The Dominant Role of the Hip in Multijoint Reflex Responses in Human Spinal Cord Injury

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THE DOMINANT ROLE OF THE HIP IN MULTIJOINT REFLEX RESPONSES IN HUMAN SPINAL CORD INJURY

by

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ABSTRACT
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Following a spinal cord injury (SCI), people often experience exaggerated reflexes, such that mild provocations can cause prolonged and uncontrolled muscle activity throughout the entire leg. These reflexes can be problematic and are known to interfere with functional tasks, such as transferring to and from a wheelchair, and they may interfere with locomotor function by prolonging muscle activity and/or inappropriately activating muscles during attempts to walk. While these multijoint reflexes have been shown to originate from several afferent cues, hip afferent input is a particularly potent sensory signal that readily triggers multijoint reflexes. The overall objective of this dissertation was to understand the role of hip sensory cues and the potential mechanisms associated with multijoint reflex behavior in human SCI. To evaluate this, a custom-built robot was used to impose movement of the legs about the hip joint. Joint torque and muscle activity were used as quantitative measures of reflex activity in SCI subjects. The findings from this suggest that the mutability of reflexes triggered by hip-mediated sensory signals is reduced. Voluntary effort and stretch-sensitive sensory feedback impart weak signals that do not significantly alter multijoint reflex patterns. Additionally, reflex behaviors presented with a distinct temporal response that has been associated with the disregulation of voltage-dependent depolarizing persistent inward currents (PICs). These results further elucidate the underlying mechanisms associated with hyperexcitable multijoint reflexes to help guide rehabilitation techniques for controlling unwanted muscle activity and for increasing functional gains in human SCI.

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CHAPTER 1

Introduction and Background

1.1 INTRODUCTION

According to the National Spinal Cord Injury Statistical Center (NSCISC 2010), approximately 262,000 Americans are living with potentially debilitating neurological deficits that develop after spinal cord injury (SCI). Despite advances in treatment and rehabilitation after SCI, most of the SCI population is regarded as having less than functional motor skills. Among the multitude of complications that arise following SCI, a common problem that develops is spastic motor reflex behaviors. A range of stimuli, such as changes in body posture, temperature or light touch, can initiate involuntary muscle activity throughout the legs. If severe enough, these aberrant reflexes impact routine activities of daily living, such as transferring or independent dressing, with the potential danger of falling out of a wheelchair (Little et al. 1989). Moreover, these reflexes are likely to interfere with walking by prolonging or inappropriately activating muscles. Since most injuries occur before the age of 40, and the percentage of individuals diagnosed with incomplete SCI is rising, it is important to increase functional outcomes in SCI to increase quality of life.

The scope of this dissertation was to further understand how unwanted reflex behaviors interact with supraspinal and sensory feedback. This chapter discusses spastic motor behaviors in human SCI, with a focus on extensor spasms, modulation of reflexes by the central nervous system (CNS), and potential mechanisms that have been associated with involuntary reflex activity often observed in chronic SCI.

1.2 SPASTIC REFLEXES IN SPINAL CORD INJURY

1.2.1 Spasticity in Chronic SCI

Spasticity is a disabling condition that develops within a variety of neurological disorders, including SCI, and affects approximately 65-78% of SCI patients (Maynard et al. 1990). The defining characteristics of spasticity are frequently related to the definition set by Lance (1980) that states: “Spasticity is a motor disorder characterized by a velocity-dependent increase in tonic stretch reflexes (muscle tone) with exaggerated tendon jerks, resulting from hyperexcitability of the stretch reflex, as one component of the upper motor neurone syndrome”. In a clinical setting, the spastic syndrome is often characterized by exaggerated short-latency reflexes and muscle hypertonia. The enhanced reflex activity was thought to be responsible for the increased tone, which leads to the spastic movement disorder (Dietz 2002). However, since there are many features of the upper motoneuron (UMN) syndrome, there are likely to be other changes to the CNS that affect movement in SCI, which presents challenges to spasticity management during rehabilitation.

Over recent years, the use of the term spasticity has posed concern since the accepted definition does not always support clinical observations. First, several studies have reported that clinical examination in patients who were thought to have spasticity have no signs of hyperreflexia (Sinkjaer et al. 1993; Shindler-Ivens and Shields 2004). These reports of increased muscle tone were attributed to structural changes in the muscle that was related to contracture (Sinkjaer et al. 1993). Thus, a change in muscle properties, and not only hyperreflexia, has been argued to result in the spasticity seen in patients

(Gracies 2005a, b). Second, the term ‘spasticity’ is often used interchangeably with ‘Upper Motor Neuron Syndrome’ in the clinical setting (Ivanhoe and Reistetter 2004). The spastic syndrome is often clinically characterized by exaggerated reflexes and hypertonia, but various clinical signs of the UMN syndrome, such as clonus, spasms, and hyperreflexia, which do not necessarily share a common pathophysiology with one other, have often also been attributed to spasticity. Lastly, differences in spasticity in patients with different lesion sites, such as stroke or cerebral palsy, suggests that reflex mechanisms other than hyperexcitable stretch reflexes are responsible for spastic muscle behavior in SCI (Burne et al. 2005; Engsberg et al. 2000; Schmit et al. 2000). As a result, it is important to understand the nature of the spastic condition following a neurological injury in order to accurately prescribe interventions.

1.2.2 Spastic Motor Behaviors

Three distinct types of spastic reflexes can evolve after SCI: clonus, flexor spasms and extensor spasms (Benz et al. 2005) with differing mechanisms underlying each type. Clonus is a rhythmic, involuntary contraction between agonist and antagonist muscles in distal joints (Gottlieb and Agarwal 1977). After SCI or stroke, for example, increased motoneuron excitability causes instability of the stretch reflex at distal joints, such that when a stretch reflex is elicited, there is an ensuing self re-excitation of spinal motoneurons in the pathway causing rhythmic oscillations about a single joint (Hidler and Rymer 1999; Rossi et al. 2003). Flexor and extensor spasms are unique in that they generate coordinated responses across multiple joints. Flexor spasms consist of coordinated flexion of the legs across the joints and are typically triggered by tactile,

nociceptive or stretch-related afferents (Schmit et al. 2000; Shahani and Young 1971). While flexor spasms in SCI likely result from a reorganization of the common flexion reflex pathway (Deutsch et al. 2005; Shahani and Young 1971), the neural organization of extensor spasms has been less clear, although they can be readily initiated through hip-mediated sensory feedback (Schmit and Benz 2002; Steldt and Schmit 2004).

Clinical observations on individuals with SCI noted that hip sensory cues could readily trigger bilateral leg extension when patients shifted from a sitting to supine position (Macht and Kuhn 1948; Little et al. 1989). These observations were later supported by experimental findings. Controlled unilateral leg extension movements in SCI subjects result in a characteristic pattern of hip flexion, knee extension and ankle plantarflexion (Schmit and Benz 2002). Moreover, Schmit and Benz (2002) were also the first to document a specificity of extensor spasms to imposed hip movements. Quickly bringing the leg in the opposite direction, into full hip flexion, resulted in slightly different responses, constituting hip extension, knee flexion and ankle plantarflexion. Dynamic stretching of hip muscles can also coordinate muscle activity throughout the leg in SCI subjects. The reflex responses triggered by continuous sinusoidal oscillations about the hip joint yielded alternating flexion and extension muscle activity patterns that were timed with the speed of imposed movement (Steldt and Schmit 2004). Furthermore, information from hip sensory cues of the contralateral leg are integrated with multijoint reflex amplitude, such that moving the legs in an anti-phase pattern (180º out of phase), similar to locomotion, amplifies reflex output (Onushko and Schmit 2007).

In addition to hip sensory cues, multijoint reflex activity can originate from knee proprioceptors and ankle load afferents. Rapid extension of the knee joint will cause a

stereotypic pattern of hip flexion, knee extension and ankle plantarflexion (Wu et al. 2005). However, reflex amplitudes are greater if the hip is in an extended posture versus a flexed posture, suggesting that hip proprioceptors still influence the multijoint reflexes. Ankle load afferents are also a trigger of multijoint reflex activity. Rapid ankle dorsiflexion when the hip is in an extended posture produces reflex patterns that resemble those triggered from hip extension movement alone (Wu and Schmit 2006). From the aforementioned studies, extensor reflexes are likely attributed to the activation of complex, organized interneuronal networks rather than hyperexcitable stretch reflexes that appear with spasticity.

1.3 MODULATION OF REFLEX ACTIONS

1.3.1 Muscle Afferent Feedback

In normal control of movement, reflexes are modulated through sensory feedback during motor tasks to provide appropriate reactions in response to task demands. For instance, the strength of the stretch reflex in the ankle modulates over different phases of the step cycle, presumably to provide resistance to unexpected perturbations, compensation for ground irregularities, and stabilization of the limb (Capaday and Stein 1987; Kearney et al. 1999; Sinkjaer et al. 2000; see review by Zehr and Stein 1999). In humans, the soleus H-reflex excitability is larger during the stance phase than during the swing phase of walking (Capaday and Stein 1986; Crenna and Frigo 1987; Kearney et al. 1999; Yang et al. 1991). In particular, Sinkjaer et al. (2000) investigated the mechanisms that were responsible for altered soleus EMG excitability. Muscle lengthening of the

plantarflexors was abruptly prevented during the stance phase, causing an unloading of the limb that resulted in a short latency drop in soleus EMG activity. The short latency implied spinal pathways are affected and that sensory feedback from muscle afferents is responsible for modulating reflexes at the ankle during walking. Similarly, proprioceptive feedback in response to unpredictable acceleration perturbations applied while walking on a treadmill modulate ankle muscle activity through stretch reflex pathways (Dietz et al. 1987), further demonstrating the mutability of reflex actions compensating for changes in walking conditions.

Observations of reflex modulation in muscles of the hip have also been examined in human walking. For instance, tendon jerk reflexes of the hamstrings muscle were shown to have motion-dependent feedback on the phases of the gait cycle (Van de Crommert et al. 1996). Rapid stretch of the hamstrings during early stance elicits the highest stretch reflex amplitude, which is thought to aid control of deceleration of hip flexion and knee extension at the end of swing (Van de Crommert et al. 1996). Furthermore, analogous reflex modulation of the quadriceps tendon jerk reflex is observed during human treadmill walking as well (Dietz et al. 1990; Larsen et al. 2006). The quadriceps tendon reflex, elicited by a motorized tendon reflex hammer, shows decreased reflex excitability during the early stance phase of gait. Functionally, the quadriceps help to support body weight during the early portion of stance for which in the case of ground irregularities, the increased reflex gain would likely prevent yielding of the knee.

1.3.2 Hip Afferent Feedback

Stretch-sensitive feedback from hip muscles has been established as a critical proprioceptive input for locomotor circuits within the spinal cord. Hip motion has been shown to be the controlling signal in the rhythmic generation of central pattern generators (CPGs) (Andersson and Grillner 1983, see review by Kiehn 2006). CPGs are fundamental neural circuits located within the spinal cord that coordinate and generate rhythmic and complex muscle activation patterns during locomotor behaviors, such as walking (Andersson and Grillner 1983; Forrsberg et al. 1980; Grillner and Zangger 1979, for review see Kiehn 2006). CPGs in the mammalian spinal cord are distributed within the lumbar segments to control muscles of the hindlimb during locomotion (Grillner and Zangger 1979). Specifically, rostral lumbar segments are believed to behave as the 'lead oscillator' for generating rhythmic hindlimb movement and entraining CPGs of knee and ankle muscles during locomotion (Kiehn 2006). Furthermore, stretch-sensitive feedback from muscles of the hip is necessary for controlling phase transitions during walking. The transition from stance to swing occurs when the hip is extended beyond a critical angle in the step cycle (Andersson and Grillner 1983; Grillner and Rossignol 1978; Pang and Yang 2000); although limb unloading has also been shown to be essential for swing initiation (Conway et al. 1987; Duysens and Pearson 1980; Gossard et al. 1994; Pang and Yang 2000; Pearson et al. 1992). In spinalized cats stepping on a treadmill, preventing the limb from reaching hip extension terminates stepping in that limb, while extending the limb beyond a critical angle induces a flexion burst to initiate swing (Grillner and Rossignol 1978). In the decerebrate cat preparation (a preparation in which descending cortical information is removed so as not to influence motor patterns) the importance of

hip afferent feedback is further supported. Several studies have demonstrated that stretch-sensitive hip proprioceptive feedback resets or entrains alternating flexor and extensor patterns during fictive locomotion, with bursting activity from the flexor motoneuron synchronized with hip extension (Andersson and Grillner 1983; Kriellaars et al. 1994; Hiebert el al. 1996). Similar responses to those observed in decerebrate or spinal cats have also been described in human walking. For example, during supported walking in human infants, pulling the leg back into extension aids in initiating hip flexion for swing, while preventing the leg from extending delays the swing phase (Pang and Yang 2000). Together, the aforementioned studies provide strong evidence that sensory feedback from the hip shapes the pattern of muscle activity by reflex pathways to motoneurons and contributes to the activation of muscles in an on-going manner.

1.3.3 Supraspinal Influences on Spinal Excitability

In healthy individuals, spinal reflexes are modulated via brainstem pathways as well as through direct corticospinal input to inhibitory or excitatory interneurons to simplify motor control during voluntary movements (Iles and Pisini 1992). For example, Sherrington and Hering (1897) provided an early demonstration of supraspinal control over simple reflex pathways. In primates, they were able to electrically stimulate the motor cortex to cause muscle contraction of one muscle while showing simultaneous active inhibition of the antagonist muscle. Using transcranial electrical stimulation of the motor cortex in humans, Iles and Roberts (1987) demonstrated that the soleus motoneuron receives a short-latency inhibition in addition to a second, long-lasting inhibition during voluntary contraction, where the second inhibitory effect is attributed to

presynaptic mechanisms. Furthermore, Hultborn et al. (1987) were able to show supraspinal pathways have selective control over presynaptic inhibition of Ia terminals. At the onset of a voluntary contraction, the contracting muscle has decreased presynaptic inhibition, whereas increased presynaptic inhibition was seen in motoneurons of relaxed muscles, suggesting that cortical input aids with selective activation of movements by changing stretch reflex gain in accordance with the task requirements. Other studies have also demonstrated that transmission of cortical signals through the corticospinal tract is modulated during walking in humans (Capaday et al. 1999; Pertersen et al. 1998, 2001). For instance, Petersen et al. (2001) demonstrated that a weak transcranial magnetic stimulus could impede corticospinal contributions to EMG activity in leg muscles during walking in humans, which they largely attributed to inhibition of corticomotoneuronal cells. The role of descending input on spinal networks is important for proper control of voluntary movements. Lesions to the corticospinal tract can result in abnormal control of movement and/or a reorganization of reflex pathways (Bareyre et al. 2004; Brunnstrom 1970; Dietz et al. 2009; Hultborn 2003; Valero-Cabre et al. 2004).

1.4 POTENTIAL MECHANISMS OF INVOLUNTARY REFLEX ACTIVITY AND SPASMS AFTER SCI

1.4.1 Fusimotor Drive

In the early 1960s it was popular belief that increased fusimotor drive, leading to increased muscle spindle sensitivity, contributed to exaggerated spastic stretch reflexes and increased muscle tone in patients with spasticity (Rushworth 1960). It was thought

that the increased muscle tone observed in human spasticity shared the same mechanism as that seen with decerebrate rigidity from experiments done on cats (Burke 1983); however, factors such as the lack of suppression over inhibitory reflex pathways in addition to fusimotor overactivity have been shown to play a role in decerebrate rigidity (Rymer et al. 1979). Early investigation of the influence of fusimotor overactivity in spasticity has shown that there is an increase in the stretch reflex amplitude compared to its electrical analogue, the H-reflex, which suggested that muscle spindle sensitivity was not a contributing factor to the responses (Ashby and Verrier 1976). Studies using microneurography, a technique that provides direct recording from muscle Ia afferents (Hagbarth and Vallbo 1968), have shown that there is no change in muscle spindle afferents in people with spasticity, further suggesting that fusimotor drive does not heavily contribute to spastic reflexes (Hagbarth et al. 1973; Wilson et al. 1999).

1.4.2 Reciprocal Inhibition from Ia Afferents

In patients with SCI, an increase in the excitability of reflex pathways is often seen and is thought to be due to the disruption of descending control over spinal inhibitory interneurons. As mentioned previously, excitatory input from descending pathways, such as corticospinal, rubrospinal and vestibulospinal tracts can actively excite spinal inhibitory interneurons (Crone and Nielsen 1994; Hultborn et al. 1987; Yanagisawa and Tanaka 1978). The reciprocal Ia inhibitory pathway, for example, receives input from descending and segmental afferents. In healthy individuals, this pathway ensures that the antagonist muscle remains relaxed during activation of the agonist muscle at the onset of voluntary movement (Crone et al. 1987; Crone and Nielsen

1994). However, in patients with spasticity, reduced reciprocal inhibition may contribute to the development of hyperexcitable reflexes as a result of diminished descending input. Several studies have reported a reversal of this normal relationship, in which the antagonist muscle is excited giving rise to the term “reciprocal facilitation” (Crone et al 2003; Myklebust et al. 1982; Xia and Rymer 2005). Crone et al. (2003) were able to show that in SCI patients a short latency facilitation of the soleus H-reflex was present, rather than the inhibition reported in healthy controls, and further demonstrated that the increased reflex activity of the ankle positively correlated with antagonist facilitation. This suggested that reduced reciprocal Ia inhibition was a likely cause of hyperreflexia often observed in spastic patients. However, Xia and Rymer (2005) posed a different rationale for reciprocal facilitation observed in ankle muscles. They suggested that reflex facilitation was a consequence of reflex pathway reorganization due to neuronal sprouting, in which pre-existing excitatory pathways can overpower inhibitory ones, as shown in animal models (Jankowska and McCrea 1983; Lundberg et al. 1977) and implicated in reflex facilitation observed in cerebral palsy (Myklebust et al. 1982).

1.4.3 Recurrent Inhibition

Impaired modulation of recurrent inhibition in the absence of full descending control has also been investigated in spastic patients. Recurrent inhibition, mediated by Renshaw cells, provides negative feedback signals to motoneurons of the contracting muscle and facilitates, through disinhibition, motoneurons of the antagonist, thereby helping to stabilize motoneuron firing rates during muscle contraction. This spinal mechanism is also modulated through descending motor pathways including

recticulospinal and corticospinal tracts (Mazzocchio and Rossi 1997; Rossi et al. 1992). While recurrent inhibition is impaired during voluntary movements in patients with upper motoneuron disease (Katz and Pierrot-Deseilligny 1982), in traumatic SCI there have been reports of increased recurrent inhibition (Katz and Pierrot-Deseilligny 1982). However, it is generally thought that impaired modulation of recurrent inhibition does not have a role in spasticity, and its functional consequences on control of movement are unclear (Nielsen et al. 2007).

1.4.4 Multisegmental Reflexes

Involuntary, multisegmental reflexes affecting the hip, knee and ankle are also thought to result from the loss of supraspinal control. The flexion withdrawal reflex has been shown to be under strong supraspinal influence in mammals (Baldissera et al. 1981), supported by observations in which the flexor reflex is facilitated in spinalized preparations and is suppressed in the decerebrate preparation (Rothwell 1994). In human SCI, group II, III and IV muscle afferents, joint mechanoreceptors and cutaneous receptors can elicit flexion reflexes, resulting in characteristic reflex patterns of hip and ankle flexion (Schmit et al. 2000, 2002). Since supraspinal centers receive input from those afferents via ascending tracts, it is thought that the disruption of this input can lead to a disregulation of the response to peripheral stimuli (Baldissera et al. 1981). Similar to flexion reflexes, extensor reflexes can be elicited through cutaneous stimulation (Hagbarth 1960) as well as stretch-sensitive input (Schmit and Benz 2002; Wu et al. 2005). The exact mechanisms of extensor spasms are still unclear, but it is thought that

extensor reflexes, similar to flexion reflexes, are inappropriate manifestations of existing spinal pathways (Burke 1988).

1.4.5 Plateau Potentials

Motoneuron excitability is largely regulated through brainstem derived neuromodulatory input. Specifically, serotonin (5-HT) and norepinephrine (NE) strongly regulate intrinsic motoneuron excitability (Hounsgaard et al. 1988; Lee and Heckman 1999). They allow the motoneuron to create sustained depolarizations (i.e. plateau potentials) (Perrier and Hounsgaard 2003), that arise from slow activating voltage-dependent persistent inward Na^{+} and Ca^{2+} currents. Persistent inward currents (PICs) give motoneurons active membrane properties by allowing sustained motoneuron firing in the absence of direct synaptic inputs from supraspinal origins or sensory afferents, and aid in amplification of excitatory synaptic currents (see review by Heckman et al. 2008). Plateau potentials and the associated PICs are thought to be functionally important for motor control by allowing the motoneuron to steadily fire without constant descending input (Hounsgaard et al. 1988).

Injury to the spinal cord results in the immediate paralysis of the muscles innervated by the motoneurons that are located below the injury site. In the rat SCI model, disruption of 5-HT and NE neuromodulators derived from supraspinal centers contributes to the loss in motoneuron excitability, along with the loss of PICs, thereby decreasing motoneuron excitability so greatly such that minimal reflex activity is seen in response to strong inputs (Bennett et al. 1999, 2004; Miller et al. 1996). However, despite the continued absence of 5-HT and NE months post-injury, PICs redevelop and reacquire

amplitudes as large as or larger than those observed previous to spinal injury (Bennett et al. 2001, 2004). Moreover, the depolarizing actions of PICs have been shown to be difficult to terminate after injury due to alterations in inhibitory input (Boulenguez et al. 2010), leading to excessive motoneuron firing. For example, in the chronic spinalized rat, brief, innocuous afferent stimulation will produce vigorous responses throughout the tail muscles that last several seconds after removal of the stimulus (Bennett et al. 1999, 2001, 2004). This timely reemergence of PICs with involuntary spasms in chronic SCI suggests that PICs may contribute to the long-lasting reflex responses observed in chronic SCI.

Similar long-lasting reflex behaviors occur in human SCI, further implicating the idea that PICs play a central part in spasms. In human SCI, the onset of involuntary muscles spasms often takes weeks to develop, paralleling the time course of hyperreflexia and simultaneous reemergence of PICs shown in animal SCI models (Bennett et al. 2001). PIC-like properties have also been observed in muscle spasms in human SCI. For example, spontaneous and self-sustained firing of motoneurons during muscle spasms continues for seconds to minutes after the stimulus in humans with chronic SCI (> 8 months) (Gorassini et al. 2004). Studies done by Hornby et al. (2003, 2006) have also provided evidence that abnormal PIC behaviors are likely to be involved with muscle spasms in SCI subjects. In addition to self-sustained motoneuron firing, PIC behavior can manifest in a phenomenon called windup, in which brief trains (< 4-6 s) of stimuli to the motoneuron progressively facilitate motoneuronal excitability (Bennett et al. 1998). This time-dependent facilitation in motoneuron output from repeated electrical stimulation (Hornby et al. 2003) or stretch-mediated (Hornby et al. 2006) input is observed in

hyperactive flexion and stretch reflexes, exhibiting a similar time constant of windup observed in spinal neurons of animals with PICs (Bennett et al. 1998).

1.5 Specific Aims

1.5.1 Aim 1: Effects of Volitional Effort on Multijoint Spastic Reflexes of the Legs during Assisted Bilateral Hip Oscillations in Human Spinal Cord Injury

The purpose of this study was to investigate whether volitional commands affect spastic reflex behaviors during single-joint 'assist' movements in clinically incomplete SCI subjects. Bilateral hip oscillations were imposed to the legs of 10 SCI subjects, while the subjects were asked to either assist the movements or remain relaxed during the imposed movements. Joint torque and electromyogram (EMG) measurements were used to compare activity patterns during the voluntary-assisted movements and reflex responses to the imposed hip oscillations. Additionally, 10 neurologically healthy subjects completed the same protocol, and their torque and EMG recordings were compared to those of the SCI subjects. We hypothesized that the timing of torque and EMG patterns during voluntary-assisted movements in SCI subjects would resemble spastic reflex activity during the passive hip motion, suggesting that spastic reflex activity determines motor coordination in SCI.

1.5.2 Aim 2: Bilateral Hip Oscillations Modulate Reflex Responses in Human Spinal Cord Injury

In this study, we investigated whether stretch-related sensory feedback from muscles at the knee or ankle joints would modulate spastic reflex activity during imposed hip oscillations. To test interactions of hip-mediated reflexes and reflexes from other joints, patellar tendon perturbations (knee sensory feedback) and vibration of the Achilles tendon (ankle sensory feedback) were applied during different phases of imposed hip oscillations (i.e. during hip flexion or hip extension). Joint torque and EMG recordings were compared to a control condition in which no sensory perturbation was applied. We hypothesized that hip sensory cues would dominate spastic reflex activity patterns, and sensory feedback from the knee and ankle would have little effect on multijoint reflex patterns.

1.5.3 Aim 3: Bilateral Oscillatory Hip Movements Induce Windup of Multijoint Lower Extremity Spastic Reflexes in Chronic Spinal Cord Injury

In the last experiment, we sought to identify a potential underlying mechanism associated with prolonged, involuntary muscle spasms. We investigated whether windup behavior would be observed in multijoint reflexes during imposed hip oscillations. We hypothesized that a non-linear increase in joint torque and EMG amplitude would increase during successive hip oscillations, implicating a role for persistent inward currents in spasms.

CHAPTER 2

Effects of Volitional Effort on Multijoint Spastic Reflexes of the Legs during Assisted Bilateral Hip Oscillations in Human Spinal Cord Injury

2.1 INTRODUCTION

Following spinal cord injury (SCI), people often experience exaggerated reflexes such that mild sensory provocations can cause prolonged and uncontrolled responses in muscles deficient of voluntary control. These reflexes are associated with spastic motor behaviors, which include clonus, flexor spasms, and extensor spasms, and these behaviors are common complications that evolve in chronic SCI (Benz et al. 2005). Particular to SCI, extensor spasms occur more frequently than other spastic motor behaviors, with prevalence in 82% of individuals with SCI (Little et al. 1989). Additionally, extensor spasms are readily initiated through hip afferent input, such as shifting from a sitting to supine position (Little et al. 1989; Macht and Kuhn 1948) or by extension of the leg (Schmit and Benz 2002). These spastic reflexes can be particularly disabling by interfering with intended movement plans, such as transfers (Sköld et al. 1999).

The extent to which spasms or other hyperexcitable reflex behaviors interfere with volitional movement after SCI is largely unknown. After SCI, it has been reported that there is a reduction of spinal inhibitory mechanisms due to a weakened cortical drive that may contribute to hyperexcitable reflexes (Crone et al. 2003; Heckman 1994). In the case of incomplete spinal injuries, residual supraspinal signals likely exhibit limited influence over motor unit recruitment leading to poor motor selection and reduced neuronal activation levels during intended movements (Dimitrijevic et al. 1984; Maegele et al. 2002). Further, active ankle dorsiflexion movements can produce undesired activation of the antagonist plantarflexors, although the effects vary by the nature of the

injury and recovery (Corcos et al. 1986). In a similar manner, isokinetic movements using a motor to impose motion to a joint during 'active assist' results in increased activation of antagonist muscles in people with stretch reflex hyperexcitability (Knutsson et al. 1997; Lum et al. 2004). Similar changes in SCI could contribute to the loss of specificity of volitional movements, increased co-contraction and multijoint reflex responses. Because many elements may be involved with impaired motor output in SCI, investigating how spastic reflex behaviors affect voluntary movements will aid rehabilitative approaches for enhancing functional motor coordination in human SCI.

In order to test spinal reflexes in people with SCI, single joint movements are often applied using a motorized robotic apparatus and measuring the subsequent joint torque or muscle activity. For example, imposed joint movements can produce undesired stretch reflex activation of the antagonist muscles, although the effects vary by the nature of the injury and recovery (Burke et al. 1970; Corcos et al. 1986). Single joint movements can also trigger multijoint reflex responses resembling flexor or extensor spasms in people with SCI (Schmit et al. 2000; Schmit and Benz 2002; Wu et al. 2005). Interestingly, measurements of spastic reflexes have focused primarily on circumstances in which subjects are instructed to relax, making it difficult to determine whether these hyperexcitable reflexes might be suppressed by voluntary effort. In stroke survivors, isokinetic movements using a motor to impose motion to a joint during 'active-assist' have been tested, with results indicating increased activation of antagonist muscles in people with stretch reflex hyperexcitability (Knutsson et al. 1997; Lum et al. 2004). We postulated that similar effects might occur during active-assist tasks in people with SCI. Effects could involve stretch reflexes, as observed in subjects with stroke, as well as

multijoint reflexes, such as those that underlie flexor and extensor spasms. Such impairments in reflex regulation could contribute to loss of specificity of muscle activation during volitional movements.

In the current study, we measured muscle activity and joint torque patterns during single-joint 'assist' movements in clinically incomplete SCI study participants. Ten individuals with motor incomplete SCI were asked to either assist robot-controlled, bilateral sinusoidal hip oscillations or to remain relaxed during the imposed movements. Responses from SCI subjects were compared to neurologically intact subjects completing the same tasks. We expected the timing patterns would differ between SCI and noninjured controls. Further, we hypothesized that muscle activity and joint torque patterns during 'assist' movements would resemble patterns produced under passive conditions in chronic SCI subjects. Similarities between the patterns of EMG and torque measurements during 'assist' movements and reflex responses to imposed hip movements would signify that spastic reflexes affect volitional motor coordination following SCI.

A preliminary account of this work has been previously published in abstract form (Onushko and Schmit 2008).

2.2 METHODS

2.2.1 Study Participants

Ten subjects with chronic SCI (mean age: 40.2 yrs) participated in this study. All SCI participants had motor incomplete injuries (American Spinal Cord Injury Association (ASIA) Classification C or D) with a cervical (6 subjects), thoracic (3 subjects) or lumbar (1 subject) level of lesion. At the time of this study, four of the ten subjects were taking antispastic medication (baclofen) to reduce the frequency and intensity of their spasms. The clinical features of each subject are described in Table 2-1. Additionally, ten study participants (age range: 23-54 yrs; mean age: 34 yrs) with no reported neurological damage were recruited for this study as control subjects. Exclusion criteria for this study included: motor complete injury, significant medical complications due to skin breakdown, urinary tract infection, other secondary infections, respiratory failure, heterotopic calcification, or other concurrent illnesses limiting the capacity to conform to study requirements, significant osteoporosis, or the inability to give informed consent. Informed consent was obtained prior to study participation and all procedures were conducted in accordance with the Helsinki Declaration of 1975 and approved by the Institutional Review Board of Marquette University.

Table 2-1 Subject Clinical Characteristics

Subject	Injury Level*	ASIA Score	Passive Hip Torque (Nm)†	Age (years)	Time Post Injury (yr)	Medications
S1	T2	C	8.4 / 7.5	70	3	Baclofen, Gabapentin, Ditropan, Imipramine, Sanctura, Botox
S2	T12	C	9.0 / 3.7	27	4	Oxybutinin, Metoprolol
S3	C5-6	C	61.1 / 74.6	41	25	None
S4	C5-6	C	31.4 / 55.9	40	23	None
S5	C5-6	C	33.1 / 55.2	43	17	Baclofen, Detrol, Darvocet
S6	C6	D	2.6 / 4.9	26	4	Ditropan XL
S7	C6-7	D	76.8 / 97.7	48	23	None
S8	T6-7	D	8.3 / 8.1	57	7	Baclofen, Morphine Sulfate, Gabapentin, Nefazodone, Escitalopram Oxalate
S9	L2	D	14.4 / 5.4	35	4	Triptolite, Baclofen
S10	C3	D	9.7 / 10.0	55	3	Baclofen, Gabapentin, Oxybutinin, Duloxetine HCL

* Neurological injury level: C = cervical; T = thoracic; L = lumbar

† Peak flexion torque / peak extension torque

Manual muscle testing (MMT) was used to evaluate the relative muscle strength of SCI subjects to establish general motor function under volitional effort. Scores were given between 0 (no perceivable muscle contractile activity) and 5 (limb position was maintained with full resistance through complete joint range of motion). Half scores (i.e. + or -) were given to graded muscles (Kendall 1983). All tests were completed by a licensed physical therapist prior to beginning the experiment. Subject specific information is provided in Table 2-2. During manual muscle testing, four subjects (S3, S4, S5 and S7) in particular demonstrated strong reflex responses (i.e. spasms). As a result, MMT scores ≥ 3 may reflect the subject's volitional control of muscle contraction in combination with spastic reflex activity. Additionally, subjects S8 and S10 could ambulate independently with the use of a standard walker while all other participants were independent with manual wheelchair propulsion. All subjects were independent with transfers.

Table 2-2 Manual Muscle Test Scores Left (L) and right (R) leg manual muscle test scores for each SCI subject.

	Hip Adductors		Hip Flexors		Hip Extensors		Knee Flexors		Knee Extensors		Dorsi-flexors		Plantar-flexors	
Subject	*L*	*R*	*L*	*R*	*L*	*R*	*L*	*R*	*L*	*R*	*L*	*R*	*L*	*R*
S1	0	0	0	1	0	0	0	1	0	4	0	3+	0	4
S2	2-	0	2-	0	0	0	0	0	0	0	0	0	0	0
S3*	3	3	4	4-	4	4-	4	4+	4	4+	4	3+	3	4
S4	1	1	1	2-	0	0	1	1	2	2	0	0	1	1
S5	NA	NA	1	1	NA	NA	NA	NA	1	2	1	3	1	1
S6	0	4+	0	4	NA	NA	0	4	0	5	0	4	0	5
S7*	3	4-	3-	3-	3+	3-	3-	3	4	3	3+	2-	2	2
S8†	3+	4+	3+	4-	4	4	4	4-	4+	4+	4+	4+	4	4
S9	0	4-	3+	4-	5	3+	2-	3-	3-	5	0	4	2-	4+
S10†	3-	3-	3-	3-	3	3	3+	3+	5	5	5	5	4-	4+

*From observation, subjects presented with spasticity. Manual muscle test scores (>3) may reflect spastic responses in addition to volitional effort.

†Subjects ambulated with the aid of a walker.

2.2.2 Test Apparatus

Bilateral hip isokinetic hip flexion and extension were imposed on the study participants using the apparatus pictured in Figure 2-1A. Study participants lay supine with both legs secured within custom-designed, adjustable leg braces. Since multijoint extensor reflexes are triggered by imposed hip movements (Schmit and Benz 2002), the knee and ankle joints were held isometrically within the leg braces in slightly flexed positions (approximately 22° knee flexion and 13° ankle plantarflexion) to isolate the reflex responses to hip joint rotations. The leg braces were attached to a novel robotic apparatus that used servomotor drive systems (Kollmorgen, Northampton, MA) to generate oscillatory movements of the legs about the hip joints. Torque transducers (S. Himmelstein and Company, Hoffman Estates, IL) measured sagittal plane hip, knee and

ankle torque from both legs while hip position was monitored using optical encoders (US Digital, Vancouver, WA). Custom-written LabVIEW software (National Instruments Corp., Austin, TX) was used to control hip trajectory and acquire torque and position signals. All signals were low-pass filtered (500 Hz) prior to data acquisition and sampled at 1,000 Hz using a data acquisition card (National Instruments Corp., Austin, TX) and a PC.

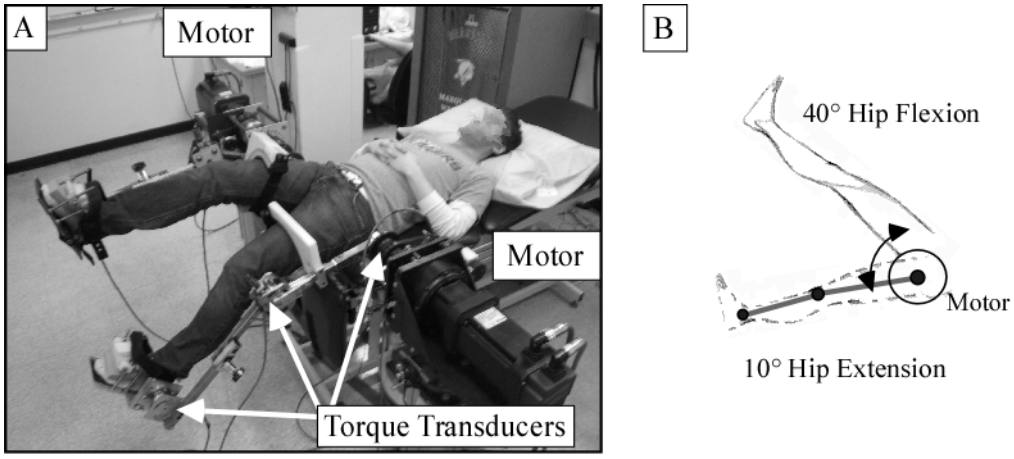


Figure 2-1 Experimental Setup. (A) Experimental apparatus used to impose bilateral hip oscillations and record sagittal plane torques during hip movements. Two servomotor systems were used to impose movements about the hips. Joint torques were recorded using torque transducers aligned with the hip, knee and ankle joints of both legs. (B) Each leg was moved through 50° of rotation about the hip (40° hip flexion to 10° hip extension).

Surface electromyograms (EMGs) were recorded bilaterally from the adductors (Add) vastus medialis (VM), vastus lateralis (VL), rectus femoris (RF), medial hamstrings (MH), medial head of the gastrocnemius (MG), soleus (Sol), and the tibialis anterior (TA) from all subjects. Disposable, pre-gelled Ag/AgCl recording electrodes (Vermed Medical Inc., Bellows Falls, VT) were applied to cleansed, lightly abraded skin over each muscle belly. EMG signals were amplified ($\times$ 1,000) and bandpass filtered (10-1000 Hz) (Bortec Medical AMT-16; Calgary, Alberta, Canada) prior to sampling.

All signals were low-pass filtered (500 Hz) and sampled at 1,000 Hz using a data acquisition card (National Instruments Corp., Austin, TX) and a personal computer. Custom LabVIEW software (National Instruments Corp.) was used for acquiring EMG, joint torque and hip angle data.

2.2.3 Experimental Protocol

With the subjects lying supine, oscillations of the legs about the hips were applied using the servomotor apparatus. The legs were moved in an alternating manner (180° out of phase) through a 50° range of motion (40° of hip flexion to 10° of hip extension) for 10 cycles. The knee and ankle joints of both legs were held isometrically within the leg braces in slightly flexed positions (knee: 22° flexion; ankle: 15°dorsiflexion). Three movement frequencies (0.25, 0.50 and 0.75 Hz) were performed to test for the effects of movement velocity. To assess the influence of supraspinal input on spastic reflex excitability, SCI study participants were asked to either assist the imposed hip motions ("assisted") or to remain inactive during the imposed hip oscillations ("unassisted"). Subjects received no verbal cueing or feedback during the trials regarding their

performance. Control subjects were asked to perform the same tests during the imposed hip oscillations. The tests were presented in random order and repeated three times for each movement frequency and level of participation using a block design (total of 18 tests). Two to five minutes were allowed between tests to prevent fatigue following assisted movements. Torque and EMG recordings were measured throughout the movements.

Concluding the experiment, two additional hip movements were performed to approximate the biomechanical properties of the leg and leg brace. Specifically, the torque due to gravity, passive joint resistance, and the mass moment of inertia were estimated (details are provided in 2.2.4 Data Analysis section). Slow (2º/sec), incremental (5º steps) movements were imposed about the hip to estimate the torque contribution due to gravity and passive joint resistance. The combined inertial constant of the leg and leg brace was determined for each leg by rapid (1.5 Hz) hip oscillations (10 cycles) imposed to each leg, moving the leg through a 15º hip range of motion (25º to 10º hip flexion). Moving the leg within midrange minimized the chance of eliciting spastic reflexes during the movement. Subjects were asked to remain as relaxed as possible during these hip movements. EMG measurements were used to ensure subjects remained relaxed during the additional hip movements.

2.2.4 Data Analysis

The biomechanical properties of the leg and leg brace were determined to calculate the torque due to active muscle contraction (active torque). To account for the biomechanical properties of the leg and leg brace, the measured torque data were

corrected for the gravitational torque, torque due to passive joint resistance, inertial torque and a torque estimate of a mechanical artifact within the system. The general equation used to calculate the active torque for each joint, τ_{joint}, is given by:

$$\tau_{joint} = \tau_{measured} - \tau_{passive/gravity} - \tau_{inertia} - \tau_{artifact}$$

where $\tau_{measured}$ is the recorded torque for a specific joint, $\tau_{passive/gravity}$ is a combined torque measurement that approximates the torque produced by the passive joint resistance as well as the gravitational forces acting on the leg, $\tau_{inertia}$ is the calculated torque attributed to the inertial properties of the leg and leg brace, and $\tau_{artifact}$ is a torque estimate of the mechanical artifact within the system that was not attributed to the biomechanical properties of the leg and leg brace.

The technique used to calculate the torque due to active muscle contraction is described in detail in previous studies (Onushko and Schmit 2007; Steldt and Schmit 2004). Briefly, the $\tau_{passive/gravity}$ was estimated by fitting a polynomial curve (torque vs. hip angle) to the trials in which the leg was moved in slow incremental steps through the range of motion. Torque data from the hold periods, when no EMG was observed, were used to estimate the combined gravitational and passive joint resistance. The polynomial function was then used to estimate the $\tau_{passive/gravity}$ during the movement trials and was subtracted from the measured torque. In a similar manner, the inertial coefficient was estimated from the trials with rapid oscillations in midrange. First the $\tau_{passive/gravity}$ was subtracted. Then the hip angular acceleration was calculated and the inertial coefficient was estimated using a regression. This inertial coefficient was

then applied to the test trial acceleration data to calculate $\tau_{inertia}$, which was then subtracted from the measured torque.

The relatively small (<5 Nm), repeatable torque artifact, $\tau_{artifact}$, was consistently observed with a similar pattern across all tests in non-injured subjects when subjects were instructed to relax during each hip oscillation condition. While the exact source of this artifact was unknown, it was likely associated with flexion of the brace itself, which likely created a "whipping" effect at the end of the range of motion. The mechanical artifact was estimated using an ensemble average of the passive torque recordings from the control subjects. Since neurologically intact individuals did not produce muscle activity during the oscillations (verified by EMG), the remaining torque well-represented the artifact within the system. This torque was subtracted from the measured torque to remove the artifact. After subtraction of $\tau_{passive/gravity}$, $\tau_{inertia}$ and $\tau_{artifact}$, the remaining torque signal reflected the active joint torque produced by the subject. All torque data were low-passed filtered (5 Hz) using a 2nd order Butterworth filter (filtered forward and backward to ensure no phase delay).

EMG signals were processed to obtain the timing and level of muscle activity during the experimental procedures. All surface EMG recordings were bandstop filtered (59-61 Hz) to remove line noise and bandpass filtered (10-350 Hz) using fourth-order Butterworth filters (Matlab; The Mathworks Inc., Natick, MA). For analysis, the root-mean square (RMS) of the EMG data was calculated using a 100-ms moving window. Only the middle eight cycles were used for the analyses of EMG data, since the responses during this time consistently exhibited suprathreshold activity (threshold = mean of

baseline + 3 SD). The first and last cycles were excluded from further analysis due to an inconsistency and incompleteness in the responses.

PHASE ANALYSIS: A phase analysis was performed on the active torques to determine the overall timing of activity with respect to the hip position during the imposed hip movements. The phase analysis was also performed on the RMS EMG data. For each subject, EMG threshold values were calculated (mean of the baseline + 3 standard deviations) from EMG measurements of the respective muscle recorded during a quiescent period when the legs were held stationary with the hip in a neutral position.

The timing of the responses was quantified using the circular statistics method outlined by Batschelet (1981). This technique has been applied previously and is described in detail elsewhere (Onushko and Schmit 2007). Briefly, torque signals were separated into flexion and extension components and the cycle period of the data was normalized to 360°, where 0° represents full hip flexion and 180° represents full hip extension. An example of the vector representation for hip flexion torque data for a single cycle is shown in Figure 2-2. A normalized vector (unit vector) was identified for each of the middle eight cycles of movement of each trial and the vectors were then averaged across the three trials (total of 24 cycles) for the hip, knee and ankle of both legs. The resulting mean vector was defined by its magnitude (range: 0-1) and phase angle. Note that if the phasing of the vectors was identical for all cycles, the mean vector magnitude would be 1. If phasing was random, the mean vector magnitude would be near 0. To examine the phasing across subjects, the mean vector for each subject was again normalized to a magnitude of 1 and then the mean vector was calculated across subjects

within a group. To identify whether there was a significant difference of the mean angle across subject groups, the Watson-Williams Test was used for statistical analysis on the timing of torque generation at each joint ($\alpha = 0.05$). A Bonferroni correction was performed for multiple comparisons. The phase analysis was also performed on the EMG measurements using a similar approach.

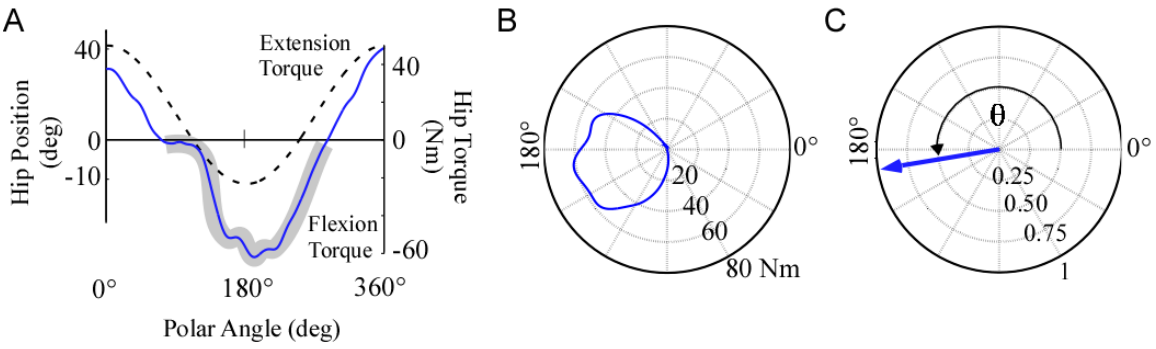


Figure 2-2 Polar Coordinate Conversion. Example of the conversion of the torque data into the vector and phase angle representation. (A) One cycle of hip position (dotted trace) and corresponding hip torque response (solid trace) plotted against the polar angle. The gray shaded background on the torque response represents hip flexion torque. (B) Hip flexion torque plotted in polar coordinates, where 180° represents the position of the hip in full extension (i.e. -10°) and 0° represents the position of the hip in full flexion (i.e. 40°). (C) A unit vector (r=1) was used to represent the normalized magnitude of the peak torque, and the polar angle, θ, represents the timing of the peak torque generation with respect to the position of the hip for one movement cycle (i.e. 0-360°). Hip extension torque was calculated in a similar manner (not shown).

AMPLITUDE ANALYSIS: The peak flexion and extension torques for the hip, knee and ankle were determined to identify whether volitional effort and movement frequency affected the amplitude of the responses. Hip, knee and ankle torque signals were separated into flexion and extension components and the peak torque was identified for each cycle of movement. The mean peak torque for hip, knee and ankle flexion and

extension was calculated for each test across all three trials, discarding data from the first and last movement cycles.

Since torque responses cannot account for coactivation of muscles at the joints, surface EMG recordings of individual muscles were used for further interpretation of the responses. Muscle activation was quantified by calculating the mean integrated area for each EMG signal for every cycle for which the muscle was active, across all three trials (total of 24 cycles). To account for differences between the three movement frequencies, the mean area was normalized to the cycle time of its respective movement frequency. The data for each muscle was then normalized to the mean of each subject's entire EMG data based on both assisted and unassisted tests.

2.2.5 Statistical Analysis

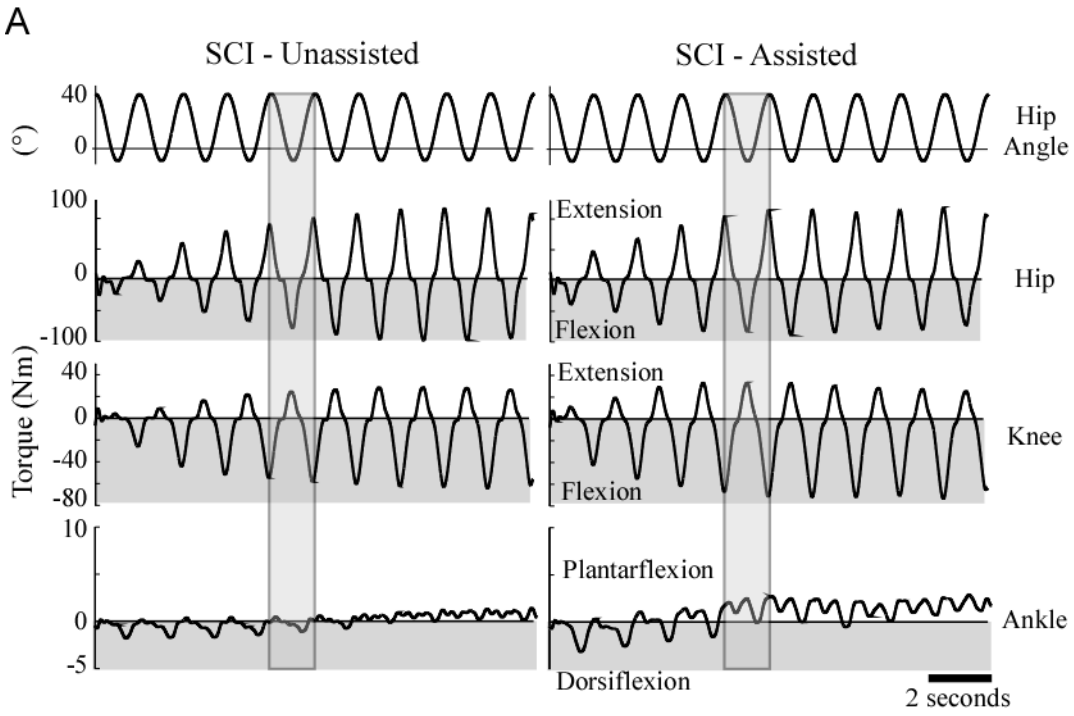
Separate univariate ANOVA was used to compare the effects of movement frequency (0.25, 0.50, and 0.75 Hz), supraspinal input (i.e. assisted vs. unassisted tests), and subject (random factor within each group) on the torque and EMG amplitude. Because not all subjects produced reflex responses to unassisted movements, a higher variance was present within the SCI subject group resulting in a non-normal distribution of the unassisted torque and EMG data. Prior to conducting the statistical analysis, the torque and EMG data for all subjects were first transformed (square-root) to obtain a normal distribution. The Bonferroni correction method was used for the post hoc pairwise comparisons among the movement frequencies. Significance was accepted at $p<0.05$.

2.3 RESULTS

The aim of this study was to examine how spastic reflexes affect volitional control during bilateral hip movements in individuals with motor incomplete SCI. Torque and EMG measurements were used to compare coordination patterns during the assisted movements to responses during unassisted hip oscillations in order to examine the interactions of reflex behavior and voluntary effort. Representative responses during the assisted and unassisted tests from SCI subject S7 are illustrated in Figure 2-3 (left column). Under the unassisted condition, imposed bilateral hip oscillations elicited multijoint spastic reflex responses that were consistent with previous findings (Onushko and Schmit 2007; Steldt and Schmit 2004). Typical responses to unassisted hip movements consisted of hip extension and knee flexion torque when the leg moved into hip flexion, and hip flexion and knee extension torque when the leg moved into hip extension (Figure 2-3A). EMG recordings were consistent with the torque measurements. Hip flexor (represented by RF) and knee extensor (VL and VM) muscles were active during hip extension, and hip extensor/knee flexor (MH) muscle activity occurred during leg movement into hip flexion (Figure 2-3B). Torque and EMG measurements during assisted hip movements revealed that volitional effort did not appear to greatly affect the pattern of movement in comparison to the reflex responses during unassisted hip oscillations. In some subjects (S3, S4, S5, and S7), the amplitude of the response also appeared to be unaffected by voluntary efforts (e.g. see Figure 2-3, left column).

Overall, a majority of the SCI subjects (8/10) produced characteristic torque patterns during assist movements that resembled spastic reflex patterns during unassisted

hip oscillations. Maximum hip and knee torques coincided with the hip angle at the extreme ranges of motion (i.e. 40° flexion or 10° extension). Not all subjects exhibited activity at the ankle, creating greater variability in ankle activity patterns among the subjects; however, for those SCI subjects who demonstrated activity in the ankles during unassisted trials (n = 3), ankle plantarflexion occurred during hip flexion and ankle dorsiflexion generally coincided with full hip extension. The control subjects recruited for this study generated consistent torque and EMG patterns during assisted trials (subject example, Figure 2-3 right column). Unassisted oscillation of the legs about the hips elicited no responses from any control subject.



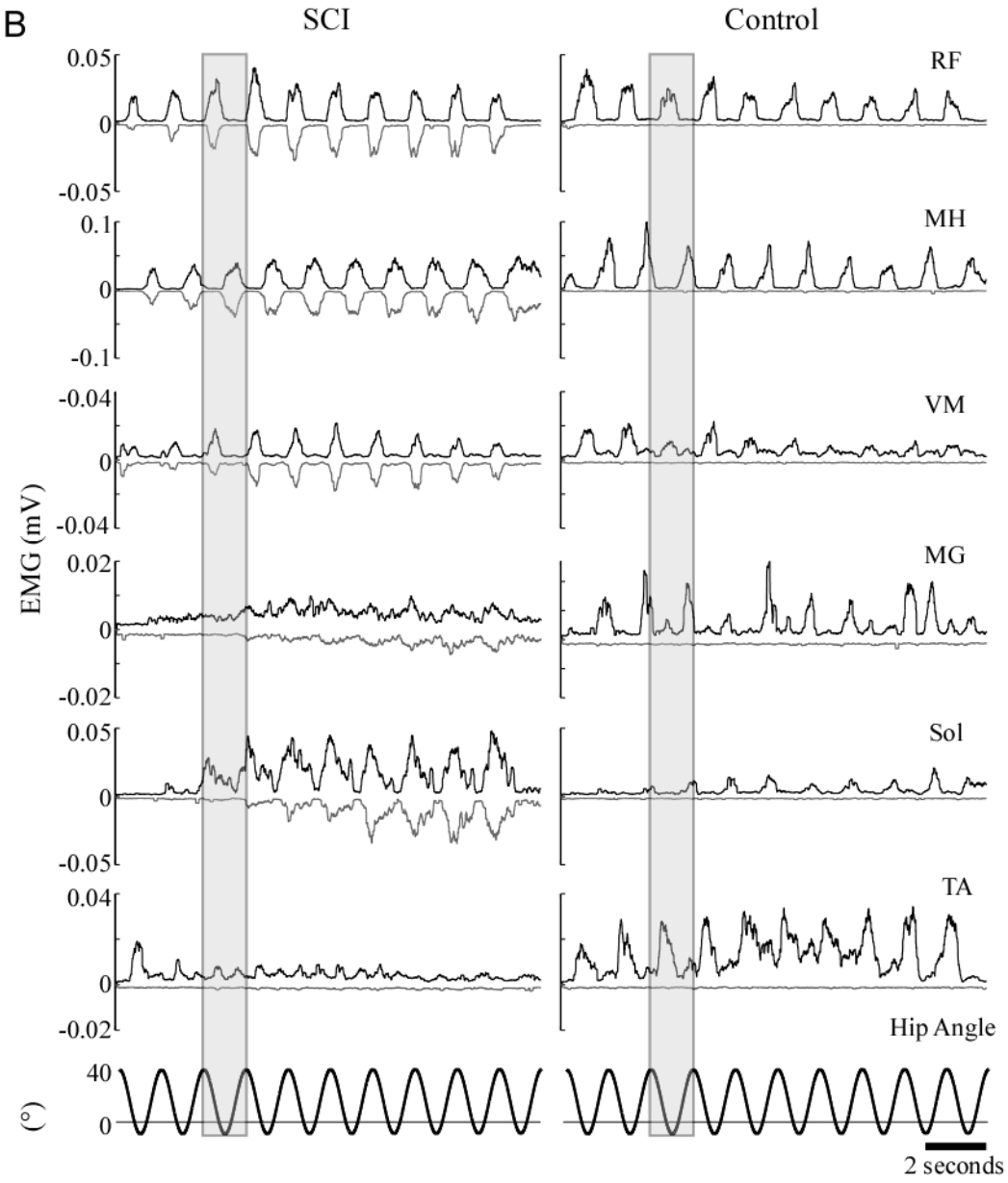


Figure 2-3 Representative Single SCI Subject Data. (A) Representative active torque traces during assisted and unassisted tests from SCI subject S7. Left column, SCI-unassisted; right column, SCI-assisted. Shaded areas represent flexion torques; unshaded areas represent extension torques. (B) EMG traces from SCI and control subject. Left column, representative EMG traces during assisted (positive rectified) and unassisted (negative rectified) tests from SCI subject S7. Right column, representative EMG traces from a neurologically intact control subject. Activity during unassisted and assisted movements for SCI subjects did not differ in timing with respect to the position of the hip.

2.3.1 Phasing Patterns during Assisted and Unassisted Hip Movements

The timing of the responses during bilateral hip oscillations in SCI subjects was similar between assisted and unassisted conditions, as evidenced by similar torque and EMG patterns during both test conditions. Figure 2-4 illustrates the average hip and knee torque generated from each SCI subject during both conditions. SCI subjects generated the most torque near the end ranges of movement (i.e. near full hip flexion/extension), whereas control subjects (95% confidence interval of all controls illustrated by a thick gray line in Figure 3-3) showed peak activity during the mid-range of hip movements (peak hip flexion and extension torque occurring approximately at 13° and at 22° of hip flexion, respectively). Two SCI subjects (S8 and S9), however, produced torque patterns that more closely followed the pattern of control subjects during assisted trials (indicated by the blue dotted lines in Figure 2-4), and no responses were noted during unassisted trials.

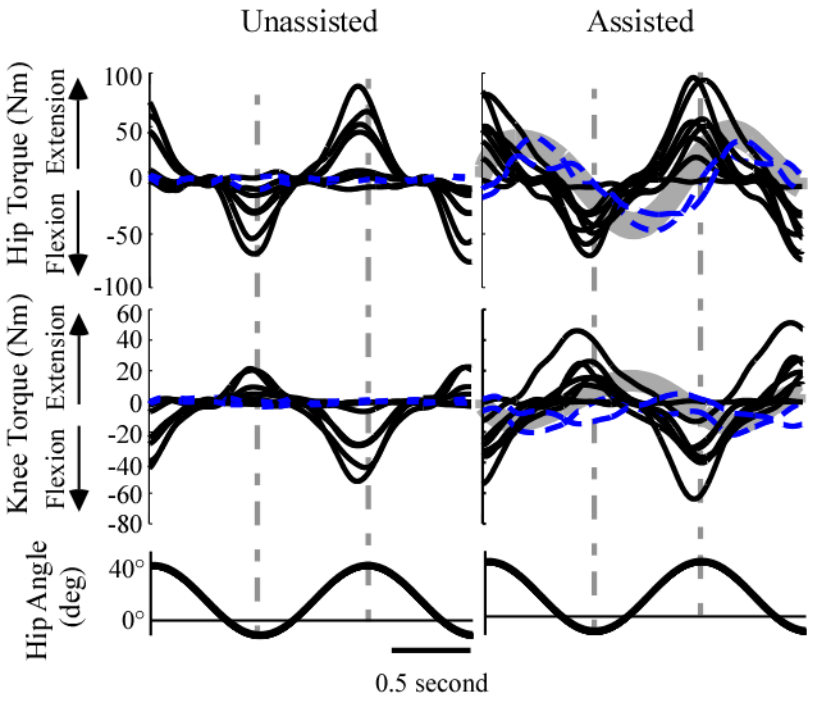


Figure 2-4 Average Torque Patterns during Assisted and Unassisted Hip Oscillations. Hip and knee torque responses of SCI subjects during unassisted and assisted bilateral hip oscillations (0.75 Hz shown only). Each solid black line represents the average response from a single SCI subject. Under the assisted condition, the thick gray line represents the 95% confidence interval of hip and knee torque responses from control subjects. Vertical dashed lines indicate the peak hip flexion and extension portions of the hip movement. Typically, the responses in SCI subjects occurred at the time when the hip reached full extension or full flexion. The dashed blue lines illustrate the responses from subjects S8 and S9, who generated patterns that more closely followed the pattern produced by the control subjects. In addition, these subjects did not produce responses under unassisted conditions.

Coordination of the lower extremities was not greatly affected when SCI subjects assisted the movements, and furthermore, it was not consistent with the patterns of muscle activity observed in neurologically intact subjects when performing the same task. The mean resultant vector for the torque measurements was used to ascertain the overall timing of muscle activity with respect to the position of the right hip during bilateral movements (refer to Figure 2-5A). With respect to the hip's position, the timing of peak

torque generation was not significantly different between assisted and unassisted tests for SCI subjects for any movement frequency (Watson-Williams Test, $P>0.05$). However, the timing of peak hip torque generation was significantly phase advanced for SCI subjects compared to the control group during the assisted task for all three frequencies (Watson-Williams Test, 0.75 Hz: flexion $P<0.001$ and extension $P<0.005$; 0.50 Hz: flexion $P<0.001$ and extension $P<0.001$; 0.25 Hz: flexion $P<0.001$ and extension $P<0.005$). Peak knee flexion/extension torque followed similar trends for the 0.75 and 0.50 Hz movement frequencies, but only approached significance during 0.25 Hz tests (Watson-Williams Test, flexion/extension for 0.75, 0.50 and 0.25: $P<0.001$, $P<0.005$, and $P>0.05$, respectively). Response patterns from ankle torque produced mixed results, yielding no significant phasing patterns in any test group (plantarflexion and dorsiflexion, $P>0.05$).

In general, the EMG results across SCI subjects showed little difference in phasing between the assisted and unassisted movements, corroborating the torque phasing results (Figure 2-5B). On average, EMG responses for the RF, VM, and VL muscles were phased with hip extension (RF, VM and VL mean hip angles: 9.2, 9.9 and 9.1°, respectively, of hip extension for assisted tests; mean hip angles: 10, 8.9, and 9.6° of hip extension for unassisted tests), and MH muscle activity typically coincided with hip flexion (mean hip angle for assisted and unassisted tests: 38.4° and 35.9°, respectively). Because not all subjects had volitional control over all muscles and not all subjects had reflexes during unassisted movements, a higher variability was present within the EMG data. On account of this variability, the statistical analysis comparing the EMG phasing patterns yielded no statistical significance for most of the muscles tested. However, MH

muscle activity was found to be directionally dependent for SCI subjects during assisted hip movements and was phased differently in comparison to MH activity of the control group (Watson-Williams Test, $P<0.005$ for all movement frequencies), which was at approximately 24° of hip flexion during the movements for the control group. Additionally, vasti muscle activity tended to coincide with the time near full hip extension (~ 9° hip extension) in both assisted and unassisted tests for the SCI group. Differences in the timing of muscle activity were noted for VL (Watson-Williams Test, $P<0.005$ for 0.75 Hz movement frequency only) and VM (Watson-Williams Test, $P<0.05$ for 0.50 Hz movement frequency only) EMG between SCI and control groups during assisted movements.

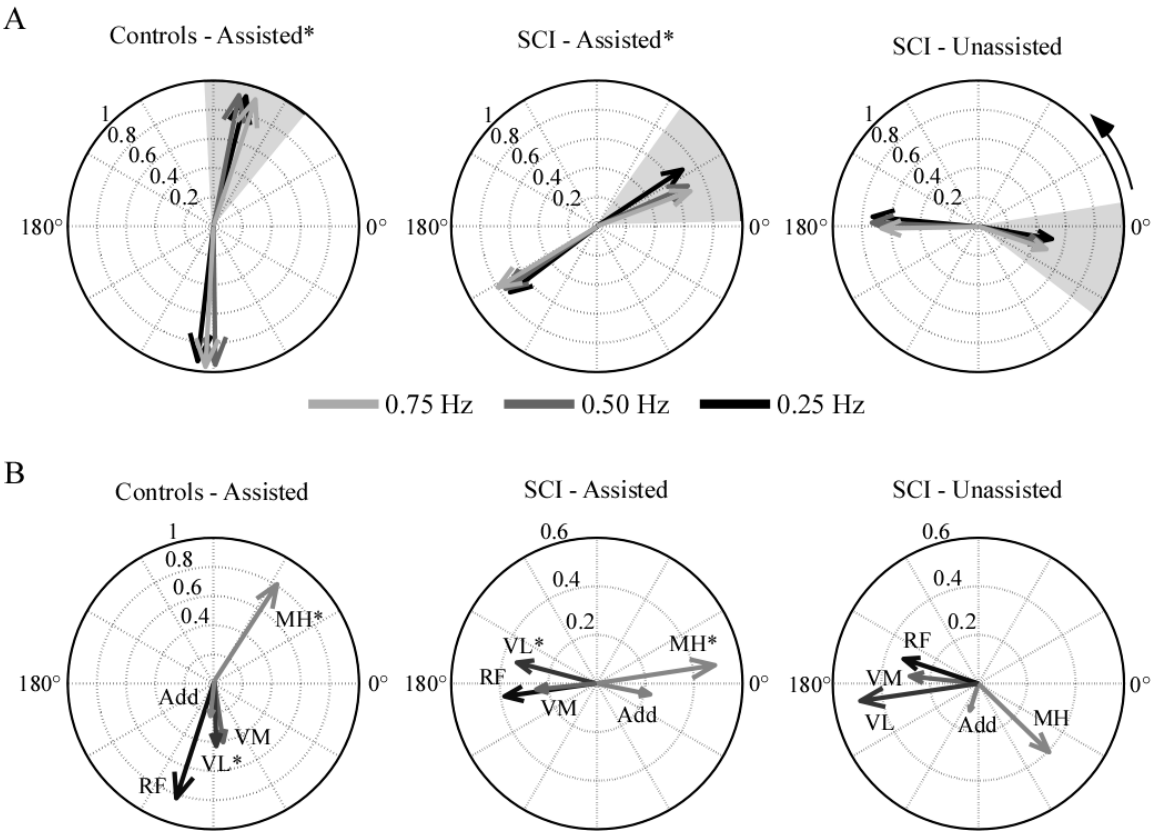


Figure 2-5 Average Torque and EMG Vectors. To obtain the timing of muscle activity during bilateral hip movements, the mean resultant vectors were plotted on a polar plane during assisted (SCI and control subjects) and unassisted (SCI only) tests. Full hip flexion corresponds to 0° and full hip extension corresponds to 180° on the polar plots. Hip extension torques are indicated within the shaded portion, and hip flexion torques are displayed over unshaded areas. The direction of hip motion is counterclockwise on the polar plane and is indicated by the curved arrow. Vector magnitudes (radius, *r*) represent the repeatability ($r = 1$ is identical phase) of the measurements during the hip movements. Arrows represent the mean direction of the torque or EMG measurement with respect to the position of the hip throughout one cycle of movement. (A) Group mean right hip extension and flexion torques for all three movement frequencies. Significant differences were found from the resultant peak torque vectors between controls and SCI during the assisted test condition (denoted by asterisks; $P < 0.005$). (B) Resultant vectors for the group average EMG of the right leg muscles for the 0.75 Hz movement frequency only. The timing of MH and VL muscle activity significantly varied between the SCI and control groups for the assisted task (denoted by asterisks; $P < 0.005$). No significant differences were found between SCI subjects during assisted and unassisted tests for any EMG phase data.

The timing of peak activity during the assisted hip movement task appeared to be influenced by the level of spastic reflex excitability present in subjects who participated in this study. Subjects who demonstrated greater spastic reflex activity (S3, S4, S5, and S7) were unable to alter the timing of muscle activity, and moreover, the pattern of muscle activity was similar between assisted and unassisted hip movements. Subjects who did not exhibit spastic reflex activity (S8, S9) (i.e. no reflex responses elicited during unassisted hip movements or MMT) were able to better coordinate movements in the lower extremities such that the EMG and torque patterns more closely resembled the coordination patterns generated by control subjects. Figure 2-6 illustrates the disparity in the timing of peak hip torque production among three subjects (control, SCI subjects S10 and S4). Representative of subjects who demonstrated greater spastic reflex activity, subject S4 produced reflex patterns during assisted hip movements that resembled hip-triggered reflex patterns during the imposed unassisted hip movements. The generation of peak torque typically occurred at the extreme ranges of the hip movements (i.e. 40° hip flexion or 10° hip extension) under both test conditions. Subject S10, one of two subjects who ambulated with the aid of a walker, did not generate multijoint reflexes during unassisted movements. Interestingly, even though subject S10 did not have spastic reflexes, the timing of peak hip torque was still delayed in comparison to the control subjects during the assisted movements.

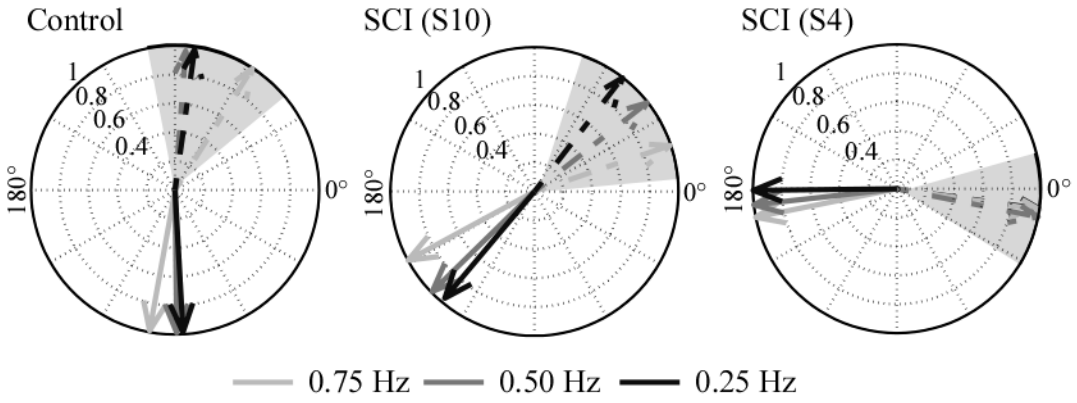


Figure 2-6 Torque Vector Comparison during Assisted Hip Movements. Normalized peak hip flexion (unshaded background) and extension torque (shaded background) for three different subjects (control, S10 and S4) during assisted hip movements. Full hip flexion corresponds to 0° and full hip extension corresponds to 180° on the polar plots. Spastic reflex excitability appeared to influence the timing of muscle activity during assisted tasks, such that the timing of peak torque production was unaltered between assisted and unassisted hip movements in subjects demonstrating greater spastic reflex activity.

2.3.2 Response Magnitude to Hip Movements

Although the pattern of activity was similar between the assisted and unassisted conditions, voluntary effort during assisted hip movements augmented responses overall. The magnitude of hip and knee torques across the test group were greater during volitional attempts in comparison to reflex responses during unassisted trials (ANOVA, hip flexion $P<0.001$; hip extension $P<0.001$; knee flexion $P<0.005$; knee extension $P<0.001$). The response at the ankle produced mixed results. Peak ankle plantarflexion torque was greater during assisted tests across SCI subjects (ANOVA, $P<0.001$), and was not sensitive to the movement frequency (ANOVA, $P>0.05$). There was no difference in peak dorsiflexion torque between assisted and unassisted tests (ANOVA, $P>0.05$), but peak dorsiflexion torque was sensitive to the movement frequency (ANOVA, $P<0.05$)

producing greater torque during the 0.75 Hz movement frequency compared to the 0.50 Hz trials. Peak hip and knee torque responses were not greatly affected by the frequency of the movement. EMG amplitude was also larger during the assisted trials as compared to the unassisted trials across SCI subjects (ANOVA, $P<0.001$). Figure 2-7 illustrates the normalized mean EMG results for the two test conditions at all three trial frequencies. In general, the movement frequency did not greatly affect the EMG amplitude. However, the EMG amplitude was greater in the 0.75 Hz movement frequency compared to the 0.25 Hz movement frequency for the Add, VL and MG muscles (ANOVA, $P<0.001$, $P<0.001$, and $P<0.05$, respectively).

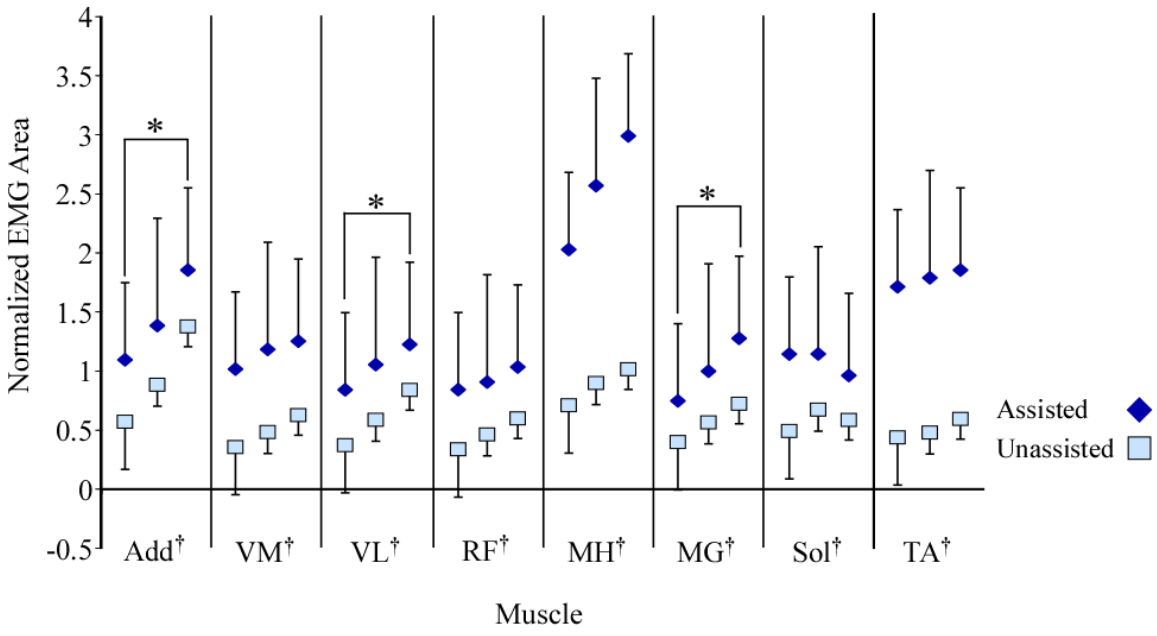


Figure 2-7 Average EMG Amplitudes. Normalized EMG group means for assisted and unassisted test conditions for all muscles of the right leg for each trial frequency (0.25, 0.50 and 0.75, from left to right for each muscle). Error bars indicate 1 SD of the group data. Asterisks above the group data indicate a significant difference between the two trial frequencies ($P<0.01$). †There was a significant difference in EMG amplitude between assisted and unassisted tests for all muscles.

2.3.3 Reflex Activity during Assisted Hip Movements

Statistically, peak hip and knee torques were not dependent on the movement frequency for either assisted or unassisted trials within SCI and control groups, but an observable trend was apparent between SCI and control subjects (refer to Figure 2-8, hip torque is shown only). The average peak hip and knee torque increased with higher movement frequencies for SCI subjects during both assisted and unassisted hip movements. This relationship could likely be due to stretch reflex activity of hip flexors and extensors during the hip movements. For instance, with increased movement velocity, peak hip extension torque may be augmented if the MH muscle was eccentrically contracting as the leg reached full flexion. Similarly, eccentric contraction of the RF muscle could also explain the increased peak hip flexion torque. In contrast to SCI subjects, control subjects demonstrated a decrease in peak hip torque with increasing movement frequency, generating concentric muscle contractions at the hip and knee during assisted trials.

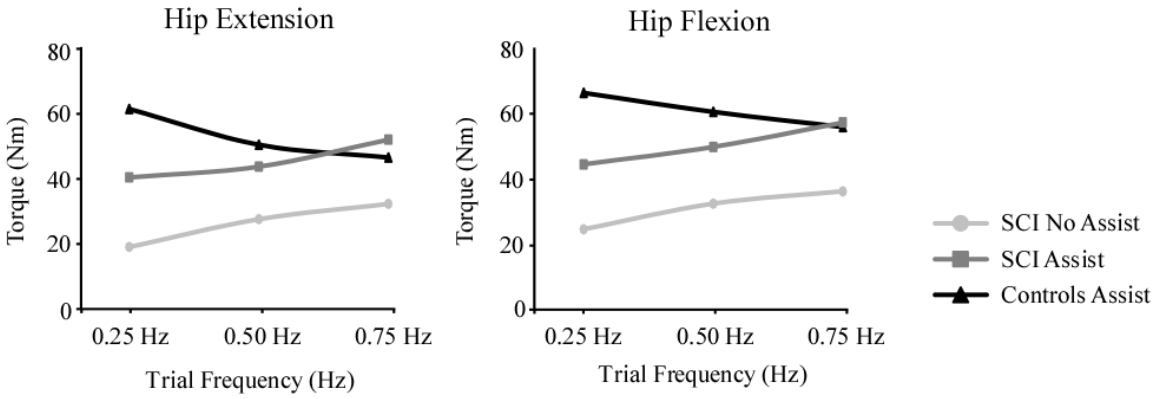


Figure 2-8 Torque vs. Frequency. Average peak hip flexion and extension torque from SCI and control subjects plotted against each movement frequency. On average, hip torque amplitude was greater during assisted trials compared to unassisted trials for SCI subjects. However, under both conditions torque magnitude increased linearly as the movement frequency was increased. The opposite was seen for the control group during assisted movements, with peak torque decreasing with increasing movement frequency.

2.4 DISCUSSION

Results from this study showed that multijoint spastic reflexes appeared to influence the timing and magnitude of muscle activity during assisted movements of the legs in people with motor incomplete SCI. The patterns of torque and EMG data from SCI subjects during assisted hip motion were ill-timed with respect to the position of the hip in comparison to neurologically healthy subjects performing the same task. Moreover, EMG and torque patterns from SCI subjects during assisted movements resembled multijoint reflex activity generated in response to imposed, unassisted hip oscillations.

2.4.1 Changes in Reflex Regulation of Movements in Chronic SCI

During imposed hip movements, stretch reflex responses were observed in muscles at the hip during the active-assist task in SCI subjects. In the normal control of movement, stretch reflex excitability is modulated by supraspinal influences over inhibitory pathways (Gottlieb and Agarwal 1980), so that voluntary motor commands can utilize reflex pathways to meet functional demands. The amplitude of the soleus H-reflex, for example, is greatly modulated during human locomotion. Reflex amplitude is greatest during the stance phase providing increased ankle stiffness and assisting with torque generation at push off, and during the swing phase it is greatly inhibited to avoid dragging the toes over the ground (Capaday and Stein 1986). However, in spastic muscles there is deficient modulation of stretch reflex excitability, and it has been reported that there is a decrease in inhibition at the onset of movement (Crone et al. 2003; Jones and Yang 1994), which has been suggested to contribute to movement impairment in people with spasticity (Crone et al. 2003; Jones and Yang 1994; Morita et al. 2001). Hemiparetic stroke patients, for example, show increased stretch reflex activity in the antagonist muscle during voluntary, concentric knee movements (Knutsson et al. 1997), and when compared to passive movements, stretch reflex activity of the antagonist muscle was greater with active paretic knee movements (Fleuren et al. 2009). In the current study, SCI subjects demonstrated similar actions during the active-assist hip oscillations. For example, imposed leg movements into hip extension generated stretch of hip flexors resulting in facilitation of hip flexion rather than inhibition. Although no direct measure of voluntary drive was quantified in the present study, it does suggest that remaining descending drive increases reflex excitability in incomplete SCI subjects and

these individuals have limited capacity to inhibit antagonist activity during voluntary single-joint movements.

Without effective descending input to modulate reflex excitability, sensory feedback to spinal centers plays an essential role in regulating the amplitude and coordination of muscle activity. For instance, moving the legs in a motion kinematically similar to human locomotion with body weight fully supported is not enough to generate sufficient muscle activity in healthy or in SCI subjects, but in combination with ankle-foot loading, appropriate leg muscle activity can be generated in SCI subjects during stepping (Dietz et al. 2002; Ferris et al. 2004; Gordon et al. 2009; Harkema et al. 1997; Maegele et al. 2002). In the current study, it is likely that reflex responses were generated during the active-assist task, since reflex patterns were similar to the reflex responses observed during the imposed, passive hip movements. And while it has been postulated that these reflex responses overlap with spinal networks associated with locomotion (Onushko and Schmit 2007; Schmit and Benz 2002; Steldt and Schmit 2004) because of the lack of feedback from other sensory cues, perhaps such as loading, it is possible that spinal motor pools did not receive sufficient feedback for appropriate muscle recruitment during the assisted hip oscillations resulting in stereotypical reflex activity throughout the leg muscles. Maegele et al. (2002) showed a similar phenomenon in SCI subjects. When SCI subjects attempted single-joint movements, multijoint flexion or extension movements were observed throughout the legs. However, when these subjects attempted stepping while bearing partial body weight, they observed appropriate muscle activation patterns and level (Maegele et al. 2002).

Changes in spinal neuronal excitability due to alterations of intrinsic properties of motoneurons (Eken et al. 1989; Gorassini et al. 2001; Heckman et al. 2008) or interneurons (Edgley and Jankowska 1987; Theiss et al. 2007) could also have contributed to the observed coordination of muscle activity during the 'active-assist' task. Spinal neuronal excitability is modulated by descending monoaminergic input (Hounsgaard et al 1988; Lee and Heckman 1999); however, after complete spinal injuries resulting in the removal of descending inhibitory control, motoneurons respond or react to a wider variety of sensory inputs (Hyngstrom et al. 2008). With respect to motoneuronal excitability, it has been suggested that a disregulation of voltage dependent depolarizing persistent inward currents (PICs) may be involved in long-lasting spastic reflex activity in spinalized rats (Bennett et al. 2001, 2004) and in human SCI (Gorassini et al. 2004; Hornby et al. 2003, 2006). Changes in interneuronal excitatory and inhibitory networks may indirectly affect alpha-motoneuronal excitability involved with the emergence of long-lasting motor outputs in chronic SCI (Jankowska and Hammer 2002). Thus, from the current study, while study participants may have partial descending drive onto the spinal cord, it still could be insufficient to regulate reflex excitability as a result of the changes of intrinsic neuronal properties. Consequently, voluntary efforts could not adjust the timing of the responses, and instead external sensory cues elicited a set pattern of uncontrolled multijoint reflex behaviors.

2.4.2 Possible Involvement of Spinal Networks for the Generation of Locomotion

In chronic SCI, multijoint reflexes are associated with organized interneuronal spinal networks that produce coordinated muscle activity in the lower extremities, and

these networks may incorporate interneuronal circuitry that is involved with the spinal control of locomotion (Onushko and Schmit 2007; Schmit and Benz 2002; Steldt and Schmit 2004). As seen in cats, spinal locomotor networks receive sensory cues from hip proprioceptors that originate from stretching hip flexor musculature (Grillner and Rossignol 1978; Hiebert el al. 1996; Lam and Pearson 2001). These same sensory signals are suggested to be important for eliciting muscle activity during locomotion in humans (SCI: Dietz et al. 2002; Dobkin et al. 1995; Ferris et al. 2004; infants: Pang and Yang 2000, 2001). From the current study, multijoint reflexes were elicited during the imposed hip oscillations from most subjects during the unassisted task, and the pattern of activity resembled previous findings (Onushko and Schmit 2007; Steldt and Schmit 2004), further supporting the idea that spastic reflex pathways involve organized interneuronal networks. Additionally, the results from this study demonstrated that spastic reflex activity responds to sensory feedback (e.g. hip proprioception) for the appropriate timing of muscle activity, while supraspinal signals were responsible for controlling the amount of activation during locomotor-like activities. Since sensory feedback and supraspinal drive are both important for shaping locomotor rhythms from spinal locomotor networks, the findings from this study may implicate the possible involvement of locomotor reflex pathways with spastic reflex pathways in humans.

2.4.3 Interaction between Spasticity and Assisted Hip Movements

The findings from the current study suggest that multijoint reflexes in patients with spasticity interfere with concentric activation of muscles at the hip during a dynamic motor task. While spasticity has been largely evaluated in the resting limb, it is often

associated with disordered control of movement. The traditional definition of spasticity attributes it to an exaggeration of tendon reflexes that are associated with hyperexcitable stretch reflexes and muscle hypertonia (Lance, 1980). With regards to the established definition of spasticity, some investigators have suggested that hyperexcitable stretch reflex activity minimally contributes to motor impairments during voluntary tasks, and have concluded that spasticity is not a prime contributor to movement disorders associated with injury to the CNS. For instance, reflex gain modulation is reduced or similar to that of non-injured individuals during voluntary contraction of muscles in the arm (Burne et al. 2005) or ankle (Ada et al. 1998) of hemiparetic stroke subjects, implying that spasticity is a disorder of the resting limb and is not responsible for motor disorders. In contrast, others have reported that stretch reflex activity in the antagonist muscle increases during voluntary concentric contraction of the agonist muscle (Knutsson et al. 1980, 1997; Lum et al. 2004). Observations from the current study are in accordance with the latter studies, supporting the idea that spastic reflex activity hinders the control of voluntary movements. Even though the origin of spasticity may be different between SCI and stroke, owing to differences in lesion sites and the reorganization of sensorimotor pathways, in this study volitional drive enhanced muscle activity without correcting phasing errors and thus adversely affected the coordination of bilateral leg movements.

2.4.4 Clinical Implications for Rehabilitation

The assessment of muscle strength is an important part of physical examination in SCI individuals, since it can be used to detect improving or worsening neurological

function and can help in the planning for rehabilitation (Aitkens et al. 1989). In particular, muscle strength of hip flexors, hip extensors and hip abductors has been shown to be an important determinant for assessing walking ability in SCI patients (Kim et al. 2004). Muscle strength assessments using conventional techniques, such as the manual muscle test, are not sensitive to small changes in muscle strength (Bolliger et al. 2008; Herbison et al. 1996; Kim et al. 2004). Furthermore, tests are often performed isometrically with little relation to functional movements, and they do not take into consideration how volition may affect the intensity of reflex excitability in people with spasticity. From the current study, the data showed that manual muscle testing yielded no conclusive results linking strength and the ability to coordinate hip movement during the imposed hip movements. However, these findings are limited since four of the subjects were taking baclofen, which may have affected reflex excitability and influenced the responses observed during the hip movements. Nonetheless, data from this study suggest that other techniques may be required to discriminate reflex from voluntary activity at the hip when assessing individuals with motor incomplete SCI.

Regulating reflex behaviors in a temporally appropriate manner using external sensory cues may be beneficial for recovering locomotor function. Spastic reflexes can be elicited through hip afferent cues to produce strong responses throughout muscles in the lower extremities, but the abnormal timing of the responses may require additional sensory cues to correct inappropriate muscle activity. Alternatively, pharmacological interventions may also be necessary for some individuals to partially suppress the increased intrinsic excitability, particularly in interneuronal circuits involved with multijoint reflexes. In doing so, it may deter abnormal timing while maintaining

sufficient torque production through reflex output to improve locomotor function during rehabilitation. One of the primary concerns with this approach is that patients with profound weakness may rely on the reflexive muscle activity to achieve functional movement. Observations from the current and previous studies show that spastic reflex activity generates strong torque responses in the lower extremities (Onushko and Schmit 2007; Wu et al. 2005, 2006), and if all reflex function was reduced, it may prevent individuals with SCI from having enough strength to undergo locomotor training.

In conclusion, sensory information from the periphery can shape motor output when volitional drive is diminished, such as in human chronic SCI. Because the timing of muscle activity was similar between assisted movements and passively imposed hip movements, we concluded that spastic reflex activity likely influences coordination during voluntary leg movements in individuals with motor incomplete SCI. Additionally, volitional drive may serve primarily as a gain modulator of the level of motoneuronal activation in SCI, and thus may serve to enhance spastic, multijoint reflexes during attempts at movement.

CHAPTER 3

Bilateral Hip Oscillations Modulate Reflex Responses in Human Spinal Cord Injury

3.1 INTRODUCTION

In individuals with spinal cord injury (SCI), spastic reflex responses from stretch-sensitive afferents of the hip flexors results in uncontrolled activity throughout the lower extremities (Schmit and Benz 2002). Distinctive involuntary bilateral leg extension responses are often seen when SCI patients shift from a sitting to a supine position (Macht and Kuhn 1948; Little et al. 1989), with responses lasting for tens of seconds (Benz et al. 2005). In incomplete SCI, remaining supraspinal input seems to have limited influence over multijoint reflexes (Maegele et al. 2002; Onushko et al. 2010). In addition to hip afferent cues, knee proprioceptors and ankle load afferents have also been shown to trigger multijoint reflexes; however, hip extension appears to be a necessary condition for the spasms to be triggered with knee and ankle afferents slightly increasing excitation of the characteristic multijoint responses (Wu et al. 2005; Wu and Schmit 2006). The objective of this study was to identify the extent to which stretch-sensitive sensory feedback from the knee or ankle modulates reflex activity at the hip.

Spastic reflex activity in individuals with SCI has been traditionally classified under the general term of spasticity, which is characterized as velocity-dependent, hyperexcitable stretch reflexes (Lance 1980). However, spastic reflexes also manifest as flexor or extensor spasms (Benz et al. 2005), which involve more than hyperexcitable reflexes from stretch at a single joint. Especially in subjects with SCI, single-joint motion can trigger multijoint responses throughout the leg, including muscles that are not being stretched (Onushko and Schmit 2007; Schmit et al. 2000; Schmit and Benz 2002; Steldt and Schmit 2004). Particular to SCI, extensor spasms develop more frequently than

flexor spasms, with a prevalence of approximately 87% of individuals with chronic SCI (Barolat and Maiman 1987; Little et al 1989). Extensor spasms can be debilitating to individuals by interfering with transfers and sleep or cause pain (Little et al. 1989; Sjölund 2002). Even though these multijoint reflexes have been shown to originate from several afferent cues, the neural mechanisms associated with controlling these reflexes are not yet fully understood. Gaining insight into the behavior of these multi-segmental reflexes will help guide rehabilitation techniques for managing unwanted muscle activity.

It is thought that the disruption of descending spinal pathways causes fundamental changes in reflex regulation in human chronic SCI. For example, it has been reported that in chronic SCI, there is diminished modulation of Ia input by presynaptic mechanisms (Faist et al. 1994 and 1999; Yang and Wheelan 1993) as well as increased recurrent inhibition (Shefner et al. 1992). This loss of modulation over spinal inhibitory pathways likely contributes to inappropriate reflex modulation that has been reported in SCI (Crone et al. 2003; Hultborn 2003). Thus, the loss of inhibitory drive to targeted reflex pathways might result in the expression of reflexes that would normally be manifested only during functional tasks. In particular, stretch of the hip flexors may have an important functional role in walking and disinhibition of this reflex could contribute to extensor spasms in people with SCI.

During walking, afferent feedback from changes in hip position operates through spinal reflex pathways to regulate muscle activity throughout the leg in a functionally appropriate manner (Dietz et al. 2002; Grillner and Rossignol 1978; Knikou and Rymer 2002). Stretch-sensitive hip afferents are important for initiating limb flexion during the swing phase of gait as demonstrated in spinalized (Grillner and Rossignol 1978) and

decerebrate (Hiebert et al. 1996) cats, and in human infants during treadmill walking (Pang and Yang 2000). However, in human SCI, stretching of hip flexor muscles produces inappropriate facilitation of knee extension and ankle plantarflexion (Schmit and Benz 2002), both of which can be debilitating during functional movements. Because of their importance in walking and the associated disinhibition of reflexes after SCI, hip proprioceptors might play a unique role in the expression of spasms in human SCI.

In the present study we investigated whether stretch-related sensory feedback from the knee or ankle would modulate hip-triggered reflex activity during bilateral hip oscillations. Joint torque and EMG were measured from 9 SCI subjects while patellar tendon tap perturbations or vibration of the Achilles tendon were applied at different phases of the hip cycle (i.e. during hip flexion or hip extension) throughout the imposed hip movements. Measurements during the sensory perturbations were compared to a control condition, in which no sensory perturbation was applied, to test whether sensory input applied to distal muscles could alter lower extremity muscle activity. We hypothesized that joint torque and EMG reflex modulation would be predominantly dependent on hip afferent input even with added sensory feedback.

3.2 METHODS

3.2.1 Study Participants

Nine subjects with chronic SCI (mean age: 43.1 yrs) participated in this study. Seven SCI participants were classified as having incomplete injuries (American Spinal

Cord Injury Association (ASIA) Classification B, C or D) with eight subjects having cervical and one subject with thoracic level of lesion. One subject was classified as a complete injury (ASIA Classification A). This subject's data were excluded from the group analysis since this subject did not present with spasms in the lower extremities. At the time of this study, two of the nine subjects were taking antispastic medication (baclofen) to reduce the frequency and intensity of their spasms, and two of the nine subjects were community ambulators (S2 and S9). The clinical features of each subject are described in Table 3-1. Additionally, the Ashworth Scale (an ordinal scale from 0, no increased tone, to 5, increased tone where passive range of motion is difficult) was used to quantify the severity of spasticity (refer to Table 3-2). Exclusion criteria for this study included: significant medical complications due to skin breakdown, urinary tract infection, other secondary infections, respiratory failure, heterotopic calcification, or other concurrent illnesses limiting the capacity to conform to study requirements, significant osteoporosis, or the inability to give informed consent. Informed consent was obtained prior to study participation and all procedures were conducted in accordance with the Helsinki Declaration of 1975 and approved by the Institutional Review Board of Marquette University.

Table 3-1 Subject clinical characteristics

Subject	Injury Level*	ASIA Score	Age (years)	Time Post Injury (yr)	Medications
S1	C6	D	32	10	Ditropan XL
S2	C4-5	D	52	8	Baclofen
S3	C3	D	55	3	Baclofen, Gabapentin, Oxybutinin, Duloxetine HCL
S4	C5-6	C	45	28	None
S5	C6	C	33	9	None
S6	C6-7	D	48	23	None
S7	T5-T6	A	43	25	None
S8	C5	B	50	30	None
S9	C6	D	30	9	None

Table 3-2 Ashworth Scale Spasticity Scores (Right leg scores given only)

Subject	Hamstrings	Quadriceps	Gastrocnemius	Soleus
S1	2	1	2	2
S2	na	na	na	na
S3	3	1	4	4
S4	5	1	3	4
S5	5	3	4	4
S6	4	3	3	2
S7	2	1	1	1
S8	na	na	5	5
S9	3	3	4	4

3.2.2 Experimental Setup

Subjects lay supine on a therapy plinth with their legs supported in custom-built leg braces. The custom-built leg braces were adjustable to accommodate a wide range of leg lengths. The legs were supported in leg braces by a strap securing the thigh, a strap securing the heel and a clamp over the dorsum of the foot to secure the foot to the plate at the end of the leg brace. The knee and ankle joints were held isometrically for these tests since extensor spasms can be triggered through stretching of hip musculature (Benz and Schmit, 2002). The leg braces were attached to two servomotor drive systems (Kollmorgen, Northhampton, MA) that were used to impose hip oscillations to the subject's legs (Figure 3-1A). Custom-developed LabVIEW programs (National Instruments Corp., Austin, TX) and a data acquisition card (National Instruments Corp., Austin, TX) were used to control the hip oscillations. The system was equipped with reaction torque transducers (S. Himmelstein and Company, Hoffman Estates, IL) that aligned with the hip, knee and ankle anatomical joint axes of rotation for each leg. During the hip oscillations, the hip angle was measured using optical encoders (US Digital, Vancouver, WA) that were affixed to the respective motor shafts.

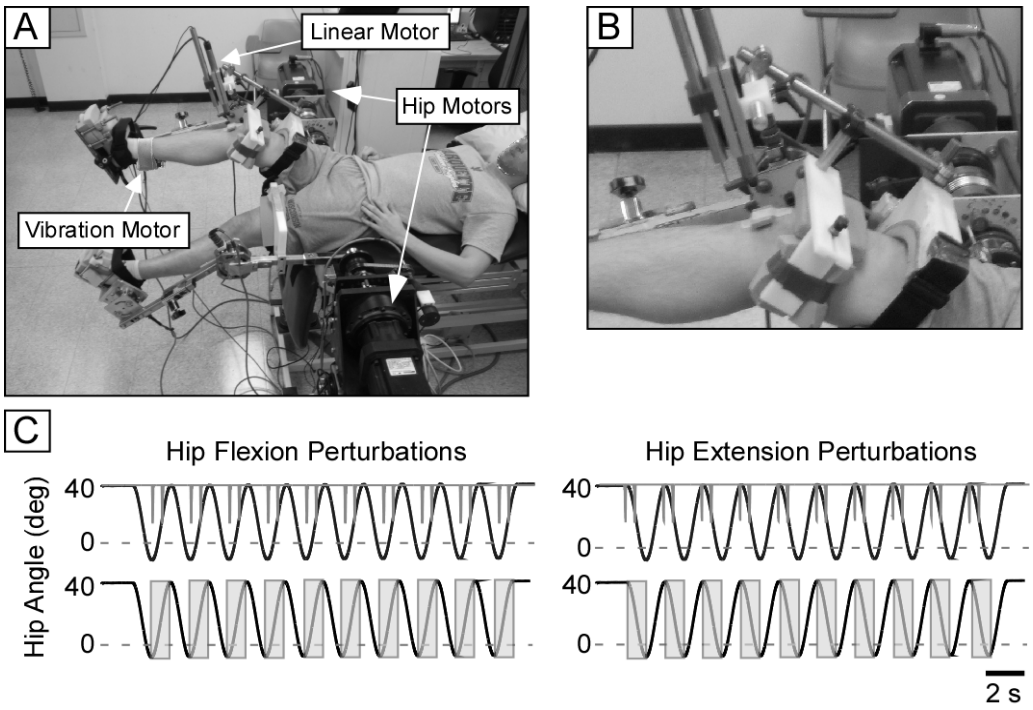


Figure 3-1 Experimental Setup. (A) Experimental apparatus used to impose bilateral hip oscillations and record sagittal plane torques during hip movements. Two servomotor systems were used to impose movements about the hips. A linear motor was aligned with the patellar tendon and a small motor was strapped around the subject's ankle to vibrate the Achilles tendon. (B) The tip of the linear motor was placed approximately 2 inches from a rubber pad that was taped to the tendon, and hit the patellar tendon at approximately 90°. The knee was braced with padding to minimize leg movement relative to the linear motor. (C) Patellar tendon tapping and Achilles tendon vibration perturbations were applied every half-cycle (i.e. either when the hip was flexing (left panels) or extending (right panels)).

Patellar tendon reflexes were elicited using a motorized reflex hammer (see Figure 3-1A). The motorized hammer consisted of a LinMot P series linear motor (P01-23x16/70x70) that was powered by a LinMot E1010 amplifier (LinMot Inc., Delavan, WI). The linear motor was mounted on the leg brace for the right leg (refer to Figure 3-1B). A small rubber tip (12 mm diameter) was screwed into the end of the motor shaft.

After palpating the knee to locate the tendon, the motor was set up so that the rubber tip was placed approximately 5 cm from the patellar tendon with an impact angle at approximately 90°. Additionally, a rubber pad (28 x 31 mm) was taped to the subject's tendon to evenly distribute the force from the motorized tapper to the tendon (refer to Figure 3-1B). A custom-written LabVIEW program was used to control the velocity (5 Volt biphasic square wave) of the linear motor. A linear variable differential transformer (LVDT; Accusens Series 2000 DC-EC, Measurement Specialists Inc., Hampton, VA) coupled to the linear motor shaft was used to measure the displacement of the motor during tendon tap perturbations.

A custom-made vibration system was used to apply an 80 Hz vibration to the subject's Achilles tendon. A small DC motor (Faulhaber Minimotor SA; GMBH & Co. KG, Schönaich, Germany) was used to rotate two eccentric weights that were attached to the motor's shaft (one weight on each end of the motor) to produce the vibration. The motor and weights were encased within a plastic tube, which was securely strapped onto the subject's ankle (Figure 3-1A). A custom-written LabVIEW program was used to send a 5-volt step input (4800 RPM) to control the motor velocity.

Surface electromyograms (EMGs) were collected from 7 muscles on each leg: rectus femoris (RF), vastus medialis (VM), vastus lateralis (VL), medial hamstrings (MH), medial gastrocnemius (MG), and tibialis anterior (TA). Disposable, pregelled Ag/AgCl recording electrodes (Vermed Inc., Bellows Falls, VT) were attached to the muscle belly of each muscle in a bipolar arrangement. Prior to electrode placement, the skin was cleaned and lightly abraded with a gauze pad doused with alcohol. EMG signals

were amplified (×1000) and band-pass filtered (10-1000 Hz; Bortec Biomedical Ltd., Calgary, AB, Canada) prior to data collection.

3.2.3 Experimental Protocol

Isokinetic hip flexion and extension movements were imposed to the legs of the subjects using the servomotor apparatus pictured in Figure 3-1A. The legs were moved in an alternating manner (i.e. 180° out of phase) through a 50° range of motion (from 40° of hip flexion to 10° hip extension) for ten continuous cycles at 50 °/s. The knee and ankle joints were held isometrically within the leg braces (knee: 45° flexion; ankle: 15° dorsiflexion). To test whether hip-triggered reflex patterns could be altered through sensory feedback from other joints, one of two perturbations was applied during different phases of the hip oscillations (refer to Figure 3-1C): (1) patellar tendon tapping during movement of the right leg into hip extension ("tap-ext") or hip flexion ("tap-flx") or (2) 80 Hz vibration of the Achilles tendon during movement of the right leg into hip extension ("vibe-ext") or hip flexion ("vibe-flx"). For each half of a cycle of the hip oscillations (i.e. flexion or extension), two tendon tap perturbations were applied (2 Hz, 25 ms pulse duration) using the motorized reflex hammer for tendon tap tests, and for vibration tests, Achilles tendon vibration was applied throughout the duration of the half-cycle (5 seconds) (refer to Figure 3-1C). A control condition was also performed in which no perturbation was applied during the hip oscillations ("control"). Additionally, to test whether remaining descending drive had an effect on spastic reflex responses with the added sensory input, subjects were asked to either assist the imposed hip oscillations ("active") or to remain relaxed during the oscillations ("passive"). Each condition was

presented in a random order and repeated three times using a block design (total of 30 tests). Torque and EMG were recorded throughout the duration of the hip movements.

At the end of the experiment, two additional hip oscillation tests were performed to estimate the torque due to gravity, passive joint resistance and inertial properties of the participant's legs. The legs were moved slowly through the 50º range at 2 º/s in 5º increments, pausing for five seconds between each increment, to measure the combined torque contribution from gravity and passive joint resistance. Then, inertia was estimated from measurements during which each leg was quickly oscillated (1.5 Hz) within a midrange of the hip (25º to 10º hip flexion) to minimize the potential for eliciting reflexes during the test.

3.2.4 Active Muscle Torque

The biomechanical properties of the leg and leg brace were estimated and used to correct the measured hip, knee and ankle joint torque using the method described in Chapter 2, section 2.2.4. Briefly, all recorded torque data were low-pass filtered (8 Hz) using a 2nd order Butterworth filter in Matlab (*filfilt* function; The Mathworks, Inc., Natick, MA). The torque due to gravity and passive resistance were estimated by fitting a 3rd order polynomial curve to the torque measurements taken during the slow, incremental hip movements. Those polynomial coefficients were then used to calculate the gravitational and passive resistance torques from the measured data. The inertial properties of the leg and leg brace were calculated by subtracting the gravitational and passive resistance torque, and then estimating the inertial constant of the leg and leg brace using a linear regression analysis. The mechanical artifact within the system

(approximately < 5Nm) was estimated using an ensemble average of torque measurements recorded from neurologically healthy subjects from the experiment in Chapter 2 (refer to section 2.2.4). The torque produced by active muscle contraction was calculated by subtracting the gravitational/passive torque, inertial torque, and artifact from the measured trial torque data, and was used for all subsequent analyses.

3.2.5 Peak Torque

The peak torque was used to determine the overall amplitude of the response to the hip oscillations. The peak flexion and extension torques for the hip, knee and ankle of the right leg were found for each cycle of hip movement. For knee extension torque during trials with patellar tendon taps, the reflex torque response from patellar tendon taps was not used for the peak torque analysis; rather, the peak torque from the movement was used for the peak torque analysis. The mean peak flexion and extension torques for each joint were calculated for each cycle (i.e. 1-9) across subjects. The peak torque at each joint (hip, knee or ankle) for each subject was normalized to the grand mean of that subject's torque data across test conditions (e.g. the peak hip torque was normalized to the average peak hip torque for the 10 test conditions × 3 trials) for comparison across subjects. During the progression of the study, the hip and knee joint from subject S3 became misaligned with the torque transducers in the apparatus, and his torque data were excluded from group analysis.

3.2.6 Surface EMG

Surface EMG signals from the right leg of SCI subjects were analyzed to obtain the level of muscle activity during the experimental procedures. All surface EMG recordings were band-stop filtered to remove line noise (59-61 Hz) and artifact from the 80 Hz vibration (79-81 Hz plus harmonics), and band-pass filtered (20-300 Hz) using 4^{th} order Butterworth filters (*filtfilt* command, Matlab). For analysis, the root-mean square (RMS) of the EMG data was calculated using a 100-ms moving window. At the end of the hip movements, the leg remained in full flexion providing continuous afferent feedback to spinal neurons from hip extensor muscles. Since we were interested in responses during dynamic hip movements, the last cycle was excluded from analysis. Muscle activation was quantified by calculating the mean integrated area of the RMS EMG signal for every cycle for which the muscle was active, across all three trials (total average of 27 cycles). Additionally, the magnitude of the reflex responses during patellar tendon tapping trials was also calculated. The peak-to-peak (p-p) amplitude, measured as the difference between the largest positive and negative peak, was calculated for the RF and VM muscles and then averaged across subjects. The RF and VM data from each subject were inspected by eye to ensure reflex responses were included and artifact was excluded from the analysis. For all EMG measurements, the data for each muscle for each subject was normalized to the mean of that subject's entire EMG data based on all experimental conditions.

3.2.7 Statistical Analysis

Individual univariate ANOVAs were used to identify the effects of Achilles tendon vibration and patellar tendon tapping on peak torque and EMG areas (significance level, $\alpha = 0.05$). Peak flexion and extension torque for the hip, knee and ankle were analyzed separately using univariate ANOVAs to compare test condition (vibe-ext, tap-ext, vibe-flx, tap-flx and control), effort (active or passive) and subject (random factor). A similar analysis was done on the average MH EMG areas. A univariate ANOVA was used to test for differences among test conditions (vibration, tendon tapping, or control), timing of stimulus (movement of hip into flexion or extension), effort (active or passive), and subject (random factor). Similarly, a univartiate ANOVA was also used to compare MG areas between the phase of Achilles tendon vibration (i.e. vibe-ext, vibe-flx or control) and effort (active or passive). Statistical comparisons were also made between the p-p reflex amplitude of the RF and VM during patellar tendon tapping trials. The response to the first and second tendon tap of each two-tap series per cycle were compared within each muscle (i.e. RF and VM) across subjects (main factors: tap number grouped with tendon tap phase (e.g. 1^{st} tap during movement of the leg into extension), effort (active or passive); random factor: subject). Prior to conducting the statistical analysis on the p-p RF and VM data, the data were first transformed (natural log) to obtain a normal distribution. The Bonferroni correction method was used for post-hoc pairwise comparisons between the test conditions (significance level, $\alpha = 0.05$).

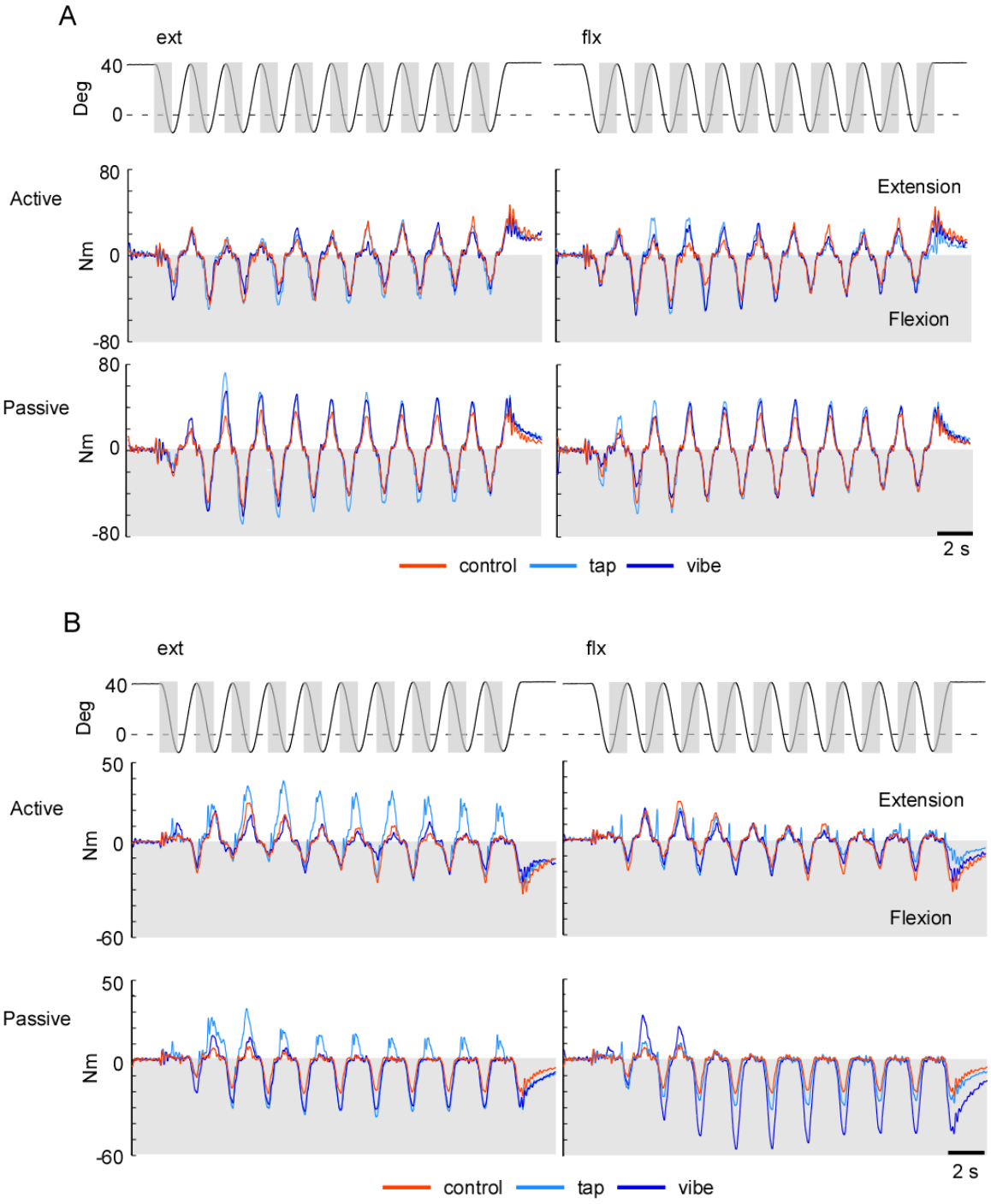
3.3 RESULTS

In general, Achilles tendon vibration and patellar tendon tapping did not greatly affect the responses produced at the hip joint, but they did modulate responses at the ankle and knee joints, respectively (see Figure 3-2). For example, Figure 3-2B shows an increase in knee extension torque when the tendon tapping was applied during movement of the hip into extension, whereas when the tendon tapping was applied during hip flexion, the knee torque pattern resembled the pattern during the control and vibration conditions. Similarly, RF and VM EMG activity also increased during trials in which tendon tapping was applied during hip extension compared to the other conditions (Figure 3-2C). Additionally, the example data shown in Figure 3-2 are representative of typical responses observed between active and passive trials. Responses during the passive trials were typically lower in amplitude and tended to lessen with the repeated hip oscillations even with added sensory input (n = 6/8); however, two subjects (S4 and S5) demonstrated responses that were similar in amplitude between passive and active trials.

3.3.1 Torque Responses

From the group results, the hip flexion and extension torque amplitudes were fairly consistent throughout the hip oscillations during the active trials (Figure 3-2). During the passive trials, the pattern of peak torque was similar to the active condition, but the amplitude was significantly lower for both hip flexion and extension torque (ANOVA, $p = 0.000$). Neither vibration nor tendon tapping, nor the phase of the stimulus (i.e. flexion or extension) significantly altered the peak hip extension torque

(ANOVA, p = 0.934). Similarly, peak hip flexion torque during vibration and tendon tapping trials was not significantly different from the control condition (ANOVA, p = 0.842).



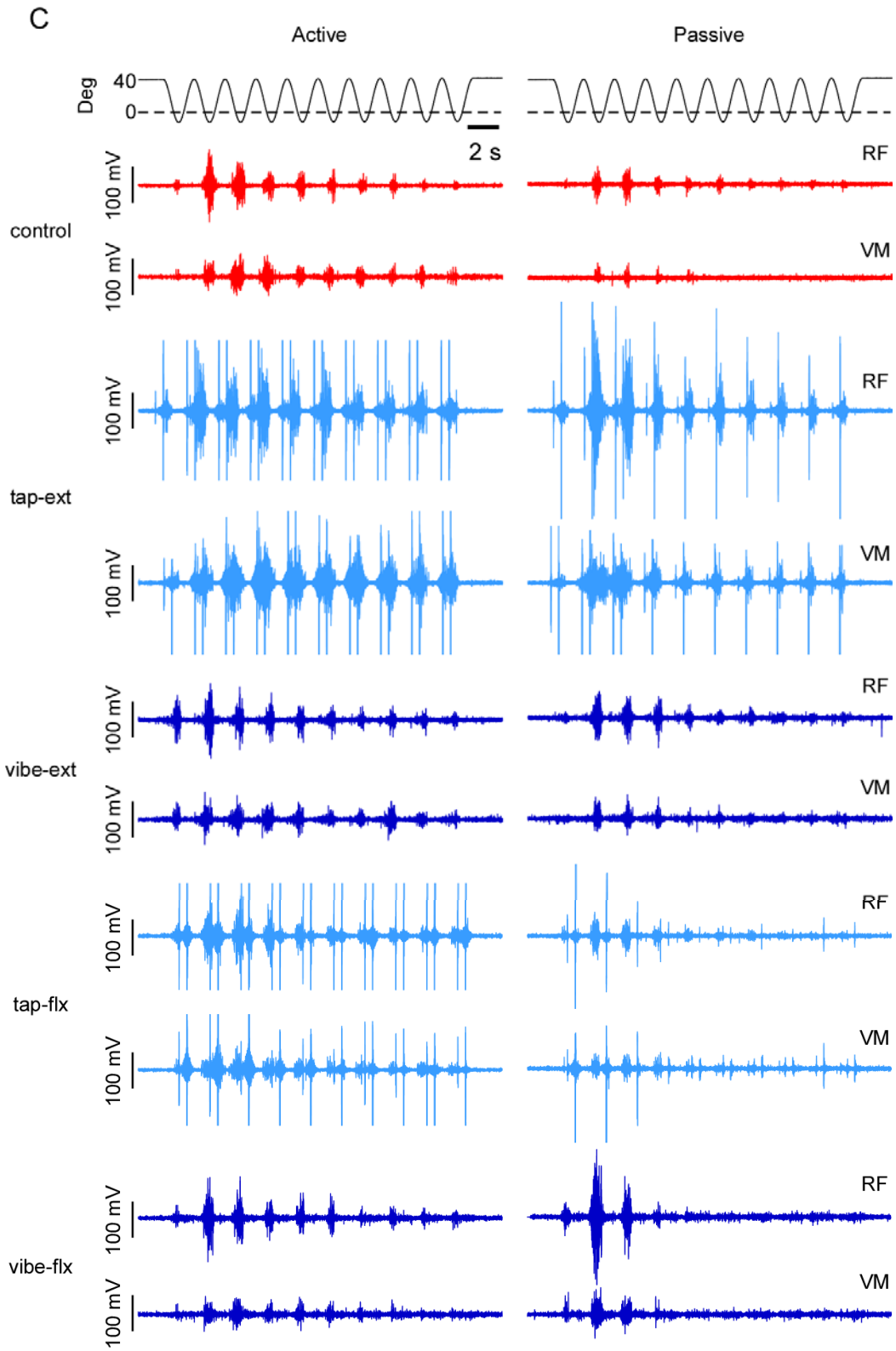

C
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Figure 3-2 Single SCI Subject Example Responses. Example hip torque (A), knee torque (B), and EMG (C) data from subject S6 representing responses from all test conditions. For the hip (A) and knee (B) torque traces, the panels on the left are trials where vibration or tendon tapping were applied during hip extension and the panels on the right are trials where the stimulus was applied during movement of the leg into hip flexion. (C) Peak RF and VM reflex amplitudes are not displayed in the figure in order to emphasize EMG bursting.

The direction of hip movement affected the responses measured at the knee and ankle joints when vibration or tendon tapping was applied. Figure 3-3 illustrates the difference in peak knee extension torque amplitude among the conditions for the active trials. The peak knee extension torque amplitude during the tap-ext condition was significantly greater compared to the other conditions (ANOVA, $p = 0.006$). While peak knee flexion torque amplitude showed variability among the test conditions, no significant difference was found among test conditions (ANOVA, $p = 0.432$). Achilles tendon vibration also had an effect on peak ankle plantarflexion torque. Peak ankle plantarflexion torque amplitudes were significantly greater for the vibration trials compared to the control condition (post-hoc: vibe-ext vs. control, $p = 0.013$; vibe-flx vs. control, $p = 0.031$), but not when compared with the tendon tapping conditions (post-hoc: vibe-ext vs. tap-ext, vibe-ext vs. tap-flx, vibe-flx vs. tap-flx and vibe-flx vs. tap-ext, $p = 1.000$). However, peak ankle dorsiflexion torque amplitudes did not differ among the test conditions (ANOVA, $p = 0.803$). Additionally, the peak amplitude for both knee and ankle were greater during the active trials compared to the passive trials (ANOVA, $p = 0.000$).

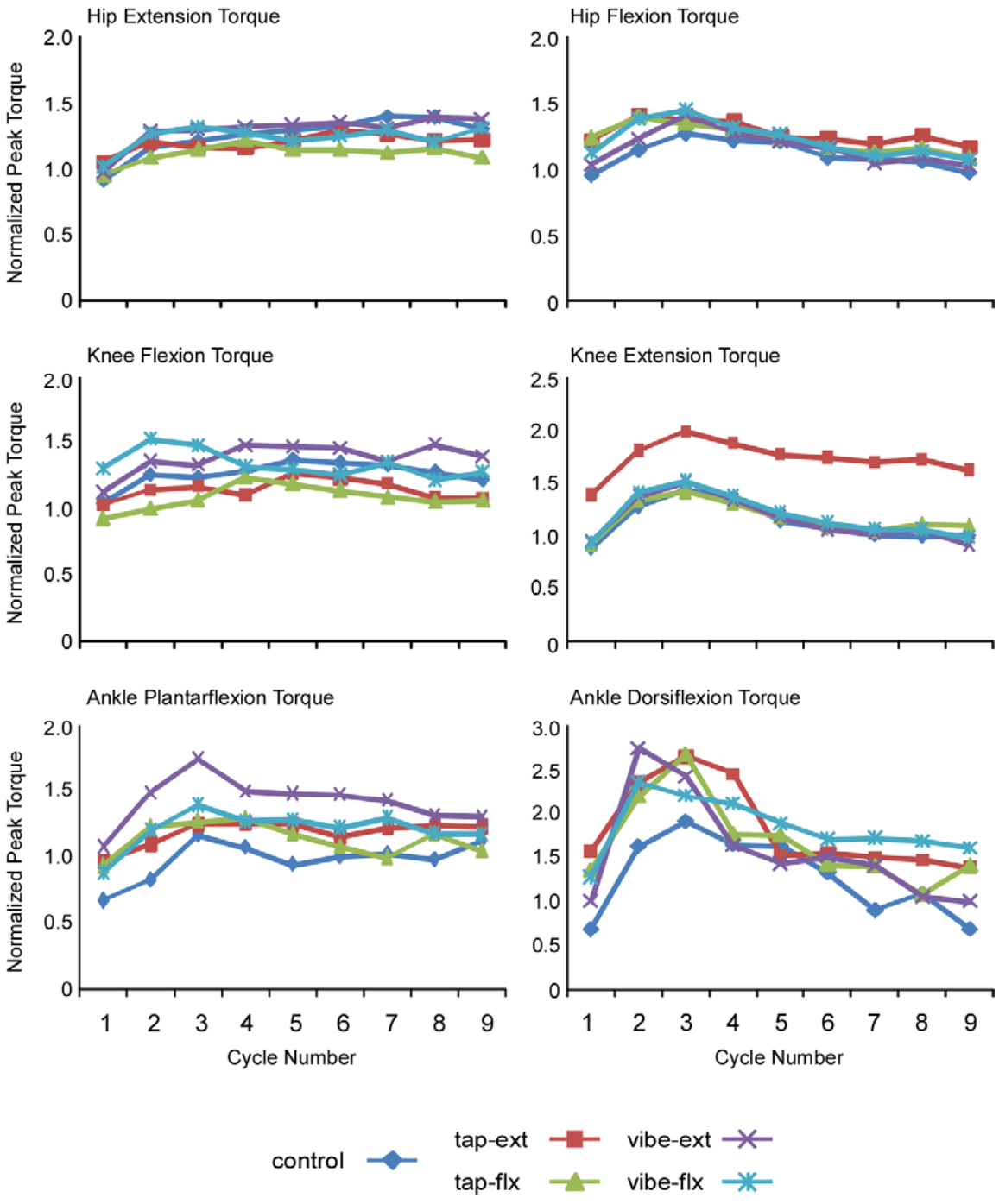


Figure 3-3 Average Peak Torque. Group average of normalized peak hip flexion/extension, knee flexion/extension and ankle plantarflexion/dorsiflexion torque per oscillation cycle (active condition only). Error bars are not provided in order to show the trends more clearly. Averages for active trials are > 1 since peak torque data were normalized to the average of active and passive trials.

3.3.2 EMG Responses

MH EMG activity showed differential effects with respect to the timing (flexion vs. extension) of the stimulus, but not to the type of stimulus (tap vs. vibration). Figure 3-4 illustrates the average MH EMG area during active and passive trials across the test conditions. Average MH EMG activity was greater when tendon tapping or vibration was applied when the hip was flexing compared to when the stimulus was applied when the hip was extending (post-hoc: flx vs. ext, $p = 0.010$). Although, the trend in the MH EMG activity appeared to show that vibration or tendon tapping during hip flexion was greater than the control condition, no significant difference was found (post-hoc: flx vs. control, $p = 0.067$). When subjects assisted the movements, MH EMG activity was greater compared to the passive trials (ANOVA, $p = 0.020$).

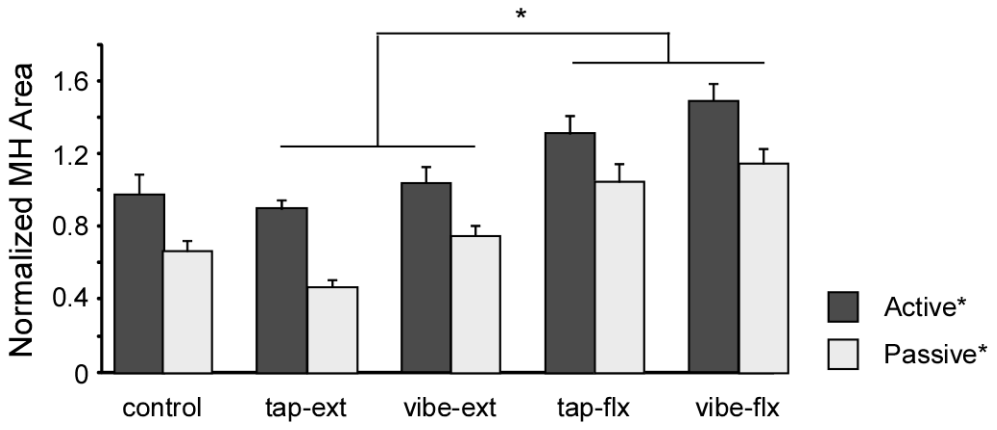


Figure 3-4 Average MH EMG Areas. Group average MH EMG area during hip oscillations during active and passive trials. MH EMG was modulated by hip position – tendon tapping and vibration increased MH activity when they were applied during hip flexion (significantly different, $p < 0.05$, from other conditions) and active trials had larger responses than passive trials ($p < 0.05$).

The peak-to-peak (p-p) RF and VM reflex responses during the tendon tapping trials were also modulated by the position of the hip, movement direction and number of the tap (first vs. second). Figure 3-5 illustrates the average p-p response from the 1st and 2nd tendon tap from the two-tap series per movement cycle. The average p-p RF response for the 2nd tap was significantly greater compared to the 1st tap for the tap-ext condition, but not for the tap-flx condition (post-hoc: tap-ext, $p = 0.000$; tap-flx, $p = 0.926$). The average p-p VM response between the two tendon taps showed the opposite effect as seen in the p-p RF response. The average 2nd tap p-p VM response was significantly larger compared to the 1st tap only for the tap-flx condition (post-hoc: tap-flx, $p = 0.001$; tap-ext, $p = 0.184$). Additionally, the p-p VM reflex amplitude from the 1st tendon tap perturbation was significantly lower in the tap-flx condition compared to the tap-ext condition ($p = 0.035$). Although not statistically significant, the p-p RF reflex amplitude from the 1st tendon tap perturbation for the tap-ext condition was on average smaller than the 1st tendon tap response in the tap-flx condition. Voluntary effort did not have an effect on the p-p RF or VM responses (ANOVA, $p = 0.682$ and $p = 0.810$, respectively).

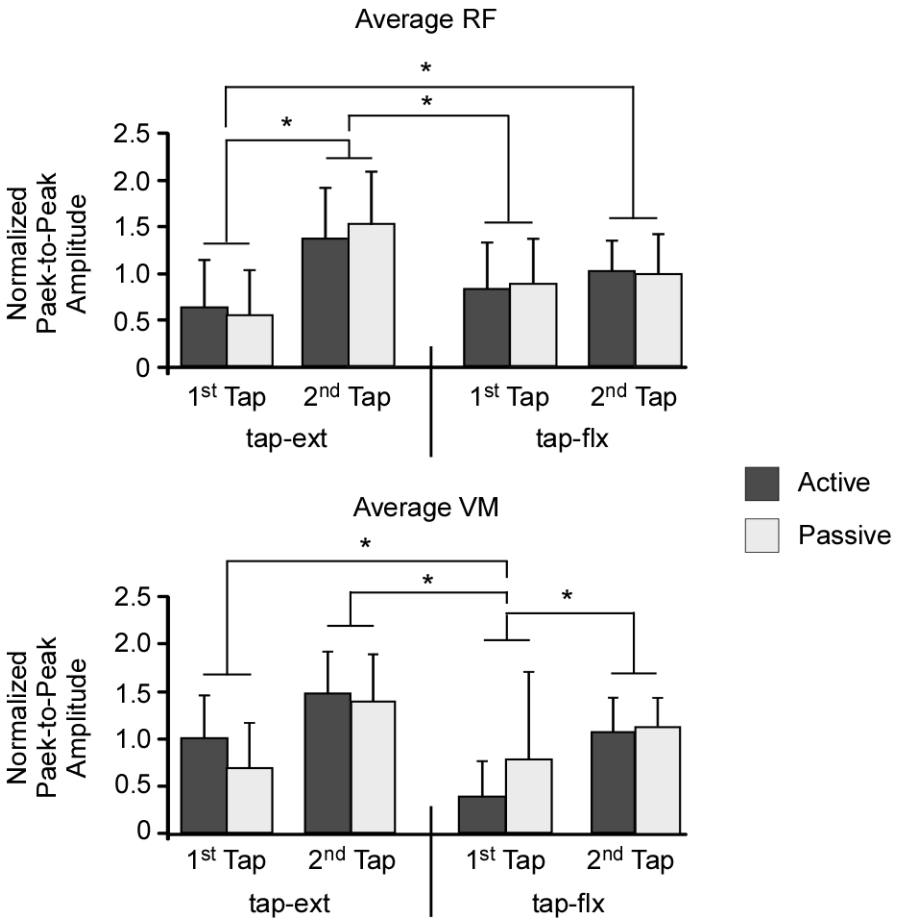
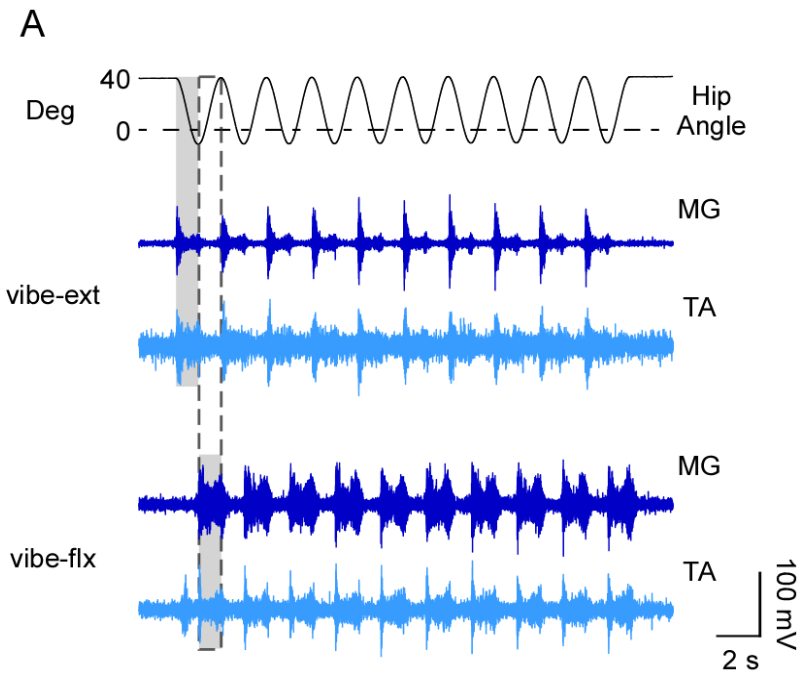


Figure 3-5 Peak-to-Peak Quadriceps EMG. Group average normalized peak-to-peak (p-p) RF and VM EMG during tendon tapping trials. Asterisks represent statistical significance between comparisons ($p < 0.05$).

Muscle activity at the ankle during Achilles tendon vibration trials also showed differences in activation patterns relative to the movement of the hip. Figure 3-6A shows typical MG and TA EMG responses from subject S3 during a passive (i.e. subject was not assisting the movements) trial with vibration applied during movement of the leg into hip flexion (vibe-flx) and into hip extension (vibe-ext). The response in the MG muscle to vibration resulted in a tendon vibration reflex (TVR), which was observed in 6 out of 8

subjects. Application of the vibration during hip extension movements resulted in a different MG EMG TVR response, where an initial burst was seen at the beginning of the cycle, but then rapidly decayed as the hip movement continued to full extension. TA EMG followed a similar trend as the MG EMG, but wasn't as strongly activated. The statistical analysis showed that MG EMG area was significantly greater when the vibration was applied during the flexion portion of the hip oscillations compared to the extension portion and control conditions (post-hoc: vibe-flx vs. vibe-ext, p = 0.000; vibe-flx vs. control, p = 0.000) (Figure 3-6B). Additionally, when subjects assisted the movements, the average MG response was greater than the passive condition (ANOVA, p = 0.002).



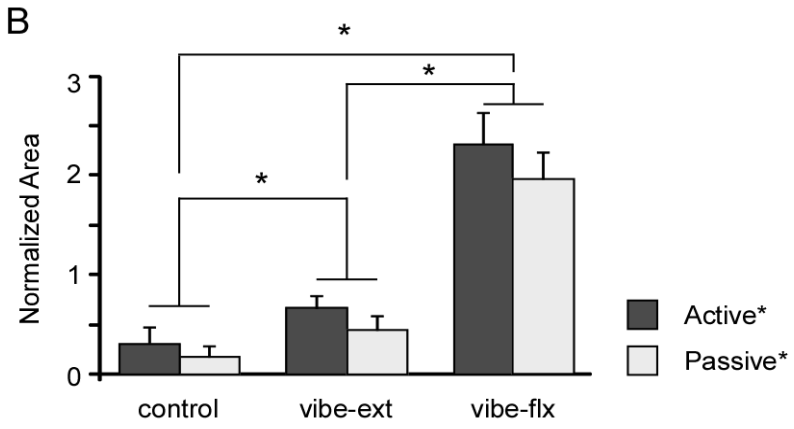


Figure 3-6 Ankle EMG Modulation. (A) Example ankle EMG data from subject S3 during passive hip oscillations with Achilles tendon vibration. (B) Average MG EMG area comparing control, vibe-ext, and vibe-flx between active and passive tests. Asterisks represent significant differences between active and passive, and between test conditions ($p < 0.05$).

3.4 Discussion

In summary, the results from this study demonstrate that hip-mediated sensory signals strongly modulate activity in distal muscles of the leg and appear to play a primary role in the generation of multijoint reflexes of the leg. Even though reflex responses to sensory stimuli applied at the knee or ankle were observed local to the knee or ankle, the movement direction of the hip had an effect on those responses. The reflex response amplitude to tendon tap perturbations measured from the VM muscle was dependent on the timing of the perturbation with respect to the hip's motion, showing greater reflex amplitude when the tendon tap perturbation was applied during hip extension than hip flexion. Moreover, TVR responses in the ankle also showed a hip movement dependency. Applying the vibratory stimulus when the hip was flexing

increased the MG EMG activity to a greater extent than when vibration was applied during hip extension. The predominance of hip afferents in the generation of multijoint reflex responses was also supported by the absence of a modulatory effect of tendon tap and plantarflexor vibration on the ongoing multijoint response to hip oscillation. Specifically, the results from this study showed reflex activity at the hip joint was weakly modulated by Ia afferent feedback from the knee (tendon tap) or ankle (vibration). MH EMG did show increased activity that was hip movement dependent, but no significant differences were seen in the hip torque responses between the timing of the sensory perturbations with hip position.

3.4.1 Hip Afferent Feedback and Reflex Modulation

The results from this study are consistent with the idea that stretch-related hip afferent feedback plays a unique role in the modulation of spastic muscle activity throughout the leg. Clinical reports on SCI patients from as early as the 1940s had first implicated the importance of the hip as a trigger for initiating multijoint reflexes (Macht and Kuhn 1948). Controlled experimental studies have since supported the clinical observations by demonstrating that characteristic multijoint reflex patterns heavily depend on the hip's position (Onushko and Schmit 2007; Schmit et al. 2000; Schmit and Benz 2002; Steldt and Schmit 2004). In previous studies investigating sensory triggers of multijoint reflexes, rapid knee extension elicited similar reflex patterns as those produced from stretching the hip, but responses were substantially enhanced if the hip was in an extended posture rather than flexed (Wu et al. 2005). Ankle load afferents have also been

shown to elicit multijoint reflexes resembling the extensor reflex pattern (Wu and Schmit 2006). Ankle (plantarflexor) load release produces a multijoint reflex response similar to hip extension, but is rarely triggered with the hip in a flexed posture. Conversely, holding the leg in an extended posture while dorsiflexing the ankle caused increased excitability of hip flexor muscles (Wu and Schmit 2006). The results observed in the current study showed that hip position provided strong control signals that modulated responses at the ankle and knee during Achilles tendon vibration and patellar tendon taps, respectively. However, neither sensory perturbation had a robust response at the hip, suggesting that stretch-related Ia feedback from distal leg muscles do not strongly modulate hip activity in SCI.

Applying vibration at the Achilles tendon elicited a tonic vibratory response in ankle plantarflexor muscles. Vibration applied to muscle or tendon strongly activates Ia afferent firing of the homonymous muscle (Agarwal and Gottlieb 1980; Burke et al. 1976; Roll et al. 1989). In the current study, muscle activation from the vibration appeared to a have a tonic component with it that varied with the movement direction of the hip. At the onset of vibration, there was a phasic burst of EMG activity that was present in both the tap-flx and tap-ext conditions (refer to Figure 3-6). A difference in the tonic component of the MG EMG was seen between the conditions, in which activity remained elevated compared to the tap-ext condition (refer to Figure 3-6). In SCI, the tonic component of the vibration reflex was previously shown to depend on input from segmental interneurons or supraspinal pathways, while the initial burst is dependent on motoneuronal excitability (Dimitrijevic et al. 1977). Results from the present study

support these findings, since it appeared that the tonic component of the vibration reflex was dependent on the hip's position.

Following cerebral or spinal lesions, there is impaired modulation over spinal inhibitory mechanisms. For example, it has been reported that hip-mediated sensory input modulates the soleus H-reflex in individuals with SCI, in which hip extension facilitates the H-reflex and hip flexion provides inhibition (Knikou and Rymer 2002; Knikou et al. 2006). Additionally, several studies have reported that people with spasticity have altered reciprocal inhibition (Crone et al. 1994, 2003; Xia and Rymer 2005). In normal control of movement, reciprocal Ia inhibition from the agonist muscle inhibits activity in the antagonist muscle at the onset of and during movements (Tanaka 1974; Yanagisawa 1976; Crone et al. 1987), but in spastic patients the pathway transmission is decreased, resulting in facilitation of the antagonist muscle (Crone et al. 1994, 2003; Xia and Rymer 2005). Consistent with previous findings, the hamstrings muscle activity observed in the current study showed a similar behavior. Patellar tendon taps delivered when the hamstrings muscles were being stretched (i.e. tap-flx condition) compared to the other conditions (i.e. control or tap-ext) resulted in increased MH EMG activity in both the active and passive cases. In contrast, an interesting observation from the current study was that reflexive hamstrings muscle activity could be suppressed (inhibited) by rotation of the hip into extension (refer to Figure 3-2). One explanation for this is that activation of the quadriceps muscles through patellar tendon taps produces too weak of inhibitory feedback to modulate the antagonist muscle (i.e. hamstrings) and other hip flexor muscles, such as the iliopsoas, may provide stronger inhibitory cues that modulate hamstrings muscles.

3.4.2 Organization of Multijoint Reflexes and Locomotor Pathways

Sensory feedback associated with changes in hip position and limb loading greatly contribute to reflex modulation during locomotion. Hip proprioceptors are important for initiating the transition from stance to swing during treadmill walking in spinalized cats (Grillner and Rossignol 1978). In addition to hip proprioception, afferent input from ankle load receptors also regulates the stance-to-swing transition. During locomotion, keeping the limb loaded prolongs the stance phase and prevents the limb from advancing (Conway et al. 1987; Duysens and Pearson 1980; Whelan et al. 1995). Furthermore, a similar behavior of hip proprioception and ankle load afferent integration has been established in human infants (Pang and Yang 2000) corroborating results from animal work that spinal cord circuitry can modify locomotor patterns under altered states of supraspinal influences.

Similar patterns of coordinated muscle activity have been observed in SCI patients during assisted treadmill walking. Bringing the hip into extension augments limb flexion through stretch-sensitive afferent input during body weight-supported treadmill walking (Dietz et al. 2002). Additionally, limb loading is a critical sensory cue to produce appropriate muscle activity patterns in SCI patients during treadmill walking (Harkema et al. 1997). It has been postulated that the pathways involved with multijoint reflexes observed during imposed hip oscillations share common pathways with spinal circuits for locomotion, since coordinated reflex patterns are strongly tied to hip sensory signals (Onushko and Schmit 2007; Schmit and Benz 2002; Steldt and Schmit 2004; Wu et al. 2005; Wu and Schmit 2006, 2010). However, integration of hip and ankle proprioceptive sensory feedback with multijoint reflexes in human SCI is less clear. Commonly in

spastic patients, extension of the leg causes increased ankle plantarflexion activity (Schmit and Benz 2002; Steldt and Schmit 2004), which was also noted in the current study. Furthermore, from the current study we saw that even though the plantarflexors turned on with vibration, plantarflexion was increased to a greater extent during hip flexion than movement of the leg into hip extension. If the interneuronal pathways of these reflexes overlap with reflex pathways involved with locomotion, then we'd expect the ankle to have a greater response to vibration during hip extension to be appropriately timed with push-off at the end of stance. However, hip flexion can be enhanced in SCI by eliminating dorsiflexion torque at the ankle following unilateral hip extension movement, which is analogous to limb unloading during the stance phase of gait (Wu and Schmit 2006). Similarly, the hip extension moment can be augmented if ankle loading is applied during the stance phase during body weight-supported treadmill stepping (Gordon et al. 2009). The differences observed in this study compared to other studies could be due to differences in the type of afferent feedback information. During treadmill walking, loading of the limb generates Golgi Ib input from the tendons, which is a strong modulator of hip flexor activity (Conway et al. 1987; Pang and Yang 2000), while in the current study, tendon vibration primarily increased muscle Ia afferent firing (Burke et al. 1976; Roll et al. 1989).

Hip proprioceptive input has been shown to be a critical sensory signal for tuning rhythmic locomotor behaviors, such as walking, in the mammalian spinal cord. Central pattern generators (CPGs) distributed within the lumbar cord are organized in an functionally appropriate manner. In the segments where hip motoneurons lie, hip movement entrains rhythm-generating networks of the knee and ankle (Kiehn 2006).

Furthermore, destroying or disconnecting these networks from lower spinal regions severely impairs locomotor capacity in rats and cats (Langlet et al. 2005; Magnuson et al. 2005). While the organization of rhythm-generating neuronal networks in the human spinal cord are largely unknown, the results from the current study do allude to the importance of hip sensory feedback for generating rhythmic, coordinated activity in the lower extremities.

3.4.3 Functional Considerations

A common phenomenon observed in patients with spasticity is reflex coupling of hip flexor and knee extensor muscle activity, which is termed 'spastic stiff-legged gait'. During late stance, when the hip is extended, exaggerated knee extension activity often occurs with hip flexor activity. Abnormal coupling of the knee extensor muscles impairs gait in these individuals by decreasing the knee flexion velocity at toe-off (Goldberg et al. 2003; Lewek et al. 2007). In SCI and stroke, extension of the hip produces prolonged reflex activity in the hip flexors and knee extensors under passive movements (Schmit and Benz 2002; Steldt and Schmit 2004) and during volitional movements (Lewek et al. 2006, 2007). In the current study, tendon reflex amplitudes of the VM and RF were larger when patellar tendon perturbations were delivered while the leg was moving into hip extension. Furthermore, the VM reflex responses were dominated by hip movements (increased reflex amplitude on 2nd tap compared to 1st tap, refer to Figure 3-5). Even though the VM muscle does not receive stretch-related input from hip movement, the initial stretch of the VM from the 1st tap may have increased excitability in that MN pool, resulting in a larger response to the 2nd tap. Alternatively, stretch-related input from the

hip may increase excitability in quadriceps motor pool increasing reflex activity at the knee. The data suggest that the integration of group I afferent input from the hip and knee are poorly modulated in spastic SCI, possibly due to decreased control over presynaptic inhibition of Ia afferents (Faist et al. 1999). However, group II hip afferents are also thought to partly account for the increased knee extensor excitability in spastic gait as seen in stroke patients (Lewek et al. 2007).

Rehabilitation efforts commonly employed in the clinic to improve locomotor function in SCI patients have a large focus on managing inappropriate reflex activity through pharmacological agents. Anti-spastic medication, such as Baclofen, is commonly prescribed to reduce reflex activity in spastic patients. Considering the results from the present study, reflex excitability is strongly modulated by hip-mediated signals. Thus, treatment of SCI patients using antispatic agents might impair the reflex regulation of gait and could have detrimental effects on function.

CHAPTER 4

Bilateral Oscillatory Hip Movements Induce Windup of Multijoint Lower Extremity Spastic Reflexes in Chronic Spinal Cord Injury

4.1 INTRODUCTION

Involuntary muscle spasms are a prominent component of the spastic syndrome that develops in chronic human spinal cord injury (SCI). Individuals with SCI will typically present with uncontrolled hyperexcitable reflexes, such as flexor and extensor spasms, in which brief stimuli can evoke forceful, prolonged responses throughout muscles in the lower extremities lasting from a few seconds to tens of seconds (Benz et al. 2005). Particular to extensor spasms, hip proprioceptive stimuli elicit long-lasting, coordinated responses of hip flexion, knee extension and ankle plantarflexion (Schmit and Benz 2002), and typically occur when patients shift from a seated to supine position (Macht and Kuhn 1948) or by extension of the leg (Schmit and Benz 2002). Although not recognized as part of the extensor spasm, from previous studies, flexing the leg will also cause a coordinated response generally composing hip extension, knee flexion and ankle extension with responses equally as long as seen in extensor spasms (Schmit and Benz 2002). Extensor spasms, which are common in SCI patients, are functionally relevant since they not only interfere with patient mobility but also cause pain and discomfort (Little et al. 1989). Accordingly, it is important to understand the underlying mechanisms associated with these prolonged responses in order to prescribe appropriate treatment options to improve function.

The neuropathophysiology behind the long-lasting nature of involuntary muscle spasms in human SCI still remains unclear, but one hypothesis is that changes in the intrinsic electrical properties of spinal circuitry contributes to the spastic syndrome (Bennett et al. 2004; Nielsen et al. 2007). In an intact nervous system, descending

monoaminergic input from the brainstem to the spinal cord modulates spinal neuronal excitability through the facilitation of persistent inward currents (PICs) found in both motoneurons (MNs) (Hounsgaard et al. 1988; Lee and Heckman 1999) and motor-related interneurons (INs) (Chen et al. 2001; Theiss et al. 2007). In mammals, it is believed that PICs are primarily generated by dendritic voltage-dependent Na^{+} and Ca^{2+} channels, that when activated by a brief excitatory stimulus, can amplify synaptic currents (Hultborn et al. 2003; Lee and Heckman 2000) and produce plateau potentials resulting in sustained firing even after the stimulus is removed (Bennett et al. 1998a; Crone et al. 1988; Schwindt and Crill 1980). PICs are associated with a phenomenon called "windup" in which repeated excitatory inputs can facilitate MN activity without increases in synaptic input (Bennett et al. 1998a; Russo and Hounsgaard 1994; Svirskis and Hounsgaard 1997).

Immediately following spinal injury in animals, MN excitability is greatly decreased with concomitant loss of PICs (Bennett et al. 1999; Hounsgaard et al. 1988; Hyngstrom et al. 2008b) and this has been associated with an acute interruption of monoaminergic inputs to the cord. Despite the loss of descending drive, a re-emergence of PICs occurs weeks to months following injury along with a concurrent increase in MN excitability and emergence of spasms (Bennett et al. 2001b; Li and Bennett 2003). Constitutive activity in serotonergic (Murray et al. 2010) and adrenergic (Rank et al. 2010) receptors are, in part, responsible for PIC recovery and the abnormal regulation of recovered PICs is likely to be involved in the long-lasting muscle spasms observed in chronic spinalized animals (Bennett et al. 2001b; Bennett et al. 2004; Li et al. 2004; Murray et al. 2010). These changes in MN properties in spinal-injured animals are consistent with clinical descriptions of involuntary spasms in human SCI (Collins et al.

2001; Gorassini et al. 2004; Hornby et al. 2006; Hornby et al. 2003; Nickolls et al. 2004). Alterations in intrinsic properties of INs, which have not been investigated as extensively as MNs, also demonstrate electrophysiological characteristics similar to PICs in MNs (Russo and Hounsgaard 1996; Smith and Perrier 2006; Theiss et al. 2007; Ziskind-Conhaim et al. 2008), including windup (Russo and Hounsgaard 1994). Since MNs receive much of their synaptic input from INs and IN axons show the ability to regenerate post SCI (Fenrich and Rose 2009), it is plausible that changes in IN PICs would have downstream effects on motor behaviors as well, especially since the spastic syndrome is in large part comprised of long-lasting, multijoint flexor and extensor spasms. Thus, after spinal injury, changes in the excitability of interneuronal networks may affect reflex muscle activity and the associated emergence of long-lasting motor output (Jankowska and Hammer 2002).

In human SCI, windup has been documented with repeated stretch of the ankle (Hornby et al. 2006) and in flexor reflex responses to repeated, brief stimulation of the skin of the foot (Hornby et al. 2003), implicating a role for PICs in the clinical symptoms of clonus and flexor spasms. However, windup behavior has not been explored in extensor spasms and it is plausible that underlying plateau potentials may play a large role in the prolonged, non-linear responses associated with this spastic reflex. In the current study we investigated whether the multijoint reflexes associated with imposed hip movement in human SCI (i.e. extensor spasms (Schmit and Benz 2002)) would exhibit windup. We hypothesized that windup responses in joint torque and muscle activity would occur throughout the lower extremities. Specifically, peak torque and muscle

activity acquired through surface electromyograms (EMGs) would be greater with subsequent hip movements in a non-linear manner, implicating PICs in extensor spasms.

Results from this study have been published in abstract form (Onushko et al. 2008).

4.2 METHODS

4.2.1 Study Participants

Participants included ten individuals with chronic (> 6 months), clinically complete (American Spinal Injury Association [ASIA] classification A) and incomplete SCI (ASIA B, C or D). During the time of this study, three subjects were prescribed anti-spastic medication (e.g. baclofen, a GABA agonist) to control the frequency and intensity of their spasms. Subject information is summarized in Table 4-1. Exclusion criteria for this study included: lower extremity nerve injury or injury below spinal cord segments innervating the hip region, significant medical complications due to skin breakdown, urinary tract infection, heterotopic calcification, significant osteoporosis, other concurrent illnesses limiting the capacity to conform to study requirements, or the inability to give informed consent. Additionally, five subjects with no reported neurological injury volunteered for this study as control subjects. Informed consent was obtained prior to study participation and all procedures were conducted in accordance with the Helsinki Declaration of 1975 and approved by the Institutional Review Board of Marquette University.

Table 4-1. Subject Clinical Characteristics

Subject	Age, yr	Injury Level	ASIA Score	Post Injury Duration, yr	Medication(s)
A	64	C8-T1	A	6	Baclofen
B	48	C6-7	A	24	None
C	42	C5	B	27	None
D	23	T8	B	6	Ditropan
E	41	C5-6	C	25	None
F	40	C5-6	C	23	None
G	70	T2	C	3	Baclofen, Neurontin, Ditropan, Imipramine, Sanctura, Botox
H	43	C5-6	C	17	Baclofen, Detrol, Darvocet
I	26	C6	D	4	Ditropan XL
J	48	C6-7	D	23	None

Injury Level (column three): C = Cervical spinal level; T = thoracic spinal level

4.2.2 Experimental Setup

Bilateral hip oscillations were imposed on the study participants using the apparatus described in Chapter 2 (refer to Figure 2-1A). Briefly, the system utilized two servomotor drive systems (Kollmorgen, Northhampton, MA) that were controlled through custom-developed LabVIEW software (National Instruments Corp., Austin, TX), and custom-built knee-ankle braces that were fastened to the servomotor apparatus. The custom-built knee-ankle braces were adjustable to accommodate a wide range of leg lengths. The legs were supported in leg braces by a strap securing the thigh, a strap securing the heel and a clamp over the dorsum of the foot to secure the foot to the plate at the end of the leg brace. The system was equipped with reaction torque transducers (S. Himmelstein and Company, Hoffman Estates, IL) that aligned with the hip, knee and ankle anatomical joint axes of rotation for each leg. The hip angle was measured using optical encoders (US Digital, Vancouver, WA) that were affixed to its respective motor shaft.

Disposable, pre-gelled Ag/AgCl surface electrodes (Vermed Medical Inc., Bellows Falls, VT) were used to measure electromyograms (EMG) of the participant's legs. Electrodes were placed in a bipolar arrangement over the cleaned muscle belly of the following muscles: vastus medialis (VM) and lateralis (VL), rectus femoris (RF), medial hamstrings (MH), medial gastrocnemius (MG), soleus (Sol) and tibialis anterior (TA) muscles of both legs. EMG signals were amplified ($\times$ 1,000), band-pass filtered (10-1,000 Hz) (Bortec Medical AMT-16; Calgary, Alberta, Canada) and sampled (1,000 Hz) using the same data acquisition card (National Instruments Corp., Austin, TX) and PC used to acquire the torque/position signals.

4.2.3 Experimental Protocol

Multijoint reflexes were elicited through controlled sinusoidal hip oscillations imposed by the robotic apparatus while subjects were instructed to remain relaxed. Hip oscillations were repeated for ten consecutive cycles to elicit windup of reflex responses. The range of hip motion was approximately 50° for all tests (40° of hip flexion to 0-10° of hip extension; hip extension was dependent on the subject's range of motion) (refer to Figure 2-1B in Chapter 2). Furthermore, we investigated whether the facilitation of multijoint spastic reflexes would be altered by varying stretch-related synaptic input by oscillating the legs about the hip joint bilaterally (bilateral 180° out of phase (OUT), bilateral in phase (INP)) and unilaterally (unilateral leg movements with the contralateral leg (i.e. left leg) held fixed either at end range hip flexion (FLEXED) or at end range hip extension (EXTENDED)). Hip oscillations were performed at two movement frequencies (0.50 and 0.75 Hz) to test for velocity dependence. Tests were presented in random order

and were repeated three times for each condition (total of 24 tests), with two to five minutes allowed between tests to minimize adaptation of the reflexes. At the completion of the experiment, two additional hip movements were performed to estimate the torque due to gravity, passive joint resistance and inertial properties of the participant's legs. The torque contribution from gravity and passive joint resistance was approximated by slowly moving each leg through the entire range of motion at 2 °/s in 5° increments, pausing for five seconds between each increment. The inertial properties of the leg and leg brace were estimated by rapidly oscillating (1.5 Hz) each leg from 25° to 10° of hip flexion for ten cycles. Oscillating the hip within midrange minimized the potential for eliciting reflexes. For all tests, EMG and torque data were acquired for the duration of the movements.

4.2.4 Data Analysis

Active muscle torques for the hip, knee and ankle joints were corrected for the biomechanical properties of the leg and leg brace using a method previously described (Onushko and Schmit 2007). All torque data were low-passed filtered (5 Hz) using a 2nd order Butterworth filter (*filtfilt* function in Matlab; The Mathworks, Inc., Natick, MA). The torque due to gravity and passive resistance were estimated by fitting a 3rd order polynomial curve to the torque measurements taken during the slow, incremental hip movements, and then those polynomial coefficients were used to calculate the gravitational and passive resistance torques from the measured trial data (i.e. INP, OUT, etc. hip oscillations). The inertial properties of the leg and leg brace were calculated by subtracting the gravitational/passive resistance torque, and then estimating the inertial

constant of the leg and leg brace using a linear regression analysis. Additionally, a mechanical artifact was present within the system that was not associated with biomechanical properties of the legs. The artifact was estimated using an ensemble average of torque measurements recorded from neurologically healthy subjects who completed the study in its entirety (using the said method to subtract the inertial and gravity/passive torque components prior to ensemble averaging). Since the healthy control subjects do not experience responses to the imposed hip oscillations, the ensemble torque provided a good estimate of the artifact (on average < 5Nm). The active muscle torque (i.e. torque produced by active muscle contraction) was calculated by subtracting the gravitational/passive torque, inertial torque, and artifact from the measured trial torque data, and was used for all subsequent analyses.

To identify windup of the multijoint reflex responses to the movements, peak flexion and extension torques from the hip, knee and ankle joints were found for the first seven consecutive hip oscillations. Torque data were separated into flexion and extension components before identifying the peak torque per cycle. The peak torque data per cycle were normalized to the mean of the seven cycles for each subject and then averaged across all subjects. The data were normalized to account for differences in the amplitude of the spastic reflex activity among the subjects. Differences in the peak torques from repeated hip oscillations were statistically compared using a multifactor ANOVA (main factors: stretch number (cycles 1-7), frequency (0.50 and 0.75 Hz) and movement type (INP, OUT, FLEXED, and EXTENDED); random factor: subject) and Bonferroni correction to determine differences among successive stretches ($\alpha = 0.05$).

Windup was identified within the EMG data using a similar technique. EMG data were first notch filtered (59-61 Hz) to remove line noise and then band-pass filtered (10-300 Hz) using 4th-order Butterworth filters (Matlab; The Mathworks, Inc.). For analysis, the root-mean square (RMS) of the filtered EMG data was calculated using a 100-ms sliding window. The peak EMG response was found per cycle for the first seven successive hip oscillations during the time when the muscle was active for the movement cycle. EMG data for a single trial of any particular muscle were excluded if the peak EMG did not exhibit suprathreshold activity for at least two consecutive movement cycles (threshold = mean of baseline + 3 SD). Differences in the peak torques from repeated hip oscillations were statistically compared using a multifactor ANOVA (main factors: stretch number, frequency and movement type; random factor: subject). Bonferroni correction was used for post hoc comparisons between individual stretches. The level of significance was set at $\alpha = 0.05$ for all tests.

4.3 RESULTS

In general, a non-linear increase was observed in the torque and EMG recordings, followed by either saturation or depression in the response. Figure 4-1 illustrates an example of multijoint windup from a single subject. Hip flexor and knee extensor torques increased from the first movement of the leg into hip extension and the amplitude of the response to each subsequent stretch exceeded the previous response through the third stretch, which then modulated to the same or lower level for the remainder of the hip oscillations (Fig. 4-1A). Muscle activity from the RF and VM followed a similar trend as

the torque data, increasing in amplitude during the initial two stretches. Although RF and VM activity slightly decreased following the fifth stretch, EMG activity modulated at a heightened level compared to the first stretch for the duration of the movements (Fig. 4-1A). Hip extensor and knee flexor torques successively increased in amplitude for the first two stretches of the leg into hip flexion and remained elevated for the duration of the trial (Fig. 4-1B). Activity at the ankle joint increased with repeated hip perturbations as well (Fig. 4-1C), although reflex responses in the ankle were more variable than responses observed at the other joints. MG and Sol activity generally remained above baseline, increasing in activity by the second rotation of the leg into hip extension, but little modulation was observed during the hip oscillations. Additionally, TA EMG increased above baseline level in response to successive stretches of the hip, but it was modulated by the hip movements, with peak TA EMG coinciding with hip extension. Knee (VM) and ankle (Sol,) muscles (i.e. muscles not crossing the hip joint) exhibited a delay in activation in response to stretches of the hip, in which reflex activity was not elicited until after the 2nd or 3rd hip oscillation in this subject (refer to Fig. 4-1A and C).

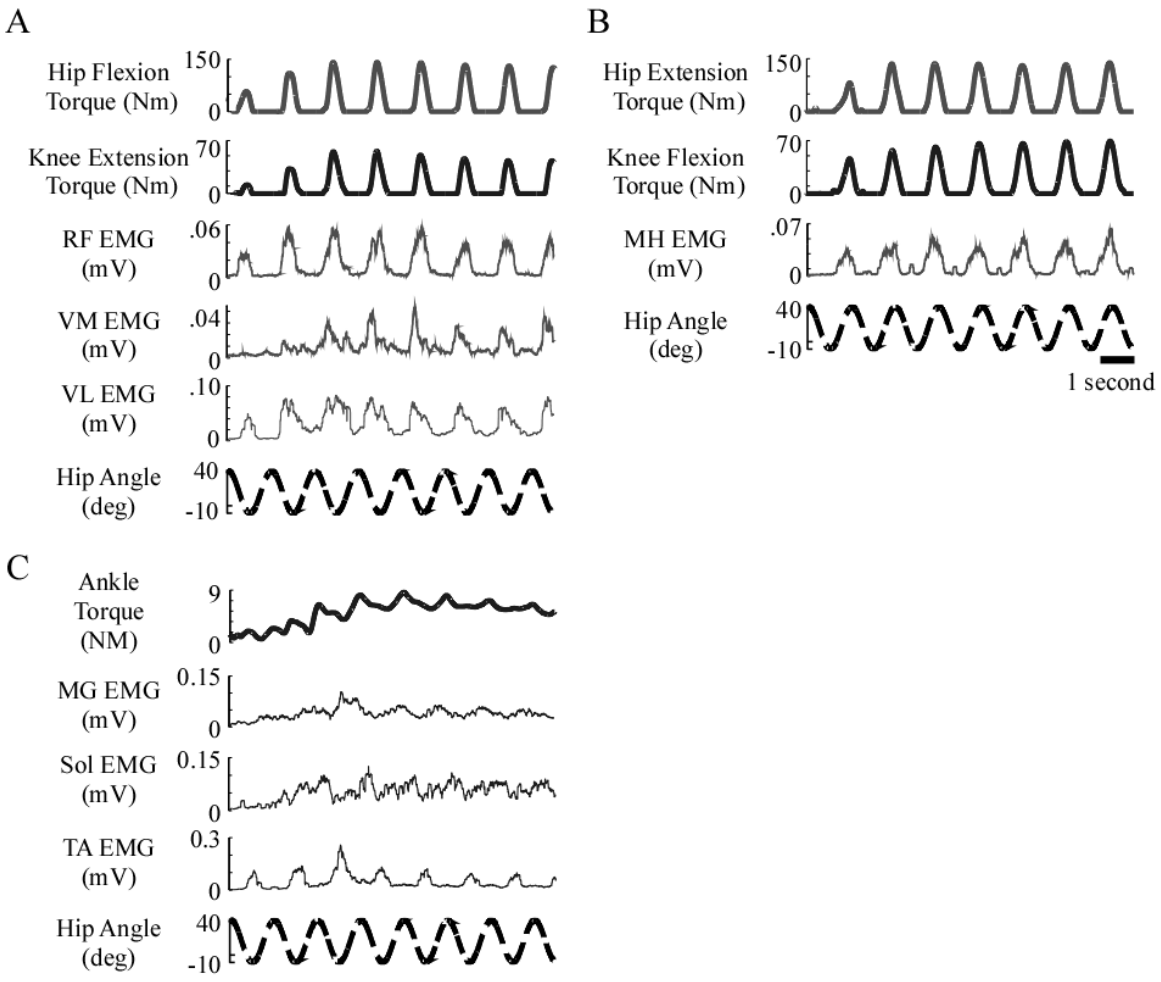


Figure 4-1 Single Subject Example of Multijoint Windup. Example of multijoint reflex responses (right leg only) from a SCI study participant (subject E) during the out of phase hip oscillations at 0.75 Hz for the first 7 cycles. (A) Windup of hip flexors/knee extensor torque (half-wave rectified) and RMS EMG data. (B) Hip extensor/knee flexor torque (half-wave rectified) and RMS EMG data during reflex responses. (C) Reflex activity of ankle dorsiflexor/plantarflexor torque and EMG data. Increasing positive torque values represents plantarflexion and decreasing positive torque values represents dorsiflexion.

The effects at the hip joint from repeatedly moving the hips are illustrated in Figure 4-2 for all movement conditions across all subjects. There were significant differences among the peak hip torque data with subsequent hip extensor stretches (hip flexion torque: ANOVA, $P < 0.001$), and hip flexor stretches (hip extension torque:

ANOVA, $P < 0.001$). Additionally, hip extension torque typically remained elevated after the 3rd or 4th stretch, whereas hip flexion torque typically decreased after the 4th stretch. Although more variability was observed within the EMG data, peak RF and MH activity also showed significant differences with subsequent stretches (RF: ANOVA, $P < 0.01$; ANOVA, MH: $P < 0.001$). Except for the OUT condition, peak RF EMG increased through the first two stretches, and peak MH EMG increased until the second stretch, but generally remained elevated through subsequent stretches (refer to Figure 4-2).

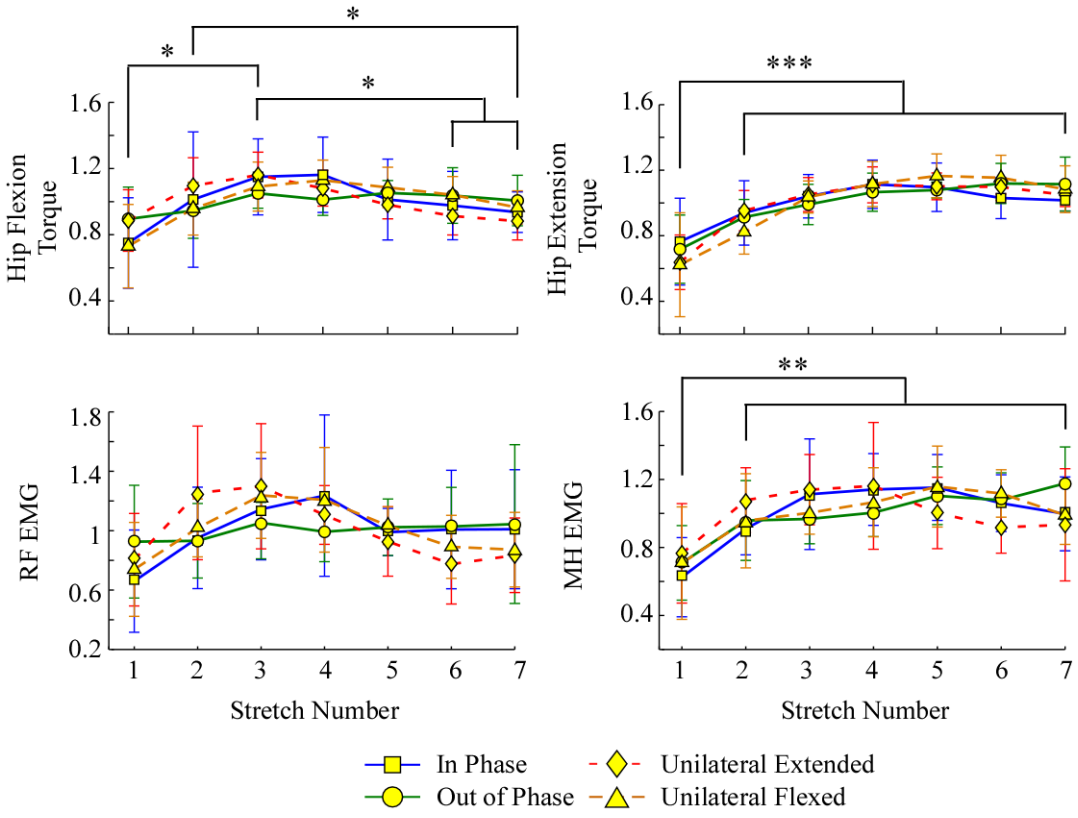


Figure 4-2 Windup of Hip Torque and EMG. Average peak hip torque (top panels) and peak RF and MH RMS EMG (bottom panels) for each of the movement types for the 0.75 Hz movement frequency (right leg data only). Peak data were normalized to the mean of each subject's data. Significant differences ($P < 0.05$) between individual responses to stretches are indicated by asterisks (* $P<0.05$; ** $P<0.01$; *** $P<0.001$). There were no statistically significant differences among the movement types ($P > 0.05$).

Reflex responses were also observed at the knee joint in response to repeated hip movements. Normalized peak knee torque responses showed significant differences with subsequent hip movements (knee extension and flexion torques: ANOVA, $P < 0.001$) (Figure 4-3). Knee extension torque increased through the first three consecutive joint rotations during the OUT, INP and FLEXED hip movements, and in general, the reflex amplitude was slightly elevated during the subsequent stretches. Knee extension torque during the EXTENDED movement condition elicited a slightly different pattern, where the activity rapidly increased during the second hip perturbation and then gradually declined throughout the remaining hip movements. Because RF is a biarticular muscle that acts at the hip and knee joint, it is possible that knee extensor torques are a result of RF stretch-related activity; however, since VM also demonstrated significant increased activity through the repeated hip oscillations (Bonferroni, 1st response vs. 4th response, $P = 0.002$), RF may have only partially contributed to knee extension torques in some cases. Significant increases in the group mean peak VL EMG activity were not observed (ANOVA, $P = 0.456$). A continuing increase in knee flexion torque (refer to Fig. 3, top right) occurred in response to repeated hip extensor stretches and followed a similar pattern as the responses observed in hip extension torques (refer to Figure 4-2, top right), which might be accounted for by MH activity.

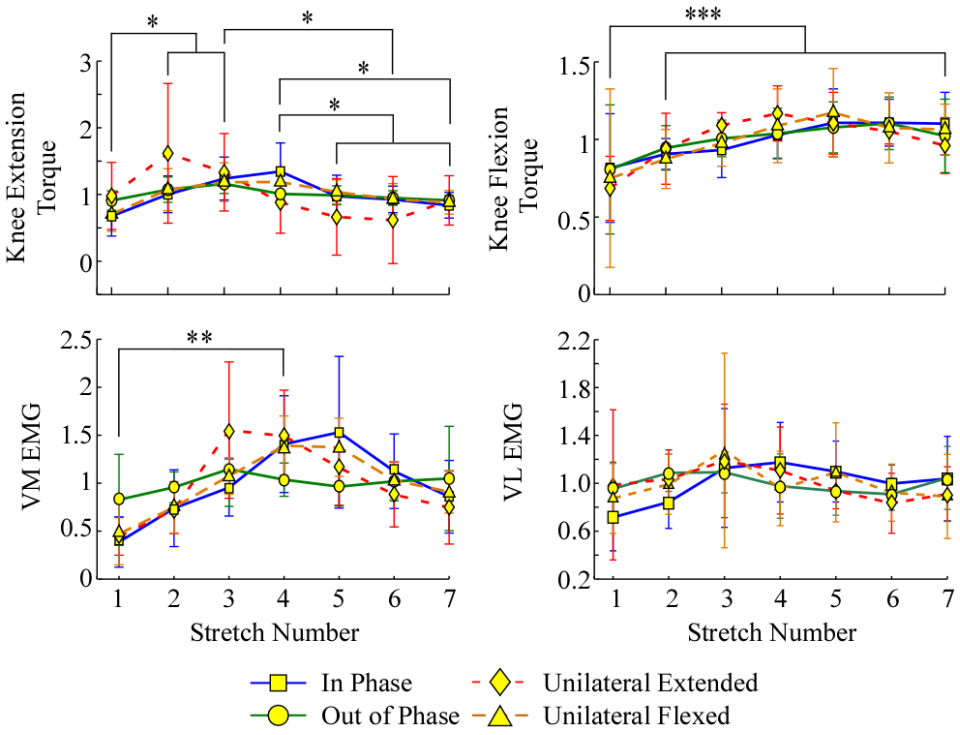


Figure 4-3 Windup of Knee Torque and EMG. Average peak knee torque (top panels) and peak VM and VL RMS EMG (bottom panels) for each of the movement types for the 0.75 Hz movement frequency. Peak data were normalized to the mean of each subject's data set. Significant differences indicated by asterisks (* P<0.05; ** P<0.01; *** P<0.001) between individual responses to stretches (P < 0.05). There were no statistically significant differences among the movement types (P > 0.05).

Increased activity at the ankle was also noted during repeated stretches at the hip. Figure 4 illustrates normalized group mean ankle torque and EMG responses. Significant increases in plantarflexion torque (ANOVA, P < 0.001) were observed during the repeated stretches. Due to higher variability, the increased dorsiflexion torque during the initial two stretches was not significant (Bonferroni, P = 1.000). More notably though, responses in dorsiflexion torque began to decrease following the second hip oscillation (Bonferroni, 1st response vs. 5th through 7th response, P < 0.045), while plantarflexor

torque continually increased (Bonferroni, 1st response vs. 2nd through 7th response, P < 0.001). MG and Sol EMG activity did not fully resemble the pattern of plantarflexion torque, likely as a result of greater variability within the EMG data (Fig. 4-4A). TA activity followed a similar pattern to the dorsiflexion torque, with an increase in activity over the course of the first few consecutive hip oscillations followed by a depression in reflex activity (Fig. 4-4B).

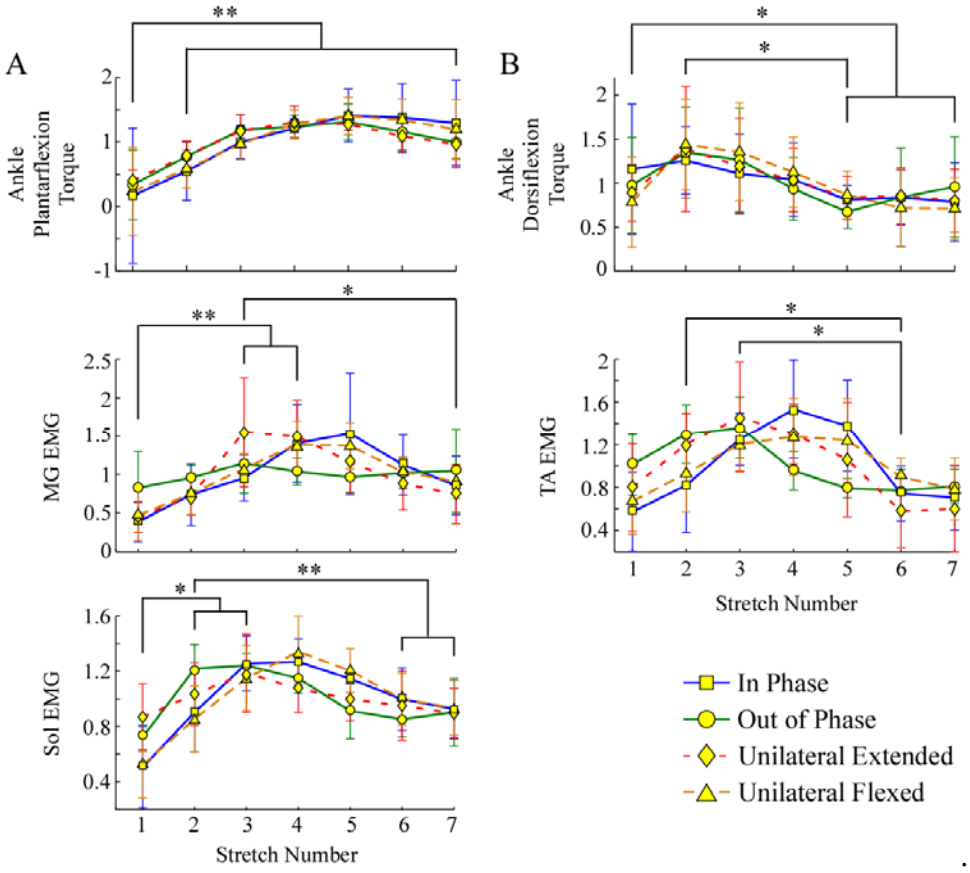


Figure 4-4 Windup of Ankle Torque and EMG. Average peak torque and RMS EMG from the (A) ankle plantarflexor and (B) ankle dorsiflexor data for each of the movement types for the 0.75 Hz movement frequency. Peak data were normalized to the mean of each subject's data set. Significant differences indicated by asterisks (* P<0.05; ** P<0.01; *** P<0.001) between individual responses to stretches (P < 0.05). There was no statistically significant difference among the movement types (P > 0.05).

The type of hip movement did not significantly affect windup amplitude during repeated hip oscillations (torque: $P = 0.558$; EMG: $P = 0.999$). However, out of phase hip movements tended to elicit greater responses (for VM, VL, RF, MG, and TA EMG) during the first stretch in comparison to the other movement types, and the peak EMG responses were smaller over the first few stretches during the in phase hip movements compared to the other movement conditions, which was more noticeable in hip flexor/knee extensor responses than hip extensor/knee flexor responses. Further, compared to the bilateral hip movements, responses during unilateral hip perturbations tended to increase more rapidly during the initial hip oscillations. Multijoint reflex windup was not sensitive to the movement frequency, with no significant effects on the amplitude of the joint torques (ANOVA, $P > 0.565$) or EMG activity (ANOVA, $P > 0.999$).

Increased muscle activity was also observed in the non-moving leg of 3 SCI subjects (E, F, and J) during the unilateral tests. Figure 4.5 shows sample RMS EMG and torque data of the non-moving leg from subject J during the EXTENDED (panel A) and FLEXED (panel B) movement conditions. The muscle activity pattern in the non-moving leg resembled the windup seen in the oscillating leg, and was also observed in the recorded ankle muscles. Additionally, the hip, knee and ankle torque also exhibited an increase in amplitude with subsequent stretches of the contralateral (i.e. oscillating) leg (Fig. 4.5). Subjects E and F showed similar windup responses in the non-moving leg (data are not shown).

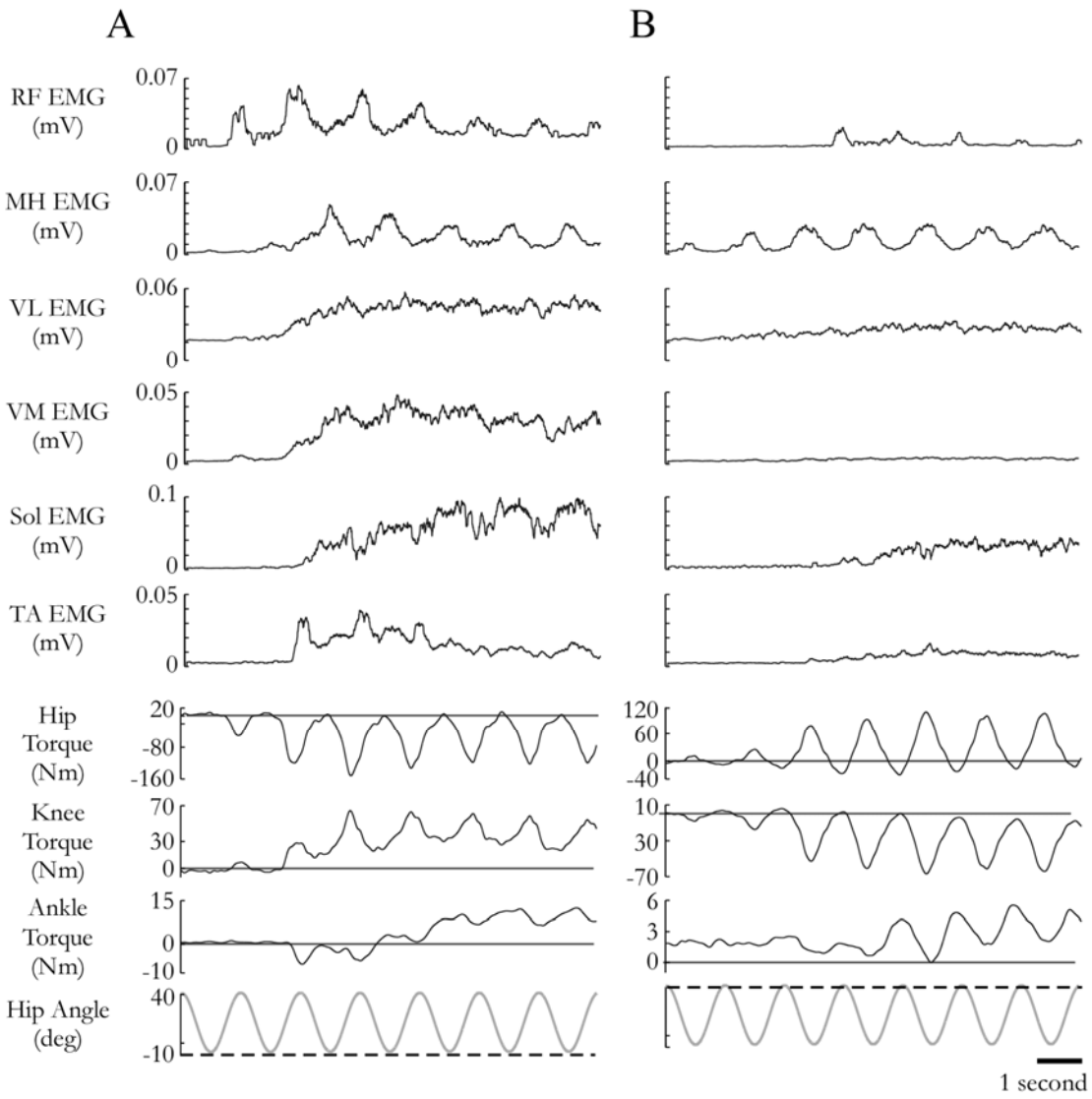


Figure 4-5 Single Subject Example of Windup in Non-Moving Leg. Example RMS EMG and torque data from the stationary leg (i.e. leg not being oscillated) from subject J during the unilateral tests for the 0.75 Hz movement frequency. (A) EXTEDNED test - the left leg was held at 10° hip extension (dotted line in bottom panel) while the right leg oscillated from 40° hip flexion to 10° hip extension. (B) FLEXED test - the left leg was held at 40° hip flexion (dotted line in bottom panel) during contralateral leg oscillations. For all torque traces, flexion torques are decreasing values and extension torques are increasing values.

4.4 DISCUSSION

In the present study, repeated stretches of hip musculature in subjects with chronic SCI induced motor responses consistent with windup not only in the primary muscles stretched, but also in muscles throughout the lower extremities. Subsequent stretching produced the windup response that was manifested as whole limb spasms of increasing EMG magnitude and joint torque. Although the spastic syndrome post SCI is likely multi-factorial, involving changes in both cellular intrinsic electrical properties and reflex pathway excitability (e.g. changes in reflex recruitment or alterations in pre-synaptic inhibition) (Nielsen et al. 2007), the temporal characteristics of multijoint reflex activation seen in this study were consistent with previous examples of stretch-reflex windup (Hornby et al. 2006) and windup of flexor reflexes (Hornby et al. 2003). In addition to being the first study to quantify windup in muscles not being stretched (VM, VL, MG, Sol, and TA), this study is the first to demonstrate in humans differences in temporal patterns of windup between functionally different muscle groups, suggesting a fundamental difference in intrinsic properties of neural pathways for muscles that have different general functions. The findings from this study further support a strong role of changes in the intrinsic electrical properties of motor-related circuitry in spastic reflex activity such as extensor spasms.

4.4.1 Possible Cellular Mechanisms of SCI Windup Response

Much of the current evidence regarding alterations in PIC regulation following injury to the nervous system has focused on MNs (Gorassini et al. 2004; McPherson et al.

2008; Mottram et al. 2009). The time course of windup responses measured during repeated hip oscillations in the current study was similar to windup observed in MNs exhibiting PICs (Bennett et al. 1998a, 1998b; Svirkis and Hounsgaard 1997). The stimulus interval is critical in generating windup responses to repeated stretches of the hind limb, as shown in the decerebrate cat MN, in which 3- to 6-second intervals generate a progressively larger amplitude response (Bennett et al. 1998a). Similar inter-stimulus intervals were used in the current study to elicit windup (0.66 and 1 second). MNs, the "final common pathway," make an attractive candidate for accounting for the windup response, but since the channels involved in windup are voltage sensitive, windup may also depend on interneuronal inputs to the MN.

Changes in the regulation of the intrinsic electrical properties of locomotor or stretch-related IN populations due to SCI could result in IN participation in the windup response. For example, locomotor-related INs have firing patterns that rely heavily on the regenerative intrinsic electrical properties of the cell (Kettunen et al. 2005; Kiehn 2006; Kyriakatos and El Manira 2007). Moreover, they receive and integrate stretch-related input from hip afferents (Edgley and Jankowska 1987; Harrison and Jankowska 1985; Kriellaars et al. 1994). In this study, stretching of the hip musculature by repeated hip movements, similar to a locomotor pattern (i.e. out of phase movements), could have provided an excitatory drive to locomotor-related INs leading to a windup response or entrainment of CPGs in the human spinal cord. Windup of INs is supported in the data from three subjects where windup was produced in the stationary leg (refer to Fig. 4-5). In addition, the delay in the muscle activity of the ankle could also suggest that subthreshold windup occurs in INs, since it has been shown that MNs need activation of

PICs to fire repetitively and exhibit windup (Bennett et al. 1998b; Lee and Heckman 2001). Repeated stretching of hip musculature may have induced subthreshold depolarizations that windup in the INs first, and then proceed to windup MNs of ankle muscles. Additionally, of the subjects who exhibited windup in muscles of the ankle, four subjects showed no EMG response during the first stretch of the hip (for the out of phase hip movement). While multijoint windup still may be occurring through windup of MNs and/or INs, n=4 isn't strong enough to rule out that other sensory pathways (e.g. cutaneous afferents) may be eliciting these responses in the other subjects. However, a recent study done in the spinalized cat revealed a broadening of the movement-related receptive fields of the MNs, in which stretch input from hip muscles generates large currents in knee and ankle extensor MN pools (Hyngstrom et al. 2008), which complements the findings from the current study.

The loss of group Ia IN activity could also indirectly contribute to windup of MNs (Nielsen et al. 2007). Although thought to be primarily ionotropic in nature, Ia inhibition appears to be important for modulating PIC amplitude and the loss of Ia IN activity has been shown to enable windup in MNs in a decerebrate preparation (Hyngstrom et al. 2007). In SCI and other neurological disorders, it has been recognized that Ia inhibitory pathways can be disrupted and or even lost (Nielsen et al. 2007). Loss of reciprocal inhibition in neurological conditions such as SCI would prevent stretch-related inhibition of PIC activity in MNs, thereby facilitating more PIC-related behaviors such as windup. However, the distribution of the monosynaptic Ia afferent and corresponding INs onto MNs is relatively focused (Eccles et al. 1957; Nichols 1999) and would not likely have resulted in the multi-segmental responses seen in the present study. For example, Ia input

would not be carried from stretch of the hip flexors onto muscles of the ankle, which SCI subjects demonstrated in this study (Figure 4-4).

Alternatively, recent evidence has shown that the hyperactivity observed in chronic spinal injury may be due to a lowered expression of the potassium-chloride cotransporter (KCC2) in MNs and spinal networks (Boulenguez et al. 2010). In the intact cord, KCC2 keeps the intracellular chloride ion concentration low; however, Boulenguez *et al.* (Boulenguez et al. 2010) have shown that KCC2 is decreased in chronic spinal-injured rats limiting chloride transport out of the cell which, compared to the intact spinal cord, results in a depolarizing shift in the equilibrium potential of chloride below the level of the lesion. The authors suggest that the shift to a more positive membrane potential could enable sensory inputs to produce long-lasting depolarizations which, in turn, activates PICs. If a similar mechanism occurs in human SCI, it is possible that while repeatedly moving the hips, stretch-related input may have been sufficient to initiate PICs in MNs or INs, and over the course of several hip movements, weaker sensory synaptic information could have initiated PICs in muscles not being stretched. However, the amount of non stretch-related input was not measured and it is unknown what contribution it had on the observed responses.

Finally, but not necessarily exclusively, Group II interneuronal pathways carry stretch-related information from the whole limb (Jankowska 1992, 2001) and have been implicated previously in the widening of a given MN's "movement receptive field" in acute spinal cord injury (Hyngstrom et al. 2008a). Group II INs receive convergent information from other afferent pathways (Jankowska and Hammer 2002) that could also contribute to windup. The multi-segmental stretch-related distribution of the windup

response in the current study, the delay in the windup response in muscles not being stretched, and the lack of velocity sensitivity in the magnitude of the response supports the role for the involvement of Group II IN in windup of human spastic reflexes. PIC behavior is necessarily regulated in the neurologically intact state, and thus it is likely that SCI disrupts control of PICs in *both* MNs and INs. Due to the indirect nature of the measurements of windup in this human study, we are limited in determining the relative contribution of each cell population (i.e. interneuron vs. motoneuron) to windup.

While PICs make a strong candidate for the windup observed during imposed hip movements in the current study, other mechanisms may also play a role in maintained muscle activity that was observed in SCI subjects. For example, after SCI there is increased concentration of extracellular glutamate that results in the upregulation of glutamate receptor expression (Liu et al. 1991), which could increase neuronal excitability through synaptic inputs. In addition, NMDA receptors have been shown to induce windup in dorsal horn neurons in response to nociceptive inputs (Mendell 1966, for review see Daw et al. 1993), and windup can be reduced by blocking NMDA receptors through the application of specific NMDA antagonists (Daw et al. 1993). While it is possible that in the current study, NMDA receptors could have contributed to the windup phenomena, NMDA currents have been reported to only last for up to 500 ms in spinal neurons (Dale and Grillner 1986) and previous studies in human SCI have shown that prolonged spastic reflex responses last several seconds after the end of the movement (Hornby et al. 2006), suggesting NMDA receptors play only a minor role in neuronal hyperexcitability in spastic reflex activity. Furthermore, exogenous application of NMDA after spinal cord transection in rats was shown to decrease rather than increase spinal

reflex sensitivity (Krenz and Weaver 1998), suggesting other mechanisms are likely to contribute to exaggerated reflex behaviors post-SCI.

4.4.2 The Functional Consequences of Uncontrolled PIC Behavior

The role of PICs in neurosystem behavior is still unclear, but evidence suggests that PICs are important for increasing the gain of the MN pool through amplification of synaptic input and bistable firing behavior of certain cell types. The modulation of PICs is multi-factorial, involving supraspinal centers and local inhibitory circuitry (Heckman et al. 2008). Loss of regulation of PICs, as may occur post SCI, could interfere with volitional control of movement by causing uncontrollable spasms or by causing large errors during repetitive movements that induce windup (e.g. during walking). From a rehabilitation standpoint, this would suggest that it might be beneficial to restore the regulation of PICs. Although, if stretch of the hip is entraining CPG pathways in human SCI, the additional excitability due to windup of these pathways may be useful for providing patients with limb propulsion. Future studies could investigate ways to facilitate inhibitory pathways either through electrical stimulation or pharmacology, with the caveat that PICs provide additional excitability needed to complete motor tasks such as transferring or walking (Dietz 2008). Importantly, methods that more clearly delineate excitability of INs from MNs are needed to determine their relative contributions to the windup response. This information could more precisely direct treatment strategies.

4.4.3 Differential Electrical States Related to Patterns of Muscle Function

An unexpected result from this study was that the temporal pattern of the windup response differed between muscle groups. While the windup between strict "flexor" and "extensor" groups were not literally identified in our study, there were two distinct groupings based on coincident activity and temporal changes in the response consisting of 1) hip flexors, ankle dorsiflexors and knee extensors and 2) hip extensors, knee flexors and ankle plantarflexors. Recent work by Endo and Kiehn (Endo and Kiehn 2008) has demonstrated that an asymmetry in conductances exist between extensor and flexor MN pools, in which they have shown that extensor MN pools are dominated more by inhibition than flexor MN pools in the neonatal mouse spinal cord. While we found an asymmetric response in different muscle groups within our study, the asymmetry in Endo and Kiehn's work is different from the windup responses seen during the hip movements. During repeated stretches of the hip flexors, hip flexion, knee extension (particularly in the EXTENDED condition) and ankle dorsiflexion torque typically decreased following the third hip perturbation (see Figs. 2-4). In contrast, hip extension, knee flexion and ankle plantarflexion torques generally remained elevated throughout the course of the hip oscillations when hip extensor muscles were stretched (see Figs. 2-4). This difference in flexor and extensor muscle groups seen in the current study could be due to the fact that tests were done in chronic SCI subjects, since the loss of supraspinal inhibition may result in more disinhibition of the extensor MN pool than the flexor MN pools. However, the asymmetry in the windup responses were consistent with previous studies investigating windup in human SCI. Depression in active muscle torque responses during repeated stretches of hip flexor muscles is consistent with the time course of windup of

the flexion reflex in human SCI, where the responses decrease following the second or third stimulus (Hornby et al. 2003). Stretch of the hip extensor muscles resulted in hip extension, knee flexion and ankle plantarflexion torques that generally remained elevated throughout the course of the hip oscillations (see Figs. 2-4) reflecting the time course of stretch reflex windup (Hornby et al. 2006). EMG activity from RF, MH, and VM followed a similar trend as the torque data, but muscle activity at the ankle, however, was more variable and the pattern of facilitation of the ankle EMG recordings was not as consistent as the torque data. Sol and MG EMG activity rapidly increased through the initial three stretches, but then decreased to levels slightly above the initial response (Fig. 4A), similar to TA activity (Fig. 4B). Since the ankle was held at a constant joint angle, variance among subjects could be due to excitatory polysynaptic pathways and lack of Ia IN inhibition.

4.4.4 Intersubject Variability

While the small number of subjects participating in the study might limit the interpretation, we believe that the consistency of the response across the heterogeneous clinical presentation actually increases the generalizability of the results. In particular, it is interesting that, regardless of completeness of injury (i.e. complete or incomplete) and level of injury, the windup response occurred throughout the lower extremities in response to stretch of a single joint. Additionally, in all of the subjects, we show a delay in activation (ankle muscles) that could suggest a subthreshold windup or involvement of windup in interneurons. However, no conclusive evidence is available regarding level of

injury, completeness of injury or anti-spasticity medication due to the limited sample size.

CHAPTER 5

Reflex Responses during Bilateral Hip Movements in Healthy Individuals

5.1 INTRODUCTION

In the normal control of movement, supraspinal and sensory input are integrated to modulate spinal excitability during movements. Supraspinal input modulates spinal stretch reflex excitability through inhibitory pathways (Gottlieb and Agarwal 1980) so that reflex pathways can be appropriately utilized during voluntary movements. For instance, the soleus H-reflex is modulated over the gait cycle, in which the reflex amplitude is greatest during the stance phase compared to the swing phase (Capaday and Stein 1986). Stretch reflexes have been shown to play a role in the control of walking in man. Specifically, stretch feedback from the hamstrings muscles contributes to EMG activation at the end of the swing phase likely aiding in decelerating the limb (Duysens et al. 1998; Van der Crommert et al. 1996). However, in spinal cord injury (SCI), even partial loss of descending tracts results in deficient modulation over reflex pathways. It has been suggested that after SCI, as a result of weakened cortical influences on spinal neurons, there is a reduction in spinal inhibitory mechanisms (Crone et al. 2003; Heckman 1994). This loss of descending input is thought to largely contribute to the hyperexcitable reflexes observed in people with SCI.

From the SCI data presented in this dissertation, imposed hip movements elicited coordinated multijoint responses throughout the legs, suggesting that stretch-sensitive sensory feedback from the hip largely controls muscle activity in people with SCI. The purpose of this study was to examine whether afferent feedback from hip muscles affects stretch reflex responses in neurologically healthy individuals. The experimental protocol was similar to the protocol described in Chapter 3. Bilateral hip oscillations were

imposed to the legs of neurologically intact (NI) subjects while they remained relaxed during the movements. Since passive stretch of the leg about the hip has not been shown to elicit reflexes in NI subjects (Onushko and Schmit 2007), patellar tendon taps were delivered during the hip movements in order to examine whether stretch reflexes would be facilitated or depressed by the hip movements in NI subjects. Data from spinal cord injured subjects were taken from Chapter 3 of the dissertation and were used for comparison with the NI data.

5.2 METHODS

Eight individuals (age range: 21-31 years) with no reported neurological damage participated in this study. Informed consent was obtained prior to study participation and all procedures were conducted in accordance with the Helsinki Declaration of 1975 and approved by the Institutional Review Board of Marquette University.

The subjects were positioned in the robotic apparatus as detailed in Chapter 3 of this dissertation. Briefly, the subjects lay supine in the robotic apparatus with their legs supported in custom-built leg braces (refer to Chapter 3, Figure 3-1A). The knee and ankle joints were set at fixed angles. The leg braces were attached to servomotor systems (Kollmorgen, Northhampton, MA) that were used to impose sinusoidal hip oscillations of the subjects' legs. A linear motor (LinMot Inc., Delavan, WI) was attached to the right leg brace to deliver controlled patellar tendon tap perturbations during hip movements (refer to Chapter 3 for setup, Figure 3-1B). Reaction joint torque of the hips, knees and ankles were measured with torque transducers (S. Himmelstein and Company, Hoffman

Estates, IL) incorporated into the system. Additionally, surface electromyograms (EMGs) were collected from the following muscles of the right leg: rectus femoris (RF), vastus medialis (VM), vastus lateralis (VL), medial hamstrings (MH), medial gastrocnemius (MG), soleus (Sol) and tibialis anterior (TA). EMG signals were amplified (x 1000), anti-aliased filtered (500 Hz low-pass analog filter) and band-pass filtered (10-1000 Hz; Bortec Biomedical Ltd., Calgary, AB, Canada) prior to data collection. Custom-written LabVIEW programs (National Instruments Corp., Austin, TX) and a data acquisition card (National Instruments Corp., Austin, TX) were used to control the hip oscillations, the motorized reflex hammer, and acquire all signals. All signals were sampled at 1000 Hz and were stored on a PC for processing.

5.2.1 Experimental Protocol

The subjects' legs were oscillated about the hip joint through a sinusoidal trajectory (0.50 Hz) while subjects remained relaxed during the movements. The legs were moved from 40° hip flexion to 10° hip extension in an alternating manner (i.e. 180° out of phase, similar to walking) for 10 continuous cycles. To test whether hip movements modulate stretch reflex responses in NI subjects, patellar tendon taps were delivered during imposed sinusoidal hip movements. Two tendon tap perturbations were delivered to the right patellar tendon either during movement of the right leg into hip flexion ("tap-flx") or hip extension ("tap-ext") (i.e. 2 taps per half cycle). A control condition ("control") was also done in which hip movements were performed without the tendon tap perturbations. The test conditions (control, tap-flx and tap-ext) were repeated three times for a total of 9 tests.

At the completion of the experiment, two additional hip movement tests were performed to estimate the inertial and gravitational components of the leg and leg brace. First, the each leg was quickly (1.5 Hz) oscillated from 25° to 10° hip flexion to estimate the inertial component. Second, each leg was moved slowly (2°/s) through the entire range of motion (40° hip flexion to 10° hip extension) pausing every 5° for 5 seconds to estimate the gravitational and passive joint resistance of the leg for each subject.

5.2.2 Data Analysis

Active muscle torque data were calibrated using the same procedure outlined in Chapter 2, section 2.2.4, of this dissertation. Briefly, the gravitational, passive joint resistances, and inertial components, estimated from the additional hip movements, were subtracted from the torque measurements from the experimental trials using the following equation:

$$\tau_{\text{joint}} = \tau_{\text{measured}} - \tau_{\text{passive/gravity}} - \tau_{\text{inertia}} - \tau_{\text{artifact}}$$

The gravitational and passive joint resistance torques ($\tau_{gravity/passive}$) were estimated by fitting a 3rd order polynomial to the slow, incremental hip movements. The polynomial coefficients were then used to calculate the gravitational and passive joint resistance during the measured trial torques. The inertial torque ($\tau_{inertia}$) was calculated by first subtracting the gravity/passive torque component, and then estimating the inertial contribution of the leg and leg brace using a linear regression analysis. The mechanical artifact ($\tau_{artifact}$) within the system (approximately < 5 Nm) was estimated using an

ensemble average of torque measurements recorded from neurologically healthy subjects from the experiment in Chapter 2 (refer to section 2.2.4).

The peak-to-peak (p-p) reflex amplitude of the RF and VM EMG was identified from the tendon tap perturbations for each consecutive hip oscillation (cycles 1-9). The average p-p reflex response from the 1st and 2nd tendon tap perturbation in the two-tap series was first calculated separately. To reduce variability of the reflex amplitude across subjects, the average p-p amplitude per subject was then normalized to the grand mean of each subject's data set for the individual muscle (i.e. grand average of the reflex amplitude for all three conditions, trials, cycles and tap perturbations). The normalized p-p reflex amplitude from the 1st and 2nd tendon tap perturbation was then averaged across subjects per hip cycle for each muscle.

The peak knee extension and flexion torques in response to the tendon tap perturbations were also identified for the first 9 continuous hip movements. Torque data were first divided into flexion and extension components prior to finding the peak torque. For each subject, the average peak flexion or extension torque data per cycle were then normalized to the grand average of all three trials and conditions per subject. The normalized data were then averaged across subjects per hip movement cycle.

5.3 RESULTS

Passive rotation of the legs in NI subjects did not elicit reflex activity in the lower extremities. Figure 5-1 illustrates example torque responses recorded from the hip, knee and ankle during passive hip movements for a single SCI and NI subject. This

observation in NI subjects is in contrast to what was observed in subjects with SCI, in which reflex responses typically increased with successive hip oscillations (Figure 5-1, and also refer to Chapter 4).

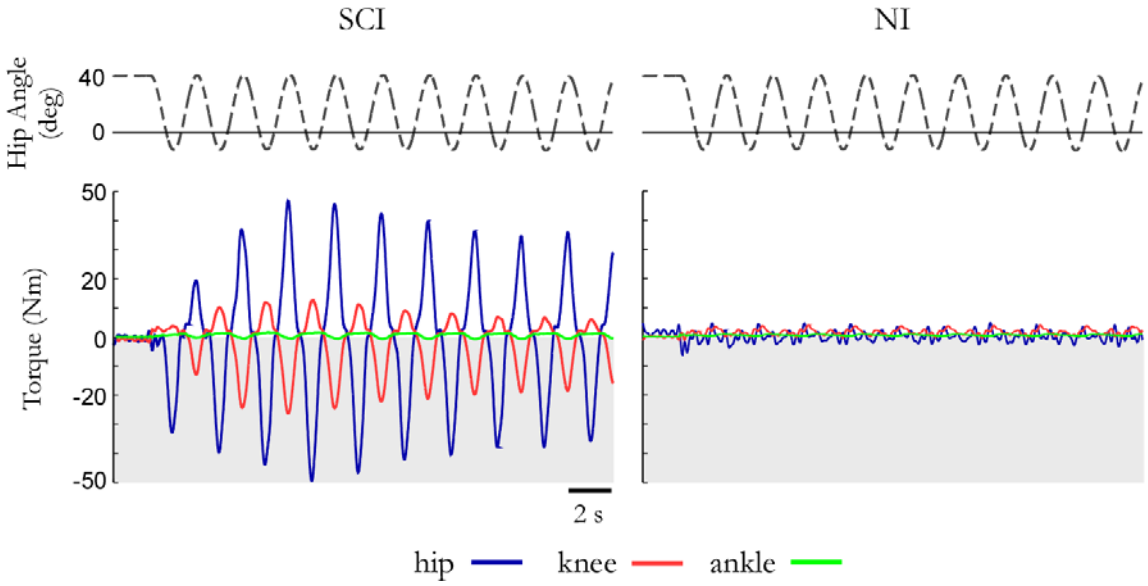


Figure 5-1 Torque Responses during Passive Hip Movements. Example hip, knee and ankle torque responses during passive hip oscillations from a single neurologically intact (NI) subject and single subject with spinal cord injury (SCI; subject S6 from Chapter 3). Shaded area represents flexion torques and non-shaded area represents extension torques.

The addition of patellar tendon taps during the hip movements elicited different reflex response patterns to the 2nd tendon tap (within the two taps per half-cycle of hip movement) in the RF and VM muscles between NI and SCI subjects. Figure 5-2A illustrates typical reflex responses from the RF muscle from an SCI and NI subject for the tap-ext test condition. In general, the reflex response to the 2nd tendon tap perturbation during movement of the leg into hip extension decreased after the first few hip cycles in

NI subjects, whereas in SCI subjects the amplitude increased over the first 3 successive hip movements (indicated by the arrows in Figure 5-2A). In contrast, the reflex amplitude in response to the 1st tendon tap perturbation was similar between SCI and NI subjects. The reflex response in both the RF and VM muscle tended to decrease with consecutive hip movements (Figure 5-2B). Additionally, the pattern of reflex responses did not appear to be altered by the timing of the patellar tendon taps with the hip movements in NI subjects, while it did influence the pattern in SCI subjects. For the tap-ext and tap-flx conditions, the p-p reflex amplitude decreased with successive cycles in NI subjects; however, in SCI subjects, the tap-ext condition appeared to elicit larger reflex responses than the tap-flx condition. Interestingly, in NI and SCI subjects the p-p reflex response amplitude from the 2nd tendon tap (within each half-cycle of hip movement) was larger than the p-p amplitude from the 1st tendon tap for the tap-ext and tap-flx conditions (Figure 5-3).

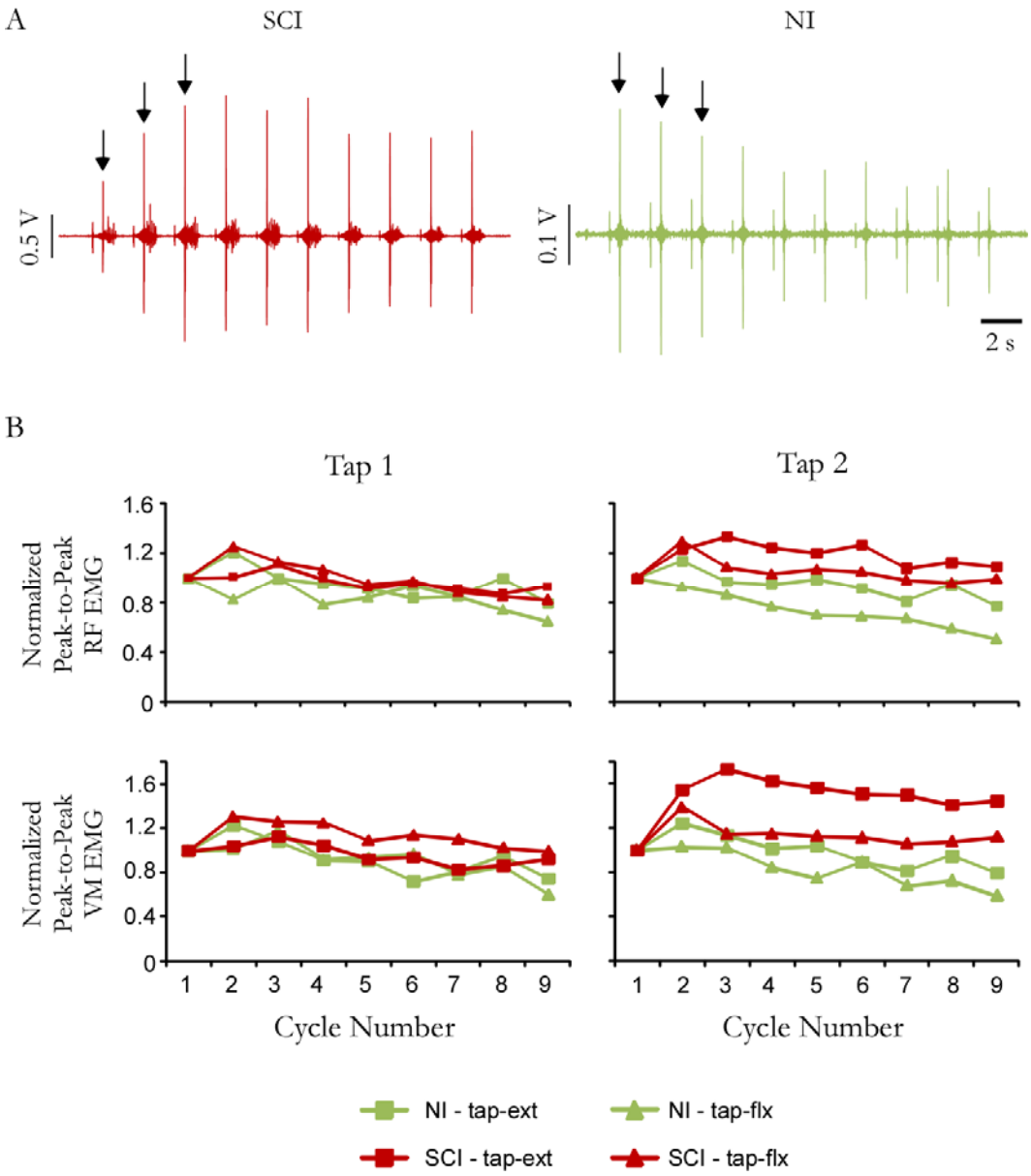


Figure 5-2 Reflex Response to Tendon Tap Perturbation. (A) Single subject example of the RF EMG during patellar tendon tap perturbations delivered during the hip movements (tap-ext condition) from a neurologically intact (NI) subject and subject with spinal cord injury (SCI; subject S6 from Chapter 3). Arrows indicate the reflex response to the 2nd tendon tap in the two-tap sequence. (B) Normalized average peak-to-peak reflex amplitude from the RF and VM EMG to patellar tendon taps during hip movements across conditions. The peak-to-peak reflex amplitude per cycle was normalized to the first response for each condition.

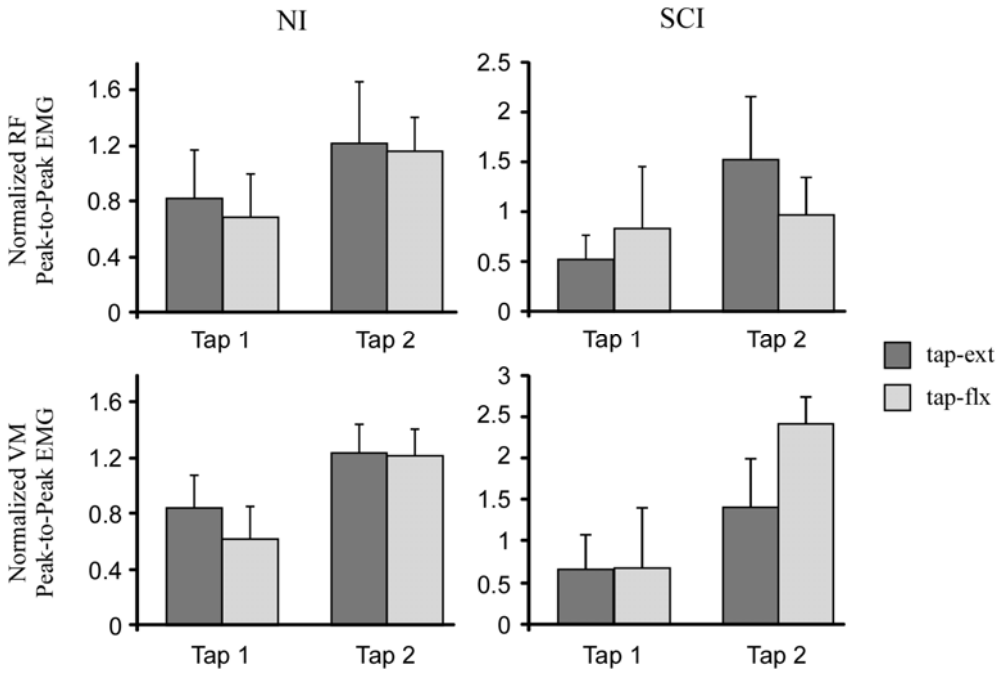


Figure 5-3 Average Peak-to-Peak EMG Reflex Response. Normalized peak-to-peak RF (top panels) and VM (bottom panels) of SCI and NI subjects. The peak-to-peak amplitude of the reflex response is shown for the 1st and 2nd tendon tap during the imposed hip movements.

Peak knee torques followed a similar trend as the EMG in NI subjects. The peak knee flexion and extension torques for all test conditions are illustrated in Figure 5-4. Under the control condition (i.e. hip movement alone), NI subjects did not generate any reflexes. Reflex responses to tendon tap perturbations during the hip oscillations resulted in increased knee extension torque for both the tap-ext and tap-flx conditions compared to the control condition in NI subjects. In addition, similar to the EMG data, a decrease in peak knee extension torque was observed in NI subjects in response to the tendon taps (Figure 5-4). No significant change was observed in the knee flexion torque since the peak torques across all conditions were similar (Figure 5-4).

For comparison, the peak torque from SCI subjects are provided in Figure 5-4 with the reflex responses from NI subjects. In SCI subjects, movement of the hips alone triggered reflex responses throughout the legs. The peak torque from the hip-triggered reflexes are only shown in Figure 5-4, and not the peak reflex torque solely due to the tendon tap (i.e. hip-triggered reflexes vs. reflexes from patellar tendon taps). The addition of patellar tendon tap perturbations influenced the hip-triggered knee extension reflex torque in SCI subjects. Particularly for the tap-ext condition, knee extension torque increased over the initial few hip movement cycles and then remained at an elevated level for the remaining cycles. In NI subjects, reflex responses typically decreased with repeated tendon tap perturbations for both the tap-ext and tap-flx conditions. In addition, peak reflex responses increased over the first 3-4 cycles in SCI subjects, which is similar to the windup of multijoint reflexes that were observed in Chapter 4 of this dissertation.

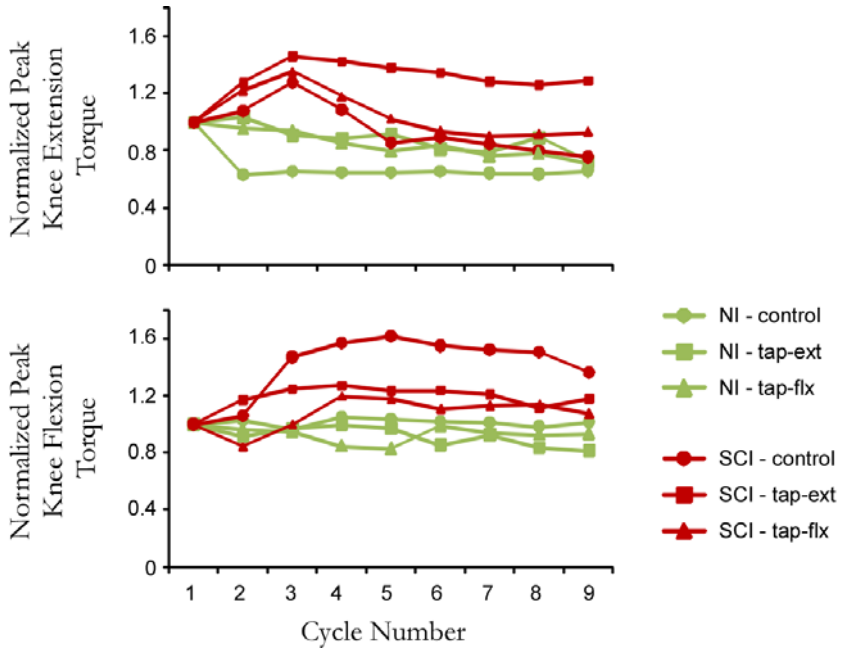


Figure 5-4 Peak Knee Torque per Hip Cycle. Average peak knee extension (top) and flexion (bottom) torque for SCI and NI subjects for each test condition. The peak torque per cycle was normalized to the value of the first peak.

5.4 DISCUSSION

In summary, hip-mediated sensory signals do not strongly modulate stretch reflexes at the knee joint in NI subjects. In contrast to what was observed in SCI subjects, the reflex amplitude to repeated patellar tendon tap perturbations decreased over successive hip movements in NI subjects, whereas an increase in amplitude was observed in SCI subjects over the first few cycles of the imposed hip movements. Furthermore, in NI subjects the reflex response amplitude to tendon tap perturbations was not dependent on the timing of the perturbation with respect to the hip's motion (see Figure 5-3 right panel and Figure 5-4). However, one common result between NI and SCI subjects that

was observed was an increase in the reflex response amplitude in both the RF and VM in response to the 2nd tendon tap within each half-cycle of hip movement (refer to Figure 5-3). These results taken together further support the idea that intact descending input strongly regulates spinal inhibitory mechanisms, since NI subjects did not display the same responses as SCI subjects.

Repeated patellar tendon taps resulted in a decrease in reflex response amplitude in NI subjects, and not as the increase in amplitude as was observed in the SCI subjects. The rate depression seen in reflex amplitude for NI subjects is consistent with post-activation depression of stretch reflexes of the ankle (Crone and Nielsen 1989; Grey et al. 2008; Hultborn et al. 1996). Post-activation depression results from a reduction in transmitter release from primary muscle spindles (Ia) to MNs due to repetitive excitation of Ia fibers (Crone and Nielsen 1989; Curtis and Eccles 1960; Hultborn et al. 1996). Curtis and Eccles (1960) demonstrated that in cat lumbar spinal MN recordings, the magnitude of Ia excitatory postsynaptic potentials (EPSPs) was facilitated with short (< 50 ms) inter-stimulus intervals and depressed at longer intervals (> 100 ms), which is consistent with the present data. The patellar tendon taps were delivered every 2 seconds during the hip movements (2 taps every half-cycle of hip movement), resulting in a depression in reflex amplitude over the 10 hip cycles in NI subjects. However, the inter-stimulus interval between taps within each cycle of hip movement (500 ms) was brief enough which could have caused a slight excitation in MN output resulting in a larger response from the 2nd tendon tap perturbation (refer to Figure 5-3). Post-activation depression was not observed in the SCI subjects, but rather, facilitation was generally observed with repeated stimuli (refer to Figure 5-2 right panels). It has been reported that

spastic patients have decreased post-activation depression (Grey et al. 2008; Nielsen et al. 1995); however, this mechanism has only been studied at a single joint and doesn't fully describe the long-lasting nature of spastic reflexes. While it may play a role in the increased stretch reflex activity seen with spasticity, the data presented in Chapter 4 of this dissertation provides evidence that the increase in reflex output from repetitive hip stretches could also be a result of abnormal intrinsic electrical properties of MNs and INs.

Even though hip proprioceptive feedback was not shown to strongly modulate the stretch reflex at the knee in NI subjects in the current study, the role of hip afferent feedback is still important for functional movements. During walking, the hip provides critical sensory cues for initiating the transition of the limb from the stance phase to the swing phase (Pang and Yang 2000), and input from hip afferents can reset or entrain flexor and extensor bursting patterns in hip muscles during locomotor-type movements (Andersson and Grillner 1983; Kriellaars et al. 1994; Hiebert el al. 1996). In addition to stretch sensory input from the hip, sensory cues from other sources, such as load afferents and cutaneous inputs, have been shown to play an essential role in regulating activation and coordination of muscle activity in animals (Andersson and Grillner 1983; Conway et al. 1987; Gossard et al. 1994; Grillner and Rossignol 1978; Hiebert et al. 1996; Kriellaars et al. 1994). However, in humans these reflex pathways are tightly controlled through descending inhibitory commands allowing proper regulation over spinal neuronal excitability. In conclusion, after a spinal injury, the balance between sensory feedback and descending commands is altered, which can result in abnormal sensory processing and increased spinal neuronal excitability, and ultimately improper coordination of functional movement.

CHAPTER 6

Integration of Results

6.1 SUMMARY OF RESULTS

6.1.1 Brief Summary

The scope of the work presented in this dissertation encompassed the extent that sensory signals from the hip joint interact with voluntary commands and sensory feedback from other joints in the lower extremities in people with spinal cord injury (SCI). In Chapters 2 and 3, the data showed that multijoint reflexes triggered by hip sensory cues remained largely unaltered by descending drive and by stretch-sensitive knee and ankle afferent input. In addition, sensory signals arising from the hip modulated the reflex activity produced by Achilles tendon vibration and patellar tendon tap perturbations. Furthermore, sensory signals from the hip initiated windup in muscles that were not directly involved with the movements (e.g. MG and VM). Taken together, these findings implicate hip-mediated sensory signals as strong modulators of muscle activity throughout the legs, and they appear to play a significant role in setting motoneuronal and/or interneuronal excitability of other neuron pools.

6.1.2 Do Multijoint Reflex Pathways Overlap with Spinal Locomotor Pathways?

These studies further support the concept that the extensor reflex response to hip movements is related to hip proprioception as a locomotor control signal. From the results presented within the current studies, the patterns of the reflex response resembled a fundamental element of locomotion - alternating flexion and extension. In the majority of the muscles recorded, alternating rhythmic patterns were observed during the imposed

movements, with hip flexor responses occurring near hip extension and hip extensor responses occurring during hip flexion movements. Similarly, during treadmill walking, extending the hip produces hip flexion in SCI patients (Dietz et al. 2002; Dobkin et al. 1995). The role of hip proprioceptors for controlling locomotion has also been extensively studied in the cat, showing stretch-sensitive hip afferents control phase transitions of the hip as well as set the rhythm for locomotor behaviors (Kiehn 2006).

From the results of the current studies, muscles distal to the hip, however, did not consistently exhibit activity patterns resembling locomotion. For example, the vasti (VM and VL) were generally active during movement into hip extension, which would correspond to the late stance/early swing in the normal gait cycle. In particular, hip proprioceptive signals were shown to govern knee extensor responses during patellar tendon tap perturbations (Chapter 3). Greater reflex responses in the VM muscle were seen during hip extension. This abnormal coupling between the hip flexors and knee extensors is similar to what has been observed in stroke gait (Lewek et al. 2007). In a previous study combined hip and knee movements in a pattern similar to locomotion resulted in muscle activity that modulated in a more locomotor-appropriate manner (Wu and Schmit 2010). Maintaining the knee in an isometric position, limiting any contribution of knee afferents to multijoint reflexes, might account for the differences observed in the current study.

Sensory feedback alone, however, cannot produce functional locomotion in human SCI, and descending input is still essential for excitatory drive and high level coordination. Descending pathways providing monoaminergic drive is necessary for properly regulating spinal neuron excitability, as implicated from the multijoint windup

observed in the data presented in Chapter 4. In humans with incomplete SCI, corticospinal tract function is highly correlated with walking ability and the amount of descending tracts may be a factor in locomotor function (Thomas and Gorassini 2005). From the data presented in Chapter 2, torque and EMG activity both exhibited windup patterns when SCI subjects were asked to assist the hip movements as well as during the passively imposed hip movements (refer to Figure 6-1). These results suggest that peripheral sensory inputs, and not remaining descending drive, strongly regulates the spinal excitability.

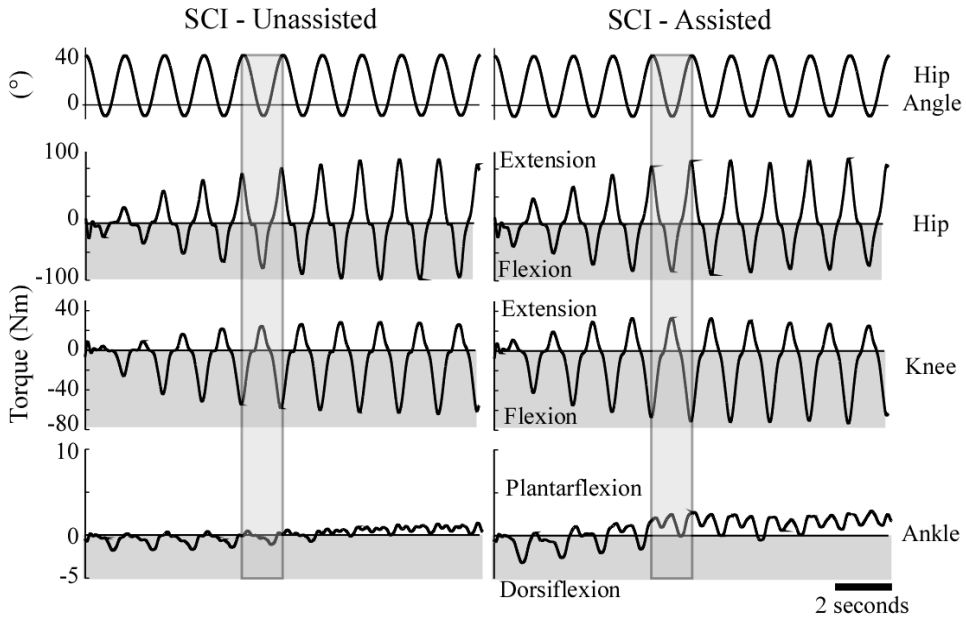


Figure 6-1 Sample torque data from an SCI subject (from Chapter 2). No change in the windup pattern was evident when SCI subjects assisted the hip movements compared to the unassisted hip movements.

6.1.3 Considerations for Rehabilitation

The results from this study suggest that novel strategies for managing unwanted reflex behaviors might be developed in order to improve function post-SCI. Typically SCI patients exhibiting spastic motor behaviors are treated with pharmacological agents, such as baclofen or tizanidine, to curtail unwanted reflex activity. Baclofen is one of the most common anti-spastic medications to treat spasticity in people with various types of neurological injuries, but it has not consistently been shown to be effective for improving function in people with spasticity (Taricco et al. 2000). Furthermore, suppression of hyperactive reflexes through anti-spastic drugs in people with stroke has resulted in no significant increase in the control of movement (McLellan 1977). In line with this, data from Chapter 3 show that knee extensor muscles are influenced by sensory feedback from the hip. VM EMG activity was greater during hip extension, which would be inappropriate reflex activity for normal gait, such that abolishing it could lead to better control of walking. However, spastic activity, especially of the knee extensors, is sometimes used to maintain upright stance during gait in people with SCI.

It has been suggested that suppressing hyperactive reflexes makes people too weak to perform useful movements. In stroke patients, for example, increased tone in extensor muscles may help them to support their body during walking (Berger et al. 1984). Voluntary muscle activation is normally augmented by reflexes, resulting in a net facilitation to the motoneuron pool, helping to increase motor output from voluntary commands (Gandevia et al. 1990). Moreover, baclofen, for instance, targets receptors in the spinal cord where sensory fibers terminate (Yong 1977) having a global effect on spinal excitability. If patients with spasticity are treated with anti-spastic drugs to

suppress reflexes, it may limit the motor output needed for functional movements. Perhaps, more localized spasticity treatments are needed to focus decreased sensory feedback. For instance, in patients with extensor spasms, reducing excitability of neurons innervated by hip muscles may aid with creating appropriate levels of activation of hip muscles while not impeding activity in distal muscles.

6.2 FUTURE STUDIES

One of the limitations to the studies presented in this dissertation is the lack of a quantitative measure of remaining descending signals from the SCI subjects. Transcranial magnetic stimulation (TMS), a common technique for eliciting corticospinal output from the motor cortex, could potentially be used to assess the excitability of corticospinal neurons (Burke et al. 1993) or cortical inhibitory circuits (Davey et al. 1994) in SCI patients to relate these measures to torque and EMG output during hip-triggered reflex activity. For example, the single-pulse TMS technique, a technique used to disrupt sustained EMG activity to measure cortical inhibition (Chen et al. 1999), in conjunction with spinal reflex conditioning stimulation could be used to assess the role of descending inhibitory drive during spastic reflex activity. This information would provide a better understanding of the role of cortical drive in modulating spinal reflex excitability during volitional movements.

A common pattern that is observed in spastic gait is an abnormal coupling of hip flexor and knee extensor activity when the hip moves into full extension. As mentioned earlier, this activity pattern was seen in the current studies, with intensified knee extensor

activity when patellar tendon tap perturbations were delivered during hip extension movements (Chapter 2). This abnormal activity impedes gait by prolonging knee extensor muscle activity prior to the swing phase. Previous studies have shown that sensory input from group Ib afferents (Golgi tendon organs) have strong ties with hip sensory cues. For example, in human infants during treadmill walking, loading the limb prolongs the stance phase and delays swing initiation (Pang and Yang 2000), similar to what has been described for the decerebrate cat (Conway et al. 1987; Duysens and Pearson 1980). Although findings in neurologically healthy subjects have suggested corticospinal input is important for modulating reflex activity between ankle and hip muscles during walking (Iglesias et al. 2008), the role of loading appears to be important for reinforcing the locomotor pattern of muscle activity in human SCI (Dietz et al. 2002; Gordon et al. 2009; Harkema et al. 1997). To elucidate the integration of ankle load and hip-mediated afferent input during locomotor movements in SCI subjects, patellar tendon perturbations could be delivered with ankle loading at different phases of the gait cycle during body weight-supported treadmill walking. Information from this would imply that ankle load afferent feedback is a critical sensory cue for modulating appropriate activity of the quadriceps muscles.

REFERENCES

Ada, L., W. Vattanaslip, N. O'Dwyer, J. Crosbie (1998). *Does spasticity contribute to walking dysfunction after stroke?* J Neurol Neurosurg Psychiatry **64**: 628-635.

Agarwal, G.C. and G.L. Gottlieb (1980). *Effect of vibration of the ankle stretch reflex in man*. Electroencephalogr Clin Neurophysiol **49**: 81-92.

Andersson, O. and S. Grillner (1983). *Peripheral control of the cat's step cycle*. Acta Physiol Scan **118**: 229-39.

Ashby, P. and M. Verrier (1976). *Neurophysiologic changes in hemiplegia. Possible explanation for the initial disparity between muscle tone and tendon reflexes*. Neurology **26:** 1145–1151.

Baldissera, F., H. Hultborn and M. Illert (1981). *Integration in spinal neuronal systems*. In: Handbook of Physiology. The Nervous System. Motor Control. Bethesda, MD.

Bareyer, F.M., M. Kerschensteiner, O. Raineteau, T.C. Mettenleiter, O. Weinmann, and M.E. Schwab (2004). *The injured spinal cord spontaneously forms a new intraspinal circuit in adult rats*. Nat Neurosci **7**: 269-277.

Barolat, G. and D. Maiman (1987). *Spasms in spinal cord injury: A study of 72 subjects*. J Am Paraplegia Soc. **10**: 35-38.

Batschelet, E. (1981). *Circular Statistics in Biology*. New York: Academic Press

Davey, N.J., P. Romaiguere, D.W. Maskill and P.H. Ellaway (1994). *Suppression of voluntary motor activity revealed using transcranial magnetic stimulation of the motor cortex in man*. J Physiol **477**: 223-235.

Bennett, D.J., H. Hultborn, B. Fedirchuk and M. Gorassini (1998a). *Short-term plasticity in hindlimb motoneurones of decerebrate cats*. J Neurophysiol **80**: 2038-2045.

Bennett, D.J., H. Hultborn, B. Fedirchuk and M. Gorassini (1998b). *Synaptic activation of plateaus in hindlimb motoneurons of decerebrate cats*. J Neurophysiol **80**: 2023-2037.

Bennett, D.J., M. Gorassini, K. Fouad, L. Sanelli, Y. Han and J. Cheng (1999). *Spasticity in rats with sacral spinal cord injury*. J Neurotrauma **16**: 69-84.

Bennett, D.J., Y. Li and M. Siu (2001a). *Plateau potentials in sacrocaudal motoneurons of chronic spinal rats, recorded in vitro*. J Neurophysiol **86**: 1955-1971.

Bennett, D.J., Y. Li, P. Harvey and M. Gorassini (2001b). *Evidence for plateau potentials in tail motoneurons of awake chronic spinal rats with spasticity*. J Neurophysiol **86**: 1972-1982.

Bennett, D.J., L. Sanelli, C. Cooke, P. Harvey and M. Gorassini (2004). *Spastic long-lasting reflexes in the awake rat after spinal cord injury*. J Neurophysiol **91**: 2247-2258.

Benz, E., T.G. Hornby, R. Bode, R.A. Scheidt and B.D. Schmit (2005). *A physiological based clinical measure for spastic reflexs in spinal cord injury*. Arch Phys Med Rehab **86**: 52-59.

Berger, W., G. Horstmann and V. Dietz (1984). *Tension development and muscle activation in the leg during gait in spastic hemi-paresis: independence of muscle hypertonia and exaggerated stretch reflexes*. J Neurol Neurosurg Psychiatry **47**: 1029-1033.

Boulenguez, P., S. Liabeuf, R. Bos, H. Bras, C. Jean-Xavier, C. Brocard, A. Stil, P. Darbon, D. Cattaert, E. Delpire, M. Marsala and L. Vinay (2010). *Down-regulation of the potassium-chloride cotransporter KCC2 contributes to spasticity after spinal cord injury*. Nat Med **16**: 302-U397.

Brunnstrom, S. (1970). *Movement therapy in hemiplegia: a neurophysiological approach*. New York: Harper and Row Publishers.

Burke D., K.E. Hagbarth, L. Lofstedt, and B.G. Wallin (1976). *The responses of human muscle spindle endings to vibration of non-contracting muscles*. J Physiol (London) **261**:673–693.

Burke, D. (1983). *Critical examination of the case for or against fusimotor involvement in disorders of muscle tone.* Adv Neurol **39**: 133-150.

Burke, D. (1988). *Spasticity as an adaptation to pyramidal tract injury*. Adv Neurol **47**: 401-423.

Burke, D., R. Hicks, S. C. Gandevia, J Stephen, I. Woodforth and M. Crawford (1993). *Direct comparison of corticospinal volleys in human subjects to transcranial magnetic and electrical stimulation*. J Physiol (London) **470**: 383-393.

Burne, J., V. Carleton, N. O'Dwyer (2005). *The spasticity paradox: movement disorder or disorder of resting limbs?* J Neurol Neurosurg Psychiatry **76**:47-54.

Capaday, C. and R.B. Stein (1986). *Amplitude modulation of the soleus H-reflex in the human during walking and standing*. J Neurosci **6**: 1308-1313.

Capaday, C. and R.B. Stein (1987). *Difference in the amplitude of the human soleus H-reflex during walking and running*. J Physiol **392**: 513-522.

Capaday, C.. A. Lavoie, H. Barbeau, C. Schneider and M. Bonnard (1999). *Studies on the corticospinal control of human walking. I. Responses to focal transcranial magnetic stimulation of the motor cortex*. J Neurophysiol **81**: 129-139.

Chen, D.F., R.D. Theiss, K. Ebersole, J.F. Miller, W.Z. Rymer and C.J. Heckman (2001). *Spinal interneurons that receive input from muscle afferents are differentially modulated by dorsolateral descending systems*. J Neurophysiol **85**: 1005-1008.

Chen, R., A.M. Lozano and P. Ashby (1999). *Mechanism of the silent period following transcranial magnetic stimulation. Evidence from epidural recordings*. Exp Brain Res **128**: 539-542.

Collins, D.F., D. Burke and S.C. Gandevia (2001). *Large involuntary forces consistent with plateau-like behavior of human motoneurons*. J Neurosci **21**: 4059-4065.

Conway, B.A., H. Hultborn and O. Kiehn (1987). *Proprioceptive input resets central locomotor rhythm in the spinal cat*. Exp Brain Res **68**: 643-56.

Corcos, D., G. Gottlieb, P. Penn, B. Myklebust and G. Agarwal (1986). *Movement deficits caused by hyperexcitable stretch reflexes in spastic humans*. Brain **109**:1043-1058.

Crenna, P. and C. Frigo (1987*). Excitability of the soleus H-reflex arc during walking and stepping in man*. Exp Brain Res **66**: 49-60.

Crone, C., H. Hultborn, B. Jespersen and J. Nielsen (1987). *Reciprocal Ia inhibition between ankle flexors and extensors in man*. J Physiol **389**:163-185.

Crone, C., H. Hultborn, O. Kiehn, L. Mazieres and H. Wigstrom (1988). *Maintained changes in motoneuronal excitability by short-lasting synaptic inputs in the decerebrate cat*. J Physiol (London) **405**: 321-343.

Crone, C. and J.B. Nielsen (1989). *Methodological implications of the post-activation depression of the soleus H-reflex in man*. Exp Brain Res **78**:28-32.

Crone, C. and J. Nielsen (1994). *Control of disynaptic reciprocal inhibition in humans*. Acta Physiol Scan **152**: 351-363.

Crone, C., J. Nielsen, N. Petersen, M. Ballegaard and H. Hultborn (1994). *Disynaptic reciprocal inhibition of ankle extensors in spastic patients*. Brain **117**:1161–1168.

Crone, C., L. Johnsen, F. Biering-Sorensen and J. Nielsen (2003). *Appearance of reciprocal facilitation of ankle extensors from ankle flexors in patients with stroke or spinal cord injury*. Brain **126**:495-507.

Dale, N. and S. Grillner (1986). *Dual-component synaptic potentials in the lamprey mediated by excitatory amino acid receptors*. J Neurosci **6**:2653-2661.

Davey, N.J., P. Romaiguere, D.W. Maskill and P.H. Ellaway (1994). *Suppression of voluntary motor activity revealed using transcranial magnetic stimulation or the motor cortex in man*. J Physiol (London) **477**: 223-235.

Daw, N.W., P.S.G. Stein and K. Fox (1993). *The role of NMDA receptors in information processing*. **16**:207-222.

Deutsch, K.M., T.G. Hornby and B.D. Schmit (2005). *The intralimb coordination of the flexor reflex response is altered in chronic human spinal cord injury*. Neurosci Lett **380**: 305-10.

Dietz, V., J. Quintern, and M. Sillem (1987). *Stumbling reactions in man: significance of proprioceptive and pre-programmed mechanisms*. J Physiol (London) **386**: 149-163.

Dietz V., M. Discher, M. Faist and M. Trippel (1990). *Amplitude modulation of the human quadriceps tendon jerk reflex during gait*. Exp Brain Res **82**: 211–213.

Dietz, V., R. Müller and G. Colombo (2002). *Locomotor activity in spinal man: significance of afferent input from joint and load receptors*. Brain **125**:2626-2634.

Dietz, V. (2002). *Proprioception and locomotor disorders*. Nat Rev **3**: 781-790.

Dietz, V. and S.J. Harkema (2004). *Locomotor activity in spinal cord-injured persons*. J Appl Physiol **96**: 1954-1960.

Dietz, V (2008). *Spasticity-spastic movement disorder*. Spinal Cord **46**: 588-588.

Dietz, V., S. Grillner, A. Trepp, M. Hubli, and M. Bolliger (2009). *Changes in spinal reflex and locomotor activity after a complete spinal cord injury: a common mechanism?* Brain **132**: 2196-2205.

Dimitrijevic, M.R., W.A. Spencer, J.V. Trontelj and M. Dimitrijevic (1977). *Reflex effects of vibration in patients with spinal cord lesions*. Neurology **27**: 1078-1086.

Dimitrijevic, M., M. Dimitrijevic, J. Fagnel and A. Sherwood (1984). *Suprasegmentally induced motor unit activity in paralyzed muscles of patients with established spinal cord injury*. Ann Neurol **16**: 216-221.

Dobkin, B., S. Harkema, P. Requejo and V. Edgerton (1995). *Modulation of locomotor-like EMG activity in subjects with complete and incomplete spinal cord injury*. J NeuroEng Rehabil **9**:183-190.

Duysens, J. and K.G. Pearson (1980). *Inhibition of flexor burst generation by loading ankle extensor muscles in walking cats*. Brain Res **187**: 321-32.

Duysens, J., B.M.H. van Wezel, H.W.A.A. van de Crommert, M. Faist and J.G.M. Kooloos (1998). *The role of afferent feedback in the control of hamstrings activity during human gait*. Eur J Morpho **36**: 293-299.

Eccles, J., R. Eccles and A. Lundberg (1957). *The convergence of monosynaptic excitatory afferents on to many different species of alpha motoneurones*. J Physiol (London) **137**: 22-50.

Edgley, S. and E. Jankowska (1989). *An interneuronal relay for group I and II muscle afferents in the midlumbar segments of the cat spinal cord*. J Physiol (Lond) **389**:647-674.

Eken, T., H. Hultborn and O. Kiehn (1989). *Possible functions of transmitter-controlled plaeau-potentials in alpha-motoneurones*. In: Prog Brain Res, pp 257-267. Amsterdam: Elsevier.

Endo, T. and O. Kiehn (2008). *Asymmetric operation of the locomotor central pattern generator in the neonatal mouse spinal cord*. J Neurophysiol **100**: 3043-3054.

Engsberg, J., S. Ross, K. Olree and T. Park (2000). *Ankle spasticity and strength in children with spastic diplegia cerebral palsy.* Devices Med Child Neurol **42**: 42-47.

Faist, M., D. Mazevet, V. Dietz and E. Pierrot-Deseilligny (1994). *A quantitative assessment of presynaptic inhibition of Ia afferents in spastics: differences in hemiplegics and paraplegics*. Brain **117**: 1149-1455.

Faist M., M. Ertel, W. Berger and V. Dietz (1999). *Impaired modulation of quadriceps tendon jerk reflex during spastic gait: differences between spinal and cerebral lesions*. Brain **122**: 567-579.

Fenrich K.K. and P.K. Rose (2009). *Spinal interneuron axons spontaneously regenerate after spinal cord injury in the adult feline*. J Neurosci **29**: 12145-12158.

Ferris, D., K. Gordon, J. Beres-Jones and S. Harkema (2004). *Muscle activation during unilateral stepping occurs in the non-stepping limb of humans with clinically complete spinal cord injury*. Spinal Cord **42**:14-23.

Forrsberg, H., S. Grillner and J. Halberstain (1980). *The locomotion of the low spinal cat I: coordination within a hindlimb*. Acta Physiol Scand **180**: 269-81.

Gandevia, S.C., G. Macefield, D. Burke and D. K. McKenzie (1990). *Voluntary activation of human motor axons in the absence of muscle afferent feedback*. Brain **113**: 1563-1581.

Goldberg, S.R., S. Ounpuu and S.L. Delp (2003). *The importance of swing-phase initial conditions in stiff-knee gait*. J Biomech **36**: 1111-1116.

Gorassini, M., M. Knash, P. Harvey, D.J. Bennett and J. Yang (2004). *Role of motoneurons in the generation of muscle spasms after spinal cord injury*. Brain **127**: 2247-2258.

Gorassini, M., J. Yang, M. Siu and D.J. Bennett (2001*). Intrinsic activation of human motoneurons: possible contribution to motor unit excitation*. J Neurophysiol **87**:1850-1858.

Gordon, K.E., M. Wu, J.H. Kahn, Y.Y. Dhaher, and B.D. Schmit (2009). *Ankle load modulates hip kinetics and EMG during human locomotion*. J Neurophysiol **101**: 2062-2076.

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

Gottlieb, G. and G. Agarwal (1977). *Physiological clonus in man*. Exp Neurol **54**: 616-621.

Gracies, J. M. (2005a). *Pathophysiology of spastic paresis I: Paresis and soft tissue changes*. Muscle Nerve **31**: 535-551.

Gracies, J. M. (2005b). *Pathophysiology of spastic paresis II: Emergence of muscle overactivity*. Muscle Nerve **31**: 552-571.

Grey, M.J., K. Klinge, C. Crone, J. Lorentzen, F. Biering-Sorensen, M. Ravnborg and J.B. Nielsen (2008). *Post-activation depression of Soleus stretch reflexes in healthy and spastic humans*. Exp Brain Res **185**:189-197.

Grillner, S. and S. Rossignol (1978). *On the initiation of the swing phase of locomotion in chronic spinal cats*. Brain Res **146**: 269-277.

Grillner, S. and P. Zangger (1979). *On the central generation of locomotion in the low spinal cat*. Exp Brain Res **34**: 241-61.

Hagbarth, K. E. (1960). *Spinal withdrawal reflexes in the human lower limb*. J Neurol Neurosurg Psychiatry **23**: 222-227.

Hagbarth, K.E. and A.B. Vallbo (1968). *Discharge characteristics of human muscle afferents during muscle stretch and contraction*. Exp Neurol **22**: 674-694.

Hagbarth, K.E., G. Wallin and L. Lofstedt (1973). *Muscle spindle responses to stretch in normal and spastic subjects*. Scand J Rehabil Med **5:** 156–159.

Harkema, S.J., S.L. Hurley, U.K. Patel, P.S. Requejo, B.H. Dobkin and V.R. Edgerton (1997). *Human lumbosacral spinal cord interprets loading during stepping*. J Neurophysiol **77**: 797–811.

Heckman, C.J. (1994). *Alterations in synaptic input to motoneurons during partial spinal cord injury*. Med Sci Sport Exer **26**:1480-1490.

Heckman, C.J., M. Johnson, C. Mottram and J. Schuster (2008). *Persistent inward currents in spinal motorneurons and their influence on human motoneuron firing patterns*. Neuroscientist **14**: 264-275.

Hidler, J.M. and Z.W. Rymer (1999). *A simulation study of reflex instability in spasticity: origins of clonus*. IEEE Trans Rehab Eng **7**: 327-340.

Hiebert, G.W., P.J. Whelan, A. Prochazka and K.G. Pearson (1996). *Contribution of hind limb flexor muscle afferents to the timing of phase transitions in the cat step cycle*. J Neurophysiol **75**: 1126-1137.

Hornby, T.G., W.Z. Rymer, E.N. Benz and B.D. Schmit (2003). *Windup of flexion reflexes in chronic human spinal cord injury: A marker for neuronal plateau potentials?* J Neurophysiol **89**: 416-426.

Hornby, T.G., V. Tysseling-Mattiace, E. Benz and B.D. Schmit (2004). *Contribution of muscle afferents to prolonged flexion withdrawal reflexes in human spinal cord injury*. J Neurophysiol **92**:3375-3384.

Hornby, T.G., J. Kahn, M. Wu and B.D. Schmit (2006). *Temporal facilitation of spastic stretch reflexes following human spinal cord injury*. J Physiol (London) **571**: 593-604.

Hounsgaard, J., H. Hultborn, B. Jespersen and O. Kiehn (1988). *Bistability of alpha-motoneurons in the decerebrate cat and in the acute spinal cat after intravenous 5-hydroxytryptophan.* J Physiol (London) **405**: 345-367.

Hultborn, H. and J. Malmsten (1983). *Changes in segmental reflexes following chronic spinal cord hemisection in the cat I. Increased monosynaptic and polysynaptic ventral root discharges*. Acta Physiol Scand **119**:405-422.

Hultborn, H., S. Meunier, E. Pierrot-Deseilligny and M. Shindo (1987). *Changes in presynaptic inhibition of Ia fibres at the onset of voluntary contraction in man*. J Physiol **389**: 757-772.

Hultborn, H., M. Illert, J. Nielsen, A. Paul, M. Ballegaard and H. Wiese (1996). *On the mechanism of the post-activation depression of the H-reflex in human subjects*. Exp Brain Res **108**:450-462.

Hultborn, H. (2003). *Changes in neuronal properties and spinal reflexes during development of spasticity following spinal cord lesions and stroke: studies in animal models and patients*. J Rehabil Med Suppl **41**: 46–55.

Hultborn, H., M.E. Denton, J. Wienecke and J.B. Nielsen (2003). *Variable amplification of synaptic input to cat spinal motoneurones by dendritic persistent inward current*. J Physiol (London) **552**: 945-952.

Hultborn, H., R. Brownstone, T. Toth and J. Gossard (2004). *Key mechanisms for setting the input-output gain across the motoneuron pool*. Prog Brain Res **143**:77-95.

Hyngstrom, A., M. Johnson, J. Schuster and C.J. Heckman (2008a). *Movement-related receptive fields of spinal motoneurons with active dendrites*. J Physiol (London) **586**: 1581-1593.

Hyngstrom, A., M.D. Johnson and C.J. Heckman (2008b). *Summation of excitatory and inhibitory synaptic inputs by motoneurons with highly active dendrites*. J Neurophysiol **99**: 1643-1652.

Hyngstrom, A., M.D. Johnson, J.F. Miller and C.J. Heckman (2007). *Intrinsic electrical properties of spinal motoneurons vary with joint angle*. Nat Neurosci **10**: 363-369.

Iglesias, C., J.B. Nielsen and V. Marchand-Pauvert (2008). *Corticospinal inhibition of transmission in propriospinal-like neurons during human walking*. Eur J Neurosci **28**: 1351-1361.

Iles, J.F. and R.C. Roberts (1987). *Inhibition of monosynaptic reflexes in the human lower limb*. J Physiol (London) **385**: 69-87.

Iles, J.F. and A. Smith (1988). *Excitation of quadriceps motoneurons from the motor cortex in man*. J Physiol (London) **406**: 147 P.

Ivanhoe, C.B. and T.A. Reistetter (2004). *Spasticity: The misunderstood part of the upper motor neuron syndrome*. Am J Phys Med Rehabil **83**(suppl):S3–S9.

Jankowska, E. and D.A. McCrea (1983). *Shared reflex pathways from Ib tendon organ afferents and Ia muscle spindle afferents in the cat*. J Physiol (London) **338**: 99-111.

Jankowska, E. (1992). *Interneuronal relay in spinal pathways from proprioceptors*. Prog Neurobiol **38**: 335-378.

Jankowska, E. (2001). *Spinal interneuronal systems: identification, multifunctional character and reconfigurations in mammals*. J Physiol (London) **533**: 31-40.

Jankowska, E. and I. Hammer (2002). *Spinal interneurons; how can studies in animals contribute to the understanding of spinal interneuronal systems in man?* Brain Res Rev **40**: 19-28.

Katz, R. and E. Pierrot-Deseilligny (1982). *Recurrent inhibition of alpha-motoneurons in patients with upper motor neuron lesions*. Brain **105**: 103-124.

Kearney, R.E., M. Lortie and R.B. Stein (1999). *Modulation of stretch reflexes during imposed walking movements of the human ankle*. J Neurophysiol **81**: 2893-2902.

Kendall, F. (1983). *Muscles: Testing and Function, with Posture and Pain*. 3 Edition: Lippincott Williams & Wilkins.

Kettunen, P., A. Kyriakatos, K. Hallen and A. El Manira (2005). *Neuromodulation via conditional release of endocannabinoids in the spinal locomotor network*. Neuron **45**: 95-104.

Kiehn, O. (2006). *Locomotor circuits in the mammalian spinal cord*. Ann Rev Neurosci **29**: 279-306.

Knikou, M. and W.Z. Rymer (2002). *Effects of changes in hip joint angle on H-reflex excitability in humans*. Exp Brain Res **143**:149–159.

Knikou, M., D. Chaudhuri, E. Kay, and B.D. Schmit (2006). *Pre- and post-alpha motoneuronal control of the soleus H-reflex during sinusoidal hip movements in human spinal cord injury*. Brain Res **1103**: 123-139.

Knutsson, E. and A. Martensson (1980). *Dynamic motor capacity in spastic paresis and its relation to prime mover dysfunction, spastic reflexes and antagonist co-activation*. Scand J Rehabil Med **12**:93-106.

Knutsson, E., A. Martensson and L. Gransberg (1997). *Influences of muscle stretch reflexes on voluntary velocity-controlled movements in spastic paraparesis*. Brain **120**:1621-1633.

Krenz, N.R. and L.C. Weaver (1998). *Effect of spinal cord transection on N-Methyl-D-Aspartate receptors in the cord*. J Neurotrauma **15**:1072-1036.

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

Kyriakatos, A. and A. El Manira (2007). *Long-term plasticity of the spinal locomotor circuitry mediated by endocannabinoid and nitric oxide signaling*. J Neurosci **27**: 12664-12674.

Lam, T. and K.G. Pearson (2001). *Proprioceptive modulation of hip flexor activity during the swing phase of locomotion in decerebrate cats*. J Neurophysiol **86**: 1321-1332.

Lance, J. (1980) *Spasticity: Disordered Motor Control*. In: (R Feldman RY, W Koella, ed), pp 185-203. Chicago, IL: Yearbook Medical Publishers

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

Larsen, B., N. Mrachacz-Kersting, B.A. Lavoie and M. Voigt (2006). *The amplitude modulation of the Quadricepts H-reflex in relation to the knee joint action during walking*. Exp Brain Res **170**: 555-566.

Lee, R.H. and C.J. Heckman (1999). *Enhancement of bistability in spinal motoneurons in vivo by the noradrenergic alpha1 agonist methoxamine*. J Neurophysiol **81**: 2164-2174.

Lee, R.H. and C.J. Heckman (2000). *Adjustable amplification of synaptic input in the dendrites of spinal motoneurons in vivo*. J Neurosci **20**: 6734-6740.

Lewek, M.D., B.D. Schmit, T.G. Hornby and Y.Y. Dhaher (2006). *Hip joint position modulates volitional knee extensor muscle activity after stroke*. Muscle Nerve **34**: 767-774.

Lewek, M.D., T.G. Hornby, Y.Y. Dhaher and B.D. Schmit (2007). *Prolonged quadriceps activity following imposed hip extension: a neurophysiological mechanism for stiff-knee gait?* J Neurophysiol **98**: 3153-3162.

Li, Y. and D.J. Bennett (2003). *Persistent sodium and calcium currents cause plateau potentials in motoneurons of chronic spinal rat*s. J Neurophysiol **90**: 857-869.

Li, Y.R., M.A. Gorassini and D.J. Bennett (2004). *Role of persistent sodium and calcium currents in motoneuron firing and spasticity in chronic spinal rats*. J Neurophysiol **91**: 767-783.

Little, J.W., P. Micklesen, R. Umlauf and C. Britell (1989). *Lower-extremity manifestations of spasticity in chronic spinal-cord injury*. Am J Phys Med Rehab **68**: 32-36.

Liu, D. W. Thangnipon and D.J. McAdoo (1991). *Excitatory amino acids rise to toxic levels upon impact injury to the rat spinal cord*. Brain Res **547**:344-348.

Lum, P., C. Pattern, D. Kothari and R. Yap (2004). *Effects of velocity on maximal torque production in poststroke hemiparesis*. Muscle Nerve **30**: 732-734.

Lundberg, A., K. Malmgren and E.D. Schomburg (1977). *Cutaneous facilitation of transmission in reflex pathways from Ib afferents to motoneurons*. J Physiol (London) **265**: 763-780.

Macht, M. and R. Kuhn (1948). *The occurrence of extensor spasm in the patients with complete transection of the spinal cord*. New Engl J Med **238**: 311-314.

Maegele, M., S. Müller, A. Wernig, V. Edgerton and S. Harkema (2002). *Recruitment of spinal motor pools during voluntary movements versus stepping after human spinal cord injury*. J Neurotrauma **19**: 1217-1229.

Magnuson, D.S., R. Lovett, C. Coffee, R. Gray and Y. Han (2005). *Functional consequences of lumbar spinal cord contusion injuries in the adult rat*. J Neurotrauma **22**: 529-543.

Maynard, F.M., R.S. Karunas and W.P. Waring III (1990). *Epidemiology of spasticity following traumatic spinal cord injury*. Arch Phys Med Rehabil **71**: 566-569.

Mazzocchio, R. and A. Rossi (1997). *Involvement of spinal recurrent inhibition in spasticity. Further insight into the regulation of Renshaw cell activity*. Brain **120**: 991-1003.

McLellan, D.L. (1977). *Co-contraction and stretch reflexes in spasticity during treatment with baclofen*. J Neurol Neurosurg Psychiatry **40**: 30-38.

McPherson, J.G., M.D. Ellis, C.J. Heckman and J.P.A. Dewald (2008). *Evidence for increased activation of persistent inward currents in individuals with chronic hemiparetic stroke*. J Neurophysiol **100**: 3236-3243.

Mendell, L.M. (1966). *Physiological properties of unmyelinated fiber projection to the spinal cord*. Exp Neurol **16**:316-332.

Miller, J., D. Paul, W.Z. Rymer and C.J. Heckman (1995). *5-HT1B/1D agonist CGS-12066B attenuates clasp knife reflexes in the cat*. J Neurophysiol **74**: 453-456.

Miller, J.F., K.D. Paul, R.H. Lee, W.Z. Rymer and C.J. Heckman (1996). *Restoration of extensor excitability in the acute spinal cat by the 5-HT2 agonist DOI*. J Neurophysiol **75**: 620-680.

Mottram, C.J., N.L. Suresh, C.J. Heckman, M.A. Gorassini and W.Z. Rymer (2009). *Origins of abnormal excitability in biceps brachii motoneurons of spastic-paretic stroke survivors*. J Neurophysiol **102**: 2026-2038.

Murray, K.C., A. Nakae, M.J. Stephens, M. Rank, J. D'Amico, P.J. Harvey, X. Li, R.L.W. Harris, E.W. Ballou, R. Anelli, C.J. Heckman, T. Mashimo, R. Vavrek, L. Sanelli, M.A. Gorassini, D.J. Bennett and K. Fouad (2010). *Recovery of motoneuron and locomotor function after spinal cord injury depends on constitutive activity in 5-HT2C receptors*. Nat Med **16**: 694-U699.

Myklebust, B.M., G. Gottlieb, R.D. Penn and G. Agarwal (1982). *Reciprocal excitation of antagonist muscles as a differentiating feature in spasticity*. Ann Neurol **12**: 367-374.

Nichols, T.R. (1999). *Receptor mechanisms underlying heterogenic reflexes among the triceps surae muscles of the cat*. J Neurophysiol **81**: 467-478.

Nickolls, P., D.F. Collins, R.B. Gorman, D. Burke and S.C. Gandevia (2004). *Forces consistent with plateau-like behaviour of spinal neurons evoked in patients with spinal cord injuries*. Brain **127**: 660-670.

Nielsen, J., C. Crone, T. Sinkjaer, E. Toft and H. Hultborn (1995). *Central control of reciprocal inhibition during fictive dorsiflexion in man*. Exp Brain Res **104**: 99-106.

Nielsen, J., N. Petersen and C. Crone (1995). *Changes in transmission across synapses of Ia afferents in spastic patients*. Brain **118**:995-1004.

Nielsen, J., C. Crone and H. Hultborn (2007). *The spinal pathophysiology of spasticity – from a basic science point of view*. Acta Physiol **189**: 171-180.

NSCISC (2010). Annual Report for the Model Spinal Cord Injury Care Systems. Birmingham, AL, National Spinal Cord Injury Statistical Center.

Onushko, T. and B.D. Schmit (2007). *Reflex response to imposed bilateral hip oscillations in human spinal cord injury*. J Neurophysiol **98**: 1849-1861.

Onushko, T., A. Hyngstrom and B.D. Schmit. *Bilateral oscillatory hip movements induce windup of multi-joint lower extremity spastic reflexes in chronic spinal cord injury*. In: Society for Neuroscience. Washington DC: 2008.

Onushko, T. and B.D. Schmit (2008). *Coordinated muscle activity of the legs during assisted bilateral hip oscillations in human spinal cord injury*. Biomed Sci Instrum **44**: 286-291.

Onushko, T. and B.D. Schmit (2010). *Effects of multijoint spastic reflexes of the legs during assisted bilateral hip oscillations in human spinal cord injury*. Arch Phys Med Rehabil **91**: 1225-1235.

Pang, M., and J. Yang (2000). *The initiation of the swing phase in human infant stepping: importance of hip position and leg loading*. J Physiol (London) **528**: 389-404.

Pang, M. and J. Yang (2001). *Interlimb co-ordination in human infant stepping*. J Physiol (London) **533**: 617-625.

Pearson, K.G., J.M. Ramirez and W. Jiang (1992). *Entrainment of the locomotor rhythm by group Ib afferents from ankle extensor muscles in spinal cats*. Exp Brain Res 90: 557-566.

Perrier, J.F. and J. Hounsgaard (2003). *5-HT2 receptors promote plateau potentials in turtle spinal motoneurons by facilitating an L-type calcium current*. J Neurophysiol **89**: 954-959.

Petersen, N., L.O.D. Christensen and J. Nielsen (1998). *The effect of transcranial magnetic stimulation on the soleus H-reflex during human walking*. J Physiol (London) **513**: 599-610.

Petersen, N.J.E. Butler, V. Marchand-Pauvert, R. Fisher, A. Ledebt, H.S. Pyndt, N.L. Hansen and J.B Nielsen (2001). *Suppression of EMG activity by transcranial magnetic stimulation in human subjects during walking*. J Physiol (London) **537**: 651-656.

Rothwell, J. C. (1994). *Control of human voluntary movement*. 2nd Ed. London: Chapman and Hall.

Roll, J.P., J.P. Vedel and E. Ribot (1989). *Alteration of proprioceptive messages induced by tendon vibration in man: a microneurographic study*. Exp Brain Res **76**: 213–222.

Rossi, A., R. Mazzocchio and C. Scarpini (1990). *Clonus in man: a rhythmic oscillation maintained by a reflex mechanism*. Electroencephalogr Clin Neurophysiol **75**: 56-63.

Rossi, A., B. Chi and V. Vecchione (1992). *Supraspinal influences on recurrent inhibition in humans. Paralysis of descending control of Renshaw cells in patients with mental retardation*. Electroencephalogr Clin Neurophysiol **85**: 419-424.

Rushworth, G. (1960). *Spasticity and rigidity: an experimental study and review*. J Neurol Neurosurg Psychiatry **23**: 99-118.

Russo, R.E. and J. Hounsgaard (1996). *Plateau-generating neurones in the dorsal horn in an in vitro preparation of the turtle spinal cord*. J Physiol (London) **493**: 39-54.

Russo, R.E. and J. Hounsgaard (1994). *Short-term plasticity in turtle dorsal horn neurons mediated by L-type Ca2+ channels*. Neuroscience **61**: 191-197.

Schindler-Ivens, S.M. and R K. Shields (2004). *Soleus H-reflex recruitment is not altered in persons with chronic spinal cord injury*. Arch Phys Med Rehabil **85**: 840-847.

Rymer, W.Z., J.C. Houk, and P.E. Crage (1979). *Mechanisms of the clasp-knife reflex studied in an animal model*. Exp Brain Res **37**:93-113.

Schmit, B.D., A. McKenna-Cole and W.Z. Rymer (2000). *Flexor reflexes in chronic spinal cord injury triggered by imposed ankle rotation*. Muscle Nerve **23**: 793-803.

Schmit, B.D. and E.N. Benz (2002). *Extensor reflexes in human spinal cord injury: activation by hip proprioceptors*. Exp Brain Res **145**: 520-527.

Schmit, B.D., E. Benz and W. Z. Rymer (2002). *Afferent mechanisms for the reflex response to imposed ankle movement in chronic spinal cord injury*. Exp Brain Res **145**: 40-49.

Schmit, B.D., T.G. Hornby, V. Tysseling-Mattiace and E.N. Benz (2003). *Absence of local sign withdrawal in chronic human spinal cord injury*. J Neurophysiol **90**: 3232-3241.

Schwindt, P. and W. Crill (1980). *Role of a persistent inward current in moto-neuron bursting during spinal seizures*. J Neurophysiol **43**: 1296-1318.

Shahani, B. and R. Young (1971). *Human flexor reflexes*. J Neurol Neurosurg Psychiatry **34**: 616-627.

Shefner J.M., S.A. Berman, M. Sarkarati and R.R. Young (1992). *Recurrent inhibition is increased in patients with spinal cord injury*. Neurology **42**: 2162–2168.

Sherrington, C.S. and E.H. Hering (1897). *Antagonistic muscles and reciprocal innervations*. Fourth note. P R Soc London **62**: 183-187.

Sinkjaer, T., E. Toft, K. Larsen, S. Andreassen and H.J. Hansen (1993). *Non-reflex and reflex mediated ankle joint stiffness in multiple sclerosis patients with spasticity*. Muscle Nerve **16**: 69-76.

Sinkjaer, T., J.B. Andersen, M. Ladouceur, L.O.D. Christensen and J.B. Nielsen (2000). *Major role for sensory feedback in soleus EMG activity in the stance phase of walking in man*. J Physiol (London) **523**: 817-827.

Sjölund, B.H. (2002). *Pain and rehabilitation after spinal cord injury: the case of sensory spasticity?* Brain Res Rev **40**:250-256.

Sköld, C., R. Levi and A. Seiger (1999). *Spasticity after traumatic spinal cord injury: nature, severity, and location*. Arch Phys Med Rehab **80**: 1548-1557.

Smith, M. and J.F. Perrier (2006). *Intrinsic properties shape the firing pattern of ventral horn interneurons from the spinal cord of the adult turtle*. J Neurophysiol **96**: 2670-2677.

Steldt, R. and B.D. Schmit (2004). *Modulation of coordinated muscle activity during imposed sinusoidal hip movements in human spinal cord injury*. J Neurophysiol **92**: 673-685.

Svirskis, G. and J. Hounsgaard (1997). *Depolarization-induced facilitation of a plateau-generating current in ventral horn neurons in the turtle spinal cord*. J Neurophysiol **78**: 1740-1742.

Tanaka, R. (1974). *Reciprocal Ia inhibition during voluntary movements in man.* Exp Brain Res **21**: 529-540.

Taricco, M, R. Adone, C. Pagliacci and E. Telaro (2000). *Pharmacological interventions for spasticity following spinal cord injury*. Cochrane Database of Systematic Reviews.

Theiss, R.D., J.J. Kuo and C.J. Heckman (2007). *Persistent inward currents in rat ventral horn neurones*. J Physiol (London) **580**: 507-522.

Valero-Cabre, A., J. Fores and X. Navarro (2004). *Reorganization of reflex responses mediated by different afferent sensory fibers after spinal cord transection.* J Neurophysiol **91**: 2838-2848.

Van de Crommert, H.W.A.A., M. Faist, W. Berger and J. Duysens (1996). *Biceps femoris tendon jerk reflexes are enhanced at the end of the swing phase in humans*. Brain Res **734**: 341-344.

Whelan, P.J., G.W. Hiebert and K.G. Pearson (1995). *Stimulation of the group I extensor afferents prolongs the stance phase in walking cats*. Exp Brain Res **103**: 20-30.

Wilson, L.R., S.C. Gandevia, J.T. Inglis, J.M. Gracies and D. Burke (1999). *Muscle spindle activity in the affected upper limb after a unilateral stroke*. Brain **122**: 2079-2088.

Wu, M., T.G. Hornby, J. Hilb and B.D. Schmit (2005). *Extensor spasms triggered by imposed extension in chronic human spinal cord injury*. Exp Brain Res **162**: 239-249.

Wu, M. and B.D. Schmit (2006). *Spastic reflexes triggered by ankle load release in human spinal cord injury*. J Neurophysiol **96**: 2941-2950.

Wu, M. and B. D. Schmit (2010). *Reflex response to combined hip and knee motion in human chronic spinal cord injury*. J Rehabil Res Dev **47**: 117-132.

Xia, R. and Z. Rymer (2005). *Reflex reciprocal facilitation of antagonist muscles in spinal cord injury*. Spinal Cord **43**: 14-21.

Yanagisawa, N. and R. Tanaka (1978). *Reciprocal Ia inhibition in spastic paralysis in man.* Electroen Clin Neuro **34**: 521-526.

Yang, J.F. and P.J. Whelan (1993). *Neural mechanisms that contribute to cyclical modulation of the soleus H-reflex in walking in humans*. Exp Brain Res **95:** 547–556.

Young, R. R. (1977). *Baclofen*. Arch Neurol **34**: 422.

Zehr, E. P. and R.B. Stein (1999). *What functions do reflexes serve during human locomotion?* Prog Neurobiology **58**: 185-205.

Ziskind-Conhaim, L., L.Y. Wu and E.P. Wiesner (2008). *Persistent sodium current contributes to induced voltage oscillations in locomotor-related Hb9 interneurons in the mouse spinal cord*. J Neurophysiol **100**: 2254-2264.

APPENDIX A: MODIFICATION OF MULTIJOINT WINDUP

An additional experiment was conducted to test whether inhibitory input from Ia firing of an antagonist muscle group (i.e. quadriceps) through patellar tendon tapping would decrease spastic reflexes observed in the hamstrings muscles, or if sensory input from non-related pathways (i.e. vibration of the Achilles tendon) would affect reflex activity in human spinal cord injury (SCI). As described in Chapter 4, the loss of inhibitory input from Ia interneurons could be an important factor for the modulation of persistent inward currents (PICs) post-SCI. In the decerebrate cat model, PIC amplitude has been shown to be tightly controlled through input from reciprocal Ia inhibitory interneurons (Hyngstrom et al. 2007). Additionally, recent evidence in SCI rat models have demonstrated that motoneuron excitability in chronic injuries may result from changes in gene expression of the receptors important for regulating motoneuron excitability (Boulenguez et al. 2010; Murray et al. 2010). These latter studies support observations of increased reflex excitability in response light cutaneous inputs in human SCI.

The same nine subjects who participated in the experiment described in Chapter 3, also participated in this study, and the experimental instrumentation used for this study was the same as described in Chapter 3 of this dissertation. A brief description of the experiment and results are provided.

Nine subjects with incomplete (8) or complete (1) chronic SCI were transferred onto a therapy plinth and lay supine with their legs supported in custom-built leg braces, which were attached to servomotor drive systems (Kollmorgen, Northhampton, MA). The knee and ankle joints were held isometrically for this experiment and spastic

hamstring muscle reflex activity was triggered by hip sensory cues associated with leg flexion. To elicit spastic reflex activity, the servomotor drive systems imposed three sinusoidal oscillations of the right leg about the hip joint. The leg was moved between 40º hip flexion and 10º hip extension at 0.50 Hz, with the last movement ending in 40º hip flexion to keep the hamstrings stretched. At the end of the third oscillation, one of two perturbations was applied: (1) patellar tendon tapping (2 Hz, 10 taps) ("tap") or (2) 80 Hz vibration of the Achilles tendon (5 s duration) ("vibe"). A control condition was also performed in which no perturbation was applied at the end of the hip oscillations ("control"). Additionally, to test whether remaining supraspinal input had an effect on spastic reflex responses with vibration or tendon tapping, subjects were asked to either assist the imposed hip oscillations ("active") or to remain relaxed during the oscillations ("passive"). Subjects were instructed to relax at the end of the third oscillation. A total of 18 trials were performed using a randomized block design.

Surface electromyograms (EMGs) were collected from seven muscles on each leg: rectus femoris (RF), vastus medialis (VM), vastus lateralis (VL), medial hamstrings (MH), medial gastrocnemius (MG), soleus (Sol) and tibialis anterior (TA). EMG signals were amplified (x 1000) and band-pass filtered (10-1000 Hz; Bortec Biomedical Ltd., Calgary, AB, Canada) prior to data collection. Torque measurements were also recorded through torque transducers (S. Himmelstein and Company, Hoffman Estates, IL) that were integrated within the system.

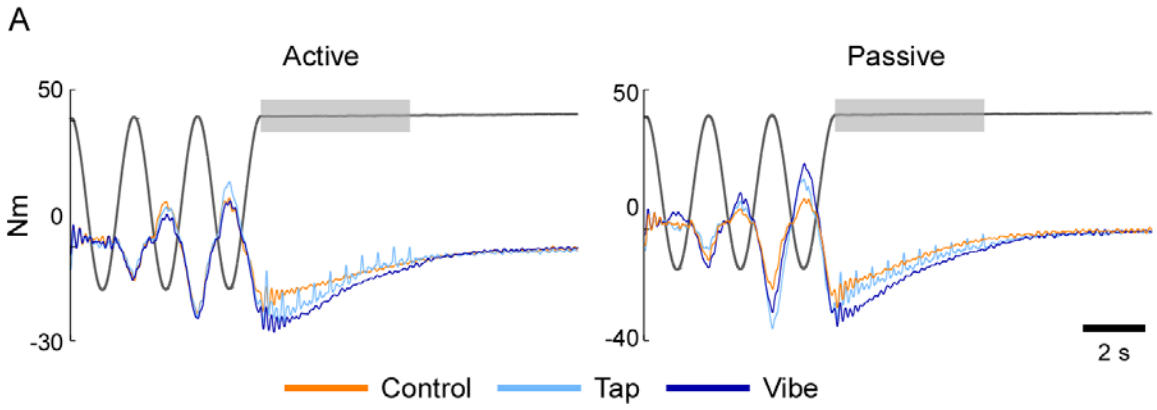
The torque data were calibrated using the same method described in Chapter 2, section 2.2.4. Additionally, the data from the one complete SCI subject was excluded from analysis since no tendon reflexes could be elicited and spastic reflexes were not

elicited during the hip movements. At the end of the movement, the decline in the torque response was quantified by determining the time constants at 63% and 37% (τ_{63} and τ_{37}, respectively) of the torque amplitude at the start of the pause period (i.e. when the hip reached full flexion in the third oscillation). The Friedman's Analysis of Variance non-parametric statistical test was used to compare the time constant values among the applied sensory perturbations with the control condition and between active and passive tests. The time constants at 63% and 37% were analyzed as separate statistical tests. Significance level was set to $\alpha = 0.05$.

Spastic reflex activity in knee torque and EMG data are provided in Figure 7-1A and B from a single SCI subject. The reflex activity patterns generated from the hip oscillations elicited windup in torque and EMG responses, but the ensuing activity at the end of the movement varied among the subjects. Four (S4, S5, S6, and S8) of the eight subjects demonstrated prolonged activity at the end of the hip oscillations that resembled the response illustrated in Figure 7-1. Three of the subjects (S1, S2, and S3) demonstrated inconsistent responses, in which little or no reflex activity was seen during passive tests. In active tests, a rapid decline in activity was observed a short delay (< 500 ms) after the end of the movement. One subject (S9) demonstrated a unique response to the tendon tap perturbations applied at the end of the movements. Following the 3rd or 4th tendon tap, a strong myoclonus activity developed throughout both legs (refer to Figure 7-1C).

Overall, no systematic change in the responses to tendon tap perturbations or vibration was seen across the tested subjects. Torque results indicated no significant alteration in response to patellar tendon taps or Achilles tendon vibration (time constants, Friedman's test: τ_{63}, $p = 0.267$; τ_{37}, $p = 0.437$), as did the MH EMG (τ_{63}, $p = 0.089$; τ_{37},

p = 0.265). While PICs have been shown to be highly sensitive to inhibitory input, specifically Ia (Hyngstrom et al. 2007), sensory input to the antagonist muscle (tendon tapping or vibration) following the hip oscillations did not alter the prolonged reflex responses in SCI subjects. Recent animal work has shown that a specific serotonin receptor type (5-HT2C) spontaneously remains open after spinal injury. The abnormal expression of this receptor allows for large persistent inward currents to occur, which is consistent with the findings from this study.



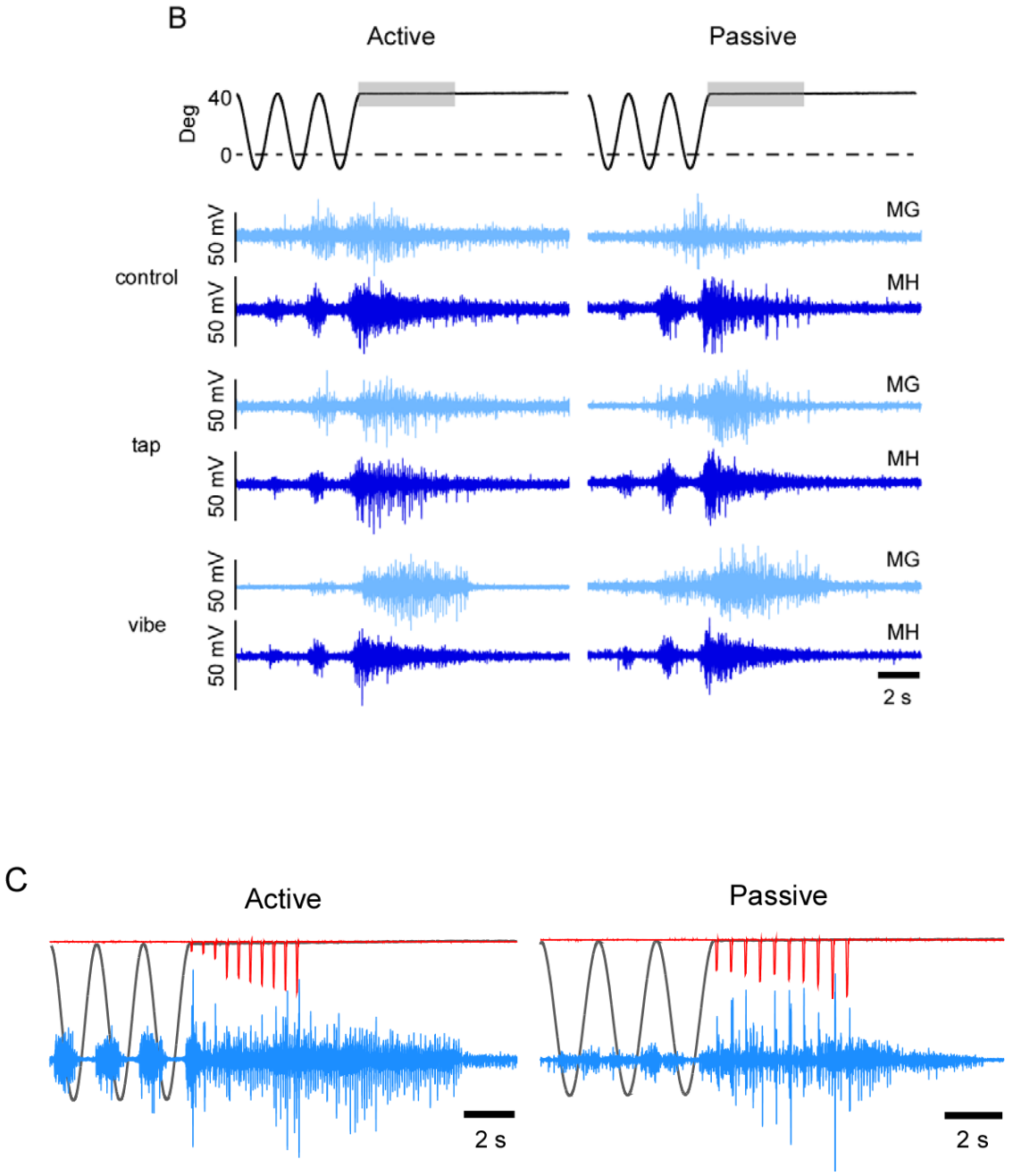


Figure 7-1 Example SCI Data of Windup Response to Sensory Input. Example data from subject SCI subject (S4, refer to Table 3-1)) showing windup of torque (A), MG and MH EMG (B) during the hip oscillations, and prolonged muscle activity at the end of the movements. The shaded area represents the time when vibration or tendon tap perturbations were applied. (C) Example of myoclonus, measured in the MH EMG, which developed in one SCI subject (S9) in response to the tendon tap perturbations.