THESIS

NEUROMUSCULAR ELECTRICAL STIMULATION: GETTING AMPED UP TO PREVENT EXERCISE RESISTANCE

Submitted by

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ABSTRACT

NEUROMUSCULAR ELECTRICAL STIMULATION: GETTING AMPED UP TO PREVENT EXERCISE RESISTANCE

Purpose: Exercise resistance (ER) is characterized by the absence of exercise induced improvements in fat metabolism following a meal. The prolonged sedentary behavior between successive workouts is what contributes to this lack of health improvements typically associated with exercise. The suggested energy expenditure (EE) threshold for avoiding ER is the equivalent of walking ~8,500 steps/day. Population data indicate that the typical adult walks 5,000 steps/day. Neuromuscular Electrical Stimulation (NMES) evokes skeletal muscle contractions and increases EE. This study aims to determine the feasibility of using NMES to increase total daily EE by the equivalent of 3,500 steps, thereby meeting the theoretical threshold sufficient to prevent ER. Methods: Fourteen recreationally active males and females (7/7) underwent measures of resting EE, with and without NMES, and EE while walking 8,500 steps on a treadmill. The duration of NMES sufficient to increase EE to match 3,500 steps was calculated, and then verified with measures of EE while walking 5,000 steps after a bout of NMES. Results**:** Bland–Altman statistics of agreement were used to assess concordance between the EE associated with walking 8,500 steps (286 ± 64 kcal; mean ± SD), and the EE associated with walking 5,000 steps after NMES (293 ± 65 kcal). The mean difference between the EE values was 7 kcal, and the 95% limits of agreement were -39 to 53 kcal. Implications**:** These preliminary data suggest NMES can be used to increase total daily EE by the equivalent of 3,500 steps, thereby meeting the theoretical threshold to prevent ER.

ACKNOWLEDGEMENTS

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I have many people I would like to thank for getting me this far in my career. To past and present members of the Integrative Biology Laboratory: thanks for your continued support and encouragement. To my family: thanks for always believing in me and for teaching me to ask, “how can I help?” It has gotten me far in life. To Hannah: thank you for putting up with late nights and plucking out the gray hairs this project has sprouted on my head. To Dr. Bell: thank you for teaching me how to ask questions and explore the field of physiology. To my committee members: thank you for taking interest in my research and helping me grow as a writer. To HES: thank you for being my family these past 6 years.

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REVIEW OF LITERATURE

Exercise Resistance

Habitual exercise bestows many benefits to health [1-3]. These benefits are mediated via cumulative effects of repeated activity, and/or as acute effects from the most recent exercise bout; they can improve numerous physiological systems across multiple regulatory levels [4]. One of the notable metabolic benefits of regular exercise is increased post meal triglyceride clearance [5]. This has clinical relevance because an acute impairment in triglyceride clearance can lead to cardiovascular dysfunction, problems with glucose clearance, may increase blood pressure, and is predictive of future cardiovascular disease [6]. These temporary impairments can be insignificant for long term health when sporadic, but if they become chronic, the likelihood of developing metabolic syndrome increases [7]. In normal instances, acute bouts of exercise can remedy the physiological and metabolic impairments associated with decreased physical activity and poor post meal triglyceride clearance. However, there are some instances in which the acute effects of exercise do not bestow full health benefits; this is known as “exercise resistance” and usually occurs if consecutive exercise bouts are separated by several days of inactivity.

Exercise resistance has garnered appreciable attention in recent scientific literature [8]. The condition is characterized by a distinctive lack of improvement across various physiological systems, such as postprandial fat metabolism and insulin sensitivity, despite completion of acute exercise [9-12]. An understanding of the underlying mechanisms and factors contributing to exercise resistance is crucial, not only for refining exercise prescriptions, but also for tailoring interventions aimed at optimizing metabolic outcomes. Generally, exercise improves human health. However, if extended physical inactivity counteracts the benefits of exercise, then under certain conditions, the perceived cost-benefit of exercise may become problematic. To identify strategies to prevent exercise resistance, we must first consider the current state of knowledge pertaining to exercise resistance, explore the implications for metabolic health, and seek potential avenues for targeted interventions.

Dr. Edward Coyle has provided considerable insight into the mechanisms of exercise resistance. In one of his studies, daily step count was identified as a significant determinant of the postprandial plasma triglyceride response the day following an acute exercise bout [13]. The total area under the triglyceride curve (AUC_T) was lowest (i.e. most beneficial) when study participants walked approximately 8,500 steps/day and exercised for one hour at 65% of maximal oxygen uptake ($\dot{V}O_2max$). Participants taking fewer than 5,000 steps/day displayed a 16% – 19% decrease in fat oxidation (estimated using respiratory exchange ratio) and a 22% decrease in postprandial plasma triglyceride clearance compared to those walking 8,500 steps/day, thereby contributing to a greater AUC_T (i.e. an unhealthy response). This work implies that a low number of daily steps attenuates the metabolic improvement typically seen following an acute bout of exercise.

These results have been replicated in short-term training interventions. Two groups of study participants underwent the same short-term training regimen, but one group was assigned a high daily step count while the other was assigned a lower step count. Training coupled with a low daily step count failed to improve fat metabolism or show favorable changes in heart rate or perceived exertion during exercise; conversely, these variables improved when training was paired with a higher daily step count [14]. Individuals with a low step count did not show improvements in postprandial plasma triglyceride clearance after acute exercise. Besides attenuating the benefits of acute exercise, these data suggest that exercise resistance may also negatively impact the response to short-term training.

Noteworthy, there appears to be some controversy as to how best to demonstrate exercise resistance. For example, in one study, postprandial metabolism following one hour of soccer was compared when the game was played after two rest days (i.e. no activity) or after two days that each included one hour of soccer. Postprandial triglyceride AUC_T and insulin sensitivity was improved for the soccer players who played three matches in a row [15]. However, when postprandial triglyceride was expressed as incremental AUC, only the group that played one game of soccer improved triglyceride clearance. This raises a controversial question in the field of post-prandial triglyceride clearance: which is the more clinically relevant measure of triglyceride clearance, AUC_T or incremental AUC [16, 17]? In favor of AUC_T, it could be argued that the absolute amount of circulating triglycerides over a longer period of time, rather than the relative increase, represents a greater contribution to the development of cardiovascular disease [18]. Furthermore, most studies demonstrating exercise resistance have expressed the postprandial triglyceride clearance as AUC_T while only a few used incremental AUC [9, 13, 14, 19-21]. For future studies of exercise resistance, it may be good practice to measure both incremental AUC and AUC_T.

Considering the clinical relevance of exercise resistance, avenues to understand the best mechanism to target for intervention are being pursued. While the full scope of exercise resistance's cellular etiology remains unclear, there have been suggestions regarding appropriate physiological targets. One such target is skeletal muscle lipoprotein lipase (mLPL), a key determinant of plasma triglyceride clearance; reduced skeletal muscle contractile activity leads to decreased mLPL activation [22, 23]. Thus, increasing muscle contractions throughout the day with activities like walking may be one mechanism to prevent exercise resistance. Another mechanism involves Adenosine Monophosphate-activated protein Kinase (AMPK), often referred to as the "master metabolic regulator," responsible for various anabolic and catabolic processes and mitochondrial biogenesis within skeletal muscle [24]. Previous research has found that training status can alter the skeletal muscle expression of AMPK and thus, sensitivity to insulin following an acute bout of exercise [25]. This suggests that brief bouts of physical inactivity in habitual exercisers can prevent acute exercise mediated improvements in carbohydrate metabolism. While AMPK activation appears linked to exercise intensity, less intense activity over a prolonged period can also enhance AMPK activation [24] . Interventions aimed at preventing exercise resistance should potentially target one or both mechanisms to achieve optimal results in overcoming its formation.

Through the research of Dr. Coyle and others, the threshold required to avoid exercise resistance is 8,500 steps/day plus one hour of exercise. Unfortunately, epidemiological data suggest that the average step count per adult in the US is approximately 5,000 steps/day [26]. This 3,500-step misalignment between the recommended threshold and actual physical activity levels is suggestive of the potential prevalence of exercise resistance on a broad scale.

Populations at Risk for Exercise Resistance

Cancer survivors are susceptible to cancer related fatigue, “a distressing, persistent, subjective sense of physical, emotional, and/or cognitive tiredness or exhaustion related to cancer or cancer treatment that is not proportional to recent activity and interferes with functioning,” and can lead to lower physical activity levels [27]. Due to this fatigue, cancer survivors represent a challenging population to study and are likely at higher risk for exercise resistance. One study has looked at the daily step count of cancer patients, and while it was estimated to be around 8,000 steps/day [28]. However, some cancer survivors experience a positive energy balance due to cancer related fatigue, leading to lower levels of daily activity. This suggests a higher amount of energy expenditure may be needed to overcome exercise resistance [29]. Furthermore, cancer survivors have varied success reaching standard physical activity guidelines [30]. These cumulative side-effects imply cancer survivors are more susceptible to exercise resistance and should be a considered population for interventions.

Individuals with low occupational physical activity are also at heightened risk of exercise resistance. Individuals in occupations such as teaching, law, tech support, and medicine typically involve prolonged sedentary periods of 7 to 9 hours daily, contributing to higher mortality and obesity rates [31, 32]. This sedentary behavior promotes exercise resistance and underscores the importance of physical activity during leisure time. Some individuals in these roles adopt "weekend warrior" habits, engaging in physical activity outside of work hours, which can reduce mortality risk [33]. However, prolonged sitting for 2 to 4 days can nullify the benefits of such activity, failing to improve post-prandial triglyceride clearance and potentially compromising metabolic health [20].

Exercise Snacks

Considering the environment of the typical workplace, it's challenging for individuals to sufficiently increase physical activity during working hours to combat exercise resistance. "Exercise snacks" offer a potentially feasible solution [34]. These are brief bouts (< 5-minutes) of exercise performed at regular intervals (e.g. hourly) aimed at interrupting prolonged sedentary behavior and increasing muscle contractions. There have been numerous implementations of exercise snacks with varying success rates. There are various reasons hindering the achievement of sustained physical activity adequate to surpass the threshold of exercise resistance. The most cited, include a perceived shortage of time and insufficient facilities to support physical activity throughout the day [35, 36]. Thus, exercise snacks have been proposed as an intervention for avoiding exercise resistance.

One example of an exercise snack that can prevent exercise resistance is brief bouts of high-intensity exercise, performed hourly, on a portable inertial pedal ergometer. One study had participants engage in repeated ergometer sprints at the end of every hour during the workday. This regimen was shown to mitigate the metabolic consequences of prolonged sedentary hours [19]. The hourly sprint condition decreased post-prandial triglyceride AUC_T (by 31%) and increased fat oxidation (by 43%). The authors suggested that these brief interruptions of sedentary behavior might be adequate to prevent exercise resistance. However, a limitation of this exercise snack is that it requires purchasing equipment costing around $60, potentially limiting its feasibility based on socio-economic status.

Several studies have suggested that exercise snacks could be used to target populations at a greater risk of exercise resistance. One study found that exercise snacks prior to meals can improve the glycemic response for individuals with type 2 diabetes [37]. Additionally, a review has proposed prescribing exercise snacks to specific cancer patients to potentially mitigate the cardiometabolic dysregulation associated with cancer treatment and recovery [38]. Finally, research has shown that stair stepping as an exercise snack is effective for individuals with overweight or obesity, leading to lower postprandial insulin and free fatty acid levels [39].

There are several proposed future directions for exercise snacks. Some of the clinically relevant directions include, “i) the feasibility of exercise snacks across various populations, ii) the comparison of intensities of exercise snacks, and iii) the psychological implications of exercise snacks” [34]. Among these, the comparison of exercise snack intensities stands out as particularly intriguing for preventing exercise resistance. Most studies on exercise snacks typically assess bouts of high intensity exercise which might not be sustainable for all populations. Moreover, adherence to behavior changes is often higher when participants find the experience pleasurable and perceive it as easy to integrate into their daily schedules [40]. For populations already struggling to improve cardiometabolic health through exercise alone, recommending high-intensity exercise snacks could be discouraging. Thus, identifying exercise snacks that are more tolerable may be crucial for individuals seeking to make long-term behavior changes.

Although exercise snacks offer immediate health benefits, their effectiveness in preventing exercise resistance remains unstudied. Considering that sedentary behavior in the workplace often contributes to exercise resistance, the idea of using exercise snacks to address this is appealing. However, exercise snacks have limitations. For example, portable options like the pedal ergometer mentioned earlier can be expensive and may not be suitable for all settings. Exercise snacks that produce noise could disrupt workplace focus and might not be permitted. While activities like stair climbing and sprinting are more accessible, they may attract unwanted attention during work hours, be disruptive to meetings, or might be incompatible with time sensitive projects. Thus, some exercise snacks still pose barriers to successful implementation [41]. Given these limitations, further research should explore alternative strategies to prevent exercise resistance that are more feasible.

Neuromuscular Electrical Stimulation as a Potential Alternative to Exercise Snacks

Neuromuscular electrical stimulation (NMES) is a therapeutic method that delivers electrical charges to skeletal muscles, inducing contractions. It's commonly employed in post-surgery recovery to counter muscle atrophy resulting from prolonged bed rest [42]. NMES tolerability hinges on factors like intensity, duration, and frequency of stimulation. Moreover, it's widely used across various populations with no reported adverse effects [42-46], suggesting its versatility and ability to be utilized throughout the day at tolerable intensities. NMES serves as a highly portable, commercially available, and cost-effective intervention, with proven benefits for skeletal muscle activity.

Additionally, existing evidence supports NMES as an alternative to exercise snacks. NMES has been shown to increase energy expenditure at rest and enhance post-meal glucose clearance [47, 48]. Moreover, a study demonstrated that a single 30-minute NMES session targeting specific muscle groups reduced total cholesterol, plasma triglycerides, and plasma free fatty acids [49]. These findings indicate that NMES can positively influence various metabolic outcomes during periods of sedentary behavior. Thus, NMES could potentially serve as a pseudo-exercise snack that is delivered at a lighter intensity over an extended period. Furthermore, NMES is capable of boosting muscle contractility, increasing caloric expenditure and favorably modifying mLPL and AMPK, proposed targets to counter exercise resistance.

There have not been any studies looking specifically at NMES and the potential activation of mLPL or AMPK in humans but there are some data in other mammalian models to indicate an increase in mLPL with electrical stimulation [50]. Human studies observing the response to chronic NMES use found an increase in Type I slow twitch muscle fibers and Type IIa fast oxidative and glycolytic intermediate fiber types in human skeletal muscle [44, 51]. These types of muscle fibers have an increased capacity for oxidative metabolic systems and are fatigue resistant, suggesting they have improved capacity for triglyceride clearance [52, 53]. A review conducted on enzymatic changes with chronic NMES use also indicates an increase in oxidative enzymes in muscle tissue, further justifying that this intervention can improve triglyceride clearance [54].

While NMES has been used to increase energy expenditure, it is unknown if this intervention could prevent exercise resistance. Certainly, NMES increases muscle contraction. While research regarding NMES is not new, its potential use to prevent exercise resistance is currently unexplored.

NEUROMUSCULAR ELECTRICAL STIMULATION: GETTING AMPED UP TO PREVENT EXERCISE RESISTANCE

Introduction

Emerging evidence suggests prolonged sedentary behavior between successive workouts can diminish the metabolic benefits of acute bouts of exercise [9, 13, 14, 20, 21]. Termed exercise resistance, this phenomenon is characterized by the absence of acute exercise-mediated improvements in fat metabolism. Poor fat metabolism can heighten the risk of developing chronic cardio-metabolic diseases like obesity, hyperglycemia, hypertension, and dyslipidemia [7, 18]. While regular aerobic exercise is typically considered one of the best preventive measures against cardio-metabolic diseases [4, 5], approximately 15-20% of individuals fail to improve their metabolic health with exercise alone [55]. Exercise resistance has been proposed as a potential explanation. That is, extended sedentary time between workouts may have diminished the protective effects usually imparted by acute exercise.

Recent data suggest that the energy expenditure threshold for avoiding exercise resistance is equivalent to walking approximately 8,500 steps [8]. Unfortunately, United States population data indicate that the average adult only walks 5,000 steps per day [12, 26, 32]. This suggests a deficit of 3,500 steps, which may contribute to exercise resistance. The biggest perceived barrier to surpassing the 8,500 steps/day threshold, and preventing exercise resistance, is insufficient time [35, 40]. Accordingly, it appears critical to identify an effective and time-efficient method for increasing energy expenditure by the equivalent of 3,500 steps. In this regard, neuromuscular electrical stimulation (NMES) may be a suitable intervention. NMES devices are easy to use and deliver electricity to targeted skeletal muscles to evoke contractions [42]. Recent studies have shown that NMES can be used to increase energy expenditure and simulate aerobic exercise while remaining seated [47, 48]. Collectively, these observations imply that NMES could potentially increase daily energy expenditure in a time-efficient manner while preventing the onset of exercise resistance.

Given the prevalence of exercise resistance and the need to overcome a deficit of 3,500 steps to reduce the risk of exercise resistance, we propose that NMES could theoretically increase energy expenditure by the equivalent of 3,500 steps, thereby meeting the step threshold component sufficient at preventing exercise resistance. To know the feasibility of such an intervention, we must know how long NMES should be applied to accomplish an energy expenditure equal to 3,500 steps. Assuming the average adult walks 5,000 steps per day, the specific aim of this research is to determine the feasibility of using NMES to match the caloric expenditure of walking 3,500 steps, making their energy expenditure equal to the energy expenditure of 8,500 steps.

Methods

Participant Enrollment Process and Screening

This study was conducted according to the guidelines of the Declaration of Helsinki and was approved by the Institutional Review Board of Colorado State University (Protocol #4308, 22 March 2023). All participants provided written informed consent prior to beginning the study.

Adult males and females in Fort Collins, Colorado were invited to participate in this study. Inclusion criteria included age 18-40 years (inclusive), being regularly active (more than 30 minutes of exercise, 3 days per week over the previous 12 months), and an ability to complete at least one hour of light treadmill exercise (i.e. walking). Exclusion criteria included presence of an injury that may hinder a participants' ability to exercise, and/or a history of cardiopulmonary disorder that may be contra-indicative to exercise. Participants completed a screening visit that involved informed consent, a screening questionnaire, measures of body composition via dual energy x-ray absorptiometry (Hologic, Discovery W, QDR Series, Bedford, Massachusetts, USA) and a maximal oxygen uptake test on a treadmill as reported in previous studies [56-58].

Experimental Design

Participants underwent three experimental visits throughout the duration of their enrollment. The aim of the first experiment was to quantify metabolic rate (Parvo Medics TrueOne 2400, Salt Lake City, Utah, USA) using previously described methods [57] with and without the application of NMES (AUVON 4 Output TENS unit muscle simulator, Peachtree Corners, Georgia, USA). The second experimental visit consisted of walking 8,500 steps on a treadmill while energy expenditure was measured. The final visit consisted of self-administered NMES during rest that was immediately followed by a 5,000-steps of walking on a treadmill. For each visit, participants arrived at the laboratory in the morning after a 12-hour fast, abstention from caffeine and alcohol for 20 hours, and abstention from vigorous exercise for 24 hours.

Experimental Visit 1 Protocol

After arriving at the lab and confirming their abstention period, participants were prepared for metabolic assessment. Metabolic measures were completed using indirect calorimetry. For metabolic measures, participants rested supine on a bed with a ventilated hood placed over their head. Oxygen uptake ($\dot{V}O_2$), Carbon Dioxide production ($\dot{V}CO_2$), respiratory exchange ratio ($\dot{V}CO_2$/ $\dot{V}O_2$), energy expenditure (kcal/min), and ventilation ($\dot{V}E$) were recorded by the metabolic cart and averaged every 30 seconds for a 45-minute period with the first 15 minutes of recorded data being excluded as participants habituated to the ventilated hood. Heart rate was measured with a remote cardiac monitoring device (OMRON Healthcare, INC., Illinois, USA). Following this period, a 15-minute break was granted for using the bathroom and for applying the NMES pads. Neuromuscular electrical stimulation was administered with two electrical stimulation devices. These devices each have four outputs allowing for the stimulation of the Vastus Lateralis, Vastus Medialis, and two heads of the Gastrocnemius. Participants were instructed to select their own intensity of the stimulation ranging from 27.5 mA – 550 mA with a symmetrical bi-phasic square waveform stimulation pattern. These specific skeletal muscles were selected due to their size (thereby potentially facilitating a bigger increase for metabolic rate compared with smaller muscles), because they were relatively superficial, and because previous published studies of NMES used these locations [47]. Participants spent another 45-minutes under the ventilated hood while NMES was applied at a self-selected intensity for a 1-to-1 second contraction-to-relaxation ratio.

Experimental Visit 2 Protocol

Participants arrived at the lab in the morning having walked less than 1,000 steps (self-reported) and were instrumented for indirect calorimetry and pedometry (3D pedometer, NESKLA, Sacramento, California, USA) before walking 8,500 steps at a self-selected pace on a horizontal treadmill. For metabolic measures while walking, participants were outfitted with a mask connected to a metabolic cart making the same measures recorded during rest. Walking heart rate was measured using short range telemetry (Polar T31, Bethpage, New York, United States). Events were marked on the metabolic cart when 3,500, 5,000, and 8,500 steps were accrued to measure the time taken to walk each distance and measure the energy expended for each step count.

Experimental Visit 3 Protocol

Based on the previous two visits, a calculation was made to determine the duration of NMES needed to evoke an energy expenditure equal to walking 3,500 steps. This was calculated with the following equation:

$$Time\ of\ NMES\ (\mathrm{min}) = \frac{Energy\ Expenditure\ of\ 3{,}500\ steps\ (kcal)}{Rate\ of\ NMES\ Energy\ Expendiuutre\ (kcal * min^{-1})}\ .$$

Participants then underwent measures of metabolic rate while NMES was applied at the same self-selected intensity previously used during the first experimental protocol for the calculated amount of time. On completion, participants then walked 5,000 steps on the treadmill while energy expenditure was measured. The total kilocalories of each activity were added together to determine whether these two activities combined could match the total kilocalories of walking 8,500 steps.

Statistical Analysis

All data, unless otherwise stated, are presented as mean and standard deviation. Statistical analyses were performed using dedicated, commercially available software (SigmaStat for Windows 3.5, Systat Software, Inc., Chicago, Illinois, USA). Differences in metabolic measures, heart rate, and total kilocalories between rest and NMES administration were examined with a paired t-test. Differences in energy expenditure while walking or while walking following a calculated dose of NMES were explored using a one-way analysis of variance (ANOVA) with repeated measures. Where appropriate, Tukey Honest Significant Difference post-hoc analysis was utilized to determine significance between conditions. The level of statistical significance was set at $p < 0.05$. A Bland-Altman Limits of Agreement plot (BA LoA) [59, 60] for agreement was utilized, to assess concordance between the energy expended for a calculated time of NMES and walking 5,000 steps and the energy expended while walking 8,500 steps. This plot assesses the degree of agreement between two techniques (5,000 steps + NMES vs. 8,500 steps) measuring the same variable (energy expenditure). Including a BA LoA plot allows one to determine whether two methods of measure produce similar results, thus allowing comparison for the similarity of energy expended while walking 8,500 steps or while walking 5,000 steps following a calculated dose of NMES.

Results

Participants

The progress of participants from screening to enrollment, to completion is depicted in the Consolidated Standards of Reporting Trials (CONSORT) flow diagram (Figure 1). Seventeen participants were screened, two participants dropped from the study due to not meeting the physical activity requirements (1 male) or being unable to tolerate fasting for 12 hours (1 male). A single participant completed all the study visits except for the last one (1 female). As a result, fifteen participants (7 male/8 female) were included in analyses made for the first two visits and fourteen participants (7 male/7 female) were included in all analyses. Selected physiological characteristics are presented in Table 1.

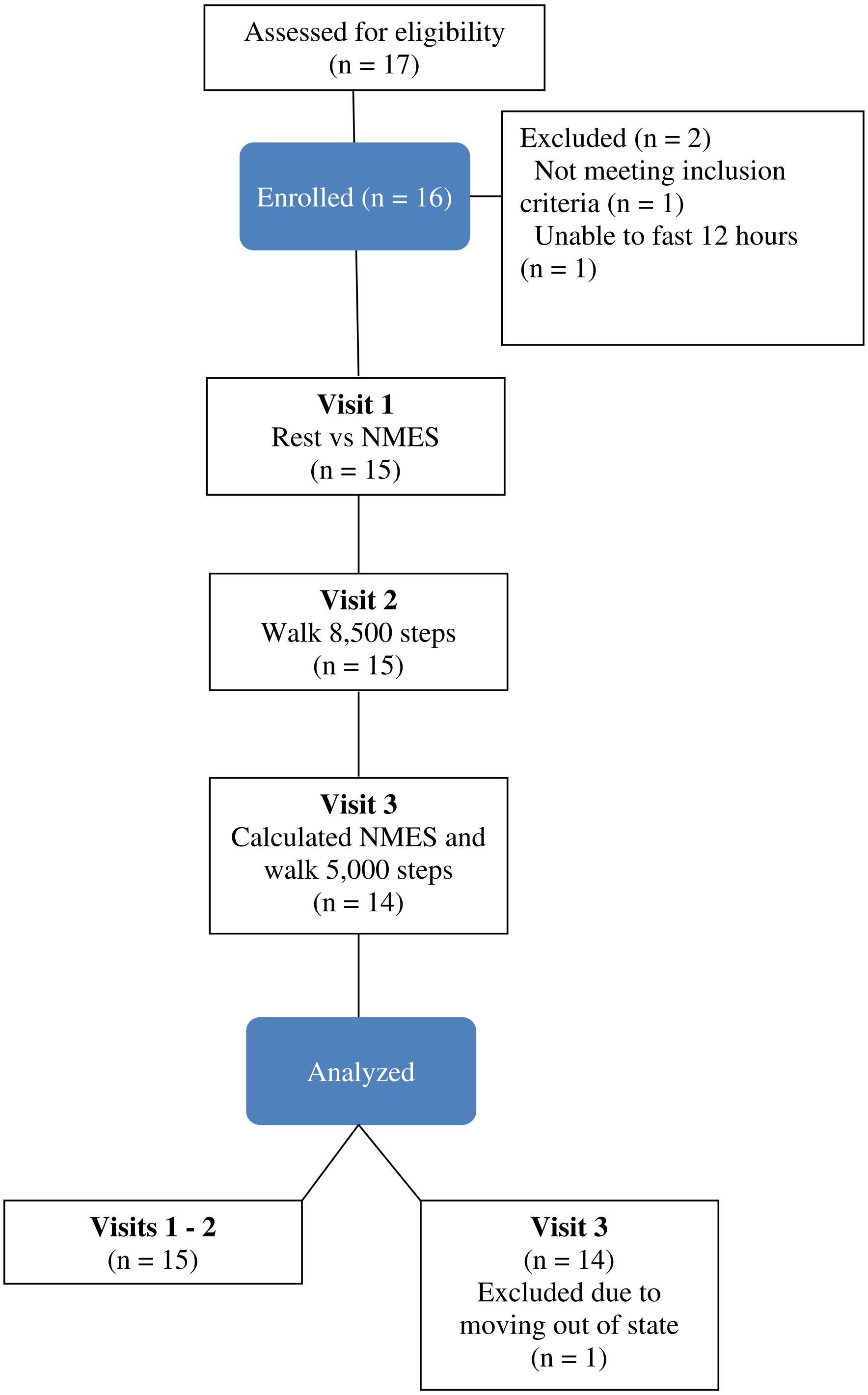

Figure 1. Consolidated Standards of Reporting Trials (CONSORT) flow diagram.

Table 1. Selected physiological characteristics. Data are mean ± SD. BMI: Body Mass Index, $\dot{V}O_2$max: Maximal oxygen uptake.

Participant Characteristics		
Variable	**Mean ± SD**	**Range**
Sex (M/F)	7/8	-
Height (cm)	170 ± 9	154 – 185
Age (years)	24 ± 2	20 - 27
Total Mass (kg)	70.13 ± 11.28	53.16 – 90.06
%Fat	24.76 ± 4.93	17.90 – 34.60
BMI (kg/m^2)	23.9 ± 2.6	20.4 – 28.8
Lean Mass (kg)	50.67 ± 8.93	38.04 – 65.93
$\dot{V}O_2$max (ml/kg/min)	48.1 ± 7.9	36.1 – 63.8
$\dot{V}O_2$max (L/min)	3.4 ± 0.8	2.1 – 4.8

Energy Expenditure

Figure 2 depicts the energy expended during 30 minutes of rest and 30 minutes of NMES. NMES significantly increased energy expenditure above rest (Rest vs. NMES: 33.2 ± 3.6 vs. 35.7 ± 4.3 kcal; p= <0.001). Furthermore, when converting the total energy expenditure into the metabolic rate (kcal/min) NMES significantly increased metabolic rate (Rest vs. NMES: 1.1 ± 0.1 vs. 1.2 ± 0.1 kcal/min; p = <0.001). Figure 3 depicts the energy expended while walking 3,500 steps, 5,000 steps, 8,500 steps, and 5,000 steps following a calculated dose of NMES respectively. All conditions significantly increased the total kilocalories expended compared to 3,500 (all $p < 0.001$), 8,500 steps and 5,000 steps following a calculated dose of NMES significantly increased total kilocalorie expenditure compared to 5,000 steps (both $p < 0.001$).

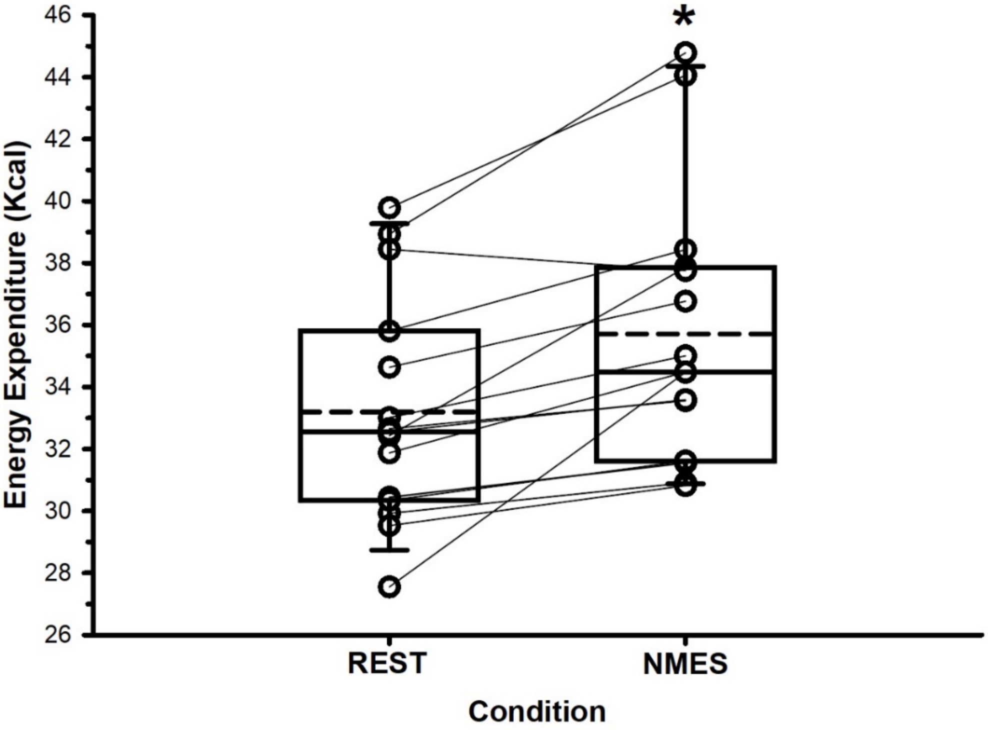

Figure 2. Energy expenditure during 30-minutes of rest and during 30-minutes of NMES. For all box plots, dashed lines depict the mean, the solid middle line depicts the median, the outer edges of the box plot represent the 25th and 75th percentiles, and the error bars extend to the 5th and 95th percentiles of the data. Individuals are depicted with open circles with a line connecting between conditions. * Depicts different from rest (*p* < .05).

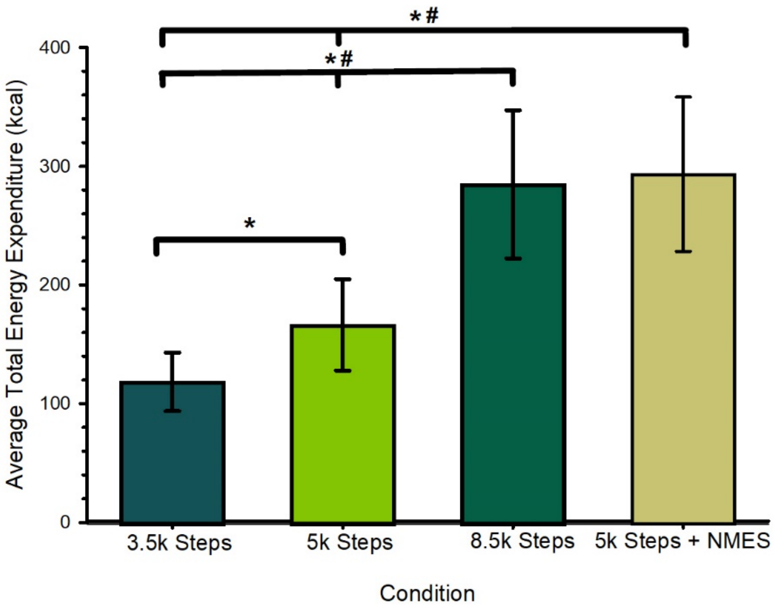

Figure 3. Energy Expenditure of different step amounts and 5,000 steps following a calculated dose of NMES. Data are depicted as mean and SD. * Depicts difference from 3,500 steps energy expenditure ($p < .05$), and # depicts difference from 5,000 steps energy expenditure ($p < .05$).

Bland-Altman Plot for Agreement

Figure 4 depicts a Bland-Altman Limit of Agreement (BA LoA) plot for assessing concordance between the caloric expenditure of walking 5,000 steps following a calculated dose of NMES (EE1) and walking 8,500 steps (EE2). Bias analysis (average between differences in measures) was 7.1 kilocalories with an SD of 23.6 kilocalories. This suggests a small bias of higher energy expenditure measured with EE1. Upper and lower limits for agreement analysis were set to be 1.96 × SD higher and lower than the bias analysis (53.4 and -39.3 kilocalories respectively.) Scatter analysis shows that there is concordance between 13 of 14 measurements in energy expenditure between 8,500 steps and 5,000 steps following a calculated dose of NMES.

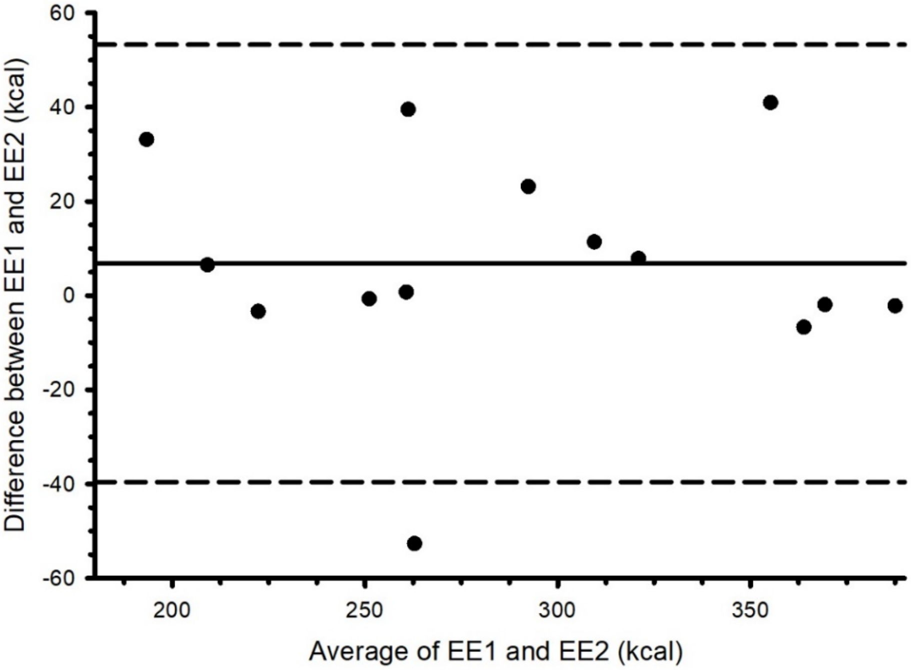

Figure 4. Bland-Altman Limit of Agreement Plot assessing concordance between 5,000 steps following a calculated dose of NMES (EE1) and 8,500 steps (EE2). The solid line represents the bias analysis, dashed lines depict the agreement analysis, and scatter analysis are represented by the filled in circles.

Heart Rate and Gas Exchange

Figure 5 depicts heart rate (A), oxygen uptake (B), respiratory exchange ratio (RER) (C), and carbon dioxide production. NMES significantly increased heart rate compared to rest (Rest vs. NMES: 55 ± 8 vs. 59 ± 8 bpm; $p = 0.017$). NMES significantly increased oxygen uptake compared to rest (Rest vs. NMES: 3.39 ± 0.38 vs.

3.63 ± 0.42 ml/kg/min; $p < 0.001$). NMES significantly increased carbon dioxide production compared to rest (Rest vs. NMES: 2.64 ± 0.23 vs. 2.86 ± 0.27 ml/kg/min; $p = 0.001$). There was no difference of RER between rest and NMES (Rest vs. NMES: 0.79 ± 0.04 vs. 0.799 ± 0.04; $p = 0.160$).

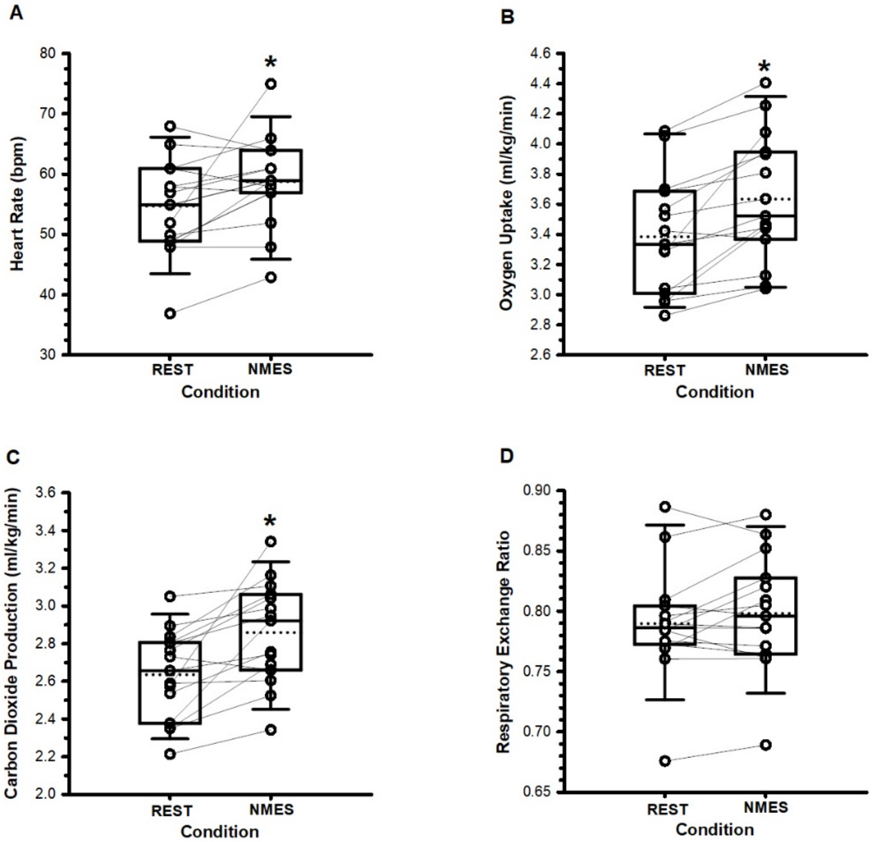

Figure 5. Measures of Heart rate (A), Oxygen uptake (B), Carbon Dioxide Production (C), and RER (D) during rest and during NMES. Dashed lines depict the mean for each condition. * Depicts different from rest ($p < .05$).

Intensity of Stimulation

Self-selected intensities of NMES on targeted skeletal muscles are shared on Table 2. There were no statistical differences between skeletal muscle groups and self-selected intensities (P = 0.166).

Table 2. Self-selected intensity of NMES on skeletal muscle groups. Data are mean ± SD.

Intensity of NMES	
Muscle Group	**Estimated Intensity (mA)**
Right Quadriceps	302 ± 69
Right Gastrocnemius	270 ± 96
Left Quadriceps	297 ± 82
Left Gastrocnemius	280 ± 81

Time of Activities

Table 3 depicts the average time taken to accomplish walking 3,500 steps, 5,000 steps, and 8,500 steps, as well as the calculated amount of time NMES was applied. Self-administration of NMES took significantly longer than walking 3,500 and 5,000 steps ($p < 0.001$) but not walking 8,500 steps ($p < 0.299$).

Table 3. Duration of Activities. Data are mean ± SD. * represents statiscal differences from 3,500 steps and $^{\#}$ represents statistical difference from 5,000 steps.

Duration of Activity	
Activity	**Time (minutes)**
3,500 steps	31 ± 2
5,000 steps	44 ± 3
8,500 steps	75 ± 7 *
Calculated NMES	100 ± 15 * $^{\#}$

Discussion

This study uses a novel approach to assess the feasibility of increasing energy expenditure to match the energy expenditure to an amount that can counter the detrimental effects of sedentary behavior. One of the key insights from this study is the ability of NMES to effectively match the energy expenditure associated with walking 3,500 steps in a convenient manner, thereby providing a means to disrupt prolonged periods of physical inactivity. Specifically, 100 minutes of NMES is comparable to the energy expenditure of walking 3,500 steps. Thus, it appears feasible that NMES can match the energy expenditure of 3,500 steps in a convenient manner.

The rate of energy expenditure at rest with NMES in our study was 1.2 kcal/min, consistent with metabolic rates reported in previous NMES studies (range: 1.1 - 1.8 kcal/min) [47, 61-63]. These collective data suggest that NMES increases resting energy expenditure. Accompanying this increase, heart rate was also accelerated, again aligning our findings with previous NMES research [62]. Noteworthy, the modest increase in heart rate (4 bpm) with no change in resting sinus rhythm, indicates that NMES does not impose undue cardiovascular stress. Moreover, research on the impact of NMES on cardiovascular function has elicited improvements in heart rate and blood pressure after a 2-week period [64] in a manner similar to exercise training.

The increase in metabolic rate was mediated by an increase in both VO_2 and VCO_2; another finding in our study that is consistent with previous work [47, 61-63]. However, in contrast to other studies, the magnitude of increase in both VO_2 and VCO_2 meant that the ratio of these variables (i.e. RER) was unchanged. Several other studies have reported that NMES increased RER and blood lactate, indicating a shift towards carbohydrate metabolism [65], but this is not a consistent observation across all studies [61]. The variability in the RER response could be attributed to differences in experimental design, including the combination of NMES with other perturbations, such as ingestion of glucose [47].

It is unclear if exercise resistance could be prevented with increased energy expenditure per se, or if the energy expenditure increase requires skeletal muscle contraction. This could be explored in future studies by administering thermogenic pharmaceuticals, such as the beta-adrenergic receptor agonist isoproterenol [66-69], to semi-recumbent, motionless adults. If exercise resistance persists despite beta-adrenergic stimulation, the interpretation would be that increased energy expenditure alone may not effectively prevent exercise resistance.

Reduced mLPL activity is another proposed mechanism contributing to exercise resistance [9, 13, 23, 70]. While no human studies demonstrate altered mLPL activity with electrical stimulation, low-frequency stimulation increases mLPL activity in rodents [22], particularly in slow-twitch oxidative muscle fibers. Despite this potential for enhancing triglyceride clearance, translation to human studies remains unexplored. Additionally, the impact of electrical stimulation on fat metabolism is debated [54, 71]. Some chronic NMES studies suggest enhanced fat metabolism via increased oxidative enzymes, while others report changes in glycolytic enzymes without improvements in oxidative enzymes. Clearly, future NMES research incorporating mLPL and metabolic enzyme activities appears warranted.

NMES predominantly recruits large motor units, contrasting with typical/physiological motor unit recruitment patterns [72]. These larger motor units, predominantly composed of fast-twitch muscles (Type IIx), rely on glycolytic pathways [52, 53], likely explaining NMES's consistent improvement in glucose metabolism [71]. Chronic NMES induces shifts away from fast-twitch, highly glycolytic Type IIx fibers towards Type IIa and Type I fibers, bolstering oxidative and glycolytic capacities [44, 50, 51, 54]. In the context of sport, this transition does not appear to compromise athletic performance for runners, weightlifters, hockey, rugby, soccer, basketball, and volleyball players [73-80]. Furthermore, NMES training improves fine motor skills, dexterity, balance, and can yield gains akin to conventional strength training for clinical populations [81-83]. Collectively, in addition to potentially preventing exercise resistance, these data suggest NMES can improve training responses for both athletic and clinical populations.

Combining NMES with other exercises can enhance its effects, offering synergistic benefits across different athletic activities. Integrating NMES with additional interventions, such as cycling, increases energy expenditure more than NMES or cycling exercise alone [84]. Stimulating inactive muscles with NMES during exercise snacks could further enhance muscle contractility, potentially preventing exercise resistance better than NMES alone. This could theoretically prevent exercise resistance to a greater extent than using NMES or exercise snacks alone. However, research on NMES and energy expenditure primarily focuses on lower body stimulation, neglecting upper body stimulation [62, 85]. This likely stems from contraindications of NMES use in certain body regions. For instance, NMES should not be used within the axial anatomy and is not recommended for those who are pregnant [86]. Contrarily, research investigating the effect of NMES on pregnant and post-partum females indicate that it is beneficial for relieving pregnancy symptoms during and following pregnancy [87]. There are still understudied muscle groups in the upper body suitable for NMES targeting, warranting further investigation for feasible targets in various populations.

Literature on exercise resistance has primarily focused on young populations, neglecting older adults. Given that metabolism declines with age and older adults tend to have lower daily step counts, they may be more susceptible to exercise resistance [2, 10, 88-92]. In this regard, NMES has been shown to prevent muscle atrophy, improve cardiorespiratory fitness, and improve balance in older adults [45, 49, 82]. These studies indicate that NMES already has benefits for older populations and can be used by this demographic. While the current study found that 100 minutes of NMES matched the energy expenditure of 3,500 steps in young adults, its applicability to older adults remains uncertain. Older adults may find NMES fatiguing, potentially making it unsuitable for preventing exercise resistance in this demographic. Regardless, more research is needed to assess the formation of exercise resistance and its reversibility in older adults before the application of NMES is considered.

Another demographic underrepresented in exercise resistance are elite athletes. Elite endurance athletes, for example, typically exhibit improved lipid metabolism and may be less susceptible to exercise resistance compared to elite powerlifters [93]. However, during tapering and detraining periods, athletes can become more susceptible to exercise resistance. Detraining studies suggest that elite athletes lose cardiovascular and metabolic gains at a faster rate than irregular exercisers [94], with data from elite rowers indicating reduced triglyceride clearance in skeletal muscle during detraining periods [95]. Furthermore, studies confirm that endurance athletes have the same post-prandial lipidemia response as untrained individuals with only 2.5 days of no exercise [96, 97]. Monitoring metabolic responses in elite athletes during detraining is crucial for maintaining peak performance and preserving metabolic gains. Furthermore, athletes of different backgrounds and sexes have different body compositions depending on the demands of their athletic event. However, existing literature lacks exploration of how body composition influences the increased energy expenditure induced by NMES in combating exercise resistance. Since NMES operates by conducting electric currents through tissue, the electric resistance of targeted tissue types may impact the efficacy of NMES to increase energy expenditure [98-100]. When considering tissues to stimulate with NMES for increasing energy expenditure, the resistance of the tissue might affect the increase in energy expenditure. Future studies should investigate this aspect to enhance understanding of NMES effectiveness across diverse athlete populations.

Limitations

Firstly, this project was a feasibility study and did not directly assess the use of NMES to prevent exercise resistance. We aim to address this in a future project by incorporating NMES on low activity days prior to acute exercise, replicating conditions in exercise resistance experiments. Additionally, our study participants were young, resembling those in previous exercise resistance research. There appears to be a need to explore exercise resistance in older populations and elite athletes.

One strength of our study is the inclusion of females, a population that has been underrepresented in previous NMES research. Potential effects of menstrual phase or contraceptive methods were not considered and there is some evidence to indicate that menstrual phase can alter neuromuscular performance, specifically force production, in young female athletes [101]. NMES appears to evoke significant increases in energy expenditure in females (Rest vs NMES in females: 32.0 ± 2.8 vs. 33.3 ± 2.4 kcal; $p = 0.01$). However, there were notable sex differences in absolute energy expenditure (Sex within NMES, Male vs Female: 38.6 ± 4.3 vs. 33.2 ± 2.4 kcal; $p = 0.01$). To account for this difference, we considered the influence of fat-free mass, noting a positive correlation between NMES energy expenditure and fat-free mass ($r = 0.790$, $p = 0.001$).

However, when normalizing absolute energy expenditure to fat-free mass, sex differences persisted, with females exhibiting a stronger metabolic response (Male vs Female: 0.678 ± 0.058 vs. 0.747 ± 0.063 kcal/kg FFM; $p = 0.03$). These findings suggest the need for further investigation into potential sex differences in NMES responsiveness. One additional and potentially confounding factor to consider pertains to the self-selection of NMES intensity; this may be different between the sexes.

Several strengths of our study stem from the use of commercially available NMES products. By using commercially available products, this study is more ecologically valid. Additionally, targeting superficial muscles that are easily accessible may enhance NMES efficacy for novices to the technique. Finally, including estimated stimulation intensities provides achievable targets for individuals seeking to match the energy expenditure of 3,500 steps in 100 minutes.

Future studies aiming to assess exercise resistance should address the previously mentioned gaps. This includes evaluating its prevalence across different demographics such as age, athletic level, activity types, and sexes to identify susceptible populations. Research should also investigate the metabolic effects of exercise snacks on exercise resistance and determine if certain snacks are more effective at prevention. Additionally, studies examining NMES on exercise resistance should assess whether muscular contraction and increased energy expenditure adequately prevent resistance and target proposed resistance mechanisms. The data presented in this study lay the groundwork for future research by exploring the optimal duration of NMES.

In conclusion, our feasibility study demonstrated that NMES effectively increased energy expenditure without changing substrate preference. Using statistics of agreement, we suggest that around 100 minutes of continuous NMES can match the energy expenditure of walking 3,500 steps. These results imply that NMES could prevent exercise resistance for the average adult American, thereby providing impetus for a future investigation.

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