Effect of Treadmill Exercise on Bone Mass in Female Rats

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Abstract: Increasing peak bone mass at skeletal maturity, minimizing bone loss during middle age and after menopause, and increasing bone mass and preventing falls in advanced age are important measures for preventing osteoporotic fractures in women. Exercise has generally been considered to have a positive influence on bone health. This paper reviews the effects of treadmill exercise on bone in young, adult, ovariectomized, and osteopenic female rats. Treadmill exercise increases cortical and cancellous bone mass of the tibia as a result of increased bone formation and decreased bone resorption in young and adult rats. The increase in lumbar bone mass seems to be more significant when long-term exercise is applied. Treadmill exercise prevents cancellous bone loss at the tibia as a result of suppressed bone resorption in ovariectomized rats, and increases bone mass of the tibia and mechanical strength of the femur, as a result of suppressed bone resorption and increased bone formation in osteopenic rats after ovariectomy. Treadmill exercise transiently decreases the serum calcium level as a result of accumulation of calcium in bone, resulting in an increase in serum 1,25-dihydroxyvitamin D3 level and a decrease in serum parathyroid hormone level. We conclude that treadmill exercise may be useful to increase bone mass in young and adult rats, prevent bone loss in ovariectomized rats, and increase bone mass and bone strength in osteopenic rats, especially in the long bones at weight-bearing sites. Treadmill exercise may have a positive effect on the skeleton in young, and adult, ovariectomized, and osteopenic female rats.

Key words: female rat, mechanical loading, treadmill running exercise

Introduction

Osteoporosis is one of the most serious health problems in elderly women; osteoporotic fractures are much more common in elderly women than in elderly men. Current strategies for the prevention of osteoporotic fractures in women focus on increasing peak bone mass at skeletal maturity, minimizing bone loss during middle-age and after menopause, and normalizing bone turnover, increasing bone mass, and preventing falls in advanced age. Exercise has generally been considered to have a positive influence on the skeleton and subsequently to be useful in the prevention or treatment of osteoporosis.

Rats have been used to predict the skeletal efficacy of some interventions including exercise in humans. In
the opinion of Frost and Jee [7], rats can model human skeletal status. In detail, the same mechanisms control gains in bone mass (longitudinal growth and modeling drifts) and losses (Basic Multicellular Units [BMU]-based remodeling), in young and aged rats and humans. In young rats, the same biologic mechanisms increase bone mass as in children. Longitudinal growth adds spongiosa and length to cortical bone. Modeling drifts add cortical bone and can thicken trabeculae. BMU-based remodeling can either conserve or remove bone touching marrow, but does not add significantly to it. Longitudinal growth becomes trivial or stops in adult rats. In aged rats, the same biological mechanisms control conservation of bone as in human adults: BMU-based remodeling. It causes the same kinds and pattern of bone losses next to marrow as in humans. It can either conserve bone or accelerate its removal.

Gains in bone mass (longitudinal growth and modeling drifts) and losses (BMU-based remodeling) may respond to exercise in terms of increased mechanical loading in young and aged rats and human. However, because the cortical bones of rats lack the Haversian based remodeling observed for human cortical bone, the efficacy of exercise in rats may not translate in every detail to human.

Running on the treadmill is one form of exercise. In research on osteoporosis treatment, treadmill exercise has been utilized to test the effects of exercise on bone mass, bone metabolism, and bone strength in rats. When the rat, a tetrapedal animal, is running on a treadmill, there is greater mechanical loading on the appendicular bones than on the axial bones. Namely, the tibia and femur of rats receive much more mechanical loading than the lumbar spine during running on the treadmill. On the other hand, when a bipedal human is running, both the axial bones and the appendicular bones in the lower extremities receive mechanical loading. Thus, the effects of running exercise on the lumbar spine in rats cannot be applicable to human.

It has been documented that weight-bearing bones like the tibia and femur have higher sensitivity to treadmill exercise than less weight-bearing bones like the lumbar spine in rats [10, 12, 14, 27], and the efficacy of treadmill exercise in terms of weight-bearing activity of long bones has been demonstrated in a variety of rat models, i.e., young female rats (Iwamoto et al. [14]), young male rats (Bourrin et al. [4]), ovariectomized rats (Barengolts et al. [2], Yeh et al. [28]), adult female rats (Yeh et al. [16]), rats with kidney disease (Darnley et al. [6]), heavy ion particle irradiated rats (Fukuda et al. [8]), rats taking caffeine (Huang et al. [9]), rats with bone loss caused by immobilization (Kannus et al. [15]), rats consuming alcohol (Reed et al. [18]), orchidectomized rats (Tuukkanen et al. [19]), and rats discontinuing parathyroid hormone (PTH) administration (Yamamoto et al. [20]). In this paper, we review the effects of treadmill exercise on bone mass, bone metabolism, and/or bone strength of the appendicular long bones and axial bones in young, adult, ovariectomized, and osteopenic female rats.

**Effect of Treadmill Exercise on Bone Mass**

*Young rats*

Yeh et al. [25] showed that treadmill exercise (20 m/min, 1 h/day, 5 days/week for 6 weeks) in 6-week-old Sprague-Dawley (SD) rats initially increased and then decreased bone resorption and continuously increased bone formation, resulting in an increase in periosteal bone formation and, subsequently, an increase in cortical bone area of the tibia. Iwamoto et al. [12] demonstrated that treadmill exercise (24 m/min, 1 h/day, 5 days/week for 12 weeks) in 4-week-old SD rats increased cancellous bone volume/total tissue volume (BV/TV) in the proximal and distal tibia, but not in the lumbar vertebrae, and that the response of cancellous bone to treadmill exercise was greater in the distal tibia than in the proximal tibia.

The mechanism for this greater response of cancellous BV/TV in the distal tibia than in the proximal tibia remains uncertain; however, it may be attributable to the location and diameter of the bone tissue to some extent. The distal tibia is likely to receive more mechanical loading than the proximal tibia during treadmill exercise, because the distal tibia is situated farther away from the body of the rat. This distal location increases the total load on the distal tibia as compared with the proximal tibia. Additionally, the diameter of the distal tibia is smaller than that of the proximal tibia, and therefore, the distal tibia supports greater weight per surface area during treadmill exercise. These findings suggest that high strain magnitude may be more effective than low strain magnitude in producing a positive effect on cancellous BV/TV. Because treadmill exercise did not
affect cancellous BV/TV and bone formation in the lumbar spine, despite the increased cortical bone formation, it has been generally accepted that cortical bone could be more sensitive to treadmill exercise than cancellous bone.

Iwamoto et al. [14] reported that treadmill exercise (25 m/min, 1 h/day, 5 days/week for 11 weeks) in 6-week-old Wistar rats increased the tibial bone mineral content (BMC) (measured by dual-energy X-ray absorptiometry [DXA]) and length of the femur, but did not increase the lumbar vertebral BMC. Treadmill exercise stimulated longitudinal bone growth and increased BMC at weight-bearing sites, but did not alter bone mineral density (BMD) (measured by DXA). These findings suggest that treadmill exercise during the growth period increases the size of long bones in the longitudinal and/or radial direction, maintaining bone density, and that weight-bearing activity is important to increase the bone mineral content of long bones by short-term treadmill exercise in young rats.

With regard to the effect of treadmill exercise on calcitropic hormones, it is accepted that exercise promotes a positive calcium balance and increases skeletal mass as indicated by total body calcium largely as a result of an increase in serum 1,25-dihydroxyvitamin D3 level and enhancement of intestinal calcium absorption in rats (exercise program: 25 m/min, 1 h/day, 5 days/week for 13 weeks) [22]. Yeh et al. [23] and Iwamoto et al. [14] suggested that treadmill exercise (Yeh et al., 25 m/min, 1 h/day, 5 days/week for 6 weeks; Iwamoto et al., 25 m/min, 1 h/day, 5 days/week for 11 weeks) in 6-week-old SD and Wistar, respectively rats stimulated bone formation, resulting in an increased demand for minerals that was satisfied by an increase in serum 1,25-dihydroxyvitamin D3 level and increased intestinal absorption of calcium, and that the increase in calcium absorption suppressed the serum PTH level.

Mosekilde et al. [16] showed that extremely long-term treadmill exercise (2 km running/day, 5 days/week, for 10 months) in 2-month-old Fischer rats had an anabolic effect on the skeleton, i.e., cross-sectional area and mechanical strength of the lumbar vertebral body (dominated by cancellous bone), and femoral diaphys- ial cross-sectional area (cortical bone), during attainment of peak bone mass and during the consolidation phase. These results suggest that the anabolic effects on the lumbar vertebrae seem to be more significant when long-term exercise is applied. One possible explanation for these positive effects on the lumbar vertebrae may be the systemic actions of calcitropic hormones that act more slowly on bones than increased mechanical loading.

Thus, treadmill exercise is considered to be useful to increase cortical bone area and cancellous BV/TV of the tibia, mainly by increasing bone formation in young female rats. However, Iwamoto et al. [13] demonstrated that deconditioning (stopping exercise) for 4 weeks in 4-week-old treadmill-exercise-trained SD rats (exercise program: 24 m/min, 1 h/day, 5 days/week for 8 weeks) resulted in decreases in cortical bone area and cancellous BV/TV of the tibia and bone formation parameters to levels not significantly different from those in rats without treadmill exercise. Namely, treadmill-exercise-trained young rats, when deconditioned, lost the benefits gained through exercise. Yeh et al. [24] similarly demonstrated that ash weight and calcium content of the femur gained through treadmill exercise (20 m/min, 1 h/day, 5 days/week for 8 weeks) was lost during 4 weeks of deconditioning as a result of a decline in bone formation and an increase in bone resorption in 12-week-old SD rats. Thus, continued exercise is required to maintain bone mass gained through exercise.

Adult rats

Yeh et al. [26] showed that treadmill exercise (17 m/min, 1 h/day, 5 days/week for 16 weeks) in 14-month-old SD rats increased cancellous BV/TV of the tibia as a result of increased bone formation and suppressed bone resorption, and inhibited age-related loss of cancellous BV/TV of the lumbar vertebrae as a result of suppressed bone resorption. Chen et al. [5] showed that treadmill exercise (17 m/min, 1 h/day, 5 days/week for 16 weeks) in 14-month-old SD rats induced accumulation of cortical bone of the tibia as a result of stimulated peristomal bone formation and suppressed endocortical bone resorption. These two studies also demonstrated that cortical bone was more sensitive than cancellous bone to stimulation by mechanical loading. Yeh et al. [27] showed that treadmill exercise (17 m/min, 1 h/day, 5 days/week for 16 weeks) in 14-month-old SD rats increased BMD (measured by DXA) of the tibia and lumbar spine, with a less significant increase in the lumbar spine. They suggested that the response
of bone to mechanical loading differed between not only the position of the direct weight-bearing site or locomotion without weight-bearing, but also the type of bone tissue (cancellous or cortical). The benefit of exercise may not be limited to adult rats.

Yeh et al. [30] showed that low-dose growth hormone administration in 14-month-old SD rats initially potentiated the response of cortical bone formation of the tibia to treadmill exercise (17 m/min, 1 h/day, 5 days/week for 9 weeks), but long-term administration (16 weeks) of low-dose growth hormone did not enhance the effect of exercise on cortical bone area. They also demonstrated the importance of insulin-like growth factor (IGF)-I in skeletal formation by using systemic treatment with low-dose growth hormone and local stimulation by treadmill exercise (17 m/min, 1 h/day, 5 days/week for 9 weeks) in 14-month-old SD rats [31]. Both growth hormone administration and treadmill exercise increased the circulating level of IGF-I, but only exercise resulted in an increase in long bone IGF-I, which was associated with greater bone formation as indicated by increased serum osteocalcin level and bone formation rate. Growth hormone administration to 14-month-old SD rats did not enhance the treadmill-exercise-induced increase in IGF-I concentration in the serum or skeleton.

Conversely, Oxlund et al. [17] showed that high-dose growth hormone administration to 18-month-old Wistar rats increased bone formation rate and cross-sectional area of cortical bone of the femur with an increase in serum IGF-I concentration, while mild treadmill exercise (8 m/min, 1 h/day, 5 days/week for 73 days) did not influence bone formation rate of cortical bone of the femur. However, high-dose growth hormone administration and mild treadmill exercise in combination modulated and further increased the formation and strength of cortical bone of the femur.

Because both exercise and growth hormone might have an effect on both bone formation and resorption in cortical bone [31], the dose of growth hormone and the intensity of exercise could affect their effect on cortical bone, alone or in combination. However, IGF-I may be both a systemic and local mediator of exercise-induced bone formation in growth hormone-treated rats.

Ovariectomized adult rats

It is established that ovariectomy increases bone resorption and formation, resulting in marked cancellous bone loss in rats. Ovariectomy-induced cortical bone loss is more significant in adult rats than in young rats [21]. The preventative effects of treadmill exercise on bone in ovariectomized rats have been reported [2, 3]. Barengolts et al. [2, 3] showed that treadmill exercise (21 m/min, 40 min or 1 h/day, 4 or 5 days/week for 3 months) in 9-month-old ovariectomized SD rats prevented the loss of ash weight of the tibia and femur, but not the lumbar vertebrae, and increased the mechanical properties of the femur, and also prevented loss of cancellous BV/TV of the tibia as a result of suppressed bone resorption and stimulated bone formation. These results suggest that treadmill exercise has beneficial effects on bone mass as well as mechanical properties at weight-bearing sites during the early stage after ovariectomy.

Osteopenic adult rats

The therapeutic effects of treadmill exercise on bone in osteopenic rats produced by feeding for 2 to 3 months after ovariectomy have been reported [1, 10, 28, 29]. Barengolts et al. [1] examined the effects of treadmill exercise (21 m/min, 40 min/day, 1 day/week for 3 months and 21 m/min, 50 min/day, 4 days/week for 3 months) on bone mass in 12-month-old osteopenic SD rats caused by feeding for 3 months after ovariectomy. They showed that treadmill exercise prevented the loss of ash weight of the tibia, but not the lumbar vertebrae, and the deterioration of bone strength of the femur.

Yeh et al. [28] examined the effects of treadmill exercise (18 m/min, 45 min/day, 5 days/week for 4 months) and 17\(\beta\)-estradiol (E\(_2\)) replacement on tibial bone mass in 12-month-old osteopenic SD rats produced by feeding for 2 months after ovariectomy. They showed that E\(_2\) replacement attenuated the loss of cancellous BV/TV of the tibia as a result of suppressed bone formation and resorption, while treadmill exercise attenuated the loss of cancellous BV/TV of the tibia and prevented the loss of calcium content of cortical bone of the tibia as a result of suppressed bone resorption and sustained bone formation. The effects of E\(_2\) replacement and exercise were independent and additive. These results suggest that E\(_2\) replacement primarily influences cancellous bone of the tibia, while positive
adaptation to exercise occurs in both cancellous and cortical bone of the tibia. Yeh et al. [29] also showed that the combined intervention of E2 replacement and treadmill exercise (18 m/min, 45 min/day, 5 days/week for 7 weeks) in 12-month-old osteopenic SD rats produced by feeding for 2 months after ovariectomy could prevent the development of osteopenia; while the bone-conserving effect of E2 replacement was primarily on the lumbar vertebrae, exercise primarily affected long bones such as the femur. Both interventions had an additive effect on bone mass.

Iwamoto et al. [10] examined the effects of quantitative application of treadmill exercise with calcium supplementation (1.4% calcium diet vs. 0.5% calcium diet) on BMD (measured by DXA) of the tibia and lumbar spine in 37-week-old osteopenic Wistar rats produced by feeding a low calcium diet for 14 weeks after ovariectomy. In this rat model, the running speed of 12 m/min corresponded to a moderate level, whereas that of 18 m/min corresponded to a high level, when the onset of blood lactate accumulation was used to determine the intensity of running exercise. They showed that treadmill exercise (12 m/min, 1 h/day, 5 days/week for 12 weeks) increased the mechanical strength of the femur, as a result of suppressed bone resorption and increased bone formation, and that the beneficial effects of treadmill exercise were recognized at weight-bearing sites only when an optimal level of exercise was applied. Treadmill exercise (12 m/min, 1 h/day, 5 days/week for 12 weeks) improved calcium balance and increased BMD of the tibia as a result of an increase in 1,25-dihydroxyvitamin D3 level and a decrease in serum PTH level [11]. High-intensity and long-duration exercise (18 m/min, 1 h/day, 5 days/week for 12 weeks and 12 m/min, 2 h/day, 5 days/week for 12 weeks, respectively) was not required to increase bone density in 37-week-old osteopenic Wistar rats. Treadmill exercise has beneficial effects on established osteopenia in ovariectomized rats.

Conclusions

Treadmill exercise may be useful for increasing bone mass in young and adult rats, for preventing bone loss in ovariectomized rats, and for increasing bone mass and bone strength in osteopenic rats, especially in the appendicular long bones at weight-bearing sites. However, the increase in lumbar bone mass seems to be more significant when long-term exercise is applied. Rats trained with treadmill exercise, when deconditioned, lose the benefits gained through exercise. Thus, continued exercise is needed to maintain bone mass gained through exercise.

References

13. Iwamoto, J., Yeh, J.K., and Aloia, J.F. 2000. Effect of


