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Vastus lateralis oxygenation and blood volume measured by near-infrared spectroscopy during whole body vibration

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Summary

The purpose of this study was to investigate the effects of whole body vibration (WBV) on oxygenation of vastus lateralis muscle during squatting exercise. Eighteen male subjects [mean age, 27·3 ± 6·0 (SD) years; mean height, 171·8 ± 4·9 cm; mean weight, 64·4 ± 6·1 kg] performed squatting exercise on a vibration platform for 3 min with and without vibration, and changes in oxygenation of the vastus lateralis muscle were determined by near-infrared spectroscopy. The muscle oxygenation levels and total haemoglobin and myoglobin levels (total Hb/Mb) decreased during squatting exercise with and without vibration. After exercise, the muscle oxygenation level and total Hb/Mb rapidly increased from the minimum value during exercise and remained constant for latter 10 min. The muscle oxygenation levels with vibration from 90 to 180 s after the start of squatting exercise were significantly lower than those without vibration. Total Hb/Mb with vibration from 90 s after the squatting exercise to 540 s were significantly higher than those without vibration. This study demonstrated that WBV exercise affects the oxygenation level of vastus lateralis muscle and reduces muscle oxygenation level compared to that with no WBV. Therefore, WBV exercise may be an efficient training stimulus for muscle deoxygenation.

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Introduction

Whole body vibration (WBV) is a new neuromuscular training method using physiological response to vibration stimuli and has been tested in several fields (Bosco *et al.*, 1998, 1999, 2000; Rittweger *et al.*, 2000; Rubin *et al.*, 2001). It has been hypothesized that mechanical stimulation in the form of vibration to the human body is an efficient way to improve muscle strength, body balance and the mechanical component of bone. As for short-term effects of WBV exercise on muscle performance, previous studies have shown significant increases in isometric leg extension strength and in power and jump height after a single training session (Bosco *et al.*, 1999, 2000) and also after 10 days of training (Bosco *et al.*, 1998). As for the long-term effects on muscle performance, Torvinen *et al.* (2002, 2003) reported significant increases in jump height after 4 and 8 months of WBV training.

It has been suggested that the effects of WBV on muscle performance are elicited via reflex muscle activation (Rittweger *et al.*, 2000). This neuromuscular response has been named 'tonic vibration reflex (TVR)' and is able to cause an increase in recruitment of motor units through activation of muscle spindle and polysynaptic pathways (De Gail *et al.*, 1966; Matthews *et al.*, 1966; Seidel *et al.*, 1998). A significantly higher level of electromyographic activity of the vastus lateralis muscle was found in a half squat position during WBV than that in a non-vibrating condition (Cardinale *et al.*, 2003). An increase in the level of muscle activity caused by WBV affects the cardiovascular system. Rittweger *et al.* (2001) reported that oxygen uptake during standing and squatting exercise with WBV was greater than that without WBV. Therefore, it is important to clarify the effects of WBV on muscle oxygen metabolism.

The study of human muscle energetics has undergone radical change in recent years due to the development of non-invasive measurement techniques, including near-infrared spectroscopy (NIRS). In NIRS, the differential absorption properties of oxygenated and deoxygenated haemoglobin (Hb) and myoglobin (Mb), especially wavelengths of 760 and 840 nm, are used to evaluate muscle oxygen content (Chance *et al.*, 1992; Hamaoka *et al.*, 1992; Shiga *et al.*, 1997). NIRS detects the

balance between oxygen supply and oxygen utilization in human skeletal muscle and enables measurement of muscle oxygenation in vessels, capillaries and intracellular sites (Chance *et al.*, 1992; Hamaoka *et al.*, 1992). Measurement of skeletal muscle oxygenation by using NIRS might, therefore, be an indication of localized muscle activities. However, there has been no study on changes in muscle oxygenation during WBV.

The purpose of this study was to investigate the effects of WBV on oxygenation of the vastus lateralis muscle during and after squatting exercise in healthy males using NIRS. It was hypothesized that WBV would result in greater muscle oxygenation decrease than that without WBV, since WBV causes an increase in oxygen consumption due to an increase in the level of muscle activity.

Methods

Subjects

Eighteen males aged 20–39 years [mean height, 171·8 ± 4·9 (SD) cm; mean weight, 64·4 ± 6·1 kg] volunteered for this study. None of them were engaged in regular organized physical activities or in sports or strength training, and none of them had chronic exposure to WBV as a result of their occupation. All of the subjects were fully informed about the nature of the experiments, including possible risks and gave written informed consent for participation in the study. The protocol was accepted by the local ethics committee.

Procedure

First, for estimation of the effects of WBV, vibrating loading was carried out on a WBV platform (Galileo 900; Novotech, Pforzheim, Germany) in the standing position.

Next, vibration loading was carried out during squatting exercise. The vibration frequency was set to 15 Hz, and the feet were placed 25 cm apart. Thus, the amplitude of vibration was 2·5 mm (5 mm from top to bottom).

The study protocols for the two conditions were the same except that the vibration platform vibrated during the squatting exercise with WBV. Squatting exercise was performed from almost complete extension of the knees to an angle of 60 ° for 3 min. Each subject stood on the platform in bare feet and was instructed to move down over a period of 1 s, keep a steady state for 1 s, and then move up over a period of 1 s. Measurements with and without vibration were performed at random.

Near-infrared spectroscopy

Changes in muscle oxygenation were measured using a near-infrared spectrometer (HEO-200, OMRON Inc., Kyoto, Japan). The NIRS probe consisted of a light source and an optical detector, with a distance of 3·0 cm between the light source and detector providing sensory input for the unit. A pair of two-wavelength light-emitting diodes, with wavelengths of 760 and 840 nm, was used as the light source. The probe was placed over the belly of right vastus lateralis muscle. The distance from base of the patella to the centre of the probe was 11·5 ± 1·1 cm. The depth of penetration of the light from the surface of the skin was 1·5–2 cm (Okada *et al.*, 1995; Shiga *et al.*, 1997). Changes in deoxygenated Hb and Mb (Δ deoxy Hb/Mb) and in total Hb and Mb (Δ total Hb/Mb) were calculated according to the method of Shiga *et al.* (1997) as follows.

Tissue oximetry using NIRS is based on the modified Beer–Lambert law. The following equations are used as basic equations:

$$\Delta \mathrm{OD}_{840} = \mathrm{k1}\Delta[\mathrm{HbO_2}] + \mathrm{k1'}\Delta[\mathrm{Hb}], \tag{1}$$

$$\Delta \mathrm{OD}_{760} = \mathrm{k2}\Delta[\mathrm{HbO_2}] + \mathrm{k2'}\Delta[\mathrm{Hb}], \tag{2}$$

where $\Delta \mathrm{OD}_{840}$ and $\Delta\ \mathrm{OD}_{760}$ indicate changes in optical density at 840 and 760 nm, respectively, and Δ [$\mathrm{HbO_2}$] and Δ [Hb] denote changes in the concentrations of oxy Hb and deoxy Hb, respectively. The coefficients k1, k1′, k2 and k2′ are assumed to be constant, although they are dependent upon absorption and scattering when the concentrations vary greatly. The following equations are obtained from Eqs. (1) and (2):

$$\Delta[\mathrm{HbO_2}] = \mathrm{K}\left[\Delta \mathrm{OD}_{840} - \left(\frac{\mathrm{k1'}}{\mathrm{k2'}}\right)\Delta \mathrm{OD}_{760}\right], \tag{3}$$

$$\Delta[\mathrm{Hb}] = \mathrm{K}\left(\frac{\mathrm{k2}}{\mathrm{k2\prime}}\right)\left[\left(\frac{\mathrm{k1}}{\mathrm{k2}}\right)\Delta \mathrm{OD}_{760} - \Delta \mathrm{OD}_{840}\right], \tag{4}$$

where

$$\mathrm{K} = \frac{\mathrm{k2'}}{(\mathrm{k1}/\mathrm{k2'}) - (\mathrm{k1'}/\mathrm{k2})} = \frac{1/\mathrm{k2}}{(\mathrm{k1}/\mathrm{k2}) - (\mathrm{k1'}/\mathrm{k2'})}.$$

Δ BV, which indicates changes in blood volume, i.e., changes in total amount of haemoglobin, is obtained as the sum of Δ [$\mathrm{HbO_2}$] and Δ [Hb]:

$$\begin{aligned}\Delta \mathrm{BV} &= \Delta[\mathrm{HbO_2}] + \Delta[\mathrm{Hb}] \\ &= \mathrm{K}\left[\left(1 - \frac{\mathrm{k2}}{\mathrm{k2\prime}}\right)\Delta \mathrm{OD}_{840} + \left(\frac{\mathrm{k2}}{\mathrm{k2'}}\right)\left(\frac{\mathrm{k1}}{\mathrm{k2}} - \frac{\mathrm{k1'}}{\mathrm{k2'}}\right)\right]\Delta \mathrm{OD}_{760}.\end{aligned} \tag{5}$$

The ratios k1/k2, k1′/k2′ and k2/k2′ can be experimentally determined by measuring the ratios of optical density changes in a fully muscle oxygenated state (oxy) and a fully muscle deoxygenated state (deoxy),

$$\frac{\mathrm{k1'}}{\mathrm{k2'}} = \frac{\Delta \mathrm{OD}_{840}}{\Delta \mathrm{OD}_{760}} \text{ in deoxy}, \tag{6}$$

$$\frac{\mathrm{k1}}{\mathrm{k2}} = \frac{\Delta \mathrm{OD}_{840}}{\Delta \mathrm{OD}_{760}} \text{ in oxy}, \tag{7}$$

$$\frac{k2}{k2'} = \frac{\Delta OD_{760} \text{ in oxy}}{\Delta OD_{760} \text{ in deoxy}}.$$

K is tentatively considered to be 1, although this coefficient can be determined if a differential path length involved in k2 is available.

According to Shiga *et al.* (1997), the average coefficients k1/k2, k1′/k2′ and k2/k2′ were 1·35, 0·661 and 0·587, respectively. Thus, the following equations were obtained.

$$\Delta\text{deoxy Hb/Mb} = 0 \cdot 80\Delta OD_{760} - 0 \cdot 59\Delta OD_{840},$$

$$\Delta\text{total Hb/Mb} = 0 \cdot 41\Delta OD_{840} + 0 \cdot 14\Delta OD_{760}.$$

The mean thickness of the fat layer at the NIRS measurement sites determined by using a B-mode ultrasonic apparatus fitted with a 5-MHz linear array transducer (LOGIQ α100, Yokogawa Medical Inc., Tokyo, Japan) was 5·5 ± 0·9 mm. Since there was no difference between fat layers in the subjects, we did not correct the influence of a subcutaneous fat layer on measurement. Changes in deoxy Hb/Mb and total Hb/Mb were measured throughout the experimental period.

Before the squatting exercise, the arteries in the right leg were occluded by inflating a cuff on the thigh to a pressure of 300 mmHg until a minimum level of deoxy Hb/Mb was reached for calibration in the standing position (Chance *et al.*, 1992; Hamaoka *et al.*, 1992; Shiga *et al.*, 1997). Then the cuff was released, and the subject remained in a resting state. Squatting exercise was started after the changes in Hb and Mb signals obtained by NIRS were stable for 30 s without WBV, and squatting exercise was continued for 3 min with and without WBV. The measurements were continued for 10 min after exercise in the standing position. The period between the two conditions was sufficiently long to allow adequate rest so that all measurement values returned to premeasurement values.

Physiological measurements

Changes in heart rate, blood pressure and arterial oxygenation were measured under the two conditions in 9 of the 18 subjects. Heart rate was measured using a telemetry electrocardiograph (Ds-2202; Fukuda Densi, Tokyo, Japan). Blood pressure was measured by non-invasive measurements in the left upper arm (HR-500; Fukuda Densi). Arterial oxygenation was monitored using a pulse oxymeter (Biox 3740 Pulse Oxymeter; Ohmeda, USA) from the tip of the right index finger. All measurements were performed during squatting exercise and continued for 10 min after exercise in the standing position by a unit of 1 min.

Analysis

A value reflecting the baseline of the amount of change in deoxy Hb/Mb was defined as the muscle oxygenation level. The deoxy Hb/Mb signal measured by this device does not indicate the absolute values of tissue oxygenation. We therefore normalized the muscle oxygenation level using the arterial occlusion method at rest (Hamaoka *et al.*, 1996; Rundell *et al.*, 1997). We judged visually on the display connected to the NIRS device whether a minimal level of deoxy Hb/Mb had been reached. The value at rest when measurement started was standardized as 100%, and the minimum value recorded during arterial occlusion was 0%. Total Hb/Mb indicated a raw value (OD). The values of muscle oxygenation level and Δ total Hb/Mb were averaged by a unit of 30 s.

Statistical analysis was performed using StatView for Windows (Version 5·0, SAS Institute Inc., California, USA). Changes in muscle oxygenation level, total Hb/Mb, heart rate, blood pressure and arterial oxygen saturation were analysed, and comparisons with and without vibration were made using Student's paired *t*-test. The significance level was set at less than 5% in statistical tests.

Results

We measured oxygenation of the right vastus lateralis muscle in a state in which the leg was lightly placed on the platform in the standing position with and without vibration to investigate the effect of the NIRS reading due to movements of the optical probe. Muscle oxygenation (OD) with and without vibration were 0·017 ± 0·007 and 0·002 ± 0·005, respectively. Muscle oxygenation (OD) with vibration was significantly higher than that without vibration.

The muscle oxygenation level and total Hb/Mb decreased in both conditions during the squatting exercise. After exercise, the muscle oxygenation level and total Hb/Mb rapidly increased and remained constant for latter 10 min. The muscle oxygenation levels with vibration were significantly lower than those without vibration at 90, 120, 150 and 180 s after the start of squatting exercise, and the differences were 5·9 ± 9·6, 6·6 ± 7·7, 5·0 ± 9·4 and 6·1 ± 9·7%, respectively ($P<0·05$) (Fig. 1). Total Hb/Mb with vibration from 90 s after squatting exercise to 540 s were significantly higher than those without vibration ($P<0·05$) (Fig. 2). Heart rate with vibration at only 30 s after squatting exercise was higher than that without vibration ($P<0·05$) (Fig. 3). There was no difference between changes in systolic and diastolic blood pressure with and without vibration (Fig. 4). Average oxygen saturation rates with and without vibration were maintained at 98·2 ± 0·2 and 98·4 ± 0·2%, respectively, and did not show a significant difference.

Discussion

Recent developments in optical instrumentation using near-infrared light have made it possible to directly and non-invasively measure changes in relative oxygenation in Mb and Hb (Hamaoka *et al.*, 1992). NIRS has been utilized to evaluate tissue oxygenation and function in resting and exercising skeletal muscle (Chance *et al.*, 1992; Hamaoka *et al.*, 1992), and

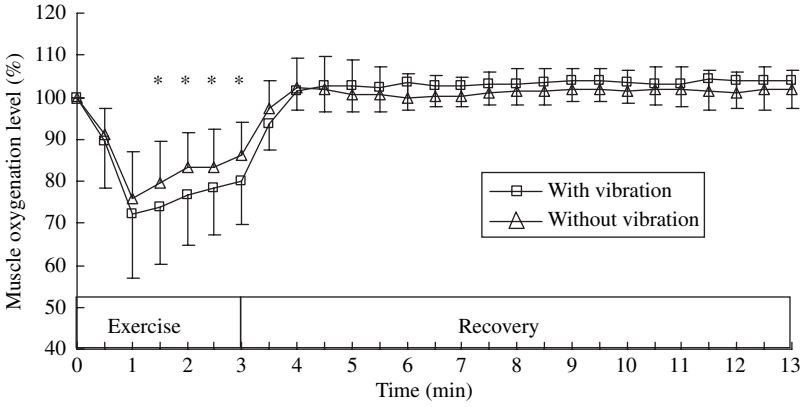

Figure 1 Changes in Δ muscle oxygenation level (mean ± SD) during squat exercise with and without vibration and recovery. *, statistically significant difference ($P<0{\cdot}05$, Student's paired t-test) from those at the same time with vibration.

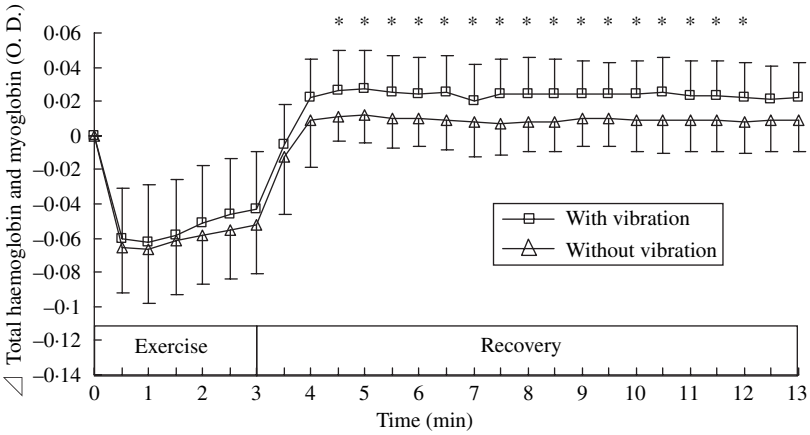

Figure 2 Changes in Δ total haemoglobin and myoglobin (mean ± SD) during squat exercise with and without vibration and recovery. *, statistically significant difference ($P<0{\cdot}05$, Student's paired t-test) from those at the same time with vibration.

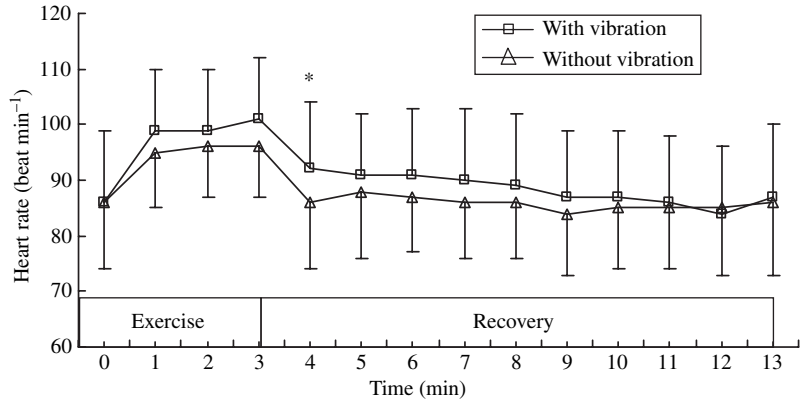

Figure 3 Changes in heart rate (mean ± SD) during squat exercise with and without vibration and recovery. *, statistically significant difference ($P<0{\cdot}05$, Student's paired t-test) from those at the same time with vibration.

it has been found to be useful for examining muscle oxygenation continuously. However, there has been no study on changes in muscle oxygenation during WBV. We therefore investigated the changes in muscle oxygenation during WBV.

In this study, the oxygenation level in the vastus lateralis muscle with vibration showed a significant decrease compared with that without vibration. Muscle oxygenation level is affected by the balance between oxygen utilization and oxygen supply in human skeletal muscle (Chance *et al.*, 1992; Hamaoka *et al.*, 1992). Therefore, the reason for the decrease in muscle oxygenation was one of the following three patterns: oxygen supply was stable and oxygen utilization increased, oxygen utilization was stable and oxygen supply decreased, or oxygen utilization increased and oxygen supply decreased.

Mechanical vibration applied to the muscle belly, tendon or whole body had been shown to elicit a TVR (De Gail *et al.*, 1966; Matthews *et al.*, 1966; Hagbarth & Eklund, 1985; Seidel *et al.*, 1998). Muscle spindle Ia afferents have been indicated to be the major determinant of this vibration-induced neuromuscular activation leading to TVR. It has been suggested that TVR is

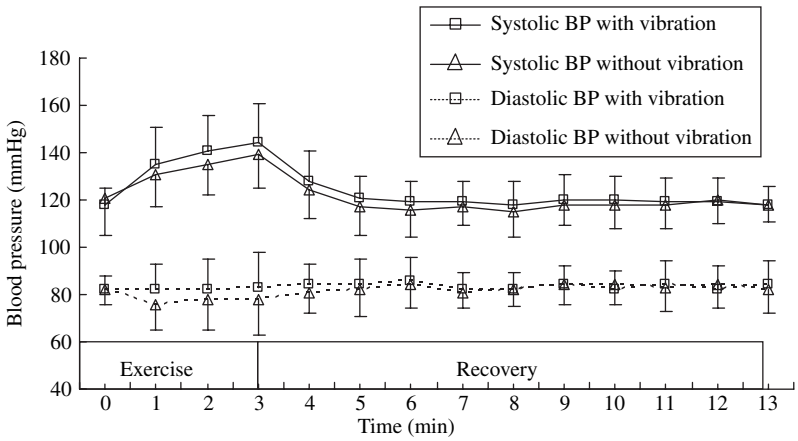

Figure 4 Changes in blood pressure (mean ± SD) during squat exercise with and without vibration and recovery.

elicited by WBV at a frequency of 1–30 Hz (Seidel *et al.*, 1998). The TVR response may increase recruitment of motor units via activation of muscle spindles and polysynaptic pathways, the effect of which is seen as a temporary increase in the level of muscle activity (De Gail *et al.*, 1966). Cardinale *et al.* (2003) reported that a significantly higher level of activity of the vastus lateralis muscle was found in a half squat position during WBV than in a non-vibrating condition. Therefore, the reason for the decrease in muscle oxygenation with WBV was thought to be increase in oxygen utilization.

In the present study, there was no difference between changes in total Hb/Mb with and without vibration during squatting exercise, but total Hb/Mb with vibration were significantly higher than those without vibration from 90 s after the squatting exercise to 540 s. Kerschan-Schindle *et al.* (2001) reported that muscular blood circulation in the calf and thigh significantly increased after one bout of WBV exercise. These results suggest that WBV causes an increase in blood volume after squatting exercise, but WBV does not affect peripheral circulation during squatting exercise. Therefore, oxygen delivery did not meet the oxygen demand in the muscles, and the mismatching between oxygen delivery and demand during WBV was larger than that without WBV. Indeed, Rittweger *et al.* (2001) reported that oxygen uptake during standing and squatting in a vibrating condition was greater than that in a non-vibration condition. Rittweger *et al.* (2003) also reported that time to exhaustion during squatting exercise was significantly shorter with vibration than without vibration, suggesting that WBV imposes more stress on neuromuscular structures.

In conclusion, this study demonstrated the effects of WBV exercise on oxygenation level of the vastus lateralis muscle and showed that muscle oxygenation level decrease during a vibration condition. More research on WBV is needed to clarify the mechanisms of muscle metabolism.

Acknowledgments

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