

Research article

Effects of the Drop-set and Reverse Drop-set Methods on the Muscle Activity and Intramuscular Oxygenation of the Triceps Brachii among Trained and Untrained Individuals

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Abstract

Influence of different load exercise to muscle activity during subsequent exercise with 75% of one repetition maximum (RM) load among trained and untrained individuals was verified. Resistance-trained men who were involved in resistance training ($n = 16$) and healthy young men who did not exercise regularly ($n = 16$) were recruited for this study. Each subject performed bench pressing with a narrow grip exercise using two different training set methods, the drop-set (DS) (3 sets \times 2-10 repetitions with 95-75% of 1RM) and the reverse drop-set (RDS) (3 sets \times 3-10 repetitions with 55-75% of 1RM). The mean concentric contraction power, root mean square (RMS) of electromyography (EMG), area under the oxygenated hemoglobin (Oxy-Hb) curve, and time constant for muscle oxygen consumption ($TcVO_{2mus}$) values of the triceps brachii were measured during and after the DS and RDS. The trained group demonstrated significantly higher mean muscle power (242.9 ± 39.6 W vs. 215.8 ± 31.7 W), RMS of EMG (86.4 ± 10.4 % vs. 68.3 ± 9.6 %), and area under the Oxy-Hb curve (38.6 ± 7.4 % \cdot sec vs. 29.3 ± 5.8 % \cdot sec) values during the DS than during the RDS ($p < 0.05$). However, in the untrained group none of the parameters differed significantly for both the DS and RDS. Furthermore, a negative correlation was detected between the area under the Oxy-Hb curve and muscle thickness ($r = -0.51$, $p < 0.01$). Long-term effects of DS on muscle strengthening and hypertrophy will be explored in further research.

Key words: Drop-set, resistance exercise, hypertrophy, hypoxia, NIRS.

Introduction

A previous study detected local increases in arterial diameter and blood flow after eight weeks of resistance exercise (Okamoto et al., 2009), resulting in blunted hypoxic muscle stimulation. Van Wessel et al. (2010) reported that the increase in muscle oxidative capacity produced by resistance training has a negative impact on muscle hypertrophy. Furthermore, Adams et al. (1993) reported that motor unit recruitment is reduced in people who do not regularly perform resistance training. In order to ensure successful muscular strengthening and hypertrophy, it is necessary to assess how much mechanical and metabolic stress was induced to the muscle by the exercise protocol.

The drop-set method (DS) and the reverse drop-set method (RDS) are exercise methods, which athletes often use for increasing muscle strength and hypertrophy. The DS is an exercise protocol, in which resistance exercise is initially performed with a higher load and then gradually decreased. On the contrary, the RDS is an exercise method in which the load used is gradually increased. The DS is based on the physiological phenomenon that high-threshold motor units can be activated more easily once they have been recruited (Gorassini et al., 2002). Strong muscle contraction during the DS leads to mechanical capillary compression, resulting in restricted blood flow to muscles and the induction of acute intramuscular hypoxia. It is assumed that marked muscle hypertrophy is induced when muscles are simultaneously subjected to metabolic and mechanical stimuli (Schoenfeld, 2013). However, no previous study has compared the effects of the DS and RDS on intramuscular hypoxia and motor unit recruitment among trained and untrained individuals.

Therefore, the aim of this study was to verify the influence of different load exercise to the muscle activity during subsequent exercise with 75% of one repetition maximum (RM) load among trained and untrained individuals. The triceps brachii muscle, which plays a major role in the bench press exercise, was used in the study. The bench press is essential for many athletes looking to increase upper body strength (Dunnick et al., 2015). For the assessment of the effects of exercise training protocols, non-invasive and practical evaluation methods such as near-infrared spectroscopy (NIRS) and surface electromyography (EMG) are useful alternatives to muscle biopsy and magnetic resonance spectroscopy (Tanimoto et al. 2006). We hypothesized that the DS might be more effective at activating high-threshold motor units and inducing intramuscular acute hypoxia.

Methods

Subjects

Sixteen resistance-trained men who were involved in a resistance training [trained group; mean age: 21.9 ± 2.6 years; mean height: 1.73 ± 0.05 m; mean body weight: 68.2 ± 9.1 kg; mean 1RM during bench pressing with a narrow grip (BPN): 61.5 ± 14.8 kg; mean triceps brachii

thickness: 4.7 ± 5.1 cm] and 16 healthy young men who did not exercise regularly (untrained group; mean age: 22.7 ± 2.9 years; mean height: 1.74 ± 0.04 m; mean body weight: 62.6 ± 8.3 kg; mean 1RM during BPN: 35.4 ± 7.5 kg; mean triceps brachii thickness: 3.6 ± 0.9 cm) were recruited from among the students at Aino University. The inclusion criteria for the trained group consisted of at least 1 year's experience of resistance training, participating in a resistance training program at least 3 days a week, and performing triceps brachii exercises at least once a week. Subjects who reported any musculoskeletal injuries of the upper extremities in the year before the test were excluded. All subjects were instructed to refrain from vigorous physical activity within 12 hours of each session (Maehlum et al. 1986). Before participating in the study, the subjects were informed about the study procedures and any possible risks both verbally and in writing before signing informed consent forms. A priori data and a power analysis were used to detect the sample size. A minimum sample of 14 subjects was calculated based on detecting a difference of concentric muscle power, area under the Oxy-Hb curve, and RMS of EMG with 80% power and 5% significance. The sample size was calculated with the G Power software (version 3.1.4). The ethics committee of Aino University approved the study, which was conducted according to the most recent declaration of Helsinki.

Exercise protocols of the DS and RDS

All 1RM and BPN testing were performed using a press bench and a standard 20-kg Olympic barbell. Each subject lay with their back on the press bench and both feet on the floor. An electrogoniometer (DTS2D goniometer; Noraxon, Arizona, USA) was used to prevent compensatory horizontal abduction in the shoulder joint during the DS and RDS. The electrogoniometer was attached to the radial side of the right forearm and the lateral side of the upper right arm. They were asked to place their upper arms so that they were perpendicular to their body, flex their elbow joints to 90 degrees, and grasp the barbell, which was held in a fixed stand. They lifted the barbell from this starting position to full extension and then re-

turned to the starting position. This triceps brachii concentric/eccentric contraction cycle was performed at a metronome-controlled tempo of one second per concentric contraction and one second per eccentric contraction. The subjects were instructed to perform the concentric phase of each repetition as fast as they could by pushing the barbell to complete extension as rapidly and explosively as possible. More than one week later after 1RM testing, the subjects performed BPN exercises using two different training set methods, the DS and RDS. The DS and RDS exercise were separated by intervals of at least 1 week. The order of the DS and RDS exercises was randomized for each subject. The DS and RDS protocols are shown in Figure 1. In order to achieve identical higher-volume workloads during both training set methods, load and the number of sets were determined (Schoenfeld 2010).

Triceps brachii concentric contraction power measurements

In both the DS and RDS, the peak power of each repetition during the concentric phase of 75% 1RM load exercise was assessed with a FiTROdyne Powerlyzer (Fitrodyne; Fitronic, Bratislava, Slovakia), which was attached to the barbell using a tether. The FiTROdyne unit uses the tether displacement time and manually entered load data to calculate power values. Jennings et al. demonstrated that this procedure exhibited high test-retest reliability (intraclass correlation coefficient; ICC: 0.86) when it was used to assess muscular power during a multiple-joint exercise (Jennings et al., 2005). The first of the 10 repetitions was different from other nine in that the barbell was lifted from the fixed stand during this repetition, so the data for the first repetition were excluded. The mean peak power was calculated from the remaining 9 repetitions. On two separate days, five untrained subjects performed 10 bench press repetitions with narrow grip using 75% of 1RM load. The repeatability of the mean peak power measurements was assessed in a pilot study. The test-retest ICC for the mean peak torque of the triceps brachii muscle was 0.89.

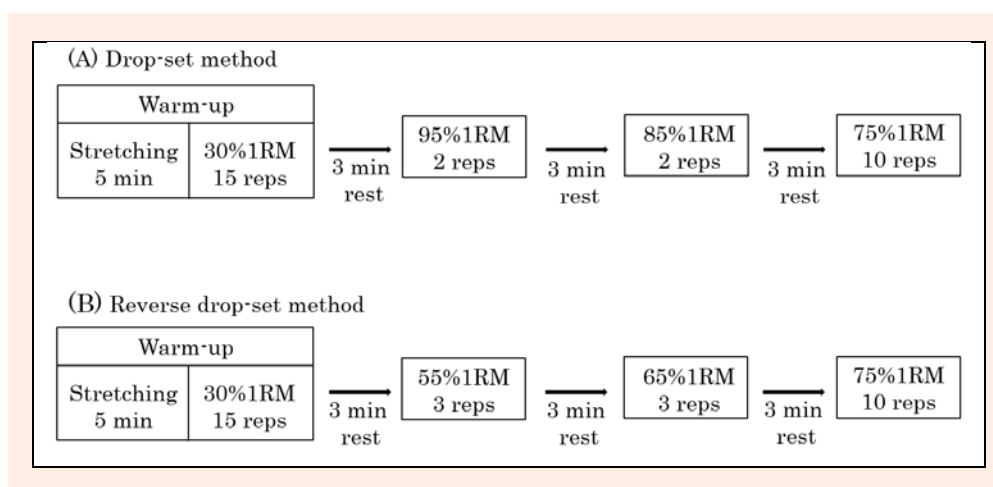


Figure 1. Exercise protocols of the (A) drop-set and (B) reverse drop-set methods. Both exercise protocols consist of same exercise volume (A: $95\% \times 2 \text{ reps} + 85\% \times 2 \text{ reps} + 75\% \times 10 \text{ reps} = 6250$, B: $55\% \times 3 \text{ reps} + 65\% \times 3 \text{ reps} + 75\% \times 10 \text{ reps} = 6250$).

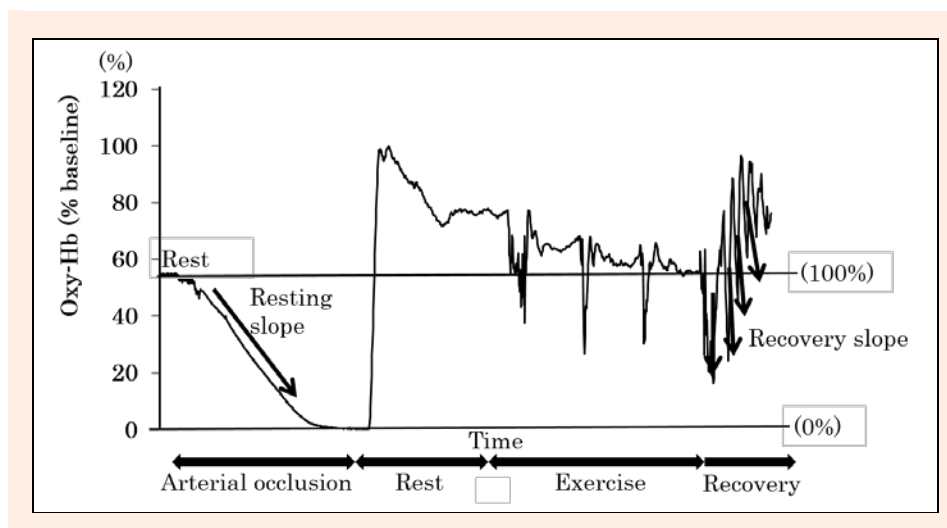


Figure 2. An example of the changes in Oxy-Hb levels detected by NIRS. The Oxy-Hb level recorded at rest was defined as 100%, and the minimum Oxy-Hb plateau level recorded after 6 minutes' arterial occlusion was defined as 0%. Furthermore, repeated arterial occlusion was performed after the exercise, and the time it took for the oxygen consumption rate to return to 63% of its resting value was calculated.

Peripheral muscle oxygenation measurements

A near-infrared continuous-wave spectrometer (HB14-2; ASTEM Co., Ltd., Kanagawa, Japan) was used to measure peripheral muscle oxygenation, the area under the Oxy-Hb curve, and the recovery time constant for muscle oxygen consumption ($TcVO_{2mus}$) (Hamaoka et al., 2001) in the right triceps brachii muscle during and after each exercise. Figure 2 shows a typical example of the Oxy-Hb dynamics detected in the right triceps brachii muscle during exercise and repeated arterial occlusion. The wavelength of the emitted light ranged between 750~850 nm, and the relative concentration of Oxy-Hb in the target tissue was quantified according to the Beer-Lambert law (Chance et al. 1992). The distance between the incident point and the detector was 30 mm. The laser emitter and detector were fixed in place with sticking tape. The NIRS signals were stored in a personal computer.

The NIRS signals recorded during exercise do not always reflect the absolute levels of intramuscular oxygenation, so the changes in the oxygenation of working skeletal muscles are expressed relative to the overall changes in the monitored signal according to the arterial occlusion method (Hamaoka et al., 2001). In this study, the Oxy-Hb level observed at rest was defined as 100%, and the minimum Oxy-Hb plateau level induced by arterial occlusion was defined as 0%. On the other side, a pressure cuff was placed around the proximal portion of the upper arm and was manually inflated to 250 mmHg until the minimum plateau level of Oxy-Hb was obtained (Bae et al., 2000). The area under the Oxy-Hb curve was used to examine the reduction in the intramuscular oxygen level induced over three sets, as described by Manfredini et al. (Manfredini et al., 2015). Figure 2 shows an example of the Oxy-Hb changing of relative oxygenation level.

$TcVO_{2mus}$ was obtained via repetitive brief arterial occlusion after the completion of each exercise. The arterial blood flow occlusion was terminated soon after the Oxy-Hb concentration reached an almost constant level. A previous study showed that the Oxy-Hb values recorded during arterial occlusion can be used as a direct index

of VO_{2mus} (Hamaoka et al., 1996). All VO_{2mus} data are shown as percentages of the resting value. $TcVO_{2mus}$ was calculated from the obtained VO_{2mus} data according to the formula shown below:

$$y = A(1 - e^{-kt})$$

In this formula, y represents the relative value of VO_{2mus} during arterial occlusion in the rest period following the exercise, A represents the total change in VO_{2mus} between the value seen at the end of the exercise and the value recorded after the subject has recovered, k is a rate constant ($1/k = Tc$), and t is time. This formula was used to express the time needed for the oxygen consumption rate to return to 63.2% of its resting value.

Electromyographic signal recording measurement

The muscle activity of the long head of the triceps brachii was recorded at a sample rate of 1000 Hz using an electromyographic (EMG) system (Myosystem 1200, Noraxon U.S.A. Inc., AZ, USA). Bipolar surface EMG electrodes (model: M-150Ag/AgCl, Nihon Kohden Inc., Tokyo, Japan) were used to measure EMG signals from the long head of the triceps brachii during the exercises. Based on the Surface Electromyography for the Non-Invasive Assessment of Muscles (SENIAM) recommendations (Hermens et al., 2000), pairs of EMG electrodes were placed along the muscle midline. The bipolar surface EMG electrodes were placed in line with the muscle fibers. The center-to-center distance between each pair of electrodes was 2.5 cm. Prior to the lead placement, the patient's skin was shaved, wiped using skin preparation gel (Nihon Kohden Inc., Tokyo, Japan), and cleaned with alcohol wipes. A reference electrode was placed over the acromioclavicular joint. All of the recorded inter-electrode resistance values were below 10 k Ω . Myoelectric signals were relayed from the bipolar electrodes to a TeleMyo device (TeleMyo 2400T, Noraxon U.S.A. Inc., AZ, USA). The raw EMG signals were amplified and filtered 20-500Hz using commercially available software (MyoResearch XP, Noraxon U.S.A. Inc., AZ, U.S.A.).

EMG amplitude (root-mean square: RMS) was measured from EMG signals: (1) during maximal voluntary contraction (MVC) measurements, RMS was calculated based on a 500 ms time window centered on the highest force value, (2) during the DS and RDS protocols, RMS was calculated for each repetition based on a 500 ms time window centered on the highest value. All RMS measurements were normalized to pre-exercise MVC.

Maximal voluntary contraction

MVC was determined for right elbow extension. During baseline measurements, a torque-angle curve was constructed on an individual basis to determine the optimal elbow joint angle to be used in all subsequent measurements. MVC determination implicated 3 isometric contraction trials, with at least 3 seconds of durations, each separated by 1 minute of recovery. Participants were instructed to exert their maximum force as fast as possible.

Muscle thickness measurements

Ultrasound imaging was used to obtain muscle thickness measurements. Compared with the gold standard measurement method; i.e., magnetic resonance imaging, the reliability and validity of ultrasound for determining muscle thickness has been reported to be very high (Reeves et al., 2004). A trained technician performed all of the tests using an ultrasound imaging unit (Noblus; Hitachi Medical Inc., Tokyo, Japan). Water-soluble transmission gel was applied to each measurement site, and a 2.5 MHz ultrasound probe was placed perpendicular to the tissue interface without depressing the skin. The images were saved to a hard drive. Muscle thickness dimensions were obtained by measuring the distance from the subcutaneous adipose tissue-muscle interface to the muscle-bone interface (Abe et al., 2000). Measurements were obtained at the muscle belly of the triceps brachii. On two separate days, a trained technician performed triceps brachii muscle thickness measurements of 5 untrained subjects to assess the repeatability of the ultrasound measurements in a pilot study. The test-retest ICC for the triceps brachii muscle was 0.91.

Statistical analysis

All data are expressed as means \pm standard deviation (SD) values. All statistical analyses were performed using SPSS for Windows version 21.0 (SPSS Statistics 21.0; IBM, Tokyo, Japan). The test-retest reliability of the peak power and muscle thickness measurements was evaluated using ICC. All tests and measurements were found to be reliable (their ICC ranged from 0.81 to 0.89, and no significant differences were detected between the mean test-retest values). A 2-way [training experience (more than one year vs. none) \times training protocol (DS vs. RDS)] mixed-measures analysis of variance (ANOVA) with the Greenhouse-Geisser correction was used to analyze the differences in mean peak power, the area under the Oxy-Hb curve, $TcVO_{2mus}$, and RMS of EMG during 75% of 1RM exercise. To analyze the differences in RMS of EMG within same training experience groups, a 2-way [exercise load (55% - 75% of 1RM or 95% - 75% of

1RM) \times training protocol (DS vs. RDS)] repeated-measures ANOVA was used. When statistically significant differences were detected, Bonferroni pairwise comparisons were performed. Pearson's correlation coefficients were calculated for the relationships between the muscle oxygenation level and muscle thickness, and between the muscle oxygenation level and muscle power during the DS. An alpha level of 0.05 was used to determine statistical significance.

Results

Mean muscle power of triceps brachii concentric contraction at a load of 75% of 1RM

In the trained group, the mean muscle power generated during concentric contractions of the triceps brachii at a load of 75% of the subject's 1RM was significantly higher during the DS than during the RDS (Figure 3). Furthermore, the mean muscle power of the trained group was significantly higher than that of the untrained group.

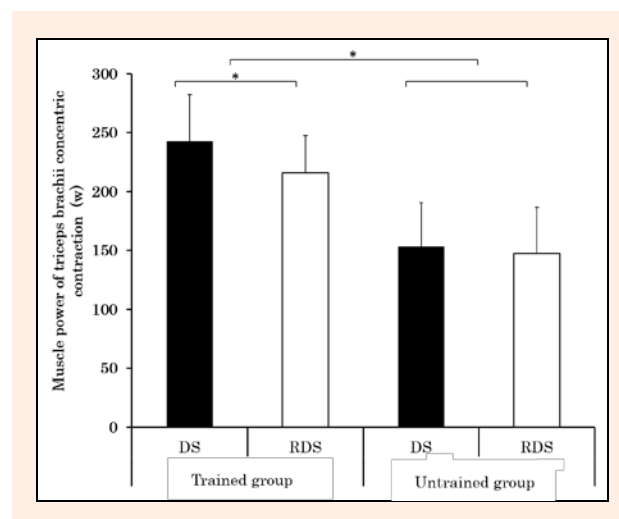


Figure 3. Mean muscle power of triceps brachii muscle during concentric contractions using a load of 75% of the subject's 1RM in the trained and untrained groups during the drop-set (DS) and reverse drop-set (RDS) methods. Means \pm SD (n = 16 for each group) values are shown. * Significant difference (p < 0.05)

RMS of EMG recorded during the DS and RDS

Figure 4 shows the RMS values recorded in the triceps brachii EMG during the exercises. In the trained group, significantly higher RMS of EMG values during 75% of 1RM load exercise were recorded during the DS than during the RDS. However, there was no significant difference in RMS of EMG value during 75% of 1RM load exercise between DS and RDS in the untrained group.

Area under the Oxy-Hb curve during the DS and RDS

Figure 5 shows a typical example of the area under the Oxy-Hb curve. In the trained group, the mean area under the Oxy-Hb curve value for the DS was significantly higher than that for the RDS. Furthermore, the mean area under the Oxy-Hb curve value of the trained group was significantly lower than that of untrained group (Figure 6). Furthermore, a negative correlation was detected be-

tween the area under the Oxy-Hb curve and muscle thickness among the whole study population ($r = -0.51$, $p < 0.01$, $n = 32$) (Figure 7).

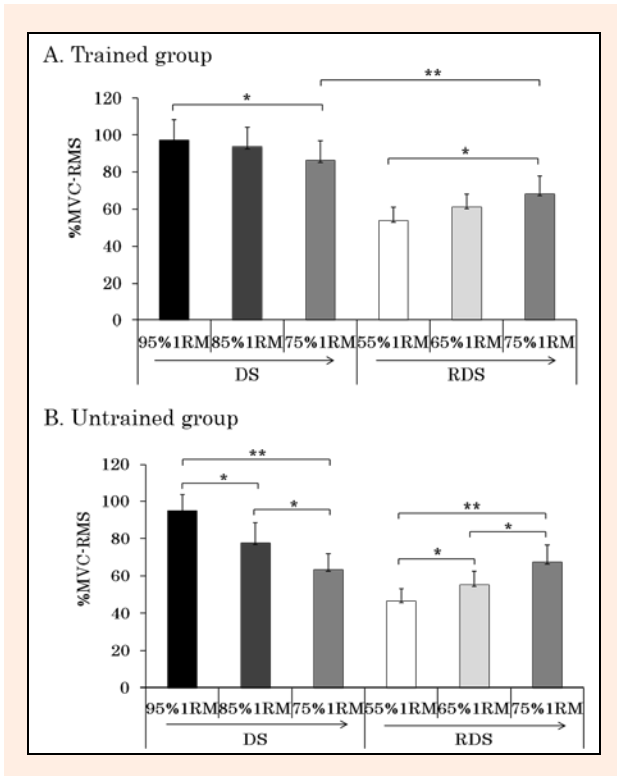


Figure 4. %Maximal voluntary contraction (MVC)-Root mean square (RMS) values of the trained (A) and untrained (B) groups during the drop-set method (DS) and reverse drop-set (RDS) methods. Mean \pm SD ($n = 16$ for each group) values are shown. *Significant difference ($p < 0.05$). **Significant difference ($p < 0.01$)

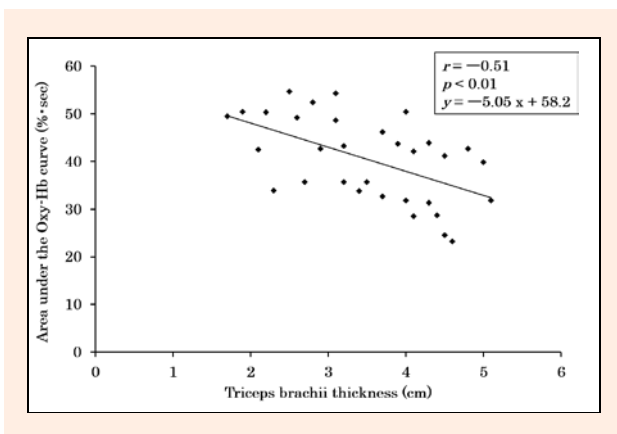


Figure 7. Correlation between the area under the Oxy-Hb curve during the DS and triceps brachii muscle thickness among the 32 subjects

TcVO_{2mus} after the DS or RDS

The mean TcVO_{2mus} values for the DS and RDS did not differ significantly, but the mean TcVO_{2mus} of the trained group was significantly faster than that of untrained group. In the trained group, mean TcVO_{2mus} values of 51.4 ± 8.9 sec and 54.6 ± 12.1 sec were recorded during the DS and RDS, respectively, whereas the equivalent values for the untrained group were 62.2 ± 11.6 sec and 67.3 ± 13.5 sec, respectively.

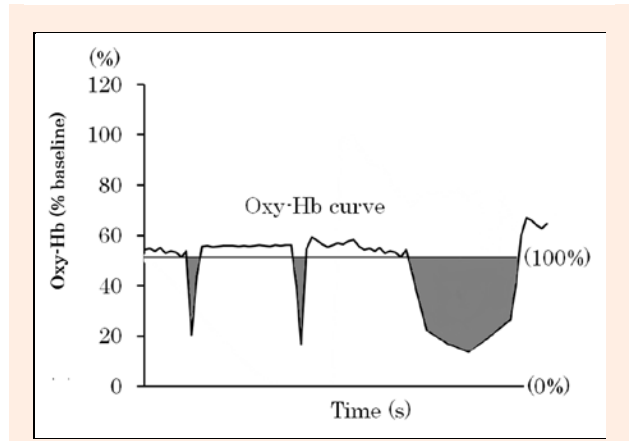


Figure 5. Typical example of an area under the Oxy-Hb curve. The Oxy-Hb level observed at rest was defined 100%, and the minimum Oxy-Hb plateau level induced by arterial occlusion was defined as 0%. The mean Oxy-Hb level was calculated every five seconds during exercise. The area under the Oxy-Hb curve was obtained from the product of the time (sec) and the mean Oxy-Hb (%). The gray regions represent the area under the Oxy-Hb curve.

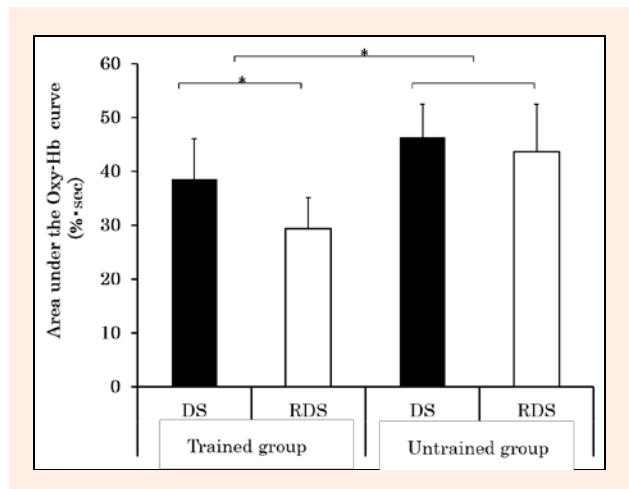


Figure 6. Area under the Oxy-Hb curve values of the trained and untrained groups during the drop-set (DS) and reverse drop-set (RDS) methods. Mean \pm SD ($n = 16$ for each group) values are shown. *Significant difference ($p < 0.05$)

Discussion

The aim of this study is to verify the influence of different load exercise to the muscle activity during subsequent exercise with 75% of 1RM load among trained and untrained individuals. The main findings of this study are that in the trained group higher concentric muscle power, larger area under the Oxy-Hb curve, and higher RMS values were recorded in the triceps brachii EMG during the DS than during the RDS. In the untrained group, none of the parameters differed significantly between the DS and RDS. Furthermore, it was found that the response of muscular tissue to changes in intramuscular oxygenation during exercise varies according to muscle thickness.

In the trained group, the triceps brachii muscle exhibited greater concentric power during the DS than during the RDS. Furthermore, higher RMS of EMG values during 75% of 1RM load exercise were recorded during the DS than during the RDS in the trained group. As RMS

values of EMG can be used an index of muscle fiber recruitment (Temfemo et al., 2007), this result agrees with the differences in muscle power detected in this study. Thus, it seems that the DS protocol increased the maximal firing rate of muscular motor units and recruited high-threshold motor units. Gorassini et al. (2002) reported that subjecting muscles to greater electrical stimulation increased the firing rate of their motor units just after the administration of the stimulus. Therefore, it is not surprising that the DS induced increases in the motor unit firing frequency and the recruitment of high-threshold motor units in the trained group. In the untrained group, no difference in muscle power was detected between the DS and RDS. Untrained individuals usually only place small loads on their muscles during activities of daily living so they might be able to recruit a limited number of motor units during exercise (Zucker, 1973).

As intramuscular hypoxic stimulation during exercise is likely to promote muscle hypertrophy (Takarada et al., 2000; Schott et al., 1995), the extent of the intramuscular hypoxic stimulation induced during the DS and RDS was investigated using the area under the Oxy-Hb curve. In the trained group, the mean area under the Oxy-Hb curve was higher during the DS than during the RDS. This might be explicable by the difference of RMS of EMG values during 75% of 1RM load exercise between the DS and RDS. The effects of the DS and RDS on muscle strength and hypertrophy require further investigation in a longitudinal study.

The blood vessel compression that occurs during strong muscle contractions will result in a greater area under the Oxy-Hb curve. However, the mean area under the Oxy-Hb curve was significantly smaller in the trained group than in the untrained group. This might be explained by the faster oxygen consumption recovery speed of the trained group (as indicated by their lower $TcVO_{2mus}$ values). Fryer et al. (2015) reported the same result concerning greater oxygen recovery in muscle tissue among elite athletes. The physical characteristics of the trained group, e.g., they probably had thicker and harder blood vessels and a higher capillary density, might have contributed to their lower $TcVO_{2mus}$ values (Tesch et al., 1988). In addition, the trained subjects' thick and hard vessels might not have been compressed much during muscle contractions. If this were true, then the intramuscular blood flow of the trained subjects might not have been limited to any great extent during the exercises. Furthermore, a negative correlation was detected between the area under the Oxy-Hb curve and triceps brachii muscle thickness among the whole study population (Figure 7). The greater RMS of EMG and area under the Oxy-Hb curve values of the untrained group might have had a positive impact on muscle hypertrophy, whereas the smaller values of the trained group might have a blunted one (van Wessel et al., 2010). However, the correlation coefficient between the area under the Oxy-Hb curve and triceps brachii muscle thickness was -0.51. It is necessary to verify this correlation using a number of subjects in our future research. As it is assumed that the reaction of muscles to the exercise is different between weight-bearing and non-weight-bearing muscles (Zhang et al. 2010), the

results of this study might be limited in an upper extremity muscle.

Conclusion

Muscle activity and acute intramuscular hypoxia during the DS and RDS were assessed among trained and untrained subjects. The DS induced greater muscle activation and intramuscular hypoxia in people who have been regularly performing resistance exercises for more than one year rather than the RDS. The thicker a person's muscles are, the more resistant they are to the induction of acute intramuscular hypoxia during muscle contraction is a suggested possibility. On the other hand, no mechanical or metabolic differences were detected between the DS and RDS among the subjects who had not participated in regular resistance training. Limited motor unit recruitment and an undeveloped microcirculation were considered to explain the latter results. The DS in which resistance exercise is initially performed with a higher load may increase the muscle activity and intramuscular hypoxia during subsequent exercise with 75% of 1RM load among trained individuals. This might have had a positive impact on muscle strengthening and hypertrophy. The long-term effects of DS on muscle strengthening and hypertrophy will be investigated in further research.

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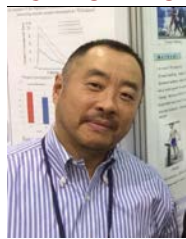
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Key points

- The DS induced greater motor unit activation and intramuscular hypoxia in people who have been regularly performing resistance exercises for more than one year than the RDS.
- No mechanical or metabolic differences were detected between the DS and RDS among the subjects who had not participated in regular resistance training.
- The thicker a person's muscles are, the more resistant they are to the induction of acute intramuscular hypoxia during muscle contraction.

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