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REPETITIVE HINDLIMB MOVEMENT USING INTERMITTENT ADAPTIVE NEUROMUSCULAR ELECTRICAL STIMULATION IN AN INCOMPLETE SPINAL CORD INJURY RODENT MODEL

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Abstract

The long-term objective of this work is to understand the mechanisms by which electrical stimulation based movement therapies may harness neural plasticity to accelerate and enhance sensorimotor recovery after incomplete spinal cord injury (iSCI). An adaptive neuromuscular electrical stimulation (aNMES) paradigm was implemented in adult Long Evans rats with thoracic contusion injury (T8 vertebral level, 155 ± 2 Kdynes). In lengthy sessions with lightly anesthetized animals, hip flexor and extensor muscles were stimulated using an aNMES control system in order to generate desired hip movements. The aNMES control system, which used a pattern generator/pattern shaper structure, adjusted pulse amplitude to modulate muscle force in order to control hip movement. An intermittent stimulation paradigm was used (5-cycles/set; 20-second rest between sets; 100 sets). In each cycle, hip rotation caused the foot plantar surface to contact a stationary brush for appropriately timed cutaneous input. Sessions were repeated over several days while the animals recovered from injury. Results indicated that aNMES automatically and reliably tracked the desired hip trajectory with low error and maintained range of motion with only gradual increase in stimulation during the long sessions. Intermittent aNMES thus accounted for the numerous factors that can influence the response to NMES: electrode stability, excitability of spinal neural circuitry, non-linear muscle recruitment, fatigue, spinal reflexes due to cutaneous input, and the endogenous recovery of the animals. This novel aNMES application in the iSCI rodent model can thus be used in chronic stimulation studies to investigate the mechanisms of neuroplasticity targeted by NMES-based repetitive movement therapy.

Keywords

Spinal cord injury; Plasticity; Rehabilitation; Neuromuscular electrical stimulation; Rat; Adaptive control; Spinal contusion

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Introduction

Traditional physical therapy after spinal cord injury (SCI) often uses repetitive movement to strengthen muscles, prevent joint contractures, and retrain the nervous system to do specific tasks. In the past decade, repetitive motor retraining has also been increasingly used to promote functional recovery of sensorimotor function after SCI by targeting use-dependent plasticity of the central nervous system (Barbeau, et al., 1999, Edgerton, et al., 2008, Lynskey, et al., 2008, Thuret, et al., 2006).

Motorized bicycles and robotic assist devices like the Lokomat® have been used to provide passive exercise for repetitive movement therapy in people with SCI resulting in decreased spasticity and ameliorating the habituation of the H-reflex (Colombo, et al., 2000, Kiser, et al., 2005, Rosche, et al., 1997). Other exercise paradigms, such as therapist-assisted partial body weight supported treadmill training, have been shown to promote recovery of locomotor function in people with incomplete SCI (Behrman, et al., 2006, Dobkin, et al., 2006). Combining elicitation of the flexion withdrawal reflex using electrical stimulation with treadmill walking therapy appears to further enhance functional mobility and speed, decrease effort and improve intralimb coordination during unstimulated overground locomotion (Barbeau, et al., 2002, Field-Fote, 2001, Field-Fote and Tepavac, 2002, Postans, et al., 2004, Thrasher, et al., 2006). However, the flexion withdrawal reflex habituates and hence repetitive elicitation of the reflex can be problematic. Neuromuscular electrical stimulation (NMES) that uses low levels of current to directly activate motoneuron axons, is another approach to produce muscle contractions in an appropriate sequence. NMES has been used as a neuroprosthesis for replacing lost coordinated motor function in people with complete SCI, for example for grasping (Mangold, et al., 2005, Peckham, et al., 2001), standing and posture (Abbas and Gillette, 2001), or rhythmic leg movement (Riess and Abbas, 2000, Riess and Abbas, 2001, Stein, et al., 2002). It has also been used as therapy to provide gait training or electrical stimulation controlled cycling in people with chronic incomplete SCI. In these studies, NMES therapy resulted in improved recovery of overground locomotion, as indicated by measures such as walking speed (Donaldson, et al., 2000, Page, et al., 2007, Stein, et al., 2002, Thrasher, et al., 2006).

The mechanisms by which NMES therapy results in improved function remain poorly understood (Dobkin, 2003, Lynskey, et al., 2008, Rushton, 2003). NMES provides afferent input to the cord, both through direct electrical activation of the sensory nerve fibers as well as indirectly when the muscles contract and activate embedded sensory endings. Afferent activation, in particular its timing, plays an important role in activation of spinal neural circuitry. Neuronal activation, working through activity-dependent gene regulation and calcium signaling, plays a very important role in dendritic structural plasticity (Kalb, 1994, Wayman, et al., 2008, Wong and Ghosh, 2002). Dendrites, in turn, integrate synaptic information and dendritic morphology affects the ability of the synaptic inputs to activate the soma. In addition, electrical stimulation of the muscles results in up-regulation of factors such as brain derived neurotrophic factor that can be retrogradely transported to the motoneuron soma (Funakoshi, et al., 1995, Sagot, et al., 1998). Direct electrical activation of the nerves can also result in up-regulation of neurotrophic factors (Al-Majed, et al., 2000, Al-Majed, et al., 2004). Thus, there are several putative mechanisms that may mediate NMES-induced plasticity of the central nervous system.

Investigation of the underlying neuroanatomical and molecular mechanisms of neuroplasticity following NMES therapy after spinal cord injury may help determine the appropriate time at which therapy should be initiated post injury and the length and duration of therapy sessions. Current non-invasive diagnostic and imaging techniques do not permit such investigations in people. An appropriate animal model for NMES therapy after incomplete SCI (iSCI) would

enable conduction of studies that test specific hypotheses relating NMES therapy with alteration of synaptic strengths for spinal reflexes, changes in dendritic arborization and up-regulation of neurotrophic factors in motoneurons.

Since rodent models are being regularly used to investigate the effects of traumatic SCI and the effects of therapeutic intervention at the molecular, synaptic and cellular levels (Bouyer, 2005, Edgerton, et al., 2008, Gazula, et al., 2004, Metz, et al., 2000, Scheff, et al., 2003, Skinner, et al., 1996), we recently developed a spinal cord injury rodent model for NMES therapy. The procedures for electrode implantation were established, the stimulation parameters that yield sufficient joint torques and joint movement determined, and the functional reliability of implanted electrodes for up to 8 weeks in rats with spinal transections examined (Ichihara, et al., 2009, Jung, et al., 2009a). These procedures were also implemented in rodents with iSCI in whom some hindlimb movement is spared post injury (Jung, et al., 2009b). The endogenous sensorimotor recovery of rodents with iSCI poses challenges for maintenance and stability of implanted electrodes. Not only were we able to maintain the electrodes in rodents with iSCI but we also established the capability of reliably performing repetitive hindlimb movements over multiple days using an open loop NMES control algorithm. Daily open loop NMES therapy in rodents with iSCI promoted recovery of inter-hindlimb coordination during overground locomotion (Jung, et al., 2009b). However, the open loop control approach requires manual tuning of stimulation parameters for each muscle being stimulated to account for muscle-specific non-linear recruitment properties. Additionally, during the course of the open loop NMES, as the muscles fatigue the stimulation parameters are not automatically adjusted. Hence, a repeated movement pattern cannot be obtained over multiple cycles of activation. This deficiency of open loop NMES control can be overcome with an adaptive NMES (aNMES) system. An adaptive pattern generator/pattern shaper (PG/PS) controller design, derived from one we have used for application of adaptive NMES using surface electrodes in people with complete SCI (Riess and Abbas, 2000, Riess and Abbas, 2001), was implemented to allow the hip joint angle to follow a pre-specified trajectory. The capabilities of the aNMES system to automatically adjust stimulation of an antagonist/agonist muscle pair and track a desired hip angle trajectory were evaluated in uninjured anesthetized rodents (Kim, et al., 2009).

The objective of the current study was to implement and determine the ability to utilize the aNMES system to provide repetitive movement of hip joints in rodents with incomplete thoracic spinal cord injury with implanted electrodes. The study assessed the ability of the aNMES system to automatically and reliably track a desired hip trajectory during lengthy stimulation sessions during which cutaneous sensory input was provided and that were repeated over several days. Good performance of the aNMES controller demonstrated its ability to account for muscle fatigue during the lengthy sessions, any spinal reflexes due to the cutaneous input, alterations in recruitment properties and the endogenous recovery of the animals post injury that can influence the excitability of spinal neural circuitry.

Materials and Methods

Data were obtained during lengthy sessions of repeated movement of 5 hindlimbs from incomplete thoracic (T8 vertebral level) spinal cord contused Long Evans adult rats weighing 250-300gms. Data obtained from a lengthy open loop stimulation session and a lengthy aNMES continuous stimulation session were also analyzed from an additional uninjured (intact) rat that was part of a previous study (Kim, et al., 2009). Rats were individually housed in an AAALAC accredited university animal care facility with a 12-hour light/dark cycle, with access to food and water ad libitum. All procedures followed guidelines established by the Institutional Animal Care and Use Committee of Arizona State University.

Electrode Implantation and Surgery for Spinal Contusion Injury

In previous work, we developed techniques to implant stimulating electrodes near the motor points of hindlimb muscles (Ichihara, et al., 2009), examined the long-term stability of their performance in a paraplegic rodent model (Jung, et al., 2009a) and iSCI rodent model (Jung, et al., 2009b) and established stimulation paradigms for providing NMES using adaptive control algorithms to generate hip movement in an acute, intact, anesthetized rodent preparation (Kim, et al., 2009). The electrode implantation procedures utilized in the current study followed the established methodology. A brief overview is provided below.

Rats were anesthetized with Sodium Pentobarbital (35mg/kg i.p.) supplemented with isoflurane gas (1- 1.5%). 2.0 cc of 0.9% sodium chloride was administered every 2 hours to avoid dehydration. Toe pinch and visual monitoring of respiration were used as indicators of adequate anesthesia. Under aseptic conditions, gas-sterilized, custom, monopolar intramuscular stimulating electrodes were implanted close to the motor points of flexor and extensor muscles of the hip (iliacus (IL) and biceps femoralis anterior head (BF_h) respectively) and a ground (reference) electrode was sutured to muscles on the back. The stimulating electrode assembly was custom-made from Teflon-coated stainless steel helical coiled (for stress relief) lead wire (Cooner Wire, pn#AS632) with a 1-2 mm bare tip and a nylon suture connected near the bare tip. The lead wires of the stimulating electrodes and the reference electrode were routed subcutaneously to an incision on the back where they were connected to the leads from the head connector (Omnetics, Inc.). The twitch threshold current (minimum current required to elicit visible muscle activation) was determined using a hand-operated stimulator (bursts of 200 μ sec cathodic pulses at 1 Hz). Electrodes were considered to have acceptable placement when the twitch threshold current was between 0.5-1mA. Proper placement was obtained in each of the two hindlimbs in two rats and one hindlimb in the third rat.

During the same surgical session, the spinal cord at thoracic level T9 was exposed by dorsal laminectomy of the T8 vertebra. An incomplete spinal contusion injury was performed at the T9 spinal level using standardized force application (Infinite Horizon Impactor, 155 ± 2 Kdyne, mean $\pm$ 1SD). The injury site was rinsed with isotonic saline solution and absorbable 5-0 sutures were used to suture musculature surrounding the injury site. The outer skin incision was closed with surgical staples, which were removed 7 days post surgery. Animals were allowed to recover on heating pads and body core temperature monitored until awake. All animals received daily doses of cefazolin antibiotic (33.3 mg/kg s.c.) and buprenorphine analgesic (0.02-0.05 mg/kg s.c.) for the first 7 days post injury. Bladders were expressed twice daily for the duration of the study. Regular weight checks and urinalysis were also performed and, if necessary, supplemental NutricalTM for weight maintenance or antibiotic regimen for bladder infection treatment was utilized.

Adaptive NMES (aNMES) controller

The goal of the aNMES controller was to generate repeatable hip flexion/extension movements in the hindlimbs that mimicked patterns observed during locomotion. To provide aNMES, we used a modified version of the PG/PS controller that we had previously developed (Abbas and Chizeck, 1995), used in people with complete SCI (Riess and Abbas, 2000) and in acute experiments in intact anesthetized rodents (Kim, et al., 2009). In the current implementation, the aNMES control system had up to four pattern shapers (2 per hip joint) to provide electrical stimulation to the IL and BF_h muscles to generate hip movements. The amplitude of each pulse in the pulse sequence was iteratively adjusted automatically by the adaptive algorithm to drive the limb through a pre-specified cyclic trajectory. Details of the PG/PS control system are provided in (Kim, et al., 2009). A brief overview is provided below.

The PG/PS controller consists of two major components: the pattern generator (PG) and the pattern shaper (PS). The PG provides the timing for the movement pattern in the form of an oscillatory signal at the desired movement frequency. This signal is filtered by the PS, a single-layer neural network, to determine the specific stimulation pattern (sequence of current amplitude values) used to activate a muscle. The stimulation pattern is automatically customized by iteratively adapting weights on the neurons of the PS network to reduce the error between the desired hip angle and the measured hip angle.

The single-layer network forms a set of output basis functions, each of which is in the form of a raised cosine. The basis functions, which are outputs of neurons in the network, are time-shifted with respect to each other. The output of the PS, $z(t)$, is a weighted summation of the output of the basis functions

$$z(t) = \sum_{j=1}^m w_j(t) y_j(t)$$

where $y_j(t)$ is the output of the basis function j at time t , m is the number of basis functions, $w_j(t)$ is the output weight for basis function j . The network output value, $z(t)$ which has an output range from 0 to 1, was scaled by the maximum stimulation intensity (stimulus pulse amplitude) to be applied for each muscle being recruited by the stimulation.

In our implementation, the number of basis functions (neurons in the PS network) was set to be equal to the number of time steps across a desired movement period at which the output weights are updated. The period of the desired cyclic movement was set to 960 ms and the controller update (adjustment of weights) occurred every 40 ms (update period T). The number of basis functions ‘ m ’ was hence set to 24 ($=960/40$) and one basis function reached its peak activity level during each time step in the movement period. The width of the individual basis function was set such that the basis function covered 3 time steps, therefore it contributed to the stimulation levels at each of those time steps.

The PS modifies the output weights, $w_j(t)$, in order to scale the contribution of each basis function to the PS output time series using an on-line learning algorithm. Given the inherent delays between muscle stimulation and movement, an error in movement pattern at a given time can be attributed to the errors in past stimulation values. Therefore the PS learning algorithm uses the error at the current time step to change the weights on basis functions that were active at prior time steps:

$$\Delta w_j(t) = \eta e(t) \sum_{k=1}^n \frac{1}{n} y_j(t - kT)$$

where $\Delta w_j(t)$ is the change in output j of the PS network, η is a constant learning rate, $e(t)$ is the error measured between the desired and measured hip angle, T is the controller update period, and n is the number of past activation values used by the algorithm. In this study, η was set at 0.002 and n was set at 7. Since this study required control of two muscles for each hindlimb (agonist and antagonist), the learning algorithm was implemented with one PS per muscle to be stimulated (i.e. 2 per joint), and the error term used for updating the weight for activating the antagonist muscle was negative of the error term for the agonist. For implementing reciprocal stimulation of the flexors and extensors of the left and right hindlimbs, four PS units functioned simultaneously.

Experimental Protocol and Data Analysis

Incomplete spinal cord injury was characterized using the Basso, Beattie and Bresnahan (BBB) open field locomotor rating scale (Basso, et al., 1996). The BBB open field locomotor assessment scale was used to score locomotor recovery on days 1-7 following injury, and weekly on days 14 and 21 post injury. Rats were allowed to walk freely over a 4ft. × 4ft. flat level area for 4 minutes, and locomotor ability was scored on a 21 point scale by 2 observers.

Evaluation of aNMES was done once a day on days 11-13, 15-18 post injury for Rat A, 8-13 dpi for Rat B and 9-11, 15 and 16 dpi for Rat C. To perform aNMES, the animals were lightly anesthetized (titrated to slightly less than 1% isoflurane) to help them relax their hindlimbs but still respond to mild toe pinch. The animals were placed on a platform such that the head, forelimbs, and midline of the torso were supported. The platform was designed to restrain excessive movement of the head and forelimbs while both unloaded hindlimbs were attached to lightweight external custom goniometers to measure the hip flexion/extension angle (Figure 1). In each movement cycle, hip rotation caused the foot plantar surface to contact a stationary brush for appropriately timed cutaneous input. The animal's head connector was engaged with a male mating cable connected to an isolated current pulse stimulator (FNS16 - CWE, Inc., Ardmore, PA).

The twitch threshold current at 40 μ sec pulse width/phase of a biphasic pulse was determined individually per electrode prior to every NMES session. An intermittent stimulation paradigm developed in our previous study was used for aNMES (Kim, et al., 2009). 100 sets of five one second movement cycles, with 20-second rest periods between sets for a total of 500 cycles and 8 minutes of stimulation "ON" time were applied. Based on stimulation pulse parameters determined previously, pulse trains of biphasic, cathodic-first, 40 μ sec/phase pulses were delivered at 75 Hz (Jung, et al., 2009a). The adaptive controller specified pulse amplitude for each pulse (maximum value allowed was 5mA) in the pulse train, with the objective of generating a movement pattern that matched a desired hip angle trajectory derived from 3D kinematic gait analyses of an intact rat walking on a treadmill at 21m/min. In order to measure hip angle, an external rotary angle sensing potentiometer (Digi-key, PVS1A103A01) assembled to function as a goniometer was used. The goniometer was designed to allow 3 degrees-of-freedom of movement at the hip and offered little resistance to rotation. The components attached onto the leg weighed approximately 1 gram and therefore did not constitute a significant load on the leg. The potentiometer was configured as a voltage divider and the output voltage was sampled at 25 Hz by a PC using a DAQ board (model PCI-MIO-16E-1, National Instrument; Austin, TX).

Data analysis

In order to evaluate the tracking performance of the controller for any given cycle, the error was calculated between the desired and the measured angles ($n=24$ points) on a cycle-by-cycle basis. The percent root-mean-squared error (%RMSE) was calculated as the root mean square error between the actual joint angle and the desired movement pattern calculated as a percent of movement range for each set of 5-cycle sets within the trial. To quantify the level of stimulation input during a given movement cycle the electric charge (coulomb, C) delivered during a movement cycle was calculated as the total summation of the product of the width and amplitude of the cathodic phases of each pulse during a cycle. A fatigue index for each day was calculated as the difference in the average charge of the 11th-20th set and the 91st-100th set normalized by the average charge of the 11th-20th set. This variable was used as an indicator of muscle fatigability since only pulse amplitude was varied during stimulation.

Results

Characterizing iSCI using a locomotor rating scale

The BBB scores at 7, 14 and 21 days post-injury (dpi) were 6, 6.5 and 7 for Rat A and 7, 9.5 and 13 for Rat C respectively while the scores were 4.5 and 6 on 7 dpi and 14 dpi for Rat B.

Continuous open loop and adaptive stimulation sessions in an intact anesthetized rat

Figures 2 A and B show the desired and measured hip angle trajectories and stimulation pulse amplitudes for the first and last ten cycles of a continuous open loop stimulation session of a single hindlimb in an intact anesthetized rat. These data indicate that the measured joint angle trajectory profile did not match the desired angle profile when an open loop stimulation paradigm is used and that the range of motion is considerably reduced by the end of the stimulation session due to muscle fatigue. In contrast, figures 2C and D show that, unlike stimulation with an open loop paradigm, aNMES adjusts the stimulation pulse amplitude such that the measured joint angle profile matches the desired profile and the range of motion can be maintained until the end of the stimulation session. However, when used in a continuous mode the automatic adjustment results in a precipitous increase in charge delivered per cycle after approximately 20 cycles due to muscle fatigue (Figure 2E).

Tracking a desired hip trajectory over a lengthy session in an iSCI rat using intermittent aNMES

Figures 3A, B and C show the desired and measured hip angle trajectories, stimulation pulse amplitudes, and error in tracking the desired trajectory for the 1st, 2nd and 100th sets of cyclic movement for one joint in a single stimulation session in Rat A. The twitch thresholds at 40µsec/phase pulse width measured prior to starting the stimulation session were 1.55 mA for the extensor and 2.3 mA for the flexor. On initiation of aNMES (Figure 3A), the hip angle was at a neutral position of zero degrees. As on-line learning progressed, the controller increased stimulation pulse amplitudes to the flexor (black) and extensor (gray) muscles. However, during the first 5 cycles, pulse amplitudes never exceeded twitch thresholds of the flexor or extensor, hence no movement was observed (top row, solid black line). This is also reflected in the time series of the error. In the 2nd set (Figure 3B), pulse amplitude to the flexor and extensor muscles increased. By the third cycle of the 2nd set, the adaptive controller had increased pulse amplitude to the extensor to a level that substantially exceeded its twitch threshold, which is reflected in the fact that movement is observed in the direction of extension and there is a low error (<40) during the extension phase. However, the pulse amplitude to the flexor was apparently still less than twitch threshold during this cycle, because the joint only returned to the neutral position, which would occur due to the passive effect of gravity. In the 100th set (Figure. 3C), the tracking performance was very good. In the first and second cycle of the 100th set the error was higher; however through online learning the controller adjusted the pulse amplitude and the timing of the stimulation pulses so that good tracking was achieved. The peak pulse amplitude to the flexor and extensor had increased to the algorithm set maximum of 5 mA but unlike an open loop controller, the adaptive PG/PS controller adjusted the pulse amplitudes along the movement cycle. The pulse amplitude profile for the flexor indicates that a lower charge was needed to adjust the measured trajectory to the desired trajectory during the flexion phase of the gait cycle.

Thus, with aNMES the joint angular excursion can be maintained very well over a lengthy stimulation session as indicated by the low error even after 8 minutes of stimulation ON time.

Tracking performance and stimulation levels applied to the flexor and extensor muscles over multiple days

Figure 4A depicts the tracking performance of the adaptive PG/PS control system during stimulation sessions conducted on seven consecutive days in Rat A. The percent RMSE for tracking the trajectory from each set of all seven individual stimulation sessions (dotted) and the mean percent RMSE across the seven stimulation sessions (solid line) are shown. During each of the seven sessions, the adaptive PG/PS system started out with a high percent RMSE ($>30\%$) in the first set then rapidly decreased to less than 10% after 3 sets and maintained good performance as indicated by the consistently low percent RMSE. Figures 4B and C show the charge per set during the stimulation sessions for the flexor and extensor muscles, respectively. The average charge over the 100 sets of 5 cycles each for all seven individual stimulation sessions (dotted) and the average charge across the seven stimulation sessions (solid) are shown. On some days the charge (stimulation levels) did not increase while on other days there was a gradual increase. No other stimulation parameters were varied during aNMES and were kept the same from day-to-day. A systematic relationship between increase in charge and day post injury was not apparent. Even though the charge applied increased over each of the stimulation sessions (maximum 7 mC), the RMSE remained low ($<10\%$). Thus, good tracking performance can be achieved in sessions repeated over several days without precipitous increase in charge.

Similar responses were obtained from Rats B and C (not illustrated). The fatigue index for all three rats varied across days for all muscles and ranged from 1.05 to -0.23 with positive indicating increase, zero indicating no change and negative fatigue index indicating a decrease of charge from the 11-20th set to the 91st-100th set. A systematic relationship to days post injury was not apparent.

Reciprocal activation of the left and right hindlimbs

Figure 5 shows the hip angle trajectories during reciprocal stimulation of the left and right hindlimb for Rat A on a single stimulation day. The figure shows the desired (dotted) and measured (solid) trajectories of the left (top) and right (bottom) hip joint during aNMES in the 1st set (panels A and D), 2nd set (panels B and E) and the 100th set (panels C and F). The right hip angle trajectory was set to be 180° (0.5 second s) out of phase with the left hip angle trajectory in order to produce a left-right alternating hip movement pattern. The adaptive controller was able to provide appropriate stimulation in order to achieve this reciprocal pattern without any pre-set manual tuning. Panel D shows that the right hip flexor was recruited in the 2nd cycle followed by the right hip extensor in the 5th cycle of the first set while the left hip flexor and left hip extensor were recruited in the 2nd cycle of the second set as shown in panel B and the left hip flexor attained the range of motion in the first cycle of the 3rd set (data not shown). This recruitment sequence reflects the twitch thresholds observed before the stimulation session began. The twitch thresholds for the right extensor and flexor were 0.9mA and 0.45mA. The left extensor and flexor had higher twitch threshold of 1.55mA and 2.3mA respectively. The higher twitch thresholds for the muscles of the left hip reflect the later recruitment of those muscles. Within a flexor/extensor pair, the muscle with the lower twitch threshold was recruited first. As compared to the open loop stimulation paradigm responses observed in a previous study (Jung, et al., 2009b), the range of motion was maintained even at 8 minutes of stimulation ON time with the intermittent aNMES stimulation. Unlike in the open loop stimulation paradigm, the measured trajectory showed a smooth transition from flexion to extension because the stimulation provided by aNMES was automatically graded to match the movement pattern for the desired trajectory that was obtained from an intact rat walking on a treadmill.

Consistency of range of motion and RMSE

Figure 6 shows the mean percent range of motion (ROM, black) and the percent RMSE (gray) $\pm$ standard deviation obtained for sets 4-100 for all three rats for stimulation sessions repeated over 6 days. The top and middle panels show these variables for reciprocal stimulation of the left and right hip joint muscles in Rats A and B, respectively. The third panel shows the same data for stimulation of the left hip muscles in Rat C. The figure shows that ROM is maintained above 75% across multiple stimulation days (total of 18 stimulation days) for all joints. On the 1st stimulation day for the right hip joint of Rat A (top row, column 2), there was a low ROM and high RMSE. This indicates that a good trajectory matching was not obtained. The RMSE for the left hindlimb of Rat B (middle row, column 1) was high for all 7 days of stimulation even though the ROM was maintained above 75% indicating that even though the range of motion was maintained, the shape of the trajectory could likely not be well maintained. On the 3rd stimulation day for the right hindlimb of the Rat B (middle row, column 2), there was a low ROM while the RMSE was $<10\%$. This indicates that most of the trajectory matched but the peak excursion did not and was confirmed by inspection of the trajectory during offline analysis. On the following days, ROM increased and RMSE decreased indicating that the problem was related to the individual session. For Rat C, the RMSE was $>10\%$ on days 1, 2 and 4 even though the ROM was maintained on the first and second day of single joint stimulation. Rat C had an average twitch threshold of 3.29 ± 1.34 for the extensor and 3.31 ± 1.31 for the flexor which resulted in high pulse amplitudes required to recruit the muscle to produce the prescribed range of motion. Overall, consistent trajectory tracking as indicated by the RMSE and range of motion were obtained over multiple days.

Discussion

This study establishes the capability of an aNMES control system to provide reciprocal NMES for multiple sessions over a number of days in rodents with iSCI. In the aNMES control system, manual tuning was not required and it was able to track a pre-defined movement pattern and allow appropriately timed cutaneous input. Intermittent aNMES thus accounted for the numerous factors that can influence the response to NMES: electrode stability, excitability of spinal neural circuitry, non-linear muscle recruitment, fatigue, spinal reflexes due to cutaneous input, and the endogenous recovery of the animals. This novel aNMES application in the iSCI rodent model can thus be used in chronic stimulation studies to investigate the mechanisms of neuroplasticity targeted by NMES-based repetitive movement therapy.

Providing multi-input, single-output control

In the present study, for the first time, the aNMES control system was implemented with four pattern shaper modules for bilateral agonist/antagonist pairs of muscles. This was also the first implementation of an adaptive controller that used chronically implanted electrodes in an iSCI rodent model. Each controller was able to perform on-line learning to accommodate the properties of the electrode-muscle interface and the muscle recruitment properties. The controller was able to, in real-time, customize the stimulation to both the agonist and antagonist muscles for control of movement of a single joint, thus successfully achieving multi-input single-output control.

Sensor anchoring and electrode stability

An adaptive or feedback control approach requires sensors to provide feedback to the controller. In this study, we used a customized goniometer system that allowed rotation along three degrees-of-freedom at the hip to avoid imposing movement constraints. One axis of rotation of the goniometer was aligned with the greater trochanter of the femur to measure hip flexion/extension. The lower link of the goniometer system was attached at the ankle. On stimulation day 1, Rat A had a low %ROM and high %RMSE for the right hindlimb (Figure

6A, right panel) because the goniometer system was loosely attached at the ankle of the right hindlimb which resulted in slippage such that full range of movement registered as only partial movement. Therefore, it is essential that the sensor is attached properly at the ankle.

The problems in trajectory matching observed in the instances described above instances were however, not due to systematic loss of electrode stability. The animals had a mild thoracic spinal cord injury and electrodes implanted during the same surgical session. By 7 days post injury, the animals were able to voluntarily produce movement of two or three joints although no stepping movement had initiated, as indicated by the low BBB score. This voluntary movement could have impacted the electrode stability. In addition, during aNMES conducted each day there was sustained repeated movement over long durations (500 cycles). However, in most instances the electrodes remained stable throughout the study and we were able to obtain reliable functionality from the electrodes. The current data thus further supports our previous studies that indicated that implanted electrodes were stable in the 7-14 day post surgery period (Jung, et al., 2009b). This ability to initiate aNMES early after injury would be critical in studies designed to evaluate the opportune time post injury at which therapy should be initiated. In people, rehabilitative training initiated soon after iSCI appears to result in improved functional outcomes (Norrie, et al., 2005, Scivoletto, et al., 2005). Other studies in rodent models have shown that reparative molecular changes take place in the endogenous nervous system in the first few weeks post injury (Bareyre and Schwab, 2003). Thus, early intervention would allow examination of the role of aNMES based movement therapy in influencing molecular changes.

Ability to counter muscle fatigue

Muscle fatigue can be a significant limiting factor when using electrical stimulation for therapeutic purposes. Muscles fatigue very quickly with electrical stimulation due to the fact that the larger more fatigable fibers have lower stimulation thresholds and higher electrical stimulation frequencies are needed in order to achieve a fused contraction with synchronous muscle fiber activation (McNeal, 1976). There are a number of methods for prolonging repetitive movements to overcome muscle fatigue, such as altering the stimulation pattern (Maladen, et al., 2007) or using strategies that modulate both pulse amplitude and frequency (Chou, et al., 2008). In the clinic, open loop strategies are often used because of easy implementation. The stimulation pulse parameters (pulse width and frequency) chosen in this study were those that yield fused muscle contractions and only one stimulation parameter, pulse amplitude, changed. The stimulation paradigm utilized the adaptive PG/PS control system to provide aNMES (8 minutes total stimulation ON time) to a small subset of the muscles involved in locomotion (hip flexor and extensor of each hindlimb) to follow a specific movement pattern obtained from the 3D kinematic analysis of an intact rat walking on a treadmill. In our previous studies on intact uninjured rats, we evaluated the performance of an open loop controller and the adaptive PG/PS controller in a continuous paradigm (100 cycles ON) as well as an intermittent paradigm (100 sets of 5 cycles ON, 20 second rest period) (Kim, et al., 2009). In the continuous open loop stimulation paradigm, the ROM decreased as the muscles fatigued quickly (Figures 2A and B). When the adaptive PG/PS controller was applied in continuous mode, the ROM was maintained but the charge per cycle increased precipitously after about 20 cycles (Figures 2C-E). In contrast, in the intermittent mode the ROM could be maintained and the charge per cycle reached a plateau and remained constant throughout the trial (not shown). Therefore, a stimulation paradigm of 100 sets of 5 cycles of stimulation followed by a 20 second rest period was selected for the present study. Although muscle fatigability can vary across days due to factors such as the physiological status of the animal on the day of the study and sensor placement it is not systematically related to the aNMES stimulation parameters. With the intermittent stimulation paradigm in this study, lengthy sessions could

be performed on most days with consistent ROM and only gradual increases in charge, indicating the ability of the stimulation paradigm to avoid substantial muscle fatigue.

The other advantage of the adaptive PG/PS control system over the open loop stimulation strategies is the ability of the control system to adjust the amplitude of stimulation current injected so that the movement follows the desired cyclic trajectory which can have a complex shape. Thus, this approach could be used to provide different patterns of joint movement.

Effects of cutaneous spinal reflexes and endogenous recovery on trajectory control

It has been suggested that cutaneous input to the plantar surface of the foot plays an important role in recovery of stepping after spinal cord injury (Muir, 1999). Keeping that in mind, we added a brush to the platform so that the plantar surface of the foot would touch the bristles during the extension phase of the cycle. As the rats started regaining sensorimotor function as reflected in the BBB, some of the reflexes returned. While a comprehensive study was not conducted, qualitative observation of the stimulation sessions indicated that under the low level of anesthesia, there were instances during the stimulation sessions when the rat flexed its hindlimbs upon contact with the bristles. This resulted in a high error for that cycle of movement. However, in these instances, the controller adjusted the stimulation parameters and a low error was restored within a few cycles (2-3 cycles).

As indicated by the changes in BBB score, the animals underwent motor recovery over the period during which controller evaluation sessions occurred. With endogenous recovery, the sensitivity and gain of the spinal reflexes could have been altered. However, changes in spinal excitability, if present, did not impair the quality of control provided by the aNMES system as evidenced by the consistency across the week of recovery.

Potential for translation to neuromotor therapy in people with iSCI

In human subjects, the adaptive PG/PS controller has been used to control isometric muscle force (Abbas and Triolo, 1997) and to provide transcutaneous electrical stimulation to activate the quadriceps muscle to generate movements of the lower leg against gravity (Riess and Abbas, 2000, Riess and Abbas, 2001). In a recent study, the adaptive PG/PS controller was extended to utilize an agonist/antagonist pair of muscles to control cyclic movements in a single hindlimb with implanted electrodes in an intact anesthetized rodent model and its ability to generate accurate and repeatable hip movements during a single session with acutely implanted electrodes was evaluated (Kim, et al., 2009). In this study, we have extended the investigation of the efficacy of PG/PS control system to an iSCI rodent model for neuromuscular stimulation therapy that uses reciprocal control of both hindlimbs. The results showed that the adaptive PG/PS control system was able to track the desired hip movement pattern by automatically adjusting stimulation delivered to hip flexor and extensor muscles of each hindlimb and also that tracking performance remained consistent for several sessions distributed over multiple days. This implementation of aNMES could be extended to provide more intense interventions by stimulating more muscles of each hindlimb, changing the movement patterns during a stimulation session or increasing the length of stimulation sessions. These results suggest that the adaptive PG/PS control system would be a very useful tool when running long term stimulation studies to investigate the therapeutic window and understanding the effects of NMES-based repetitive movement therapy on sensorimotor function and the underlying physiological mechanisms. Since, the feasibility of utilizing transcutaneous aNMES in humans has already been demonstrated, knowledge gained from the pre-clinical studies could be expeditiously translated for promoting sensorimotor recovery in people with iSCI.

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References

1. Abbas JJ, Chizeck HJ. Neural network control of functional neuromuscular stimulation systems: computer simulation studies. *IEEE Trans Biomed Eng* 1995;42:1117–1127. [PubMed: 7498916]
2. Abbas JJ, Gillette JC. Approaches to control standing posture using electrical stimulation. *IEEE Control Systems Mag* 2001;21:80–90.
3. Abbas JJ, Triolo RJ. Experimental evaluation of an adaptive feedforward controller for use in functional neuromuscular stimulation systems. *IEEE Trans Rehab Eng* 1997;5:12–22.
4. Al-Majed AA, Brushart TM, Gordon T. Electrical stimulation accelerates and increases expression of BDNF and trkB mRNA in regenerating rat femoral motoneurons. *Eur J Neurosci* 2000;12:4381–4390. [PubMed: 11122348]
5. Al-Majed AA, Tam SL, Gordon T. Electrical stimulation accelerates and enhances expression of regeneration-associated genes in regenerating rat femoral motoneurons. *Cell Mol Neurobiol* 2004;24:379–402. [PubMed: 15206821]
6. Barbeau H, Fung J, Leroux A, Ladouceur M. A review of the adaptability and recovery of locomotion after spinal cord injury. *Prog Brain Res* 2002;137:9–25. [PubMed: 12440356]
7. Barbeau H, McCrea DA, O'Donovan MJ, Rossignol S, Grill WM, Lemay MA. Tapping into spinal circuits to restore motor function. *Brain Res Brain Res Rev* 1999;30:27–51. [PubMed: 10407124]
8. Bareyre FM, Schwab ME. Inflammation, degeneration and regeneration in the injured spinal cord: insights from DNA microarrays. *Trends Neurosci* 2003;26:555–563. [PubMed: 14522149]
9. Basso DM, Beattie MS, Bresnahan JC, Anderson DK, Faden AI, Gruner JA, Holford TR, Hsu CY, Noble LJ, Nockels R, Perot PL, Salzman SK, Young W. MASCIS evaluation of open field locomotor scores: effects of experience and teamwork on reliability. Multicenter Animal Spinal Cord Injury Study. *J Neurotrauma* 1996;13:343–359. [PubMed: 8863191]
10. Behrman AL, Bowden MG, Nair PM. Neuroplasticity after spinal cord injury and training: an emerging paradigm shift in rehabilitation and walking recovery. *Phys Ther* 2006;86:1406–1425. [PubMed: 17012645]
11. Bouyer LJ. Animal models for studying potential training strategies in persons with spinal cord injury. *J Neurol Phys Ther* 2005;29:117–125. [PubMed: 16398944]
12. Chou LW, Kesar TM, Binder-Macleod SA. Using customized rate-coding and recruitment strategies to maintain forces during repetitive activation of human muscles. *Phys Ther* 2008;88:363–375. [PubMed: 18174446]
13. Colombo G, Joerg M, Schreier R, Dietz V. Treadmill training of paraplegic patients using a robotic orthosis. *J Rehabil Res Dev* 2000;37:693–700. [PubMed: 11321005]
14. Dobkin B, Apple D, Barbeau H, Basso M, Behrman A, Deforge D, Ditunno J, Dudley G, Elashoff R, Fugate L, Harkema S, Saulino M, Scott M. Weight-supported treadmill vs over-ground training for walking after acute incomplete SCI. *Neurology* 2006;66:484–493. [PubMed: 16505299]
15. Dobkin BH. Do electrically stimulated sensory inputs and movements lead to long-term plasticity and rehabilitation gains? *Curr Opin Neurol* 2003;16:685–691. [PubMed: 14624077]
16. Donaldson N, Perkins TA, Fitzwater R, Wood DE, Middleton F. FES cycling may promote recovery of leg function after incomplete spinal cord injury. *Spinal Cord* 2000;38:680–682. [PubMed: 11114775]
17. Edgerton VR, Courtine G, Gerasimenko YP, Lavrov I, Ichiyama RM, Fong AJ, Cai LL, Otoshi CK, Tillakaratne NJ, Burdick JW, Roy RR. Training locomotor networks. *Brain Res Rev* 2008;57:241–254. [PubMed: 18022244]
18. Field-Fote EC. Combined use of body weight support, functional electric stimulation, and treadmill training to improve walking ability in individuals with chronic incomplete spinal cord injury. *Arch Phys Med Rehabil* 2001;82:818–824. [PubMed: 11387589]

19. Field-Fote EC, Tepavac D. Improved intralimb coordination in people with incomplete spinal cord injury following training with body weight support and electrical stimulation. *Phys Ther* 2002;82:707–715. [PubMed: 12088467]
20. Funakoshi H, Belluardo N, Arenas E, Yamamoto Y, Casabona A, Persson H, Ibanez C. Muscle-derived neurotrophin-4 as an activity-dependent trophic signal for adult motor neurons. *Science* 1995;268:1495–1499. [PubMed: 7770776]
21. Gazula VR, Roberts M, Luzzio C, Jawad AF, Kalb RG. Effects of limb exercise after spinal cord injury on motor neuron dendrite structure. *J Comp Neurol* 2004;476:130–145. [PubMed: 15248194]
22. Ichihara K, Venkatasubramanian G, Abbas JJ, Jung R. Neuromuscular electrical stimulation of the hindlimb muscles for movement therapy in a rodent model. *Journal of Neuroscience Methods* 2009;176:213–224. [PubMed: 18848960]
23. Jung R, Belanger A, Kanchiku T, Fairchild M, Abbas JJ. Neuromuscular stimulation therapy after incomplete spinal cord injury promotes interlimb coordination during locomotion. *J. Neural Eng* 2009b;6:055010. [PubMed: 19721184]
24. Jung R, Ichihara K, Venkatasubramanian G, Abbas JJ. Chronic Neuromuscular electrical stimulation of paraletic hindlimbs in a rodent model. *Journal of Neuroscience Methods*. 2009a doi:10.1016/j.jneumeth.2009.06.043. Epub 9 July 2009.
25. Kalb RG. Regulation of motor neuron dendrite growth by NMDA receptor activation. *Development* 1994;120:3063–3071. [PubMed: 7720552]
26. Kim SJ, Fairchild MD, Iarkov Yarkov A, Abbas JJ, Jung R. Adaptive control of movement for neuromuscular stimulation-assisted therapy in a rodent model. *IEEE Trans Biomed Eng* 2009;56:452–461. [PubMed: 19272884]
27. Kiser TS, Reese NB, Maresh T, Hearn S, Yates C, Skinner RD, Pait TG, Garcia-Rill E. Use of a motorized bicycle exercise trainer to normalize frequency-dependent habituation of the H-reflex in spinal cord injury. *J Spinal Cord Med* 2005;28:241–245. [PubMed: 16048142]
28. Lynskey JV, Belanger A, Jung R. Activity-dependent plasticity in spinal cord injury. *J Rehabil Res Dev* 2008;45:229–240. [PubMed: 18566941]
29. Maladen RD, Perumal R, Wexler AS, Binder-Macleod SA. Effects of activation pattern on nonisometric human skeletal muscle performance. *J Appl Physiol* 2007;102:1985–1991. [PubMed: 17272410]
30. Mangold S, Keller T, Curt A, Dietz V. Transcutaneous functional electrical stimulation for grasping in subjects with cervical spinal cord injury. *Spinal Cord* 2005;43:1–13. [PubMed: 15289804]
31. McNeal DR. Analysis of a model for excitation of myelinated nerve. *IEEE Trans Biomed Eng* 1976;23:329–337. [PubMed: 1278925]
32. Metz GA, Curt A, van de Meent H, Klusman I, Schwab ME, Dietz V. Validation of the weight-drop contusion model in rats: a comparative study of human spinal cord injury. *J Neurotrauma* 2000;17:1–17. [PubMed: 10674754]
33. Muir GD. Locomotor plasticity after spinal injury in the chick. *J Neurotrauma* 1999;16:705–711. [PubMed: 10511243]
34. Norrie BA, Nevett-Duchcherer JM, Gorassini MA. Reduced functional recovery by delaying motor training after spinal cord injury. *J Neurophysiol* 2005;94:255–264. [PubMed: 15985696]
35. Page SJ, Levine P, Strayer J. An electric stimulation cycling protocol for gait in incomplete spinal cord injury. *Arch Phys Med Rehabil* 2007;88:798–800. [PubMed: 17532906]
36. Peckham PH, Keith MW, Kilgore KL, Grill JH, Wuolle KS, Thrope GB, Gorman P, Hobby J, Mulcahey MJ, Carroll S, Hentz VR, Wiegner A. Efficacy of an implanted neuroprosthesis for restoring hand grasp in tetraplegia: a multicenter study. *Arch Phys Med Rehabil* 2001;82:1380–1388. [PubMed: 11588741]
37. Postans NJ, Hasler JP, Granat MH, Maxwell DJ. Functional electric stimulation to augment partial weight-bearing supported treadmill training for patients with acute incomplete spinal cord injury: A pilot study. *Arch Phys Med Rehabil* 2004;85:604–610. [PubMed: 15083437]
38. Riess J, Abbas JJ. Adaptive neural network control of cyclic movements using functional neuromuscular stimulation. *IEEE Trans. Rehab. Eng* 2000;8:42–52.

39. Riess J, Abbas JJ. Adaptive control of cyclic movements as muscles fatigue using functional neuromuscular stimulation. *IEEE Trans Neural Syst Rehabil Eng* 2001;9:326–330. [PubMed: 11561670]
40. Rosche J, Paulus C, Maisch U, Kaspar A, Mauch E, Kornhuber HH. The effects of therapy on spasticity utilizing a motorized exercise-cycle. *Spinal Cord* 1997;35:176–178. [PubMed: 9076869]
41. Rushton DN. Functional electrical stimulation and rehabilitation--an hypothesis. *Med Eng Phys* 2003;25:75–78. [PubMed: 12485788]
42. Sagot Y, Rosse T, Vejsada R, Perrelet D, Kato AC. Differential effects of neurotrophic factors on motoneuron retrograde labeling in a murine model of motoneuron disease. *J Neurosci* 1998;18:1132–1141. [PubMed: 9437033]
43. Scheff SW, Rabchevsky AG, Fugaccia I, Main JA, Lumpp JE Jr. Experimental modeling of spinal cord injury: characterization of a force-defined injury device. *J Neurotrauma* 2003;20:179–193. [PubMed: 12675971]
44. Scivoletto G, Morganti B, Molinari M. Early versus delayed inpatient spinal cord injury rehabilitation: an Italian study. *Arch Phys Med Rehabil* 2005;86:512–516. [PubMed: 15759237]
45. Skinner RD, Houle JD, Reese NB, Berry CL, Garcia-Rill E. Effects of exercise and fetal spinal cord implants on the H-reflex in chronically spinalized adult rats. *Brain Res* 1996;729:127–131. [PubMed: 8874885]
46. Stein RB, Chong SL, James KB, Kido A, Bell GJ, Tubman LA, Belanger M. Electrical stimulation for therapy and mobility after spinal cord injury. *Prog Brain Res* 2002;137:27–34. [PubMed: 12440357]
47. Thrasher TA, Flett HM, Popovic MR. Gait training regimen for incomplete spinal cord injury using functional electrical stimulation. *Spinal Cord* 2006;44:357–361. [PubMed: 16249784]
48. Thuret S, Moon LD, Gage FH. Therapeutic interventions after spinal cord injury. *Nat Rev Neurosci* 2006;7:628–643. [PubMed: 16858391]
49. Wayman GA, Davare M, Ando H, Fortin D, Varlamova O, Cheng HY, Marks D, Obrietan K, Soderling TR, Goodman RH, Impey S. An activity-regulated microRNA controls dendritic plasticity by down-regulating p250GAP. *Proc Natl Acad Sci U S A* 2008;105:9093–9098. [PubMed: 18577589]
50. Wong RO, Ghosh A. Activity-dependent regulation of dendritic growth and patterning. *Nat Rev Neurosci* 2002;3:803–812. [PubMed: 12360324]

Fig.1. Schematic of experimental setup for aNMES control system

Electrical stimulation is provided to flexors and extensors of the hip muscles via implanted electrodes. An external goniometer is attached to the hindlimb to measure hip angle. The rat is positioned such that the plantar surface of the foot touches the bristles of a brush at the peak of the extension phase. The errors between the measured hip angles (θ_1 and θ_2) and the corresponding desired angles for the hip trajectory are calculated. The weights of the basis functions of four single layered neural network pattern shapers (PS), one for each muscle, are adjusted online based on these errors to control the stimulator output. The pattern generator provides the fixed desired trajectory of movement. (See text for further details)

Fig. 2. Continuous open loop and adaptive NMES sessions in an intact anesthetized rat
 A. Open loop (1-10 cycles), B. Open loop (91-100 cycles), C. Adaptive (1-10 cycles), D. Adaptive (91-100 cycles) and E. Charge delivered per cycle by the adaptive control system to the flexor muscle during the continuous stimulation session. In each panel the top, middle and bottom rows show the hip angle trajectories (desired (dotted), measured (solid)), pulse amplitude (flexor (black), extensor (gray)) respectively. (See text for further details)

Fig. 3. Hip angle trajectories, pulse amplitude and error during a stimulation session for one rat
 A. 1st set (0-5 seconds), B. 2nd set (26-31 seconds) and C. 100th set (2455-2460 seconds). In each panel the top, middle and bottom rows show the hip angle trajectories (desired (dotted), measured (solid)), pulse amplitude (flexor (black), extensor (gray)), and error respectively. Each set consists of 5 cycles of stimulation. The data illustrate that joint angular excursion can be maintained till the end of the stimulation session (8 minutes stimulation ON time). (See text for further details)

Fig. 4. Average tracking performance and average charge per set applied to the flexor and extensor muscle during a stimulation session in a single joint

A. Tracking performance of the adaptive PG/PS control system per set during a stimulation session. During each of the seven stimulation sessions, the adaptive PG/PS system started out with a high percent RMSE in the first set but rapidly decreased to less than 10 percent after 3 sets and maintained good performance (indicated by low tracking error) in each of the seven sessions. The percent RMSE from each set of all seven individual stimulation sessions (dotted) and the mean percent RMSE across the seven stimulation sessions (solid thick line) is shown. The horizontal axis shows the set number. B and C. Charge per set during the stimulation sessions for the flexor and extensor muscles respectively. The average charge over 5 cycles

for each set for all seven individual stimulation sessions (dotted) and the average charge across the seven stimulation sessions (solid) are shown. Even though the charge applied increases over each of the stimulation sessions, the RMSE remains low indicating good tracking performance. Days 1-3 correspond to 11 dpi -13 dpi and days 4-7 correspond to 15 dpi-18dpi.

Fig. 5. Hip angle trajectories during reciprocal adaptive PG/PS stimulation of the left and right hip flexor and extensor muscles

Desired (dotted) and measured (solid) joint angle trajectories for the 1st (panels A and D), 2nd (panels B and E) and the 100th (panels C and F) set of 5 cycle stimulation are shown. The data show the ability of the controller to provide stimulation such that both flexors and extensors are activated to produce the desired trajectory and maintain reciprocal adaptive control of the hip joint angles.

Fig. 6. Average percent range of motion (ROM) and percent RMSE per day (n=5 joints)
 The mean percent range of motion (black) and the percent RMSE (gray) $\pm$ standard deviation for each day for sets 4-100. The top and middle panels show the percent range of motion and percent RMSE for reciprocal joint stimulation, the third panel shows the same data for single joint stimulation. The data illustrate the ability to perform adaptive control of the joints reliably over multiple days (see text for details).