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# Skeletal Effects of a Long Term Submaximal Exercise Programme on Standardbred Yearlings

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ABSTRACT. The effects of a 26 week treadmill exercise programme on metacarpal bone quality in Standardbreds was assessed by ultrasound velocity measurement and single photon absorptiometry. Weekly bone measurements began at around 6 months of age and training started when the horses were exactly 12 months old. Exercise intensity and duration gradually increased and consisted of trotting with some $V_{200}$ interval work. Rates of change in bone quality parameters were compared to pre-training and also to rates of change in a control group. Results indicated possible trends towards increasing bone quality associated with exercise. However, the changes were small and often not of statistical significance. It is likely that more intense exercise is required for more demonstrable changes. The results are consistent with studies in other species where the skeletal effects of submaximal exercise have ranged from no detectable change to small increases in bone mass.

*Key words.* Bone density; bone mineral; elastic modulus; horses; photon absorptiometry; ultrasound velocity.

## INTRODUCTION

Over 200 years ago anatomists and surgeons became aware of an apparent relationship between mechanical stress and bone structure. In 1892 the German anatomist Julius Wolff concluded, in what is now referred to as Wolff's Law, that "every change in the function of a bone ... is followed by certain definite changes in their internal architecture, and equally definite secondary alterations in their external conformation, in accordance with mathematical laws".[10]

It has long been known that disuse and immobilisation, due to a variety of factors, result in bone loss. Less is understood about the effects of prolonged training, although it is generally recognised that physical activity can prevent bone atrophy and may lead to bone hypertrophy. Work examining the effects of exercise on bone has been conducted in both humans and animals. In animals, much of the work has involved rats and mice, and the results have varied greatly. This variation probably stems from differences in the duration and intensity of training and different ages of animals used.[2]

Inhibition of bone growth due to strenuous exercise in young animals has been documented for young growing mice,[11] immature rats[23] and young roosters.[15] Despite small increases in bone mass in some animals, reductions in longitudinal and circumferential bone growth were usually observed. Bone hypertrophy, however, has been reported in most animal studies of the effects of exercise on both immature and mature skeletons.[12,19,20]

There have been several long term studies of submaximal exercise in other species. Small increases in bone weight and mineral content were observed in mice that were exercised lightly over a 12 month period.[1] In immature swine it was concluded that prolonged exercise (12 months) had a significant effect on the quantity, but not on the quality of bone.[25] However, another study was unable to demonstrate absolute increases in tibial bone mineral content in beagle dogs exercised on a carousel treadmill for 71 weeks.[14]

Most human studies have examined the hypertrophic effects of exercise in normal adults and the reduction or prevention of involutional bone loss associated either with senescence or the menopause. Many of the studies on normal adults have involved static comparisons between athletes or exercising individuals and an age-matched control population. These have reported increases in bone mass in the exercising individuals.[5,9] In one of the few prospective experimental studies, a significant increase in tibial bone mineral content was demonstrated in infantry recruits over an intensive 14 week training period.[13] Other human studies have concentrated on senile and post-menopausal osteoporosis. These have demonstrated that light to moderate regular exercise can arrest[24] and even reverse[22] bone mineral loss.

There are few data concerning the skeletal effects of training in horses. In a 90 day study of low intensity exercise in yearlings, a small, non-significant increase in bone mineral content was demonstrated using a photometric radiographic technique.[17] Another study used compression testing to measure elastic modulus and ultimate breaking strength in the excised bones of 12 ponies, 6 of which were exercised by trotting 16 km per day for 4 months.[21] Bone samples from exercised ponies tended to be stiffer and have a greater ultimate breaking strength than those from the non-exercised controls.

Since the development of accurate and precise noninvasive techniques for the assessment of bone quality in the horse,[16] the skeletal effects of exercise have been examined more closely. Short term submaximal exercise resulted in very little alteration in bone strength in adult Standardbreds.[7] However, a 12 week maximal exercise programme was shown to produce a significant increase in bone ultrasound velocity, indicative of a reduction in bone porosity.[7]

In summary, experimental evidence in humans and animals, including horses, suggests that three factors are critical in determining the skeletal effects of exercise. These are the duration and intensity of training and the age of animal involved. Low intensity exercise has similar effects in both the immature and mature skeleton. These range from no effect in short duration studies to small increases in bone mass in longer term programmes (e.g. 6 months or more). High intensity exercise produces significant increases in bone mass in mature animals, however it may result in a reduction in longitudinal bone growth in some weight bearing bones of immature animals.

The objective of this study was to expand on previous work on bone development during growth in horses[3] and to investigate the skeletal effects of long term submaximal exercise in young horses using noninvasive assessment techniques.

## MATERIALS AND METHODS

### *Experimental animals*

A group of 6 Standardbred foals (5 colts and 1 filly) was used. Bone measurements began at 5 to 6 months of age. Another group of 5 Standardbreds had been kept under exactly the same conditions in the previous year as part of skeletal maturity investigations.[3] These horses had received no forced exercise. Changes in this group were used as a basis for comparison with changes in the exercised group. The foals were kept in large yards so that, apart from treadmill work which began at 12 months of age, their level of exercise was limited.

### *Measurements*

Techniques for the examination of the exercised foals were identical to those for the skeletal maturity investigations.[3] Weekly measurements of growth rate included girth, bodyweight and mid-metacarpal circumference. Plasma alkaline phosphatase and plasma osteocalcin were measured at weekly intervals to assess bone turnover.[8] In addition, ultrasound velocity measurement, single photon absorptiometry and radiographic photodensitometry were performed according to methods described previously.[8]

The measurement of ultrasound velocity through the third metacarpal bone can give a good indication of bone strength and density. The apparent transverse velocity ($C_a$), is derived by measuring the distance between two ultrasound transducers and the time of flight of the beam through the bone. However, the fastest path for the sound is in fact via the cortex (i.e. around the medullary cavity). This path length can be estimated and $C_a$ can be corrected to estimate the transverse cortical bone ultrasound velocity ($C_b$).[16] A detailed analysis of the ultrasonic trace allows excellent estimates of the medulla to cortex ratio (R) and the cortical cross sectional area (CSA) to be made.[16] Ultrasonic measurements were made each week.

Single photon absorptiometry is a technique developed to measure bone mineral content (BMC) noninvasively.[4] The BMC measurement can be combined with the ultrasonic estimate of cross sectional area so that bone mineral density (BMD) can be calculated (BMD = BMC/CSA). Using a bone model and incorporating values for BMD and $C_b$, the compact bone density or specific gravity (CBD) and a modulus of elasticity (E) can be estimated.[6,8] Measurements of BMC, and subsequent calculations of BMD, CBD and E, were made each month.

Radiographic photodensitometry involves measuring the optical density of bone from a radiograph and comparing it to the optical density of a known thickness of a standard material (aluminium stepwedge). Radiographic photodensitometry is less accurate and precise than single photon absorptiometry, and only estimates the amount of bone mineral present, not bone density. Radiographic photodensitometry was performed monthly.

*Exercise programme*

All 6 foals underwent a 26 week submaximal exercise programme on a high speed treadmill (Beltalong, Euroa, Vic.). The angle of the treadmill was set at 3 degrees. Training began when each foal was exactly 12 months old and the horses trained 3 days per week in Weeks 1 and 2, and thereafter 5 days per week.

During the first four weeks, training progressed from 5 min at 3.5 m $s^{-1}$ on 3 days per week, to 10 min at 4 m $s^{-1}$ on 5 days per week. These speeds constituted a steady trot. During Week 5 an incremental exercise test was performed and a heart rate monitor (PEH 100 Horse Tester, Finland) was used. The format of the test was as follows: 120 s at 3.5 m $s^{-1}$; 75 s at 5 m $s^{-1}$; 60 s at 7, 8, 9 and 10 m $s^{-1}$. From these tests $V_{200}$ and $V_{160}$ values for each foal were calculated using linear regression analysis. Exercise tests were performed in Weeks 10, 15, 20 and 25 to continually update the working value for each foal's $V_{200}$ and to quantify their change in fitness. Speeds of 9 and 10 m $s^{-1}$ were only used when necessary, and the tests generally ended when heart rates reached 210 bpm so that the foals were not unduly fatigued.

From Weeks 5 to 8, trotting exercise was performed on 3 days per week. This began with 12 min at 4 m $s^{-1}$ and progressed to 20 min at 4.5 m $s^{-1}$ by the end of week 8. The foals ran single intervals at their respective $V_{200}$ on 2 days each week, between the trotting days. $V_{200}$ generally constituted a steady canter in these foals. The distance progressed from 400 to 800 m by Week 8. Each interval was preceded and followed by 5 min of trotting. Training progressed under this format until Week 12, by which time each foal completed 20 min at 4.5 m $s^{-1}$ on the trotting days and single $V_{200}$ intervals of 1 200 m on the other two.

The foals began to run two $V_{200}$ intervals in Week 13, and by Week 20 they were completing $2\times1\,000$ m intervals interrupted with a total of 10 min trotting at 4.5 m $s^{-1}$. The trotting days were now 20 min at 5 m $s^{-1}$. This format remained unaltered except that in Week 21 the foals began triple intervals, beginning with $3\times400$ m and ending with 3×800 m in Week 26. Throughout the programme, intervals were run at speeds equivalent to the current $V_{200}$ for each horse.

*Statistical analysis*

Statistical analysis of exercise effects was based on comparison of slopes for all parameters, before and after onset of training (i.e. pre-12 months old and post-12 months old periods). The Student's paired *t*-test was used. This was also performed for the previous year's foals, which had received no forced exercise. The pre-12 months slopes were compared for the two sets of foals, as were the post-12 months slopes, using Student's two sample *t*-test. Finally, the slope changes, the difference between pre- and post-slopes, were compared for the two groups of foals using Student's two sample *t*-test. Results were considered to be statistically significant when $p<0.05$.

## RESULTS

During the exercise programme the foals' muscularity appeared to increase. They became far more tractable and subjective assessment of their increasing capacity for work indicated that athletic ability may have improved. This was perhaps in conflict with the finding that the cardiac response to the standardised exercise test did not improve. When measured for all foals, $V_{200}$ and $V_{160}$ remained unchanged throughout the exercise programme. Mean ($\pm$ SD) values for $V_{200}$ were 7.6 ($\pm$0.9) m s$^{-1}$ at Week 5, 7.6 ($\pm$0.8) m s$^{-1}$ at Week 10, 7.4 ($\pm$0.5) m s$^{-1}$ at Week 15, 7.8 ($\pm$0.7) m s$^{-1}$ at Week 20 and 7.6 ($\pm$0.7) at Week 25. It was not possible to rigidly adhere to the training programme for all horses, and some flexibility was required as minor clinical problems arose. One foal was retired prematurely, due to splints and low grade osteochondrosis, but this was not until the end of Week 23.

The growth rate parameters for which regression lines were calculated were girth, mid-metacarpal circumference and body weight. There were no significant changes in the slopes of the regression lines pre- and post-training for either girth or body weight. There was, however, a reduction in the slope for mid-metacarpal circumference ($p<0.02$). This was also noted for the non-exercised foals. No significant differences were found between the two groups of foals for either the pre-12 month slopes or the post-12 month slopes for any of the three parameters. The slope changes were not significantly different between the two groups.

Plasma alkaline phosphatase did not significantly change with exercise, nor was there any difference in the slope change at 12 months of age between the two groups of foals. There was no difference between the slope changes for the 2 groups for plasma osteocalcin, however there was a significant reduction ($p<0.001$) in the slope for osteocalcin in the exercised group. A similar reduction was found in the non-exercised foals but this was not significant.

Radiographic optical density did not change significantly with exercise, but the increase in slope with exercise was significantly ($p<0.05$) lower than at the corresponding age in the non-exercised foals.

There were no significant changes in the slopes of the regression lines with exercise for any of the following: $C_a$, $C_b$, BMC, CSA, BMD, CBD, E and R (Table 1). This was also the case for pre- and post-12 month slopes for the non-exercised foals, except that there was a significant ($p<0.05$) decrease in R. However, the trends were of interest. Whereas the slopes of the regressions for $C_a$, $C_b$, BMD, CBD, E and R all decreased after 12 months in the non-exercised foals, these slopes all increased slightly in the exercised horses. The slope of the CSA regression increased in the first group, while it decreased in the exercised group. BMC increased at a slightly greater rate in the exercised foals.

Next the slope changes in the exercised foals were compared with the changes in slope at the same period (i.e. 12 months old) in the non-exercised group (Table 2). The slope of $C_a$ increased at this time for the

Table 1. *Comparison of slopes pre- and post-exercise for all bone quality parameters in the exercised group*

Data represents the mean (± SD) of the differences between pre- and post-exercise slopes (Post-Pre) for the 6 foals. The Student's paired *t*-test was used to test the significance of these slope changes

| Parameter | Slope change Mean | SD | *p*-value |
|---|---|---|---|
| $C_a$ | 0.1802 | 0.3017 | 0.20 |
| $C_b$ | 0.3276 | 0.4067 | 0.10 |
| CSA | −0.000534 | 0.001102 | 0.28 |
| R | 0.000021 | 0.000091 | 0.59 |
| BMC | 0.001303 | 0.002232 | 0.21 |
| BMD | 0.000378 | 0.000659 | 0.22 |
| CBD | 0.000244 | 0.000389 | 0.18 |
| E | 0.006299 | 0.010110 | 0.19 |
| Optical density | −0.000859 | 0.002680 | 0.47 |
| Alkaline phosphatase | −0.6011 | 1.1228 | 0.25 |
| Osteocalcin | −0.9732 | 0.3407 | <0.001 |

exercised foals, and it decreased in the other group. This difference was significant at the level $p=0.05$. A similar trend was seen for $C_b$ and was significant at $p<0.05$. The medulla to cortex ratio (R) increased in the exercised foals, but decreased in the non-exercised ones, and this difference was also significant ($p<0.05$). This same trend was observed for BMD, CBD and E, but without significance. The small decrease in CSA with exercise was not significantly different from the small increase in the other foals and there was no change in BMC.

## DISCUSSION

Previous studies in other species have shown that exercise can clearly have significant skeletal effects. These effects are dependent on the age of the animal and the intensity and duration of training.[2] It appears, from this and other experiments,[7,17,21] that exercise intensity is critical to the production of changes in bone quality in the horse.

The cardiovascular effects of exercise were closely monitored in the young Standardbreds. It is generally assumed that as adult animals become fitter, the speed at which they can travel, for a given heart rate, will increase. Arbitrary measures of this were the velocities at which each foal's heart rate reached 200 bpm ($V_{200}$) and 160 bpm ($V_{160}$). $V_{200}$ is the more commonly used parameter, but $V_{160}$ may have been a more appropriate measurement in these foals since the majority of training, except for the intervals, would have been at around this heart rate. The results indicated that $V_{200}$ and $V_{160}$ remained unchanged throughout the exercise programme. This is in accord with previous studies on submaximal exercise in Standardbred yearlings where no change in $V_{200}$ was detected from 12 to 20 months of age.[18] The reason for this is probably twofold. First, as foals grow, their cardiac stroke volume increases and so their cardiac output increases, mainly to compensate for an expanding muscle mass. Secondly, there may be an increase in the efficiency of oxygen utilisation by muscle and consequently an increase in the arteriovenous oxygen difference at a given level of submaximal work.[18] The fact that the foals increased in muscularity and could handle the intervals, suggested that a training effect may have existed.

The skeletal effects of exercise in these young horses were small. There was no significant alteration between pre- and post-exercise slopes for any of the parameters, except for plasma osteocalcin which decreased significantly. Whereas the slopes of the regressions for $C_a$, $C_b$, R, BMD, CBD and E all decreased after 12 months in the non-exercised foals, these slopes all increased slightly in the exercised group. The previous year's foals had been kept under exactly the same conditions and were fed in the same way. The seasonal conditions were also similar. It was decided to use this group as controls and to compare the change in slope at the 12 months of age point. The

Table 2. *Comparison of slope changes (mean ± SD) pre- and post-12 months between non-exercised and exercised foals for all growth rate and bone quality parameters*

A positive *t*-value indicates an increase in the slope change for the exercised group

| Parameters $\bar{x}$ ± SD | Non-exercised foals Slope change ($n$=5) | Exercised foals Slope change ($n$=6) | *t*-value | *p*-value |
|---|---|---|---|---|
| Girth | −0.019 ±0.012 | −0.010 ±0.010 | 1.31 | 0.22 |
| Mid-MC3 circumference | −0.0044 ±0.0024 | −0.0043 ±0.0028 | 0.05 | 0.96 |
| Bodyweight | 0.0222 ±0.1635 | 0.0362 ±0.0495 | 0.20 | 0.85 |
| $C_a$ | −0.2256 ±0.2899 | 0.1802 ±0.3017 | 2.26 | 0.05 |
| $C_b$ | −0.2422 ±0.3459 | 0.3276 ±0.4067 | 2.47 | 0.04 |
| CSA | 0.002654 ±0.004678 | −0.000534 ±0.001102 | −1.63 | 0.14 |
| R | −0.000135 ±0.000106 | 0.000021 ±0.000091 | 2.63 | 0.03 |
| BMC | 0.000140 ±0.000106 | 0.001303 ±0.002232 | 0.87 | 0.40 |
| BMD | −0.000227 ±0.000416 | 0.000378 ±0.000659 | 1.77 | 0.11 |
| CBD | −0.000228 ±0.000399 | 0.000244 ±0.000389 | 1.98 | 0.08 |
| E | −0.003966 ±0.006004 | 0.006299 ±0.010110 | 0.20 | 0.08 |
| Optical density | 0.010617 ±0.008903 | −0.000859 ±0.002680 | −3.03 | 0.01 |
| Alkaline phosphatase | −0.1635 ±0.6622 | −0.6011 ±1.1228 | −0.76 | 0.46 |
| Osteocalcin | −0.6078 ±0.6443 | −0.9732 ±0.3407 | −1.21 | 0.26 |

general trend was for the changes (i.e. increases) in slope to be greater in the exercised group, with the most significant increases being for $C_a$, $C_b$ and R. The changes in $C_a$ and $C_b$ indicate a trend to reduction in bone porosity. The levels of significance of the increases in BMD ($p$=0.11), CBD ($p$=0.08) and E ($p$=0.08), although outside the accepted significance level, were nevertheless worthy of note. The change in slope of radiographic optical density was lower ($p$<0.05) than in the non-exercised group. This is difficult to explain in the light of the increases in the other parameters. The accuracy of this technique is questionable. Differences between changes in biochemical parameters were negligible.

This was the first study in young horses in which combined ultrasonic and photon absorptiometric measurements have been used to monitor the skeletal effects of submaximal exercise. Despite the absence of strong statistical evidence of change for most parameters, there was a trend towards increasing bone strength and density associated with exercise. The results were consistent with other more superficial studies in the horse where either no effect,[7] or very small increases in bone mass,[17,21] were detected as a result of submaximal exercise. These findings were also consistent with studies of submaximal exercise in other species such as mice,[1] pigs,[25] dogs[14] and humans.[5] We feel that more intense exercise, or perhaps an even longer programme, would be required for more demonstrable changes.

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