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Bampouras, TM, Reeves, ND, Baltzopoulos, V and Maganaris, CN (2017) Interplay between body stabilisation and quadriceps muscle activation capacity. Journal of Electromyography and Kinesiology, 34. pp. 44-49. ISSN 1050-6411

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Title

Interplay between body stabilisation and quadriceps muscle activation capacity

Introduction

Isometric torque produced by maximal voluntary contraction (MVC) of the quadriceps is routinely used to assess knee joint function in various populations, such as clinical (e.g. [Hart et al., 1984; Souza et al., 2009]) and aged populations (e.g. [Reeves et al., 2004; Thompson et al., 2013]), as well as to assess the impact of various interventions (e.g [Labrunée et al., 2012; Stock and Thompson., 2014]). The result of this assessment depends on the quadriceps muscle size of the subject, as well as their ability to voluntarily activate the muscles tested [Bampouras et al., 2006; Kent-Braun and Le Blanc, 1996], with the latter factor being linked to the subject's stabilisation on the dynamometer seat [Hart et al., 1984; Magnusson et al., 1993].

Stabilisation and muscle activation are strongly interlinked, as a more stable segment will allow for higher muscle activation and, consequently, muscle force and torque generation. During quadriceps muscle strength assessment, stabilising the subject and, in particular, their pelvis on the dynamometer seat, allows for greater fixation of the rectus femoris origin [Hart et al., 1984]. This facilitates activation and hence greater force production by the rectus femoris muscle, contributing to increased quadriceps torque, as the rectus femoris accounts for ~17% of quadriceps torque [McNair et al., 1991]. Similarly, if the pelvis is not adequately stabilised, the bicep femoris muscle is likely to become more active to contribute more substantially towards ensuring stabilisation of the pelvis [van Wingerden et al., 2004]. In turn, this will reduce the force generated by the quadriceps muscle, through increased reciprocal neural inhibition by the hamstrings contracting to stabilise the pelvis [Hamm and Alexander, 2010], as well as the increased antagonistic muscle torque.

The activation of the tested agonist muscle, however, can potentially be enhanced through a different path. During an isometric quadriceps MVC, it is common for subjects to simultaneously contract a number of other muscles, remote to the tested muscle, to achieve

maximum torque [Jacobsen et al., 2012]. When hands were used to hold onto the dynamometer and the back was fixed to it, knee extensors torque was higher by 6.4 % compared to when only the back was fixed, which, in turn, was higher by 7.5% than when no stabilisation at all was used [Magnusson et al., 1993]. The activation of those remote muscles may augment the tested muscle's activation capacity, and subsequent torque produced, through a phenomenon termed concurrent activation potentiation (CAP) [Ebben, 2006; Ebben et al., 2008]. CAP is underpinned by the theory of motor overflow, which suggests that when a motor area is active, other areas are affected by that activation [Hoy et al., 2004]. In the primary motor cortex, which controls movements of the face, arms, and legs [Donohue and Sanes, 1994], activation of one area would also result in higher activation of the others. Indeed, this theory has been supported by studies reporting that contraction of remote muscles to the tested quadriceps, e.g. jaw and arms, results in higher knee extensor torque [Ebben et al., 2008]. Interestingly, remote voluntary contractions can also augment the agonistic muscle's torque by increasing stabilisation (for example, hands gripping onto the dynamometer, [Magnusson et al., 1993]; the Valsalva manoeuvre increasing intra-thoracic pressure through activation of various torso muscles, stabilising the core, [Harman et al., 1998]) or by directly increasing agonistic muscle activation capacity. Clearly, being able to distinguish between stabilisation and activation effects on torque

Clearly, being able to distinguish between stabilisation and activation effects on torque generation is important for avoiding erroneous conclusions in studies comparing muscle function assessment. Therefore, the aim of the present study was to determine the effect of stabilisation and muscle activation capacity on the quadriceps maximum voluntary isometric torque, by manipulating subject stabilisation configurations on a dynamometer seat and inclusion of simultaneous remote voluntary contractions.

Methods

Subjects

Following Institutional ethics approval, nine healthy, active males (mean \pm SD: age 28.7 \pm 6.8 years, stature 1.78 \pm 0.08 m, body mass 89.3 \pm 13.0 kg) free from any musculoskeletal injuries gave written, informed consent to participate in the study. To reduce variability in performance, all subjects were familiarised with the experimental procedures [Button and Behm, 2004] and visited the laboratory on a single occasion for testing.

Isometric knee extension strength measurement

Each subject's isometric knee extension strength was initially determined by performing two maximum voluntary contractions (MVCs). For those MVCs, the subjects were sat in the chair of a custom-made dynamometer [Bampouras et al., 2012] with the hip, knee and ankle joint angles at 90°. The lever arm and the bed of the dynamometer was very rigid, while the restraints allowed for better fixation of the pelvis and the subjects' body compared to commercially available dynamometers. Straps were positioned over the pelvis to prevent extraneous movement, while the tested right leg was securely strapped, above the lateral malleolus, to a force-transducer (KAP, E/200 Hz, Bienfait B.V. Haarlem, The Netherlands). If the coefficient of variation (calculated as standard deviation / average * 100) between the two MVCs was <5%, the two MVCs were averaged, otherwise a third MVC was performed and the closest two were averaged (average MVC).

Subsequently, subjects performed an MVC under four different conditions, in a randomised, counterbalanced order, which were:

- a) an MVC as described above (Typical MVC),
- b) an MVC as the Typical MVC but with the addition of exerting maximal handgrip force (Handgrip MVC),

c) an MVC where the subjects were instructed to isolate the contraction to their leg muscles only (Knee extension MVC) with the rest of the muscles relaxed, and d) an MVC where there were no restraining straps on the pelvis (Unrestrained MVC) (Figure 1). During all MVCs, subjects were asked to exert as much force as possible against the ankle strap and had their arms crossed over their chest, which were allowed to contract during effort (apart from the Knee extension MVC). Adequate rest between trials was provided.

FIGURE 1a, 1b, 1c ABOUT HERE

With the subjects sat in the dynamometer and their leg relaxed, the force trace was zeroed removing any passive force due to passive tension of the muscle-tendon unit of the knee extensors. Real-time force readings were displayed online and recorded (Matlab, The Mathworks, Natick, MA). Force recorded was the mean force during the plateau phase and 500ms prior to stimulus application (see *Muscle activation capacity measurement* below). The perpendicular distance from the centre of the knee joint to the point where force was applied (at the level of the ankle, at right angles to the longitudinal axis of the lower leg) was measured and multiplied by that force to provide torque, which was used for further analysis. *Handgrip strength*

Handgrip strength was assessed with the use of a dynamometer (Takei Scientific Inst. Co. Ltd, Niigata, Japan). The subject, sat in the dynamometer chair, held the handgrip dynamometer in the same position as they would have it during the Handgrip MVC. They then squeezed the dynamometer as hard as they could and the maximum value achieved was recorded. Similarly to the MVC procedures, two trials were performed, unless the coefficient of variation was > 5%, in which case a third contraction was performed and the closest were averaged.

Muscle activation capacity measurement

Two 7 x 12.5-cm self-adhesive carbon rubber electrodes (Versa-Stim, ConMed, New York, USA) were placed on the proximal and distal regions of the quadriceps muscle group. The greater size and placement of the electrodes aimed to induce the highest possible knee extension torque generation. Two stimuli of 200-µs pulse width and 10-ms inter-stimulus gap (doublets) were generated by a constant current electrical stimulator (model DS7, Digitimer stimulator, Welwyn, Garden City, UK) and applied at rest and at increments of 50mA, with the voltage set at 300 V. The stimulation intensity that resulted in generating one third of the average MVC torque [Bampouras et al., 2012] was recorded and used for the experiment. Subsequently, the subjects performed the four difference MVC conditions and a doublet was applied at the plateau phase of each MVC (superimposed) and approximately 4 seconds after the superimposed twitch and while the subject was relaxed (resting) (Figure 2). Electrical stimuli application was displayed online along with the force signal. Muscle activation capacity was quantified from the superimposed and resting twitch torque using the interpolated twitch technique according to the equation ((1 – (superimposed twitch torque))*100.

FIGURE 2 ABOUT HERE

Electromyography (EMG) measurement

Two surface Ag-AgCl electrodes of 10mm diameter each were placed in a bipolar configuration on flexor carpi radialis, biceps brachii, triceps brachii (long head), deltoid, pectoralis major, sternocleidomastoid, rectus abdominis, external oblique, vastus lateralis, biceps femoris (long head), and latissimus dorsi muscles to obtain EMG signals. The placement area was prepared by shaving and alcohol cleansing and all electrodes were placed

perpendicular to the muscle fibres, with a centre-to-centre distance of 20mm and on the right handside, except for the vastus lateralis where the contralateral muscle was used. These muscles were selected as the more likely muscles to contract during the MVCs described above and, thus, provide an indication of muscle activity during contractions as well as adherence to instructions for the Leg MVC.

EMG was collected at a sampling rate of 2000Hz, and filtered with a high- and low-pass filter of 10 and 500Hz, respectively. The signal was subsequently smoothed using root mean square over 30ms (Aqknowledge, Biopac Systems, Santa Barbara, California) and a mean value from a 500ms window was taken during the plateau phase of the MVC and prior to the application of the twitch. As no comparison between subjects or muscles was to be conducted and testing took place in a single session, no EMG normalisation was performed.

Statistical analysis

Normality of distribution of the data was checked using Shapiro-Wilk test and subsequently confirmed for handgrip strength, torque and muscle activation capacity but not for EMG. Consequently, separate one-way repeated measures ANOVAs were used for torque and muscle activation capacity to compare differences between the four MVC conditions, followed by dependent t-test for pairwise comparisons when differences were found. In addition, a dependent t-test was used to compare handgrip strength performed on its own and during Handgrip MVC. Friedman's test was used to compare EMG between conditions for all muscles followed by Wilcoxon test where differences were found. Holm-Bonferroni adjustment was used for all pairwise comparisons and the adjusted p values are presented for these comparisons.

Effect sizes (ES) were calculated for significantly different comparisons to provide an indication of the magnitude of the effect, with 0.8, 0.5 and 0.2 representing large, moderate and small effects for parametric tests effects sizes and 0.5, 0.3 and 0.1 representing large,

moderate and small effects for non-parametric tests effects sizes [Fritz et al., 2012]. For all statistical analysis IBM SPSS Statistics v 22 was used. Data are presented as means \pm SD, unless otherwise stated. Statistical significance level was set at p < 0.05.

Results

Average MVC torque from the two initial MVCs was 298.1 ± 56.7 Nm, while submaximal stimulation intensity was 372 ± 123.0 mA.

A significant overall difference was found for torque between the four conditions (p = 0.001). Subsequent analysis revealed that Typical MVC was significantly higher than Leg MVC (p = 0.008, ES = 1.4) and Unrestrained MVC (p = 0.004, ES = 1.7), while Handgrip MVC was also significantly higher than Leg MVC (p = 0.034, ES = 1.0) and Unrestrained MVC (p = 0.008, ES = 1.3) (Figure 3). In addition, handgrip strength was not significantly different (p = 0.282) between handgrip performed on its own (44.4 \pm 6.4 kg) and Handgrip MVC (41.6 \pm 6.1 kg).

FIGURE 3 ABOUT HERE

Muscle activation capacity was significantly different between conditions (p = 0.001), with higher activation for Typical MVC compared to Leg MVC (p = 0.020, ES = 0.7) and Unrestrained MVC (p = 0.002, ES = 1.1), and higher activation for Handgrip MVC compared to Unrestrained MVC (p = 0.001, ES = 1.0) (Figure 4). No other differences for activation were found.

FIGURE 4 ABOUT HERE

EMG differences between conditions were seen for the flexor carpi radialis, biceps brachii, triceps brachii (long head) and external oblique muscles only (Figure 5). For the flexor carpi radialis, Leg MVC was lower than Typical MVC (p = 0.036, ES = 0.9), Handgrip MVC (p = 0.036, ES = 0.8) and Unrestrained MVC (p = 0.036, ES = 0.8). For the biceps brachii, Leg

MVC was lower than Handgrip MVC (p = 0.036, ES = 0.8) and Unrestrained MVC (p = 0.036, ES = 0.8). For the triceps brachii (long head), Leg MVC was lower than Typical MVC (p = 0.012, ES = 0.9). Finally, for the external oblique muscle, both Leg MVC (p = 0.036, ES = 0.8) and Unrestrained MVC (p = 0.036, ES = 0.8) were lower than Handgrip MVC.

FIGURE 5a, 5b, 5c, 5d ABOUT HERE

Discussion

The aim of the study was to examine the effect of subject stabilisation and muscle activation capacity on knee joint torque developed during an isometric MVC, by distinguishing the effect of each component through manipulation of stabilisation configurations and inclusion of remote voluntary contractions. The results suggest that although both stabilisation and activation capacity play an important role in torque generation, stabilisation of the involved segments plays the major role which will in turn allow fuller activation of the muscle. When the handgrip was added to the Typical MVC, no statistically significant change in torque or muscle activation was observed. Our results agree with CAP literature showing that when bilateral handgrip was added to knee extension, torque from an isometric contraction [Ebben et al., 2009] or dynamic contraction [Cherry et al., 2010] did not change, suggesting no beneficial effect of handgrip on knee extensor torque. Similarly to the present study, handgrip strength was also not significantly reduced during the knee extension (Cherry et al., 2010; Ebben et al., 2008]. These findings contradict expectations of increased activation and subsequent torque due to increased H-reflex activity and motor-evoked potentials induced by the additional handgrip contraction (Dowman and Wolpaw, 1988; Péréon et al., 1995]. One possible reason for this contradiction is the contraction of the handgrip-related muscles not being sufficient to excite further the difficult to activate (possibly due to its higher content of type II muscle fibres; [Johnson et al., 1973]) quadriceps muscle [Behm et al., 2002], as suggested by the very similar activation values during Typical MVC and Handgrip MVC. A second possible reason relates to the action performed with the handgrip. When the arms were used to grab the dynamometer seat, an increase in knee extension torque was seen, attributed to a better-fixed torso [Magnusson et al., 1993]. However, the handgrip contraction used in the present study and Ebben et al. [2008] and Cherry et al. [2010] studies, does not appear to substantially contribute towards stabilising the torso. Therefore, although excitatory

responses may take place during the grip, these do not assist in further stabilising the pelvis during knee extension. This notion is supported by findings that gripping the dynamometer seat or the pelvic strap during knee extension had no effect on quadriceps torque [Kramer, 1990], most likely due to the fact that both actions offered the same stabilising effect to the torso.

When the subjects were requested to focus on contracting the knee extensors only (Knee extension MVC) while still restrained by the dynamometer belt, activation was reduced by 17.5%, while torque was reduced by 24.2% when compared to the Typical MVC values. Knee extension MVC EMG data suggests that subjects 'engaged less' the flexor carpi radialis, biceps brachii, triceps brachii (long head) and external oblique muscles, during the Knee extension MVC. Interestingly, the rest of the EMG data showed no difference between any of the conditions. This could mean that other muscles were not required for the contraction, and hence, they remained 'quiet' throughout all conditions, or they were crucial to the contraction and therefore they were activated to achieve the task required in all conditions, regardless of the instruction. Whichever the reason, the lack of difference in EMG activity between conditions for the rest of the muscles studied, precludes them as contributors to the muscle activation capacity changes.

When the pelvis and tested right thigh restraints were removed (Unrestrained MVC), the reduction in activation (22.0%) and torque (29.6%) compared to typical MVC, was higher than the respective reduction in activation and torque seen in Leg MVC condition, although not statistically significantly so. Given that the subjects' EMG in the measured muscles during Unrestrained MVC was equal or higher than the corresponding EMG values during Leg MVC, it is reasonable to assume that CAP did not augment knee extensor torque, as otherwise activation and torque would be higher in the Unrestrained condition. Collectively, these findings suggest that pelvis and tested thigh stabilisation is the major factor determining

the knee extensor torque produced during an isometric knee extension, and optimal stabilisation subsequently facilitates muscle activation enabling maximum possible force generation by the tested muscles.

The subjects in the present study were familiar with maximal isometric contraction, as per Typical MVC. However, the Leg MVC condition inevitably contained two potentially conflicting instructions ('push as hard as possible against the ankle strap' and 'use only your leg muscles, relax the other ones'). This could have presented a limitation to the force generation during this condition as the opposing instruction requirement could impact negatively on maximum force generation [Marchant, 2010]. In addition, during the Unrestrained MVC, there was a tendency for subjects to lift off the dynamometer seat. However, they all maintained a position similar to the Typical MVC. It is likely that this position was maintained by voluntary activation reduction to prevent further lifting off the chair, which supports further the concept of the need for stabilisation first to enable maximum voluntary activation of the muscles.

In conclusion, the present findings suggest that although stabilisation and activation are interlinked, stabilisation of the pelvis during an isometric knee extension is a priority in order to allow maximum voluntary activation of the quadriceps muscle. These results further reinforce the need for close attention to stabilisation during dynamometry-based knee joint functional assessment.

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Figure captions

Figure 1. Experimental set up for the four maximum voluntary contraction conditions.

Typical MVC (Fig 1a): subjects were asked to exert as much force as possible against the ankle strap; Handgrip MVC (Fig 1b): as the Typical MVC but with the addition of exerting maximal handgrip force; Knee extension MVC (same as Fig 1a): subjects were asked to isolate the contraction to their leg muscles only with the rest of the muscles relaxed; Unrestrained MVC (Fig 1c): as the Typical MVC but without restraining straps on the pelvis. EMG and electrical stimulation electrodes have been omitted for clarity.

Figure 2. A schematic diagram demonstrating the application of the stimulus during the maximum voluntary contraction (MVC) for all four conditions. A: initiation of MVC; B: MVC plateau phase (force and EMG measurements); C: superimposed stimulus; D: resting stimulus. The arrows indicate application of the stimulus.

Figure 3. Isometric knee extension torque in all four different conditions (Typical MVC, subjects sat in the dynamometer chair with straps over the pelvis and tested right thigh; Handgrip MVC, as the Typical MVC but with the addition of exerting maximal handgrip force; Knee extension MVC, subjects were instructed to contract their knee extension muscles only with the rest of the muscles relaxed; Unrestrained MVC, no restraining straps on the pelvis and tested thigh). Values are means and SD. Significant differences with Typical MVC are indicated by an asterisk, while significant differences with Handgrip MVC are indicated by a dagger symbol.

Figure 4. Quadriceps activation capacity in all four different conditions (Typical MVC, subjects sat in the dynamometer chair with straps over the pelvis and tested right thigh; Handgrip MVC, as the Typical MVC but with the addition of exerting maximal handgrip force; Knee extension MVC, subjects were instructed to contract their knee extension muscles only with the rest of the muscles relaxed; Unrestrained MVC, no restraining straps on the pelvis and tested thigh). Values are means and SD. Significant differences with Typical MVC are indicated by an asterisk, while significant differences with Handgrip MVC are indicated by a dagger symbol.

Figure 5. Mean quadriceps EMG in all four different conditions (Typical MVC, subjects sat in the dynamometer chair with straps over the pelvis and tested right thigh; Handgrip MVC, as the Typical MVC but with the addition of exerting maximal handgrip force; Knee extension MVC, subjects were instructed to contract their knee extension muscles only with the rest of the muscles relaxed; Unrestrained MVC, no restraining straps on the pelvis and tested thigh) for flexor carpi radialis (Panel A), biceps brachii (Panel B), triceps brachii (long head) (Panel C) and external oblique muscles (Panel D). Values are means and SD. Significant differences are indicated by a, difference with Typical MVC; b, difference with Handgrip MVC; c, difference with Leg MVC; d, difference with Unrestrained MVC.