Trans-Tibial Amputation as a Model to Evaluate the Role of Cutaneous Sensation, Proprioception and Muscular Strength on Balance Performance

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<u>Abstract</u>

Each year, the United States reports around 185,000 limb amputations (Owings & Kozak, 1998). By the year 2050, amputation's prevalence is expected to double 2005' prevalence, affecting close to 3.6 million of individuals (Ziegler-Graham et al., 2008).

The purpose of this research project was to investigate the role of cutaneous sensation, proprioception and muscular strength on balance performance in unilateral traumatic trans-tibial amputees (TTA) under two different conditions, quiet stance and squatting. We proposed that cutaneous sensation, proprioception and strength are reduced in the non-amputated side of TTA. In addition, the center of pressure velocity (COPv), the root-mean-square displacement (RMSd) and the root-mean-square velocity (RMSv) were expected to increase on the non-amputated side. The last hypothesis was that the decrease of balance performance in traumatic TTA is due at least in part to reduced cutaneous information, proprioception and strength.

Seven traumatic TTA (6 M/1F, age: M = 36.0, SD = 12.8 years old) and seven able-bodied controls (6M/1F, age: M = 39.9, SD = 8.1 years old), matched for sex, age, and level of physical activity, volunteered to participate in this project. Balance assessment was conducted through the analysis of center of pressure (COP). The evaluation was performed on the non-amputated limb of traumatic TTA and a randomly selected limb in able-bodied controls during single-legged stance. The test included three random conditions: 1-standing still with eyes open (EO), 2standing still with eyes closed (EC), and 3- squatting with EO. Three additional measurements on the same limb as balance included: touch pressure sensation (TPS), proprioception, and muscular strength. The study revealed significant reduction of COPv and RMSv for the medial-lateral (ML) direction in amputees as compared to controls. Muscular strength also evidenced significant differences for the knee and the ankle joints with lower peak-torque-to-body-weight in knee flexors (FLX) and ankle dorsal-flexor (D-FLX) muscles compared to knee extensors (EXT) and ankle plantar-flexor (P-FLX) muscles respectively. Significant correlations were observed between COP variables and muscular strength, in particular to ankle strength.

From our study we can conclude that balance is altered in amputees, with lower values in COPv and RMSv on the sound limb of amputated individuals as compared to able-bodied controls. This decrease in COP variables may represent better balance in amputees that could be explained, at least in part, by amputees relying more on their sound limb on a day-to-day basis during ambulation and standing. However, lower values of COP variables do not necessarily indicate better balance performance as a decrease of COP variables may be related to a reduced ability to control balance.

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Abbreviations

- AAS: Adjusted activity score
- ABS: Area of the base of support
- AKA: Above-knee amputee
- ANOVA: Analysis of variance
- AP: Anterior-posterior
- BKA: Below-knee amputee
- CNS: Central nervous system
- COG: Center of gravity
- COM: Center of mass
- COP: Center of pressure
- COPv: Center of pressure velocity
- CSA: Cross-sectional area
- D-FLX: Dorsal-flexor
- EC: Eyes closed
- EO: Eyes open
- EXT: Extensors
- FLX: Flexors
- HAP: Human activity profile

LLA: Lower limb amputation

MAS: Maximum activity score

ML: Medial-lateral

P-FLX: Plantar-flexor

PT-BW: Peak torque to body weight

RJP: Reproduction of joint position

RMS: Root-mean-square

RMSd: Root-mean-square displacement

RMSv: Root-mean-square velocity

TADM: Threshold assisted detection of motion

TDPM: Threshold detection of passive motion

TPS: Touch pressure sensation

TTA: Trans-tibial amputee

Introduction

Amputation is a surgical procedure, which, in the past 15 years, has risen due to diseases such as diabetes and peripheral vascular disease (Gregg et al., 2014; Malyar et al., 2013), and also due to traumatic injuries (Fergason et al., 2010). Commonly, the motor and somatosensory system reorganizes, and the changes in the representation of the mental corporal schema seem to be related to the changes imposed by the limb loss (Chen et al., 2002; Chen et al., 1998; Geurts et al., 1991; Karl et al., 2001; Kavounoudias et al., 2005).

The loss of large portions of tissue from a body segment leads to changes of different body systems (Chen et al., 2002; Chen et al., 1998). The missing information from the amputated segment is also associated with the loss or reduction of sensation (Kavounoudias et al., 2005; Kosasih & Silver-Thorn, 1998), altered proprioceptive information (Eakin et al., 1992; Kavounoudias et al., 2005), and changes in muscular strength (Isakov et al., 1996b; Moirenfeld et al., 2000; Nadollek et al., 2002; Pedrinelli et al., 2002; Renstrom et al., 1983a) as well as changes in balance performance (Dornan et al., 1978; Duclos et al., 2009; Duclos et al., 2007; Fernie & Holliday, 1978; Gauthier-Gagnon et al., 1986; Geurts et al., 1991; Hermodsson et al., 1994; Isakov et al., 1992). Despite the fact that numerous studies have evaluated the effect of amputation on multiple variables, there is no clear explanation of how these variables influence balance control.

The following literature review contains some epidemiological data related to amputation. We also present different aspects related to: TPS, proprioception and muscular strength. These variables are all commonly accepted measures studied in amputated individuals. We discuss the changes affecting not only the amputated side, but also the non-amputated side. We also considered other subjects like balance response under static and dynamic conditions, and the influence of vision on balance performance.

Chapter 1 – Review of Literature.

1.1 Epidemiology of Amputation.

Each year, the United States reports around 185,000 limb amputations (Owings & Kozak, 1998). The National Health Interview Survey in 1996 estimated that 1.2 to 1.6 million persons in the US lived with a limb amputation (Adams et al., 1999). According to Ziegler-Graham et al. (2008) the forthcoming prevalence of limb loss for the year 2050 in the US will be more than double the estimated prevalence for 2005 (about 3.6 million in 2050). Statistical data from 2005 indicated that 54% were amputations related to dysvascular disease among older adults and 45% of the amputation procedures were related to trauma. The remaining 1% percent corresponded to cancer-related amputations. Independent of the origin, amputation imposes a significant health, social and economic burden (Clarke et al., 2003; King et al., 1998; Moulik et al., 2003). In developed countries, non-traumatic etiology is considered the main cause of lower limb amputations (LLA) (Manchester et al., 1989; Ziegler-Graham et al., 2008) compared to developing countries, which report trauma as the primary cause of LLA (Collin & Collin, 1995) affecting both combatants (Fernie & Holliday, 1978; Islinger et al., 2000), and civilians (Meade & Mirocha, 2000). Moreover, trauma related amputation affects a significant number of younger individuals generating a great impact on life among young, previously healthy individuals (Laupland et al., 2005).

1.1.1 Non-Traumatic Amputations.

Non-traumatic lower-extremity amputation is a condition that increases with aging, affecting elderly people (Dillingham et al., 2002; Ziegler-Graham et al., 2008). It is considered the primary cause of morbidity and mortality among individuals with diabetes and dysvascular disease diagnosis (Dillingham et al., 2002; Ebskov et al., 1994; Ziegler-Graham et al., 2008). From all dysvascular amputation procedures, around 60% were major limb amputations. According to the authors, among amputees from all etiologies, 65% of the procedures were performed on the lower extremity (Ziegler-Graham et al., 2008).

Major non-traumatic limb amputations have a high correlation with diabetes. Ziegler-Graham et al. (2008) estimated that two-thirds from a total of 54% of dysvascular amputees were linked to a diabetes diagnosis in the US. In the future, an increase in major limb amputations is expected considering the expected increase of diabetes prevalence from 2.8% in 2000 to 4.4% in 2030 (Ebskov et al., 1994; Wild et al., 2004).

1.1.2 Trauma-Related Amputations.

Trauma is considered the second leading cause of amputations in developed countries (Dillingham et al., 2002; Owings & Kozak, 1998). According to some studies, which included amputees from different etiologies trauma-related amputation affect more the upper limbs (Dillingham et al., 2002; Ziegler-Graham et al., 2008). However, the study by Barmparas et al. (2010) who analyzed only traumatic amputees, reported that 59% of single extremity amputations affected the lower extremity, most of them below knee level. The other 41% affected the upper extremity. They also reported a higher frequency

of LLA in pedestrians and motorcyclists compared to motor vehicle occupants whom displayed more upper extremity amputations (Barmparas et al., 2010). Traumatic amputations related to age and sex report controversial results. According to Dillingham et al. (2002), amputation increased with age for all, traumatic and non-traumatic cases and was independent of sex and race. Laupland et al. (2005) also reported a higher incidence of amputation in the elderly, and Ziegler-Graham et al. (2008)indicated that men had five times higher risk of trauma-related amputations compared to women. Note that Ebskov et al. (1994) described two major peaks for male trauma-related amputation: one at the ages 20 to 29 and the second from 70 to 79 years. Female traumatic amputees showed a single peak only for the ages 70 to 79.

1.2 Sensation in Amputees.

Different studies reported changes in sensorimotor representation in traumatic and nontraumatic amputees (Braune & Schady, 1993; Chen et al., 1998; Geurts et al., 1992; Simoes et al., 2012). Changes in the sensorimotor representation may also lead to a cortical and neural structural reorganization of the non-amputated side (Simoes et al., 2012). Those changes could explain in part why the non-amputated side is affected in both vascular and traumatic amputees.

The loss of anatomical structures implies the deprivation or altered sensation information from musculoskeletal, articular and cutaneous tissues (Geurts et al., 1992). The amputation could generate significant changes on afferent information related to proprioception and cutaneous sensation (Kavounoudias et al., 2005).

1.2.1 Proprioception.

Proprioception is defined as "the sensory information from the muscles, tendons, or joints about limb position and movement" (Gaither, 2008), or the sensory awareness of body position essential for motor control (Lackie, 2010). The ability is derived from the neural afferent information related to joint motion, spatial localization and force generation sensations processed by the central nervous system (CNS) (Lephart & Fu, 2000). The most frequent methods used to assess the perception of joint movements and positions are the threshold detection of passive motion (TDPM) and the reproduction of joint position (RJP). The TDPM measurement is based on the perception of joint motion. The joint to be evaluated is passively displaced at a very low angular speed, and individuals indicate the perception of the joint displacement using a control device. The test measures the difference between the starting angle and the angle where motion is perceived. The RJP procedure also involves the passive movement of the joint from the starting point to a pre-set target angle. The joint is held at this target angle for a few seconds and then it is repositioned to the starting angle. After that, the participants are then asked to bring the joint to the target angle. The evaluation of this test involves measuring the difference between the pre-set target angle and the angle reached by the voluntary displacement.

1.2.1.1 Amputated Limb Vs Non-Amputated Side.

Eakin et al. (1992) and Liao & Skinner (1995) studied the TDPM in unilateral lower limb amputees. Liao & Skinner (1995) evaluated the TDPM in vascular and traumatic belowknee amputees (BKA) at a speed displacement of 0.4°/second while the participants were seated. Eakin et al. (1992) evaluated traumatic and cancer related above-knee amputees (AKA) (Nakagawa et al., 1993) at the speed of 0.5°/second while the participant was standing. Both Eakins's and Liao's studies reported higher TDPM values in the amputated limb compared to the non-amputated limb (sound limb of amputees). Kavounoudias et al. (2005) also evaluated the TDPM under non-weight bearing conditions in traumatic and non-traumatic (vascular) BKA amputees. The participants were seated while the joint was displaced at the speed of 0.7°/second. However, in their results Kavounoudias et al. (2005) reported no difference in TDPM between the amputated and the non-amputated limb. In their studies, Eakin et al. (1992) and Liao & Skinner (1995) also included the assessment of the RJP. Both studies displayed no difference between the amputated and the non-amputated side of unilateral lower limb amputees.

1.2.1.2 Amputees Vs Able-Bodied Controls.

Liao & Skinner (1995) and Kavounoudias et al. (2005) not only compared the amputated limb to the non-amputated limb side. They also included able-bodied controls to make the comparisons. They reported that the TDPM in the non-amputated side of amputees was higher when compared to able-bodied controls. Moreover, Kavounoudias et al. (2005) separately compared vascular and traumatic amputees to able-bodied controls. When the sound limb of amputees was compared to able-bodied controls, both traumatic and vascular amputees exhibited significant differences in TDPM at knee but not at the ankle joint level. Knee joint in amputees showed higher TDPM values compared to controls. Liao's study compared RJP between the amputated limb and able-bodied controls, reporting no difference among these two groups. Despite the absence of differences in RJP, significant differences are evidenced in TDPM in both limbs of amputees. These results support the existence and involvement of different mechanisms in the proprioceptive regulation of joint motor control (Kavounoudias et al., 2005). Based on the results, Kavounoudias, et al. (2005) suggested that amputees compensate the absence of proprioceptive information from missing anatomical structures. They could gather additional information from other sources or using other means such as the displacement of adjacent joints, changes on superficial and deep pressure sensors and modifications on soft tissues receptors. Liao & Skinner (1995) also suggested that muscle spindles could be more susceptible to perceive small changes of joint angles and muscle length than able to reproduce the angle of joint repositioning.

1.2.2 Cutaneous Sensation.

Cutaneous sensation is defined as the perception originating from receptors of the skin. Touch perception includes "several partially independent senses" such as thermal sensation (warmth – cold), cutaneous touch-pressure (superficial – deep), vibration, pain, itch, and "movement across the skin" (Kalat, 2013). Different tests used to evaluate cutaneous sensation are usually applied on pressure-tolerant and pressure-sensitive areas and may be influenced by the loss of tissues in amputees (Murdoch, 1969). In amputees, sensory impairment could be influenced by factors like age and time since amputation (Kosasih & Silver-Thorn, 1998).

1.2.2.1 Amputated Limb Vs Non-Amputated Side.

The procedures used to assess sensation in amputees varied between different studies (Kavounoudias et al., 2005; Kosasih & Silver-Thorn, 1998). In these studies, amputees evidenced an altered perception of the cutaneous sensory information. Kosasih & Silver-Thorn (1998) compared sensation between the amputated and the non-amputated side of traumatic and non-traumatic amputees. They evaluated symmetry and asymmetry perception of light touch, deep pressure, vibration, and pinprick sensation in amputees. To standardize the assessment perception of these parameters they included cotton swabs, a tuning fork and, the sharp and dull ends of a safety pin. Only deep pressure did not include an instrument, but they used the examiners' thumb to evaluate the sensation. "Normal" sensation was established as the ability to perceive or not, the applied stimulus. Results were reported as the comparison in sensation perceived on the amputated side compared to the non-amputated side (sensation amputated/sensation non-amputated). A "normal/normal" condition indicated a normal sensation on both sides. Three more combinations were also presented in the study (impaired/normal, normal/impaired and impaired/impaired). Out of 16 participants, 7 displayed altered sensation in the nonamputated side and were excluded from additional analysis. Among the participants who evidenced non-altered sensation in the non-amputated side (8 traumatic and 1 cancer related amputee) the most affected sensation on the amputated side was pin prick (67% overall participants). Altered pin prick sensation was also related to age and time since the amputation. Sensory impairment affected 60% of middle aged participants (45 - 59 years) and 100% of participants aged 60 years old or more. All the participants with 10 years or less since the amputation or those with 21 year or more also revealed a sensory

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impairment of 100% and 75% respectively. Moreover, these participants evidenced an altered perception of light touch and vibration perception (11% of the participants for each group). The results did not evidence alterations in deep pressure sensation. Researchers suggest that the deterioration of the sensory perception on the non-amputated limb seems to be related to different conditions such as bilateral trauma, diabetic polyneuropathy, and or prior vascular surgery (Kosasih & Silver-Thorn, 1998).

Kavounoudias et al. (2005) reported different results when comparing the amputated and the non-amputated extremities depending on the origin of the amputation. They indicated that traumatic, but not vascular amputees exhibited significant differences in TPS between tibial sites. Traumatic amputees displayed higher TPS levels on the amputated side compared to the non-amputated side.

1.2.2.2 Non-Amputated Side Vs Able-Bodied Controls.

The study by Kavounoudias, et al. (2005) also compared the TPS between the sound limb of amputated individuals and able-bodied controls, reporting that cutaneous sensation impairment also affected the non-amputated side. Due to significant differences in age and time since the amputation between vascular and traumatic amputees, they were analysed independently. When comparing only the non-amputated side of traumatic amputees to able-bodied controls, the TPS demonstrated higher values. This difference was significant only at the plantar site, not at the tibial site. The results for vascular amputees did not display differences in TPS when they were compared to able-bodied controls. However, vascular amputees evidenced significant differences for the testing site, with lower TPS thresholds at the tibial site compared to plantar site.

1.3 Muscular Changes in Amputees.

Muscle tissue has been considered as an effector "organ" responsible for generating the force necessary to develop joint movement and body displacement. Amputation, regardless of the origin causes the loss of different tissues mainly musculoskeletal tissue involved in the generation of muscular force.

1.3.1 Muscle Atrophy.

Different methods have been used to describe muscular changes in amputees in order to explain the reduction of muscular strength (Isakov et al., 1996a; Renstrom et al., 1983a; Schmalz et al., 2001). These methods ranged from low to high technology devices including: measuring tape (Isakov et al., 1996a; Renstrom et al., 1983a), biopsy analysis, computed tomography, magnetic resonance image measurements (Renstrom et al., 1983a) and ultrasound techniques (Schmalz et al., 2001).

Renstrom et al.(1983a) used BKA due to vascular and non-vascular origin to describe muscle atrophy associated with an amputation. The authors performed muscular biopsies of the vastus lateralis of the amputated and the sound limb sides and compared the muscles' fiber distribution. They also studied the effect of amputation on the crosssectional area (CSA) of the thigh using computed tomography and on the whole thigh perimeter using a measuring tape. The authors conducted the evaluation of the CSA and the thigh perimeter assessment at the same level where they performed the muscular biopsies. The biopsies result showed no differences in the fiber type distribution between the amputated and the sound limb. However, there was a trend to a reduction in fiber type I (from 38% to 33% approximately) and a trend of increments in fiber type II (from 62% to 67% approximately). Conversely, the analysis of muscle fiber II subtypes showed significant differences in the percentage of fiber distribution. The amputated side evidenced with a smaller fraction of type IIA fibers and a bigger fraction of type IIB and IIC fibers. When they evaluated the CSA, the whole thigh evidenced lower values in the amputated side, representing 86% of the CSA of the sound limb side. The compromise of quadriceps muscles was higher than hamstring muscles evidencing a higher reduction of CSA; quadriceps muscles exhibited a 66% and hamstring muscles 80% of the CSA of the non-amputated leg muscles respectively.

Different studies indicated that the measuring tape might not detect large differences in muscles' CSA (Lexell et al., 1983; Young et al., 1980). However, the results reported by Renstrom et al. (1983a) revealed that the difference between the level of atrophy measured by computed tomography and the level of atrophy using the measuring tape was less than 2%. Both measurements suggested a reduction in muscular CSA in the amputated side compared to the non-amputated side.

Another method used to evaluate muscular changes between the amputated, and the nonamputated side was ultrasonography showing significant reductions in muscular thickness and CSA in the amputated side (Schmalz et al., 2001). However, the CSA indicated a higher compromise with a mean reduction of 21% compared to the muscle thickness that evidenced a mean reduction of 13%. This compromise affected specific

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muscular groups, with significant changes in rectus femoris, vasti and sartorius but no difference in muscle thickness and CSA for the gracilis, semitendinosus and biceps femoris muscles.

1.3.2 Muscle Strength.

By contracting the muscle fibres, it is possible to develop the force necessary to produce the movement (acceleration - deceleration) of different body segments. Muscular strength is influenced by the level of muscle mass (Gopalakrishnan et al., 2010; Hurley, 1995; Kasper et al., 2002). In amputees, sarcopenia and the associated reduction of muscular fibre CSA (Renstrom et al., 1983b) could lead a reduction of force generation (Isakov et al., 1996b; Moirenfeld et al., 2000; Pedrinelli et al., 2002; Renstrom et al., 1983b). Changes in muscular strength may influence the rehabilitation outcomes and eventually the adaptation of prosthetic use in amputated individuals (Isakov et al., 1996b). The measurement of muscle strength can be performed through different methods including dynamometers. Dynamometers are considered the most reliable method to evaluate the force (Bandy & McLaughlin, 1993; de Carvalho Froufe Andrade et al., 2013; Holmback et al., 1999; Orri & Darden, 2008).

1.3.2.1 Amputated Limb vs. Non-Amputated Limb.

Various studies compare muscular strength levels between the amputated and the nonamputated extremity in amputees to evaluate the level of strength compromise. These studies evaluated amputees from traumatic and non-traumatic etiologies (Isakov et al., 1996b; Isakov et al., 1996a; Pedrinelli et al., 2002; Renstrom et al., 1983b). One single study evaluated only traumatic amputees (Moirenfeld et al., 2000). Independent of the origin and level of amputation, most of the studies reported significant reduction of thigh muscles strength in the amputated side including peak torque (Isakov et al., 1996b; Moirenfeld et al., 2000), isometric force (Isakov et al., 1996b), maximum bending moment, total work and total power (Pedrinelli et al., 2002).

The following studies evaluated BKA, performing the assessments at different isokinetic speeds: Renstrom et al. (1983b) evaluated strength at 30°, 60° and 120°/second, Pedrinelli et al. (2002) at 60°/sec and 180°/second, Isakov et al. (1996a) at 60°/sec and Moirenfeld et al. (2000) at 120°/second. All the studies, except Isakov et al. (1996b; 1996a) evaluated strength (peak torque) during consecutive concentric knee FLX and knee EXT, but also included the eccentric isokinetic strength measurement of the same muscles. Isakov et al. (1996b; 1996a) evaluated knee FLX and EXT muscles using eccentric isokinetic strength measurement. Isakov et al. (1996b; 1996a) and Renstrom et al. (1983b) also assessed isometric strength.

In general when the amputated side was compared to the non- amputated side, different studies revealed significant reductions of concentric and eccentric knee FLX and EXT strength. Other significant differences are related to the assessed muscular group. Knee EXT muscles evidenced higher reduction of isometric (Renstrom et al., 1983b) and isokinetic muscular strength compared to knee FLX muscles (Moirenfeld et al., 2000; Renstrom et al., 1983b). However, Pedrinelli et al. (2002) suggested the opposite; they indicated that peak bending moment reductions were more evident for the knee FLX than for knee EXT muscles when comparing the amputated side compared to the non-

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amputated side. Despite reporting significant differences between the amputated side and the non-amputated side, Isakov et al. (1996a) described no differences between knee FLX and knee EXT muscles.

Moirenfeld et al. (2000) also evaluated the isokinetic endurance using a fatigue index. The fatigue index was calculated as the difference in total work from the first ten and the last ten repetitions, divided by the total work during the first 10 repetitions. The amputated limb evidenced lower levels of the fatigue index than the sound limb. This deficit was statistically significant for EXT muscles but not for FLX muscles. In another study by Nadollek et al. (2002) they evaluated a different muscular group and also used a different method of assessment. Using a manual dynamometer, they evaluated the hip abductor muscles of each limb in traumatic and vascular BKA. The assessment included the peak force of maximum isometric abduction of the hip. However, Nadollek et al. (2002) did not report significant differences in strength measurements between the amputated and the non-amputated limb.

The length of the amputated residual extremity was an additional factor that yielded a significant role on strength of BKA. In a supplementary report by Isakov et al. (1996b), concentric, eccentric and isometric strength for the knee FLX and EXT muscles displayed lower values in those amputees with shorter remnant limb (less than 15 cm) than those with the butt end length higher than 15 cm. However, Pedrinelli et al. (2002) did not demonstrate a relationship between force reduction and those amputated individuals with shorter residual limb length.

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Renstrom et al. (1983b) evaluated the effect of muscle atrophy on strength, reporting significant correlations with the CSA for the amputated limb. They reported a significant correlation between CSA and muscular strength for knee FLX and EXT, when the participants were using the prosthesis and only for knee EXT during isometric contractions. No correlations were revealed between CSA and muscular strength for the non-amputated side. The authors indicated that the large reduction in muscular strength compared to the slow progress in muscular atrophy suggested the existence of concurrent factors, other than muscular atrophy (e.g.: reflex inhibition). These factors could explain the reduction of muscular strength on the non-amputated side.

1.3.2.2 Non-Amputated Limb vs. Able-bodied Controls.

When they evaluated the strength levels of amputees Pedrinelli et al. (2002) not only compared the amputated limb to the sound limb, but they also compared the sound limb of amputated individuals to limbs of able-bodied controls. Amputees showed significant reductions for all the measures at the different speeds assessed in maximum bending moment, total work and maximum power. With these results, the authors concluded that the use of the non-amputated limb as reference to evaluate the muscular strength of the amputated side in amputees is an inadequate comparison. To our knowledge, no other study compared the level of muscular strength of the amputated limb in TTA to the level of muscular strength in able-bodied controls.

1.4 Balance Control.

Balance is an expression used to describe all the postural changes in order to maintain the projection of the body's center of mass (COM) within the limit area of the base of support (ABS) (Mooren, 2012). Balance control is a multi-faceted motor skill influenced by the coordinated activation of extremities and trunk muscles (Horak et al., 1997). The balance-control system also maintains a particular body orientation and stability under static and dynamic conditions (Deliagina et al., 2012).

The CNS plays a crucial role in balance control by integrating the sensory input information coming from different structures and tissues (Figure 1). Experimental animal models reported that in addition to the motor cortex, basic mechanisms for postural balance control are located at lower levels of CNS, within the brainstem and cerebellum (Deliagina et al., 2007; Deliagina et al., 2012). After processing this information, the CNS generates a coordinated series of motor responses, adjusting the orientation and position of different body segments (Massion, 1998).

The CNS system is capable of controlling posture using different strategies including: anticipatory responses, compensatory responses, or combination of both (Maki & McIlroy, 1997). The anticipatory strategy, also called "predictive" strategy, seems to imply the voluntary activation of various muscles as an expected response to potential changes in posture. The second is the compensatory or "reactive" strategy. This may include the muscular response and the associated postural adjustments that follow an unpredicted perturbation of balance (Maki & McIlroy, 1997). Thus, postural control should not be considered only as an automatic response, but also as a motor skill that could be learnt or trained (Horak et al., 1997).

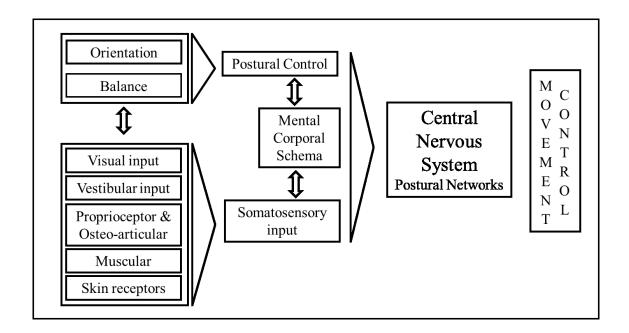


Figure 1. Organization of the postural control system. The diagram summarizes the principal structures involved in postural control. The CNS integrates and processes the afferent information to generate an efferent response (muscular activation). Modified from Massion, 1994.

1.4.1 Afferent Information Input.

Most studies evaluating balance control assess the role of vision, vestibular, and proprioceptive information. Different sensors register the gravity force and other forces, which typically occur during motion. The signals are transmitted and, integrated by the CNS and then compensated by the coordinated activation of different muscular groups. These internally processed signals then lead to anticipatory postural responses to maintain balance. These signals in their large majority are produced by the action of the voluntary muscles activity (Bloem et al., 2000; Massion, 1994; Mergner & Rosemeier, 1998). These anticipatory adjustments play a significant role in the feed-forward and feed-back response that helps to maintain balance (Dietz et al., 1993). The somatosensory input from muscles, joints and cutaneous receptors represents one of the primary sources of information for balance control. Proprioceptive sensors which provide information related to muscle length (muscle spindle fibres), muscular tension (Golgi tendon organ) and the articular angle position (Ruffini's ending receptors), allow the spatial location of different body segments (Gandevia & Burke, 1992; Mohapatra et al., 2012). Some studies considered that proprioceptive information from muscles which control the ankle joint play a significant role in body position changes (Barbieri et al., 2008; Nakagawa et al., 1993).

The influence of visual information on balance has been evaluated mainly from two different perspectives. The first one is related to the effect of the visual fields and the visual object-motion perception on postural sway (Dijkstra et al., 1994; Previc et al., 1993). According to Dijkstra et al. (1994) the visual information from peripheral and central visual fields equally affects postural sway. In addition they suggest the existence of a dynamic coupling between the moving visual environment and postural sway and the slight retinal displacement that occurs under moving visual environment does not explain the changes in postural sway. The second perspective used is the evaluation of COM modifications associated to changes of the ground-reaction forces when switching from EO to EC condition. By measuring the center of gravity (COG) displacement or the COP displacement, calculated from ground reaction forces (Deliagina et al., 2007), various studies reported that COP displacement is more evident under EC condition (Baltich et al., 2014; Chen et al., 2014; Kanekar et al., 2014; Tsai et al., 2008).

When the visual information is reduced or absent, the vestibular system plays a significant role in the perception of changes of body inertia specially the head angular

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acceleration (Mergner & Rosemeier, 1998). The semicircular canal systems provide more information related to angular acceleration while the otolith systems provide more information related to linear acceleration. For balance control, the information from these two subsystems has to be combined. Some authors considered that the information from these signals is not ideal due to the delay generated by the synthesis process (Mergner & Rosemeier, 1998). Some others considered that vestibular information might not represent a major source of information during early postural change response to balance perturbations. The vestibular system has been shown to induce the activation of the hip strategy (Winter, 1995). This strategy involves the muscular activation sequence from the proximal-to-distal body segments and also the activation of the trunk muscles (Allum et al., 1993; Horak et al., 1994; Mergner & Rosemeier, 1998).

1.4.2 Center of Pressure.

1.4.2.1 Definitions.

It is important to define first the COM and COG, which are completely different from the COP (Palmieri et al., 2002).

The COM could be defined as the point where the total mass of the body is concentrated, representing the sum of the "weighted average" of all the different body segments COM locations (Newman, 2008; Winter, 2009). This center is considered the point of action of all external forces and it is used to analyze and understand body "translational motion" (Newman, 2008). The COG is defined as the vertical projection of the COM from the floor (Palmieri et al., 2002; Winter, 2009). The center can be modified by the position

and/or the displacement of different body segments. In addition, it is also the point of reference for the postural control system to adjust and maintain balance (Winter, 2009).

The COP is a point that represents the mean weighted average from the area of support, where all the ground reaction forces act. Winter (1995) considered the COP as the vector position of the vertical ground reaction forces. The position of this vector represents also the projection of the muscular forces required to maintain balance (Winter, 2009; Winter et al., 1990). The COP reveals the course followed by the COM. The path followed by the COM can be correlated to the path followed by the COP yet, as explained before they are entirely different (Palmieri et al., 2002).

The displacements and adjustments of the COP during quiet standing are due mainly to the activity of the ankle, hip and trunk muscles leading to body sway. During quiet stance, the body sways sideways between the lower extremities and, forward and backward swivelling around the ankle joints. Changes in the COP through body sway help to control the body COM. When the somatosensory system perceives the anterior displacement of the body COM, the COP is displaced forward toward the edge of the area of base support, in front of the COM. The coordinated activation of different muscular groups, which modify the COP, reduces or stops the displacement of the COM and reverses its forward translation. The muscular activation also generates the displacement of the COM backwards and now the COP follows the COM in the opposite direction and the entire process is repeated now for the posterior direction (Winter, 1995, 2009; Winter et al., 1990).

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The main objective of the process described before is to maintain the COM within the base of support. However, greater displacements of the COM are associated with greater displacements of the COP. This activity also implies a higher trend to reach a point where the COM is displaced outside the ABS. Outside this area, it is necessary to adjust the posture in order to maintain the body balance (Horstmann & Dietz, 1990; Winter, 1995, 2009). During single-legged stance, the COP is located within the area of contact of the foot. During two-legged stance the COP moves within a zone located between the two feet and is influenced by the relative distribution of body weight (Winter, 2009; Winter et al., 1990).

1.4.2.2 Center of Pressure Measurement.

The COP can be measured using a force plate or a pressure mat system. Data from the force plate measures the force in three dimensions but only the vertical components are necessary to calculate the COP (Winter, 2009). The pressure sensing systems also measure the vertical component of the ground reaction force derived from the area in contact with the feet (Orlin & McPoil, 2000).

COP data is recorded as a successive coordinate points (x and y) system related to time. Table 1 summarizes the most common measurements used to analyze the COP performance in able-bodied individuals and also in amputees (Abrahamova & Hlavacka, 2008; Buckley et al., 2002; Isakov et al., 1992; Palmieri et al., 2002).

Conditions like neurological diseases, musculoskeletal disturbances or any modification of the systems which provide or integrate the information could generate an inappropriate

response of body effectors. The loss of tissues in lower-limb amputees and the related information also affects body posture and may predispose to balance impairment.

1.4.3 Balance in Amputees.

To minimize the displacement of the COM above the base of support, it is necessary the continuous control by the COP. The upright position is a less stable condition which requires a greater control of the COM and also demands a higher activity from the centers of control (Korr, 1975). The study of balance in amputees has included many different conditions and settings trying to explain how balance is affected in these individuals.

COP VARIABLE	DESCRIPTION
Maximum amplitude	Maximum absolute displacement of the COP from its average point.
Minimum amplitude	Minimum absolute displacement of the COP from its average point.
Peak-to-peak amplitude	Difference between the maximum and minimum amplitudes of COP.
Mean amplitude of COP	The average value over all data points collected in a trial.
Total excursion - Displacement	Sum of distance between COP successive points.
COP velocity	Total displacement traveled by the COP over time.
Root-mean-square amplitude	Standard deviation of the COP position.
Root-mean-square velocity	Standard deviation of the COP instantaneous velocity.
Spectral analysis	Detect what frequencies existed in the data, related to a particular sensory system.
Time-Frequency analysis	Study the frequency characteristics over time, associated to a specific sensory system

 Table 1. Definitions of the most common measurements used to analyse the COP (Palmieri et al., 2002).

1.4.3.1 Two Limb Stance: Amputees vs. Able-bodied Controls.

A considerable number of research studies on balance in amputees performed assessments during double-legged stance and almost all of them reported increments in postural sway in amputated individuals when compared to able-bodied controls. Different parameters were used to measure balance performance including the COP excursion range and the sum of the squares deviations from the mean COP in the anterior-posterior (AP) (Shapiro, 2013) and ML direction (Buckley et al., 2002), the mean speed of sway in AP and ML direction (Dornan et al., 1978; Fernie & Holliday, 1978), and the root mean square of the COP velocity (Geurts et al., 1991). In this section, we will use the term "postural sway" as a general term that includes all the variables used in different studies to evaluate balance.

Static Balance.

In BKA, significant differences have been evidenced for postural sway, with higher values in amputees as compared to able-bodied controls. Although the test was performed during bipedal position, these differences were even larger under EC conditions (Dornan et al., 1978; Fernie & Holliday, 1978; Isakov et al., 1992). When compared to able-bodied controls, all BKA (traumatic and vascular) displayed significant differences only for ML directions under EO condition. Significant differences were reported for both AP and ML direction only under EC condition (Hermodsson et al., 1994). Amputated individuals and vascular amputees in particular revealed significant increments in the standard deviation of the COP position (Hermodsson et al., 1994). When compared separately to able-bodied controls under EO condition, vascular amputees displayed

significant differences in ML direction and traumatic amputees only evidenced significant differences in AP direction.

The standing time in balance was also impaired in BKA amputees. Hermodsson et al. (1994) evaluated balance in amputees from different etiologies during double-legged stance compared to able-bodied controls. All the amputated participants and vascular amputees in particular displayed a significant shorter standing time compared to able-bodied controls (Hermodsson et al., 1994). Significant reduction in standing time was also evident in vascular amputees when compared to traumatic amputees. No significant difference was revealed for standing time between traumatic amputees and able-bodied controls (Hermodsson et al., 1994).

However, Gauthier-Gagnon et al. (1986) reported different results compared to other studies. They demonstrated reductions of postural sway in AKA and BKA individuals from traumatic and non-traumatic origin. They measured and compared the mean surface of sway of amputees under two different rehabilitation programs and able-bodied controls. Either under EO or EC, able-bodied controls exhibited a mean surface of sway of 14 ± 13 cm² representing almost 2% of the ABS. The initial assessment of both, the experimental and the conventional rehabilitation groups displayed a reduced sway surface area of 1.9 ± 1.3 cm² and 3.3 ± 1.0 cm² respectively. Despite an increase of sway surface area after the rehabilitation program, these values were still significantly reduced in both groups when compared to able-bodied controls. The change represented less than 0.3% of the total ABS leading to a less stable condition.

In AKA individuals, it was expected that the proximal location of the amputation imposed a greater demand not only to the remaining structures (stump and sound limb) but also to the different systems responsible for controlling the posture and balance. Considering the greater loss of tissue, researchers expected more changes in balance performance in AKA individuals. However, results seem controversial. In some cases, AKA demonstrated no differences in balance performance when compared to able-bodied controls (Dornan et al., 1978; Fernie & Holliday, 1978). In other cases, AKA revealed significant higher levels of postural sway in AP and ML direction when compared to able-bodied individuals (Buckley et al., 2002; Geurts et al., 1991). It is drawn the attention to an interesting result observed when comparing AKA and BKA individuals. Despite the smaller extremity level of amputation, BKA described higher levels of postural sway when compared to AKA, or when compared to able-bodied controls (Dornan et al., 1978; Fergason et al., 2010).

Dynamic Balance.

One study not only evaluated balance under static conditions, they included also dynamic balance assessment in amputees. Buckley et al. (2002) separately evaluated dynamic postural sway in traumatic AKA and BKA individuals using a modified single axis stabilimeter. Results evidenced reduced values for time in balance in amputees when compared to able-bodied controls. The number of board contacts was reported to be similar in both groups. The comparisons between amputees and able-bodied individuals did not indicate differences in the results for any of the tested conditions (platform tilt in AP and ML directions). However, more board contacts were displayed on the amputated side compared to the non-amputated side in amputees. The authors also stated that able-

bodied individuals did not report significant differences in the number of board contacts between both sides. They suggested that increments in board contacts on the amputated side might be used as an additional source of somatosensory information. This information could improve amputees' response to balance perturbations. Despite the trend to increments in mean time spent during board contact (more evident in AP task), the results indicated no differences when comparing amputees and able-bodied individuals.

Influence of Vision on Balance.

The influence of visual information is the most common factor involved in balance performance assessment. Independent of the amputation level or the etiology, visual information seems to exert an important role in balance performance in amputees (Dornan et al., 1978; Fernie & Holliday, 1978; Geurts et al., 1991; Isakov et al., 1992). In general, under EC condition, amputated individuals displayed a less stable condition when compared to able-bodied controls (Buckley et al., 2002; Dornan et al., 1978; Fernie & Holliday, 1978; Geurts et al., 1991; Isakov et al., 1992). Despite reporting no changes in the weight-bearing distribution, Isakov et al. (1992) reported that postural sway was increased while the participants were blindfolded.

The ratio EO to EC of the mean speed of sway of AKA and BKA evaluated by Fernie & Holliday (1978) described significant differences when amputees were compared to ablebodied controls. Both groups of amputees evidenced lower ratio values. This difference was even greater for AKA, which exhibited the lowest ratio values. The lower ratio EO to

EC in amputees may indicate the importance that visual information plays on the control of balance in those individuals (Fernie & Holliday, 1978).

1.4.3.2 Two Limb Stance: Amputated vs. Non-Amputated Side.

Nadollek et al. (2002) compared the amputated limb to the non-amputated limb during two-legged stance in non-traumatic BKA. The measurements included the standard deviation of ML and AP COP excursion and the percentage of weight-bearing distribution during quiet stance or standing still while distributing evenly the body weight (even stance). Due to the absence of differences between quiet and even stance, the authors reported the comparisons using only the quiet stance condition. The authors stated that the non-amputated side supported significantly more weight compared to the amputated side during quiet stance. During double-legged stance under EO condition amputees displayed higher displacements of COP in AP direction under the nonamputated limb compared to amputated limb. Moreover, under EC condition the nonamputated limb indicated a trend to larger displacements in AP direction. The analysis of ML direction did not display differences between extremities or significant differences related to eyes condition. However, the authors reported a trend of higher values in ML direction when amputated limb side was compared to the non-amputated side.

Duclos et al. (2009; 2007) described the postural asymmetry in LLA, reporting a shift of the COP position to the non-amputated side. These studies described the effect of muscle vibration (trapezius - gluteus medius) (Duclos et al., 2007) and the effect of isometric neck muscle contraction (Duclos et al., 2009) on standing posture and balance. The

authors described an involuntary leaning of the body weight induced by these stimuli. However, the leaning direction was not dependant on the side where the stimulus was applied. Independent of the stimulus, amputees reported higher values of RMSv compared to able-bodied controls, both before and after the stimulus. However, RMSv was not affected by the stimulus.

1.4.3.3 Single Limb Stance: Amputees vs. Able-bodied Controls.

Hermodsson et al. (1994) evaluated balance during 1-legged stance. In general amputees failed to maintain an upright position on one leg for 30 seconds on the non-amputated side (5 out of 18 vascular, 11 out of 18 traumatic amputees). Many of the able-bodied individuals were also unable to maintain balance on one leg for 30 seconds (19 out of 27 controls). Among all the participants who were able to stand for 30 seconds no significant differences in COP sway was observed for AP and ML direction when vascular and traumatic amputees were compared to controls.

When the single-legged stance was performed on the amputated side, almost all of amputated participants failed to maintain an upright position (16 out of 18 vascular, 12 out of 18 traumatic amputees). Therefore time in balance was the parameter used to evaluate balance performance in those participants who were not able to stand for 30 seconds. Comparison included the non-amputated limb to the right side of able-bodied controls and the amputated limb to the left side of able-bodied controls. The analysis of standing time during single-leg support revealed significant differences between groups. When comparing all amputees together to able-bodied controls, amputees evidenced

significant shorter standing times in both the sound limb and the amputated side. The comparisons within amputated individuals also showed significant differences, with reduced standing time in vascular amputees compared to traumatic amputees. When comparing amputated individuals separately to able bodied controls, significant differences were described only for vascular amputees with lower standing time values. Due to the level of difficulty, the single-legged stance assessment under EC condition was excluded from the study.

Chapter 2 – Research Design and Methods

2.1 Rationale, Objectives, and Hypotheses.

2.1.1 Rationale.

Balance performance is one of the parameters evaluated and trained in amputated individuals. The reductions of balance performance have been reported in various studies (Hermodsson et al., 1994; Isakov et al., 1992; Nadollek et al., 2002). However there are many factors associated that can affect balance performance. Sensation information from mechanoreceptors, exteroceptors and, muscular strength modify the ability to maintain balance. Some parameters used to evaluate proprioception like TDPM, have displayed opposite results from different studies (Kavounoudias et al., 2005; Liao & Skinner, 1995). In addition the amputation cause have evidenced that sensation perception could be modified differently due to amputee's etiology. Most of the studies evaluated amputees during double-legged stance and, few studies evaluated the sound limb of amputees which also indicated changes in muscular strength (Pedrinelli et al., 2002), changes in proprioception (Kavounoudias et al., 2005; Liao & Skinner, 1995) and altered balance performance (Hermodsson et al., 1994). Also, to our knowledge, the role of cutaneous sensation, proprioception, and strength on balance has not been directly studied in amputees. Based on these studies reports we wanted to evaluate the influence of cutaneous sensation, proprioception, and muscular strength on balance performance. Traumatic BKA were used as a model to evaluate and explain the different mechanisms involved in balance control.

2.1.2 Objectives and Hypotheses.

The purpose of this research project was to investigate the role of cutaneous sensation, proprioception, and strength on balance control in unilateral TTA. In this study, we evaluated balance during quiet stance and balance after squatting. The specific objectives were:

• Evaluate the effect of traumatic trans-tibial amputation on: cutaneous sensation, proprioception, and strength and COP displacement during quiet stance and after balance perturbation (squats).

Hypothesis number 1: The cutaneous sensation, proprioception and strength in the sound limb of traumatic TTA are reduced as a consequence of the amputation.

Hypothesis number 2: The COP variables are increased during balance assessment on the sound limb in traumatic TTA during one-legged quiet standing and after squatting.

• Study the role of cutaneous sensation, proprioception, and strength on balance. Hypothesis number 3: Reduction in balance in amputees is due at least in part to reduced cutaneous information, proprioception and strength.

Chapter 3 – Research Design and Methods.

3.1 Participants.

Two groups of participants were recruited to contribute in the current study: Seven traumatic BKA and seven able-bodied individuals. Each group included 6 males and 1 female. Participants' age was lower than 60 years old in order to reduce the effects of aging process on balance performance. Indeed, according to the results by Buckley et al. (2002), individuals aged 60 and over show a reduction in balance. Able-bodied healthy individuals and traumatic BKA were matched for age, sex and level of physical activity. Traumatic amputees exhibited an age M = 40.1, SD = 10.6 years, height M = 168.5, SD = 9.5 cm and body mass M = 88.4, SD = 22.2 kg. The time since the amputation for traumatic amputees was M = 42.0, SD = 22.3 months. Amputees with less than 1 year since the amputation were excluded from the study. The control group showed an age M = 39.8, SD = 8.1 years, height M = 173.1, SD = 4.7 cm, body mass M = 76.3, SD = 10.8 kg.

Exclusion criteria for both groups were: major visual deficiency, impaired balance or middle-inner ear pathology or any medical condition that could affect mobility (nervous deficiency, major cardiac or respiratory disease, motor or cognitive disability), diabetes mellitus, peripheral vascular disease or any sensorimotor deficit. All the participants read and signed an informed consent form approved by the ethics committee prior to their participation in the study.

3.2 Balance Assessment.

Balance assessment was conducted during static (quiet standing) and dynamic (squatting) conditions. Three test conditions were randomly performed, while standing on one leg: 1-Standing still with EO, 2-Standing still with EC and 3-Squat with EO. Amputated individuals stood on the sound limb and controls stood on a randomly selected limb while flexing the contralateral side. The amputated individuals wore their prosthesis during the whole procedure. The squat maneuver was selected as part of the conditions to evaluate balance. The knee FLX demanded the motion and the activation of ankle muscles involved in balance control. On the other hand, the internal disturbance of balance does not require the use of equipment to produce a perturbation of equilibrium.

The duration for each test trial was set at 45 seconds, and a total of 3 good trials for EO and up to 5 trials under EC conditions were recorded. We considered a good trial when the individuals kept the tested limb within the area of assessment of the pressure mat, without any surface contact with the non-tested limb. In addition to the previous conditions, during dynamic balance assessment participants were asked to perform a squat between 30°- 40° of knee FLX then, come up and stay still to consider the test valid.

As a guide for the squat two dots were placed on the wall at the eye level, 1.5 m in front of the participant, one when the knees were completely extended and the other dot when the knee reached 35° of FLX. An electro-goniometer (Noraxon U.S.A. Inc., Scottsdale, AZ - US) was set on the tested limb to control the amplitude of the squat and to ensure that participants reached the required knee FLX. The electrogoniometer had two parts; one was located on the distal portion of the thigh aligned with the longitudinal axis. The

second part was placed at the proximal portion of the shank along the longitudinal axis (Figure 2). There was no measure for the duration of the squats but it was approximately one second. Participants performed a few trials before the test to ensure they were able to bend the knee and reach the angle range within the time requested to perform a squat.



Figure 2. Participant on MatScan[®] system with the electro-goniometer.

3.3 Questionnaires.

The level of physical activity and the level of muscular fatigue were evaluated using different questionnaires. The assessment of these variables was not part of the main

objectives of this research. However, we used this information to ensure that both groups were matched in terms of level of physical activity and also to avoid the potential influence of muscular fatigue on balance assessment.

3.3.1 Physical Activity Assessment.

The level of physical activity was evaluated by using two different questionnaires. The Human Activity Profile (HAP) (Shapiro, 2013) is a self-administered questionnaire that measures the level of physical activity in healthy and impaired individuals (Polese et al., 2013; Teixeira-Salmela et al., 1999). The other questionnaire, the Tegner scale is frequently used to evaluate the level of work, physical activity or sports-related activities of individuals with knee dysfunction (Gordon et al., 2010; Sonnery-Cottet et al., 2014; Steadman et al., 2014).

The HAP evaluates the level of physical activity by presenting to the participant a series of activities with an increasing level of difficulty (Appendix A). The participant checks a selection-box up to the level of physical activity according to different parameters. These parameters include the options "Still Doing" if the participant could complete the task without help. "Have stopped doing this activity" is another option for the person was able to complete the task before but not at the current time and "Never did this activity" if never performed the action. The activities include daily life activities from "Getting in and out of chairs or bed without assistance" to high-performance activities like "Running or jogging 3 miles in 30 minutes or less". Two values are calculated from the test. The Maximum Activity Score (MAS) which is the highest level of activity still performed and the Adjusted Activity Score (AAS). The AAS results from the subtraction of MAS minus

the number of "stopped doing activities" registered below the MAS level. Events marked as 'never done' are not counted (Harbo et al., 2012).

The Tegner score ranges from 0 to 10 (Appendix B). The lowest value indicates "Sick leave or disability" and the highest value indicates the participation in sports ("Competitive sports"). The scale considers not only sports activities but also the individual's occupation.

3.3.2 Muscular Fatigue.

During the balance assessment, we used the Borg Scale of Global Muscle Fatigue to evaluate the participants' perception of fatigue at the lower-limb level (Appendix C). The fatigue scale ranges from 0 to 10 where 0 means "No fatigue" and 10 means the maximum level of perceived fatigue ("Very, very strong fatigue") (Hampton et al., 2014). The Borg Scale was used during balance assessment at six different moments: one was previous to starting each condition and one at the end of each condition (Figure 3). Between each assessment conditions, participants rested for 3 minutes sitting on a chair.

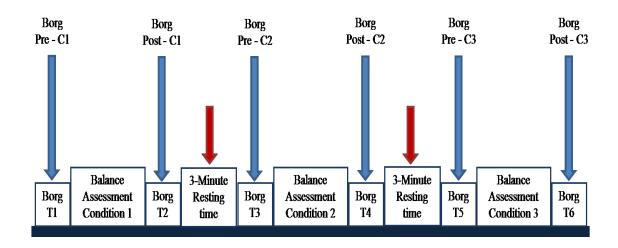


Figure 3. Experimental procedures for balance assessment. Participants performed three random conditions for balance assessment. The Borg scale of global muscle fatigue was measured six times (T1 to T6), previous to the starting and after the end of each condition.

3.4 Equipment and Measurements.

During balance assessment we measured COP position. In addition to balance assessment, we also evaluated three other measures which included: TPS, proprioception, and muscular strength. The first variables evaluated were related to sensitivity (cutaneous sensation followed by proprioception), then balance performance, and finally muscle strength. The purpose of this sequence was to avoid the potential effects of the strength measurement (soreness, fatigue or pain) on the other variables. All assessments were performed on the sound limb in amputees and, on a randomly selected limb in ablebodied controls. Studies have reported differences between the dominant and nondominant extremities (Kiss, 2012; Lee et al., 2012; Mezaour et al., 2009). However, it may be difficult to determine the dominant side in amputees. It is probable that amputees may have changed the dominant side after the amputation procedure. It is possible that the amputated extremity was the dominant limb and, the amputees started to use the previously non-dominant limb (sound limb) as the new dominant limb. Due to these reasons we decided not to match the sides in terms of dominance between the study group and the participants in the control group. Instead, we randomized the limb evaluated in the able-bodied participants.

3.4.1 Balance: Center of Pressure.

During both the static and dynamic balance tests, the COP position was recorded using pressure distribution from a MatScan[®] System (TekScan, Inc. South Boston, MA, USA) at a sampling frequency of 100 Hz (Figure 2). This is a portable equipment which is considered a reliable instrument and can provide objective measurements of the foot plantar forces and pressures (Zammit et al., 2010).

An electrogoniometer was used to confirm the participants performed the squat within 35±5° of knee FLX. The electrogoniometer activity was recorded with a TeleMyo 2400T G2 system (Noraxon U.S.A. Inc. Scottsdale, AZ, USA) at a sample rate of 1500 Hz and synchronized with the MatScan[®] using the trigger box (Trigger Box -TS-100, MatScan[®]).

3.4.2 Cutaneous Sensation: Touch Pressure.

To measure TPS we used Semmes-Weinstein monofilaments (North Coast Medical Inc. Gilroy, CA. USA). The filaments varied in numbers from 1.65 to 6.65 equating to log 10 of the force in milligrams required to bend the filament. The TPS was registered on two different sites. The first place evaluated was the tibial site (Figure 4 left panel), 10 cm below the anterior tibial spine on the medial aspect of the limb. The second site was located under the foot (Figure 4 right panel) on the head of the third metatarsal bone of the tested limb. The assessment was set in this order due to the skin thickness. The lower skin thickness at the tibial site facilitated the assessment and made it easier for the participant to learn the task. The tibial site is considered a pressure-tolerant area supporting part of amputees' body while wearing their prosthesis (Lee et al., 2005), and the plantar site was recommended by the equipment manufacturer as a suggested site to evaluate foot plantar sensation. Also, these sites were previously studied in amputees (Kavounoudias et al., 2005).



Figure 4. Touch pressure sensation. The tibial site (left panel) was located on the medial aspect of the limb 10 cm below the anterior tibial spine. The plantar sole site (right panel) was located on the head of 3rd metatarsal bone.

We used the staircase and the limit methods to evaluate the TPS (Cornsweet, 1962; Jones, 1989). Gross sensation was determined using first the thinnest filament and, continued with an incremental stimulus until delimiting the threshold stimulus. We stopped when the individual indicated the perception of pressure sensation from the progressive stimulus. Using the limits method allowed us to determine a more accurate sensation measurement. The step size was estimated using three filaments above and three

filaments below the threshold filament established by the staircase method (7 filaments in total). Three ascending/descending series of stimulus followed using these seven filaments. The test began with the smallest filament (from the group of seven) and progressively moved up until the participant perceived the touching sensation. We registered the number of the perceived filament and started the descending series. The descending series was initiated with the first filament perceived in the previous ascending series and progressively decreased to the filament exerting a lower pressure. This procedure continued until the participant did not perceive the touch pressure. The registered number corresponded to the first filament not perceived in the descending series using the same set of seven filaments. The recorded measurements from the ascending and descending steps were used to estimate the level of cutaneous sensation.

The filaments do not require a constant calibration process, which facilitates their use. In addition, a systematic review by Jerosch-Herold (2005) and a more recent study by Tracey et al. (2012) suggested that Semmes-Weinstein monofilaments have a high validity and reliability as a mean to evaluate touch threshold sensation. However, Collins et al. (2010) suggest that this method is more reliable when measured by a single researcher.

3.4.3 Proprioception: Joint Threshold Assisted Detection Motion.

To evaluate proprioception we measured the joint threshold assisted detection motion (TADM) using a custom-made proprioceptive apparatus. This measurement is similar to the TDPM but it was performed while the participant was bearing the body weight without hand contact. Under these conditions, the procedure required the body balance

control and, the activation of voluntary muscles during the standing position prevented the displacement of the joints to be fully passive. The device consisted of two platforms, one was fixed and the other one was mobile. The tested limb (non-amputated side in amputees and randomly selected side in able-bodied controls) was located on the moving platform during the assessment. Total body-weight was equally distributed between both lower extremities with one foot on each platform (Figure 5). Arms were crossed in front of the chest to avoid the potential bias in joint proprioception from the information that upper-limb support could provide.



Figure 5. Proprioception machine. The left panel shows the right foot on the fixed platform attached to a load-cell; the left foot is on the mobile platform. The right panel shows a position where the mobile platform is up resulting in FLX of the joints.

A load cell was placed under the fixed platform to ensure the equal distribution of the body weight. We measured first, the force exerted by the total body weight when the participant stood with feet together on the fixed platform supported by the load cell. The registered value from the load cell was used as a reference to evenly distribute the body weight $(50 \pm 10\%)$ between the two platforms when the participant stood placing a foot on each platform. The platform was randomly displaced up or down from the starting position of 20° of knee FLX. The platform assisted the joints displacement, moving the tested knee at a very slow speed of 0.7° /s, as used in a previous study (Kavounoudias et al., 2005). Participants were instructed to stop the limb-motion transfer system only when they perceived the assisted knee joint displacement using an On-Off motor-control device. In addition, to avoid the potential use of auditory information from the activation and displacement of the platform, the participants wore a headphone set to hear a white noise sound while they performed the test. We also prevented the individuals to gather visual information by covering the top part of the equipment and setting the participants' position looking straight forward to a distant point on the wall.

Participants performed twelve random trials (6 knee FLX and 6 knee EXT) with at least 10 good trials. A test trial was adequate when the participant maintained the body weight evenly distributed (50% of the total body weight $\pm 10\%$). Proprioception was estimated using the joint angular displacement. The ascending or the descending movements of the platform during the test involved the displacement of all joints either in FLX or EXT respectively. Although all the joints were rotated with platform motion, only the knee angle was computed from the measurement of the vertical linear displacement.

Most studies reported that threshold detection of motion is a reliable technique to evaluate knee proprioception under non-weight bearing conditions (Ageberg et al., 2007; Arockiaraj et al., 2013; Boerboom et al., 2008; Courtney et al., 2013; Nagai et al., 2012; Reider et al., 2003); only few studies reported that threshold detection of motion was a reliable method to measure proprioception under weight-bearing conditions at the knee (Nagai et al., 2013) and ankle joints (Deshpande et al., 2003).

3.4.4 Muscular Strength: Peak Torque to Body Weight.

We used a KinCom[®] - KC125AP dynamometer (Chattex Corporation, Chattanooga, TN, USA) to evaluate muscular strength. The use of the dynamometer allowed us to set and control a particular combination of contraction and angular speed for muscle strength measurement. We registered the peak torque and computed the peak torque to body weight (PT-BW) of concentric FLX /concentric EXT contractions of the knee and ankle (Figure 6). The knee joint was evaluated before the ankle joint, due to a greater level of difficulty observed during the pilot test, to perform the test when the ankle joint was evaluated first. A gravity compensation procedure was performed before each joint assessment. Setting positions for joint assessment were performed according to the manufacturer's manual. To set the tested range of motion, we first evaluated the full range of motion of the joints and moved back 5° in each direction. Participants were strapped to the seat during the test in order to reduce the body motion and to obtain the best performance. They were verbally encouraged to exert their best effort while performing the test.

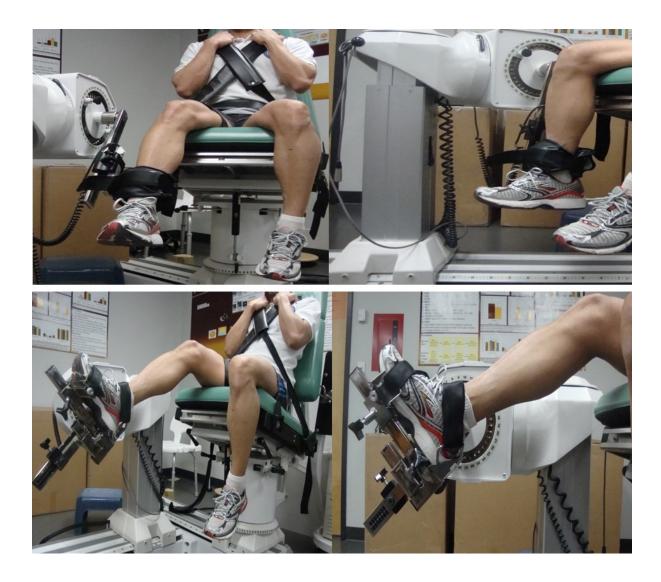


Figure 6. Muscular strength measurement settings. Knee (top panels) and ankle (bottom panels).

For the knee joint, individuals were seated with the back reclined at 75° and the knee in 90° of FLX. To assess the ankle joint the individuals were also seated with the back reclined at 60°, and the knee was flexed until the ankle adopted a neutral position. For both joints assessment, the bottom seat position was elevated at 15° from a horizontal line

parallel to the ground level. For each joint, participants had a warm-up set of ten repetitions at 100°/second. For the recorded set, the velocity was established at 60°/second as performed in a previous study (Renstrom et al., 1983b). Participants were asked to pull and push as hard as possible in concentric FLX and EXT direction for six complete cycles within the range of motion for each joint.

In general isokinetic dynamometers including the Kin-Com, exhibited the highest level of validity and reliability for strength measurement. A review of the literature by Nitschke (1992) supports this statement by analysing several studies that used different isokinetic dynamometers and settings to evaluate strength.

3.5 Procedures.

All participants first read and signed a consent form previous to the evaluation. We then evaluated the level of physical activity using the HAP and Tegner questionnaires. We also registered the record of antecedents to ensure the absence of previous injuries or any other exclusion criteria.

The first measurement performed was the TPS. We started at the tibial site and then proceeded to the plantar site. The second test was proprioception. Participants performed 12 random trials, 6 in FLX and 6 in EXT. The next test evaluated was balance performance during single-leg stance. The test included three randomized trial conditions: standing still with EO, standing still with EC and balance performance after squat with EO. During balance assessment, we applied the Borg Scale of Global Muscle Fatigue before and after each condition. Participants rested for three minutes after each balance condition was tested, to reduce the effect of muscle fatigue on balance performance. The last evaluation performed was muscular strength. We evaluated first the knee and then the ankle joint during concentric FLX and concentric EXT contractions.

3.6 Data Analysis.

To estimate TPS, we calculated the total median value from the three ascending and the three descending measurements for each tested site. The joint TADM was calculated using the data from the custom made proprioceptive apparatus. We measured the difference between the starting position angle (20° of knee FLX) and the knee angle where the participant perceived the joint displacement, in either knee FLX or knee EXT direction. The mean threshold for knee FLX or knee EXT was computed from a total of six trials in FLX and six trials in EXT direction, respectively.

The PT-BW was the measurement used to evaluate muscular strength either in FLX or in EXT direction. It was recorded over the 6 cycles performed during the test using the Kin-Com[®] dynamometer.

For the balance assessment, the MatScan[®] system indicated the position of the COP for each measured frame. A period of thirty seconds from the beginning of the stand trials and after the squats was used to evaluate COP variables. Similar studies that measured the COP, used recording trial periods which ranged from 30 to 60 seconds (Buckley et al., 2002; Dornan et al., 1978; Duclos et al., 2009; Duclos et al., 2007; Fernie & Holliday, 1978; Hermodsson et al., 1994; Isakov et al., 1992; Nadollek et al., 2002). We analyzed the COP variables after the squat (not during the test or during the recuperation phase) because we wanted to evaluate the 'long-term' effect of the reactive response to balance perturbation and also the potential effect of the voluntary muscles anticipatory response as suggested by Winter (1995). A script allowed us to establish the offset threshold point during the squat using the electrogoniometer data. The point indicated the angle where the knee joint reached 5% of the maximum angular speed during knee EXT (Figure 7). Using the COP data we computed the COPv, RMSd and the RMSv for both the AP and the ML directions in all the conditions. Some authors consider the velocity as one of the most reliable methods to measure sway (Cornilleau-Peres et al., 2005; Lafond et al., 2004; Lin et al., 2008; Raymakers et al., 2005) . Moreover, according to Geurts et al. (1993), and Pinsault & Vuillerme (2009) RMSv is less affected during EO condition. These measurements allowed us to analyse independently, the AP and the ML direction

and the potential difference in these two components of the COP variables. Data was averaged over the 3 recorded trials for each condition and each participant.

The MatScan[®] system consists of a 50.8 cm per 49.9 cm mat with a matrix of 43.59 cm per 36.88 cm. The sensel density is 1.4 per cm² and, a total of 2288 sensels are located in the matrix. Using a script we were able to do vector analysis decomposition and evaluate each direction independently for all computed variables.

Using the row sensor and the column sensor position multiplied by the respective row and column sensor spacing, we determined the exact location of the COP in the matrix location. This measurement of the COP position in the matrix is indicated in cm for both, the AP (matrix rows) and the ML (matrix columns) directions. We used the following formula to compute the COP location for each individual frame:

 $COP_i = SLi * SS$

where SL = sensor location, SS = sensor spacing and, *i* is the frame number.

The COPd was computed from the sum of the absolute value of the differences between the COP locations in the matrix of two consecutive frames in the test. Dividing COPd by time we were able to obtain COPv.

$$\operatorname{COPd} = \sum_{i=1}^{n-1} (|\operatorname{COP}_{i+1} - \operatorname{COP}_i|)$$

$$COPv = \frac{COPd}{t}$$

where n = total number of frames analyzed and, t = time.

The RMSd and the RMSv were calculated according to the following formulas:

$$\text{RMSd} = \sqrt[2]{\frac{1}{n} \sum_{i=1}^{n} (\text{COP}_i - \overline{\text{COP}})^2}$$

$$RMSv = \sqrt[2]{\frac{1}{n} \sum_{i=1}^{n} (COPv_i - \overline{COPv})^2}$$

All of the formulas were used separately for the AP or the ML directions.

In Hermodsson et al. (1994), they analysed the time in balance for those participants who were not able to keep balance during single-legged stance. Most of the participants in our study were not able to stand with EC for 45 seconds, but we were not able to determine the time in balance. For these reasons, we did not analyze standing still condition under EC.

Knee Angle During Squat Condition

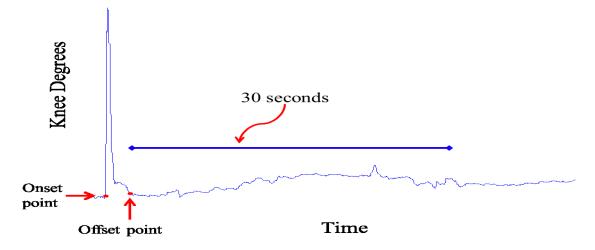


Figure 7. Knee angle during dynamic (squat) balance assessment. The COP analysis was performed using the data collected for a period of 30 seconds after the end of the squat (offset point).

3.7 Statistics.

We reported descriptive statistical values for the level of physical activity of all research participants. Muscular fatigue was analysed using a three-way repeated measures analysis of variance (ANOVA). We evaluated one between factor (group: amputees/controls) and two within factors; one factor related to the period of assessment (pre-test/ post-test) and one factor for all the conditions (standing EO, standing EC and squatting). This analysis was performed in order to ensure that muscular fatigue did not influence balance results. Cutaneous sensation was analysed using a two-way repeated measures ANOVA with one between factor related to the group: amputees/controls and one within factor related to the group: amputees/controls and one within factor related to the tested site: tibial/plantar sole.

Proprioception was analysed using a two-way repeated measures ANOVA with one between factor (group: amputees/controls) and one within factor (direction: FLX/EXT).

We also analysed the PT-BW at knee and ankle joints. Two two-way repeated measures ANOVA were performed for FLX and EXT strength separately with one between factor related to group: amputees/controls and one within factor related to joint: knee/ankle. Two other two-way repeated measures ANOVA analyzed the knee and ankle separately. The ANOVA included the same between factor related to group (amputees/controls) and a different within factor related to direction: FLX/EXT.

For strength, we also included the analysis of muscular strength ratios using one two-way repeated measures ANOVA. This included one between factor related to the group: amputees/controls and one within factor related to the joint: knee FLX/EXT ratio or ankle D-FLX/P-FLX ratio.

To analyse the COP variables we performed two-way repeated measures ANOVA. Two two-way repeated measures ANOVA were related to the direction (AP and ML separately). The analysis included one between factor related to group (amputees/controls) and one within factor related to the condition: stand /squat. The other two two-way repeated measures ANOVA were related to condition (stand and squat separately). Again, the analysis included one between factor related to group: amputees/controls and one within factor related to the direction: AP and ML separately. Pearson's correlations were performed to evaluate the relationship between cutaneous sensation, proprioception, muscular strength and balance performance (COPv, RMSv, RMSd) during quiet standing and after squatting. All the correlations were performed using the total quantity of participants' data pooled together.

A p-value ≤ 0.05 was used as the evaluation criteria to consider that a variable outcome evidenced significant differences.

<u>Chapter 4 – Results.</u>

In this section we first ensure that the level of physical activity was similar for both groups and also that muscular fatigue did not affect the evaluation of the different conditions. We then summarize the results from the different measurements and compare them between and within groups (means and SD).

4.1 Level of Physical Activity.

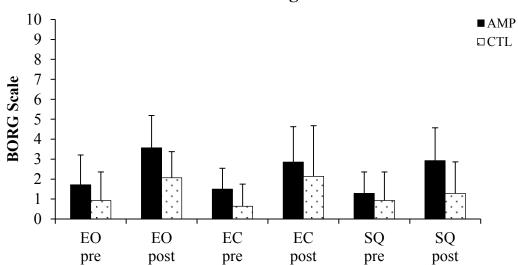
The results showed by the HAP questionnaire in amputees evidenced a M = 80.2, SD = 14.8 points and a M = 74.7, SD = 15.7 points for the MAS and the AAS respectively. Able-bodied participants displayed a M = 93.5, SD = 2.5 points for both, the MAS and the AAS. The mean level of activity showed by amputees using the Tegner score was M = 3.0, SD = 1.0 points and, the same test showed a M = 3.8, SD = 1.0 points in controls.

4.2 Level of Muscular Fatigue.

Results for muscular fatigue are presented in Figure 8. The repeated measures ANOVA indicated no difference between groups (F = 2.30, p = 0.15), conditions (F = 1.29, p = 0.29), or the interactions: condition by group (F = 0.19, p = 0.83), time by group (F = 1.32, p = 0.27), condition by time (F = 0.56, p = 0.58) or condition by time by group (F = 0.99, p = 0.39).

However, the results revealed significant differences for the assessment time (F = 23.71, p < 0.001). The results evidenced lower values during pre-test conditions (stand EO pre: M = 1.32, SD = 1.46; stand EC pre: M = 1.07, SD = 1.12; squat pre: M = 1.11, SD = 1.23)

compared to post-test values conditions (stand EO post: M = 2.82, SD = 1.61; stand EC post: M = 2.50, SD = 2.13; squat post: M = 2.11, SD = 1.77).



Muscular Fatigue Level

Figure 8. Mean values of muscular fatigue levels. The figure shows the level of muscular fatigue for both groups during pre-test and post-test periods for standing with EO, standing with EC and squat conditions.

4.3 Balance.

Results for balance (COPv, RMSv, and RMSd) are summarized in Figures 9, 10, and 11.

4.3.1 AP and ML Directions.

For the AP direction, no difference was demonstrated for group, condition, or interactions in COPv, RMSd or RMSv. The COPv evidenced an F = 3.10, p = 0.11 for group, an F =1.15, p = 0.31 for condition and, for the interaction an F = 0.58, p = 0.46. The RMSd displayed an F = 1.83, p = 0.20 for group, an F = 0.24, p = 0.63 for condition and, for the interaction an F = 0.04, p = 0.84. The last one corresponded to the RMSv which evidenced an F = 3.68, p = 0.08 for group, an F = 0.01, p = 0.92 for condition and, for the interaction an F = 0.85, p = 0.82.

When evaluated the ML direction, RMSd showed no difference between groups (F = 1.10, p = 0.32). However, there were significant differences related to group in COPv (F = 6.73, p = 0.02) and RMSv (F = 6.10, p = 0.03), with higher mean values in COPv in controls (squat ML: M = 2.81, SD = 0.38 cm/s; stand ML: M = 3.15, SD = 0.57 cm/s) compared to traumatic amputees (squat ML: M = 2.20, SD = 0.73 cm/s; stand ML: M = 2.15, SD = 0.63 cm/s). Controls also described higher mean values for RMSv (squat ML: M = 4.49, SD = 1.67 cm/s; stand ML: M = 4.39, SD = 0.93 cm/s) compared to amputees (squat ML: M = 3.02, SD = 0.91 cm/s). COPv, RMSd and RMSv did not report differences related to condition (COPv: F = 1.63, p = 0.23; RMSd: F = 0.002, p = 0.97; RMSv: F = 0.08, p = 0.79) or group by condition interaction for the ML direction (COPv: F = 4.23, p = 0.06; RMSd: F = 0.76, p = 0.40; RMSv: F = 0.002, p = 0.97).

4.3.2 Standing and Squatting.

Other comparisons for COP variables included the separate evaluation of the conditions (standing and squatting). The results revealed no difference between groups for the squat condition (COPv: F = 3.57, p = 0.09; RMSd: F = 1.64, p = 0.23; RMSv: F = 3.10, p = 0.11) nor the stand condition (COPv: F = 3.20, p = 0.10; RMSd: F = 0.87, p = 0.37; RMSv: F = 1.98, p = 0.18).

However, all COP variables displayed significant differences related to the AP and ML directions under the squat condition (COPv: F = 4.79, p = 0.05; RMSd: F = 36.47, p < 0.001; RMSv F = 8.50, p = 0.01). The squat condition in AP direction revealed higher mean values of COPv (M = 2.83, SD = 0.94 cm/s), RMSd (M = 0.72, SD = 0.15 cm) and RMSv (M = 5.02, SD = 2.78 cm/s) when compared to squat ML direction (COPv: M = 2.53, SD = 0.63 cm/s; RMSd: M = 0.60, SD = 0.11 cm; RMSv: M = 3.83, SD = 1.54 cm/s). The direction effect for the stand condition evidenced similar results but, revealed significant differences only for the RMSd (F = 23.46, p < 0.01) and the RMSv (F = 6.42, p = 0.03). The stand AP direction exhibited higher mean values (RMSd: M = 0.75, SD = 0.21 cm; RMSv: M = 5.19, SD = 2.78 cm/s) compared to stand ML direction (RMSd: M = 0.60, SD = 0.14 cm; RMSv: M = 3.71, SD = 1.14 cm/s). Despite revealing no difference in COPv values (F = 4.46, p = 0.06) between AP and ML directions, the results indicated a trend to significant higher mean values in the AP direction (M = 3.20, SD = 1.39 cm/s) compared to the ML direction (M = 2.68, SD = 0.78 cm/s).

The interaction direction by group also indicated significant differences only for the squat condition in the RMSd (F = 5.62, p = 0.04) but not for COPv (F = 0.74, p = 0.40) or RMSv (F = 1.52, p = 0.24). Controls evidenced higher mean values (squat AP: M = 0.78, SD = 0.18 cm; squat ML: M = 0.61, SD = 0.14 cm) when compared to amputated individuals (squat AP: M = 0.65, SD = 0.07 cm; squat ML: M = 0.57, SD = 0.14 cm). For the stand condition, the interaction direction by group did not report significant differences for any of the COP variables (COPv: F = 0.12, p = 0.73; RMSd: F = 0.01, p = 0.91; RMSv F = 0.002, p = 0.96).

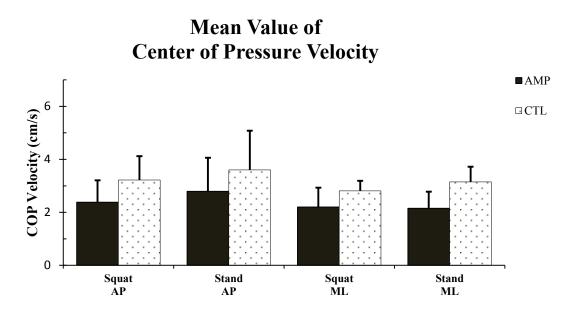


Figure 9. Mean values of the COPv for AP and ML direction during dynamic (squat) and static (stand) conditions with EO. Significant differences between amputees and able-bodied controls were evidenced only for the ML direction.

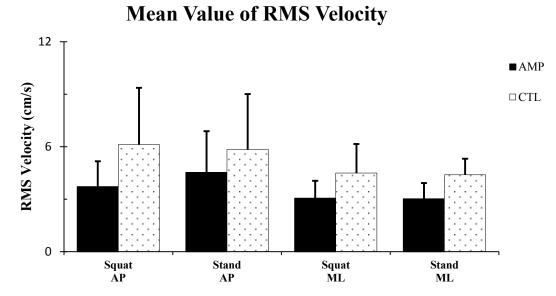


Figure 10. Mean values of the RMSv for AP and ML direction during dynamic (squat) and static (stand) conditions with EO. Significant differences between amputees and able-bodied controls were evidenced only for the ML direction.

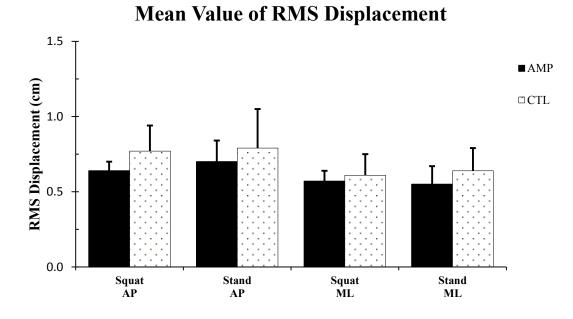


Figure 11. Mean values of the RMSd for AP and ML direction during dynamic (squat) and static (stand) conditions with EO. No significant differences were observed between amputees and able-bodied controls.

4.4 Cutaneous Sensation.

Figure 12 displays the TPS values for both groups at both the tibial and plantar sites, presented as a number representing the force in grams (log10) required to bend the filament. No difference was described related to group (F = 1.40, p = 0.26), site (F = 2.52, p = 0.14) or site by group interaction (F = 2.23, p = 0.16).

4.5 Proprioception.

The mean values of the knee angular displacement during proprioception assessment are presented in Figure 13. No difference in mean proprioception values were evidenced

related to the group (F = 0.34, p = 0.57), direction (F = 0.78, p = 0.40) or the direction by group interaction (F = 0.71, p = 0.42).

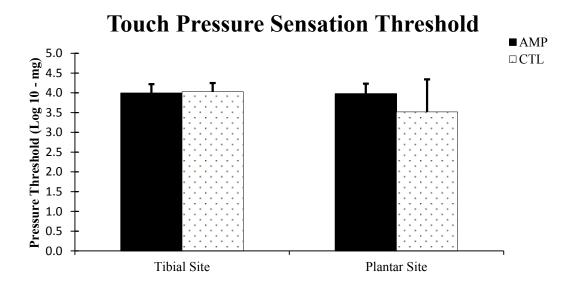


Figure 12. Median values of TPS thresholds at tibial and plantar sites.

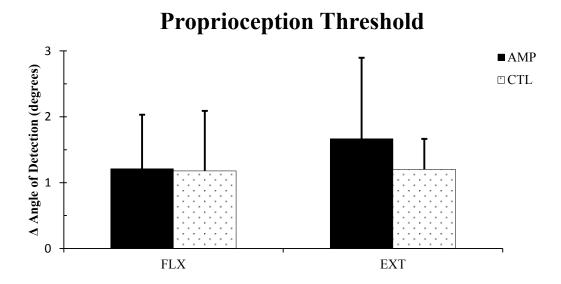


Figure 13. Mean values for knee angular displacement measurements during FLX and EXT motion for the proprioception procedure.

4.6 Muscular Strength.

Figure 14 shows the strength values for knee and ankle joints in FLX and EXT directions for both, traumatic amputees and able-bodied controls. When evaluating separately the strength at the knee and at the ankle joints, the study revealed no difference between groups in both joints (knee: F = 0.81, p = 0.39; ankle: F = 1.30, p = 0.28). Similar results were described for the interaction direction by group with no difference revealed for both joints (knee: F = 1.0, p = 0.34; ankle: F = 0.008, p = 0.93). However the direction effect evidenced significant differences at the knee (F = 9.8, p = 0.01) and at the ankle joint (F = 16.68, p = 0.002). The knee displayed a higher mean PT-BW for EXT muscles (M = 1.52, SD = 0.29 N/Kg) compared to knee FLX (M = 1.17, SD = 0.39 N/Kg). At the ankle joint similar results were described, and P-FLX muscles showed higher PT-BW (M = 0.72, SD = 0.27 N/Kg) as compared to ankle D-FLX muscles which revealed lower mean values (M = 0.56, SD = 0.21 N/Kg).

When evaluating separately the directions, the study revealed no difference between groups, neither for the FLX (F = 2.27, p = 0.16) nor EXT (F = 0.40, p = 0.54) direction. No differences were described for the joint by group interaction (FLX: F = 0.16, p = 0.69; EXT: F = 0.95, p = 0.35). However, the results evidenced significant difference for joint effect. They displayed an F = 13.26 (p = 0.03) for joint FLX and, an F = 257.75 (p < 0.001) for joint EXT muscles. Knee FLX (M = 1.17, SD = 0.39 N/Kg) and knee EXT (M = 1.52, SD = 0.29 N/Kg) muscles showed significant higher values than ankle D-FLX (M = 0.56, SD = 0.21 N/Kg) and ankle P-FLX (M = 0.72, SD = 0.27 N/Kg) muscles respectively.

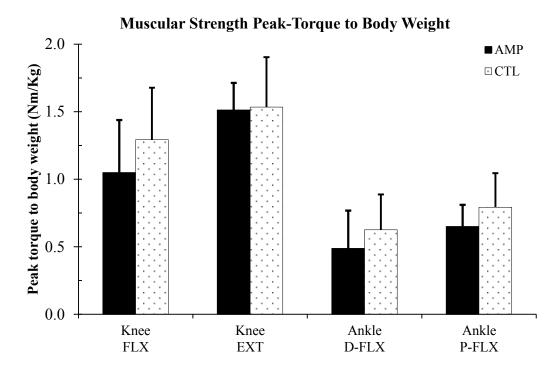


Figure 14. Mean values of the PT-BW at the knee and the ankle during isokinetic concentric FLX and EXT.

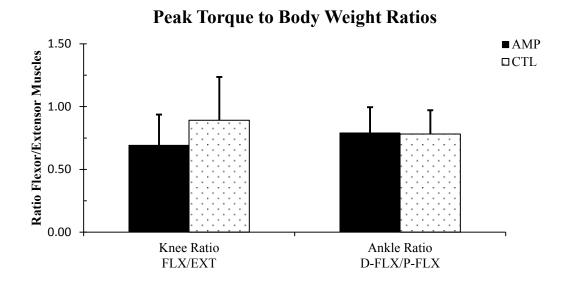


Figure 15. Mean values of the ratios for knee and ankle FLX/EXT muscles during isokinetic concentric motion.

Strength ratios are presented in Figure 15. When comparing joints' ratios, the results revealed no difference related to group (F = 1.40, p = 0.26), joint (F = 0.002, p = 0.97) or the joint by group interaction (F = 0.84, p = 0.38).

4.7 Correlations.

Pearson correlations were performed between COP variables and the other variables that could potentially affect balance. All the correlations performed between COP and all the study variables (TPS, proprioception, and muscular strength) are presented in Appendix D and the correlation graphs in Appendix E (Tables 3, 4, 5). Pearson coefficients revealed significant correlation levels for muscular strength, in particular ankle strength for the P-FLX direction and, strength ratios. Table 2 summarizes the coefficients that evidenced significant statistical values ($p \le 0.05$) or a trend to significant correlations.

COP	Variable	Condition	Pearson Correlation	p-value
	TPS tibial site	Squat AP	0.54	0.06 [†]
COD		Squat AP	0.64	0.02
COPv	Ankle P-FLX	Squat ML	0.60	0.03
		Stand AP	0.70	0.01
	Ankle P-FLX	Squat AP	0.63	0.02
RMSd	AIIKIE F-FLA	Stand AP	0.53	0.05
	Knee FLX/EXT ratio	Stand AP	0.52	0.06 ⁺
		Stand ML	0.53	0.06 ⁺
		Squat AP	0.55	0.06 ⁺
RMSv		Squat ML	0.61	0.03
	Ankle P-FLX	Stand AP	0.68	0.01
	Ankle D-FLX/P-FLX ratio	Squat AP	-0.58	0.04
		Squat ML	-0.61	0.03

Table 2. Significant correlations between the COP (COPv, RMSd, RMSv) and other variables. († non-significant difference).

<u>Chapter 5 – Discussion.</u>

The first goal of our study was to evaluate cutaneous sensation, proprioception, muscular strength, and balance in the non-amputated side of TTA compared to a randomly selected limb in able-bodied controls; the second was to use the amputation condition as a model to evaluate the role of cutaneous sensation, proprioception and muscular strength on balance performance. In our study, the only variable that showed differences between groups was balance. Amputees demonstrated reduced values in COPv and RMSv compared to able-bodied controls for ML direction.

The other variable that evidenced significant differences was muscular strength, but these differences were related to the joint and direction tested. The results displayed lower strength level at the ankle joint compared to the knee and also reduced muscular strength in knee FLX and ankle D-FLX compared to knee EXT and ankle P-FLX muscles respectively. As for correlations the main variable that indicated a relationship with balance was muscular strength predominantly at the ankle level, and strength ratios.

Given the results obtained from our study, we are able to suggest that the differences displayed between groups for the variables of COP are best explained by muscular strength. However, given the fact that our study does not contain a large number of participants, the aforementioned suggestion may not necessarily fully explain this difference.

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5.1 Balance.

As hypothesized, balance was affected in amputees in our study. The loss of information from anatomical structures demanded several adaptations on different systems in order to maintain balance. As a result, we expected an increment of COP variables in amputees. Our expectation for reduced balance was also based in part on the existing literature. An increase in amputees' COP variables was described by diverse studies during doublelegged stance (Buckley et al., 2002; Dornan et al., 1978; Fernie & Holliday, 1978; Geurts et al., 1991; Hermodsson et al., 1994; Isakov et al., 1992; Nadollek et al., 2002). Various studies also associated the reduction of COPv (Le Clair & Riach, 1996) as well as RMSd and RMSv (Geurts et al., 1993) with better balance performance.

The results that we present for COP variables are opposed to the ones proposed in our hypothesis. Although we expected increments of the COP variables in amputees based on the results that similar studies reported in the literature, we described reduced values of COP variables in TTA when compared to able-bodied controls. It should be noted however that our study was performed during single-legged stance on the non-amputated side. In our study it is possible that the greater use and greater reliance of the non-amputated side on a daily basis in TTA could lead to a greater stability. The non-amputated side may therefore be more stable and strong. The increment in AP direction for COP excursion on the sound limb side compared to the amputated side during double-legged stance (Nadollek et al., 2002) may be related to the trend described in amputees to displace their body weight to the non-amputated side (Duclos et al., 2009; Duclos et al., 2007; Engsberg et al., 1992; Isakov et al., 1992).

To our knowledge, only Hermodsson et al. (1994) compared single-legged stance on the non-amputated limb of TTA to able-bodied controls. Despite not reporting differences in COP amplitude for AP and ML directions on the non-amputated limb, the authors revealed significant shorter time in balance for all amputees together when compared to able-bodied controls. Vascular amputees in particular showed significant shorter standing time when compared alone to able-bodied, or when compared to traumatic amputees. It is possible that the sound limb of traumatic amputees in our study was not as affected as those of vascular amputees described by Hermodsson's study.

It should be noted that, similar to our study, a greater mean surface of sway has been displayed in able-bodied controls when compared to traumatic and non-traumatic amputees (Gauthier-Gagnon et al., 1986). Despite revealing lower values of mean sway surface and a greater ABS, amputees used only a small proportion of the ABS (0.3%) compared to the proportion used by able-bodied controls (2%). According to the authors, this proportion of the ABS represents the area of stability, and therefore a lower proportion of use of this area could be interpreted as a less stable condition. In addition to this, a reduction in the activity of muscles about the ankle joint has been associated with a reduction in postural sway (Gurfinkel et al., 1995; Gurfinkel et al., 1979). This result may also explain in part, why lower values in COP variables do not necessarily indicate a better balance performance. There are similar results showed within a study by Davids et al. (1999), where individuals with anterior cruciate ligament injury displayed lower COP variables compared to controls. We should also consider the idea proposed by Duclos et al. (2009) suggesting that the CNS uses this strategy as a mechanism to obtain more information for balance control.

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The study presented by Buckley et al. (2002) also included double-legged stance, but instead they performed a dynamic balance assessment using a stabilimeter. In our study, we also evaluated dynamic balance. However, we studied COP variables (COPv, RMSd and RMSv) and different conditions (squatting during single-legged stance). In the study by Buckley et al. (2002), reduced values for time in balance in TTA compared to controls was reported, meaning amputees were less stable during dynamic balance assessment. The other variables measured in Buckley's study demonstrate that the differences in the number of board contacts were only significant for ML direction under blindfolded conditions, with more board contacts on the amputated side. Despite the absence of differences, related to the groups, the size effect suggested a trend to higher mean time of board contacts for AP condition (Buckley et al., 2002).

5.2 Cutaneous Sensation.

In our study, cutaneous sensation did not reveal differences related to group, site or site by group interaction. However, other studies such as Kosasih & Silver-Thorn (1998), and Kavounoudias et al. (2005) reported reduced levels of sensation in amputees. Both Kavounoudias et al. (2005) and Kosasih & Silver-Thorn (1998) compared the amputated side to the non-amputated side. However, only Kavounoudias et al. (2005) compared sensation of the non-amputated to able-bodied control group.

In our study, we used the same sites and methods as Kavounoudias et al. (2005) to assess TPS. The absence of differences between groups in our study may be associated with the fact that Kavounoudias et al. (2005) evaluated amputees from traumatic and non-

traumatic etiologies. It is described that amputations related to diabetes and vascular diseases are the most frequent etiologies of non-traumatic amputations which frequently exhibit an altered cutaneous sensitivity. These diseases also lead to the concomitant development of sensory neuropathy which can influence not only cutaneous sensation but also proprioception (van Deursen & Simoneau, 1999). However, when Kavounoudias et al. (2005) compared vascular and traumatic amputees separately, they reported that only traumatic amputees showed significant higher TPS values specifically at the plantar site.

5.3 Proprioception.

The assessment of proprioception in our study did not demonstrate differences between groups, directions or direction by group interaction. This does not correspond to our expectations and to some of the existing studies (Eakin et al., 1992; Kavounoudias et al., 2005; Liao & Skinner, 1995). These studies described reductions in proprioceptive information in amputees. Eakin et al. (1992), Kavounoudias et al. (2005) and, Liao & Skinner (1995) measured proprioception in LLA from diverse etiologies and levels (AKA and BKA or BKA alone) using different settings (seated or standing) and various speeds which, ranged from 0.4°/second to 0.7°/second.

Eakin et al. (1992) and Liao & Skinner (1995) reported higher TDPM values in the amputated limb compared to the non-amputated limb. These results are opposite to those of Kavounoudias et al. (2005), who reported non-significant differences in TDPM.

Similar to our study, Liao & Skinner (1995) and Kavounoudias et al. (2005) compared the non-amputated side to controls. Both of them revealed higher TDPM in the nonamputated side compared to able-bodied controls. Liao & Skinner used a speed of $0.4^{\circ}/s$ and pooled traumatic and vascular amputees, which may have led to different results. However, Kavounoudias et al. (2005) used the same speed as in our study $(0.7^{\circ}/s)$ and evaluated vascular and traumatic amputees separately. Their results indicated higher TDPM values on the non-amputated side at the knee, but not at the ankle joint for both, traumatic and vascular amputees when compared to controls. The absence of difference for proprioception in our study may be related to the settings used in our experiment. In our study we evaluated the level of proprioception using TADM, a procedure similar to the TDPM. Note that Kavounoudias et al. (2005) performed their evaluation while the participants were seated while we performed the assessments with the participants bearing their body-weight. The amputated individuals from our study may have perceived much faster the joint displacement due to the weight-bearing condition because of the related "pre-activation" of different muscles involved in balance control during doublelegged stance. It is possible that the participants also obtained proprioceptive information from the activation of different muscles involved in balance maintenance. Also, the procedure in our study implied the displacement of multiple joints: the ankle, the knee and the hip. All these structures could provide proprioceptive information and may explain the absence of differences in our study. Also, the fact that the standing position is more challenging for the balance system may have influenced the results.

5.4 Muscular Strength.

When we evaluated muscular strength, our results revealed no significant difference between the groups. This is not what we expected and is different from other studies which showed a reduction in muscular strength in amputees.

Studies that compared the amputated side to the non-amputated side reported reduced muscular strength in the amputated side (Isakov et al., 1996b; Moirenfeld et al., 2000; Pedrinelli et al., 2002). The aforementioned result suggests that muscular strength might not be affected in the non-amputated limb.

We have considered the possibility of "a training effect". It has been reported that amputees support more body-weight on the non-amputated side (Duclos et al., 2009; Duclos et al., 2007; Gauthier-Gagnon et al., 1986; Isakov et al., 1992). The second reason could be the level of physical activity. Although it was not part of the main objectives of this study, we evaluated the level of physical activity. The participants in our research were classified as physically active according to the AAS scale in the HUMAP questionnaire. The increased, constant use of the non-amputated limb may have an effect on muscular strength in amputees resulting in smaller or no difference between the nonamputated extremity and the selected limb assessed in the able-bodied-control group.

Note, however that Pedrinelli et al. (2002) not only compared the amputated side to the non-amputated side from different etiologies. They also evaluated muscular strength on the non-amputated side in amputees and compared it to that of able-bodied controls. In their study, they described a reduction in muscular strength of the non-amputated side. Pedrinelli et al. (2002) recruited traumatic and non-traumatic amputees to evaluate

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various parameters. Testing two different speeds, they evaluated muscular strength and muscular resistance. They used a set of 4 repetitions at the speed of 60°/second to evaluate muscular strength. They also tested 6 from a maximum of 20 repetitions at a speed of 180°/second, to evaluate muscular resistance. All of the measurements were relative to concentric/concentric contractions during knee FLX and EXT direction. It is possible that the different settings used by Pedrinelli might be more sensitive to the amputation. In addition, Pedrinelli et al. (2002) included amputees from mixed etiologies in their study and we included only amputees from traumatic origin. The inclusion of vascular amputees in Pedrinelli's study may have led to the decrease in strength reported in amputees.

5.5 Correlations.

For correlations, we evaluated the role of cutaneous sensation, proprioception and muscular strength on balance performance. To our knowledge, the literature has not reported any studies that compared at the same time TPS, proprioception and muscular strength against performance in tasks involving balance.

In our study, the only variable that revealed significant correlations with the COP was muscular strength, predominantly about the ankle joint. The increments in COP variables and, the level correlation between COP variables and muscular strength at the ankle level allow us to suggest the importance of muscle strength at the ankle in balance maintenance. It seems also important to keep an adequate balance between D-FLX and P-FLX muscles. The ankle strategy is one of the most important mechanisms involved in

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balance maintenance (Winter, 1995, 2009; Winter et al., 1990) and muscular strength changes may explain in part the variations in balance performance.

We did not report significant correlations between TPS, proprioception and balance. A possible explanation could be an adaptation of the CNS, similar to that proposed for balance (Duclos et al., 2009). The CNS might use the proprioceptive information from the remaining receptors in the amputated extremity and the information originated from the receptors in the sound limb to compensate the proprioception deficit. Another possibility is related to the sample size. These factors might probably influence the statistical analysis and the determination of correlations among the variables.

The correlations between strength and COP variables should be taken carefully due to the controversial interpretations of COP variables (Palmieri et al., 2002). Considering the studies where reduced COP variables indicate a greater postural control (Baier & Hopf, 1998; Baloh et al., 1998; Davids et al., 1999; Geurts et al., 1993; Le Clair & Riach, 1996), a positive correlation will display a less stable condition when muscular strength increases. On the other hand, if we consider the study by Davids et al. (1999), the same correlation for muscular strength will indicate that muscular strength increments would be associated to a more stable position during postural challenge. This correlation seems to be apparently more logical. Moreover, individuals with greater strength levels may increase the oscillation levels in order to have a better control of their balance.

Chapter 6. Limitations.

In our study, we did not include the limb dominance as a parameter to evaluate balance in TTA. Not considering the limb dominance is a limitation for our study. We omitted this factor due to the possibility that it could have been affected by the amputation. We considered different scenarios including one where the dominant limb may have been amputated. As a result and subsequent to surgery, the individuals could have started to use the non-amputated limb as a dominant limb. This situation could mislead the results related to the limb dominance.

The evaluation of proprioception was performed under assisted conditions. The parameters of evaluation demanded participants to support their body weight during the proprioception task. Keeping the body in an upright position involves a coordinated action of various muscle groups. Thus, we can consider that there is a "pre-activation" response of the muscles during the evaluation of proprioception. This "pre-activation" can also modify the assessment of proprioception during joints' displacement. In addition, during proprioception assessment, the test was performed without hand contact on the support. We chose this position because we did not want participants to hold part of their body weight, even though the standing position also involved a balance task. The purpose of these settings was to evaluate proprioception under a condition the closest to a real life situation where the proprioception information is more relevant during an upright position which demands a higher level of muscular activation.

We consider the total number of participants in the current study as a limitation. We were able to match only seven TTA, which volunteered to participate in the study. The limited number of participants restricted the possibility to perform additional statistical tests to

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measure the potential effect of other factors. In addition, the absence of differences could be due to a reason other than that specified in the hypothesis. An unknown process may underlie the relationship between COP measurements and the other variables.

If we could repeat the study, it would be ideal to include the evaluation of amputees in different periods of the rehabilitation process to evaluate the effect of this intervention on balance performance.

<u>Chapter 7 – Conclusions.</u>

Balance and postural control are considered a multifactorial condition. Different studies have included the assessment of cutaneous sensation, proprioception, and muscular strength separately.

Our study reports a reduction of different COP parameters in amputees and changes in muscular strength related to the direction of the assessment. Reductions of COPv and RMSv in amputees were shown to be significant particularly for the ML direction, contrary to most studies that reported increments in COP variables when amputees were compared to able-bodied controls. It is possible that the reduced COP values are due in part to the fact that amputees rely more on their sound limb on a day-to-day basis during ambulation and standing. Yet, reduced values of COP variables do not necessarily indicate an increase in balance performance.

The correlations for COP variables mainly related to muscular strength, in particular to ankle strength, indicating the influence of muscle strength in balance performance. However, we can assume that this correlation is logical only if the increase in COP variables indicates an increased balance.

Based on our results and also on the literature review we can state the importance of a muscle strengthening program. This program must be accompanied by activities of body-weight translation and body weight control to distribute as evenly as possible, the body weight between both extremities.

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Appendix A

Human Activity Profile

C - 1	
Code:	
Couc.	

Date:

Instructions:

This brochure contains elements describing tasks of daily life. Read each statement and mark an X in the column indicating either you are *Still Doing This Activity*, you *Have Stopped Doing This Activity* or *Never Did this task*. Use the following instructions to guide you in your choice of responses:

- Draw an X in the column that "Still Doing" if you have completed this task without help the last time or opportunity you have had need.
- Draw an X in the column **"Have stopped doing this activity"** if you have completed the task in the past but would not be able to do the same today if you have the chance.
- Draw an X in the column "Never did this activity" if you've never undertaken this task.

Code: _____ Date: _____

		Still Doing This Activity	Have Stopped Doing This Activity	Never Did This Activity
1.	Getting in and out of chairs or bed (without assistance)			
2.	Listening to the radio			
3.	Reading books, magazines, or newspapers			
4.	Writing (letters, notes)			
5.	Working at a desk or table			
6.	Standing (for more than 1 minute)			
7.	Standing (for more than 5 minutes)			
8.	Dressing or undressing (without assistance)			
9.	Getting clothes from drawers or closets			
10.	Getting in or out of a car (without assistance)			
11.	Dining at a restaurant			
12.	Playing cards/table games			
13.	Taking a bath (no assistance needed)			
14.	Putting on shoes, stockings, or socks (no rest or break needed)			
15.	Attending a movie, play, church event, or sports activity			
16.	Walking 30 yards (27 meters)			
17.	Walking 30 yards (nonstop)			
18.	Dressing/undressing (no rest or break needed)			
19.	Using public transportation or driving a car (99 miles or less)			
20.	Using public transportation or driving a car (100 miles or more)			
21.	Cooking your own meals			
22.	Washing or drying dishes			

Code: _____ Date: _____

	Still Doing	Have Stopped	Never Did
	This Activity	Doing This Activity	This Activity
23. Putting groceries on shelves			
24. Ironing or folding clothes			
25. Dusting/polishing furniture or polishing a car			
26. Showering			
27. Climbing 6 steps			
28. Climbing 6 steps (nonstop)			
29. Climbing 9 steps			
30. Climbing 12 steps			
31. Walking ¹ / ₂ block on level ground			
32. Walking ½ block on level ground (nonstop)			
33. Making a bed (not changing sheets)			
34. Cleaning windows			
35. Kneeling, squatting to do light work			
36. Carrying a light load of groceries			
37. Climbing 9 steps (nonstop)			
38. Climbing 12 steps (nonstop)			
39. Walking ¹ / ₂ block uphill			
40. Walking ¹ / ₂ block uphill (nonstop)			
41. Shopping (by yourself)			
42. Washing clothes (by yourself)			
43. Using public transportation or driving a car			
(100 miles or more)			
44. Cooking your own meals			
45. Washing or drying dishes			
46. Putting groceries on shelves			
47. Ironing or folding clothes			

Code: _____ Date: _____

	Still Doing	Have Stopped	Never Did
	This Activity	Doing This	This Activity
		Activity	
48. Dusting/polishing furniture or polishing a car			
49. Showering			
50. Climbing 6 steps			
51. Climbing 6 steps (nonstop)			
52. Climbing 9 steps			
53. Climbing 12 steps			
54. Walking ¹ / ₂ block on level ground			
55. Walking ¹ / ₂ block on level ground (nonstop)			
56. Making a bed (not changing sheets)			
57. Cleaning windows			
58. Kneeling, squatting to do light work			
59. Carrying a light load of groceries			
60. Climbing 9 steps (nonstop)			
61. Climbing 12 steps (nonstop)			
62. Walking ¹ / ₂ block uphill			
63. Walking ¹ / ₂ block uphill (nonstop)			
64. Shopping (by yourself)			
65. Washing clothes (by yourself)			
66. Walking 1 block on level ground			
67. Walking 2 blocks on level ground			
68. Walking 1 block on level ground (nonstop)			
69. Walking 2 blocks on level ground (nonstop)			
70. Scrubbing (floors, walls or cars)			
71. Making a bed (changing sheets)			
72. Sweeping			
73. Sweeping (5 minutes nonstop)			
74. Carrying a large suitcase or bowling (one			
game)			
75. Vacuuming carpets			

Code: _____ Date: _____

76. Vacuuming carpets (5 minutes nonstop)	
77. Painting (interior/exterior)	
78. Walking 6 blocks on level ground	
79. Walking 6 blocks on level ground (nonstop)	
80. Carrying out the garbage	
81. Carrying a heavy load of groceries	
82. Climbing 24 steps	
83. Climbing 36 steps	
84. Climbing 24 steps (nonstop)	
85. Climbing 36 steps (nonstop)	
86. Walking 1 mile	
87. Walking 1 mile (nonstop)	
88. Running 110 yards (100 meters) or playing	
softball/baseball	
89. Dancing (social)	
90. Doing calisthenics or aerobic dancing (5	
minutes nonstop)	
91. Mowing the lawn (power mower, but not a	
riding mower)	
92. Walking 2 miles	
93. Walking 2 miles (nonstop)	
94. Climbing 50 steps (2 ¹ / ₂ floors)	
95. Shoveling, digging, or spading	
96. Shoveling, digging, or spading (5 minutes	
nonstop)	
97. Climbing 50 steps	
98. Walking 3 miles or golfing 18 holes without	
a riding cart	
99. Walking 3 miles (nonstop)	
100. Swimming 25 yards	
101. Swimming 25 yards (nonstop)	
<u> </u>	

Code: _____ Date: _____

	Still Doing This Activity	Have Stopped Doing This Activity	Never Did This Activity
102. Bicycling 1 mile			
103. Bicycling 2 miles			
104. Bicycling 1 mile (nonstop)			
105. Bicycling 2 miles (nonstop)			
106. Running or jogging ¹ / ₄ mile			
107. Running or jogging ¹ / ₂ mile			
108. Playing tennis or racquetball			
109. Playing basketball/soccer (game play)			
110. Running or jogging ¹ / ₄ mile (nonstop)			
111. Running or jogging ¹ / ₂ mile (nonstop)			
112. Running or jogging 1 mile			
113. Running or jogging 2 miles			
114. Running or jogging 3 miles			
115. Running or jogging 1 mile in 12 minutes or			
less			
116. Running or jogging 2 miles in 20 minutes or			
less			
117. Running or jogging 3 miles in 30 minutes or			
less			

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Code: _____ Date: _____

Age: _____ Gender: M or F

HAP SUMMARY SCORE GRID

Primary Score	Score	Percentile
MAS		
AAS		
Activity Age		

Fitness	Low	Fair	Average & Above
Classification			

Activity	Impaired	Moderately Active	Active
Classification			

Energy Analysis	Score	Percentile
EEP		
LEC		
Dyspnea Scale		

Appendix B

Tegner Activity Score

TEGNER ACTIVITY SCORE

Check the box beside the activity which best describes the level at which the patient participates. (Please check only one box).		
participates. (1	rease check only one box).	
Level 10	Competitive sports: Soccer – national & international elite	
Level 9	□ Competitive sports: Soccer – lower divisions, Ice hockey, Wrestling and/or Gymnastics	
Level 8	Competitive sports: Squash or badminton, Athletics (jumping, etc.) and/or Downhill skiing	
Level 7	 Competitive sports: Tennis, Athletics (running), Motocross/speedway, Handball and/or Basketball <u>OR</u> Recreational sports: Soccer, Ice hockey, Squash, Athletics (jumping, etc.) and/or Cross-country track 	
<u>Level 6</u>	□ Recreational sports: Tennis/badminton, Handball, Basketball, Downhill skiing and/or Jogging at least 5X per week	
Level 5	 Work: Heavy labor (e.g., Building, Forestry) <u>OR</u> Competitive sports: Cycling or Cross-country skiing <u>OR</u> Recreational sports: Jogging on uneven ground at least twice weekly 	
Level 4	❑ Work: Moderately heavy labor (e.g., Truck driving, Heavy domestic work) <u>OR</u> Recreational sports: Cycling, Cross-country skiing, and/or Jogging on even ground at least twice weekly	
Level 3	 Work: Light labor (e.g., Nursing) <u>OR</u> Competitive and Recreational Swimming <u>OR</u> Walking in forest possible 	
Level 2	Work: Light labor OR Walking on uneven ground possible but impossible to walk in forest	
Level 1	Sedentary work <u>OR</u> Walking on even ground possible	
Level 0	□ Sick leave or disability because of knee problems	

Appendix C

Modified Borg Scale

Modified Borg Scale				
0	Nothing at all			
0.5	Extremely weak (just noticeable)			
1	Very weak			
2	Weak (light)			
3	Moderate			
4	Somewhat strong			
5	Strong (heavy)			
6				
7	Very strong			
8	8			
9	9			
10	Extremely strong (almost max)			

Appendix D

Pearson Correlations Coefficients

Variable	Pearson Correlation	Squat AP	Squat ML	Stand AP	Stand ML
TPS tibial	CC	0.54	0.49	0.29	0.39
	p-Value	0.05	0.09	0.31	0.16
TPS plantar	CC	-0.11	-0.16	0.06	-0.08
Tr S plaittai	p-Value	0.72	0.59	0.83	0.78
Propriogontion FL V	CC	0.26	0.31	0.29	0.24
Proprioception FLX	p-Value	0.42	0.33	0.34	0.43
Proprioception EXT	CC	-0.15	-0.08	-0.20	-0.14
	p-Value	0.65	0.80	0.51	0.65
Knee PTBW FLX	CC	0.32	0.18	0.43	0.34
	p-Value	0.28	0.55	0.12	0.24
Knee PTBW EXT	CC	0.36	0.18	0.44	0.08
	p-Value	0.23	0.55	0.12	0.77
Ankle PTBW P-FLX	CC	0.64	0.60	0.70	0.45
AllKICFIDWF-FLA	p-Value	0.02	0.03	0.01	0.10
Ankle PTBW D-FLX	CC	0.52	0.49	0.49	0.39
AllKICFIDW D-FLA	p-Value	0.07	0.09	0.07	0.17
Knee PTBW FLX/EXT	CC	0.16	0.11	0.18	0.32
ratio	p-Value	0.60	0.72	0.54	0.26
Ankle PTBW	CC	-0.14	-0.13	-0.30	-0.20
D-FLX/P-FLX ratio	p-Value	0.66	0.67	0.29	0.48

Table 3. Pearson correlations between COPv and other variables under the squat and the stand conditions for AP and ML direction.

Variable	Pearson Correlation	Squat AP	Squat ML	Stand AP	Stand ML
TPS tibial	CC	0.10	0.03	0.25	0.32
	p-Value	0.74	0.91	0.38	0.26
TDC aleater	CC	-0.05	-0.12	0.20	0.24
TPS plantar	p-Value	0.86	0.70	0.49	0.40
Proprioception FLX	CC	0.11	-0.06	0.29	0.27
	p-Value	0.73	0.85	0.34	0.37
Drannia contian EVT	CC	-0.13	-0.09	0.01	-0.10
Proprioception EXT	p-Value	0.69	0.79	0.96	0.75
Knee PTBW FLX	CC	0.29	0.00	0.40	0.41
KIECFIDW FLA	p-Value	0.34	0.99	0.15	0.15
Knee PTBW EXT	CC	0.07	-0.35	0.30	-0.02
KIECTIDW LAI	p-Value	0.81	0.24	0.30	0.95
Ankle PTBW P-FLX	CC	0.63	0.31	0.53	0.38
Ankle PIBW P-FLA	p-Value	0.02	0.30	0.05	0.18
Ankle PTBW D-FLX	CC	0.42	0.09	0.44	0.24
	p-Value	0.16	0.76	0.11	0.42
Knee PTBW FLX/EXT	CC	0.36	0.39	0.20	0.43
ratio	p-Value	0.23	0.19	0.49	0.13
Ankle PTBW	CC	-0.30	-0.37	-0.09	-0.20
D-FLX/P-FLX ratio	p-Value	0.32	0.21	0.77	0.48

Table 4. Pearson correlations between RMSd and other variables under the squat and the stand conditions for AP and ML direction.

Variable	Pearson Correlation	Squat AP	Squat ML	Stand AP	Stand ML
TPS tibial	CC	0.05	0.01	0.24	0.40
1PS tiolai	p-Value	0.86	0.97	0.41	0.15
TDS plantar	CC	-0.20	-0.13	0.17	0.00
TPS plantar	p-Value	0.52	0.68	0.57	0.99
Proprioception FLX	CC	0.00	0.03	0.36	0.30
	p-Value	0.99	0.93	0.23	0.33
Proprioception EXT	CC	-0.25	-0.24	-0.18	-0.13
	p-Value	0.44	0.45	0.56	0.66
Knoo DTDW ELV	CC	0.20	0.14	0.52	0.43
Knee PTBW FLX	p-Value	0.50	0.64	0.06	0.12
Knee PTBW EXT	CC	-0.25	-0.48	0.53	0.11
KIECPIDWEAI	p-Value	0.41	0.10	0.05	0.72
Ankle PTBW P-FLX	CC	0.36	0.20	0.68	0.45
	p-Value	0.23	0.51	0.01	0.11
Ankle PTBW D-FLX	CC	0.07	-0.09	0.48	0.37
	p-Value	0.81	0.77	0.08	0.19
Knee PTBW FLX/EXT	CC	0.55	0.61	0.17	0.39
ratio	p-Value	0.05	0.03	0.55	0.17
Ankle PTBW	CC	-0.58	-0.61	-0.28	-0.20
D-FLX/P-FLX ratio	p-Value	0.04	0.03	0.33	0.49

Table 5. Pearson correlations between RMSv and other variables under the squat and the stand conditions for AP and ML direction.

Appendix E

Correlation Graphs

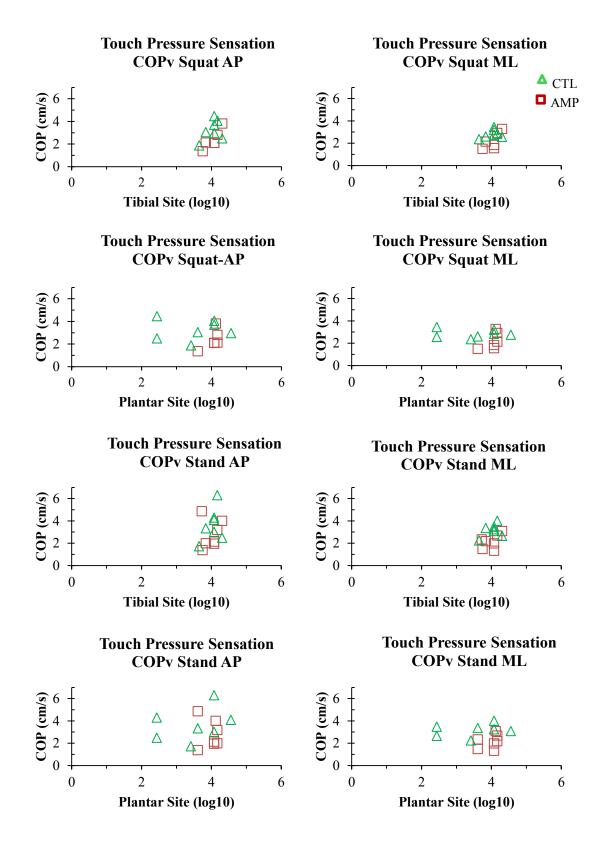


Figure 16. Correlation graphs for COP velocity and TPS.

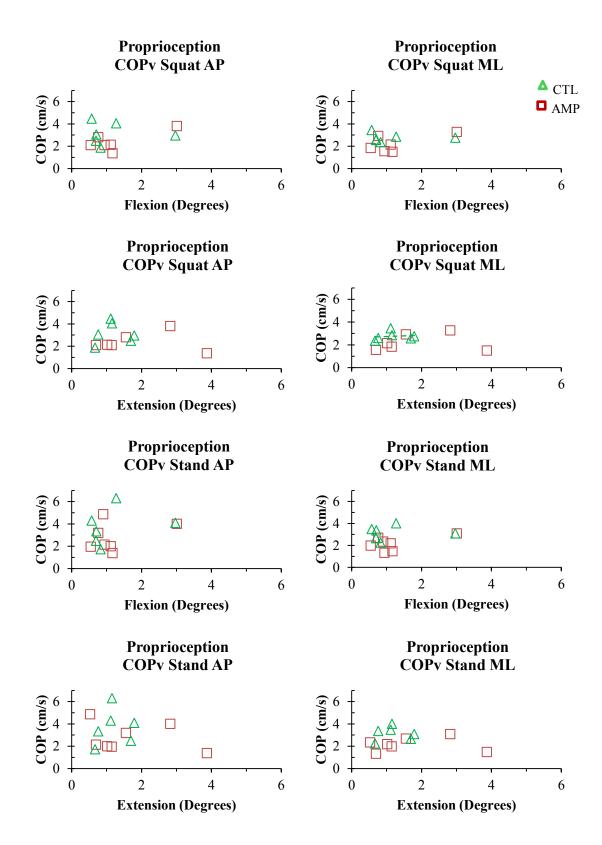


Figure 17. Correlation graphs for COP velocity and proprioception.

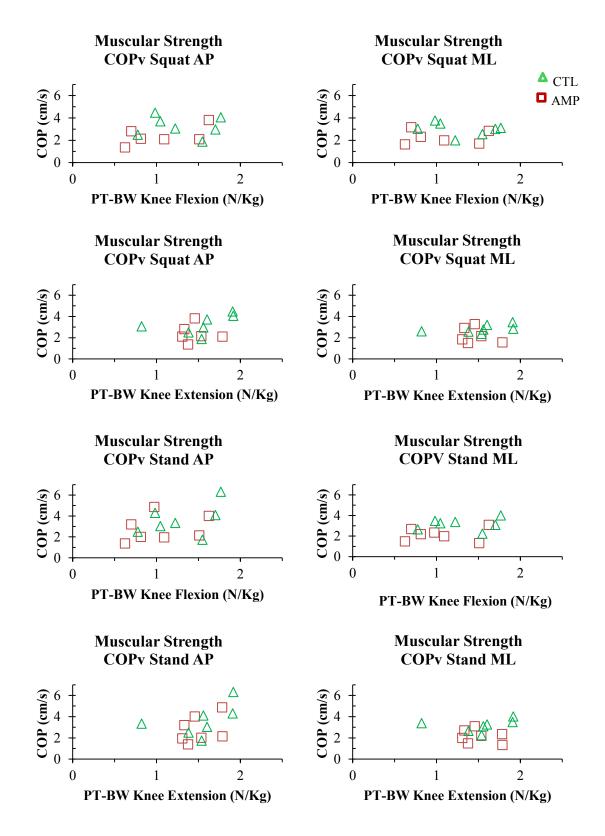


Figure 18. Correlation graphs for COP velocity and knee muscular strength.

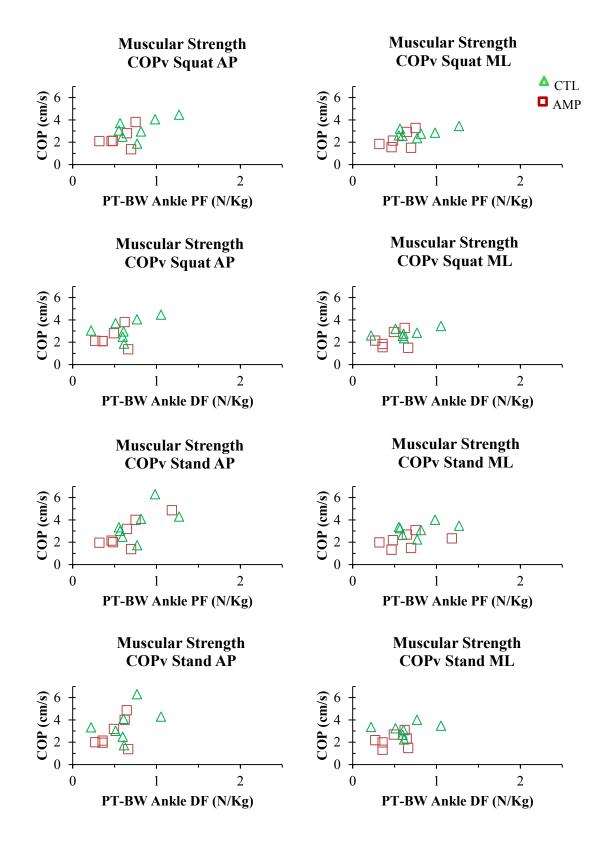


Figure 19. Correlation graphs for COP velocity and ankle muscular strength.

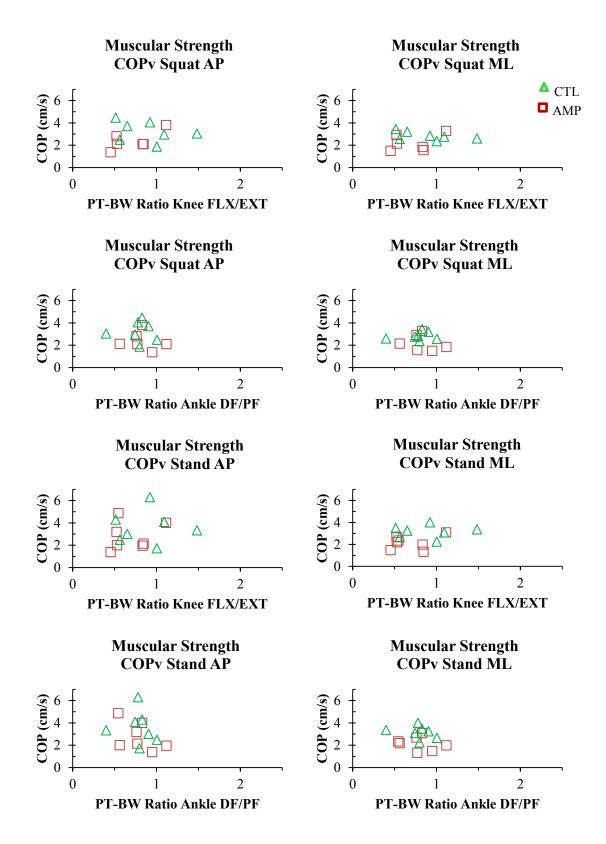


Figure 20. Correlation graphs for COP velocity and muscular strength ratios.

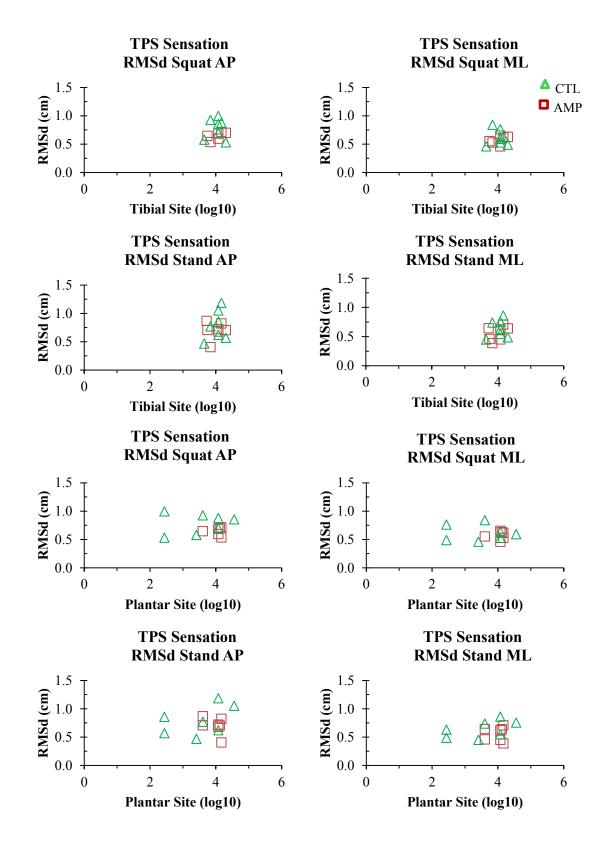


Figure 21. Correlation graphs for RMS displacement and TPS.

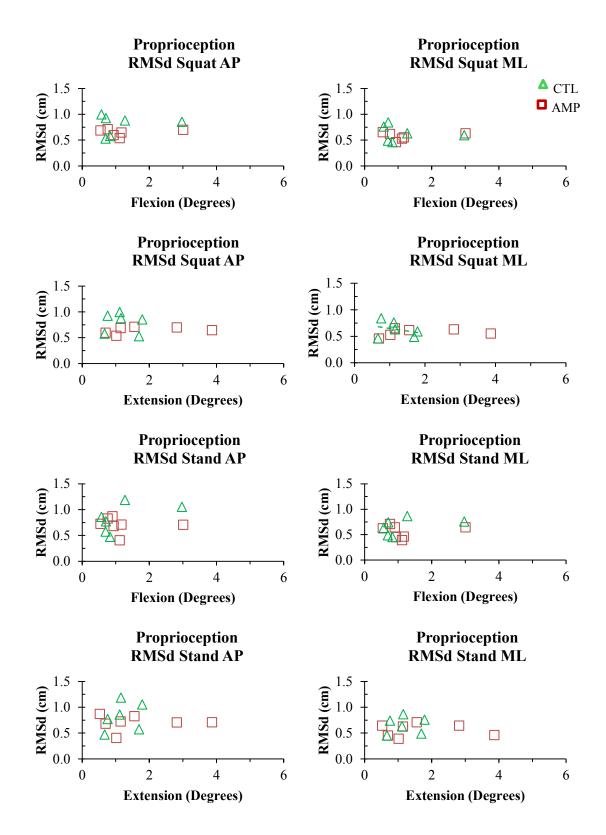


Figure 22. Correlation graphs for RMS displacement and proprioception.

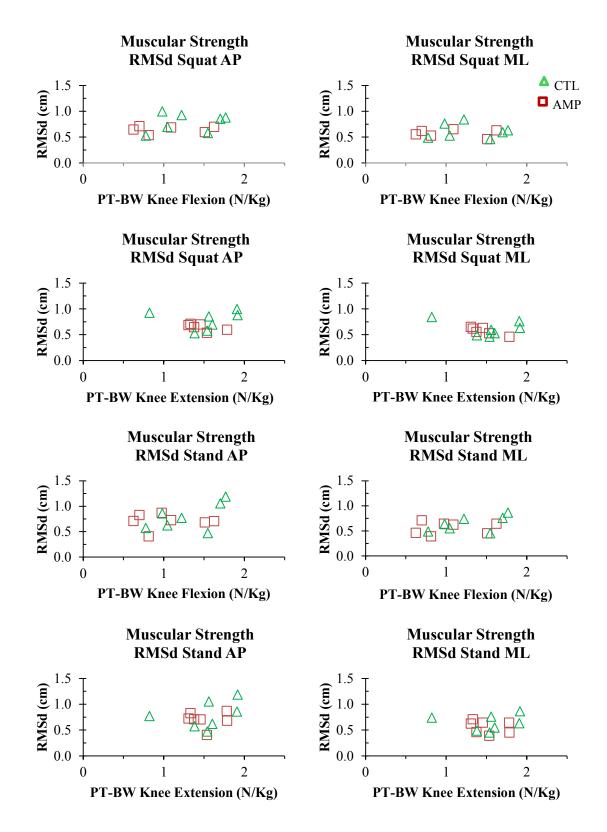


Figure 23. Correlation graphs for RMS displacement and knee muscular strength.

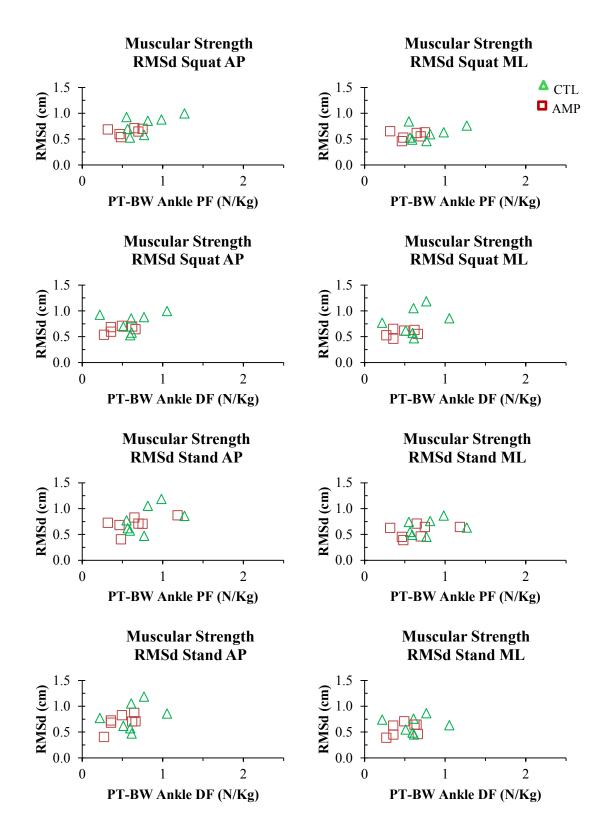


Figure 24. Correlation graphs for RMS displacement and ankle muscular strength.

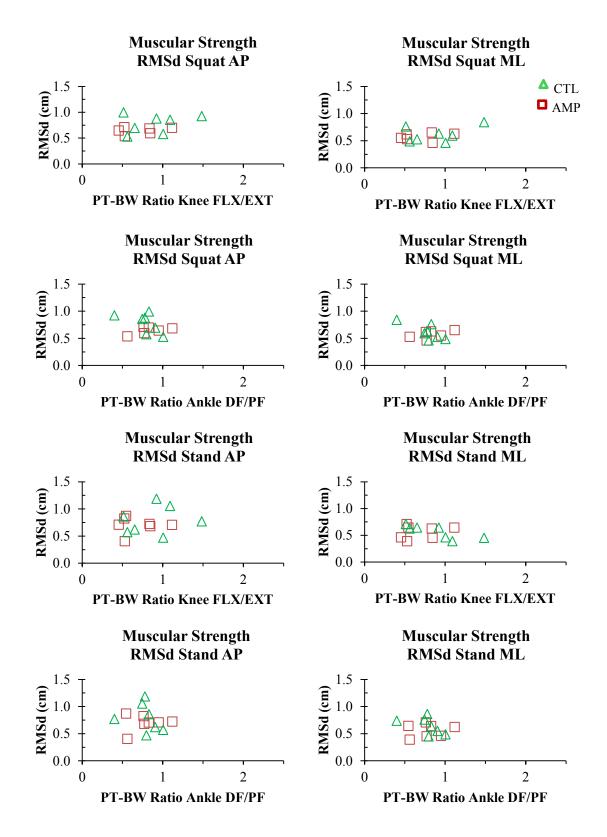


Figure 25. Correlation graphs for RMS displacement and muscular strength ratios.

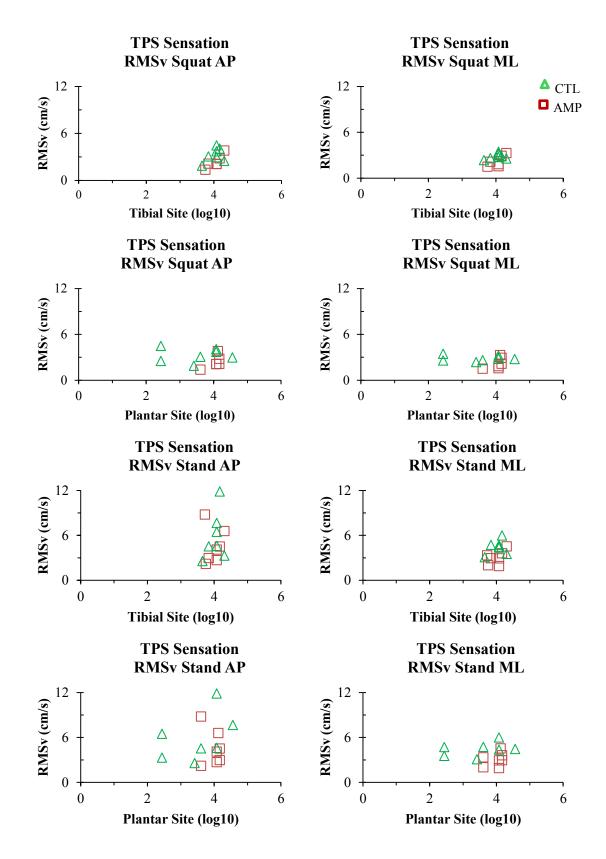


Figure 26. Correlation graphs for RMS velocity and TPS.

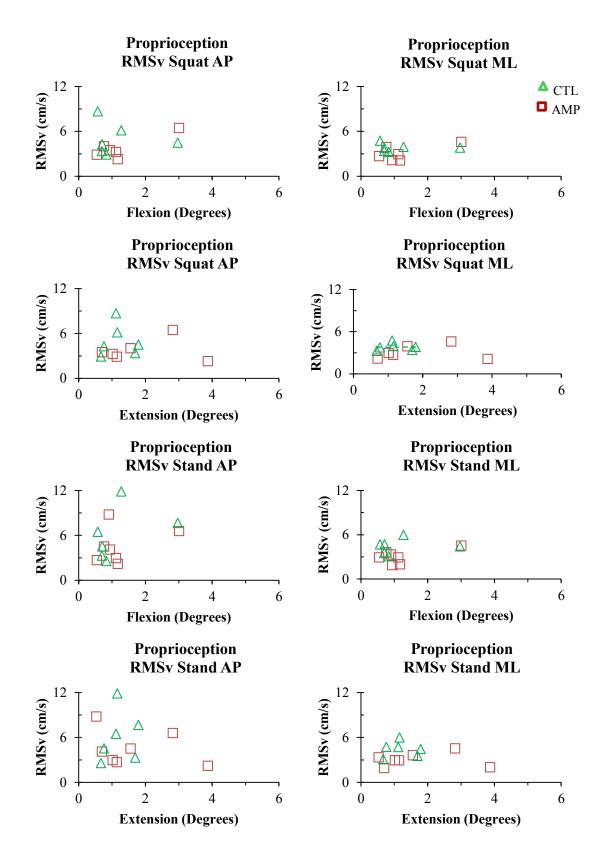


Figure 27. Correlation graphs for RMS velocity and proprioception.

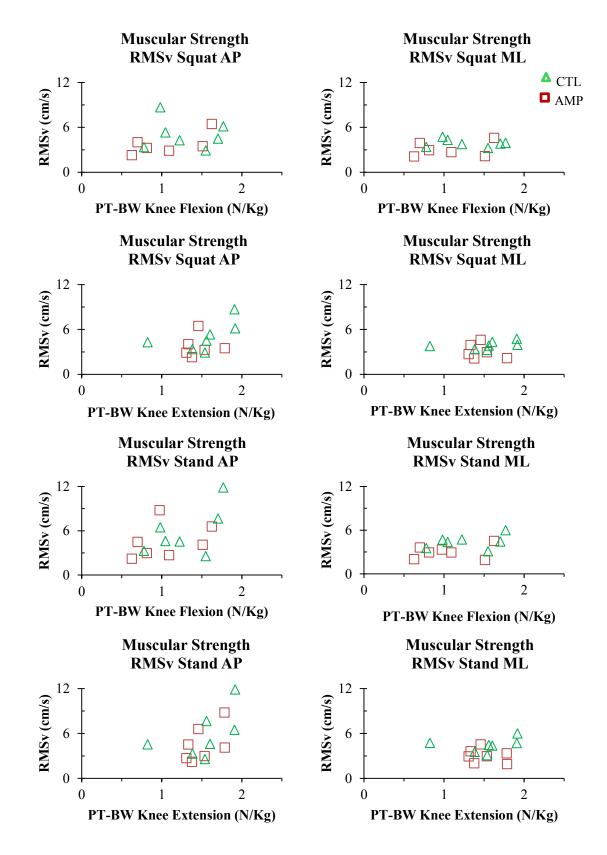


Figure 28. Correlation graphs for RMS velocity and knee muscular strength.

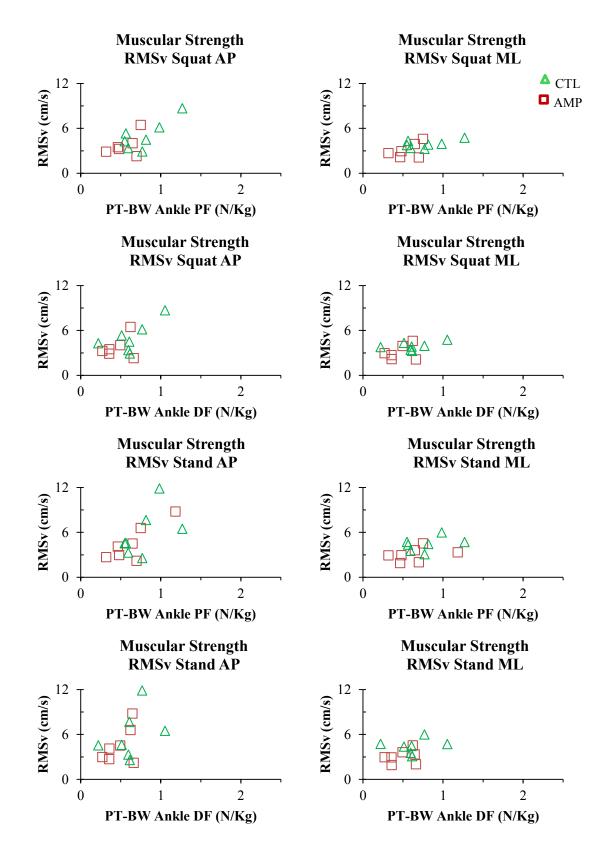


Figure 29. Correlation graphs for RMS velocity and ankle muscular strength.

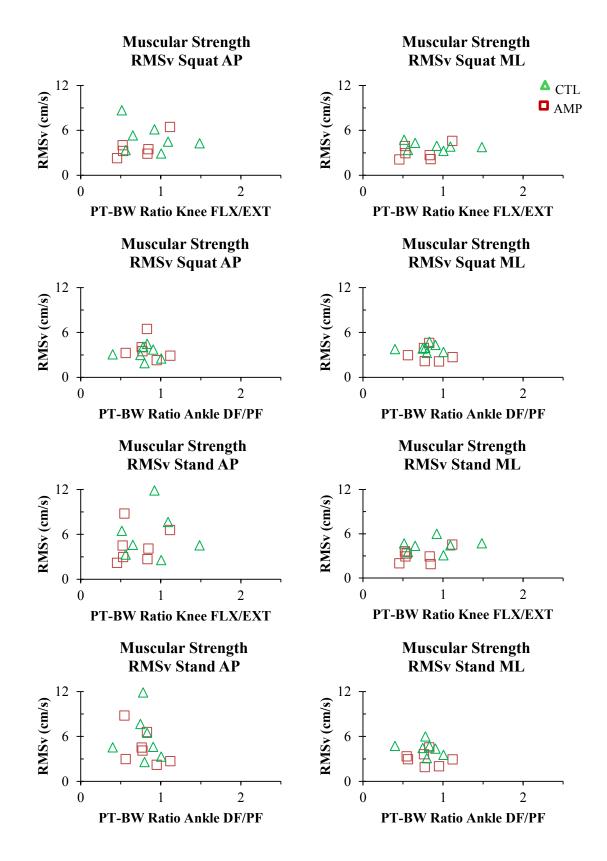


Figure 30. Correlation graphs for RMS velocity and muscular strength ratios.