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Effects of three neuromuscular electrical stimulation methods on muscle force production and neuromuscular fatigue

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This study compared the acute responses of three neuromuscular electrical stimulation (NMES) methods on muscle torque-time integral (TTI) and neuromuscular fatigue. Narrow-pulse (0.2 ms; NP), wide-pulse (1 ms; WP), and tendon vibration superimposed onto wide-pulse (WP + VIB)-NMES conditions were applied to sixteen healthy individuals ($n = 16$) in three separate sessions in a randomized order. Stimulation intensity was set to elicit 20% of maximal voluntary contraction (MVC); the stimulus pattern comprised four sets of 20 repetitions (5 s On and 5 s Off) with a one-minute inter-set interval. TTI was measured for each NMES condition and MVC, voluntary activation (VA), peak twitch torque ($Peak_{twitch}$), and peak soleus (EMG_{SOL}), medial (EMG_{MG}), and lateral gastrocnemius (EMG_{LG}) electromyography were measured before and immediately after each NMES condition. TTI was higher during WP + VIB (19.63 ± 6.34 MVC.s, mean difference = 3.66, $p < 0.001$, Cohen's d = 0.501) than during WP (15.97 ± 4.79 MVC.s) condition. TTI was higher during WP + VIB (mean difference = 3.79, $p < 0.001$, Cohen's d = 0.626) than during NP (15.84 ± 3.73 MVC.s) condition. MVC and $Peak_{twitch}$ forces decreased ($p \leq 0.001$) immediately after all conditions. No changes were observed for VA ($p = 0.365$). EMG_{SOL} amplitude reduced ($p = 0.040$) only after NP, yet EMG_{LG} and EMG_{MG} amplitudes decreased immediately after all conditions ($p = 0.003$ and $p = 0.013$, respectively). WP + VIB produced a higher TTI than WP and NP-NMES, with similar amounts of neuromuscular fatigue across protocols. All NMES protocols induced similar amounts of peripheral fatigue and reduced EMG amplitudes.

KEYWORDS

motor unit recruitment, muscle fatigue, muscle force, muscle stimulation, muscle strength, rehabilitation, stimulus pulse width, triceps surae

1 | INTRODUCTION

Neuromuscular electrical stimulation (NMES) elicits muscle contractions by involuntarily activating muscles, stimulates the physiological benefits, attenuates muscle mass loss, strength, and functional capacity as a rehabilitative tool.[1] NMES recruits motor units in a non-physiological order when conventional (0.05–0.4 ms) narrow-pulse (NP) stimuli are used.[2] NP typically causes the direct depolarization of motor axons and activates a large proportion of fast-fatigable motor units.[3] This type of recruitment results in rapid muscle fatigue, which is a limiting factor for clinical practice due to its limited ability to increase training volume.[3,4] It has been argued that increasing training volume is the most effective way to induce muscular and health adaptations in resistance-type exercises.[5] Thus, it is of clinical importance to develop NMES methods that can delay muscle fatigue and increase torque-time integral (TTI).

The use of wide-pulse (1 ms, WP) stimulation appears to be an alternative method due to the indirect (i.e., central) recruitment of motor units[3] or asynchronous activation of motor units via spinal reflex pathways.[6,7] The involvement of spinal reflexes in electrically-induced contractions favors the recruitment of lower-threshold (i.e., fatigue-resistant) motor units, which could delay muscle fatigue.[8] Also, although debatable,[9] it has been suggested that WP stimuli may trigger the development of persistent inward currents (PICs),[10] through repetitive activation of Ia afferents resulting in the production of extra forces.[6,11] Collectively, these findings led researchers to hypothesize that WP could produce larger TTI (i.e., less fatigue and more force) than NP. However, WP has been shown to elicit greater fatigue than NP,[12] suggesting that WP by itself may not be a viable option to delay fatigue and increase TTI.

Tendon vibration (VIB) has been suggested as a possible strategy to enhance force production whilst minimizing muscle fatigue by producing greater excitation of the fatigue-resistant motor units,[13] especially when coupled with WP.[2,14,15] VIB evokes a tonic vibration reflex through both spinal and supraspinal pathways by generating trains of Ia-afferent signals to the spinal cord to induce an excitation of homonymous motor neurones, and possibly promoting the development of PICs.[7,15,16] When activated, PICs can amplify and prolong synaptic input and create sustained depolarization, leading to the increased and sustained physiological recruitment of motor units and thus increasing muscle force output.[17] Consequently, it is possible that VIB superimposed onto WP (WP + VIB) could increase muscular TTI and delay neuromuscular fatigue in comparison with WP alone.

To date, studies have not found clear differences in muscular TTI and fatigue between WP with and without the inclusion of VIB in quadriceps muscles.[2] However, several studies have reported large extra forces in triceps surae muscle after adding VIB[15] but with no direct comparison to other NMES methods.[14,16] Therefore, it is possible that WP + VIB may produce higher TTI and delay fatigue in triceps surae muscles. The objectives of this study were to compare the responses of three NMES methods on plantar flexor TTI and neuromuscular fatigue. It was hypothesized that the use of WP + VIB would increase TTI and delay neuromuscular fatigue after stimulation when compared with NP and WP.

2 | MATERIALS AND METHODS

2.1 | Participants

Sixteen healthy adults (13-males) with no neurological or musculoskeletal disorders volunteered to participate in this study (mean ± SD, age: 29.9 ± 2.57 years.; height: 172.6 ± 9.27 cm; body mass: 75.1 ± 15.51 kg; BMI: 25.1 ± 4.29 kg/m^2). Participants refrained from vigorous exercises (48 h) and consuming stimulants (12 h) before testing. Participants were given detailed information about the procedures and risks of participation, reading, and signing their informed consent. This study was approved by the university's human research ethics committee (1900000372), and it was designed as an experimental crossover study.

2.2 | Procedures

Participants attended the laboratory on four occasions. Each testing session was separated by at least 48 h, but no longer than a week. In the first session, participants were familiarized with NMES methods (NP, WP, and WP + VIB$_)$ and performed maximal voluntary isometric contractions (MVC) of the ankle plantar flexors to ensure they could tolerate the protocols. In the following sessions, participants performed the three experimental conditions in a randomized order. All neuromuscular tests were performed immediately before (Pre) and after (Post) each of the experimental conditions. Participants were seated with hip flexion at 55°, knee extension at 0°, ankle dorsiflexion at 5°. The thigh and trunk were secured to the dynamometer chair and the ankle joint aligned with the centre of dynamometer rotation. A standardized warm-up protocol (six-submaximal isometric contractions at 40%, 60%, and 80% of perceived maximal effort as warm-up)

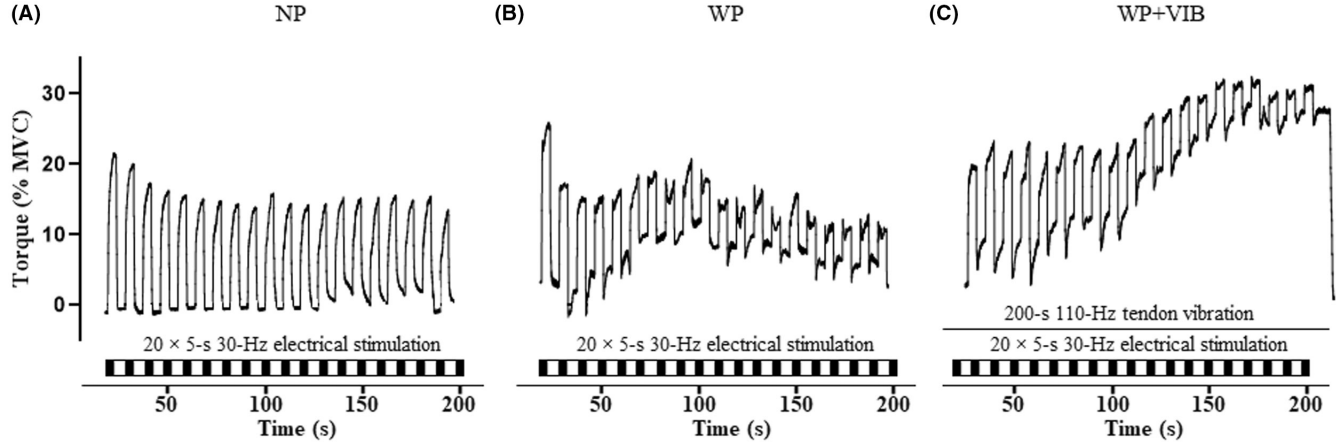

FIGURE 1 Illustration of the torque trace obtained from a single participant during the first set for the following experimental NMES conditions: (A) NP, (B) WP, and (C) WP + VIB. Panel A and B showed a torque trace of contractions elicited by narrow- and wide-pulse NMES without the superimposition of vibration. Panel C showed contractions when tendon vibration was superimposed to wide-pulse NMES. Note that torque trace went back (or close) to baseline during the off phase of the duty cycle of panels A and B. In panel C, it was clear how the vibration elicited sustained contractions during the off phase of the duty cycle that progressively got larger with repetition helping to increase the total torque-time integral

was performed before testing on the isokinetic dynamometer (Biodex Medical Systems,). Three MVCs separated by 30 s rests were performed before and one MVC after each NMES condition.

2.3 | Neuromuscular electrical stimulation and tendon vibration protocols

Electrical stimulation was delivered by a high-voltage-constant-current electrical stimulator (400 V, DS7AH-1580-2010, Digitimer Ltd.,) via 2–7.5 × 10 cm rectangular self-adhesive neurostimulation electrodes (Axelgaard, PALS,). The proximal electrode (cathode) was placed transversely over the prominent bulge ~10 cm distal to the popliteal fossa and the distal electrode (anode) was placed transversely over the musculotendinous junction (gastrocnemius-Achilles) of the triceps surae. Electrodes were carefully placed on the skin to elicit the greatest twitch response with the lowest stimulation intensity; the same electrode placement was maintained in the following sessions by marking the electrode margins with a pen.

NP stimulation pattern consisted of repeated 30 Hz trains (0.033 s inter-pulse interval) of 150 narrow (0.2 ms) square-wave pulses. WP consisted of the same stimulation pattern as NP with a wider pulse (1 ms). Single-train duration was 5 s and the inter-train interval 5 s (duty cycle: 5 s On and 5 s Off). The stimulation protocol comprised four sets of 20 trains with a 1 min rest between sets. This between-set resting time was provided to ensure the muscle had recovery time and to mimic the rest period of a resistance training program. NMES intensity was chosen using a 1 s 30 Hz train of electrical stimulation that could elicit a contraction of 20% MVC for all experimental conditions. The WP + VIB condition consisted of the same stimulation protocol as WP with tendon vibration (sinusoidal wave) applied simultaneously by a vibrator (Vibrasens Proprioceptive Technology, France) at 110 Hz and constant amplitude (1 mm peak-to-peak vibration) using a VB115 hand-free applicator. The Achilles tendon vibration elicited involuntary contractions in thirteen out of the sixteen participants during the off phase of the duty cycle in WP + VIB condition. Vibration was initiated 5 s before the first train of electrical stimulation, delivered and maintained throughout the whole set but stopped in the rest interval between sets. The vibrator was positioned distally, fixed at the posterior aspect of the Achilles tendon at the level of the medial malleolus and strapped around the ankle. This position was then marked on the skin to avoid changes in placement during the stimulation.

2.4 | Data collection and analysis

2.4.1 | Peak torque and electrically evoked torque-time integral

During the three experimental sessions, plantar flexion peak torque assessed during MVCs measured fatigue and normalized the torque elicited by NMES. Peak torque was defined as the maximum torque produced over a 500 ms window and included the plateau phase

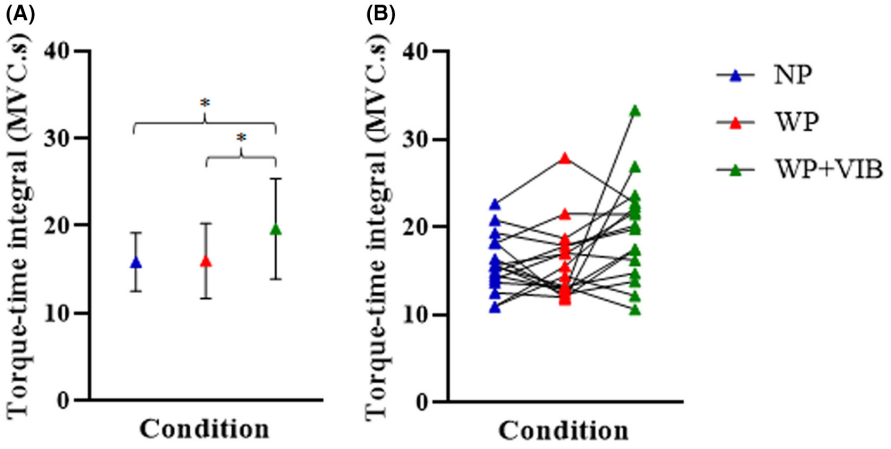

FIGURE 2 Normalized torque-time integral (TTI). A) Group results measured during the three NMES conditions (NP, WP, WP + VIB) (mean ± SD). B) Individual results. *Significant difference from WP and NP ($p < 0.05$)

after at least 250 ms rise time above baseline. TTI was used to measure the training volume received by the muscle in each NMES condition (Figure 1). TTI calculated the torque-time trace as the area under the curve, from the onset of the first stimulation train to the final evoked contraction of each set. TTI of each of the four-sets was summed to provide a total TTI measure; this was normalized by the peak torque value recorded before NMES, accounting for variations in peak torque between sessions.

2.4.2 | Voluntary activation level and peak twitch torque

Voluntary activation (VA) was used to estimate the amount of central fatigue induced by each protocol and was assessed using the interpolated twitch technique through a single supramaximal electrical stimulus during MVC. VA reflects the central nervous system's ability to drive muscle, quantified as a percentage using: VA(%) = (1–superimposed twitch/potentiated twitch) × 100.[18] A superimposed twitch was delivered at the MVC plateau phase, and a potentiated twitch evoked by the same stimulus in the potentiated relaxed muscle 3 s after MVC. A resting twitch was evoked by a single stimulus in the unpotentiated relaxed muscle 3 s before MVC. To obtain information about peripheral/muscle fatigue, peak twitch ($Peak_{twitch}$) torque was measured before and after each experimental condition. To determine the $Peak_{twitch}$ torque before the experiment began, an electrical stimulus was delivered to the muscle belly of the right Gastrocnemii every 10 s while the current was increased from 20 to 99 mA in 10 mA increments until a plateau in the maximum $Peak_{twitch}$ was observed. The current was increased 20% after observing the plateau and this intensity was maintained throughout the session.

2.4.3 | Muscle activity recording

Electromyography (EMG) recorded the myoelectrical activity of plantar flexor muscles during MVCs and was sampled at an analog-to-digital conversion rate of 4000 Hz using a Quad Bio Amplifier (PowerLab System, ADInstruments,). The skin was carefully shaved, abraded, and cleaned with 70% alcohol before electrode placement. Bipolar electrodes (N-00-S/25, Ambue blue sensor-N, Ambu Australia,) with a 1-cm inter-electrode distance were attached to the skin over the muscle's belly parallel to the direction of muscle fibers, following SENIAM recommendations. The EMG signal was bandpass-filtered (10–500 Hz), and peak EMG was retained for analysis.[19] Muscle activity was calculated as the root mean square of the EMG amplitude, measured for the soleus (EMG_{SOL}), lateral (EMG_{LG}), and medial gastrocnemius (EMG_{MG}) muscles over the same time as the torque measurements. Ankle torque measurements and EMG data were simultaneously recorded using LabChart version-8.1.16 Software (PowerLab System, ADInstruments,) at the same analog-digital conversion rate.

2.5 | Statistical analysis

Statistical analyses were performed using Jamovi-1.1.9.0 software (version-11, jamovi Project,), with significance set at $p \leq 0.05$. TTI was compared within and between conditions using a two-way repeated-measures ANOVA (condition vs. set). MVC, VA, and $Peak_{twitch}$ were

compared within and between conditions using a two-way repeated-measures ANOVA (condition vs. time). EMG_{SOL}, EMG_{LG}, and EMG_{MG} were separately analyzed by two-way repeated-measures ANOVA (condition vs. time), α-value set at 0.05. Tukey Post Hoc tests were performed when significant two-way interaction effects were found. Values were reported as the mean and standard deviation (mean ± SD).

3 | RESULTS

3.1 | Torque-time integral (TTI)

No significant interaction ($p = 0.629$) was observed between condition and sets for TTI; nonetheless, a significant condition effect ($p = 0.009$, $F_{2,3} = 5.54$, $\eta p2 = 0.270$) was observed (Figure 2). TTI was higher during WP + VIB (19.63 ± 6.34 MVC.s, mean difference = 3.66, $p < 0.001$, Cohen's d = 0.501) than during WP (15.97 ± 4.79 MVC.s) condition. TTI was higher during WP + VIB (mean difference = 3.79, $p < 0.001$, Cohen's d = 0.626) than during NP (15.84 ± 3.73 MVC.s) condition.

3.2 | Maximal voluntary isometric contraction (MVC) torque and peak twitch ($Peak_{twitch}$) torque

No interaction effect ($p = 0.988$) was observed for MVC torque; however, a significant time effect ($p \leq 0.001$, $F_{1,15} = 22.38$, $\eta p2 = 0.599$) was found. Reductions were found after all conditions. WP + VIB reduced MVC torque (12.45 ± 6.99 Nm), WP reduced MVC torque (12.19 ± −0.45 Nm), and NP reduced the MVC torque (12.7 ± 3.81 Nm; see Table 1). Additionally, no interaction effect ($p = 0.795$) was observed for $Peak_{twitch}$ torque; nonetheless, a significant time effect ($p \leq 0.001$, $F_{1,15} = 22.68$, $\eta p2 = 0.602$) was found. Reductions were found after all conditions. WP + VIB reduced $Peak_{twitch}$ (1.78 ± 0.77 Nm;), WP reduced $Peak_{twitch}$ (2.79 ± 0.83 Nm;), and NP reduced $Peak_{twitch}$ (2.44 ± 0.23 Nm; see Table 1).

3.3 | Voluntary activation (VA) level and muscle activity (EMG) amplitude

VA levels showed neither significant interaction ($p = 0.365$, $F_{2,30} = 1.04$, $\eta p2 = 0.065$) nor time effects ($p = 0.092$). However, a significant interaction ($p = 0.038$, $F_{2,30} = 3.65$, $\eta p2 = 0.196$) was observed for EMG_{SOL} amplitude. Post hoc analysis showed a reduction in EMG_{SOL} amplitude in only NP condition ($p = 0.040$). No interaction effects ($p = 0.103$ and $p = 0.065$) were observed for EMG_{LG} and EMG_{MG} amplitudes, yet significant time effects were observed, indicating EMG_{LG} ($p = 0.003$, $F_{1,15} = 12.55$, $\eta p2 = 0.456$) and EMG_{MG} ($p = 0.013$, $F_{1,15} = 8.04$, $\eta p2 = 0.349$) reduced similarly across all conditions (see Table 1).

TABLE 1 Maximal voluntary isometric contraction, peak twitch torque, voluntary activation level and surface EMG amplitudes

	PRE	POST
Measure	**Mean ± SD**	**Mean ± SD**
MVC (Nm)		
NP	133.32 ± 26.78	120.59 ± 22.96
WP	131.48 ± 24.63	119.21 ± 25.1
WP + VIB	125.97 ± 29.95	113.42 ± 22.96
Peaktwitch (Nm)		
NP	27.28 ± 4.38	24.84 ± 4.14
WP	27.22 ± 4.46	24.42 ± 3.63
WP + VIB	24.72 ± 6.43	22.94 ± 5.67
VA (%)		
NP	94.88 ± 4.72	93.12 ± 6.37
WP	94.78 ± 4.70	93.45 ± 5.86
WP + VIB	93.92 ± 5.89	93.87 ± 5.22
RMS EMGSOL		
NP	0.35 ± 0.29	0.19 ± 0.10*
WP	0.28 ± 0.18	0.21 ± 0.16
WP + VIB	0.20 ± 0.11	0.25 ± 0.22
RMS EMGLG (mV)		
NP	0.36 ± 0.21	0.22 ± 0.09
WP	0.31 ± 0.16	0.21 ± 0.07
WP + VIB	0.25 ± 0.12	0.27 ± 0.16
RMS EMGMG (mV)		
NP	0.38 ± 0.18	0.28 ± 0.10
WP	0.32 ± 0.16	0.22 ± 0.11
WP + VIB	0.23 ± 0.10	0.27 ± 0.19

Abbreviations: EMG, electromyography; EMG_{LG}, electromyography of lateral gastrocnemius; EMG_{MG}, electromyography of medial gastrocnemius; EMG_{SOL}, electromyography of soleus; mV, millivolt; MVC, maximal voluntary isometric contraction; $Peak_{twitch}$, peak twitch torque; RMS, root mean square; VA, voluntary activation level.

*Significant difference within condition from Pre to Post ($p < 0.05$).(Mean ± SD) at Pre and Post for NP, WP, and WP + VIB-NMES conditions.

4 | DISCUSSION

This study investigated the acute responses of three NMES methods (WP + VIB, WP, and NP) on TTI, MVC, $Peak_{twitch}$, VA, and EMG. We investigated whether Achilles tendon

vibration superimposed onto WP increases TTI of the ankle plantar flexors, leading to less fatigue than the WP and NP conditions. The augmentation in TTI was higher during WP + VIB; therefore, the tendon vibration superimposed onto WP provided additional benefits perhaps not derived from stimulations alone. Neuromuscular fatigue, as revealed by the reduction in MVC and $Peak_{twitch}$ forces with no changes in VA levels, was largely the result of peripheral rather than central changes and was similar in all NMES conditions. Similarly, EMG_{LG} and EMG_{MG} amplitudes reductions followed all conditions, yet EMG_{SOL} decreased only in the NP condition.

The higher TTI observed during WP + VIB than only WP or NP could be advantageous in clinical practice, allowing muscles to produce greater tension for longer periods thus evoking chronic increases in muscle mass, strength, and health outcomes as training volume has been argued to be the most important variable to promote these adaptations.[5] This result was consistent with previous observations of higher force levels (≤50% MVC) elicited by tendon vibration applied simultaneously with electrical stimulation in healthy participants.[14] The augmented TTI might be attributed to the development of PICs in motor neurone dendrites. PICs amplify and prolong synaptic input and generate sustained depolarisation of α-motor neurons, leading to increased recruitment of fatigue-resistant motor units and maximizing the use of reflexive pathways.[11,14,17] Thus, the development of PICs could have increased muscle force generation capacity during electrical stimulation and caused tonic vibration reflexes to occur in between muscle-evoked contractions when only superimposed tendon vibration was applied.

The current results showed that MVC forces decreased similarly after all NMES conditions. These reductions were not influenced by the values obtained before protocols, as these values were not significantly different ($p = 0.912$) and were reliable between days of intervention (ICC = 0.974). Reducing EMG amplitudes mean neuromuscular fatigue is similar between the three conditions. However, the WP + VIB condition elicited the same fatigue with a greater training volume, possibly because the recruitment of fatigue-resistant motor units in addition to higher-threshold motor units was sufficient to elicit strong muscle contractions, contributing to higher force levels.[20] WP + VIB may be beneficial to achieving further muscular training with the same level of fatigue caused by other conditions. Our results can not explicitly explain how WP + VIB could provide fatigue-attenuation benefits. Although it has been shown that tendon vibration significantly reduced the synchrony of motor unit activity, decreasing the rate of muscle fatigue,[21] the present results should be interpreted with caution since WP + VIB did not show considerable muscle fatigue attenuation. In addition, reductions in MVC strength of approximately 9%–29% were reported due to prolonged electrically stimulated isometric contractions of the ankle plantar flexors,[12,22] and knee extensors,[23] and these results are similar or higher than the reductions in MVC observed in the present study (~9%–10%). Potential discrepancies in MVC force loss may arise from using different waveforms, lower stimulation frequencies or different duty cycle ratios, muscle groups, or the level of activation to only 20% MVC in this study, compared with each individual's maximal tolerated current level.[24]

Almost identical reductions in MVC forces were observed following NMES conditions. Our findings support the origin of NMES-induced fatigue to be predominantly peripheral, evidenced by decreased $peak_{twitch}$ torque with no changes in VA.[25] $Peak_{twitch}$ torque is commonly used to evaluate peripheral fatigue, and our findings showed reduced $peak_{twitch}$ torque after NMES protocols, supporting the work of prior studies linking peripheral fatigue induced by NMES with a comparable MVC force loss.[8,26] Reductions in muscle force may be caused by skeletal muscle fatigue from local changes or processes at or distal to the neuromuscular junction,[10] such as metabolic changes.[12,27] A prior study evaluating neuromuscular fatigue and contractile changes during electrical stimulations observed impairment of excitation-contraction coupling, which led to muscle contractile failure.[28] This condition causes peripheral muscle fatigue in different forms, such as reduced adenosine triphosphate (ATP) and phosphocreatine supply and accumulated lactate and intracellular hydrogen ion concentration (H+).[27] Furthermore, prolonged periods of NMES can cause a reduction in Ca^{2+} release from the sarcoplasmic reticulum, thus causing an impairment in excitation-contraction coupling processes, which could decrease muscle force production.[27] The results of the present study support the hypothesis that when NMES protocols are performed at low force levels (20% MVC) and for prolonged durations (~13 min), contractile alterations were the main contributors to muscle fatigue.

VA was measured to assess the completeness of muscle activation during voluntary contractions before and after the experimental protocols.[18] This study did not find significant changes in VA levels, consistent with a previous study,[12] yet changes in EMG amplitudes were significant following NMES conditions. Reductions in EMG amplitudes were in accord with previous work, in which neuromuscular fatigue was ascribed to central and peripheral factors,[22] as shown by the significant decline in EMG_{SOL} (−11%)[22] and current reduction in EMG_{SOL} (−43.4%) following NP. However, reductions in EMG amplitude in isolation (as observed in the present study) cannot be taken as evidence of central fatigue as changes in EMG might also be affected by peripheral factors if not normalized

by the M-wave amplitude.[22] Thus, because there was no change in VA, these findings suggest reductions in central drive did not contribute to reductions in MVC torque. In conclusion, WP+VIB protocol allowed for a greater TTI response to electrical stimulation for the plantar flexor muscles. Also, all NMES protocols produced the same amount of neuromuscular fatigue. Whether or not this would result in an improved training effect should be explored in future research.

5 | PERSPECTIVE

This study was designed to determine the acute effects of three NMES interventions on muscle force production and neuromuscular fatigue in healthy individuals. This study has shown that the protocol of moderate-frequency wide-pulse width NMES coupled with high-frequency tendon vibration allowed for a greater amount of muscular work, elicited by the increase in TTI compared with the use of wide-pulse or narrow-pulse NMES alone. The study has also shown that all NMES protocols elicited the same amount of voluntary fatigue experienced after the stimulation session for the triceps surae muscle. The results of this study indicate that the peripheral rather than central processes are the main mechanisms of neuromuscular fatigue after all NMES protocols. These results suggest that the use of tendon vibration superimposed onto wide-pulse NMES should be used when the goal is to maximize muscle force production during NMES training and rehabilitation protocol in future clinical research. This study lays the groundwork for future clinical practice using the tendon vibration superimposed onto wide-pulse NMES in patient populations who are immobilized or unable to exercise. The findings will be of interest to the fields of rehabilitation and disease relating to sport, exercise, and physical activity.

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CONFLICT OF INTEREST

The authors declare no pecuniary or other personal interest, direct or indirect, in any matter that raises or may raise a conflict of interest.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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