Lack of Evidence for Non-Local Muscle Fatigue and Performance Enhancement in Young Adults

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Abstract
Post-activation performance enhancement (PAPE) is an improvement to voluntary muscle performance following a conditioning activity. There is evidence of fatigue resistance deficits in non-exercised muscles following unilateral fatigue exercise of a contralateral muscle. The purpose of this study was to determine if a unilateral conditioning exercise protocol could induce PAPE in a contralateral, non-exercised muscle in young healthy adults. Thirty-two recreationally trained (n = 16) and athletically trained (n = 16) participants (16 males; age: 22.9 ± 2.03 years; height: 1.81 ± 0.06 m; weight: 82.8 ± 9.43 kg, and 16 females; age: 23.1 ± 2.80 years; height: 1.67 ± 0.07 m; weight: 66.4 ± 11.09 kg) were randomly allocated into two groups (dominant or non-dominant limb intervention). The experimental intervention, involved a conditioning exercise (4-repetitions of 5-seconds knee extension maximal voluntary isometric contractions: MVIC) with either the dominant (DOM) (n = 16) or non-dominant (ND) (n = 16) knee extensors with testing of the same (exercised) or contralateral (non-exercised) leg as well as a control (no conditioning exercise: n = 32) condition. Testing was performed before, 1-minute and 10-minutes after a high intensity, low volume, conditioning protocol (2 sets of 2x5-s MVIC). Pre- and post-testing included MVIC force and F100 (force developed in the first 100 ms: a proxy measure of rate of force development) and unilateral drop jump (DJ) height and contact time. There were no significant MVIC peak force or EMG nor DJ height or contact time interactions (intervention x limb dominance x time). The pre-test (0.50 ± 0.13) dominant leg MVIC F100 forces exceeded (p = 0.02) both improvements and null effects within their studies (Halperin et al., 1983; Sjogaard et al., 1986), or central fatigue (motoneuron: (Bellemare and Garzaniti, 1988;
Stephens and Taylor, 1972), spinal cord: (Bigland-Ritchie et al., 1986; Maton and Gamet, 1989), and brain (Bergstrom and Hultman, 1988; Bigland-Ritchie et al., 1982). Although the effects of fatigue and potentiation on subsequent performance are intertwined (Behm, 2004; Behm et al., 2004; Rassier and MacIntosh, 2000), the scientific literature on PAPE-related neural mechanisms is not conclusive. Strong evidence for neural mechanisms are evident with crossover or NLMF studies (Halperin et al., 2015). Only two studies whose stated purpose was to examine crossover effects of a unilateral conditioning exercise reported potentiation of the conditioned leg but significant impairments of the homologous, contralateral limb (Andrews et al., 2016) or no contralateral effect (Wong et al., 2020). In contrast, while the objective of the Hamilton and Behm (2017) study was to examine non-local muscle fatigue with a unilateral fatiguing intervention (2 x 100-s MVIC), they found evidence of augmented contralateral MVIC force (PAPE). More studies are necessary to determine the characteristics of the conditioning protocol to elicit contralateral PAPE and the possible neural contributions with the use of a crossover limb design.

Cross education training studies have demonstrated a crossover effect from the dominant to the non-dominant side (Andrushko et al., 2018; Farthing et al., 2005), however, the crossover effects from non-dominant to dominant side are usually found to be weaker or absent (Farthing, 2009; Farthing et al., 2005). It has been hypothesized that since the dominant limb may be more proficient at learning a new task than the non-dominant limb greater information can be transferred (Parlow and Kinsbourne, 1989, Stoddard and Vaid, 1996). However, there are also studies that show a symmetry in the direction of crossover (Coombs et al., 2016; Ben Othman et al., 2019). The greatest asymmetry tends to occur with more novel tasks and thus less challenging task of coordination such as isometric strength tests may show less asymmetry (Farthing, 2009). To this point, there have been no studies investigating if there are differences in the crossover direction for PAPE studies. Such findings could contribute to the identification of mechanisms associated with asymmetrical responses to conditioning stimuli. Implications of this study could impact training and rehabilitation, where a stronger or non-injured muscle could be conditioned in order to potentiate contralateral weak or injured muscle(s) and improve the effectiveness of the rehabilitation exercises.

The objective of this exploratory study was to examine the crossover (non-local) effects using voluntary conditioning stimuli between dominant and non-dominant legs on unilateral MVIC, and jump performance of the knee extensors among resistance- and recreationally-trained participants. Based on Hamilton and Behm (2017), it was hypothesized that there would be performance enhancements in the conditioned, exercised knee extensors as well as the contralateral non-exercised knee extensors.

Methods

Participants

A purposeful sample of 32 recreationally and athletically trained participants (16 males; age: 22.9 ± 2.03 years; height: 1.81 ± 0.06 m; weight: 82.8 ± 9.43 kg, and 16 females; age: 23.1 ± 2.80 years; height: 1.67 ± 0.07 m; weight: 66.4 ± 11.09 kg) volunteered to take part in the study. Of the 32 participants, 16 were considered recreationally trained (regularly participating in physical activity for recreational purposes), and 16 were considered athletically trained (an athlete on a varsity or provincial sports team). The dominant leg of each participant was determined by asking which leg they would use to kick a soccer ball. Each participant was required to complete a physical activity readiness questionnaire (PAR-Q) with no positive responses and read and sign the informed consent form before participating in the study. Exclusion criteria included any neurological conditions or serious musculoskeletal injuries in the past year. The Interdisciplinary Committee on Ethics in Human Research (ICEHR) of the Memorial University of Newfoundland approved the study (Approval #: 20171234-HK).

Research design

The study followed a two-group mixed design. In order to examine the influence of limb dominance, participants were randomly assigned to either the dominant (DOM) conditioned or the non-dominant conditioned (ND) group. Based on the tendency for greater potentiation in strength trained individuals (Gourgoulis et al., 2003; Hrysomallis and Kidgell, 2001; Scott and Docherty, 2004), the randomization was controlled to ensure an equal number of recreationally trained and athletically trained participants in each group. Participants attended the lab on four different days, separated by a minimum of 48 hours, to complete four different sessions in a random order. The four sessions included one intervention and control day per leg. The conditioning exercise was always performed on the dominant leg for the DOM group and always performed on the non-dominant leg for the ND group. The rest on control days was equal to the length of the intervention. A testing protocol was performed immediately before the voluntary conditioning intervention, and 1- and 10-minutes after the intervention. The tests consisted of a unilateral maximum voluntary isometric contraction (MVIC) of the knee extensors, and single-leg drop jump (DJ). Due to practical reasons related to equipment preparation and time constraints, the testing protocol always began with the MVIC followed by the DJ.

Experimental protocol

Each day followed the same protocol. First, participants were prepared with electromyography (EMG) electrodes for the vastus lateralis (VL), and biceps femoris (BF). VL was chosen as an indicator of changes to the agonist muscle group, while BF was chosen in order to detect if there was any change in antagonist co-contraction during the study. Based on SENIAM recommendations (Hermens et al., 1999), the midpoint of each muscle, halfway between the anterior superior iliac spine and the top of the patella for VL and halfway between the gluteal fold and the popliteal space for the BF, and the lateral condyle of the tibia were marked to identify where the two muscle points and the ground respectively, would be located for EMG electrodes. Each location was prepared by shaving any hair, removing
dead skin cells with an abrasive pad, and cleaning the area with an alcohol swab. Researchers placed two recording electrodes (1-cm 162 Ag/AgCl; MediTrace 133, Kendall, Technical products Toronto, Ontario, Canada) at each of the specified muscle locations, two centimetres apart, parallel to the muscle fibre, and one recording electrode was placed on the fibular lateral condyle, acting as the ground electrode. Once the resting EMG was checked for a low noise signal (<0.05 mV amplitude), the participant began their warm-up. Electrode placement was traced with a permanent marker to allow for consistent placement between days. With both groups combined (D-ND and ND-D), reliability using Cronbach alpha intraclass correlation coefficients (ICC: Cohen 1988) showed coefficients of 0.68 and 0.74 for VL EMG and 0.73 and 0.85 for BF EMG for the contralateral and conditioned legs respectively.

The warm-up consisted of five minutes on a cycle ergometer (Monark Ergomedic 828E Exercise Test Cycle, Monark Exercise AB, Vansbro, Sweden) with 1 kilopond of resistance at 70 revolutions per minute, followed by 10; 5-second isometric knee extension contractions at approximately 50% of their perceived maximal intensity on the leg to be tested. One minute after the warm-up, the pre-test protocol began with the MVIC test, followed by the DJ test. EMG was recorded for both of these tests. Immediately following the pre-test, either the intervention or control protocol was performed. Upon completion of the intervention or control, a 1- and 10-minute post-test was performed. Using a random allocation, participants completed four sessions (Figure 1).

**Conditioning protocol**

Four different sessions were performed for each participant:

1) **Dominant control day**: Testing protocols performed on the dominant leg, 6 minutes and 20 seconds rest between pre-test and 1-minute post-test.

2) **Non-dominant control day**: Testing protocols performed on the non-dominant leg, 6 minutes and 20 seconds of rest between pre-test and 1-minute post-test

3) **Intervention and testing were performed on the same leg**: The dominant leg received both the conditioning exercise intervention and the testing for the DOM group and similarly, the non-dominant leg performed the conditioning intervention and was also tested for the ND group.

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**Figure 1.** Experimental design.
4) Intervention and testing were performed on separate (contralateral) legs. The conditioning intervention was performed on the dominant leg with testing on the non-dominant leg for the DOM group. The non-dominant leg received the conditioning intervention and the dominant leg was tested for the ND group.

The conditioning intervention consisted of four repetitions of five-second knee extension MVICs. The same equipment setup was used as with the testing MVIC. Participants were instructed to push as hard and fast as possible against the strap and were given verbal encouragement during each repetition. In order to ensure fatigue was not induced, one-minute of rest was given after the first and third repetition, and three-minutes of rest was given after the second repetition. The total duration of the intervention was 5 minutes and 20 seconds.

Testing protocol
The testing protocol consisted of two different unilateral exercises, a knee extension MVIC, and DJ test.

Knee Extension Maximum Voluntary Isometric Contraction (MVIC)
For the MVIC, the participant was seated on a specially made table with an adjustable backrest (constructed by Technical Services of Memorial University of Newfoundland). Researchers positioned participants so that when they were in an upright, seated, comfortable position, the backrest was flush against their back, and the crease of their knees was on the edge of the table. Restraints were put in place across the participant's chest and thighs to stabilize them. The tested leg was inserted in a strap, attached to a chain that was hooked into a load cell containing a Wheatstone bridge strain gauge (Omega Engineering Inc., LCCA 250, St. Eustache, Quebec, Canada) so that the knee was flexed at approximately 90° (visual inspection with goniometer). During the four second MVICs, the participant would cross their arms over their chest and be instructed to push against the strap as hard and fast as possible against the strap and were given verbal encouragement during each repetition. In order to ensure fatigue was not induced, one-minute of rest was given after the first and third repetition, and three-minutes of rest was given after the second repetition. The total duration of the intervention was 5 minutes and 20 seconds.

Peak force and instantaneous strength (F100: the force produced in the first 100 milliseconds of the MVIC: Grabow et al., 2018; Low et al., 2019) was recorded. EMG of the VL and BF was recorded, filtered using a bandpass filter and the root mean square of the rectified EMG over a one second period at the peak of the MVIC (500 ms before and after the peak force) was calculated using AcqKnowledge III software (Biopac Systems Inc., Holliston, MA). With both groups combined (DOM and ND), reliability using ICC (Cohen 1988) showed coefficients of 0.93 and 0.95 for MVIC force and 0.78 and 0.81 for F100 for the contralateral and conditioned legs respectively.

Unilateral Drop Jump (DJ)
For the unilateral DJ, the participant was positioned on top of a step with three pairs of risers underneath, totaling 25.4 cm in height. Participants stood on the untested leg, with the single leg to be tested positioned in front of the step, over the force plate (Advanced Mechanical Technology Inc; BP400600HF-2000, Watertown MA, USA) and arms in the akimbo position. Participants were instructed to drop onto the force plate passively, then once their foot made contact with the force plate, to jump straight up as quickly and as high as possible, before once again making contact with the force plate on the landing. Two unilateral DJ trials were performed; if participants did not follow the proper DJ protocol (i.e. jumped up rather than directly down from the platform or did a double hop upon landing), they were required to repeat the trial. If the second jump height was 5% better than the first jump, a third attempt was performed.

Initial contact time and flight time measured by the force plate were recorded from the highest jump. Using flight time, jump height was determined. Jump height = \( \frac{1}{2} g (t/2)^2 \), where \( g = 9.81 \text{ m/s}^2 \), \( t = \text{flight time in the air (s)} \). With both groups combined (DOM and ND), reliability using ICC (Cohen 1988) showed coefficients of 0.94 and 0.93 for DJ jump height and 0.82 and 0.84 for DJ contact time for the contralateral and conditioned legs respectively.

Statistical analyses
Statistical analyses were conducted in R (v 4.0.2; R Core Team, https://www.r-project.org/) and using the R software environment for statistical computing (Version 4.0.3). A linear mixed model using the lme4 package (Bates et al., 2015) was used across the following interaction variables 3 times (Pre-test, 1-min post, 10-min post) x 3 intervention leg variables (control, dominant and non-dominant) x 2 test legs variables (dominant, and non-dominant). Random intercepts by participant and random slopes for time were incorporated in the model. The package lmerTest (Kuznetsova et al., 2017) was used to produce type III ANOVA tables with Satterthwaite’s method. Significance was set at \( p \leq 0.05 \). If significant main or interaction effects were found, a simple planned contrast analysis was performed. Estimated marginal means and standardized (Cohen’s) effect sizes (d) were calculated using the emmeans package (Lenth, 2020) with effect sizes qualitatively interpreted as: trivial <0.2, small 0.2 ≤ d < 0.5; medium 0.5 ≤ d < 0.8; large: d ≥ 0.8 (Cohen 1988). Reliability was assessed with intraclass correlation coefficients (Cohen, 1988).

Analysis for this study was not pre-registered and treated as exploratory. The analysis of inferential statistics from the data generated from the participants should be treated as highly unstable local descriptions of the relations between model assumptions and data to acknowledge the inherent uncertainty in drawing generalised inferences from single samples (Amrhein et al., 2019). While \( p \) values and effect sizes are provided, we also consider the implications of all results compatible with these data, from the lower limit to the upper limit of interval estimates, with the greatest interpretive emphasis placed on the point estimates. Further, qualitative description of results based on visualisation of the data are also offered. All data and code utilised is presented in the supplementary materials.
Results

MVIC Peak Force
There were no significant main effects or interactions for MVIC peak force (Figure 2).

MVIC F100
A significant (p = 0.02) time x intervention leg demonstrated that the pre-test (0.50 ± 0.13) dominant leg normalized MVIC F100 forces exceeded both post-test (0.46 ± 0.13) and post-10 min (0.46 ± 0.13) by a small magnitude 8.7% (d = 0.31). There was also a significant (p = 0.02) time x intervention leg x testing leg intervention (Figure 2). However, when observing the individual interactions the control condition was as likely to demonstrate small to large magnitude changes as were the dominant and non-dominant legs. Cohen d effect size interaction are given at the Table 1.

Table 1. MVIC F100 Cohen d effect size interactions.

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<thead>
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<tr>
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<td>0.62</td>
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DOM: Dominant leg, ND: Non-dominant leg. CONTRA: testing of the contralateral leg, IPSIL: testing of the conditioned exercised leg.

MVIC EMG
There were no significant interactions for either the VL or BF EMG activity. A main effect for the intervention leg (p = 0.03) indicated that the dominant normalized VL EMG (0.37 ± 0.19) exceeded the non-dominant VL EMG (0.34 ± 0.23) by a trivial magnitude 8.8% (d = 0.19). The BF EMG exhibited a main effect for testing leg (p < 0.0001) with the contralateral leg (0.07 ± 0.03) exhibiting a moderate magnitude 22.2% (d = 0.5) less normalized EMG activity than the exercised leg (0.09 ± 0.05) (Figure 2).

Drop Jump (DJ) Contact Time
There were no significant leg x time interactions (Figure 2). There was a significant main effect of time on DJ contact time (p < 0.004). Contrasts revealed that compared to the pre-test, contact time increased (decreased performance) 3.7% at 1-minute (d = 0.22), and 3.4% at 10-minutes post-test (d = 0.21). A main effect for the intervention leg (p = 0.048) illustrated that the dominant leg (0.33 ± 0.03-s) exhibited a small magnitude 3.1% (d = 0.25) longer DJ contact time than the non-dominant leg (0.32 ± 0.05-s).

Drop Jump (DJ) Height
There were no significant leg x time interactions (Figure 2). A main effect for the intervention leg (p=0.04) illustrated that the dominant leg (0.13 ± 0.04 m) exhibited a small magnitude 8.3% (d = 0.28) higher DJ height than the non-dominant leg (0.12 ± 0.03 m).

Discussion

A major finding of this study was the lack of evidence for voluntary muscle performance enhancement (PAPE) or NLMF with the contralateral (non-exercised) legs. The conditioning activity induced fatigue with the F100 in the conditioned leg but no significant changes in the contralateral leg. These lack of contralateral (non-local) performance enhancements and NLMF are in accord with a recent meta-analytical review (Behm et al., 2021), which determined that when all neuromuscular fatigue measures from the reviewed studies were combined (muscle strength, power, and endurance measures), only trivial effects were evident.

Although the effect size estimate in the NLMF meta-analytical review (Behm et al., 2021) was close to zero, there were a number of studies that did exhibit significant NLMF with single discrete maximal contractions (Martin and Rattey, 2007; Ben Othman et al., 2019; Halperin et al., 2015; Kawamoto et al., 2014; Sidhu et al., 2014). However the significant NLMF effects observed in these studies were counterbalanced by more studies showing no significant changes. Thus the overall evidence for NLMF was too inconsistent to garner more than a trivial magnitude effect.

Two prior studies whose intention was to investigate contralateral effects of a unilateral conditioning exercise reported potentiation of the conditioned leg but significant deficits with the homologous, contralateral limb (Andrews et al., 2016) or no contralateral effect (Wong et al., 2020) respectively. Wong et al. (2020) used a 6-second MVIC as the conditioning stimulus and reported PAPE effects with the conditioned limb but not crossover effects. Andrews et al. (2016) used Bulgarian split squats (5 repetitions at 50% of 1-repetition maximum [1RM], 2 repetitions at 70% 1RM and 1 repetition at 90% 1RM with 3-min rest periods between sets) as the conditioning exercise and found post-intervention deficits to the countermovement jump and no change to the DJ in the contralateral limb. They suggested that the post-warm-up cooling due to inactivity (conditioning activity was performed by the other leg) may have reduced subsequent contralateral performance. Secondly, the combination of maximal (MVIC testing) or near maximal (Bulgarian squats conditioning intervention) contractions can lead to greater central fatigue than a submaximal effort (Behm, 2004). Without the benefit of the muscle potentiation effects of myosin phosphorylation (Houston et al., 1985; Houston and Grange, 1990; Hamada et al., 1997) to counterbalance fatigue effects, the significant NLMF-induced impairments in muscle endurance with recreationally trained individuals. It is not clear why two 100-s MVICs (intention of the conditioning exercise was to
elicit fatigue) elicited MVIC PAPE effects. Hamilton and Behm (2017) postulated that the enhancement was due to enhanced neural mechanisms such as firing frequency, synchronization and recruitment of higher order motor units. The lack of contralateral PAPE in the present study is in accord with Wong and colleagues (2020), but the literature using a unilateral conditioning exercise to induce contralateral PAPE is still scarce. Hence, this initial variability in the literature points to the need for further studies to examine the effect of varying conditioning exercise durations and workload to possibly elicit crossover PAPE effects.

In the Behm et al. (2021) NLMF meta-analytical review, between-study heterogeneity was high. It might be argued that a relatively similar scenario is illustrated with the present study in Figure 2. Whereas the mean changes in all measures do not achieve significance, there are many examples of individual participants with substantial

**Figure 2. Means, standard deviations (bolded) and individual participant data (shaded) plots for all measures.** X axis: Testing occurred at pre-test, and 1-min and 10-min post-test. CON: Control, D: Dominant leg, ND: Non-dominant leg Column titles: CONTRA: testing of the contralateral leg, IPSIL: testing of the conditioned exercised leg.
performance enhancement either at immediate post-test or 10-min post-test. Similar to the Behm et al. review (2021), these few examples of contralateral PAPE were counterbalanced by those individuals experiencing either no change or deficits (i.e. between participant heterogeneity). This finding is not uncommon in the PAPE literature (Chaouachi et al., 2011). For example, Chaouachi et al.’s (2011) initial analysis of their data did not present significant PAPE effects, but their further analysis demonstrated that a significant number of participants did experience PAPE but at different time points.

There are a few studies, that had relatively clear evidence of performance enhancements without decrements of the conditioned muscle (Baker, 2003; Gourgoulis et al., 2003; Gullich and Schmidtbleicher, 1996; Young et al., 1998), which contrast with the more common PAPE-type studies that also report fatigue-related decrements (Jensen and Ebben, 2003; Jones and Lees, 2003; Scott and Docherty, 2004). The main reason for this performance variability was thought to be related to the co-existence of fatigue and potentiation (Behm, 2004; Behm et al., 2004; Duthie et al., 2002; Rassier and MacIntosh, 2000). The magnitude of fatigue-induced force impairments can be substantial but of short duration, whereas PAPE-induced force increases may be less significant but persist for a longer duration (Chiu et al., 2003). As fatigue diminishes, performance enhancements can emerge. While this emergence may have occurred with some participants in the present study, the effect was not consistent. Furthermore, fatigue effects were more predominant when monitoring the F100 of the conditioned leg. This sparsity and variability in the literature points to the need for future research to examine the effect of varying conditioning exercise durations and workloads to possibly elicit crossover PAPE effects.

Limitations
As with all studies, limitations are inherent. Although the athletic and recreationally active participants were allocated under controlled randomized conditions, the strength of each group (DOM and ND) was not monitored and differences in the groups were later observed, which contributed to heterogeneity. Although the 32 participants (16 per group) were sufficient to achieve statistical power according to an “a priori” statistical power analysis, greater numbers of participants could have increased that power and permitted further comparisons of the effect of the participant’s trained state on PAPE. Although including groups of recreationally and athletically trained participants provides a broader subject base, the lack of training status homogeneity of the participants could have increased data variability adversely affecting the ability to achieve significant findings. While the initial five second pre-test MVC might contribute as a conditioning activity, it was included in the control session and thus differences between the experimental and control conditions would only be attributed to the four MVICs conditioning protocol.

Conclusion
The present study did not provide significant or consistent NLMF and non-local PAPE effects. The lack of either effect (NLMF or PAPE) may due to the co-existence of fatigue and potentiation. For future research to find more convincing findings of non-local potentiation, it is important to continue to explore the coexistence of potentiation and fatigue, particularly as it relates to unconditioned limbs. Therefore, it is important for future non-local potentiation studies, that participant characteristics that contribute to potentiation be better understood and that study designs implement a method for better individualizing conditioning and testing protocols.

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References


Key points
- The present study did not provide significant or consistent NLMF and non-local PAPE effects.
- The lack of either effect (NLMF or PAPE) may due to the co-existence of fatigue and potentiation.
- The lack of contralateral performance enhancements and NLMF are in accord with the recent meta-analytical review (Behm et al., 2021), which determined that when all neuromuscular fatigue measures from the reviewed studies were combined (muscle strength, power, and endurance measures), only trivial effects were evident.

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