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**Author Manuscript**

*Int J Obes (Lond)*. Author manuscript; available in PMC 2009 September 1.

## Published in final edited form as:

*Int J Obes (Lond)*. 2008 September ; 32(9): 1348–1354. doi:10.1038/ijo.2008.111.

## **Whole-body vibration slows the acquisition of fat in mature female**

## **rats**

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## **Abstract**

**Objective—**To evaluate the effects of whole-body vibration on fat, bone, leptin and muscle mass.

**Methods/Design—**Thirty 7-month-old female 344 Fischer rats were randomized by weight into three groups (baseline, vibration or control; *n*=7–10 per group). Rats in the vibration group were placed inside individual compartments attached to a Pneu-Vibe vibration platform (Pneumex, Sandpoint, ID, USA) and vibrated at 30–50 Hz (6mm peak to peak) for 30 min per day, 5 days per week, for 12 weeks. The vibration intervention consisted of six 5-min cycles with a 1-min break between cycles.

**Results—**There were significant body composition differences between the whole-body vibration and the control groups. The whole-body vibration group weighed approximately 10% less (mean  $\pm$ s.d.;  $207 \pm 10$  vs  $222 \pm 15$  g,  $P<0.03$ ) and had less body fat  $(20.8 \pm 3.8$  vs  $26.8 \pm 5.9$  g,  $P<0.05$ ), a lower percentage of body fat  $(10.2 \pm 1.7 \text{ vs } 12 \pm 2.0\% , P < 0.05)$ , and lower serum leptin levels  $(1.06$ ± 0.45 vs 2.27 ± 0.57 ng ml<sup>-1</sup>, *P*<0.01) than the age-matched controls. No differences were observed for total lean mass, bone mineral content (BMC), bone mineral density (BMD), insulin-like growth factor-I (IGF-I) or soleus (SOL) and extensor digitorum longus (EDL) mass or function. Regional high-resolution dual-energy X-ray absoptiometry scans of the lumbar spine (L1-4) revealed that the whole-body vibration group had significantly greater BMC (0.33  $\pm$  0.05 vs 0.26  $\pm$  0.03 g, *P*<0.01) and BMD  $(0.21 \pm 0.01 \text{ vs } 0.19 \pm 0.01 \text{ gcm}^{-2}, P<0.01)$  than the control group. No differences between the groups were observed in the amount of food consumed.

**Conclusion—**These findings show that whole-body vibration reduced body fat accumulation and serum leptin without affecting whole body BMC, BMD or lean mass. However, the increase in vertebral BMC and BMD suggests that vibration may have resulted in local increases in bone mass and density. Also, whole-body vibration did not affect muscle function or food consumption.

## **Keywords**

weight gain; fat mass; leptin; lean mass; bone

## **Introduction**

Nearly 66% of Americans are overweight or obese.<sup>1,2</sup> Obesity is an important co-morbidity factor in many common diseases such as heart disease, diabetics and cancer.<sup>3–5</sup> Although weight control is a commonly reported behavior, the prevalence of obesity is increasing in the

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United States<sup>5</sup> in part, because traditional approaches for treating overweight and obese adults, which have focused on individual weight loss have had limited success.  $6,7$ 

Prevention has been suggested as the most important approach to decreasing obesity.<sup>8</sup> Physical activity increases energy metabolism, contributes to a healthy energy balance and can be used to increase lean mass and bone mass while decreasing fat mass.9 Physical activity is also beneficial in attenuating or improving chronic disease conditions.<sup>10</sup> However, despite the proven benefits of physical activity, more than 50% of US adults do not get enough physical activity to provide health benefits; and 24% are not active at all in their leisure time.<sup>2</sup>

Recently, whole-body vibration has been proposed as a potential alternative, or adjuvant, to exercise. Whole-body vibration has been reported to increase energy metabolism through an increased oxygen uptake to values comparable to moderate walking.<sup>11,12</sup> Additionally, standing on a vibration platform at a frequency of 26 Hz and amplitude of 6mm increased heart rate response, Borg rating of perceived exertion and blood lactate concentrations to levels found when performing moderate exercise.<sup>11,12</sup> These data suggest that the increase in energy expenditure with whole-body vibration is of sufficient magnitude to impact body composition. In support of this finding, Rubin *et al.*13 report that short bursts of low-intensity vibration in young mice reduce the differentiation of precursor cells to adipocytes, suggesting a plausible mechanism by which mechanical vibration may prevent fat accumulation.

Although obesity is associated with negative health outcomes, weight loss is also not without risks.<sup>14</sup> Weight loss is associated with bone and muscle loss, potentially increasing the risk for sarcopenia and osteoporosis.<sup>15</sup> Thus, weight reduction may accelerate bone and muscle loss associated with menopause.<sup>16</sup>

Whole-body vibration training has been promoted as a method of improving bone mass  $17-$ 24 and muscle strength.24,25 However, to our knowledge, the long-term effect of vibration on body composition has not been investigated. Thus, the purpose of this study was to determine whether whole-body vibration could promote weight loss while maintaining or improving bone and muscle mass in mature rats.

## **Methods**

This study was approved by the Institutional Animal Care and Use Committee at the Oregon State University.

#### **Animals**

Seven-month-old virgin female Fisher 344 rats (Harlan Laboratories, Indianapolis, IN, USA) were used. Each animal was housed separately in a translucent polycarbonate cage with enclosed filters on the sides and tops of the cages. The colony room was maintained on a 14/10 h light–dark cycle, with temperature maintained at  $21-23$  °C. The attending veterinarian inspected the animals once a day for overall health. The author (GFM) also inspected the animals once a day and noted their health and any signs of distress; including changes in daily cleaning behavior, food and water consumption, activity level, shedding and porphyrin secretion around the eyes and nose.

#### **Food and diet**

The animals had free access to water and a standard commercial diet (Teklad Rodent Chow, no. 8604 Harlan Teklad, Madison, WI, USA) containing 1.46% calcium, 0.99% phosphorus and 4.96 IU per gram of vitamin  $D_3$ . Food intake was determined over weeks  $3-4$ ,  $7-8$  and 11–12.

#### **Experimental design**

The animals were randomized by weight into one of three groups (8–10 per group): (a) baseline, (b) vibration or (c) control. The baseline group was sacrificed at initiation of the study for analysis of body composition, bone mass and muscle function. For vibration treatment, animals in the vibration group were placed in individual  $13 \times 28 \times 15$ cm compartments attached to a Pneu-Vibe vibration platform (Pneumex, Sandpoint, ID, USA). This group received 30 min of whole-body vibration each day, 5 days per week, for 12 weeks.

The vibration intervention consisted of six 5-min cycles at an amplitude of 6mm. Each minute of vibration consisted of 35 s at 30 Hz, followed by 20 s at 40 Hz, followed by 5 s at 50 Hz. The cycle was repeated. After 5 min of vibration, the animals were given a 1-min rest period as recent data suggest that high-frequency loading regimes in combination with the insertion of rest periods may augment osteogenesis.26 The non-vibration animals remained in their cages, which were placed approximately 4m from the vibration platform.

#### **Whole-body scans**

Whole-body bone mineral content (BMC), whole-body bone mineral density (BMD), lean and fat mass, percentage of body fat and femoral and lumbar spine (L1-L4) BMC and BMD were determined by dual-energy X-ray absorptiometry using small animal software (Model 4500- A, Hologic Inc., Waltham, MA, USA). The rats were anesthetized with 2–3% isoflurane delivered in 100% oxygen (1.5 l min−<sup>1</sup> ) via a nose cone during the scanning procedure. The least significant changes for whole body and spine BMD for rats in our laboratory are 0.003 and 0.006 gcm<sup>-2</sup> and less than 1% for whole body composition (lean muscle mass and fat mass) at 95% confidence level, respectively.

#### **Muscle mass and function**

Following the scans, soleus (SOL) and extensor digitorum longus (EDL) muscles were excised from the anesthetized animals. The muscles were mounted between an isometric force transducer and an immovable post via the tendons, and suspended in a temperature-controlled (22 °C) bicarbonate buffer that was continuously bubbled with 95%  $O_2$  and 5%  $CO_2$ .<sup>27</sup> Supramaximal stimuli (200 μs pulses at 60 Hz for SOL and 120 Hz for EDL) were delivered to platinum electrodes flanking the muscle. Data were collected with muscles at their optimal length for tetanic tension. At the conclusion of the experiment, muscle mass was determined and the muscle's physiological cross-sectional area was calculated as described previously.27

#### **Micro-computed tomography**

After the removal of the muscles, the animals were euthanized by exsanguination. The right tibia, femur and lumbar vertebrae (L1-4) were removed, stored in 70% alcohol at 4 °C. Microcomputed tomography was used for non-destructive three-dimensional evaluation of vertebral cancellous bone architecture.<sup>28</sup> The lumbar vertebrae were scanned using a Scanco  $\mu$ CT40 scanner (Scanco Medical AG, Basserdorf, Switzerland) at a voxel size of  $20 \times 20 \times 20$  µm. Analysis of the lumbar vertebra included the entire region of secondary spongiosa between the cranial and caudal growth plates at a threshold of 265. Direct cancellous bone measurements included (1) cancellous bone volume/tissue volume (volume of total tissue occupied by cancellous bone,), (2) trabecular thickness (mean thickness of individual trabeculae, μm), (3) trabecular number (number of trabeculae within the sampling site, 1mm−<sup>1</sup> ) and (4) trabecular separation (the distance between trabeculae, μm).

#### **Serum Leptin and IGF-I**

Serum leptin was measured using a DSL-10-23100 ACTIVE Human Leptin Enzyme-Linked Immunosorbent (ELISA) kit according to the manufacturer's instructions (Diagnostic Systems

Laboratories, INC. Webster, TX, USA). Serum insulin-like growth factor-I (IGF-I) was measured using a rat IGF-I radioimmunoassay kit according to the manufacturer's instructions (Diagnostics Systems Laboratories, Inc., Webster, TX, USA).

#### **Statistical analyses**

Data are expressed as mean  $\pm$  s.d. One-way analysis of variance (ANOVA) was used to evaluate the effects of treatment (baseline, control, vibration) on total body mass, lean and fat mass, percentage of body fat, BMC, BMD, muscle mass and function, leptin and IGF-1 levels. When the ANOVA was significant, a Bonferroni *post-hoc* test was used to evaluate differences among the three treatment groups. A *t*-test was used to evaluate the effects of treatment (control, vibration) on food consumption, lumbar BMC and BMD, vertebral cancellous bone architecture and serum leptin levels. A *P*-value of less than0.05 was considered significant for all effects.

## **Results**

The rats adjusted quickly to the vibration, were healthy, and tolerated the vibration well. Subsequent to the first two vibration sessions the animals exhibited no signs of distress as evidenced by normal condition of their tail and fur, minimal urine and feces excretion during vibration and the absence of porphyrin secretion around the animal's eyes. Furthermore, no difference was observed in the amount of food and water consumed between the control and vibration groups (Table 1).

#### **Whole-body composition**

The control and vibrated rats weighted more than the baseline group at the end of the study. (Table 1). However, at termination of the study, the control group weighed more than the vibration group. The control and vibration groups had greater lean mass than the baseline group. Differences in lean mass were not detected between the control and vibration groups (Table 1). The vibration group had less body fat and a lower percentage of body fat than the agematched control and baseline groups (Figure 1).

#### **Bone mass and architecture**

Dual-energy X-ray absorptiometry analysis revealed that the vibration and control groups had greater BMC and BMD than the baseline group. However, no differences were observed between the control and vibration groups for total body BMC or BMD (Table 2). Regional high-resolution dual-energy X-ray absorptiometry scans of the lumbar spine (L1-4) revealed that the vibration group had greater BMC and BMD than the control group.

Micro-computed tomography analysis of cancellous bone in the first lumbar vertebra revealed no differences between the whole-body vibration and control groups in cancellous bone volume/tissue volume, trabecular number, trabecular thickness or trabecular separation (Table 3).

#### **Muscle mass**

The mass of the SOL and the EDL was similar for both vibration and control groups (Tables 3 and 4). In addition, no differences were observed in twitch force or kinetics (twitch contraction time and  $\perp$  relaxation time) or in absolute or specific (per unit muscle cross-sectional area) tetanic force.

#### **IGF-I**

No differences in serum levels of IGF-I were observed between the vibration and non-vibration groups. However, serum levels of IGF-I were lower in both the vibration and non-vibration groups compared with the baseline group (Table 1).

#### **Leptin**

Serum leptin levels were lower in the vibration group compared with the age-matched control group (Figure 2). Serum leptin levels in the baseline group were not evaluated.

## **Discussion**

Over the course of the study the vibrated rats were healthy, tolerated the vibration well and exhibited no obvious signs of distress following adaptation to vibration. Twelve weeks of whole-body vibration resulted in significant body composition differences between the vibration and control groups. The whole-body vibration group weighed less, had less body fat and had a lower overall percentage of body fat than the age-matched non-vibration group. In addition, the vibrated rats had lower serum leptin levels than the control animals. However, differences in food intake were not observed between the two groups.

Serum leptin concentrations are strongly correlated with body fat and body mass index (BMI).  $29$  Leptin acts on peripheral tissue and increases the inflammatory response by stimulating the production of tumor necrosis factor-α, interleukin-6, and interleukin-12. As a consequence, elevated leptin levels are considered to be an independent risk factor for increased cardiovascular disease, prostate cancer and breast cancer.  $30,31$  Thus, the observed large reduction in serum leptin levels suggests that whole-body vibration may have beneficial effects on disease risk that are disproportionately large compared with the relatively small reduction in body weight.

Epidemiologic, cross-sectional and prospective correlation studies suggest an essential role for physical activity in weight-loss maintenance, and *post-hoc* analysis of prospective trials shows a clear dose–response relationship between physical activity and weight maintenance.5 The mechanism responsible for the reduced fat accumulation in the vibration group is not known. Whole-body vibration has been proposed to increase energy expenditure via repetitive muscle contractions.5 Whole-body vibration has been reported to upregulate energy metabolism through an increased oxygen uptake comparable to moderate walking  $11,12$  and corresponding increases in heart rate response, rating of perceived exertion and blood lactate levels expected when performing moderate exercise.<sup>11,12</sup> Thus, increased energy expenditure without a concomitant increase in food consumption could explain the reduced weight gain in vibrated rats.

Previous whole-body vibration studies showing effects on bone mass and muscle strength have used frequencies ranging from 20 to 90 Hz.25,32–34 Recently, Rubin *et al*. 13 demonstrated that growing mice receiving 15 weeks of high-frequency, low-magnitude mechanical vibration at 90 Hz, 0.2*-g* peak acceleration developed far fewer fat cells than controls not receiving the treatment. Similar to our study, Rubin *et al*. reported no difference in food intake between the two groups of animals. This suggests that vibration does not affect appetite in spite of decreased leptin levels and that an increase in energy expenditure may attenuate or decrease fat cell accumulation. Moreover, at the termination of the Rubin *et al*. study,13 the vibrated mice had nearly 28% less fat in the torso than control animals. In addition, levels of fatty compounds linked to type 2 diabetes, such as triglycerides and free fatty acids, were reduced by 43 and 39%, respectively, in the livers of the vibrated mice.<sup>13</sup> The present study extends this work by showing that vibration also reduces fat accumulation in mature rats fed a normal diet.

Whole-body-vibration has been reported to increase bone mass in disabled children,  $34$  young women with low bone mass  $32$  and postmenopausal women.  $33$  Thus, whole-body-vibration may potentially increase bone mass similar to increases associated with weight gain by mimicking the increased skeletal loading associated with increased weight. We did not detect an increase in total body bone mass in this study. However, the increase in vertebral BMC and BMD suggests that vibration may have resulted in local increases in bone mass and density. Importantly, our findings indicate that prevention of weight gain by vibration had no negative impact on muscle or bone mass.

Vibration reduced fat mass but not lean mass. Even though the whole-body vibration group weighed less, had less fat mass and a lower percentage of body fat, no differences were observed for lean mass and bone mass between the control and whole-body vibration group. Some studies have suggested that up to 30% of weight loss because of energy restriction may be due to a reduction of lean mass.  $36$  Thus, it is possible that a weight reduction program supplemented with whole-body vibration may positively affect body composition by increasing the rate of fat loss while preserving lean mass. Further research is needed to test this possibility.

No differences in lean mass between the whole-body vibration and control groups are consistent with our finding that vibration had no effect on the mass or function of the oxidative SOL and glycolytic EDL muscles. There was a tendency for EDL mass and tetanic force to be reduced by 5–6% in the vibration group. Although our statistical power for this comparison was low (0.42–0.45), these findings deserve further investigation because whole-body vibration has been proposed to augment strength gains during human resistance training exercise programs.24,25

Previous whole-body vibration studies  $37,38$  have reported acute increases in plasma concentrations of growth hormone and serum IGF-I. In our study, whole-body vibration had no effect on IGF-I levels. However, both the whole-body vibration and control groups had lower IGF-I levels than the baseline group. This is consistent with previous research showing serum IGF-I levels to decline with age in a predictable manner in healthy individuals. After reaching a peak during the pubertal growth spurt, mean circulating IGF-I levels decrease twofold by the third decade and continue to steadily decline with age.<sup>39</sup> As our treatment groups were 12 weeks older than the baseline group, we attribute the lower IGF-I levels in the older rats to the natural effects of aging.

In conclusion, these studies show that whole-body vibration reduced age-related increases in fat mass in mature rats. This effect occurred without changes in lean body mass, bone mass or muscle mass and function and without alterations in food intake. These findings suggest that whole-body vibration exercise suppresses the accumulation of fat in young adult rats. Further studies are necessary to determine whether whole-body vibration can prevent fat accumulation in humans. If successful, this research could have implications in cases such as preventing childhood obesity and other metabolic disorders associated with weight gain.

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#### **Figure 1.**

Whole-body vibration reduced total body fat (a) and percent body fat (b). Values are mean  $\pm$ s.d. for total body fat (**a**) and percent body fat (**b**), the vibration group had less fat mass and percent body fat than the baseline and control group. \**P*<0.05.



#### **Figure 2.**

Effects of whole-body vibration on serum leptin. Values are mean  $\pm$  s.d. The vibration group had lower serum leptin levels than the control group.  $*P \leq 0.01$ .

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#### **Table 1**

#### Food consumption, total body mass, total body lean mass and serum IGF-I levels



Abbreviation: IGF-I, insulin-like growth factor-I. Groups sharing a common superscript do not differ significantly from one another (*P*>0.05). Values are mean  $\pm$  s.d.

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## Total body and lumbar spine (L1-4) bone mineral content (BMC) and bone mineral density (BMD)



Abbreviations: BMC, bone mineral content; BMD, bone mineral density. Groups sharing a common superscript do not differ significantly from one another ( $P > 0.05$ ). Values are mean  $\pm$  s.d.

#### **Table 3**

## Cancellous bone volume and architecture in the first lumbar vertebra as determined by microcomputed tomography



#### **Table 4**

## Muscle weight and isometric contractile properties



Abbreviation: CSA cross-sectional area. Values are mean  $\pm$  s.d.