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THE EFFECT OF HIP AND KNEE JOINT ANGLES ON THE MUSCLE-TENDON
UNIT DURING ELECTRICAL STIMULATION

DISSERTAÇÃO

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DEDICATION

I dedicate this dissertation to my heavenly Father, my mum and dad, Ms. Zélia and Mr. Hélio, and my loving daughter Milena.

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LIST OF ABBREVIATIONS

θ_p : Pennation angle

L_f : Fascicle length

TACD: Tendon-aponeurosis complex displacement

NMES: Neuromuscular electrical stimulation

RF: Rectus femoris

VL: Vastus lateralis

VM: Vastus medialis

VI: Vastus intermedius

QF: Quadriceps femoris

SK60: seated with knee at 60°

SK20: seated with knee at 20°

LK60: lying with knee at 60°

LK20: lying with knee at 20°

ANOVA: Analysis of Variance

ICC: Intra-class Correlation

RESUMO EXPANDIDO

Introdução: O comprimento muscular é um dos principais determinantes da capacidade de geração de força das fibras musculares¹. O torque varia expressivamente com a manipulação do ângulo articular. Alguns estudos relatam que o ângulo articular influencia o torque evocado e a eficiência da corrente (torque/amplitude da corrente) durante a estimulação elétrica neuromuscular (EENM)²⁻⁴. Tipicamente, o torque extensor do joelho é maior em ~60° de flexão em uma posição sentada. A arquitetura muscular (i.e., espessura, ângulo de penação [θ_p] e comprimento do fascículo [C_f]) e o deslocamento do complexo tendão-aponeurose (DCTA) têm sido estudados para explicar mecanismos da geração de torque durante a manipulação do comprimento muscular⁵⁻⁷. No entanto, até o momento, não foram estudados os efeitos dos ângulos do quadril e do joelho durante a EENM no comportamento da unidade miotendínea dos constituintes do quadríceps femoral (QF): o biarticular reto femoral (RF) e os monoarticulares vasto lateral (VL), vasto medial (VM) e vasto intermédio (VI)^{2,8}.

Até onde sabemos, existe apenas um estudo crônico que trata da EENM em diferentes ângulos articulares, o qual mostrou melhores resultados para a EENM realizada em um maior comprimento muscular⁹. No entanto, uma posição estendida do joelho foi previamente recomendada, apesar do pequeno tamanho de efeito relatado¹⁰. Vários estudos têm aplicado a EENM do QF escolhendo aleatoriamente o ângulo articular, como sentado ou deitado, com os joelhos estendidos ou flexionados em diferentes ângulos, ou mesmo sem descrição completa^{11,12}. Os clínicos devem estar cientes da configuração articular a fim de otimizar as respostas musculares aos programas de exercícios isométricos.

Objetivos: O objetivo principal deste estudo foi investigar o efeito dos ângulos do joelho (60° ou 20°) e do quadril (0° ou 85°) no torque evocado e na eficiência da corrente da EENM. Nós também avaliamos a arquitetura muscular (θ_p e C_f) em repouso e durante contração e o DCTA de cada componente do QF para investigar a contribuição deles.

Hipóteses: Nossa hipótese inicial foi que durante a EENM seria obtido maior torque extensor com o joelho a 60° na posição sentada, porém com amplitude de corrente proporcionalmente menor, portanto, melhor eficiência da corrente. Também levantamos a hipótese de que o θ_p seria menor e o C_f seria maior quando o joelho estivesse em 60° para todos os componentes do quadríceps (RF, VL, VM e VI), mas para o RF, o quadril em 0° diminuiria o θ_p e aumentaria o C_f ainda mais. Além disso, esperávamos que o TACD fosse mais pronunciado em posições com maior torque.

Métodos: Vinte homens hígidos com idade $24,0 \pm 4,6$ participaram de cinco sessões separadas por sete dias entre cada uma delas: uma sessão de familiarização e quatro sessões experimentais para testar quatro combinações diferentes de ângulos do quadril e do joelho durante EENM: quadril a 85° (sentado) e joelho a 60° (SJ60); quadril a 85° e joelho a 20° (SJ20); quadril a 0° (deitado) e joelho a 60° (DJ60); e quadril a 0° e joelho a 20° (DJ20). Oito contrações evocadas foram necessárias para realizar a ultrassonografia dos quatro componentes do QF (duas contrações para cada um). Os participantes foram questionados sobre condições de saúde e foi realizada a caracterização demográfica e antropométrica. Na familiarização foi verificado se os participantes toleravam amplitude de corrente suficiente para gerar um torque evocado $\geq 30\%$ da contração voluntária máxima (CVM).

Os valores de CVM, torque evocado, eficiência da corrente, amplitude da corrente, espessura muscular, θ_p , C_f , e DCTP foram reportados por meio de média \pm desvio

padrão. Para o θ_p e o C_f , as análises foram realizadas com os valores de repouso e em contração, bem como com a mudança relativa (%). A ANOVA unidirecional de medidas repetidas com fator “*posicionamento*” (DJ60, SJ60, DJ20, SJ20) foi aplicada para verificar diferenças entre posições para a CVM, o torque evocado, a eficiência da corrente, a amplitude da corrente e o DCTA. A ANOVA bidirecional (“*posicionamento*” [4 níveis: DJ60, SJ60, DJ20, and SJ20] *versus* “*intensidade*” [2 níveis: repouso and contração evocada]) com medidas repetidas no fator *posicionamento* foi aplicada para verificar diferença entre posições para o θ_p e o C_f . Quando uma diferença significativa foi detectada, o teste *post-hoc* de Tukey foi aplicado. O limiar de significância foi estabelecido em $P < 0,05$. Todas as análises foram realizadas usando o STATISTICA 23.0 (STATSOFT Inc., Tulsa, Oklahoma, EUA) e o software GRAPHPAD PRISM 8.3.0 (San Diego, CA, EUA) foi utilizado para o design gráfico.

Resultados: O torque evocado e a eficiência da corrente foram maiores para o DJ60 e o SJ60 em comparação com o DJ20 e o SJ20 ($p < 0,001$). O QF (média de todos os músculos), o VL e o VM apresentaram menor θ_p e maior C_f em DJ60 e SJ60, enquanto o reto femoral demonstrou influência do ângulo do quadril, uma vez que em DJ60 houve menor θ_p e maior C_f do que em todas as outras posições ($p < 0,001 - 0,05$). O vasto intermédio se comportou semelhante aos demais vastos ($p < 0,001$), exceto pela falta de diferença no θ_p entre SK60 em comparação com DJ20 e SJ20 ($p = 0,25$ e $0,30$, respectivamente). A TACD foi maior para o SJ60 em comparação com o DJ60 ($p < 0,001$), apesar do mesmo torque.

Discussão: Os principais achados deste estudo foram: 1) o torque evocado extensor do joelho e a eficiência da corrente foram maiores em 60° de flexão do joelho comparado com em 20°, sem diferença de acordo com o ângulo do quadril; 2) O QF teve menor θ_p and greater C_f em 60° de flexão de joelho. 3) O DCTA foi menor em DJ60 comparado com SJ60 apesar do mesmo torque. Estes novos achados são importantes para ajudar fisioterapeutas e treinadores físicos a desenvolverem estratégias mais efetivas quando aplicarem EENM. Nossos resultados estão de acordo com relatos anteriores que encontraram maior torque evocado a 60° de flexão do joelho em comparação com posições mais estendidas^{3,4}. Um ângulo articular escolhido com cautela permite atingir o torque alvo com menor amplitude de corrente e, com isso, com menos desconforto sensorial.

Apenas dois estudos avaliaram a arquitetura de todos os constituintes do QF *in vivo*, mas eles não aplicaram ENM ou avaliaram diferentes ângulos articulares^{8,13}. Nosso principal achado foi que o QF demonstrou um padrão em que as posições com o joelho a 60° apresentavam θ_p menor e maior C_f quando comparadas às posições com o joelho a 20°. Assim, sugere-se que em DJ60 e o SJ60 o QF foi colocado em uma melhor configuração para geração de torque, ou seja, melhor aproveitamento da força muscular e comprimento ideal do sarcômero^{1,14}. O QF apresentou um DCTA menor em DJ60 comparado com SJ60, apesar do mesmo torque evocado, indicando que o aumento da tensão passiva em DJ60 limitou o alongamento tendíneo durante a contração⁶. O aumento da tensão do complexo tendão-aponeurose em condições de alongamento permite contrações mais fortes com menor esforço devido à melhor transmissão de força do músculo para o osso⁷.

Conclusão: A EENM gera um torque maior a 60° de flexão do joelho, comparado a 20°, independentemente do ângulo do quadril. A arquitetura de cada constituinte do quadríceps demonstrou um comportamento único de acordo com o ângulo do quadril e do joelho, mas predominaram um menor θ_p e um maior C_f nas posições com maior torque (SJ60 e DJ60). Uma posição mais alongada enrijece o complexo tendão-aponeurose, como demonstrado por um DCTA menor em DJ60 em comparação com SJ60, o que provavelmente contribuiu

para uma transmissão otimizada da força e um torque ligeiramente mais alto para o DJ60. Clínicos devem preferencialmente usar NMES em DJ60 ou DJ60 para fins de fortalecimento.

Palavras-chave: Estimulação elétrica neuromuscular; Relação ângulo-torque; Arquitetura muscular; Complexo tendão-aponeurose; Quadríceps.

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TITLE PAGE**THE EFFECT OF HIP AND KNEE JOINT ANGLES ON THE MUSCLE-TENDON UNIT DURING NEUROMUSCULAR ELECTRICAL STIMULATION**

Running title: Joint angle and electrical stimulation

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ABSTRACT

Neuromuscular electrical stimulation (NMES) is recommended to counteract muscle atrophy and for strengthening. However, the influence of hip and knee angles during quadriceps femoris (QF) NMES is poorly investigated. We aimed to investigate the effect of knee and hip angle on NMES-evoked torque and current efficiency. We secondarily assessed the QF architecture at rest and during contraction, and the tendon-aponeurosis complex displacement (TACD). Twenty men aged 24.0 ± 4.6 years received NMES in four positions: hip at 85° (seated) and knee at 60° (SK60); hip at 85° and knee at 20° (SK20); hip at 0° (lying) and knee at 60° (LK60); and hip at 0° and knee at 20° (LK20). NMES-evoked torque and current efficiency (evoked torque/current amplitude) were recorded. Ultrasonography of the QF was performed to measure pennation angle (θ_p), fascicle length (L_f), and TACD. Evoked torque and current efficiency were greater for LK60 and SK60 compared to LK20 and SK20 ($p < 0.01$). The QF (all muscles), vastus lateralis, and medialis showed lower θ_p and higher L_f at LK60 and SK60, while rectus femoris demonstrated influence of hip angle, since in LK60 there was lower θ_p and higher L_f than in all other positions ($p < 0.05$). The vastus intermedius was similar to the other vasti, except for a lack of difference in θ_p between SK60 compared to LK20 and SK20. TACD was greater for SK60 compared to LK60 ($p < 0.001$) despite the same torque. These findings suggest that clinicians should apply NMES preferably at 60° of knee flexion.

Keywords: Neuromuscular Electrical Stimulation; Angle-torque relationship; Muscle architecture; Tendon-aponeurosis complex; Quadriceps.

INTRODUCTION

Muscle length is a major determinant of force generation capacity of muscle fibers^{1,2}. Maximal voluntary contraction (MVC) varies expressively with the manipulation of joint-angle due to changes in muscle length and moment arm³⁻⁵. In addition, some studies have reported that joint angle has an effect on evoked torque induced by neuromuscular electrical stimulation (NMES)⁶⁻⁹. Typically, knee extensor torque is greater at ~60° of flexion (0° being fully extended) in a seated position^{5,8,10,11}.

Muscle architecture (i.e., muscle thickness, pennation angle [θ_p], and fascicle length [L_f]) and the tendon-aponeurosis complex displacement (TACD) have been studied to explain mechanisms of torque generation during muscle length manipulation¹²⁻¹⁵. Muscle architecture is the arrangement of muscle fibers relative to the force axis, a strong determinant of muscle function^{16,17}. A steeper θ_p and shorter L_f are found at shortened compared to more elongated positions¹². Moreover, an increase in θ_p is expected with a concomitant reduction in L_f during isometric contraction^{12,18,19}. The TACD indicates the elongation from the deep aponeurosis to the distal free tendon in response to transmission of muscle force to bones¹³. Muscle and tendon-aponeurosis complex behavior have been assessed during NMES^{20,21}. However, to date, the effects of hip and knee angles during NMES have not been studied on the muscle-tendon behavior of the constituents of the quadriceps femoris (QF): the biarticular rectus femoris (RF), and the monoarticular vastus lateralis (VL), medialis (VM), and intermedius (VI)^{7,22}.

NMES is commonly applied to counteract the harmful effects of atrophy and for strengthening purposes^{23,24}. Although QF is the most frequently stimulated muscle²³, the influence of hip and knee angles on evoked torque and current efficiency (torque/current amplitude) during NMES is not well established. Scott et al.⁸ compared three knee angles (30°, 60°, and 90°) and found higher absolute evoked torque at 60°, which suggests a similar pattern to that observed during MVC⁵. Lastly, Maffiuleti et al.⁶ and Bampouras et al.⁷

showed greater evoked torque of knee extension when lying compared to a seated position while maintaining the knee at 90°. These studies only evaluated the knee or the hip separately and they did not assess the current efficiency, a parameter that should be optimized to attain higher torque with lower discomfort and could also be dependent on muscle length ^{25,26}.

JUSTIFICATIVE

Notably, the only clinical trial available showed better results for NMES performed at longer muscle length ²⁷. However, a knee extended position (shortened QF) was previously recommended in situations where patients are unable to tolerate NMES with flexed knees or if a dynamometer is unavailable, despite the reported small effect size for QF strengthening compared to previous protocols with flexed knee ²⁸. Chronic studies have applied NMES with a random joint angle choice, such as seated or lying, with knees extended or flexed at different angles, or even with an incomplete description of lower limb position ²⁹⁻³¹. This approach is questionable in view of the pivotal physiological principles of the force-length and angle-torque relationships ¹. Physical therapist and athletic trainers should be aware of joint configuration in order to optimize muscular responses to isometric-based exercise programs.

OBJECTIVES

Therefore, the primary aim of this study was to investigate the effect of knee (60° or 20°) and hip (0° or 85°) angle on NMES-evoked torque and current efficiency. We secondarily assessed the muscle architecture (θ_p and L_f) at rest and during contraction, and the TACD of each QF constituent during NMES to investigate their contribution to torque production.

HYPOTHESIS

Our primary hypothesis was that during NMES greater knee extensor torque would be obtained in the seated position with the knee at 60° of flexion with a proportionally lower current amplitude, and, thus, better current efficiency. We also hypothesized that at rest and during the plateau of the evoked contraction, θ_p would be lower and L_f would be greater when the knee is at 60° for all quadriceps components (RF, VL, VM, and VI), although for RF, the hip at 0° would decrease the θ_p and increase the L_f even more. Moreover, we expected that the TACD would be more pronounced in positions with greater torque for at least one of the QF constituents (RF, VL, VM, and VI).

MATERIAL AND METHODS

Participants

Twenty men (mean \pm SD age: 24.0 \pm 4.6 years, body mass: 77.0 \pm 9.3 kg, height: 177.6 \pm 6.3 cm) with no known neuromuscular disorders and not engaged in systematic lower limb strengthening or sport competitions in the previous 6 months volunteered to participate. Sample size ($n = 20$) was determined a priori using G* POWER (v 3.13; University of Trier, Germany) based on evoked torque found at three knee angles (30°, 60°, and 90°) by Scott et al. (2019). The level of significance was set at $p = 0.05$, a power ($1 - \beta$) = 0.80, and an effect size = 0.75. Subjects were informed about the purposes, benefits, and risks before enrollment and all agreed to participate and signed the consent form. Approval was obtained (protocol number 94388718.8.0000.8093; Appendix III) from the Research Ethics Committee of the University of Brasília/Faculty of Ceilândia in accordance with the Helsinki Declaration of 1975.

Experimental design

Participants took part in five sessions at least 7 days apart, during the day. The first session was for familiarization. Participants were asked about health conditions and

demographic and anthropometric characterization was performed. Motor point localization of the VL and VM, and a pre-session preparation: 2 MVC and 2 evoked contractions in each position were carried out, in order to verify that participants tolerated enough current amplitude to generate an evoked torque $\geq 40\%$ of their MVC. Each experimental session lasted ~ 3 hrs during which one of four different combinations of lower limb position was randomly tested during quadriceps NMES: hip at 85° (seated) and knee at 60° (SK60°); hip at 85° and knee at 20° (SK20°); hip at 0° (lying) and knee at 60° (LK60°); and hip at 0° and knee at 20° (LK20°). We instructed subjects not to ingest alcohol or stimulants (e.g., caffeine, chocolate, and dietary performance supplements), respectively, for 24 and 6 hours before each visit, and not to participate in strenuous activities in the prior 48 hours. The primary outcomes were the evoked torque (absolute and normalized by MVC) and the current efficiency (torque/current amplitude). Secondary outcomes were the muscle architecture at rest and during NMES (θ_p and L_f) and the TACD of the four QF constituents (Fig. 1).

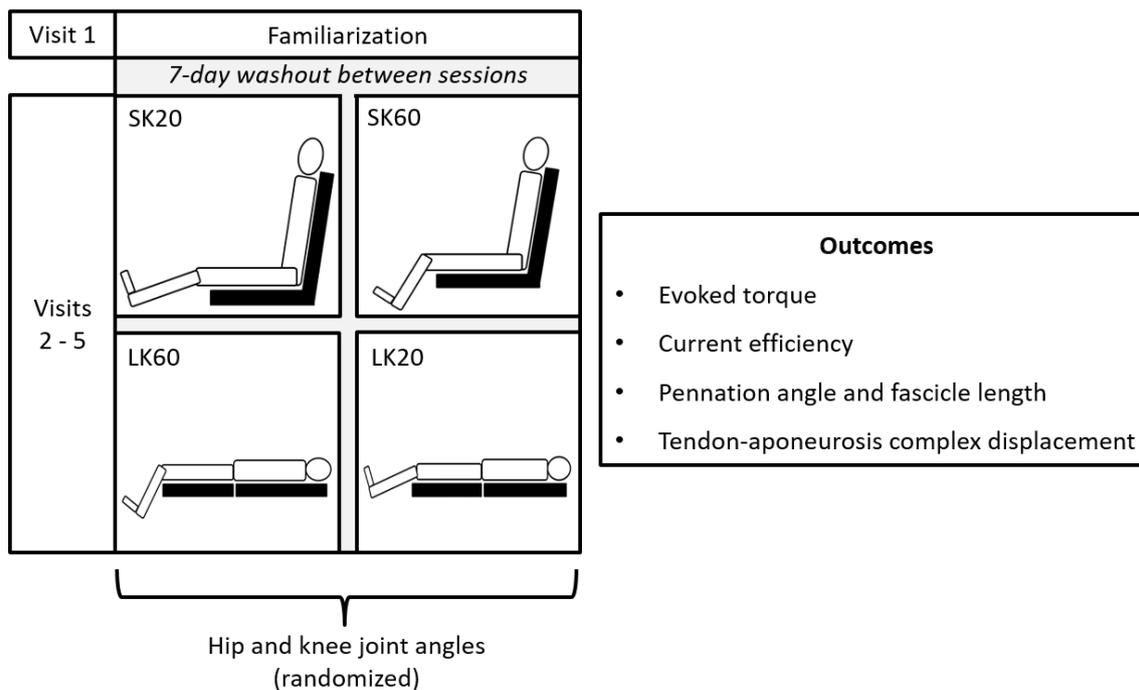


Figure 1 – Experimental design: Participants took part in five sessions at least 7 days apart; a familiarization and four experimental sessions, to test four different combinations of hip and knee joint angles randomly during NMES-evoked contraction of knee extension. 8 evoked contractions were necessary to perform ultrasonography of the four quadriceps constituents (2 contractions for each one). The primary outcomes were the evoked torque (absolute and normalized by maximal voluntary contraction) and the current efficiency (torque/current amplitude). Secondary outcomes were the muscle architecture at rest and during NMES (pennation angle and fascicle length) and the tendon-aponeurosis complex displacement of the four quadriceps constituents. Legend: NMES: neuromuscular electrical stimulation; LK60: lying with knee at 60°; SK60: seated with knee at 60°; LK20: lying with knee at 20°; SK20: seated with knee at 20°.

Torque

The positions (SK60, SK20, LK60, and LK20) were tested while participants were seated on the chair of a dynamometer (SYSTEM 4; BIODEX MEDICAL SYSTEMS, Shirley, New York) to measure extensor torque of the right knee. The equipment axis was visually aligned with the knee flexion-extension axis, i.e., the lateral epicondyle of the femur. The knee and hip angles were adjusted with a goniometer and the lever arm of the dynamometer transducer was firmly attached 2-3 cm above the lateral malleolus with a strap. Subjects were firmly stabilized to the chair with belts across the chest and pelvic girdle to minimize body movements. Resting torque was recorded in each position and used for subsequent gravity correction due to the weight of the limb or other force, such as the passive tension of the structures that cross the knee⁷. A warm up of 6 submaximal isometric contractions of 5 s and a rest interval of 10 s was performed at the beginning of each session at percentages of the maximum perceived effort (50% [x3], 75% [x2], and 90% [x1]). Prior to the recording of the evoked torque during NMES, participants performed 2 MVCs of the right knee extensors separated by a 2min rest. During each MVC, participants were encouraged verbally to perform maximally and received visual feedback of the torque produced.

NMES

A neuromuscular electrical stimulator (NEURODYN 2.0, IBRAMED, SP, Brazil) was connected to two isolated cables and a pair of self-adhesive electrodes of 25cm² applied over the motor points of the VL and VM as described by Botter et al.³². NMES was applied using a pulsed current with a frequency = 100 Hz, phase duration = 400 μ s, rise time = 3 s, on time = 4 s, decay time = 3 s, and off time = 1 min. Current amplitude was rapidly increased to reach the maximum tolerable amplitude achieved in the familiarization. Eight contractions were performed to allow all ultrasonographic recordings described in the

section below. Subjects were instructed to fully relax during the NMES to enable measurement only of evoked torque. The mean evoked torque of the first three evoked contractions was recorded and presented as absolute (N.m) and relative (a percentage of MVC) values. The mean current amplitude (mA) was used for calculation of current efficiency, as the ratio between absolute evoked torque and current amplitude ²⁵.

Muscle architecture

The θ_p and L_f were obtained using an ultrasound device (M-TURBO®, SONOSITE, Bothell, WA, USA) in B mode with a linear transducer of 7.5 MHz and width of 40 mm. Visualization depth was set at 6 cm. A custom-made device held the transducer, preventing it from moving on the thigh surface. For each QF constituent, two video recordings were obtained and the mean of the calculations was considered. The transducer was positioned in the longitudinal plane of the muscle in parallel with the direction of the fascicles. Proper alignment was achieved when multiple fascicles were traced without interruption, for this, the transducer was allowed to tilt in relation to the skin to adapt to the three-dimensional configuration of the fascicles. The lateral compartments of the bipennated RF and VL, VM, and VI were evaluated, respectively, at the percentages 50%, 60%, 75%, and 80% of the thigh length, from proximal to distal, considering the distance between the medial aspect of the anterior superior iliac spine and the superior border of the patella, as adapted from Blazevich et al. ²². For the VI, although it could be seen on the same window as the RF or VL during contraction, VI visualization could be partially lost, in which case it was recorded more distally. Confirmation of each muscle location, when necessary, was performed with the probe in transverse plane, allowing visualization of the transition from one muscle to another. The RF and VI were visualized on the anterior aspect of the thigh, while the VL and VM were visualized, respectively, on the lateral and medial aspects. Video files recorded during evoked contractions were stored on the device itself and

transferred to a computer for processing in public domain software (ImageJ software v. 1.46; National Institutes of Health, Bethesda, Maryland). Muscle thickness was considered as the distance between the superficial and deep aponeurosis of each muscle. The θ_p was calculated considering the angle between the deep aponeurosis and the fascicles. The L_f was directly measured wherever possible, or in cases where the fascicles extended beyond the visible field of view, linear extrapolation was applied, as well as an equation with a reported error of 2-7%³³. Figure 2 shows an example of measurement at rest and during evoked contraction of muscle architecture and the TACD.

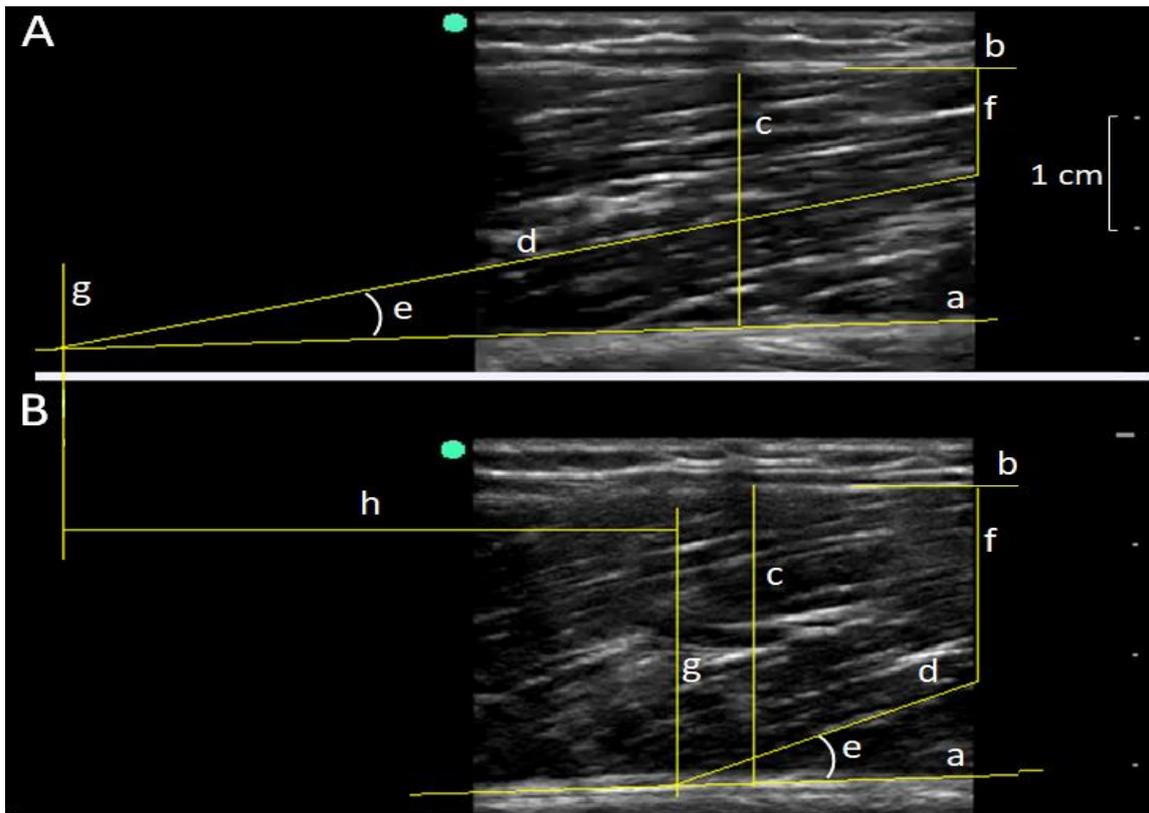


Figure 2 - Muscle ultrasonography: An ultrasound analysis of the vastus medialis at rest (A) and during NMES-evoked contraction (B). (a) deep aponeurosis; (b) superficial aponeurosis; (c) muscle thickness; (d) fascicle with extrapolation or not; (e) pennation angle; (f) distance between end of fascicle visualization and superficial aponeurosis; (g) lines indicating the cross point between fascicle and deep aponeurosis; (h) tendon-aponeurosis complex displacement. Legend: NMES: neuromuscular electrical stimulation.

Tendon-aponeurosis complex displacement

The longitudinal TACD was assessed, in each QF constituent, using the same files obtained for muscle architecture. A vertical line was traced over the cross point between the fascicle and the deep aponeurosis at rest and the displacement in centimeters was measured considering the final position of the cross point during the evoked torque. A hypoechoic mark was used to correct any unavoided movement of the transducer. Moreover, to correct overestimation of the TACD due to any knee joint angular rotation, we performed ultrasonographic recordings of all QF constituents during passive motion of the knee from 60° to 0° in both seated and lying positions, and placed a digital goniometer (GN360; Miotec®, Porto Alegre/RS, Brazil) (GN360) on the lateral aspect of the knee during NMES. Only the values corrected for angular rotation were reported¹³. A single examiner conducted all measurements.

Statistical Analysis

Values of MVC, evoked torque, current efficiency, current amplitude, muscle thickness, θ_p , L_f , and TACD are reported as mean \pm standard deviation (SD). For θ_p and L_f , analyses were performed with the rest and contracting (during NMES) values, as well as their relative (%) change (rest to contraction). Although individual QF constituents have previously been referred to as “quadriceps” as a form of generalization (Aagard et al., 2001), for better understanding, in the present study, QF constituents are referred to as RF, VL, VM, and VI, and the mean of their values is referred to as QF. Repeated measures one-way ANOVA with a within-subject of “*positioning*” (LK60, SK60, LK20, and SK20) was applied to verify differences between positions for MVC, evoked torque, current efficiency, current amplitude, and TACD. A two-way ANOVA (“*positioning*” [4 levels: LK60, SK60, LK20, and SK20] by “*intensity*” [2 levels: rest and evoked contraction]) with repeated measures on the *positioning* factor was applied to verify differences between positions for

θ_p and L_f . When a significant difference was detected, a Tukey *post-hoc* test was applied to identify the differences. Effect sizes and statistical power were calculated. Effect size was determined using partial eta squared (η_p^2): small ($\eta_p^2 = 0.01$), medium ($\eta_p^2 = 0.06$), and large ($\eta_p^2 = 0.14$) effects. Intra-class correlation (ICC) of evoked torque (all 8 contractions performed in each position), muscle architecture, and TACD (2 repeated analyses of 40 random assessment in any position, 10 for each QF constituent), was classified as: insufficient < 0.8 , moderate = $0.8 - 0.9$, and high > 0.9 . The significance threshold was set at $P < 0.05$ for all procedures. All analyses were performed using STATISTICA 23.0 (STATSOFT Inc., Tulsa, Oklahoma, USA) and the software GRAPHPAD PRISM 8.3.0 (San Diego, CA, USA) was used for graphics design.

RESULTS

MVC, Evoked torque, current efficiency, and current amplitude

Table 1 shows the mean \pm SD as well as statistical significances on *post-hoc* analysis for MVC, absolute and normalized evoked torque, current efficiency, and current amplitude. A significant main effect was found for MVC ($F_{3, 57} = 102.97$, $p < 0.001$, η_p^2 : 0.84, power: 1.0), absolute evoked torque ($F_{3, 57} = 30.42$, $p < 0.001$, η_p^2 : 0.61, power: 1.0), and current efficiency ($F_{3, 57} = 18.15$, $p < 0.001$, η_p^2 : 0.48, power: 0.99). In the *post-hoc* analysis, LK60 and SK60 showed greater MVC ($p < 0.001$), absolute evoked torque ($p < 0.001$), and current efficiency ($p < 0.01$) than LK20 and SK20, with no differences between positions with the same knee angle, respectively: SK60 vs. LK60: $p = 0.71$, 0.99, and 0.062; SK20 vs. LK20: $p = 0.99$, 0.98, and 0.55. For normalized evoked torque, there was no main effect ($F_{3, 57} = 1.52$, $p = 0.21$, η_p^2 : 0.07, power: 0.38). For current amplitude, a significant main effect was found ($F_{3, 57} = 12.74$, $p < 0.001$, η_p^2 : 0.40, power: 0.99). A lower current amplitude was found at LK20 compared to SK60 ($p < 0.001$), SK20 ($p < 0.05$), and LK60 ($p < 0.001$). LK60 did not differ from SK60 and SK20 ($P = 0.45$), but SK60 was greater

than SK20 ($P < 0.05$). ICC was high for evoked torque at SK60 (0.92), SK20 (0.97) and LK20 (0.97), and moderate (0.89) at LK60.

Table 1. Maximal voluntary contraction, absolute evoked torque, normalized evoked torque, current efficiency, and intensity at different hip and knee angles

	LK60	SK60	LK20	SK20
MVC (N.m)	216.45 ± 43.10	206.53 ± 47.70	96.96 ± 17.47 ^{a,b}	95.63 ± 24.22 ^{a,b}
Absolute evoked torque (N.m)	172,53 ± 62,50	155,58 ± 59,49	71,38 ± 23,25 ^{a,b}	67.01 ± 26.0 ^{a,b}
Normalized evoked torque (%)	80.81 ± 25.40	74.69 ± 22.73	72.53 ± 17.38	69.47 ± 19.07
Current efficiency (N.m/mA)	2.16 ± 0.65	1.81 ± 0.64	1.15 ± 0.41 ^{a,b}	0.92 ± 0.33 ^{a,b}
Intensity (mA)	81.46 ± 22.00	87.25 ± 25.21	64.16 ± 17.49 ^{a,b}	75.65 ± 28.35 ^{a,c}

Values are reported as mean ±SD. Legend: LK60°: lying with knee at 60°; SK60°: seated with knee at 60°; LK20°: lying with knee at 20°; SK20°: seated with knee at 20°; MVC: Maximal voluntary contraction. ^aP< 0.05 vs. LK60; ^bP< 0.05 vs. SK60; ^cP< 0.05 vs. LK20

Table 2. Muscle thickness, fascicle length, and pennation angle of rectus femoris, vastus lateralis, vastus medialis, and vastus intermedius at rest and during neuromuscular electrical stimulation in four lower limb positions

	LK60		SK60		LK20		SK20	
	Rest	NMES	Rest	NMES	Rest	NMES	Rest	NMES
Rectus femoris								
MT	2.64 ± 0.39 (14.14 ± 3.03%)	3.0 ± 0.49	2.46 ± 0.36 (3.38 ± 3.10%)	2.91 ± 0.48	2.47 ± 0.41 (31.41 ± 6.96%)	3.19 ± 0.63	2.46 ± 0.42 (26.32 ± 3.74%)	3.10 ± 0.61
θ_p	11.21 ± 2.91 (27.23 ± 30.79)	14.0 ± 4.15	13.20 ± 2.98 (43.36 ± 33.51%)	18.58 ± 4.92	13.64 ± 3.22 (39.87 ± 27.13%)	18.64 ± 4.16	13.78 ± 3.32 (55.34 ± 45.51%)	21.23 ± 7.62
L_f	15.09 ± 4.47 (-5.71 ± 27.48)	13.67 ± 4.26	11.79 ± 3.09 (-14.29 ± 20.79%)	9.77 ± 2.27	11.87 ± 3.15 (-8.52 ± 27.28%)	10.54 ± 2.86	10.75 ± 3.60 (-9.26 ± 29.01%)	9.44 ± 3.51
Vastus lateralis								
MT	2.42 ± 0.26 (9.82 ± 3.46%)	2.66 ± 0.48	2.50 ± 0.29 (9.07 ± 2.44%)	2.73 ± 0.37	2.22 ± 0.32 (21.53 ± 2.94%)	2.7 ± 0.47	2.31 ± 0.26 (16.63 ± 3.92%)	2.67 ± 0.31
θ_p	10.99 ± 1.98 (39.85 ± 38.08)	15.01 ± 3.54	11.81 ± 2.05 (28.56 ± 29.61%)	15.15 ± 4.39	13.22 ± 2.22 (40.90 ± 42.44%)	18.43 ± 5.14	13.83 ± 2.73 (51.49 ± 40.70%)	20.37 ± 5.03
L_f	12.89 ± 2.02 (-17.43 ± 22.11)	10.46 ± 2.46	13.08 ± 2.84 (-13.08 ± 30.46%)	11.18 ± 3.75	10.79 ± 2.17 (-14.10 ± 23.92%)	9.29 ± 3.74	10.17 ± 1.67 (-19.25 ± 17.58%)	8.09 ± 1.64
Vastus medialis								
MT	2.28 ± 0.35 (27.20 ± 2.17%)	2.91 ± 0.58	2.49 ± 0.43 (14.39 ± 3.25%)	2.84 ± 0.55	2.37 ± 0.39 (25.22 ± 3.99%)	2.93 ± 0.45	2.49 ± 0.38 (16.66 ± 3.16%)	2.87 ± 0.38
θ_p	11.14 ± 2.13 (56.55 ± 50.40)	16.87 ± 4.50	11.70 ± 3.27 (59.68 ± 32.92)	18.32 ± 4.98	16.41 ± 3.47 (32.79 ± 19.52%)	21.30 ± 2.80	15.05 ± 5.18 (58.41 ± 37.30%)	22.49 ± 5.64
L_f	11.67 ± 2.41 (-11.61 ± 23.29)	10.11 ± 2.59	12.78 ± 2.29 (-25.41 ± 19.92%)	9.33 ± 2.25	9.32 ± 1.97 (-14.41 ± 19.46%)	7.70 ± 1.42	10.35 ± 2.97 (-23.28 ± 25.33%)	7.53 ± 1.86
Vastus intermedius								
MT	2.32 ± 0.32 (2.57 ± 2.17%)	2.36 ± 0.28	2.25 ± 0.34 (11.84 ± 5.78%)	2.46 ± 0.33	2.09 ± 0.39 (9.13 ± 3.38%)	2.26 ± 0.38	1.84 ± 0.34 (31.86 ± 5.04%)	2.37 ± 0.26
θ_p	11.53 ± 1.91 (54.94 ± 23.18)	17.69 ± 3.25	13.73 ± 3.36 (42.89 ± 36.22%)	19.22 ± 5.45	14.64 ± 3.01 (46.59 ± 27.73%)	21.03 ± 4.23	14.10 ± 3.57 (59.00 ± 27.64%)	21.75 ± 3.51
L_f	11.18 ± 1.2 (-26.48 ± 10.28)	8.22 ± 1.45	10.69 ± 1.67 (-21.18 ± 17.81%)	8.30 ± 1.73	8.73 ± 1.26 (-22.61 ± 16.21%)	6.68 ± 1.44	8.99 ± 1.50 (-20.23 ± 15.61%)	7.04 ± 1.17

Values are expressed as mean ± SD. Legend: SK20°: seated with knee at 20°; SJ60°: seated with knee at 60°; LK20°: lying with knee at 20°; LK60°: lying with knee at 60°; MT: muscle thickness; θ_p : pennation angle; L_f : fascicle length.

Muscle architecture

Table 2 shows the mean \pm SD for muscle thickness, θ_p , and L_f of the RF, VL, VM, and VI at rest and during NMES, as well as the percentage change, in the four lower limb positions. Figure 3 shows the mean \pm SD and statistical significances on *post-hoc* analysis of θ_p and L_f at rest and during evoked contraction, and the main effect of position, for the RF, VL, VM, VI, and QF in the four lower limb position. ICC was moderate for muscle thickness (0.87), θ_p (0.85), and L_f (0.85).

Rectus femoris

There was interaction between *positioning* and *intensity* for θ_p ($F_{3, 57} = 3.64$, $p = 0.017$, η_p^2 : 0.16, power: 0.77; (Fig. 3A). The *post-hoc* analysis showed significant differences from rest to evoked contraction in each position ($p < 0.001$), except LK60 ($p = 0.12$). Furthermore, there was no difference between positions at rest ($p = 0.19-1.0$), but during contraction LK60 had lower θ_p compared to SK60, LK20, and SK20 ($p < 0.001$). There was no interaction of factors for L_f ($F_{3, 57} = 0.18$, $p = 0.90$, η_p^2 : 0.009, power: 0.08; Fig. 3B), but position effect was significant ($F_{3, 57} = 9.64$, $p < 0.001$, η_p^2 : 0.33, power: 0.99), where the *post-hoc* analysis showed greater L_f ($p < 0.001-0.002$) for LK60 compared to all positions.

Vastus lateralis

There were no significant interactions for θ_p ($F_{3, 57} = 2.22$, $p = 0.095$, η_p^2 : 0.10, power: 0.53; Fig. 3C) or L_f ($F_{3, 57} = 0.33$, $p = 0.79$, η_p^2 : 0.017, power: 0.11; Fig. 3D). Position factor was significant for both θ_p ($F_{3, 57} = 14.23$, $p < 0.001$, η_p^2 : 0.42, power: 0.99) and L_f ($F_{3, 57} = 11.79$, $p < 0.001$, η_p^2 : 0.38, power: 0.99). The *post-hoc* analysis showed lower θ_p ($p < 0.001-0.011$) and greater L_f ($p < 0.001-0.031$) for LK60 and SK60 compared to LK20 and SK20.

Vastus medialis

There was a significant interaction for L_f of VM ($F_{3, 57} = 2.867$, $p = 0.044$, η_p^2 : 0.13, power: 0.65; Fig. 3F). The *post-hoc* analysis showed that SK60 ($p < 0.001$) and SK20 ($p < 0.001$) were different from rest to contraction, but not LK60 ($p = 0.10$) and LK20 ($p = 0.083$). Moreover, there were significant differences in LK60 and SK60 compared to LK20 and SK20 ($p < 0.05$), except LK60 vs SK20 at rest ($p = 0.25$) and SK60 vs LK20 during NMES ($p = 0.077$). There was no interaction for θ_p ($F_{3, 57} = 2.2$, $p = 0.097$, η_p^2 : 0.10, power: 0.53; Fig. 3E), but there was a significant main effect of *positioning* ($F_{3, 57} = 14.75$, $p < 0.001$, η_p^2 : 0.43, power: 0.99). The *post-hoc* analysis showed lower θ_p ($p < 0.001$) for all pairwise comparisons of LK60 and SK60 compared to LK20 and SK20.

Vastus intermedius

There was no significant effect of interaction for θ_p ($F_{3, 57} = 1.39$, $p < 0.25$, η_p^2 : 0.06, power: 0.35; Fig. 3G) or L_f ($F_{3, 57} = 1.707$, $p = 0.15$, η_p^2 : 0.08, power: 0.42; Fig. 3H), but position factor was significant for θ_p ($F_{8, 03} = 8.03$, $p < 0.01$, η_p^2 : 0.29, power: 0.98) and L_f ($F_{3, 57} = 22.905$, $p < 0.001$, η_p^2 : 0.54, power: 1.0). L_f was greater at LK60 and SK60 than at LK20 and SK20 ($p < 0.001$). However, for θ_p , LK60 was greater than LK20 ($p < 0.001$) and SK20 ($p < 0.001$), but SK60 was not ($p = 0.25$ and 0.30 , respectively).

Quadriceps femoris

Considering the QF, there was an effect of interaction for θ_p ($F_{3, 57} = 6.45$, $p < 0.001$, η_p^2 : 0.25, power: 0.95; Fig. 3I). Although the *post-hoc* showed that increasing θ_p from rest to contraction was highly significant for all positions ($p < 0.001$), the delta changes for all positions were different: LK60 = $4.674 \pm 2.33^\circ$, SK60 = $5.205 \pm 2.24^\circ$, LK20 = $5.374 \pm 1.66^\circ$, and SK20 = $7.269 \pm 2.65^\circ$, where a complementary one-way ANOVA showed that it was higher at SK20 compared to LK60 ($p < 0.001$), SK60 ($p = 0.009$), and LK20 ($p = 0.02$). Furthermore, a lower θ_p ($p < 0.001$ - 0.01) was found for LK60

and SK60 compared to LK20 and SK20, both at rest and during contraction, besides a lower θ_p during NMES of LK60 compared to SK60 ($p = 0.001$), and LK20 compared to SK20 ($p = 0.014$). There was no interaction for L_f ($F_{3, 57} = 1.03$, $p = 0.38$, $\eta_p^2: 0.05$, power: 0.26; Fig. 3J), but a *positioning* effect was found ($F_{3, 57} = 24.57$, $p < 0.001$, $\eta_p^2: 0.56$, power: 1.0). The *post-hoc* analysis showed lower θ_p ($p < 0.001$) and greater L_f ($p < 0.001$) for LK60 and SK60 compared to LK20 and SK20, besides a greater θ_p at SK60 than at LK60 ($p = 0.008$).

Summarizing the time effect (rest vs contraction), for all constituents, individually and grouped, as expected, θ_p increased ($F_{3, 57} = 7.59-30.21$, $p < 0.001$, $\eta_p^2: 0.28-0.61$, power: 0.98-1.00) and L_f reduced ($F_{3, 57} = 9.64-24.57$, $p < 0.001$, $\eta_p^2: 0.33-0.56$, power: 0.99-1.0) from rest to evoked contraction. For the relative change in θ_p and L_f , for the majority of muscles, there was no main effect ($F_{3, 57} = 0.34-2.22$, $p = 0.09-0.88$, $\eta_p^2: 0.009-0.10$, power: 0.08-0.53), except for the θ_p of VM, but without significance in the *post-hoc* analysis ($F_{3, 57} = 2.89$, $p = 0.043$, $\eta_p^2: 0.13$, power: 0.66).

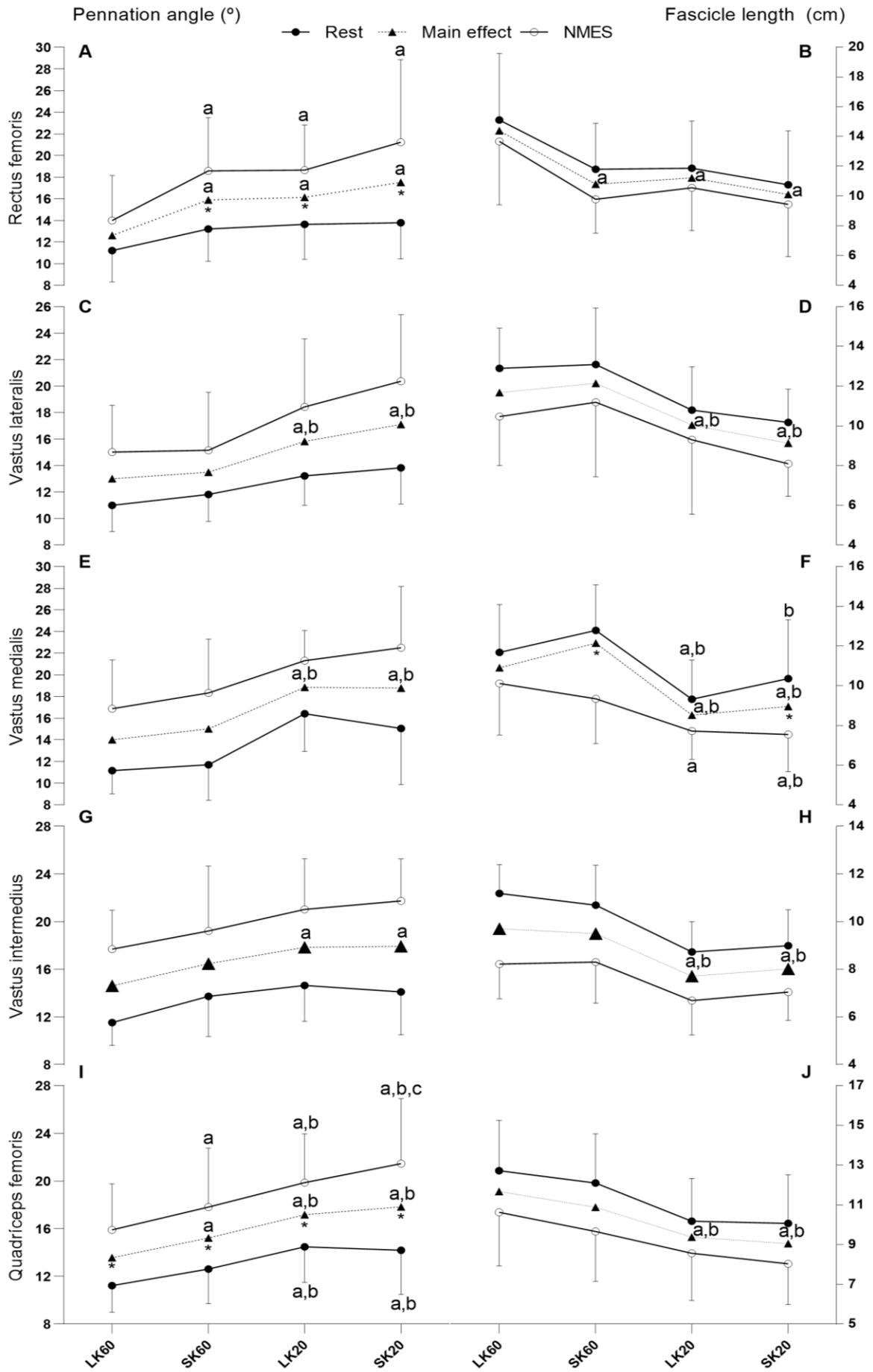


Figure 3 – Muscle architecture changes of the quadriceps femoris according to hip and knee angles at rest and during NMES: Pennation angle (left y axis) and fascicle length (right y axis) of all constituents of the quadriceps femoris individually and grouped at rest, during NMES (continuous lines), and main effect of position (dotted lines). Data are presented as mean \pm SD. (A-B): *Rectus femoris*; (C-D): *Vastus lateralis*; (E-F): *Vastus medialis*; (G-H): *Vastus intermedius*; (I-J): Quadriceps muscle. Legend: NMES: neuromuscular electrical stimulation; LK60: lying with knee at 60°; SK60: seated with knee at 60°; LK20: lying with knee at 20°; SK20: seated with knee at 20°. Statistically significant differences: ^adifferent from LK60; ^bdifferent from SK60; ^cdifferent from LK20. Asterisks (*) indicate significant differences from rest when there is a position by time effect; $p \leq 0.05$. (n=20 per group).

Tendon-aponeurosis complex displacement

Figure 4 shows the mean \pm SD and statistical significances on *post-hoc* analysis of the TACD for RF, VL, VM, VI, and QF in the four lower limb position. A significant effect of *positioning* was found for the VL ($F_{3,57} = 11.53$, $p < 0.001$, $\eta_p^2: 0.37$, power: 0.99), VM ($F_{3,57} = 5.0$, $p = 0.003$, $\eta_p^2: 0.20$, power: 0.89), VI ($F_{3,57} = 2.86$, $p = 0.044$, $\eta_p^2: 0.13$, power: 0.65), and QF ($F_{3,57} = 14.86$, $p < 0.001$, $\eta_p^2: 0.15$, power: 0.99), but not for the RF ($F_{3,57} = 1.24$, $p = 0.30$, $\eta_p^2: 0.06$, power: 0.31). For the VL, SK60 had greater TACD than SK20 ($p < 0.001$), LK60 ($p = 0.016$), and LK20 ($p < 0.001$). For the VM, SK60 had greater TACD than LK20 ($p = 0.002$). For the VI, SK60 had greater TACD than SK20 ($p = 0.025$). Finally, for the QF, SK60 had greater TACD than SK20 ($p < 0.001$), LK60 ($p < 0.001$), and LK20 ($p < 0.001$) (other comparisons: $p = 0.089-0.93$). The ICC for TACD was high (0.94).

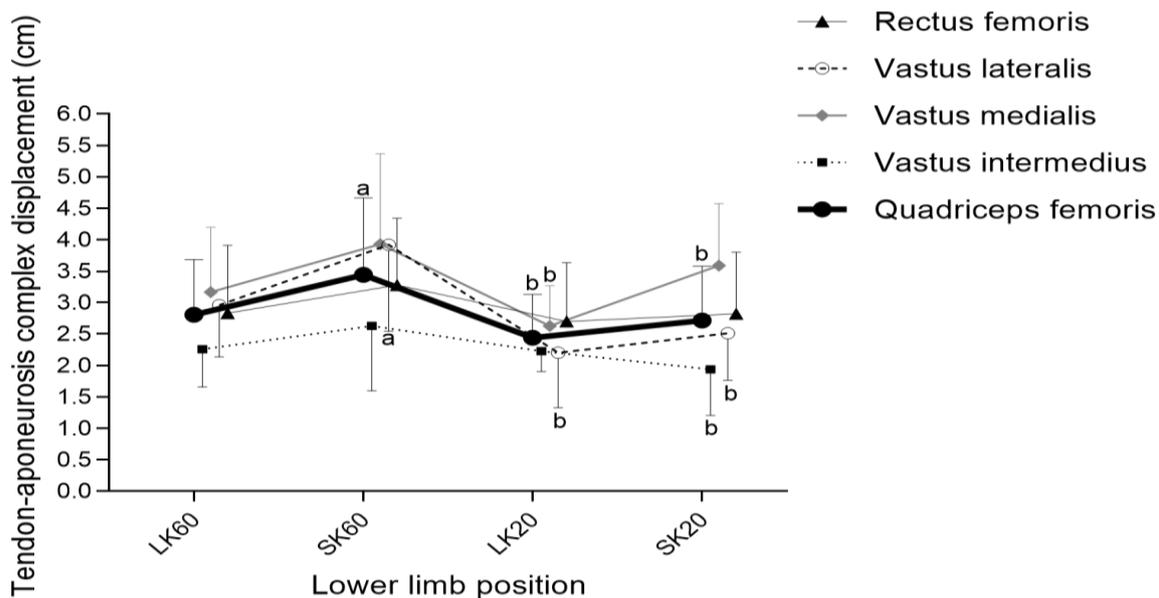


Figure 4 - Tendon aponeurosis complex displacement of the quadriceps femoris according to hip and knee angles at rest and during NMES: Tendon-aponeurosis complex displacement of all quadriceps femoris constituents individually and grouped (bold line). Data are presented as mean \pm SD and are significant at $p < 0.05$. Legend: NMES: neuromuscular electrical stimulation; SK60: seated with knee at 60°; SK20: seated with knee at 20°; LK20: lying with knee at 20°; LK60: lying with knee at 60°. Statistically significant differences: ^adifferent from LK60; ^bdifferent from SK60; ^cdifferent from LK20.

DISCUSSION

The main findings of this study were: 1) knee extensor evoked torque and current efficiency were higher at 60° of knee flexion compared to 20°, with no difference according to the hip angle (0° or 85°); 2) QF had lower θ_p and greater L_f at 60° of knee flexion; 3) TACD was lower at LK60 compared to SK60 despite the same torque. These new findings may help physical therapists and athletic trainers to develop more effective strategies when applying NMES by positioning the knee at 60° when the goal is to induce higher knee extensor torque.

Evoked torque and current efficiency

We clearly demonstrated that NMES applied at LK60 and SK60 produces greater absolute evoked torque and current efficiency compared to LK20 and SK20 (Table 1). Our results are in agreement with previous reports that found greater evoked torque at 60° of knee flexion compared to more extended positions (15° and 30°)^{8,9}. Other studies have shown that NMES is more fatiguing at 90° of knee flexion than at 15°³⁴ and at 65° compared to 90° or 20°³⁵, which was explained by greater pre-fatiguing torque in the more flexed position of Lee et al.³⁴ and in the midrange position of Marion et al.³⁵.

In addition, we investigated the current efficiency, which is a function of the evoked torque and current amplitude²⁵. To the best of our knowledge, this outcome has not previously been evaluated according to hip and knee angles. Current efficiency was greater at LK60 and SK60, demonstrating that at these angles NMES allows the generation of higher absolute torque with lower current amplitude^{25,26}. Our protocol required the maximal tolerated amplitude with the goal of achieving the maximal tolerated evoked torque. However, at a chosen percentage of the MVC, a cautiously chosen joint angle could allow the achievement of the targeted torque with lower current amplitude and, by doing so, with less sensory discomfort, which is a common limitation of NMES²⁴. Therefore, it

is possible to suggest the application of NMES at 60° of knee flexion unless contraindicated by disease or not tolerated.

Muscle architecture

Only two studies have assessed the architecture of all QF constituents *in vivo*, but they did not apply NMES or assess different joint angles^{22,36}. Our main finding was that QF (Fig. 3I, J) demonstrated a pattern where positions with the knee at 60° presented a lower θ_p and greater L_f compared to positions with the knee at 20°. Thus, it is suggested that LK60 and SK60 placed the QF at a better architectural configuration for torque generation, i.e., improved harnessing of muscle force and ideal sarcomere length^{1,16}. Individually, this optimum pattern according to knee angle was found in the VL, VM, and largely in the VI, while for the RF, it was more dependent on hip angle (Fig 4 A-H). In contrast, LK20 and SK20 presented an increased θ_p and shorter L_f , which, respectively, attenuates the transmission of force to the tendon-aponeurosis complex³⁶ and reduces force production, according to the force–length relationship². Interestingly, with some exceptions (θ_p of RF, L_f of VM, and θ_p of QF; Fig. 3A, F, I), the change from rest to contraction did not differ and the percentage change was the same in different positions, despite differences in torque and muscle architecture, which limits these findings to explain mechanisms for torque production.

Rectus femoris

We clearly demonstrated the effect of hip angle on the RF (Fig. 3A, B), as expected for the only biarticular constituent of QF. The θ_p was lower and L_f greater at LK60 than in all other positions, since LK60 was the most stretched position assessed here. A lack of difference between SK60 and LK20 probably occurred because each position shortened the RF in one joint and lengthened it in another, but the similarity between LK20 and SK20 indicate that, beyond a certain angle within the range of motion, no significant reduction

occurs in fiber length, but only a slack in the muscle-tendon unit ¹². This is supported by Herzog et al. ², who predicted that, when the hip is flexed, RF force ceases before full knee extension is reached. Considering θ_p at rest, there were no differences between any positions, in contrast to the lower θ_p at LK60 that occurred during contraction, possibly because θ_p changes due to joint rotation may be more pronounced if a muscle is contracting ³⁷.

Vastus lateralis, medialis, and intermedius

The VL and VM, monoarticular constituents of QF, showed the clear effect of knee angle, i.e., lower θ_p and greater L_f when the knee was at 60° compared to 20° (Fig. 3C-F). Unexpectedly, the L_f of the VM did not change from rest to contraction at LK60 and LK20. Grob et al. ³⁸ reported that VM insertion expands over the entire length of the VI aponeurosis and on the medial edge of the biarticular RF. Furthermore, inter- and extramuscular connective tissues are a source of interaction between synergistic muscles ³⁹. This evidence suggests that VM architecture may be influenced by the hip angle. For the VI, only LK60, but not SK60, demonstrated lower θ_p compared to positions with the knee at 20° (Fig. 3G). Since the VI is surrounded by the superficial QF muscles (RF, VL, and VM), it may be compressed due to space constraints ¹⁶, therefore, when the RF is stretched at LK60, it compresses the VI, reducing its θ_p .

Tendon-aponeurosis complex

The QF had a lower TACD at LK60 than at SK60 (Fig. 4) despite the same absolute evoked torque, indicating that the increased passive tension at LK60 limited the tendinous elongation during contraction ¹⁴. Increased tension of the tendon-aponeurosis complex in stretched conditions allows stronger contractions with less effort due to better force transmission from muscle to bone ¹⁵. This was demonstrated by Maffiulet et al. ⁶ and Bampouras et al. ⁷, who found greater knee extensor evoked torque in the lying compared

to the seated position with the knee at 90°. In the present study, we only found a non significant increase in evoked torque and current efficiency at LK60 compared to SK60. Fukutani et al.¹⁴ found higher triceps sural torque at a longer muscle length compared to a neutral position. However, for the QF, a more stretched position at 90° of knee flexion decreased voluntary and evoked torque compared to 60°^{5,8}. These discrepancies in the literature indicate a limit between improving force transmission while avoiding a mismatch between contractile filaments of muscle fibers².

According to Massey et al.³⁶, stiffer tendons also cause less muscle shortening at the same absolute force. Supporting this concept, we found that the increase in θ_p of the RF was not significant at LK60 (Fig. 3A), and neither was the decrease in L_f of the VM at LK60 and LK20, which correspond to the lower TACD at LK60. On the other hand, larger fascicle shortening may be necessary to eliminate the slack of the tendon-aponeurosis complex in more shortened positions¹², which explains why at LK20 and SK20 the TACD of QF was the same as at LK60 despite torque dissimilarities. Suydam et al.⁴⁰ showed that a lengthened Achilles tendon (after rupture repair) reduces the ability to generate adequate triceps surae muscle output, requiring increased muscle shortening for compensation, but without efficient load transmission and leading to atrophy. Similarly, it is possible that a muscle in a shortened position needs to contract more without proper load, limiting the stimulus for strengthening and hypertrophy.

Some limitations should be addressed in the present study. Our ultrasound had a probe width of 40 mm, which limited visualization of the entire muscle fascicles. However, this limitation is commonly demonstrated and there are reliable methods to overcome this issue, such as equations for L_f stimulations³³. Despite this limitation, we showed moderate and high ICCs for our outcomes. Another limitation is related to the nature of the NMES-induced contractions assessed by ultrasound whose visualization is challenging due to

muscle deformation and reduced control of contraction velocity. Finally, our results are limited to our population and a single session of NMES.

PERSPECTIVES

The present study provides novel application of sports medicine basis for the optimal adjustment of hip and knee angles during NMES. Importantly, once the knee angle dictates the knee extensor torque, it should be maintained at the ideal angle of 60°, regardless of the hip angle, unless contraindicated by joint disease, musculoskeletal injury or discomfort. Therefore, physical therapists and athletic trainers may use the seated or lying position according to the clinical setting. Further studies are necessary to elucidate how the knee and hip angles influence the short and long-term adaptations of muscle architecture and tendon-aponeurosis complex following NMES training programs.

CONCLUSIONS

NMES generate greater torque at 60° of knee flexion, compared to 20°, regardless of the hip angle. Each quadriceps constituent demonstrated unique behavior according to hip and or knee angle, but a greater L_f and lower θ_p were predominant for positions with greater torque (SK60 and LK60). A more elongated position stiffens the tendon-aponeurosis complex, as demonstrated by a lower TACD at LK60 compared to (SK60), which probably contributed to optimized transmission of force and slightly higher torque for LK60. Clinicians should preferably use NMES at SK60 or LK60 for strengthening purposes.

CONFLICT OF INTEREST

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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APPENDICES

Appendix I – Submission proof

Scandinavian Journal of Medicine & Science in Sports - PROOF



The effect of hip and knee joint angles on the muscle-tendon unit during neuromuscular electrical stimulation

Journal:	<i>Scandinavian Journal of Medicine and Science in Sports</i>
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Keywords:	Neuromuscular Electrical Stimulation, Angle-torque relationship, Muscle architecture, Tendon-aponeurosis complex, Quadriceps

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Appendix II - Author Guidelines for submission

Sections

- 1. Submission**
- 2. Aims and Scope**
- 3. Manuscript Categories and Requirements**
- 4. Preparing the Submission**
- 5. Editorial Policies and Ethical Considerations**
- 6. Author Licensing**
- 7. Publication Process After Acceptance**
- 8. Post Publication**
- 9. Editorial Office Contact Details**

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6. Main text;
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Author Guidelines Updated 27 November 2019

Appendix III - Research ethics committee approval

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PARECER CONSUBSTANCIADO DO CEP

DADOS DO PROJETO DE PESQUISA

Título da Pesquisa: Efeitos agudos da estimulação elétrica neuromuscular em diferentes ângulos do quadril e do joelho nas adaptações neuromiotendíneas e no torque extensor do joelho em adultos jovens saudáveis

Pesquisador: Jonathan Galvão Tenório Cavalcante

Área Temática:

Versão: 1

CAAE: 94388718.8.0000.8093

Instituição Proponente: Universidade de Brasília Faculdade de Ceilândia

Patrocinador Principal: Financiamento Próprio

DADOS DO PARECER

Número do Parecer: 2.799.049

Apresentação do Projeto:

Trata-se de um projeto mestrado do Programa de Pós Graduação em Ciências da Reabilitação, da Faculdade de Ceilândia – Universidade de Brasília, do mestrando Jonathan Galvão Tenório Cavalcante, e demais pesquisadores envolvidos: Karenina Arrais Guida Modesto, Rita de Cássia Marqueti Durigan, Nicolas Babault e João Luiz Quagliotti Durigan (orientador). Tem como Área de Estudo (Grandes Áreas do Conhecimento (CNPq): Grande Área 4. Ciências da Saúde e o Propósito Principal do Estudo (OMS): Clínico. O pesquisador deixa claro quais são os problemas ou Condições de saúde (Plasticidade musculotendínea, Força muscular, Relação comprimento-tensão, Atividade elétrica muscular) vinculando ao CID: M62.9 - Transtorno muscular não especificado e, M62 - Outros transtornos musculares.

Os pesquisadores informam que:

“Trata-se de um estudo observacional de delineamento transversal. As variáveis independentes são: 1) o posicionamento do membro inferior: angulação da articulação do joelho em 20° ou 60° com o quadril em 0° ou 80° (respectivamente, indivíduos deitados ou sentados com leve inclinação); 2) a estimulação elétrica neuromuscular para obtenção do torque evocado. As variáveis dependentes serão a contração voluntária máxima (CVM), o torque evocado eletricamente (TEE), a atividade elétrica de superfície, a arquitetura muscular (espessura muscular, ângulo de penação e comprimento fascicular) e o alongamento do complexo tendão-aponeurose do quadríceps e a

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Continuação do Parecer: 2.799.049

rigidez, módulo de Young e área de secção transversa do tendão patelar. Os torques gerados na CVM e no TEE serão comparados entre os diferentes ângulos do joelho, enquanto que as demais variáveis serão comparadas em duas situações: em repouso entre os diferentes ângulos do joelho e em repouso versus em contração.”

Ainda descrevem no projeto que “A estimulação elétrica neuromuscular (NMES) é um recurso não-invasivo que ativa o músculo esquelético artificialmente e, dessa forma, perpassa o trajeto natural da contração muscular (Trimble & Enoka, 1991). A NMES é amplamente usada na reabilitação para auxiliar na recuperação de força, reeducação muscular, redução de atrofia muscular e redução de limitações funcionais (Salvini et al., 2012; Vaz et al., 2013, Durigan et al., 2014a,b; Oliveira et al., 2018a; Almeida et al., 2018), bem como também é usada entre indivíduos saudáveis e atletas para o aumento de força muscular e desempenho esportivo (Bax et al., 2005; Billot et al., 2010).”

Objetivo da Pesquisa:

Objetivo Primário:

- Investigar o efeito da estimulação elétrica neuromuscular – NMES, em diferentes ângulos do quadril e do joelho no torque extensor, na atividade eletromiográfica, na arquitetura muscular, no complexo tendão-aponeurose e nas propriedades tendíneas dos componentes do quadríceps.

Objetivos específicos

Analisar o torque de extensão do joelho durante a estimulação elétrica neuromuscular tendo como intervenção controle a contração voluntária isométrica máxima;

Analisar a atividade elétrica dos três componentes superficiais do quadríceps (vasto lateral, reto femoral e vasto medial) em repouso e durante contração máxima, bem como do antagonista bíceps femoral;

Analisar a arquitetura muscular (ângulo de penação, comprimento fascicular e espessura muscular) dos quatro componentes do quadríceps femoral em repouso e durante contração máxima;

Analisar a rigidez do complexo tendão-aponeurose do tendão quadricipital em repouso e durante as manobras de contração máxima;

Testar as variáveis acima em quatro posturas diferentes: joelho em 0°, 60° e 90°, estando o quadril em 80°, e ambos o joelho e o quadril em 0° (ou seja, em extensão, estando o indivíduo em decúbito dorsal).

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Avaliação dos Riscos e Benefícios:

RISCOS

- Reações alérgicas e/ou queimadura a nível tecidual nas regiões selecionadas para avaliação da excitabilidade neuromuscular inerente à estimulação elétrica;
- Lesão neuromuscular ou tendínea decorrente da estimulação elétrica;
- Constrangimento durante as avaliações;
- Fadiga muscular.

BENEFÍCIOS:

- Os participantes receberão, de forma impressa ou por email, os resultados dos exames de imagem e eletromiográficos realizados e serão explicados o significado e as repercussões conforme a literatura atual;
- Os indivíduos terão a força dos extensores do joelho avaliada por meio de um dinamômetro isocinético e saberão se se encontram na faixa esperada para a sua faixa etária e altura;
- No caso de que alterações musculoesqueléticas sejam observadas, mesmo que estas impossibilitem o indivíduo de participar do estudo, será realizada avaliação fisioterapêutica mais aprofundada e orientações serão fornecidas quanto ao tratamento e a necessidade de consulta médica.
- Após finalização do estudo e publicação, cada participante receberá a versão em PDF do artigo, bem como terão livre acesso à dissertação no acervo da Biblioteca da Universidade de Brasília.

Medidas de proteção de risco e outros aspectos éticos

- Serão utilizados materiais de uso comum em pesquisas e na prática clínica, como gel condutor, gaze e eletrodo de carbono, os quais não possuem histórico de proporcionar reações adversas ao contato com a epiderme íntegra;
- O risco de lesão pela corrente da estimulação elétrica será minimizado por meio do incremento controlado da intensidade do estímulo e pela observação da reação muscular e da presença de hiperemia na pele;
- Será respeitada a autonomia do participante durante todo o processo, podendo o mesmo se recusar a qualquer momento, caso não se sinta apto a realizar os procedimentos. Os resultados serão transmitidos de forma apropriada, adaptada à condição física, emocional e intelectual do indivíduo;
- Será garantido o sigilo profissional e todos os participantes serão identificados por números, sendo ocultados seus nomes dos formulários de avaliação;

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- As contrações musculares serão separadas por um período suficiente de repouso para evitar fadiga muscular.

Comentários e Considerações sobre a Pesquisa:

“O tamanho da amostra foi determinado do por meio do software G* Power (versão 3.13; Universidade de Trier, Alemanha): 20 sujeitos serão recrutados, considerando o pico de torque extensor voluntário 60° de flexão do joelho como a variável principal, de acordo com estudo previamente publicado (Ruiter et al., 2008), o qual mostra uma média de 107.3 Nm ± 20.7. Para isso, o nível de significância foi estabelecido em 5% (p= 0,05), um power (1- β) = 0,80 e um tamanho de efeito = 0,90. Os indivíduos serão recrutados pelo método de amostragem não probabilística de conveniência, por meio de panfletos a serem distribuídos na Universidade e por meio de convite verbal.”

Os pesquisadores deixam claro os critérios de inclusão e exclusão:

Critério de Inclusão:

Serão incluídos indivíduos idade entre 18 e 30 anos, do sexo masculino, hígidos e com índice de massa corpórea (IMC) 18,5 - 24,9 kg/m²; não participantes de treino sistemático de fortalecimento dos membros inferiores nos últimos 12 meses; praticantes ou não de atividades esportivas recreativamente; fisicamente ativos de acordo com o Questionário Internacional de Atividade Física (IPAQ); e com alcance de torque mínimo de 30% da contração voluntária isométrica máxima durante a NMES.

Critério de Exclusão:

Serão excluídos aqueles que apresentarem: dor em qualquer dos procedimentos; edema, lesão dérmica, limitação da amplitude de movimento articular, deformidade ou amputação em qualquer parte dos membros inferiores; histórico de luxação patelar ou trauma nos membros inferiores ou tronco que comprometa os resultados; condições que afetem a morfologia musculotendínea ou a excitabilidade neuromuscular como diabetes mellitus tipo II, hipercolesterolemia familiar, doença neuromuscular e cardiopatia grave; déficit cognitivo, doença psiquiátrica, dependência química ou problemas comportamentais que inviabilizem a cooperação com os procedimentos (Dudley-Javoroski et al., 2010).

A pesquisa terá um custo de R\$280.503,75, e o pesquisador deixa claro que esse custo refere-se, na maior parte aos equipamentos cedidos pela Faculdade da Ceilândia (FCE) da Universidade de

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Brasília, e os menores gastos serão arcados pelos pesquisadores.

Considerações sobre os Termos de apresentação obrigatória:

Todos os documentos foram adequadamente apresentados.

Conclusões ou Pendências e Lista de Inadequações:

Não há pendências.

Considerações Finais a critério do CEP:

Protocolo de pesquisa em consonância com a Resolução 466/12 do Conselho Nacional de Saúde. Cabe ressaltar que compete ao pesquisador responsável: desenvolver o projeto conforme delineado; elaborar e apresentar os relatórios parciais e final; apresentar dados solicitados pelo CEP ou pela CONEP a qualquer momento; manter os dados da pesquisa em arquivo, físico ou digital, sob sua guarda e responsabilidade, por um período de 5 anos após o término da pesquisa; encaminhar os resultados da pesquisa para publicação, com os devidos créditos aos pesquisadores associados e ao pessoal técnico integrante do projeto; e justificar fundamentadamente, perante o CEP ou a CONEP, interrupção do projeto ou a não publicação dos resultados.

Este parecer foi elaborado baseado nos documentos abaixo relacionados:

Tipo Documento	Arquivo	Postagem	Autor	Situação
Informações Básicas do Projeto	PB_INFORMAÇÕES_BASICAS_DO_PROJETO_1109531.pdf	23/07/2018 13:21:53		Aceito
Outros	Curriculo_Nicolas_Babault_ingles.pdf	23/07/2018 13:21:18	Jonathan Galvão Tenório Cavalcante	Aceito
Outros	Carta_resposta_membro_estrangeiro.pdf	23/07/2018 13:19:48	Jonathan Galvão Tenório Cavalcante	Aceito
Projeto Detalhado / Brochura Investigador	Projeto_Detalhado.docx	16/07/2018 18:24:30	Jonathan Galvão Tenório Cavalcante	Aceito
Outros	Curriculo_Lattes_Karenina_Arrais.pdf	16/07/2018 18:22:28	Jonathan Galvão Tenório Cavalcante	Aceito
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Outros	Curriculo_Lattes_Jonathan_Galvao.pdf	16/07/2018 18:20:36	Jonathan Galvão Tenório Cavalcante	Aceito

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Outros	termo_de_concordancia_de_instituicao_participante.pdf	16/07/2018 18:19:25	Jonathan Galvão Tenório Cavalcante	Aceito
Declaração de Pesquisadores	termo_de_responsabilidade_e_compromisso_do_pesquisador.pdf	16/07/2018 18:18:23	Jonathan Galvão Tenório Cavalcante	Aceito
Folha de Rosto	folhaDeRosto.pdf	16/07/2018 18:16:25	Jonathan Galvão Tenório Cavalcante	Aceito
Outros	cartaencaminhprojeto_ao_cepfce.docx	28/06/2018 11:08:53	Jonathan Galvão Tenório Cavalcante	Aceito
Orçamento	Planilha_de_orcamento.doc	28/06/2018 02:24:55	Jonathan Galvão Tenório Cavalcante	Aceito
TCLE / Termos de Assentimento / Justificativa de Ausência	TCLE_Jonathan_Galvao.doc	28/06/2018 02:22:51	Jonathan Galvão Tenório Cavalcante	Aceito
Cronograma	Cronograma.docx	28/06/2018 02:21:44	Jonathan Galvão Tenório Cavalcante	Aceito
Outros	termo_instituicao_proponente.jpg	28/06/2018 02:19:23	Jonathan Galvão Tenório Cavalcante	Aceito

Situação do Parecer:

Aprovado

Necessita Apreciação da CONEP:

Não

BRASILIA, 03 de Agosto de 2018

**Assinado por:
Dayani Galato
(Coordenador)**

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Appendix IV - Research ethics committee approval

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Information provided by (Responsible Party):

João Luiz Q. Durigan, University of Brasilia