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THE INFLUENCE ON BREAKING FORCE OF OSTEOPOROSIS FOLLOWING FRACTURE OF THE TIBIAL SHAFT IN RATS

By

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INTRODUCTION

Loss of mineral in a fractured bone and in neighboring bones is a frequently-encountered phenomenon. In cases of tibial shaft fracture in humans, *Nilsson* (1966) found that there was a loss of about 25 per cent in the cancellous bone at the distal end of the adjacent femur.

In rats similar changes have been shown to occur by *Bauer* (1954) and by *Bohr* (1955).

It is the object of the present work to study the osteoporosis which appears in the proximal end of the rat tibia following fracture of the tibial shaft. The bones are evaluated in terms of ash weight loss and of decrease in resistance to compressional fracture.

MATERIALS AND METHODS

Twenty female Wistar rats were used. The weight of the rats was 252 ± 14 grams (range 223-271). This weight was chosen to minimize the influence of rat growth during the period of the experiment, and no significant growth was observed during this time.

The left tibial shaft of each rat was manually fractured under anaesthesia. The right tibias were to serve as controls. Four weeks later the rats were sacrificed and both tibias were removed and cleaned. The proximal end of each tibia was then cut away and the rest of the bone discarded. In each case, the end retained accounted for exactly one fifth of the original bone length. Since the fracture area was located near the middle of the shaft, it was always excluded from the test specimen.

The compression tests were carried out on a Riehle 5000-Kg Materials Testing Machine, operated on its lowest scale (0.250 Kg) and calibrated to an accuracy of 0.5 per cent. This is a standard industrial testing instrument. It has a motor-driven head, which presses the specimens down against a scale platform. A system of levers transmits the platform displacement to an indicator dial which reads in kilograms of force.

To ensure that the specimen was held in the proper position during the test it was mounted in the way shown in Figure 1. The force from the testing machine was transmitted to the sample through two plastic disks, which were free to slide in a plastic sleeve. It was necessary to provide two flat parallel surfaces on the bone so that the force from the disks should be transmitted evenly to the ends of the sample and directly along its longitudinal axis. The condylar end of each specimen was therefore flattened slightly with a fine grinding wheel, and then the cut end of the shaft was carefully ground parallel with it. To ensure that the cut end of the shaft should not fracture before the epiphyseal end, a fine wire was bound around the shaft to reinforce it. The breaking force of a bone varies with its degree of hydration (*Evans & Lebow* 1952). Prior to testing, the samples were hydrated in saline solution under negative pressure for one hour. This assured a thorough and constant water content.

Each sample was then tested under compression and the initial failure point was recorded. The initial failure point represents the highest resistance the specimen could offer to compression. If the test was carried beyond this point, the resisting force decreased to a lower range of values as the bone gradually collapsed. When the test was halted at initial failure, no definite fracture was discernible by eye. Just beyond this point, however, the bone could be clearly seen to fracture at a point just below the epiphyseal line. After the breaking force determination each sample was ashed for 48 hours at 550° C, and the ash weight determined.

RESULTS

The average ash weight and breaking force of the injured and control sides are presented in Table 1. The ash weight was found to be significantly less on the injured side ($P < 0.01$). The breaking force of the injured side was also found to be significantly less than the uninjured side ($P < 0.001$). The percentage loss in breaking force was significantly higher than the percentage loss in ash weight ($P < 0.001$). Although ash weight and breaking force form a regression when samples from both injured and control sides are included, there was no such regression within each group considered separately (Figure 2).

In Figure 3, the loss in ash weight is plotted against the ash weight of the normal (right-side) tibia sample ($P < 0.01$). Figure 4 is an analogous plot of the decrease in breaking force versus the breaking force of the normal side ($P < 0.001$).

The weight of the animals was not related to the ash weight of the

Table 1. Ash weight and breaking force. Average $\pm$ SD.

	Ash weight (mg)	Breaking force (kg)
Normal side	62.2 $\pm$ 9.7	19.1 $\pm$ 4.5
Fracture side	52.1 $\pm$ 7.8	10.3 $\pm$ 2.8
Per cent Loss	15.4 $\pm$ 10.8	44.4 $\pm$ 14.3

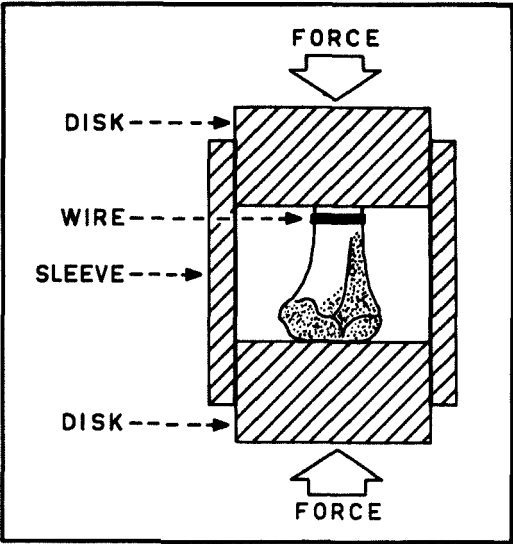


Figure 1. Bone sample mounted for testing (Cutaway view).

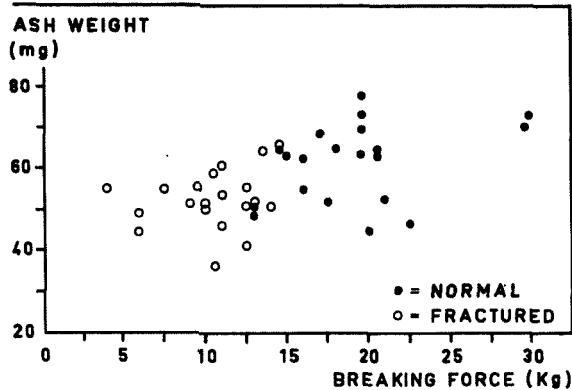


Figure 2. Ash weight of tibia samples versus breaking force.

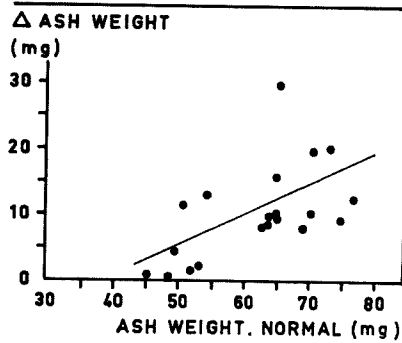


Figure 3. Ash weight difference between samples from the normal and injured sides versus ash weight of the normal side.

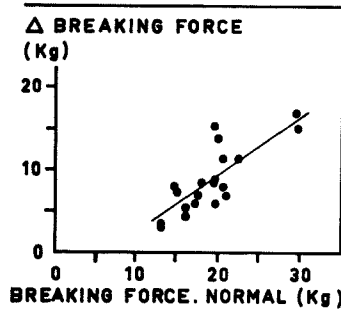


Figure 4. Breaking force difference between samples from the normal and injured sides versus breaking force of the normal side.

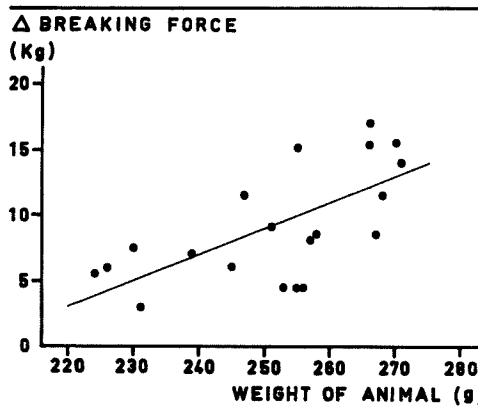


Figure 5. Difference in breaking force versus weight of animal.

uninjured side or to the loss of ash weight, nor was it related to the breaking load of the uninjured side. The loss of breaking force, however, was significantly ($P < 0.001$) correlated to the weight of the animals. Extrapolation of the regression in Figure 5 to zero seems to indicate that below a body weight of 200 grams no change in the breaking load occurs after fracture. Additional experiments including a wider range of animal weights will be necessary before any conclusions can be made about this finding.

DISCUSSION

The loss in ash weight observed in the present experiment confirms the presence of post-traumatic osteoporosis in the experimental limbs. This osteoporosis is associated with a relatively large fall in resistance to fracture. Since the specimens were not of uniform diameter and since each was largely cancellous bone, no estimation of stress (breaking force per unit cross section) could be made. *Nilsson & Saville* (1967) demonstrated a retardation of growth of the skeleton in fractured legs of growing rats. In the present study no significant growth could be demonstrated in the rats. It must therefore be assumed that the samples from the two sides were of equal size. Thus the observed breaking force difference must be attributed to a loss in bone quality.

Reports in the literature vary regarding the possibility of establishing a direct relationship between mineral content and bone strength. In studies on intact human femur, *Vose & Kubala* (1959) were able to demonstrate a relation between breaking stress and mineral content, where mineral content was measured by an X-ray absorption method. On the other hand *Melick & Miller* (1966), investigating age changes, observed a significant fall in the breaking stress of uniformly machined human tibia samples, but were unable to detect any change in specific ash weight.

In studies on rats, *Saville & Smith* (1966) found that femur density, calcium per unit volume, breaking force and breaking stress are linear functions of body weight, and therefore of each other (*Saville & Smith* 1966, *Smith & Saville* 1966). In bipedal animals, where the weight bearing of the femora was increased, it was found that breaking force and stress had risen to higher values than could be accounted for on the basis of a density increase alone (density expressed either as weight per unit volume or calcium per unit volume).

In studying the effects of a rachitogenic diet on rats, *deWeir et al.*

(1949) found a relationship between the breaking stress and ash weight of the femur of animals from various diet groups. Within each diet group, no such relationship could be found.

The findings of the present work are similar in that a relation between breaking force and ash weight is found only when samples from both the injured and the control side are included. The discrepancy between the ash-weight and breaking-force losses indicates that factors other than loss in mass alone are responsible for the deterioration in the mechanical quality of the bone.

In the graphs (Figures 3 and 4) in which the losses in ash weight and breaking force are plotted, respectively, against the normal-side values for each rat, both regressions have significant intercepts. Thus, when the loss in ash weight and breaking force between the normal and fracture side is zero, the bone still has a certain ash weight and breaking force. This seems to indicate that a certain fraction of the bone sample tested, presumably the cortical bone, to a lesser degree the participates in the process of resorption.

SUMMARY

Tibial shaft fractures in rats were found to cause a loss of bone in the proximal end of the tibia. This loss was found to be associated with a loss of resistance to longitudinal compression of the proximal end of the bone. This change in quality was found to be greater than could be accounted for by loss of mass only.

RESUME

On a découvert que les fractures du corps du tibia chez les rats causent une perte d'os dans la partie proximale du tibia. Celle-ci doit être associée à une diminution de la résistance de la partie proximale de l'os à la compression longitudinale. Cette modification de la qualité osseuse est en réalité plus importante qu'on aurait pu s'y attendre, compte tenu de la seule perte de masse.

ZUSAMMENFASSUNG

Es wurde gefunden, dass Tibiaschaftbrüche bei Ratten einen Knochenverlust am proximalen Ende der Tibia hervorriefen. Dieser Verlust war mit einem Widerstandsverlust gegen longitudinale Kompression des

proximalen Endes des Knochens verbunden. Diese Veränderung in der Beschaffenheit war grösser als man aus dem Verlust der Masse allein erwarten konnte.

REFERENCES

- Bauer, G. C. H. (1954) Rate of bone salt formation in a healing fracture determined in rats by means of radiocalcium. *Acta orthop. scand.* **23**, 169-191.
- Bohr, H. H. (1959) Studies on fracture healing. *J. Bone Jt Surg.* **37 A**, 327-337.
- Evans, F. G. & Lebow, M. (1952) The strength of human compact bone as revealed by engineering technics. *Amer. J. Surg.* **83**, 326-331.
- Melick, R. A. & Miller, D. R. (1966) Variations of tensile strength of human cortical bone with age. *Clin. Sci.* **30**, 243-248.
- Nilsson, B. E. R. (1966) Post-traumatic osteopenia. A quantitative study of the bone mineral mass in the femur following fracture of the tibia in man using Americium-241 as a photon source. *Acta orthop. scand.* Suppl. 37.
- Nilsson, B. E. R. & Saville, P. D. (1968) Influence of growth and trauma on bone mass and mineral turnover in rats. *Acta orthop. scand.* **39**, 273-280.
- Saville, P. D. & Smith, R. E. (1966) Bone density, breaking force and leg muscle mass as functions of body weight in bipedal rats. *Amer. J. Physiol. Anthropol.* **25**, (in press).
- Smith, R. E. & Saville, P. D. (1966) Bone breaking stress as a function of weight-bearing in bipedal rats. *Amer. J. Physiol. Anthropol.* (in press).
- deWeir, J. B., Bell, G. H. & Chambers, J. W. (1949) The strength and elasticity of bones in rats on a rachitogenic diet. *J. Bone Jt Surg.* **31 B**, 444-451.
- Vose, G. P. & Kubala, A. L. (1959) Bone strength—its relation to X-ray determined ash content. *Hum. Biol.* **31**, 261-270.