Effects of Whole Body Vibration Treatment on Tibial Bone of Ovariectomized Rats Assessed by In Vivo Micro-CT

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Introduction: Whole-body vibration (WBV) treatment has shown to stimulate bone formation rates in rats and mice and may serve as a potential treatment for osteoporosis [1,2]. However, while bone formation rates have shown to respond markedly, the reported effects of WBV on trabecular bone microstructure and bone mass are small and often non-significant. Due to the cross-sectional design of these studies, detection of small differences is limited and it is presently unclear if changes in bone microstructure over time indeed occur. With recently introduced in vivo micro-CT techniques, longitudinal assessment of bone structure in vivo in small animals has become possible. Potentially, this technique could lead to a more sensitive detection of structural changes. Therefore, our goal was to assess the effects of WBV on the microstructure of ovariectomized rats over time using such in vivo micro-CT equipment.

Materials and Methods: Fourteen 6-month old virgin Wistar rats were ovariectomized and divided into a WBV (n=7) and ovariectomy (OVX) (n=7) group. Rats were left untreated for 8 weeks to develop osteopenia. Then, the WBV group was placed twice a day on a vertical vibration platform for 20 minutes, five days a week for six weeks at 90 Hz and 0.3g. Follow-up in vivo CT-scans were made at week 0, 8, 10, 12 and 14 using an in vivo micro-CT scanner (vivaCT 40, Scanco Medical AG, Bassersdorf, Switzerland). A 6 mm micro CT-scan with an isotropic resolution of 15 microns was made of the proximal tibia and a 3.15 mm micro CT-scan of the diaphysis was made with an isotropic resolution of 30 microns. Rats were sacrificed at 14 weeks by cervical dislocation. Image processing of the metaphyseal and epiphyseal trabecular bone included Gaussian filtering and segmentation. The same filtering and segmentation values were used for every measurement of each animal. All follow-up images were registered to the baseline image to ensure equal orientation. From every baseline and follow-up CT-scan, the metaphyseal and epiphyseal trabecular bone was manually selected and bone structural parameters (bone volume fraction (BV/TV), connectivity density (Conn.D), structure model index (SMI), trabecular number, thickness and separation (Tb.N, Tb.Th, Tb.Sp)) were automatically determined. From the diaphysis, cortical thickness was determined for all measurements. An ANOVA with repeated measures was used to determine differences between the groups at each time point using the previous time point as reference.

Results: Metaphysis: At week 8, both groups displayed loss of BV/TV, Conn.D, Tb.N and Tb.Th and an increase in SMI and Tb.Sp, indicating the development of osteopenia (figure 1). Between 8 and 12 weeks, no significant differences were found between the groups. Between 12 and 14 weeks, BV/TV, Tb.N and Tb.Sp were significantly more favorable in the WBV group, but differences were very small (see figure 2a for zoomed BV/TV graph). Interestingly, a student t-test at 14 weeks showed no significant differences at all, indicating the increased sensitivity to determine significant differences as a result of longitudinal measurements.

Epiphyseal: At week 8, both groups displayed loss of BV/TV, Conn.D and Tb.N and an increase in SMI and Tb.Sp, indicating the development of osteopenia (figure 3). Changes were smaller than in the metaphysis and Tb.Th was not significantly decreased. After eight weeks, Tb.Th linearly increased in the WBV group, while it followed a nonlinear path in the OVX group, ending at similar values. Between week 12 and 14 BV/TV and SMI significantly became more favorable in the WBV group, although this effect was very small.

Diaphysis: Cortical thickness increased slightly after ovariectomy (figure 2b). No significant effect of WHB was found on cortical thickness over time.

Discussion: In this study, effects of daily WBV treatment on tibial bone of OVX rats were analyzed longitudinally. For the first time, effects of OVX on the trabecular bone in the epiphysis were determined longitudinally and were found to be smaller than in the metaphysis. Effects of WBV on bone microstructure and mass were very small and did not take place until after four weeks of treatment. Differences were so small that they could be detected only with the repeated measures statistical method and not by student t-tests. Effects in metaphysis and epiphysis were similar. Given the minor differences between both groups, we conclude that WBV does not lead to substantial improvement of the osteoporotic bone structure in the tibiae of rats after 6 weeks of treatment. The results suggest, however, that longer periods of WBV treatment could lead to a more pronounced response in the trabecular bone.


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Figure 1: Average percentage change in structural parameter and upper standard deviation at all time points in the metaphyseal, proximal tibia.

Figure 2: (left) Zoomed graph of BV/TV in the metaphyseal, proximal tibia between week 12 and 14, (right) Average percentage change in cortical thickness and upper standard deviation at all time points.

Figure 3: Average percentage change in structural parameter and upper standard deviation at all time points in the epiphyseal, proximal tibia.