Article Classification:
Research article

MULTIPLE-SET RESISTANCE TRAINING: IMMEDIATE CHANGES IN MUSCLE THICKNESS, ECHO-INTENSITY, FORCE PRODUCTION, AND PERCEPTION OF EFFORT

Paulo Marchetti[1], Rex Ruswick[1], Colin Stas[1] & Charles Lopes[2]

[1]Resistance Training Laboratory. Department of Kinesiology, California State University-Northridge, Northridge, CA, United States of America
[2]School of Physical Education, Pontifical Catholic University of Campinas (PUCCAMP), Brazil, Campinas, Brazil

OPEN ACCESS

Submitted: 26 September 2025
Accepted: 25 November 2025

ORCID
Paulo Marchetti
https://orcid.org/0000-0002-2016-936X
Rex Ruswick
https://orcid.org/0009-0004-7457-0089

Cite this article as:
Marchetti, P., Ruswick, R., Stas, C., Lopes, C. (2025). Multiple-set resistance training: immediate changes in muscle thickness, echo-intensity, force production, and perception of effort. *Journal of Applied Sports Sciences*, *9*(2), pp. 114 - 125. DOI: *10.37393/JASS.2025.09.02.8*

ABSTRACT

This investigation examined how performing multiple-set resistance training (RT) acutely influences maximal repetitions (MNR), muscle thickness (MT), echo intensity (EI), peak force (PF), and perceived exertion (RPE) in recreationally trained subjects. Twenty-one recreationally resistance-trained subjects (16 males and five females) performed a unilateral biceps curl with eight sets of 10RM, and 2-minute rest. During each set, MNR was recorded, and RPE_{set} *was assessed. PF at 90° elbow flexion and ultrasound measurements of the elbow flexors (MT and EI) were obtained before and immediately after each set of the RT session. Session RPE (sRPE) was reported 15 minutes after completion of the RT session. One-way ANOVAs were used to examine differences across sets for all dependent variables. Findings showed a progressive rise in RPEset, MT, and EI across sets, while both MNR and PF declined (p < .05). In summary, performing multiple RT sets reduced MNR and PF but elevated RPEset, EI, and MT, with MT stabilizing after the fifth set. Coaches should prioritize not only the chronic effects of RT but also the effective prescription and control of each RT session by implementing multiple sets per exercise, monitoring physical performance and perceived exertion to regulate neuromuscular fatigue, emphasizing high-intensity loading, and ensuring adequate recovery to maximize muscle responses after each RT session.*

Keywords: *neuromuscular fatigue, muscle performance, muscle pump, muscle image, muscle performance.*

INTRODUCTION

Resistance training (RT) is well established as an effective strategy to enhance skeletal muscle function, improve performance, and support overall health (American College of Sports, 2009). Evidence indicates that multiple-set RT protocols are typically more effective than single-set approaches for stimulating hypertrophy and strength gains, likely because they impose greater metabolic stress, mechanical loading, and neuromuscular demand (Schoenfeld et al., 2019; Schoenfeld et al., 2017). Although the long-term benefits of multiple-set RT are well documented, less is known about the immediate, set-by-set effects of training volume. In a recent study (Marchetti et al., 2023), resistance-training sessions with varying set numbers and non-equalized volume were examined for their effects on muscle thickness, force output, and perceived exertion in recreationally trained subjects. Fifteen subjects performed unilateral biceps curl exercises under four different protocols: (i) G4: 4 sets of 10RM; (ii) G8: 8 sets of 10RM; (iii) G12: 12 sets of 10RM; and (iv) G16: 16 sets of 10RM. The variables analyzed included the average number of repetitions (ANR), total number of repetitions (TNR), time under tension (TUT), muscle thickness (MT), peak force (PF), and rating of perceived exertion (RPE), assessed pre- and post-intervention. Their findings showed that average repetitions (ANR) decreased as the number of sets increased (notably in G8, G12, and G16 compared with G4). In contrast, the total number of repetitions (TNR) rose progressively with greater set volume. TUT increased across protocols, both between the first and last set and across conditions at the final set. PF significantly decreased from pre- to post-test in all protocols, with additional between-group differences at post-test (G4 × G12, G4 × G16, and G8 × G16). MT significantly increased across all protocols, whereas session RPE did not differ among them. In summary, higher-volume RT protocols resulted in a lower ANR but greater total repetitions and TUT, along with a more pronounced reduction in force production. Nevertheless, all RT protocols elicited significant increases in MT, with no differences in session RPE. Such acute responses to RT—namely alterations in MT, EI, and PF—provide insight into the physiological processes that may drive long-term adaptations. This study is novel in that it examines the progression of these variables during an RT session, rather than simply comparing pre- and post-session measurements. Nonetheless, the progressive set-by-set impact during a single training session remains poorly understood (Abe et al., 2014; Schoenfeld, 2010, 2013; Schoenfeld & Contreras, 2014). These short-term changes depend on several training variables—such as load intensity, rest duration, repetition number, cadence, and particularly the total sets performed within a session (Krieger, 2010; Radaelli et al., 2015). Acute increases in MT likely reflect exercise-induced swelling, a temporary response linked to cell hydration and mechanical loading (Abe et al., 2014; Loenneke et al., 2011). Similarly, alterations in EI, assessed via ultrasound imaging, can indicate acute changes in muscle composition, fluid distribution, and intramuscular edema (Stock & Thompson, 2021; Wong et al., 2020a). Functional markers, such as force production, provide complementary information on neuromuscular fatigue and recovery, reflecting the immediate impact of RT on performance capacity (Pareja-Blanco et al., 2020). A clearer understanding of these acute set-by-set responses may help clarify the mechanisms of adaptation and inform optimal RT prescriptions. Accordingly, this study sought to analyze how multiple RT sets acutely affect MNR, MT, EI, PF, and RPE in recreationally trained subjects. We hypothesized that (1) MNR and PF would decline with successive sets, (2) RPE would rise progressively, and (3) both MT and EI would show cumulative increases across sets. Clarifying how training volume is affected by preceding sets may improve understanding of acute structural and functional changes in muscle, with practical implications for designing RT sessions to optimize performance.

METHODOLOGY

Subjects

The number of subjects was determined in a previously conducted pilot study, based on an effect size difference of 0.75, a significance level of 5%, and a power of 80% derived from the muscle thickness of subjects with characteristics similar to those in the present study. (Eng, 2003). Twenty-one resistance-trained subjects were recruited to this study (16 males and five females; age 24 ± 2.5 years, total body mass 75.1 ± 11.4 kg, height 171.7 ± 8.8 cm). All subjects were regularly engaged in an RT program and familiar with the unilateral bicep curl exercise. They had 2 ± 1 years' experience (3 ± 1 sessions/week, two exercises per muscle group, and four sets per exercise). All subjects had no previous surgery or history of injury with residual symptoms (pain) in the spine or upper limbs within the last year. The maximal external load (10RM) performed was 13.8 ± 5.4 kg. The subjects were informed of the risks and benefits of the study prior to any data collection. All subjects provided written informed consent prior to participation, and the Institutional Review Board approved the study (#FY23-222).

Protocol

A within-subjects design was applied to compare the acute effects of multiple sets on the maximal number of repetitions, muscle thickness, echo intensity, peak force, and rating of perceived exertion following an RT session in recreationally trained subjects. Subjects attended two RT sessions in the laboratory and refrained from performing any upper-body exercise for at least 48 hours prior to testing. In the first RT session, anthropometric data (height, weight, and arm length) were collected, and each subject was asked to identify their preferred writing arm, which was considered the dominant arm (Maulder & Cronin, 2005). Subjects performed a familiarization and specific warm-up protocol consisting of one set of 15 repetitions without external load, followed by one set of 10 repetitions with a 5-kg load, with a 1-minute rest interval between sets. Subsequently, all subjects performed a multiple maximum-repetition test (10RM) for the unilateral biceps curl. For the unilateral biceps curl, subjects stood in front of a cable-pulley machine, holding the handle with a supinated grip. Each repetition consisted of lifting the weight stack from full elbow extension to full elbow flexion (concentric phase) and then returning to full elbow extension (eccentric phase). The 10RM test was conducted in accordance with the guidelines of the National Strength and Conditioning Association (NSCA) to determine each subject's initial training load. The load was progressively increased until subjects reached their maximum capacity to perform 10 repetitions with proper technique. The movement velocity was self-selected, and exercise technique was directly supervised by a Certified Strength and Conditioning Specialist (CSCS) research assistant to ensure correct performance. During the second RT session, subjects performed a specific warm-up consisting of one set of 10 repetitions at 50% of their 10RM. The subjects rested for five minutes before completing eight sets of 10RM (Repetition in reserve, RIR=0-1) with a 2-minute rest between sets. During each set, the MNR was recorded, and the rating of perceived exertion (RPE) was collected. Maximal voluntary isometric contraction (PF) at 90° elbow flexion and ultrasound measurements (MT and EI) of the elbow flexors were obtained before and immediately after each set of the RT session. Following a 15-minute rest period, subjects were asked to report their session RPE.

Measurements

Maximal number of repetitions (MNR): The MNR was recorded for all sets.

Muscle Thickness (MT): Ultrasound images were used to measure MT. A US technician performed all measurements using a portable ultrasound imaging unit (Hitachi Noblus; Hitachi Medical Corporation, Tokyo, Japan). Following a generous application of a water-soluble transmission gel (Cskin, Medics Medical Products LLC., NY, USA) to the measured site, a 7.5-MHz linear array probe

(L55 Probe) was placed perpendicular to the tissue interface without depressing the skin. Equipment settings were optimized for image quality (depth: 8 cm; brightness / B-mode gain: 35) according to the manufacturer's user manual and held constant across all sessions. MT was determined by measuring the distance from the subcutaneous adipose tissue–muscle interface to the muscle–bone interface, following the methods described by Abe et al. (2014). Measurements were taken on the dominant arm over the elbow flexors while the subjects were in a standing position. For the elbow flexors, measurements were taken at 60% distal between the lateral epicondyle of the humerus and the acromion process of the scapula. To maintain consistency between pre- and post-intervention testing, each site was marked with ink. To further ensure measurement accuracy, at least three images were taken at each muscle site. If measurements were more than 1mm apart, a fourth image was obtained and averaged. When the image's quality was deemed satisfactory, it was saved to a hard drive. The test-retest ICC was 0.96–0.98, and the intra-rater reliability was 0.96–0.97.

Echo Intensity (EI): The same images used to define muscle thickness were used to quantify EI. EI was assessed using an automated grayscale analysis that ranged from 0 (black) to 255 (white) (arbitrary unit, A.U.). A rectangular region of interest box was drawn as large as possible to encompass the entire cross-sectional area. Care was taken to ensure that fascia along the borders of the elbow flexors, subcutaneous adipose tissue, and the bone region were not included in the analyses. EI was measured twice, and the mean value was used for further analysis. The test-retest ICC was 0.97.

Peak Force (PF): The maximal voluntary isometric contraction (MVIC) was measured by a digital load cell acquisition system (FM-204-1000K, Shenzhen Aermanda Technology Co. Ltd., Shenzhen, Guangdong, China (Capacity: 1000Kgf / Resolution: 0.01kgf). To perform the MVIC, all subjects were positioned standing in front of the cable-pulley machine, with a supinated grip on a handle. All subjects performed three MVICs at 90º of elbow flexion before (pre-test) and immediately after all sets. The 90º elbow position was selected because it represents the sticking point for the elbow flexors. Each MVIC was performed for 3-sec and 10-sec rest intervals. The maximum value (PF) was used for further analysis. The test-retest ICC ranged from 0.95 to 0.98.

Rating of Perceived Exertion (RPE): The rating of perceived exertion was assessed after each set (RPE_{set}) and 15 minutes after the RT session (sRPE) using the CR-10 scale, in accordance with the recommendations of Sweet et al. (Sweet et al., 2004). Subjects were asked to use an arbitrary unit (A.U.) on the scale to rate their overall effort after each set and RT session. A rating of 0 indicated no effort, and a rating of 10 indicated maximal effort and the most stressful exercise ever performed.

Statistical Analyses

The normality and homogeneity of variances were confirmed by Shapiro-Wilk and Levene's tests, respectively. The mean, standard deviation (SD), 95% confidence interval ($CI_{95\%}$), and delta percentage (Δ%) were calculated. One-way ANOVAs were used to detect differences across sets for the following variables: MNR, MT, EI, RPE_{set}, and PF. Post-hoc comparisons were conducted using the Bonferroni test when appropriate. The magnitude of differences was further evaluated using standardized effect sizes based on Cohen's *d*, which were qualitatively interpreted as follows: <0.35 – trivial; 0.35–0.8 – small; 0.8–1.5 – moderate; >1.5 – large for recreationally trained (Cohen, 1988). An alpha of 5% was used to determine statistical significance.

RESULTS

The sRPE reported by the subjects 15-min post-exercise was 8.37 ± 1.34 A.U.

For MNR, significant reductions were observed across sets ($p < .001$), as presented in Table 1and Figure 1a.

Table 1. *Comparison between sets [p-value, delta percentage (Δ%), effect size, and 95% confidence intervals ($CI_{95\%}$)] for the maximal number of repetitions.*

Comparison	*p*-value	Δ%	Effect Size	$CI_{95\%}$
Set 1 x Set 5	.009	17.0%	6.0 (large)	0.28 / 3.14
Set 1 x Set 6	.001	23.0%	7.3 (large)	0.69 / 3.87
Set 1 x Set 7	<.001	28.0%	7.7 (large)	0.99 / 4.62
Set 1 x Set 8	<.001	31.5%	9.4 (large)	1.45 / 4.83

For PF, significant reductions were observed across sets ($p < .005$), as presented in Table 2 and Figure 1b.

Table 2. *Comparison between pre-test and sets [p-value, delta percentage (Δ%), effect size, and 95% confidence intervals ($CI_{95\%}$)] for peak force.*

Comparison	*p*-value	Δ%	Effect Size	$CI_{95\%}$
Pre-Test x Set 1	.017	5.8%	0.9 (large)	1.21 / 20.70
Pre-Test x Set 2	.003	9.1%	1.4 (very large)	4.26 / 30.10
Pre-Test x Set 3	.002	12.5%	2.1 (very large)	6.64 / 40.20
Pre-Test x Set 4	<.001	15.3%	2.5 (very large)	13.10 / 44.50
Pre-Test x Set 5	<.001	17.2%	2.9 (very large)	16.10 / 48.60
Pre-Test x Set 6	<.001	18.3%	3.0 (very large)	19.30 / 49.60
Pre-Test x Set 7	<.001	19.4%	3.3 (very large)	20.30 / 52.50
Pre-Test x Set 8	<.001	21.4%	3.6 (very large)	24.00 / 56.40

For RPE_{set}, significant increases were observed across sets ($p < .001$), as presented in Table 3 and Figure 1c.

Table 3. *Comparison between sets [p-value, delta percentage (Δ%), effect size, and 95% confidence intervals ($CI_{95\%}$)] for RPE_{set}.*

Comparison	*p*-value	Δ%	Effect Size	$CI_{95\%}$
Set 1 x Set 2	<.001	12.6%	2.8 (very large)	-1.77 / -0.32
Set 1 x Set 3	<.001	18.8%	4.5 (very large)	-2.56 / -0.67
Set 1 x Set 4	<.001	25.0%	7.2 (very large)	-3.25 / -1.32
Set 1 x Set 5	<.001	27.3%	8.2 (very large)	-3.52 / -1.62
Set 1 x Set 6	<.001	28.1%	9.2 (very large)	-3.83 / -1.60
Set 1 x Set 7	<.001	30.3%	10.2 (very large)	-4.24 / -1.76
Set 1 x Set 8	<.001	30.3%	10.2 (very large)	-4.16 / -1.83
Set 3 x Set 4	<.001	7.6%	2.7 (very large)	-1.18 / -0.15

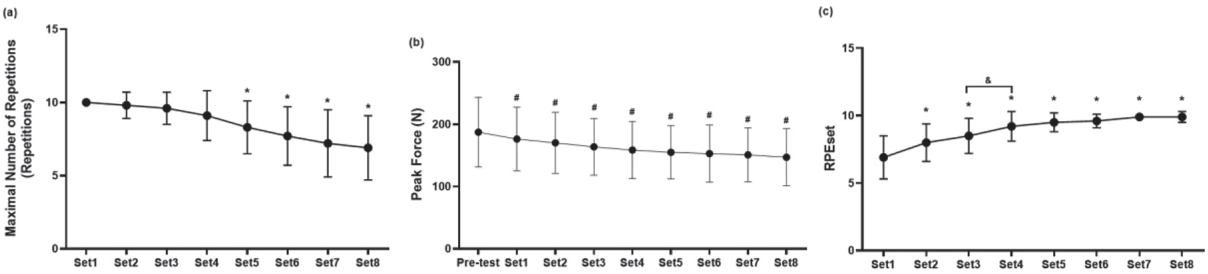

Figure 1. *Mean ± standard deviation of (a) maximal number of repetitions, (b) peak force, and (c)* RPE_{set}.

***Legend**: #Significant difference pre-test and set; *Significant difference between set 1 and all sets; &Significant difference between sets.*

For MT, significant increases were observed across sets ($p < .05$), as presented in Table 4 and Figure 2a.

Table 4. *Comparison between pre-test and sets and between sets [p-value, delta percentage (Δ%), effect size, and 95% confidence intervals* ($CI_{95\%}$)*] for muscle thickness.*

Comparison	*p*-value	Δ%	Effect Size	$CI_{95\%}$
Pre-Test x Set 1	<.001	6.9%	1.6 (very large)	-3.75 / -1.19
Pre-Test x Set 2	<.001	10.1%	2.4 (very large)	-4.82 / -2.71
Pre-Test x Set 3	<.001	11.8%	2.9 (very large)	-5.81 / -3.18
Pre-Test x Set 4	<.001	13.1%	3.1 (very large)	-6.54 / -3.69
Pre-Test x Set 5	<.001	15.3%	3.8 (very large)	-7.72 / -4.47
Pre-Test x Set 6	<.001	15.5%	3.8 (very large)	-7.69 / -4.73
Pre-Test x Set 7	<.001	15.1%	3.8 (very large)	-7.66 / -4.37
Pre-Test x Set 8	<.001	16.8%	4.3 (very large)	-8.42 / -4.96
Set 1 x Set 2	.006	3.4%	0.8 (large)	-2.35 / -0.25
Set 4 x Set 5	.009	2.5%	0.5 (small)	-1.79 / -0.16

For EI, significant increases were observed across sets ($p < .001$), as presented in Table 5 and Figure 2b.

Table 5. *Comparison between pre-test and sets and between sets [p-value, delta percentage (Δ%), effect size, and 95% confidence intervals* ($CI_{95\%}$)*] for echo intensity.*

Comparison	*p*-value	Δ%	Effect Size	$CI_{95\%}$
Pre-Test x Set 1	.002	13.8%	2.6 (very large)	-20.90 / -3.46
Pre-Test x Set 2	.001	20.0%	3.8 (very large)	-32.10 / -5.76
Pre-Test x Set 3	<.001	25.4%	5.1 (very large)	-41.50 / -10.00
Pre-Test x Set 4	<.001	26.7%	5.6 (very large)	-42.20 / -12.90
Pre-Test x Set 5	<.001	28.2%	6.5 (very large)	-42.50 / -16.90
Pre-Test x Set 6	<.001	29.3%	6.8 (very large)	-43.80 / -18.90
Pre-Test x Set 7	<.001	33.3%	7.6 (very large)	-51.50 / -24.00
Pre-Test x Set 8	<.001	37.1%	9.7 (very large)	-57.60 / -31.60
Set 1 x Set 7	.024	22.6%	1.04 (moderate)	-49.59 / -1.55
Set 1 x Set 8	<.001	27.0%	1.40 (moderate)	-56.45 / -8.41
Set 6 x Set 7	.015	5.6%	1.1 (large)	-11.90 / -0.77

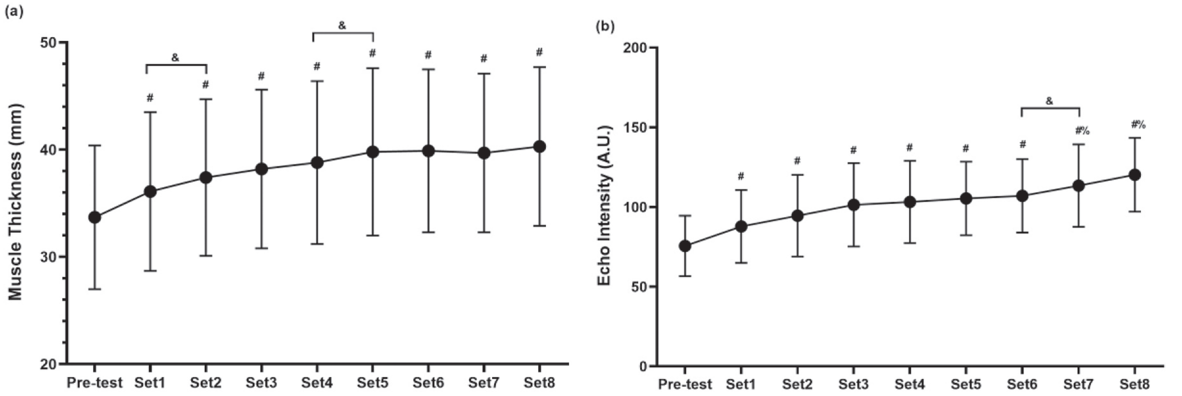

Figure 2. *Mean ± standard deviation of (a) MT and (b) EI.*
***Legend**: $^{\#}$Significant difference pre-test and sets; $^{\&}$Significant difference between sets; $^{\%}$ Significant difference between set 1.*

DISCUSSION

This study examined the acute effects of multiple resistance-training sets on MNR, MT, EI, PF, and RPE in recreationally trained subjects. Key outcomes indicated that MNR declined noticeably after the fifth set, PF dropped steadily with set progression, RPEset rose consistently, MT expanded initially but stabilized after the fifth set, and EI continued to increase throughout. These results provide new insights into the acute response to RT sessions. To the best of the authors' knowledge, no previous study has examined the acute dose–response of cumulative set-by-set effects in recreationally trained subjects.

An initial analysis of the multiple-set RT session revealed cumulative acute effects across all sets when muscular failure was reached (RIR = 0–1; sRPE = 8.3 A.U.). PF and RPE_{set} were affected from the first set, whereas a significant reduction in MNR was observed only at the fifth set. We hypothesized that MNR would progressively decrease set by set, and the results confirmed a 17% reduction at the fifth set and a 31.5% reduction by the end of all sets. Similarly, PF was expected to decline progressively, and the data indicated a 5.8% reduction after the first set and a 21.4% reduction by the final set. Notably, only after a 17.2% decrease in force production did MNR significantly decline (5th set).

Neuromuscular fatigue during RT typically presents as declining force output, fewer repetitions per set, and shorter time under tension, influenced by both peripheral and central factors. (Debold & Westerblad, 2024; Westerblad & Allen, 2003). Peripheral factors include metabolite accumulation and impaired excitation–contraction coupling, which compromise calcium handling and cross-bridge formation. Central factors—such as reduced corticospinal excitability and diminished motoneuron output—further limit voluntary activation and exacerbate performance declines (Behrens et al., 2023; Enoka & Duchateau, 2016; Gandevia, 2001). With repeated high-intensity sets, depletion of phosphocreatine and greater reliance on glycolysis lead to the buildup of Pi, H^+, and lactate. These byproducts impair cross-bridge cycling and reduce calcium sensitivity in the myofibrils, thus decreasing contractile efficiency with each additional set of a multiple-set RT session (Debold & Westerblad, 2024; Westerblad & Allen, 2003). Consistent with our hypothesis, RPEset rose steadily from the initial set onward. The results showed an initial 12.6% increase after the first set and a 30.3% increase by the end of all sets. RPE_{set}

captures the link between physiological stress and performance by quantifying perceived effort during each set (Alexiou & Coutts, 2008; Halperin & Emanuel, 2019; Marchetti, 2023; Weilharter et al., 2024). In short, these findings indicate that perceived exertion closely mirrors the progressive neuromuscular and metabolic demands of a multiple-set RT session. The consistent rise in RPE_{set} underscores its utility as a practical, non-invasive marker of cumulative fatigue and physiological stress during multiple sets and high-intensity RT sessions.

Continuing the analysis of the dependent variables, two muscle assessments were conducted to evaluate muscle morphology (MT) and muscle quality (EI) after multiple sets. RT exercises with multiple sets have been shown to elicit varying degrees of acute cell swelling, depending on factors such as the number of sets and repetitions, fatigue level, training volume, and intensity (Marchetti et al., 2023; Schoenfeld, 2013; Sullivan et al., 2023). Moreover, RT performed to momentary mechanical failure (10RM, RIR = 0–1) reduces intramuscular adenosine triphosphate (ATP) and creatine phosphate (CP) levels, while concomitantly increasing inorganic phosphate (Pi), adenosine diphosphate (ADP), and adenosine monophosphate (AMP) accumulation. This is accompanied by enhanced glycolytic flux, hypoxia, and reactive oxygen species production (Chen et al., 1996; Schoenfeld, 2013; Schoenfeld & Contreras, 2014; Sjøgaard et al., 1985; Usher-Smith et al., 2009). Collectively, these metabolic and mechanical stressors contribute to cell swelling and elevated intramuscular pressure. In this study, MT and EI were assessed before and immediately after each set to examine the extent of acute cell swelling. We hypothesized that both MT and EI would progressively increase on a set-by-set basis. For MT, the results partially supported this hypothesis: a 16.8% increase (a very large effect size) was observed from the pre-test to the last set. However, no significant changes occurred after the fifth set. Thus, despite the cumulative effect of multiple sets to momentary mechanical failure, the acute expansion in muscle size due to cellular swelling appears to be limited. Passive connective tissues, such as the endomysium, play a critical role in maintaining structural support and influencing the passive mechanical response of skeletal muscle. Because of their viscoelastic nature, these tissues deform under load but partially recover when the load is released (Nordin & Frankel, 2021). Nevertheless, their capacity for elongation is limited. The endomysium, which encases individual muscle fibers, contributes substantially to passive stiffness and force transmission within skeletal muscle. Research on intramuscular connective tissue indicates that passive muscle structures tolerate approximately 8–12% strain before collagen disruption occurs (Purslow, 2002). In contrast, our hypothesis concerning EI was supported, with grayscale pixel brightness rising steadily across sets. EI has been associated with cell swelling and has recently been proposed as a non-invasive indicator to differentiate whether acute changes in MT arise from intracellular fluid shifts or extracellular fluid accumulation (Wong et al., 2020b; Yitzchaki et al., 2019). In this study, EI increased by 37.1% from pre-test to the last set, reflecting a substantial influx of fluids into the muscle tissue during multiple sets. Given the restricted expansion of passive tissues (as suggested by MT plateauing) and the ongoing rise in EI, intramuscular pressure is likely to become more pronounced after the fifth set. Thus, performing multiple sets to near failure elevated both MT and EI, although MT stabilized after the fifth set, whereas EI continued to rise. These results imply that passive tissue limitations restrict further muscle expansion, thereby increasing intramuscular pressure during high-volume RT. Several limitations should be acknowledged. The modest sample size and multiple comparisons may have reduced statistical power, but the effect-size analysis provided meaningful insights. Second, lactate and other metabolic by-products were not measured, which limits insights into the mechanisms of neuromuscular fatigue. Finally, caution is warranted in generalizing these findings to other exercises, training protocols, or populations such as youth, competitive athletes, or older adults.

CONCLUSIONS

In summary, this study shows that multiple-set RT sessions trigger specific acute neuromuscular and morphological adaptations in recreationally trained subjects. Based on the authors' knowledge, this is the first study to report that training volume is affected by preceding sets and may improve understanding of the acute structural and functional changes in the muscle pump, with practical implications for designing RT sessions to optimize performance. More precisely, MNR and PF decreased with multiple sets, whereas both set-based and session RPE rose, reflecting heightened neuromuscular fatigue and perceived effort. In contrast, MT expanded early but stabilized after the fifth set, suggesting a ceiling effect for acute swelling responses. EI, however, continued to increase, suggesting possible changes in muscle quality associated with neuromuscular fatigue and intramuscular fluid shifts. To the authors' knowledge, this is the first study to identify a potential physical limitation in muscle response alongside a consistent increase in acute muscle swelling following multiple sets in trained men. Overall, these results suggest that multiple-set RT sessions reduce repetition capacity and force output over time, while simultaneously elevating perceived exertion and inducing short-term morphological and qualitative changes in muscle tissue. For strength and conditioning professionals, the data imply that performing more than five sets may not further enhance acute swelling responses. Instead, prioritizing sets with higher intensity and adequate recovery might be more effective for hypertrophy and strength gains. Nonetheless, combining high-intensity exercise with multiple sets elevates intramuscular pressure, potentially enhancing protein signaling via mechanotransduction pathways. Thus, optimizing RT outcomes requires a careful balance between training volume, intensity, and recovery periods. Monitoring perceived exertion and performance decline across sets may provide useful feedback for tailoring training volume and recovery strategies. Future research should investigate the long-term effects of different set configurations on muscle growth and strength adaptations and re-examine the trends observed in this study using a larger participant sample to provide more specific and generalizable recommendations.

PRACTICAL APPLICATIONS

Coaches should prioritize not only the chronic effects of RT but also the effective prescription and control of each RT session by implementing multiple sets per exercise, monitoring physical performance and perceived exertion to regulate neuromuscular fatigue, emphasizing high-intensity loading, and ensuring adequate recovery to maximize muscle responses after each RT session. Below are some practical applications:

- This study indicates that muscle thickness increases only up to approximately five sets, suggesting a ceiling effect for acute swelling. Thus, performing three to five sets per exercise appears sufficient to maximize the immediate muscle stimulus, while additional sets may provide minimal added benefits.

- Progressive declines in repetition capacity and peak force highlight the value of tracking performance loss. Reductions in reps may serve as a practical reference to terminate the exercise and prevent the accumulation of non-productive volume.
- The steady increase in the number of sets and session RPE suggests that subjective effort is a useful real-time indicator of neuromuscular fatigue. When RPE values exceed target levels, adjustments in load, rest intervals, or total sets may help maintain training quality.
- Since acute muscle swelling does not increase beyond the fifth set, high-intensity loading continues to stimulate mechanotransductive pathways. Therefore, increasing the number of sets could be an interesting strategy.
- Elevations in muscle swelling and reductions in force output following multiple sets suggest meaningful neuromuscular fatigue and muscle quality changes. High-volume sessions should therefore be followed by appropriate recovery strategies, such as alternating heavy and light days or reducing volume in subsequent sessions.

REFERENCES

Abe, T., Loenneke, J. P., Thiebaud, R. S., & Loftin, M. (2014). Morphological and functional relationships with ultrasound-measured muscle thickness of the upper extremity and the trunk. *Ultrasound*, *22*(4), 229-235. https://doi.org/10.1177/1742271X14554678

Alexiou, H., & Coutts, A. J. (2008). The validity of the session-RPE method for monitoring training load in elite female soccer players. *International Journal of Sports Physiology and Performance*, *3*(3), 320–330.

American College of Sports, M. (2009). American College of Sports Medicine position stand. Progression models in resistance training for healthy adults. *Med Sci Sports Exerc*, *41*(3), 687-708. https://doi.org/10.1249/MSS.0b013e3181915670

Behrens, M., Gube, M., Chaabene, H., Prieske, O., Zenon, A., Broscheid, K. C., Schega, L., Husmann, F., & Weippert, M. (2023). Fatigue and Human Performance: An Updated Framework. *Sports Med. (Auckland, N.Z.)*, *53*(1), 7–31.

Chen, Y. L., Wolin, M. S., & Messina, E. J. (1996). Evidence for cGMP mediation of skeletal muscle arteriolar dilation to lactate *J Appl Physiol.*, *81*(1), 349–354.

Cohen, J. (1988). *Statistical power analysis for the behavioral sciences* Lawrence Erlbaum Associates.

Debold, E. P., & Westerblad, H. (2024). New insights into the cellular and molecular mechanisms of skeletal muscle fatigue: the Marion J. Siegman Award Lectureships. *Am J Physiol Cell Physiol*, *327*(4), C946-C958. https://doi.org/10.1152/ajpcell.00213.2024

Eng, J. (2003). Sample Size Estimation: How many individuals should be studied? *Radiol.*, *227*(2), 309-313.

Enoka, R. M., & Duchateau, J. (2016). Translating Fatigue to Human Performance. *Med Sci Sports Exerc*, *48*(11), 2228-2238. https://doi.org/10.1249/MSS.0000000000000929

Gandevia, S. C. (2001). Spinal and supraspinal factors in human muscle fatigue. *Physiol Rev.*, *81*(4), 1725-1789.

Halperin, I., & Emanuel, A. (2019). Rating of Perceived Effort: Methodological Concerns and Future Directions. *Sports Med.*, 1-9. https://doi.org/10.1007/s40279-019-01229-z

Krieger, J. W. (2010). Single vs. multiple sets of resistance exercise for muscle hypertrophy: a meta-analysis. *J Strength Cond Res*, *24*(4), 1150–1159. https://doi.org/10.1519/JSC.0b013e3181d4d436

Loenneke, J. P., Wilson, J. M., Wilson, G. J., Pujol, T. J., & Bemben, M. G. (2011). Potential safety issues with blood flow restriction training. *Scand J Med Sci Sports*, *21*(4), 510-518. https://doi.org/10.1111/j.1600-0838.2010.01290.x

Marchetti, P. H. (2023). *Strength Training Manual: Applied Science* (3 ed.). Kendall Hunt Publishing Company.

Marchetti, P. H., Samaniego, A., Talamantez, R., Lopes, C. R., & Martinez, V. G. F. (2023). Acute effects of different number of sets and non-equalized volume on muscle thickness, peak force, and physical performance in recreationally-trained participants. *Journal of Kinesiology and Wellness*, *12*(1), 10–19.

Maulder, P., & Cronin, J. (2005). Horizontal and vertical jump assessment: reliability, symmetry, discriminative and predictive ability. *Phys Ther Sport*, *6*(2), 74–82. https://doi.org/10.1016/j.ptsp.2005.01.001

Nordin, M., & Frankel, V. H. (2021). *Basic Biomechanics of the Musculoskeletal System* (5 ed.). Wolters Kluwer.

Pareja-Blanco, F., Rodriguez-Rosell, D., Aagaard, P., Sanchez-Medina, L., Ribas-Serna, J., Mora-Custodio, R., Otero-Esquina, C., Yanez-Garcia, J. M., & Gonzalez-Badillo, J. J. (2020). Time Course of Recovery From Resistance Exercise With Different Set Configurations. *J Strength Cond Res*, *34*(10), 2867–2876. https://doi.org/10.1519/JSC.0000000000002756

Purslow, P. P. (2002). The structure and functional significance of variations in the connective tissue within muscle. *Comp Biochem Physiol A Mol Integr Physiol*, *133*(4), 947–966. https://doi.org/10.1016/s1095-6433(02)00141-1

Radaelli, R., Fleck, S. J., Leite, T., Leite, R. D., Pinto, R. S., Fernandes, L., & Simao, R. (2015). Dose-response of 1, 3, and 5 sets of resistance exercise on strength, local muscular endurance, and hypertrophy. *J Strength Cond Res*, *29*(5), 1349–1358. https://doi.org/10.1519/JSC.0000000000000758

Schoenfeld, B. J. (2010). The mechanisms of muscle hypertrophy and their application to resistance training. *J Strength Cond Res*, *24*(10), 2857–2872.

Schoenfeld, B. J. (2013). Potential mechanisms for a role of metabolic stress in hypertrophic adaptations to resistance training. *Sports Med.*, *43*(3), 179–194. https://doi.org/10.1007/s40279-013-0017-1

Schoenfeld, B. J., & Contreras, B. (2014). The Muscle Pump: Potential Mechanisms and Applications for Enhancing Hypertrophic Adaptations. *Strength Cond J.*, *36*(3), 21–25. https://doi.org/10.1097/SSC.0000000000000021

Schoenfeld, B. J., Contreras, B., Krieger, J., Grgic, J., Delcastillo, K., Belliard, R., & Alto, A. (2019). Resistance Training Volume Enhances Muscle Hypertrophy but Not Strength in Trained Men. *Med Sci Sports Exerc.*, *51*(1), 94-103.

Schoenfeld, B. J., Grgic, J., Ogborn, D., & Krieger, J. W. (2017). Strength and Hypertrophy Adaptations Between Low- vs. High-Load Resistance Training: A Systematic Review and Meta-analysis. *J Strength Cond Res*, *31*(12), 3508–3523. https://doi.org/10.1519/JSC.0000000000002200

Sjøgaard, G., Adams, R. P., & Saltin, B. (1985). Water and ion shifts in skeletal muscle of humans with intense dynamic knee extension. *Am J Physiol.*, *248*(2), R190-196.

Stock, M. S., & Thompson, B. J. (2021). Echo intensity as an indicator of skeletal muscle quality: applications, methodology, and future directions. *Eur J Appl Physiol*, *121*(2), 369–380. https://doi.org/10.1007/s00421-020-04556-6

Sullivan, K., Ferreira, P. R., Martinez, V. G. F., & Marchetti, P. H. (2023). Muscle Thickness, Echo-Intensity, Peak Force, Time Under Tension, Total Load Lifted, and Perception of Effort Comparisons Between Two Abdominal Crunch Resistance Training Protocols in Recreationally-Trained Participants. *Int J Exerc Sci*, *16*(6), 538–549. https://www.ncbi.nlm.nih.gov/pubmed/37621710

Sweet, T. W., Foster, C., McGuigan, M. R., & Brice, G. (2004). Quantitation of resistance training using the session rating of perceived exertion method. *J Strength Cond Res*, *18*(4), 796–802.

Usher-Smith, J. A., Huang, C. L., & Fraser, J. A. (2009). Control of cell volume in skeletal muscle. *Biol Rev Camb Philos Soc.*, *84*(1), 143-159.

Weilharter, F., Rewitz, K., Halperin, I., & Wolff, W. (2024). The relationship between prescribed ratings of perceived exertion and force production in repeated isometric contractions. *Psychol Sport Exerc*, *73*, 102657. https://doi.org/10.1016/j.psychsport.2024.102657

Westerblad, H., & Allen, D. G. (2003). Cellular mechanisms of skeletal muscle fatigue. *Adv Exp Med Biol*, *538*, 563–570; discussion 571. https://doi.org/10.1007/978-1-4419-9029-7_50

Wong, V., Spitz, R. W., Bell, Z. W., Viana, R. B., Chatakondi, R. N., Abe, T., & Loenneke, J. P. (2020a). Exercise induced changes in echo intensity within the muscle: a brief review. *J Ultrasound*, *23*(4), 457–472. https://doi.org/10.1007/s40477-019-00424-y

Yitzchaki, N., Kuehne, T. E., Mouser, J. G., & Buckner, S. L. (2019). Can changes in echo intensity be used to detect the presence of acute muscle swelling? *Physiol Meas*, *40*(4), 045002.

Corresponding author:
Paulo Marchetti
Resistance Training Laboratory
Department of Kinesiology,
California State University-Northridge,
Northridge, United States of America
E-mail: dr.pmarchetti@gmail.com