



Original Research

Muscle Architecture and Strength Changes Induced by Different Resistance Training Frequencies and Detraining

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ABSTRACT

International Journal of Exercise Science 15(4): 1661-1679, 2022. The purpose of the present study was to investigate muscle thickness and strength outcomes of the quadriceps femoris induced by different resistance training (RT) frequencies and detraining. In addition, muscle architecture (MA) parameters were also assessed. Twenty-seven healthy resistance-trained subjects (men, $n = 17$; women, $n = 10$; 20.8 ± 1.9 years; RT experience = 3.3 ± 1.6 years) volunteered to participate in this study. One leg of each subject was randomly allocated into the 2 sessions per week condition (2x) and the contralateral leg was then placed in the 4 sessions per week condition (4x). There were 16 RT sessions in 2x and 4x. After 4 weeks, 4x were divided into 2 other conditions: more 4 weeks with 2x(4x (+2x)) and detraining (4x (+Det)). Muscle thickness (MT), fascicle length (FL), pennation angle (PA) of the quadriceps muscles and one-repetition maximum for unilateral knee extension ($1RM_{KE}$) were evaluated. A significant increase of $1RM_{KE}$ in 2x, 4x, and 4x (+2x) and a decrease in 4x (+Det) was observed (all $p < 0.05$). The MA showed similar results in most dependent variables for MT, FL and PA. Specifically 4x (+Det) condition demonstrated antagonistic results when compared to the 4x (+2x) in MT of rectus femoris ($p = 0.001$) and increased FL in vastus intermedius ($p = 0.001$).

KEY WORDS: Muscle thickness; fascicle length; pennation angle; regional difference; regional hypertrophy; muscle hypertrophy

INTRODUCTION

Resistance training (RT) is considered as the most effective method to develop muscle strength and hypertrophy, and to change muscle architecture (MA) parameters (e.g., increases in pennation angle and fascicle length) (1, 21, 53). The magnitude of such muscular adaptations is related to the adequate configuration of training variables. Training frequency is, therefore, considered a determinant variable in the hypertrophic response to a given RT program (13, 22).

Resistance training frequency refers to the number of sessions performed during a specific period, usually described on a weekly basis. From a hypertrophic standpoint, frequency can be characterized as the number of sessions per week (sessions \cdot wk⁻¹) in which the same muscle group is trained (11).

In this context, higher training frequencies can help to accumulate greater volumes of training, which may in turn enhance the hypertrophic response (40). However, systematic reviews with meta-analysis showed no significant difference between higher and lower frequency on a volume-equated basis for both muscle strength (18) and hypertrophy (40). Therefore, higher weekly RT frequencies seems to exert an influence on muscle strength and hypertrophy gains only when it results in higher total load lifted (TLL – sets \times repetitions \times load [kg]) compared to low frequencies (40, 52). In fact, it has been shown that increases in muscle strength and hypertrophy are strongly dependent on TLL of RT. Accordingly, studies have shown greater increments in muscle strength and hypertrophy for high TLL protocols when compared to low TLL ones (12, 35, 41). Conversely, equalized TLL RT protocols have not shown differences in muscle strength and hypertrophy responses in spite of distinct manipulations of RT variables (4, 24, 45).

Although it is well established that the weekly RT frequency does not influence the morphofunctional adaptations on a volume-equated basis, to the best of the authors' knowledge, it is still unknown whether the application of a specific number of RT training sessions (e.g., 16 sessions), performed in mesocycles that differ in the number of weeks (duration) and weekly frequency employed (8 weeks with 2 sessions \cdot wk⁻¹ versus 4 weeks with 4 sessions \cdot wk⁻¹, both culminating in 16 RT sessions), result in different responses regarding muscle strength and hypertrophy.

In addition, RT periodization might encompass periods of reduced training load due to competitions or changes in individuals' daily routine. Usually, reduced resistance training (RRT) is a strategy in which both training volume and/or frequency are decreased but intensity is maintained. Thus, different RRT strategies may be organized as an attempt to avoid detraining effects (e.g. muscle atrophy and strength decrease) and to maintain previously acquired morphofunctional adaptations (10, 30, 33).

In order to avoid detraining effects, several studies that have investigated RRT used a low training frequency (one or two sessions \cdot wk⁻¹) associated with a reduction of ~30– 60% in training volume (8, 20, 45, 46). For example, Trappe et al. (46) used a RRT scheme during 12 weeks, 1 session \cdot wk⁻¹ with a reduction of ~50% in total volume. The authors found maintenance in the quadriceps cross sectional area (QCSA) and maximal dynamic strength (one repetition maximum [1-RM]) in the leg extension exercise after the RRT period while a 5% reduction in QCSA and 11% in 1RM were observed in the cessation training group. In another study, Tavares et al. (45) conducted a experiment which training volume was reduced by ~50% with 1 or 2 sessions \cdot wk⁻¹ for 8 weeks following a 8-week (3 sessions \cdot wk⁻¹) RT programme. No significant changes were observed in half-squat 1-RM and QCSA for both RRT groups after the RRT period,

while the ceased training group demonstrated a decrease in half-squat 1-RM (-22.6%) and QCSA (-5.4%) when compared to the initial 8-week RT period.

Although several studies have investigated the RRT in different populations, to the best of the authors' knowledge, no study has investigated the effect of RRT on strength and hypertrophy in resistance-trained subjects. In light of these topics, the aim of the present investigation was two-fold. Firstly, to compare the effects of 2 different mesocycles on muscle strength and hypertrophy in resistance-trained subjects: one composed of 16 RT sessions distributed over 8 weeks with a frequency of 2 sessions \cdot wk⁻¹ (2x) versus 16 RT sessions distributed over 4 weeks with a frequency of 4 sessions \cdot wk⁻¹ (4x). Secondly, after a 4x period, to compare the effects of an additional 4 week-period of RRT with a frequency of 2 sessions \cdot wk⁻¹ and a ~50% reduction in RT volume versus a detraining period on muscle strength and hypertrophy. As a secondary aim, we compared the effects of these protocols on some muscle architecture parameters. Our hypothesis was that 2x and 4x RT schemes would yield similar muscle strength and hypertrophy adaptations, since both RT interventions were conducted on a volume-equated basis. Moreover, we hypothesized that morphofunctional adaptations would be preserved in the RRT condition, whereas the detraining condition would display significant reductions in both strength and muscle morphology.

METHODS

Participants

Twenty-seven healthy resistance-trained subjects (men, $n = 17$; women, $n = 10$; 20.8 ± 1.9 years [range 19 to 25 years]; 1.73 ± 9.8 cm; total body mass = 73.2 ± 11.7 kg; RT experience = 3.3 ± 1.6 years [range 2.6 to 4.0 years]; RT frequency = 4.4 ± 0.7 sessions per week; number of sets performed per week for quadriceps before the study commencement = 27.4 ± 12.3 sets per week) volunteered to participate in this study. The sample size was justified by a priori power analysis based on a pilot study where the vastus lateralis muscle thickness (MT) was assessed as the outcome measure with a target effect size difference of 0.75, an alpha level of 0.05, and a power ($1 - \beta$) of 0.80 (15). All subjects were resistance-trained; performing RT on a minimum of 3 days per week for at least 1 year at the university's RT gym. All subjects regularly performed (minimum frequency of once a week) the exercise utilized in the training intervention and in the strength tests for at least 1 year before entering the study. Moreover, subjects were free from any existing musculoskeletal disorders; history of injury with residual symptoms (pain, "giving-away" sensations) in the trunk and lower limbs within the last year and stated they had not taken anabolic steroids or any other illegal agents known to increase muscle size currently and for the previous year. Thus, participation in the study required that the subjects answered negatively to all questions on the Physical Activity Readiness Questionnaire (PAR-Q). All participants read and signed an informed consent prior to participating in the study. This study was approved by the University's research ethics committee and was conducted in accordance with the Declaration of Helsinki and the ethical standards of the International Journal of Exercise Science (32).

Protocol

The present study followed a randomized and longitudinal design by the dominant limb and gender. Parallel groups are not suited to test the problem at hand because differences in strength and MA parameters between groups may be due to between-subject variability and not to changes in training frequency and detraining (37). This study did not employ the use of a control group. Instead, a within-subjects design was used where pre-training (Pre) assessments of muscle morphology and strength were compared to those observed following 4 or 8 weeks of resistance training and detraining (Post) (28). In this case, a within-subject design greatly reduces the between-subject variance in the statistical model (37). Thus, in order to reduce inter-subjects variability, a unilateral design was used. Each participant's leg was considered as an experimental unit. One leg of each subject was randomly allocated in the 2x (13 dominant and 14 nondominant legs) and the contralateral leg was then allocated in the 4x (13 dominant and 14 nondominant legs). In both conditions, the same number of training sessions were completed: 16 RT sessions distributed over 8 weeks with a frequency of 2 sessions wk^{-1} (2x), and 16 RT sessions distributed over 4 weeks with a frequency of 4 sessions wk^{-1} (4x).

After four weeks, 4x was divided into two conditions: 4 more weeks of RRT with training frequency of 2x (8 more RT sessions) or detraining. Experimental conditions were as follows: two-session- wk^{-1} (2x); four-session- wk^{-1} (4x); four-session- wk^{-1} + two-session- wk^{-1} (4x (+2x)); four-session- wk^{-1} + detraining (4x (+Det)). In the first week, volunteers attended 2 familiarization sessions in the laboratory. The subjects refrained from all physical exercises other than activities of daily living for at least 48 hours before the first familiarization session. In the first session, the anthropometric and MA parameters of quadriceps femoris muscles were evaluated. Then, volunteers were familiarized with the maximal dynamic strength test for unilateral knee extension (1RM_{KE}). The following day (24 hours after), volunteers were familiarized with standard procedures adopted in the RT session, such as body position, cadence, range of motion, rest, etc. and performed a maximal dynamic strength test for unilateral knee extension (1RM_{KE}). The training intervention period lasted 8 weeks and MA and 1RM_{KE} evaluations were performed after four and eight weeks of RT.

1-RM test: the unilateral 1-RM test in leg extension exercise (1RM_{KE}) was performed following the recommendations described by Scarpelli (37). Initially, participants performed a general warm-up consisting of five minutes in a cycle ergometer (Schwinne; AC Sport, Vancouver, WA) at 60–70 rpm and 50 W, followed by two sets of specific warm-up. The first set consisted of 8 repetitions with 50% of the estimated 1-RM, and the second set comprised 3 repetitions with 70% of the estimated 1-RM with a 2-min rest between warm-up sets. After the warm-up, the 1-RM test was initiated. Participants had up to 5 attempts to reach their 1-RM load on each leg, with a rest of 3 min between attempts. The greatest load lifted was considered as the 1-RM load. The CV% and the typical error of the measurement (TEM) for the 1RM_{KE} was 3.05% (CI 95% = 1.90 to 4.20%) and 0.5 kg (CI 95% = 0.3 to 0.7 kg), respectively.

The RT protocol consisted of 12 sets of unilateral knee extension exercise (Portico Fitness Inc., Itatiba, SP, Brazil). Subjects were instructed to refrain from any additional resistance-type

training for the lower limbs during the study. They were positioned seated with the hip and knee flexed at 90° and were instructed to perform all repetitions from 90 degrees of knee flexion to 0 degrees of knee extension. At the beginning of each session, subjects performed a specific warm-up of 2 sets of 8 repetitions with a load between 40 and 60% of 10RM (evaluated in the second familiarization session). Each set involved 8–12RM with 60 seconds of rest afforded between sets. All sets were performed to the point of momentary concentric muscular failure. The external load (kg) was adjusted, as needed, on successive sets to ensure that subjects achieved failure in the target repetition range. The cadence of repetitions was conducted in a controlled fashion, with concentric and eccentric actions of approximately 1.5 seconds, for total repetition duration of approximately 3 seconds. All routines were directly supervised by the research assistants to ensure the correct performance of the respective routines. Attempts were made to progressively increase the load lifted each week while maintaining the target repetition range. The adherence to the program was 100% for all conditions.

During the first four weeks of RT (2x and 4x intervention period), five subjects withdrew because of personal reasons not related to the current study. Additionally, during the last four weeks of RT (RRT and detraining period), one subject withdrew because of patellar tendinopathy. Therefore, data from 44 legs were included in the statistical analysis of 2x ($n = 22$) and 4x ($n = 22$), and data from 21 legs were included in the statistical analysis of 4x (+2x) ($n = 10$) and 4x (+Det) ($n = 11$).

Total load lifted (TLL): the TLL was calculated from training logs filled out by research assistants for every RT session ($TLL = \text{sets} \times \text{repetitions} \times \text{external load [kg]}$). The accumulated TLL was the sum of all RT weeks (1 to 16th session). Additionally, an additional statistical analysis was performed for the calculation of the TLL. Each condition was divided in two: from the 1 to 8th session vs 9 to 16th session for the 2x and 4x conditions, as well as the TLL of the 9 to 16th session vs 17 to 24th session for the 4x (+2x) condition. The 2x(4x (+2x) condition initiated from the 9th session because it started after 4 weeks (see methods). Only repetitions performed through a full range of motion were included for analysis. The data were expressed in kilograms (kg).

Muscle architecture: for muscle architecture parameters were measured using B-mode ultrasound LOGIC L3, (General Electric Healthcare®, Wauwatosa, Wisconsin, USA), with a 45-mm, 12.0-MHz linear-array probe and a water-soluble transmission gel (Mercur S.A. – Body Care, Santa Cruz do Sul, RS, Brazil). To obtain the images the subjects lay supine with their legs fully extended and their muscles relaxed. The transducer was orienting perpendicular and without pressing to the skin. The measurements were done after the participant rested in a horizontal position for 20 min to allow fluid shifts to stabilize (28). The muscle thicknesses (MT), fascicle length (FL), and pennation angle (PA) (Figure 1) of each muscle of the quadriceps femoris was measured at pre-intervention, post-four-week, and post-eight-week.

The MT, FL and PA assessment was conducted following previous procedures by Ema et al (14). Measurements were performed at least in two regions (distal and proximal) of the rectus femoris (RF), vastus lateralis (VL), vastus medialis (VM), and vastus intermedius (VI). For VI,

measurements were also performed in the medial and lateral regions (14). The measurement positions were assigned along the thigh length, considered the distance from the lateral condyle of the femur to the palpable center of the greater trochanter (Figure 1). The measurement regions were: RF – 70% and 50%, VL – 55% and 35%, VM – 35% and 15%, VI – 55% and 35% (Figure 1).

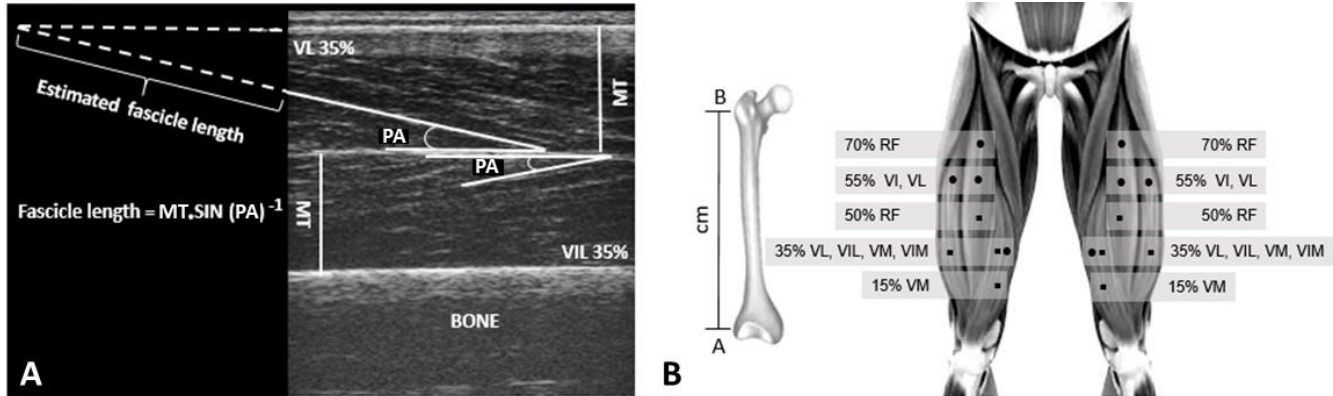


Figure 1. (A) Muscle architecture measurements: MT = muscle thickness; PA = pennation angle; FL = fascicle length; SIN = sine. (B) Positions of measurements imaging for ultrasonography: A = lateral condyle; B = greater trochanter; VL = vastus lateralis; VM = vastus medialis; RF = rectus femoris; VI = vastus intermedius; • = proximal point; ▪ = distal point.

In the post-intervention of four and eight weeks, images were obtained 72 hours after the last training session. To maintain consistency between the testing, each site was marked with henna ink (reinforced every week). The images were collected in duplicates, with an interval of 10s between them and the reproducibility (internal consistency) of ultrasound measurements was examined utilized the software MicroDicom® (version 3.2.7).

Statistical Analysis

The normality and homogeneity of the variances were verified using the Shapiro-Wilk and Levene tests, respectively. Prior to analysis, all data were log-transformed for analysis to reduce bias arising from non-uniformity error (heteroscedasticity). The mean, standard deviation (SD), 90% and 95% confidence intervals (CI) were used after data normality was assumed. To compare mean values of the accumulated TLL in 2x vs 4x a paired *t*-test was used (condition effect). The same procedure was used for the comparison between the TLL of 1 to 8th session vs 9 to 16th session for each training frequency (time effect) and TLL of 9 to 16th session vs 17 to 24th session in condition 4x (+2x) (time effect). A repeated measures analysis of variance (ANOVA) was used to compare time effect (pre vs post) and conditions (2x vs 4x, *n* = 22) for 1RM_{KE}, muscle thickness, fascicle length and pennation angle in each muscle (RF, VL, VM, VI). As a result of our experimental design (division of condition 4x - *n* = 11), another ANOVA was performed separately to compare time effect (pre vs post) and conditions 4x (+2x) vs 4x (+Det) in same variables. Post hoc comparisons were performed with the Bonferroni correction. Assumptions of sphericity were evaluated using Mauchly's test. Where sphericity was violated (*p* < 0.05), the Greenhouse-Geisser correction factor was applied. To assess whether the observed differences could be considered real, changes were compared to their calculated smallest worthwhile change (SWC) for all dependent variables (1RM_{KE}, muscle thickness, fascicle length and

pennation angle in each muscle) (44). SWC was calculated by the formula ($SWC = TEM \times 1.96 \times \sqrt{2}$) (50). We defined an individual as “responding” to training with a response greater than 1SWC from zero for increases in dependent-variables; if not, he was considered the nonresponder. Percentage of legs exceeding the SWC were calculated for all dependent-variables (28). All analyses were conducted in SPSS-22.0 software (IBM Corp., Armonk, NY, USA). The adopted significance was $p \leq 0.05$. The figures were formatted in GraphPad Prism version 6.0 software (La Jolla, CA, USA) following the assumptions for continuous data (51).

RESULTS

1-RM test: a significant increase of $1RM_{KE}$ in the pre vs post comparison for 2x ($\Delta = 33.0\%$, $p = 0.001$, $ES = 0.96$), 4x ($\Delta = 26.7\%$, $p = 0.001$, $ES = 0.86$) and 4x (+2x) ($\Delta = 22.5\%$, $p = 0.002$, $ES = 0.81$) was observed. In condition 4x (+Det), $1RM_{KE}$ presented a significant decrease ($\Delta = -16.7\%$, $p = 0.021$, $ES = 0.60$) (Figure 2A). In contrast, 100% of sample in the 4x (+Det) condition presented significant reductions in 1RM values (Figure 2B).

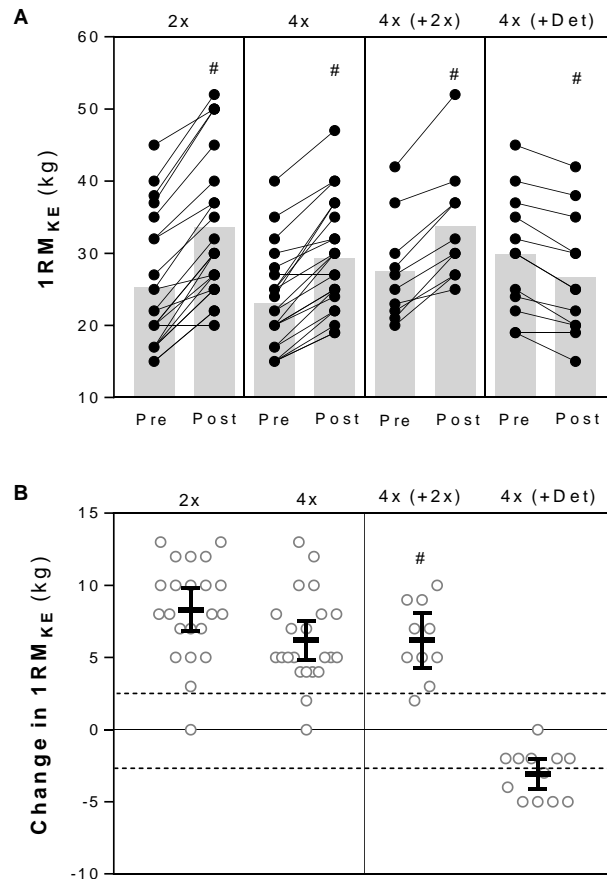


Figure 2. One repetition maximum test in knee extension ($1RM_{KE}$) for each training frequency at pre and post of resistance training. Bars are means and circles are individual values (2A). Mean with 95% CI of individual absolute changes (kg) in $1RM_{KE}$ in relation to pre values (2B). Dashed line indicates the $SWC = 2.8$ kg. Legend: 2x = two session $\cdot wk^{-1}$; 4x = four session $\cdot wk^{-1}$; 4x (+Det) = four session $\cdot wk^{-1}$ + detraining; 4x (+2x) = four session $\cdot wk^{-1}$ + two session $\cdot wk^{-1}$; # $p < 0.05$ vs 4x (+Det).

TLL: No significant difference was noted between conditions 2x and 4x for accumulated TLL ($p = 0.713$, $2x = 51934 \pm 19884$ kg vs $4x = 49804 \pm 18212$ kg) (Figure 3A). Both conditions presented increase in TLL from 1 to 8th session to 9 to 16th session ($2x = 15.4\%$, $p = 0.01$; $4x = 16.5\%$, $p = 0.01$). In condition 4x (+2x), there was also an increase in the TLL of the 9 to 16th session vs 17 to 24th session ($\Delta = 15.5\%$, $p = 0.01$) (Figure 3B).

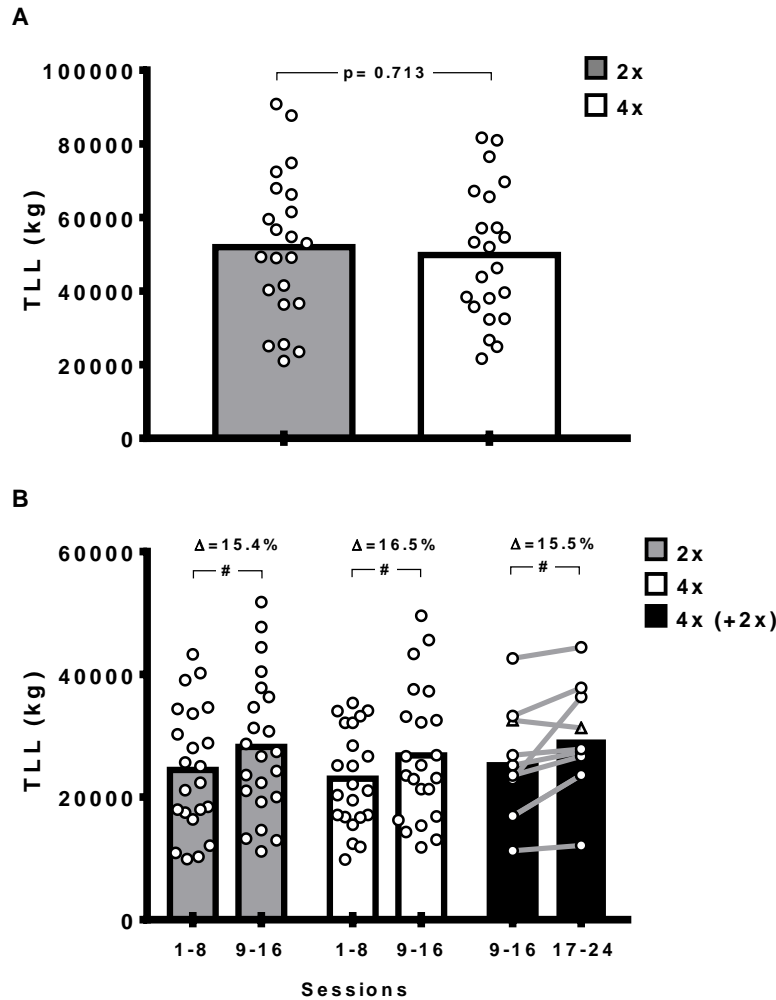


Figure 3. Weekly total load lifted (TLL) for each training frequency (2x vs 4x) at 16 sessions of RT (3A) and 1 to 8th session vs 9 to 16th session (4B). Black columns represented subjects in the 4x condition who maintained RT for two session per week (4x (+2x)), 9 to 16th session vs 17 to 24th session and line with circles demonstrate individual analysis. Triangle represented negative individual response in TLL (3B).

Muscle architecture: the RF showed an increase in MT in the distal and proximal region for all experimental conditions except 4x(+Det). A significant decrease was observed to 4x (+Det) in the RF proximal region ($p = 0.002$) (Table 1). Similarly, a significant increase was observed for VL in distal region (all $p < 0.05$), except for the 4x (+Det) condition ($p = 0.182$). For VM, both regions (distal and proximal) increased MT after training period in 2x ($p = 0.022$) and 4x ($p = 0.018$), however, there was no improvement for 4x (+2x) ($p > 0.05$) and a significant decrease for 4x (+Det) ($p < 0.05$). The distal (medial) region of the VI showed an increase in conditions 2x, 4x

and 4x (+ 2x) (all $p < 0.05$) and a reduction in 4x (+Det) ($p = 0.028$) (Table 1). Similarly, individual analysis demonstrated a larger percentage of legs experienced changes in MT of the RF PROXIMAL (2x = 63.6%; 4x = 68.%, 4x (+2x) = 80.0%) and VL DISTAL (2x = 68.2%; 4x = 54.5%, 4x (+2x) = 60.0%). In contrast, a low percentage of participants demonstrated changes in MT of the RF PROXIMAL (0.0%), RF DISTAL (9.7%), VL PROXIMAL (18.2%), VL DISTAL (36.4%), VI PROXIMAL (27.3%) and, VI DISTAL (36.4%) in 4x (+Det) condition (Table 2).

The RF muscle showed an increase in FL in the distal region for all experimental conditions (all $p < 0.05$), and decreased in the detraining condition ($p = 0.014$, mean difference = -18.6 mm, 95%CI = -31.5 to -9.4 mm) (Table 1). However, a decrease in FL was observed in the VM PROXIMAL and VM DISTAL for conditions 2x and 4x ($p < 0.05$). Although no statistical differences were observed in 4x (+2x), a low percentage of participants experienced changes for the FL in VM PROXIMAL (40%, mean difference = -13.5 mm, 95%CI = -27.3 to 0.3 mm) and VM DISTAL (40%, mean difference = -3.9 mm, 95%CI = -9.1 to 1.2 mm). Similar adaptations between conditions have been found regarding the decrease in FL of the VI in distal lateral and distal medial regions for 2x (all $p < 0.05$), distal lateral for 4x ($p = 0.002$), proximal ($p = 0.018$) and distal medial ($p = 0.032$) for 4x (+2x). However, increased in FL of VI were observed in detraining for proximal ($p = 0.004$) and distal medial regions ($p = 0.009$) (Table 1). These results are also supported by comparison 4x (+2x) vs 4x (+Det) in FL of the VI proximal ($p = 0.001$) and distal ($p = 0.001$).

A significant increase in PA was noted for RT_{PROXIMAL} in 4x and 4x(+2x) and a decrease in PA of the RF DISTAL was observed. The PA of the VL DISTAL increased for all training frequencies (2x or 4x or 4x (+ 2x)). A significant decrease for PA of the VL PROXIMAL ($p = 0.009$, mean difference = -2.8°, 95%CI = -5.1 to -0.5°) was observed for the 4x (+Det) condition (Table 1). Similarly, individual analysis displayed a higher percentage of legs presenting changes in PA of the VL DISTAL (2x = 50.0%; 4x = 40.9%, 4x (+2x) = 60.0%) and a low percentage in 4x (+Det) (36.4%) (Table 2). Both conditions (2x and 4x) increased the PA of the VM DISTAL (all $p < 0.05$). The 4x (+Det) condition resulted in a decreased in the PA of the VM DISTAL ($p = 0.013$, mean difference = -3.5°, 95%CI = -6.5 to -0.5°) and this change was significantly different when compared to 4x (+2x) ($p = 0.001$) (Table 2). Furthermore, regardless of the weekly training frequency (2x or 4x) the VI in distal lateral and distal proximal regions increased PA (all $p < 0.05$). Detraining caused a decrease in PA of the regions proximal and distal medial of the VI (all $p < 0.05$) (Table 1).

Table 1. Architecture values of muscles RF, VL, VM and VI for each resistance training frequency.

Muscle	Condition	Region	Time	Muscle thickness (mm)	Fascicle length (mm)	Pennation angle (°)
RF	2x	proximal	pre	24.9 ± 2.6	98.0 ± 17.7	15.0 ± 3.4
			post	26.7 ± 2.5 [#]	103.8 ± 16.6	14.9 ± 2.3
		distal	pre	22.6 ± 2.5	108.3 ± 9.1	11.6 ± 2.3
			post	24.2 ± 2.2 [#]	127.5 ± 8.3 [#]	10.9 ± 1.7
	4x	proximal	pre	24.5 ± 2.0	102.8 ± 21.6	15.1 ± 1.4
			post	26.0 ± 2.5 [#]	101.6 ± 14.6	16.9 ± 1.8 [#]
		distal	pre	22.5 ± 2.2	112.8 ± 14.8	10.9 ± 1.0

VL	4x (+2x)	proximal	post	23.9 ± 2.2 [#]	122.9 ± 10.8 [#]	11.6 ± 1.6	
			pre	25.2 ± 1.7	88.7 ± 16.0	15.8 ± 1.7	
			post	27.2 ± 2.4 [#]	104.6 ± 25.3	16.7 ± 1.3 [#]	
		distal	pre	21.7 ± 2.3	110.4 ± 19.8	11.6 ± 0.8	
			post	24.3 ± 1.5 [#]	149.9 ± 19.4 [#]	11.5 ± 0.6	
			pre	26.5 ± 2.7	104.9 ± 10.4	14.4 ± 1.8	
	4x (+Det)	proximal	post	23.6 ± 2.2 [#]	110.9 ± 9.6	13.4 ± 1.6	
			pre	22.8 ± 2.4	119.7 ± 12.3	11.1 ± 1.3	
			post	21.2 ± 1.5	101.1 ± 10.1 [#]	9.6 ± 0.8 [#]	
		2x	proximal	pre	24.0 ± 2.9	92.1 ± 13.9	16.8 ± 3.3
				post	24.9 ± 2.9	98.3 ± 19.4	15.6 ± 2.5
		4x	distal	pre	23.0 ± 3.8	79.8 ± 9.7	15.5 ± 1.9
post	25.6 ± 3.2 [#]			81.3 ± 12.5	17.4 ± 2.0 [#]		
proximal	pre		24.7 ± 2.5	89.1 ± 12.9	15.8 ± 2.1		
	post		25.5 ± 3.1	91.7 ± 8.7	16.9 ± 2.0		
4x (+2x)	distal	pre	24.7 ± 2.1	82.3 ± 11.7	16.1 ± 1.6		
		post	26.0 ± 2.7 [#]	88.6 ± 8.7	17.6 ± 2.1 [#]		
		pre	25.7 ± 1.7	96.5 ± 5.3	16.4 ± 2.0		
	proximal	post	24.5 ± 3.1	108.0 ± 12.2	13.1 ± 1.6		
		pre	22.3 ± 1.9	93.8 ± 9.5	15.5 ± 1.5		
		post	24.4 ± 2.3 [#]	82.6 ± 7.9	16.7 ± 1.3 [#]		
4x (+Det)	proximal	pre	24.9 ± 4.6	86.7 ± 2.9	17.9 ± 1.1		
		post	24.9 ± 1.6	86.4 ± 5.6	15.1 ± 1.2 [#]		
		pre	24.7 ± 4.7	85.2 ± 3.5	17.3 ± 3.3		
	distal	post	25.0 ± 3.2	73.7 ± 3.0 [#]	18.8 ± 0.6		
		pre	26.3 ± 5.1	100.7 ± 22.7	15.1 ± 2.1		
		post	23.5 ± 4.7 [#]	82.6 ± 13.6 [#]	15.3 ± 3.1		
VM	2x	proximal	pre	26.3 ± 5.1	100.7 ± 22.7	15.1 ± 2.1	
			post	23.5 ± 4.7 [#]	82.6 ± 13.6 [#]	15.3 ± 3.1	
			pre	28.7 ± 5.0	85.5 ± 14.5	20.9 ± 4.6	
		distal	post	26.0 ± 3.3 [#]	58.2 ± 6.9 [#]	28.2 ± 6.8 [#]	
			pre	27.6 ± 4.1	105.6 ± 12.6	15.6 ± 2.7	
			post	23.4 ± 3.8 [#]	89.5 ± 13.8 [#]	15.1 ± 2.1	
	4x	proximal	pre	29.4 ± 5.3	82.4 ± 9.7	21.7 ± 4.0	
			post	26.9 ± 3.3 [#]	67.5 ± 6.8 [#]	27.4 ± 6.5 [#]	
			pre	23.7 ± 4.1	89.8 ± 6.0	14.9 ± 2.3	
		distal	post	23.4 ± 3.5	76.3 ± 13.6	17.9 ± 3.3	
			pre	28.0 ± 3.6	68.5 ± 8.0	25.7 ± 6.7	
			post	29.2 ± 5.1	64.6 ± 5.5	28.4 ± 7.6	
4x (+2x)	proximal	pre	23.6 ± 3.5	90.7 ± 20.5	15.4 ± 1.9		
		post	22.7 ± 2.2 [#]	89.2 ± 17.3	15.1 ± 2.8		
		pre	27.1 ± 3.0	66.9 ± 7.2	25.5 ± 3.2		
	distal	post	26.4 ± 3.9 [#]	66.5 ± 5.2	22.0 ± 4.5 [#]		
		pre	22.7 ± 4.3	88.8 ± 4.7	15.2 ± 1.8		
		post	22.9 ± 3.5	89.9 ± 4.3	14.2 ± 2.6		
VI	2x	proximal	pre	19.0 ± 2.7	85.4 ± 4.1	12.6 ± 1.6	
			post	17.2 ± 2.3	73.1 ± 5.7 [#]	14.2 ± 1.1 [#]	
	distal (lateral)	pre	19.0 ± 2.7	85.4 ± 4.1	12.6 ± 1.6		
		post	17.2 ± 2.3	73.1 ± 5.7 [#]	14.2 ± 1.1 [#]		

4x	distal (medial)	pre	13.0 ± 2.8	65.8 ± 3.6	11.6 ± 1.2
		post	15.0 ± 2.0 [#]	57.5 ± 4.7 [#]	13.7 ± 1.5 [#]
	proximal	pre	22.1 ± 3.4	96.9 ± 17.5	13.5 ± 2.7
		post	23.4 ± 2.7	95.1 ± 9.9	14.2 ± 1.4
4x (+2x)	distal (lateral)	pre	18.6 ± 3.7	84.1 ± 5.2	12.8 ± 1.2
		post	17.2 ± 1.8	78.3 ± 3.4 [#]	14.5 ± 0.9 [#]
	distal (medial)	pre	12.6 ± 2.4	64.1 ± 6.3	10.1 ± 1.2
		post	15.1 ± 2.0 [#]	60.4 ± 10.7	11.9 ± 1.3 [#]
4x (+Det)	proximal	pre	24.6 ± 1.2	102.1 ± 5.2	14.1 ± 1.6
		post	23.3 ± 3.4	84.2 ± 5.4 [#]	16.8 ± 1.2 [#]
	distal (lateral)	pre	17.7 ± 2.5	81.5 ± 12.4	12.6 ± 1.0
		post	16.6 ± 2.3	83.0 ± 9.9	11.3 ± 2.1
4x (+Det)	distal (medial)	pre	12.4 ± 2.2	63.5 ± 4.1	11.0 ± 1.3
		post	15.0 ± 2.3 [#]	56.1 ± 5.7 [#]	13.6 ± 0.9 [#]
	proximal	pre	21.6 ± 2.6	86.7 ± 5.3	14.1 ± 1.1
		post	22.5 ± 2.9	103.0 ± 6.9 [#]	12.2 ± 1.2 [#]
distal (lateral)	pre	16.2 ± 1.7	73.6 ± 5.1	12.2 ± 1.0	
	post	18.6 ± 1.8	79.0 ± 13.5	13.4 ± 1.6	
distal (medial)	pre	14.0 ± 1.4	55.8 ± 6.8	12.9 ± 0.7	
	post	10.6 ± 1.6 [#]	64.2 ± 7.0 [#]	10.1 ± 1.0 [#]	

Legend: 2x = two session·wk⁻¹; 4x = four session·wk⁻¹; 4x (+Det) = four session·wk⁻¹ + detraining; 4x (+2x) = four session·wk⁻¹ + two session·wk⁻¹; # Significantly different than the corresponding pre value (*p* < 0.05).

Table 2. Mean with 95%CI of individual absolute changes in muscle thickness, fascicle length and pennation angle for RF, VL, VM and VI and percentage of participants exceeding the SWC for regional architecture changes between conditions 2x (*n* = 22), 4x (*n* = 22), 4x (+2x) (*n* = 10) and 4x (+Det) (*n* = 11).

Condition	Proximal			Distal			
	MD [95%CI]	SWC	%↑SWC	MD [95%CI]	SWC	%↑SWC	
MT (mm)							
RF	2x	1.1 [0.2 to 2.0]	0.47	63.6 (14)	1.6 [0.4 to 3.8]	0.39	50.0 (11)
	4x	1.5 [0.3 to 2.7]		68.2 (15)	1.4 [0.1 to 2.7]		40.9 (9)
	4x (+2x)	2.0 [0.4 to 3.6]		80.0 (8)	2.6 [0.7 to 4.4]		70.0 (7)
	4x (+Det)	-2.9 [-5.1 to -0.7]§		0.0 (0)	-1.6 [-4.8 to 1.6]§		9.7 (1)
VL	2x	0.9 [-1.9 to 2.9]	0.39	36.4 (8)	2.6 [1.2 to 4.0]	0.47	68.2 (15)
	4x	0.8 [-2.1 to 3.7]		31.8 (7)	1.3 [0.4 to 1.9]		54.5 (12)
	4x (+2x)	-1.2 [-4.0 to 1.6]		70.0 (7)	2.1 [0.5 to 3.7]		60.0 (6)
	4x (+Det)	0.1 [-2.2 to 2.4]		18.2 (2)	0.3 [-3.5 to 4.1]		36.4 (4)
VM	2x	-2.8 [-4.7 to -0.9]	0.44	63.6 (14)	-2.7 [-0.7 to -4.7]	0.64	54.5 (12)
	4x	-4.2 [-6.6 to -1.8]		77.3 (17)	-2.5 [-0.2 to -4.8]		50.0 (11)
	4x (+2x)	-0.3 [-2.7 to 2.1]		30.0 (3)	1.2 [-2.2 to 4.2]		40.0 (4)
	4x (+Det)	-0.9 [-2.4 to 0.6]		45.5 (5)	-0.7 [-3.6 to 2.2]		36.4 (4)
VI	2x	0.2 [-1.8 to 2.2]	0.42	45.5 (10)	0.1 [-1.1 to 1.3]	L:0.55 M:0.33 X̄: 0.44	29.5 (7)
	4x	1.3 [-1.6 to 4.2]		50.0 (11)	0.6 [-1.9 to 3.1]		34.1 (8)
	4x (+2x)	-1.3 [-2.7 to 0.1]		80.0 (8)	0.7 [-3.7 to 4.9]		60.0 (6)
	4x (+Det)	0.9 [-3.5 to 5.1]§		27.3 (3)	-0.5 [-3.9 to 3.1]		36.4 (4)

FL (mm)							
RF	2x	5.8 [-3.7 to 9.5]		59.1 (13)	19.2 [2.2 to 36.0]		50.0 (11)
	4x	-1.2 [-4.3 to 3.1]	3.41	36.4 (8)	10.1 [1.4 to 18.8]	4.60	40.9 (9)
	4x (+2x)	15.9 [-6.9 to 22.8]		80.0 (8)	39.5 [18.7 to 60.3]		30.0 (3)
	4x (+Det)	6.0 [-12.5 to 18.5]		63.6 (7)	-18.6 [-31.5 to -9.4]§		81.8 (9)
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VL	2x	6.2 [-6.3 to 18.7]		45.5 (10)	1.5 [-11.0 to 14.0]		54.5 (12)
	4x	2.6 [-6.6 to 11.8]	2.80	45.5 (10)	6.3 [-10.4 to 23.0]	2.27	59.1 (13)
	4x (+2x)	11.5 [-4.2 to 27.2]		60.0 (6)	-11.5 [-28.5 to 5.5]		40.0 (4)
	4x (+Det)	-0.3 [-9.2 to 8.5]		63.6 (7)	-11.5 [-19.7 to -3.3]		9.7 (1)
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VM	2x	-18.1 [-26.3 to -9.9]		31.8 (7)	-27.3 [-41.1 to -13.5]		18.2 (4)
	4x	-16.1 [-25.3 to -6.9]	6.15	18.2 (4)	-14.9 [-27.9 to -1.9]	2.44	22.7 (5)
	4x (+2x)	-13.5 [-27.3 to 0.3]		40.0 (4)	-3.9 [-9.1 to 1.2]		40.0 (4)
	4x (+Det)	-1.5 [-5.5 to 3.5]		9.1 (1)	-0.4 [-4.3 to 3.5]		36.4 (4)
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VI	2x	1.1 [-8.6 to 10.8]		59.1 (13)	-10.3 [-17.5 to -3.1]		31.8 (7)
	4x	-1.8 [-10.4 to 12.2]	3.55	40.9 (9)	-4.7 [-8.7 to -0.7]	L:4.21 M:2.61 X̄:3.41	29.5 (7)
	4x (+2x)	-17.9 [-31.7 to -4.1]		10.0 (1)	-2.9 [-4.8 to -0.9]		50.0 (5)
	4x (+Det)	16.3 [2.5 to 30.1]§		63.6 (7)	6.9 [-12.0 to 18.9]§		54.5 (6)
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PA (°)							
RF	2x	-0.1 [-2.1 to 2.0]		31.8 (7)	-0.7 [-4.1 to 2.4]		40.9 (9)
	4x	1.8 [0.6 to 3.0]#	0.39	54.5 (12)	0.7 [-1.8 to 2.5]	0.39	50.0 (11)
	4x (+2x)	0.9 [0.2 to 1.6]		20.0 (2)	-0.1 [-1.4 to 1.4]		60.0 (6)
	4x (+Det)	-1.0 [-4.2 to 2.2]		36.4 (4)	-1.5 [-2.7 to -0.3]§		18.2 (2)
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VL	2x	-1.2 [-3.0 to 0.6]		50.0 (11)	1.9 [0.6 to 3.2]		50.0 (11)
	4x	-1.1 [-2.9 to 0.6]	0.44	63.6 (14)	1.5 [0.3 to 2.7]	0.36	40.9 (9)
	4x (+2x)	-3.3 [-7.0 to 0.4]		40.0 (4)	1.2 [0.2 to 2.2]		60.0 (6)
	4x (+Det)	-2.8 [-5.1 to -0.5]		9.1 (1)	1.5 [-2.4 to 5.4]		36.4 (4)
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VM	2x	0.2 [-2.4 to 2.2]		36.4 (8)	7.3 [4.3 to 10.1]		86.4 (19)
	4x	-0.5 [-1.9 to 0.9]	0.42	50.0 (11)	5.7 [3.4 to 8.0]	0.44	63.6 (14)
	4x (+2x)	3.0 [-2.1 to 8.1]		30.0 (3)	2.7 [-2.5 to 7.9]		60.0 (6)
	4x (+Det)	-0.3 [-5.6 to 5.0]		63.6 (7)	-3.5 [-6.5 to -0.5]§		18.2 (2)
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VI	2x	-1.0 [-4.3 to 2.3]		36.4 (8)	1.8 [-2.5 to 6.1]		54.5 (12)
	4x	0.7 [-2.2 to 3.5]	0.36	63.6 (14)	1.7 [-2.3 to 5.7]	L:0.44 M:0.36 X̄: 0.40	40.9 (9)
	4x (+2x)	2.7 [-2.2 to 7.5]		40.0 (4)	0.6 [-2.0 to 3.2]		30.0 (3)
	4x (+Det)	-1.9 [-6.2 to 3.4]		54.5 (6)	-0.8 [-4.9 to 3.3]		72.7 (8)
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Legend: MT = muscle thickness; FL = fascicle length; PA = pennation angle; SWC = Smallest worthwhile change; %↑SWC = percentual exceeding SWC; MD [95% CI] = mean difference with 95% confidence interval; 2x = two session·wk⁻¹; 4x = four session·wk⁻¹; 4x (+Det) = four session·wk⁻¹ + detraining; 4x (+2x) = four session·wk⁻¹ + two session·wk⁻¹; # = *p* < 0.05 vs 2x; § = *p* < 0.05 vs 4x (+2x).

DISCUSSION

To the authors' knowledge, this is the first study comparing the effects of two different mesocycles on muscle strength and muscle architecture parameters in resistance-trained subjects: one composed of 16 RT sessions distributed over 8 weeks with a frequency of 2 sessions·wk⁻¹ (2x) versus 16 RT sessions distributed over 4 weeks with a frequency of 4

sessions $\cdot\text{wk}^{-1}$ (4x). Secondly, after a 4x period, we compared the effects of an additional 4 week-period of RRT with a frequency of 2 sessions $\cdot\text{wk}^{-1}$ and a $\sim 50\%$ reduction in RT volume versus a detraining period on muscle strength and muscle architecture parameters. Regarding the first experimental intervention (2x versus 4x), the main finding of this study was that performing 16 training sessions in a more concentrated (4x) or more distributed (2x) fashion promotes a similar increase in morphofunctional outcomes.

It is well established in the literature that RT programs carried out in different frequencies (e.g. 1-6 sessions $\cdot\text{wk}^{-1}$) induce positive effects on muscle strength and hypertrophy outcomes in resistance-trained subjects (11, 23, 36, 42, 45, 52). Therefore, the present study expands on previous findings by providing direct evidence that performing 16 RT sessions in 8 weeks with a frequency of 2 sessions $\cdot\text{wk}^{-1}$ is as efficient as performing the same 16 sessions, but in 4 weeks with a frequency of 4 sessions $\cdot\text{wk}^{-1}$ to promote positive changes on morphofunctional responses. Accordingly, we found similar increases in 1RM_{KE} between 2x and 4x protocols (33% and 26.7%, respectively). Moreover, only 1 and 2 legs presented absolute changes below the SWC for 2x and 4x conditions, respectively. For MT, both 2x and 4x results in similar increases for RF proximal (7.2% and 6.1%) and distal (7.1% and 6.2%), VL distal (11.3% vs 5.3%) and VI distal (medial) (15.4% vs 19.8%). Moreover, the percentage of legs exceeding the SWC for these measurements was 34.1–68.2% and 29.5–68.2% for 2x and 4x, respectively. Taken together, these results suggest a homogeneity in both the mean and individual responses between conditions for 1RM and MT. Probably, the similar results observed for muscle strength and hypertrophy outcomes are due to the equal TLL observed between the 2 experimental conditions (51934 kg vs 49804 kg for 2x and 4x, respectively). Thus, the present findings essentially reflected recent meta-analytic data that reported no significant difference between higher and lower frequency on a volume-equated basis for both muscle strength (18) and hypertrophy (40). In this context, it is important to emphasize that the increments in muscle strength and mass seem to be dependent on TLL of RT (17). In fact, a clear dose–response relationship has been reported between TLL and both muscle strength (35) and hypertrophy (41). Thus, consistent with the meta-analytic data, RT volume appears to be more important than RT frequency for promoting muscular adaptations (40). In other words, when RT volume-load employed is high enough, the morphofunctional adaptations occur, even if the volume-load is distributed in a more concentrated (4x) or distributed (2x) scheme.

Another interesting aspect of the TLL results is that the ΔTLL between 1 to 8th session 9 to 16th session presented increments in both 2x and 4x conditions ($\Delta = 15.4\%$; $\Delta = 16.5\%$, respectively). Thus, these findings provide direct evidence that both RT conditions result in similar capacities to accumulate TLL over the weeks. That is, the capacity to accumulate more TLL over the weeks was not negatively influenced by the greater concentrated RT volume/frequency condition (4x group). Although this study has consistently demonstrated that a higher RT volume/frequency is well tolerated with no detrimental effects, it is plausible that this greater TLL, when achieved through high RT volume/frequency protocol (4x), may culminate in detrimental effects if executed for a longer time frame (more than 4 weeks). However, Schoenfeld et al. (39) reported a greater increase in MT of RF and VL with 45 weekly sets per muscle group versus 9 and 27

weekly sets for 8 weeks in resistance-trained men. This higher RT volume of 45 sets was performed throughout 3 weekly sessions, with 15 sets per muscle group per session being performed. Furthermore, Radaelli et al. (34) reported a greater increase in elbow flexor MT with 30 weekly sets per muscle group versus 6 or 18 sets for 6 months in untrained individuals. Taken together, these previous studies (34, 38) demonstrated that higher volumes are well tolerated even when executed for longer periods and even when performed by untrained subjects. However, other studies have shown that higher volumes do not result in greater morphofunctional adaptations when compared to lower volumes (5, 37) and/or lower volumes/frequencies (7, 19). It is, therefore, evident that further investigation in trained individuals for longer time periods is warranted to better elucidate this topic.

On the other hand, the RT interventions did not result in an increase in the MT of VL proximal, VI proximal and distal (lateral) and a decrease in the MT of the VM proximal and distal. Similarly, changes in the other muscle architecture parameters (FL and PA) showed markedly inter- and intramuscular heterogenic responses. In general, some muscles showed an increase in FL without changes in PA (RF distal in 2x and 4x); a decrease in FL without changes in PA (VM proximal in 2x and 4x); an increase in PA without changes in FL (RF proximal [4x], VL distal [2x and 4x], and VI distal medial [4x]); and an increase in PA with a concomitant decrease in FL (VM distal and VI distal lateral [2x and 4x], and VI distal medial [2x]).

Although inhomogeneous results were observed regarding the parameters and muscles assessed, both RT protocols showed similar results. Moreover, the modality of exercise (multi- vs single-joint), type of muscle actions, range of motion and the velocity of execution, variables that are known to influence the quadriceps architecture outcomes (16, 29, 47), were standardized and held constant in both experimental conditions. Therefore, it is plausible to assume that the inter- and intramuscular differences are due to the mechanical effects of the exercise adopted (knee extension machine) and not due to the training load (volume and frequency) itself.

In fact, the inhomogeneous architecture changes of the quadriceps femoris induced by the knee extension exercise is well established (14, 29, 43, 48). For example, in the present study, the RF was the only muscle that presented an increase in MT proximal and distal regions. In the other muscles that showed an increase in MT, this occurred only in the distal region of them. This is consistent with several studies that demonstrated: (i) the increases in anatomical cross sectional area and MT were more prominent in RF than in the vasti (14, 29, 43, 48); (ii) the knee extension exercise results in greater relative hypertrophy in the quadriceps muscles in the distal region than in the proximal region (14, 29, 43, 48).

These results may be linked to regional differences in muscle activation during the pre-scribed exercise mode. Wakahara et al. (49) suggested regional differences in muscle hypertrophy after RT could be attributable to the region-specific muscle activation during the exercise. It has been observed that muscle activation in the distal region of RF during isokinetic knee extension exercise was higher than that in the proximal region (3). Therefore, although we have no data for regional differences in muscle activation and their associations with regional differences in

muscle hypertrophy, the differences within a muscle in muscle activation during the knee extension training might account for the inter and intra-muscle differences in the training-induced changes of morphological.

In regards to the second experimental intervention of the present study, our results support the hypothesis that RRT performed twice a week, with a reduction of ~50% in RT volume (4x (+2x)) was able to maintain previous RT-induced muscle adaptations. However, the cessation of training (4x (+Det)) led to decreases in morphofunctional responses. The positive morphological and functional adaptations to RT are reversed in any population when training ceases (10, 30, 45). Our findings are consistent with the previous research, which demonstrated that RT cessation beyond 4 weeks results in a significant reduction in morphofunctional adaptations (8, 26, 45). On the other hand, following the 4x (+2x) period, MT and 1RM_{KE} gains were retained. Total training volume of 4x (+2x) was reduced by ~50% when compared with the volume performed during the 4x intervention. Our findings are similar to other studies that reduced the training volume by ~30-60% in the RRT period (8, 20, 45, 46).

To the best of the authors' knowledge, this is the first study that investigated the effect of RRT on strength and MA parameters in resistance-trained subjects. Interestingly, the findings of the present study demonstrate that the RRT period was not only able to maintain the positive morphofunctional adaptations acquired in the 4x period, but also provided additional increases on muscle strength and morphological parameters in resistance-trained subjects. The 4x (+2x) promote increases in the 1RM_{KE}; in the MT of RF proximal and distal, VL distal and VI distal (medial); in the FL of RF distal and in the PA of the RF proximal, VL distal, VI proximal and distal medial. Besides that, the decrease observed in the MT of VM proximal and distal during the 4x intervention did not occur in the 4x (+2x) period.

Another relevant finding is that the Δ TLL between 9 to 16th session, representing the 4x period) and 17 to 24th session presented increments in 4x (+2x) period ($\Delta = 15.5\%$). Thus, these findings provide direct evidence that the RRT condition result in an improvement in the capacity to accumulate TLL over the weeks. These results are especially interesting for tapering strategies aimed at increasing or maintaining muscle strength and hypertrophy. It is, therefore, evident that further investigation in trained individuals for longer time periods is warranted to better elucidate this topic.

Our study provides some practical insights that should be considered. First, as 2x and 4x produced similar changes in the assessed parameters, it is recommended that the utilization of a RT with a more concentrated (4x) or more dispersed (2x) fashion should take into account individual time commitment and preferences, as well the specific needs and time availability for each phase of the periodization program. Second, RRT periods was not only able to maintain the positive morphofunctional adaptations, but also provide additional increases on muscle strength and hypertrophy in resistance-trained subjects. Hence, providing deloading phases aimed to promote recovery, can facilitate a continuous improvement in neuromuscular performance and muscular hypertrophy. This hypothesis requires further investigation.

The present study is not without limitations. The unilateral training model employed in the present study may favor the occurrence of cross-education, which may lead to neurally-induced strength gains in untrained contralateral muscles (25). However, it is plausible to hypothesize that cross-education effects have been minimized in our design due to the following factors: (i) Systematic reviews with a meta-analysis demonstrated an average muscle strength increase of ~10% when undergoing cross-education in untrained subjects (27, 31). The muscle strength gains of the present study are 3.2 times (~30.6%) greater than the gains induced by cross-education, which may rule out cross-education as a factor driving our training-induced adaptations; (ii) the participants of the present study were deemed as resistance-trained individuals, as they had 3.3 ± 1.6 years of resistance training experience. Cross-education is less likely to occur in trained individuals than untrained ones; (iii) the advantages of using a within-subject design outgain those of a between-subject design. Biological variability (between-subject design) has a greater effect on muscle strength and hypertrophy gains than cross-education.; (iv) a within-subject design is very effective in controlling biological variability as between-leg responses are equally affected by biological variability.

Finally, in the present study, the participants were instructed to maintain their typical dietary habits throughout the experimental period but we did not attempt to determine nutritional intake, which may have influenced results between conditions. However, the within subject design would have helped to minimize any potential variations attributed to this variable.

In conclusion, this study provides evidence that performing 16 RT sessions in 8 weeks with a frequency of 2 sessions $\cdot\text{wk}^{-1}$ is as efficient as performing the same 16 sessions, but in 4 weeks with a frequency of 4 sessions $\cdot\text{wk}^{-1}$ to promote positive changes on morphofunctional responses. Additionally, the findings of the present study demonstrate that the RRT period was not only able to maintain the positive morphofunctional adaptations acquired, but also provided additional increases on muscle strength and hypertrophy in resistance-trained subjects.

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