

these rats survived through the first 2 weeks and resumed growth during the third week. At autopsy 3 weeks later, one of the rats which had lost weight showed severe connective tissue scarring of the kidney surface.

Two of the rats which had received the stock diet for a 5-week period before being placed on the deficient diet lost weight during the second week of the experiment and both rats had connective tissue kidney scars at necropsy.

The data suggest that rats which were fed the stock diet during the preliminary feeding trial were somewhat less susceptible to choline-deficiency disease than those which were fed the choline-deficient diet supplemented with relatively high levels of choline.

Summary. Weanling rats receiving a diet severely deficient in choline required approximately 5 mg of choline chloride per rat daily to prevent the fatal hemorrhagic kidney disease. Choline deficiency was produced within 7 to 14 days at any time during the rapid growing period when choline was omitted from the diet. Weanling rats fed a stock diet for periods of 3 or 5 weeks were somewhat less susceptible to choline deficiency than rats fed the diet low in choline and supplemented with 20 mg of choline chloride daily for similar periods.

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Alterations in X-Ray Diffraction Pattern of Rat Tibia in Rickets.*

C. I. REED AND B. P. REED.

From the Department of Physiology, University of Illinois, Chicago Colleges.

The use of diffractograms in the study of molecular structure has been discussed extensively by Clark.¹ Numerous studies on bone by this technic have confirmed that there is a characteristic pattern most nearly agreeing with the type pattern of dahlite. Not many reports have given consideration to the possibility that this characteristic pattern may undergo physiological alteration. Roseberry, Hastings and Morse² described briefly some age changes in cultures of osseous

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¹ Clark, G. L., *Applied X-Rays*, 3rd, New York, 1940, McGraw-Hill Book Co.

² Roseberry, H. H., Hastings, A. Baird, and Morse, J. K., *J. Biol. Chem.*, 1931, **90**, 395.

tissue. Clark and Mrgudich³ described characteristic changes in rickets, quantitatively related to the degree of severity.

There was no fundamental change in chemical composition, since the lattice spacings were not altered. Both inorganic and organic elements were disorganized as evidenced by the much darker central area with its sharply defined boundaries and by the extension of the orientation arcs, sometimes forming a complete circle.

Several other reports have been made of qualitative alterations in diffraction patterns of bones under various experimental conditions.⁴

In an effort to study the influence of healed rickets on the subsequent behavior of bone, our laboratory first made a study of mechanical efficiency, already reported.⁵ A summary of findings in the first 5 series of those experiments is presented in Table I. From this it will be seen that in all groups on a rachitic diet for 2-3 weeks, there were no significant differences in mechanical efficiency as manifested by breaking stress, regardless of the duration of the recovery period. Significant differences were seen only when the period of rachitic feeding was extended to 6 weeks.

These same bones were used in the present study. Sections of cortex were ground from the tibial mid-shaft adjacent to the point of fracture. These were reduced to a thickness of 0.05 to 0.1 mm and mounted on a Hayes diffraction unit before a pinhole 0.025 inch in diameter. Exposure was made at 40 mm film distance to a Cu-target, G. E. X-ray tube operating at 35 kv, 22 ma for 2½ hours.

Analysis of over 500 diffractograms shows that in any long bone the most striking evidence of orientation is found in one large ring

TABLE I.
Comparison of Mechanical Resistance of Rat Tibiæ.

Series	Age on rachitic diet	Days on diet	Days on healing diet	Age killed, days	
I	25	20	65	110	Not significant
II	34	41	42	117	Significant
III	24	19	72	115	Not significant
IV	24	18	275	317	" "
V	23	37	42	102	Significant

In the last column is indicated the difference in mechanical efficiency between control and experimental animals, as determined by breaking stress.

³ Clark, G. L., and Mrgudich, J. N., *Am. J. Physiol.*, 1934, **108**, 74.

⁴ Clark, G. L., Bucher, C. S., and Lorenz, O., *Radiology*, 1931, **17**, 482; Henschen, C., *Schwiz. Med. Woch.*, 1937, **67**, 153, 182; **202**, 223; Reynolds, L., Corrigan, K. E., Hayden, H. S., Macy, I. G., and Hunscher, H. A., *Am. J. Roentgenol. Rad. Ther.*, 1938, **30**, 103, 286.

⁵ Schiller, A. A., and Struck, H. C., *Proc. Soc. Exp. Biol. and Med.*, 1941, **46**, 198.

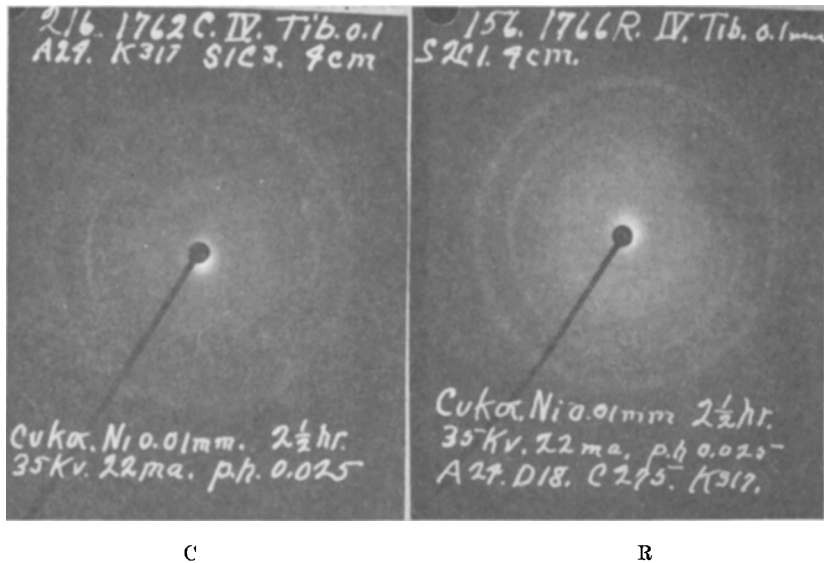


FIG. 1.

with a diameter of 40 mm and a d value 3.34 Åu. This agrees with all previous observations. This ring was sharply arced in the long axis of the bone. The mean total length of the two arcs for all of the control animals at the time of sacrifice was 231° as compared to a mean of 340° for the experimental animals. This last computation was made without regard to the period on the healing diet.

A comparison of two films is presented in Fig. 1. C was taken from a control rat in series IV and R from an experimental animal in the same series. Despite the fact that this series was on a good healing diet for 275 days the rachitic pattern is clearly evident. Table I shows that mechanical efficiency of the bones in this series was not impaired. It is not clear whether this disorientation is a physiological handicap in any other respect. The point will be investigated by some other technic.

The age of this particular group at death would be comparable to 35 years for the human.

If these findings apply to the human it must be concluded that the rachitic pattern of bone is never eradicated completely. Whether its persistence has any bearing on the behavior of bone in later life remains to be determined.