

Effects of osteocytes on vibration-induced reflex muscle activity in postmenopausal women

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Background/aim: To assess whether osteocytes have an effect on reflex myoelectrical activity during whole-body vibration (WBV) in postmenopausal women.

Materials and methods: Participants were classified into 2 groups: the low bone mineral density (BMD) group (n = 37) and normal BMD group (n = 43). Hip BMD was measured using dual-energy X-ray absorptiometry. Surface electromyography data recorded from the adductor longus muscle were processed to obtain vibration-induced reflex myoelectrical activity. Changes in plasma sclerostin (SOST) levels with WBV were expressed as a standardized vibration-induced SOST index.

Results: The standardized vibration-induced SOST index was 1.03 ± 0.24 in the low BMD group and 0.99 ± 0.33 in the normal BMD group. For plasma SOST levels, no group-by-time interaction was found. The resting myoelectrical activities of adductor muscles increased significantly during WBV in both groups. However, there was no significant difference in the main effects of WBV on resting myoelectrical activity between the groups. The standardized vibration-induced plasma SOST index was found to be a significant independent predictor of the standardized vibration-induced reflex myoelectrical activity of the adductor muscle in both groups.

Conclusion: This study suggests that osteocytes serve as mechanoreceptors of reflex electromyography during WBV.

Key words: Plasma sclerostin protein, human, bone density, electromyography

1. Introduction

Whole-body vibration (WBV), as a method of exercise training, is becoming increasingly popular in physical therapy, rehabilitation, and professional sports, and is increasingly used in beauty and wellness applications due to its beneficial effects on the neuromusculoskeletal system. These benefits include the enhancement or improvement of strength and power of muscles, balance coordination, and bone mass, or at least the prevention of loss of strength and power of muscles, balance coordination, and bone mass (1–4).

Little is known about the physiological mechanisms underlying the effects of WBV on neuromuscular performance, although the presence of reflex muscle activity during WBV has been shown (4,5). The “tonic vibration reflex” (TVR) is the most commonly cited mechanism to explain the effects of WBV on neuromuscular

performance, although there is no conclusive evidence that TVR occurs (4–6). In locally applied vibration studies, it has been reported that direct vibration applied to a muscle or tendon stimulates muscle spindles, causing TVR to occur. As highlighted by these studies, muscle spindle discharges are sent to the motoneurons through Group Ia afferents during muscle or tendon vibration. There, they activate reflex arcs that cause the muscle to contract (3,4,6,7). However, it has been reported that the sensitivity of the muscle spindle decreases or does not increase and that presynaptic inhibition occurs in Group Ia afferent fibers with vibration (6,8–14).

The bone myoregulation reflex (BMR) is another neurological mechanism used to explain the effects of vibration on neuromuscular performance. BMR is a reflex mechanism in which osteocytes exposed to cyclic mechanical loading induce muscle activity. Osteocytes

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embedded in the bone matrix are interconnected by numerous dendritic processes, forming a wide, mechanosensitive cellular network. Osteocytes exposed to cyclic mechanical loading send mechanical input signals to the central nervous system, influencing the neuronal regulation of muscle activity (11,15–17). This study was designed to test the role of BMR in the increase in myoelectrical activity during WBV (vibration-induced reflex myoelectrical activity) in postmenopausal women.

We hypothesize that osteocytes play a role in increasing myoelectrical activity during WBV. We further hypothesize that because the number of osteocytes is reduced during postmenopausal osteoporosis, the effects of osteocytes on vibration-induced reflex myoelectrical activity may be weaker in postmenopausal women with osteoporosis than in postmenopausal women without osteoporosis. The aim of this study was to assess whether osteocytes affect reflex myoelectrical activity during WBV in postmenopausal women.

2. Materials and methods

2.1. Subjects

Ethical approval was obtained from the Institutional Review Board (Ethics Committee of İstanbul University, İstanbul Medical Faculty, 2010/363-86). The study was performed in accordance with the principles of the Declaration of Helsinki. All participants were volunteers and provided written informed consent.

In total, 397 postmenopausal outpatients who underwent bone densitometry measurements on

suspicion of osteoporosis were screened at our hospital. Of them, 133 met the inclusion criteria; 130 postmenopausal women who voluntarily agreed to participate in the study were assessed for eligibility. According to the criteria presented in Table 1, 34 of these individuals were excluded. Specifically, we excluded 6 subjects who felt dizzy during WBV, 3 whose blood samples were not taken, 6 whose blood samples had hemolyzed, and 1 who could not relax during electromyography (EMG) recordings. In total, there were 80 participants in the study.

2.2. Procedures

This was a prospective, controlled, double-blind, clinical trial. Vibration-induced reflex muscle activity was evaluated using surface EMG (SEMG). To examine the effects of osteocytes on vibration-induced reflex muscle activity, hip bone mineral density (BMD) and plasma sclerostin (SOST) levels were measured.

Osteocytes, which are mechanosensitive bone cells, are the “receptors” of BMR (17). SOST, a mechanosensitive protein, is produced almost exclusively by osteocytes. Its expression in adult bone is regulated by mechanical strain (18–20). The plasma SOST level indicates osteocyte activity (21). WBV can cause changes in plasma SOST levels in adult women (22). In this study, changes in plasma SOST level with WBV were used as a tool to demonstrate changes in osteocyte activity during WBV.

To examine the effects of osteocytes on vibration-induced reflex muscle activity, hip BMD was used as a bone-related second variable. The number of osteocytes is reduced during postmenopausal osteoporosis (23,24).

Table 1. Inclusion and exclusion criteria.

Inclusion criteria

1. Postmenopausal women between the ages of 45 and 65.
2. Participants with total hip or femoral neck T-score less than or equal to -2.0 or participants with normal hip and lumbar bone mineral density (T-score greater than or equal to -1.0).

Exclusion criteria

1. Early menopause.
2. Metabolic bone disease (secondary osteoporosis, osteomalacia, Paget’s disease), fracture history, or medication that could affect the musculoskeletal system.
3. Muscle, tendon, joint, vascular, or dermatological diseases in the lower extremities.
4. Postural abnormalities (e.g., scoliosis, kyphosis).
5. Systemic diseases:
 - a. Hypertension (diastolic > 85 mmHg, systolic > 135 mmHg).
 - b. Heart diseases (e.g., coronary heart disease, conduction or rhythmic problems, pacemaker).
 - c. Infectious disease.
 - d. Endocrine disease, such as diabetes mellitus.
 - e. Renal calculi.
6. Neurologic disorders: peripheral neuropathy, poliomyelitis squal.
7. Vertigo.
8. Cognitive dysfunction.

To test the validity of our hypotheses, participants were classified into 2 groups on the basis of their T-score for BMD: the low BMD group (n = 37) and the normal BMD group (n = 43).

According to the World Health Organization (WHO) criteria, a normal BMD is defined as higher than 1 standard deviation below the young adult women reference mean (T-score greater than or equal to -1) (25). Thus, the normal BMD group was defined by a BMD of the hip and lumbar region that was not more than 1 standard deviation below the reference mean for young healthy women. The low BMD group was defined by a BMD of the total hip or femoral neck that was more than 2 standard deviations below the reference mean for young healthy women (T-score less than or equal to -2). Due to densitometric measurement error, mistakes in the classification of participants into the normal BMD and low BMD groups may have occurred. Participants with T-scores between -1 and -2 were not included to avoid any effects of measurement error.

Hip BMD was measured once prior to the trial. SEMG analyses were done before and during WBV. Plasma SOST level measurements were done before and after WBV.

2.2.1. Bone mineral density

Left hip and lumbar BMD were measured using an XR-46 bone densitometer machine (Norland, Malvern, PA, USA). The precision error is <1.2 % for lumbar AP, spine, and hip.

2.2.2. Whole-body vibration

WBV was performed using a My5 Power Plate (Performance Health Systems, London, UK). Two sets of WBVs (amplitude: 2 mm, frequency: 40 Hz, acceleration: 2.7 g, set duration: 30 s) were used for all participants. There was a rest period of 10 s between sets.

The participants were barefoot, and no sponge or foam was placed between the vibration platform and their feet. Participants stood upright with their knees locked on the vibration platform. This position is used because it exposes the lower extremity bones to maximum mechanical load while preventing stretching of the extrafusal and intrafusal muscle fibers in the adductor muscles. In most studies of WBV, vibration has generally been applied in a squatting or semisquatting position; however, intramuscular tension is high in such positions (6,26).

Maintaining static balance in a standing position may be difficult during WBV, and muscle contractions in the lower leg may occur to maintain balance. To prevent such instability, 2 precautions were taken. First, the base of support (distance between the heels) was adjusted. Two white strips were pasted 30 cm apart on the vibration platform, and participants were asked to place their feet on these 2 white strips. As a second precaution, participants were asked to hold the handles of the WBV equipment.

2.2.3. Surface electromyography

SEMG recordings (POWERLAB Data Acquisition System, AD Instruments, Oxford, UK) were obtained from the adductor longus muscle. SEMG was sampled at a frequency of 2 kHz.

Self-adhesive, disposable, Ag/AgCl disk electrodes 10 mm in diameter (Kendall ARBO, Covidien AG, Mansfield, MA, USA) were used. Prior to electrode placement, the skin was cleaned with alcohol wipes, lightly abraded, and, if necessary, shaved. To place electrode pairs on the left adductor longus muscles, participants were asked to lie in a supine position and keep about 15° of abduction in each hip. Electrode pairs were placed 5 cm caudal to the tuberculum pubicum on the left adductor longus muscle belly. To test the accuracy of electrode pair locations, participants were asked to lie in a supine position with both knees at 90° flexion and each hip at 15° abduction. Then participants were asked to adduct their hips against resistance, which applied pressure against the medial aspect of the distal end of the thighs in the direction of the abduction. During this clinical test, SEMG activity in the left adductor longus muscle was assessed to see if it increased. To prevent the snap end of the cable from swinging, it was fixed to the body. In all cases, electrodes were placed by the same laboratory technician.

SEMG recordings were obtained in 3 positions: in a supine lying position, in a resting standing position, and during WBV. While SEMG recordings were being obtained in these positions, participants were asked to relax and not to make voluntary contractions in their lower extremity muscles. For that purpose, participants were given relaxation training while in the lying position and the resting standing position. This training was provided with verbal feedback from the researcher who monitored the SEMG recordings on the screen. The same verbal feedback was given during WBV to preserve the relaxed state of the participants. The first set of WBV was used to familiarize participants with the process.

The data were processed offline with a computer. All SEMG analyses were conducted using LABCHART7 software (ver. 7.3.3; POWERLAB System, AD Instruments). All SEMG recordings and analyses were conducted by the same researcher.

The first and last 1.5 s of the exercise data were discarded to eliminate vibration onset and finish effects (artifacts), and the remaining 27 s were analyzed further.

Myoelectrical activity measurements recorded via SEMG could be affected by motion artifacts and 50 Hz noise due to electromagnetic fields from electrical equipment, such as unshielded power lines, switching equipment, fluorescent lights, and network cables (27,28). Such artifacts can be eliminated using an appropriate band-pass filter and full-wave rectification without losing valuable

information on the motor unit response to vibration in SEMG data (29). In this study, after band-pass filtering at 60–1000 Hz, the SEMG signal was rectified (Figure).

Root-mean-square values (RMS) were calculated from the filtered SEMG signal. RMS amplitude, calculated from the SEMG data recorded during WBV, was defined as “vibration-induced RMS”. RMS amplitude calculated from the SEMG data recorded in the resting standing position was defined as “RMS in resting standing”.

Fast Fourier transform (FFT) analysis was used to determine the frequency components of the recorded signals. Power spectral analyses were conducted using Hanning windows. The FFT length was set to 2048 points. Because a peak at the vibration frequency (40 Hz) during WBV was clearly visible in SEMG signal spectrograms, the amplitude of this peak, observed during WBV, was defined as “vibration-induced peak amplitude”. An amplitude at 40 Hz was also calculated from the SEMG data recorded in lying and resting standing positions. The amplitude at 40 Hz calculated from the SEMG data recorded in the resting standing position was defined as “amplitude in resting standing”.

To standardize the changes during vibration in myoelectrical activity (RMS and peak amplitude), the following formulae were used:

Standardized vibration-induced RMS (SVI RMS) = vibration-induced RMS / RMS in resting standing,

Standardized vibration-induced peak amplitude (SVI Amp) = vibration-induced peak amplitude / amplitude in resting standing.

SVI myoelectrical activities (SVI RMS and SVI Amp) were accepted as indicators of vibration-induced reflex myoelectrical activity.

2.2.4. SOST levels

In a previous study on healthy young adult women, we determined that the plasma SOST level peaked at the 10th minute after WBV (22). To standardize the change occurring after vibration in the plasma SOST level, the following formula was used:

SVI SOST index = SOST level (10th minute after WBV) / SOST level (before WBV).

The SVI SOST index was accepted as an indicator of the response of osteocytes to WBV. A high plasma SOST level was reported to indicate an increase in osteocyte activity (21). Thus, a high SVI SOST index was accepted as indicating an increase in osteocyte activity.

Blood samples were obtained by inserting an intravenous cannula into the antecubital vein. Blood was collected using EDTA tubes and centrifuged (400 × g, 15 min) within 15 min of collection. Aliquots of plasma were added to microcentrifuge tubes and stored at –80 °C.

Plasma SOST levels were measured in a blinded fashion by the same researcher. Plasma SOST levels were measured

using a Human Sclerostin ELISA Kit (CUSABIO, Wuhan, P.R. China). The minimum detectable concentration of human SOST was typically <0.012 ng mL⁻¹. None of the measured SOST values in our participants was below the limit of detection of this assay. The intraassay precision error was <8%.

2.3. Blinding

All participants were told that the WBV could affect muscle performance. However, participants had no knowledge regarding their BMD or plasma SOST level during the SEMG recordings. All measurements (SEMG recordings, BMD, and plasma SOST levels) were made by independent assessors blinded to other measurements of the participants. Thus, this was a double-blind study because both the researchers and the participants were blinded in terms of the effects of bone on the vibration-induced reflex muscle activity.

2.4. Statistical analyses

All data were analyzed for normality of distribution using the Kolmogorov–Smirnov test. Continuous variables were summarized as mean ± standard deviation or median (minimum–maximum). The Wilcoxon test was used to analyze differences in RMS data between the resting standing position and during WBV. The Wilcoxon test was used to analyze differences in peak amplitude data between the resting standing position and during WBV. Comparisons of the 2 groups in terms of age, age at menopause, and body mass index (BMI) were performed using an unpaired t-test. Comparison of the 2 groups in terms of a percent change in RMS and peak amplitude was performed using a Mann–Whitney U test. The percent change in RMS and peak amplitude for each individual subject was calculated. The percentage of change for RMS was determined by subtracting the RMS in resting standing from the vibration-induced RMS, dividing by the RMS in resting standing, and multiplying by 100. The percentage of change for peak amplitude was determined by subtracting the peak amplitude in resting standing from the vibration-induced peak amplitude, dividing by the peak amplitude in resting standing, and multiplying by 100. SOST levels were analyzed using a 2 × 2 (group (low and normal BMD) × time (pre- and post-WBV)) ANOVA appropriate for multiple dependent variables with repeated measures. The assumptions of the repeated measures ANOVA (normal distribution, homogeneity of variances, sphericity) were confirmed. Differences in the distribution of participants whose plasma SOST level increased or decreased between the groups were analyzed by using the chi-squared test with Yates correction.

To perform multiple linear regression (MLR) analysis, the distributions of data (SVI RMS and SVI Amp) were fitted to a normal distribution using logarithmic (log₁₀) transformation. MLR analysis was performed to detect

independent predictors of SVI RMS and SVI Amp and to determine confounding effects between potentially independent predictors. A step-wise method was used to construct MLR models. A variable was entered into the model if the probability of its score statistic was less than the entry value (0.05), and it was removed if the probability was greater than the removal value (0.1). Multicollinearity was tested with variance inflation factor and condition index; autocorrelation was tested with Durbin-Watson statistics.

$P < 0.05$ was considered to indicate statistical significance. The software package used for data management was PASW Statistics, version 18.

3. Results

The mean ages of the participants were 56.8 ± 6.5 years in the low BMD group and 55.8 ± 4.7 years in the normal BMD group ($P = 0.463$); the mean ages at menopause were 46.7 ± 3.9 years and 48.3 ± 4.5 years ($P = 0.092$); and the mean BMIs were $30.9 \pm 4.1 \text{ kg m}^{-2}$ and $33.9 \pm 4.5 \text{ kg m}^{-2}$ ($P = 0.003$), respectively.

The mean hip BMD and plasma SOST levels of both groups at trial entry are given in Table 2.

The mean plasma SOST levels at the 10th minute after WBV were $1.34 \pm 0.44 \text{ ng mL}^{-1}$ in the low BMD group and $1.09 \pm 0.42 \text{ ng mL}^{-1}$ in the normal BMD group. For plasma SOST levels, no group-by-time interaction was

found ($F(1,78) = 1.636$, $P = 0.205$). The plasma SOST level increased in 19 (51.4%) participants, but decreased in 18 (48.6%) participants in the low BMD group. These rates were 44.2% and 55.8%, respectively, in the normal BMD group. There was no significant difference in the distribution of participants whose plasma SOST level increased or decreased between the groups (continuity correction chi-squared = 0.173, $P = 0.678$).

The mean SVI-SOST index was 1.03 ± 0.24 in the low BMD group and 0.99 ± 0.33 in the normal BMD group.

Mean data for resting myoelectrical activity of the adductor longus muscle, measured in the lying and resting standing position, are given Table 3. The resting myoelectrical activities were significantly higher during WBV compared with the resting standing position in both groups. However, there was no significant difference in percent change in resting myoelectrical activities during WBV between the groups ($P > 0.05$) (Tables 4 and 5).

A peak at the vibration frequency was clearly visible in unprocessed SEMG signal spectrograms. A band-pass filter and rectification proved effective in removing artifacts and revealed a prominent peak at the vibration frequency in the adductor longus muscle in all participants (Figure).

MLR analysis indicated that the SVI-SOST index was an important predictor of the SVI RMS and the SVI Amp in both groups. However, age, age at menopause, and BMI were not significant predictors. As shown in Table 6,

Table 2. Participants' characteristics at trial entry: the left hip bone mineral density (BMD) and plasma SOST level. Data are expressed as mean \pm standard deviation.

	Low BMD Group (n = 37)	Normal BMD Group (n = 43)
Femur neck BMD (g cm^{-2})	0.681 ± 0.043	0.971 ± 0.075
Trochanter BMD (g cm^{-2})	0.584 ± 0.070	0.800 ± 0.088
Total hip BMD (g cm^{-2})	0.770 ± 0.061	1.049 ± 0.090
Plasma SOST level (ng mL^{-1})	1.32 ± 0.38	1.18 ± 0.49

Table 3. Characteristics at trial entry: the resting myoelectrical activity of m. adductor longus. Data are expressed as median (minimum-maximum).

SEMG data	Low BMD group (n = 37)		P-value	Normal BMD group (n = 43)		P-value
	During supine lying	During resting standing		During supine lying	During resting standing	
RMS (μV)	1.17 (0.59–11.20)	2.74 (0.84–9.77)	0.008	1.25 (0.72–7.46)	2.65 (0.74–7.13)	0.001
Amplitude at 40 Hz (μV)	0.04 (0.02–0.38)	0.09 (0.02–0.39)	0.010	0.04 (0.02–0.23)	0.09 (0.02–0.26)	0.001

Table 4. Effects of WBV for myoelectrical activities of m. adductor longus in both groups. Data are expressed as median (minimum–maximum).

SEMG data	Low BMD group (n = 37)		P-value	Normal BMD group (n = 43)		P-value
	During resting standing	During WBV		During resting standing	During WBV	
RMS (μ V)	274.0 (84.0–977.0)	1011.0 (164.0–3088.0)	<0.001	265.0 (74.0–713.0)	1012.0 (224.0–2304.0)	<0.001
Amplitude at 40 Hz (μ V)	9.2 (2.2–38.7)	108.0 (8.5–853.0)	<0.001	8.9 (2.4–26.1)	124.0 (12.1–353.0)	<0.001

Table 5. Comparison of the 2 groups in terms of percent change in RMS and peak amplitude of m. adductor longus during WBV. Data are expressed as median (minimum–maximum).

SEMG data	Low BMD group (n = 37)	Normal BMD group (n = 43)	P-value
RMS (%)	276.1 (2.6–2090.1)	215.1 (54.5–1220.1)	0.696
Amplitude at 40 Hz (%)	1008.8 (31.2–19,904.7)	1022.3 (181.2–5497.2)	0.779

Table 6. Multiple linear regression models for standardized vibration-induced (SVI) myoelectrical activity for each group. Logarithmic (Log) transformation was applied to the dependent variables.

Group	Dependent variable	Independent variable	R ²	F	P	D–W	B	SE	P-value
Low BMD (n = 37)	Log [SVI RMS]	SVI-SOST	0.72	110.7	<0.001	1.6	0.51	0.04	<0.001
	Log [SVI Amp]	SVI-SOST	0.79	156.6	<0.001	2.0	0.98	0.07	<0.001
Normal BMD (n = 43)	Log [SVI RMS]	SVI-SOST	0.75	112.4	<0.001	2.3	0.56	0.05	<0.001
	Log [SVI Amp]	SVI-SOST	0.83	171.6	<0.001	1.9	1.04	0.08	<0.001

D–W: Durbin–Watson coefficient. B: Unstandardized regression coefficient. SE: Standard error.

the coefficients of determination (R²) and the regression coefficients (B) of the low BMD group were the same or similar to those of the normal BMD group. These findings indicated that there were similarities between the groups in terms of the effects of SVI-SOST index on variances of the SVI Amp and SVI RMS.

4. Discussion

Many studies in recent years have aimed to determine the effects of WBV on neuromuscular performance in elderly people (3,30). Motor unit synchronization was found to take place during WBV in the present study, as in previous studies. This synchronization results from vibration-induced reflex muscle activity (4,5). We also report

several novel findings. The first is that osteocytes play a role in vibration-induced reflex myoelectrical activity in postmenopausal women. The second is that the effect of osteocytes on vibration-induced reflex myoelectrical activity in postmenopausal women with normal BMD is similar to the effect of osteocytes on vibration-induced reflex myoelectrical activity in postmenopausal women with low BMD.

Previous studies reported that myoelectrical activity increased during vibration (5,11,15–17,29). In other WBV studies, the squatting or semisquatting positions have been preferred to evaluate effects of vibration on muscular performance in the lower extremities (6,26). Because of voluntary muscle contractions, myoelectrical

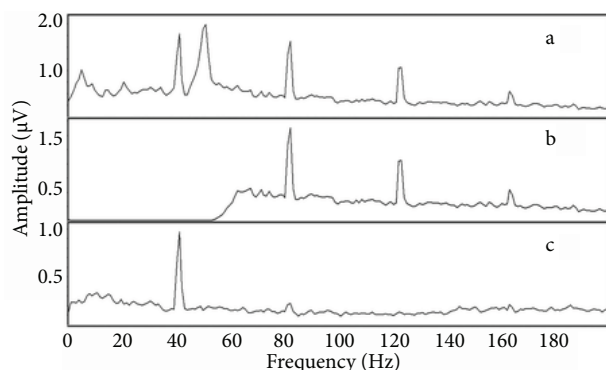


Figure 3. Frequency spectrograms of SEMG signals recorded during WBV. **a)** Unprocessed SEMG signal. **b)** Filtered SEMG signal and elimination of artifacts. **c)** Rectified SEMG signal showing a prominent peak at the vibration frequency (40 Hz) due to synchronization.

activity increases in such positions. However, to assess the reflex effects of osteocytes exposed to vibration on an increase in myoelectrical activity, it is important to avoid voluntary muscle contractions. With that in mind, participants were trained in relaxation and appropriate posture techniques. Participants were asked to stand in an upright posture to minimize exposure to probable movement in lower extremity joints during WBV and to prevent resulting muscle contractions. However, it is difficult to maintain postural balance during WBV. An unstable posture triggers postural control mechanisms, including muscle contractions, to avoid loss of balance during WBV. According to the laws of physics, an object is in balance if its center and line of gravity are above its base of support (31). Thus, 2 precautions were taken to maintain postural stability during WBV: the base of support was adjusted, and participants were asked to hold the front handle of the WBV device. The results of this study showed an increase in myoelectrical activity of the adductor longus muscle without voluntary contractions during WBV. If the increase in myoelectrical activity during WBV was primarily due to postural control mechanisms or voluntary muscle contraction, then the frequency of the SEMG signal would occur randomly throughout the power spectrum (5). However, as shown in the Figure, frequency spectrograms of the SEMG signal recorded during WBV displayed a prominent peak at the vibration frequency in both groups. The emergence of a prominent peak at the vibration frequency was attributed to the increase in motor unit activity synchronized with vibration frequency (5,29).

The “receptor” of BMR is the osteocyte (17). In the present study, the response of osteocytes to vibration stimuli was evaluated by means of plasma SOST levels,

because SOST is produced almost exclusively by osteocytes. Additionally, SOST expression is regulated by mechanical strain in adult bone, and plasma SOST levels increase after WBV (18–20,22). The results of this study showed that the changes in plasma SOST level with the vibration stimulus were an important predictor of the reflex myoelectrical response of the adductor longus muscle to the vibration stimulus in both groups. Positive regression coefficients indicated that as the response by osteocytes to the vibration stimulus increased, so did myoelectrical activity during vibration. Consequently, our data suggest that osteocytes have an effect on vibration-induced reflex muscle activity.

The second finding is that the effect of osteocytes on vibration-induced reflex myoelectrical activity in postmenopausal women with a normal BMD is similar to their effect on vibration-induced reflex myoelectrical activity in postmenopausal women with a low BMD. The coefficients of determination and the regression coefficients of the low BMD group were the same as or similar to those of the normal BMD group. These results suggest similarities between the 2 groups in terms of the effects of the SVI-SOST index on the variance of SVI-myoelectrical activities. More importantly, no significant difference was found in the main effects of WBV on resting myoelectrical activities between the groups. According to our second hypothesis, because the number of osteocytes is reduced during postmenopausal osteoporosis, the effects of osteocytes on vibration-induced reflex myoelectrical activity are weaker in postmenopausal women with osteoporosis than in postmenopausal women without osteoporosis. However, our findings do not support this hypothesis.

Why was there no significant difference between the low and normal BMD groups in terms of the effects of osteocytes on vibration-induced reflex myoelectrical activity? The second hypothesis is based on the following points. The “receptor” of BMR is the osteocyte (17). Numbers of osteocytes decrease during the development of postmenopausal osteoporosis (23,24). According to the WHO criteria, the diagnosis of postmenopausal osteoporosis is based on the T-score of BMD (25). Thus, to test the validity of the second hypothesis, postmenopausal women were categorized on the basis of their T-score for BMD. This method of classification assumes that BMD decreases simultaneously with the number of osteocytes. Although the numbers of osteocytes have been reported to decrease during the development of postmenopausal osteoporosis, no significant difference in the main effects of WBV for plasma SOST level between the groups was found in the present study. This finding indicated that a decrease in the number of osteocytes did not necessarily occur simultaneously with the loss of BMD. If a decrease in the number of osteocytes occurred simultaneously with

the loss of BMD, a significant difference in the main effects of WBV on plasma SOST level between the groups would be expected.

Osteocyte apoptosis plays a central role in signaling the activation and maintenance of bone remodeling. Osteocytes thus play a role in the loss of BMD during osteoporosis. Osteocyte apoptosis may precede the onset of loss of BMD (32–35). In that case, the loss of osteocytes would be more pronounced than the loss of BMD. On the other hand, osteocyte numbers per bone tissue volume could be higher in osteoporotics than in the healthy, because osteoblasts produce less bone matrix per cell (24). In that case, the loss of BMD would be more pronounced than the loss of osteocytes. In short, the changes in the number of osteocytes may not be coherent or parallel the changes in BMD during the postmenopausal period.

Although osteocytes play a crucial role in the loss of BMD, the diagnosis of osteoporosis and the assessment of fracture risk is based on the T-score of BMD in current medical practice (25,32–35). The primary goal of diagnosis and treatment of osteoporosis is to reduce the risk of or prevent atraumatic fractures (25). However, the T-score of BMD is not sufficiently reliable for assessment of fracture risk. Even with T-scores that are normal or close to normal, patients with atraumatic fractures may be seen in medical practice. In contrast, T-scores that are very low may be seen in patients without osteoporotic fractures (24,36). Therefore, reliable assessment of fracture risks cannot be carried out based only on T-scores; new methods, such as FRAX and FRISK scores, should also be considered (37–39). Briefly, classifying postmenopausal women

based on the T-score for BMD alone is not a sufficiently reliable method. Differences between the changes in the numbers of osteocytes and changes in BMD during the postmenopausal period could explain why there was no significant difference between the low and normal BMD groups in terms of the effects of osteocytes on vibration-induced reflex myoelectrical activity.

This study suggests that osteocytes serve as a mechanoreceptor of reflex EMG during WBV. Osteocytes are known to be sensitive to mechanical stimulation and to control the matrix formation process through local and neurological mechanisms in response to such stimulation. Osteocytes subjected to mechanical stimulation are considered to cause changes within only bone tissue (40,41). However, our findings suggest that osteocytes subjected to mechanical stimulation also affect muscle activity. Our findings also show that it is possible to assess in vivo the response of the bone to mechanical loading using electrophysiological methods. This may facilitate development of novel methods of assessing the resistance of bone to mechanical loads and the fracture risk. Further research is needed to investigate this.

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