Retraining walking after spinal cord injury: functional gains and neuroplasticity

by

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ABSTRACT

Physical training can affect the excitability of spinal reflexes in a training-specific manner in uninjured humans. Therefore, the first part of this thesis examined the changes in the excitability of a polysynaptic and a monosynaptic reflex in an ankle plantarflexor, after contrasting forms of walking retraining in humans with chronic incomplete spinal cord injury: 1) Endurance training, which emphasized walking speed and distance, and 2) Precision training, which emphasized skilled stepping movements over ground. We determined that both reflexes responded in a training-specific manner, wherein the excitability of only the polysynaptic reflex was reduced after Endurance training. Furthermore, the reduced excitability of the polysynaptic reflex before Endurance training and the decrease in the excitability of the monosynaptic reflex across all the training were related to improvements in walking ability.

Over ground powered exoskeletons are a recent development and have provided individuals with motor complete spinal cord injuries the ability to walk. Therefore, the second part of this thesis examined the changes in walking ability, other functional capabilities and neuroplasticity after walking retraining using the ReWalk powered exoskeleton. We determined that individuals who could not walk before the training were able to walk using the ReWalk after the training. Furthermore, some individuals who could walk before the training improved their walking ability without the ReWalk, after the training. We also observed improvements in other functional capabilities such as improved postural stability and reduced physiologic exertion during walking, and some neuroplasticity within sensory and motor pathways. Finally, field tests of walking in the ReWalk were performed in- and outdoors, and revealed that while some tasks

were possible in the ReWalk, others were not, either due to the ReWalk's design features or safety issues.

Spinal reflexes play a significant role in the regulation of locomotion in humans. Maladaptive neuroplasticity after spinal cord injury leads to the hyperexcitability of spinal reflexes, resulting in spasticity. Before studying the effects of any form of intervention on spinal pathways that, for example, share the Ia-motoneuronal pathway, we first need to establish the variability of the reflexes across multiple days. Therefore, in the third part of this thesis, we determined that spinal reflexes that share the Ia-motoneuronal pathway exhibit differential changes in day-to-day variability within and across individuals with chronic incomplete spinal cord injury.

PREFACE

Chapter 1 - Introduction: The literature review in Chapter 1 is my original work, and revised using feedback provided by the Supervisor.

Chapter 2 – Training-specific neural plasticity in spinal reflexes after incomplete spinal cord injury: A version of Chapter 2 of this thesis has been published as **Khan AS, Patrick SK, Roy FD, Gorassini MA, and Yang JF**. Training-Specific Neural Plasticity in Spinal Reflexes after Incomplete Spinal Cord Injury. *Neural Plast* 2016: 6718763, 2016. I was involved with data analysis and statistics, preparing the manuscript, and share co-authorship with the Supervisor, Supervisory Committee members, and S.K. Patrick. Research ethics approval for the research project in Chapter 2 was provided by the Health Research Ethics Board at the University of Alberta and Alberta Health Services, Pro00003873.

Chapter 3 – Training to walk in the ReWalk exoskeleton after incomplete and complete spinal cord injury: I was involved with the collection and analysis of some of the data, organizing and summarizing the data and its statistics, and the write up of Chapter 3 which has been revised using feedback from D.C. Livingstone, C.L. Hurd, J.E. Misiaszek, Supervisory Committee member M.A. Gorassini, P.J. Manns, R.B. Stein, and the Supervisor. Research ethics approval for the research project in Chapter 3 was provided by the Health Research Ethics Board at the University of Alberta, Pro00036789.

Chapter 4 – Reliability of spinal reflexes reflecting the Ia-motoneuronal pathway after incomplete spinal cord injury: I was involved with data collection, analysis and statistics, and the write up of Chapter 4 which has been revised using feedback provided by the Supervisor. Research ethics approval for the research project in Chapter 4 was provided by the Health Research Ethics Board at the University of Alberta, Pro00031413.

Chapter 5 - Discussion: The discussion in Chapter 5 is my original work, and revised using feedback provided by the Supervisor.

To my parents.

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CHAPTER 1: INTRODUCTION

1.1 Preface

This thesis examines the changes in neuroplasticity, walking ability and functional capabilities before and after different methods of retraining walking after incomplete and complete spinal cord injury (SCI) in humans. Spinal reflexes play a pivotal role in the regulation of walking. Therefore, we determined the effects of contrasting forms of walking retraining on spinal reflexes, in people with incomplete SCI. However, to also understand the effects of retraining walking after complete SCI, the effects of walking retraining were determined using an over ground powered exoskeleton. These exoskeletons are a recent development, have enabled individuals with severe incomplete or complete SCI to regain the ability to walk over ground, and therefore its effects on walking function, neuroplasticity and other functional capabilities were also determined. Lastly, we examined the excitability and consistency of various spinal reflexes in individuals with incomplete SCI.

1.2 Introduction to spinal cord injury

1.2.1 The spinal cord: anatomy and levels

The spinal cord is one of the two constituents of the central nervous system (CNS), the other being the brain, and is housed within the spinal canal of the vertebrae column. Spinal nerves pass through openings between each pair of vertebrae (i.e., the intervertebral foramina), bifurcate (now termed 'roots') and enter the spinal cord on the dorsal and ventral sides to bring in sensory information and send out motor commands, respectively.

The vertebrae column is composed of 7 cervical (C), 12 thoracic (T), 5 lumbar (L), 5 fused sacral (S) vertebrae (i.e., the sacrum) and 4 fused coccygeal (CO) vertebrae (i.e., the coccyx), with a single spinal nerve entering the spinal cord below its respective vertebra, except for the C1 vertebra which has 2 spinal nerves, each entering above and below it, and the coccyx which has only 1 spinal nerve. The spinal cord stretches from the C1 – L1 vertebrae, but spinal nerves continue to travel down the spinal canal as a collection, termed the cauda equina, and exit through the lumbar and sacral vertebrae. Since there are 8 cervical, 12 thoracic, 5 lumbar, 5

sacral and 1 coccygeal nerves entering the vertebral column, therefore there are 31 spinal segments that make up the spinal cord.

Injury at or above the T12 vertebra damages the spinal cord and results in 'upper motoneuron injury', while injury below the T12 vertebra damages the cauda equina and results in 'lower motoneuron injury'. However, the exact level resulting in upper motoneuron injury can vary between T10 and T12, and that the T12 cut off is not always the case. Due to the discrepancy between the location where the spinal cord ends and where the lumbar and sacral spinal nerves enter the vertebrae column, the distinction between the skeletal and neurological levels of injury is crucial (see below).

1.2.2 Spinal cord injury: complete vs incomplete

The completeness and severity of SCI is determined from sensory and motor tests, performed according to the International Standards for Neurological and Functional Classification of Spinal Cord Injury, developed by the American Spinal Injury Association (ASIA) (Burns et al. 2012; Maynard et al. 1997). Completeness of SCI (i.e., complete or incomplete) is determined by testing for the presence or absence of sensory and motor function at the S4-5 segments (i.e., sacral sparing): sensation at the anal mucocutaneous junction and in the rectum, and contraction of the external anal sphincter. A SCI is complete if sensation and motor function at the S4-5 segments are absent, otherwise the injury is incomplete. After testing for sacral sparing, sensory and motor exams are performed to determine the sensory, motor and neurological levels of injury.

The sensory exam determines the integrity of light touch and pinprick sensation, bilaterally along the 28 dermatomes. A piece of cotton, and the dull and sharp ends of a safety pin are used to determine if the sensation is absent (score: 0), impaired (score: 1) or normal (score: 2). For light touch sensation, a score of 0 represents no sensation, and a score of 1 represents sensation different from that on the face or another unaffected region of the skin. For pinprick sensation, a score of 0 represents the inability to distinguish the dull and sharp ends of the safety pin, while a score of 1 represents heightened pain (hyperpathia) or a painful response to an otherwise non-painful stimuli (allodynia), and 2 is normal sensation. The sensory level of injury is defined as the most caudal spinal segment with normal sensation of light touch and pinprick, bilaterally.

The motor exam determines the strength of 5 key muscles of the upper and lower limbs on both sides by assigning a score from 0-5 along a 6-point scale depending on the strength of the muscle (0 – total paralysis to 5 – normal strength). Upper limb muscles (and corresponding spinal segment) include: elbow flexors (C5), wrist extensors (C6), elbow extensors (C7), finger flexors, i.e., distal phalanx of middle finger (C8), and little finger abductor (T1). The lower limb muscles include: hip flexors (L2), knee extensors (L3), ankle dorsiflexors (L4), long toe extensors (L5) and ankle plantarflexors (S1). The motor level of injury is defined as the most caudal spinal segment with a motor score of ≥3/5 (defined as full range of motion against gravity), bilaterally, and that all more rostral segment muscles score 5 (normal).

In the case of complete SCI only, the Zone of Partial Preservation is defined as the spinal segments below the neurological level of injury with partial sensory or motor functions. The severity of the SCI is defined using the ASIA Impairment Scale (AIS) grade on a 5-point ordinal scale from A-E. Motor and sensory-complete SCI is defined as AIS grade A, while motor-complete and sensory-incomplete as AIS grade B. AIS grades C and D are defined as motor- and sensory-incomplete injuries, with motor scores of <3 and ≥3 on >50% of the 5 key muscles below the neurological level of injury, respectively. AIS grade E implicates normal sensory and motor function (Sipski et al. 2004).

The level of injury influences the functional impairments after the injury. Cervical injuries affect the stability of the trunk, and function of the upper and lower extremities resulting in tetraplegia or tetraparesis (also known as ‘quadriplegia’ or quadriparesis). Injuries along the length of the thoracic segments affect the trunk’s stability below the injury, and function of the lower extremities resulting in paraplegia or paraparesis.

1.2.3 Causes: traumatic vs non-traumatic

The mechanism behind traumatic SCI is an external force that factures the vertebrae column. The fracture, in turn, exerts pressure on the spinal cord, which causes mechanical deformation of the neural tissue (Ahuja et al. 2017; Miele et al. 2012). Deformation of the spinal cord can take place through tension, torsion or compression of the neural tissue. Hyperflexion, hyperextension and dislocation of the vertebrae result in tension. Extreme rotation leads to torsion, while axial

compression results in burst fracture, causing compression (Miele et al. 2012). Common causes of traumatic SCI are motor vehicle accidents, falls, sports and violence (Ahuja et al. 2017).

In contrast to traumatic SCI, non-traumatic SCI results from an internal disease process rather than external force to the vertebrae column (Ahuja et al. 2017; Grassner et al. 2016). Common causes of non-traumatic SCI are infections (e.g., spinal epidural abscess), inflammatory disease (e.g., transverse myelitis), autoimmune disease (e.g., multiple sclerosis), vascular disorders (e.g., ischemia, hemorrhage), motoneuron disease (i.e., Amyotrophic lateral sclerosis), cervical myelopathy, spinal stenosis, neoplastic disease (e.g., tumors) and genetic disorders (e.g., spinal muscular atrophy) (Grassner et al. 2016; New et al. 2014).

1.2.4 Statistics and demographics

The global incidence for traumatic SCI range from 15 to 40/million(M)/year (Lee et al. 2014), and for non-traumatic SCI range from 6 to 76/M/year (New et al. 2014). Furthermore, the incidence is the highest in North America for traumatic (40/M/year) and non-traumatic SCI (76/M/year), and are similar between Canada and the USA (Lee et al. 2014; New et al. 2014). In Canada, the annual cost of traumatic SCI is $2.67 billion, and life-time cost for an individual with the injury ranges from $1.47M (paraparesis) to $3.03M (tetraplegia) (Krueger et al. 2013).

The distribution of traumatic SCI across Canada mirrors the distribution of the population in the different provinces. That is, injuries are scattered across British Columbia, Alberta and Saskatchewan where the population is also wide spread, whereas injuries are confined to specific regions in Ontario, Quebec and Manitoba where the population is geographically confined to certain areas (Cheng et al. 2017). Facilities providing acute care and rehabilitation are located across the following eight provinces: British Columbia, Alberta, Saskatchewan, Manitoba, Ontario, Quebec, New Brunswick and Nova Scotia (Noonan et al. 2017).

The incidence of traumatic SCI in Canada ranges from 3.6 to 109.3/M/year (Jazayeri et al. 2015). The injuries are 1) cervical in majority, 2) predominantly caused by motor vehicle collisions, falls, sports and violence, and 3) approximately 3 times more common in males compared to females (Cheng et al. 2017; Couris et al. 2010; Dryden et al. 2003; Pickett et al. 2006). Sex differences also exist between the cause of the injuries, and these vary by age. For example, in

Alberta and British Columbia, incidence of traumatic injury due to motor vehicle collision are more common among males, while those due to falls are more common in older females (Dryden et al. 2003; Lenehan et al. 2012).

1.2.5 Sequence of interventions

Acute management of SCI is beyond the scope of this thesis, however, a summary is provided on the four key interventions during the acute management of traumatic SCI (see Ahuja et al. (2017) for details on the following). Apart from the primary damage to the spinal cord, further damage is avoided by maintaining the spine in a rigid posture along the craniospinal axis by placing the SCI patient on a flat surface, with a cervical collar. To prevent ischemic damage, hemodynamic factors are controlled by providing the patient with sufficient blood and oxygen supply, by maintaining systemic and mean arterial blood pressures, and by medication for deep vein thrombosis to avoid thromboembolism. Next, inflammatory reactions secondary to the mechanical damage to the spinal cord are addressed by providing the patient with methylprednisolone treatment (i.e., non-steroidal anti-inflammatory drug treatment), beginning as early as ≤8 hours of injury and lasting for 24 hours. The efficacy of treatment with methylprednisolone, however, is not fully clear and under debate (see Juknis et al. (2012) for details). Finally, surgery is performed to decompress the spinal cord, realign the fractured spine and stabilize it using metal implants and screws (≤8 or <24 hours after the injury).

1.2.6 Medical and other problems

Complications after SCI are numerous, and arise during the acute (Hagen 2015) and chronic (Sezer et al. 2015) stages of the injury. Medical complications are beyond the scope of this thesis, and therefore only a brief summary is provided on the following select number of complications and its management (see Juknis et al. (2012) for details on the following): pulmonary complications, autonomic complications, thrombosis, bone loss, hypercalcemia, heterotopic ossification and pressure ulcers.

Pulmonary complications after SCI arise from injuries along the cervical levels, resulting in bronchoconstriction, increase in mucus secretion, risk of developing respiratory infections (e.g., pneumonia, influenza), and phrenic nerve damage. Excess mucus is excreted by chest

percussions and postural drainage, while respiratory infections can be avoided by vaccinations on a yearly basis. Phrenic nerve damage and the resulting impairment and/or loss of function in the respiratory muscles and breathing is addressed by mechanical ventilation and phrenic nerve pacing.

Autonomic complications after SCI include autonomic dysreflexia (AD) and orthostatic hypotension. AD is observed in individuals with SCI above the T6 level, and more so in those with injuries that are complete. The trigger for AD is a noxious stimulus below the level of injury, which induces a sympathetic response of high blood pressure and vasoconstriction below the injury. This is counteracted by reduced heart rate and vasodilation above the level of injury to reduce the high blood pressure. Main symptoms of AD include sweating, headache and flushing, and is managed by sitting the patient upright, and removing the source of the noxious stimulus, e.g., compacted stool, full bladder, uncomfortable clothing, pressure ulcer or an ingrown toenail. Hypertensive medication is provided if the AD persists despite alleviating the noxious stimulus. Orthostatic hypotension is triggered by a sudden shift to an upright posture resulting in reduced blood pressure, and is managed by having the patient rise slowly when sitting up, wearing compression stocking, tilt table standing and medication.

Loss of mobility of SCI, especially after complete injuries, can lead to deep vein thrombosis and pulmonary embolism, which can be managed using compressive stocking and pneumatic compression, anticoagulant drugs and inferior vena cava filters.

Bone loss is observed one to two years after SCI, after which the rate of bone loss plateaus. Major sites of bone loss are the distal femur and proximal tibia. Management of bone loss and the resulting osteoporosis involves care in avoiding stress to the bone, e.g., during transfers and exercises, performing weight-bearing exercises, e.g., standing, treadmill walking and functional electrical stimulation (FES-) assisted cycling, and medication that inhibit bone resorption by the osteoclast bone cells.

Hypercalcemia, i.e., increase in blood calcium levels, results from the increase in bone resorption by the osteoclasts and from reduced secretion of calcium through the kidneys. Hypercalcemia is managed by hydration, loop diuretics and medication that inhibits the resorption of bone by osteoclasts.

Heterotopic ossification (HO) results in the formation of bone tissue at sights that otherwise do not have any ossified bone present, predominantly at the joints. Common joints affected by HO are the shoulders, elbows and knees, and the most common being the hips. Management of HO include exercises to improve the joints' range of motion, stretching, medication, radiation therapy and surgery.

Lastly, pressure ulcers, also known as bed sores, result from prolonged pressure on soft tissue that is located between a bony prominence and the external force, and leads to localized tissue damage. Common areas of the body affected by pressure ulcer are the lower back (i.e., sacrum), ischial tuberosity, greater trochanter, malleoli, heels and the back of the head (i.e., occiput). Management of pressure ulcer requires relieving pressure at these hot sports by shifting the body's weight every 2 hours, skin care, avoiding friction across the skin, balanced nutrition, and use of bed mattresses and wheelchair cushions that can distribute pressure.

1.2.7 Priorities after injury

After SCI, priorities for functional recovery are similar among those with tetraplegia and paraplegia, with the main exception of arm and hand function being the top priority in the former (Anderson 2004a; Simpson et al. 2012). Both groups prioritize sexual function, bowel and bladder function, stability of the trunk, walking, pain and sensation as important. The priorities are similar between those injured <3 years ago compared to those injured >3 years ago. Almost all of those affected by SCI (96.5%) recognize the importance of exercise for functional recovery (Anderson 2004a).

1.3 Neural control of walking

Among the top priorities after SCI is regaining the ability to walk (Simpson et al. 2012). However, before the effects of walking retraining on neuroplasticity and functional capabilities after SCI in humans can be determined, it is necessary to understand the neural control of walking. Walking is governed by three levels of neural control: supraspinal, spinal and sensory.

1.3.1 Supraspinal control: role of cortical and subcortical structures

Control of walking at supraspinal levels involves: 1) cortical structures including the primary motor cortex and the corticospinal tract, adjacent cortical areas including the supplementary motor area and premotor area, visual, parietal and temporal areas, and 2) subcortical structures including the midbrain (i.e., mesencephalic locomotor region, red nucleus), hindbrain (i.e., vestibular nucleus, reticular formation), cerebellum and basal ganglia (Field-Fote et al. 2017; Fukuyama et al. 1997; Suzuki et al. 2004).

1.3.1.1 Cortical structures

Corticospinal tract

Considerable information is available on the role of the primary motor cortex and the corticospinal tract in walking because of its accessibility for study. The corticospinal tract (CST) originates from the primary motor cortex and is involved in the generation of skilled walking movements, and activating the spinal locomotor circuitry. Evidence for the role of the CST in skilled motor tasks comes from training to make skilled dorsiflexion movements in humans, which increased the maximum motor evoked potential (MEP_{max}) from the tibialis anterior (TA) in response to transcranial magnetic stimulation (TMS) which activates the CST at the level of the motor cortex, but not after transcranial electric stimulation (TES) which activates the CST at the level of the axons (Perez et al. 2004). Damage to the CST in quadrupeds does not result in observable problems during level ground walking, but deficits are seen in skilled walking (Bolton et al. 2006). Furthermore, recordings from the forelimb area in the motor cortex of cats (Drew 1988) trained to walk on the rungs of a ladder of progressively smaller widths showed higher levels of activity as the stepping task required higher precision (Beloozerova et al. 2010). In humans, injury to the motor cortex, such as from stroke (Jorgensen et al. 1995), or from direct damage to the CST (Nathan 1994) leads to difficulty in walking. Contribution of the motor cortex and CST to walking is demonstrated by the task-dependent differences in the characteristics of the MEP recruitment curves, measured from the TA and SOL, during treadmill walking (early swing and early stance) and during isometric contractions (dorsi- and plantarflexion), which implicate the involvement of the circuitry within the motor cortex in modulating the activity of the CST during walking (Capaday et al. 1999). Contribution of the

motor cortex and CST to walking is also demonstrated by subthreshold TMS-induced cortical inhibition, which suppressed the TA electromyogram (EMG) by ~20% of the unperturbed EMG signal (Petersen et al. 2001). This suppression was not observed in response to subthreshold TES (Petersen et al. 2001), signifying that muscle activity is being activated at the level of the motor cortex and the CST.

Other cortical areas

Walking involves the integration of information from different cortical areas. To study the role of different cortical areas in walking, brain imagining techniques are used, e.g., functional magnetic resonance imaging, single photon emission computed tomography, either while a person is walking or imagines walking, or after walking. Role of sensory cortical areas was demonstrated during walking on a treadmill, which led to higher activation in the sensorimotor and the supplementary motor area, whereas imagining walking activated the primary and the supplementary motor areas only (Miyai et al. 2001). Walking over ground for 4 min was associated with higher levels of activity in the supplementary motor area, visual cortex and temporal lobe, in addition to the primary sensorimotor area, in uninjured participants after the walking [subcortical structures were also activated (see *Section 1.3.1.3 Subcortical structures*)] (Fukuyama et al. 1997). Differences in cortical activation are also observed between walking and running. Training to imagine walking and running revealed significantly greater activity in the parahippocampal and fusiform gyri (cortical areas in the temporal lobe) during walking, and the cerebellum (a subcortical structure) during running (Jahn et al. 2004), which suggests that the two tasks are controlled by different structures.

1.3.1.3 Subcortical structures

Rubrospinal, vestibulospinal and reticulospinal tracts

Motor tracts besides the CST, i.e., the rubrospinal, vestibulospinal and reticulospinal tracts, also play a role in walking, and originate from subcortical areas in the midbrain (red nucleus) and hind brain (vestibular nucleus, reticular formation). Early work on the role of the rubrospinal and vestibulospinal tracts in locomotion comes from Orlovsky and others, using thalamic cats walking with the hindlimbs on a treadmill, and the forelimbs resting on a stationary platform

(Orlovsky 1972a; b). Recordings made from the red nucleus to measure the activity of neurons in the rubrospinal tract (Orlovsky 1972a), and from the Deiter's nucleus (i.e., the vestibular nucleus) to measure neuronal activity in the vestibulospinal tract (Orlovsky 1972b) revealed high activity during the flexion and extension phases of the cat's hindlimb locomotion, respectively (i.e., in a phase-dependent manner). Furthermore, elevating the treadmill to increase the load on the hindlimb extensors reduced the activity of the rubrospinal neurons (Orlovsky 1972a), and increased the activity of the vestibulospinal neurons (Orlovsky 1972b), during the stance phase. Early work on the role of the reticulospinal tract in locomotion comes from Drew et al. (1986) using uninjured adult cats that have been trained to walk on a treadmill (Drew et al. 1986). Using recordings from the reticulospinal neurons in the medial reticular formation, and the flexor and extensor muscles of all four limbs, a relationship was observed between activity in the reticulospinal neurons and activity of the flexor and/or extensor muscles during the walking (Drew et al. 1986). It should be noted that the contribution from these tracts to locomotion in humans may not be as much compared to the contribution from the CST, since the prognosis for walking after incomplete SCI and stroke is poor if the integrity of the CST, estimated using MEP responses, is reduced (see Barthelemy et al. (2011) for review and details).

Cerebellum

The cerebellum also plays a role in walking and motor learning. The cerebellum is active after 4 min of over ground walking (Fukuyama et al. 1997), and shows higher activity during imagined running compared to imagined walking and standing (Jahn et al. 2004). From the studies by Orlovsky and others (Orlovsky 1972a; b), removing the cerebellum in the thalamic cats abolished the phase-dependent modulation of the rubrospinal (Orlovsky 1972a) and vestibulospinal tracts (Orlovsky 1972b), indicating its involvement with descending motor pathways. Stimulating the white matter along the midline of the cerebellum, i.e., the axons in the Hook bundle that originate in the fastigial nuclei, in decerebrate cats also induced locomotor activity in the fore- and hindlimbs on a moving treadmill, or enhanced the extensor activity in the limbs as part of postural adjustment (Mori et al. 1998). In the same study, Mori et al. (1998) showed that the cerebellar-induced locomotion was similar to that induced by stimulating the mesencephalic locomotor region (MLR), and that stimulating both resulted in faster locomotion compared to the speed after stimulating either regions separately.

The cerebellum is also involved in the motor learning, e.g., when adapting the legs to walk at different speeds on a split-belt treadmill. Compared to uninjured humans, those with global degeneration of the cerebellum were unable to adapt the step length, duration of double support and the phase-lag between the legs by the end of the split-belt walking, even though all participants were able to coordinate the legs well enough to walk on the treadmill (Morton and Bastian 2006).

Basal ganglia

The basal ganglia is involved in initiating movement through the direct pathway and inhibiting movement through the indirect pathway (see Grillner et al. (2005) for review, and details on the following). Cortical and thalamic activation of the directly pathway of the basal ganglia inhibits the pallidum (i.e., the output of the basal ganglia), thereby disinhibiting the MLR, which initials locomotion (Amemiya and Yamaguchi 1984; Yamaguchi 1992). Similarly, cortical activation of the indirect pathway of the basal ganglia disinhibits the subthalamic nucleus, which also receives direct activation from the cortex. The resultant activation of the subthalamic nucleus activates the pallidum, which inhibits the MLR. Furthermore, individuals with Parkinson's disease, wherein levels of dopamine in the basal ganglia are reduced (Lotharius and Brundin 2002), exhibit freezing of gait that is characterized by "brief, episodic absence or marked reduction of forward progression of the feet despite the intension to walk", and increasing the levels of dopamine reduces the freezing of gait (see Nutt et al. (2011) for review and details). This further implicates the role of the basal ganglia in the control of locomotion in humans. Direct evidence for the role of the MLR in initiating movement is observed in the decerebrate and spinal cat: stimulating the MLR induced alternating movements in the forelimbs on a moving and stationary treadmill (Amemiya and Yamaguchi 1984), in addition to fictive locomotion, measured using needle EMG (Yamaguchi 1992) and nerve recordings (Amemiya and Yamaguchi 1984) of the flexor and extensor muscles.

1.3.2 Spinal control: role of a central pattern generator

1.3.2.1 Animal studies

Early evidence for a central pattern generator (CPG), capable of producing and maintaining the alternating and rhythmic movements of the limbs during locomotion, comes from studies on animals with loss of supraspinal input over the spinal circuitry through decortication, decerebration and spinalization. In decerebrated, lower-thoracic spinalized cat, with the tested hindlimb fixed using steel clamps, the tibialis anterior and medial gastrocnemius muscles displayed rhythmic out-of-phase contractions, with and without deafferentation of the spinal roots (i.e., sensory feedback) (Brown 1911). Rhythmic alternating activity in flexor and extensor nerves and muscles is also observed in the forelimbs of a decerebrated and lower-thoracic spinalized cat, and induced by the movement of the treadmill belt or stimulation of the mid brain MLR (Amemiya and Yamaguchi 1984; Yamaguchi 1992). Fictive locomotion is also observed in the rat (Cazalets et al. 1992; 1994), lamprey (McPherson and Kemnitz 1994) and frog embryo (Soffe 1993). In these studies, sensory stimulation of the skin or the application of neurotransmitters, e.g., glutamate and serotonin, induced rhythmic activity in the motoneuron (MN) or myotome (Soffe 1993), and fictive locomotion in the ventral roots (Cazalets et al. 1992; McPherson and Kemnitz 1994). Furthermore, the fictive locomotion is also modulated by the inhibitory neurotransmitter GABA, which is able to reduce the activity in the ventral roots (Cazalets et al. 1994). For details on the cellular mechanisms of the CPG, see Kiehn et al. (2010) for review. In contrast, evidence for a CPG in non-human primates is less clear, with stepping-like movements over a treadmill in the spinalized monkey observed in some but not in others [see Vilensky and O'Connor (1998) for review].

1.3.2.2 Human studies

Evidence for a CPG in humans comes from observations of rhythmic alternating movements of the trunk and leg muscles (Nadeau et al. 2010) and stepping movements after complete and incomplete SCI (Bussel et al. 1988; Calancie et al. 1994; Holmes 1915; Kuhn 1950) wherein descending cortical drive from the developed CST over the spinal circuitry is interrupted (Nathan 1994), and from infants (Peiper 1963; Yang et al. 1998a) wherein the cortical drive is weak because the CST is still developing (Huttenlocher 1979).

The involuntary movements after SCI involve: repetitive movements in the spinal extensor and abdominal muscles (Nadeau et al. 2010), flexor or extensor muscles of the leg, as well as alternating movements of the legs on both sides, are slow and reach frequencies of 0.6 or 0.9 Hz (Bussel et al. 1988; Calancie et al. 1994; Holmes 1915; Nadeau et al. 2010). These movements are typically observed in the supine position, and modulated by sensory stimulation at the foot (Bussel et al. 1988), by pin prick stimulation to the inner surface of the thigh or perineum (Holmes 1915), by changing the position of the neck and toes, and emptying the bladder (Calancie et al. 1994) and by extending the hip or pinching the skin on the back (Nadeau et al. 2010). Repetitive movements of the leg muscles are also observed during sleep (Lee et al. 1996), but not always (Bussel et al. 1988; Calancie et al. 1994). Lastly, the stepping movements after SCI are also observed during standing or treadmill walking with 60% of body weight-support, are able to adapt to the speed of the treadmill, and stop once the legs are completely unloaded (Calancie et al. 1994). Infants as young as 1 year of age or less are also capable of performing stepping movements on a treadmill with alternating activity in the flexor and extensor muscles, and are able to adjust the walking speed to that of the treadmill (Yang et al. 1998a).

Further, evidence of a CPG network in adult humans comes from studies on magnetic and epidural electrical stimulation of the spinal cord in uninjured and SCI participants, respectively. In uninjured participants, spinal electromagnetic stimulation between the T11-T12 vertebrae (3 Hz at 50% of the maximum stimulator output [MSO]) induced alternating flexion and extension movements of the hip and knee, respectively, that were not observed when stimulation was applied between the T12-L1, L1-L2 and L2-L3 vertebrae (Gerasimenko et al. 2010). Similarly, in participants with motor and sensory complete SCI (AIS A), epidural electrical stimulation of the L2 spinal segment (25-50 Hz between 5-9 V) induced alternating flexor-extensor stepping-like movements in the leg (Dimitrijevic et al. 1998). Therefore, both findings suggesting that the human CPG is located between the lower thoracic and higher lumbar spinal segments.

The human CPG network is capable of interacting with sensory feedback. In the same study on electromagnetic stimulation of the spinal cord in uninjured participants by Gerasimenko et al. (2010), vibration to the hip flexor and knee extensor tendons during the spinal stimulation augmented the leg movements compared to the movements without the tendon vibration (Gerasimenko et al. 2010), suggesting that sensory feedback plays a role in modulating the

output of the CPG network. As described in the next section, sensory feedback is crucial in coordinating the step cycle and avoiding obstacles in order to maintain balance while walking.

1.3.3 Sensory control: role of reflex afferents

Sensory feedback from afferents of the leg muscles (Ia afferents), tendons (Ib afferents) and skin (cutaneous afferents) play a role in walking by coordinating the phases of the step cycle and the transition between the phases, clearing obstacles that could otherwise result in loss of balance while walking (Nielsen and Sinkjaer 2002; Pearson 2008; Zehr and Stein 1999), and generating the EMG activity in the leg muscles that contribute to the walking (Dietz et al. 1992; Pearson 2004; Sinkjaer et al. 2000; Sinkjaer et al. 1996; Van Wezel et al. 1997). In humans, the role of sensory feedback has been studied by either superimposing the studied sensory input (e.g., electrically stimulating a nerve) or removing the sensory input (e.g., unloading the limb) (Nielsen and Sinkjaer 2002).

1.3.3.1 Ia afferents

Ia afferents originate from stretch receptors (i.e., muscle spindles), located within skeletal muscles, and relay sensory feedback on muscle stretch. As the Ia nerve endings wrap around the muscle fibers within the spindles, stretch to the muscle fibers activates the monosynaptic Ia-motoneuron reflex loop (Magladery et al. 1951) which leads to contraction of the stretched muscle (Grey et al. 2008). During locomotion, Ia afferents in the plantarflexors play a role in the production of force during the stance phase to aid in propelling the body forward.

Evidence for the role of Ia afferents in the production of force comes from studies on the excitability of the stretch reflex (Grey et al. 2008) and its electrophysiologic analogue: the Hoffmann reflex (H-reflex) (Magladery et al. 1951) in the ankle plantarflexors. Throughout the stance and swing phases, the excitability (i.e., gain) of the stretch reflex and H-reflex is modulated in a phase-dependent manner (Capaday and Stein 1987; Sinkjaer et al. 1996; Yang et al. 1991). This is observed during walking and running (Capaday and Stein 1987). During the swing phase, the leg is off the ground, the gain of the reflex pathway is low and minimal EMG activity is recorded from the plantarflexors (Capaday and Stein 1987; Sinkjaer et al. 1996). During the stance phase, the leg is on the ground as it is supporting the body's weight, the gain

of the reflex pathway increases (Capaday and Stein 1987; Sinkjaer et al. 1996; Yang et al. 1991) and the ankle plantarflexors undergo lengthening contraction (Capaday and Stein 1987). The increased gain of the reflex pathway in conjunction with muscle lengthening produce reflexive torque (Grey et al. 2008; Stein and Kearney 1995) necessary to push the body forwards.

1.3.3.2 Ib afferents

Ib afferents originate from muscle tendons as the Ib nerve endings wrap around the tendon's collagen fibers, and relay sensory feedback on muscle tension (i.e., load) (Dietz et al. 1992). The effects of Ib afferents are task-dependent, providing negative and positive feedback depending on whether the muscle is at rest (e.g., quiet sitting), or active during a task that is non-functional (e.g., stretch reflex, standing) or functional (e.g., locomotion) (Pearson and Collins 1993; Stephens and Yang 1996). During walking, Ib afferents play a role in modulating the duration of the stance phase and the activity of the extensor muscles, as shown in animal work (see Pearson (2008) for review and details on the following). In cats spinalized at the T13 level, electrical stimulation of the plantaris nerve (composed of Ia and Ib afferents) enhanced the amplitude and duration of the EMG activity from the medial gastrocnemius (Pearson and Collins 1993). However, this effect was not observed when the plantaris muscle was vibrated, thereby ruling out the contribution of Ia afferents to the positive feedback on the medial gastrocnemius muscle (Pearson and Collins 1993).

Evidence for the role of Ib afferents during human walking comes from studies on the effects of loading and unloading during the stance phase of walking, on the duration of the stance phase and activity in the leg extensor muscles. Work with infants ≤12 months of age illustrate the effect of loading during supported treadmill stepping (Pang and Yang 2000; Yang et al. 1998b): 1) when load is applied by pressing down at the hips, the duration of the stance phase and the step cycle is increased, and the stance-to-swing transition occurs at a more extended hip angle than in steps without loading, and 2) when the load is reduced during walking by sliding the foot backwards or forwards, the duration of the stance phase is reduced (backwards disturbance) or the stance-to-swing transition is prompted at a hip angle of higher flexion (forward disturbance) compared to steps without the disturbance. In adults, applying load during the stance phase of walking, using weights placed at the hips, increased the: 1) amplitude of the EMG activity in the soleus (SOL, ankle plantarflexor) and vastus lateralis (knee extensor), 2) duration of the SOL

EMG activity, and 3) the duration of the stance phase, while having the opposite effect during unloading by lifting the body (Stephens and Yang 1999). The inhibitory effect of unloading on SOL EMG during walking is attributed to reduced positive feedback from Ib afferents, and not from the reciprocal inhibition of the SOL from the common peroneal nerve (during stretch of TA), nor from Ia afferents. Blocking transmission in these nerves did not affect the inhibition of the SOL EMG during unloading (Sinkjaer et al. 2000).

1.3.3.3 Cutaneous afferents

Cutaneous afferents [i.e., A-β sensory fibers (van der Laan et al. 2000)] originate from sensory receptors distributed within the dermis of the skin, and play a role in obstacle avoidance and maintaining balance during walking. Role of cutaneous reflexes during locomotion was studied in spinal cats walking on a treadmill, wherein stimulation of the foot dorsum resulted in a flexor reflex in the entire leg during the swing phase, and in knee extension during the stance phase (Forssberg et al. 1975). Evidence for the role of cutaneous afferents during human walking comes from studies on the effects of electrical stimulation of three nerves supplying the foot, i.e., the sural, posterior tibial and superficial peroneal nerves, on 1) reflex responses from muscles and 2) changes in the joint angles, of the legs.

The posterior tibial nerve innervates the medial and plantar surfaces of the foot. Stimulation of the posterior tibial nerve during the stance-to-swing transition, early and mid swing activates the TA, and induces dorsiflexion (Duysens et al. 1990; Yang and Stein 1990; Zehr et al. 1997). Similarly, knee flexors are activated during mid swing, and induces flexion at the knee (Zehr et al. 1997). These flexor responses serve to move the foot away from an obstacle. In contrast, stimulating the posterior tibial nerve during the late swing inhibits the TA (Van Wezel et al. 1997; Yang and Stein 1990), activates the SOL and gastrocnemii, and induces plantarflexion (Zehr et al. 1997). These responses ensure the foot is placed on the ground as the leg enters the stance phase. Stimulation during early stance also activates the gastrocnemii and induces plantarflexion, thereby placing the foot on the ground, while stimulation during mid stance does not induce any response from the gastrocnemii and plantarflexion is reduced (Duysens et al. 1990). This ensures that the foot is stable on the ground while the contralateral leg is in swing phase. Lastly, stimulating the posterior tibial nerve during late stance inhibits the SOL (Yang and Stein 1990), and ensures that the leg is stable to prevent loss of balance.

The sural nerve innervates the lateral border of the foot. Similar to the posterior tibial nerve, stimulating the sural nerve during early swing activates the TA (van der Laan et al. 2000), and induces dorsiflexion (Van Wezel et al. 1997), hence serving to move the foot away from an obstacle. Stimulating it during late swing inhibits the TA (van der Laan et al. 2000) and induces plantarflexion (Van Wezel et al. 1997), serving to place the foot on the ground for stability. Stimulating the sural nerve during mid swing also induces flexor responses in the leg, i.e., knee and ankle flexion (Zehr et al. 1998), again serving to move the leg away from an obstacle. During late stance, stimulating the sural nerve induces flexor responses in the leg joints, i.e., hip, knee and ankle flexion, and ankle eversion (Zehr et al. 1998), serving to move the leg away from an obstacle and promoting the transition from stance to swing phase.

The superficial peroneal nerve innervates the foot dorsum. Stimulating the superficial peroneal nerve during early and late swing phase activates the knee flexors and induces flexion at the knee, and inhibits the TA and induces plantarflexion (Van Wezel et al. 1997; Zehr et al. 1997). During the mid and late swing, stimulating the superficial peroneal nerve also activates the triceps surae (i.e., SOL, gastrocnemii) (Zehr et al. 1997). Given that the superficial peroneal nerve innervates the foot dorsum, these responses serve to move the foot away from the obstacle.

1.4 Problems with walking after spinal cord injury

SCI results in the disruption of descending drive (Nathan 1994) and maladaptive neuroplasticity (Roy and Edgerton 2012), resulting in reduced strength of the muscles (Kim et al. 2004) and spasticity (Hubli et al. 2012), both of which lead to impaired walking ability (Hubli et al. 2012; Kim et al. 2004). Therefore, in order to study the effects of retraining walking after SCI, it is pivotal to first understand how walking is affected by the impaired descending drive and spasticity.

1.4.1 Altered descending drive and impaired walking

The effect of SCI on the integrity of descending pathways (Nathan 1994) is reflected in the: 1) reduced strength of the affected muscles, 2) reduced common drive to the motoneuron, and 3) reduced amplitude, and higher threshold and latency of the TMS-induced MEP responses. The

reduced descending drive is associated with reduced walking ability and maladaptive neuroplasticity in the spinal cord, as detailed below.

Muscle strength after SCI is estimated based on the patient's ability to contract a muscle and move the joint through a full range of motion against varying levels of resistance (Maynard et al. 1997). Muscle strength is reduced after the injury (Gorassini et al. 2009; Kim et al. 2004; Yang et al. 2011), and the degree of reduction is associated with reduced walking speed (Kim et al. 2004; Yang et al. 2011).

The altered descending drive after SCI reduces the common drive to the motoneuron. During walking, measures of common synaptic drive to muscles can be estimated using coherence and cross-correlation between two simultaneously recorded EMG signals, and represent the association between the two signals in the frequency and time domains. EMG signals recorded from the TA during walking in SCI participants showed reduced coherence and cross-correlation, compared to the uninjured participants (Barthelemy et al. 2010). Consequently, the altered descending drive after SCI leads to impaired walking ability. In the study by Barthelemy et al. (2010), reduced coherence between the TA EMG signals correlated with reduced elevation of the toe during the early-to-mid swing phase of walking (Barthelemy et al. 2010).

Compared to uninjured participants, reduced integrity of the CST after SCI is reflected in: 1) higher MEP threshold (Barthelemy et al. 2010; Cariga et al. 2002) and latency (Barthelemy et al. 2010; Cariga et al. 2002; Ertekin et al. 1998) and 2) reduced amplitude, or complete abolishment of the MEP (Barthelemy et al. 2010; Bjerkefors et al. 2015; Ellaway et al. 2004; Ertekin et al. 1998). Reduced amplitude of the MEP from the TA is associated with reduced walking ability, i.e., decrease in walking speed and reduced elevation of the toe during the early-to-mid swing phase of walking (Barthelemy et al. 2010).

The altered supraspinal drive after SCI is associated with neuroplasticity within the spinal cord, including: 1) heightened excitability of the motoneuron (Gorassini et al. 2004), and 2) reduced reciprocal inhibition (Boorman et al. 1996), presynaptic inhibition (Faist et al. 1994), polysynaptic inhibition (Jones and Yang 1994; Norton et al. 2008) and post-activation depression (Hultborn et al. 1996; Nielsen et al. 1993b), resulting in hyperexcitability of spinal reflexes that constitute spasticity.

1.4.2 Spasticity and impaired walking

A common sequelae to the altered descending drive is spasticity, manifesting as hyperexcitable spinal reflexes and involuntary muscle contractions called spasms (Benz et al. 2005). The severity of spasticity in a person can vary throughout the day (i.e., in the short-term) (Little et al. 1989), and across days (i.e., in the long-term) (Priebe et al. 1996). The effects of spasticity can be problematic, affecting the activities of daily living, sleep, transfers, skin integrity, pain (Johnson et al. 1998; Levi et al. 1995; Little et al. 1989; Skold et al. 1999), however, spasticity can also be used to advantage, for example in changing clothes, relieving pressure and during transfers (Little et al. 1989). Finally, similar to muscle weakness, spasticity after SCI is also associated with reduced walking ability. For example, hyperexcitability of a cutaneomuscular reflex in the TA, measured in the supine position, was correlated with lower walking speed after SCI (Hubli et al. 2012).

Hyperexcitable spinal reflexes can be mono- or polysynaptic, and the reflex excitability is attributed to maladaptive neuroplasticity within the nervous system (i.e., central factors) and changes in muscle-tissue properties, resulting from changes in: muscle-fiber type (Olsson et al. 2006), composition of the connective tissue (Olsson et al. 2006) and 3) increase in joint stiffness (de Vlugt et al. 2012; Mirbagheri et al. 2001) (i.e., peripheral factors).

1.4.2.1 Monosynaptic reflexes

Reflex manifestations of the monosynaptic Ia-motoneuronal pathway include the stretch and H-reflexes.

Stretch reflex and H_{max}/M_{max} ratio

Rapid stretch to the ankle plantarflexors elicits the stretch reflex, whose onset latency is consistent with a monosynaptic reflex pathway (Magladery et al. 1951), and becomes hyperexcitable after SCI (Grey et al. 2008). The monosynaptic Ia-motoneuron pathway is also activated by surface electrical stimulation of the muscle's peripheral nerve, thus bypassing the muscle spindles (Magladery et al. 1951; Schindler-Ivens and Shields 2000). Stimulating the nerve elicits the monosynaptic H-reflex and the direct motor (M-) response (Capaday and Stein 1987; Schindler-Ivens and Shields 2000). At progressively higher stimulus intensities, the peak-

to-peak amplitude of the H-reflex increases until the maximum H-reflex (H_{max}) is reached and decreases afterwards, while the amplitude of the M-response increases to a plateau (i.e., M_{max}) (Stein et al. 2007). The increase in the amplitude of the H-reflex is attributed to the activation of Ia afferents and recruitment of MNs, which increase the size of the orthodromic volleys that reach the muscle. As the stimulus intensity continues to increase, activation of MN at the level of the axon generates antidromic volleys which collide with the orthodromic volleys and reduce the size of the H-reflex. The H_{max}/M_{max} ratio represents the maximal recruitment of MNs through activation of Ia afferents, and increases after SCI (Little and Halar 1985; Shemesh et al. 1977). The increase in the H_{max}/M_{max} ratio indicates an increase in the recruitment of the MNs due to reduced inhibition of the Ia afferents and increase in the excitability of the MN after SCI [see D'Amico et al. (2014) and Roy and Edgerton (2012) for review], and therefore could potentially be used to gauge the excitability of the Ia-motoneuronal pathway.

Clonus

The stimulus necessary to induce clonus is an abrupt stretch to the muscle, and results in involuntary, repetitive muscle contractions in the muscle after SCI (Benz et al. 2005). Studies on the central mechanisms for clonus suggest the role of an intrinsic spinal circuitry generating the alternating rhythmic muscle contractions (Beres-Jones et al. 2003; Dimitrijevic et al. 1980b; Walsh 1976). In contrast, studies on peripheral mechanisms suggest that clonus results from hyperexcitable stretch reflexes repeatedly activated by the recurrent stretch of the muscle (Hidler and Rymer 1999; Rossi et al. 1990; Wallace et al. 2012), and exacerbated by the increase in joint stiffness (de Vlugt et al. 2012). Differences in the mechanisms behind clonus suggest that spasticity is mediated by factors that are both internal and external to the nervous system.

1.4.2.2 Polysynaptic reflexes

Reflex manifestations of hyperexcitable polysynaptic pathways after SCI include: spasms, hyperexcitable flexor reflexes, and reduced rate-dependent depression of the H-reflex.

Spasms and flexor reflexes

Muscle spasms are involuntary, sustained contractions of the flexor and/or extensor muscles, typically induced by innocuous stimulation of the skin (e.g., rubbing, cold touch, vibration, pin

prick) and/or proprioceptive stimulation (e.g., movement of the limb), and can last ≥1 second (Benz et al. 2005; Gorassini et al. 2004; Little et al. 1989; Norton et al. 2008). Similar responses in the form of hyperexcitable flexor reflexes are also induced by electrical stimulation of cutaneous nerves innervating the foot on the lateral (Shahani and Young 1971), dorsal (Butler et al. 2006), and medial-plantar surfaces (Jones and Yang 1994; Norton et al. 2008). The heightened reflex excitability is attributed to reduced inhibition of polysynaptic pathways (Bos et al. 2013; Jones and Yang 1994; Norton et al. 2008), and increased excitability of the motoneuron (Boulenguez et al. 2010; Gorassini et al. 2004).

Rate-dependent depression of the H-reflex

H-reflexes induced repetitively at interpulse intervals of <5 seconds, are reduced in amplitude compared to the first unconditioned H-reflex, and the amount of depression increases as the interpulse interval decreases (Stein et al. 2007). This form of inhibition is known as the rate-dependent depression of the H-reflex (RDD) and may represent the strength of inhibitory pathways within the spinal cord (Bos et al. 2013; Butler et al. 2006; Kakinohana et al. 2012; Kapitza et al. 2012). After SCI, RDD is reduced (Schindler-Ivens and Shields 2000). Mechanisms of reduced RDD are of central and peripheral origins, and may involve: 1) reduced post-activation depression of the Ia afferents (Grey et al. 2008; Hultborn et al. 1996), 2) reduced presynaptic inhibition (Kakinohana et al. 2012; Kapitza et al. 2012), 3) reduced polysynaptic inhibition (Bos et al. 2013; Butler et al. 2006), 4) increased excitability of the motoneuron (Boulenguez et al. 2010), 5) dopaminergic pathways (Liu et al. 2010), and 6) dysregulation of the gap junction protein connexin-36 (Yates et al. 2008). Furthermore, the reduced RDD is correlated with higher fatigability of the muscle (Schindler-Ivens and Shields 2000).

1.5. Training to restore walking

1.5.1 Predicting long-term walking function immediately after spinal cord injury

Numerous factors immediately after SCI can predict walking ability in the future: injury severity including the AIS grade, muscle strength, balance, sensation, reflexes, spasticity and age. In contrast, the cause of the injury, i.e., traumatic or non-traumatic, has no effect on the prognosis of walking, whereas conflicting findings point to the effects of sex-based differences on long-

term walking ability. Lastly, only a limited number of studies report the factors that predict improvements in walking after a physical rehabilitation program to retrain walking (e.g., walking on treadmill and over ground) after SCI. For review and details, see Burns et al. (2012) and Scivoletto and Di Donna (2009).

1.5.1.1 Injury severity and grade

Incomplete injuries including AIS grades C and D within 72 hr or 1-month after injury predict better walking function at discharge or 6 months post injury, compared to complete injuries and AIS grades A and B. At 1-month post injury, ≤25% of individuals with motor-complete SCI improved in scores on the Lower Extremity Motor Score (LEMS), and the scores on the Walking Index for Spinal Cord Injury Version 2 (WISC-II) and the 10-Meter Walk Test (10MWT) at self-selected speed, compared to ≥76% among those with incomplete injury, at 6 months post injury (Wirz et al. 2006). Individuals with AIS grade D injury within 72 hours after injury were more likely to improve walking ability compared to AIS C at the time of discharge (Burns et al. 1997; Kay et al. 2007), which in turn were more likely than AIS A and B (Kay et al. 2007). Furthermore, individuals classified as AIS C at the time of admission were also more likely to reclassify to AIS D at the time of discharge, compared to AIS A, B and D, which predominantly remained at the same grade (Scivoletto et al. 2004a).

1.5.1.2 Muscle strength and balance

Muscle strength immediately after injury can predict future walking function. A score of 2/5 on the Manual Muscle Test (MMT) of the stronger quadricep muscles within 1 week after onset of Frankel grade C SCI (analogous to AIS C SCI) predicted home and community walking at 2-months post injury (Crozier et al. 1992).

Muscle strength and balance during the chronic stage of SCI can also predict current walking capabilities. For example, MMT score of the less affected hip flexor muscles at ≥1 year after motor-incomplete SCI predicted walking speed and distance (normalized to the length of the leg) (Kim et al. 2004). Furthermore, higher scores on the LEMS of the proximal muscles, Upper Extremity Muscle Strength, and the Berg Balance Scale are associated with higher scores on the

10MWT, 6-Minute Walk Test (6MWT), Walking Index for Spinal Cord Injury Version 1 (WISCI) and the Timed Up and Go test (TUG) (Scivoletto et al. 2008).

1.5.1.3 Sensation, reflexes and spasticity

The ability to discriminate between light touch and pinprick sensation, absence and presence of certain spinal reflexes, and reduced spasticity are associated with greater walking ability. Presence of pinprick-light touch discrimination at >50% of L2-S1 dermatomes within 72 hours after AIS grade B SCI predicted walking at 26 and 52 weeks after injury (Oleson et al. 2005), which supports the role of sensory feedback from the skin in the control of walking after SCI (Bussel et al. 1988; Holmes 1915). The presence of the crossed-adductor and patellar reflexes at ≤20 days after motor-complete and incomplete SCI predicted higher reclassification to motor-incomplete status at >12 months after injury (Calancie et al. 2004). Furthermore, absence of the deep plantar reflex at the onset of injury predicted better ambulation at discharge from rehabilitation 6-8 weeks after injury (Ko et al. 1999). Lastly, lower scores on the Modified Ashworth Scale (MAS) from the leg muscles after chronic SCI correlated with higher scores on TUG and 6MWT (Scivoletto et al. 2008). These findings highlight that the excitability of spinal reflexes and spasticity after SCI also affects walking function (Hubli et al. 2012).

1.5.1.4 Age

Younger age (<50 years) at the time of injury is associated with a better prognosis of walking function. Improvements in the ASIA motor scores and scores on the WISCI at discharge from inpatient rehabilitation were higher in younger SCI participants (Scivoletto et al. 2003). A higher portion of the younger participants with AIS grade C injury became ambulatory at discharge, compared to the older participants (Burns et al. 1997). Similarly, in a different study, a greater portion of the younger participants with AIS grade D injury became ambulatory at discharge (Kay et al. 2007). The total number of reclassifications, and the number of reclassifications from AIS grades A, B and C to D were higher among the younger participants at discharge (Scivoletto et al. 2004a). Lastly, lower age is associated with higher scores on the 6MWT among participants with chronic SCI (Scivoletto et al. 2008). Therefore, lower age at the onset of SCI is associated with a better prognosis for walking function in the future, possibly due to a lower

frequency of complications during hospitalization in the younger participants (Scivoletto et al. 2003).

1.5.1.5 Cause: traumatic vs non-traumatic

The cause of SCI, whether traumatic or non-traumatic, did not affect long-term walking ability after AIS grade A, B, C and D injuries. The reclassification of AIS grade at discharge from acute rehabilitation did not differ between those with traumatic and non-traumatic injuries (Scivoletto et al. 2004a). The duration and cost of inpatient rehabilitation was higher for traumatic injury, however, the motor score on the Functional Independence Measure (FIM) scale at admission and the change in the motor score, when normalized to the duration of the inpatient rehabilitation, did not differ between those with traumatic and non-traumatic injuries (McKinley et al. 2001).

1.5.1.6 Sex-based differences

Effects of a participant's sex on the prognosis of their motor function after SCI are conflicting. Scivoletto et al. (2004b) report no sex-based differences in changes to the ASIA motor scores and scores on the WISCI between admission to and discharge from inpatient rehabilitation, whereas Sipski et al. (2004) observed sex-based differences: 1) the increase in ASIA motor scores between onset of injury and 1-year follow up after motor-complete SCI was higher in women, 2) the ASIA motor scores at 1-year follow up after motor-incomplete SCI was higher in women, and 3) the FIM motor scores at discharge from in- and outpatient rehabilitation were higher in women after motor-incomplete SCI, and in men after motor-complete SCI. However, in the same study, sex-based differences were non-significant for the: 1) reclassification of the AIS grade and 2) FIM motor scores at 1-year follow up (Sipski et al. 2004).

1.5.1.7 Predicting walking ability after walking retraining

Individuals with SCI respond differently to walking retraining (Louie et al. 2015; Yang and Musselman 2012), and therefore, it is necessary to improve our understanding of the factors that affect the outcome of walking retraining before the training itself (Kim et al. 2004; Yang et al. 2011). However, studies on the use of pretraining measures to predict improvements in walking ability after walking retraining during the acute and chronic stages of SCI are limited. After combining body weight-supported treadmill training (BWSTT) with manual assistance, body

weight-supported treadmill training with the Lokomat (henceforth, referred to as Lokomat training) and over ground walking for 3 months after motor-incomplete SCI (average ± SD of onset of injury: 1.36 ± 1.23 years), Winchester et al. (2009) concluded, based on participants with AIS C SCI for ≤ 5 years, the following four measures were able to predict over ground walking speed after the training: 1) pretraining 10MWT at a self-selected speed, 2) spasticity, 3) voluntary bowel and bladder voiding and 4) the square root of the time since injury. Similarly, retraining walking for an average duration of 4.5 months using BWSTT and, when possible, over ground walking in participants with motor-incomplete SCI for ≥ 7 months, Yang et al. (2011) concluded that the pretraining MMT score of 30/80 or higher in the hip flexor and extensor, hip abductor and adductor, knee flexor and extensor, and the ankle dorsiflexor and plantarflexor muscles, bilaterally, predicted improvements in over ground walking speed after the training.

1.5.2 Over ground walking

Retraining walking over ground >1 year after SCI, i.e., during the chronic stage of the injury, improves walking ability. Over ground walking training for 8-15 weeks improved the over ground walking speed (Gollie et al. 2017; Sharp et al. 2014). Similarly, 13 weeks each of over ground walking training, BWSTT and conventional physical therapy (e.g., walking, balance, stretching, aerobics) significantly improved over ground walking speed and distance (Alexeeva et al. 2011). Over ground walking training combined with FES to the common peroneal nerve, BWSTT with and without the FES, and Lokomat training, each for 12 weeks, led to significant improvements in walking speed and distance, with the largest effect size observed after the FES-assisted over ground walking training (Field-Fote and Roach 2011). A systematic review of randomized controlled trials comparing the effects of overground walking training with BWSTT, and overground walking training with robotic-assisted gait training revealed overground walking training as a superior method of walking training in terms of walking speed and distance, although the participants in the thirteen trials included in the review were in the acute and chronic stages of SCI (Mehrholz et al. 2017).

1.5.3 Skilled over ground walking

Some of the tasks necessary in daily life involve skilled over ground walking, e.g., navigating uneven surfaces outdoors such as ramps, slopes, grass and concrete, stepping over obstacles,

stepping up and down stairs and curbs, walking in crowded environments, while carrying objects and in narrow spaces, and using the intersection to cross a street (Musselman and Yang 2007). Physical training programs incorporating skilled over ground walking, in- and outdoors, improve walking and reduce the dependence on walking aid. In a case-study of a single participant with AIS D injury at the C5-6 level, BWSTT, over ground walking and skilled over ground walking for 9 weeks enhanced the self-selected and fast-paced over ground walking speed, daily step count and WISCI-II score (Behrman et al. 2005). In another case-study (Musselman et al. 2009), 4 participants with AIS C injuries spanning C5 to L1 levels, underwent 3 months of BWSTT and 3 months of skilled over ground walking training (stepping up stairs and curbs, stepping over obstacles, walking on uneven surfaces [i.e., slopes], opening and closing doors, walking in crowded and narrow spaces, walking while carrying objects and crossing intersections). Both training methods improved the 10MWT and 6MWT, however, the improvements were greater after the skilled training (Musselman et al. 2009).

Skilled over ground walking training in a specific task, e.g., stepping over obstacles, also improves over ground walking function. In a 2 × 2 cross-over design, training to step over obstacles along a 10 m pathway for 30 min improved the 10MWT and TUG (i.e., balance) scores, compared to walking back and forth without the obstacles for the same duration (wash out period: 2 days) (Pramodhyakul et al. 2013). In a different study using the same design, training to step over obstacles and on to targets over ground, versus BWSTT each for 2 months (wash out period: 2 months) significantly improved the 10MWT, 6MWT and the Spinal Cord Injury-Functional Ambulation Profile (SCI-FAP) scores (i.e., walking skill) after both forms of training (Yang et al. 2014).

1.5.4 Body weight-supported treadmill training with manual assistance

An alternative to retraining walking over ground is walking on a treadmill, with body weight-support provided by an overhead harness and a system to offset the body's load onto the legs. Walking is performed using active volition of the leg muscles and manual assistance provided by trainers on each side. The manual assistance involves lifting the foot (i.e., knee flexion and ankle dorsiflexion) during the swing phase to avoid toe-drag, and extending the knee during the stance phase to prevent knee buckling (Thomas and Gorassini 2005; Wernig et al. 1999). The goals of BWSTT are to promote safe walking on the treadmill with proper leg movements (i.e., no toe-

drag and knee buckling) for longer durations with minimum rests, whilst reducing the body weight-support to enable the legs to withstand higher loads, and increasing the treadmill speed (Yang et al. 2014). Early work on BWSTT showed that walking on the treadmill with body-weight support compared to no weight support, in participants with spasticity and paresis, improved the timing and amplitude of the EMG activity from the leg muscles, the joint kinematics and speed, as well as reduced fatigue during the treadmill walking (Visintin and Barbeau 1989). BWSTT also improves walking speed and distance on the treadmill with reduced need for body weight-support, and improves walking speed and distance over ground with reduced need for assistance using walking aids and trainers, after chronic SCI (≥ 7 months post injury) (Hicks et al. 2005; Thomas and Gorassini 2005; Trimble et al. 2001). Note that among the four participants in the Trimble et al. (2001) study, only one was 3 months post injury, however the improvements were observed across all participants.

Studies comparing the effects of BWSTT with conventional physical therapy and over ground walking suggest that the effects on walking outcome are comparable between the three forms of training. Fast-paced over ground walking speed improved significantly after BWSTT, over ground walking and conventional physical therapy in chronic SCI participants (≥ 1 year post injury), however, a greater proportion of the participants from the over ground walking and BWSTT groups became community walkers after the training (over ground walking speed post training >0.4 m/s) (Alexeeva et al. 2011). BWSTT when combined with FES to the common peroneal nerve for 12 weeks led to significant improvements in over ground walking speed and distance, while BWSTT without FES for the same period improved the walking speed only, in chronic SCI participants (≥ 1 year post injury) (Field-Fote and Roach 2011). However, as mentioned previously (Section *1.5.2 Over ground walking*), in the recent systematic review by Mehrholz et al. (2017), BWSTT did not compare favorably in terms of walking speed and distance compared to over ground walking training, although the review was based on data obtained from participants during the acute and chronic stages of SCI.

Reports on the efficacy of BWSTT compared to skilled over ground walking after chronic SCI (≥ 7 months post injury) are mixed. In the study by Musselman et al. (2009), improvement in the 10MWT and 6MWT were higher after the skilled walking training than BWSTT (training at ≥ 0.9 year post injury) (Musselman et al. 2009). In contrast, BWSTT led to a greater improvement

in 6MWT compared to the skilled training in the study by Yang et al. (2014) (training at ≥ 7 months post injury).

1.5.5 Lokomat assisted body weight-supported treadmill training

The Lokomat (Hocoma, Volketswil, Switzerland) is a driven gait orthosis which provides assistance with stepping movements during BWSTT, as an alternative to the manual assistance provided by trainers. Stepping assistance in the Lokomat is provided at the hip and knee joints, with the feet resting on the Lokomat's footplates. The stepping assistance can be adjusted by setting variable levels of guiding force, i.e., assistance, from the motors at the hip and knee joints during the swing and stance phases (Varoqui et al. 2014). In contrast, the Lokomat can also be programed to provide resistance against the stepping movements in a velocity-dependent manner, thereby engaging muscle activity during the training (Lam et al. 2015). The 'resistance mode', however, is not commercially available and applied by Lam et al. (2015) only.

Reports on Lokomat training to improve over ground walking after SCI are mixed. Walking speed and distance either improved (Varoqui et al. 2014; Wirz et al. 2005) or were unaffected (Knikou 2013; Niu et al. 2014) by Lokomat training. The duration of training was similar between the studies reporting improvements [8 weeks (Wirz et al. 2005), 12 weeks (Varoqui et al. 2014)] or no change in the walking outcomes [4 weeks (Niu et al. 2014), 1.5 – 3.5 months (Knikou 2013)], measured using the scores on the 10MWT and 6MWT. In the study by Field-Fote and Roach (2011), over ground walking with FES to the common peroneal nerve, and BWSTT with and without the FES, each for 12 weeks, led to significant improvements in the 10MWT and 6MWT, while Lokomat training did not affect either measure (Field-Fote and Roach 2011). When compared with training in an over ground powered exoskeleton (i.e., the Ekso), measures of postural stability and balance remained the same or worsened after Lokomat training, but improved after training in the Ekso (Chisholm et al. 2017). Furthermore, EMG signals from the spinal extensor and abdominal muscles during Lokomat training were similar to pretraining values and to those recorded while lying supine, and higher when walking in the Ekso (Chisholm et al. 2017).

Engagement of volitional muscle activity during Lokomat training is essential in achieving functional gains post training, and is possible when training is performed in the Lokomat's

'resistance mode'. Training in the Lokomat with or without resistance to the leg movements, each for 3 months, led to higher scores on the 10MWT, 6MWT and SCI-FAP in both groups, with greater improvement in the SCI-FAP scores observed in resistance group (Lam et al. 2015). In contrast, fast-paced walking speed during the 10MWT was greater after strength training compared to Lokomat training with passive movement of the legs (Labruyere and van Hedel 2014).

1.5.6 Walking in over ground powered exoskeleton

Individuals with SCI that are unable to improve over ground walking after over ground walking training, BWSTT and Lokomat training are likely to benefit from retraining walking in an over ground powered exoskeleton (Bryce et al. 2015; Lajeunesse et al. 2015; Louie et al. 2015; Miller et al. 2016). Three over ground powered exoskeletons are approved for use in North America: the ReWalk (ReWalk Robotics, Inc., Marlborough, MA) and the Indego (Parker Hannifin Co., Macedonia, OH) are approved for rehabilitation and home use, while the Ekso (Ekso Bionics, Richmond, CA) is approved for use in rehabilitation only. All three devices are worn outside the clothing, and secured using straps at the torso, thigh and lower leg. All three devices allow the user to transition from sit-to-stand and stand-to-sit, and to step in the forwards direction on level ground. The functions are initiated by forward or backward weight-shifts of the trunk, or by selecting the function on an external remote control. These signals are received by an onboard computer (housed in a backpack or incorporated into the device's pelvic band) that controls the motors at the hip and knee. During standing or walking, the weight of the exoskeleton is not supported by the user, rather it is transferred down to the foot plates of the device, which are placed either inside the shoes (ReWalk and Indego) or outside the shoes (Ekso). The exoskeletons differ in other functions. For example, the ReWalk and Indego have functions for stepping up and down stairs, while the Ekso and Indego can provide variable levels of assistance by the motors during the walking. In contrast, only the Indego can provide FES at the trunk and at the hip and knee extensors, while only the Ekso incorporates an overhead tethering system to prevent falls.

Using over ground powered exoskeletons to retrain overground walking is a major advancement in the field of locomotor training, which has enabled individuals with motor-complete and incomplete SCI to walk on smooth surfaces indoors (Esquenazi et al. 2012; Farris et al. 2014;

Hartigan et al. 2015; Kozlowski et al. 2015; Kressler et al. 2014; Yang et al. 2015) and uneven surfaces outdoors (Platz et al. 2016). However, only a few users could ascend and descend stairs, and required complete dependence on the trainer while performing these function (Platz et al. 2016). Walking speeds achieved in the exoskeletons varied across the different models: 0.03 – 0.71 m/s in the ReWalk (Benson et al. 2015; Esquenazi et al. 2012; Spungen et al. 2013b; Talaty et al. 2013; Yang et al. 2015; Zeilig et al. 2012), 0.063 – 0.45 m/s in the Indego (Evans et al. 2015; Farris et al. 2014; Hartigan et al. 2015), and 0.11 – 0.33 m/s in the Ekso (Kozlowski et al. 2015; Kressler et al. 2014). Higher walking speeds in the exoskeletons correlated with higher number of training sessions, lower level of injury and higher age at the time of training (Louie et al. 2015).

Over ground powered exoskeletons are a relatively new development in rehabilitation technology and therefore can be improved in terms of usability and durability. Usability can be improved by addressing design features to reduce some of the risk of adverse events encountered during the training, e.g., falls (Kolakowsky-Hayner et al. 2013) and skin abrasions (Benson et al. 2015; Yang et al. 2015), and by improving the functions of stepping up and down stairs, as only a fraction of the users could perform these tasks (Platz et al. 2016). Furthermore, issues regarding the durability of powered exoskeletons also need improvement (Benson et al. 2015; Zeilig et al. 2012). Lastly, reports on the satisfaction with using over ground powered exoskeletons are mixed. While some users are satisfied with the device (Platz et al. 2016; Stampacchia et al. 2016), others report it did not meet their expectations (Benson et al. 2015).

1.6 Neuroplasticity and other benefits induced by training

1.6.1 Spinal and supraspinal plasticity

1.6.1.1 Uninjured individuals

Neuroplasticity at the spinal and supraspinal levels responds in a training-specific manner after different forms of athletic and skill training, resulting in reduced reflex excitability after certain forms of training, and heightened excitability after others. The training-specific neuroplasticity in uninjured individuals is compared across contrasting forms of training: power, endurance, skill and strength training. Power training (e.g., sprinting) entails strong and quick contractions, and is

in contrast with the prolonged and repetitive muscle contractions involved in endurance training (e.g., long distance running). Skill training (e.g., ballet dancing, slacklining), in contrast to endurance and power training, requires slow and precise muscle contractions. Lastly, strength training is achieved through resistance training (e.g., calf raises) and isometric contractions. Before discussing the effects of physical training on neuroplasticity after SCI, a brief overview of the training-specific effects on neuroplasticity in uninjured individuals is provided in this subsection.

Power- and endurance training induce opposite changes in reflex excitability and spinal inhibitory pathways. Power trained athletes exhibit lower H_{max}/M_{max}, whereas endurance trained athletes show the opposite effect (Casabona et al. 1990; Maffiuletti et al. 2001). Recurrent inhibition is greater in power- compared to endurance trained athletes, while RDD of the H-reflex is higher in endurance- compared to power trained athletes (Earles et al. 2002). Spinal plasticity after endurance training is also task-specific: training to walk backwards on a treadmill reduced the excitability of the H-reflex during walking, without affecting the H_{max}/M_{max} during sitting (Schneider and Capaday 2003; Ung et al. 2005).

Skill training induces spinal and supraspinal plasticity, resulting in reduced spinal reflex excitability and strengthened descending cortical drive. The H_{max}/M_{max} was lower and RDD of the H-reflex was higher (i.e., more depression) in ballet dancers compared to endurance- and power trained athletes, and the untrained participants (Goode and Van Hoven 1982; Nielsen et al. 1993a). However, the ballet dancers had the least amount of reciprocal inhibition compared to that in the athletes and untrained groups (Nielsen et al. 1993a). Training the ankle muscles to perform skilled dorsi- and plantarflexion movements enhanced the TMS-induced MEP responses (Perez et al. 2004). However, in the same study, the strength of cortical interneuronal inhibitory pathways (i.e., short-latency intracortical inhibition) was minimally affected, while no change was observed in the excitatory pathways (i.e., intracortical facilitation) (Perez et al. 2004).

Strength training heightens the excitability of spinal reflexes. Resistance training of the leg muscles (e.g., calf raises) and isometric plantarflexion exercises increased the H/M_{max} (i.e., H-reflex at submaximal stimulus intensity) compared to untrained individuals, without affecting the H_{max}/M_{max} (Aagaard et al. 2002; Lagerquist et al. 2006).

1.6.1.2 Individuals with spinal cord injury

Physical training after SCI induces neuroplasticity at the spinal and supraspinal levels, resulting in reduced hyperexcitability of spinal reflexes and strengthened cortical drive to the weakened muscles, respectively. The training-induced neuroplasticity is also observed in conjunction with improvements in over ground walking. However, unlike the training-specific effects observed in uninjured individuals, similar studies with individuals with SCI are limited (Adams and Hicks 2011; Manella and Field-Fote 2013). Chapter 2 addresses the training-specific changes in spinal reflexes after SCI, and therefore a brief overview of the effects of physical training on spinal and supraspinal neuroplasticity is provided in the following.

Body weight-supported treadmill training with manual assistance

BWSTT induces spinal and supraspinal neuroplasticity, in conjunction with improvements in over ground walking. A single session of BWSTT reduced the H_{max}/M_{max} during the swing and stance phases of treadmill walking (Trimble et al. 2001). In contrast, 4 months of the training enhanced the RDD of the H-reflex without any effects on the H_{max}/M_{max} (Trimble et al. 1998). Changes in the excitability of the H-reflexes from both studies were associated with greater over ground walking speed at a self-selected and fast pace after the training (Trimble et al. 2001; Trimble et al. 1998). BWSTT for 10-28 weeks reduced the power of the EMG signal within the clonus frequency range from the leg muscles, and increased the over ground walking speed and scores on the WISCI II (Gorassini et al. 2009). In the same study, BWSTT for 10-23 weeks increased the MEP_{max} and slope of the MEP recruitment curve, without affecting the MEP threshold (Thomas and Gorassini 2005). The percent increase in MEP_{max} correlated with higher positive change scores on the 6MWT and WISCI II (Thomas and Gorassini 2005).

Body weight-supported treadmill training in the Lokomat

Reports of neuroplasticity after Lokomat training are numerous, however the relationship between the neuroplasticity and improvements in functional walking is unclear. Lokomat training reduced the reflex stiffness at the ankle joint after 4 weeks of training (Mirbagheri et al. 2015), and the excitability of the H-reflex during walking after 1.5-3.5 months of training (Knikou 2013), in conjunction with reduced body weight-support and stepping assistance, and

higher walking speed on the treadmill (Knikou 2013; Mirbagheri et al. 2015). In different reports on the same set of participants and training protocol from the Knikou (2013) study, the RDD of the H-reflex was enhanced (Knikou and Mummidisetty 2014), the excitability of a flexor reflex was altered (Smith et al. 2014), and presynaptic, Ia and Ib inhibition were increased (Knikou and Mummidisetty 2014; Knikou et al. 2015). The training, however, did not affect clinical spasticity (i.e., frequency of spasms) and over ground walking function (i.e., 6MWT, TUG scores) (Knikou 2013). These findings suggest that the effects of training in the Lokomat on hyperexcitable reflexes are variable, wherein either some reflexes are altered (e.g., H- and flexor reflexes) while others remain the same (e.g., spasms).

Walking in over ground powered exoskeleton

Reports on the effects of retraining walking in an over ground powered exoskeleton on reflex excitability after SCI are limited. Only one study examined changes in reflex excitability after training in the Ekso. Compared to pre training, changes in the H_{max}/M_{max} at mid training (i.e., after 9 sessions) and post training (i.e., after 18 sessions) were minimal. However, the participants with lower H_{max}/M_{max} at pre, mid and post training walked for a longer duration in the Ekso than those with higher ratios (Kressler et al. 2014).

Combination of walking training methods

Neuroplasticity is induced at the spinal and supraspinal levels after walking retraining using BWSTT, Lokomat training, over ground walking training and skilled over ground walking training, in different combinations in separate studies. Manual assisted BWSTT with and without FES to the common peroneal nerve, Lokomat training and over ground walking training with the FES, each for 3 months, reduced the duration of extensor spasms and clonus, and increased the threshold for inducing clonus using the foot drop test. The reduced duration of the spasm and higher threshold for inducing clonus correlated with the increase in over ground walking speed after the training (Manella and Field-Fote 2013). In a different study, training using BWSTT with and without FES to: the TA, peroneal muscles and the common peroneal nerve, and using the Lokomat enhanced the early (60-120 ms after stimulation onset) and diminished the late response (120-450 ms after stimulation onset) of a cutaneomuscular reflex in the supine position, and improved the scores on the Mobility subscale of the Spinal Cord Independence Measure

Version I (Hubli et al. 2012). Over ground walking combined with FES to the common peroneal nerve reduced the reflex stiffness of the ankle joint (Mirbagheri et al. 2002). Lastly, skilled over ground walking training (i.e., stepping over obstacles and onto targets) and BWSTT, each for 2 months in a 2 × 2 cross-over design (washout period: 2 months) reduced the excitability of a cutaneomuscular reflex in the TA, enhanced the inhibition of the TA MEP following conditioning stimulation to the common peroneal nerve, and heightened the TA MEP_{max} (Zewdie et al. 2015). Increase in the MEP_{max} correlated with the increase in the 6MWT and 10MWT, but not the SCI-FAP scores (Zewdie et al. 2015). These findings suggest that combining more than one form of physical training can also induce neuroplasticity, and improve walking ability.

Physical training without stepping movements can also induce neuroplasticity. Active bicycling for 20 min improved the over ground walking speed and enhanced the RDD of the H-reflex (Phadke et al. 2009). Furthermore, tilt table standing for 4 weeks reduced extensor spasms, measured using the Spinal Cord Assessment Tool for Spasticity, by the end of training (Adams and Hicks 2011). These findings suggest that neuroplasticity can be induced by other forms of physical training that does not require the retraining of stepping movements, and has implications for rehabilitation interventions for individuals with complete or severe incomplete injuries.

1.6.2 Other benefits

Over ground walking and skilled over ground walking, and BWSTT promote psychologic and physiologic well being. Over ground walking training and BWSTT lead to improvements in the satisfaction with life and physical function (Hicks et al. 2005), and reduced depression as measured by the Center for Epidemiologic Studies-Depression Scale (Yang et al. 2014). The effort to walk over ground is reduced after over ground walking training and BWSTT with FES to the common peroneal nerve (Kressler et al. 2013). Combination of BWSTT, over ground walking training with and without FES, skilled over ground walking and conventional physical therapy strengthens the upper and lower extremity muscles (Alexeeva et al. 2011; Behrman et al. 2005; Buehner et al. 2012; Field-Fote and Roach 2011; Senthilvelkumar et al. 2015), increases confidence in balance (Musselman et al. 2009; Yang et al. 2014) and improves postural stability and balance (Alexeeva et al. 2011; Buehner et al. 2012; Harkema et al. 2012; Pramodhyakul et al. 2013). Lastly, in a case-study on a single individual with AIS D injury at C5-6 level, over

ground walking training, skilled over ground walking training and BWSTT also improved light touch sensation and motor level impairment (Behrman et al. 2005).

Reports on the effects of Lokomat training on other physiologic benefits are limited. Lokomat training led to improvements in the strength of the lower limb muscles (Field-Fote and Roach 2011; Wirz et al. 2005), reduced intrinsic stiffness at the ankle (Mirbagheri et al. 2015) and reduced pain (Labruyere and van Hedel 2014). However, its effects on postural stability, measured using the Timed Up and Go test, are negligible. Briefly, the TUG test measures the time taken to: stand up from a chair, walk 3 m, turn around, walk back to the chair, turn around and sit back on the chair, and relies on maintaining balance throughout the test. A few studies report improvements in the scores on the TUG test after Lokomat training (Varoqui et al. 2014; Wirz et al. 2005), while the majority report no change (Duffell et al. 2015; Knikou 2013; Niu et al. 2011; Niu et al. 2014). Furthermore, when balance is quantified using body sway and limits of stability, the balance measures remained the same or worsened after Lokomat training (Chisholm et al. 2017). Finally, walking in the Lokomat requires less physiologic exertion compared to walking without the Lokomat on the treadmill, in terms of metabolic equivalents, heart rate, and Rating of Perceived Exertion from 0-10 (Fenuta and Hicks 2014).

Retraining walking using over ground powered exoskeletons provides physiologic benefits, in addition to providing those with severe SCI the ability to walk again (Louie et al. 2015; Miller et al. 2016). Walking retraining in powered exoskeletons promotes physiologic exertion (Benson et al. 2015; Evans et al. 2015; Zeilig et al. 2012) and, therefore, can be used as a means of exercise. The exertion required to walk in powered exoskeletons is lower than walking using knee-ankle-foot-orthoses (Farris et al. 2014), and is reduced by the end of training (Spungen et al. 2013b). Walking in powered exoskeletons improves postural balance (Chisholm et al. 2017; Kozlowski et al. 2015) and sleep (Kozlowski et al. 2015), and reduces pain (Kozlowski et al. 2015; Kressler et al. 2014; Stampacchia et al. 2016) and body fat composition (Karelis et al. 2017; Spungen et al. 2013a). The effects of walking in powered exoskeletons on bowel function and spasticity are mixed. Some suggest improvements in bowel function (Kozlowski et al. 2015) and reduction in spasticity (Benson et al. 2015; Esquenazi et al. 2012; Kozlowski et al. 2015; Stampacchia et al. 2016), while others report no effect on bowel function (Platz et al. 2016) or spasticity (Kressler et al. 2014; Platz et al. 2016). Given that overground powered exoskeletons are a new

development, the effects of retraining walking in these devices on neuroplasticity in the sensory and motor domains is not well clear. Furthermore, since some powered exoskeletons are now approved for home use, the efficacy of these devises in the home and community needs further exploration.

1.7 Bibliography for Chapter 1

Aagaard P, Simonsen EB, Andersen JL, Magnusson P, and Dyhre-Poulsen P. Neural adaptation to resistance training: changes in evoked V-wave and H-reflex responses. *Journal of applied physiology (Bethesda, Md : 1985)* 92: 2309-2318, 2002.

Adams MM, and Hicks AL. Comparison of the effects of body-weight-supported treadmill training and tilt-table standing on spasticity in individuals with chronic spinal cord injury. *The journal of spinal cord medicine* 34: 488-494, 2011.

Ahuja CS, Wilson JR, Nori S, Kotter MRN, Druschel C, Curt A, and Fehlings MG. Traumatic spinal cord injury. *Nature reviews Disease primers* 3: 17018, 2017.

Alexeeva N, Sames C, Jacobs PL, Hobday L, Distasio MM, Mitchell SA, and Calancie B. Comparison of training methods to improve walking in persons with chronic spinal cord injury: a randomized clinical trial. *The journal of spinal cord medicine* 34: 362-379, 2011.

Amemiya M, and Yamaguchi T. Fictive locomotion of the forelimb evoked by stimulation of the mesencephalic locomotor region in the decerebrate cat. *Neurosci Lett* 50: 91-96, 1984.

Anderson KD. Targeting recovery: priorities of the spinal cord-injured population. *Journal of neurotrauma* 21: 1371-1383, 2004a.

Barthelemy D, Grey MJ, Nielsen JB, and Bouyer L. Involvement of the corticospinal tract in the control of human gait. *Prog Brain Res* 192: 181-197, 2011.

Barthelemy D, Willerslev-Olsen M, Lundell H, Conway BA, Knudsen H, Biering-Sorensen F, and Nielsen JB. Impaired transmission in the corticospinal tract and gait disability in spinal cord injured persons. *Journal of neurophysiology* 104: 1167-1176, 2010.

Behrman AL, Lawless-Dixon AR, Davis SB, Bowden MG, Nair P, Phadke C, Hannold EM, Plummer P, and Harkema SJ. Locomotor training progression and outcomes after incomplete spinal cord injury. *Physical therapy* 85: 1356-1371, 2005.

Beloozerova IN, Farrell BJ, Sirota MG, and Prilutsky BI. Differences in movement mechanics, electromyographic, and motor cortex activity between accurate and nonaccurate stepping. *Journal of neurophysiology* 103: 2285-2300, 2010.

Benson I, Hart K, Tussler D, and van Middendorp JJ. Lower-limb exoskeletons for individuals with chronic spinal cord injury: Findings from a feasibility study. *Clin Rehabil* 2015.

Benz EN, Hornby TG, Bode RK, Scheidt RA, and Schmit BD. A physiologically based clinical measure for spastic reflexes in spinal cord injury. *Archives of physical medicine and rehabilitation* 86: 52-59, 2005.

Beres-Jones JA, Johnson TD, and Harkema SJ. Clonus after human spinal cord injury cannot be attributed solely to recurrent muscle-tendon stretch. *Experimental brain research* 149: 222-236, 2003.

Bjerkefors A, Squair JW, Chua R, Lam T, Chen Z, and Carpenter MG. Assessment of abdominal muscle function in individuals with motor-complete spinal cord injury above T6 in response to transcranial magnetic stimulation. *Journal of rehabilitation medicine* 47: 138-146, 2015.

Bolton DA, Tse AD, Ballermann M, Misiaszek JE, and Fouad K. Task specific adaptations in rat locomotion: runway versus horizontal ladder. *Behavioural brain research* 168: 272-279, 2006.

Boorman GI, Lee RG, Becker WJ, and Windhorst UR. Impaired "natural reciprocal inhibition" in patients with spasticity due to incomplete spinal cord injury. *Electroencephalography and clinical neurophysiology* 101: 84-92, 1996.

Bos R, Sadlaoud K, Boulenguez P, Buttigieg D, Liabeuf S, Brocard C, Haase G, Bras H, and Vinay L. Activation of 5-HT2A receptors upregulates the function of the neuronal K-Cl cotransporter KCC2. *Proceedings of the National Academy of Sciences of the United States of America* 110: 348-353, 2013.

Boulenguez P, Liabeuf S, Bos R, Bras H, Jean-Xavier C, Brocard C, Stil A, Darbon P, Cattaert D, Delpire E, Marsala M, and Vinay L. Down-regulation of the potassium-chloride

cotransporter KCC2 contributes to spasticity after spinal cord injury. *Nature medicine* 16: 302-307, 2010.

Brown TG. The Intrinsic Factors in the Act of Progression in the Mammal. *Proceedings of the Royal Society of London Series B, Containing Papers of a Biological Character* 84: 308-319, 1911.

Bryce TN, Dijkers MP, and Kozlowski AJ. Framework for Assessment of the Usability of Lower-Extremity Robotic Exoskeletal Orthoses. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 94: 1000-1014, 2015.

Buehner JJ, Forrest GF, Schmidt-Read M, White S, Tansey K, and Basso DM. Relationship between ASIA examination and functional outcomes in the NeuroRecovery Network Locomotor Training Program. *Archives of physical medicine and rehabilitation* 93: 1530-1540, 2012.

Burns AS, Marino RJ, Flanders AE, and Flett H. Clinical diagnosis and prognosis following spinal cord injury. *Handbook of clinical neurology* 109: 47-62, 2012.

Burns SP, Golding DG, Rolle WA, Jr., Graziani V, and Ditunno JF, Jr. Recovery of ambulation in motor-incomplete tetraplegia. *Archives of physical medicine and rehabilitation* 78: 1169-1172, 1997.

Bussel B, Roby-Brami A, Azouvi P, Biraben A, Yakovleff A, and Held JP. Myoclonus in a patient with spinal cord transection. Possible involvement of the spinal stepping generator. *Brain : a journal of neurology* 111 (Pt 5): 1235-1245, 1988.

Butler JE, Godfrey S, and Thomas CK. Depression of involuntary activity in muscles paralyzed by spinal cord injury. *Muscle & nerve* 33: 637-644, 2006.

Calancie B, Molano MR, and Broton JG. Tendon reflexes for predicting movement recovery after acute spinal cord injury in humans. *Clin Neurophysiol* 115: 2350-2363, 2004.

Calancie B, Needham-Shropshire B, Jacobs P, Willer K, Zych G, and Green BA. Involuntary stepping after chronic spinal cord injury. Evidence for a central rhythm generator for locomotion in man. *Brain : a journal of neurology* 117 (Pt 5): 1143-1159, 1994.

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

Capaday C, and Stein RB. Difference in the amplitude of the human soleus H reflex during walking and running. *The Journal of physiology* 392: 513-522, 1987.

Cariga P, Catley M, Nowicky AV, Savic G, Ellaway PH, and Davey NJ. Segmental recording of cortical motor evoked potentials from thoracic paravertebral myotomes in complete spinal cord injury. *Spine* 27: 1438-1443, 2002.

Casabona A, Polizzi MC, and Perciavalle V. Differences in H-reflex between athletes trained for explosive contractions and non-trained subjects. *European journal of applied physiology and occupational physiology* 61: 26-32, 1990.

Cazalets JR, Sqalli-Houssaini Y, and Clarac F. Activation of the central pattern generators for locomotion by serotonin and excitatory amino acids in neonatal rat. *The Journal of physiology* 455: 187-204, 1992.

Cazalets JR, Sqalli-Houssaini Y, and Clarac F. GABAergic inactivation of the central pattern generators for locomotion in isolated neonatal rat spinal cord. *The Journal of physiology* 474: 173-181, 1994.

Cheng CL, Noonan VK, Shurgold J, Chen J, Rivers CS, Hamedani HK, Humphreys S, Bailey C, Attabib N, Mac-Thiong JM, Goytan M, Paquet J, Fox R, Ahn H, Kwon BK, and Fourney DR. Geomapping of Traumatic Spinal Cord Injury in Canada and Factors Related to Triage Pattern. *Journal of neurotrauma* 2017.

Chisholm AE, Alamro RA, Williams AM, and Lam T. Overground vs. treadmill-based robotic gait training to improve seated balance in people with motor-complete spinal cord injury: a case report. *Journal of neuroengineering and rehabilitation* 14: 27, 2017.

Couris CM, Guilcher SJ, Munce SE, Fung K, Craven BC, Verrier M, and Jaglal SB. Characteristics of adults with incident traumatic spinal cord injury in Ontario, Canada. *Spinal cord* 48: 39-44, 2010.

Crozier KS, Cheng LL, Graziani V, Zorn G, Herbison G, and Ditunno JF, Jr. Spinal cord injury: prognosis for ambulation based on quadriceps recovery. *Paraplegia* 30: 762-767, 1992.

D'Amico JM, Condliffe EG, Martins KJ, Bennett DJ, and Gorassini MA. Recovery of neuronal and network excitability after spinal cord injury and implications for spasticity. *Frontiers in integrative neuroscience* 8: 36, 2014.

de Vlugt E, de Groot JH, Wisman WH, and Meskers CG. Clonus is explained from increased reflex gain and enlarged tissue viscoelasticity. *Journal of biomechanics* 45: 148-155, 2012.

Dietz V, Gollhofer A, Kleiber M, and Trippel M. Regulation of bipedal stance: dependency on "load" receptors. *Experimental brain research* 89: 229-231, 1992.

Dimitrijevic MR, Gerasimenko Y, and Pinter MM. Evidence for a spinal central pattern generator in humans. *Annals of the New York Academy of Sciences* 860: 360-376, 1998.

Dimitrijevic MR, Nathan PW, and Sherwood AM. Clonus: the role of central mechanisms. *Journal of neurology, neurosurgery, and psychiatry* 43: 321-332, 1980b.

Drew T. Motor cortical cell discharge during voluntary gait modification. *Brain Res* 457: 181-187, 1988.

Drew T, Dubuc R, and Rossignol S. Discharge patterns of reticulospinal and other reticular neurons in chronic, unrestrained cats walking on a treadmill. *Journal of neurophysiology* 55: 375-401, 1986.

Dryden DM, Saunders LD, Rowe BH, May LA, Yiannakoulias N, Svenson LW, Schopflocher DP, and Voaklander DC. The epidemiology of traumatic spinal cord injury in Alberta, Canada. *Canadian journal of neurological sciences* 30: 113-121, 2003.

Duffell LD, Brown GL, and Mirbagheri MM. Interventions to Reduce Spasticity and Improve Function in People With Chronic Incomplete Spinal Cord Injury: Distinctions Revealed by Different Analytical Methods. *Neurorehabilitation and neural repair* 29: 566-576, 2015.

Duysens J, Trippel M, Horstmann GA, and Dietz V. Gating and reversal of reflexes in ankle muscles during human walking. *Experimental brain research* 82: 351-358, 1990.

Earles DR, Dierking JT, Robertson CT, and Koceja DM. Pre- and post-synaptic control of motoneuron excitability in athletes. *Medicine and science in sports and exercise* 34: 1766-1772, 2002.

Ellaway PH, Anand P, Bergstrom EM, Catley M, Davey NJ, Frankel HL, Jamous A, Mathias C, Nicotra A, Savic G, Short D, and Theodorou S. Towards improved clinical and physiological assessments of recovery in spinal cord injury: a clinical initiative. *Spinal cord* 42: 325-337, 2004.

Ertekin C, Uludag B, On A, Yetimalar Y, Ertas M, Colakoglu Z, and Arac N. Motor-evoked potentials from various levels of paravertebral muscles in normal subjects and in patients with focal lesions of the spinal cord. *Spine* 23: 1016-1022, 1998.

Esquenazi A, Talaty M, Packel A, and Saulino M. The ReWalk powered exoskeleton to restore ambulatory function to individuals with thoracic-level motor-complete spinal cord injury. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 91: 911-921, 2012.

Evans N, Hartigan C, Kandilakis C, Pharo E, and Clesson I. Acute Cardiorespiratory and Metabolic Responses During Exoskeleton-Assisted Walking Overground Among Persons with Chronic Spinal Cord Injury. *Top Spinal Cord Inj Rehabil* 21: 122-132, 2015.

Faist M, Mazevet D, Dietz V, and Pierrot-Deseilligny E. A quantitative assessment of presynaptic inhibition of Ia afferents in spastics. Differences in hemiplegics and paraplegics. *Brain : a journal of neurology* 117 (Pt 6): 1449-1455, 1994.

Farris RJ, Quintero HA, Murray SA, Ha KH, Hartigan C, and Goldfarb M. A preliminary assessment of legged mobility provided by a lower limb exoskeleton for persons with paraplegia. *IEEE transactions on neural systems and rehabilitation engineering : a publication of the IEEE Engineering in Medicine and Biology Society* 22: 482-490, 2014.

Fenuta AM, and Hicks AL. Metabolic demand and muscle activation during different forms of bodyweight supported locomotion in men with incomplete SCI. *BioMed research international* 2014: 632765, 2014.

Field-Fote EC, and Roach KE. Influence of a locomotor training approach on walking speed and distance in people with chronic spinal cord injury: a randomized clinical trial. *Physical therapy* 91: 48-60, 2011.

Field-Fote EC, Yang JF, Basso DM, and Gorassini MA. Supraspinal Control Predicts Locomotor Function and Forecasts Responsiveness to Training after Spinal Cord Injury. *Journal of neurotrauma* 34: 1813-1825, 2017.

Forssberg H, Grillner S, and Rossignol S. Phase dependent reflex reversal during walking in chronic spinal cats. *Brain Res* 85: 103-107, 1975.

Fukuyama H, Ouchi Y, Matsuzaki S, Nagahama Y, Yamauchi H, Ogawa M, Kimura J, and Shibasaki H. Brain functional activity during gait in normal subjects: a SPECT study. *Neurosci Lett* 228: 183-186, 1997.

Gerasimenko Y, Gorodnichev R, Machueva E, Pivovarova E, Semyenov D, Savochin A, Roy RR, and Edgerton VR. Novel and direct access to the human locomotor spinal circuitry. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 30: 3700-3708, 2010.

Gollie JM, Guccione AA, Panza GS, Jo PY, and Herrick JE. Effects of Overground Locomotor Training on Walking Performance in Chronic Cervical Motor Incomplete Spinal Cord Injury: A Pilot Study. *Archives of physical medicine and rehabilitation* 98: 1119-1125, 2017.

Goode DJ, and Van Hoven J. Loss of patellar and Achilles tendon reflexes in classical ballet dancers. *Arch Neurol* 39: 323, 1982.

Gorassini MA, Knash ME, Harvey PJ, Bennett DJ, and Yang JF. Role of motoneurons in the generation of muscle spasms after spinal cord injury. *Brain : a journal of neurology* 127: 2247-2258, 2004.

Gorassini MA, Norton JA, Nevett-Duchcherer J, Roy FD, and Yang JF. Changes in locomotor muscle activity after treadmill training in subjects with incomplete spinal cord injury. *Journal of neurophysiology* 101: 969-979, 2009.

Grassner L, Marschallinger J, Dünser MW, Novak HF, Zerbs A, Aigner L, Trinka E, and Sellner J. Nontraumatic spinal cord injury at the neurological intensive care unit: spectrum, causes of admission and predictors of mortality. *Therapeutic Advances in Neurological Disorders* 9: 85-94, 2016.

Grey MJ, Klinge K, Crone C, Lorentzen J, Biering-Sorensen F, Ravnborg M, and Nielsen JB. Post-activation depression of soleus stretch reflexes in healthy and spastic humans. *Experimental brain research* 185: 189-197, 2008.

Grillner S, Hellgren J, Menard A, Saitoh K, and Wikstrom MA. Mechanisms for selection of basic motor programs--roles for the striatum and pallidum. *Trends in neurosciences* 28: 364-370, 2005.

Hagen EM. Acute complications of spinal cord injuries. *World journal of orthopedics* 6: 17-23, 2015.

Harkema SJ, Schmidt-Read M, Lorenz DJ, Edgerton VR, and Behrman AL. Balance and ambulation improvements in individuals with chronic incomplete spinal cord injury using locomotor training-based rehabilitation. *Archives of physical medicine and rehabilitation* 93: 1508-1517, 2012.

Hartigan C, Kandilakis C, Dalley S, Clausen M, Wilson E, Morrison S, Etheridge S, and Farris R. Mobility Outcomes Following Five Training Sessions with a Powered Exoskeleton. *Topics in Spinal Cord Injury Rehabilitation* 21: 93-99, 2015.

Hicks AL, Adams MM, Martin Ginis K, Giangregorio L, Latimer A, Phillips SM, and McCartney N. Long-term body-weight-supported treadmill training and subsequent follow-up in persons with chronic SCI: effects on functional walking ability and measures of subjective well-being. *Spinal cord* 43: 291-298, 2005.

Hidler JM, and Rymer WZ. A simulation study of reflex instability in spasticity: origins of clonus. *IEEE transactions on rehabilitation engineering : a publication of the IEEE Engineering in Medicine and Biology Society* 7: 327-340, 1999.

Holmes G. The goulstonian lectures on spinal injuries of warfare: delivered before the Royal College of Physicians of London. *British medical journal* 2: 815-821, 1915.

Hubli M, Dietz V, and Bolliger M. Spinal reflex activity: a marker for neuronal functionality after spinal cord injury. *Neurorehabilitation and neural repair* 26: 188-196, 2012.

Hultborn H, Illert M, Nielsen J, Paul A, Ballegaard M, and Wiese H. On the mechanism of the post-activation depression of the H-reflex in human subjects. *Experimental brain research* 108: 450-462, 1996.

Huttenlocher PR. Synaptic density in human frontal cortex - developmental changes and effects of aging. *Brain Res* 163: 195-205, 1979.

Jahn K, Deutschlander A, Stephan T, Strupp M, Wiesmann M, and Brandt T. Brain activation patterns during imagined stance and locomotion in functional magnetic resonance imaging. *Neuroimage* 22: 1722-1731, 2004.

Jazayeri SB, Beygi S, Shokraneh F, Hagen EM, and Rahimi-Movaghar V. Incidence of traumatic spinal cord injury worldwide: a systematic review. *European spine journal : official publication of the European Spine Society, the European Spinal Deformity Society, and the European Section of the Cervical Spine Research Society* 24: 905-918, 2015.

Johnson RL, Gerhart KA, McCray J, Menconi JC, and Whiteneck GG. Secondary conditions following spinal cord injury in a population-based sample. *Spinal cord* 36: 45-50, 1998.

Jones CA, and Yang JF. Reflex behavior during walking in incomplete spinal-cord-injured subjects. *Experimental neurology* 128: 239-248, 1994.

Jorgensen HS, Nakayama H, Raaschou HO, and Olsen TS. Recovery of walking function in stroke patients: the Copenhagen Stroke Study. *Archives of physical medicine and rehabilitation* 76: 27-32, 1995.

Juknis N, Cooper JM, and Volshteyn O. The changing landscape of spinal cord injury. *Handbook of clinical neurology* 109: 149-166, 2012.

Kakinohana O, Hefferan MP, Miyanohara A, Nejime T, Marsala S, Juhas S, Juhasova J, Motlik J, Kucharova K, Strnadel J, Platoshyn O, Lazar P, Galik J, Vinay L, and Marsala M. Combinational spinal GAD65 gene delivery and systemic GABA-mimetic treatment for modulation of spasticity. *PloS one* 7: e30561, 2012.

Kapitza S, Zorner B, Weinmann O, Bolliger M, Filli L, Dietz V, and Schwab ME. Tail spasms in rat spinal cord injury: changes in interneuronal connectivity. *Experimental neurology* 236: 179-189, 2012.

Karelis AD, Carvalho LP, Castillo MJ, Gagnon DH, and Aubertin-Leheudre M. Effect on body composition and bone mineral density of walking with a robotic exoskeleton in adults with chronic spinal cord injury. *Journal of rehabilitation medicine* 49: 84-87, 2017.

Kay ED, Deutsch A, and Wuermser LA. Predicting walking at discharge from inpatient rehabilitation after a traumatic spinal cord injury. *Archives of physical medicine and rehabilitation* 88: 745-750, 2007.

Kiehn O, Dougherty KJ, Hagglund M, Borgius L, Talpalar A, and Restrepo CE. Probing spinal circuits controlling walking in mammals. *Biochemical and biophysical research communications* 396: 11-18, 2010.

Kim CM, Eng JJ, and Whittaker MW. Level walking and ambulatory capacity in persons with incomplete spinal cord injury: relationship with muscle strength. *Spinal cord* 42: 156-162, 2004.

Knikou M. Functional reorganization of soleus H-reflex modulation during stepping after robotic-assisted step training in people with complete and incomplete spinal cord injury. *Experimental brain research* 228: 279-296, 2013.

Knikou M, and Mummidisetty CK. Locomotor training improves premotoneuronal control after chronic spinal cord injury. *Journal of neurophysiology* 111: 2264-2275, 2014.

Knikou M, Smith AC, and Mummidisetty CK. Locomotor training improves reciprocal and nonreciprocal inhibitory control of soleus motoneurons in human spinal cord injury. *Journal of neurophysiology* 113: 2447-2460, 2015.

Ko HY, Ditunno JF, Jr., Graziani V, and Little JW. The pattern of reflex recovery during spinal shock. *Spinal cord* 37: 402-409, 1999.

Kolakowsky-Hayner S, Crew J, Moran S, and Shah A. Safety and feasibility of using the ekso bionic exoskeleton to aid ambulation after spinal cord injury. *Journal of spine* S4: 8, 2013.

Kozlowski AJ, Bryce TN, and Dijkers MP. Time and effort required by persons with spinal cord injury to learn to use a powered exoskeleton for assisted walking. *Topics in Spinal Cord Injury Rehabilitation* 21: 110-121, 2015.

Kressler J, Nash MS, Burns PA, and Field-Fote EC. Metabolic responses to 4 different body weight-supported locomotor training approaches in persons with incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 94: 1436-1442, 2013.

Kressler J, Thomas CK, Field-Fote EC, Sanchez J, Widerstrom-Noga E, Cilien DC, Gant K, Ginnety K, Gonzalez H, Martinez A, Anderson KD, and Nash MS. Understanding therapeutic benefits of overground bionic ambulation: exploratory case series in persons with chronic, complete spinal cord injury. *Archives of physical medicine and rehabilitation* 95: 1878-1887 e1874, 2014.

Krueger H, Noonan VK, Trenaman LM, Joshi P, and Rivers CS. The economic burden of traumatic spinal cord injury in Canada. *Chronic diseases and injuries in Canada* 33: 113-122, 2013.

Kuhn RA. Functional capacity of the isolated human spinal cord. *Brain : a journal of neurology* 73: 1-51, 1950.

Labruyere R, and van Hedel HJ. Strength training versus robot-assisted gait training after incomplete spinal cord injury: a randomized pilot study in patients depending on walking assistance. *Journal of neuroengineering and rehabilitation* 11: 4, 2014.

Lagerquist O, Zehr EP, and Docherty D. Increased spinal reflex excitability is not associated with neural plasticity underlying the cross-education effect. *Journal of applied physiology (Bethesda, Md : 1985)* 100: 83-90, 2006.

Lajeunesse V, Vincent C, Routhier F, Careau E, and Michaud F. Exoskeletons' design and usefulness evidence according to a systematic review of lower limb exoskeletons used for functional mobility by people with spinal cord injury. *Disability and rehabilitation Assistive technology* 1-13, 2015.

Lam T, Pauhl K, Ferguson A, Malik RN, Krassioukov A, and Eng JJ. Training with robot-applied resistance in people with motor-incomplete spinal cord injury: Pilot study. *Journal of rehabilitation research and development* 52: 113-129, 2015.

Lee BB, Cripps RA, Fitzharris M, and Wing PC. The global map for traumatic spinal cord injury epidemiology: update 2011, global incidence rate. *Spinal cord* 52: 110-116, 2014.

Lee MS, Choi YC, Lee SH, and Lee SB. Sleep-related periodic leg movements associated with spinal cord lesions. *Movement disorders : official journal of the Movement Disorder Society* 11: 719-722, 1996.

Lenehan B, Street J, Kwon BK, Noonan V, Zhang H, Fisher CG, and Dvorak MF. The epidemiology of traumatic spinal cord injury in British Columbia, Canada. *Spine* 37: 321-329, 2012.

Levi R, Hultling C, and Seiger A. The Stockholm Spinal Cord Injury Study: 2. Associations between clinical patient characteristics and post-acute medical problems. *Paraplegia* 33: 585-594, 1995.

Little JW, and Halar EM. H-reflex changes following spinal cord injury. *Archives of physical medicine and rehabilitation* 66: 19-22, 1985.

Little JW, Micklesen P, Umlauf R, and Britell C. Lower extremity manifestations of spasticity in chronic spinal cord injury. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 68: 32-36, 1989.

Liu H, Skinner RD, Arfaj A, Yates C, Reese NB, Williams K, and Garcia-Rill E. L-Dopa effect on frequency-dependent depression of the H-reflex in adult rats with complete spinal cord transection. *Brain research bulletin* 83: 262-265, 2010.

Lotharius J, and Brundin P. Pathogenesis of Parkinson's disease: dopamine, vesicles and alpha-synuclein. *Nature reviews Neuroscience* 3: 932-942, 2002.

Louie DR, Eng JJ, and Lam T. Gait speed using powered robotic exoskeletons after spinal cord injury: a systematic review and correlational study. *Journal of neuroengineering and rehabilitation* 12: 82, 2015.

Maffiuletti NA, Martin A, Babault N, Pensini M, Lucas B, and Schieppati M. Electrical and mechanical H(max)-to-M(max) ratio in power- and endurance-trained athletes. *Journal of applied physiology (Bethesda, Md : 1985)* 90: 3-9, 2001.

Magladery JW, Porter WE, Park AM, and Teasdall RD. Electrophysiological studies of nerve and reflex activity in normal man. IV. The two-neurone reflex and identification of certain action potentials from spinal roots and cord. *Bulletin of the Johns Hopkins Hospital* 88: 499-519, 1951.

Manella KJ, and Field-Fote EC. Modulatory effects of locomotor training on extensor spasticity in individuals with motor-incomplete spinal cord injury. *Restorative neurology and neuroscience* 31: 633-646, 2013.

Maynard FM, Jr., Bracken MB, Creasey G, Ditunno JF, Jr., Donovan WH, Ducker TB, Garber SL, Marino RJ, Stover SL, Tator CH, Waters RL, Wilberger JE, and Young W. International Standards for Neurological and Functional Classification of Spinal Cord Injury. American Spinal Injury Association. *Spinal cord* 35: 266-274, 1997.

McKinley WO, Seel RT, Gadi RK, and Tewksbury MA. Nontraumatic vs. traumatic spinal cord injury: a rehabilitation outcome comparison. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 80: 693-699; quiz 700, 716, 2001.

McPherson DR, and Kemnitz CP. Modulation of lamprey fictive swimming and motoneuron physiology by dopamine, and its immunocytochemical localization in the spinal cord. *Neurosci Lett* 166: 23-26, 1994.

Mehrholz J, Harvey LA, Thomas S, and Elsner B. Is body-weight-supported treadmill training or robotic-assisted gait training superior to overground gait training and other forms of

physiotherapy in people with spinal cord injury? A systematic review. *Spinal cord* 55: 722-729, 2017.

Miele VJ, Panjabi MM, and Benzel EC. Anatomy and biomechanics of the spinal column and cord. *Handbook of clinical neurology* 109: 31-43, 2012.

Miller LE, Zimmermann AK, and Herbert WG. Clinical effectiveness and safety of powered exoskeleton-assisted walking in patients with spinal cord injury: systematic review with meta-analysis. *Medical devices (Auckland, NZ)* 9: 455-466, 2016.

Mirbagheri MM, Barbeau H, Ladouceur M, and Kearney RE. Intrinsic and reflex stiffness in normal and spastic, spinal cord injured subjects. *Experimental brain research* 141: 446-459, 2001.

Mirbagheri MM, Kindig MW, and Niu X. Effects of robotic-locomotor training on stretch reflex function and muscular properties in individuals with spinal cord injury. *Clin Neurophysiol* 126: 997-1006, 2015.

Mirbagheri MM, Ladouceur M, Barbeau H, and Kearney RE. The effects of long-term FES-assisted walking on intrinsic and reflex dynamic stiffness in spastic spinal-cord-injured subjects. *Neural Systems and Rehabilitation Engineering, IEEE Transactions on* 10: 280-289, 2002.

Miyai I, Tanabe HC, Sase I, Eda H, Oda I, Konishi I, Tsunazawa Y, Suzuki T, Yanagida T, and Kubota K. Cortical mapping of gait in humans: a near-infrared spectroscopic topography study. *Neuroimage* 14: 1186-1192, 2001.

Mori S, Matsui T, Kuze B, Asanome M, Nakajima K, and Matsuyama K. Cerebellar-induced locomotion: reticulospinal control of spinal rhythm generating mechanism in cats. *Annals of the New York Academy of Sciences* 860: 94-105, 1998.

Morton SM, and Bastian AJ. Cerebellar contributions to locomotor adaptations during splitbelt treadmill walking. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 26: 9107-9116, 2006.

Musselman KE, Fouad K, Misiaszek JE, and Yang JF. Training of walking skills overground and on the treadmill: case series on individuals with incomplete spinal cord injury. *Physical therapy* 89: 601-611, 2009.

Musselman KE, and Yang JF. Walking tasks encountered by urban-dwelling adults and persons with incomplete spinal cord injuries. *Journal of rehabilitation medicine* 39: 567-574, 2007.

Nadeau S, Jacquemin G, Fournier C, Lamarre Y, and Rossignol S. Spontaneous motor rhythms of the back and legs in a patient with a complete spinal cord transection. *Neurorehabilitation and neural repair* 24: 377-383, 2010.

Nathan PW. Effects on movement of surgical incisions into the human spinal cord. *Brain : a journal of neurology* 117 (Pt 2): 337-346, 1994.

New PW, Cripps RA, and Bonne Lee B. Global maps of non-traumatic spinal cord injury epidemiology: towards a living data repository. *Spinal cord* 52: 97-109, 2014.

Nielsen J, Crone C, and Hultborn H. H-reflexes are smaller in dancers from The Royal Danish Ballet than in well-trained athletes. *European journal of applied physiology and occupational physiology* 66: 116-121, 1993a.

Nielsen J, Petersen N, Ballegaard M, Biering-Sorensen F, and Kiehn O. H-reflexes are less depressed following muscle stretch in spastic spinal cord injured patients than in healthy subjects. *Experimental brain research* 97: 173-176, 1993b.

Nielsen JB, and Sinkjaer T. Afferent feedback in the control of human gait. *Journal of electromyography and kinesiology : official journal of the International Society of Electrophysiological Kinesiology* 12: 213-217, 2002.

Niu X, Patel C, Varoqui D, and Mirbagheri MM. The effect of robot-assisted lokomotor training on gait recovery: a multivariate analysis. *Conference proceedings : Annual International Conference of the IEEE Engineering in Medicine and Biology Society IEEE Engineering in Medicine and Biology Society Annual Conference* 2011: 3542-3545, 2011.

Niu X, Varoqui D, Kindig M, and Mirbagheri MM. Prediction of gait recovery in spinal cord injured individuals trained with robotic gait orthosis. *Journal of neuroengineering and rehabilitation* 11: 42, 2014.

Noonan VK, Chan E, Santos A, Soril L, Lewis R, Singh A, Cheng CL, O'Connell C, Truchon C, Paquet J, Christie S, Ethans K, Tsai E, Ford MH, Drew B, Linassi G, Bailey C, and Fehlings MG. Traumatic Spinal Cord Injury Care in Canada: A Survey of Canadian Centres. *Journal of neurotrauma* 2017.

Norton JA, Bennett DJ, Knash ME, Murray KC, and Gorassini MA. Changes in sensory-evoked synaptic activation of motoneurons after spinal cord injury in man. *Brain : a journal of neurology* 131: 1478-1491, 2008.

Nutt JG, Bloem BR, Giladi N, Hallett M, Horak FB, and Nieuwboer A. Freezing of gait: moving forward on a mysterious clinical phenomenon. *The Lancet Neurology* 10: 734-744, 2011.

Oleson CV, Burns AS, Ditunno JF, Geisler FH, and Coleman WP. Prognostic value of pinprick preservation in motor complete, sensory incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 86: 988-992, 2005.

Olsson MC, Kruger M, Meyer LH, Ahnlund L, Gransberg L, Linke WA, and Larsson L. Fibre type-specific increase in passive muscle tension in spinal cord-injured subjects with spasticity. *The Journal of physiology* 577: 339-352, 2006.

Orlovsky GN. Activity of rubrospinal neurons during locomotion. *Brain Res* 46: 99-112, 1972a.

Orlovsky GN. Activity of vestibulospinal neurons during locomotion. *Brain Res* 46: 85-98, 1972b.

Pang MY, and Yang JF. The initiation of the swing phase in human infant stepping: importance of hip position and leg loading. *The Journal of physiology* 528 Pt 2: 389-404, 2000.

Pearson KG. Generating the walking gait: role of sensory feedback. *Prog Brain Res* 143: 123-129, 2004.

Pearson KG. Role of sensory feedback in the control of stance duration in walking cats. *Brain research reviews* 57: 222-227, 2008.

Pearson KG, and Collins DF. Reversal of the influence of group Ib afferents from plantaris on activity in medial gastrocnemius muscle during locomotor activity. *Journal of neurophysiology* 70: 1009-1017, 1993.

Peiper A. *Cerebral function in infancy and childhood*. Plenum Pub Corp, 1963.

Perez MA, Lungholt BK, Nyborg K, and Nielsen JB. Motor skill training induces changes in the excitability of the leg cortical area in healthy humans. *Experimental brain research* 159: 197-205, 2004.

Petersen NT, Butler JE, Marchand-Pauvert V, Fisher R, Ledebt A, Pyndt HS, Hansen NL, and Nielsen JB. Suppression of EMG activity by transcranial magnetic stimulation in human subjects during walking. *The Journal of physiology* 537: 651-656, 2001.

Phadke CP, Flynn SM, Thompson FJ, Behrman AL, Trimble MH, and Kukulka CG. Comparison of single bout effects of bicycle training versus locomotor training on paired reflex depression of the soleus H-reflex after motor incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 90: 1218-1228, 2009.

Pickett GE, Campos-Benitez M, Keller JL, and Duggal N. Epidemiology of traumatic spinal cord injury in Canada. *Spine* 31: 799-805, 2006.

Platz T, Gillner A, Borgwaldt N, Kroll S, and Roschka S. Device-Training for Individuals with Thoracic and Lumbar Spinal Cord Injury Using a Powered Exoskeleton for Technically Assisted Mobility: Achievements and User Satisfaction. *BioMed research international* 2016: 8459018, 2016.

Pramodhyakul W, Wattanapan P, Siritaratiwat W, Eungpinichpong W, and Amatachaya S. Immediate effects of obstacle crossing training in independent ambulatory patients with spinal cord injury. *Spinal cord* 51: 379-383, 2013.

Priebe MM, Sherwood AM, Thornby JI, Kharas NF, and Markowski J. Clinical assessment of spasticity in spinal cord injury: a multidimensional problem. *Archives of physical medicine and rehabilitation* 77: 713-716, 1996.

Rossi A, Mazzocchio R, and Scarpini C. Clonus in man: a rhythmic oscillation maintained by a reflex mechanism. *Electroencephalography and clinical neurophysiology* 75: 56-63, 1990.

Roy RR, and Edgerton VR. Neurobiological perspective of spasticity as occurs after a spinal cord injury. *Experimental neurology* 235: 116-122, 2012.

Schindler-Ivens S, and Shields RK. Low frequency depression of H-reflexes in humans with acute and chronic spinal-cord injury. *Experimental brain research* 133: 233-241, 2000.

Schneider C, and Capaday C. Progressive adaptation of the soleus H-reflex with daily training at walking backward. *Journal of neurophysiology* 89: 648-656, 2003.

Scivoletto G, and Di Donna V. Prediction of walking recovery after spinal cord injury. *Brain research bulletin* 78: 43-51, 2009.

Scivoletto G, Morganti B, Ditunno P, Ditunno JF, and Molinari M. Effects on age on spinal cord lesion patients' rehabilitation. *Spinal cord* 41: 457-464, 2003.

Scivoletto G, Morganti B, and Molinari M. Neurologic recovery of spinal cord injury patients in Italy. *Archives of physical medicine and rehabilitation* 85: 485-489, 2004a.

Scivoletto G, Morganti B, and Molinari M. Sex-related differences of rehabilitation outcomes of spinal cord lesion patients. *Clin Rehabil* 18: 709-713, 2004b.

Scivoletto G, Romanelli A, Mariotti A, Marinucci D, Tamburella F, Mammone A, Cosentino E, Sterzi S, and Molinari M. Clinical factors that affect walking level and performance in chronic spinal cord lesion patients. *Spine* 33: 259-264, 2008.

Senthilvelkumar T, Magimairaj H, Fletcher J, Tharion G, and George J. Comparison of body weight-supported treadmill training versus body weight-supported overground training in people with incomplete tetraplegia: a pilot randomized trial. *Clin Rehabil* 29: 42-49, 2015.

Sezer N, Akkus S, and Ugurlu FG. Chronic complications of spinal cord injury. *World journal of orthopedics* 6: 24-33, 2015.

Shahani BT, and Young RR. Human flexor reflexes. *Journal of neurology, neurosurgery, and psychiatry* 34: 616-627, 1971.

Sharp KG, Gramer R, Butler L, Cramer SC, Hade E, and Page SJ. Effect of overground training augmented by mental practice on gait velocity in chronic, incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 95: 615-621, 2014.

Shemesh Y, Rozin R, and Ohry A. Electrodiagnostic investigation of motor neuron and spinal reflex arch (H-reflex) in spinal cord injury. *Paraplegia* 15: 238-244, 1977.

Simpson LA, Eng JJ, Hsieh JT, Wolfe, and the Spinal Cord Injury Rehabilitation Evidence Research Team DL. The health and life priorities of individuals with spinal cord injury: a systematic review. *Journal of neurotrauma* 29: 1548-1555, 2012.

Sinkjaer T, Andersen JB, Ladouceur M, Christensen LO, and Nielsen JB. Major role for sensory feedback in soleus EMG activity in the stance phase of walking in man. *The Journal of physiology* 523 Pt 3: 817-827, 2000.

Sinkjaer T, Andersen JB, and Larsen B. Soleus stretch reflex modulation during gait in humans. *Journal of neurophysiology* 76: 1112-1120, 1996.

Sipski ML, Jackson AB, Gomez-Marin O, Estores I, and Stein A. Effects of gender on neurologic and functional recovery after spinal cord injury. *Archives of physical medicine and rehabilitation* 85: 1826-1836, 2004.

Skold C, Levi R, and Seiger A. Spasticity after traumatic spinal cord injury: nature, severity, and location. *Archives of physical medicine and rehabilitation* 80: 1548-1557, 1999.

Smith AC, Mummidisetty CK, Rymer WZ, and Knikou M. Locomotor training alters the behavior of flexor reflexes during walking in human spinal cord injury. *Journal of neurophysiology* 112: 2164-2175, 2014.

Soffe SR. Two distinct rhythmic motor patterns are driven by common premotor and motor neurons in a simple vertebrate spinal cord. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 13: 4456-4469, 1993.

Spungen AM, Asselin P, Fineberg DB, Harel NY, Kornfeld S, and Bauman WA. Beneficial changes in body composition after exoskeletal-assisted walking: Implications for improved metabolic function. *Topics in Spinal Cord Injury Rehabilitation* 19: 8-9, 2013a.

Spungen AM, Asselin P, Fineberg DB, Kornfeld S, and Harel NY. Exoskeletal-assisted walking for persons with motor-complete paraplegia. In: *Force Sustainment: Rehabilitation, Regeneration and Prosthetics for Re-Integration to Duty Meeting Proceedings STO-MP-HFM-228, Paper 6* Neuilly-sur-Seine, France: Science and Technology Organization, 2013b, p. 6-1 - 6-14.

Stampacchia G, Rustici A, Bigazzi S, Gerini A, Tombini T, and Mazzoleni S. Walking with a powered robotic exoskeleton: Subjective experience, spasticity and pain in spinal cord injured persons. *NeuroRehabilitation* 39: 277-283, 2016.

Stein RB, Estabrooks KL, McGie S, Roth MJ, and Jones KE. Quantifying the effects of voluntary contraction and inter-stimulus interval on the human soleus H-reflex. *Experimental brain research* 182: 309-319, 2007.

Stein RB, and Kearney RE. Nonlinear behavior of muscle reflexes at the human ankle joint. *Journal of neurophysiology* 73: 65-72, 1995.

Stephens MJ, and Yang JF. Loading during the stance phase of walking in humans increases the extensor EMG amplitude but does not change the duration of the step cycle. *Experimental brain research* 124: 363-370, 1999.

Stephens MJ, and Yang JF. Short latency, non-reciprocal group I inhibition is reduced during the stance phase of walking in humans. *Brain Res* 743: 24-31, 1996.

Suzuki M, Miyai I, Ono T, Oda I, Konishi I, Kochiyama T, and Kubota K. Prefrontal and premotor cortices are involved in adapting walking and running speed on the treadmill: an optical imaging study. *Neuroimage* 23: 1020-1026, 2004.

Talaty M, Esquenazi A, and Briceno JE. Differentiating ability in users of the ReWalk(TM) powered exoskeleton: an analysis of walking kinematics. *IEEE International Conference on Rehabilitation Robotics : [proceedings]* 2013: 6650469, 2013.

Thomas SL, and Gorassini MA. Increases in corticospinal tract function by treadmill training after incomplete spinal cord injury. *Journal of neurophysiology* 94: 2844-2855, 2005.

Trimble MH, Behrman AL, Flynn SM, Thigpen MT, and Thompson FJ. Acute effects of locomotor training on overground walking speed and H-reflex modulation in individuals with incomplete spinal cord injury. *The journal of spinal cord medicine* 24: 74-80, 2001.

Trimble MH, Kukulka CG, and Behrman AL. The effect of treadmill gait training on low-frequency depression of the soleus H-reflex: comparison of a spinal cord injured man to normal subjects. *Neurosci Lett* 246: 186-188, 1998.

Ung RV, Imbeault MA, Ethier C, Brizzi L, and Capaday C. On the potential role of the corticospinal tract in the control and progressive adaptation of the soleus h-reflex during backward walking. *Journal of neurophysiology* 94: 1133-1142, 2005.

van der Laan L, Boks LM, van Wezel BM, Goris RJ, and Duysens JE. Leg muscle reflexes mediated by cutaneous A-beta fibres are normal during gait in reflex sympathetic dystrophy. *Clin Neurophysiol* 111: 677-685, 2000.

Van Wezel BM, Ottenhoff FA, and Duysens J. Dynamic control of location-specific information in tactile cutaneous reflexes from the foot during human walking. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 17: 3804-3814, 1997.

Varoqui D, Niu X, and Mirbagheri MM. Ankle voluntary movement enhancement following robotic-assisted locomotor training in spinal cord injury. *Journal of neuroengineering and rehabilitation* 11: 46, 2014.

Vilensky JA, and O'Connor BL. Stepping in nonhuman primates with a complete spinal cord transection: old and new data, and implications for humans. *Annals of the New York Academy of Sciences* 860: 528-530, 1998.

Visintin M, and Barbeau H. The effects of body weight support on the locomotor pattern of spastic paretic patients. *The Canadian journal of neurological sciences Le journal canadien des sciences neurologiques* 16: 315-325, 1989.

Wallace DM, Ross BH, and Thomas CK. Characteristics of lower extremity clonus after human cervical spinal cord injury. *Journal of neurotrauma* 29: 915-924, 2012.

Walsh EG. Clonus: beats provoked by the application of a rhythmic force. *Journal of neurology, neurosurgery, and psychiatry* 39: 266-274, 1976.

Wernig A, Nanassy A, and Muller S. Laufband (treadmill) therapy in incomplete paraplegia and tetraplegia. *Journal of neurotrauma* 16: 719-726, 1999.

Winchester P, Smith P, Foreman N, Mosby JM, Pacheco F, Querry R, and Tansey K. A prediction model for determining over ground walking speed after locomotor training in persons with motor incomplete spinal cord injury. *The journal of spinal cord medicine* 32: 63-71, 2009.

Wirz M, van Hedel HJ, Rupp R, Curt A, and Dietz V. Muscle force and gait performance: relationships after spinal cord injury. *Archives of physical medicine and rehabilitation* 87: 1218-1222, 2006.

Wirz M, Zemon DH, Rupp R, Scheel A, Colombo G, Dietz V, and Hornby TG. Effectiveness of automated locomotor training in patients with chronic incomplete spinal cord injury: a multicenter trial. *Archives of physical medicine and rehabilitation* 86: 672-680, 2005.

Yamaguchi T. Muscle activity during forelimb stepping in decerebrate cats. *The Japanese journal of physiology* 42: 489-499, 1992.

Yang A, Asselin P, Knezevic S, Kornfeld S, and Spungen AM. Assessment of In-Hospital Walking Velocity and Level of Assistance in a Powered Exoskeleton in Persons with Spinal Cord Injury. *Top Spinal Cord Inj Rehabil* 21: 100-109, 2015.

Yang JF, and Musselman KE. Training to achieve over ground walking after spinal cord injury: a review of who, what, when, and how. *The journal of spinal cord medicine* 35: 293-304, 2012.

Yang JF, Musselman KE, Livingstone D, Brunton K, Hendricks G, Hill D, and Gorassini M. Repetitive mass practice or focused precise practice for retraining walking after incomplete spinal cord injury? A pilot randomized clinical trial. *Neurorehabilitation and neural repair* 28: 314-324, 2014.

Yang JF, Norton J, Nevett-Duchcherer J, Roy FD, Gross DP, and Gorassini MA. Volitional muscle strength in the legs predicts changes in walking speed following locomotor training in people with chronic spinal cord injury. *Physical therapy* 91: 931-943, 2011.

Yang JF, and Stein RB. Phase-dependent reflex reversal in human leg muscles during walking. *Journal of neurophysiology* 63: 1109-1117, 1990.

Yang JF, Stein RB, and James KB. Contribution of peripheral afferents to the activation of the soleus muscle during walking in humans. *Experimental brain research* 87: 679-687, 1991.

Yang JF, Stephens MJ, and Vishram R. Infant stepping: a method to study the sensory control of human walking. *The Journal of physiology* 507 (Pt 3): 927-937, 1998a.

Yang JF, Stephens MJ, and Vishram R. Transient disturbances to one limb produce coordinated, bilateral responses during infant stepping. *Journal of neurophysiology* 79: 2329-2337, 1998b.

Yates C, Charlesworth A, Allen SR, Reese NB, Skinner RD, and Garcia-Rill E. The onset of hyperreflexia in the rat following complete spinal cord transection. *Spinal cord* 46: 798-803, 2008.

Zehr EP, Komiyama T, and Stein RB. Cutaneous reflexes during human gait: electromyographic and kinematic responses to electrical stimulation. *Journal of neurophysiology* 77: 3311-3325, 1997.

Zehr EP, and Stein RB. What functions do reflexes serve during human locomotion? *Progress in neurobiology* 58: 185-205, 1999.

Zehr EP, Stein RB, and Komiyama T. Function of sural nerve reflexes during human walking. *The Journal of physiology* 507 (Pt 1): 305-314, 1998.

Zeilig G, Weingarden H, Zwecker M, Dudkiewicz I, Bloch A, and Esquenazi A. Safety and tolerance of the ReWalk exoskeleton suit for ambulation by people with complete spinal cord injury: a pilot study. *The journal of spinal cord medicine* 35: 96-101, 2012.

Zewdie ET, Roy FD, Yang JF, and Gorassini MA. Facilitation of descending excitatory and spinal inhibitory networks from training of endurance and precision walking in participants with incomplete spinal cord injury. *Prog Brain Res* 218: 127-155, 2015.

CHAPTER 2: TRAINING-SPECIFIC NEURAL PLASTICITY IN SPINAL REFLEXES AFTER INCOMPLETE SPINAL CORD INJURY

A version of this chapter has been published.

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2.1 Introduction

Intensive motor training induces task-specific changes in the excitability of spinal reflexes in people without injuries to the central nervous system (Koceja et al. 2004; Zehr 2006). For example, resistance training of the plantarflexor muscles over many weeks increases the excitability of the soleus (SOL) Hoffmann (H) reflex (Aagaard et al. 2002; Lagerquist et al. 2006), whereas balance training on unstable surfaces for a few weeks does the opposite (Behrens et al. 2015; Gruber et al. 2007; Keller et al. 2012). Furthermore, professional athletes who spend years training intensively in their sport show sport-specific reflex excitability. For example, ballet dancers and athletes training in explosive leg sports (e.g., sprinting) show small H-reflexes and tendon reflexes in the SOL (Casabona et al. 1990; Goode and Van Hoven 1982; Nielsen et al. 1993a), whereas athletes training in endurance sports, such as middle and long distance running, show large H-reflexes (Rochcongar et al. 1979). Thus, motor training alters reflexes in a task-specific way.

Individuals with spinal cord injury (SCI) can exhibit abnormally exaggerated reflex excitability, for example, in the tendon or H-reflex of the soleus (SOL) (Taylor et al. 1984) and in the cutaneous or cutaneomuscular reflex elicited by stimulation of peripheral nerves at the ankle (Roby-Brami and Bussel 1987). Whether this increased reflex excitability is useful for, or detrimental to, functional movements remains uncertain (Dietz and Sinkjaer 2007; Nielsen et al. 2007). Some suggest that exaggerated reflex excitability might be useful because it can augment hand grip, aid movements such as transfers, and help maintain muscle mass and strength (Reyes et al. 2015), although this is largely based on expert opinion (Dietz 2003; Dietz and Sinkjaer 2007).

As with uninjured individuals, intensive motor training after SCI, such as walking on a treadmill, is associated with changes in spinal reflexes such as reduced clonus (Gorassini et al. 2009; Manella and Field-Fote 2013), stretch reflex amplitude (Mirbagheri et al. 2015), and flexor reflex excitability (Hubli et al. 2012; Smith et al. 2014). It remains to be determined if these individuals also show training-specific spinal plasticity in response to different forms of exercises.

We studied two forms of walking training in people with incomplete SCI, one emphasizing walking for as long and as fast as possible on a treadmill, Endurance Training, and the other emphasizing stepping over obstacles and precisely onto targets overground, Precision Training. We previously reported functional improvements in both walking endurance and skill (Yang et al. 2014) and strengthening of the corticospinal pathways to the tibialis anterior (TA) motoneurons (Zewdie et al. 2015) after both forms of training.

Here, we report the neural plasticity induced in spinal circuits from cutaneomuscular afferents in the foot to the SOL motoneurons and their antagonist, the TA, in the same individuals but during the functional task of walking. We examined two reflexes during treadmill walking: the cutaneomuscular reflex (CMR), induced by stimulating the posterior tibial nerve (PTN) (Jones and Yang 1994; Yang and Stein 1990), and clonus, induced by the natural recurrent activation of monosynaptic Ia inputs by the walking (Rossi et al. 1990; Wallace et al. 2012). Portions of this data have been presented in abstract form (Khan et al. 2014).

2.2 Materials and methods

2.2.1 Participants

Participants were recruited through a website (http://www.scialberta.ca), through clinicians and from personal contact in the community. Inclusion criteria include non-progressive SCI between C1–L1 neurological levels acquired ≥7 months before enrolment, with the ability to provide informed consent, walk independently for ≥5 meters using walking aids and/or braces, and attend training sessions 5 days/week. Exclusion criteria include severe head injury, cognitive impairment, or other comorbidities that preclude participation in an intensive training program or

transcranial magnetic stimulation (TMS, one of the outcome measures of the training (Zewdie et al. 2015)).

Ethical approval was provided by the Health Research Ethics Board at the University of Alberta and Alberta Health Services, Pro00003873. Written consent was obtained from all participants.

2.2.2 Experimental design

Participants were block randomized (block size = 4) to begin Endurance or Precision Training for two months (Phase 1), followed by two months of rest (Rest 1), and then crossed over to the other exercise regimen for another two months of training (Phase 2), followed by another 2 months of rest (Rest 2; Figure 2.1). Reflexes were measured twice at baseline (~1 week apart) and once after each period of training and rest (see *Section 2.2.4 Measurements*). Overground walking speed, distance, skill, and spinal reflexes were measured at the same time points (at black arrows in Figure 2.1).

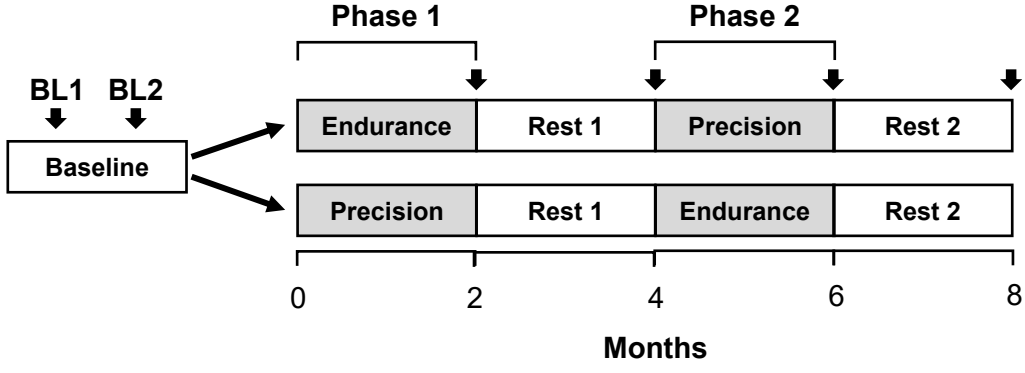

Figure 2.1: Experimental design.

Training consisted of two months of each of Endurance and Precision Training, in a crossover design with rest periods interspersed. Participants were randomly allocated to start with either Endurance or Precision Training. Reflex and walking measures were taken twice at baseline (BL; ~1 week apart) and after each training and rest period (solid vertical arrows).

2.2.3 Training

Target training frequency was ~1 hour/day, 3–5 days/week. Details of the training procedures, published previously (Yang et al. 2014), are briefly described below.

2.2.3.1 Precision training

Participants trained to step over obstacles without touching them and onto circular targets by obscuring the whole target with the foot, along a 15 m straight hallway with ~15 obstacles and targets spaced ~1 m apart. The difficulty of the initial training course was determined by trial and error, and it varied daily based on errors made on the previous day, aiming for an error rate of 20%. An error was defined as contact with an obstacle or failing to obscure a target completely. With improvement, the difficulty was increased by reducing support provided by walking aids, increasing obstacle height and width, increasing the size of the circular targets (making it more difficult to completely cover the target), and disallowing extra steps between obstacles and targets. Not all participants reached this last level of difficulty.

2.2.3.2 Endurance training

Participants were trained to walk for as long and as fast as possible on a level treadmill with minimal rests. Body-weight support (BWS) and manual stepping assistance were provided only if necessary. The initial treadmill speed was set to be higher than the baseline of overground walking speed (see 10-Meter Walk Test (10MWT) in Table 2.1). Progression in training is comprised of reducing BWS, manual stepping assistance, and rests, while increasing walking speed and distance.

2.2.4 Measurements

All measures were taken on a non-training day to minimize the effects of fatigue on the results of reflexes and the walking outcomes.

Table 2.1: Participant characteristics

Endurance Training in Phase 1							
Participant code-gender	**Age (yr)**	**Neurological level of injury**	**Cause of injury**	**Time since injury (yr)**	**10MWT (m/s)**	**6MWT (m)**	**SCI-FAP score**
P1-F	49	C6	MVA	2.5	0.05	17	649
P2-M	24	T6	MVA	1	0.37	114	119
P3-M	25	T4	MVA	1.1	0.13	48	235
P4-M	57	C5	MVA	34.9	0.90	298	59
P5-M	48	T12	Fall	0.7	0.23	119	140
P6-M	65	C3	MVA	1.2	0.05	19	864
P7-M	60	C3	Bull attack	0.6	0.47	172	286
P8-M	46	C6	MVA	7.3	0.14	43	308
P9-F	63	C4	MVA	6	1.13	278	10
P10-F	45	T2	Surgical clot	2.3	0.33	115	133
Mean (SD)	**48.2 (14.4)**			**5.8 (10.5)**	**0.38 (0.37)**	**122 (101)**	**280 (273)**
Precision Training in Phase 1							
Participant code-gender	**Age (yr)**	**Neurological level of injury**	**Cause of injury**	**Time since injury (yr)**	**10MWT (m/s)**	**6MWT (m)**	**SCI-FAP score**
P11-M	43	T12	Sport	17.5	1.27	392	10
P12-M	63	C4	Sport	20	0.82	254	31
P13-F	21	C6	MVA	1	0.25	67	262
P14-M	61	C5	Sport	2.4	0.90	292	14
P16-M	34	C4	Gun shot	1.2	1.10	337	8
P17-M	32	T2	Fall	0.8	0.16	53	231
P18-F	41	T12	Infection	3.5	0.17	38	602
P19-F	50	T6	Tumor	1.8	0.20	67	165
P20-M	52	T10	Surgical bleed	1.4	0.37	120	117
Mean (SD)	**44.1 (13.8)**			**5.5 (7.6)**	**0.6 (0.4)**	**180 (138)**	**160 (192)**

F: female; M: male; C: cervical; T: thoracic; MVA: motor vehicle accident; 10MWT: 10-Meter Walk Test; 6MWT: 6-Minute Walk Test; SD: standard deviation; SCI-FAP: Spinal Cord Injury-Functional Ambulation Profile.

2.2.4.1 Instrumentation

Bipolar surface electromyograms (EMG) were recorded from the TA, SOL, and abductor hallucis brevis (AHB) muscles using disposable Ag/AgCl electrodes (Kendall H59P, Mansfield, MA), placed 2 cm apart center-to-center. EMG signals were amplified and band-passed filtered between 10 and 1,000 Hz (AMT-8 EMG System, Bortec Biomedical Ltd., Calgary, AB, Canada). Knee angles were recorded using electrogoniometers bilaterally (Biometrics Ltd., type K100, Newport, UK) in the sagittal plane. All signals were digitized at 5 kHz (Axon Instruments, Digidata 1322A, Molecular Devices, Sunnyvale, CA) and stored for offline analyses.

2.2.4.2 Cutaneomuscular reflex and clonus

Reflexes were measured during walking on a level treadmill, with the speed and BWS being held constant across all testing sessions. The CMR was measured from the SOL and TA muscles in the more spastic leg on the basis of the participant's self-report and opinion of the clinical staff. The CMR was recorded in response to triple pulse stimulation (200 Hz, 0.3 ms pulse width; Digitimer, DS7A, Hertfordshire, England) of the posterior tibial nerve at the level of the medial malleolus, delivered pseudorandomly at intervals between 2 and 5 sec to reduce predictability. The posterior tibial nerve was chosen because (i) it is a mixed nerve that allows recording of an M-wave during movement to ensure that responses are compared at similar effective stimulus intensities (i.e., matched M-wave size) and (ii) stimulus intensity can be compared between days for participants that have impaired sensation. The nerve was first located with a hand-held probe, as we have done in the past (Yang and Stein 1990), and once an optimal location was identified, the probe was replaced with a disposable 1 cm diameter Ag/AgCl adhesive electrode. The anode was a 5 cm × 10 cm disposable adhesive electrode (Axelgaard Manufacturing Co., Fallbrook, CA), placed on the lateral aspect of the ankle joint. Both electrodes were held in place with additional skin tape. Stimuli were applied at a few intensities around 1.5× motor threshold (MT) of the AHB muscle (typically at 1.5× MT and then one intensity above and one below 1.5× MT) to ensure similar stimulus intensities (i.e., matched M-waves) throughout walking and across different experiment days (see *Section 2.2.5 Data analyses*), as done in the past (Jones and Yang 1994; Yang and Stein 1990). The intensity of 1.5× MT was chosen to be consistent with previous reports of this reflex (Yang and Stein 1990). Motor threshold, estimated in standing, was defined as the stimulus intensity eliciting an M-wave with 50% probability. A minimum of 3 to 4 trials

(~3 min each) were required in order to provide a sufficient number of sweeps to match the M-wave size during the walking cycle.

A single trial of ~3 min walking without stimulation (i.e., unperturbed walking) was also recorded. EMG from this control trial was subtracted from the EMG trace obtained during the stimulation trials to reveal the reflex response. The SOL EMG during the entire length of the unperturbed walking trial was also used to estimate clonus ((Gorassini et al. 2009); see *Section 2.2.5 Data analyses*).

2.2.4.3 Over ground walking

Self-selected walking speed was estimated using the 10MWT (van Hedel et al. 2005) (time to walk the middle 10 m of a 14 m straight track). The 6-Minute Walk test (6MWT) measured the maximum distance covered while walking back and forth along a straight 30 m track for 6 min (Guyatt et al. 1985). The Spinal Cord Injury-Functional Ambulation Profile (SCI-FAP), which contains 7 timed tasks during walking (carpet, up-and-go, walking over and around obstacles, up and down stairs, up and down a curb, walking while carrying a bag, and walking while opening door), measured walking skill (Musselman et al. 2011). All measures have been validated for people with SCI (Musselman and Yang 2014; van Hedel et al. 2005).

2.2.5 Data analyses

2.2.5.1 Cutaneomuscular reflex

Reflexes were analyzed using custom-written MatLab programs (The MathWorks, Inc., Natick, MA). The EMG responses were sorted according to the time of occurrence of the stimulus within the walking stride. The beginning of a walking cycle was set to be near mid-swing on the stimulated side, based on a threshold crossing of the knee goniometer signal. Because the duration of each stride can vary, the strides were first normalized in time from the beginning of one cycle to the beginning of the next cycle and then divided into 8 bins of equal duration, using the MatLab software. Reflexes elicited with AHB M-wave amplitudes (peak-to-peak) within a target range were analyzed to control for stimulus intensity during movement, as pioneered by Capaday and Stein (1986). The target range was selected after examining the distribution of M-wave amplitudes for each participant before and after training, with the range being restricted to

differences in the upper and lower limits of <20% of the maximum M-wave (M_{max}), consistent with previous reports (Yang and Stein 1990). Reflex responses in each bin were full-wave rectified, low-pass filtered at 200 Hz (dual-pass, zero phase, 2nd-order, digital Butterworth filter), time-locked to the stimuli, and averaged.

EMG traces during unperturbed walking were similarly divided into 8 equal bins and subtracted from EMG responses with stimulation to reveal the reflex response (Jones and Yang 1994; Yang and Stein 1990). All acceptable reflexes were analyzed, unless the subtraction was deemed unsatisfactory by visual inspection, revealed by a non-zero baseline prior to the stimulus. This ensured that the levels of EMG were matched across the control and stimulated conditions. The start and end times (i.e., window) of reflex responses were determined by visually examining the response across all 8 bins separately for each participant (e.g., Figures 2.2(a) and 2.2(b), vertical dashed lines) and kept consistent before and after training. The reflex response was the averaged EMG amplitude over the duration of the window for each bin (e.g., Figures 2.2(a) and 2.2(c) (SOL) and 2.2(b) and 2.2(d) (TA)) within a participant. These bin averages were then averaged bin-by-bin across participants (e.g., Figures 2.3(a)(i) and 2.3(b)(i)) to reveal the pattern of reflex modulation of the CMR over the step cycle. Reflex inhibition and excitation in the step cycle were also quantified using the maximum inhibition (for SOL only, e.g., Figure 2.2(c)) and excitation (for TA and SOL, e.g., Figures 2.2(c) and 2.2(d)) of the CMR, wherever it happened in the step cycle and used to correlate with the walking outcomes. Since there were both excitation and inhibition in the SOL through the cycle but only excitation in the TA, only these were quantified. As the walking outcomes have been reported previously (Yang et al. 2014), those analyses are not repeated here.

2.2.5.2 Clonus

The raw SOL EMG signal was corrected for the DC offset, rectified, and analyzed in the frequency domain using fast Fourier transform within the clonus frequency range set to 4–10 Hz in agreement with other studies (Beres-Jones et al. 2003; Gorassini et al. 2009; Rossi et al. 1990; Wallace et al. 2012; Walsh 1976). To normalize the amplitude of the clonic EMG bursts to that of the regular EMG during walking, the 4–10 Hz signal power was expressed as a fraction of the total power within 0–40 Hz, according to clonus power = (power between 4–10 Hz)/(power between 0–40 Hz). Frequencies >40 Hz were excluded to avoid power-line noise. Clonus power

from the more spastic side, that is, the side with the higher average clonus power across the baseline measures, was used in the analyses.

2.2.6 Statistical analyses

2.2.6.1 Were the two baseline measures repeatable?

The two baseline measures of reflexes taken approximately 1 week apart (i.e., BL1 and BL2 in Figure 2.1) were compared for all participants, regardless of which training they did first, to determine repeatability using the Paired Samples t-test (or Wilcoxon Signed Rank test for data that was not normally distributed). Baseline comparisons were made for the maximum inhibition, maximum excitation, and clonus, and a two-way repeated-measures ANOVA for the reflex modulation of the CMR (factor 1: time across the step cycle (i.e., bins 1–8); factor 2: time of measurement of the baseline measures (i.e., Baseline 1 and Baseline 2)). In cases where the two baseline measures were not statistically different (all CMR data), the two baseline measures were averaged to provide the best estimate for pretraining conditions. Participant characteristics (age, time since injury, 10MWT, 6MWT, and SCI-FAP) at baseline were also compared between the 2 groups (Precision or Endurance first) using an Independent Sample t-test (or Mann–Whitney U test).

2.2.6.2 Did the two types of training affect reflex excitability in different ways?

<u>CMR modulation in walking</u>

The CMR was quantified for before and after each type of training separately, regardless of when the training occurred (i.e., Phase 1 or 2). For example, for those who had Endurance Training in Phase 1, before and after training measures were those taken at baseline and at the end of Phase 1 (i.e., at end of Month 2; see Figure 2.1), respectively. For those who had Endurance Training in Phase 2, before and after training measures were those taken at the end of Rest 1 (i.e., end of Month 4) and at end of Phase 2 (i.e., end of Month 6), respectively. The CMR for each of the two groups before and after Endurance Training was then pooled across phase (i.e., Phase 1 and Phase 2) to counterbalance the possible effect of the order of training [Figure 2.1, see also (Yang et al. 2014)]. The same was done for Precision Training.

The distribution of the data was tested using the Shapiro-Wilk test for normality. To determine the effects of training on reflex excitability, the modulation of the CMR across the step cycle was compared using a 2-way repeated-measures ANOVA, with treatment effect (i.e., before and after) and time bin of the step cycle (i.e., bins 1–8) as the two factors. Missing data in the ANOVA, such as when data was excluded at certain bins due to unsatisfactory subtraction of the background EMG (see *Section 2.2.5 Data analyses*), was managed by replacing the missing value with the average reflex value for the respective bins. A Greenhouse-Geisser correction was applied if the assumption of sphericity was not satisfied (i.e., $p<0.05$ on Mauchly's test of sphericity) or if the test was not performed due to insufficient degrees of freedom as a result of a small sample size. Post hoc analysis of significant findings was performed using Tukey's Honestly Significant Difference test.

To ensure that the results were not distorted by the crossover design, two further analyses were performed. First, the changes as a result of Precision or Endurance Training were compared for Phase 1 only, using the same statistical procedures (as above). Second, to determine if the rest period between Phases 1 and 2 resulted in changes to the reflexes, a similar 2-way repeated-measures ANOVA was used to compare the measures at the beginning and at end of Rest 1 (i.e., at the end of Month 2 versus Month 4, Figure 2.1). To be complete, the comparisons were also made for Rest 2 (i.e., at the end of Month 6 versus Month 8).

Clonus power

Clonus was higher at Baseline 1 compared to Baseline 2 (Baseline 1: 0.13 ± 0.08, and Baseline 2: 0.10 ± 0.07; paired t-test: $p = 0.031$), which could be related to familiarity with the testing environment, since nervousness during Baseline 1 could have caused greater reflex excitability (Mahoney et al. 2007). Hence, we used clonus power at Baseline 2 as the more conservative estimate for clonus before Phase 1. Otherwise, comparisons were made for clonus measured before and after each form of training and collapsed across phase as described for the CMR (above). Unlike the CMR modulation, there was only one measure taken before and after each type of training, so a Paired Samples t-test (or Wilcoxon Signed Rank test) was used to determine if there was a significant change as a result of the training.

2.2.6.3 Was the reflex excitability related to walking ability?

The relationship between reflex excitability and walking ability was quantified with linear regression. The maximum inhibition of the CMR during the walking cycle and the clonus power were each examined as a function of the 3 walking measures (10MWT, 6MWT, and SCI-FAP) prior to the start of any training. Since there were strong trends, including significant relationships between clonus and walking ability, we further determined if the overall change in clonus (i.e., from Baseline 2 to the end of Month 6) was related to the overall change in walking ability. The relationship between maximum inhibition of the SOL CMR and walking ability was similarly examined for before Endurance Training, since only Endurance Training induced a significant change in the CMR inhibition (see below). The relationship between change in the maximum inhibition of the SOL CMR and change in walking ability was also examined. Significance for all tests was set to $p < 0.05$.

2.3 Results

2.3.1 Participants

Of the original 20 participants who contributed data to the previous paper (Yang et al. 2014), all completed Precision Training and 17 completed Endurance Training. One participant (P15) was excluded from the current paper because he had no muscle activity in the TA and SOL (lesion level L1). Participant characteristics are shown in Table 2.1, with the order of training indicated; no differences in participant characteristics were observed at baseline between the two groups. To facilitate comparison with our previous publication (Yang et al. 2014), participants are described using the same codes.

For measures of clonus, all 19 participants were included in our analysis of Precision Training, and 14 for Endurance Training. Two participants were excluded from analysis of Endurance Training because one had large single motor unit activity in the SOL during walking (P4), whose firing rates confounded the clonus estimate (Gorassini et al. 2004), and the other had a technical problem with the EMG recording (P18).

For the CMR, 14 data sets were included for Precision Training and 12 for Endurance Training. Three participants were excluded from the analysis of both forms of training because the stimuli

either stopped their walking (P6, P18) or induced extensive clonus (P8), making it impossible to collect this data. P4 was additionally excluded from analysis of Endurance Training due to inadequate number of responses with acceptable M-wave matches. Two additional participants were excluded from the analysis of Precision Training due to extensive clonus generated by the stimuli (P3), or poor subtraction of unperturbed EMG, resulting in uncertainty of the CMR response (P12).

The walking speeds during reflex testing ranged from 0.13 to 0.89 m/s (mean ± SD = 0.35 ± 0.18 m/s). Body-weight support was used by 6 participants, which ranged from 9 to 27 kg (mean ± SD = 20 ± 7 kg).

2.3.2 Baseline measures

The pattern of reflex modulation of the CMR across the step cycle did not differ between the two baselines (2-way repeated-measures ANOVA (factor 1: bin number; factor 2: baseline measure) and ANOVA interaction effect: SOL: $p = 0.84$ and TA: $p = 0.3$), neither did the maximum inhibition (Baseline 1: −15.5 ± 18.3 μV and Baseline 2: −16.3 ± 14.5 μV; paired t-test: $p = 0.71$) and excitation (Baseline 1: 7.4 ± 6.2 μV and Baseline 2: 6.4 ± 6.0 μV; paired t-test: $p = 0.76$) of the SOL CMR and the maximum excitation of the TA CMR (Baseline 1: 33.2 ± 24.6 μV and Baseline 2: 25.2 ± 27.5 μV; paired t-test: $p = 0.31$). Clonus power at Baseline 1 was higher than at Baseline 2 (Baseline 1: 0.13 ± 0.08 and Baseline 2: 0.10 ± 0.07; paired t-test: $p = 0.031$), so Baseline 2 was used to represent pretraining values for Phase 1 (see *Section 2.2.6 Statistical analyses* for rationale).

2.3.3 Cutaneomuscular reflexes

Representative data from participant P12 is shown in Figures 2.2(a) and 2.2(b), illustrating reflex modulation across a walking cycle for the SOL and TA muscles, respectively. Vertical dashed lines show the time window of the reflex responses used for averaging for this participant, with the averaged responses plotted in Figures 2.2(c) and 2.2(d). Average reflex window onset and offset times across participants were 44.7 ± 6 to 107.9 ± 29.4 ms for the SOL and 48.4 ± 8.1 to 101.3 ± 15.9 ms for the TA. Reflex responses during walking in the SOL muscle were dominated by inhibition while those in the TA were dominated by excitation. For this participant, the

maximum excitation and inhibition of the SOL CMR occurred in bins 1 and 6, respectively, and maximum excitation of the TA CMR in bin 8.

The repeated-measures ANOVA comparing group data of the reflex amplitude in the SOL before and after Endurance Training (Figure 2.3(a)(i)) indicated a significant interaction ($p = 0.047$) between treatment effect and time bin of the step cycle and a significant training effect ($p = 0.011$). Post hoc analysis revealed significant increase in inhibition at bins 5 ($p = 0.014$), 6 ($p = 0.0048$), and 7 ($p = 0.044$), resulting in a deeper reflex modulation after training. EMG amplitude during unperturbed walking did not change in the SOL after Endurance Training (ANOVA: interaction effect, $p = 0.5$; training effect, $p = 1$; Figure 2.3(a)(ii)), suggesting that the greater inhibition is not due to changes in the background muscle excitability. Modulation of reflex excitation in the TA showed no significant changes after Endurance Training (ANOVA: interaction effect, $p = 0.075$; training effect, $p = 0.082$; Figure 2.3(b)(i)), as with the background EMG during unperturbed walking (ANOVA: interaction effect, $p = 0.36$; training effect, $p = 0.53$; Figure 2.3(b)(ii)). Precision Training had no effect on the CMR and background EMG activity in either muscle (ANOVA, data not shown). Changes in individual participants before and after Endurance Training are shown in Figure 2.3(a)(iii). With 2 exceptions, the reflex inhibition of the SOL CMR was enhanced with training.

To determine if there was washout during the rest periods, we compared the modulation of the SOL CMR at the beginning and end of each rest period for Endurance and Precision Training, separately. No differences were found in the 2-way repeated-measures ANOVA (Rest 1: no interaction for Endurance $p = 0.38$ and Precision $p = 0.29$; Rest 2: no interaction for Endurance $p = 0.25$ and Precision $p = 0.47$). Since there was no washout, the residual effects from the first phase of training could have affected the second phase of training. Hence, we reanalyzed the reflex modulation in the SOL separately using Phase 1 data only (i.e., $n = 6$ and 7 for Endurance and Precision Training, resp.). The same trends were observed for the two forms of training: greater inhibition after Endurance Training (ANOVA: interaction effect, $p = 0.09$) and no change after Precision Training (ANOVA: interaction effect, $p = 0.61$).

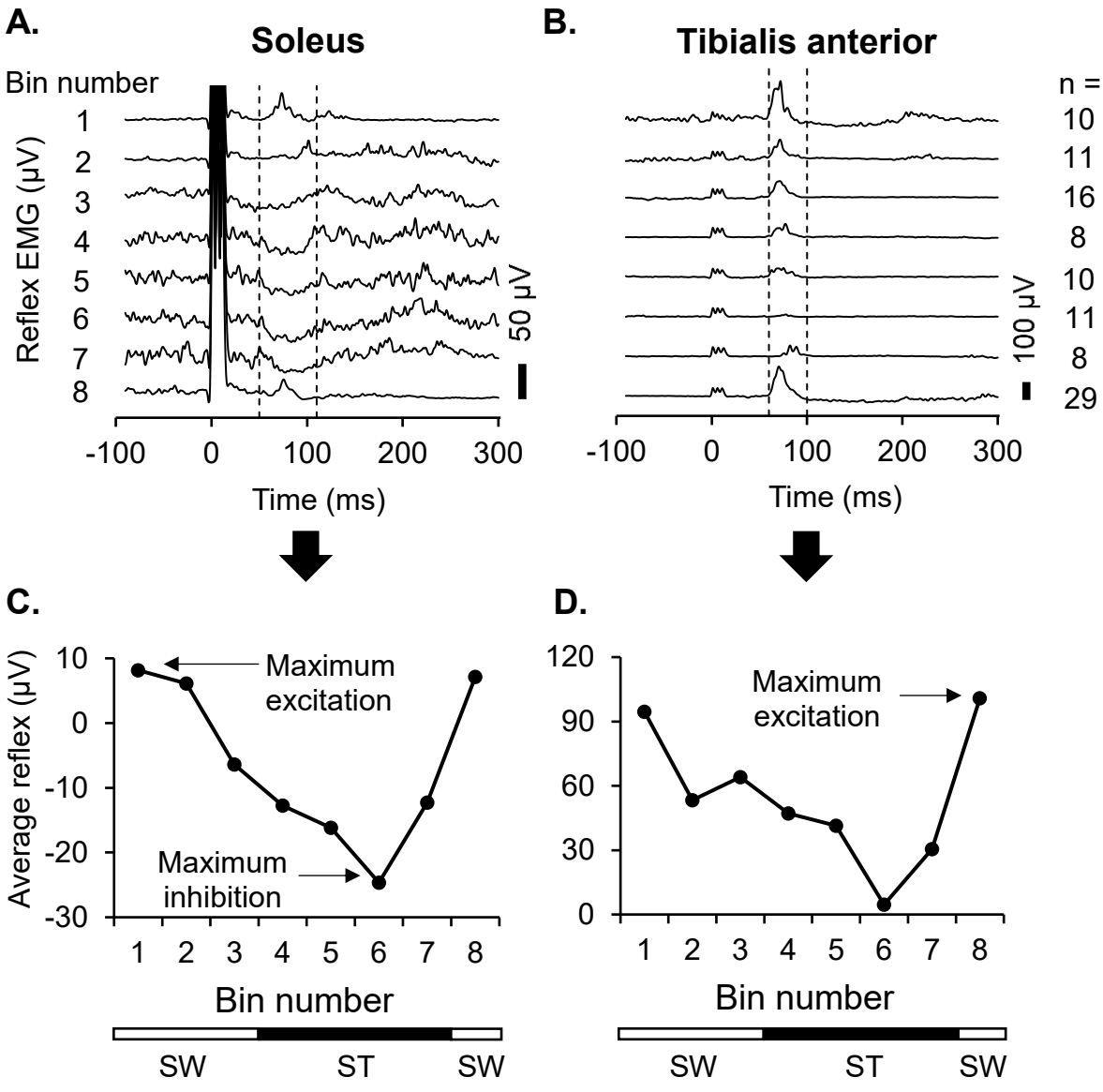
A.
Soleus
Bin number
1
2
3
4
5
6
7
8
Reflex EMG (μV)
50 μV
-100
0
100
200
300
Time (ms)
B.
Tibialis anterior
n =
10
11
16
8
10
11
8
29
100 μV
-100
0
100
200
300
Time (ms)
C.
Average reflex (μV)
10
0
-10
-20
-30
Maximum excitation
Maximum inhibition
1 2 3 4 5 6 7 8
Bin number
SW
ST
SW
D.
120
90
60
30
0
Maximum excitation
1 2 3 4 5 6 7 8
Bin number
SW
ST
SW

Figure 2.2: The cutaneomuscular reflex (CMR) during walking in a single participant.

Average EMG traces from the soleus (a) and tibialis anterior (b) muscles, time-locked to the beginning of the stimulus (time = 0), are shown for the 8 bins of the walking cycle from participant P12 after subtraction of the background EMG during unperturbed walking. The number of acceptable reflexes within each time bin (bin number) across the step cycle is indicated on the far right. Data were obtained after the first rest period (i.e., at Month 4) prior to Endurance Training. Triple pulses (200Hz; 0.3ms pulse width; at ~1.5 × motor threshold for the abductor hallucis brevis (AHB) muscle) were applied to the posterior tibial nerve behind the medial malleolus during walking on a treadmill. Responses displayed are matched for M-wave amplitudes, measured from the AHB (see *Section 2.2.5 Data analyses* for details). Responses for each of the 8 bins of the walking cycle were quantified by averaging the EMG between vertical dashed lines in (a) and (b) and plotted as a function of time in the walking cycle in (c) and (d). Correspondences of the bin numbers with stance (ST) and swing (SW) phases are shown by the solid and open bars, respectively, at the bottom of the graphs. Horizontal arrows in (c) and (d) indicate maximum inhibition and excitation.

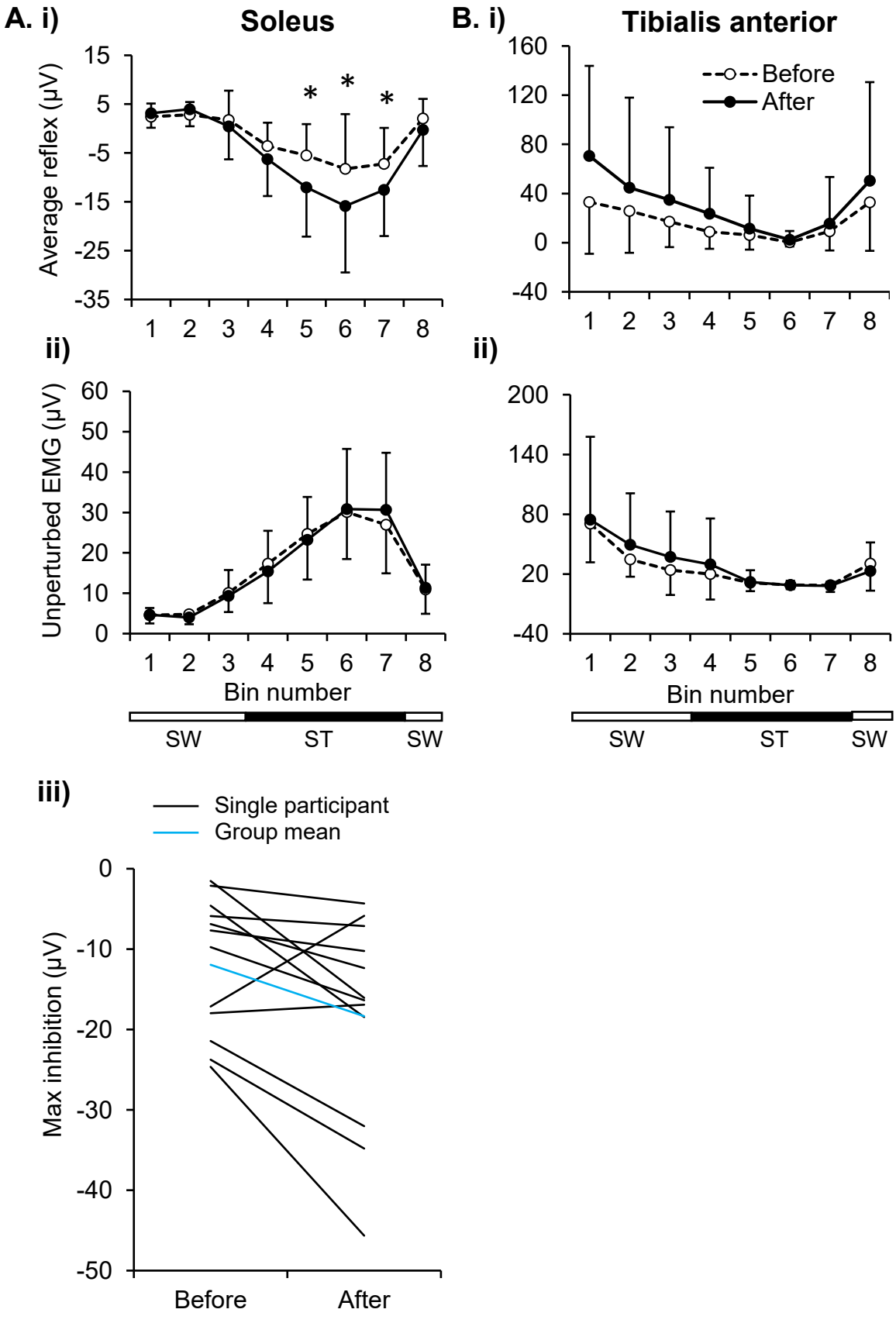
A. i)
Soleus
Average reflex (μV)
15
5
-5
-15
-25
-35
1 2 3 4 5 6 7 8
*
*
*
B. i)
Tibialis anterior
160
120
80
40
0
-40
Before
After
1 2 3 4 5 6 7 8
ii)
Unperturbed EMG (μV)
60
50
40
30
20
10
0
1 2 3 4 5 6 7 8
Bin number
SW
ST
SW
ii)
200
140
80
20
-40
1 2 3 4 5 6 7 8
Bin number
SW
ST
SW
iii)
Single participant
Group mean
Max inhibition (μV)
0
-10
-20
-30
-40
-50
Before
After

Figure 2.3: Changes in reflex excitability during walking varied with training type.

The cutaneomuscular reflex (CMR) during 8 bins in the walking cycle, averaged across participants (n = 12), are shown for the soleus (a)(i) and tibialis anterior (b)(i) muscles before and after Endurance Training. Significant differences in post hoc comparisons are indicated with *. Average EMG corresponding to the bin numbers during unperturbed walking trials are shown in (a)(ii) and (b)(ii). Error bars represent one standard deviation. ST: stance phase (solid bars); SW: swing phase (open bars). (a)(iii) Maximum (Max) inhibition of the SOL CMR before and after Endurance Training is shown for each participant, with the group mean superimposed.

2.3.4 Clonus

Clonus power changed in different ways for different participants, but those who showed a reduction in clonus also showed an improvement in walking function. Figures 2.4(a) and 2.4(b) illustrate rectified, smoothed EMG traces from the SOL muscle during unperturbed walking on a treadmill, from participants exhibiting a decrease (Figure 2.4(a)) and a small increase (Figure 2.4(b)) in clonus with training, with large and small increases in walking function (i.e., speed and distance, resp.). Since there were no differences in the way clonus responded to the 2 forms of training across participants, we collapsed the data across training type and considered the change in clonus for each participant from Baseline 2 to the end of training in Phase 2 (i.e., end of Month 6). Changes in clonus power for P17 and P13 are shown in Figure 2.4(c)(i), with changes in walking ability shown in Figures 2.4(c)(ii) and 2.4(c)(iii).

2.3.5 Relationship between reflex excitability and walking measures

The relationship between the SOL CMR and walking measures was examined for Endurance Training, since only Endurance Training induced a significant enhancement of the inhibition in the SOL CMR. Prior to the start of Endurance Training, there was a relationship between the maximum inhibition in the SOL CMR during walking and walking speed (10MWT, Figure 2.5(a)(i); $r = 0.65$; $p = 0.022$) and walking distance (6MWT, not shown; $r = 0.69$; $p = 0.013$) and a weaker relationship with walking skill (SCI-FAP, not shown; $r = 0.53$; $p = 0.078$). Improved walking speed as a result of Endurance Training (i.e., Δ10MWT) was weakly associated with enhanced inhibition in the SOL CMR (i.e., Δmax inhibition), but the correlation was not significant (Figure 2.5(a)(ii); $r = 0.36$; $p = 0.26$). No relationships were observed between the change in the max inhibition of the CMR and walking distance (Δ6MWT, not shown; $r = 0.2$; $p = 0.53$) and skill (ΔSCI-FAP, not shown; $r = 0.0095$; $p = 0.98$).

There were trends for a relationship between clonus and walking measures prior to any training (10MWT in Figure 2.5(b)(i), $r = 0.45$, $p = 0.073$; 6MWT, not shown, $r = 0.43$, $p = 0.088$). Again, there was no relationship between clonus and walking skill (SCI-FAP, not shown; $r = 0.13$; $p = 0.61$). The reduction in clonus after all training was significantly related to improvements in walking speed (Figure 2.5(b)(ii), $r = 0.698$, $p = 0.006$) and walking distance (not shown, $r = 0.59$, $p = 0.026$) but not walking skill (not shown, $r = 0.065$, $p = 0.82$). Due to the possibility of a

single outlier in the bottom right corner of Figure 2.5(b)(ii) dominating the results, regression analyses were also performed without the outlier. There remained a trend in the relationship between change in clonus and change in walking speed ($r = 0.5$, $p = 0.083$), but the relationship between change in clonus and change in walking distance was no longer significant ($r = 0.29$, $p = 0.34$).

2.4 Discussion

The primary new finding is that contrasting forms of walking training have differential effects on the excitability of the CMR. Endurance Training enhanced the reflex inhibition in the SOL during walking, while Precision Training did not. Neither form of training changed clonus in a systematic way. Yet there is a significant relationship between the degree of clonus and walking ability, as well as a relationship between improvement in walking and reduction in clonus. Increase in the CMR inhibition induced by Endurance Training was weakly related to improvement in walking. These results suggest that certain forms of walking training can reduce reflex excitability, and some aspects of the reduced reflex excitability are associated with improved walking function.

2.4.1 Methodological limitations

As with all exercise interventions, the participants cannot be blinded to the intervention. While we told participants that we did not know which method would be better, the participants could have had their own biases. This may have affected the results but likely would not favor one type of training.

The crossover design could have led to confoundedness in Phase 2 of training, as there was no washout from the training in Phase 1. However, since the trends in the results remained the same when we analyzed Phase 1 alone, we do not think this would have changed our overall findings.

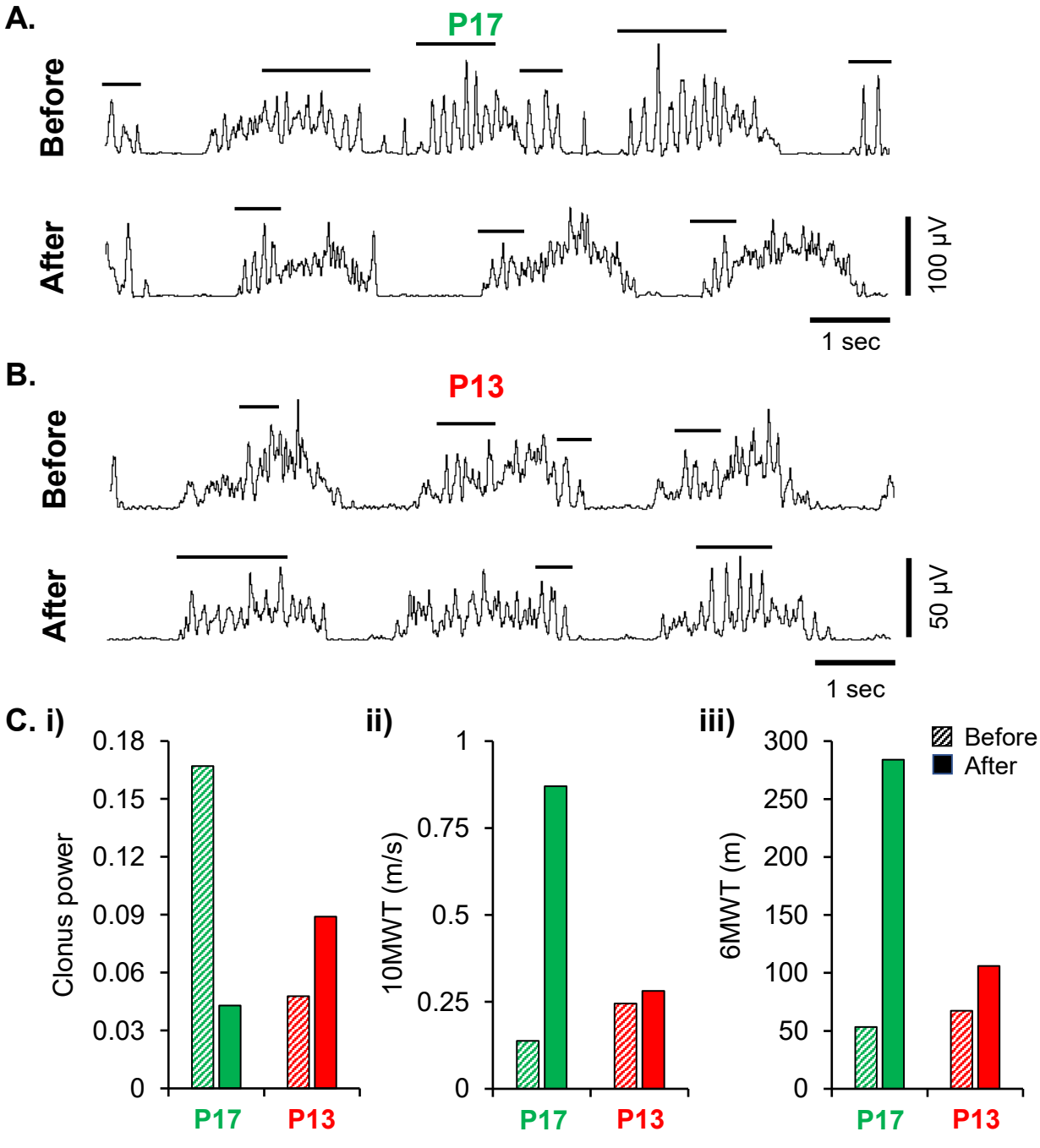
A.
P17
Before
After
100 µV
1 sec
B.
P13
Before
After
50 µV
1 sec
C. i)
Clonus power
0.18
0.15
0.12
0.09
0.06
0.03
0
P17
P13
ii)
10MWT (m/s)
1
0.75
0.5
0.25
0
P17
P13
iii)
6MWT (m)
300
250
200
150
100
50
0
Before
After
P17
P13

Figure 2.4: Clonus in the soleus muscle during unperturbed walking.

(a) and (b) Rectified and smoothed (dual-pass, zero phase, 20Hz lowpass, 2nd-order, digital Butterworth filter, for display purposes) EMG traces from the soleus muscle during unperturbed walking on a level treadmill, recorded before (i.e., at Baseline 2) and after both phases of training (i.e., at Month 6), from participants exhibiting (a) decrease and (b) a small increase in clonus after the training. (c)(i) The change in clonus over both phases of training (i.e., change from Baseline 2 to end of Month 6) and the corresponding changes in (ii) the 10-Meter Walk Test (10MWT) and (iii) 6-Minute Walk Test (6MWT) for both participants are shown for comparison. Horizontal lines indicate clonic EMG bursts.

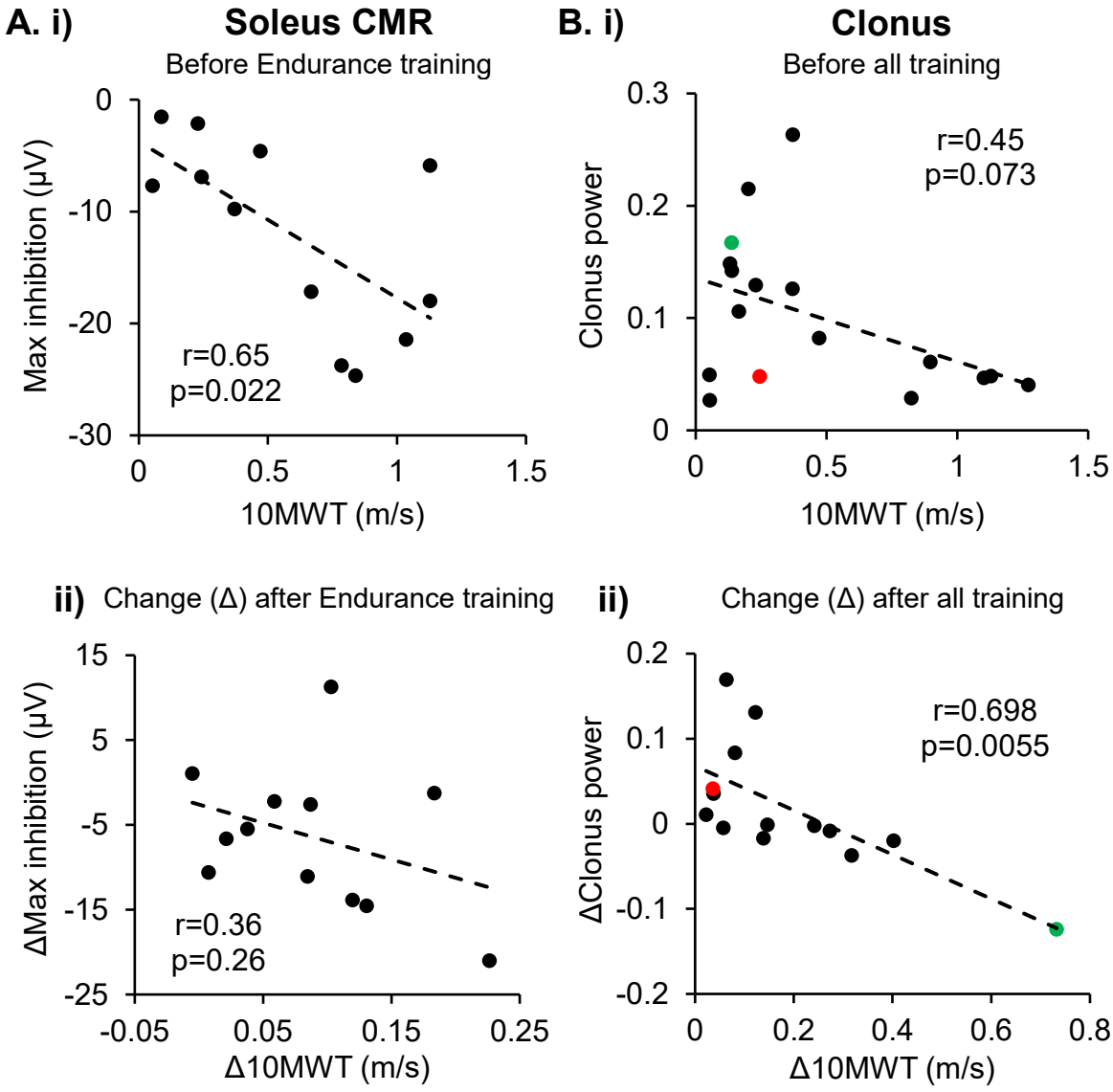

Figure 2.5: The relationship between reflex excitability and walking outcomes.

(a) The inhibition in the soleus CMR was related to walking speed before Endurance Training (i). Change in the maximum inhibition of the CMR as a result of Endurance Training was weakly related to the improvement in walking speed (ii). (b) Clonus in the soleus was weakly related to walking speed before any form of training (i.e., at baseline) (i). Reduction in clonus was significantly related to improvement in walking speed (ii). Green and red data points correspond to P17 and P13, respectively, from Figure 2.4.

2.4.2 How you train makes a difference

Different forms of training in uninjured individuals have differential effects on the functional outcome of walking and spinal reflex excitability (see *Section 2.1 Introduction*). Here, Endurance Training resulted in a small, yet significant, increase in inhibition of the SOL CMR during walking (Figure 2.3(a)) and improvements in walking speed and distance (Yang et al. 2014), whereas Precision Training only modestly improved walking (Yang et al. 2014) and did not change the reflex excitability. Interestingly, both forms of training strengthened the corticospinal tracts (CST), as measured using maximum motor evoked potentials from single-pulse TMS (Zewdie et al. 2015). Thus, neural plasticity was differentially affected at the spinal and cortical levels by the two different forms of training. Interestingly, in these injured individuals, the total duration of training to elicit a change was considerably less than the athletes mentioned in *Introduction*, although similar changes to reflex modulation have been induced in uninjured individuals training to execute unfamiliar tasks such as backward walking over a 10-day period (Schneider and Capaday 2003).

It might be argued that the change in CMR inhibition in the SOL was relatively minor, that is, about −10 μV on average (Figure 2.3(a)). However, the typical inhibition seen in uninjured individuals during the stance phase of walking is in the order of −30 μV (Jones and Yang 1994), and the pretraining inhibition in our participants was about −5 μV (Figure 2.3(a)), so an additional −10 μV is a considerable change in the direction towards the pattern of the uninjured.

The reason that Endurance Training was more effective in reducing spinal reflex excitability overall could have been because of the much greater number of steps executed during training compared to Precision Training (steps/session: Endurance ~1200; Precision ~400), or it could be that the two forms of training induced neural plasticity at different neural pathways. The two forms of training emphasized very different aspects of walking: speed and distance for Endurance Training versus skill and accuracy for Precision Training. Thus, it was not feasible to equate the number of steps executed in these two forms of training, because it would have required over 4 hours a day of Precision Training to obtain the same number of steps. Thus, while we do not know the "ingredient" that caused the reduction in reflex excitability with Endurance Training, we suggest that the more likely explanation is that the volume (i.e., number of steps) of training is especially important to induce plasticity in the reflex pathways, because

walking skill improved equally with both forms of training as measured by SCI-FAP (Yang et al. 2014). A definitive answer will require studies targeting this question specifically, with perhaps more sensitive measures of walking skill than those contained in SCI-FAP.

Two other studies have compared changes in spinal reflex excitability with different forms of walking training after SCI in humans (Adams and Hicks 2011; Manella and Field-Fote 2013). The changes in H_{max}/M_{max} ratio and clonus in the SOL muscle were compared after 4 types of training: (a) BWS treadmill training (BWSTT) with manual assistance, (b) BWSTT with functional electrical stimulation to the common peroneal nerve, (c) training in the Lokomat, and (d) overground training with functional electrical stimulation to the common peroneal nerve (Manella and Field-Fote 2013). While the excitability and duration of clonus were reduced after all forms of training, H_{max}/M_{max} ratio did not change. Thus, consistent with our study, different spinal reflexes could be differentially responsive to the training. The lack of difference between the different types of training could have been because the 4 training methods were more similar compared to the two in the current study, as all 4 training methods involved continuous stepping, with no differences in walking outcomes between the methods. In a different study, changes in clonus and H_{max}/M_{max} ratio were compared after 1 month of BWSTT and tilt table standing (Adams and Hicks 2011). Clonus and flexor spasms were reduced only after the treadmill walking, while extensor spasms were reduced after tilt table standing; no change was observed in H_{max}/M_{max} after either form of training (Adams and Hicks 2011).

2.4.3 Are training-induced changes in reflex behavior related to improvements in walking?

Many reports confirm that walking training changes reflex excitability in people with SCI, but few studies have considered its relationship with improvements in walking. BWS step training induced a reduction in SOL clonus duration and an increase in threshold angle for triggering clonus when measured in sitting (Manella and Field-Fote 2013) and lying supine (Adams and Hicks 2011), while Lokomat training induced a deeper modulation of the H-reflex throughout the walking cycle (Knikou 2013) and greater rate-dependent depression (Knikou and Mummidisetty 2014). Lokomat training, however, did not change the overground walking function as measured by the 6MWT and the timed-up-and-go test (Knikou 2013), so presumably there was no appreciable relationship between walking function and change in reflex excitability (relationship not reported in (Knikou 2013)). Only one study reported a significant relationship

between reduced clonus and improved walking speed with training (Manella and Field-Fote 2013).

Cutaneous and cutaneomuscular reflexes also change with walking training. The CMR in the TA muscle in response to posterior tibial nerve stimulation showed enhanced early excitation and reduced late excitation with 4 weeks of Lokomat training (Hubli et al. 2012). Improvements were seen in the 10MWT, but the relationship between the electrophysiological and functional changes was not reported. Flexor reflex in the TA in response to sural nerve stimulation was also reduced with Lokomat training, but changes in overground walking ability were not reported (Smith et al. 2014).

We show here that the amount of clonus in the SOL during walking is weakly related to how well the person walks (i.e., 10MWT and 6MWT) before training. Furthermore, reduction in clonus is correlated with improvement in walking outcomes (Figure 2.5(b)(ii)). Similarly, the depth of inhibition in the CMR is related to walking speed and distance before Endurance Training, and the increase in CMR inhibition as a result of Endurance Training is weakly related to the improvement in walking speed (Figure 2.5(a)(ii)). The ability to walk is multifactorial and dependent on factors such as the strength of descending input from the brain, strength and endurance of muscles, ability to balance, and excitability of spinal reflexes. Thus, the correlation between any factor and walking ability may be low. Nevertheless, we believe it is essential to understand which aspects of neural plasticity might underlie functional improvements. Indeed, it is possible that some neural plasticity is unrelated to walking function. Determining the correspondence between change in walking function and change in training-induced neural plasticity is the first step towards improving this understanding.

2.5 Conclusions

Our data, along with other converging evidence, suggests that intensive training of walking reduces the abnormal reflex excitability seen after SCI, for example, reduced excitability of the H-reflex (Knikou 2013; Knikou and Mummidisetty 2014), stretch reflex (Mirbagheri et al. 2015; Mirbagheri et al. 2002), and clonus (Adams and Hicks 2011; Manella and Field-Fote 2013) at the ankle, and the flexor reflex (Smith et al. 2014) and long-latency cutaneomuscular reflex (Hubli et al. 2012) in the lower leg. This enhanced inhibition is concurrent with training-induced

strengthening of corticospinal input to the motoneurons/interneurons (Knikou 2012; Thomas and Gorassini 2005; Zewdie et al. 2015) and better volitional control of leg muscles (Mirbagheri et al. 2005; Zewdie et al. 2015). Descending control of spinal circuits, especially from the CST, is well described for humans (Iles and Pisini 1992; Rothwell et al. 1984; Roy et al. 2014; Valls-Sole et al. 1994) including during voluntary movements (Fournier et al. 1983; Iglesias et al. 2008). It is likely that walking training strengthens the descending control, including inhibitory control of spinal circuits, which have been weakened by the injury (Zewdie et al. 2015).

2.6 Bibliography for Chapter 2

Aagaard P, Simonsen EB, Andersen JL, Magnusson P, and Dyhre-Poulsen P. Neural adaptation to resistance training: changes in evoked V-wave and H-reflex responses. *Journal of applied physiology (Bethesda, Md : 1985)* 92: 2309-2318, 2002.

Adams MM, and Hicks AL. Comparison of the effects of body-weight-supported treadmill training and tilt-table standing on spasticity in individuals with chronic spinal cord injury. *The journal of spinal cord medicine* 34: 488-494, 2011.

Behrens M, Mau-Moeller A, Wassermann F, Bader R, and Bruhn S. Effect of balance training on neuromuscular function at rest and during isometric maximum voluntary contraction. *Eur J Appl Physiol* 115: 1075-1085, 2015.

Beres-Jones JA, Johnson TD, and Harkema SJ. Clonus after human spinal cord injury cannot be attributed solely to recurrent muscle-tendon stretch. *Experimental brain research* 149: 222-236, 2003.

Capaday C, and Stein RB. Amplitude modulation of the soleus H-reflex in the human during walking and standing. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 6: 1308-1313, 1986.

Casabona A, Polizzi MC, and Perciavalle V. Differences in H-reflex between athletes trained for explosive contractions and non-trained subjects. *European journal of applied physiology and occupational physiology* 61: 26-32, 1990.

Dietz V. Spastic movement disorder: what is the impact of research on clinical practice? *Journal of neurology, neurosurgery, and psychiatry* 74: 820-821, 2003.

Dietz V, and Sinkjaer T. Spastic movement disorder: impaired reflex function and altered muscle mechanics. *The Lancet Neurology* 6: 725-733, 2007.

Fournier E, Katz R, and Pierrot-Deseilligny E. Descending control of reflex pathways in the production of voluntary isolated movements in man. *Brain Res* 288: 375-377, 1983.

Goode DJ, and Van Hoven J. Loss of patellar and Achilles tendon reflexes in classical ballet dancers. *Arch Neurol* 39: 323, 1982.

Gorassini MA, Knash ME, Harvey PJ, Bennett DJ, and Yang JF. Role of motoneurons in the generation of muscle spasms after spinal cord injury. *Brain : a journal of neurology* 127: 2247-2258, 2004.

Gorassini MA, Norton JA, Nevett-Duchcherer J, Roy FD, and Yang JF. Changes in locomotor muscle activity after treadmill training in subjects with incomplete spinal cord injury. *Journal of neurophysiology* 101: 969-979, 2009.

Gruber M, Taube W, Gollhofer A, Beck S, Amtage F, and Schubert M. Training-specific adaptations of H- and stretch reflexes in human soleus muscle. *J Mot Behav* 39: 68-78, 2007.

Guyatt GH, Sullivan MJ, Thompson PJ, Fallen EL, Pugsley SO, Taylor DW, and Berman LB. The 6-minute walk: a new measure of exercise capacity in patients with chronic heart failure. *Can Med Assoc J* 132: 919-923, 1985.

Hubli M, Dietz V, and Bolliger M. Spinal reflex activity: a marker for neuronal functionality after spinal cord injury. *Neurorehabilitation and neural repair* 26: 188-196, 2012.

Iglesias C, Nielsen JB, and Marchand-Pauvert V. Corticospinal inhibition of transmission in propriospinal-like neurones during human walking. *The European journal of neuroscience* 28: 1351-1361, 2008.

Iles J, and Pisini J. Cortical modulation of transmission in spinal reflex pathways of man. *The Journal of physiology* 455: 425-446, 1992.

Jones CA, and Yang JF. Reflex behavior during walking in incomplete spinal-cord-injured subjects. *Experimental neurology* 128: 239-248, 1994.

Keller M, Pfusterschmied J, Buchecker M, Muller E, and Taube W. Improved postural control after slackline training is accompanied by reduced H-reflexes. *Scand J Med Sci Sports* 22: 471-477, 2012.

Khan A, Patrick S, Roy FD, Gorassini M, and Yang J. Changes in spasticity-related reflexes after two forms of walking retraining in individuals with incomplete spinal cord injury. In: *Society for Neuroscience 2014 Annual Meeting*. Washington, DC: 2014.

Knikou M. Functional reorganization of soleus H-reflex modulation during stepping after robotic-assisted step training in people with complete and incomplete spinal cord injury. *Experimental brain research* 228: 279-296, 2013.

Knikou M. Plasticity of corticospinal neural control after locomotor training in human spinal cord injury. *Neural plasticity* 2012: 2012.

Knikou M, and Mummidisetty CK. Locomotor training improves premotoneuronal control after chronic spinal cord injury. *Journal of neurophysiology* 111: 2264-2275, 2014.

Koceja DM, Davison E, and Robertson CT. Neuromuscular characteristics of endurance- and power-trained athletes. *Research quarterly for exercise and sport* 75: 23-30, 2004.

Lagerquist O, Zehr EP, and Docherty D. Increased spinal reflex excitability is not associated with neural plasticity underlying the cross-education effect. *Journal of applied physiology (Bethesda, Md : 1985)* 100: 83-90, 2006.

Mahoney JS, Engebretson JC, Cook KF, Hart KA, Robinson-Whelen S, and Sherwood AM. Spasticity Experience Domains in Persons With Spinal Cord Injury. *Archives of physical medicine and rehabilitation* 88: 287-294, 2007.

Manella KJ, and Field-Fote EC. Modulatory effects of locomotor training on extensor spasticity in individuals with motor-incomplete spinal cord injury. *Restorative neurology and neuroscience* 31: 633-646, 2013.

Mirbagheri M, Tsao C, Pelosin E, and Rymer W. Therapeutic effects of robotic-assisted locomotor training on neuromuscular properties. In: *Rehabilitation Robotics, 2005 ICORR 2005 9th International Conference on*IEEE, 2005, p. 561-564.

Mirbagheri MM, Kindig MW, and Niu X. Effects of robotic-locomotor training on stretch reflex function and muscular properties in individuals with spinal cord injury. *Clin Neurophysiol* 126: 997-1006, 2015.

Mirbagheri MM, Ladouceur M, Barbeau H, and Kearney RE. The effects of long-term FES-assisted walking on intrinsic and reflex dynamic stiffness in spastic spinal-cord-injured subjects. *Neural Systems and Rehabilitation Engineering, IEEE Transactions on* 10: 280-289, 2002.

Musselman K, Brunton K, Lam T, and Yang J. Spinal cord injury functional ambulation profile: a new measure of walking ability. *Neurorehabilitation and neural repair* 25: 285-293, 2011.

Musselman KE, and Yang JF. Spinal Cord Injury Functional Ambulation Profile: a preliminary look at responsiveness. *Physical therapy* 94: 240-250, 2014.

Nielsen J, Crone C, and Hultborn H. H-reflexes are smaller in dancers from The Royal Danish Ballet than in well-trained athletes. *European journal of applied physiology and occupational physiology* 66: 116-121, 1993a.

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

Reyes R, Icarangal A, and Bertellotti G. Part 1: The Good, The Bad, and The Not-So-Ugly: Medical and Pharmacological Treatments for Spasticity http://sci.washington.edu/spasticity/index.asp.

Roby-Brami A, and Bussel B. Long-latency spinal reflex in man after flexor reflex afferent stimulation. *Brain : a journal of neurology* 110 (Pt 3): 707-725, 1987.

Rochcongar P, Dassonville J, and Le Bars R. [Modification of the Hoffmann reflex in function of athletic training (author's transl)]. *European journal of applied physiology and occupational physiology* 40: 165-170, 1979.

Rossi A, Mazzocchio R, and Scarpini C. Clonus in man: a rhythmic oscillation maintained by a reflex mechanism. *Electroencephalography and clinical neurophysiology* 75: 56-63, 1990.

Rothwell JC, Day BL, Berardelli A, and Marsden CD. Effects of motor cortex stimulation on spinal interneurones in intact man. *Experimental brain research* 54: 382-384, 1984.

Roy FD, Bosgra D, and Stein RB. Interaction of transcutaneous spinal stimulation and transcranial magnetic stimulation in human leg muscles. *Experimental brain research* 232: 1717-1728, 2014.

Schneider C, and Capaday C. Progressive adaptation of the soleus H-reflex with daily training at walking backward. *Journal of neurophysiology* 89: 648-656, 2003.

Smith AC, Mummidisetty CK, Rymer WZ, and Knikou M. Locomotor training alters the behavior of flexor reflexes during walking in human spinal cord injury. *Journal of neurophysiology* 112: 2164-2175, 2014.

Taylor S, Ashby P, and Verrier M. Neurophysiological changes following traumatic spinal lesions in man. *Journal of neurology, neurosurgery, and psychiatry* 47: 1102-1108, 1984.

Thomas SL, and Gorassini MA. Increases in corticospinal tract function by treadmill training after incomplete spinal cord injury. *Journal of neurophysiology* 94: 2844-2855, 2005.

Valls-Sole J, Alvarez R, and Tolosa ES. Vibration-induced presynaptic inhibition of the soleus H reflex is temporarily reduced by cortical magnetic stimulation in human subjects. *Neuroscience letters* 170: 149-152, 1994.

van Hedel HJ, Wirz M, and Dietz V. Assessing walking ability in subjects with spinal cord injury: validity and reliability of 3 walking tests. *Archives of physical medicine and rehabilitation* 86: 190-196, 2005.

Wallace DM, Ross BH, and Thomas CK. Characteristics of lower extremity clonus after human cervical spinal cord injury. *Journal of neurotrauma* 29: 915-924, 2012.

Walsh EG. Clonus: beats provoked by the application of a rhythmic force. *Journal of neurology, neurosurgery, and psychiatry* 39: 266-274, 1976.

Yang JF, Musselman KE, Livingstone D, Brunton K, Hendricks G, Hill D, and Gorassini M. Repetitive mass practice or focused precise practice for retraining walking after incomplete

spinal cord injury? A pilot randomized clinical trial. *Neurorehabilitation and neural repair* 28: 314-324, 2014.

Yang JF, and Stein RB. Phase-dependent reflex reversal in human leg muscles during walking. *Journal of neurophysiology* 63: 1109-1117, 1990.

Zehr EP. Training-induced adaptive plasticity in human somatosensory reflex pathways. *Journal of applied physiology (Bethesda, Md : 1985)* 101: 1783-1794, 2006.

Zewdie ET, Roy FD, Yang JF, and Gorassini MA. Facilitation of descending excitatory and spinal inhibitory networks from training of endurance and precision walking in participants with incomplete spinal cord injury. *Prog Brain Res* 218: 127-155, 2015.

CHAPTER 3: TRAINING TO WALK IN THE REWALK EXOSKELETON AFTER INCOMPLETE AND COMPLETE SPINAL CORD INJURY

3.1 Introduction

Restoring walking is of high priority for individuals with thoracic spinal cord injury (SCI) (Anderson 2004b; Brown-Triolo et al. 2002; Simpson et al. 2012). Treadmill and over ground training improve functional walking for those with residual strength in the lower extremity, but not for those with severe injuries (Dobkin et al. 2006; van Middendorp et al. 2011; Waters et al. 1994; Yang et al. 2011). Assistive devices such as reciprocating gait orthoses (Lotta et al. 1994) or functional electrical stimulation (Hesse et al. 2004) can restore standing and walking, but the energy expenditure to use them is high (Bernardi et al. 1995), making them unfeasible for daily use (Coghlan et al. 1980; Mikelberg and Reid 1981; Sykes et al. 1995). The recent emergence of powered exoskeletons (Lajeunesse et al. 2015) could change this. Three such devices have been approved in North America. The ReWalk [ReWalk Robotics, Inc., Marlborough, MA (http://rewalk.com/)] and the Indego [Parker Hannifin Co., Macedonia, OH (http://www.indego.com/indego/en/home)] are approved for rehabilitation and home use, while the Ekso [Ekso Bionics, Richmond, CA (http://eksobionics.com/)] is approved for rehabilitation only. Early findings indicated individuals with severe SCI and preserved arm strength can train in these devices with varying success. Walking speeds achieved after training range from 0.03 – 0.71 m/s in the ReWalk (Benson et al. 2016; Esquenazi et al. 2012; Spungen et al. 2013c; Talaty et al. 2013; Yang et al. 2015; Zeilig et al. 2012), 0.11 – 0.33 m/s in the Ekso (Kozlowski et al. 2015; Kressler et al. 2014), and 0.063 – 0.45 m/s in the Indego (Farris et al. 2014; Hartigan et al. 2015; Kressler et al. 2014). While these results are promising, some suggest the devices may not meet the high expectations of the users (Benson et al. 2015), and few have reported training-induced changes in the nervous system (i.e., neuroplasticity) (Kressler et al. 2014).

Here, we report initial findings from a training program with the ReWalk, focusing on the functional gains and neuroplasticity induced by the training. Also included are results from preliminary testing of the device for use in the home and community. Portions of this data have been published in abstract form (Khan et al. 2015).

3.2 Methods

3.2.1 Participants

Potential participants were screened by phone, then in-person. *Inclusion criteria:* chronic (≥1 yr), non-progressive SCI (AIS A, B, C), body weight <90 kg, thigh and lower leg length appropriate for the ReWalk, able to use forearm crutches and train for 4 days/week, approval for participation from primary care physician, and walking speed ≤0.4 m/s. *Exclusion criteria:* comorbidities that interfere with training or measurements, bone fractures within the last 2 years, low bone density (femoral neck t-score < -3), hip and knee contractures >10° flexion, ankle plantarflexion contractures, active pressure ulcers and severe spasticity. Uninjured (i.e., control) participants were also recruited for comparison of a few measures with the SCI participants.

The study was approved by the Health Research Ethics Board at the University of Alberta (Pro00036789). Written consent was obtained from all participants.

3.2.2 Experimental design

A prospective cohort study with a single, 12-week intervention was used. Measurements were taken at least twice at baseline, then at 6 and 12 weeks of training, and 2-3 months after the end of training.

3.2.3 Intervention

3.2.3.1 Standing in the standing frame

Participants who had not been standing weekly prior to training practiced standing in a standing frame (EasyStand Evolv, Morton, MN) daily for 2 weeks before ReWalk training, until one hour of standing in two 30-min bouts was tolerable.

3.2.3.2 The ReWalk exoskeleton

The ReWalk 2.0 was used with updated footplates (unilateral calf-holder) from the ReWalk 5.0 after skin abrasions on the foot were experienced by the first participant (P1). Only one participant (P2) continued to use 2.0 footplates (bilateral calf-holder) because the smallest 5.0 footplates were too large to fit inside her shoes. The ReWalk was upgraded to 5.0 towards the

end of training in P7 and P8. Velcro straps secured the torso, pelvis, and legs to the device, with footplates inside the shoes. The Velcro straps below the knee were replaced by knee brackets for P9's follow-up measures, and onwards from P10's 8th session of standing balance in the ReWalk (see *Section 3.2.3.3 Standing balance in the ReWalk*). Skin integrity at the contact points with the device and under the straps was checked before and after each training session. Padding was used as needed (Alpha Classic Gel Liner, WillowWood, Mt Sterling, OH, normally used to line amputee sockets).

The mode of operation (i.e., stand, walking, stair climbing) was initiated by the trainer/spotter with a wrist worn communicator that signaled the onboard computer. Once participants became proficient walkers, they learned to control the communicator. Motors at the hips and knees generated the steps, and a variety of adjustable parameters including swing time (0.6 - 1.5 sec), delay between steps (1 - 60 sec), and flexion angles at the hip (10 - 25°) and knee (17 - 47°) were set by a computer program. Forearm crutches were used for balance.

3.2.3.3 Standing balance in the ReWalk

Participants learned sit/stand transitions in the ReWalk, then balancing in standing. Balance tasks included, but were not limited to, lifting one crutch at a time, both crutches simultaneously, and preventing falls with the crutches. Walking began once participants could maintain balance while lifting a crutch for >30 sec.

3.2.3.4 Walking in the ReWalk

Participants started walking indoors on smooth flooring. Each step was initiated by a slight forward lean of the body (sensed by a tilt sensor), and an immediate return to upright to allow for foot clearance. Walking speed was increased by modifying the parameters of the ReWalk (see above), and by the participant initiating consecutive steps more quickly. Participants aimed to increase the walking speed and distance, and the number of uninterrupted steps in a sequence. They practiced turning while walking by pivoting on one foot during the stance phase. Stopping was practiced by allowing the foot to hit the floor during swing phase, which triggered a current surge to stop the device. When deemed safe by the therapist, they trained to walk on carpet,

ramps, asphalt, and grass, and attempted stairs and curbs. The trainer maintained contact-guard of the pelvic band from behind, with a spotter in front.

3.2.4 Measures

3.2.4.1 Progression of training

The total step count, walking distance, speed, and duration were documented during each training session. The number of consecutive steps was counted manually for each sequence of walking, and used as a measure of skill, because novice walkers often stalled the device inadvertently with inadequate toe clearance.

3.2.4.2 Walking related outcomes

Walking speed in 10 m was recorded during continuous walking (i.e., modified 10-Meter Walk Test [10MWT]), because starting and stopping the device added variability to the measure. The 6-Minute Walk Test (6MWT) was estimated by walking back and forth in a 40 m hallway. The Physiological Cost Index (PCI) was estimated during the 6MWT. The 10MWT and 6MWT have been validated for individuals with SCI (van Hedel et al. 2005). The effort of walking was estimated by the PCI (Leung et al. 2009) as:

$$\text{PCI (heart beats/m)} = \frac{\text{(Active HR - Resting HR)}}{\text{Average walking speed}}$$

Heart rate (HR: beats/min) was recorded every 30 sec with a HR monitor (Polar Bluetooth). Resting HR and Active HR were the average HR during the last 2 min of sitting, and the last 2 min of the 6MWT, respectively. Average walking speed (m/min) was from the 6MWT. If a participant had to rest before 6 min, the PCI was estimated from the distance walked. PCI was also measured for wheelchair propulsion, and walking without the ReWalk in participants who could do so. Measures from 4 uninjured participants (without the ReWalk) were obtained for comparison. Finally, the maximum walking distance without a rest, for up to 1 hour, was measured indoors on a smooth floor towards the end of training.

3.2.4.3 Balance

Sitting balance was measured on a force platform (Model OR6-7-1000, AMTI, Watertown, MA), with feet unsupported and hands crossed over the chest. Visual feedback of the center of pressure was provided, with 8 equally spaced targets in a circle (Figure 3.4A). Participants leaned as far as possible towards each target in a randomized order.

Postural sway with eyes closed was measured for a maximum of 30 sec of sitting (Harel et al. 2013; Lemay et al. 2013), or until balance was lost. Measures of balance were repeated 2-4 times on different days before training until a stable baseline was achieved. When possible, limits of stability and sway were also measured in standing with arms at the side and the position of the feet standardized. Seven uninjured participants provided data for comparison.

P10 was unable to sit with arms crossed at the level of the chest, and could sit unsupported for only a few seconds before losing balance. Therefore, for P10 only, sway was measured with arms on the side and hands resting on the abdomen at the level of the umbilicus. The duration of sway was measured during 3 bouts of unsupported sitting.

3.2.4.4 Strength of sensory pathways

Skin sensation was measured by surface electrical stimulation (Digitimer DS7A, Hertfordshire, England) of the C3-S2 sensory key points (Savic et al. 2006) from the International Standards for Neurological Classification of SCI (Maynard et al. 1997) (ISNCSCI). Stimulation frequency was 2-3 Hz, pulse width 0.5 ms, to a maximum current of 40 mA. Sensory threshold was the lowest current at which a tapping sensation was reported from 2 trials (Savic et al. 2006). Three uninjured participants provided data for comparison.

3.2.4.5 Pain

Daily rating of pain immediately before and after a training session was recorded using a numerical rating scale between zero (no pain) and 10 (worst pain imaginable) (Siddall et al. 2006). Pain over a week was estimated with the McGill Pain Questionnaire (MPQ) (Melzack 1975), filled out prior to the training on the test day.

3.2.4.6 Manual muscle strength test

The lower and upper extremity muscle strength (LEMS and UEMS) were estimated with the scale from the ISNCSCI (Maynard et al. 1997) by a physical therapist, before and after the 12-week training.

3.2.4.7 Spinal cord assessment tool for spasticity

Clonus, flexor and extensor spasms were measured bilaterally, by a physical therapist not involved in the training, using the Spinal Cord Assessment Tool for Spasticity (SCATS) (Benz et al. 2005). Two trials of each measure were performed on the same day. For P1 and P2 only, the SCATS was measured on two different days prior to the 12-week training, and once at mid-training and immediately after the training. Since we found the measures varied day-to-day in P2, all subsequent participants were measured for the SCATS weekly throughout training.

3.2.4.8 Affective measures

Effects of training in the ReWalk on depression were determined by using the total score (maximum = 27) on the Patient Health Questionnaire-9 (PHQ-9) (Wittkampf et al. 2009), filled out biweekly, prior to training on the test day. The average PHQ-9 score across multiple baseline measures was compared with the score immediately after the training.

3.2.4.9 General methods: strength of motor pathways and the cutaneomuscular reflex

All surface electromyogram (EMG) signals were measured with disposable, bipolar Ag/AgCl electrodes (Kendall H59P, Mansfield, MA), placed 2 cm apart center-to-center. Signals were amplified and filtered (10-1000 Hz) with an AMT-8 EMG system (Bortec Biomedical Ltd, Calgary, AB, Canada), digitized (5 kHz) and recorded with Axoscope (DigiData 1440A series, Molecular Devices, Sunnyvale, CA).

3.2.4.10 Strength of motor pathways

The strength of the corticospinal tracts to the trunk extensor muscles was measured in sitting with single-pulse transcranial magnetic stimulation (Magstim 200, Whitland, UK), applied with a double-cone coil positioned at the vertex. Surface EMG of back muscles were measured

bilaterally, with electrodes placed 1 & 3 cm lateral to the midline across 8 vertebral levels spanning the injury (similar to Cariga et al. (2002)). Bony landmarks were identified by a physical therapist. To ensure repeatable placements day-to-day, a photograph of the electrodes on the back was taken on the first recording day and used as reference for future recordings. Stimulus intensity was incremented in steps of 2-3% of maximum stimulator output (MSO), ranging from below motor threshold to 80% of MSO (or maximum tolerable if below 80% MSO). Five responses, time locked to the stimulus, were averaged per stimulus intensity. In all participants except for P1, P2 and P3, MEP amplitudes were also compared at different levels of background EMG by having participants hold various contraction levels during application of stimuli, such as leaning forwards, seated push-up, lifting arms, trunk extension against resistance, while pulses were induced at the maximum tolerable stimulus intensity.

3.2.4.11 Cutaneomuscular reflex

The cutaneomuscular reflex (CMR) was measured from the tibialis anterior (TA) during quiet sitting, evoked with posterior tibial nerve stimulation at the ankle (5 pulses, 300 Hz, 0.5 ms pulse width; Digitimer DS7A, Hertfordshire, UK). This mixed nerve was chosen to confirm the consistency of the motor threshold and the maximum M-wave of the abductor hallucis brevis (AHB) between testing days (Jones and Yang 1994; Yang and Stein 1990). EMG was recorded from the TA and AHB, bilaterally. Stimuli were delivered every 3 - 5 sec to allow any triggered spasms to subside. Stimulus intensity ranged from below motor threshold of the AHB to 100 mA, in 10 mA increments, to determine the reflex threshold and magnitude (see *Section 3.2.5 Data analyses*). Five stimuli were applied at each intensity.

3.2.4.12 Field tests

The training physical therapist carried out the following tests when deemed safe. Indoor tests included walking on ramps, turning and stopping in narrow spaces, opening and closing automatic and manual doors, and using an elevator. Kitchen tasks performed in a simulated kitchen included using the stove and sink, retrieving items from low and overhead cabinets, and the refrigerator. Outdoor tests included walking on uneven surfaces (i.e., concrete, grass, and asphalt).

3.2.4.13 Follow-up measures

Follow-up measures were taken at 2-3 months after the end of training for the 10MWT, 6MWT and PCI with the ReWalk and a standard walker, limits of stability and postural sway, MPQ, SCATS, PHQ-9 and SPT. Follow-up measures of the UEMS and LEMS were taken from P3, who showed changes in muscle strength after the training (see *Section 3.3 Results*).

3.2.5 Data analyses

Data from walking and clinical scores were processed in Microsoft Excel (Microsoft, Redmond, WA). Data for limits of stability and postural sway, CMR and MEP measures were analyzed using custom written codes in the LabVIEW (National Instruments Corp., TX) and MATLAB (The Mathworks, Natick, MA) programming environments.

3.2.5.1 Balance

Data from balance tests, including 3-D forces and moments from the force platform were digitized, low-pass filtered at 10 Hz (dual-pass, 2nd order, Butterworth filter), and used to calculate the instantaneous position of the center of pressure. Limits of stability was defined as the area enclosed by the position of the center of pressure at the 8 maximum leans (Figure 3.4C), expressed as a percentage of the base of support (BoS; shaded areas in Figure 3.4B):

$$\text{Limits of stability (\%BoS)} = \frac{\text{Area of limits of stability}}{\text{Area of base of support}} \times 100\%$$

Postural sway was quantified as the speed of the center of pressure excursion (total distance/duration of sway (Harel et al. 2013; Lemay et al. 2013) (Figure 3.4D), and the average sway distance (i.e., length of the polar vector for every data point) from the centroid of sway. The last baseline measure of balance was used for comparison with other time points, because the scores had stabilized by then. For P10, the duration of sway across the 3 bouts of unsupported sitting was averaged.

3.2.5.2 Manual muscle strength test

The scores on the UEMS and LEMS were summed across both sides (maximum = 50), and analyzed separately for the upper and lower extremities.

3.2.5.3 Spinal cord assessment tool for spasticity

For each item, an average score across the two trials was calculated, and then summed across the items and the legs to generate a total score for that day (maximum = 18).

3.2.5.4 Strength of motor pathways

MEP threshold was defined as the stimulus intensity eliciting an MEP at 50% probability. MEP latency was measured from the stimulus artifact to the first deflection of the MEP waveform. Threshold and latency were measured based on visual inspection by an un-blinded researcher. MEP_{max} at rest was the average peak-to-peak amplitude of the highest MEP response, typically at 80% MSO. Background EMG was defined as the average rectified EMG from 2 to 50 ms before the stimulus. The relationship between background EMG and MEP amplitude for each session were fit with a least squares straight line for each vertebral level, described by the slope and y-intercept. For each measurement session in a participant, the MEP measures (threshold, latency, MEP_{max}, and the slope and y-intercept of the least squares line) were first averaged across all spinal segments for those with incomplete injuries (P2, P3, P10, P11), and only at and above the level of injury for participants with complete injuries. This is because plasticity at levels below the injury is unlikely when the injury is complete.

3.2.5.5 Cutaneomuscular reflex

The CMR threshold was defined as the stimulus intensity eliciting a response with a 50% probability, based on visual inspection of the sweeps of the raw TA EMG. Reflex EMG at each stimulus intensity was full-wave rectified, low-pass filtered at 200 Hz, time-locked to the stimulus and averaged across 5 trials per stimulus intensity, to generate the response time series. The background EMG (2 to 90 ms before the first stimulus) was removed from these time series. The magnitude of the reflex was defined as the area under this EMG curve after background EMG removal, quantified from the start and end times of the response window. The response window was determined by visual inspection.

3.2.6 Statistics

All statistical analyses were performed using OriginPro 2018 Student Version (OriginLab Corporation, Northampton, MA). Pairwise comparisons between measures before and after training, and between measures after training and at follow up were performed using the Paired t-test or Wilcoxon Signed Rank Test, for data that were normally or not normally distributed, respectively. Pairwise comparisons before and after training include: limits of stability and postural sway (speed and distance), score on the Daily Rating of Pain immediately before and after each training session, total score on the SCATS and PHQ-9, threshold and magnitude of the CMR, and MEP measures (latency, threshold, MEP_{max}, background EMG at threshold and MEP_{max}, slope and y-intercept). Pairwise comparisons between measures at follow up and immediately after training include: 10MWT, 6MWT and PCI in the ReWalk, limits of stability and postural sway, and total score on the MPQ, SCATS and PHQ-9. To determine the effects of breaks in training (i.e., due to health-related issues or holidays), the average distance and maximum number of sequential steps across 3 sessions immediately before and after the break (except for the first session after the break, because the training duration was reduced intentionally) were also compared. Significance was set at $p<0.05$. All values are reported as mean ± 1 standard deviation.

3.3 Results

3.3.1 Participants

Forty-nine potential participants with SCI were screened (Figure 3.1) and 38 were excluded, with the most common reasons for exclusion being: declines (n=11), over weight limit (n=6), contractures in the lower extremities (n=5) and pressure sores (n=5). Eleven participants were enrolled (Figure 3.1, Table 3.1). One (P6) dropped out after two sessions of standing in the ReWalk (reason: ‘not for him’). Eight participants completed 12 weeks of training, and one (P5) completed 6 weeks of training because a car accident aggravated her elbow epicondylitis at Week 6; available data from P5 were included. Seven control participants were also recruited in order to compare the measures of balance, sensory perceptual threshold and physiological cost index (Table 3.2) with the SCI participants. Details on each of the measures are provided in the subsequent sections.

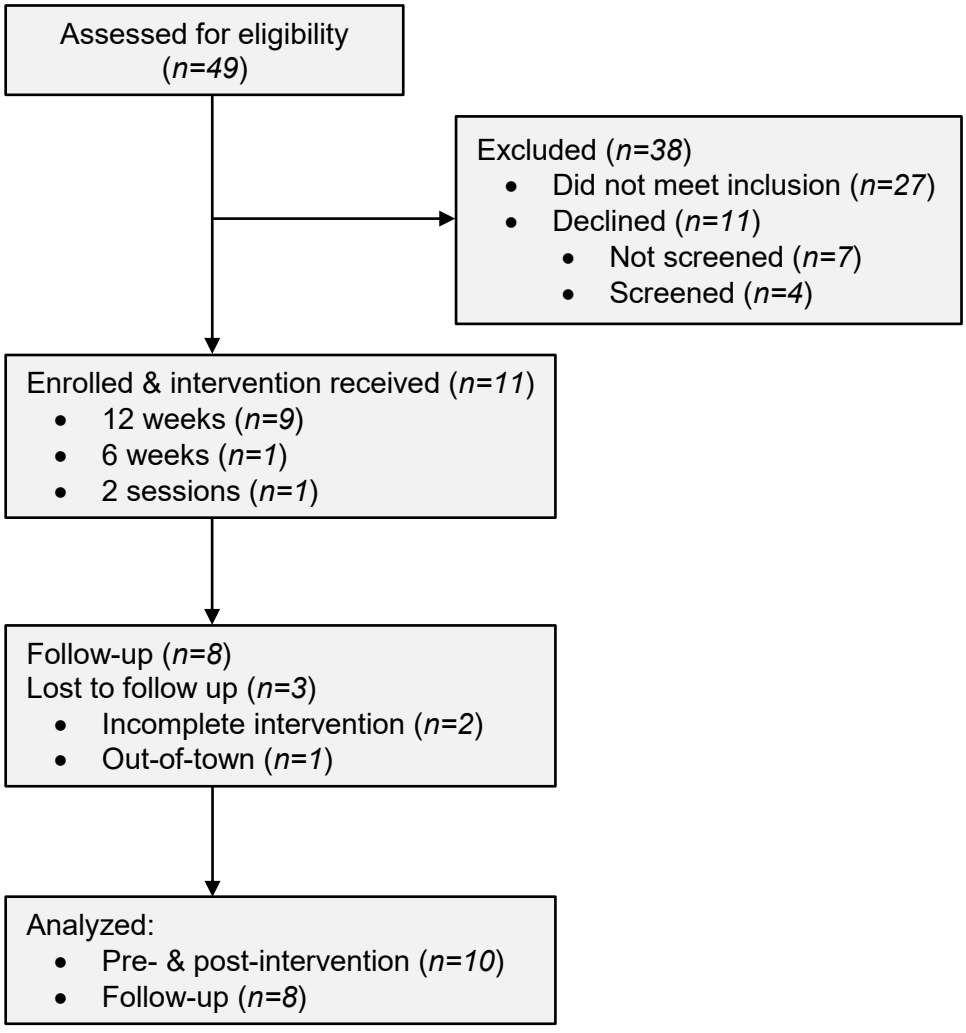

Figure 3.1: CONSORT diagram.

Flow chart summarizing the participant recruitment and participation. See text for reasons of ineligibility and drop-out.

Table 3.1: Spinal cord injury participant characteristics

Participant code-gender	Age (yr)	Neurological level of injury	AIS	Cause of injury	Time since injury (yr)	Anti-spastic medication
P1-M	35	T4	B	MVA	5	Baclofen
P2-F	25	C7	C	MVA	6	Baclofen, Tizanidine, Oxybutynin
P3-F	29	C6	C	MVA	3	Baclofen, Oxybutynin
P4-M	63	T6	A	Work	24	Baclofen, Diazepam, Oxybutynin
P5-F	46	T4	A	Sports	16	Oxybutynin
P6-M	29	T5	A	MVA	1.2	Oxybutynin
P7-M	19	T7	A	MVA	2.9	None
P8-M	46	T10	A	Work	1.4	Oxybutynin
P9-F	30	T10	A♦	MVA	2.0	Baclofen
P10-M	35	T4 ▲	C ▲	MVA	4.5	Baclofen
P11-M	56.2	C6	C or D‡	MVA	18.7	Baclofen
Mean* (SD)	**38.4 (14.0)**				**8.4 (8.1)**	

M: Male; F: Female; AIS: ASIA Impairment Scale; T: Thoracic; C: Cervical; MVA: Motor vehicle accident; SD: Standard deviation. *Mean and SD exclude P6, who dropped out after two sessions of training. ♦ ASIA exam not done, but most likely AIS grade A injury. ▲ Before syrinx. ‡Based on information provided, not on a full assessment.

Table 3.2: Control participant characteristics

Experiment	N	Age range	Mean age (SD)
Limits of stability in sitting	7	23-62	34.6 (15.7)
Limits of stability in standing	7	21-62	34.3 (15.9)
Sway during sitting	7	23-62	34.6 (15.7)
Sway during standing	4	21-28	24.8 (2.9)
Sensory perceptual threshold	3	25-60	37.7 (19.4)
Physiological cost index	4	25-60	40.5 (17.3)

N: sample size.

3.3.2 Progression of training

Three participants (P1, P4 and P7) required the standing frame training. The progress in walking training, shown in Figure 3.2, is a running average of 5 sessions starting from Session #3. Most completed >40 sessions with an average frequency of 3.63 ± 0.44 sessions/week. By the end of training, all participants walked for about 1 hour, achieving >1500 steps (Figure 3.2A), a distance of about 1 km (Figure 3.2B), at speeds of about 0.3 to 0.6 m/s (Figure 3.2C), with maximum bouts of 500-2000 uninterrupted steps (Figure 3.2D). All participants were able to walk indoors on smooth floors, make turns as needed, and occasionally walk outdoors (except P4 because of unfavorable winter weather). None found the adjustment to outdoor conditions difficult. Nine participants (P1, P2, P3, P5, P7, P8, P9, P10, P11) learned to walk on carpet, and seven (P1, P2, P3, P7, P8, P9, P10) learned to use the wrist worn ReWalk controller. All participants required assistance with donning the device, while some also required assistance with doffing.

3.3.3 Walking related outcomes

Walking speed over 10 m (Figure 3.3A), distance in the 6MWT (Figure 3.3B), and maximum walking distance in an hour (Figure 3.3C) at the end of training are shown in Figure 3.3, with mean ± SD being: 0.41 ± 0.12 m/s, 144.20 ± 37.76 m, 1338.44 ± 648.31 m, respectively. Since P3 became an independent walker after training, and P2 was an independent walker prior to training, their 10MWT, 6MWT and PCI without the ReWalk are also shown. Both walked further with the ReWalk (Figure 3.3B) and at a lower effort (Figure 3.3D) compared to without the ReWalk. The PCI in the ReWalk averaged across participants was 1.54 ± 0.86 beats/m, compared to 0.48 ± 0.11 beats/m for wheelchair propulsion, and 0.60 ± 0.02 beats/m for over ground walking in the uninjured participants.

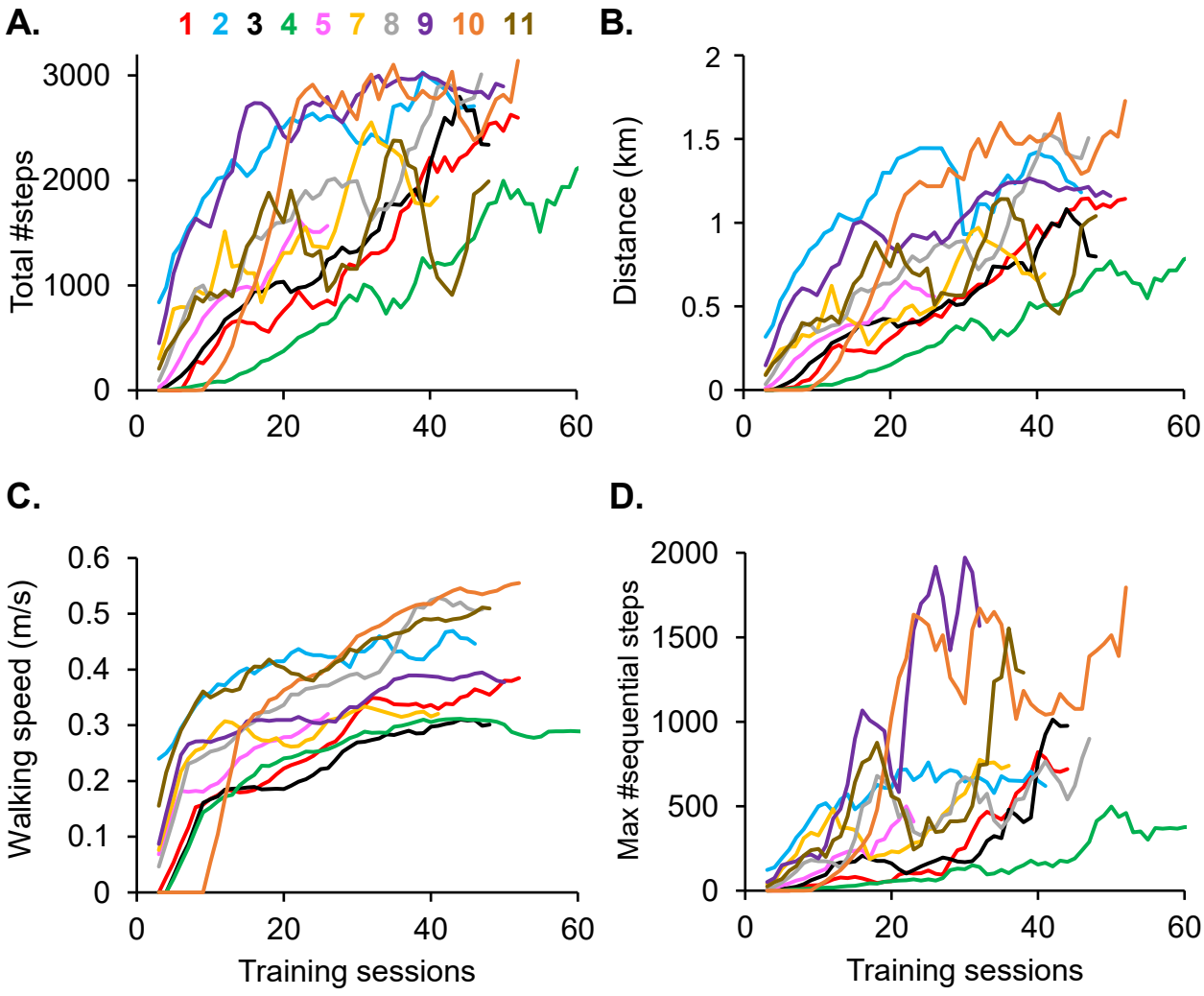

Figure 3.2: Training progression.

(a) Total number of steps taken, (b) total distance walked, (c) average walking speed, and (d) maximum number of sequential steps taken in a single walking bout, shown only until a plateau in performance is met (d). Lines represent sliding averages across 5 sessions for clarity, displayed starting at Session #3.

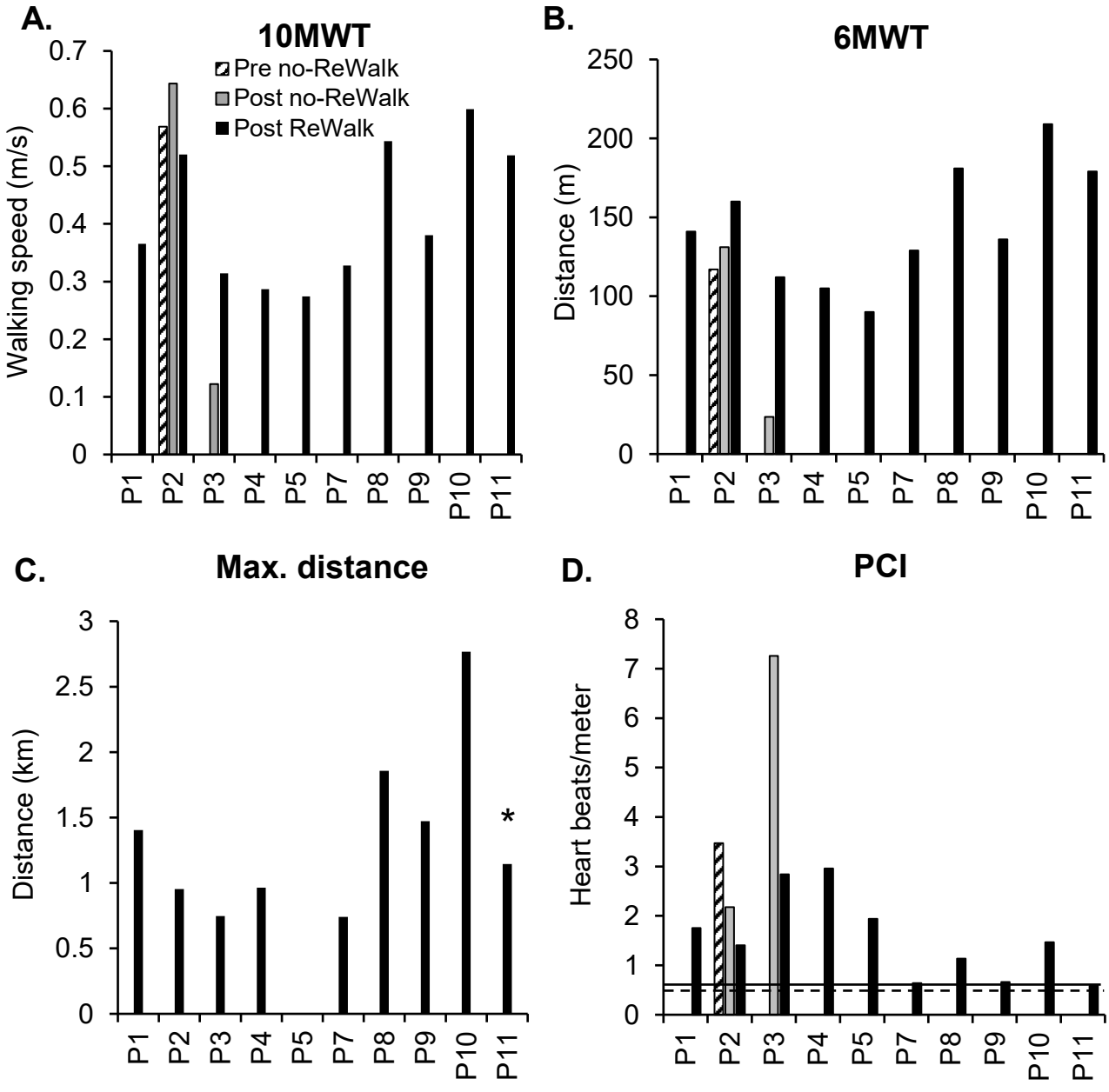
A.
10MWT
Pre no-ReWalk
Post no-ReWalk
Post ReWalk
Walking speed (m/s)
0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
P1
P2
P3
P4
P5
P7
P8
P9
P10
P11
B.
6MWT
Distance (m)
0
50
100
150
200
250
C.
Max. distance
Distance (km)
0
0.5
1
1.5
2
2.5
3
*
D.
PCI
Heart beats/meter
0
1
2
3
4
5
6
7
8

Figure 3.3: Walking ability at the end of training.

(a) Walking speed over 10 m (10MWT). (b) Distance walked during the 6-Minute Walk test (6MWT). (c) Maximum (Max.) distance walked in the ReWalk without a rest. (d) The effort of walking was measured by the Physiological Cost Index (PCI) while performing the 6MWT. In individuals who could walk without the ReWalk, the same measures while walking without the ReWalk (no-ReWalk) are shown for comparison before (Pre) and after (Post) training. Dashed and solid horizontal lines represent the PCI during wheelchair propulsion in SCI participants, and walking in uninjured participants (n=4), respectively. * ReWalk not working properly.

3.3.4 Balance

ReWalk training enlarged the limits of stability (LoS) (Figure 3.4C) and reduced the postural sway (Figure 3.4D). Since P2 and P11 were the highest functioning and their LoS during sitting did not change after training compared to before training (P3: 30.4 to 26.3 %BoS, P11: 7.2 to 11.1 %BoS), we show their LoS during standing only (blue traces in Figure 3.4C). Scores are shown for all SCI and uninjured participants in the line graphs (Figures 3C and D - bottom). LoS during sitting tended to increase after the training (6.85 ± 9.59 to 9.03 ± 10.71 %BoS, p=0.13), similar to the average sway speed (calculated from 21 sec of data – longest time all could sit without falling) which showed a trend for reduction (3.18 ± 2.18 to 2.00 ± 1.41 cm/s, p=0.061). In contrast, the average sway distance during sitting did not change after the training (0.37 ± 0.21 to 0.31 ± 0.34 cm, p=0.30). For P10, the average duration of sway across each of the 3 bouts did not change as a result of the training (1.37 ± 0.42 to 1.54 ± 1.29 sec). Finally, since P11 was the only participant who could stand for 30 seconds, measures of sway were also measured in standing, which did not change as a result of the training; sway speed: 3.14 to 3.60 cm/s (control: 1.89 ± 0.24 cm/s), sway distance: 1 to 1.13 cm (control: 0.5 ± 0.13 cm).

3.3.5 Strength of sensory pathways

Sensory perceptual threshold changed in four participants (P1, P4, P8, P10) below the level of the injury (Figure 3.5A). Average values from uninjured participants are shown for comparison (mean - black line, 2SD - dashed line). Details of changes are available in Table 3.3.

3.3.6 Pain

Daily rating of pain was reduced immediately after training across 5 participants (P1, P2, P3, P8, P10), and did not change in P4 and P5, who had minimal pain before training, and in P7, P9 and P11 (Figure 3.5B). Total scores from the MPQ are shown as a function of the cumulative training steps because this is our best estimate of intervention dosage, given that participants differed in their rate of learning to walk. No consistent change was seen in the MPQ scores over time (Figure 3.5C).

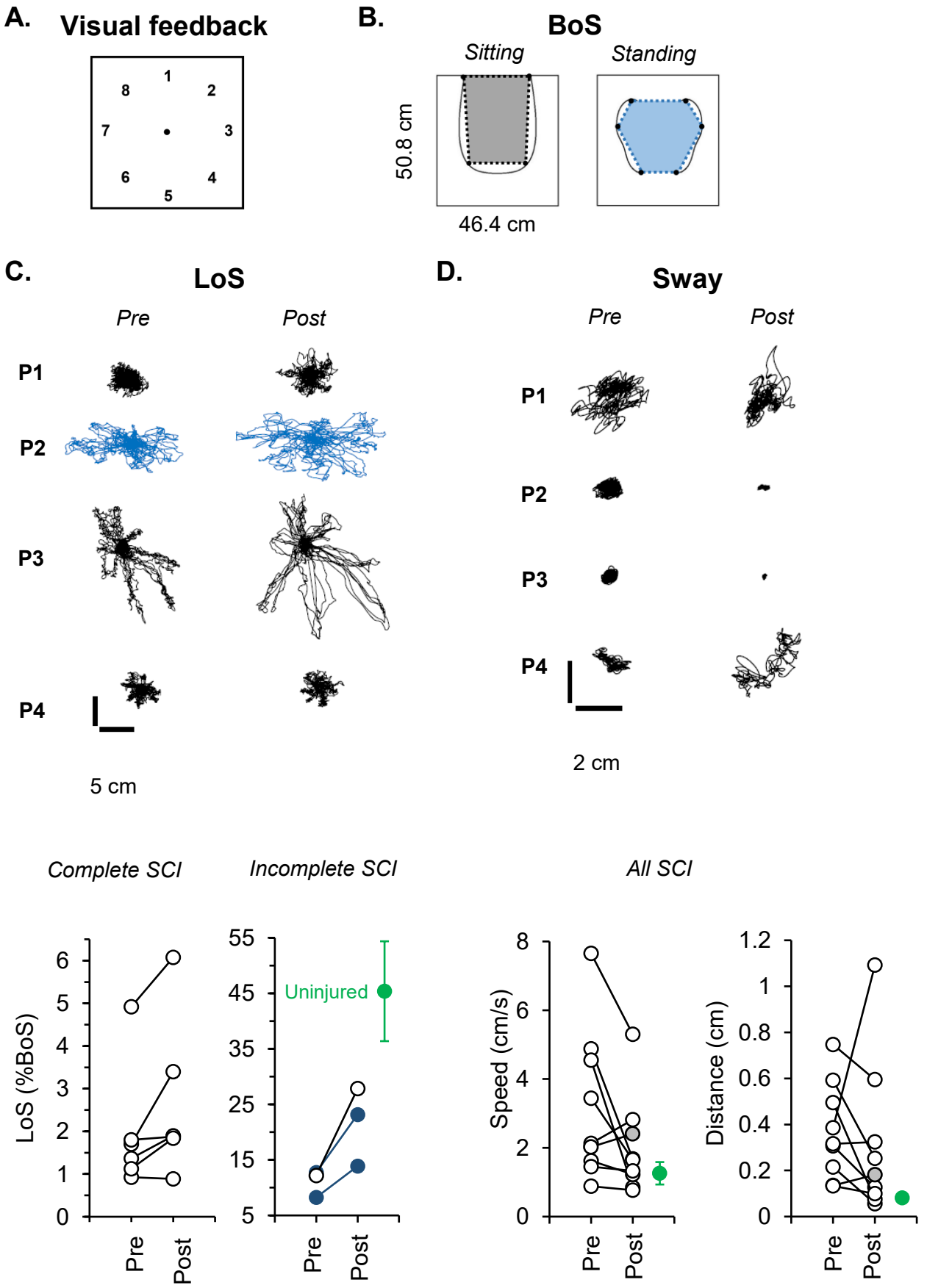
A.
Visual feedback
1
2
3
4
5
6
7
8
B.
BoS
Sitting
Standing
50.8 cm
46.4 cm
C.
LoS
Pre
Post
P1
P2
P3
P4
5 cm
D.
Sway
Pre
Post
P1
P2
P3
P4
2 cm
Complete SCI
Incomplete SCI
All SCI
LoS (%BoS)
Uninjured
Speed (cm/s)
Distance (cm)
Pre
Post

Figure 3.4: Sitting and standing balance before and after training.

(a) Visual feedback of the center of pressure (CoP, center dot) and a schematic of the 8 targets around the CoP. (b) Estimate of the base of support (BoS, shaded areas) on force platforms (rectangular outlines) during sitting and standing. Dotted lines represent the shape of the BoS. (c) Limits of stability (LoS) and (d) sway trajectory for a few participants before (Pre) and after (Post) training in sitting (black traces) and standing (blue traces), showing examples of change (P1, P2, P3) and no change (P4). For the purposes of plotting only, the traces have been down sampled for clarity. Individual scores for all SCI participants before and after training are shown in the line graphs (bottom), with average scores for uninjured participants (n=7) shown for comparison (green, vertical bars = 1 standard deviation). Gray circles are P5's mid-training values, because she only finished 6 weeks of training (see text for details). Blue circles are P2's and P11's measures in standing.

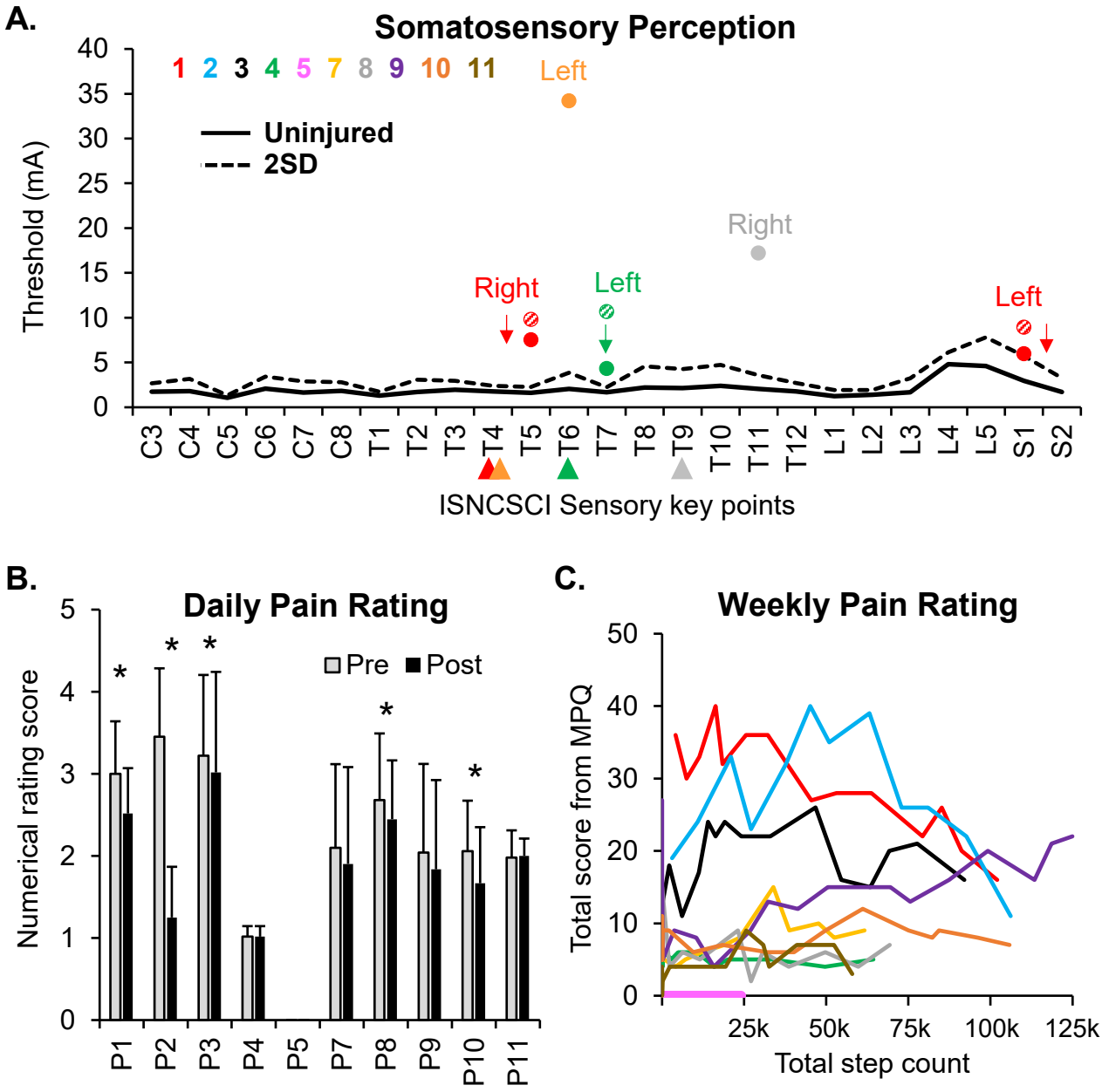
A.
Somatosensory Perception
1 2 3 4 5 7 8 9 10 11
Left
Uninjured
2SD
Right
Right
Left
Left
Threshold (mA)
40
35
30
25
20
15
10
5
0
C3 C4 C5 C6 C7 C8 T1 T2 T3 T4 T5 T6 T7 T8 T9 T10 T11 T12 L1 L2 L3 L4 L5 S1 S2
ISNCSCI Sensory key points
B.
Daily Pain Rating
Pre
Post
Numerical rating score
5
4
3
2
1
0
P1 P2 P3 P4 P5 P7 P8 P9 P10 P11
C.
Weekly Pain Rating
Total score from MPQ
50
40
30
20
10
0
25k 50k 75k 100k 125k
Total step count

Figure 3.5: Changes in somatosensory sensation and neuropathic pain.

(a) Sensory perceptual threshold across the C3-S2 sensory dermatomes. Only dermatomes in which there was a change in threshold as a result of training are shown. P1 and P4 showed a reduction in threshold from before (hatched circles) to after training (solid circles), whereas P8 and P10 developed sensation that was not present at baseline (i.e., threshold >40 mA, see Table 3.3). The neurological level of injury is shown with inverted triangles at the bottom. Thresholds from uninjured participants are shown for comparison (n=3, left and right sides averaged): solid line - mean, dashed line - 2 standard deviations (SD). (b) Average daily numerical ratings of neuropathic pain within a single session, before (Pre) and after (Post) training. (c) Total score from the McGill Pain Questionnaire (MPQ), measured weekly before the training session, shown as a function of the total number of cumulative steps in training. * $p<0.05$.

Table 3.3: Sensory perceptual threshold (mA) for P1, P4, P8 and P10

Sensory key point	BL 1	BL 2	Mid	Post	Follow up
P1, Right					
T1	1.76	1.89	3.03	2.31	2.41
T2	2.9	5.54	2.58	3.25	2.36
T3	2.31	2.71	2.04	2.69	1.66
T4	2.08	1.98	2.05	3.07	1.55
T5*	NS	9.8	7.96	7.52	4.44
P1, Left					
S1*	NS	8.93	7.21	5.98	NS
S2	5.71	5.26	3.68	5.45	4.64
P4, Left					
T4	1.54	1.5	2.09	2.92	2.29
T5	5.15	3.53	3.5	3.8	3.35
T6	3.05	2.72	3.34	3.25	3.02
T7*	13.5	7.85	8.82	4.34	5.18
P8, Right					
T8	2.54	2.44	3.23	2.53	2.56
T9	2.83	2.57	2.98	3.04	3.5
T10	1.97	2.2	2.15	2	1.69
T11*	NS	NS	17.1	17.2	NS
P10, Left					
T3	2.24	1.24	2.29	2.17	2.08
T4	1.59	1.81	2.37	2.28	2.3
T5	15.4	8.97	6.84	19.9	6.79
T6*	NS	NS	NS	34.2	28.1

T: Thoracic; S: Sacral; BL: Baseline; Mid: Middle of training; Post: Immediately after training; NS: No sensation. *Sensory key points where sensation was improved after training.

3.3.7 Muscle strength

Only P3 showed overall improvements in muscle strength: immediately after the training, the total UEMS score from both sides increased from 30 to 37, and the total LEMS score increased from 23 to 25. In contrast, P2 showed a decrease of 1 in the total scores on the UEMS (45 to 44) and LEMS (34 to 33), however, the score for the left ankle dorsiflexor increased from 1 (palpable/visible contractions) to 3 (movement against gravity). Finally, changes in muscle strength were also observed in P11, wherein the UEMS score increased from 33 to 37, and the LEMS score increased from 27 to 30. No other changes were seen.

3.3.8 Spinal cord assessment tool for spasticity

No consistent changes were observed in the total SCATS score throughout the training. Furthermore, the changes immediately after the training were non-significant compared to before training (pre: 2.34 ± 2.87, post: 2.55 ± 3.36, p=0.81).

3.3.9 Affective measures

Similar to the SCATS, changes in the total score from the PHQ-9 throughout the training were inconsistent, and were non-significant immediately after the training compared to before training (pre: 4.95 ± 4.07, post: 4.00 ± 4.38, p=0.16).

3.3.10 Strength of motor pathways

Representative data from participant P8 are shown in Figure 3.6, which demonstrate no obvious changes in the MEPs, despite the fact that he showed excellent walking speeds and distances by the end of training, and some reduction in sensory perceptual threshold. Across participants, the threshold, latency, MEP_{max}, and the slope and y-intercept of the relationship between the MEP amplitude and background EMG did not change from baseline to after training, as shown in Table 3.4. MEP thresholds were difficult to measure in P11, either due to powerline noise or no MEP responses observed at threshold in the spinal extensor muscles at rest.

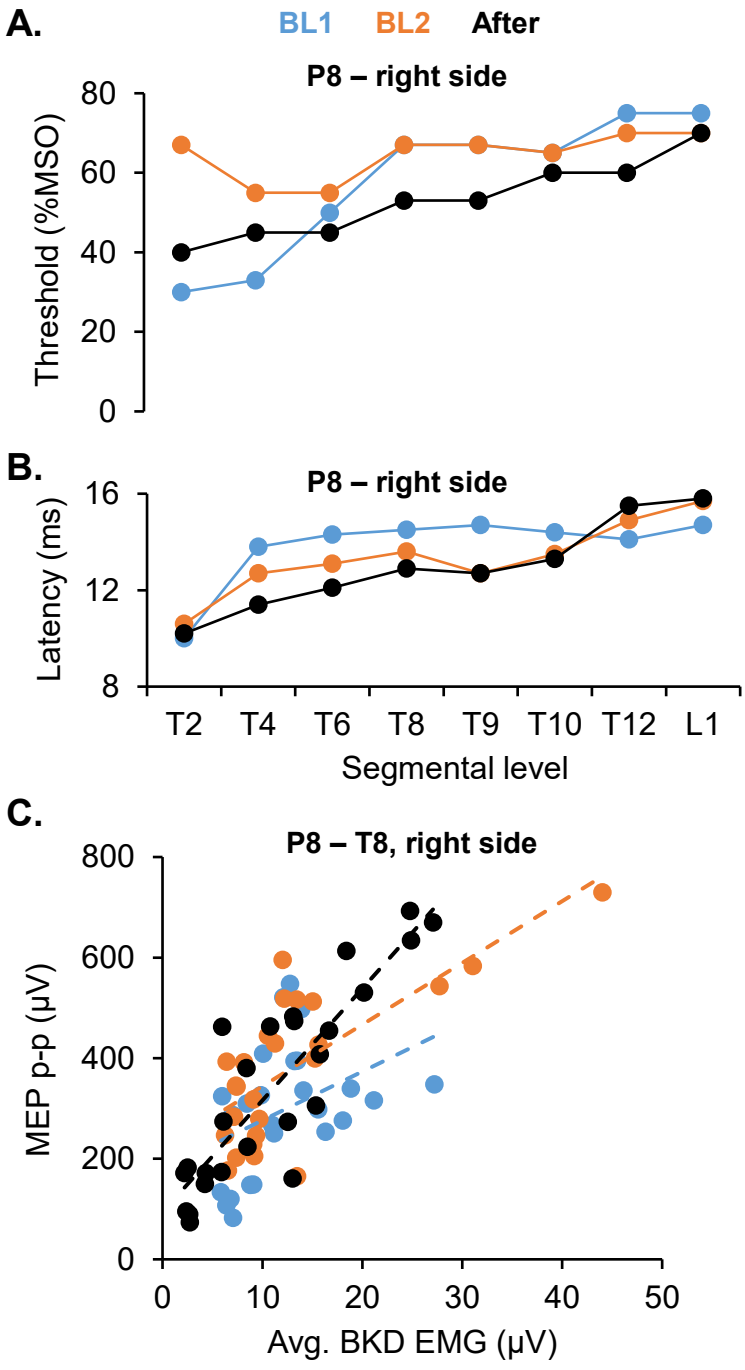

Figure 3.6: Motor evoked potentials in a single participant.

Training in the ReWalk did not affect the strength of the corticospinal tracts to the extensor muscles of the trunk. Representative data for motor evoked potentials (MEP): (a) threshold and (b) latency, and (c) for the relationship between the peak-to-peak (p-p) amplitude of the MEPs and average (Avg.) background (BKD) EMG from P8, before (baseline [BL] 1 & 2) and after training.

Table 3.4: MEP measures induced by TMS

Measure*	Baseline♦ (mean ± SD)	Post (mean ± SD)	N	P-value
Latency (ms)	12.8 ± 3.9	13.1 ± 3.7	10	0.39
	12.8 ± 4.7	13.0 ± 4.6	10	0.63
Threshold (%MSO)‡	55.5 ± 9.4	55.6 ± 7.8	9	0.96
	57.5 ± 11.2	58.1 ± 9.2	9	0.65
BKD @ threshold (μV)	4.9 ± 2.1	6.8 ± 3.6	9	0.09
	5.2 ± 2.3	5.2 ± 3.0	9	0.95
MEP_{max} (μV)	126.3 ± 122.8	151.5 ± 199.6	10	0.32
	257.6 ± 423.4	240.3 ± 533.5	10	0.32
BKD @ MEP_{max} (μV)	5.4 ± 3.3	6.7 ± 3.6	10	0.12
	4.8 ± 2.7	4.7 ± 2.4	10	0.93
Slope	10.6 ± 3.8	10.8 ± 7.5	7	0.93
	10.6 ± 8.1	10.5 ± 6.4	7	0.96
Y-intercept	169.1 ± 110.9	191.3 ± 170.6	7	0.78
	169.3 ± 176.4	107.0 ± 104.4	7	0.39

MEP: Motor evoked potential; TMS: Transcranial magnetic stimulation; SD: Standard deviation; Post: Immediately after training; N: sample size; MSO: Maximum stimulator output; BKD: Background EMG. *Top and bottom values from right and left sides, respectively, ♦Average of Baseline 1 and 2. ‡Excludes P11: thresholds were difficult to measure.

3.3.11 Cutaneomuscular reflex

Reflex data were available from 7 participants, because three were excluded for the following reasons: P1 - technical issues, P5 - no motor response from the AHB, and P8 – no reflex response from the TA. No significant changes in the CMR measures were observed: the reflex threshold as follows, right side pre: 18.33 ± 9.82 mA, post: 15.43 ± 7.3 mA, p=0.49, left side pre: 22.93 ± 12.22 mA, post: 16.43 ± 9.88 mA, p=0.089; reflex magnitude follows, right side pre: 122.08 ± 134.21 mV·s, post: 100.94 ± 108.08 mV·s, p=0.28, left side pre: 101.04 ± 90.95 mV·s, post: 93.64 ± 95.40 mV·s, p=0.66.

3.3.12 Observations from field tests

Indoor field tests were completed by all but P4, who was not proficient enough to perform all the indoor field tests, and P5, who dropped out at mid-training before all indoor tests were completed. All other participants could maneuver between kitchen counters, retrieve items from the refrigerator and overhead cabinets, handle cooking utensils on the counter and stove, and wash dishes at the sink. Maneuvering in tight spaces was challenging for all. Participants used a swing-through gait or pivoted on one leg at a time without engaging the motors in those situations. The following tasks were difficult or impossible: stopping near counters/appliances, reaching that required trunk flexion (such as reaching objects below waist level), standing from sitting in a low chair, opening doors that swing towards the participant.

Only P1, P2 and P7 attempted stairs. P2 and P7 could ascend/descend a short flight of stairs with contact guard and assistance, while P1 had a near fall on stairs. The stair function was not attempted with the remaining participants because of safety concerns. Only P2 ascended and descended a curb with assistance. Only P3 attempted to walk across an uncontrolled intersection, and shop in a grocery store with contact guard. Lastly, only P11 attempted to walk up and down a concrete spiral ramp, without any stoppage in walking while on the ramp.

3.3.13 Follow-up measures

No significant differences were observed between measures immediately after training and at follow up (Table 3.4). For P3, the follow up walking measures without the ReWalk, of the 10MWT (0.14 m/s), 6MWT (33.5 m) and PCI (7.84 beats/m) were similar to those immediately

after the training (compare follow-up values with values in Figure 3.3). Furthermore, at follow up, P3's total score on the UEMS decreased from 37 to 34, and the total score on the LEMS decreased from 25 to 22. The follow up measures of walking function without the ReWalk and muscle strength were taken only from P3 because she became a walker without the ReWalk after the training. Among the 4 participants that showed improvements in sensation immediately after the training (Figure 3.5A), only 2 participants (P1, P8) lost the improvements at follow up (Table 3.3). For P1's sensation, the improvement was retained on the right side, but lost on the left side. Lastly, P10's average duration of sway across the 3 bouts of unsupported sitting was reduced at follow up (0.59 ± 0.23 sec), compared to immediately after the training (1.54 ± 1.29 sec).

3.3.14 Adverse events and technical issues

Skin abrasions were experienced by P1, P4, P7, P10 and P11 at locations under the straps and the pelvic band at the sacrum and iliac crest, at the coccyx, under the knee bracket on the medial side of the knee, and at the base of the 5th metatarsal (when P1 used the ReWalk 2.0 footplates), and at the heel.

A thigh beam on the ReWalk 2.0 fractured just above the knee on 3 separate occasions. Falls were prevented by the trainer and spotter in those instances. Near falls occurred occasionally, so the presence of a spotter and trainer was important. One controlled fall was experienced by P2 and P4, wherein the trainer broke the fall.

The trainer behind the participant also experienced musculoskeletal injuries: biceps muscle strain precipitated by breaking a fall, first degree sprain of the knee during the controlled fall, and a bruised shin when the participant leaned too far forward during stance phase, causing the swing leg to hit the trainer at the time of toe-off.

3.3.15 Effects of breaks in training

There were 19 breaks in training among 9 participants, related to either skin or health issues, holidays, trainer injury or device breakage. Seventeen breaks were analyzed to determine if resumption of training was associated with a reduction in performance. In P4, one break was not analyzed because there were repeated skin problems over a couple of weeks. In P10, one break was not analyzed because the break took place during standing training in the ReWalk, and

therefore its effect on walking was not measurable. The 17 breaks ranged in duration from 0.86 to 8.29 weeks, and did not affect the walking distance (pre: 634.17 ± 419.60 m, post: 718.06 ± 489.57 m, p=0.19) and maximum number of sequential steps (pre: 442.63 ± 376.67, post: 625.01 ± 680.59, p=0.14).

Table 3.5: Comparison between measures at post training and follow-up

Measure	Post (mean ± SD)	Follow-up (mean ± SD)	N	P-value
10MWT (m/s)	0.44 ± 0.12	0.43 ± 0.10	7	0.53
6MWT (m)	149.0 ± 36.8	143.1 ± 38.2	8	0.18
PCI (beats/m)	1.50 ± 0.96	1.49 ± 0.84	8	0.88
LoS (%BoS)	7.72 ± 9.49	8.64 ± 12.67	7	0.3
Sway speed (cm/s)	1.96 ± 1.45	1.64 ± 0.72	7	0.81
Sway distance (cm)	0.29 ± 0.27	0.31 ± 0.18	7	0.77
MPQ	11.71 ± 6.32	12.57 ± 7.23	7	0.47
SCATS	2.75 ± 3.72	2.81 ± 3.56	8	0.69
PHQ-9	5.33 ± 4.27	5.00 ± 4.15	6	0.47

Post: Immediately after training; Follow-up: 2 to 3 months after training; SD: Standard deviation; N: Sample size; 10MWT: 10-Meter Walk Test; 6MWT: 6-Minute Walk Test; PCI: Physiological cost index; LoS: Limits of stability; MPQ: McGill Pain Questionnaire; SCATS: Spinal Cord Assessment Tool for Spasticity; PHQ-9, Patient Health Questionnaire-9.

3.4 Discussion

All participants who completed training learned to walk in the ReWalk for about 1 km without a rest by the end of training. The rate of learning varied between participants. Improvements were also seen in balance, sensory perception, and neuropathic pain in some. Adverse events and/or technical issues were encountered.

3.4.1 Training schedule for walking proficiency

Participants trained a minimum of 3 times/week, except during unavoidable breaks which did not adversely affect the progression of training.

Different aspects of walking improved at different rates. Most participants showed a plateau in walking speed after 30 sessions (Figure 3.2C), whereas walking distance continued to improve (Figure 3.2B). Improvement in walking skill (Figure 3.2D) was more variable, requiring 10-50 sessions to plateau. The exceptions were P2, who was a walker prior to training improved more quickly than others, and P4 who continued to improve after 60 sessions. By the end of training, 7 participants (P1, P2, P3, P7, P8, P10, P11) could walk without assistance, while 3 participants (P4, P5 [who completed only 6 weeks of training], P9) still required occasional physical correction to maintain balance while walking. Overall, we suggest a minimum of 40 sessions of training are necessary for most participants to reach their walking potential in the ReWalk. The differences in the rate of progression of training could possibly have resulted from a combination of: 1) the participants' accommodation to the ReWalk (i.e., learning to use the device), 2) training-induced neuroplasticity, 3) injury severity, 4) motivation and 5) age.

3.4.2 Walking distances and speeds achieved with training

At the end of training, participants walked at speeds of about 0.3 to 0.6 m/s (Figure 3.3A), which is comparable to other reports of powered exoskeletons for over ground walking (Benson et al. 2015; Esquenazi et al. 2012; Farris et al. 2014; Hartigan et al. 2015; Kozlowski et al. 2015; Kressler et al. 2014; Spungen et al. 2013b; Zeilig et al. 2012), except for the Rex (not yet approved for North America), which is much slower (Barbareschi et al. 2015). All of our participants achieved the walking speed of a 'supervised walker' for people with SCI (i.e., about 0.34 m/s) (van Hedel and Group 2009).

The effort of walking (i.e., PCI) in the ReWalk for our participants was 1.54 ± 0.86 beats/m, or 3.2 times that of wheelchair propulsion, 2.6 times that of walking in the uninjured participants (Figure 3.3D), and about 7 times lower than walking with FES and bracing (~11 beats/m) (Stein et al. 2005). Oxygen consumption reported by others was 11.2 ± 1.7 ml/kg/min (or 31% VO^2_{max}, at a speed of at 0.22 ± 0.11 m/s) for the ReWalk (Asselin et al. 2015), and 11.5 ± 1.4 ml/kg/min (or 51.5-63.2% VO^2_{max}, at a speed of 0.27 ± 0.05 m/s) for the Indego (Evans et al. 2015), compared to 16.19 ml/kg/min for FES walking (or 70% VO^2_{max}, speed not reported) (Jacobs et al. 1997). Thus, walking in powered exoskeletons is not exceptionally energy demanding and may be feasible for some individuals with SCI (Biering-Sorensen et al. 2009; Evans et al. 2015; Morse et al. 2008; Phillips et al. 2004).

3.4.3 Neuroplasticity induced by the training

Participants experienced other benefits. For example, prior to training, P3 was a non-walker who could stand only with assistance of one person, and used a sliding board for transfers. After training, she could walk independently at a slow speed using a standard walker, became independent in standing pivot transfers, and muscle strength was improved in the arms and legs. Most participants showed improvements in sitting or standing balance (Figure 3.4), which means the training resulted in transferable gains to other tasks.

Sensory function is rarely reported in intervention studies for walking. Reports of improved sensory function after chronic SCI have mostly involved training of hand function concurrently with FES (Beekhuizen and Field-Fote 2008), or training walking concurrently with either FES (Possover 2014) or robotic resistance (Chisholm et al. 2015). The improvements we observed were achieved by motor training alone, and occurred in dermatomes just below the injury level in addition to more distal regions with partial sparing of sensation (Figure 3.5A). We speculate that these improvements may have been driven by the need to attend to all residual sensory input to successfully walk in the ReWalk. Tracking sensory function in response to all forms of walking training would be valuable in the future.

Neuropathic pain was reduced after each training session in most participants who had high levels of pain before the training (Figure 3.5B), as has been reported for training in the Ekso (Kozlowski et al. 2015; Kressler et al. 2014), but long-term reduction was rare (Figure 3.5C). Anecdotally, one participant with pre-existing and constant musculoskeletal low back pain reported the pain was gone after training.

3.4.4 Limitations of the device

The ReWalk 2.0 is an early version of powered exoskeletons for walking. We document the following problems which may motivate future design improvements. The Velcro straps securing the body to the ReWalk caused skin abrasions in some, even when well padded, and the straps were difficult to manipulate for individuals with limited hand function. Furthermore, the knee brackets, although padded, also led to skin abrasions and therefore may require further padding. Inserting the feet in the shoes was difficult for most of our participants because of limited trunk control. Participants preferred not to use the ReWalk controls because it caused discomfort at the wrist while using the crutches. We suggest placing the controls on the crutches, thereby also avoiding the need to lift one crutch to change functions. The stair function was unsafe, but we understand this has been revised in ReWalk 6.0, which we did not have access to. The device could be made more convenient for rehabilitation by a simpler system for changing the pelvic band to fit different individuals. A narrower device width that fits a standard wheelchair would be more convenient. Fall prevention remains entirely up to the training staff, and to our knowledge, has not been addressed by any over ground powered exoskeleton except the Rex, which is stable, but much heavier and slower (Barbareschi et al. 2015). The battery life became a limitation during the test for the maximum walking distance in P11, measured during the end of training. The battery life lasted approximately 45 min, and the issue remained unresolved despite replacing the battery unit and the ReWalk unit. Finally, our low enrollment of 11 out of 49 individuals was partly because we restricted the study to individuals with chronic SCI, many of whom have secondary problems, precluding their participation. The intensive training and testing schedule may also have been a deterrent.

3.4.5 Clinical implications

The ReWalk is a promising device to train walking in individuals with severe SCI and good upper extremity strength. The manpower required for training is substantial (i.e., 40 sessions with 2 trainers), balanced by considerable benefits such as: walk for long distances indoors and outdoors at a reasonable effort, improved sitting balance in many, and improved muscle strength and skin sensation in some. While limitations remain, we feel that powered exoskeletons such as the ReWalk are making walking possible for many who previously were restricted to a wheelchair for mobility. We hope continued improvements to the devices will make them increasingly feasible for daily use in these individuals.

3.5 Bibliography for Chapter 3

Anderson KD. Targeting recovery: priorities of the spinal cord-injured population. *J Neurotrauma* 21: 1371-1383, 2004b.

Asselin P, Knezevic S, Kornfeld S, Cirnigliaro C, Agranova-Breyter I, Bauman WA, and Spungen AM. Heart rate and oxygen demand of powered exoskeleton-assisted walking in persons with paraplegia. *J Rehabil Res Dev* 52: 147-158, 2015.

Barbareschi G, Richards R, Thornton M, Carlson T, and Holloway C. Statically vs dynamically balanced gait: Analysis of a robotic exoskeleton compared with a human. *Conf Proc IEEE Eng Med Biol Soc* 2015: 6728-6731, 2015.

Beekhuizen KS, and Field-Fote EC. Sensory stimulation augments the effects of massed practice training in persons with tetraplegia. *Archives of physical medicine and rehabilitation* 89: 602-608, 2008.

Benson I, Hart K, Tussler D, and van Middendorp JJ. Lower-limb exoskeletons for individuals with chronic spinal cord injury: findings from a feasibility study. *Clin Rehabil* 30: 73-84, 2016.

Benson I, Hart K, Tussler D, and van Middendorp JJ. Lower-limb exoskeletons for individuals with chronic spinal cord injury: Findings from a feasibility study. *Clin Rehabil* 2015.

Benz EN, Hornby TG, Bode RK, Scheidt RA, and Schmit BD. A physiologically based clinical measure for spastic reflexes in spinal cord injury. *Archives of physical medicine and rehabilitation* 86: 52-59, 2005.

Bernardi M, Canale I, Castellano V, Di Filippo L, Felici F, and Marchetti M. The efficiency of walking of paraplegic patients using a reciprocating gait orthosis. *Paraplegia* 33: 409-415, 1995.

Biering-Sorensen F, Hansen B, and Lee BS. Non-pharmacological treatment and prevention of bone loss after spinal cord injury: a systematic review. *Spinal cord* 47: 508-518, 2009.

Brown-Triolo DL, Roach MJ, Nelson K, and Triolo RJ. Consumer perspectives on mobility: implications for neuroprosthesis design. *J Rehabil Res Dev* 39: 659-669, 2002.

Cariga P, Catley M, Nowicky AV, Savic G, Ellaway PH, and Davey NJ. Segmental recording of cortical motor evoked potentials from thoracic paravertebral myotomes in complete spinal cord injury. *Spine* 27: 1438-1443, 2002.

Chisholm AE, Peters S, Borich MR, Boyd LA, and Lam T. Short-term cortical plasticity associated with feedback-error learning after locomotor training in a patient with incomplete spinal cord injury. *Physical therapy* 95: 257-266, 2015.

Coghlan JK, Robinson CE, Newmarch B, and Jackson G. Lower extremity bracing in paraplegia--a follow-up study. *Paraplegia* 18: 25-32, 1980.

Dobkin B, Apple D, Barbeau H, Basso M, Behrman A, Deforge D, Ditunno J, Dudley G, Elashoff R, Fugate L, Harkema S, Saulino M, Scott M, and Spinal Cord Injury Locomotor Trial G. Weight-supported treadmill vs over-ground training for walking after acute incomplete SCI. *Neurology* 66: 484-493, 2006.

Esquenazi A, Talaty M, Packel A, and Saulino M. The ReWalk powered exoskeleton to restore ambulatory function to individuals with thoracic-level motor-complete spinal cord injury. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 91: 911-921, 2012.

Evans N, Hartigan C, Kandilakis C, Pharo E, and Clesson I. Acute Cardiorespiratory and Metabolic Responses During Exoskeleton-Assisted Walking Overground Among Persons with Chronic Spinal Cord Injury. *Top Spinal Cord Inj Rehabil* 21: 122-132, 2015.

Farris RJ, Quintero HA, Murray SA, Ha KH, Hartigan C, and Goldfarb M. A preliminary assessment of legged mobility provided by a lower limb exoskeleton for persons with paraplegia. *IEEE transactions on neural systems and rehabilitation engineering : a publication of the IEEE Engineering in Medicine and Biology Society* 22: 482-490, 2014.

Harel NY, Asselin PK, Fineberg DB, Pisano TJ, Bauman WA, and Spungen AM. Adaptation of computerized posturography to assess seated balance in persons with spinal cord injury. *The journal of spinal cord medicine* 36: 127-133, 2013.

Hartigan C, Kandilakis C, Dalley S, Clausen M, Wilson E, Morrison S, Etheridge S, and Farris R. Mobility Outcomes Following Five Training Sessions with a Powered Exoskeleton. *Topics in Spinal Cord Injury Rehabilitation* 21: 93-99, 2015.

Hesse S, Werner C, and Bardeleben A. Electromechanical gait training with functional electrical stimulation: case studies in spinal cord injury. *Spinal cord* 42: 346-352, 2004.

http://eksobionics.com/. Ekso Bionics Ekso Bionics. http://eksobionics.com/.

http://rewalk.com/. ReWalk 6.0 – Home ReWalk Robotics. http://rewalk.com/.

http://www.indego.com/indego/en/home. Indego – Powering People Forward | Parker Indego Parker Hannifin Corp. http://www.indego.com/indego/en/home.

Jacobs PL, Klose KJ, Guest R, Needham-Shropshire B, Broton JG, and Green BA. Relationships of oxygen uptake, heart rate, and ratings of perceived exertion in persons with paraplegia during functional neuromuscular stimulation assisted ambulation. *Spinal Cord* 35: 292-298, 1997.

Jones CA, and Yang JF. Reflex behavior during walking in incomplete spinal-cord-injured subjects. *Experimental neurology* 128: 239-248, 1994.

Khan A, Livingstone D, Misiaszek J, Stein RB, Gorassini M, Manns P, and Yang J. Improving walking and inducing neuroplasticity after chronic SCI by training in the ReWalk exoskeleton. In: *Society for Neuroscience 2015 Annual meeting*. Chicago, IL: 2015.

Kozlowski AJ, Bryce TN, and Dijkers MP. Time and effort required by persons with spinal cord injury to learn to use a powered exoskeleton for assisted walking. *Topics in Spinal Cord Injury Rehabilitation* 21: 110-121, 2015.

Kressler J, Thomas CK, Field-Fote EC, Sanchez J, Widerstrom-Noga E, Cilien DC, Gant K, Ginnety K, Gonzalez H, Martinez A, Anderson KD, and Nash MS. Understanding

therapeutic benefits of overground bionic ambulation: exploratory case series in persons with chronic, complete spinal cord injury. *Archives of physical medicine and rehabilitation* 95: 1878-1887 e1874, 2014.

Lajeunesse V, Vincent C, Routhier F, Careau E, and Michaud F. Exoskeletons' design and usefulness evidence according to a systematic review of lower limb exoskeletons used for functional mobility by people with spinal cord injury. *Disability and rehabilitation Assistive technology* 1-13, 2015.

Lemay JF, Gagnon D, Duclos C, Grangeon M, Gauthier C, and Nadeau S. Influence of visual inputs on quasi-static standing postural steadiness in individuals with spinal cord injury. *Gait Posture* 38: 357-360, 2013.

Leung AK, Wong AF, Wong EC, and Hutchins SW. The Physiological Cost Index of walking with an isocentric reciprocating gait orthosis among patients with T(12) - L(1) spinal cord injury. *Prosthet Orthot Int* 33: 61-68, 2009.

Lotta S, Fiocchi A, Giovannini R, Silvestrin R, Tesio L, Raschi A, Macchia L, Chiapatti V, Zambelli M, and Tosi C. Restoration of gait with orthoses in thoracic paraplegia: a multicentric investigation. *Paraplegia* 32: 608-615, 1994.

Maynard FM, Jr., Bracken MB, Creasey G, Ditunno JF, Jr., Donovan WH, Ducker TB, Garber SL, Marino RJ, Stover SL, Tator CH, Waters RL, Wilberger JE, and Young W. International Standards for Neurological and Functional Classification of Spinal Cord Injury. American Spinal Injury Association. *Spinal cord* 35: 266-274, 1997.

Melzack R. The McGill Pain Questionnaire: major properties and scoring methods. *Pain* 1: 277-299, 1975.

Mikelberg R, and Reid S. Spinal cord lesions and lower extremity bracing: an overview and follow-up study. *Paraplegia* 19: 379-385, 1981.

Morse LR, Stolzmann K, Nguyen HP, Jain NB, Zayac C, Gagnon DR, Tun CG, and Garshick E. Association between mobility mode and C-reactive protein levels in men with chronic spinal cord injury. *Archives of physical medicine and rehabilitation* 89: 726-731, 2008.

Phillips SM, Stewart BG, Mahoney DJ, Hicks AL, McCartney N, Tang JE, Wilkinson SB, Armstrong D, and Tarnopolsky MA. Body-weight-support treadmill training improves blood glucose regulation in persons with incomplete spinal cord injury. *Journal of applied physiology (Bethesda, Md : 1985)* 97: 716-724, 2004.

Possover M. Recovery of sensory and supraspinal control of leg movement in people with chronic paraplegia: a case series. *Archives of physical medicine and rehabilitation* 95: 610-614, 2014.

Savic G, Bergstrom EM, Frankel HL, Jamous MA, Ellaway PH, and Davey NJ. Perceptual threshold to cutaneous electrical stimulation in patients with spinal cord injury. *Spinal cord* 44: 560-566, 2006.

Siddall PJ, Cousins MJ, Otte A, Griesing T, Chambers R, and Murphy TK. Pregabalin in central neuropathic pain associated with spinal cord injury: a placebo-controlled trial. *Neurology* 67: 1792-1800, 2006.

Simpson LA, Eng JJ, Hsieh JT, Wolfe, and the Spinal Cord Injury Rehabilitation Evidence Research Team DL. The health and life priorities of individuals with spinal cord injury: a systematic review. *Journal of neurotrauma* 29: 1548-1555, 2012.

Spungen AM, Asselin P, Fineberg DB, Kornfeld S, and Harel NY. Exoskeletal-assisted walking for persons with motor-complete paraplegia. In: *Force Sustainment: Rehabilitation, Regeneration and Prosthetics for Re-Integration to Duty Meeting Proceedings STO-MP-HFM-228, Paper 6* Neuilly-sur-Seine, France: Science and Technology Organization, 2013b, p. 6-1 - 6-14.

Spungen AM, Asselin PK, Fineberg DB, Kornfeld SD, and Harel NY. Exoskeletal-assisted walking for persons with motor-complete paraplegia. In: *North Atlantic Treaty Organization (NATO), Research and Technology Organization, Human Factors and Medicine Panel, HFM-228*. Italy: 2013c.

Stein RB, Hayday F, Chong S, Thompson AK, Rolf R, James KB, and Bell G. Speed and efficiency in walking and wheeling with novel stimulation and bracing systems after spinal cord injury: a case study. *Neuromodulation* 8: 264-271, 2005.

Sykes L, Edwards J, Powell ES, and Ross ER. The reciprocating gait orthosis: long-term usage patterns. *Archives of physical medicine and rehabilitation* 76: 779-783, 1995.

Talaty M, Esquenazi A, and Briceno JE. Differentiating ability in users of the ReWalk(TM) powered exoskeleton: an analysis of walking kinematics. *IEEE International Conference on Rehabilitation Robotics : [proceedings]* 2013: 6650469, 2013.

van Hedel HJ, and Group ES. Gait speed in relation to categories of functional ambulation after spinal cord injury. *Neurorehabil Neural Repair* 23: 343-350, 2009.

van Hedel HJ, Wirz M, and Dietz V. Assessing walking ability in subjects with spinal cord injury: validity and reliability of 3 walking tests. *Archives of physical medicine and rehabilitation* 86: 190-196, 2005.

van Middendorp JJ, Hosman AJ, Donders AR, Pouw MH, Ditunno JF, Jr., Curt A, Geurts AC, Van de Meent H, and Group E-SS. A clinical prediction rule for ambulation outcomes after traumatic spinal cord injury: a longitudinal cohort study. *Lancet* 377: 1004-1010, 2011.

Waters RL, Adkins R, Yakura J, and Vigil D. Prediction of ambulatory performance based on motor scores derived from standards of the American Spinal Injury Association. *Arch Phys Med Rehabil* 75: 756-760, 1994.

Wittkampf K, van Ravesteijn H, Baas K, van de Hoogen H, Schene A, Bindels P, Lucassen P, van de Lisdonk E, and van Weert H. The accuracy of Patient Health Questionnaire-9 in detecting depression and measuring depression severity in high-risk groups in primary care. *General hospital psychiatry* 31: 451-459, 2009.

Yang A, Asselin P, Knezevic S, Kornfeld S, and Spungen AM. Assessment of In-Hospital Walking Velocity and Level of Assistance in a Powered Exoskeleton in Persons with Spinal Cord Injury. *Top Spinal Cord Inj Rehabil* 21: 100-109, 2015.

Yang JF, Norton J, Nevett-Duchcherer J, Roy FD, Gross DP, and Gorassini MA. Volitional muscle strength in the legs predicts changes in walking speed following locomotor training in people with chronic spinal cord injury. *Physical therapy* 91: 931-943, 2011.

Yang JF, and Stein RB. Phase-dependent reflex reversal in human leg muscles during walking. *Journal of neurophysiology* 63: 1109-1117, 1990.

Zeilig G, Weingarden H, Zwecker M, Dudkiewicz I, Bloch A, and Esquenazi A. Safety and tolerance of the ReWalk exoskeleton suit for ambulation by people with complete spinal cord injury: a pilot study. *The journal of spinal cord medicine* 35: 96-101, 2012.

CHAPTER 4: RELIABILITY OF SPINAL REFLEXES REFLECTING THE Ia-MOTONEURONAL PATHWAY AFTER INCOMPLETE SPINAL CORD INJURY

4.1 Introduction

The development of spasticity is a common sequela of spinal cord injury (SCI), manifesting as hyperexcitable tendon, stretch and Hoffmann (H-) reflexes, flexor and cutaneomuscular reflexes, clonus and spasms (see Roy and Edgerton (2012) and D'Amico et al. (2014) for review). Spasticity impacts various aspects in the life of individuals with SCI (Johnson et al. 1998) in negative and positive ways. Negative effects of spasticity include interfering with activities of daily living, sleep, transfers, health (e.g., pain, skin problems) (Little et al. 1989; Skold et al. 1999) and mobility (Corcos et al. 1986; Hubli et al. 2012; Krawetz and Nance 1996). In contrast, some of the positive effects include assistance in functional activities such as transfers, hand grip and maintaining muscle mass (Dietz 2003; Reyes et al. 2015).

Numerous interventions are now undertaken to address spasticity (see Roy and Edgerton (2012), Elbasiouny et al. (2010) and D'Amico et al. (2014) for review). A few of these include: 1) physical training, e.g., passive cycling (Kiser et al. 2005) and treadmill walking with body-weight support (Adams and Hicks 2011; Gorassini et al. 2009; Manella and Field-Fote 2013) or with the Lokomat (Hubli et al. 2012; Knikou 2013; Smith et al. 2014), 2) pharmacological treatment, e.g., baclofen (Lazorthes et al. 1990), gabapentin (Gruenthal et al. 1997), clonidine (Nance 1994), tizanidine (Mirbagheri et al. 2010), cyproheptadine (Wainberg et al. 1990), zolmitriptan (D'Amico et al. 2013), and L-dopa (Eriksson et al. 1996), 3) surface (Mirbagheri et al. 2002) and epidural electrical stimulation (Pinter et al. 2000), 4) denervation using botulinum toxin (Al-Khodairy et al. 1998; Fried and Fried 2003), dantrolene sodium (Glass and Hannah 1974) or phenol (Gunduz et al. 1992), and 5) surgery, e.g., dorsal rhizotomy (Reynolds et al. 2014).

One way to evaluate the efficacy of any intervention to reduce the excitability of hyperexcitable reflexes after SCI is to compare the magnitude of the reflex response before and after the intervention. A reliable assessment of the change in the reflex response after the intervention relies on the consistency of the reflex response across multiple baseline measures before the intervention. However, the majority of the studies on interventions that aim to reduce the

excitability of hyperexcitable reflexes after SCI measure the reflex response either once or multiple times on a single day before the intervention, without first establishing the reliability of the reflex response across multiple days (Adams and Hicks 2011; Eriksson et al. 1996; Gruenthal et al. 1997; Hubli et al. 2012; Kiser et al. 2005).

Given the importance of the reliability of reflex measures before any intervention that may affect the excitability of hyperexcitable reflexes, we sought to determine the test-retest reliability of three commonly used spinal reflexes, 1) the rate-dependent depression of the H-reflex, 2) H_{max}/M_{max} (both measured at rest during sitting), and 3) clonus (measured during walking), all of which involve the Ia-motoneuron reflex pathway, after chronic, motor-incomplete SCI. We further determined the relationship between these reflexes.

4.2 Methodology

4.2.1 Participants

Inclusion criteria: 1) non-progressive, chronic (>7 months) motor-incomplete SCI between C1 and L1 neurological levels, 2) able to provide informed written consent, 3) able to walk for 3 minutes on a treadmill with or without body-weight support. Exclusion criteria: head injury, cognitive impairment, musculoskeletal abnormalities and/or other comorbidities besides SCI that would impede reflex measurements during sitting and walking. For comparison with the SCI participants, reflexes were also measured from neurologically intact, uninjured participants (i.e., control participants).

The Health and Research Ethics Board at the University of Alberta, approved this study (Pro00031413). All participants provided informed written consent prior to enrollment.

4.2.2 Experimental design

Reflexes were measured from the soleus muscle (SOL), because the ankle plantarflexors commonly become hypertonic after SCI (Nielsen et al. 1993b; Schindler-Ivens and Shields 2000). In order to establish the test-retest reliability, reflexes were measured 2 – 5 times across different days (once per day) in the SCI and control participants. In some participants, not all reflexes were recorded each time (see *Section 4.3.1,* and Table 4.3 for details). Furthermore, to

determine the relationship between the reflexes, additional data from participants with only a single measure of a reflex were also included (Table 4.3). H_{max}/M_{max} and rate-dependent depression of the H-reflex (henceforth referred to as 'RDD') were measured at rest during sitting, and clonus was measured during approximately 3 minutes of undisturbed walking on a level treadmill. Clonus was recorded last to avoid the recurrent muscle stretches during walking (Capaday and Stein 1987) from affecting the excitability of the stretch reflex pathway (Nielsen et al. 1993b), which may confound the measures of H_{max}/M_{max} and RDD, particularly since different participants may have different walking abilities. Reflexes were measured from the more spastic side in the SCI group, as determined by the participant's self-report or opinion of the clinical staff, and from the right side in the control participants.

4.2.3 Measurements

4.2.3.1 Instrumentation

Bipolar surface electromyogram (EMG) signals of the SOL were recorded using disposable Ag/AgCl electrodes (Kendall H59P, Mansfield, MA), placed 2 cm center-to-center, caudal to the muscle belly of the lateral gastrocnemius and lateral to the midline of the Achilles tendon. A ground electrode was placed between the recording electrodes and the knee (site of stimulation - see below). EMG signals were amplified, band-pass filtered between 10 – 1,000 Hz (AMT-8 EMG system, Bortec Biomedical Ltd, Calgary, AB, Canada) and digitized at 5 kHz (Molecular Devices, Digidata 1322A, Sunnyvale, CA).

To stimulate the posterior tibial nerve (PTN), the cathode (single, disposable Ag/AgCl electrode) was placed at the popliteal fossa at the optimal site of stimulation, as determined by a hand-held probe. To ensure proximity of the cathode to the PTN, the electrode was pressed against the knee using padding, and wrapped around the knee using Nylatex Wraps (DJO Global, Vista, CA). A 50 × 100 mm or 50 × 90 mm self-adhesive electrode (Axelgaard Manufacturing Co., Ltd., Fallbrook, CA) was placed at the anterior aspect of the thigh as the anode. Pulse width of 1 ms was used across all participants. In one SCI participant, pulse width of 0.5 ms was used by mistake. Data from this participant were included because the amplitude of H-reflexes evoked using 0.5 and 1 ms pulse widths are not considered different (Panizza et al. 1989).

4.2.3.2 H-reflexes at rest during sitting

The tested leg was positioned with knee and ankle angles at approximately 30° of flexion and approximately 15° of plantarflexion, respectively. The non-tested leg rested on the ground with knee and ankle angles at approximately 90°. Participants with SCI sat in their wheelchairs, whereas control participants sat in a comfortable chair. H- and M-recruitment curves were obtained from stimulation to the PTN at a regular rate of 0.5 Hz or slower (Ishikawa et al. 1966). Note that the H- and M-recruitment curves were measured at 0.5 Hz stimulation in 3 (P2, P9, P10) of the SCI participants. Stimulus intensity was slowly increased from below threshold for the H-reflex and M-response until the H-reflex disappeared, and the M-response plateaued. The maximum amplitude of the H-reflex (H_{max}) and M-response (M_{max}) were determined from the H- and M-recruitment curves, to estimate the H_{max}/M_{max}.

RDD was measured at 0.1, 0.2, 0.3, 0.5, 1, 2 and 5 Hz, using 15 stimuli at each rate to the PTN, at a stimulus intensity generating 50% H_{max}, confirmed by the amplitude of the first H-reflex. A second trial of RDD at 0.1 or 0.2 Hz was repeated after all the frequency trials to determine the stability of the responses over time. Since during the first 7 participants (P1, P2, P5, P6, P9, P10, P11), we determined that the H-reflexes at 0.1 Hz showed more variability than those at 0.2 Hz, and the 5 Hz stimulation induced spasms in some participants, the trials recorded at 0.1 and 5 Hz were excluded from the analysis for those participants, and were subsequently not recorded in the remaining participants. H-reflexes could not be measured in P13 due to swelling in the whole leg, whereas clonus was measured.

4.2.3.3 Clonus during walking

Clonus was estimated during treadmill walking. SOL EMG signals were measured during a single trial of approximately 3 minutes of undisturbed walking on a level treadmill at the participant's self-selected walking speed. The speed chosen by the participant in the first session was used on subsequent sessions. For the SCI participants, body-weight support was provided as needed; the majority used the harness for safety only (see *Section 4.3 Results*).

4.2.4 Data analysis

Data were analyzed using custom written codes in the Matlab programing environment (The MathWorks Inc., MA).

4.2.4.1 H-reflexes

The H-reflex and M-response were quantified using the peak-to-peak (p-p) amplitudes of the responses from unrectified EMG. RDD was estimated from the average p-p amplitude of the last 8 reflexes at 0.3, 0.5, 1 and 2 Hz, normalized to that at 0.2 Hz, because reflexes across the last 8 stimuli are stable during RDD (Ishikawa et al. 1966). The 0.2 Hz trial was selected as a reference for normalization because RDD at 0.2 Hz is absent in people with SCI (Schindler-Ivens and Shields 2000), and because of the amplitude of H-reflexes at 0.1 Hz was more variable. Nevertheless, in uninjured people, RDD is present at 0.2 Hz (Schindler-Ivens and Shields 2000), however, there was no significant difference ($p=0.13$) in the absolute values of the average of the last 8 H-reflexes at 0.2 Hz between our sample of SCI (1.54 ± 1.27 mV) and control participants (2.07 ± 0.70 mV). Furthermore, since in our sample of control participants, inhibition during RDD was largest at 2 Hz (24.7%), we only compared RDD at the 2 Hz trial [henceforth, referred to as the '0.5/5 ratio' (Achache et al. 2010)].

Two parameters were examined to ensure consistency of stimulating and recording conditions within and across sessions for the H-reflex recordings: the size of the M_{max} across recording days, and the background EMG just prior to eliciting the reflex for all trials. H_{max}/M_{max} and RDD from sessions with unmatched M_{max} were excluded from analyses. That is, values of the M_{max} were visually compared across days for consistency, and data were excluded if the value of the M_{max} was different from those recorded on the other days. H_{max}/M_{max} and RDD were also excluded if the background EMG was high (i.e., greater than EMG signal noise: ~5 μV or greater) at the H_{max}. Background EMG and M-responses were also examined across the 15 sweeps during RDD at 0.2 and 2 Hz trials. If background EMG was high (~5 μV or greater) or the M-responses were not consistent within and across the individual sweeps during the two trials, then H-reflexes at those sweeps were excluded from analyses.

4.2.4.2 Clonus

The entire length of the 3 min of unrectified SOL EMG signal during walking was corrected for DC bias, rectified and analyzed using fast Fourier transform between the clonus frequency range of 4-10 Hz (Beres-Jones et al. 2003; Dimitrijevic et al. 1980a; Gorassini et al. 2009; Rossi et al. 1990; Wallace et al. 2012; Walsh 1976). The power of the EMG signal in the clonus range was normalized to the power of the EMG between 0 and 40 Hz, as previously done (Khan et al. 2016), according to:

$$\text{Clonus Power} = \frac{\text{SOL EMG power between 4-10 Hz}}{\text{SOL EMG power between 0-40 Hz}}$$

Contamination from power-line noise was avoided by limiting the frequency analysis to < 40 Hz. The frequency analysis between the clonus frequency range and between 0 and 40 Hz was performed in steps of 0.5 Hz, and determined the power of the SOL EMG signal that was present during both the stance as well as the swing phases of the walking. Walking trials were excluded from analyses if the raw SOL EMG signal was contaminated with single motor unit activity, observed during visual inspection of the EMG signal, whose firing rate (Gorassini et al. 2004) can confound the EMG signal power within the clonus frequency range.

4.2.5 Statistical analyses

4.2.5.1 Differences between injured and uninjured participants

In participants with more than one measurement time point, the multiple measures across the different days were first averaged to obtain the best estimate for that person. Similarly, age at the time of each experiment was averaged across days. Group comparisons for age, RDD, H_{max}/M_{max} and clonus were performed using the Independent samples t test or Mann-Whitney U test for normally and non-normally distributed data, respectively.

4.2.5.2 Variability of reflexes

The variability of reflex measures was estimated in two ways: 1) variability across *all* participants, and 2) variability between days for *each* participant. Variability was quantified using the coefficient of variation:

$$\text{Coefficient of variation (CV)} = \frac{\text{Standard deviation (SD)}}{\text{Mean}} \times 100\%$$

In the SCI and control groups, the variability of reflex measures between each group was determined in order to compare the spread of the reflex values in each group ($CV_{between}$), and was estimated using the group mean and SD of the reflex measures.

In the SCI group, the variability of reflex measures within each participant was estimated using the mean and SD of the reflex measures across all multiple days for that participant (CV_{Within}).

Lastly, to quantify the consistency of the stimulating and recording conditions between days, the between-day variability of the M_{max} within each SCI and control participant was also estimated using the mean and SD of the M_{max} across all multiple days for that participant, and then averaged across participants for each group (CV_{Mmax}).

4.2.5.3 Relationship between reflexes

In the SCI group, linear correlation between the average magnitude of the different reflexes across participants was determined using the Pearson's correlation analysis. Significance was set at $p<0.05$. Reported values are mean ± 1 SD.

4.3 Results

4.3.1 Participants

Reflexes were measured from 13 SCI participants (Table 4.1) and 5 age-matched, control participants (Table 4.2). In both groups, acceptable reflexes were obtained on a single day or across 2-4 days (Table 4.3). Within the SCI group, all 13 participants provided data for the estimate of correlation between reflexes, and all but two (* in Table 4.1) provided data for the estimate of test-retest reliability. All control participants provided data for both estimates.

Table 4.1: Spinal cord injury participant characteristics

Participant code-gender	Age (yr)	Neurological level of injury	Cause of injury	Time since injury (yr)	Side of reflex testing	Anti-spastic medication
P1-M	28.6	T4	MVA	4.6	L	None
P2-M	45.5	C6	MVA	7.2	R	None
P3-M	55.7	C6-7	MVA	18.2	R	Baclofen
P4-F	54.1	T2	Surgical clot	11.0	R	None
P5-M	67.9	C4	Sports	24.6	L	None
P6-M	52.1	T10	Surgical bleed	1.4	R	Baclofen
P7-M	64.8	T12	Bull attack	33.3	R	None
P8-F	57.2	C6	MVA	10.5	R	Baclofen
P9-M*	47.9	T12	Fall	0.7	R	Baclofen
P10-M	64.9	C3	MVA	1.1	L	Baclofen
P11-M*	47.8	T12	MVA	10.8	L	None
P12-F	30.7	C6	MVA	4.2	R	Yes‡
P13-M	49.6	C4	Work injury	1.3	R	Baclofen
Mean (SD)	**51.3 (11.9)**			**9.9 (10.0)**		

M: Male; F: Female; C: Cervical; T: Thoracic; MVA: Motor vehicle accident; R: Right; L: Left; SD: Standard deviation; * Provided data only for the estimate of correlation between reflexes. ‡ Name of anti-spastic medication not available.

Table 4.2: Control participant characteristics

Participant code-gender	Age (yr)
C1-F	55.2
C2-F	46.1
C3-M	32.1
C4-F	52.4
C5-F	58.7
Mean (SD)	**48.9 (10.5)**

F: Female; M: Male; SD: Standard deviation.

Table 4.3: Number of reflex measures obtained across all participants

Spinal cord injury participants			
Participant code	RDD	H_{max}/M_{max}	Clonus
P1	3	3	3
P2	2	2	2
P3	3	3	4
P4	2	2	3
P5	2	3	4
P6	2	2	2
P7	2	3	0
P8	3	3	0
P9	1	1	0
P10	1	1	2
P11	1	1	0
P12	1	2	3
P13	0	0	2
Control participants			
Participant code	RDD	H_{max}/M_{max}	Clonus
C1	3	3	3
C2	3	3	1
C3	2	2	3
C4	2	3	3
C5	2	2	0

RDD: Rate-dependent depression of the H-reflex.

4.3.2 Differences between injured and uninjured participants

Age of the SCI participants that contributed data to the estimates of correlation between reflexes (51.3 ± 11.9 yr) and test-retest reliability (51.9 ± 13.0 yr) was not significantly different from that of the control participants (48.9 ± 10.5 yr); p=0.7 and p=0.65, respectively. Average walking speeds on the treadmill was 0.23 ± 0.15 m/s in the SCI group, and 0.60 ± 0.17 m/s in the control group. In the SCI group, only P2 required body-weight support during walking (50 lb, which was held constant across days). All other SCI participants used the harness for safety without weight support, except P3 and P13, who felt safe without the harness.

Representative data for RDD, H_{max}/M_{max} and clonus from single participants in the SCI (P2) and control (C3) groups are illustrated in Figure 4.1. In P2, H-reflexes evoked at 0.2-1 Hz are variable and did not exhibit RDD, whereas RDD is observed at 2 Hz only (Figure 4.1A). In contrast, C3 showed RDD at 0.3-2 Hz. Despite the greater excitability of the H-reflexes during RDD in P2, the excitability of the H_{max}/M_{max} is lower in P2 compared to C3 (Figure 4.1B). Rectified and smoothed (20 Hz low-pass, 2^{nd} order, dual pass, digital Butterworth filtered) SOL EMG (down sampled by 20 data points for clarity) from P2 depicts clonus as rhythmic bursts at the end of the regular SOL EMG (horizontal lines in Figure 4.1C), which are not observed in C3 (Figure 4.1C).

Individual data of reflex values (Figure 4.2A-C), and the H_{max} and M_{max} values (Figure 4.2D-E) from all participants are presented in Figure 4.2, with the group average and median represented by the filled and unfilled horizontal lines, respectively. Higher numbers indicate increased reflex excitability. Thus, the higher 0.5/5.0 ratio of RDD at the 2 Hz stimulation indicated that the SCI group showed higher reflex excitability compared to the control group (SCI: 58.6 ± 32.5%, control: 24.7 ± 15.3%, p = 0.044 - Figure 4.2A). H_{max}/M_{max} did not differ significantly between the two groups (SCI: 54.7 ± 19.8%, control: 62.6 ± 7.4%, p = 0.25 - Figure 4.2B), nor did clonus (SCI: 0.064 ± 0.054, control: 0.038 ± 0.004, p = 0.60 – Figure 4.2C).

Differences between the SCI and control participants were also observed in the spread of the reflex values. Using the group average and SD of the individual data in Figure 4.2, the $CV_{between}$ was determined to estimate the between-group variability of the reflexes in each group. The

$CV_{Between}$ of RDD was similar between the two groups, while the $CV_{Between}$ of H_{max}/M_{max} and clonus was higher among the SCI compared to the control participants (Figure 4.3).

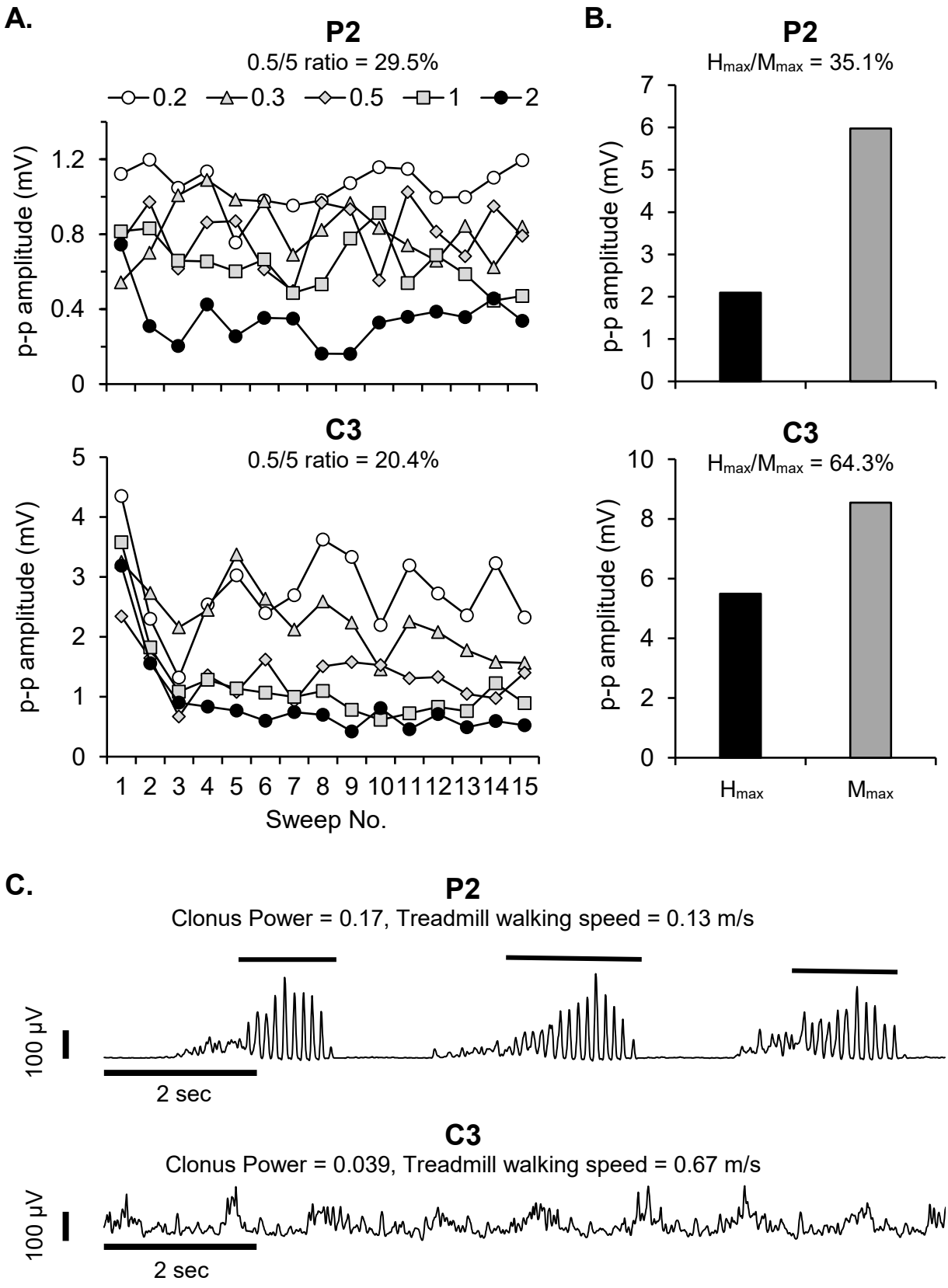
A.
P2
0.5/5 ratio = 29.5%
0.2
0.3
0.5
1
2
p-p amplitude (mV)
C3
0.5/5 ratio = 20.4%
Sweep No.
B.
P2
Hmax/Mmax = 35.1%
C3
Hmax/Mmax = 64.3%
Hmax
Mmax
C.
P2
Clonus Power = 0.17, Treadmill walking speed = 0.13 m/s
100 µV
2 sec
C3
Clonus Power = 0.039, Treadmill walking speed = 0.67 m/s

Figure 4.1: Reflexes from single participants.

Reflexes are shown for a participant with spinal cord injury (P2) and without injury (i.e., control) (C3). (a) The rate-dependent depression (RDD) of the peak-to-peak (p-p) amplitude of the H-reflex from the soleus muscle (SOL) at 0.2, 0.3, 0.5, 1 and 2 Hz stimulation of the posterior tibial nerve behind the knee (pulse width = 1 ms, stimulus intensity = 50% H_{max}), across 15 stimuli. RDD was quantified at 2 Hz using the 0.5/5 ratio (see text for rationale and details). (b) The p-p amplitudes of the maximum H-reflex (H_{max}) and M-response (M_{max}), recorded during the H- and M-recruitment curves, were used to estimate the H_{max}/M_{max} from the SOL. (c) The rectified and smoothed (20 Hz low-pass, 2nd order, dual pass, digital Butterworth filtered) SOL electromyogram (EMG) trace (down sampled by 20 data points for clarity). Clonus, estimated using the SOL EMG signal power within the clonus frequency range (see text for details), is observed as rhythmic bursts at the end of the regular SOL EMG trace in P2, indicated using horizontal lines above the EMG. RDD and H_{max}/M_{max} were measured during quiet sitting, while clonus was measured during undisturbed walking on a level treadmill. Clonus Power and speed of walking of each participant is indicated above the respective EMG trace.

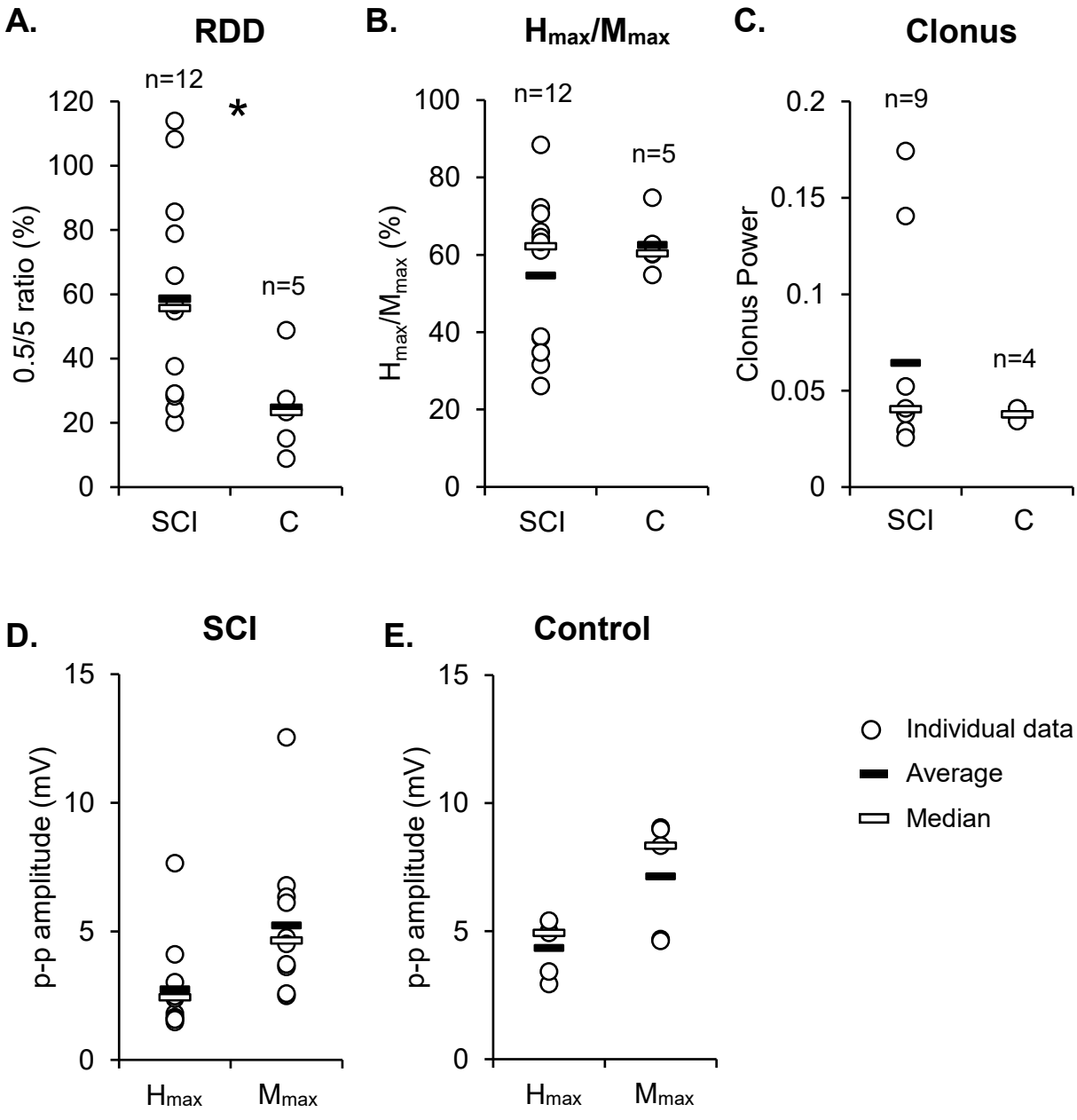

Figure 4.2: Differences in the excitability of spinal reflexes.

Data from single participants (unfilled circles), and as group average (filled horizontal line) and median (unfilled horizontal line) for the (a) rate-dependent depression of the H-reflex at 2 Hz (0.5/5 ratio), (b) H_{max}/M_{max} and (c) clonus (Clonus Power), are shown for comparison between participants in the spinal cord injury (SCI) and control (C) groups, along with the H_{max} and M_{max} values for the (d) SCI and (e) Control groups. Higher numbers indicate greater reflex excitability. * $p<0.05$

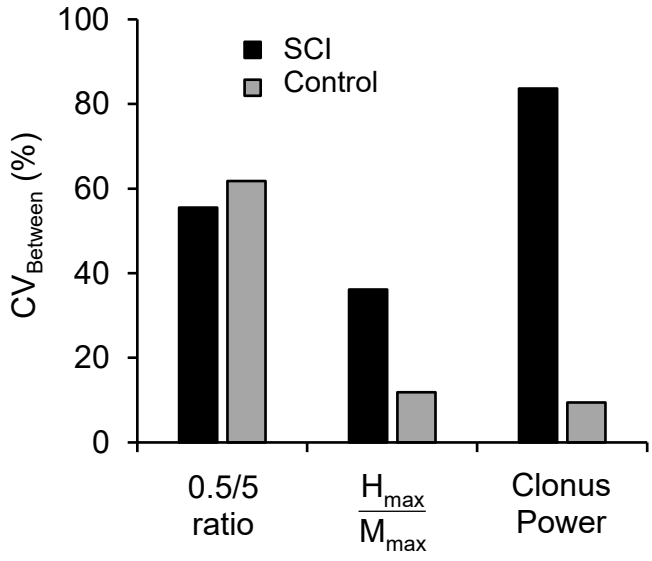

Figure 4.3: Variability of spinal reflexes.

Between-group coefficient of variation ($CV_{Between}$) of the rate-dependent depression of the H-reflex at 2 Hz (0.5/5 ratio), H_{max}/M_{max} and clonus (Clonus Power) was determined using the average and standard deviation of the individual data presented in Figure 4.2, in order to estimate the spread of the data in the spinal cord injury (SCI) and control groups.

4.3.3 Variability of reflexes

Day-to-day differences in the reflexes within a participant in the SCI and control groups are illustrated in the left and right panels of Figure 4.4, respectively. In the SCI group, all three reflexes exhibited variable excitability across days in some participants. In contrast, the reflexes were stable across days in the control participants. Using the mean and SD of the individual data from each SCI participant, the CV_{Within} was determined for each participant (Figure 4.5). In the SCI group, within-participant variability (i.e., CV_{Within}) was highest for RDD, followed by clonus, and then the H_{max}/M_{max} (Figure 4.5). As shown in Figure 4.4, the differences in the day-to-day variability of the reflexes in both groups of participants were not due to differences in the stimulating and recording conditions between days, as estimated using the CV_{Mmax}. The CV_{Mmax} was low across days in participants with SCI (8.9 ± 4.7%) and the control participants (11.2 ± 3.2%). Thus, the within-participant variability observed in the SCI group likely reflects real differences in reflex excitability across days.

Finally, in order to estimate the changes in the reliability of a reflex response with greater number of measures of the reflex across days, the CV_{Within} was determined separately across 2 days (Day 1 + Day 2, Day 1 + Day 3, Day 2 + Day 3) and across 3 days (Day 1 + Day 2 + Day 3) within the same SCI participant, and then averaged across participants. This analysis was performed to determine whether increasing the number of measures of the same reflex affects the within-participant variability of the reflex, and included data from SCI participants with ≥ 3 measures of RDD (n=3), H_{max}/M_{max} (n=5) and clonus (n=5). Across all three reflexes, the CV_{Within} across any of the two days did not appear to be different compared to that across all three days, based on the criteria that the CV_{Within} across all three days was between the range of CV_{Within} measured across any of the two days (statistics not performed due to small sample size): RDD, 51.1%, 40.6%, 14.3% vs 35.1%; H_{max}/M_{max}, 23.2%, 13.4%, 14.7% vs 18.2%; clonus, 20.0%, 24.6%, 31.1% vs 29.5%.

4.3.4 Relationship between reflexes

No significant correlations were observed between any of the reflex measures in the SCI participants, however, a trend was observed between higher H_{max}/M_{max} and reduced RDD (Figure 4.6). Interestingly, in Figure 4.2, from the 5 SCI participants with high RDD (i.e., 0.5/5

ratio < 40% in P1, P2, P3, P4, P5), 3 participants (P2, P3, P4) also had low H_{max}/M_{max}, with only 1 of these 3 participants (P3) taking Baclofen to control his spasticity.

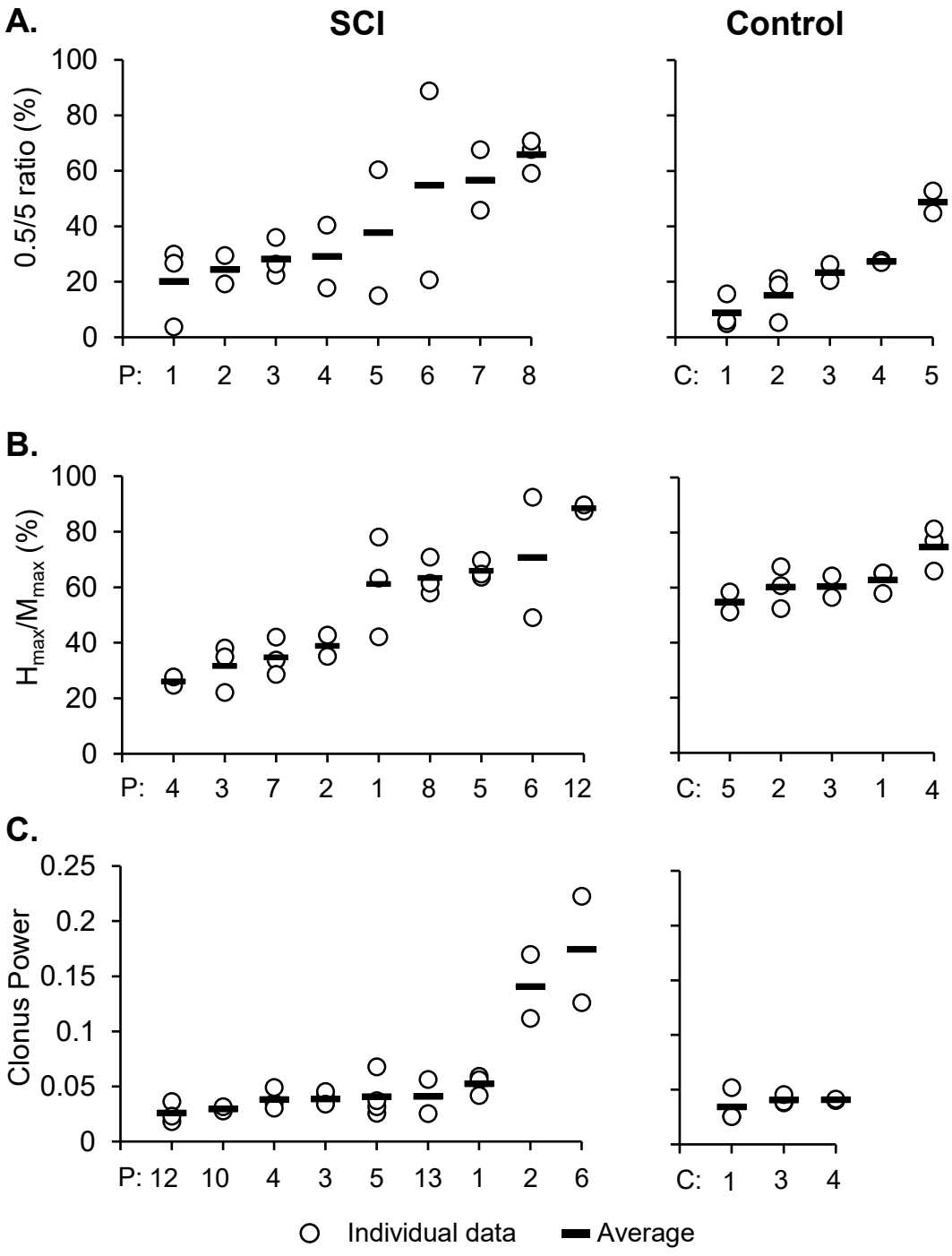
A.
SCI
Control
0.5/5 ratio (%)
100
80
60
40
20
0
P: 1 2 3 4 5 6 7 8
C: 1 2 3 4 5
B.
H_{max}/M_{max} (%)
100
80
60
40
20
0
P: 4 3 7 2 1 8 5 6 12
C: 5 2 3 1 4
C.
Clonus Power
0.25
0.2
0.15
0.1
0.05
0
P: 12 10 4 3 5 13 1 2 6
C: 1 3 4
Individual data
Average

Figure 4.4: Day-to-day differences in spinal reflexes.

Reflex measures on a single day (circles) and as averages across days (filled horizontal line) are shown for participants in the spinal cord injury (SCI) and control groups, illustrating the day-to-day variability of the (a) rate-dependent depression of the H-reflex at 2 Hz (0.5/5 ratio), (b) H_{max}/M_{max} and (c) clonus (Clonus Power). Participants are numbered at the bottom of each plot as listed in Tables 4.1 and 4.2. Data is shown for participants with ≥ 2 reflex measures: see Table 4.3 for details.

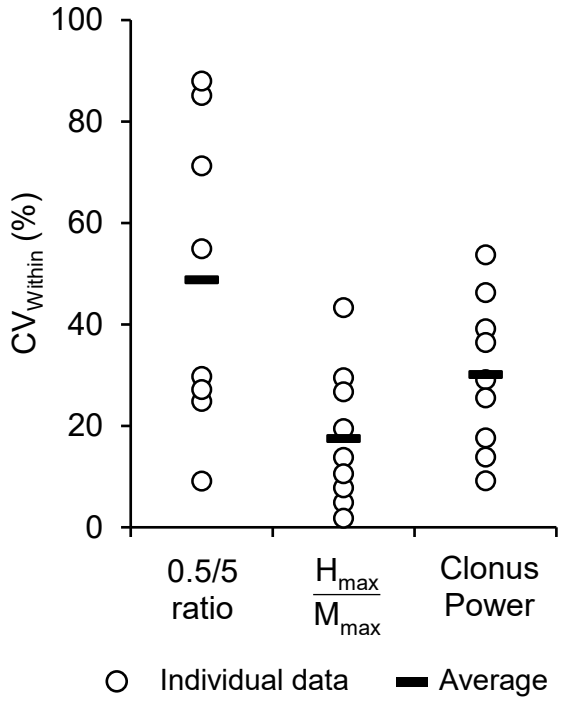

Figure 4.5: Within-participant variability of spinal reflexes.

Within-participant coefficient of variation (CV_{Within}) of the rate-dependent depression of the H-reflex at 2 Hz (0.5/5 ratio), H_{max}/M_{max} and clonus (Clonus Power) measured across 2-4 days, from single participants (open circles) and as averages across participants (filled horizontal line), in the spinal cord injury group. CV_{Within} was determined using the average and standard deviation of the individual data for each spinal cord injured participant in Figure 4.4.

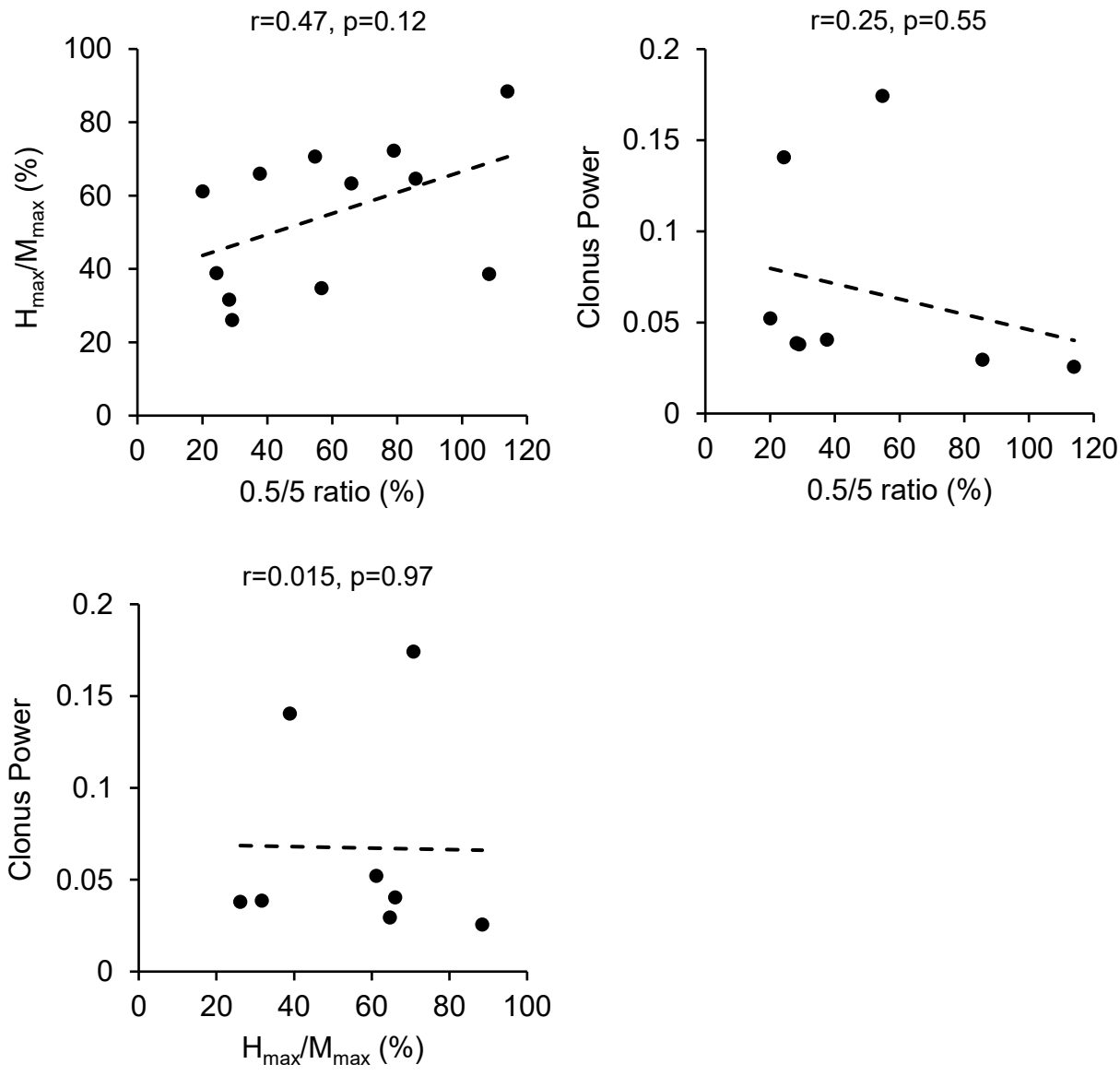

Figure 4.6: Relationship between spinal reflexes.

Correlation between the rate-dependent depression of the H-reflex at 2 Hz (0.5/5 ratio), H_{max}/M_{max} and clonus (Clonus Power) in the spinal cord injury group. Each dot represents either a single measure or an average of multiple measures, within a participant - see Table 4.3 for details. Pearson's correlation coefficient (r) and significance (p) are indicated above each plot.

4.4 Discussion

The main findings of this study are that the three reflexes that involve the Ia–MN pathway: RDD, H_{max}/M_{max} and clonus, showed differences in day-to-day variability within, and consistency across SCI participants.

4.4.1 Methodological consideration

Variability of the reflex measures between days in the SCI participants is unlikely a result of differences in the repeatability of the experimental procedure, because the same reflexes were stable across days in the control participants. Furthermore, the stimulating and recording conditions across days were controlled by maintaining a consistent M_{max} across days within each participant, estimated using the coefficient of variation of the M_{max}. Spasticity is known to vary considerably day-to-day and within a day in people with SCI (Little et al. 1989). We did not control for the time of day of the experiment, which may have affected the variability (Little et al. 1989; Mahoney et al. 2007). Lastly, approximately half of the participants in the SCI group were taking anti-spastic medication, which could also have affected the variability of the reflexes.

Changes in musculoskeletal properties, i.e., peripheral factors, after SCI (Olsson et al. 2006) can also affect reflex excitability, and were not considered in this study. For example, increase in stiffness of the ankle joint after SCI (Lorentzen et al. 2012; Mirbagheri et al. 2001) is associated with increase in clonus (de Vlugt et al. 2012). However, such changes take place over a longer time period [e.g., 44 weeks, (Schindler-Ivens and Shields 2000)]. It is unlikely that such changes could have taken place across the maximum time interval between reflex recordings (3.4 weeks) in our group of SCI participants and, therefore, to have affected the excitability of the spinal reflexes.

4.4.2 Variability of reflexes: importance of multiple measures

Within-participant variability of RDD was higher, compared to that for clonus and H_{max}/M_{max} (Figure 4.5). Furthermore, the within-participant variability of each of the three reflexes did not vary when compared across two days (i.e., Day 1 + Day 2, Day 1 + Day 3, Day 2 + Day 3) versus three days (i.e., Day 1 + Day 2 + Day 3), suggesting that the reflexes are stable by the

second measure. However, due to the small sample size of the participants with ≥ 3 measures of RDD, H_{max}/M_{max} and clonus, this finding needs to be confirmed with a larger sample size and statistics. Regardless of the mechanism behind the day-to-day differences in the reflex measures, it is therefore recommended that reflexes be measured at least two times on different days. Doing so will establish the reliability of the reflex response before interventions are undertaken to address changes in spasticity as a result of the interventions, in people with SCI.

4.4.3 Relationship between reflexes

4.4.3.1 Different Ia-MN reflexes reflect different neuronal mechanisms

It is likely that different interneuronal pathways affect the excitability of the H_{max}/M_{max}, RDD and clonus. The H_{max}/M_{max} estimates the monosynaptic recruitment of MNs from the activation of Ia afferents at increasing strength of stimulation of a peripheral nerve (Magladery et al. 1951). However, given that the p-p amplitude of the H-reflex was analyzed a few milliseconds after the first deflection of the H-reflex in the EMG waveform, it is possible that the H_{max}/M_{max} ratio measured in the current study may also involve polysynaptic pathways. In contrast, transmission along the Ia-MN pathway is dampened when the Ia afferents are stimulated at high frequencies (Schindler-Ivens and Shields 2000), leading to RDD (Bos et al. 2013; Boulenguez et al. 2010; Schindler-Ivens and Shields 2000). The source of the inhibition is unclear and could include: post-activation depression of the Ia terminals (Hultborn et al. 1996), presynaptic inhibition of the Ia terminals (Kakinohana et al. 2012; Kapitza et al. 2012), and inhibition within polysynaptic interneuronal pathways (Bos et al. 2013; Butler et al. 2006), all of which are affected after SCI (Faist et al. 1994; Jones and Yang 1994; Nielsen et al. 1993b). Since the depression of the H-reflex during RDD is frequency-dependent and observed after multiple repetitive stimulations, it is possible that putative polysynaptic inhibitory pathways are being recruited throughout the consecutive multiple stimuli.

Interestingly, clonus did not correlate with the H_{max}/M_{max}, despite the presumed role of the Ia-MN stretch reflex pathway in the generation of clonus (Hidler and Rymer 1999; Rossi et al. 1990; Wallace et al. 2012) and the H_{max}/M_{max} (Magladery et al. 1951). Since clonus did not correlate with the H_{max}/M_{max}, it is therefore likely that clonus may arise from a central mechanism, e.g., an intrinsic spinal circuitry, similar to a central pattern generator for walking

(Dimitrijevic et al. 1998; Gerasimenko et al. 2010; Vilensky and O'Connor 1998), that produces the rhythmic, clonic muscle contractions, which are unaffected by peripheral factors (Beres-Jones et al. 2003; Dimitrijevic et al. 1980a; Walsh 1976).

Maladaptive neuroplasticity within spinal pathways after SCI is implicated in the hyperexcitability of these reflexes: reduced presynaptic inhibition of Ia afferents (Faist et al. 1994), reduced reciprocal inhibition (Boorman et al. 1996), increased reciprocal facilitation (Crone et al. 2003), reduced polysynaptic inhibition (Jones and Yang 1994; Norton et al. 2008), and increased excitability of the MN (Boulenguez et al. 2010; Gorassini et al. 2004; Murray et al. 2010). However, some inhibitory pathways may be affected more so than others in different individuals with SCI, manifesting as an increase in excitability of some reflexes but not others, as reported here. Based on these findings, it is therefore necessary to have a better understanding of the underlying mechanisms behind spinal reflexes, in order to determine how they are affected in people with SCI.

4.4.3.2 Task-dependent differences in reflex excitability

There is considerable evidence that movement changes reflex excitability (Stein and Thompson 2006). Task-dependent changes in the H-reflex and clonus may have produced the differences in the excitability of these reflexes during sitting at rest and walking on a treadmill, respectively. For example, the H_{max}/M_{max} in the SOL increases as the background contraction increases, and decreases with further increase in background activity (Stein et al. 2007). Similarly, RDD is reduced during background contraction in sitting, and becomes negligible in standing (Stein et al. 2007). H-reflexes also show changes during functional movements, e.g., standing vs walking, and walking vs running. Excitability of the H-reflex in the SOL is reduced during walking compared to standing while matching for background EMG under the two tasks (Capaday and Stein 1986). The excitability of the H-reflex is further reduced during running compared to walking, when compared at similar levels of background EMG (Capaday and Stein 1987). Future studies should compare the excitability of the H_{max}/M_{max} and RDD in sitting with clonus when it is also measured in sitting, e.g., using the foot drop test (Manella and Field-Fote 2013; Wallace et al. 2012), in order to rule out the effects of walking on the excitability of the pathways that govern clonus.

4.5 Conclusion

Three spinal reflexes, all of which involve the Ia-MN pathway, displayed differences in their day-to-day variability within participants, and consistency across participants after SCI. Therefore, we recommend that, prior to any intervention that addresses hyperexcitable reflexes, these reflexes be measured at least two times across several days, until a stable baseline is reached. Doing so will allow for a reliable assessment of the change in the reflex response after the intervention.

4.6 Bibliography for Chapter 4

Achache V, Roche N, Lamy JC, Boakye M, Lackmy A, Gastal A, Quentin V, and Katz R. Transmission within several spinal pathways in adults with cerebral palsy. *Brain : a journal of neurology* 133: 1470-1483, 2010.

Adams MM, and Hicks AL. Comparison of the effects of body-weight-supported treadmill training and tilt-table standing on spasticity in individuals with chronic spinal cord injury. *The journal of spinal cord medicine* 34: 488-494, 2011.

Al-Khodairy AT, Gobelet C, and Rossier AB. Has botulinum toxin type A a place in the treatment of spasticity in spinal cord injury patients? *Spinal cord* 36: 854-858, 1998.

Beres-Jones JA, Johnson TD, and Harkema SJ. Clonus after human spinal cord injury cannot be attributed solely to recurrent muscle-tendon stretch. *Experimental brain research* 149: 222-236, 2003.

Boorman GI, Lee RG, Becker WJ, and Windhorst UR. Impaired "natural reciprocal inhibition" in patients with spasticity due to incomplete spinal cord injury. *Electroencephalography and clinical neurophysiology* 101: 84-92, 1996.

Bos R, Sadlaoud K, Boulenguez P, Buttigieg D, Liabeuf S, Brocard C, Haase G, Bras H, and Vinay L. Activation of 5-HT2A receptors upregulates the function of the neuronal K-Cl cotransporter KCC2. *Proceedings of the National Academy of Sciences of the United States of America* 110: 348-353, 2013.

Boulenguez P, Liabeuf S, Bos R, Bras H, Jean-Xavier C, Brocard C, Stil A, Darbon P, Cattaert D, Delpire E, Marsala M, and Vinay L. Down-regulation of the potassium-chloride cotransporter KCC2 contributes to spasticity after spinal cord injury. *Nature medicine* 16: 302-307, 2010.

Butler JE, Godfrey S, and Thomas CK. Depression of involuntary activity in muscles paralyzed by spinal cord injury. *Muscle & nerve* 33: 637-644, 2006.

Capaday C, and Stein RB. Amplitude modulation of the soleus H-reflex in the human during walking and standing. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 6: 1308-1313, 1986.

Capaday C, and Stein RB. Difference in the amplitude of the human soleus H reflex during walking and running. *The Journal of physiology* 392: 513-522, 1987.

Corcos DM, Gottlieb GL, Penn RD, Myklebust B, and Agarwal GC. Movement deficits caused by hyperexcitable stretch reflexes in spastic humans. *Brain : a journal of neurology* 109 (Pt 5): 1043-1058, 1986.

Crone C, Johnsen LL, Biering-Sorensen F, and Nielsen JB. Appearance of reciprocal facilitation of ankle extensors from ankle flexors in patients with stroke or spinal cord injury. *Brain : a journal of neurology* 126: 495-507, 2003.

D'Amico JM, Condliffe EG, Martins KJ, Bennett DJ, and Gorassini MA. Recovery of neuronal and network excitability after spinal cord injury and implications for spasticity. *Frontiers in integrative neuroscience* 8: 36, 2014.

D'Amico JM, Li Y, Bennett DJ, and Gorassini MA. Reduction of spinal sensory transmission by facilitation of 5-HT1B/D receptors in noninjured and spinal cord-injured humans. *Journal of neurophysiology* 109: 1485-1493, 2013.

de Vlugt E, de Groot JH, Wisman WH, and Meskers CG. Clonus is explained from increased reflex gain and enlarged tissue viscoelasticity. *Journal of biomechanics* 45: 148-155, 2012.

Dietz V. Spastic movement disorder: what is the impact of research on clinical practice? *Journal of neurology, neurosurgery, and psychiatry* 74: 820-821, 2003.

Dimitrijevic M, Nathan P, and Sherwood A. Clonus: the role of central mechanisms. *Journal of Neurology, Neurosurgery & Psychiatry* 43: 321-332, 1980a.

Dimitrijevic MR, Gerasimenko Y, and Pinter MM. Evidence for a spinal central pattern generator in humans. *Annals of the New York Academy of Sciences* 860: 360-376, 1998.

Elbasiouny SM, Moroz D, Bakr MM, and Mushahwar VK. Management of spasticity after spinal cord injury: current techniques and future directions. *Neurorehabilitation and neural repair* 24: 23-33, 2010.

Eriksson J, Olausson B, and Jankowska E. Antispastic effects of L-dopa. *Experimental brain research* 111: 296-304, 1996.

Faist M, Mazevet D, Dietz V, and Pierrot-Deseilligny E. A quantitative assessment of presynaptic inhibition of Ia afferents in spastics. Differences in hemiplegics and paraplegics. *Brain : a journal of neurology* 117 (Pt 6): 1449-1455, 1994.

Fried GW, and Fried KM. Spinal cord injury and use of botulinum toxin in reducing spasticity. *Physical medicine and rehabilitation clinics of North America* 14: 901-910, 2003.

Gerasimenko Y, Gorodnichev R, Machueva E, Pivovarova E, Semyenov D, Savochin A, Roy RR, and Edgerton VR. Novel and direct access to the human locomotor spinal circuitry. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 30: 3700-3708, 2010.

Glass A, and Hannah A. A comparison of dantrolene sodium and diazepam in the treatment of spasticity. *Paraplegia* 12: 170-174, 1974.

Gorassini MA, Knash ME, Harvey PJ, Bennett DJ, and Yang JF. Role of motoneurons in the generation of muscle spasms after spinal cord injury. *Brain : a journal of neurology* 127: 2247-2258, 2004.

Gorassini MA, Norton JA, Nevett-Duchcherer J, Roy FD, and Yang JF. Changes in locomotor muscle activity after treadmill training in subjects with incomplete spinal cord injury. *Journal of neurophysiology* 101: 969-979, 2009.

Gruenthal M, Mueller M, Olson WL, Priebe MM, Sherwood AM, and Olson WH. Gabapentin for the treatment of spasticity in patients with spinal cord injury. *Spinal cord* 35: 686-689, 1997.

Gunduz S, Kalyon TA, Dursun H, Mohur H, and Bilgic F. Peripheral nerve block with phenol to treat spasticity in spinal cord injured patients. *Paraplegia* 30: 808-811, 1992.

Hidler JM, and Rymer WZ. A simulation study of reflex instability in spasticity: origins of clonus. *IEEE transactions on rehabilitation engineering : a publication of the IEEE Engineering in Medicine and Biology Society* 7: 327-340, 1999.

Hubli M, Dietz V, and Bolliger M. Spinal reflex activity: a marker for neuronal functionality after spinal cord injury. *Neurorehabilitation and neural repair* 26: 188-196, 2012.

Hultborn H, Illert M, Nielsen J, Paul A, Ballegaard M, and Wiese H. On the mechanism of the post-activation depression of the H-reflex in human subjects. *Experimental brain research* 108: 450-462, 1996.

Ishikawa K, Ott K, Porter RW, and Stuart D. Low frequency depression of the H wave in normal and spinal man. *Experimental neurology* 15: 140-156, 1966.

Johnson RL, Gerhart KA, McCray J, Menconi JC, and Whiteneck GG. Secondary conditions following spinal cord injury in a population-based sample. *Spinal cord* 36: 45-50, 1998.

Jones CA, and Yang JF. Reflex behavior during walking in incomplete spinal-cord-injured subjects. *Experimental neurology* 128: 239-248, 1994.

Kakinohana O, Hefferan MP, Miyanohara A, Nejime T, Marsala S, Juhas S, Juhasova J, Motlik J, Kucharova K, Strnadel J, Platoshyn O, Lazar P, Galik J, Vinay L, and Marsala M. Combinational spinal GAD65 gene delivery and systemic GABA-mimetic treatment for modulation of spasticity. *PloS one* 7: e30561, 2012.

Kapitza S, Zorner B, Weinmann O, Bolliger M, Filli L, Dietz V, and Schwab ME. Tail spasms in rat spinal cord injury: changes in interneuronal connectivity. *Experimental neurology* 236: 179-189, 2012.

Khan AS, Patrick SK, Roy FD, Gorassini MA, and Yang JF. Training-Specific Neural Plasticity in Spinal Reflexes after Incomplete Spinal Cord Injury. *Neural Plast* 2016: 6718763, 2016.

Kiser TS, Reese NB, Maresh T, Hearn S, Yates C, Skinner RD, Pait TG, and Garcia-Rill E. Use of a motorized bicycle exercise trainer to normalize frequency-dependent habituation of the H-reflex in spinal cord injury. *The journal of spinal cord medicine* 28: 241-245, 2005.

Knikou M. Functional reorganization of soleus H-reflex modulation during stepping after robotic-assisted step training in people with complete and incomplete spinal cord injury. *Experimental brain research* 228: 279-296, 2013.

Krawetz P, and Nance P. Gait analysis of spinal cord injured subjects: effects of injury level and spasticity. *Archives of physical medicine and rehabilitation* 77: 635-638, 1996.

Lazorthes Y, Sallerin-Caute B, Verdie JC, Bastide R, and Carillo JP. Chronic intrathecal baclofen administration for control of severe spasticity. *Journal of neurosurgery* 72: 393-402, 1990.

Little JW, Micklesen P, Umlauf R, and Britell C. Lower extremity manifestations of spasticity in chronic spinal cord injury. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 68: 32-36, 1989.

Lorentzen J, Grey MJ, Geertsen SS, Biering-Sorensen F, Brunton K, Gorassini M, and Nielsen JB. Assessment of a portable device for the quantitative measurement of ankle joint stiffness in spastic individuals. *Clin Neurophysiol* 123: 1371-1382, 2012.

Magladery JW, Porter WE, Park AM, and Teasdall RD. Electrophysiological studies of nerve and reflex activity in normal man. IV. The two-neurone reflex and identification of certain action potentials from spinal roots and cord. *Bulletin of the Johns Hopkins Hospital* 88: 499-519, 1951.

Mahoney JS, Engebretson JC, Cook KF, Hart KA, Robinson-Whelen S, and Sherwood AM. Spasticity Experience Domains in Persons With Spinal Cord Injury. *Archives of physical medicine and rehabilitation* 88: 287-294, 2007.

Manella KJ, and Field-Fote EC. Modulatory effects of locomotor training on extensor spasticity in individuals with motor-incomplete spinal cord injury. *Restorative neurology and neuroscience* 31: 633-646, 2013.

Mirbagheri MM, Barbeau H, Ladouceur M, and Kearney RE. Intrinsic and reflex stiffness in normal and spastic, spinal cord injured subjects. *Experimental brain research* 141: 446-459, 2001.

Mirbagheri MM, Chen D, and Rymer WZ. Quantification of the effects of an alpha-2 adrenergic agonist on reflex properties in spinal cord injury using a system identification technique. *Journal of neuroengineering and rehabilitation* 7: 29, 2010.

Mirbagheri MM, Ladouceur M, Barbeau H, and Kearney RE. The effects of long-term FES-assisted walking on intrinsic and reflex dynamic stiffness in spastic spinal-cord-injured subjects. *Neural Systems and Rehabilitation Engineering, IEEE Transactions on* 10: 280-289, 2002.

Murray KC, Nakae A, Stephens MJ, Rank M, D'Amico J, Harvey PJ, Li X, Harris RLW, Ballou EW, Anelli R, Heckman CJ, Mashimo T, Vavrek R, Sanelli L, Gorassini MA, Bennett DJ, and Fouad K. Recovery of motoneuron and locomotor function after spinal cord injury depends on constitutive activity in 5-HT(2C) receptors. *Nature medicine* 16: 694-700, 2010.

Nance PW. A comparison of clonidine, cyproheptadine and baclofen in spastic spinal cord injured patients. *The Journal of the American Paraplegia Society* 17: 150-156, 1994.

Nielsen J, Petersen N, Ballegaard M, Biering-Sorensen F, and Kiehn O. H-reflexes are less depressed following muscle stretch in spastic spinal cord injured patients than in healthy subjects. *Experimental brain research* 97: 173-176, 1993b.

Norton JA, Bennett DJ, Knash ME, Murray KC, and Gorassini MA. Changes in sensory-evoked synaptic activation of motoneurons after spinal cord injury in man. *Brain : a journal of neurology* 131: 1478-1491, 2008.

Olsson MC, Kruger M, Meyer LH, Ahnlund L, Gransberg L, Linke WA, and Larsson L. Fibre type-specific increase in passive muscle tension in spinal cord-injured subjects with spasticity. *The Journal of physiology* 577: 339-352, 2006.

Panizza M, Nilsson J, and Hallett M. Optimal stimulus duration for the H reflex. *Muscle & nerve* 12: 576-579, 1989.

Pinter MM, Gerstenbrand F, and Dimitrijevic MR. Epidural electrical stimulation of posterior structures of the human lumbosacral cord: 3. Control Of spasticity. *Spinal cord* 38: 524-531, 2000.

Reyes R, Icarangal A, and Bertellotti G. Part 1: The Good, The Bad, and The Not-So-Ugly: Medical and Pharmacological Treatments for Spasticity http://sci.washington.edu/spasticity/index.asp.

Reynolds RM, Morton RP, Walker ML, Massagli TL, and Browd SR. Role of dorsal rhizotomy in spinal cord injury-induced spasticity. *Journal of neurosurgery Pediatrics* 14: 266-270, 2014.

Rossi A, Mazzocchio R, and Scarpini C. Clonus in man: a rhythmic oscillation maintained by a reflex mechanism. *Electroencephalography and clinical neurophysiology* 75: 56-63, 1990.

Roy RR, and Edgerton VR. Neurobiological perspective of spasticity as occurs after a spinal cord injury. *Experimental neurology* 235: 116-122, 2012.

Schindler-Ivens S, and Shields RK. Low frequency depression of H-reflexes in humans with acute and chronic spinal-cord injury. *Experimental brain research* 133: 233-241, 2000.

Skold C, Levi R, and Seiger A. Spasticity after traumatic spinal cord injury: nature, severity, and location. *Archives of physical medicine and rehabilitation* 80: 1548-1557, 1999.

Smith AC, Mummidisetty CK, Rymer WZ, and Knikou M. Locomotor training alters the behavior of flexor reflexes during walking in human spinal cord injury. *Journal of neurophysiology* 112: 2164-2175, 2014.

Stein RB, Estabrooks KL, McGie S, Roth MJ, and Jones KE. Quantifying the effects of voluntary contraction and inter-stimulus interval on the human soleus H-reflex. *Experimental brain research* 182: 309-319, 2007.

Stein RB, and Thompson AK. Muscle reflexes in motion: how, what, and why? *Exercise and sport sciences reviews* 34: 145-153, 2006.

Vilensky JA, and O'Connor BL. Stepping in nonhuman primates with a complete spinal cord transection: old and new data, and implications for humans. *Annals of the New York Academy of Sciences* 860: 528-530, 1998.

Wainberg M, Barbeau H, and Gauthier S. The effects of cyproheptadine on locomotion and on spasticity in patients with spinal cord injuries. *Journal of neurology, neurosurgery, and psychiatry* 53: 754-763, 1990.

Wallace DM, Ross BH, and Thomas CK. Characteristics of lower extremity clonus after human cervical spinal cord injury. *Journal of neurotrauma* 29: 915-924, 2012.

Walsh EG. Clonus: beats provoked by the application of a rhythmic force. *Journal of neurology, neurosurgery, and psychiatry* 39: 266-274, 1976.

CHAPTER 5: GENERAL DISCUSSION

5.1 Thesis summary

This thesis examined the changes in neuroplasticity and functional capabilities before and after walking retraining in people with chronic, complete and incomplete spinal cord injury (SCI). Chapters 2 and 3 compared the changes in spinal reflexes, walking function and other functional capabilities after walking retraining, while Chapter 4 examined the changes in spinal reflexes before training. A brief summary of each study is provided in the following.

In Chapter 2, we showed that mono- and polysynaptic spinal reflexes respond in a training-specific manner, and that the excitability of the reflexes before and the change in the excitability after training are related to walking function. In Chapter 3, we showed that individuals with severe SCI, particularly those with motor complete injuries, can learn to walk over ground in the ReWalk powered exoskeleton, while those who could walk before the training further improved their walking ability without the ReWalk after the training. Finally, in Chapter 4, we showed that spinal reflexes that involve the Ia-motoneuronal (Ia-MN) pathway: rate-dependent depression (RDD) of the Hoffmann (H-) reflex, H_{max}/M_{max} ratio and clonus exhibit differences in the test-retest reliability across days, and varied independently of each other, after chronic, motor-incomplete SCI.

5.2 Individuals with SCI respond differently to walking retraining

5.2.1 Injury related factors

Clinical manifestations of SCI are heterogeneous across individuals after the injury. Within each grade of the American Spinal Injury Association Impairment Scale, the manifestations vary depending on the mechanism and the level of injury, which lead to a variety of syndromes (see Burns et al. (2012) and Miele et al. (2012) for review). Mechanisms of injury that result in SCI include hyperflexion, hyperextension, rotation, dislocation and compression (Miele et al. 2012), and each can lead to a variety of syndromes, including central cord syndrome, Brown-Sequard syndrome and anterior cord syndrome. Each syndrome produces characteristic deficits that reflect damage to specific spinal pathways, resulting in a wide range of deficits within the motor and sensory domains: anterior cord syndrome results in impairments in pain and temperature,

and motor output; central cord syndrome leads to greater impairment in upper limbs, compared to lower limbs; and Brown-Sequard syndrome results in loss of proprioception and motor control on the ipsilateral side, and loss of pain and temperature on the contralateral side (Burns et al. 2012). With such a range of deficits, it is therefore well established that the motor recovery also varies considerably across individuals with SCI (see Scivoletto and Di Donna (2009) for review). In terms of the level of injury, higher neurological level of injury leads to greater loss of function compared to a lower level of injury. Cervical injuries affect arm, hand, trunk and leg function, while a lower level of injury would primarily affect leg function (see Burns et al. (2012) and Scivoletto and Di Donna (2009) for review).

5.2.2 Non-injury related factors

Factors other than the above injury related factors also affect walking ability. Better prognosis for walking is associated with younger age at the time of injury (Burns et al. 1997; Scivoletto et al. 2004a; Scivoletto et al. 2008), lower spasticity (Scivoletto et al. 2008), better balance (Scivoletto et al. 2008), presence of the crossed-adductor and patellar reflexes (Calancie et al. 2004) and the absence of deep plantar reflex (Ko et al. 1999) and the ability to differentiate between light touch and pinprick sensation (Oleson et al. 2005).

5.3 The same training method does not work for everyone

Optimizing the 'perfect' locomotor rehabilitation program for any individual with SCI, to yield the best possible outcome for functional walking, depends on many factors: form of training, dosage of training, time between onset of injury and admission into rehabilitation, muscle strength, initial walking speed, spasticity, time since injury, and physiologic functions such as bowel and bladder voiding, and is detailed in the following sections.

5.3.1 Form of training

The form of training can affect walking outcome, depending on the stepping movements that are being emphasized during the training. Walking training that emphasizes precise stepping movements affects the skilled aspect of walking, whereas training that maximizes the number of steps during body weight-supported treadmill training (BWSTT) with manual assistance affects walking endurance, speed and distance (Musselman et al. 2009; Yang et al. 2014). Furthermore,

walking training that emphasizes stepping movements when performed using different training methods: BWSTT with and without functional electrical stimulation (FES) to the common peroneal nerve, walking in the Lokomat, and walking overground with the FES, can also lead to differences in the improvements in walking speed and distance (Field-Fote and Roach 2011). Significant increase in walking speed was observed after all forms of training, except for the Lokomat training, as per the time effect, however, the time × training interaction was not significant (Field-Fote and Roach 2011), suggesting that the effects on the improvements in walking speed after all forms of training were the same (Field-Fote and Roach 2011; Manella and Field-Fote 2013). In contrast, distance improved after the FES-assisted walking training over ground and on the treadmill (i.e., time effect), and that the improvements in distance were significantly higher after the overground walking (i.e., time × training interaction) (Field-Fote and Roach 2011).

5.3.2 Dosage of training

Dosage of training also affects walking outcome. The number of steps taken during training to make skilled walking movements is lower compared to the number of steps taken during BWSTT, which emphasises walking speed and distance (Yang et al. 2014). In contrast, BWSTT with complete stepping assistance, for example, when provided using the Lokomat, is not limited by time and trainer fatigue (Colombo et al. 2000), and therefore is capable of producing greater number of steps compared to BWSTT with manual assistance . However, such form of training does not emphasis volitional activation of the leg muscles, and therefore may not always yield improvements in walking outcome (Field-Fote and Roach 2011; Knikou 2013), although other reports suggest otherwise (Hornby et al. 2005; Wirz et al. 2005).

5.3.3 Injury related factors

Various injury related factors that differ across individuals with SCI also affect the outcome of walking retraining. A total score of 30/80 or more on the Manual Muscle Test, measured bilaterally from eight muscles on each leg prior to training, predicted improvements in walking speed after BWSTT with manual assistance (Yang et al. 2011). Walking speed after BWSTT, overground walking and walking in the Lokomat in participants with acute and chronic SCI (≤ 5 years post injury) was also predicted by the initial over ground walking speed at a self-selected

pace, spasticity, voluntary bowl and bladder voiding and the square root of the time since injury, before the training (Winchester et al. 2009). However, individuals with complete injuries or those with incomplete injuries and insufficient strength in the lower extremities are more likely to benefit from walking in over ground powered exoskeletons, wherein greater number of training sessions, lower level of injury and higher age were associated with greater walking speed in the exoskeleton (Louie et al. 2015).

5.4 Spinal excitability affects walking ability

5.4.1 Role of spinal reflexes during walking

To better understand the significance of spinal reflexes and its role during walking, we need a better understanding of the mechanisms behind spinal reflexes after SCI. Spinal reflexes play a major role during walking, wherein reflexes from Ia, Ib and c fibers serve functional significance during the walking, in order to maintain balance and generate muscle activity (Zehr and Stein 1999). Ia afferents induce stretch reflexes during the stance phase of walking (Capaday and Stein 1987; Sinkjaer et al. 1996; Yang et al. 1991), thereby generating force necessary for the forward propulsion during the stance phase (Grey et al. 2008; Stein and Kearney 1995). Ib afferents increase the duration of the stance phase by activating the extensor muscles of the knee and ankle (Stephens and Yang 1999). Finally, cutaneous and cutaneomuscular fibers are involved in obstacle avoidance by inhibiting and activating the leg flexors and extensors, to allow the leg and foot to step over the obstacle during walking (Van Wezel et al. 1997; Zehr et al. 1997; Zehr et al. 1998).

5.4.2 Changes in spinal reflexes after walking retraining

Our understanding of the interaction between spinal reflexes and walking can be improved by comparing the changes in spinal reflexes from before to after walking retraining, and correlating that change with the change in walking outcomes. Walking retraining on the treadmill enhanced the RDD of the H-reflex in a single person with SCI (Trimble et al. 1998), whereas a single session of the training also reduced the H_{max}/M_{max} ratio (Trimble et al. 2001), with both studies reporting an improvement in walking speed at a self-selected and fast pace (Trimble et al. 2001; Trimble et al. 1998). BWSTT also reduced clonus, improved the walking speed and improved

the scores on the Walking Index for Spinal Cord Injury II (Gorassini et al. 2009). Furthermore, BWSTT with and without FES to the common peroneal nerve, walking in the Lokomat, and walking overground with the FES reduced the duration of extensor spasms and increased the threshold angle for eliciting clonus, which correlated with higher walking speed (Manella and Field-Fote 2013). BWSTT with and without FES and training in the Lokomat enhanced the early and reduced the late components of a cutaneomuscular reflex in the tibialis anterior muscle (TA), and improved the score on the Spinal Cord Independence Measure Version II (Hubli et al. 2012). However, the improvements in walking function are not always observed in conjunction with changes in spinal reflex excitability. Lokomat training changed the excitability of the monosynaptic H-reflex during walking (Knikou 2013) and improved the RDD of the H-reflex during sitting (Knikou and Mummidisetty 2014), without changing the overground walking ability (Knikou 2013). Furthermore, our understanding of spinal reflexes after SCI is confounded by the lack of repeatability and consistency between the different reflexes, suggesting that different interneuronal pathways govern the excitability of different spinal reflexes, e.g., RDD (Bos et al. 2013; Butler et al. 2006; Hultborn et al. 1996; Kakinohana et al. 2012; Kapitza et al. 2012), H_{max}/M_{max} (Magladery et al. 1951) and clonus (Beres-Jones et al. 2003; Dimitrijevic et al. 1980a; Hidler and Rymer 1999; Rossi et al. 1990; Wallace et al. 2012; Walsh 1976). Therefore, to better understand the relationship between reflexes and walking outcomes after physical training after SCI, 1) the reliability of the reflex measures must be established before the training, and 2) the changes in the reflexes must be compared after a single form of training, rather than a combination of training methods. Doing so will reduce the confounding effects of the variability of the reflex measure across participants, and that of other training methods.

5.4.3 Neuroplasticity within spinal pathways

Changes in the excitability of spinal reflexes reflect neuroplasticity within inhibitory and excitatory pathways. Training in the Lokomat enhanced the RDD of the H-reflex and strengthened the presynaptic inhibition (Knikou and Mummidisetty 2014), and Ia and Ib inhibition (Knikou et al. 2015). Skilled overground walking training increased the suppression of the TA motor evoked potential (MEP) when the transcranial magnetic stimulation (TMS) was conditioned by common peroneal nerve stimulation (Zewdie et al. 2015). In the same study, the excitability of a cutaneomuscular reflex in the TA, induced by posterior tibial nerve stimulation

at the ankle, was reduced after skilled over ground walking training and BWSTT with manual assistance (Zewdie et al. 2015). Similarly, Lokomat training induced a change in the conditioning effect of TMS over the TA motor cortex, on the sural nerve-induced cutaneous reflex, by switching the reflex response from excitation to inhibition (Hajela et al. 2013). Therefore, physical training after SCI results in an increase in spinal inhibition and as a result reduces the hyperexcitability of the spinal pathways, which is a common sequela of SCI.

5.4.4 Role of supraspinal input

As evident from the above discussion, spinal reflexes and neuroplasticity within spinal pathways are related to changes in walking ability after SCI, however the contribution of supraspinal pathways and peripheral factors to the improvements in walking cannot be ruled out. Two months of BWSTT with manual assistance enhanced the TMS-induced MEP_{max} in the TA and improved the walking speed and distance (Zewdie et al. 2015). Furthermore, BWSTT with manual assistance, in addition to higher MEP_{max}, also increased the slope of the MEP recruitment curve and the duration of the MEP silent period (Thomas and Gorassini 2005). The increase in the MEP_{max} correlated with increase in walking distance and score on the Walking Index for Spinal Cord Injury II (Thomas and Gorassini 2005). Strength of leg muscles before training (Yang et al. 2011) and improvements in postural stability after training (Yang et al. 2014) are also related to improvements in walking ability after the training (Yang et al. 2014; Yang et al. 2011). Therefore, physical training after SCI strengthens the supraspinal drive over the spinal cord, and may work in conjunction with the reduced hyperexcitability of the spinal cord, to improve the walking ability that is observed after the training.

5.5 Changes in walking ability: an interaction between spinal and supraspinal neuroplasticity and the locomotor CPG?

Neuroplasticity in the spinal cord, induced from walking retraining after SCI, results in changes in the excitability of spinal reflexes and improvements in walking function (Gorassini et al. 2009; Hubli et al. 2012; Manella and Field-Fote 2013; Trimble et al. 2001; Trimble et al. 1998). Therefore, it is possible that the spinal pathways generating these reflexes also interact with the pathways of a presumed locomotor central pattern generator (CPG) for walking (Gerasimenko et al. 2010; Jankowska et al. 1967). The modulation of spinal reflexes during walking illustrates

this interaction, wherein the amplitude of H-reflexes (Capaday and Stein 1987), stretch reflexes (Sinkjaer et al. 1996), cutaneous and cutaneomuscular reflexes (Van Wezel et al. 1997; Yang and Stein 1990; Zehr et al. 1997; Zehr et al. 1998) undergo a phase-dependent change in amplitude throughout the step cycle. Presumably, the changes in the amplitude of these reflexes reflect the changes in the balance between excitation and inhibition exerted by the CPG network over the flexor and extensor MNs (Gerasimenko et al. 2010; Jankowska et al. 1967).

Similarly, the changes in the excitability of spinal reflexes are also modulated by descending input, as it is well established that activating supraspinal pathways using voluntary contraction (Fournier et al. 1983) or transcranial magnetic stimulation (Iglesias et al. 2008; Valls-Sole et al. 1994) affects the excitability of various spinal pathways. These effects manifest as an increase or decrease in the excitability of inhibitory and excitatory spinal pathways such as presynaptic inhibition (Valls-Sole et al. 1994), Ib inhibition (Fournier et al. 1983) and heteronymous facilitation (Iglesias et al. 2008), and are observed at rest (i.e., while lying down) (Valls-Sole et al. 1994), during voluntary contraction (Fournier et al. 1983) and during walking (Iglesias et al. 2008). Furthermore, retraining walking strengthens supraspinal pathways and improves walking function (Thomas and Gorassini 2005; Zewdie et al. 2015), presumably by restoring the balance between excitation and inhibition within the locomotor CPG network (Gerasimenko et al. 2010), that is affected after SCI (Bussel et al. 1988; Calancie et al. 1994; Holmes 1915).

5.6 Future directions

To improve our understanding of the contribution of spinal reflexes (Zehr and Stein 1999) to the changes in walking ability resulting from walking retraining after SCI (Manella and Field-Fote 2013), we need to further determine the contribution of the different spinal reflexes, i.e., reflexes that are both monosynaptic (Capaday and Stein 1987; Yang et al. 1991) and polysynaptic (Stephens and Yang 1999; Yang and Stein 1990; Zehr et al. 1997; Zehr et al. 1998), and reflexes that originate from Ia (Capaday and Stein 1987; Yang et al. 1991), Ib (Stephens and Yang 1996) and c fibers (Zehr et al. 1997; Zehr et al. 1998), to the changes in walking ability after the training. Doing so will enable us to determine the contribution of the different reflexes and whether one reflex affects the changes in walking ability more so than others. Furthermore, since the changes in the excitability of spinal reflexes reflects neuroplasticity within excitatory and inhibitory pathways (Zewdie et al. 2015), therefore, future studies should compare changes in the

strength of excitatory and inhibitory pathways after the walking retraining (Hubli et al. 2012; Knikou and Mummidisetty 2014; Knikou et al. 2015; Zewdie et al. 2015). Doing so will improve our understanding of the contribution of spinal pathways towards the changes in walking ability, and enable us to ascribe these changes with specific neuroplastic changes within the spinal cord. Finally, the effects of supraspinal inputs on spinal reflexes (Gorassini et al. 2009; Thomas and Gorassini 2005; Zewdie et al. 2015) and its effects on the excitatory and inhibitory pathways require further understanding, in order to determine the contribution of both spinal and supraspinal neuroplasticity on the walking ability after SCI.

5.7 Bibliography for Chapter 5

Beres-Jones JA, Johnson TD, and Harkema SJ. Clonus after human spinal cord injury cannot be attributed solely to recurrent muscle-tendon stretch. *Experimental brain research* 149: 222-236, 2003.

Bos R, Sadlaoud K, Boulenguez P, Buttigieg D, Liabeuf S, Brocard C, Haase G, Bras H, and Vinay L. Activation of 5-HT2A receptors upregulates the function of the neuronal K-Cl cotransporter KCC2. *Proceedings of the National Academy of Sciences of the United States of America* 110: 348-353, 2013.

Burns AS, Marino RJ, Flanders AE, and Flett H. Clinical diagnosis and prognosis following spinal cord injury. *Handbook of clinical neurology* 109: 47-62, 2012.

Burns SP, Golding DG, Rolle WA, Jr., Graziani V, and Ditunno JF, Jr. Recovery of ambulation in motor-incomplete tetraplegia. *Archives of physical medicine and rehabilitation* 78: 1169-1172, 1997.

Bussel B, Roby-Brami A, Azouvi P, Biraben A, Yakovleff A, and Held JP. Myoclonus in a patient with spinal cord transection. Possible involvement of the spinal stepping generator. *Brain : a journal of neurology* 111 (Pt 5): 1235-1245, 1988.

Butler JE, Godfrey S, and Thomas CK. Depression of involuntary activity in muscles paralyzed by spinal cord injury. *Muscle & nerve* 33: 637-644, 2006.

Calancie B, Molano MR, and Broton JG. Tendon reflexes for predicting movement recovery after acute spinal cord injury in humans. *Clin Neurophysiol* 115: 2350-2363, 2004.

Calancie B, Needham-Shropshire B, Jacobs P, Willer K, Zych G, and Green BA. Involuntary stepping after chronic spinal cord injury. Evidence for a central rhythm generator for locomotion in man. *Brain : a journal of neurology* 117 (Pt 5): 1143-1159, 1994.

Capaday C, and Stein RB. Difference in the amplitude of the human soleus H reflex during walking and running. *The Journal of physiology* 392: 513-522, 1987.

Colombo G, Joerg M, Schreier R, and Dietz V. Treadmill training of paraplegic patients using a robotic orthosis. *Journal of rehabilitation research and development* 37: 693-700, 2000.

Dimitrijevic M, Nathan P, and Sherwood A. Clonus: the role of central mechanisms. *Journal of Neurology, Neurosurgery & Psychiatry* 43: 321-332, 1980a.

Field-Fote EC, and Roach KE. Influence of a locomotor training approach on walking speed and distance in people with chronic spinal cord injury: a randomized clinical trial. *Physical therapy* 91: 48-60, 2011.

Fournier E, Katz R, and Pierrot-Deseilligny E. Descending control of reflex pathways in the production of voluntary isolated movements in man. *Brain Res* 288: 375-377, 1983.

Gerasimenko Y, Gorodnichev R, Machueva E, Pivovarova E, Semyenov D, Savochin A, Roy RR, and Edgerton VR. Novel and direct access to the human locomotor spinal circuitry. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 30: 3700-3708, 2010.

Gorassini MA, Norton JA, Nevett-Duchcherer J, Roy FD, and Yang JF. Changes in locomotor muscle activity after treadmill training in subjects with incomplete spinal cord injury. *Journal of neurophysiology* 101: 969-979, 2009.

Grey MJ, Klinge K, Crone C, Lorentzen J, Biering-Sorensen F, Ravnborg M, and Nielsen JB. Post-activation depression of soleus stretch reflexes in healthy and spastic humans. *Experimental brain research* 185: 189-197, 2008.

Hajela N, Mummidisetty CK, Smith AC, and Knikou M. Corticospinal reorganization after locomotor training in a person with motor incomplete paraplegia. *BioMed research international* 2013: 516427, 2013.

Hidler JM, and Rymer WZ. A simulation study of reflex instability in spasticity: origins of clonus. *IEEE transactions on rehabilitation engineering : a publication of the IEEE Engineering in Medicine and Biology Society* 7: 327-340, 1999.

Holmes G. The goulstonian lectures on spinal injuries of warfare: delivered before the Royal College of Physicians of London. *British medical journal* 2: 815-821, 1915.

Hornby TG, Zemon DH, and Campbell D. Robotic-assisted, body-weight-supported treadmill training in individuals following motor incomplete spinal cord injury. *Physical therapy* 85: 52-66, 2005.

Hubli M, Dietz V, and Bolliger M. Spinal reflex activity: a marker for neuronal functionality after spinal cord injury. *Neurorehabilitation and neural repair* 26: 188-196, 2012.

Hultborn H, Illert M, Nielsen J, Paul A, Ballegaard M, and Wiese H. On the mechanism of the post-activation depression of the H-reflex in human subjects. *Experimental brain research* 108: 450-462, 1996.

Iglesias C, Nielsen JB, and Marchand-Pauvert V. Corticospinal inhibition of transmission in propriospinal-like neurones during human walking. *The European journal of neuroscience* 28: 1351-1361, 2008.

Jankowska E, Jukes MG, Lund S, and Lundberg A. The effect of DOPA on the spinal cord. 5. Reciprocal organization of pathways transmitting excitatory action to alpha motoneurones of flexors and extensors. *Acta physiologica Scandinavica* 70: 369-388, 1967.

Kakinohana O, Hefferan MP, Miyanohara A, Nejime T, Marsala S, Juhas S, Juhasova J, Motlik J, Kucharova K, Strnadel J, Platoshyn O, Lazar P, Galik J, Vinay L, and Marsala M. Combinational spinal GAD65 gene delivery and systemic GABA-mimetic treatment for modulation of spasticity. *PloS one* 7: e30561, 2012.

Kapitza S, Zorner B, Weinmann O, Bolliger M, Filli L, Dietz V, and Schwab ME. Tail spasms in rat spinal cord injury: changes in interneuronal connectivity. *Experimental neurology* 236: 179-189, 2012.

Knikou M. Functional reorganization of soleus H-reflex modulation during stepping after robotic-assisted step training in people with complete and incomplete spinal cord injury. *Experimental brain research* 228: 279-296, 2013.

Knikou M, and Mummidisetty CK. Locomotor training improves premotoneuronal control after chronic spinal cord injury. *Journal of neurophysiology* 111: 2264-2275, 2014.

Knikou M, Smith AC, and Mummidisetty CK. Locomotor training improves reciprocal and nonreciprocal inhibitory control of soleus motoneurons in human spinal cord injury. *Journal of neurophysiology* 113: 2447-2460, 2015.

Ko HY, Ditunno JF, Jr., Graziani V, and Little JW. The pattern of reflex recovery during spinal shock. *Spinal cord* 37: 402-409, 1999.

Louie DR, Eng JJ, and Lam T. Gait speed using powered robotic exoskeletons after spinal cord injury: a systematic review and correlational study. *Journal of neuroengineering and rehabilitation* 12: 82, 2015.

Magladery JW, Porter WE, Park AM, and Teasdall RD. Electrophysiological studies of nerve and reflex activity in normal man. IV. The two-neurone reflex and identification of certain action potentials from spinal roots and cord. *Bulletin of the Johns Hopkins Hospital* 88: 499-519, 1951.

Manella KJ, and Field-Fote EC. Modulatory effects of locomotor training on extensor spasticity in individuals with motor-incomplete spinal cord injury. *Restorative neurology and neuroscience* 31: 633-646, 2013.

Miele VJ, Panjabi MM, and Benzel EC. Anatomy and biomechanics of the spinal column and cord. *Handbook of clinical neurology* 109: 31-43, 2012.

Musselman KE, Fouad K, Misiaszek JE, and Yang JF. Training of walking skills overground and on the treadmill: case series on individuals with incomplete spinal cord injury. *Physical therapy* 89: 601-611, 2009.

Oleson CV, Burns AS, Ditunno JF, Geisler FH, and Coleman WP. Prognostic value of pinprick preservation in motor complete, sensory incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 86: 988-992, 2005.

Rossi A, Mazzocchio R, and Scarpini C. Clonus in man: a rhythmic oscillation maintained by a reflex mechanism. *Electroencephalography and clinical neurophysiology* 75: 56-63, 1990.

Scivoletto G, and Di Donna V. Prediction of walking recovery after spinal cord injury. *Brain research bulletin* 78: 43-51, 2009.

Scivoletto G, Morganti B, and Molinari M. Neurologic recovery of spinal cord injury patients in Italy. *Archives of physical medicine and rehabilitation* 85: 485-489, 2004a.

Scivoletto G, Romanelli A, Mariotti A, Marinucci D, Tamburella F, Mammone A, Cosentino E, Sterzi S, and Molinari M. Clinical factors that affect walking level and performance in chronic spinal cord lesion patients. *Spine* 33: 259-264, 2008.

Sinkjaer T, Andersen JB, and Larsen B. Soleus stretch reflex modulation during gait in humans. *Journal of neurophysiology* 76: 1112-1120, 1996.

Stein RB, and Kearney RE. Nonlinear behavior of muscle reflexes at the human ankle joint. *Journal of neurophysiology* 73: 65-72, 1995.

Stephens MJ, and Yang JF. Loading during the stance phase of walking in humans increases the extensor EMG amplitude but does not change the duration of the step cycle. *Experimental brain research* 124: 363-370, 1999.

Stephens MJ, and Yang JF. Short latency, non-reciprocal group I inhibition is reduced during the stance phase of walking in humans. *Brain Res* 743: 24-31, 1996.

Thomas SL, and Gorassini MA. Increases in corticospinal tract function by treadmill training after incomplete spinal cord injury. *Journal of neurophysiology* 94: 2844-2855, 2005.

Trimble MH, Behrman AL, Flynn SM, Thigpen MT, and Thompson FJ. Acute effects of locomotor training on overground walking speed and H-reflex modulation in individuals with incomplete spinal cord injury. *The journal of spinal cord medicine* 24: 74-80, 2001.

Trimble MH, Kukulka CG, and Behrman AL. The effect of treadmill gait training on low-frequency depression of the soleus H-reflex: comparison of a spinal cord injured man to normal subjects. *Neurosci Lett* 246: 186-188, 1998.

Valls-Sole J, Alvarez R, and Tolosa ES. Vibration-induced presynaptic inhibition of the soleus H reflex is temporarily reduced by cortical magnetic stimulation in human subjects. *Neuroscience letters* 170: 149-152, 1994.

Van Wezel BM, Ottenhoff FA, and Duysens J. Dynamic control of location-specific information in tactile cutaneous reflexes from the foot during human walking. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 17: 3804-3814, 1997.

Wallace DM, Ross BH, and Thomas CK. Characteristics of lower extremity clonus after human cervical spinal cord injury. *Journal of neurotrauma* 29: 915-924, 2012.

Walsh EG. Clonus: beats provoked by the application of a rhythmic force. *Journal of neurology, neurosurgery, and psychiatry* 39: 266-274, 1976.

Winchester P, Smith P, Foreman N, Mosby JM, Pacheco F, Querry R, and Tansey K. A prediction model for determining over ground walking speed after locomotor training in persons with motor incomplete spinal cord injury. *The journal of spinal cord medicine* 32: 63-71, 2009.

Wirz M, Zemon DH, Rupp R, Scheel A, Colombo G, Dietz V, and Hornby TG. Effectiveness of automated locomotor training in patients with chronic incomplete spinal cord injury: a multicenter trial. *Archives of physical medicine and rehabilitation* 86: 672-680, 2005.

Yang JF, Musselman KE, Livingstone D, Brunton K, Hendricks G, Hill D, and Gorassini M. Repetitive mass practice or focused precise practice for retraining walking after incomplete spinal cord injury? A pilot randomized clinical trial. *Neurorehabilitation and neural repair* 28: 314-324, 2014.

Yang JF, Norton J, Nevett-Duchcherer J, Roy FD, Gross DP, and Gorassini MA. Volitional muscle strength in the legs predicts changes in walking speed following locomotor training in people with chronic spinal cord injury. *Physical therapy* 91: 931-943, 2011.

Yang JF, and Stein RB. Phase-dependent reflex reversal in human leg muscles during walking. *Journal of neurophysiology* 63: 1109-1117, 1990.

Yang JF, Stein RB, and James KB. Contribution of peripheral afferents to the activation of the soleus muscle during walking in humans. *Experimental brain research* 87: 679-687, 1991.

Zehr EP, Komiyama T, and Stein RB. Cutaneous reflexes during human gait: electromyographic and kinematic responses to electrical stimulation. *Journal of neurophysiology* 77: 3311-3325, 1997.

Zehr EP, and Stein RB. What functions do reflexes serve during human locomotion? *Progress in neurobiology* 58: 185-205, 1999.

Zehr EP, Stein RB, and Komiyama T. Function of sural nerve reflexes during human walking. *The Journal of physiology* 507 (Pt 1): 305-314, 1998.

Zewdie ET, Roy FD, Yang JF, and Gorassini MA. Facilitation of descending excitatory and spinal inhibitory networks from training of endurance and precision walking in participants with incomplete spinal cord injury. *Prog Brain Res* 218: 127-155, 2015.

BIBLIOGRAPHY

Aagaard P, Simonsen EB, Andersen JL, Magnusson P, and Dyhre-Poulsen P. Neural adaptation to resistance training: changes in evoked V-wave and H-reflex responses. *Journal of applied physiology (Bethesda, Md : 1985)* 92: 2309-2318, 2002.

Achache V, Roche N, Lamy JC, Boakye M, Lackmy A, Gastal A, Quentin V, and Katz R. Transmission within several spinal pathways in adults with cerebral palsy. *Brain : a journal of neurology* 133: 1470-1483, 2010.

Adams MM, and Hicks AL. Comparison of the effects of body-weight-supported treadmill training and tilt-table standing on spasticity in individuals with chronic spinal cord injury. *The journal of spinal cord medicine* 34: 488-494, 2011.

Ahuja CS, Wilson JR, Nori S, Kotter MRN, Druschel C, Curt A, and Fehlings MG. Traumatic spinal cord injury. *Nature reviews Disease primers* 3: 17018, 2017.

Al-Khodairy AT, Gobelet C, and Rossier AB. Has botulinum toxin type A a place in the treatment of spasticity in spinal cord injury patients? *Spinal cord* 36: 854-858, 1998.

Alexeeva N, Sames C, Jacobs PL, Hobday L, Distasio MM, Mitchell SA, and Calancie B. Comparison of training methods to improve walking in persons with chronic spinal cord injury: a randomized clinical trial. *The journal of spinal cord medicine* 34: 362-379, 2011.

Amemiya M, and Yamaguchi T. Fictive locomotion of the forelimb evoked by stimulation of the mesencephalic locomotor region in the decerebrate cat. *Neurosci Lett* 50: 91-96, 1984.

Anderson KD. Targeting recovery: priorities of the spinal cord-injured population. *Journal of neurotrauma* 21: 1371-1383, 2004a.

Anderson KD. Targeting recovery: priorities of the spinal cord-injured population. *J Neurotrauma* 21: 1371-1383, 2004b.

Asselin P, Knezevic S, Kornfeld S, Cirnigliaro C, Agranova-Breyter I, Bauman WA, and Spungen AM. Heart rate and oxygen demand of powered exoskeleton-assisted walking in persons with paraplegia. *J Rehabil Res Dev* 52: 147-158, 2015.

Barbareschi G, Richards R, Thornton M, Carlson T, and Holloway C. Statically vs dynamically balanced gait: Analysis of a robotic exoskeleton compared with a human. *Conf Proc IEEE Eng Med Biol Soc* 2015: 6728-6731, 2015.

Barthelemy D, Grey MJ, Nielsen JB, and Bouyer L. Involvement of the corticospinal tract in the control of human gait. *Prog Brain Res* 192: 181-197, 2011.

Barthelemy D, Willerslev-Olsen M, Lundell H, Conway BA, Knudsen H, Biering-Sorensen F, and Nielsen JB. Impaired transmission in the corticospinal tract and gait disability in spinal cord injured persons. *Journal of neurophysiology* 104: 1167-1176, 2010.

Beekhuizen KS, and Field-Fote EC. Sensory stimulation augments the effects of massed practice training in persons with tetraplegia. *Archives of physical medicine and rehabilitation* 89: 602-608, 2008.

Behrens M, Mau-Moeller A, Wassermann F, Bader R, and Bruhn S. Effect of balance training on neuromuscular function at rest and during isometric maximum voluntary contraction. *Eur J Appl Physiol* 115: 1075-1085, 2015.

Behrman AL, Lawless-Dixon AR, Davis SB, Bowden MG, Nair P, Phadke C, Hannold EM, Plummer P, and Harkema SJ. Locomotor training progression and outcomes after incomplete spinal cord injury. *Physical therapy* 85: 1356-1371, 2005.

Beloozerova IN, Farrell BJ, Sirota MG, and Prilutsky BI. Differences in movement mechanics, electromyographic, and motor cortex activity between accurate and nonaccurate stepping. *Journal of neurophysiology* 103: 2285-2300, 2010.

Benson I, Hart K, Tussler D, and van Middendorp JJ. Lower-limb exoskeletons for individuals with chronic spinal cord injury: findings from a feasibility study. *Clin Rehabil* 30: 73-84, 2016.

Benson I, Hart K, Tussler D, and van Middendorp JJ. Lower-limb exoskeletons for individuals with chronic spinal cord injury: Findings from a feasibility study. *Clin Rehabil* 2015.

Benz EN, Hornby TG, Bode RK, Scheidt RA, and Schmit BD. A physiologically based clinical measure for spastic reflexes in spinal cord injury. *Archives of physical medicine and rehabilitation* 86: 52-59, 2005.

Beres-Jones JA, Johnson TD, and Harkema SJ. Clonus after human spinal cord injury cannot be attributed solely to recurrent muscle-tendon stretch. *Experimental brain research* 149: 222-236, 2003.

Bernardi M, Canale I, Castellano V, Di Filippo L, Felici F, and Marchetti M. The efficiency of walking of paraplegic patients using a reciprocating gait orthosis. *Paraplegia* 33: 409-415, 1995.

Biering-Sorensen F, Hansen B, and Lee BS. Non-pharmacological treatment and prevention of bone loss after spinal cord injury: a systematic review. *Spinal cord* 47: 508-518, 2009.

Bjerkefors A, Squair JW, Chua R, Lam T, Chen Z, and Carpenter MG. Assessment of abdominal muscle function in individuals with motor-complete spinal cord injury above T6 in response to transcranial magnetic stimulation. *Journal of rehabilitation medicine* 47: 138-146, 2015.

Bolton DA, Tse AD, Ballermann M, Misiaszek JE, and Fouad K. Task specific adaptations in rat locomotion: runway versus horizontal ladder. *Behavioural brain research* 168: 272-279, 2006.

Boorman GI, Lee RG, Becker WJ, and Windhorst UR. Impaired "natural reciprocal inhibition" in patients with spasticity due to incomplete spinal cord injury. *Electroencephalography and clinical neurophysiology* 101: 84-92, 1996.

Bos R, Sadlaoud K, Boulenguez P, Buttigieg D, Liabeuf S, Brocard C, Haase G, Bras H, and Vinay L. Activation of 5-HT2A receptors upregulates the function of the neuronal K-Cl cotransporter KCC2. *Proceedings of the National Academy of Sciences of the United States of America* 110: 348-353, 2013.

Boulenguez P, Liabeuf S, Bos R, Bras H, Jean-Xavier C, Brocard C, Stil A, Darbon P, Cattaert D, Delpire E, Marsala M, and Vinay L. Down-regulation of the potassium-chloride

cotransporter KCC2 contributes to spasticity after spinal cord injury. *Nature medicine* 16: 302-307, 2010.

Brown-Triolo DL, Roach MJ, Nelson K, and Triolo RJ. Consumer perspectives on mobility: implications for neuroprosthesis design. *J Rehabil Res Dev* 39: 659-669, 2002.

Brown TG. The Intrinsic Factors in the Act of Progression in the Mammal. *Proceedings of the Royal Society of London Series B, Containing Papers of a Biological Character* 84: 308-319, 1911.

Bryce TN, Dijkers MP, and Kozlowski AJ. Framework for Assessment of the Usability of Lower-Extremity Robotic Exoskeletal Orthoses. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 94: 1000-1014, 2015.

Buehner JJ, Forrest GF, Schmidt-Read M, White S, Tansey K, and Basso DM. Relationship between ASIA examination and functional outcomes in the NeuroRecovery Network Locomotor Training Program. *Archives of physical medicine and rehabilitation* 93: 1530-1540, 2012.

Burns AS, Marino RJ, Flanders AE, and Flett H. Clinical diagnosis and prognosis following spinal cord injury. *Handbook of clinical neurology* 109: 47-62, 2012.

Burns SP, Golding DG, Rolle WA, Jr., Graziani V, and Ditunno JF, Jr. Recovery of ambulation in motor-incomplete tetraplegia. *Archives of physical medicine and rehabilitation* 78: 1169-1172, 1997.

Bussel B, Roby-Brami A, Azouvi P, Biraben A, Yakovleff A, and Held JP. Myoclonus in a patient with spinal cord transection. Possible involvement of the spinal stepping generator. *Brain : a journal of neurology* 111 (Pt 5): 1235-1245, 1988.

Butler JE, Godfrey S, and Thomas CK. Depression of involuntary activity in muscles paralyzed by spinal cord injury. *Muscle & nerve* 33: 637-644, 2006.

Calancie B, Molano MR, and Broton JG. Tendon reflexes for predicting movement recovery after acute spinal cord injury in humans. *Clin Neurophysiol* 115: 2350-2363, 2004.

Calancie B, Needham-Shropshire B, Jacobs P, Willer K, Zych G, and Green BA. Involuntary stepping after chronic spinal cord injury. Evidence for a central rhythm generator for locomotion in man. *Brain : a journal of neurology* 117 (Pt 5): 1143-1159, 1994.

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

Capaday C, and Stein RB. Amplitude modulation of the soleus H-reflex in the human during walking and standing. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 6: 1308-1313, 1986.

Capaday C, and Stein RB. Difference in the amplitude of the human soleus H reflex during walking and running. *The Journal of physiology* 392: 513-522, 1987.

Cariga P, Catley M, Nowicky AV, Savic G, Ellaway PH, and Davey NJ. Segmental recording of cortical motor evoked potentials from thoracic paravertebral myotomes in complete spinal cord injury. *Spine* 27: 1438-1443, 2002.

Casabona A, Polizzi MC, and Perciavalle V. Differences in H-reflex between athletes trained for explosive contractions and non-trained subjects. *European journal of applied physiology and occupational physiology* 61: 26-32, 1990.

Cazalets JR, Sqalli-Houssaini Y, and Clarac F. Activation of the central pattern generators for locomotion by serotonin and excitatory amino acids in neonatal rat. *The Journal of physiology* 455: 187-204, 1992.

Cazalets JR, Sqalli-Houssaini Y, and Clarac F. GABAergic inactivation of the central pattern generators for locomotion in isolated neonatal rat spinal cord. *The Journal of physiology* 474: 173-181, 1994.

Cheng CL, Noonan VK, Shurgold J, Chen J, Rivers CS, Hamedani HK, Humphreys S, Bailey C, Attabib N, Mac-Thiong JM, Goytan M, Paquet J, Fox R, Ahn H, Kwon BK, and Fourney DR. Geomapping of Traumatic Spinal Cord Injury in Canada and Factors Related to Triage Pattern. *Journal of neurotrauma* 2017.

Chisholm AE, Alamro RA, Williams AM, and Lam T. Overground vs. treadmill-based robotic gait training to improve seated balance in people with motor-complete spinal cord injury: a case report. *Journal of neuroengineering and rehabilitation* 14: 27, 2017.

Chisholm AE, Peters S, Borich MR, Boyd LA, and Lam T. Short-term cortical plasticity associated with feedback-error learning after locomotor training in a patient with incomplete spinal cord injury. *Physical therapy* 95: 257-266, 2015.

Coghlan JK, Robinson CE, Newmarch B, and Jackson G. Lower extremity bracing in paraplegia--a follow-up study. *Paraplegia* 18: 25-32, 1980.

Colombo G, Joerg M, Schreier R, and Dietz V. Treadmill training of paraplegic patients using a robotic orthosis. *Journal of rehabilitation research and development* 37: 693-700, 2000.

Corcos DM, Gottlieb GL, Penn RD, Myklebust B, and Agarwal GC. Movement deficits caused by hyperexcitable stretch reflexes in spastic humans. *Brain : a journal of neurology* 109 (Pt 5): 1043-1058, 1986.

Couris CM, Guilcher SJ, Munce SE, Fung K, Craven BC, Verrier M, and Jaglal SB. Characteristics of adults with incident traumatic spinal cord injury in Ontario, Canada. *Spinal cord* 48: 39-44, 2010.

Crone C, Johnsen LL, Biering-Sorensen F, and Nielsen JB. Appearance of reciprocal facilitation of ankle extensors from ankle flexors in patients with stroke or spinal cord injury. *Brain : a journal of neurology* 126: 495-507, 2003.

Crozier KS, Cheng LL, Graziani V, Zorn G, Herbison G, and Ditunno JF, Jr. Spinal cord injury: prognosis for ambulation based on quadriceps recovery. *Paraplegia* 30: 762-767, 1992.

D'Amico JM, Condliffe EG, Martins KJ, Bennett DJ, and Gorassini MA. Recovery of neuronal and network excitability after spinal cord injury and implications for spasticity. *Frontiers in integrative neuroscience* 8: 36, 2014.

D'Amico JM, Li Y, Bennett DJ, and Gorassini MA. Reduction of spinal sensory transmission by facilitation of 5-HT1B/D receptors in noninjured and spinal cord-injured humans. *Journal of neurophysiology* 109: 1485-1493, 2013.

de Vlugt E, de Groot JH, Wisman WH, and Meskers CG. Clonus is explained from increased reflex gain and enlarged tissue viscoelasticity. *Journal of biomechanics* 45: 148-155, 2012.

Dietz V. Spastic movement disorder: what is the impact of research on clinical practice? *Journal of neurology, neurosurgery, and psychiatry* 74: 820-821, 2003.

Dietz V, Gollhofer A, Kleiber M, and Trippel M. Regulation of bipedal stance: dependency on "load" receptors. *Experimental brain research* 89: 229-231, 1992.

Dietz V, and Sinkjaer T. Spastic movement disorder: impaired reflex function and altered muscle mechanics. *The Lancet Neurology* 6: 725-733, 2007.

Dimitrijevic M, Nathan P, and Sherwood A. Clonus: the role of central mechanisms. *Journal of Neurology, Neurosurgery & Psychiatry* 43: 321-332, 1980a.

Dimitrijevic MR, Gerasimenko Y, and Pinter MM. Evidence for a spinal central pattern generator in humans. *Annals of the New York Academy of Sciences* 860: 360-376, 1998.

Dimitrijevic MR, Nathan PW, and Sherwood AM. Clonus: the role of central mechanisms. *Journal of neurology, neurosurgery, and psychiatry* 43: 321-332, 1980b.

Dobkin B, Apple D, Barbeau H, Basso M, Behrman A, Deforge D, Ditunno J, Dudley G, Elashoff R, Fugate L, Harkema S, Saulino M, Scott M, and Spinal Cord Injury Locomotor Trial G. Weight-supported treadmill vs over-ground training for walking after acute incomplete SCI. *Neurology* 66: 484-493, 2006.

Drew T. Motor cortical cell discharge during voluntary gait modification. *Brain Res* 457: 181-187, 1988.

Drew T, Dubuc R, and Rossignol S. Discharge patterns of reticulospinal and other reticular neurons in chronic, unrestrained cats walking on a treadmill. *Journal of neurophysiology* 55: 375-401, 1986.

Dryden DM, Saunders LD, Rowe BH, May LA, Yiannakoulias N, Svenson LW, Schopflocher DP, and Voaklander DC. The epidemiology of traumatic spinal cord injury in Alberta, Canada. *Canadian journal of neurological sciences* 30: 113-121, 2003.

Duffell LD, Brown GL, and Mirbagheri MM. Interventions to Reduce Spasticity and Improve Function in People With Chronic Incomplete Spinal Cord Injury: Distinctions Revealed by Different Analytical Methods. *Neurorehabilitation and neural repair* 29: 566-576, 2015.

Duysens J, Trippel M, Horstmann GA, and Dietz V. Gating and reversal of reflexes in ankle muscles during human walking. *Experimental brain research* 82: 351-358, 1990.

Earles DR, Dierking JT, Robertson CT, and Koceja DM. Pre- and post-synaptic control of motoneuron excitability in athletes. *Medicine and science in sports and exercise* 34: 1766-1772, 2002.

Elbasiouny SM, Moroz D, Bakr MM, and Mushahwar VK. Management of spasticity after spinal cord injury: current techniques and future directions. *Neurorehabilitation and neural repair* 24: 23-33, 2010.

Ellaway PH, Anand P, Bergstrom EM, Catley M, Davey NJ, Frankel HL, Jamous A, Mathias C, Nicotra A, Savic G, Short D, and Theodorou S. Towards improved clinical and physiological assessments of recovery in spinal cord injury: a clinical initiative. *Spinal cord* 42: 325-337, 2004.

Eriksson J, Olausson B, and Jankowska E. Antispastic effects of L-dopa. *Experimental brain research* 111: 296-304, 1996.

Ertekin C, Uludag B, On A, Yetimalar Y, Ertas M, Colakoglu Z, and Arac N. Motor-evoked potentials from various levels of paravertebral muscles in normal subjects and in patients with focal lesions of the spinal cord. *Spine* 23: 1016-1022, 1998.

Esquenazi A, Talaty M, Packel A, and Saulino M. The ReWalk powered exoskeleton to restore ambulatory function to individuals with thoracic-level motor-complete spinal cord injury. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 91: 911-921, 2012.

Evans N, Hartigan C, Kandilakis C, Pharo E, and Clesson I. Acute Cardiorespiratory and Metabolic Responses During Exoskeleton-Assisted Walking Overground Among Persons with Chronic Spinal Cord Injury. *Top Spinal Cord Inj Rehabil* 21: 122-132, 2015.

Faist M, Mazevet D, Dietz V, and Pierrot-Deseilligny E. A quantitative assessment of presynaptic inhibition of Ia afferents in spastics. Differences in hemiplegics and paraplegics. *Brain : a journal of neurology* 117 (Pt 6): 1449-1455, 1994.

Farris RJ, Quintero HA, Murray SA, Ha KH, Hartigan C, and Goldfarb M. A preliminary assessment of legged mobility provided by a lower limb exoskeleton for persons with paraplegia. *IEEE transactions on neural systems and rehabilitation engineering : a publication of the IEEE Engineering in Medicine and Biology Society* 22: 482-490, 2014.

Fenuta AM, and Hicks AL. Metabolic demand and muscle activation during different forms of bodyweight supported locomotion in men with incomplete SCI. *BioMed research international* 2014: 632765, 2014.

Field-Fote EC, and Roach KE. Influence of a locomotor training approach on walking speed and distance in people with chronic spinal cord injury: a randomized clinical trial. *Physical therapy* 91: 48-60, 2011.

Field-Fote EC, Yang JF, Basso DM, and Gorassini MA. Supraspinal Control Predicts Locomotor Function and Forecasts Responsiveness to Training after Spinal Cord Injury. *Journal of neurotrauma* 34: 1813-1825, 2017.

Forssberg H, Grillner S, and Rossignol S. Phase dependent reflex reversal during walking in chronic spinal cats. *Brain Res* 85: 103-107, 1975.

Fournier E, Katz R, and Pierrot-Deseilligny E. Descending control of reflex pathways in the production of voluntary isolated movements in man. *Brain Res* 288: 375-377, 1983.

Fried GW, and Fried KM. Spinal cord injury and use of botulinum toxin in reducing spasticity. *Physical medicine and rehabilitation clinics of North America* 14: 901-910, 2003.

Fukuyama H, Ouchi Y, Matsuzaki S, Nagahama Y, Yamauchi H, Ogawa M, Kimura J, and Shibasaki H. Brain functional activity during gait in normal subjects: a SPECT study. *Neurosci Lett* 228: 183-186, 1997.

Gerasimenko Y, Gorodnichev R, Machueva E, Pivovarova E, Semyenov D, Savochin A, Roy RR, and Edgerton VR. Novel and direct access to the human locomotor spinal circuitry. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 30: 3700-3708, 2010.

Glass A, and Hannah A. A comparison of dantrolene sodium and diazepam in the treatment of spasticity. *Paraplegia* 12: 170-174, 1974.

Gollie JM, Guccione AA, Panza GS, Jo PY, and Herrick JE. Effects of Overground Locomotor Training on Walking Performance in Chronic Cervical Motor Incomplete Spinal Cord Injury: A Pilot Study. *Archives of physical medicine and rehabilitation* 98: 1119-1125, 2017.

Goode DJ, and Van Hoven J. Loss of patellar and Achilles tendon reflexes in classical ballet dancers. *Arch Neurol* 39: 323, 1982.

Gorassini MA, Knash ME, Harvey PJ, Bennett DJ, and Yang JF. Role of motoneurons in the generation of muscle spasms after spinal cord injury. *Brain : a journal of neurology* 127: 2247-2258, 2004.

Gorassini MA, Norton JA, Nevett-Duchcherer J, Roy FD, and Yang JF. Changes in locomotor muscle activity after treadmill training in subjects with incomplete spinal cord injury. *Journal of neurophysiology* 101: 969-979, 2009.

Grassner L, Marschallinger J, Dünser MW, Novak HF, Zerbs A, Aigner L, Trinka E, and Sellner J. Nontraumatic spinal cord injury at the neurological intensive care unit: spectrum, causes of admission and predictors of mortality. *Therapeutic Advances in Neurological Disorders* 9: 85-94, 2016.

Grey MJ, Klinge K, Crone C, Lorentzen J, Biering-Sorensen F, Ravnborg M, and Nielsen JB. Post-activation depression of soleus stretch reflexes in healthy and spastic humans. *Experimental brain research* 185: 189-197, 2008.

Grillner S, Hellgren J, Menard A, Saitoh K, and Wikstrom MA. Mechanisms for selection of basic motor programs--roles for the striatum and pallidum. *Trends in neurosciences* 28: 364-370, 2005.

Gruber M, Taube W, Gollhofer A, Beck S, Amtage F, and Schubert M. Training-specific adaptations of H- and stretch reflexes in human soleus muscle. *J Mot Behav* 39: 68-78, 2007.

Gruenthal M, Mueller M, Olson WL, Priebe MM, Sherwood AM, and Olson WH. Gabapentin for the treatment of spasticity in patients with spinal cord injury. *Spinal cord* 35: 686-689, 1997.

Gunduz S, Kalyon TA, Dursun H, Mohur H, and Bilgic F. Peripheral nerve block with phenol to treat spasticity in spinal cord injured patients. *Paraplegia* 30: 808-811, 1992.

Guyatt GH, Sullivan MJ, Thompson PJ, Fallen EL, Pugsley SO, Taylor DW, and Berman LB. The 6-minute walk: a new measure of exercise capacity in patients with chronic heart failure. *Can Med Assoc J* 132: 919-923, 1985.

Hagen EM. Acute complications of spinal cord injuries. *World journal of orthopedics* 6: 17-23, 2015.

Hajela N, Mummidisetty CK, Smith AC, and Knikou M. Corticospinal reorganization after locomotor training in a person with motor incomplete paraplegia. *BioMed research international* 2013: 516427, 2013.

Harel NY, Asselin PK, Fineberg DB, Pisano TJ, Bauman WA, and Spungen AM. Adaptation of computerized posturography to assess seated balance in persons with spinal cord injury. *The journal of spinal cord medicine* 36: 127-133, 2013.

Harkema SJ, Schmidt-Read M, Lorenz DJ, Edgerton VR, and Behrman AL. Balance and ambulation improvements in individuals with chronic incomplete spinal cord injury using

locomotor training-based rehabilitation. *Archives of physical medicine and rehabilitation* 93: 1508-1517, 2012.

Hartigan C, Kandilakis C, Dalley S, Clausen M, Wilson E, Morrison S, Etheridge S, and Farris R. Mobility Outcomes Following Five Training Sessions with a Powered Exoskeleton. *Topics in Spinal Cord Injury Rehabilitation* 21: 93-99, 2015.

Hesse S, Werner C, and Bardeleben A. Electromechanical gait training with functional electrical stimulation: case studies in spinal cord injury. *Spinal cord* 42: 346-352, 2004.

Hicks AL, Adams MM, Martin Ginis K, Giangregorio L, Latimer A, Phillips SM, and McCartney N. Long-term body-weight-supported treadmill training and subsequent follow-up in persons with chronic SCI: effects on functional walking ability and measures of subjective well-being. *Spinal cord* 43: 291-298, 2005.

Hidler JM, and Rymer WZ. A simulation study of reflex instability in spasticity: origins of clonus. *IEEE transactions on rehabilitation engineering : a publication of the IEEE Engineering in Medicine and Biology Society* 7: 327-340, 1999.

Holmes G. The goulstonian lectures on spinal injuries of warfare: delivered before the Royal College of Physicians of London. *British medical journal* 2: 815-821, 1915.

Hornby TG, Zemon DH, and Campbell D. Robotic-assisted, body-weight-supported treadmill training in individuals following motor incomplete spinal cord injury. *Physical therapy* 85: 52-66, 2005.

http://eksobionics.com/. Ekso Bionics Ekso Bionics. http://eksobionics.com/.

http://rewalk.com/. ReWalk 6.0 – Home ReWalk Robotics. http://rewalk.com/.

http://www.indego.com/indego/en/home. Indego – Powering People Forward | Parker Indego Parker Hannifin Corp. http://www.indego.com/indego/en/home.

Hubli M, Dietz V, and Bolliger M. Spinal reflex activity: a marker for neuronal functionality after spinal cord injury. *Neurorehabilitation and neural repair* 26: 188-196, 2012.

Hultborn H, Illert M, Nielsen J, Paul A, Ballegaard M, and Wiese H. On the mechanism of the post-activation depression of the H-reflex in human subjects. *Experimental brain research* 108: 450-462, 1996.

Huttenlocher PR. Synaptic density in human frontal cortex - developmental changes and effects of aging. *Brain Res* 163: 195-205, 1979.

Iglesias C, Nielsen JB, and Marchand-Pauvert V. Corticospinal inhibition of transmission in propriospinal-like neurones during human walking. *The European journal of neuroscience* 28: 1351-1361, 2008.

Iles J, and Pisini J. Cortical modulation of transmission in spinal reflex pathways of man. *The Journal of physiology* 455: 425-446, 1992.

Ishikawa K, Ott K, Porter RW, and Stuart D. Low frequency depression of the H wave in normal and spinal man. *Experimental neurology* 15: 140-156, 1966.

Jacobs PL, Klose KJ, Guest R, Needham-Shropshire B, Broton JG, and Green BA. Relationships of oxygen uptake, heart rate, and ratings of perceived exertion in persons with paraplegia during functional neuromuscular stimulation assisted ambulation. *Spinal Cord* 35: 292-298, 1997.

Jahn K, Deutschlander A, Stephan T, Strupp M, Wiesmann M, and Brandt T. Brain activation patterns during imagined stance and locomotion in functional magnetic resonance imaging. *Neuroimage* 22: 1722-1731, 2004.

Jankowska E, Jukes MG, Lund S, and Lundberg A. The effect of DOPA on the spinal cord. 5. Reciprocal organization of pathways transmitting excitatory action to alpha motoneurones of flexors and extensors. *Acta physiologica Scandinavica* 70: 369-388, 1967.

Jazayeri SB, Beygi S, Shokraneh F, Hagen EM, and Rahimi-Movaghar V. Incidence of traumatic spinal cord injury worldwide: a systematic review. *European spine journal : official publication of the European Spine Society, the European Spinal Deformity Society, and the European Section of the Cervical Spine Research Society* 24: 905-918, 2015.

Johnson RL, Gerhart KA, McCray J, Menconi JC, and Whiteneck GG. Secondary conditions following spinal cord injury in a population-based sample. *Spinal cord* 36: 45-50, 1998.

Jones CA, and Yang JF. Reflex behavior during walking in incomplete spinal-cord-injured subjects. *Experimental neurology* 128: 239-248, 1994.

Jorgensen HS, Nakayama H, Raaschou HO, and Olsen TS. Recovery of walking function in stroke patients: the Copenhagen Stroke Study. *Archives of physical medicine and rehabilitation* 76: 27-32, 1995.

Juknis N, Cooper JM, and Volshteyn O. The changing landscape of spinal cord injury. *Handbook of clinical neurology* 109: 149-166, 2012.

Kakinohana O, Hefferan MP, Miyanohara A, Nejime T, Marsala S, Juhas S, Juhasova J, Motlik J, Kucharova K, Strnadel J, Platoshyn O, Lazar P, Galik J, Vinay L, and Marsala M. Combinational spinal GAD65 gene delivery and systemic GABA-mimetic treatment for modulation of spasticity. *PloS one* 7: e30561, 2012.

Kapitza S, Zorner B, Weinmann O, Bolliger M, Filli L, Dietz V, and Schwab ME. Tail spasms in rat spinal cord injury: changes in interneuronal connectivity. *Experimental neurology* 236: 179-189, 2012.

Karelis AD, Carvalho LP, Castillo MJ, Gagnon DH, and Aubertin-Leheudre M. Effect on body composition and bone mineral density of walking with a robotic exoskeleton in adults with chronic spinal cord injury. *Journal of rehabilitation medicine* 49: 84-87, 2017.

Kay ED, Deutsch A, and Wuermser LA. Predicting walking at discharge from inpatient rehabilitation after a traumatic spinal cord injury. *Archives of physical medicine and rehabilitation* 88: 745-750, 2007.

Keller M, Pfusterschmied J, Buchecker M, Muller E, and Taube W. Improved postural control after slackline training is accompanied by reduced H-reflexes. *Scand J Med Sci Sports* 22: 471-477, 2012.

Khan A, Livingstone D, Misiaszek J, Stein RB, Gorassini M, Manns P, and Yang J. Improving walking and inducing neuroplasticity after chronic SCI by training in the ReWalk exoskeleton. In: *Society for Neuroscience 2015 Annual meeting*. Chicago, IL: 2015.

Khan A, Patrick S, Roy FD, Gorassini M, and Yang J. Changes in spasticity-related reflexes after two forms of walking retraining in individuals with incomplete spinal cord injury. In: *Society for Neuroscience 2014 Annual Meeting*. Washington, DC: 2014.

Khan AS, Patrick SK, Roy FD, Gorassini MA, and Yang JF. Training-Specific Neural Plasticity in Spinal Reflexes after Incomplete Spinal Cord Injury. *Neural Plast* 2016: 6718763, 2016.

Kiehn O, Dougherty KJ, Hagglund M, Borgius L, Talpalar A, and Restrepo CE. Probing spinal circuits controlling walking in mammals. *Biochemical and biophysical research communications* 396: 11-18, 2010.

Kim CM, Eng JJ, and Whittaker MW. Level walking and ambulatory capacity in persons with incomplete spinal cord injury: relationship with muscle strength. *Spinal cord* 42: 156-162, 2004.

Kiser TS, Reese NB, Maresh T, Hearn S, Yates C, Skinner RD, Pait TG, and Garcia-Rill E. Use of a motorized bicycle exercise trainer to normalize frequency-dependent habituation of the H-reflex in spinal cord injury. *The journal of spinal cord medicine* 28: 241-245, 2005.

Knikou M. Functional reorganization of soleus H-reflex modulation during stepping after robotic-assisted step training in people with complete and incomplete spinal cord injury. *Experimental brain research* 228: 279-296, 2013.

Knikou M. Plasticity of corticospinal neural control after locomotor training in human spinal cord injury. *Neural plasticity* 2012: 2012.

Knikou M, and Mummidisetty CK. Locomotor training improves premotoneuronal control after chronic spinal cord injury. *Journal of neurophysiology* 111: 2264-2275, 2014.

Knikou M, Smith AC, and Mummidisetty CK. Locomotor training improves reciprocal and nonreciprocal inhibitory control of soleus motoneurons in human spinal cord injury. *Journal of neurophysiology* 113: 2447-2460, 2015.

Ko HY, Ditunno JF, Jr., Graziani V, and Little JW. The pattern of reflex recovery during spinal shock. *Spinal cord* 37: 402-409, 1999.

Koceja DM, Davison E, and Robertson CT. Neuromuscular characteristics of endurance- and power-trained athletes. *Research quarterly for exercise and sport* 75: 23-30, 2004.

Kolakowsky-Hayner S, Crew J, Moran S, and Shah A. Safety and feasibility of using the ekso bionic exoskeleton to aid ambulation after spinal cord injury. *Journal of spine* S4: 8, 2013.

Kozlowski AJ, Bryce TN, and Dijkers MP. Time and effort required by persons with spinal cord injury to learn to use a powered exoskeleton for assisted walking. *Topics in Spinal Cord Injury Rehabilitation* 21: 110-121, 2015.

Krawetz P, and Nance P. Gait analysis of spinal cord injured subjects: effects of injury level and spasticity. *Archives of physical medicine and rehabilitation* 77: 635-638, 1996.

Kressler J, Nash MS, Burns PA, and Field-Fote EC. Metabolic responses to 4 different body weight-supported locomotor training approaches in persons with incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 94: 1436-1442, 2013.

Kressler J, Thomas CK, Field-Fote EC, Sanchez J, Widerstrom-Noga E, Cilien DC, Gant K, Ginnety K, Gonzalez H, Martinez A, Anderson KD, and Nash MS. Understanding therapeutic benefits of overground bionic ambulation: exploratory case series in persons with chronic, complete spinal cord injury. *Archives of physical medicine and rehabilitation* 95: 1878-1887 e1874, 2014.

Krueger H, Noonan VK, Trenaman LM, Joshi P, and Rivers CS. The economic burden of traumatic spinal cord injury in Canada. *Chronic diseases and injuries in Canada* 33: 113-122, 2013.

Kuhn RA. Functional capacity of the isolated human spinal cord. *Brain : a journal of neurology* 73: 1-51, 1950.

Labruyere R, and van Hedel HJ. Strength training versus robot-assisted gait training after incomplete spinal cord injury: a randomized pilot study in patients depending on walking assistance. *Journal of neuroengineering and rehabilitation* 11: 4, 2014.

Lagerquist O, Zehr EP, and Docherty D. Increased spinal reflex excitability is not associated with neural plasticity underlying the cross-education effect. *Journal of applied physiology (Bethesda, Md : 1985)* 100: 83-90, 2006.

Lajeunesse V, Vincent C, Routhier F, Careau E, and Michaud F. Exoskeletons' design and usefulness evidence according to a systematic review of lower limb exoskeletons used for functional mobility by people with spinal cord injury. *Disability and rehabilitation Assistive technology* 1-13, 2015.

Lam T, Pauhl K, Ferguson A, Malik RN, Krassioukov A, and Eng JJ. Training with robot-applied resistance in people with motor-incomplete spinal cord injury: Pilot study. *Journal of rehabilitation research and development* 52: 113-129, 2015.

Lazorthes Y, Sallerin-Caute B, Verdie JC, Bastide R, and Carillo JP. Chronic intrathecal baclofen administration for control of severe spasticity. *Journal of neurosurgery* 72: 393-402, 1990.

Lee BB, Cripps RA, Fitzharris M, and Wing PC. The global map for traumatic spinal cord injury epidemiology: update 2011, global incidence rate. *Spinal cord* 52: 110-116, 2014.

Lee MS, Choi YC, Lee SH, and Lee SB. Sleep-related periodic leg movements associated with spinal cord lesions. *Movement disorders : official journal of the Movement Disorder Society* 11: 719-722, 1996.

Lemay JF, Gagnon D, Duclos C, Grangeon M, Gauthier C, and Nadeau S. Influence of visual inputs on quasi-static standing postural steadiness in individuals with spinal cord injury. *Gait Posture* 38: 357-360, 2013.

Lenehan B, Street J, Kwon BK, Noonan V, Zhang H, Fisher CG, and Dvorak MF. The epidemiology of traumatic spinal cord injury in British Columbia, Canada. *Spine* 37: 321-329, 2012.

Leung AK, Wong AF, Wong EC, and Hutchins SW. The Physiological Cost Index of walking with an isocentric reciprocating gait orthosis among patients with T(12) - L(1) spinal cord injury. *Prosthet Orthot Int* 33: 61-68, 2009.

Levi R, Hultling C, and Seiger A. The Stockholm Spinal Cord Injury Study: 2. Associations between clinical patient characteristics and post-acute medical problems. *Paraplegia* 33: 585-594, 1995.

Little JW, and Halar EM. H-reflex changes following spinal cord injury. *Archives of physical medicine and rehabilitation* 66: 19-22, 1985.

Little JW, Micklesen P, Umlauf R, and Britell C. Lower extremity manifestations of spasticity in chronic spinal cord injury. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 68: 32-36, 1989.

Liu H, Skinner RD, Arfaj A, Yates C, Reese NB, Williams K, and Garcia-Rill E. L-Dopa effect on frequency-dependent depression of the H-reflex in adult rats with complete spinal cord transection. *Brain research bulletin* 83: 262-265, 2010.

Lorentzen J, Grey MJ, Geertsen SS, Biering-Sorensen F, Brunton K, Gorassini M, and Nielsen JB. Assessment of a portable device for the quantitative measurement of ankle joint stiffness in spastic individuals. *Clin Neurophysiol* 123: 1371-1382, 2012.

Lotharius J, and Brundin P. Pathogenesis of Parkinson's disease: dopamine, vesicles and alpha-synuclein. *Nature reviews Neuroscience* 3: 932-942, 2002.

Lotta S, Fiocchi A, Giovannini R, Silvestrin R, Tesio L, Raschi A, Macchia L, Chiapatti V, Zambelli M, and Tosi C. Restoration of gait with orthoses in thoracic paraplegia: a multicentric investigation. *Paraplegia* 32: 608-615, 1994.

Louie DR, Eng JJ, and Lam T. Gait speed using powered robotic exoskeletons after spinal cord injury: a systematic review and correlational study. *Journal of neuroengineering and rehabilitation* 12: 82, 2015.

Maffiuletti NA, Martin A, Babault N, Pensini M, Lucas B, and Schieppati M. Electrical and mechanical H(max)-to-M(max) ratio in power- and endurance-trained athletes. *Journal of applied physiology (Bethesda, Md : 1985)* 90: 3-9, 2001.

Magladery JW, Porter WE, Park AM, and Teasdall RD. Electrophysiological studies of nerve and reflex activity in normal man. IV. The two-neurone reflex and identification of certain action potentials from spinal roots and cord. *Bulletin of the Johns Hopkins Hospital* 88: 499-519, 1951.

Mahoney JS, Engebretson JC, Cook KF, Hart KA, Robinson-Whelen S, and Sherwood AM. Spasticity Experience Domains in Persons With Spinal Cord Injury. *Archives of physical medicine and rehabilitation* 88: 287-294, 2007.

Manella KJ, and Field-Fote EC. Modulatory effects of locomotor training on extensor spasticity in individuals with motor-incomplete spinal cord injury. *Restorative neurology and neuroscience* 31: 633-646, 2013.

Maynard FM, Jr., Bracken MB, Creasey G, Ditunno JF, Jr., Donovan WH, Ducker TB, Garber SL, Marino RJ, Stover SL, Tator CH, Waters RL, Wilberger JE, and Young W. International Standards for Neurological and Functional Classification of Spinal Cord Injury. American Spinal Injury Association. *Spinal cord* 35: 266-274, 1997.

McKinley WO, Seel RT, Gadi RK, and Tewksbury MA. Nontraumatic vs. traumatic spinal cord injury: a rehabilitation outcome comparison. *American journal of physical medicine & rehabilitation / Association of Academic Physiatrists* 80: 693-699; quiz 700, 716, 2001.

McPherson DR, and Kemnitz CP. Modulation of lamprey fictive swimming and motoneuron physiology by dopamine, and its immunocytochemical localization in the spinal cord. *Neurosci Lett* 166: 23-26, 1994.

Mehrholz J, Harvey LA, Thomas S, and Elsner B. Is body-weight-supported treadmill training or robotic-assisted gait training superior to overground gait training and other forms of physiotherapy in people with spinal cord injury? A systematic review. *Spinal cord* 55: 722-729, 2017.

Melzack R. The McGill Pain Questionnaire: major properties and scoring methods. *Pain* 1: 277-299, 1975.

Miele VJ, Panjabi MM, and Benzel EC. Anatomy and biomechanics of the spinal column and cord. *Handbook of clinical neurology* 109: 31-43, 2012.

Mikelberg R, and Reid S. Spinal cord lesions and lower extremity bracing: an overview and follow-up study. *Paraplegia* 19: 379-385, 1981.

Miller LE, Zimmermann AK, and Herbert WG. Clinical effectiveness and safety of powered exoskeleton-assisted walking in patients with spinal cord injury: systematic review with meta-analysis. *Medical devices (Auckland, NZ)* 9: 455-466, 2016.

Mirbagheri M, Tsao C, Pelosin E, and Rymer W. Therapeutic effects of robotic-assisted locomotor training on neuromuscular properties. In: *Rehabilitation Robotics, 2005 ICORR 2005 9th International Conference on*IEEE, 2005, p. 561-564.

Mirbagheri MM, Barbeau H, Ladouceur M, and Kearney RE. Intrinsic and reflex stiffness in normal and spastic, spinal cord injured subjects. *Experimental brain research* 141: 446-459, 2001.

Mirbagheri MM, Chen D, and Rymer WZ. Quantification of the effects of an alpha-2 adrenergic agonist on reflex properties in spinal cord injury using a system identification technique. *Journal of neuroengineering and rehabilitation* 7: 29, 2010.

Mirbagheri MM, Kindig MW, and Niu X. Effects of robotic-locomotor training on stretch reflex function and muscular properties in individuals with spinal cord injury. *Clin Neurophysiol* 126: 997-1006, 2015.

Mirbagheri MM, Ladouceur M, Barbeau H, and Kearney RE. The effects of long-term FES-assisted walking on intrinsic and reflex dynamic stiffness in spastic spinal-cord-injured subjects. *Neural Systems and Rehabilitation Engineering, IEEE Transactions on* 10: 280-289, 2002.

Miyai I, Tanabe HC, Sase I, Eda H, Oda I, Konishi I, Tsunazawa Y, Suzuki T, Yanagida T, and Kubota K. Cortical mapping of gait in humans: a near-infrared spectroscopic topography study. *Neuroimage* 14: 1186-1192, 2001.

Mori S, Matsui T, Kuze B, Asanome M, Nakajima K, and Matsuyama K. Cerebellar-induced locomotion: reticulospinal control of spinal rhythm generating mechanism in cats. *Annals of the New York Academy of Sciences* 860: 94-105, 1998.

Morse LR, Stolzmann K, Nguyen HP, Jain NB, Zayac C, Gagnon DR, Tun CG, and Garshick E. Association between mobility mode and C-reactive protein levels in men with chronic spinal cord injury. *Archives of physical medicine and rehabilitation* 89: 726-731, 2008.

Morton SM, and Bastian AJ. Cerebellar contributions to locomotor adaptations during splitbelt treadmill walking. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 26: 9107-9116, 2006.

Murray KC, Nakae A, Stephens MJ, Rank M, D'Amico J, Harvey PJ, Li X, Harris RLW, Ballou EW, Anelli R, Heckman CJ, Mashimo T, Vavrek R, Sanelli L, Gorassini MA, Bennett DJ, and Fouad K. Recovery of motoneuron and locomotor function after spinal cord injury depends on constitutive activity in 5-HT(2C) receptors. *Nature medicine* 16: 694-700, 2010.

Musselman K, Brunton K, Lam T, and Yang J. Spinal cord injury functional ambulation profile: a new measure of walking ability. *Neurorehabilitation and neural repair* 25: 285-293, 2011.

Musselman KE, Fouad K, Misiaszek JE, and Yang JF. Training of walking skills overground and on the treadmill: case series on individuals with incomplete spinal cord injury. *Physical therapy* 89: 601-611, 2009.

Musselman KE, and Yang JF. Spinal Cord Injury Functional Ambulation Profile: a preliminary look at responsiveness. *Physical therapy* 94: 240-250, 2014.

Musselman KE, and Yang JF. Walking tasks encountered by urban-dwelling adults and persons with incomplete spinal cord injuries. *Journal of rehabilitation medicine* 39: 567-574, 2007.

Nadeau S, Jacquemin G, Fournier C, Lamarre Y, and Rossignol S. Spontaneous motor rhythms of the back and legs in a patient with a complete spinal cord transection. *Neurorehabilitation and neural repair* 24: 377-383, 2010.

Nance PW. A comparison of clonidine, cyproheptadine and baclofen in spastic spinal cord injured patients. *The Journal of the American Paraplegia Society* 17: 150-156, 1994.

Nathan PW. Effects on movement of surgical incisions into the human spinal cord. *Brain : a journal of neurology* 117 (Pt 2): 337-346, 1994.

New PW, Cripps RA, and Bonne Lee B. Global maps of non-traumatic spinal cord injury epidemiology: towards a living data repository. *Spinal cord* 52: 97-109, 2014.

Nielsen J, Crone C, and Hultborn H. H-reflexes are smaller in dancers from The Royal Danish Ballet than in well-trained athletes. *European journal of applied physiology and occupational physiology* 66: 116-121, 1993a.

Nielsen J, Petersen N, Ballegaard M, Biering-Sorensen F, and Kiehn O. H-reflexes are less depressed following muscle stretch in spastic spinal cord injured patients than in healthy subjects. *Experimental brain research* 97: 173-176, 1993b.

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

Nielsen JB, and Sinkjaer T. Afferent feedback in the control of human gait. *Journal of electromyography and kinesiology : official journal of the International Society of Electrophysiological Kinesiology* 12: 213-217, 2002.

Niu X, Patel C, Varoqui D, and Mirbagheri MM. The effect of robot-assisted lokomotor training on gait recovery: a multivariate analysis. *Conference proceedings : Annual International Conference of the IEEE Engineering in Medicine and Biology Society IEEE Engineering in Medicine and Biology Society Annual Conference* 2011: 3542-3545, 2011.

Niu X, Varoqui D, Kindig M, and Mirbagheri MM. Prediction of gait recovery in spinal cord injured individuals trained with robotic gait orthosis. *Journal of neuroengineering and rehabilitation* 11: 42, 2014.

Noonan VK, Chan E, Santos A, Soril L, Lewis R, Singh A, Cheng CL, O'Connell C, Truchon C, Paquet J, Christie S, Ethans K, Tsai E, Ford MH, Drew B, Linassi G, Bailey C, and Fehlings MG. Traumatic Spinal Cord Injury Care in Canada: A Survey of Canadian Centres. *Journal of neurotrauma* 2017.

Norton JA, Bennett DJ, Knash ME, Murray KC, and Gorassini MA. Changes in sensory-evoked synaptic activation of motoneurons after spinal cord injury in man. *Brain : a journal of neurology* 131: 1478-1491, 2008.

Nutt JG, Bloem BR, Giladi N, Hallett M, Horak FB, and Nieuwboer A. Freezing of gait: moving forward on a mysterious clinical phenomenon. *The Lancet Neurology* 10: 734-744, 2011.

Oleson CV, Burns AS, Ditunno JF, Geisler FH, and Coleman WP. Prognostic value of pinprick preservation in motor complete, sensory incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 86: 988-992, 2005.

Olsson MC, Kruger M, Meyer LH, Ahnlund L, Gransberg L, Linke WA, and Larsson L. Fibre type-specific increase in passive muscle tension in spinal cord-injured subjects with spasticity. *The Journal of physiology* 577: 339-352, 2006.

Orlovsky GN. Activity of rubrospinal neurons during locomotion. *Brain Res* 46: 99-112, 1972a.

Orlovsky GN. Activity of vestibulospinal neurons during locomotion. *Brain Res* 46: 85-98, 1972b.

Pang MY, and Yang JF. The initiation of the swing phase in human infant stepping: importance of hip position and leg loading. *The Journal of physiology* 528 Pt 2: 389-404, 2000.

Panizza M, Nilsson J, and Hallett M. Optimal stimulus duration for the H reflex. *Muscle & nerve* 12: 576-579, 1989.

Pearson KG. Generating the walking gait: role of sensory feedback. *Prog Brain Res* 143: 123-129, 2004.

Pearson KG. Role of sensory feedback in the control of stance duration in walking cats. *Brain research reviews* 57: 222-227, 2008.

Pearson KG, and Collins DF. Reversal of the influence of group Ib afferents from plantaris on activity in medial gastrocnemius muscle during locomotor activity. *Journal of neurophysiology* 70: 1009-1017, 1993.

Peiper A. *Cerebral function in infancy and childhood*. Plenum Pub Corp, 1963.

Perez MA, Lungholt BK, Nyborg K, and Nielsen JB. Motor skill training induces changes in the excitability of the leg cortical area in healthy humans. *Experimental brain research* 159: 197-205, 2004.

Petersen NT, Butler JE, Marchand-Pauvert V, Fisher R, Ledebt A, Pyndt HS, Hansen NL, and Nielsen JB. Suppression of EMG activity by transcranial magnetic stimulation in human subjects during walking. *The Journal of physiology* 537: 651-656, 2001.

Phadke CP, Flynn SM, Thompson FJ, Behrman AL, Trimble MH, and Kukulka CG. Comparison of single bout effects of bicycle training versus locomotor training on paired reflex depression of the soleus H-reflex after motor incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 90: 1218-1228, 2009.

Phillips SM, Stewart BG, Mahoney DJ, Hicks AL, McCartney N, Tang JE, Wilkinson SB, Armstrong D, and Tarnopolsky MA. Body-weight-support treadmill training improves blood glucose regulation in persons with incomplete spinal cord injury. *Journal of applied physiology (Bethesda, Md : 1985)* 97: 716-724, 2004.

Pickett GE, Campos-Benitez M, Keller JL, and Duggal N. Epidemiology of traumatic spinal cord injury in Canada. *Spine* 31: 799-805, 2006.

Pinter MM, Gerstenbrand F, and Dimitrijevic MR. Epidural electrical stimulation of posterior structures of the human lumbosacral cord: 3. Control Of spasticity. *Spinal cord* 38: 524-531, 2000.

Platz T, Gillner A, Borgwaldt N, Kroll S, and Roschka S. Device-Training for Individuals with Thoracic and Lumbar Spinal Cord Injury Using a Powered Exoskeleton for Technically Assisted Mobility: Achievements and User Satisfaction. *BioMed research international* 2016: 8459018, 2016.

Possover M. Recovery of sensory and supraspinal control of leg movement in people with chronic paraplegia: a case series. *Archives of physical medicine and rehabilitation* 95: 610-614, 2014.

Pramodhyakul W, Wattanapan P, Siritaratiwat W, Eungpinichpong W, and Amatachaya S. Immediate effects of obstacle crossing training in independent ambulatory patients with spinal cord injury. *Spinal cord* 51: 379-383, 2013.

Priebe MM, Sherwood AM, Thornby JI, Kharas NF, and Markowski J. Clinical assessment of spasticity in spinal cord injury: a multidimensional problem. *Archives of physical medicine and rehabilitation* 77: 713-716, 1996.

Reyes R, Icarangal A, and Bertellotti G. Part 1: The Good, The Bad, and The Not-So-Ugly: Medical and Pharmacological Treatments for Spasticity http://sci.washington.edu/spasticity/index.asp.

Reynolds RM, Morton RP, Walker ML, Massagli TL, and Browd SR. Role of dorsal rhizotomy in spinal cord injury-induced spasticity. *Journal of neurosurgery Pediatrics* 14: 266-270, 2014.

Roby-Brami A, and Bussel B. Long-latency spinal reflex in man after flexor reflex afferent stimulation. *Brain : a journal of neurology* 110 (Pt 3): 707-725, 1987.

Rochcongar P, Dassonville J, and Le Bars R. [Modification of the Hoffmann reflex in function of athletic training (author's transl)]. *European journal of applied physiology and occupational physiology* 40: 165-170, 1979.

Rossi A, Mazzocchio R, and Scarpini C. Clonus in man: a rhythmic oscillation maintained by a reflex mechanism. *Electroencephalography and clinical neurophysiology* 75: 56-63, 1990.

Rothwell JC, Day BL, Berardelli A, and Marsden CD. Effects of motor cortex stimulation on spinal interneurones in intact man. *Experimental brain research* 54: 382-384, 1984.

Roy FD, Bosgra D, and Stein RB. Interaction of transcutaneous spinal stimulation and transcranial magnetic stimulation in human leg muscles. *Experimental brain research* 232: 1717-1728, 2014.

Roy RR, and Edgerton VR. Neurobiological perspective of spasticity as occurs after a spinal cord injury. *Experimental neurology* 235: 116-122, 2012.

Savic G, Bergstrom EM, Frankel HL, Jamous MA, Ellaway PH, and Davey NJ. Perceptual threshold to cutaneous electrical stimulation in patients with spinal cord injury. *Spinal cord* 44: 560-566, 2006.

Schindler-Ivens S, and Shields RK. Low frequency depression of H-reflexes in humans with acute and chronic spinal-cord injury. *Experimental brain research* 133: 233-241, 2000.

Schneider C, and Capaday C. Progressive adaptation of the soleus H-reflex with daily training at walking backward. *Journal of neurophysiology* 89: 648-656, 2003.

Scivoletto G, and Di Donna V. Prediction of walking recovery after spinal cord injury. *Brain research bulletin* 78: 43-51, 2009.

Scivoletto G, Morganti B, Ditunno P, Ditunno JF, and Molinari M. Effects on age on spinal cord lesion patients' rehabilitation. *Spinal cord* 41: 457-464, 2003.

Scivoletto G, Morganti B, and Molinari M. Neurologic recovery of spinal cord injury patients in Italy. *Archives of physical medicine and rehabilitation* 85: 485-489, 2004a.

Scivoletto G, Morganti B, and Molinari M. Sex-related differences of rehabilitation outcomes of spinal cord lesion patients. *Clin Rehabil* 18: 709-713, 2004b.

Scivoletto G, Romanelli A, Mariotti A, Marinucci D, Tamburella F, Mammone A, Cosentino E, Sterzi S, and Molinari M. Clinical factors that affect walking level and performance in chronic spinal cord lesion patients. *Spine* 33: 259-264, 2008.

Senthilvelkumar T, Magimairaj H, Fletcher J, Tharion G, and George J. Comparison of body weight-supported treadmill training versus body weight-supported overground training in people with incomplete tetraplegia: a pilot randomized trial. *Clin Rehabil* 29: 42-49, 2015.

Sezer N, Akkus S, and Ugurlu FG. Chronic complications of spinal cord injury. *World journal of orthopedics* 6: 24-33, 2015.

Shahani BT, and Young RR. Human flexor reflexes. *Journal of neurology, neurosurgery, and psychiatry* 34: 616-627, 1971.

Sharp KG, Gramer R, Butler L, Cramer SC, Hade E, and Page SJ. Effect of overground training augmented by mental practice on gait velocity in chronic, incomplete spinal cord injury. *Archives of physical medicine and rehabilitation* 95: 615-621, 2014.

Shemesh Y, Rozin R, and Ohry A. Electrodiagnostic investigation of motor neuron and spinal reflex arch (H-reflex) in spinal cord injury. *Paraplegia* 15: 238-244, 1977.

Siddall PJ, Cousins MJ, Otte A, Griesing T, Chambers R, and Murphy TK. Pregabalin in central neuropathic pain associated with spinal cord injury: a placebo-controlled trial. *Neurology* 67: 1792-1800, 2006.

Simpson LA, Eng JJ, Hsieh JT, Wolfe, and the Spinal Cord Injury Rehabilitation Evidence Research Team DL. The health and life priorities of individuals with spinal cord injury: a systematic review. *Journal of neurotrauma* 29: 1548-1555, 2012.

Sinkjaer T, Andersen JB, Ladouceur M, Christensen LO, and Nielsen JB. Major role for sensory feedback in soleus EMG activity in the stance phase of walking in man. *The Journal of physiology* 523 Pt 3: 817-827, 2000.

Sinkjaer T, Andersen JB, and Larsen B. Soleus stretch reflex modulation during gait in humans. *Journal of neurophysiology* 76: 1112-1120, 1996.

Sipski ML, Jackson AB, Gomez-Marin O, Estores I, and Stein A. Effects of gender on neurologic and functional recovery after spinal cord injury. *Archives of physical medicine and rehabilitation* 85: 1826-1836, 2004.

Skold C, Levi R, and Seiger A. Spasticity after traumatic spinal cord injury: nature, severity, and location. *Archives of physical medicine and rehabilitation* 80: 1548-1557, 1999.

Smith AC, Mummidisetty CK, Rymer WZ, and Knikou M. Locomotor training alters the behavior of flexor reflexes during walking in human spinal cord injury. *Journal of neurophysiology* 112: 2164-2175, 2014.

Soffe SR. Two distinct rhythmic motor patterns are driven by common premotor and motor neurons in a simple vertebrate spinal cord. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 13: 4456-4469, 1993.

Spungen AM, Asselin P, Fineberg DB, Harel NY, Kornfeld S, and Bauman WA. Beneficial changes in body composition after exoskeletal-assisted walking: Implications for improved metabolic function. *Topics in Spinal Cord Injury Rehabilitation* 19: 8-9, 2013a.

Spungen AM, Asselin P, Fineberg DB, Kornfeld S, and Harel NY. Exoskeletal-assisted walking for persons with motor-complete paraplegia. In: *Force Sustainment: Rehabilitation, Regeneration and Prosthetics for Re-Integration to Duty Meeting Proceedings STO-MP-HFM-228, Paper 6* Neuilly-sur-Seine, France: Science and Technology Organization, 2013b, p. 6-1 - 6-14.

Spungen AM, Asselin PK, Fineberg DB, Kornfeld SD, and Harel NY. Exoskeletal-assisted walking for persons with motor-complete paraplegia. In: *North Atlantic Treaty Organization (NATO), Research and Technology Organization, Human Factors and Medicine Panel, HFM-228*. Italy: 2013c.

Stampacchia G, Rustici A, Bigazzi S, Gerini A, Tombini T, and Mazzoleni S. Walking with a powered robotic exoskeleton: Subjective experience, spasticity and pain in spinal cord injured persons. *NeuroRehabilitation* 39: 277-283, 2016.

Stein RB, Estabrooks KL, McGie S, Roth MJ, and Jones KE. Quantifying the effects of voluntary contraction and inter-stimulus interval on the human soleus H-reflex. *Experimental brain research* 182: 309-319, 2007.

Stein RB, Hayday F, Chong S, Thompson AK, Rolf R, James KB, and Bell G. Speed and efficiency in walking and wheeling with novel stimulation and bracing systems after spinal cord injury: a case study. *Neuromodulation* 8: 264-271, 2005.

Stein RB, and Kearney RE. Nonlinear behavior of muscle reflexes at the human ankle joint. *Journal of neurophysiology* 73: 65-72, 1995.

Stein RB, and Thompson AK. Muscle reflexes in motion: how, what, and why? *Exercise and sport sciences reviews* 34: 145-153, 2006.

Stephens MJ, and Yang JF. Loading during the stance phase of walking in humans increases the extensor EMG amplitude but does not change the duration of the step cycle. *Experimental brain research* 124: 363-370, 1999.

Stephens MJ, and Yang JF. Short latency, non-reciprocal group I inhibition is reduced during the stance phase of walking in humans. *Brain Res* 743: 24-31, 1996.

Suzuki M, Miyai I, Ono T, Oda I, Konishi I, Kochiyama T, and Kubota K. Prefrontal and premotor cortices are involved in adapting walking and running speed on the treadmill: an optical imaging study. *Neuroimage* 23: 1020-1026, 2004.

Sykes L, Edwards J, Powell ES, and Ross ER. The reciprocating gait orthosis: long-term usage patterns. *Archives of physical medicine and rehabilitation* 76: 779-783, 1995.

Talaty M, Esquenazi A, and Briceno JE. Differentiating ability in users of the ReWalk(TM) powered exoskeleton: an analysis of walking kinematics. *IEEE International Conference on Rehabilitation Robotics : [proceedings]* 2013: 6650469, 2013.

Taylor S, Ashby P, and Verrier M. Neurophysiological changes following traumatic spinal lesions in man. *Journal of neurology, neurosurgery, and psychiatry* 47: 1102-1108, 1984.

Thomas SL, and Gorassini MA. Increases in corticospinal tract function by treadmill training after incomplete spinal cord injury. *Journal of neurophysiology* 94: 2844-2855, 2005.

Trimble MH, Behrman AL, Flynn SM, Thigpen MT, and Thompson FJ. Acute effects of locomotor training on overground walking speed and H-reflex modulation in individuals with incomplete spinal cord injury. *The journal of spinal cord medicine* 24: 74-80, 2001.

Trimble MH, Kukulka CG, and Behrman AL. The effect of treadmill gait training on low-frequency depression of the soleus H-reflex: comparison of a spinal cord injured man to normal subjects. *Neurosci Lett* 246: 186-188, 1998.

Ung RV, Imbeault MA, Ethier C, Brizzi L, and Capaday C. On the potential role of the corticospinal tract in the control and progressive adaptation of the soleus h-reflex during backward walking. *Journal of neurophysiology* 94: 1133-1142, 2005.

Valls-Sole J, Alvarez R, and Tolosa ES. Vibration-induced presynaptic inhibition of the soleus H reflex is temporarily reduced by cortical magnetic stimulation in human subjects. *Neuroscience letters* 170: 149-152, 1994.

van der Laan L, Boks LM, van Wezel BM, Goris RJ, and Duysens JE. Leg muscle reflexes mediated by cutaneous A-beta fibres are normal during gait in reflex sympathetic dystrophy. *Clin Neurophysiol* 111: 677-685, 2000.

van Hedel HJ, and Group ES. Gait speed in relation to categories of functional ambulation after spinal cord injury. *Neurorehabil Neural Repair* 23: 343-350, 2009.

van Hedel HJ, Wirz M, and Dietz V. Assessing walking ability in subjects with spinal cord injury: validity and reliability of 3 walking tests. *Archives of physical medicine and rehabilitation* 86: 190-196, 2005.

van Middendorp JJ, Hosman AJ, Donders AR, Pouw MH, Ditunno JF, Jr., Curt A, Geurts AC, Van de Meent H, and Group E-SS. A clinical prediction rule for ambulation outcomes after traumatic spinal cord injury: a longitudinal cohort study. *Lancet* 377: 1004-1010, 2011.

Van Wezel BM, Ottenhoff FA, and Duysens J. Dynamic control of location-specific information in tactile cutaneous reflexes from the foot during human walking. *The Journal of neuroscience : the official journal of the Society for Neuroscience* 17: 3804-3814, 1997.

Varoqui D, Niu X, and Mirbagheri MM. Ankle voluntary movement enhancement following robotic-assisted locomotor training in spinal cord injury. *Journal of neuroengineering and rehabilitation* 11: 46, 2014.

Vilensky JA, and O'Connor BL. Stepping in nonhuman primates with a complete spinal cord transection: old and new data, and implications for humans. *Annals of the New York Academy of Sciences* 860: 528-530, 1998.

Visintin M, and Barbeau H. The effects of body weight support on the locomotor pattern of spastic paretic patients. *The Canadian journal of neurological sciences Le journal canadien des sciences neurologiques* 16: 315-325, 1989.

Wainberg M, Barbeau H, and Gauthier S. The effects of cyproheptadine on locomotion and on spasticity in patients with spinal cord injuries. *Journal of neurology, neurosurgery, and psychiatry* 53: 754-763, 1990.

Wallace DM, Ross BH, and Thomas CK. Characteristics of lower extremity clonus after human cervical spinal cord injury. *Journal of neurotrauma* 29: 915-924, 2012.

Walsh EG. Clonus: beats provoked by the application of a rhythmic force. *Journal of neurology, neurosurgery, and psychiatry* 39: 266-274, 1976.

Waters RL, Adkins R, Yakura J, and Vigil D. Prediction of ambulatory performance based on motor scores derived from standards of the American Spinal Injury Association. *Arch Phys Med Rehabil* 75: 756-760, 1994.

Wernig A, Nanassy A, and Muller S. Laufband (treadmill) therapy in incomplete paraplegia and tetraplegia. *Journal of neurotrauma* 16: 719-726, 1999.

Winchester P, Smith P, Foreman N, Mosby JM, Pacheco F, Querry R, and Tansey K. A prediction model for determining over ground walking speed after locomotor training in persons with motor incomplete spinal cord injury. *The journal of spinal cord medicine* 32: 63-71, 2009.

Wirz M, van Hedel HJ, Rupp R, Curt A, and Dietz V. Muscle force and gait performance: relationships after spinal cord injury. *Archives of physical medicine and rehabilitation* 87: 1218-1222, 2006.

Wirz M, Zemon DH, Rupp R, Scheel A, Colombo G, Dietz V, and Hornby TG. Effectiveness of automated locomotor training in patients with chronic incomplete spinal cord injury: a multicenter trial. *Archives of physical medicine and rehabilitation* 86: 672-680, 2005.

Wittkampf K, van Ravesteijn H, Baas K, van de Hoogen H, Schene A, Bindels P, Lucassen P, van de Lisdonk E, and van Weert H. The accuracy of Patient Health Questionnaire-9 in detecting depression and measuring depression severity in high-risk groups in primary care. *General hospital psychiatry* 31: 451-459, 2009.

Yamaguchi T. Muscle activity during forelimb stepping in decerebrate cats. *The Japanese journal of physiology* 42: 489-499, 1992.

Yang A, Asselin P, Knezevic S, Kornfeld S, and Spungen AM. Assessment of In-Hospital Walking Velocity and Level of Assistance in a Powered Exoskeleton in Persons with Spinal Cord Injury. *Top Spinal Cord Inj Rehabil* 21: 100-109, 2015.

Yang JF, and Musselman KE. Training to achieve over ground walking after spinal cord injury: a review of who, what, when, and how. *The journal of spinal cord medicine* 35: 293-304, 2012.

Yang JF, Musselman KE, Livingstone D, Brunton K, Hendricks G, Hill D, and Gorassini M. Repetitive mass practice or focused precise practice for retraining walking after incomplete spinal cord injury? A pilot randomized clinical trial. *Neurorehabilitation and neural repair* 28: 314-324, 2014.

Yang JF, Norton J, Nevett-Duchcherer J, Roy FD, Gross DP, and Gorassini MA. Volitional muscle strength in the legs predicts changes in walking speed following locomotor training in people with chronic spinal cord injury. *Physical therapy* 91: 931-943, 2011.

Yang JF, and Stein RB. Phase-dependent reflex reversal in human leg muscles during walking. *Journal of neurophysiology* 63: 1109-1117, 1990.

Yang JF, Stein RB, and James KB. Contribution of peripheral afferents to the activation of the soleus muscle during walking in humans. *Experimental brain research* 87: 679-687, 1991.

Yang JF, Stephens MJ, and Vishram R. Infant stepping: a method to study the sensory control of human walking. *The Journal of physiology* 507 (Pt 3): 927-937, 1998a.

Yang JF, Stephens MJ, and Vishram R. Transient disturbances to one limb produce coordinated, bilateral responses during infant stepping. *Journal of neurophysiology* 79: 2329-2337, 1998b.

Yates C, Charlesworth A, Allen SR, Reese NB, Skinner RD, and Garcia-Rill E. The onset of hyperreflexia in the rat following complete spinal cord transection. *Spinal cord* 46: 798-803, 2008.

Zehr EP. Training-induced adaptive plasticity in human somatosensory reflex pathways. *Journal of applied physiology (Bethesda, Md : 1985)* 101: 1783-1794, 2006.

Zehr EP, Komiyama T, and Stein RB. Cutaneous reflexes during human gait: electromyographic and kinematic responses to electrical stimulation. *Journal of neurophysiology* 77: 3311-3325, 1997.

Zehr EP, and Stein RB. What functions do reflexes serve during human locomotion? *Progress in neurobiology* 58: 185-205, 1999.

Zehr EP, Stein RB, and Komiyama T. Function of sural nerve reflexes during human walking. *The Journal of physiology* 507 (Pt 1): 305-314, 1998.

Zeilig G, Weingarden H, Zwecker M, Dudkiewicz I, Bloch A, and Esquenazi A. Safety and tolerance of the ReWalk exoskeleton suit for ambulation by people with complete spinal cord injury: a pilot study. *The journal of spinal cord medicine* 35: 96-101, 2012.

Zewdie ET, Roy FD, Yang JF, and Gorassini MA. Facilitation of descending excitatory and spinal inhibitory networks from training of endurance and precision walking in participants with incomplete spinal cord injury. *Prog Brain Res* 218: 127-155, 2015.