

NEURAL MECHANISMS OF FATIGUE IN HUMANS

by

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Abstract

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Neural Mechanisms of Fatigue in Humans

Thesis directed by Professor Roger M. Enoka

In the context of human performance, the term fatigue describes a variety of phenomena that limit the ability of an individual to perform a given task. Fatigue is a symptom experienced by all humans; however, the magnitude of the effect depends on such factors as the specificity of the task being performed, age, handedness, and neurological health. This dissertation examined some of the mechanisms that contribute to both perceived fatigability and performance fatigability and how they are influenced by varying acute and chronic adaptations. The first study compared [^{18}F]-FDG uptake by skeletal muscles and the amount of muscle activity as indicated by surface electromyography when young and old men performed two types of fatiguing contractions. The results indicated that young men were able to modulate muscle activation during the two tasks but old men were not. The second study compared changes in the modulation of sensory feedback in young, middle-aged, and old adults before and after practicing a task that involved the contraction of a pair of antagonistic muscles. The results highlighted the impact of fatigue on the ability to modulate afferent feedback and control muscle force across the three age groups and suggested that declines in nervous system function precede the decrease in motor function and these begin in middle age. The purpose of the third study was to compare activity of the same motor units in left-handed individuals during sustained contractions supporting two load types. This study showed that handedness does not influence the activity of single motor units during these types of fatiguing contractions. The fourth study

investigated fatigue in individuals with multiple sclerosis. This study showed no differences in force declines following a set of fatiguing contractions when compared with control subjects, however, reported levels of fatigue were significantly greater for the multiple sclerosis group. This study also indicated that intellectual capacity to be protective against declines in neuromuscular function. The results of this dissertation underscore the concept that the physiological mechanisms limiting human performance during fatiguing contractions are influenced by factors such as the specificity of the task being performed, age, and neurological health.

Dedication

This dissertation is dedicated to my Mom and Dad
for their continuous love, support, and encouragement.

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Chapter I

Review of the Literature

Introduction

In the context of human physical performance, the term fatigue is generally used to describe a wide variety of phenomena that constrain the ability of an individual to perform a given task. Fatigue is a symptom experienced by all humans, and while it has presumably evolved as a protective homeostatic mechanism to prevent overexertion, injury, and catastrophic failure (Noakes, 2012; Smirmaul, 2012; O'Connor, 2013), it is also detrimental to the quality of life in individuals with an array of neurological diseases. Additionally, otherwise healthy individuals routinely experience levels of fatigue that negatively impact daily life (Kroenke, 1988; Lewis and Wessely, 1992). The manifestation of fatigue in an individual may be influenced by factors such as the specificity of the task being performed, the age and sex of the individual, training status, and neurological health.

The topic of fatigue has been reviewed thoroughly from several different perspectives ranging from changes that occur in the brain and spinal cord, to cellular and molecular changes that occur in the muscle (Enoka and Stuart, 1992; Gandevia, 2001; Allen et al., 2008; Noakes, 2012). This review aims to summarize the topic by providing a brief historical overview of the study of fatigue and how the field has progressed over time, a description of the possible mechanisms that contribute to the manifestation of fatigue in healthy individuals, as well as those that may contribute to fatigue in individuals with multiple sclerosis.

Historical perspective on the study of fatigue

This historical overview is not intended to serve as an exhaustive review on the literature of fatigue. Rather, it is intended to highlight some of the key thought-leaders whose innovative ideas and contributions were instrumental in guiding the direction of the field. Since the early

works of Angelo Mosso, fatigue was approached as a complex, multi-dimensional concept. In an effort to simplify and better understand the many factors that contribute to fatigue, mainstream physiologists began attempting to classify discrete types of fatigue. This approach was helpful to an extent, however it became clear that this approach came with severe limitations, as it 1) gave rise to vague, confusing terminology in the literature such as central fatigue, peripheral fatigue, mental fatigue, cognitive fatigue, etc. and 2) tends to ignore interactions between subdomains of fatigue. Brenda Bigland-Ritchie, for example, was one of those who reintroduced the idea that performance fatigability could be caused at several different sites within the nervous system, the neuromuscular junction, and the muscle (Bigland-Ritchie and Woods, 1984). This more inclusive approach has important implications for better understanding the underlying physiology of fatigue in healthy individuals and those with neurologic disorders.

The Beginning

Angelo Mosso (1846-1910) was one of the first physiologists to systematically study fatigue. As a clinician and a scientist, Mosso developed his interests during a time when public health issues related to manual labor and industrial work-related accidents were becoming more prevalent, and he attributed many of these problems to fatigue. Mosso also identified fatigue as an important factor that limited the performance of soldiers in the field, contributed to infant mortality, and stunted the healthy growth and development of children.

When reflecting back on the history of the study of fatigue, and considering the current state of the field, one of the most impressive aspects of Mosso's contributions was his awareness (or at least speculation) that fatigue comprises several components and most notably that those

components may interact to influence the fatigue experienced by an individual. Mosso distinguished between two major domains of fatigue; performance fatigability and sensations of fatigability.

“On an examination of what takes place in fatigue, two series of phenomena demand our attention. The first is the diminution of the muscular force. The second is fatigue as a sensation. That is to say, we have a physical fact, which can be measured and compared, and a psychic fact, which eludes measurement.”

-Mosso, 1904 (pg. 154)

Mosso used an ergograph to study performance fatigability in humans. His device recorded the movement of the middle finger during repeated anisometric contractions performed to failure. In addition to showing the expected declines in finger displacement with repeated muscle contractions, his experiments also demonstrated that the recorded displacements of the finger yielded fatigue profiles that differed between individuals. For example, he observed that factors such as age, physical training status, and varying conditions of the task could influence task performance.

Another important contribution from Mosso was the recognition that fatigability may arise due to changes in either the muscle or the nervous system. In an experiment involving the exposed muscle of a frog, Mosso noted that irrigating the muscle with saline helped to resist reductions in force production, concluding that fatigue can occur due to the accumulation of waste product in the muscle (Mosso, 1904). Additionally, he extended Borelli’s hypothesis that fatigue involved the nervous system. He demonstrated in humans that performance fatigability involved

impairment of the activation signal sent by the nervous system in addition to impairment of the muscle itself. This conclusion was reached by observing differences recorded by an ergograph between voluntary muscle contractions and those elicited by electrical stimulation (Di Giulio et al., 2006).

Mosso also acknowledged the concept of fatigue as a sensation. These sensations of fatigue, he noted, could be induced by continued physical activity or by engaging in a prolonged cognitive task. For example, Mosso showed the effect of perceived fatigue induced by a cognitive task on performance by having one of his colleagues perform anisometric fatiguing contractions to task failure while the work performed was recorded by an ergograph. The following day, the task was repeated, but this time after the colleague had delivered a lecture, which served as a cognitive challenge. The amount of work performed by Mosso's colleague during the muscle contractions was reduced following the lecture suggesting an effect of a cognitive task on performance fatigability. Moreover, he examined the same relation in the opposite direction and found that intense physical activity resulted in impaired thinking and memory in students, lending to the idea that sensations of fatigue and performance fatigability may influence one another.

Even today our understanding of fatigue is far from complete. However, Mosso's integrative approach to the study of fatigue has established a rather comprehensive foundation for further study by acknowledging the multiple factors contributing to fatigue and considering those factors in the context of public health. This type of approach was abandoned for some time in favor of more reductionist approaches in an effort to better understand the many individual factors contributing to fatigue. However, physiologists have since realized the limitations involved with such approaches and have been working toward a more integrative understanding of fatigue.

Voluntary activation

In 1928, the idea of using electrical stimulation to study the activation of muscle by the nervous system was expanded upon by Denny-Brown in his studies of spinal reflexes. Subsequently, Merton (1954) adapted this electrical stimulation technique, known as the twitch occlusion method, in an effort to distinguish the contributions of the nervous system and muscle to observed declines in force. Merton showed that the amplitude of the muscle twitch force was inversely proportional to the level of force being produced during a voluntary contraction. Moreover, Merton did not observe any additional force induced by the applied stimulus during maximal voluntary contractions performed with the adductor pollicis muscle. He interpreted these observations as indicating that the artificial twitch imposed on the muscle was occluded by the action potentials due to voluntary drive from the central nervous system. He therefore concluded that voluntary drive to the motor neuron pool and the activation signal to the muscle was adequate, and not a limiting factor in maximal force production. These findings seemed to exclude the nervous system as a contributor to performance fatigability, at least during maximal voluntary efforts.

Lactic acid

The idea that performance fatigability was due to changes in the muscle was expanded by Fletcher and Hopkins (1907), and A.V. Hill (1932). They observed a relation between the accumulation of lactic acid in stimulated skeletal muscle and declines in performance of the muscle, particularly in the absence of oxygen. It was later demonstrated that inhalation of oxygen enhanced performance during exercise, an effect that was ascribed to increased oxygen availability to allow for an increase in cardiac output to the muscle. Their interpretation was that

active skeletal muscle produced lactic acid in response to high intensity exercise during which there was a deficit of oxygen (Hill and Flack, 1910), and it was the resulting accumulation of lactic acid that caused performance fatigability. Today, it is known that this is not the case, but at that time, these studies strongly influenced the focus of research on fatigue.

Interestingly, Hill and Flack (1910) made anecdotal notes on how subjects reported ‘the greater ease with which the work was done’ after oxygen inhalation. While this observation hinted at the interaction between performance fatigability and perceived fatigability, the role of perceptions in influencing fatigue were largely cast aside at the time.

These studies and many others made a significant contribution to investigating fatigability and the changes that occurred within the skeletal muscle itself. However, greater inquiries into the role of the nervous system in fatigue came later, in large part to the efforts of Bigland-Ritchie, who was an advocate for the idea that multiple sites along the neuromuscular pathway could contribute to processes involved with declines in the force generating capacity of a muscle.

Task dependency

Brenda Bigland-Ritchie made seminal contributions to our understanding of performance fatigability (Thomas et al., 1995). One of the more notable features of her work was that performance fatigability, or an exercise induced decline in force production, results from limitations at several possible sites in the central nervous system, the neuromuscular junction, and in the muscle. Moreover, she acknowledged that functional changes could occur at all of these sites from the onset of a task but may vary as the task progresses depending on the muscle group being used and the type of activity being performed (Bigland-Ritchie and Woods, 1984).

In addition to Bigland-Ritchie reintroducing the idea that there may be multiple concurrent sites of fatigue within the neuromuscular system, further progress was made by acknowledging the contribution of perceived fatigability and that the site of fatigue depended on the particular task being performed; a concept often referred to as task-dependency (Enoka and Stuart, 1992).

Performance fatigability, or more specifically, a reduction in the ability to maintain an expected force or achieve a maximal force, can occur due to 1) declines in the ability of the central nervous system to generate and send an adequate command signal to the spinal motor neurons, 2) changes in the activation pattern of muscles and motor units, 3) signal transmission across the neuromuscular junction, and 4) a variety of changes occurring at the level of the muscle fiber including blood flow, excitation-contraction coupling, and the availability of substrate to produce ATP (Enoka and Stuart, 1992; Kluger et al., 2013). The acute adjustments made along the neuromuscular system depend on the details of the task being performed, such as the intensity or force level, the duration of the task, whether or not the task involves isometric or anisometric force production, and the neural strategy required to perform the task (force control versus position control).

Perceived fatigability and performance fatigability in healthy individuals

The integration of these ideas and considering fatigue as a multidimensional construct has led to a contemporary model of fatigue (Kluger et al., 2013) (Figure 1). Furthermore, advancements in characterizing perceived fatigability have been made by considering both trait levels of fatigue, which are quantified by assessing the frequency of usually occurring feelings of fatigue, and state levels of fatigue, which are quantified by assessing the intensity of currently occurring feelings of fatigue during a discrete point in time (O'Connor, 2013).

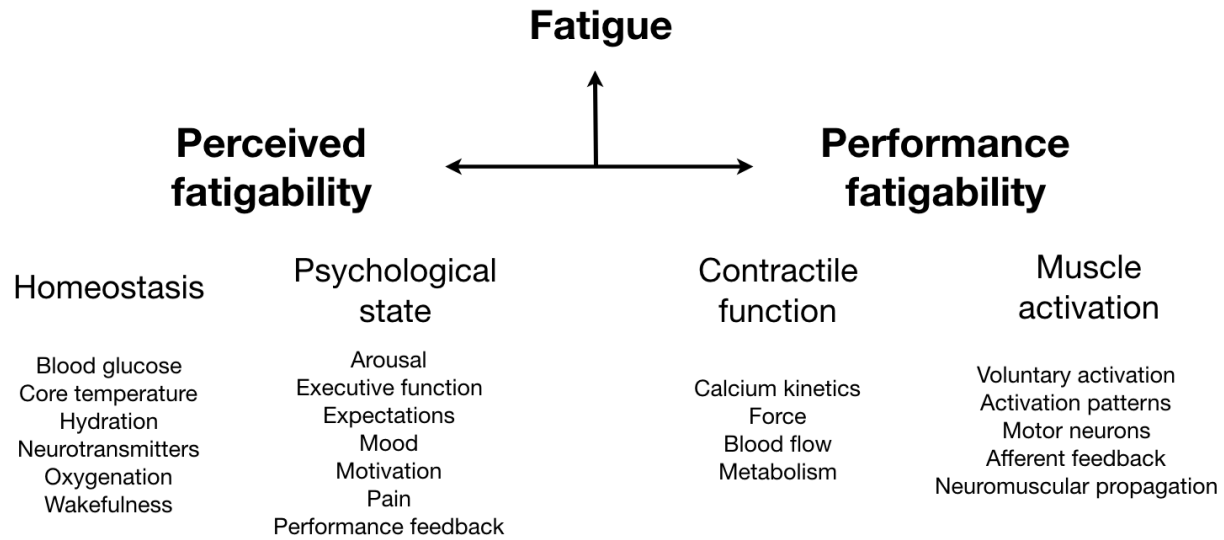


Figure 1. A contemporary model of fatigue. Fatigue is a self-described symptom that manifests from two domains; perceived fatigability, and performance fatigability. The major factors contributing to each of the domains are shown.

The following examples will introduce the idea of perceived fatigue and task dependency of performance fatigability, with an emphasis on varying force levels. Most tasks that induce fatigability, regardless of whether they are performed at high or low intensities, will involve changes at multiple sites within the neuromuscular system. However, performance during tasks at higher forces tend to be limited by changes occurring in the muscle, whereas those performed at lower forces and longer durations tend to be limited by changes in the activation signal provided by the nervous system.

Perceived fatigability

Perceived fatigability can refer to increasing sense of effort, subjective sensations of weariness, a mismatch between expended effort and actual performance, and exhaustion (Kluger et al., 2013). A consequence of performing a fatiguing contraction with one limb, for example, is an altered ability to estimate the force being produced by that limb using the non-fatigued contralateral

limb. In an experiment during which individuals performed a force-matching task with the elbow flexor muscles (McCloskey et al., 1974), a reference load of 4 kg was supported with one arm. Experimenters then adjusted the load in the other arm as requested by the subject to ‘match’ the load in the reference arm. During the fatiguing contraction, the subjects maintained the load with the reference arm for 10 min, and subjects again were asked to match the load in the other arm. Despite the reference load remaining constant, the subjects chose larger loads as the task progressed to indicate that the development of performance fatigability was accompanied by a progressive increase in perceptions of fatigability.

Perceived fatigability are particularly difficult to define given their inherently subjective nature, and that the literature contains a sizable range of terms and definitions, such as cognitive fatigue, sleepiness, or lassitude, which overlap with the concept of perceived effort. In an effort to quantify perceived fatigability, category-rating scales have been developed in which verbal expressions are anchored to numerical positions on a scale. For example, in Borg’s RPE scale, a 6 corresponds to ‘no exertion at all’, a 13 corresponds to ‘somewhat hard’, and a 20 corresponds to ‘maximal exertion’ (Borg, 1990). By anchoring numbers to well-defined levels of perception, such as at the highest and lowest parts of the scale, ratings can be compared across individuals or groups. Ratings of perceived exertion have been shown to correlate with increases in heart rate and blood lactate (Borg et al., 1987), and have thus been adopted as a useful tool to assess exertion and perceived effort during exercise.

Performance fatigability

Performance fatigability refers to the rate or magnitude of decline in an objective measure of motor performance, and occurs due to an inability for physiological adjustments to meet the demands of a task (Enoka, 2013).

The rate-limiting adjustment during a fatiguing contraction varies depending on the level of force being produced by the individual. In a similar approach used by earlier investigators to assess voluntary activation, Gandevia et al. (1996) used motor point stimulation and transcranial magnetic stimulation (TMS) during maximal voluntary contractions of the elbow flexor muscles. In the first set of experiments, the contractions were sustained for 3 min and maximal voluntary contraction force declined to 26% of its initial value. Motor point stimulation was used before, during, and after the contractions to identify the site of fatigability. The twitches used immediately following the contraction could only produce 29% of the force generated by the twitches preceding the contraction, which suggests that the fatigability experienced is due mainly to changes occurring in the muscle. However, twitches imposed during the contraction showed a progressive increase in amplitude, suggesting that the contractions were accompanied by reductions in the activation signal from the nervous system to the muscle. In a second set of experiments using the same subjects, TMS was applied intermittently throughout the contractions to activate the cortical motor neurons involved in activating the elbow flexor muscles. As with the motor point stimulation, the superimposed TMS resulted in progressive increases in the force evoked by the artificial activation. At the beginning of the contraction, TMS was able to increase force production by 1%, suggesting that the voluntary activation produced by most of the subjects was adequate to fully activate the elbow flexor muscles. However, by the end of the contraction the applied stimulus increased force production by ~10%. These experiments together show that fatigability induced by contractions sustained at higher

forces involve modest declines in voluntary activation and that most of the loss in force can be attributed to changes in muscle function.

Later experiments by Sogaard et al. (2006), examined voluntary activation during longer submaximal contractions. Subjects performed sustained contractions at 15% MVC for 43 min. Following the protocol, MVC force declined to 58% of the initial value. Resting twitch values elicited by peripheral nerve stimulation declined to 59% of the initial value, again suggesting that changes in the muscle contributed to the declines in MVC force, but to a lesser extent than that seen following sustained maximal contractions. Furthermore, superimposed twitches provided during the contraction by both nerve stimulation and TMS, showed that voluntary activation declined from 98% to 72% and from 98% to 77%, respectively. These findings highlight that performance fatigability, in this case reductions in maximal voluntary force, induced by lower force contractions involved smaller declines in muscle function and greater reductions in muscle activation by the central nervous system.

While many variables, both in the muscle and the nervous system, may contribute to declines in force and time to task failure, another example that distinguishes between contractions of differing target force is the pattern with which the nervous system activates the synergist muscles involved in a given task. Kouzaki et al. (2002) have shown muscle activity, as measured by EMG recordings, to alternate between three knee extensor muscles during low force, but not higher force sustained contractions. Subjects performed sustained contractions at 2.5, 5, 7.5 and 10% MVC force. The lower two target forces, which were sustained for 60 min, showed alternating EMG activity between the rectus femoris and the vastus lateralis and medialis muscles, and these alternations increased in frequency as the task progressed. This pattern of activity was not seen at the higher two forces. While there is considerable between-subject

variability in the frequency of alternating activity during these low-force contractions, individuals exhibiting a greater number of alternations showed smaller reductions in MVC force, suggesting that the nervous system may alternate the pattern of muscle activation as a strategy to improve task performance (Kouzaki and Shinohara, 2006).

Target force also influences the strategy employed by the nervous system during fatiguing contractions that require individuals to support loads of varying compliance. For example, it has been shown that endurance times are briefer when a compliant load is supported versus a noncompliant load, despite the net torque about the joint being identical for the two tasks (Hunter et al., 2005; Rudroff et al., 2005). Interestingly, this difference is observed for lower force contractions, whereas in higher force contractions, the endurance times are similar (Maluf and Enoka, 2005). Because not all of the available motor units have been recruited at the onset of a low force contraction, the nervous system may recruit more motor units or increase the discharge rate of active units to maintain a desired submaximal target force. The greater decline in single motor unit discharge rate when supporting a compliant load (Mottram et al., 2005) lends support to the idea that motor units are recruited more rapidly during this task, thus leading to a briefer endurance time. However, when higher forces are used to support both compliant and noncompliant loads, the endurance times are similar (Maluf et al., 2005). When target forces are at or above the upper limit of motor unit recruitment, all of the motor units are active from the start of the task. Therefore, performance fatigability, as indicated by the endurance time to maintain a submaximal target force, is modulated by the ability of the nervous system to recruit motor units.

Blood flow to a muscle during a contraction is another important factor that is influenced by the force being exerted during a task. de Ruiter et al. (2007) measured muscle oxygenation during

brief isometric contractions of the knee extensor muscles that ranged in intensity from 20-45% MVC force in 5% increments. Near-infrared spectroscopy was used to measure the relative levels of oxyhemoglobin and deoxyhemoglobin. Each contraction intensity was performed twice; once with a blood occluding cuff around the proximal thigh, and once without. The changes in oxyhemoglobin concentrations were compared under these two conditions in order to identify the force at which blood flow was completely occluded. The rectus femoris was found to be deoxygenated during contractions performed at 35% MVC force, whereas the vastus lateralis and vastus medialis muscles at 25% MVC force. When a muscle is active, increases in blood flow are necessary to maintain the required oxygen and substrate demands, as well as to remove metabolites that may reduce performance. Blood flow, therefore is thought to be adequate during low force contractions, but may contribute to fatigability at higher forces.

A substantial literature supports the idea that the level of force produced during a task influences the degree to which various adjustments within the neuromuscular system contribute to performance fatigability. It has been unclear however, whether these contributions change gradually with varying levels of force, or if there is a more discrete threshold at which the performance of a task is limited predominantly by the output of the nervous system of the contractile function of the muscle. One more recent study (Burnley et al., 2012) examined performance fatigability in the context of critical force, to suggest the existence of a threshold. Monod and Scherrer (1965) introduced the concept of critical power when they reported a hyperbolic relation between power output and endurance time. Critical power was defined as the asymptote of the hyperbola to indicate a power output that could be maintained indefinitely (Hill, 1993). Although this is not quite correct, the measurement of critical power can reasonably determine the power output at which time to task failure decreases exponentially if exceeded.

Burnley et al. (2012) used this approach to examine fatigability of the knee extensor muscles at a range of target forces above and below critical force. Times to failure for levels below critical force were sustained for 60 min, whereas those above threshold lasted from 3 to 18 min. Furthermore, trials performed above critical torque resulted in greater declines in the force producing capabilities of the muscle than for trials performed below critical force, as shown by measuring twitch force at rest. The main finding of this study was that the decline in time to task failure is not linearly proportional to target force, but rather increases rapidly when critical force is exceeded. This finding implies that there may be a discrete force level or threshold at which the sites of fatigue shift to explain these observed differences in performance, which was approximately 35% MVC force in this study.

Challenges to homeostasis can also contribute to fatigue (Kluger et al., 2013). For example, it has been shown that perturbations to core body temperature and blood glucose levels have a significant effect on performance and perceptions of fatigability.

Hyperthermia, an increase in core body temperature, has been shown to impair force production of the muscle. Increases in muscle temperature have been shown to reduce the relaxation time of the muscle twitch, which decreases muscle force by effectively shifting the force-frequency curve to the right. However, this reduction is small (~2% MVC force) compared with the reductions in output from the motor cortex as shown with TMS (Todd et al., 2005). Nybo and Nielsen (2001) also demonstrated that hyperthermia has a greater influence on the output of the central nervous system by comparing sustained (2min) MVCs of the knee extensor muscles following cycling that was performed in elevated temperatures (core temperature rose to 40 degrees C) and normal temperature environments. The MVC forces did not differ between the two groups during the first 5 s of the contraction, but declined more rapidly for the hyperthermic

group. Furthermore, declines in voluntary activation became greater throughout the duration of the MVC, as measured by stimulating the femoral nerve. The results of these studies suggest that the functional consequences of elevations in core temperature on performance fatigability are minimal in the muscle, but rather stem from a more substantial effect on the output of the nervous system during fatiguing contractions.

Another example of a challenge to homeostasis that contributes to fatigue is the decrease in blood glucose during prolonged exercise (Kluger et al., 2013). Support for this can be seen by prolonging times to failure with carbohydrate supplementation during cycling (Coyle et al., 1983). Ten trained cyclists performed two endurance tests at 70-79% $\text{VO}_{2\text{max}}$ for as long as possible. In one test they received a glucose solution, whereas in the other they received a placebo solution. Endurance time when receiving glucose supplementation was 157 min, compared with 134 min with placebo. These and other similar findings, where glucose supplementation has been shown to enhance endurance times, are typically attributed to increases in glucose availability for the muscle, or the sparing of glycogen stores in the muscle and the liver. However, it has also been demonstrated that hypoglycemia induced by prolonged cycling reduces cerebral glucose uptake, and the cerebral metabolism of oxygen, implying that performance fatigability may also arise in the brain in response to depleted levels of glucose, and not exclusively in the muscle (Nybo et al., 2003).

Performance fatigability experienced by healthy individuals may occur due to many different rate-limiting adjustments within the neuromuscular system. The factors contributing to both declines in voluntary force production and endurance time may vary depending on the activity being performed, but in healthy individuals, perceptions of fatigability often increase congruently with performance fatigability.

Perceived fatigability and performance fatigability in multiple sclerosis

Multiple sclerosis (MS) is a chronic immune-mediated disease of the central nervous system that is often disabling (Kos et al., 2008; Vucic et al., 2010). Symptoms of the disease vary from mild tingling in the limbs to more severe symptoms such as muscle spasticity, loss of vision, and paralysis (National MS Society). However, fatigue is one of the most common disabling symptoms, being reported in up to 90% of individuals with MS, with up to 65% of individuals experiencing fatigue on a daily basis (Freal et al., 1984; Fisk et al., 1994).

Fatigue in individuals with multiple sclerosis

In the early stages of disease progression, state levels of fatigue, performance fatigability, and sensations of fatigability are largely caused by changes occurring in the nervous system. In a study by Chard et al. (2004), thirteen subjects who were recently diagnosed with MS enrolled in a study 0.5 – 2.7 years following diagnosis. The subjects underwent an MRI scan at baseline and were scanned every six months for 18 months. Compared with nine healthy control subjects, the individuals with MS had lower (-5.8%) levels of white matter at baseline. However, over the course of the study it was observed that grey matter, but not white matter, declined in individuals with MS (-0.0086%/yr versus -0.0021%/yr in controls), highlighting the differences between the time course of white matter and grey matter atrophy, and that MS is a disease of the neuron and not necessarily restricted to the myelin.

Roelcke et al. (1997) characterized some of the functional changes that may contribute to fatigue by performing a study using PET imaging in 47 individuals with MS and 16 healthy control subjects to measure cerebral glucose metabolism. They reported that cerebral glucose metabolism was reduced in individuals with MS versus controls, but was not related to fatigue

severity as measured by the Fatigue Severity Scale (FSS). However, individuals with MS experiencing severe fatigue were shown to have reduced glucose metabolism in the frontal cortex and basal ganglia compared with the non-fatigued individuals with MS, which suggested dysfunction in these brain regions due to demyelination of frontal white matter. Interestingly, the individuals experiencing severe fatigue showed elevated glucose metabolism in other regions of the brain, including the cerebellar vermis and the anterior cingulate gyrus. Although it seems reasonable to speculate that a compensatory strategy is being adopted by the nervous system as the disease progresses, these brain regions are involved with attentional and cognitive tasks and this observation requires further investigation.

Other neuroimaging studies have looked for relations between the fatigue experienced by persons with MS and structural and functional measures, resulting in mixed findings. Some studies, for example, showed no relation between fatigue and brain atrophy (Bakshi et al., 1999), or lesion load (van der Werf et al., 1998). In contrast, other studies have shown a relation between the progression of brain atrophy and fatigue (Marrie et al., 2005; Tedeschi et al., 2007). This discrepancy in findings may indicate that a more rapid progression of the disease, and therefore presumably less adaptation by the nervous system, is responsible for heightened fatigue.

Two additional factors that likely play an important role in the fatigue experienced in multiple sclerosis are the changes in neuronal function due to demyelination of axons, and sensitivity to heat. In terms of the fatigue taxonomy (Figure 1), demyelination will impair the activation signal that can be sent from the spinal cord to muscle, whereas the sensitivity to heat will exacerbate challenges to homeostasis and thereby perceptions of fatigability.

Demyelination of axons that occurs with MS may lead to conduction block, which is the failure of an action potential to propagate along a structurally intact axon (Kaji, 2003). In a healthy axon, voltage-gated channels are activated at a node allowing an inward current carried by sodium to depolarize the axon, giving rise to an action potential. This current then travels down the myelinated internodal region inducing an outward capacitive current at the next node, which activates voltage-gated sodium channels. This cycle, known as saltatory conduction, is continued down the length of the axon. In a healthy axon, myelin acts to reduce capacitance and membrane conductance. However, in the case of demyelination, these properties are diminished, which results in impaired signal propagation. Conduction block can be activity dependent (Vagg et al., 1998) and may contribute to the reduced ability of the nervous system to activate skeletal muscle in individuals with multiple sclerosis.

Sensitivity to heat may also contribute to the fatigue experienced by persons with MS. While it has been shown that hyperthermia can increase performance fatigability in healthy individuals during prolonged exercise (Nybo and Nielsen, 2001; Todd et al., 2005), a debilitating worsening of symptoms, including reported symptoms of fatigue (Uhthoff's syndrome), are commonly seen in individuals with MS when exposed to heat (Vucic et al., 2009). Increases in body temperature by as little as 0.5 degrees Celsius may impair the ability for axons to effectively conduct action potentials (Rasminsky, 1973) by increasing the speed of channel gating and thereby decreasing the inward Na^+ current and the subsequent outward capacitive current at the following node (Burke et al., 2001). This is one possible mechanism to explain the temporary exacerbation of symptoms with heat.

Performance fatigability in individuals with multiple sclerosis

The consideration of task dependency is also useful in the assessment of performance fatigability in individuals with MS to identify the rate-limiting adjustments when performing various tasks. As observed in healthy individuals, the level of force produced by an individual will influence the factors contributing to fatigue, with higher target forces resulting in greater limitations in the muscle and lower forces involving in greater involvement of the nervous system.

One relevant study aimed to identify the rate-limiting adjustments during fatiguing contractions in individuals with MS using the knee extensor muscles during MVCs performed for 2 min (Skurvydas et al., 2011). This study examined maximal voluntary force, voluntary activation (250 ms train at 100Hz was superimposed on MVC) and fatigability of the muscle (stimulus used for voluntary activation applied to the resting muscle). As expected, declines in MVC force were greater for individuals with MS. However, impairment in the contractile properties of the muscle was approximately 50% of that observed in healthy controls, whereas declines in voluntary activation were ~3 times greater than that observed in healthy controls. Data for the healthy subjects were consistent with previous findings suggesting that fatigability induced by higher force contractions is mainly due to impairments in the muscle. The data for the MS subjects however, suggest a much greater involvement of the CNS as compared to declines in the contractile properties of the muscle (Skurvydas et al., 2011).

Thickbroom et al. (2006) investigated the adjustments during lower force contractions. Twenty-three subjects with MS and 15 controls produced intermittent contractions to 40% of MVC force (7 s contractions with 3 s periods of rest) for 20 min. Although healthy subjects demonstrated greater MVC forces, the declines were similar between the two groups after the task; however, the MS subjects did not show as much recovery after 10 min of rest. Ratings of perceived exertion were similar for the first 5 min, but were higher in subjects with MS for the duration of

the task. In this study, motor evoked potentials (MEPs) were elicited in the first dorsal interosseous muscle and the responses were recorded in the muscle using surface EMG. Transcranial magnetic stimulation is applied over the motor cortex to evoke a descending volley of action potentials in cortical motor neurons of a muscle. The response recorded in the muscle is used to assess the responsiveness of the pathway from the motor cortex to the muscle (Gandevia, 2001). The MEP amplitudes measured in this study, while smaller, showed a greater proportional increase in the MS group, and remained elevated for the duration of the task with little fluctuation in both groups. A similar trend was seen for the increased duration of the silent period following the MEP, which is a measurement thought to reflect changes in inhibitory and excitatory inputs to cortical motor neurons (Gandevia, 2001). These trends show that while the changes in both groups tended to be similar, the magnitude of these changes were greater in the MS subjects, suggesting that a greater increase in cortical activity is necessary to perform a similar task. These observations would also explain the more rapid increase in RPE. Moreover, there was no relation between trait levels of fatigue as reported by the modified fatigue impact scale, state levels of fatigue as reported by RPE during the task, or the declines in MVC. These findings support the idea that trait fatigue, state fatigue and performance fatigability are distinct from one another.

Evidence for the role of the nervous system in the performance fatigability experienced by individuals with MS is also supported by other studies. Rice et al. (1992) studied the knee extensor muscles in four subjects with MS and found that MVC force was 30-70% lower during isometric maximal voluntary contraction force compared with control subjects. This reduction in maximal force capacity was attributed to a decrease in the ability to activate spinal motor neurons as demonstrated by twitch interpolation, as well as atrophy of the muscle. Motor unit

firing rates were also reported to be lower in subjects with MS (8-16 Hz) during MVCs than in control subjects (24 Hz), suggesting either a reduction in descending drive to the motor neuron pool or decreased responsiveness of the motor neurons.

There has been an observation of deficits at the neuromuscular junction contributing to fatigability in individuals with MS (Patten et al., 1972). However, the authors concede that in this case study ($n = 3$), the observed deficit at the neuromuscular junction may be attributable to ‘overlap syndrome’ or the possibility of the involvement of another neurologic disease, such as myasthenia gravis. Additionally, more recent studies have confirmed that the neuromuscular junction is not impaired during exercise in MS (Kent-Braun et al., 1994; Sharma et al., 1995).

Fatigue and fatigability in individuals with multiple sclerosis

Multiple sclerosis is a chronic, debilitating neurologic disorder, with fatigue being one of the most common and disabling symptoms. Although there is generally a relation between reported sensations of fatigability and performance fatigability in healthy individuals, the relation in individuals with neurological disease, such as MS is less clear. For example, when performing isometric maximal voluntary contractions with the knee extensor muscles, 15 of the 22 individuals experiencing excessive levels of self-reported fatigue were able to produce forces within or above the range of healthy controls. Additionally, interpolated twitches resulted in little to no increase in the evoked force during maximal efforts, suggesting that the decline in performance was largely related to perceptions of effort (Stokes et al., 1988). Similarly, individuals diagnosed with chronic fatigue syndrome showed no differences from control subjects (69 vs. 68 N•m) in MVC force for the elbow flexor muscles, or voluntary activation of those muscles (99% in both groups). Subjects also performed repetitive isometric contractions at

30% MVC force for 45 min. Following the protocol, all subjects had similar declines in MVC force (~25%) and twitch force (~35%), with no significant difference between groups. Ratings of perceived exertion were also similar for the two groups at both the beginning and end of the task (Lloyd et al., 1991). While it would be reasonable to expect a greater increase in RPE in the chronic fatigue syndrome group, sensations of fatigability during exercise are thought to come about due to corollary discharge (McCloskey et al., 1974); that is, if the centrally generated motor command increases in response to the demand of a task, the subjective perception of fatigue in response to that task should increase in parallel. Further support for this idea is also seen during aerobic exercise (Morrison et al., 2008). Twelve individuals with MS and 12 healthy, age-matched and sex-matched controls performed a graded exercise test on a cycle ergometer for 8-12 min. Despite the individuals with MS reporting higher levels of fatigue (MFIS = 34) than controls ($n = 19$), no differences were observed in the rate of increase in RPE between the two groups.

One study by Steens and colleagues (2012) however, has reported that a moderate amount of the variability in perceived fatigue ($R^2 = 0.45$) can be explained by MVC force and its rate of decline during a sustained contraction (performance fatigability) of the first dorsal interosseus muscle in individuals with relapsing remitting MS. While most studies have not consistently identified such a relation in individuals with MS, this interesting observation suggests that there may be some overlap in the mechanisms responsible for these two variables, and encourages further study.

Conclusion

In healthy individuals, the relation between perceived fatigability and performance fatigability seems to suggest, perhaps erroneously, that these two subdomains of fatigue share overlapping mechanisms. However, the independence of fatigue and fatigability in individuals with neurologic disease suggests that the domains emerge from independent physiological processes. The fatigue experienced in humans, therefore, depends on the ability of the nervous system to activate skeletal muscle and the contractile properties of the muscle that influence fatigability, as well as the perceived fatigability that arise from the psychological state of the individual and the maintenance of homeostasis.

Chapter II

PET/CT imaging of age- and task-associated differences in muscle activity during fatiguing contractions

Abstract

The study compared positron emission tomography/computer tomography (PET/CT) of [^{18}F]-FDG uptake by skeletal muscles and the amount of muscle activity as indicated by surface EMG recordings when young and old men performed fatiguing isometric contractions that required either force or position control. EMG signals were recorded from thigh muscles of six young (26 ± 6 yrs) and six old men (77 ± 6 yrs) during fatiguing contractions with the knee extensors. PET/CT scans were performed immediately after task failure. Glucose uptake in 24 leg muscles, quantified as standardized uptake values, was greater for the old men after the force task and differed across tasks for the young men (force: 0.64 ± 0.3 g/ml; position: 0.73 ± 0.3 g/ml), but not the old men (0.84 ± 0.3 g/ml and 0.79 ± 0.26 g/ml) (age \times task interaction, $P < 0.001$). In contrast, the rate of increase in EMG amplitude for the agonist muscles was greater for the young men during the two contractions and there was no difference for either group of subjects in the rate of increase in EMG amplitude across the two tasks. The imaging estimates of glucose uptake indicated age- and task-dependent differences in the spatial distribution of [^{18}F]-FDG uptake by skeletal muscles during the fatiguing contractions. The findings demonstrate that PET/CT imaging of [^{18}F]-FDG uptake, but not surface EMG recordings, detected the modulation of muscle activity across the fatiguing tasks by the young men but not the old men.

Introduction

The modulation of motor unit activity during voluntary contractions varies with both the magnitude of the requisite force and the characteristics of the load against which the limb acts. When subjects performed submaximal, isometric contractions with the elbow flexor muscles, for example, the adjustments in motor unit activity to sustain the same net muscle torque differed when the wrist pulled against a rigid restraint to match a target force compared with maintaining a constant joint angle while supporting a more compliant load (Mottram et al., 2005; Rudroff et al., 2007, 2011, 2010b). The mean discharge rate of motor units in biceps brachii declined more rapidly and was accompanied by greater recruitment of additional motor units when the task required position control. Moreover, position control was associated with an increase in the variability of the motor unit discharge times, which suggested a difference between the two tasks in the amount of synaptic noise at the level of the motor neuron (Enoka, 2012). Because the amount of antagonist muscle coactivation did not differ between the two tasks in young subjects, the two control strategies required different combinations of synaptic input to sustain the same net muscle torque. Consistent with this interpretation, Baudry et al. (2009b, 2011) observed greater modulation of Ia presynaptic inhibition during position control than during force control.

Despite the different adjustments in motor unit activity during force and position control, the rates of change in concurrently recorded global measures of motor unit activity, as indicated by the amplitude of the surface electromyogram (EMG) for biceps brachii, did not differ during sustained contractions with the two control strategies (Mottram et al., 2005, Rudroff et al., 2010b). Due to the limitations of surface EMG recordings (Dideriksen et al., 2010; Farina et al., 2010; Enoka, 2012), the current study estimated the intensity and spatial distribution of muscle activity during fatiguing contractions by measuring the uptake of [^{18}F]-2-fluoro-2-deoxy-D-

glucose ($[^{18}\text{F}]$ -FDG) by skeletal muscles with positron emission tomography (PET) immediately after the contraction ended (Tashiro et al., 1999; Kemppainen et al., 2002; Fujimoto et al., 2003; Oi et al., 2003; Reinking et al., 2009; Kalliokoski et al., 2011). Because $[^{18}\text{F}]$ -FDG uptake is proportional to exercise intensity, accumulation of the radioactive tracer can be detected after the action and the spatial distribution of signal intensity used to characterize the contributions of selected muscles to the task (Pappas et al., 2001; Kemppainen et al., 2002; Fujimoto et al., 2003).

To evaluate the sensitivity of the PET measurements, the current study also compared surface EMG recordings and $[^{18}\text{F}]$ -FDG signal intensity in young and old adults during force and position control. Old adults tend to increase the amount of agonist-antagonist muscle coactivation when switching from force to position control, whereas young adults prefer to modulate afferent feedback (Baudry et al., 2009). The purpose of the current study was to compare the amount of muscle activity as indicated by PET/CT imaging of $[^{18}\text{F}]$ -FDG uptake by skeletal muscles and surface EMG recordings when young and old men performed fatiguing isometric contractions that required either force or position control.

Methods

The current study comprised two sets of experiments: 1) Endurance time experiments to establish the relative time to failure for both the force and position tasks when performed with the knee extensor muscles in a supine posture; and 2) muscle activation experiments during which the task durations were established on the first visit and the two subsequent visits were used to determine the spatial and temporal distribution of muscle activity during the two types of fatiguing contractions based on $[^{18}\text{F}]$ -FDG PET/CT imaging and surface EMG recordings. Informed consent was obtained from all participants, who reported being free from

cardiovascular and neurological disorders and participating in moderate levels of structured physical activity (2-4x/wk). The experimental procedures were approved by the Institutional Review Board at the University of Colorado Boulder and were in accordance with the *Declaration of Helsinki*.

Endurance time experiment

Endurance time was measured in three young (23 ± 4 years) and three old (72 ± 4 years) men. Each subject reported to the laboratory to perform the two types of fatiguing contractions with the knee extensors of the left leg. The two tasks required the subject to sustain an isometric contraction with the knee extensors at 25% of the maximal voluntary contraction (MVC) force for as long as possible. At this target force, blood flow is impaired but not occluded (Sadamoto et al., 1983). One fatiguing contraction required the knee extensors to pull against a rigid restraint and to match the force exerted by the leg to the target force that was displayed on a monitor (1% MVC/cm). The other fatiguing contraction required subjects to use the knee extensors to support an equivalent inertial load and to maintain the position of the leg by matching knee angle to the target displayed on the monitor ($1^\circ/\text{cm}$). The two fatiguing contractions are referred to as the force and position tasks, respectively.

Each type of fatiguing contraction was performed on a separate occasion, with one week between the two sessions. The fatiguing contractions were performed with the subject in a supine posture with the trunk-thigh angle at 3.14 rad, the left knee joint angle at 0.78 rad, and the right knee angle at 1.57 rad (Fig. 1). One strap was placed around the waist to stabilize the subject and another strap was wrapped around the ankle to connect the load to the leg. The force exerted by the leg was measured with a load cell (0-500 lb, Noraxon, Scottsdale, AZ) placed in

series with the load. The force signal was low-pass filtered (0-5 Hz) and recorded on a computer (1000 samples/s). Knee joint angle during the position task was measured with a flexible 2D goniometer sensor (Noraxon, Scottsdale, AZ) secured to the lateral aspect of the knee joint. The output of the goniometer was recorded, displayed on a monitor, and stored (1000 samples/s) on a computer. The inertial load (25% MVC force) for the position task was suspended from the ankle at the same location that the restraint was applied during the force task. The force task was terminated when the subject was not able to achieve the target force for 5 s, and the position task was ended when the subject was unable to maintain the knee angle within 0.17 rad of the target value for 5 s.

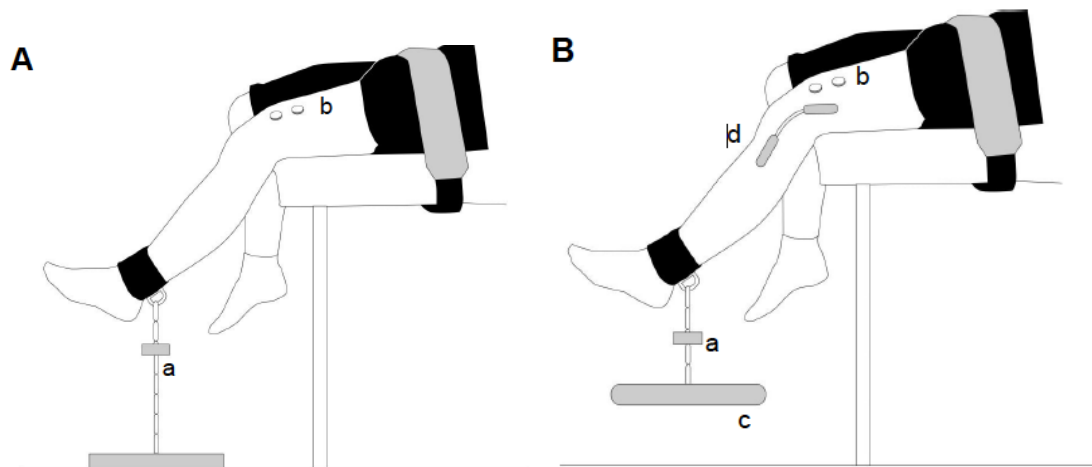


Figure 1. Experimental arrangement for the two fatiguing contractions. The subject lay in a supine position and the loads for the two types of fatiguing contractions, the force (A) and position (B) tasks, were attached to the left leg. A force transducer (a) was in series with the load during both tasks. Surface EMG electrodes (b) were placed over rectus femoris, vastus medial, vastus lateralis, and the short head of biceps femoris. The position task (B) involved supporting an inertial load (c) that was equivalent to the force exerted against the rigid restraint (A). Knee angle was measured with a goniometer (d) during the position task.

Before and immediately after each fatiguing contraction, the subject performed an MVC with the knee extensor muscles of the left leg. The initial MVC was used to determine the target force for the fatiguing contraction, and the final MVC was used to derive an index of fatigability. The MVC task comprised a 3-s increase in force from zero to maximum with the maximal force held

for ~3 s, and subjects were verbally encouraged to achieve maximal force. Subjects rested for 60 to 90 s between trials. When the peak forces achieved in two of the three trials differed by >5%, additional MVCs were performed until this criterion was met. The greatest force achieved by each subject was taken as the MVC force.

Muscle activation experiment

Six young (26 ± 6 years) and six old (77 ± 6 years) men with similar body mass (young men 77.3 ± 5.9 kg; old men 79.0 ± 6.2 kg, $P = 0.7$), who did not participate in the first experiment, were recruited for the second set of experiments and visited the laboratories on three occasions to perform MVCs and fatiguing contractions with the same approach described for the measurement of endurance time. The first visit involved determining the endurance time for the position task (Fig. 1B) with the target force set at 25% MVC. All subjects performed the position task until failure. The other two sessions involved the subject performing the two fatiguing contractions, in a randomized order, in a room adjacent to the PET/CT scanner for 90% of endurance time for the position task as determined in the first session. Immediately after the target time was achieved, the subject was placed in the PET/CT scanner to estimate [^{18}F]-FDG uptake in selected muscles.

EMG signals were recorded during the two fatiguing contractions prior to the PET/CT scans with bipolar surface electrodes (Ag-AgCl, 8-mm diameter; 20-mm distance between electrodes) that were placed over the rectus femoris, vastus medialis, vastus lateralis, and, as a representative antagonist muscle, the short head of biceps femoris (Telemyo 2400 T G2, Noraxon, Scottsdale, AZ). The electrodes were attached to the skin based on established landmarks between the

innervation zone and the end of the tendon. The EMG signals were amplified ($\times 2000$), band-pass filtered (13-1000 Hz), and recorded on a computer (2000 samples/s).

The maximal EMG for the knee extensor muscles was calculated as the average amplitude over a 0.5-s interval about the peak MVC force. The maximal EMG for the biceps femoris was calculated as the average value over a 0.5-s interval about the peak rectified EMG during maximal knee flexor activity. The maximal EMGs were recorded in the same experimental setup prior to the fatiguing contraction in each session. EMG activity during the fatiguing contractions was quantified by averaging the rectified EMG (aEMG) over the first and last 20 s of endurance time and over 20-s intervals centered about the 20, 40, 60, and 80% time points. The EMG values were normalized to the aEMG obtained during the MVC. Muscle coactivation was quantified from the EMG measurements (Falconer and Winter, 1985): $(2 \times \text{antagonist aEMG} / \text{agonist aEMG} + \text{antagonist aEMG})$.

Prior to the two sessions in which PET/CT imaging was to be performed, the subjects were required to fast for at least 4 hours, to refrain from any kind of strenuous activity for at least 1 day, and to consume water and void the bladder just before the experiment (Kemppainen et al., 2002). After the MVC had been determined, a “buffalo cap” line was placed into the antecubital vein of the right arm to deliver the tracer. A finger stick was used before the injection of [^{18}F]-FDG to determine the level of plasma glucose concentration. Approximately 2 min after the start of the fatiguing contraction, ~ 260 MBq of [^{18}F]-FDG in 5 ml of saline was infused into the vein and the sustained contractions continued thereafter for total durations of 848 ± 137 s (young men) and 751 ± 83 s (old men). Immediately after the injection of the tracer, the “buffalo cap” was removed. Once the fatiguing contraction had been sustained for the prescribed duration (90% of endurance time for the position task), the subject was moved into the scanner within 2

min and the acquisition and processing of the PET/CT images was performed following the standard protocol used in the Division of Nuclear Medicine, Department of Radiology, University of Colorado School of Medicine, Denver, CO.

The PET scans were performed with a GE Discovery ST scanner (General Electric Medical Systems, Milwaukee, WI, USA). The scanner has 24 PET detector rings of Bismuth Germinate (BGO) crystals forming 47 two-dimensional imaging planes with a sampling interval of 3.27 mm each. The PET scans were immediately preceded by CT scans for attenuation correction. Both sets of data were acquired consecutively with the subject on the same scanning table and in the same position. The feet and lower legs of the subject were secured to maintain co-registration. The lower limb was scanned from hip to feet in 2-min time frames, with 6 to 7 frames for each subject depending on the height of the individual. The data sets were reconstructed using an iterative method (OSEM) with 21 subsets and 2 iterations with a Gaussian filter. All data sets were corrected for dead-time and random coincidence. The axial and in-plane resolution of the reconstructed images was ~5 mm full-width at half maximum.

Standardized uptake values (SUV) were calculated for each muscle: $SUV = [\text{tissue radioactivity concentration} / (\text{injected dose} / \text{subject body mass})]$ (Sadato et al., 1998). Because the PET images were acquired immediately after the fatiguing contractions, the SUVs closely reflected the uptake of [^{18}F]-FDG during the sustained contractions (Kemppainen et al., 2002). Twenty-four regions of interest (ROI) were identified in the skeletal muscles of the lower limb. In the lower leg section, defined as 30% of the distance from the knee joint to the external malleolus, ROIs were located for tibialis anterior and posterior, gastrocnemius lateralis and medialis, soleus, and peroneus longus and brevis. In the thigh section, defined as 50% of the distance from the femoral head to the knee joint, ROIs were located for the knee extensors (vastus lateralis, vastus

intermedius, vastus medialis, and rectus femoris) and flexors (biceps femoris short and long head, semimembranosus, and semitendinosus). In the hip region, which was 30 mm above the femoral head, ROIs were located for adductor magnus, sartorius, gracilis, iliopsoas, tensor fasciae latae, quadratus femoris, obturator internus, and gluteus maximus, medius, and minimus. ROIs on the PET images were identified using cylinders with reference to the CT image (Fig. 2). The data were analyzed with the software package CarimasTM (Cardiac Image Analysis System), developed at the Turku PET Centre and validated by Nesterov et al. (2009).

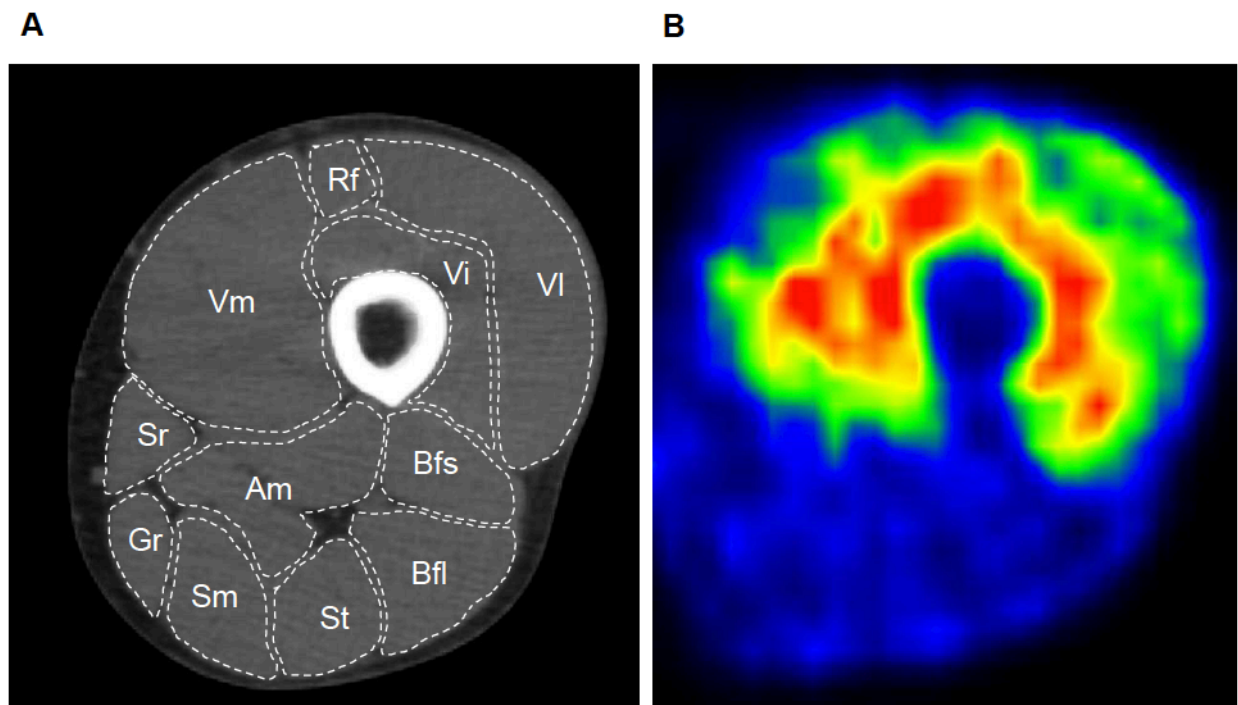


Figure 2. Transaxial CT/PET images at a mid-thigh level. Identification of leg muscles on a CT image (A) and the corresponding PET image (B) after one of the fatiguing contractions. Red denotes the greatest signal intensity followed by yellow, green, and blue. Am = adductor magnus, Bfs = biceps femoris short-head, Bfl = biceps femoris long-head, Gr = gracilis, Rf = rectus femoris, St = semitendinosus, Sm = semimembranosus, Sr = sartorius, Vi = vastus intermedius, Vl = vastus lateralis, Vm = vastus medialis.

As a translational reference for the muscle activation data, the participants wore an accelerometer (ActiGraph GT3X, Pensacola, FL) mounted at the hip to record accelerations in the vertical and horizontal directions during waking hours for seven consecutive days. The accelerometers

recorded data in 60-s intervals and also counted the number of steps. The data were downloaded onto a Microsoft Excel spreadsheet using the ActiLife software. Data recorded on the first and last days were discarded and only data sets for at least four complete days (including one weekend day) were used in the comparison, consistent with current recommendations (Mâsse et al., 2005).

Statistical analysis

The dependent variables included: endurance time for the position task, standardized uptake value for glucose in the selected muscles, and EMG activity of the principal and accessory muscles. Assessments with the Kolmogorov-Smirnov test confirmed that the distributions were normal. A two-factor, repeated-measures ANOVA (task \times age) was used to compare MVC forces and endurance times for the force and position tasks (first experiment) between young and old men. Independent t-tests were used to compare MVC forces and endurance times for the position task sustained to failure. A four-factor ANOVA (task \times age \times time \times muscle) with repeated measures on time was used to compare the changes in EMG activity and coactivation ratios. Changes in MVC force and glucose uptake were examined with a two-factor, repeated-measures ANOVA (task \times age). A repeated-measures ANOVA was also performed to test the significance of differences in glucose uptake between muscles. After a significant F -test, pairwise differences were identified using paired and unpaired t-test with Bonferroni corrections as post-hoc tests. Stepwise, linear regression analysis using forward selection was performed to examine the associations between physical activity levels (average steps/day) and SUVs. The coefficient of determination (adjusted R^2) was used to evaluate the fit of the model and guided the stepwise selection procedure. The significance level was set at $P < 0.05$. Statistical analyses

were performed with SPSS software (SPSS version 17.0). Data are reported as means \pm SD within text and tables and displayed as means \pm SEM in figures.

Results

The knee extensor MVC forces of the six men who participated in the first experiment were 500 ± 28 N for the young men and 331 ± 29 N for the old men ($P = 0.005$). The target force for the two fatiguing contractions was 125 ± 7 N for the young men and 83 ± 7 N ($P < 0.01$) for the old men. Endurance time for the force task (1105 ± 103 s) was longer than that for the position task (770 ± 94 s, $P < 0.001$) with no difference between the young and old men ($P = 0.5$). The decline in MVC force for the two groups of participants was similar (25.7 ± 13.4 % MVC, $P = 0.2$) after the two fatiguing contractions.

The muscle activation experiment compared [^{18}F]-FDG uptake and EMG activity during two types of fatiguing contractions performed with the knee extensor muscles of the left leg. The MVC force at the beginning of each session was greater for the young men (462 ± 77 N) than for the old men (354 ± 91 N, $P < 0.001$) (Table 1). The target force for the two fatiguing contractions was 115 ± 19 N for the young men and 89 ± 20 N for the old men. There was no difference in endurance time between groups for the position task (943 ± 153 s vs. 835 ± 92 s, respectively, $P = 0.166$), which was longer than that in another study when the leg was in a more elevated position (Rudroff et al., 2010a). Accordingly, the young (848 ± 137 s) and old men (751 ± 83 s) performed the fatiguing contractions prior to the PET/CT imaging for similar durations ($P = 0.166$). Moreover, the decline in MVC force immediately after the fatiguing contractions was similar for young and old men (24.5 ± 7.5 % MVC and 22.9 ± 4.3 % MVC, $P = 0.247$). However, MVC force was decreased to a greater extent after the position task had been

performed to 90% of its endurance time compared with the force task for the young men ($28.8 \pm 2.8\%$ MVC and $20.2 \pm 7.7\%$ MVC, $P = 0.02$) and for the old men ($26.1 \pm 2.6\%$ MVC and $19.6 \pm 2.4\%$ MVC, $P = 0.017$).

Table 1. MVC forces (N) for the knee extensors of young and old men before and after the force and position tasks.

Group	Position task to failure			Force (90%)			Position (90%)		
	Before	After	% Δ	Before	After	% Δ	Before	After	% Δ
Young	473 \pm 85	352 \pm 78*	-26 \pm 10	466 \pm 85	359 \pm 127*	-23 \pm 18	470 \pm 85	369 \pm 94*	-21 \pm 10
Old	346 \pm 114	243 \pm 115*	-30 \pm 11	319 \pm 83	208 \pm 72 *	-34 \pm 16	348 \pm 108	279 \pm 96*	-20 \pm 5

Mean \pm SD for MVC forces (N) of young and old men performed before and after the force and position tasks during the three sessions that comprised the second experiment: the position task to failure and the force and positions for a duration equal to 90% of endurance time for the position task. * $P < 0.05$ compared with Before MVCs.

EMG amplitude

The aEMG for the agonist muscles (rectus femoris, vastus lateralis, and vastus medialis) increased similarly during the two types of fatiguing contractions for both groups of subjects (task \times muscle \times age, $P = 0.957$), but at a greater rate for the young men (time \times age, $P = 0.009$) (Fig. 3). The aEMG for the knee extensor muscles collapsed across the force and position tasks increased from 16.4 ± 3.9 to $48.4 \pm 17.8\%$ MVC for the young men and from 18.1 ± 4.7 to $31.1 \pm 10.6\%$ MVC for the old men. The aEMG for the knee extensor muscles at the beginning of the contraction (first 30 s) was similar for the force task ($18.1 \pm 5.7\%$ MVC) and the position task ($16.4 \pm 4.9\%$ MVC) for the young and old men. The aEMG for the knee extensor muscles was similar at the start and at 20% of endurance time for the two groups, but greater for the young men at 40, 60, 80%, and 100% of endurance time (muscles \times time \times age, $P = 0.009$).

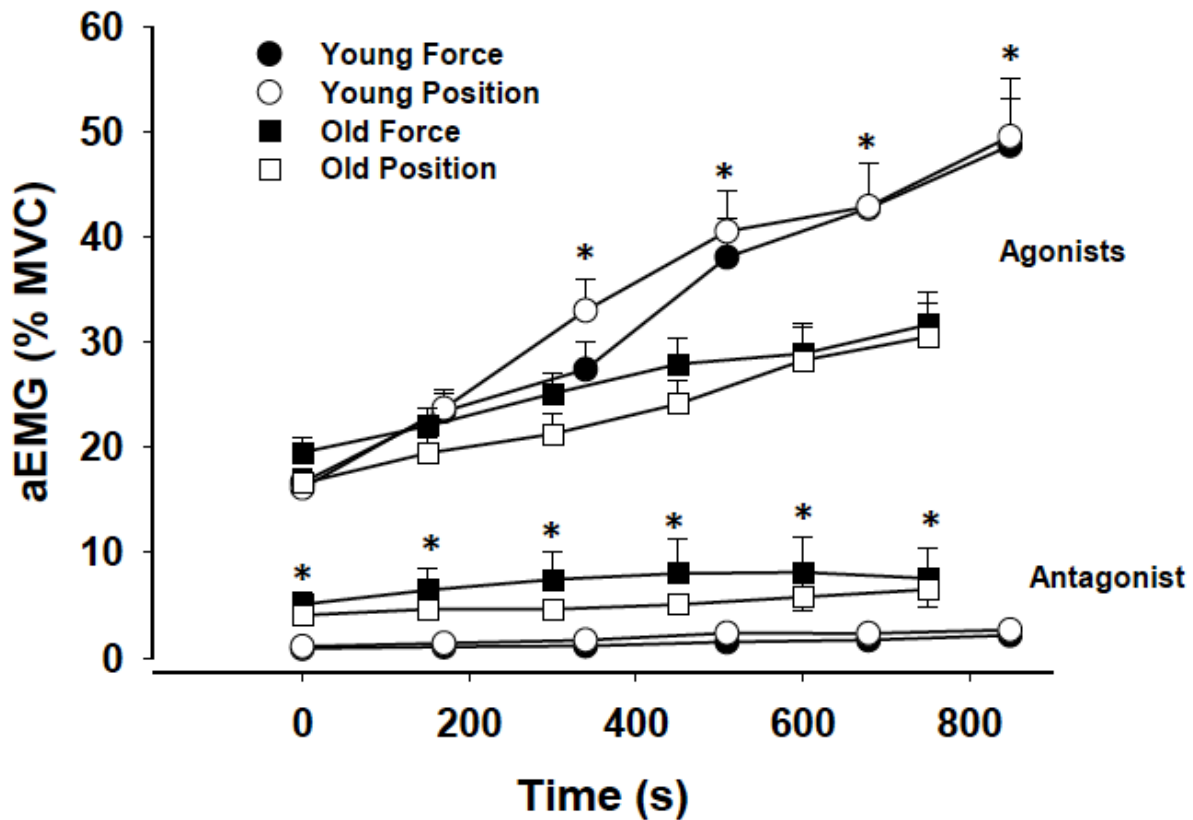


Figure 3. Change in EMG amplitude during the fatiguing contractions. Mean \pm SEM for average EMG amplitude (aEMG, normalized to the peak MVC value) for the agonist muscles (vastus medialis, vastus lateralis, rectus femoris) and the antagonist muscle (short head of biceps femoris) during the two fatiguing contractions. The aEMG was averaged over 20-s intervals for each task at 6 time points that correspond to the absolute times at start, 20, 40, 60, 80, and 100% of the prescribed contraction duration (90% of endurance time for the position task). * $P < 0.05$ between young and old.

The aEMG for the antagonist muscle (short head of biceps femoris) increased similarly during the two fatiguing contractions for both groups of subjects (time \times age, $P = 0.14$, Fig. 3). There were no differences in aEMG for the antagonist muscle between force and position tasks for either group of subjects (task \times muscle \times age, $P = 0.59$). An age \times time interaction ($P = 0.04$) indicated that aEMG for the antagonist muscle was greater at each time point for the old men relative to the young men.

The coactivation ratios did not change during the sustained contractions and were comparable between tasks at each time point during the two contractions for the young men (force task, start: 0.11 ± 0.05 , end: 0.09 ± 0.05 ; position task, start: 0.13 ± 0.07 , end: 0.10 ± 0.04) and the old men (force task, start: 0.40 ± 0.15 , end: 0.34 ± 0.20 ; position task, start: 0.39 ± 0.12 , end: 0.35 ± 0.20) (task \times age \times time, ($P = 0.47$). However, there was a significant main effect for age ($P < 0.001$) due to greater coactivation ratios for the old men in both the force and position tasks.

[^{18}F]-FDG uptake in skeletal muscles

Plasma glucose concentration immediately prior to the infusion of the [^{18}F]-FDG was similar for the young (88 ± 7 mg/dl) and old (92 ± 8 mg/dl) men, which ensured that the measurement of glucose uptake began from comparable baseline conditions for the two groups of participants. SUVs (g/ml) were calculated for three-dimensional volumes of 24 leg muscles that were identified in CT images referenced to a standardized atlas (Table 2). When collapsed across the two fatiguing contractions (force and position tasks), the average SUVs (mean \pm SD) for the 24 muscles were significantly greater for the old men (0.82 ± 0.26 g/ml) than for the young men (0.68 ± 0.32 g/ml) (age main effect, $P < 0.01$). Furthermore, average SUVs differed between the two fatiguing contractions for the young men (force: 0.64 ± 0.3 g/ml; position: 0.73 ± 0.3 g/ml), but not the old men (0.84 ± 0.3 g/ml and 0.79 ± 0.26 g/ml, respectively) (age \times task interaction, $P < 0.001$). Thus, the amount of [^{18}F]-FDG uptake was greater for the old men and they did not modulate the uptake across the two tasks (Fig. 4 and 5).

Table 2. Glucose uptake in lower limb muscles after fatiguing contractions that required either force or position control.

	Force		Position	
	Young	Old	Young	Old
Knee extensors	0.86 ± 0.41	1.15 ± 0.34**	1.14 ± 0.41††	1.14 ± 0.40
Vastus lateralis	0.88 ± 0.48	1.04 ± 0.41	1.18 ± 0.51†	1.02 ± 0.40
Vastus intermedius	1.00 ± 0.59	1.34 ± 0.21	1.23 ± 0.41	1.08 ± 0.47
Vastus medialis	0.84 ± 0.31	1.17 ± 0.29*	1.01 ± 0.31†	0.94 ± 0.19
Rectus femoris	0.73 ± 0.17	1.17 ± 0.35*	1.15 ± 0.46†	0.89 ± 0.34
Knee flexors	0.52 ± 0.15	0.69 ± 0.21**	0.60 ± 0.19††	0.64 ± 0.17
Biceps femoris short	0.69 ± 0.17	0.97 ± 0.22*	0.84 ± 0.18†	0.86 ± 0.17
Biceps femoris long	0.45 ± 0.08	0.67 ± 0.13**	0.51 ± 0.07	0.61 ± 0.10
Semimembranosus	0.47 ± 0.09	0.56 ± 0.05*	0.52 ± 0.09	0.56 ± 0.07
Semitendinosus	0.44 ± 0.09	0.56 ± 0.08*	0.51 ± 0.13†	0.54 ± 0.10
Hip muscles	0.62 ± 0.30	0.78 ± 0.30**	0.68 ± 0.33††	0.72 ± 0.16
Adductor magnus	0.52 ± 0.09	0.81 ± 0.19**	0.66 ± 0.20†	0.72 ± 0.12
Sartorius	0.48 ± 0.08	0.67 ± 0.12**	0.58 ± 0.13†	0.67 ± 0.12
Gracilis	0.48 ± 0.11	0.59 ± 0.08*	0.54 ± 0.11†	0.56 ± 0.09
Iliopsoas	0.67 ± 0.19	0.95 ± 0.64	0.67 ± 0.19	0.75 ± 0.13
Tensor fasciae latae	0.52 ± 0.07	0.71 ± 0.19*	0.54 ± 0.09	0.63 ± 0.09
Quadratus femoris	0.54 ± 0.08	0.70 ± 0.11*	0.59 ± 0.10	0.74 ± 0.18
Obturator internus	1.33 ± 0.54	1.05 ± 0.15	1.44 ± 0.55	0.85 ± 0.13*
Gluteus maximus	0.46 ± 0.08	0.61 ± 0.09*	0.51 ± 0.10	0.63 ± 0.16
Gluteus medius	0.58 ± 0.11	0.87 ± 0.25*	0.61 ± 0.11	0.75 ± 0.13
Gluteus minimus	0.60 ± 0.14	0.87 ± 0.15**	0.63 ± 0.13	0.93 ± 0.13*
Lower leg muscles	0.60 ± 0.17	0.91 ± 0.24**	0.60 ± 0.16	0.86 ± 0.19**
Tibialis anterior	0.37 ± 0.12	0.75 ± 0.41*	0.40 ± 0.19	0.65 ± 0.26
Tibialis posterior	0.66 ± 0.15	1.15 ± 0.25*	0.66 ± 0.09	1.01 ± 0.16*
Gastrocnemius lateralis	0.63 ± 0.13	0.86 ± 0.13**	0.65 ± 0.10	0.85 ± 0.15*
Gastrocnemius medialis	0.64 ± 0.15	0.81 ± 0.10*	0.63 ± 0.13	0.78 ± 0.12*
Soleus	0.66 ± 0.12	0.99 ± 0.14*	0.65 ± 0.11	0.98 ± 0.12**
Peroneus longus brevis	0.62 ± 0.31	0.78 ± 0.27**	0.63 ± 0.17	0.90 ± 0.13**

SUVs (mean ± SD) for left lower limb muscles of six young and six old men. * $P < 0.05$ and ** $P < 0.01$ between young and old men. † $P < 0.05$ and †† $P < 0.01$ between force and position tasks in young men.

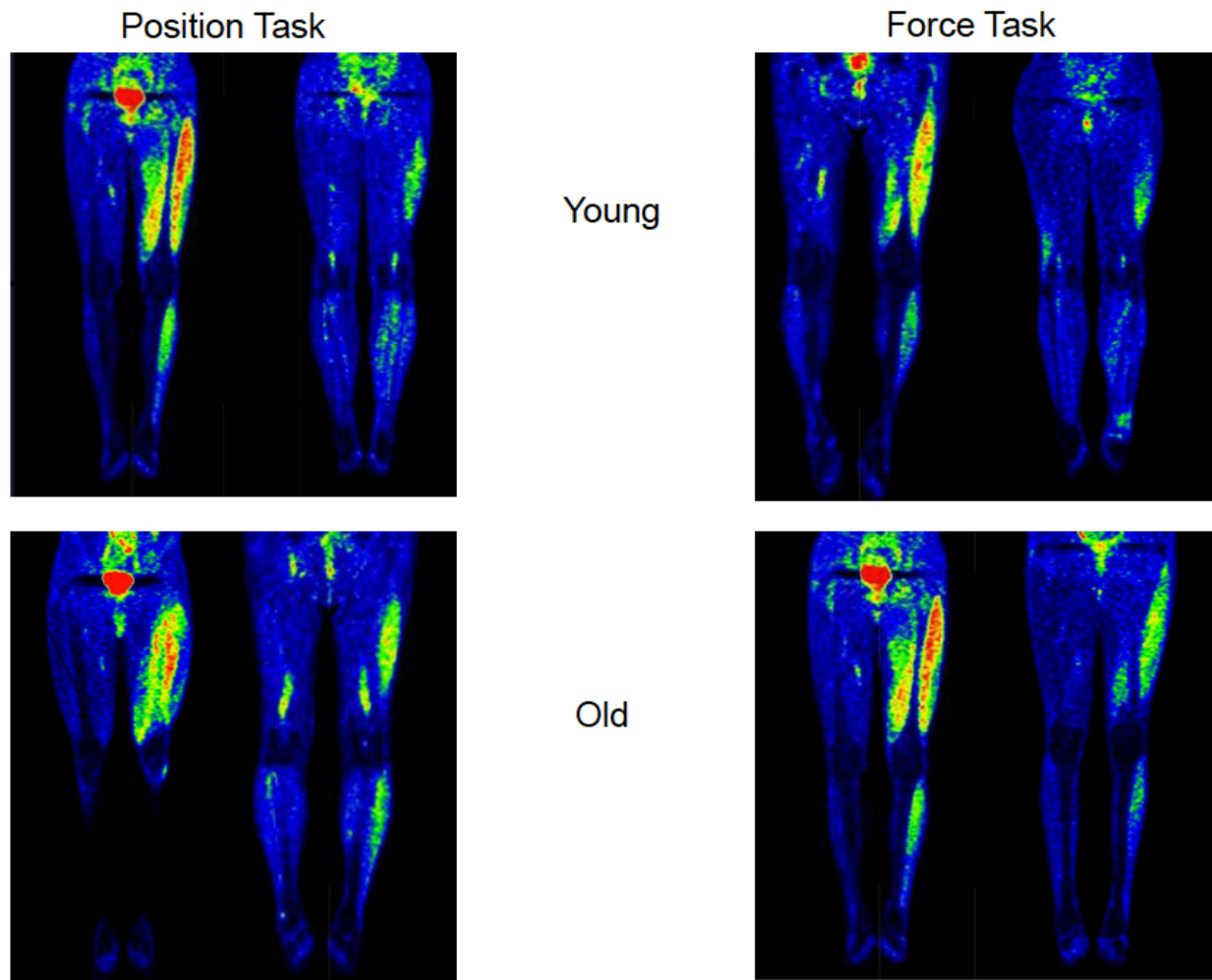


Figure 4. Lower limb PET images after position and force tasks. Longitudinal PET images (ventral [left] and dorsal [right]) taken after a young and an old man performed the force and position tasks for a similar duration. The high signal intensity (red) at the center of the pelvis resulted from accumulation of [^{18}F]-FDG in the bladder.

The group difference in [^{18}F]-FDG uptake by the 24 muscles in the leg was also apparent for the primary agonist muscles (vastus lateralis, intermedius, medius, and rectus femoris). The SUVs for the force (1.15 ± 0.34 g/ml) and position tasks (1.01 ± 0.40 g/ml) were similar for the old men and the force-task value was greater than that for the young men, but the SUVs for the young men were greater for the position task (1.14 ± 0.41 g/ml) than for the force task (0.86 ± 0.41 g/ml). Similarly, the SUV for the antagonist muscles (short and long heads of biceps femoris, semimembranosus, and semitendinosus) after the force task was greater for the old men

(0.69 ± 0.21 g/ml) than for the young men (0.52 ± 0.15 g/ml), and was greater for the position task (0.60 ± 0.19 g/ml) than the force task for the young men.

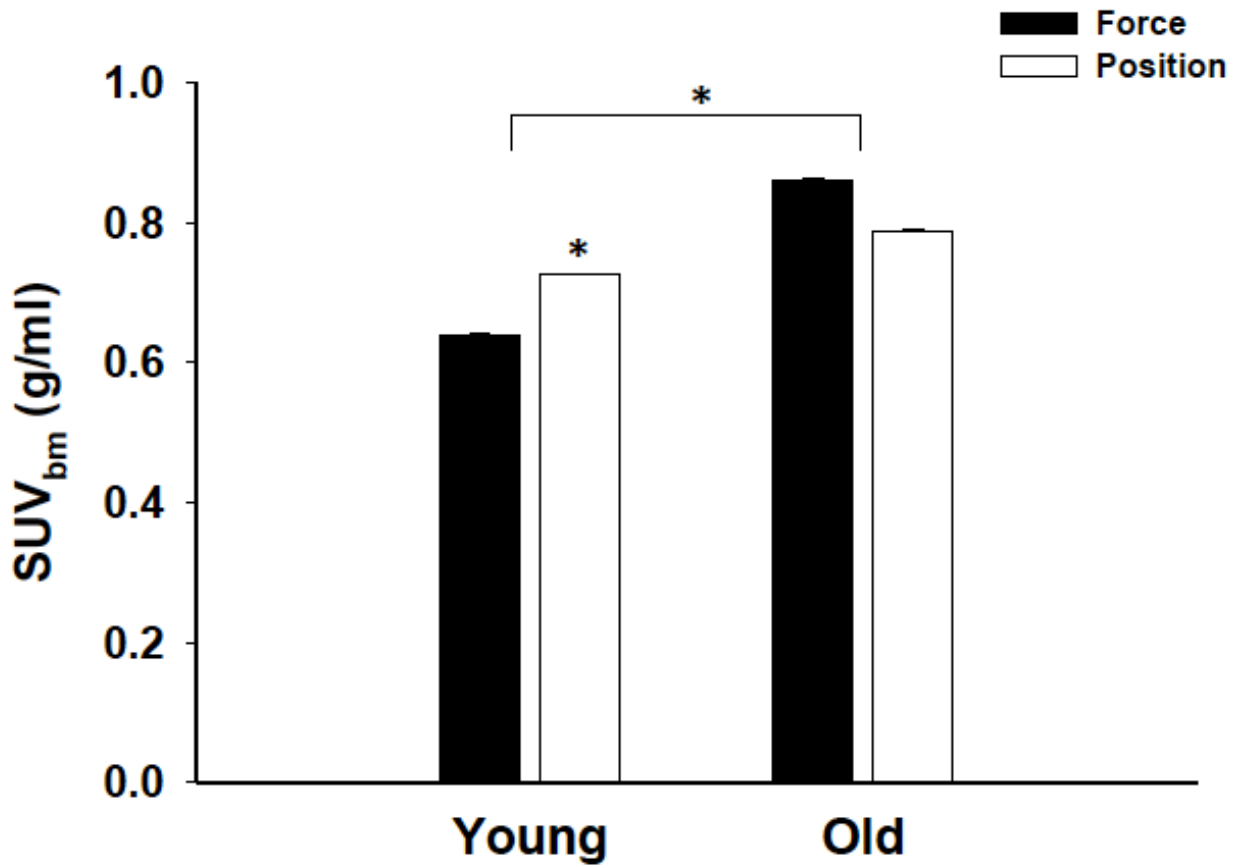


Figure 5. Glucose uptake in lower limb muscles after the two types of fatiguing contractions. Glucose uptake [SUV_{bm} (g/ml)] collapsed across 24 leg muscles was greater during the position task than the force task for young men, similar for both tasks in old men, and greater for old men than young men when collapsed across tasks. * $P < 0.01$ between force and position task for the young men and collapsed across tasks between the young and old men.

Furthermore, the SUVs for the hip muscles (adductor magnus, sartorius, gracilis, iliopsoas, tensor fasciae latae, quadratus femoris, obturator internus, and gluteus maximus, medius, minimus) was greater after the force task for the old men (0.78 ± 0.30 g/ml) than for the young men (0.62 ± 0.3 g/ml), and was greater for the position task (0.68 ± 0.33 g/ml) than the force task for the young men. In contrast, the SUVs for the lower leg muscles (tibialis anterior, posterior, gastrocnemius lateralis, medialis, soleus, and peroneus longus and brevis) were greater for the

old men after the force (0.91 ± 0.24 g/ml) and position (0.86 ± 0.19 g/ml) tasks than for the young men (force: 0.60 ± 0.17 g/ml; position: 0.60 ± 0.16 g/ml) (Fig.6). Thus, the more difficult fatiguing contraction, which required position control, was associated with greater overall levels of [^{18}F]-FDG uptake (muscle activity) for the young men but not the old men, and the old men used greater amounts of agonist, antagonist, and accessory muscle activity than young men during the force task but not the position task, except for the lower leg muscles.

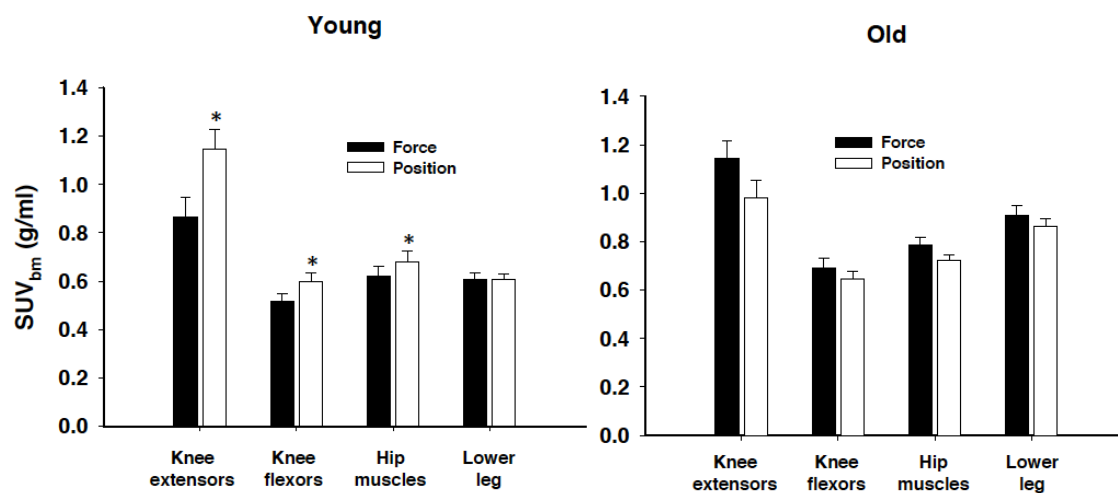


Figure 6. Glucose uptake in four lower limb muscle groups after the two types of fatiguing contractions. Glucose uptake [SUV_{bm} (g/ml)] across four muscle groups for the young and old men after completing the force and position tasks. Knee extensors: vastus lateralis, vastus intermedius, vastus medialis, and rectus femoris. Knee flexors: biceps femoris short and long heads, semimembranosus, and semitendinosus. Hip muscles: adductor magnus, sartorius, gracilis, iliopsoas, tensor fasciae latae, quadratus femoris, obturator internus, gluteus maximus, medius, and minimus. Lower leg muscles: tibialis anterior and posterior, gastrocnemius lateralis and medialis, soleus, and peroneus longus and brevis. * $P < 0.01$ between force and position tasks.

Physical activity

Young men were more physically active than the old men (9193 ± 1829 and 4393 ± 2518 avg. steps/day, $P = 0.004$). Stepwise linear regression analysis using forward selection was adopted to develop a parsimonious model using the SUV values of the leg extensors and flexors, lower leg, and hip muscles and knee extensor MVC force to predict physical activity levels (steps/day) of young and old men. The stepwise procedure converged on a model ($R^2 = 0.73$, $P < 0.001$) that

included the SUV values of the lower leg muscles (partial $r = -0.72$, $P < 0.001$) and knee extensor MVC force (partial $r = 0.55$, $P = 0.006$) (Fig. 7).

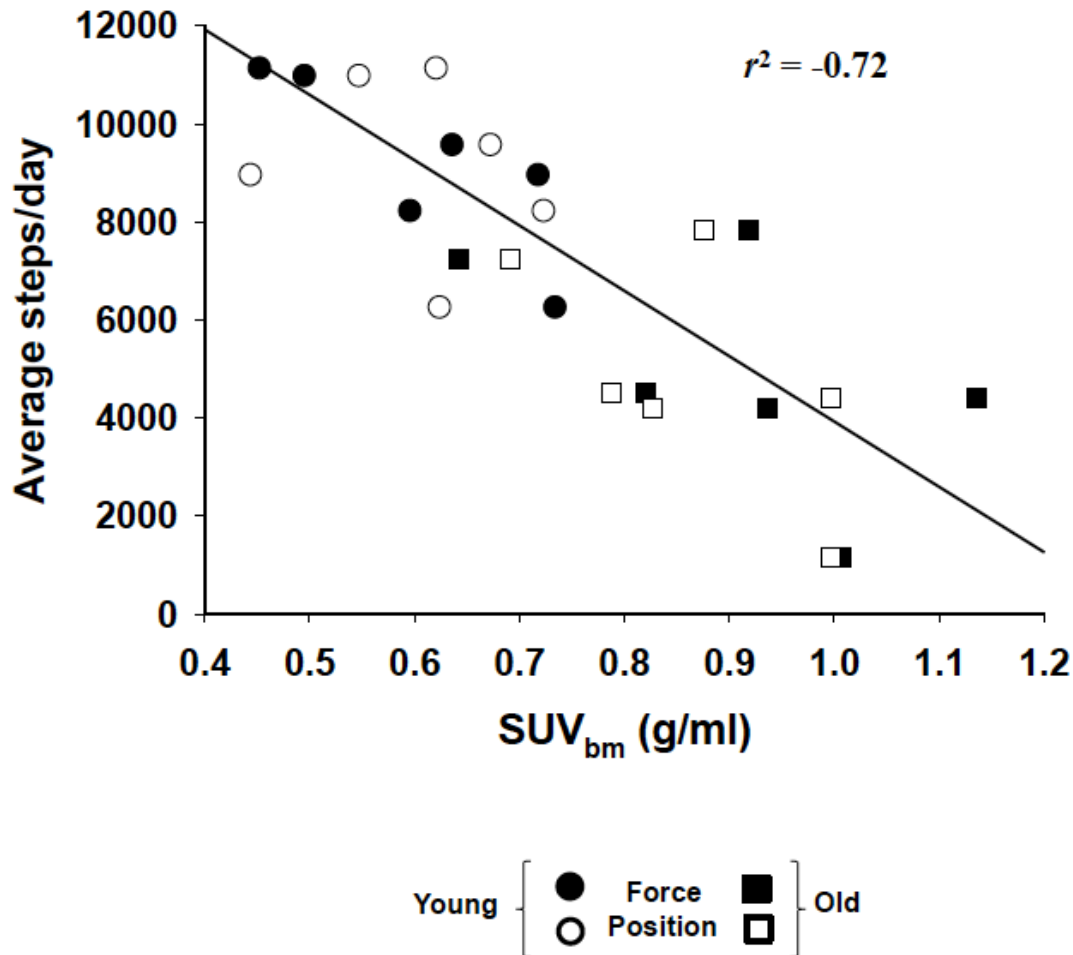


Figure 7. Prediction of daily levels of physical activity (steps/day) of young and old men. Daily levels of physical activity (steps/day) were strongly predicted by the linear combination of glucose uptake [SUV_{bm} (g/ml)] in the lower leg muscles of young and old men during the two types of fatiguing contractions and the MVC force. The lower leg muscles included tibialis anterior and posterior, lateral and medial gastrocnemius, soleus, and peroneus longus and brevis. The equation for the predicted physical activity (steps/day) was: $9614 + (-9525 \times (\text{SUV}_{\text{bm}} (\text{g/ml}) \text{ in the lower leg muscles}) + (11.225 \times \text{MVC force})$.

Discussion

The main findings of the study were that the amount of [^{18}F]-FDG uptake was greater in the leg muscles of old men than young men after performing the force task and that the young men, but not the old men, modulated [^{18}F]-FDG uptake across the two fatiguing contractions. In contrast, the rate of increase in EMG amplitude for the agonist muscles was greater for the young men during the two fatiguing contractions and there was no difference for either group of subjects in the rate of increase in EMG amplitude during the two tasks.

The surface EMG signal comprises the algebraic sum of field potentials associated with the currents that underlie muscle fiber action potentials and provides a temporal measure of muscle activity. Because muscle fiber action potentials correspond to polyphasic waveforms, the summation of the overlapping positive and negative phases of concurrent field potentials reduces the absolute amplitude of the signal (Adrian 1925). This effect is known as amplitude cancellation (Farina et al., 2004). The reduction in signal amplitude due to cancellation from the summation process increases progressively with contraction force, reaching a value of about 70% during a maximal contraction (Keenan et al., 2005; Chu et al., 2009) and increasing progressively during sustained low-force contractions (Farina et al., 2004). Amplitude cancellation reduces the sensitivity of a surface EMG signal to detect modest changes in the associated motor unit activity. For example, Mottram et al. (2005) found that sustaining a submaximal isometric contraction with position control involved greater changes in motor unit recruitment and rate coding than when the task required force control, yet there was no difference in the rate of change in the amplitude of the surface EMG signal between the two conditions.

Because an EMG signal is also influenced by the properties of the muscle fibers and the characteristics of the recording configuration in addition to the activation of the motor units by the nervous system (Farina et al., 2004; Fujimoto et al., 2003), the relative rate of change in EMG amplitude during the two types of fatiguing contractions varies across muscles and subjects (Hunter et al., 2002; Maluf et al., 2005; Rudroff et al., 2005; Klass et al., 2008). In contrast to the results in the current study, for example, we have previously found that EMG amplitude for the knee extensors increased more rapidly during the position task when the target force was 20% MVC and knee angle was 1.57 rad (Rudroff et al., 2010a). Changes in EMG amplitude when measured with bipolar surface electrodes, therefore, provide an unreliable measure of the difference in the control strategy used during these two types of fatiguing contractions.

As an alternative approach to quantifying muscle activity, the current study used PET/CT measurement of [^{18}F]-FDG uptake by skeletal muscles during the two fatiguing contractions. Pappas et al. (2001) have shown a close association between [^{18}F]-FDG uptake and muscle contraction intensity. A five-fold increase in resistance during elbow flexion, for example, increased [^{18}F]-FDG uptake in the biceps brachii by a factor of 4.9. Because [^{18}F]-FDG PET imaging relies on active muscle cells increasing glucose uptake (Holloszy et al., 1998; Ploug et al., 1984) and [^{18}F]-FDG uptake is closely correlated with exercise intensity (Fujimoto et al., 2003, Kemppainen et al., 2002; Pappas et al., 2001), it was possible to assess spatial differences in the magnitude of muscle activation across the two tasks and age groups as measured by [^{18}F]-FDG that had accumulated in the muscles during the fatiguing contractions.

The results of the current study indicating a difference in [^{18}F]-FDG uptake by the young men across the two types of fatiguing contractions are consistent with previous work showing greater

reflex responses elicited by activating group I afferents during position control relative to force control (Akazawa et al., 1983; Doemges and Rack, 1992; Maluf et al., 2007; Baudry et al., 2009b). When submaximal (20% MVC force) isometric contractions were sustained with the wrist extensors for as long as possible by requiring either force or position control, Baudry et al. (2011) observed a difference in the time course of the change in presynaptic inhibition of homonymous Ia afferent input for extensor carpi radialis during the two types of fatiguing contractions. The task-dependent modulation of synaptic input received by the involved motor neurons underlies the different adjustments in motor unit recruitment and rate coding during the two tasks (Baudry et al., 2009; Mottram et al., 2005; Rudroff et al., 2010b). The current findings indicate that the differences in motor unit activity during force and position control was manifested as greater [^{18}F]-FDG uptake by skeletal muscles during position control, at least for the young men.

The old men, however, did not exhibit a difference in [^{18}F]-FDG uptake across the two fatiguing contractions, which is consistent with a lack of modulation in the amount of Ia presynaptic inhibition when asked to perform brief isometric contractions requiring either force or position control (Baudry et al., 2010). Whereas young adults did modulate the strength of Ia presynaptic inhibition across the two load types when performing steady contractions, the old adults preferred to vary the amount of antagonist coactivation. Old adults often use greater amounts of antagonist coactivation when performing actions in which postural stability is critical (Hortobágyi and DeVita, 2006; Chu et al., 2009; Kang and Dingwell, 2009; Schmitz et al., 2009; Hortobágyi et al., 2011; Shimada, 2012), and the preference for this strategy was observed in the current study by greater [^{18}F]-FDG uptake in the knee extensors, knee flexors, and hip muscles after the force task relative to the young men and the absence of a difference across the two

tasks. Thus, the old men had greater amounts of [^{18}F]-FDG uptake after the force task than the young men, but similar uptake values after the two types of fatiguing contractions.

In contrast to the [^{18}F]-FDG uptake values for the knee extensors, knee flexors, and hip muscles, those for the lower leg muscles were greater for the old men relative to the young men after both types of fatiguing contractions. The combination of the age \times task interaction for the uptake values for the lower leg muscles after the fatiguing contractions and knee extensor MVC forces were able to predict physical activity levels of young and old men. The analysis indicated that the least physically active participants had lower knee extensor MVC forces and had greater [^{18}F]-FDG uptake in the lower leg muscles during the two fatiguing contractions. Therefore, the physical activity levels observed in the participants, which was strongly influenced by the amount of ambulation performed by the participants (McClain et al., 2007), was not related to any of the age- or task-related differences in [^{18}F]-FDG uptake during the fatiguing contractions with the knee extensor muscles. Instead, the findings indicate that [^{18}F]-FDG uptake in the lower leg muscles, which produce most of the joint power for ambulation (Hortobágyi et al., 2011), were greater in the participants with lower physical activity levels.

In conclusion, the findings of the current study demonstrate that PET/CT imaging of [^{18}F]-FDG uptake, but not surface EMG recordings, detected the modulation of muscle activity across the two types of fatiguing contractions by the young men and the greater levels of muscle activity among agonist, antagonist, and accessory muscles after the force task for old men relative to young men. The [^{18}F]-FDG measurements of muscle activity obtained with PET/CT imaging are consistent with age-associated differences in the modulation of motor unit activity and spinal reflexes during tasks that require either force or position control, but provide greater spatial

information about the magnitude of the difference in muscle activity between young and old men when performing fatiguing contractions.

Chapter III

Force steadiness during a co-contraction task can be improved with practice, but only in young adults and not middle-aged or old adults

Abstract

After practicing a co-contraction task involving lower leg muscles, young subjects improved force steadiness by reducing the amount of Ia presynaptic inhibition as indexed by D1 inhibition. Middle-aged and old adults both found the task challenging, and force steadiness even worsened for old adults after practicing the co-contraction task. Despite similar muscle strength for young and middle-aged adults, the capacity to modulate a spinal reflex pathway was reduced in middle-aged adults.

This study compared the changes in steadiness and the modulation of presynaptic inhibition of soleus Ia afferents in young, middle-aged and old adults before and after a single session of practicing a task that involved concurrent contraction of dorsiflexor and plantarflexor muscles. The hypothesis was that young subjects would be able to improve steadiness with practice by modulating Ia afferent feedback as indicated by changes in a measure of presynaptic inhibition (D1 inhibition), but that middle-aged and older subjects would exhibit a lesser ability to augment steadiness. There were no differences in steadiness between groups during an initial co-contraction trial ($P = 0.713$). Maximal voluntary contraction force for the plantarflexors was not significantly different between young and middle-aged subjects ($P > 0.05$), but it was significantly less in old subjects ($P < 0.05$). The main finding of the study was that young adults were able to improve steadiness by $\sim 19\%$ ($P < 0.001$) during a co-contraction task after 50 min of practice, whereas there was no change for the middle-aged adults, and old adults became less steady by $\sim 15\%$ ($P < 0.05$). The improvement in steadiness by young adults was accompanied by a significant reduction in the amount of Ia presynaptic inhibition as indexed by D1 inhibition ($P < 0.01$). Conversely, neither of the other two groups exhibited any change in D1 inhibition after practicing the co-contraction task. In contrast to young subjects, middle-aged and old

adults found the co-contraction task challenging and were not able to improve steadiness after practicing the low-force isometric contraction.

Introduction

Old adults tend to perform motor tasks with greater amounts of concurrent agonist and antagonist activity compared with young individuals, yet the underlying reasons for the greater coactivation remain to be identified (Hortobágyi and DeVita, 2006). Agonist–antagonist coactivation is typically considered undesirable because it increases the energetic cost of performing muscular work (Granata et al., 2004), requires activation of larger motor units to complete a task (Brown and McGill, 2008), and impairs neuromuscular performance, especially in an elderly population (Reeves et al., 2008, 2009). When performing submaximal isometric contractions, for example, old adults accommodate changes in task demands by coactivating agonist and antagonist muscles, whereas young adults tend to rely on a strategy of modulating afferent feedback (Baudry et al., 2010). It is unclear whether or not the greater coactivation exhibited by elderly adults during such conditions is due to a decline in the function of the pathways that adjust afferent input across tasks.

In a novel approach to distinguish between coactivation and the modulation of afferent input, Perez and colleagues (2007) examined the strategies used by young adults to improve steadiness after 50 min of practicing a task that involved concurrently contracting the plantarflexor and dorsiflexor muscles (co-contraction task). Fluctuations in the torque exerted about the ankle decreased across the practice session, indicating an improvement in steadiness with practice of the co-contraction task. Improved steadiness was accompanied by a decrease in the amplitude of the soleus H reflex, which was attributed to an increase in the amount of Ia presynaptic inhibition

during the co-contraction task. Improvements in steadiness, therefore, were at least partly due to modulation of afferent input that occurred during the practice contractions.

The aim of the present study was to compare the changes in steadiness and accompanying adjustments in an index of presynaptic inhibition (D1 inhibition; Mizuno et al., 1971) exhibited by young, middle-aged, and old adults after 50 min of practicing a task that required concurrent contraction of dorsiflexor and plantarflexor muscles to match a submaximal target force. The approach was based on that of Perez et al. (2007) and compared the changes in steadiness and the modulation of D1 inhibition of soleus Ia afferents in young, middle-aged, and old adults before and after a single session of practicing the co-contraction task.

Methods

Eleven young (24.5 ± 3 years old; six women), nine middle-aged (52.3 ± 8 years old; three women), and nine old adults (78.2 ± 4 years old; five women) volunteered to participate in the study after informed consent was obtained. The study conformed to the *Declaration of Helsinki* and was approved by the institutional review board at the University of Colorado Boulder.

Experimental set-up

Subjects were comfortably seated and reclined in a custom-built plantarflexion ergometer with hip, knee and ankle joints set to 120, 160 and 110 deg, respectively. The foot was securely attached to a plate that was connected to a strain-gauge force transducer (JR3 Inc., Woodland, CA, USA).

Electromyographic recordings

Electromyographic signals were recorded from soleus, medial gastrocnemius, and tibialis anterior with surface electrodes (silver–silver chloride electrodes, 8 mm in diameter; Coulbourn Instruments, Allentown, PA, USA) attached to the right leg and placed according to SENIAM recommendations (Hermens et al., 1999). The EMG signals were amplified ($\times 500$ – 5000) and bandpass filtered (13–1000 Hz) prior to sampling at 2 kHz (Coulbourn Instruments) and storage on a computer. The EMG data were normalized to values obtained during maximal voluntary contractions (MVCs).

Maximal voluntary contractions

Subjects performed MVCs with the plantarflexor muscles by gradually increasing force from zero to maximum over 3 s and maintaining this force for ~ 3 s. At least two trials were performed, with ~ 90 s of rest between successive trials. Once two peak forces within 5% of each other were obtained, the greater value was taken as the maximum and used as a reference for submaximal contractions. A single dorsiflexion MVC was performed to determine maximal EMG amplitude for tibialis anterior.

Test H reflexes and conditioned H reflexes

Responses were elicited by electrical stimulation (Grass S88K; Astra-Med, Hempstead, NY, USA; 1 ms rectangular pulse) of the target nerve in the right leg via a stimulus isolation unit (Model SIU8T; Astra-Med) that was connected to adhesive surface electrodes (Conmed, Utica, NY, USA). The intensity for the conditioning stimuli was expressed relative to motor threshold (MT), which was defined as the lowest stimulus to evoke a discernable twitch in the muscle as indicated by palpation of the tibialis anterior tendon and presence of an M-wave in the tibialis anterior EMG trace.

The test H reflex was elicited in soleus by stimulating the tibial nerve via a cathode located in the popliteal fossa and an anode slightly above the patella. The recruitment curves for the Hoffmann (H) reflex and M-wave in the soleus muscle were recorded, and the intensity of the stimulus was adjusted throughout the experiment to keep the amplitude of the test H reflex constant. Care was taken to ensure that the test H reflex was on the ascending limb of the H-reflex recruitment curve, and the initial value was kept between 10–20% of M_{\max} (Baudry et al., 2010); whenever possible, the test H reflex was accompanied by a preceding M-wave.

The level of Ia presynaptic inhibition was estimated with a conditioned H reflex that was obtained by applying an electrical stimulus to the common peroneal nerve (D1 inhibition) to activate the primary afferent depolarization interneurons converging onto the Ia afferents from the soleus (Mizuno et al., 1971). The delay between the stimuli delivered to the common peroneal nerve (conditioning stimulus) and the tibial nerve (test stimulus) was optimized (20.3 ± 1.3 ms) for each subject (Mizuno et al., 1971; Hultborn et al., 1987a,b). The intensity of the conditioning stimulus was set at $1.3 \times MT$ to standardize the relative intensity for all three groups of participants.

Test and conditioned reflexes were each obtained in two blocks of 10 reflex measurements. As the stimulus intensity had to be adjusted across conditions to keep the amplitude of the test reflex constant, the first block of 10 reflexes always included test H reflexes. The second block of test H reflexes and two blocks of conditioned H reflexes were delivered in a counterbalanced order. Twenty test and 20 conditioned reflexes were obtained across the following four conditions: rest, plantarflexion at 10% MVC force, and before and after practicing the co-contraction task.

Coactivation and co-contraction

Although the terms ‘coactivation’ and ‘co-contraction’ are often used as synonyms, a subtle but important distinction was used in the present study. ‘Coactivation’ is defined as the concurrent activation of agonist, antagonist and accessory muscles that occurs as a consequence of performing a voluntary motor task (Hortobágyi and DeVita, 2006), whereas ‘co-contraction’ is defined as the deliberate voluntary activation of both agonist and antagonist muscles as prescribed for a task (Perez et al., 2007).

Co-contraction trials required subjects to maintain a plantarflexion force of 10% MVC for several seconds and then to decrease the force to zero without reducing soleus EMG amplitude (Figure 1). This was accomplished by co-contracting tibialis anterior so that plantarflexor and dorsiflexor muscles produced equal and opposite torques. Subjects received visual feedback of the force exerted by the foot and the rectified and integrated (time constant 120 ms) soleus EMG activity during the co-contraction trials, as well as verbal feedback of soleus EMG activity. Subjects were instructed to match the force line without a reduction in soleus EMG amplitude.

Once baseline measures of the co-contraction task had been made, subjects performed a 50 min practice session that comprised 10 co-contractions of 3 min duration, each separated by 2 min of rest. Steadiness was quantified as the coefficient of variation for force; this was calculated as 10% MVC force divided by the SD for force during a 15 s window in the first 30 s of each co-contraction trial. The EMG activity for soleus, medial gastrocnemius, and tibialis anterior were also measured during this window. The 15 s window selected for analysis included no major deviations in the force exerted by the foot away from the target force.

Experimental procedure

Recording electrodes were attached over soleus, medial gastrocnemius and tibialis anterior, and

the locations of the stimulating electrodes over the tibial and common peroneal nerves were carefully determined. After subjects had performed plantarflexion and dorsiflexion MVCs, H and M recruitment curves were determined for each subject, and the conditioning stimulus was set once a suitable test H reflex had been identified. Twenty test and 20 conditioned H reflexes were then evoked both at rest and as subjects held a plantarflexion contraction at 10% MVC force. Subsequently, subjects performed two 3 min co-contraction trials before and after practicing the task. In the second co-contraction trial of each set, 20 test and 20 conditioned H reflexes were recorded.

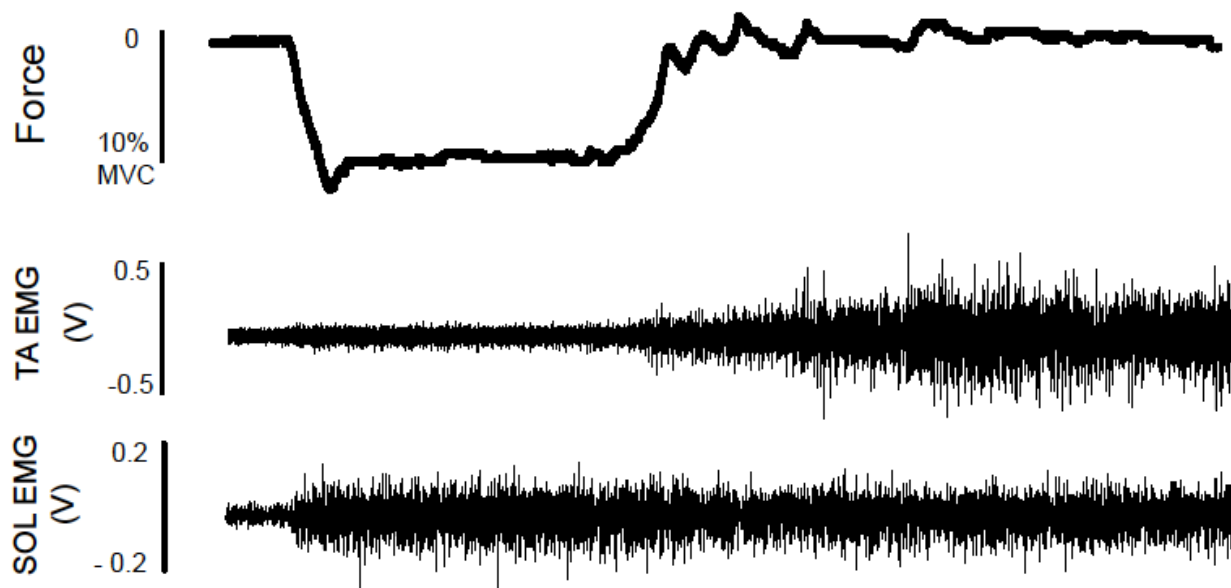


Figure 1. Representative traces showing co-contraction of dorsiflexor and plantarflexor muscles. The subjects produced a plantarflexion force equivalent to 10% MVC; the force exerted by the foot is indicated in the top trace. The subject was instructed to decrease the force to the zero line without reducing the amplitude of the soleus (SOL) EMG, indicated in the lower trace. The required decrease in force was accomplished by co-contracting the antagonist muscle (tibialis anterior; TA), which is indicated in the middle trace.

After this initial set of measurements, subjects practiced the co-contraction task for 50 min by performing 10 co-contractions of 3 min duration with 2 min of rest in between co-contractions.

In the 3-min co-contraction trial during which the reflexes were elicited after the 50 min of practice, the order of test and conditioned reflexes was counterbalanced across subjects. Subjects reported ratings of perceived exertion (RPE) on the modified Borg 10-point scale midway through the before and after co-contraction trials, with 0 corresponding to no exertion and 10 being maximal effort. The MVC force was measured again after the second set of co-contraction trials.

Data analysis

Surface EMG recordings were normalized to values obtained during the initial MVCs. Steadiness during co-contraction trials was calculated as the coefficient of variation for force measured over a 15 s window during the first 30 s of each co-contraction trial.

Statistical approach

After confirming the normality of the data with the Kolmogorov–Smirnov test, one-way ANOVAs were performed to compare the means between the three age groups. When significant differences were found, Tukey–Kramer *post hoc* tests were performed to identify the differences between groups. Variation in the means within groups was investigated using repeated-measures ANOVAs. Student’s two-tailed paired-sample *t* tests were used to examine differences within groups before and after practice. The level of statistical significance was set at $P = 0.05$ for all comparisons. Values are expressed as means \pm SD in the text and means \pm SEM in the figures. Some of these data have been presented in abstract form (Gould et al., 2012).

Results

A one-way ANOVA indicated that mean plantarflexion MVC force differed among groups ($P = 0.015$; Fig. 2). Tukey–Kramer *post hoc* tests found no significant differences in initial MVC force between young (123 ± 30 N) and middle-aged subjects (127 ± 31 N), but it was significantly less for the old group (81 ± 44 N) than for both the young ($P < 0.05$) and middle-aged groups ($P < 0.05$).

After 50 min of co-contraction practice, plantarflexion MVC force for the young subjects declined by $\sim 6\%$ (115 ± 37 N), which Student's two-tailed paired t test indicated was not statistically significant; however, MVC force declined by $\sim 17\%$ (105 ± 30 N) for the middle-aged group ($P = 0.007$) and by $\sim 20\%$ (60 ± 26 N) for the old group ($P = 0.019$).

Recruitment curves

H and M recruitment curves were obtained for all subjects. The value of M_{\max} was 5.2 ± 1.4 V for young subjects, 5.3 ± 1.7 V for middle-age subjects, and 5.4 ± 2.4 V for old subjects. There were no significant differences in M_{\max} across age groups (one-way ANOVA, $P = 0.9542$). In contrast, there were significant differences among groups for H_{\max}/M_{\max} ratios ($P = 0.0005$). Tukey–Kramer *post hoc* tests indicated that the H_{\max}/M_{\max} ratio for young subjects (0.46 ± 0.09) was significantly greater than those for the middle-aged (0.30 ± 0.13 , $P < 0.05$) and older groups (0.22 ± 0.13 , $P < 0.001$). The H_{\max}/M_{\max} ratios for the middle-aged group were not significantly different from those for the old group ($P > 0.05$).

Steadiness during co-contraction trials

The coefficient of variation for force was used as an index of steadiness and measured during co-contraction trials before and after 50 min of co-contraction practice. There were no differences

in steadiness between young ($5.0 \pm 1.5\%$), middle-aged ($5.3 \pm 1.8\%$) and old adults ($5.5 \pm 0.9\%$) during the initial co-contraction trial (one-way ANOVA, $P = 0.714$).

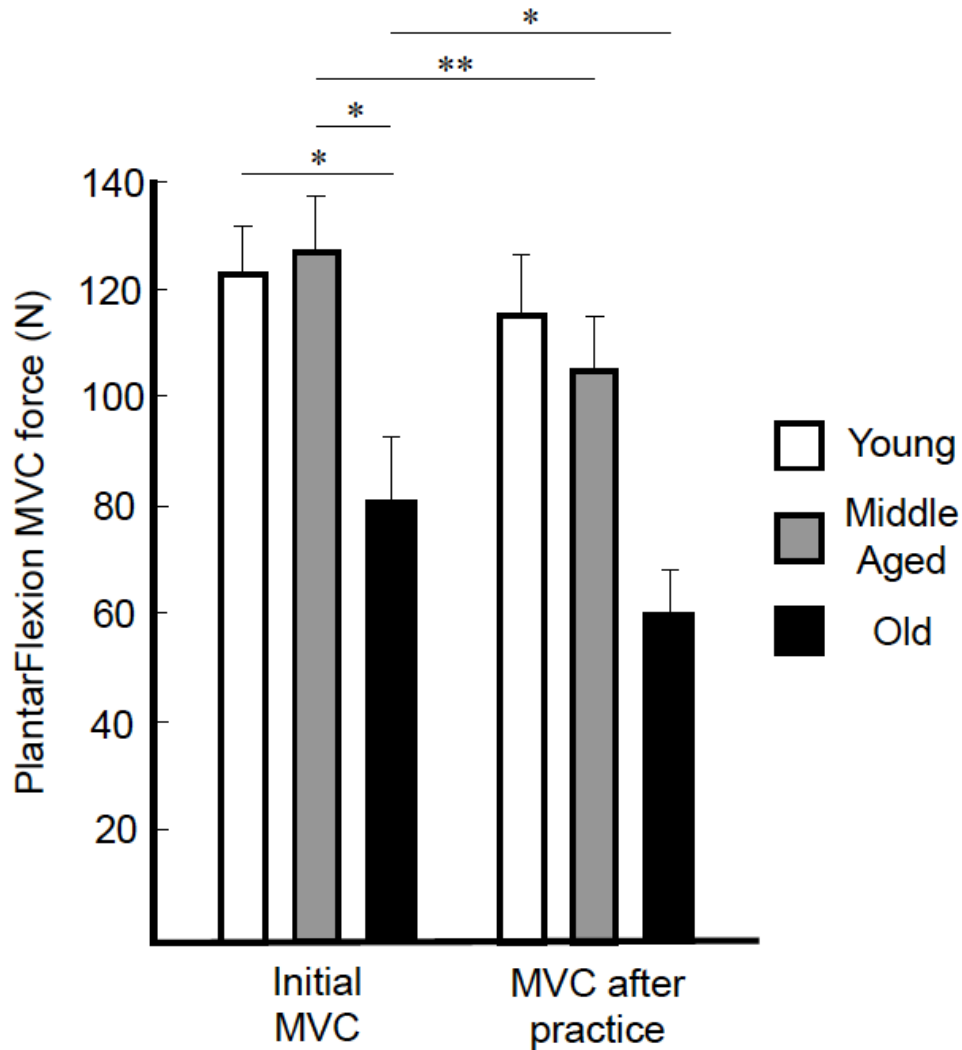


Figure 2. Maximal voluntary contraction (MVC) forces for each group, before and after practice.

Plantarflexion MVC forces before and after 50 min of co-contraction practice for young, middle-aged and old subjects. Initial MVC forces for both young and middle-aged adults were greater than those for old subjects. After practice, the MVC force declined for both the middle-aged and old subjects, but not the young subjects. * $P < 0.05$, ** $P < 0.01$.

After the practice trials, Student's two-tailed paired-sample t tests showed that young subjects improved steadiness by $\sim 19\%$ ($4.0 \pm 1.4\%$, $P = 0.005$); there was no significant change ($\sim 8\%$)

for the middle-aged group ($5.9 \pm 1.9\%$, $P = 0.279$), and steadiness was $\sim 15\%$ worse for the old group ($6.6 \pm 1.0\%$, $P = 0.020$).

Normalized EMG amplitude for soleus, tibialis anterior, and medial gastrocnemius was significantly greater for the old group than the young group during the co-contraction trials before practice ($P > 0.05$; Table 1). The EMG amplitude during the co-contraction trials before practice was greater for the middle-aged group than for the young group for two (tibialis anterior and medial gastrocnemius) of the three muscles ($P > 0.05$). Normalized EMG amplitudes (percentage MVC values before practice) were not significantly different among age groups after 50 min of practice compared with before practice, but the decline in MVC force for the middle-aged and old adults suggests a greater relative activation intensity.

Ratings of perceived exertion

A Friedman test indicated significant differences between age group means ($P < 0.05$) in RPE scores reported during the initial co-contraction trials. *Post hoc* tests showed that scores reported by the old group (2.7 ± 0.8) were significantly lower than those reported by the young group (4.3 ± 1.2 , $P < 0.05$). The scores reported by the middle-aged group (3.1 ± 1.6) were not significantly different from either the young ($P > 0.05$) or middle-aged groups ($P > 0.05$).

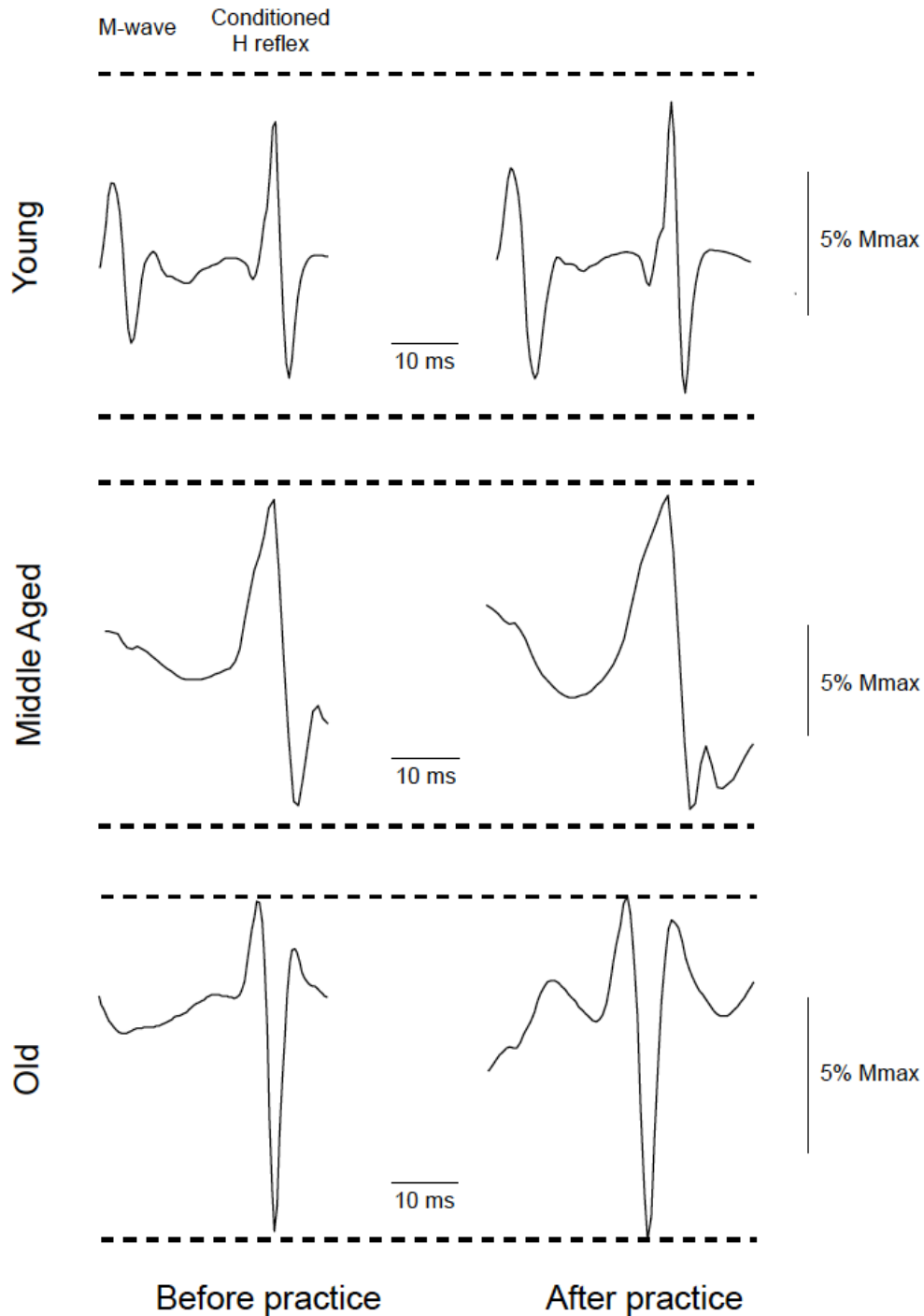


Figure 3. Representative M-waves and conditioned H reflexes. Representative traces, averaged from 20 reflex responses, for a young subject (top), a middle-aged subject (center), and an old subject (bottom) before and after practice. The dashed lines indicate the peak-to-peak amplitude of the test H reflex averaged over 20 reflex responses, whereby the greater depression in the amplitude of the conditioned H reflex, relative to the amplitude of the test reflex, indicates an increase in Ia presynaptic inhibition.

Subject group	Soleus (% MVC)		Tibialis anterior (% MVC)		Medial gastrocnemius (% MVC)	
	Before	After	Before	After	Before	After
Young	18.6 ± 11.5	17.9 ± 10.4	12.4 ± 5.7	13.7 ± 2.1	13.0 ± 4.8	13.9 ± 4.6
Middle aged	20.1 ± 10.1	18.4 ± 8.9	13.7 ± 2.1*	17.2 ± 12.7	32.7 ± 14.7*	33.1 ± 16.0
Old	34.6 ± 17.2*	43.9 ± 18.0	20.0 ± 4.0*	26.4 ± 8.7	35.0 ± 18.6*	32.0 ± 20.0

Table 1. Electromyogram amplitudes (means ± SEM) before and after practice. Normalized EMG amplitudes (expressed as a percentage of maximal voluntary contraction; % MVC) during the co-contraction trials performed before and after practice for the three age groups of participants. The EMG values were significantly greater for middle-aged and old groups compared with the young group during the initial co-contraction task (* $P < 0.05$), and there were no differences between middle-aged and old groups. There were no differences in EMG values among groups after practice.

Student's two-tailed paired samples t tests showed that the scores reported by young subjects during the co-contraction trials after practice were significantly lower (2.6 ± 1.3) than those before practice ($P = 0.0002$). Conversely, both the middle-aged (4.2 ± 1.0 , $P = 0.003$) and old groups (4.8 ± 1.2 , $P = 0.0001$) reported significantly higher scores after practice.

Test H reflexes

Repeated-measures ANOVAs indicated that M-wave amplitude differed across conditions for young subjects ($P = 0.0002$; Table 2). Tukey–Kramer *post hoc* tests showed significant differences between rest ($2.8 \pm 3.5\%$ M_{\max}) and the co-contraction trials after practice (12.7 ± 9.7 , $P < 0.001$), between 10% MVC plantar flexion ($3.2 \pm 3.2\%$ M_{\max}) and the co-contraction trials after practice ($P < 0.001$) and between the co-contraction trials before practice ($5.8 \pm 6.8\%$ M_{\max}) and those after practice ($P < 0.05$). However, there were no significant changes in M-wave amplitude across conditions for middle-aged ($P = 0.807$) or old subjects ($P = 0.241$).

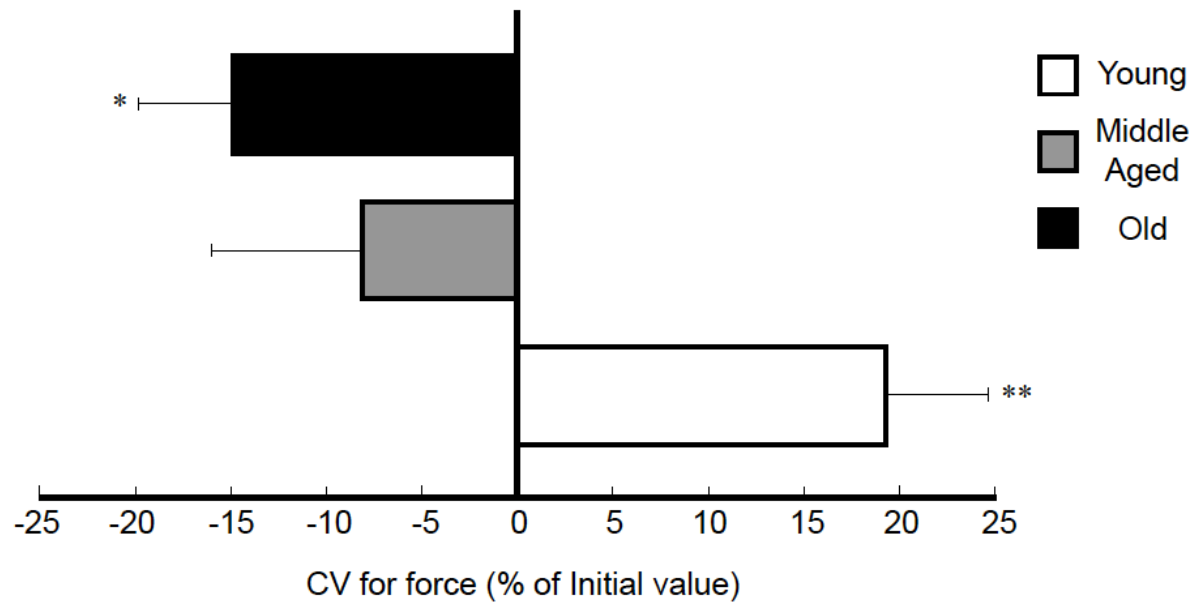


Figure 4. Changes in steadiness after practice. Change in steadiness, measured as the coefficient of variation (CV) for force during a 15 s window in the first 30 s of each co-contraction task, among groups after 50 min of practice. In young subjects, the CV for force decreased by ~19%. There was no change in middle-aged subjects. In old subjects, the CV for force increased by ~15% compared with the initial value. * $P < 0.05$, ** $P < 0.01$.

D1 inhibition

The amplitude of the D1-conditioned H reflex, when expressed as a percentage of the test H reflex, provides an index of Ia presynaptic inhibition (Fig. 3). The amount of D1 inhibition differed across conditions for young subjects (repeated-measures ANOVA, $P < 0.0001$) only (Table 3). *Post hoc* tests showed that the amount of D1 inhibition for the young group was greater (smaller-amplitude conditioned reflex) during rest ($48.1 \pm 15.7\%$) than during 10% MVC plantarflexion ($64.9 \pm 7.8\%$, $P < 0.01$) and the co-contraction trials before ($68.4 \pm 11.3\%$, $P < 0.01$) and after practice ($84.9 \pm 9.7\%$, $P < 0.001$). The amount of D1 inhibition for the young adults was significantly less during the co-contraction trials after practice than 10% MVC plantarflexion ($P < 0.01$) and the co-contraction trials before practice ($P < 0.01$). The variation in D1 inhibition across conditions was not significant for either middle-aged ($P = 0.157$) or old

subjects ($P = 0.105$), but was substantially less than the reduction in the amplitude of the conditioned H reflex exhibited by young subjects (Table 3).

One-way ANOVAs were used to compare age-group means during each task (Table 3). There were significant differences between groups in the amount of D1 inhibition between groups at rest ($P < 0.0001$). *Post hoc* tests showed that the conditioned reflex at rest was significantly more depressed in young subjects ($48.1 \pm 15.7\%$) than in middle-aged ($79.3 \pm 14.5\%$, $P < 0.001$) and old groups ($86.6 \pm 6.3\%$, $P < 0.001$), whereas there were no differences between middle-aged and old groups. Significant differences were also found between groups during plantarflexion at 10% MVC ($P < 0.0001$); conditioned reflexes were significantly more depressed in young ($64.9 \pm 7.8\%$) than in middle-aged ($85.8 \pm 8.3\%$, $P < 0.001$) and old adults ($95.0 \pm 10.2\%$, $P < 0.001$), whereas there were no differences between middle-aged and old groups ($P > 0.05$).

Subject group	Rest (% M_{\max})	10% MVC (% M_{\max})	Before (% M_{\max})	After (% M_{\max})
Young	2.8 ± 1.0	3.2 ± 0.9	5.7 ± 2.0	$12.6 \pm 2.9^{*\dagger\dagger}$
Middle aged	6.4 ± 2.4	4.9 ± 2.3	4.3 ± 1.7	5.9 ± 1.9
Old	5.3 ± 2.0	7.8 ± 2.2	11.4 ± 4.1	7.2 ± 1.8

Table 2. M-Wave amplitudes (means \pm SEM) across conditions for the three groups. There was significant variation in soleus M-wave amplitude (% M_{\max}) across the different phases of the protocol for young subjects. Significant differences existed between rest and the co-contraction trial after 50 min of practice ($*P < 0.001$), between 10% MVC force and after practice ($\dagger P < 0.001$) and between the before and after practice co-contraction trials ($\ddagger P < 0.05$). There was no significant variation in M-wave amplitude for either middle-aged or old subjects.

There were also significant differences ($P < 0.0001$) among groups before practicing the co-contraction task. *Post hoc* tests showed that conditioned reflexes for young subjects ($68.4 \pm 11.3\%$) were significantly more depressed than for middle-aged ($89.7 \pm 5.8\%$, $P < 0.001$) and old subjects ($97.2 \pm 10.8\%$, $P < 0.05$) and that there were no significant differences between middle-

aged and old groups. Significant differences were also found between groups after practicing the co-contraction task ($P = 0.033$). *Post hoc* tests showed that conditioned reflexes in young subjects ($84.9 \pm 9.7\%$) were significantly more depressed than in old adults ($97.5 \pm 14.1\%$, $P < 0.05$), and there were no significant differences between middle-aged ($85.2 \pm 9.1\%$) and either young or old subjects.

Discussion

The main finding in the present study is that young people were able to improve steadiness during a co-contraction task after 50 min of practice, whereas there was no change for the middle-aged adults, and old adults became less steady. The improvement in steadiness by young adults was accompanied by significant reductions in RPE and in the amount of Ia presynaptic inhibition as indexed by D1 inhibition. Conversely, the other two groups both exhibited a reduction in MVC force, an increase in RPE and no change in initial low levels of D1 inhibition after practicing the co-contraction task.

Changes in steadiness

There were no differences in force steadiness between groups during the co-contraction trials before the practice trials. Despite the same relative target force for all groups, the EMG amplitudes for soleus, medial gastrocnemius and tibialis anterior during the initial co-contraction trials were greater for old adults than for young adults. Furthermore, there was no statistically significant difference in the target force during the initial co-contraction trials for young and middle-aged adults, yet the middle-aged subjects used greater EMG amplitudes for medial gastrocnemius and tibialis anterior than young adults.

The changes in steadiness after 50 min of practicing the co-contraction task differed among the three groups (Fig. 4), yet there were no changes in the normalized EMG amplitudes for soleus, medial gastrocnemius or tibialis anterior (Table 1). Nonetheless, there were significant differences among the three groups in both RPE and MVC force. Young subjects did not experience any change in MVC force after the co-contraction practice trials and reported a decrease in RPE during the co-contraction task, which suggests that the task became easier for them. Conversely, the middle-aged and old groups experienced significant reductions in MVC force (Fig. 2) and reported greater RPE scores after co-contraction practice, indicating that the task became more difficult for them after the 50 min of practice. The decrease in MVC force after the practice trials was greater for the old adults than for the middle-aged adults, but the increase in RPE was similar for the two groups. The fatigue-related adjustments (decline in MVC force, increase in RPE) exhibited by the middle-aged and old adults were associated with greater EMG amplitudes during the co-contraction task (before and after practice) and an inability to modulate this activity with 50 min of practice.

Reflex modulation

Similar to the present findings, Perez et al. (2007) reported an increase in steadiness for young subjects after a similar practice protocol. The increase in steadiness was accompanied by a decrease in the peak-to-peak amplitude of the H reflex after practice, which was attributed to an increase in Ia presynaptic inhibition. In the present study, Ia presynaptic inhibition was assessed with the D1 inhibition method, which tests the excitability of the primary afferent depolarization (PAD) network by stimulating Ia afferents arising from the antagonist muscle prior to applying the test stimulus to the nerve supplying the agonist muscle (Mizuno et al., 1971). The D1 technique is presumed to provide a more valid index of Ia presynaptic inhibition than changes in

H-reflex amplitude because it assesses the excitability of the PAD interneurons.

Subject group	Rest (%)	10% MVC (%)	Before (%)	After (%)
Young	48.1 ± 4.7	64.9 ± 2.3*	68.4 ± 3.4*	84.9 ± 2.9* [§]
Middle aged	79.3 ± 4.8*	85.8 ± 2.7 [†]	89.7 ± 1.9 [‡]	85.1 ± 3.1
Old	86.6 ± 2.1*	95.0 ± 3.4 [‡]	97.2 ± 3.6 [‡]	97.4 ± 4.7 [¶]

Table 3. Changes in D1 inhibition within groups. Amplitude of D1 conditioned reflex across conditions in the three age groups. * $P < 0.05$ compared with young adults at rest, $^{\dagger}P < 0.01$ compared with young adults at 10% MVC force, $^{\ddagger}P < 0.001$ compared with young adults during the co-contraction trials before practice, $^{\S}P < 0.01$ compared with young adults during 10% MVC force and $^{\P}P < 0.01$ compared with young adults during the co-contraction trials after practice.

In the present study, D1 inhibition during the co-contraction task decreased after the practice trials for the young subjects, which may seem contrary to the findings of Perez et al. (2007). The apparent difference between studies can be explained by the approach used to elicit the test responses. The approach in the present study was to keep the amplitude of the test H reflex constant across conditions, which meant that the stimulus intensity was not constant; changes in stimulus intensity are manifest as variation in the peak-to-peak amplitude of the M-wave that precedes the H reflex (Table 2). Given that the sensitivity of the monosynaptic reflex to excitatory and inhibitory inputs depends on the amplitude of the test H reflex (Crone et al., 1990), the approach in the present study was to keep this constant across conditions (Baudry et al., 2010). If the stimulus intensity had not been adjusted between the before and after co-contraction trials, the amplitude of the H reflex would have been reduced after co-contraction practice, as observed by Perez et al. (2007). The depression in H-reflex amplitude reported by Perez et al. (2007), therefore, was probably attributable to factors other than Ia presynaptic inhibition, as they discussed. The other factors could include a change in the recruitment gain of the motor neuron pool (Kernell and Hultborn, 1990), Ib inhibition (Marchand-Pauvert et al., 2002) or recurrent inhibition (Nielsen and Pierrot-Deseilligny, 1996).

To assess the potential contribution of other adjustments to the improvement in steadiness, Perez et al. (2007) elicited motor-evoked potentials with transcranial magnetic stimulation, measured the transcranial magnetic stimulation-elicited suppression of EMG activity and calculated the coherence between electroencephalographic and EMG activities. Together, these measures provide an index of the relative responsiveness of cortical motor neurons (Gandevia et al., 1996). They found that 30 min of co-contraction practice reduced the amplitude of motor-evoked potentials in soleus and increased both transcranial magnetic stimulation-elicited suppression and coherence of soleus EMG, which suggested that the adjustments associated with the co-contraction practice included a decrease in the responsiveness of corticospinal neurons. If the decrease in H-reflex amplitude observed by Perez et al. (2007) after the co-contraction practice represents an increase in Ia presynaptic inhibition, it is difficult to explain how the subjects maintained constant EMG amplitudes during the co-contraction trials after practice. One likely explanation, as suggested by the results of the present study, is that in fact, Ia presynaptic inhibition decreased after practice, which would have provided a source of excitatory inputs to the spinal motor neurons to achieve the requisite levels of muscle activation. Such an interpretation suggests that young subjects become more reliant on feedback control after practice.

Previous work has shown that old adults often modulate afferent feedback less than young adults (Darling et al., 1987; Earles et al., 2001). For example, Baudry et al. (2010) found that young adults modulated afferent feedback to distinguish between force and position control, whereas old adults changed the amount of agonist–antagonist coactivation to accommodate the change in load compliance. Results from the present study are consistent with these observations, because old adults used greater amounts of muscle activity in soleus, medial gastrocnemius and tibialis

anterior than young adults during the co-contraction trials before practice (Table 1). However, the opportunity to improve steadiness by decreasing D1 inhibition was probably compromised by the greater fatigability exhibited by middle-aged and older groups, because Ia presynaptic inhibition often increases during fatiguing contractions (Duchateau and Hainaut, 1993) due to activation of group III and IV afferents in response to the accumulation of metabolites (Pettorossi et al., 1999). Moreover, the amount of D1 inhibition exhibited by the middle-aged and old adults was substantially less than that of the young adults, which greatly reduced the likelihood that it could be modulated across tasks.

Fewer data are available on the modulation of afferent input across conditions for middle-aged adults. In the present study, plantarflexor MVC force did not differ between young and middle-aged subjects, whereas middle-aged subjects were significantly stronger than old subjects (Figure 2). Moreover, the middle-aged and old adults both experienced an increase in RPE and a decrease in MVC force after the co-contraction practice, but differed in the effect on steadiness; it did not change for the middle-aged subjects but worsened (increase in the coefficient of variation for force) for the old subjects. The H_{\max}/M_{\max} ratios obtained during the recruitment curve for the middle-aged subjects were significantly less than for young subjects, whereas the H_{\max}/M_{\max} ratios for old subjects were significantly less than for both middle-aged and young subjects. The decline in H_{\max}/M_{\max} ratios indicates that a smaller proportion of the motor pool was activated in response to electrical stimulation of group Ia afferents, which suggests a decrease in the capacity to make corrective actions (Scaglioni et al., 2002). The absence of changes in the size of the test H reflex and the amount of D1 inhibition between rest, plantarflexion and co-contraction tasks before or after practice suggests that the middle-aged adults were similar to the old subjects in this respect. However, the relatively low levels of D1

inhibition across tasks for both the middle-aged and old subjects temper this interpretation. Given the apparent low level of D1 inhibition observed in old subjects during the initial co-contraction, a task they found more difficult to accomplish than the other two groups, coupled with the increase in RPE and decline in MVC force exhibited by this group of participants, it is not clear whether an inability to modulate D1 inhibition contributed to the decline in steadiness. The initial level of D1 inhibition in the old group suggests that modulating D1 inhibition across tasks was unlikely, presumably due to an already reduced level of Ia presynaptic inhibition. Consistent with this interpretation, the middle-aged group did not modulate D1 inhibition, did not find the protocol fatiguing, and there was no change in steadiness before and after practice. Taken together, these findings indicate that the fatigability experienced by the old subjects may have significantly contributed to their decline in steadiness.

Limitations

There are several limitations in the present study that should be acknowledged. One limiting factor was the exclusive use of D1 inhibition to estimate the level of Ia presynaptic inhibition. When the PAD network becomes saturated with multiple inputs, for example, responsiveness to the D1 conditioning stimulus declines, which is referred to as occlusion (Pierrot-Deseilligny, 1997; Baudry et al., 2010). Furthermore, the D1-inhibition approach can be compromised by changes in the gain of the reflex (Kernell and Hultborn, 1990) and postactivation depression of the H reflex (Crone and Nielsen, 1989). By using multiple measures of Ia presynaptic inhibition, such as D1 inhibition and heteronymous Ia facilitation, it is possible to circumvent these limitations. This was not possible in the present study, however, as pilot work showed that the activation of quadriceps muscles (vastus lateralis and medialis) increased during the co-contraction practice trials and compromised the use of the approach during the co-contraction

task after the practice trials. Furthermore, it was not possible to measure the responses to both types of conditioning stimuli in a single session.

In studies that use both D1 inhibition and heteronymous facilitation as independent measures of Ia presynaptic inhibition, any divergent changes in presynaptic inhibition are often explained by occlusion of the PAD network (Hultborn et al., 1987a,b; Faist et al., 1996; Aymard et al., 2001). For example, Baudry and Duchateau (2012) attributed the divergence of the change in D1 inhibition responses and the modulation of motor unit discharge probability during postural tasks to occlusion in the PAD network. Moreover, Nielsen and Kagamihara (1993) found that peaks at monosynaptic latencies in motor unit post-stimulus time histograms and heteronymous facilitation of soleus H reflex indicated an increase in Ia presynaptic inhibition during a co-contraction task compared with an isolated plantarflexion contraction, which contrast with our findings using the D1 technique. In the present study, however, there were significant changes in the amplitude of H reflexes conditioned by D1 inhibition across conditions, which suggests that the PAD network was not occluded.

A second limitation was related to the demands of the task. Given that the relative target forces in the present study were low, it was assumed that the challenges involved in the protocol would be similar for the three groups of participants. This assumption was appropriate for the young subjects, but not for the middle-aged and old adults, who found the task more challenging than anticipated. Consequently, the demands of the task confounded the adjustments exhibited by the middle-aged and old adults.

Summary

The present study compared the changes in steadiness, MVC force, RPE and the modulation of

D1 inhibition of soleus Ia afferents in young, middle-aged and old adults before and after a single session of practicing a co-contraction task. There were no differences in steadiness between groups during an initial co-contraction trial. The main finding of the study was that young people were able to improve steadiness by ~19% during a co-contraction task after 50 min of practice, whereas there was no change for the middle-aged adults, and older adults became less steady by ~15%. The improvement in steadiness by young adults was accompanied by no change in MVC force, a decrease in RPE and a significant reduction in the amount of Ia presynaptic inhibition as indexed by D1 inhibition. Conversely, the middle-aged and old adults both experienced significant decreases in MVC force, increases in RPE and no change in D1 inhibition after practicing the co-contraction task. As there was no difference in strength between young and middle-aged subjects, the findings of the present study indicate that the decline in the control of voluntary actions precedes the age-associated loss of muscle strength.

Chapter IV

Motor unit activity in biceps brachii of left-handed humans during fatiguing contractions supporting two load types

Abstract

The purpose of the current study was to compare the discharge characteristics of single motor units during sustained isometric contractions that required either force or position control with the dominant arm of left-handed individuals. There were no differences between the dominant and nondominant limbs of the 21 participants on a test of manual dexterity (grooved pegboard test) or grip strength. However, time to complete a writing test was longer ($P < 0.05$) for the nondominant arm. The target force during the two fatiguing contractions ($24.9 \pm 10.5\%$ maximal force) was identical for each biceps brachii motor unit ($n = 32$) and set at $4.7 \pm 2.0\%$ of maximal voluntary contraction (MVC) force above its recruitment threshold (range: 0.5-41.2% MVC force). Contraction duration for both tasks was set relative to the recruitment threshold of the motor unit (152 ± 84 s). The decline in MVC force immediately after the fatiguing contraction was similar for the two tasks (force: $11.1\% \pm 13.7\%$; position: $11.6\% \pm 9.9\%$). Despite similar increases ($P < 0.001$) in the amplitude of the surface-recorded electromyogram for the involved muscles during the two tasks, mean discharge rate declined more during the position task (task x time interaction, $P < 0.01$) and the variability in discharge times (coefficient of variation for interspike interval) increased only during the position task (task x time interaction, $P < 0.008$). Handedness does not influence the adjustments in biceps brachii motor unit activity during sustained submaximal contractions requiring either force or position control.

Introduction

Adjustments in motor unit activity during fatiguing contractions depend on the details of the task being performed (Baudry et al., 2009; Carpentier et al., 1998; Enoka et al., 1989; Farina et al., 2009; Pascoe et al., 2013; Rudroff et al., 2010). One of these details is the compliance of the load being supported by the involved muscles. Several studies have found that endurance times are briefer when the task requires participants to support a high-compliance load (position control) relative to one with low compliance (force control), even when the magnitude of the two submaximal loads ($\leq 30\%$ of maximum) is matched (Hunter et al., 2002; Maluf et al., 2005; Rudroff et al., 2011). A more rapid increase in the amplitude of surface electromyographic (EMG) recordings during fatiguing contractions that require position control, even when the task involves a single agonist muscle (Maluf et al., 2005), indicates that sustained position control may involve more rapid adjustments in motor unit activity. To examine this possibility, Mottram et al. (2005) recorded the discharge of 32 motor units in biceps brachii during isometric contractions sustained for the same duration (161 ± 96 s) for the two load conditions (force and position control). Consistent with previous reports (Enoka et al., 1989; Carpentier et al., 1998), mean discharge rate declined during both fatiguing contractions, but the decline was greater during position control. To sustain the identical target force during the two sustained contractions, therefore, Mottram et al. (2005) found that the rate at which additional motor units were recruited was greater during position control. Subsequent studies have confirmed that changes in motor unit recruitment and rate coding occur more rapidly during position control despite a similar net muscle torque during both tasks (Baudry et al., 2009) and that these adjustments involve differential modulation of spinal reflex pathways (Baudry et al., 2011).

These comparisons of the adjustments during force and position control have mainly been performed on the left arm of right-handed participants. However, the control of motor unit activity has been reported to differ between dominant and nondominant limbs and between left- and right-handed individuals (De Luca et al., 1986; Semmler and Nordstrom, 1995; Adam et al., 1998; Schmied et al., 1998; Bilodeau et al., 2009; Pereira et al., 2012). To assess the potential influence of handedness and dominance on endurance time during fatiguing contractions with the two load types, Gordon et al. (2012) compared the performance of left- and right-handed individuals with the dominant and nondominant arms. The target force was set at 20% of maximal elbow flexor force for each arm. Hand dominance did not influence endurance time for either group of participants. A task x time interaction, however, indicated greater variance in the ratio of the endurance times of the two tasks for the left-handed participants. Moreover, there was a statistically significant correlation between the endurance times of the two tasks for the right-handed group ($r^2 = 0.62$), but not the left-handed group ($r^2 = 0.004$).

The purpose of the current study was to compare the discharge characteristics of single motor units during sustained isometric contractions that required either force or position control with the dominant arm of left-handed individuals. The approach replicated the protocol developed by Mottram et al. (2005). Based on the findings of Gordon et al. (2012), the hypothesis was that the adjustments in the discharge characteristics of the biceps brachii motor units would not differ during the two tasks. Some of these data have previously been presented in abstract form (Cleland et al., 2013).

Methods

Twenty-one healthy adults (21.9 ± 1.9 yrs, 8 women; height: 173.3 ± 9.5 cm; body mass: 70 ± 12

kg) participated in the study after written informed consent was obtained. All subjects were left-handed (laterality quotient = -0.58 ± 0.22) as identified by the Edinburgh Handedness inventory (Oldfield, 1971) and free of neurologic disease. The experimental procedures conformed to the *Declaration of Helsinki* and were approved by the Institutional Review Board at the University of Colorado Boulder (Protocol 11-0423).

Participants visited the laboratory on two occasions. The first visit lasted ~30 min and was used to familiarize the volunteer with the details of the study, to obtain written informed consent, and to assess hand preference and performance. The second visit lasted approximately two hours and involved recording the activity of single motor units in biceps brachii (32 motor units were recorded from 21 participants) during two fatiguing isometric contractions. Load compliance varied across the two fatiguing contractions.

Hand Preference and Performance

After handedness was assessed with the Edinburgh Handedness inventory, participants performed three tests: 1) grip strength, 2) grooved pegboard test, and 3) handwriting test (Porac and Coren, 1981; Provins and Magliaro, 1993). The Edinburgh handedness inventory is a self-report questionnaire that indicates the level of preference to use the left or right limb when performing 12 different tasks. Grip strength was measured with a handheld dynamometer (Hydraulic Hand Dynamometer; Baseline Evaluation Instruments, Irvington, TX) while the participant was seated with their elbow flexed to 1.57 rad and the wrist in a neutral position. Participants were asked to increase grip force gradually from rest to maximum in ~3 s. Verbal encouragement was provided during each effort and subjects were given at least 90 s of rest between the three trials. The grooved pegboard test (Lafayette Instruments, Lafayette, IN)

requires participants to place 25 grooved metal pegs into keyhole shaped openings of varying orientation on the board as quickly as possible. Performance was quantified as the time to place pegs into all of the openings in the pegboard. The handwriting test required subjects to write out the alphabet as quickly as possible. Time to write the alphabet three times was used as a measure of hand performance. Each of the three tests (grip strength, grooved pegboard test, writing test) were performed with each hand in a randomized order and quantified as the mean of three trials.

Experimental Setup

Subjects were seated comfortably with the upper left arm perpendicular to the ground and slightly abducted from the trunk. The elbow was flexed to ~ 1.57 rad and the wrist and forearm were oriented in a neutral position between pronation and supination and placed in a wrist-hand-thumb orthosis (Orthomerica, Newport Beach, CA, USA). The orthosis was connected to a strain gauge force transducer (JR3, Woodland, CA) to measure the force transmitted to the load. The applied force was displayed on a monitor located at eye level ~ 60 cm in front of the subject. Before the start of the position task, an electrogoniometer (Biometrics, Cwmfelinfach, UK) was attached to the lateral aspect of the arm and forearm to measure elbow angle and the signal was displayed on the monitor.

Electromyographic Recordings

Electromyographic (EMG) signals were recorded from the biceps brachii, brachioradialis, and triceps brachii of the left arm using surface electrodes (silver-silver chloride electrodes, 8-mm diameter, Coulbourn Instruments, Allentown, PA, USA) arranged in a bipolar configuration and placed ~ 15 mm apart on one side of the innervation zones for each muscle. Interference EMG

signals were also recorded from the brachialis muscle of the left arm using an intramuscular fine-wire bipolar electrode. The intramuscular electrode comprised two Formvar insulated, stainless steel wires (50- μ m diameter) that were fastened together with an all purpose adhesive gel. The bipolar recording was made between the cut end of one wire and a \sim 1-2 mm region on the other wire from which the insulation was removed. A surface electrode was used as a reference. EMG signals were amplified (1000x), band-pass filtered (13–1000 Hz) (Coulbourn Instruments, Allentown, PA, USA), sampled at 2 kHz (Cambridge Electronic Design, Cambridge, England), and stored on a computer (Dell, Plano, TX, USA). Reported EMG data were normalized to values obtained during maximal voluntary contractions performed before the fatiguing contraction.

Single motor unit action potentials were recorded in the short head of the biceps brachii muscle using subcutaneous fine-wire branched bipolar electrodes that have been described in detail previously (Gydikov et al., 1986; Enoka et al., 1988; Mottram et al., 2005; Pascoe et al., 2013). Single-motor unit recordings were amplified (5,000x) and band-pass filtered (0.3–8.5 kHz; Coulbourn Instruments, Allentown, PA). The motor unit signal was sampled at 20 kHz by a Power 1401 (Cambridge Electronic Design, Cambridge, England) and stored on a computer (Dell, Plano, TX, USA) for offline analysis.

Experimental Protocol

Participants performed two submaximal isometric contractions that were sustained for a prescribed duration. The net torque about the elbow joint was the same during both contractions for each individual, but one task required the participant to maintain a constant force against a low-compliance load (force control) and the other task required the individual to maintain a

constant elbow angle (1.57 rad) while supporting a high-compliance load (position control). The two contractions were performed in a random order, but consecutively to record the same motor unit with the subcutaneous electrode. The participant rested for ~25 min before starting the second contraction.

Maximal Voluntary Contractions

Maximal voluntary contraction (MVC) force was measured before and after each fatiguing contraction. The first MVC was used to determine maximal strength, to provide a reference for motor unit recruitment threshold force, and to record peak EMG activity. The second MVC was performed after the first fatiguing contraction and the decline in peak elbow flexor force was used as an index of performance fatigability. The third MVC was performed before the beginning of the second fatiguing contraction to provide a measure of recovery. A fourth MVC was performed after the second fatiguing contraction to provide another index of performance fatigability.

The MVCs were performed with the elbow flexor muscles by gradually increasing force from rest to maximum in ~3 s and maintaining this force for ~3 s. The investigators provided strong verbal encouragement during each MVC, and the gain of the visual feedback was adjusted between contractions. At least two trials were performed with ≥ 90 s of rest between consecutive trials. If the peak forces were not within 5% of each other for the two trials, or if a participant indicated that one of the efforts were not maximal, additional MVCs were performed until those criteria were met. The greatest peak force was taken as maximum and used as a reference for the submaximal fatiguing contractions. A single MVC was performed with the elbow extensors to record peak EMG amplitude.

Motor Unit Recruitment Threshold

Participants were asked to gradually increase the elbow flexor force up to a target force of 60% MVC over the course of 10 s. Motor unit recruitment threshold was identified as the force at which a motor unit began discharging action potentials repetitively. Recruitment threshold was determined online as the force corresponding to the third spike in a consistent train of motor unit action potentials. Participants briefly matched a target force set at 3.5% of MVC force above the identified recruitment threshold to ensure that the motor unit discharged action potential repetitively. Small adjustments in target force were made to optimize the quality of the recorded signal before beginning the fatiguing contractions.

Fatiguing Contractions

Each fatiguing contraction required either force control or position control with the target force set relative to the recruitment threshold of the isolated motor unit. The force task required participants to exert an upward force with the elbow flexor muscles against a rigid restraint to match the target force. The position task required the participant to support an equivalent load (same net muscle torque) while maintaining a constant elbow angle to match the displayed target. Participants were provided with visual feedback during both the force task (gain: 3% MVC force/cm) and position task (gain: 2° elbow flexion or extension/cm) (Hunter et al., 2002; Mottram et al., 2005). The duration of each fatiguing contraction was for each participant (152 ± 84 s; range: 60 – 330 s) and was based on recruitment threshold of the isolated motor units; duration was longer for motor units with low recruitment thresholds (Mottram et al., 2005).

Rating of perceived exertion (RPE) was measured using the modified Borg scale (0–10) during the fatiguing contractions. The scale was anchored so that a 0 represented rest or no exertion, and 10 represented the strongest possible effort.

Data Analysis

Force, elbow angle, and EMG data were sampled by an analog-to-digital converter (Power 1401; Cambridge Electronic Design, Cambridge, England) and stored on a computer for offline analysis using Spike2 data acquisition/analysis software (Cambridge Electronic Design, Cambridge, England). Force signals were sampled at 1 kHz, motor unit recordings were sampled at 20 kHz, and EMG signals were sampled at 2 kHz.

EMG signals recorded from biceps brachii using the subcutaneous fine-wire electrodes were processed using an automatic decomposition algorithm in EMGLab (McGill et al., 2005). The output of the algorithm was then visually inspected to verify individual spike times and edited to correct for missing or incorrectly identified spikes based on timing of interspike intervals and the residual of the signal.

MVC force was measured as the greatest peak force exerted during one of the MVC trials. The maximal EMG amplitude during that MVC trial, which was used to normalize subsequent EMG recordings, was the average value during a 0.5-s interval centered about the peak of the rectified EMG. EMG data collected during the fatiguing contractions were quantified as the aEMG value (normalized to the peak EMG obtained during the selected MVC trial) during three 10-s intervals at the beginning, middle, and end of the task.

Statistics

Dependent t-tests were used to compare MVC forces and motor unit recruitment thresholds before and after each task. Two-factor analysis of variance was used to examine changes in dependent variables over time (3 time points; beginning, middle, and end) and differences between tasks (2 tasks; force control and position control). Dependent variables included motor unit discharge rate and variability, EMG amplitude of agonist and antagonist muscles, and ratings of perceived exertion. Alpha levels for significance were set at 0.05 for all statistical tests unless otherwise noted. Data are presented as mean \pm standard deviation (SD) in the text and as mean \pm standard error (SE) in the figures.

Results

There was no difference in the performance of the dominant and nondominant arms of the left-handed participants (Edinburgh inventory: -0.54 ± 0.22) in either time to complete the grooved pegboard test (59.1 ± 8.1 s and 62.9 ± 7.2 s, respectively) or grip strength (40 ± 12 kg and 41 ± 12 kg). However, coordination was more developed in the dominant (left) hand as indicated by the time to complete the writing test (dominant: 42.0 ± 9.0 s and 68.2 ± 16.9 s, respectively).

The force produced during the two fatiguing contractions ($24.9\% \pm 10.5\%$ MVC force) depended on the recruitment threshold of the identified isolated motor unit ($21.6\% \pm 11.7\%$ MVC force).

In order to elicit a steady discharge of motor unit action potentials, the target force was set to $4.7\% \pm 2.0\%$ MVC force above recruitment threshold of the identified motor unit. The duration of the two tasks was the same for each participant (152 ± 84 s), which resulted in similar reductions ($P = 0.77$) in MVC force immediately after each fatiguing contraction: $11.1\% \pm 13.7\%$ decline in MVC for the force task and $11.6\% \pm 9.9\%$ decline for the position task. MVC forces performed before the first fatiguing contraction (252 ± 89 N) did not differ significantly

($P = 0.41$) from MVC forces performed after ~ 25 min of rest and before the start of the second fatiguing contraction (230 ± 88 N).

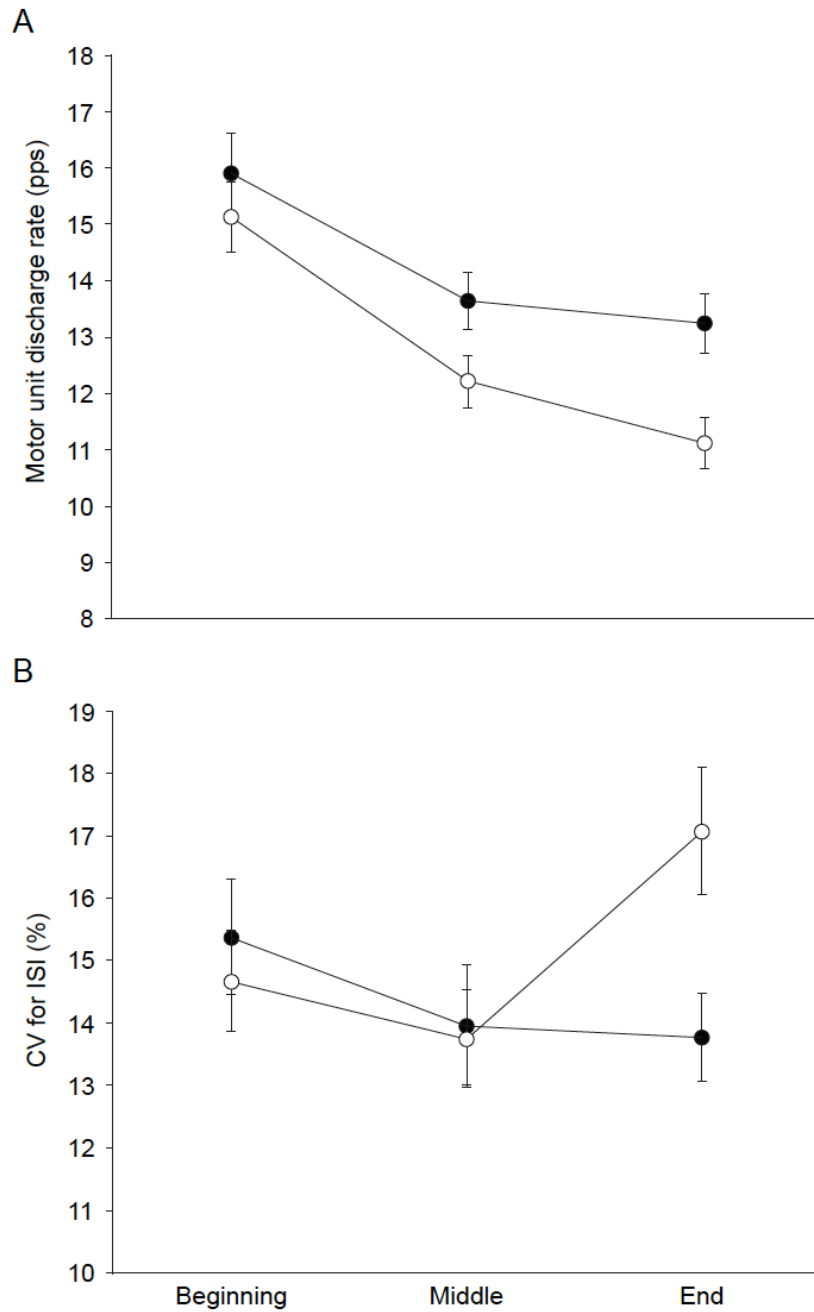


Figure 1. Motor unit discharge rate (A), discharge variability (B) (mean \pm SE, $n = 32$), were measured at the beginning, middle, and end of the contraction for the force (filled circles) and position (open circles) tasks. Discharge rate declined during both tasks ($P < 0.001$) but declined more during the position task ($P = 0.014$). Discharge rate variability increased for the position task but decreased for the force task ($P = 0.008$).

Ratings of perceived exertion (RPE) increased during both tasks (main effect for time, $P < 0.001$; Figure 2), but RPE levels were greater (main effect for time, $P = 0.021$) and there was a greater increase during the position task (time x task interaction, $P = 0.006$). The final RPE values, which were obtained at the same for two fatiguing contractions for each participant, were 7.0 ± 2.2 for the force task and 8.0 ± 1.5 for the position task, which indicates that the fatiguing contractions were not performed to task failure.

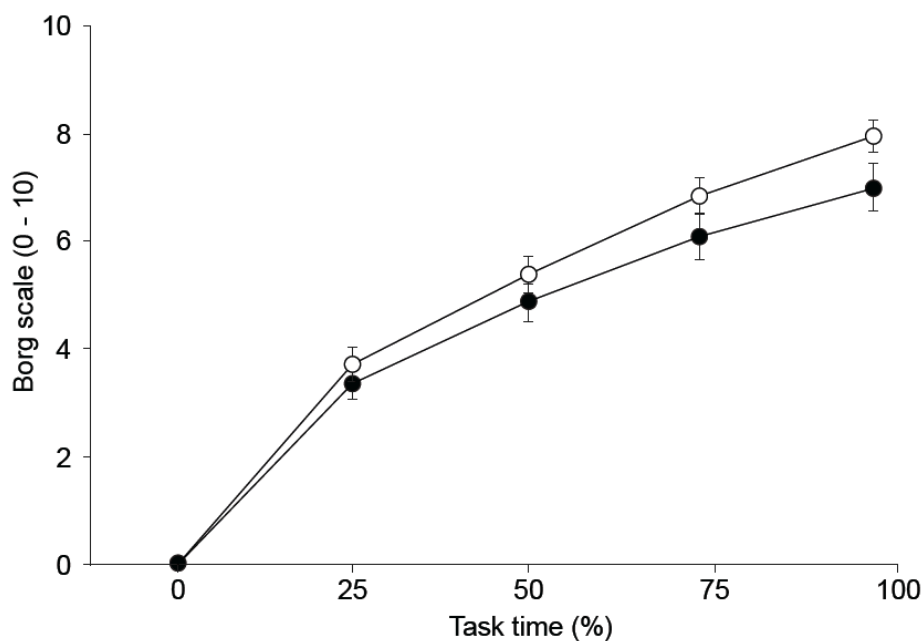


Figure 2. Rating of perceived exertion increased during both force (filled circles) and position (open circles) tasks ($P < 0.001$) but was higher during the position task ($P = 0.021$) and increased more during the position task ($P = 0.006$).

Due to the absence of a significant interaction (muscle x time) for average rectified EMG amplitudes (aEMG) of the agonist muscles (biceps brachii, brachioradialis, and brachialis) during each fatiguing contraction, the results are reported as the average values across the three agonists (Table 1). The aEMG values increased similarly ($P < 0.001$) during the force and

position tasks for both the agonist and antagonist muscles. The aEMGs at the end of both fatiguing contractions were increased significantly relative to the values at the start (% Δ) for both the agonist (force: $39 \pm 25\%$; position: $26 \pm 24\%$) and antagonist (force: $21 \pm 18\%$; position: $26 \pm 27\%$) muscles.

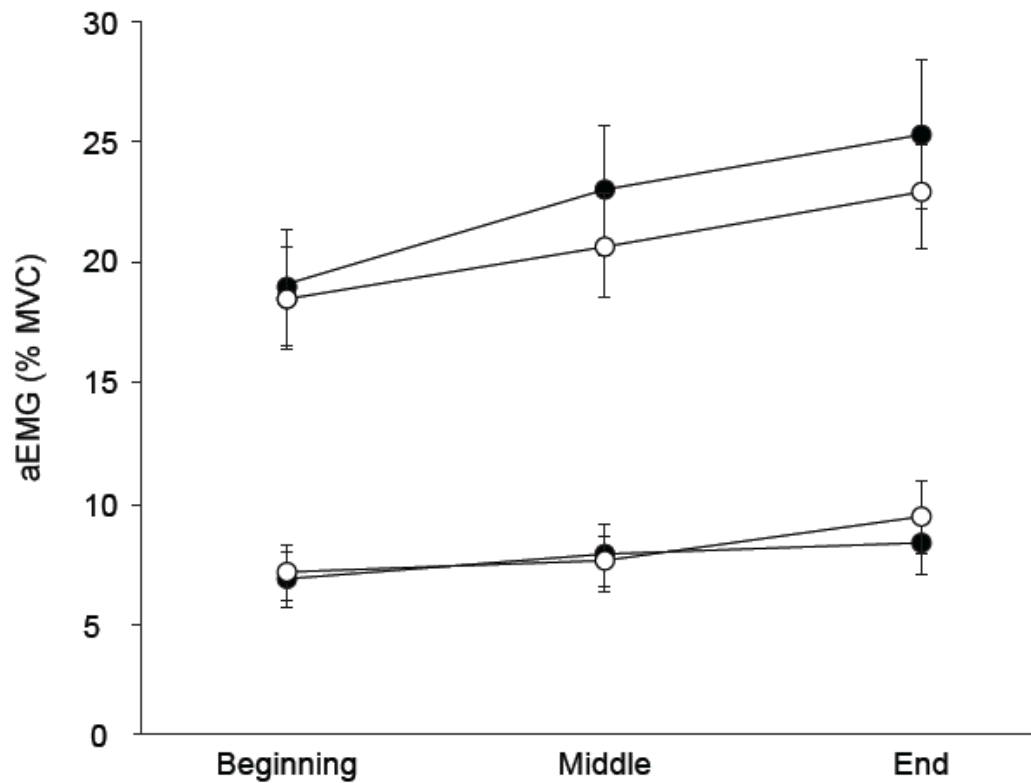


Figure 3. Surface aEMG increased for agonists ($P < 0.001$) and antagonists ($P < 0.001$), but did not differ between the two tasks.

	Force task			Position task		
	Beginning	Middle	End	Beginning	Middle	End
Biceps	14.6 ± 9.8	20.6 ± 12.4	22.8 ± 15.6	15.3 ± 8.3	18.0 ± 9.7	19.8 ± 10.8
Brachialis	25.5 ± 14.8	28.3 ± 15.6	30.4 ± 16.0	25.0 ± 14.9	26.1 ± 13.0	29.0 ± 13.6
<u>Brachioradialis</u>	16.1 ± 9.6	19.4 ± 11.0	22.0 ± 12.3	14.7 ± 7.6	17.3 ± 7.9	19.6 ± 10.0
Agonists	18.7 ± 11.4	22.8 ± 13.0	25.1 ± 14.6	18.3 ± 10.3	20.5 ± 10.2	22.8 ± 11.4*
Antagonist	6.6 ± 5.5	7.6 ± 6.2	8.1 ± 6.4*	6.9 ± 5.5	7.3 ± 5.3	9.2 ± 7.2

Table 1. Average rectified EMG amplitudes (% MVC value) for agonist (biceps brachii, brachialis, and brachioradialis) and antagonist (triceps brachii) muscles during the force and position tasks. Data presented as means ± SD.

The recordings obtained from 23 experimental sessions (2 participants volunteered for a second session) with the subcutaneous electrode were analyzed to determine the discharge times of at least one clearly identifiable motor unit. Most recordings (16) yielded a single motor unit, but it was possible to discriminate two motor units in two recordings and four motor units in one recording. The mean recruitment threshold of the 32 motor units was $21.6\% \pm 11.7\%$ MVC force (range: 0.5-41.2% MVC force). The discharge characteristics of each motor unit were analyzed during both fatiguing contractions. Consistent with the magnitude of the two loads being matched, the mean discharge rate for the 21 motor units was similar at the start of the two fatiguing contractions (Table 2). A significant task x time interaction ($P = 0.01$), however, indicated that the decline in motor unit discharge rate during the two fatiguing contractions (main effect for time, $P < 0.001$) was greater during the position task (Table 2). Moreover, a main effect for task ($P = 0.004$) indicated that discharge rates were lower during the position task.

	Force task			Position task		
	Beginning	Middle	End	Beginning	Middle	End
Discharge Rate, <u>pps</u>	15.9 ± 4.0	13.6 ± 2.8	13.2 ± 3.1*	15.1 ± 3.5	12.2 ± 2.6*	11.1 ± 2.5*
CV for ISI, %	15.4 ± 5.2	14.0 ± 5.4	13.8 ± 5.7*	14.7 ± 4.5	13.8 ± 4.4	17.1 ± 5.8*
Steadiness, CV for force, %	1.7 ± 1.3	1.5 ± 0.7	2.0 ± 0.9*	1.0 ± 0.8	1.0 ± 0.9	1.3 ± 1.3*

Table 2. Motor unit discharge characteristics and force steadiness at three time points during the force and position tasks. Data presented as means ± SD. *P < 0.05 relative to start value. pps, pulse per second; CV, coefficient of variation; ISI, interspike interval.

The variability in discharge times was quantified as the coefficient of variation for interspike interval. A significant task x time interaction ($P = 0.008$) indicated that the coefficient of variation for interspike interval decreased during the force task and increased during the position task (Table 2). Moreover, the index of force steadiness (coefficient of variation for force) increased during both fatiguing contractions (main effect for time, $P = 0.05$), and was less during the position task (main effect for task, $P = 0.05$).

Discussion

In a study on the influence of handedness and dominance on the adjustments during submaximal fatiguing contractions, Gordon et al. (2012) found significant differences between left- and right-handed individuals in how they accommodated a difference in load compliance. In that study, participants sustained an isometric contraction with the elbow flexor muscles to match a submaximal target force (20% MVC force) for as long as possible while pushing against a rigid restraint (low compliance) or supporting an inertial load (high compliance). Limb dominance did not influence endurance time for either group of participants. In contrast, endurance times for the two tasks were significantly correlated ($r^2 = 0.62$) for the right-handers, but not for the left-handers ($r^2 = 0.004$). The main finding of the current study was that adjustments in

discharge rate and the variability in discharge times of biceps brachii motor units differed during force (low-compliance load) and position (high-compliance load) control, but these adjustments were similar to those observed in right-handed individuals performing the same tasks (Mottram et al., 2005).

Handedness and Dominance

As found in the current study, the performance of voluntary actions can differ between dominant and nondominant limbs when the task exceeds some threshold level of coordination (Goble and Brown, 2007; Buckingham et al., 2012; Przbyla et al., 2012). The current findings, for example, included a difference between limbs for left-handed individuals in the time to perform a writing test but no difference in the strength of forearm-hand muscles or, surprisingly, in the time to complete a test of manual dexterity. The absence of a difference in hand strength between limbs for left-handers has been reported previously, whereas the dominant hand is slightly, but significantly, stronger in right-handers (Armstrong and Oldham, 1999). In contrast, others have found that young adults can complete the grooved pegboard test more quickly (Ruff and Parker, 1993), remove more pegs from a pegboard in 30 s (Michmata et al., 2008), and perform the Moberg Pick-Up test more quickly (Amirjani et al., 2007) with the dominant arm than the nondominant arm. Nonetheless, there are no reports of an interaction between limb dominance and handedness on tests of manual dexterity.

There are also some reports of differences in muscle activation that are associated with handedness and limb dominance. For example, De Luca et al. (1986) found that the median frequency of the surface EMG signal for a hand muscle during a fatiguing contraction decreased more for the non-dominant hand of right-handers, whereas there was no difference between

limbs for left-handers. Subsequently, Farina et al. (2003) measured the rate of change in the spectral properties of surface EMG signals obtained from the upper trapezius muscle and observed no difference due to handedness, but a more rapid decrease for the dominant side of both left- and right-handed individuals. These differences are likely attributable to adaptations in the electrical properties of the muscle fiber membranes.

The discharge characteristics of motor units can also differ between left- and right-handed individuals and between limbs, although the findings are mixed. One study found that the discharge characteristics of motor units in a hand muscle differed between limbs when individuals sustained a fatiguing contraction at 30% MVC force (Adam et al., 1998). Analysis of the cross-correlation between discharge times and muscle force indicated a longer latency for the dominant, which was attributed to differences in contractile properties of the two muscles. Motor unit synchronization, which is often used as an index of the relative strength of common input to pairs of motor neurons, was reported to be greater in a forearm muscle of the dominant arm (Schmied et al., 1994), but less in a hand muscle of the dominant arm (Semmler and Nordstrom, 1995). However, force fluctuations during steady, submaximal contractions—which are strongly correlated with the common input to the motor neuron pool (Farina and Negro 2015)—do not differ between limbs (Semmler and Nordstrom, 1995; Pereira et al., 2012).

Taken together, these results suggest that motor function can differ between limbs for actions that require some level of coordination, but that handedness does not appear to influence performance on relatively simple tasks, such as tests of muscle strength and manual dexterity. However, handedness does appear to influence perceptual judgements, intersegmental coordination, and the processing of proprioceptive feedback during arm movements (Goble and Brown, 2007; Buckingham et al., 2012; Przbyla et al., 2012).

Force and Position Control

The findings of Gordon et al. (2012) suggested that handedness also influences the performance of submaximal fatiguing contractions that required either force or position control. In contrast to right-handed participants, they found that the endurance times for the two fatiguing contractions were not correlated in left-handers. In right-handers, endurance time for the submaximal fatiguing contraction is consistently less during position control (Hunter et al., 2002; Maluf et al., 2005) and is associated with more rapid adjustments in motor unit activity (Mottram et al., 2005; Baudry et al., 2009; Rudroff et al., 2010). Moreover, the modulation of feedback transmitted by group Ia afferents differs for the tasks (Baudry et al., 2010, 2011). The finding of no consistent difference in the endurance times for the two fatiguing when performed by left-handers, therefore, suggests that there may be differences in the adjustments in motor unit activity during these two tasks due to handedness.

The findings of the current study, however, demonstrate that the decline in discharge rate and the increase in discharge variability of biceps brachii motor units in left-handers were greater during the position task. The results are consistent with a previous report on the adjustments in motor unit activity during the two types of fatiguing contractions in right-handers (Mottram et al., 2005). Taken together, the two sets of results indicate that handedness does not influence the adjustments in motor unit activity during fatiguing contractions requiring either force or position control. Thus, despite differences in the adjustments in motor unit activity during force and position control, these changes are not influenced by either handedness or dominance.

Chapter V

Association between intellectual capacity, fatigue, and neuromuscular function in persons with multiple sclerosis

Abstract

Fatigue is one of the most debilitating symptoms of multiple sclerosis (MS) and the underlying mechanisms are poorly understood. When exposed to a physical or cognitive challenge, persons with MS exhibit a decline in task performance and increased fatigue. These effects, however, can be attenuated by intellectual capacity. The purpose of the current study was to examine the influence of intelligence on the associations between trait levels of fatigue, performance fatigability, and perceived fatigability for persons with MS. We hypothesized that intellectual capacity confers some protection against heightened levels of fatigue and fatigability caused by MS. Twelve adults (1 male) with relapsing-remitting MS and 12 control (CO) subjects (1 male) were matched for age, sex, and intellectual capacity, which was estimated using the Verbal score of the Wechsler Abbreviated Scale of Intelligence. Trait fatigue was assessed using the modified fatigue impact scale (MFIS) questionnaire. Performance fatigability was measured as the decline in maximal voluntary (MVC) torque after 60 isometric contractions (10-s contraction, 5-s rest) performed at 25% MVC with the knee extensor muscles. Estimates of perceived fatigability (rating of perceived exertion; RPE), torque steadiness, and EMG activity were recorded during the fatigue protocol. Persons with MS reported greater trait fatigue via questionnaire (MS: 43.1 ± 14.4 ; CO: 11.25 ± 8.4 , $P \leq 0.001$). Initial MVC torque for the knee extensors did not differ between the two groups (MS: 112 ± 38 N•m; CO: 107 ± 44 N•m) and performance fatigability (decline in MVC torque) was similar for the two groups (MS: 15 ± 19 N•m; CO: 13 ± 16 N•m). Verbal IQ was used to control for premorbid intellectual capacity (MS: 112 ± 13 ; CO: 114 ± 10). RPE increased during the fatiguing contraction for both groups ($P < 0.001$), but was significantly greater in magnitude ($P = 0.03$) and increased more for the MS group (group x time interaction, $P = 0.05$). Coefficient of variation for torque increased during the fatigue protocol (main effect

for time, $P = 0.05$) and torque steadiness was less for the MS group (main effect for group, $P = 0.02$). Verbal IQ was correlated with the decline in torque steadiness ($r = -0.82$, $P = 0.001$).

Intellectual capacity was not associated with either the trait level of fatigue or the two measures of fatigability (performance and perceived) in persons with MS, but was associated with adjustments in muscle activation during the fatigue protocol.

Introduction

Multiple sclerosis is a neurological disorder that compromises the integrity of signaling pathways in the nervous system through acute inflammatory responses and chronic neurodegeneration.

Although the clinical progression of MS varies widely between individuals (Lublin and Reingold, 1996; Confavreux and Vukusic, 2006), one of the most common symptoms of the disease is a heightened level of fatigue experienced by patients. Symptoms of fatigue can severely limit activities of daily life and are often reported to have a more negative impact on quality of life than physical disability (Amato et al., 2001; Janardhan and Bakshi 2002). In addition to the debilitating effects of fatigue, multiple sclerosis often leads to a variety of other cognitive and motor impairments. A number of studies, however, have shown that possessing a higher premorbid intellectual capacity may offer protection against declines in cognitive function with advancing neurological disease, an idea referred to as cognitive reserve.

The concept of cognitive reserve posits that cognitive impairments develop more gradually as a neurological disease progresses in people with greater intellectual capacity (Stern, 2009).

Persons with MS who have experienced a lifetime of intellectual enrichment, for example, can achieve comparable levels of performance on behavioral tasks with less modulation of cortical

activity compared with those who have fewer years of education or lower vocabulary scores (Benedict et al., 2010; Sumowski et al., 2010).

Although current evidence for the relation between self-reported fatigue and cognitive function appears equivocal (Morrow et al., 2009), persons with MS often report that fatigue interferes with cognitive functioning (Krupp et al., 1988; Monks, 1989) and that increases in fatigue occur after experiencing a cognitive challenge (Bailey et al., 2007; Tartaglia et al., 2008).

The purpose of the current study was to examine the influence of intelligence on the associations between trait levels of fatigue, performance fatigability, and perceived fatigability in persons with MS. Given that intellectual enrichment protects against MS-related declines in cognitive function (Sumowski et al., 2009a, 2009b) and cerebral efficiency (Sumowski et al., 2010) and that fatigue is associated with greater levels cortical activity, we hypothesized that intellectual capacity may confer some protection against heightened levels of fatigue and fatigability caused by MS.

Methods

Twelve adults (38.6 ± 14.5 yrs, 1 male) with a diagnosis of relapsing-remitting multiple sclerosis and 12 control subjects (40.6 ± 14.4 yrs, 1 male) who were matched for age, sex, and intellectual capacity participated in the study after written informed consent was obtained. Participants with MS were included if they were taking a stable dose (no prescription change for ≥ 90 days) of disease-modifying medications, able to walk ≥ 100 m unassisted, complained that fatigue interfered with their quality of life, and did not experience a relapsing episode within 90 days of participating in the study. None of the participants had a history of cardiovascular incidents, seizures, or traumatic brain injuries and all of the participants were right-handed.

The experimental protocol was approved by the University of Colorado Boulder Institutional Review Board (protocol#: 12-0421) and conformed to the *Declaration of Helsinki*. The study involved one visit to the laboratory, which lasted approximately two hours, and was used to assess intellectual capacity, executive function, walking performance, perceived fatigability, and performance fatigability.

Intellectual Capacity

Intellectual capacity was assessed using the Wechsler Abbreviated Scale of Intelligence (WASI) (Wechsler, 1999). The test provides a performance score derived from: the block design and matrix reasoning subtests, a verbal score based on the vocabulary and similarities subtests, and a Full-4 scale that is a composite of the performance and verbal scores. Verbal performance is resistant to declines caused by the progression of MS (Chiaravalloti and DeLuca 2008; Lezak, 2004; Sumowski et al., 2009) and was therefore used to estimate premorbid intellectual capacity and to match control participants to those in the MS group. The Behavior Rating Inventory of Executive Function served as an additional measure of cognitive function by assessing difficulties in goal-directed behavior during activities of daily living (Roth et al., 2005).

Walking Performance

The 25-foot walk test was used to measure functional capacity of the lower extremities and was performed without the use of assistive devices. Participants were directed to one end of a 25-foot distance, which was clearly marked with white lines at both ends. The instructions were to walk as quickly as possible from one end to the other, but safely, and without slowing down until after crossing the line. Measurements were made by one of the investigators using a digital

stopwatch. The walk was performed a second time in the opposite direction, and the faster of the two times was reported.

Perceived Fatigability

Trait levels of fatigue experienced by participants were quantified using the Modified Fatigue Impact Scale (MFIS) questionnaire, which considers physical, cognitive, and social factors influencing perceived fatigability (Kos et al., 2006). Participants were asked to indicate how often fatigue has had an influence on 21 different situations over the 4 weeks immediately preceding the study. Additionally, participants were asked to report state levels of fatigue at seven time points during the fatiguing contraction experiments using the modified Borg scale (6-20) (Borg, 1987).

Performance Fatigability

Participants performed 60 intermittent isometric contractions with the knee extensor muscles to a target of 25% MVC torque. The protocol was performed with both the left and right legs in a randomized order with approximately 20 min of rest between the two trials. Each intermittent contraction was sustained for 10 s with 5 s of rest before the next contraction. MVC torque was measured before and after the fatiguing protocol.

After determining MVC torque, participants were asked to increase knee extensor torque to the target (25% MVC torque) displayed on the monitor (gain: 3% MVC force/cm) in front of them and hold it as steady as possible. The investigator verbally prompted the participant to contract for 10 s and then to relax for 5 s. Rating of perceived exertion (RPE) was measured with the

modified Borg scale (6–20) after every 10th contraction. The scale was anchored so that a 6 represented rest or no exertion, and 20 represented the strongest possible effort.

Experimental Setup

Participants were seated in an upright position with hips and knees flexed to ~1.57 rad. A strain gauge force transducer (JR3, Woodland, CA) was positioned to contact the anterior aspect of the lower leg (~8 cm above the ankle). The applied force was displayed on a monitor located at eye level ~60 cm in front of the subject. The distance between the axis of rotation for the knee joint and the horizontal plane of the lower leg that contacted the center of the force transducer was measured to calculate torque about the knee joint. The upper body was secured to the chair using 5-cm wide nylon straps across the shoulders and lap in order to minimize movement during contractions.

Electromyographic (EMG) signals were recorded from the vastus lateralis muscle using Ag-AgCl surface electrodes (Covidien, Mansfield, MA) arranged in a bipolar configuration. Three pairs of electrodes were placed over the vastus lateralis along a line between the anterior superior iliac spine and the lateral border of the patella in the presumed direction of the muscle fibers. Three reference electrodes were placed over the medial surface of the tibia. EMG signals were amplified (1000x), band-pass filtered (13–1000 Hz) (Coulbourn Instruments, Allentown, PA, USA), sampled at 2 kHz (Cambridge Electronic Design, Cambridge, England), and stored on a computer (Dell, Plano, TX, USA). Reported EMG data were normalized to values obtained during maximal isometric contractions performed before the fatigue protocol.

Maximal Voluntary Contractions

MVC torque was measured before the fatigue protocol to determine maximal knee extensor strength, to provide a reference value for calculating a target torque of 25% MVC, and to record peak EMG activity. MVC torque was measured again immediately after the fatigue protocol to assess performance fatigability. The MVCs were performed with the knee extensor muscles by gradually increasing torque from rest to maximum over ~ 3 s and maintaining this torque for ~ 3 s. The investigators provided strong verbal encouragement during each MVC, and the gain of the visual feedback was adjusted between contractions. At least two trials were performed with ≥ 90 s of rest between consecutive trials. If the peak torques were not within 5% of each other for the two trials, or if a participant indicated that one of the efforts were not maximal, additional MVCs were performed until those criteria were met. The greatest peak torque during these trials was used as the maximum value.

Data Analysis

Force and EMG data were sampled by an analog-to-digital converter (Power 1401; Cambridge Electronic Design, Cambridge, England) and stored on a computer for offline analysis using Spike2 data acquisition/analysis software (Cambridge Electronic Design, Cambridge, England). Force signals were sampled at 1 kHz, and EMG signals were sampled at 2 kHz.

The maximal EMG amplitude during the MVC trial with the maximal torque was quantified as the root-mean-square (RMS) value during a 0.5-s interval centered about the peak of the EMG and was used to normalize subsequent EMG recordings. EMG data collected during the fatigue protocol were quantified as the normalized RMS value during seven 8-s intervals for the first, 10th, 20th, 30th, 40th, 50th, and 60th contraction. Force steadiness was quantified during the same 8-s intervals as the coefficient of variation for torque.

Statistics

Dependent t-tests were used to compare dependent variables before and after the fatiguing contractions and independent t-tests were used to compare group differences of dependent variables and subject characteristics. Two-factor analysis of variance was used to examine changes in dependent variables measured over 7 time points (first contraction, and every tenth contraction up to the 60th contraction) and differences between the two groups (MS and CO). Dependent variables measured during the fatigue protocol included EMG amplitude of the vastus lateralis muscle, knee extensor torque steadiness (the coefficient of variation for torque), and ratings of perceived exertion. One of the subjects in the MS group was unable to complete all 60 contractions, so data obtained during the last performed contraction were also used for the 50th and 60th contractions. Pearson correlation coefficients were determined for all measures of physical and cognitive function in the two groups. Data are presented as mean \pm standard deviation (SD) in the text and as mean \pm standard error (SE) in the figures.

Results

Individuals with MS reported significantly greater levels of trait fatigue than the control (CO) group, as measured with the MFIS questionnaire (Figure 1A; MS: 43.1 ± 14.4 ; CO: 11.25 ± 8.4 , $P \leq 0.001$). Nonetheless, MS and CO participants were matched for verbal IQ (Table 1; MS: 112.3 ± 13.1 ; CO: 114.3 ± 9.8), and both performance IQ (MS: 119.5 ± 14.0 ; CO: 121.7 ± 11.1) and Full-4 IQ (MS: 118.1 ± 14.4 ; CO: 119.7 ± 9.7) scores were also similar between groups. Thus, the overall intellectual capacity of the MS group was similar to that for the CO group, despite the MS participants having been diagnosed with the disease for 9.4 ± 6.4 yrs (range: 0.2 – 19 years) prior to participating in the study. However, a measure of executive function (BRIEF)

was significantly greater for the MS group (MS: 121.9 ± 19.2 ; CO: 89.6 ± 17.4 , $P \leq 0.001$), which indicates lower executive function in the MS participants. Greater BRIEF scores were correlated with greater levels of trait fatigue (MFIS), but this relation was only significant ($P < 0.05$) for the CO group (Table 2).

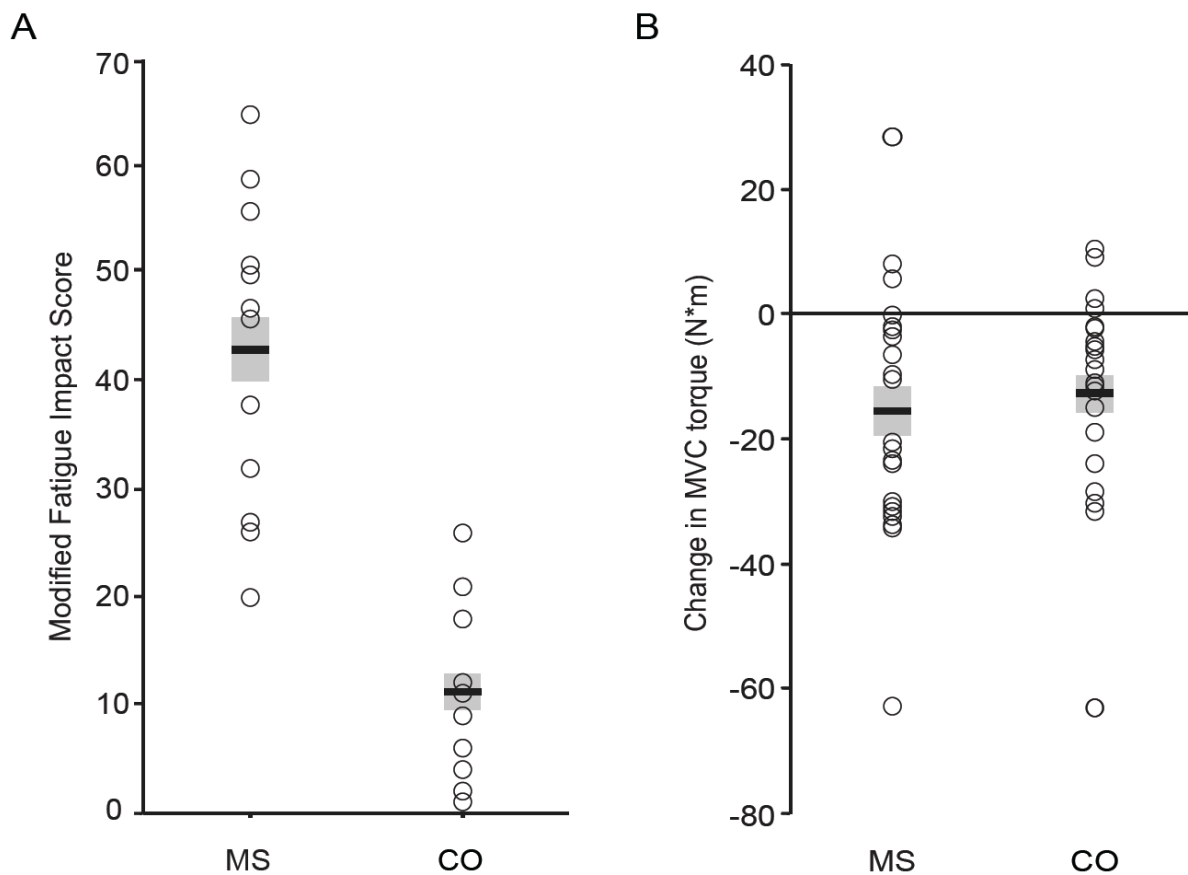


Figure 1. *Perceived trait fatigue* (A). Reported levels of fatigue (MFIS score) were greater for the MS group ($P = 0.001$). *Performance fatigability* (B). Declines in MVC torque were significant ($P = 0.001$ for both groups), and did not differ between groups ($P = 0.74$).

Table 1

Subject characteristics.

	Multiple Sclerosis (MS)	Control (CO)
	<i>n</i> = 12, 1 male	<i>n</i> = 12, 1 male
Age (yr)	38.6 ± 14.5	40.6 ± 14.4
Height (cm)	146 ± 55	170 ± 50
Body mass (kg)	52 ± 20	65 ± 23
MFIS	43.1 ± 14.4	11.25 ± 8.4†
WASI-IQ		
Verbal	112.3 ± 13.1	114.3 ± 9.8
Performance	119.5 ± 14.0	121.7 ± 11.1
Full-4	118.1 ± 14.4	119.7 ± 9.7
BRIEF	121.9 ± 19.2	89.6 ± 17.4†
Depression	5.2 ± 2.6	1.3 ± 1.7
25-foot walk (s)	3.6 ± 0.4	3.3 ± 0.4
MVC torque (N•m)	111.9 ± 37.8	107.2 ± 43.7
MVC decline (N•m)	15.5 ± 18.8	12.9 ± 15.9

MFIS, Modified Fatigue Impact Scale; WASI, Wechsler Abbreviated Scale of Intelligence; BRIEF, Behavior Rating Inventory of Executive Function; Depression, Beck Depression Inventory; MVC torque refers to knee extensor muscle strength before the fatiguing contracts, whereas MVC decline refers to the observed decrease in torque after the fatigue protocol. † $P \leq 0.001$ between MS and CO groups.

Table 2

Pearson correlation coefficients for pairs of variables are shown for both multiple sclerosis subjects and control subjects.

	Verbal IQ	Performance IQ	Full-4 IQ	BRIEF	Depression	ΔRPE	Δ MVC	Δ EMG	Δ CV for Torque
MFIS	-0.07/0.09	-0.46/-0.04	-0.26/0.04	0.42/0.70*	0.66/0.52	-0.26/0.09	-0.20/0.57*	0.12/0.45	0.09/0.33
Verbal IQ	1	0.73†/0.30	0.94‡/0.80†	-0.20/0.23	-0.64/-0.09	-0.36/0.01	0.04/0.24	-0.54/-0.01	-0.82‡/0.00
Performance IQ		1	0.92‡/0.80†	-0.50/-0.23	-0.80/-0.56	-0.24/-0.39	0.29/-0.02	-0.40/-0.14	-0.60*/0.17
Full-4 IQ			1	-0.38/0.02	-0.78/-0.37	-0.33/-0.23	0.17/0.18	-0.50/-0.08	-0.76†/0.10
BRIEF				1	0.69/0.35	0.27/0.48	0.18/0.57*	-0.06/0.34	0.14/0.48
Depression					1	-0.20/0.17	-0.33/0.75†	0.31/0.52	0.82/0.06
Δ RPE						1	0.10/0.27	0.53/-0.02	0.51/-0.03
Δ MVC							1	0.03/0.45	0.14/0.37
Δ EMG								1	0.86‡/0.69†

Pearson correlations for each pair of variables are presented in the table as MS/CO. Changes in rating of perceived exertion – Borg scale 6-20 (RPE), maximal voluntary contraction torque (MVC), electromyography for vastus lateralis (EMG), and coefficient of variation for torque (CV for Torque) refer to differences between initial and final measurements. * $P \leq 0.05$, † $P \leq 0.01$ and ‡ $P \leq 0.001$.

There were no statistically significant group differences in either the 25-foot walk test (MS: 3.6 ± 0.4 s; CO: 3.3 ± 0.4 s) or knee extensor MVC torque prior to the fatigue protocol (MS: 112 ± 38 N•m; 107 ± 44 N•m). Participants performed the fatigue protocol with both legs separately due to the possibility of disease progression having an unequal effect on both sides of the body. However, changes in EMG, RPE, coefficient of variation for torque, and declines in MVC torque did not differ between left and right legs for either group of participants, so these data were collapsed across legs. The two groups exhibited similar significant declines in MVC torque after the fatigue protocol (Figure 1B; MS: 16 ± 19 N•m; CO: 13 ± 16 N•m). The declines in MVC torque for the CO group, but not the MS group, were associated (Table 2) with greater levels of trait fatigue (MFIS) ($r = 0.57$, $P \leq 0.05$), BRIEF scores ($r = 0.57$, $P \leq 0.05$), and depression ($r = 0.57$, $P \leq 0.01$).

There was no group difference ($P = 0.49$) in the increase in normalized EMG amplitude for vastus lateralis (MS: $38 \pm 37\%$ increase; CO: $31 \pm 24\%$ increase) during the fatigue protocol (Figure 2; $P < 0.001$). Although the coefficient of variation for torque increased for both groups (MS: $40 \pm 49\%$ increase; CO: $66 \pm 187\%$ increase) during the fatigue protocol (Figure 3; main effect for time, $P = 0.05$), the MS group was less steady (main effect for group, $P = 0.02$). Increases in CV for torque were strongly correlated with increases in EMG amplitude for both MS ($r = -0.86$, $P \leq 0.001$) and CO ($r = -0.69$, $P \leq 0.01$) groups (Table 2). In addition, the increase in the coefficient of variation for torque during the fatigue protocol observed for the MS group (start: $2.5 \pm 1.6\%$; end: $3.9 \pm 4.3\%$) was inversely correlated (Table 2) with all three measures of intellectual capacity (Verbal IQ: $r = -0.82$, $P \leq 0.001$; Performance IQ: $r = -0.60$, $P \leq 0.05$; Full-4 IQ: $r = -0.76$, $P \leq 0.01$).

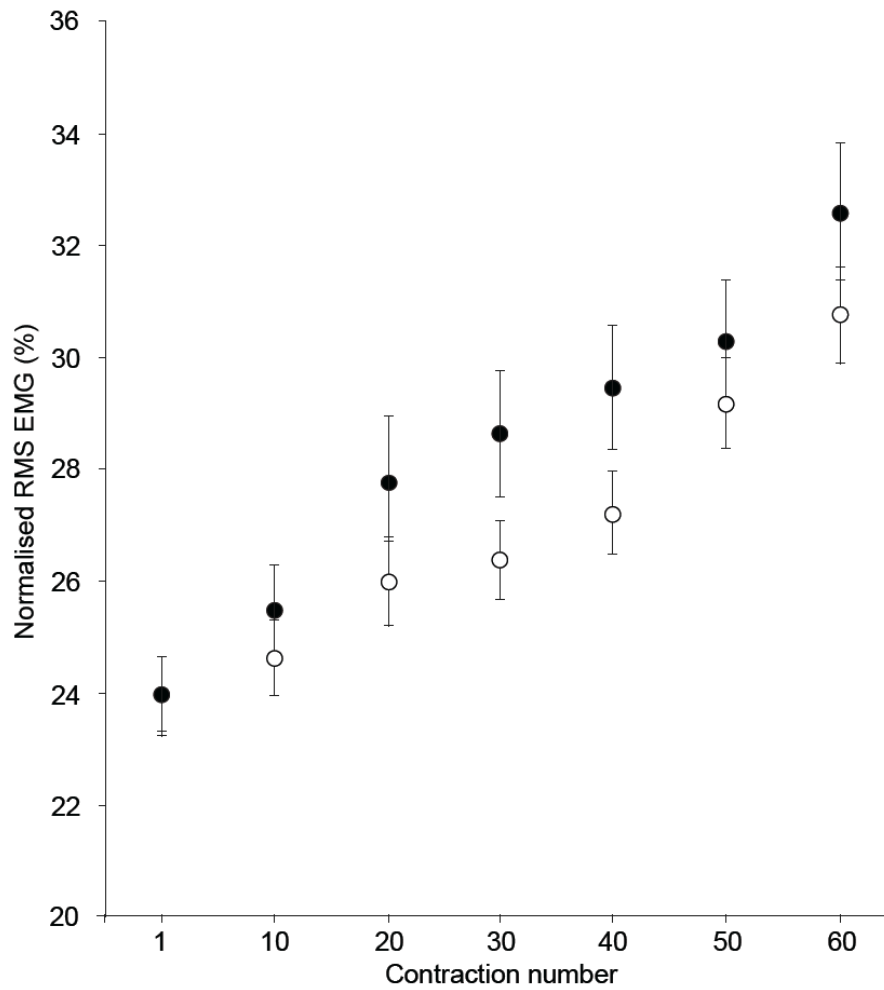


Figure 2. Normalized EMG amplitude for the vastus lateralis muscle in MS (filled circles) and CO subjects (open circles) increased during the fatigue protocol ($P < 0.001$) with no difference between groups ($P = 0.49$).

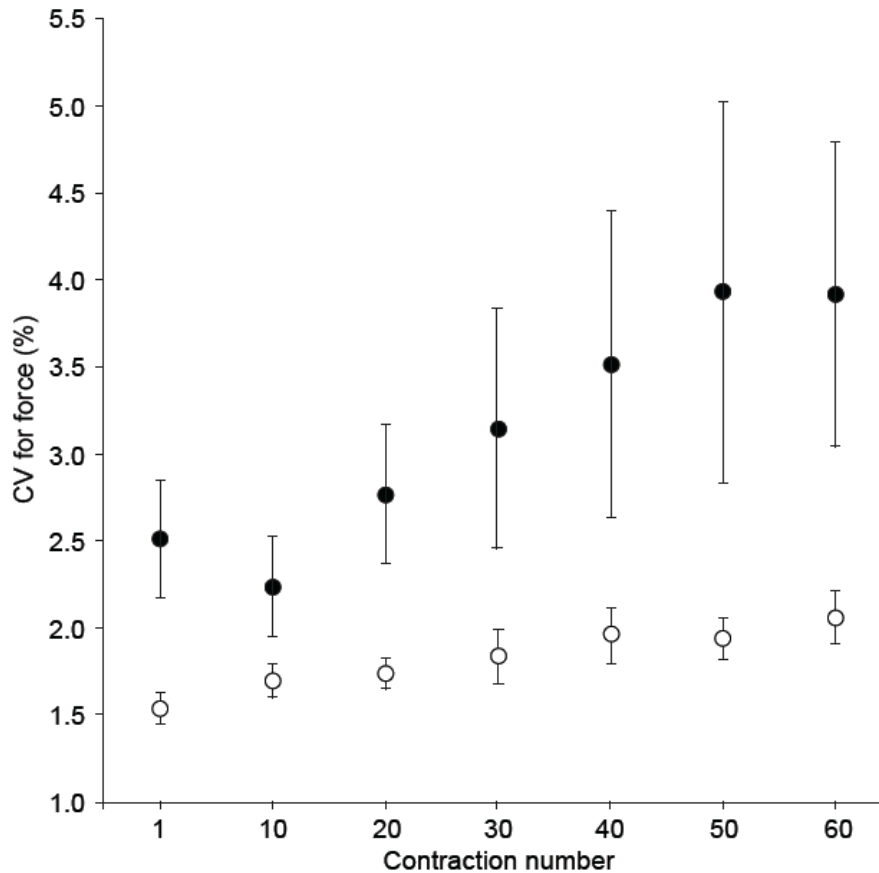


Figure 3. Coefficient of variation (CV) for torque increased during the fatigue protocol (main effect for time, $P = 0.05$) for the MS (filled circles) and CO subjects (open circles) and the MS group was less steady (main effect for group, $P = 0.02$).

RPE increased for both groups during the fatigue protocol (Figure 4; $P < 0.001$), but was significantly greater in magnitude ($P = 0.03$) and increased more for the MS group (group x time interaction, $P = 0.05$). The MS group experienced a greater incidence of reaching maximal efforts with 5 participants reaching a final RPE of 20 at the end of the fatigue protocol and only one participant for the CO group.

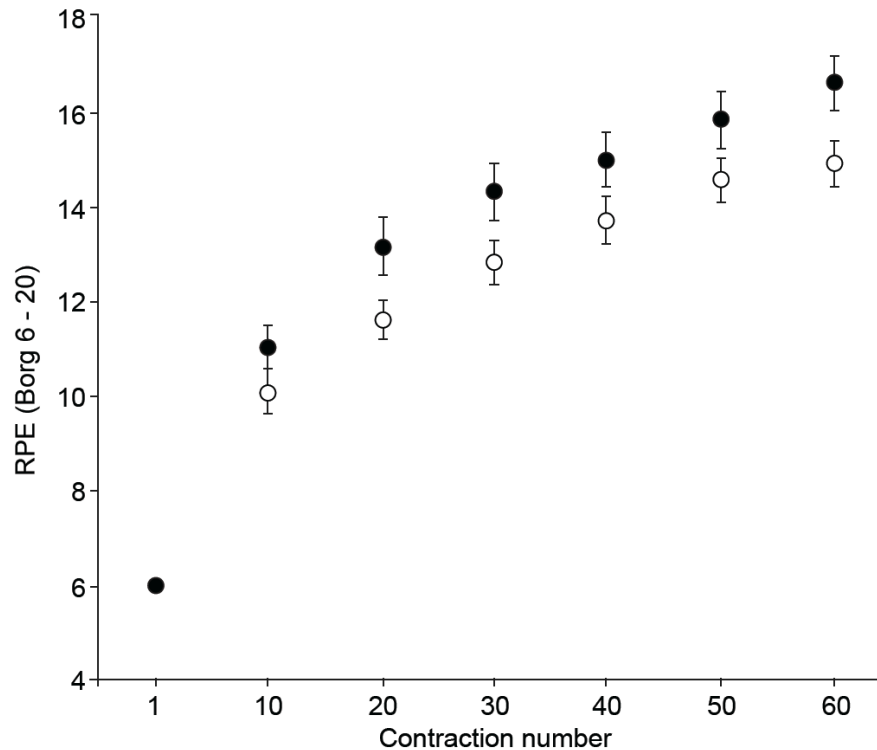


Figure 4. Rating of perceived exertion (RPE) increased more for the MS group (filled circles) during the fatigue protocol (group X time interaction, $P = 0.05$) than for the control group (open circles). Values reported as mean \pm SE.

Discussion

The current study compared the adjustments exhibited by persons with MS to healthy control subjects during a fatigue protocol that required isometric contractions with the knee extensor muscles. The two groups of participants were matched on the basis of age, sex, and premorbid intellectual capacity (Verbal IQ). Additionally, there were no group differences for Performance IQ, indicating that cognitive function did not differ between the two groups when performing the block design and matrix reasoning tests. Moreover, MVC torque for the knee extensors and time to complete the 25-foot walk test did not differ between groups. In contrast, the MS participants

reported greater trait levels fatigue (MFIS) and produced lower scores on a test of executive function (BRIEF). Although the decline in MVC torque after the fatigue protocol (performance fatigability) was similar for the two groups, the state level of fatigue (RPE) and the coefficient of variation for knee extensor torque (torque steadiness) increased more during the fatigue protocol for the MS group. Notably, there was an inverse relation between the measures of intellectual capacity and the changes in torque steadiness during the fatigue protocol for the MS group.

Fatigue and Fatigability

Fatigue has been defined as a disabling symptom in which physical and cognitive function is limited by interactions between performance fatigability and perceived fatigability (Enoka and Duchateau, in press). In the current study, fatigue was quantified as a trait characteristic with the MFIS questionnaire. As expected due to the nature of the disease, the MS participants reported greater MFIS scores (Krupp et al., 1988; Fisk et al., 1994; Alvarenga-Filho et al., 2015). Despite greater trait levels of fatigue for the MS participants, the decline in knee extensor torque after the fatigue protocol—a measure of performance fatigability—was similar for the two groups. Steens et al. (2012a) reported similar observations when they compared the decrease in MVC force after a 2-min sustained maximal contraction with a hand muscle exhibited by 20 persons with relapsing-remitting MS and 20 age- and sex-matched control subjects. Based on a multiple-regression analysis to explain the variance in the trait level of fatigue (Fatigue Severity Scale, FSS) for the MS participants, they found that 45% of the variance in the FSS score could be explained by the measure of performance fatigability (decline in MVC force) and normalized muscle strength (MVC force). When a measure of depression (Hospital Anxiety and Depression Scale questionnaire) was included in the analysis, the regression model explained 77% of variance in the trait level of fatigue (FSS score).

Despite the similar level of performance fatigability for the two groups in the current study, the level of perceived fatigability (rating of perceived exertion) increased more during the fatigue protocol for the MS participants. In another study, Steens et al. (2012b) compared the adjustments exhibited by persons with relapsing-remitting MS and control subjects during the 2-min sustained maximal contraction with a hand muscle. As in their other study (Steens et al., 2012a), performance fatigability (decline in MVC force) was similar for the two groups of participants. Nonetheless, there were significant differences between the two groups in the changes in the level of voluntary activation (twitch interpolation technique) and cortical activation as determined with functional MRI. Compared with the control subjects, the MS participants exhibited a more substantial and variable reduction in voluntary activation and less of an increase in cortical activating during the fatiguing contraction. A multiple-regression analysis indicated that control subjects who were stronger experienced greater performance fatigability, as reported in other studies (Hunter and Enoka, 2001; Keller-Ross et al., 2014), and that performance fatigability for the individuals with MS was less for those who could sustain greater levels of voluntary activation.

Based on the results of Steens et al. (2012b), the finding of a greater increase in perceived fatigability (RPE) during the fatigue protocol in the current study might be explained by a tendency for persons with MS to experience a greater reduction in voluntary activation during fatiguing contractions. Thus, it is necessary for these individuals to increase the levels of cortical activation to achieve the submaximal target torque during the fatigue protocol.

Muscle Activation and Intelligence

Due to the association between the increase in perceived fatigability (RPE) and EMG amplitude during submaximal fatiguing contractions (Hunter et al., 2002; Rudroff et al., 2007), the more rapid increase in RPE for the MS group in the current study should have been accompanied by a quicker increase in EMG amplitude during the fatigue protocol. In contrast, the increase in EMG amplitude for the vastus lateralis muscle was similar for the two groups (Figure 2). However, normalized measures of EMG amplitude are relatively insensitive to modest changes in the underlying motor unit activity (Mottram et al., 2005; Farina et al., 2008; Rudroff et al., 2013).

Nonetheless, the more rapid increase in the coefficient of variation for torque—a measure of torque steadiness—for the MS group suggests that the muscle activation signal differed between the two groups during the fatigue protocol. The synaptic inputs received by the pools of motor units involved in the fatigue task arise from three sources: a control signal received by all motor neurons, independent noise, and common noise (Farina and Negro, 2015). Although the timing of the action potentials discharged by each motor neuron is relatively independent, the control signal to produce the required muscle force (neural drive to muscle) can be identified by low-pass filtering (<10 Hz) the cumulative train of action potentials generated by the activated motor neurons. Critically, there is a strong association between the estimated neural drive to muscle and the amplitude of the force fluctuations (force steadiness) during a steady submaximal contraction (Farina et al., in press). Thus, the more rapid increase in the coefficient of variation for torque during the fatigue protocol for the MS participants in the current study suggests quantitative differences between the two groups in the generation of control signal by the nervous system during this task.

During the muscle contraction protocol, the CV for torque increased for both groups, but the increase was more rapid and more variable for the MS group. Two motor unit properties that can

influence the magnitude of fluctuations in torque are motor unit force and discharge variability (Enoka et al., 2003). While there is no reason to suspect that motor unit size or force production is different in this sample, it has been shown that motor unit discharge rates tend to be lower and more variable in persons with MS (Dorfman et al., 1989), including during maximal efforts (Rice et al., 1992) and may therefore explain the declines in steadiness seen in the MS group despite no differences in initial maximal strength or declines in MVC force (Laidlaw et al., 2000).

Findings for the current study indicated a strong correlation between the increase in torque steadiness during the fatigue protocol and intellectual capacity for the MS group, but not the control group. Studies that have examined the interactions between cognitive and motor function have shown that the ability to exert a steady force during a voluntary contraction is compromised when the individual is exposed to a cognitive challenge (Zijdwind et al., 2006), and that this effect is greater during fatiguing contractions (Lorist et al., 2002) and in older adults (Vanden Nivenet et al., 2014).

D'Orio and colleagues (2012) examined the relation between intellectual capacity and neuromuscular function in persons with MS. Measures of processing speed, executive function, memory and general intelligence were among the cognitive variables that were significantly associated with walking speed and fall frequency. General intelligence as measured by the WASI test was associated with walking speed ($r = -0.272$, $P = 0.017$) as well as frequency of falls ($r = -0.229$, $P = 0.043$). Measures of intellectual capacity were not correlated with walking performance in the current study, and time to complete the 25-foot walk did not differ between the two groups. However, intellectual capacity was correlated with the increased torque steadiness during the fatigue protocol for the MS group, which may indicate that changes in the neural control of lower limb function may precede declines in walking performance.

Conclusion

The underlying mechanisms for the trait level of fatigue in persons with MS remain largely unexplained. Although the current study found no statistically significant associations between the trait level of fatigue and measures of performance and perceived fatigability for the MS participants during a fatigue protocol, intellectual capacity was associated with adjustments in muscle activation during the fatigue protocol. This finding substantiates further inquiry into the relation between intellectual capacity and physical performance so as to inform clinical practice in the management of cognitive and physical declines in neurological diseases such as multiple sclerosis.

Chapter VI

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