Exercise speed and workload effects on muscle hypoxia in vastus lateralis muscle during squatting exercises

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ABSTRACT

Background: The study aimed to examine the effects of varying exercise speed and joint motion range (opting for either full knee extension or sustained knee flexion) on the intramuscular hypoxic environment, a key factor in muscle hypertrophy, during squat exercises. Methods: The participants were 17 healthy male and female students from Kibi International University, without back or knee pain at the time of evaluation. Two squat variations were performed: squats with full knee extension (Locked group; L group) and squats without full knee extension (non-locked group; NL group). The exercises were conducted in the following sequence with intervals: 12 s of non-locking (NL12), 12 s of locking (L12), 8 s of non-locking (NL8), 8 s of locking (L8), 4 s of non-locking (NL4), and 4 s of locking (L4). Tissue Oxygen Saturation (StO₂) in the right vastus lateralis muscle, under blood flow restriction during squat exercises, was measured using near-infrared spectroscopy. Results: The minimum StO₂ for NL12 was significantly lower than the resting StO₂ values. Similarly, the minimum StO₂ values for L12 and L8 were also significantly lower than the resting StO₂ values. The minimum StO₂ value for 8 s was significantly lower than at rest only in group L. Conclusion: When performing squat exercises, it is beneficial to reduce the exercise speed and increase the knee joint’s range of motion. This adjustment enhances the work of the quadriceps muscles, thereby creating an intramuscular hypoxic environment and promoting muscle hypertrophy.

Keywords: Sport medicine, Squatting, Blood flow restriction, StO₂, Muscle hypoxia.

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INTRODUCTION

Resistance training methodology encompasses various considerations, including the physiological response to exercise, loading volume, exercise speed, and the estimation of muscle exertion tension by the arm's length of joint moment. A physiological response induced by resistance training is the development of an intramuscular hypoxic environment. Research has demonstrated an increase in reactive oxygen species (ROS) activity in muscles under hypoxic conditions (Korthius et al., 1985). Moreover, nitric oxide, a component of ROS, has been identified as a mediator in vascular smooth muscle activation as well as muscle satellite cell activation and proliferation (Anderson, 2000).

The intramuscular environment, which seems to be associated with muscle hypertrophy, characterized by hypoxia and the accumulation of metabolic by-products, is believed to be optimized through sustained contractility and repetitive motion (work production) (Tanimoto and Ishii, 2006). In studies involving arm muscles (Bonde-Petersen, et al., 1975) and knee extensors (Koba, et al., 2004) under normal circulation conditions, continuous muscle contraction force generation at 40% MVC (Maximum Voluntary Contraction) has been shown to restrict blood inflow and outflow, leading to increased intramuscular pressure. Therefore, even without externally applied pressure, moderate-intensity resistance exercises (>40% MVC) that involve continuous force generation are expected to promote increases in muscular size and strength (Tanimoto and Ishii, 2006).

Near-infrared spectroscopy (NIRS) is a non-invasive method for measuring skeletal muscle hemodynamics. It has been reported that training the quadriceps with leg extension machines, an open kinetic chain exercise, at low-intensity (~50% of 1 Repetition Maximum), with slow movements and sustained tension generation, results in increases in both muscle size and strength following resistance training. Moreover, skeletal muscle oxygenation levels as measured by NIRS, decreased at the onset of slow exercise repetitions and were significantly higher post-exercise, compared to those in the resting state (Tanimoto and Ishii, 2006). This phenomenon likely corresponds to exercise hyperaemia, which results from muscle contraction compressing blood vessels and restricting blood flow immediately after the start of the exercise (Takemiya, et al., 1973).

Some training methods recommend squatting without full extension of the knee joint in order to maintain the muscle exertion tension (Tanimoto and Ishii, 2009). However, in the context of squatting exercises, a closed kinetic chain exercise, there is a lack of reports examining the relationship between exercise speed and the intramuscular hypoxic environment.

Therefore, this study aimed to examine the effects of altering exercise speed and joint motion range (between full knee extension or continuous knee flexion) on the intramuscular hypoxic environment, during squat exercises.

MATERIAL AND METHODS

Ethical considerations
This study was approved by the Kibi International University Research Ethics Review Committee (Approval No. 16-22). All participants were informed about the purpose and procedures of the study, and informed consent was obtained prior to their participation.
Participants and exercise training regimes

The study included 17 healthy students (7 males and 10 females) from Kibi International University, none of whom were experiencing back or knee pain at the time of evaluation. Their average body mass index was 21.98 ± 3.53.

Joint motion is defined as the rotational motion of each segment with the joint serving as the fulcrum, or the joint moment. The participants were assigned to perform two types of squats, differentiated by the arm length of the knee joint extension moment. The arm length of the knee joint extension moment is defined as the vertical distance from the knee joint, the fulcrum, to the line extending vertically from the body’s centre of gravity (Figure 1). The two squat variations included squats with full knee extension (Locked group; L group) and squats without full knee extension (non-locked group; NL group). The participants positioned their lower extremities one foot more than shoulder-width apart, with the hip joint externally rotated to angle the feet 30° outward relative to the mid-sagittal plane.

Participants were instructed to align their knees with their index toes, ensuring the knees did not extend beyond the toes. The range of motion for the knee joint in the non-locking group was maintained between 30° and 90° of flexion. The upper extremities were raised anteriorly to 90°. To ensure reproducibility of knee joint angles during the exercise, landmarks were established at the tips of the fingertips corresponding to 30° and 90° of knee flexion (Figure 1). The reason for initiating the knee flexion range of motion at 30° is based on the observation that rotatory motion occurs in the knee joint from full extension to approximately 30° of flexion (Neuman, 2009). The reason for limiting knee joint flexion to 90° is to prevent overloading the knee (Nakamura, and Ogata, 2016). The speed of each squat was quantified by varying the exercise speed in three distinct steps: 12, 8, and 4 s per squat, respectively. The exercises were performed in the following sequence with intervals: 12 s of non-locking (NL12), 12 s of locking (L12), 8 s of non-locking (NL8), 8 s of locking (L8), 4 s of non-locking (NL4), and 4 s of locking (L4). Each squat was performed for one minute. Intervals between each exercise set were maintained until tissue oxygen saturation (StO₂) fluctuations generally ceased. To ensure consistency in exercise speed and squat form, both were controlled using a metronome and verbal guidance.

Figure 1. Squatting for the locked (L) and non-locked (NL) groups.
The double circle represents the upper body’s centre of mass, the downward arrow signifies the line of the centre of mass, and the horizontal line that connects the centre of mass line to the knee joint axis represents the knee joint extension moment arm.

**Joint Range of Motion Measurement**

Joint range of motion during exercise was recorded using a two-dimensional goniometer. The intersection point of the goniometer was aligned with the axis of rotation of the knee joint’s centre (lateral condyle) on the lateral aspect of the right knee. The goniometer’s output was filtered through a high-cut filter at 30 Hz and then stored on a laptop computer via a data acquisition system (Power Lab, AD Instruments).

**Near-infrared spectroscopy measurement**

StO\(_2\) resulting from obtained by blood flow restriction to the right vastus lateralis muscle during squatting exercises, was measured NIRS (OMEGAMONITOR BOM-L1TRW, Omega Wave, Inc.).

This device uses continuous near-infrared light to measure four parameters: tissue oxygenated blood volume (Oxy-Hb), tissue deoxygenated blood volume (Deoxy-Hb), total tissue blood volume (Total-Hb), and StO\(_2\). Total-Hb and StO\(_2\) are calculated using the following formulas: Total-Hb = Oxy-Hb + Deoxy-Hb; StO\(_2\) = (Oxy-Hb / Total-Hb) × 100%. StO\(_2\) measurements were performed by transmission. The laser emitting part and the detector were covered with a special rubber sheet and secured with adhesive tape. The depth of near-infrared light penetration was set at 30 mm (Ishida, et al., 1986). The NIRS signal was recorded on a personal computer.

![Figure 2](image)

Figure 2. The upper row of the figure shows example of StO\(_2\) measurement by NIRS. The Lower row of the figure shows example of measurement of knee joint angle by goniometer corresponding to the upper row. From left to right: 12 s of non-locking (NL12), 12 s of locking (L12), 8 s of non-locking (NL8).

**Statistical analysis**

For statistical analysis, the mean of the lowest StO\(_2\) value for each exercise and the mean of the resting StO\(_2\) values before the start of each exercise (all resting values at 12, 8, and 4 seconds) were compared for each group. A one-way analysis of variance was employed to examine the main effect of each outcome across
groups. Post-hoc comparisons were conducted using the Tukey method for differences in relative weight ratios. Measurements are presented as mean ± standard deviation. Statistical analyses were conducted using IBM SPSS Statistics Base. The level of significance was set at 5%.

RESULT

Vastus lateralis StO₂
Minimum StO₂ values for NL12 were significantly lower than the resting StO₂ values. Similarly, the StO₂ minimums for L12 and L8 were also significantly lower than their respective resting values. In the group with a squat speed of 8 seconds, the minimum StO₂ value was significantly lower relative to the resting StO₂ value only in the NL group (Table 1).

Table 1. Tissue blood oxygen saturation in the vastus lateralis.

<table>
<thead>
<tr>
<th>Group</th>
<th>n</th>
<th>rest. (%)</th>
<th>12 sec. (%)</th>
<th>8 sec. (%)</th>
<th>4 sec. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NL</td>
<td>17</td>
<td>63.17 ± 5.23</td>
<td>53.04 ± 10.77*</td>
<td>54.04 ± 11.19</td>
<td>55.13 ± 12.00</td>
</tr>
<tr>
<td>L</td>
<td>17</td>
<td>64.01 ± 4.95</td>
<td>53.64 ± 10.51*</td>
<td>54.16 ± 11.17*</td>
<td>56.14 ± 11.18</td>
</tr>
</tbody>
</table>

Note. Values for each group (mean ± standard deviation). *p < .05 (vs rest.). L = locked group. NL = non-locked group.

DISCUSSIONS

Our studies results align with findings from previous research, suggesting that joint exercise performed at a reduced speed increases muscle load and creates a hypoxic environment within the muscle (Tanimoto and Ishii, 2006).

In the squat exercise of the L group, the body’s centre of gravity line approached the knee joint axis when the knee joint was fully extended. Consequently, the knee joint extension moment arm in the L group is shorter than that in the NL group (Figure 1). Consequently, it was hypothesized that the L group, with the knee joint fully extended, would be less effective than the NL group in loading the vastus lateralis muscle and, therefore, less likely to achieve an appropriate intramuscular hypoxic environment. However, against our expectations, the 8-s minimum StO₂ value was significantly lower compared to the resting value exclusively in the L group.

The work ("W") produced in the quadriceps during squatting exercise is the product of the force (a combination of the mass of the upper body, including the thighs, and the acceleration of gravity: F) and the distance (x) moved by the upper body (W = Fx). The significantly lower StO₂ at the 8-s minimum in the L group, as compared to the resting value, is thought to be attributable to the increased travel distance (x) of the upper body, including the thighs, due to the full extension of the knee joint in squats. We theorized that this increased travel distance (x) amplified the work (W) performed by the L group, thus imposing a greater load on the vastus lateralis muscle compared to the NL group.

Resistance training, as typified by squatting exercises, increases the number and size of type Iib or type Iix (Ezaki O., 2012). The results of this study can be applied to exercises for people seeking to improve lower limb muscle strength and as an effective prevention method for sarcopenia, a condition in which predominantly type II fibres atrophy.

Measurement of peripheral muscle oxygenation by NIRS often uses the transient arterial occlusion method. This is a method of calibrating StO₂ values by applying a pressure cuff to the proximal part of the extremity,
inflating it to 280-300 mmHg until a minimum plateau level of Oxy-Hb is obtained, blocking arterial blood flow, with the StO₂ value at that time set at 0% and the resting StO₂ value at 100% (Tanimoto and Ishii, 2006; Kimura, et al., 2006). However, this method was not used in this study. All StO₂ measurements were higher in the L group than in the NL group. All measurements in the L group were performed after the NL group. The vascular and haemodynamic changes in the measured muscles caused by the exercise in the NL group may have influenced the StO₂ values in the L group.

CONCLUSIONS

Our findings demonstrated that the creation of an intramuscular hypoxic environment, which leads to muscle hypertrophy, is greatly influenced by exercise speed and the amount of work generated during squatting exercises. It is therefore advisable in squat exercises to slow down the exercise speed and increase the knee joint's range of motion. This approach amplifies the workload on the quadriceps muscles, enabling an intramuscular hypoxic environment that can stimulate muscle hypertrophy.

AUTHOR CONTRIBUTIONS

Tetsuo Imano: study design, data analysis, manuscript preparation. Masaaki Nakajima: study design, data analysis.

SUPPORTING AGENCIES

No funding agencies were reported by the authors.

DISCLOSURE STATEMENT

No potential conflict of interest was reported by the authors.

ETHICS STATEMENT

The experiments completed in this study comply with the current laws of the country in which they were performed.

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REFERENCES


