

Effects of order of resistance training exercises on muscle hypertrophy in young adult men

Running head: exercise order and muscle hypertrophy

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Abstract

The purpose of the present study was to analyze the effects of the order of resistance training (RT) exercises on hypertrophy in young adult men. Thirty-six young adult men (21.9 ± 2.5 years, 72.6 ± 12.1 kg, 176.9 ± 7.4 cm, 23.1 ± 3.3 kg/m²) were randomly assigned to one of two training groups that performed a 6-week RT program in either a traditional approach starting with multi-joint exercises (MJ) following to single-joint exercises (SJ) order (MJ-SJ, $n = 19$) or an inverse order (SJ-MJ, $n = 17$). Muscle thickness of the biceps brachii and mid-thigh were assessed by ultrasound. Lean soft tissue (LST) was assessed by dual-energy X-ray absorptiometry. Both groups similarly increased ($P < 0.05$) biceps brachii thickness (MJ-SJ = +14.2%, SJ-MJ = +13.8%). Alternatively, only the MJ-SJ group presented an increase in mid-thigh thickness from pre- to post-training (MJ-SJ = +7.2%, SJ-MJ = +3.9%). Upper limbs LST (MJ-SJ = +5.2%, SJ-MJ = +7.5%) was statistically similar between conditions, and a trend for significance ($P = 0.07$) was found for trunk LST (MJ-SJ = +7.2%, SJ-MJ = +1.7%). Non-significant pre- to post-training changes were observed for lower limb LST (MJ-SJ = +0.7%, SJ-MJ = +1.8%). Our data suggest that both sequences are effective for increasing muscle hypertrophy over a short-term RT period; there may be a potentially beneficial hypertrophic effect for the mid-thigh to performing exercises in a manner that progresses from MJ to SJ exercises.

Keywords

Strength training; training variables; pre-exhaustion; quadriceps; biceps brachii, exercise order.

Introduction

The adaptations induced by resistance training (RT) are dependent on the manipulation of exercise training variables including the magnitude of load, number of sets and repetitions, frequency, rest interval, exercise selection, time under tension, muscle action, velocity of movement, and exercise order (ACSM 2009, Schoenfeld 2010). Regarding exercise order, several studies have indicated that this variable can acutely affect aspects of a RT session (ACSM 2009, Simão et al. 2012) such as total force (Sforzo and Touey 1996), the number of repetitions performed (Spreuwenberg et al. 2006, Simão et al. 2007), neuromuscular activation (Augustsson et al. 2003, Gentil et al. 2007, Brennecke et al. 2009), and hormonal responses (Rønnestad et al. 2011).

Current guidelines recommend performing multiple-joint (MJ) exercises early in a RT session followed thereafter by single-joint (SJ) exercises (ACSM 2009). The rationale for this recommendation is based on the premise that performance of MJ exercises is impaired when the involved muscles are pre-fatigued by prior SJ exercises (ACSM 2009). For example, performing the triceps pushdown would fatigue the triceps brachii, thereby impairing the ability to overload the pectoralis major during subsequent performance of bench press. However, paradoxically, athletes and practitioners usually perform exercises in the opposite order with paired sets (i.e. pre-exhaustion system) to maximize muscular adaptations (Ribeiro et al. 2017, 2018). This approach consists of performing a SJ exercise before a MJ exercise, with both exercises involving a common muscle group. For instance, exercising a muscle that will be an agonist in an ensuing MJ (e.g. for the quadriceps, performing the knee extension before the leg press) (Ribeiro et al. 2018). The theory here is that stimulation of the quadriceps would be maximized during the leg press. Another popular way to execute this system is to perform an isolation exercise that works a muscle that will be an agonist in an ensuing MJ exercise (e.g. for the triceps brachii, performing the triceps pushdown before the bench press) (Ribeiro et al. 2018). The theory behind this approach is that the fatigued synergistic muscles contribute less to the movement, thereby placing greater stress on the agonist group (Augustsson et al. 2003).

A limited number of studies have attempted to investigate the chronic influence of exercise order on markers of upper limb hypertrophy (Simão et al. 2010, Spinetti et al. 2010), with somewhat conflicting results. However, to the authors' knowledge, no previous study has investigated the effect of exercise

order on hypertrophy of the lower limb and trunk lean soft tissue (LST). Therefore, the purpose of the present study was to analyze the effects of two orders of exercises on muscular hypertrophy in young adult men. We hypothesized that both exercise order would be effective to induce increases in muscle mass, and that the order initiating by MJ exercises would elicit greater improvements compared to the order initiated by SJ exercises.

Methods

Participants

Thirty-eight participants were randomly divided into one of two training groups: a group that performed the RT program in a traditional sequence starting with MJ followed by SJ exercises (MJ-SJ), or a group that performed the RT program in a SJ to MJ order (SJ-MJ). All participants completed health history and physical activity questionnaires and were included in the study if they had no reported disease symptoms, no orthopedic injuries. Additionally participants were inactive or moderately active individuals (physical activity less than twice a week) and not been regularly engaged in any RT programme during the previous six months before the beginning of the study. Two subjects in the SJ-MJ group left the program due to personal reasons and were not included in final analyses. The final sample was composed of 36 subjects (MJ-SJ = 19; SJ-MJ = 17). Adherence to the RT program was satisfactory, with all subjects participating in $\geq 85\%$ of the total sessions (Gentil and Bottaro 2013). Measurements were performed at least three days apart from the training sessions, in the week prior to the start of the RT program for pre-training measures, and in the week following the end of the RT program for post-training measures.

Written informed consent was obtained from all subjects after receiving a detailed description of study procedures. This investigation was conducted according to the Declaration of Helsinki and was approved by the local University Ethics Committee.

Muscle thickness

Assessment of mid-thigh thickness measurement involved demarcating a point 15 cm above the upper border of the patella at the mid-thigh while for the biceps brachii thickness a point 12 cm above the ante-

ulnar fossa was demarcated (Bemben 2002). All evaluations were performed with subjects in the supine position, the legs together, and knees and elbows semi-flexed ($\sim 10^\circ$), hands supinated, and shoulders abducted at 15 cm from the body. Both measurements were performed on the right side of the body.

All images were processed using a B-mode ultrasound scanner (Ultra Vision Flip, model BF, VMI industry) with a frequency of 7.5 MHz. A generous amount of water-soluble transmitter gel was applied to each site of interest. The transducer was placed perpendicular to the point of measurement, as described elsewhere (Nogueira et al. 2009). Muscle thickness was identified in the image as the distance between the fat-muscle interface and the bone-muscle interface.

Several procedures were implemented to minimize evaluation error: 1) all evaluations were always performed by same technician; 2) all evaluations were carried out at a minimum interval of 48hrs following the last training session; 3) care was taken to ensure that the pressure exerted on the region evaluated was only the weight of the transducer; 4) subjects were instructed not to perform any type of vigorous physical activity in the 24 hours preceding the examination; and, 5) all evaluations were performed in an controlled-temperature environment (22°C to 26°C) with a relative humidity of 50% to 60%.

For reliability analysis, two measures of biceps brachii and mid-thigh were performed at the beginning of the study. The measurements showed high agreement with the mean of the differences between the two measurements being close to zero (biceps = 0.1 mm and mid-thigh = 0.3 mm). The intraclass correlation coefficient was 0.96 and 0.99 for the biceps and mid-thigh, respectively.

Lean soft tissue

Whole-body dual-energy X-ray absorptiometry (DXA) (Lunar Prodigy, model NRL 41990, GE Lunar, Madison, WI) was used to assess upper limb lean soft tissue (ULLST), lower limb (LLLST), and trunk LST (TLST). Scans were performed with the subjects lying in the supine position along the table's longitudinal centerline axis. Feet were taped together at the toes to immobilize the legs while the hands were maintained in a pronated position within the scanning region. Subjects remained motionless during the entire scanning procedure. Both calibration and analysis were carried out by a laboratory technician. Equipment calibration followed the manufacturer's recommendations. The software generated standard

lines that set apart the limbs from the trunk and head. These lines were adjusted by the same technician using specific anatomical points determined by the manufacturer. Analyses were performed by the same technician who was blinded to group identity throughout the investigation.

Resistance training program

Participants performed a 6-week RT program. Participants from both training groups (MJ-SJ, and SJ-MJ) performed the same whole-body program with 10 exercises. The routine consisted of 3 sets of 8-12 RM carried out 3 times per week on Mondays, Wednesdays, and Fridays, differing only in the order of execution of the exercises. Participants in MJ-SJ performed exercises in the following order: 1) bench press, 2) lat pulldown, 3) upright row, 4) shoulder press, 5) triceps pushdown, 6) arm curl, 7) leg press, 8) knee extension, 9) leg curl, and 10) calf raise; those in SJ-MJ performed exercises as follows: 1) arm curl, 2) triceps pushdown, 3) shoulder press, 4) upright row, 5) lat pulldown, 6) bench press, 7) calf raise, 8) leg curl, 9) knee extension, and 10) leg press. Repetitions were performed until volitional failure, herein defined as an inability to carry out the exercise with proper technique. Participants were instructed to inhale during the eccentric phase and exhale during the concentric phase while maintaining a constant velocity of movement at a ratio of approximately 1:2 seconds (concentric and eccentric phases, respectively). Participants were afforded 1 to 2 min of rest between sets and 2 to 3 min between each exercise. The initial training load was determined using procedures described elsewhere (Ribeiro et al. 2015). Briefly, this involved executing the first and second sets at the lower end of the repetition zone (8 repetitions), and then performing as many repetitions as possible until voluntary exhaustion or the inability to maintain proper technique in the third set. The same weight was used to perform all three sets of each exercise. Each subject initial training load was then determined using the following equations:

$$\text{Upper limb exercises: } FW = WT + RE/2$$

$$\text{Lower limb exercises: } FW = WT + RE$$

Where FW = final weight (kg); WT = weight used in the test (kg); RE = maximum number of repetitions performed that exceeded the lower limit (8 repetitions) in the third set.

Statistical analyses

The independent Student's *t*-test was used to compare typical characteristics at baseline. Two-way mixed-model analysis of variance was employed for intra- and inter-group comparisons. Effect size (ES) was calculated as post-training mean minus pre-training mean divided by the pooled pre-training standard deviation (Cohen 1992), where an ES of 0.00 - 0.19 was considered trivial, 0.20-0.49 was considered as small, 0.50-0.79 as moderate and ≥ 0.80 as large (Cohen 1992). Significance was accepted at $P < 0.05$.

Results

Table 1 presents the typical characteristics of participants at baseline. There was no significant difference ($P > 0.05$) between groups for any variable analyzed.

Insert Table 1

The data for muscle thickness and lean soft tissue according to group are presented in Table 2. No significant ($P > 0.05$) interactions were observed for any variable. Both groups similarly increased ($P < 0.05$) biceps brachii thickness (MJ-SJ = +14.2%, SJ-MJ = +13.8%). Alternatively, only the MJ-SJ group presented an increase in mid-thigh thickness from pre- to post-training (MJ-SJ = +7.2%, SJ-MJ = +3.9%). ULLST (MJ-SJ = +5.2%, SJ-MJ = +7.5%) was statistically similar between conditions, and a trend for significance ($P = 0.07$) was found for TLST (MJ-SJ = +7.2%, SJ-MJ = +1.7%). Non-significant pre- to post-training changes were observed for LLLST (MJ-SJ = +0.7%, SJ-MJ = +1.8%).

Table 3 presents the ES values of the training groups as well as the differences between them. A difference of a small magnitude was observed for TLST favoring MJ-SJ vs. SJ-MJ; a trivial difference was observed for the remaining outcomes.

Insert Table 2 and Table 3

Discussion

Our main finding was that statistically similar RT-induced increases in muscle mass are seen regardless of the order of exercise progression. However, despite the lack of differences between conditions from a probability standpoint, the percentage change in mid-thigh hypertrophy was markedly larger in MJ-SJ compared to SJ-MJ (MJ-SJ = +7.2%, SJ-MJ = +3.9%) and only the MJ-SJ condition showed significant pre- to post-study increases for this variable. This indicates a potentially beneficial effect to perform

exercises in a manner that progresses from MJ to SJ movements when the fitness goal is to maximize lower limb hypertrophy. To our knowledge, this is the first study to investigate the effects of different orders of exercise of lower limb hypertrophy; therefore, no direct comparisons can be made with the current literature. However, our findings appear to be supported by Augustsson et al. (2003) who investigated electromyographic activity during 1 set of the leg press with and without performing 1 set of knee extension prior to the leg press. Results indicated that performing the knee extension before the leg press impaired the performance and the mid-thigh activation during the leg press. It is important to mention that although an association exists between muscular activation and muscle growth (Wakahara et al. 2012), hypertrophy is a complex phenomenon (Schoenfeld 2010) and electromyographic signals may be influenced by multiple factors (Vigotsky et al. 2018). Whether the observed differences in mid-thigh hypertrophy in our study were related to alterations in muscle activation remains to be determined. Moreover, it should be noted that the training order of body segments was not changed; thus, it remains undetermined if results would be different in a session starting with exercise for the lower limbs. Future studies alternating the order of the exercises for upper and lower body involving MJ and SJ are needed to address this topic.

It is possible that results may be related to the different stimuli theorized to induce muscle hypertrophy including mechanical tension, muscle damage and metabolic stress (Schoenfeld 2013). Based on these stimuli, the physiological advantage for the leg press theoretically would be when it is positioned earlier in a lower limb routine. For example, peak tension in quadriceps during leg press occurs when the muscle is stretched (Escamilla et al. 2001) favoring greater muscle damage, and the muscle damage is even greater with higher loads; therefore, when this exercise is performed first, a greater absolute load can be lifted resulting in greater muscle damage. On the other hand, peak of tension in quadriceps during knee extension occurs when the muscle is shortened (Wilk et al. 1996), thus favoring greater metabolite accumulation; hence, performing the knee extension after the leg press will conceivably result in heightened metabolic stress. Therefore, MJ-SJ order might have been favorable for lower limb hypertrophy compared to SJ-MJ by optimizing mechanical tension/muscle damage and metabolic stress in the MJ and SJ exercises, respectively. Notably, this rationale only applies to training for the lower body in the current study; it would be applicable to upper body RT only if the exercises had

involved a common target muscle (e.g. chest press and pec fly), which is not the case. How these factors ultimately drive hypertrophy requires further study.

To our knowledge, this study is also the first to investigate the effects of exercise order on the increase of the TLST. From a probability standpoint, results showed that exercise order produced no significant ($P = 0.07$) differences in TLST. Considering that some researchers have proposed that null hypothesis statistical testing should not be used as the sole method to draw practical inferences (Gelman and Stern 2006, Hopkins et al. 2009, Bernards et al. 2017), it should be noted that ES analysis indicates a small but potentially practically meaningful difference between training protocols (relative ES = -0.35) favoring the sequence beginning with MJ exercises. The novelty of this analysis precludes a direct comparison with the literature, however previous studies investigating the acute effects of exercise order have shown no change in trunk muscle activity patterns (Gentil et al. 2007, Brennecke et al. 2009, Soares et al. 2016). For example, Soares et al. (2016) investigated the neuromuscular activity of pectoralis major and triceps brachii during the triceps pushdown and bench press exercises in two sequences: a SJ-MJ whereby the triceps pushdown was followed by the bench press and a MJ-SJ sequence whereby the bench press preceded the pushdown. No significant difference was noted between conditions for both the pectoralis major and triceps brachii.

Interestingly, while investigations fail to show an effect of exercise order on the pectoralis major during the bench press, there was a reported increase in activity of the synergist triceps brachii (Gentil et al. 2007, Brennecke et al. 2009), indicating a possible benefit of the SJ-MJ order for the elbow extensors. Our results do not support such a benefit, as biceps thickness and ULLST were unaffected by exercise order. This finding agrees with those of Spinetti et al. (2010) and Simão et al. (2010) who also found exercise order had no effect on triceps brachii hypertrophy. Collectively, these results suggest that once a given threshold is reached for neuromuscular activity, any further increases would have no further effects on muscle hypertrophy, making alterations in exercise order superfluous from this standpoint in upper-body hypertrophy in untrained men. In line with this suggestion, studies reported that muscle mass gains in the leg muscles required a higher volume threshold, while no difference were reported between low and high volume thresholds for the upper-body muscles in untrained men (Rønnestad et al. 2007, Bottaro et al. 2011). However, the study by Simão et al. (2010) did in fact show a benefit to SJ-MJ order

on biceps brachii hypertrophy, whereby only the groups that performed arm curl before lat-pulldown realized a significant increase in elbow flexor thickness from pre- to post-training. These conflicting findings are difficult to reconcile and warrant further study.

Despite the observed increase in mid-thigh thickness across conditions, the LLLST did not change from pre- to post-training. It is possible that discrepancies in findings can be explained, at least in part, by the use of DXA as proxy measurement of muscle mass. Although DXA is well-established as a valid modality for estimating body composition, it may lack the sensitivity to detect subtle changes in muscle tissue over relatively short time frames compared to ultrasound. In support of this hypothesis, Scanlon et al. (2014) observed a significant increase in vastus lateralis measured by ultrasound, but no changes in thigh lean tissue measured by DXA after 6 weeks of RT. Similarly, Snijders et al. (2015) found significant differences in changes in thigh muscle cross-sectional area between conditions in a RT protocol as measured by computerized tomography, but measures of leg lean mass as determined by DXA failed to reach statistical significance (Snijders et al. 2015). Discrepancies in findings between modalities may have been magnified by the possibility that RT-induced hypertrophy tends to be smaller in the lower limbs compared to both upper limb and trunk muscles (Counts et al. 2017), as well as the observation that a greater volume is necessary to achieve optimal improvements in the lower versus upper limbs (Paulsen et al. 2003, Bottaro et al. 2011, Radaelli et al. 2014).

The present study had some limitations of note. For one, the study period of 6 weeks can be considered short; it remains to be determined whether results would differ over a longer timeframe. Moreover, the study is specific to untrained young adult men and results cannot be generalized to other populations including children, older adults, women, and those with RT experience. Finally, although subjects were instructed to maintain their usual nutritional habits and abstain from any additional structured exercise, dietary intake and physical activity were not monitored during the experiment, hindering our ability to determine whether changes in nutrition or habitual physical activity occurred across the study period may have affected results.

In conclusion, our data suggest that both sequences are effective for inducing muscle hypertrophy over the course of a short-term RT period in young adult men. Results indicate the possibility of a

potentially beneficial hypertrophic effect for performing exercises in a manner that progresses from MJ to SJ movements.

Conflict of Interest

The authors have no conflicts of interest.

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Table 1. Characteristics of the participants at baseline. Data are presented as mean and standard deviation.

| | MJ-SJ (n = 19) | SJ-MJ (n = 17) | <i>P</i> -value |
|--------------------------------------|----------------|----------------|-----------------|
| Age (years) | 21.3 ± 2.8 | 22.2 ± 2.3 | 0.46 |
| Body mass (kg) | 72.3 ± 15.1 | 72.9 ± 8.4 | 0.87 |
| Height (cm) | 177.9 ± 7.3 | 175.9 ± 7.5 | 0.42 |
| Body mass index (kg/m ²) | 22.7 ± 3.8 | 23.6 ± 2.7 | 0.41 |

Note: MJ-SJ = multi-joint to single-joint exercises order. SJ-MJ = single-joint to multi-joint exercises order.

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Table 2. Muscle thickness and lean soft tissue according to groups at pre and post training (6 weeks). Data are presented as mean and standard deviation.

| | MJ-SJ (n = 19) | | | SJ-MJ (n = 17) | | | <i>P</i> -value | | |
|---------------------|----------------|------------|------------------------|----------------|------------|------------------------|-----------------|-------|-------------|
| | Pre | Post | Change (95% CI) | Pre | Post | Change (95% CI) | Time | Group | Interaction |
| Biceps brachii (mm) | 26.7 ± 5.7 | 30.5 ± 5.4 | 3.76 ± 2.9 (2.3, 5.1) | 26.8 ± 4.4 | 30.5 ± 4.9 | 3.64 ± 3.0 (2.0, 5.2) | <0.001 | 0.96 | 0.90 |
| Mid-thigh (mm) | 34.8 ± 7.4 | 37.3 ± 6.7 | 2.50 ± 2.6 (1.2, 3.7) | 35.7 ± 8.1 | 37.1 ± 6.6 | 1.40 ± 3.2 (-0.2, 3.1) | <0.001 | 0.88 | 0.29 |
| ULLST (kg) | 7.0 ± 1.5 | 7.4 ± 1.2 | 0.35 ± 1.7 (-0.5, 1.2) | 6.9 ± 0.9 | 7.5 ± 0.9 | 0.52 ± 0.2 (0.3, 0.6) | 0.03 | 0.80 | 0.76 |
| LLLST (kg) | 20.0 ± 3.1 | 20.1 ± 2.2 | 0.14 ± 2.8 (-1.2, 1.5) | 19.8 ± 1.9 | 20.2 ± 2.4 | 0.35 ± 1.0 (-0.1, 0.8) | 0.39 | 0.99 | 0.92 |
| TLST (kg) | 26.4 ± 6.4 | 28.3 ± 7.3 | 1.88 ± 5.4 (-0.7, 4.5) | 26.0 ± 1.8 | 26.4 ± 1.6 | 0.45 ± 1.1 (-0.1, 1.0) | 0.07 | 0.69 | 0.28 |

Note: MJ-SJ = multi-joint to single-joint exercises order. SJ-MJ = single-joint to multi-joint exercises order. ULLST = upper limb lean soft tissue. LLLST = lower limb lean soft tissue. TLST = trunk lean soft tissue.

Table 3. Effect sizes values according to groups.

| | MJ-SJ (n = 19) | SJ-MJ (n = 17) | Difference | Classification |
|-----------|----------------|----------------|------------|----------------|
| Biceps | 0.75 | 0.73 | -0.02 | Trivial |
| Mid-thigh | 0.32 | 0.18 | -0.14 | Trivial |
| ULLST | 0.30 | 0.43 | 0.13 | Trivial |
| LLLST | 0.06 | 0.14 | 0.08 | Trivial |
| TLST | 0.46 | 0.11 | -0.35 | Small |

Note: MJ-SJ = multi-joint to single-joint exercises order. SJ-MJ = single-joint to multi-joint exercises order. ULLST = upper limb lean soft tissue. LLLST = lower limb lean soft tissue. TLST = trunk lean soft tissue. Differences = SJ-MJ effect size minus MJ-SJ effect size. Effect size classification = 0.00 - 0.19 trivial, 0.20 - 0.49 small, 0.50 - 0.79 moderate and ≥ 0.80 large (Cohen 1992).

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