EJ, Holmes SA, Qureshy H, Johnson A, Lee D, Sherwood AM. Supported treadmill ambulation training after spinal cord injury: A pilot study. *Arch Phys Med Rehabil*. 2001;82(6):825–31. [PMID: 11387590]
24. Wernig A, Muller S, Nanassy A, Cagol E. Laufband therapy based on “rules of spinal locomotion” is effective in spinal cord injured persons. *Eur J Neurosci*. 1995;7(4):823–29. [PMID: 7620630]

25. Winchester P, McColl R, Query R, Foreman N, Mosby J, Tansey K, Williamson J. Changes in supraspinal activation patterns following robotic locomotor therapy in motor-incomplete spinal cord injury. *Neurorehabil Neural Repair*. 2005;19(4):313–24. [[PMID: 16263963](#)]
26. Colombo G, Joerg M, Schreier R, Dietz V. Treadmill training of paraplegic patients using a robotic orthosis. *J Rehabil Res Dev*. 2000;37(6):693–700. [[PMID: 11321005](#)]
27. Colombo G, Wirz M, Dietz V. Driven gait orthosis for improvement of locomotor training in paraplegic patients. *Spinal Cord*. 2001;39(5):252–55. [[PMID: 11438840](#)]
28. Pierrot-Deseilligny E, Mazevet D. The monosynaptic reflex: A tool to investigate motor control in humans. Interest and limits. *Neurophysiol Clin*. 2000;30(2):67–80. [[PMID: 10812576](#)]
29. Knikou M, Rymer Z. Effects of changes in hip joint angle on H-reflex excitability in humans. *Exp Brain Res*. 2002;143(2):149–59. [[PMID: 11880891](#)]
30. Knikou M, Conway BA. Reflex effects of induced muscle contraction in normal and spinal cord injured subjects. *Muscle Nerve*. 2002;26(3):374–82. [[PMID: 12210367](#)]
31. Miyoshi T, Hotta K, Yamamoto S, Nakazawa K, Akai M. Somatosensory graviception inhibits soleus H-reflex gain in humans during walking. *Exp Brain Res*. 2006;169(1):135–38. [[PMID: 16365752](#)]
32. Phadke CP, Wu SS, Thompson FJ, Behrman AL. Soleus H-reflex modulation in response to change in percentage of leg loading in standing after incomplete spinal cord injury. *Neurosci Lett*. 2006;403(1–2):6–10. [[PMID: 16723187](#)]
33. Field-Fote EC, Brown KM, Lindley SD. Influence of posture and stimulus parameters on post-activation depression of the soleus H-reflex in individuals with chronic spinal cord injury. *Neurosci Lett*. 2006;410(1):37–41. [[PMID: 17046161](#)]

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