References and Notes

1. The virus under investigation is one that has been found constantly associated with root and foliage symptoms of internal cork disease. This sweetpotato virosis received its name from the root symptom phase. Studies are in progress to determine the relationship of associated symptoms such as leafspot, chlorotic spot, ringspot, chlorotic ringspot, purple ringspot, feather, and oak-leaf with the typical root symptoms.
2. E. M. Hildebrand, *Sceince* 123, 506 (1956).
3. ———, *Phytopathology*, in press.
4. A chip of diseased stem tissue with a bud in the middle, and measuring about ½ in. in length, was removed with a sharp razor blade and transplanted to an identical position on the healthy plant stem. It was held in place by a ½-in. strip of Stericrepe rubber.
5. With a scalpel, vertical stab wounds are made in the upper end of a sweetpotato root. The root is bedded upright in sand. Into each wound a snug-fitting, wedge-shaped scion is inserted. The scions usually establish vascular connections in 7 to 10 days.
6. E. M. Hildebrand, in preparation.

28 June 1957

Bone Crystallites as Observed by Use of the Electron Microscope

In 1949 Wolpers (*1*) observed, by use of an electron microscope, that the mineral portion of bone consists of needle-shaped crystals 30 to 60 A wide and 400 to 1000 A long. However, later electron microscopic studies by Robinson *et al.* (*2*) indicated that bone crystals are hexagonal platelets having average dimensions of 500 A long by 250 A wide by 100 A thick. Robinson's concept has been accepted by most workers in this country for several years. In 1953 Schwarz and Pahlke (*3*) interpreted electron micrographs to indicate that the calcareous (mineral) deposits in bone are spindle-shaped particles 150 to 1300 A long. From 1953 to 1957 Finean and Engstrom (*4*) presented evidence from x-ray diffraction studies indicating that bone contains rod-shaped, apatite crystallites 40 to 75 A wide and about 200 A long. Recently, Fernandez-Moran and Engstrom (*5, 6*) observed a predominance of rod- or needle-shaped apatite particles 30 to 40 A wide and about 200 A long in electron microscopic studies of undecalcified bone sections (human, rat, hen, and fish).

Electron microscopic studies of bone have been made in this laboratory (*7*). Sections (6 μ thick) of frozen-dried, methacrylate-embedded, undecalcified bone were obtained by routine methods (*8*). These were re-embedded in methacrylate with the desired orientation and sectioned at 90° to the plane of the 6-μ section. Satisfactory sections were obtained by using a diamond knife (*9*) in a Porter-Blum microtome (*10*). The sections were mounted on a grid coated with a carbon membrane and examined, using a 100-kv beam in an RCA EMU-3 electron microscope. The thickness of the sections was nominally 250 A. However, for best results it was necessary to study selected areas which may have been somewhat thinner than the average section thickness.

Inspection of many sections of normal, mature, cortical bone taken from humans and dogs showed an abundance of rod-shaped particles situated in groups or bundles in the plane of the section (Fig. 1). In other sites they appeared to be less regularly arranged, probably because of their oblique orientation. From considerations of relative densities, these are considered to be the inorganic crystallites. These crystallites were about 50 A thick and ordinarily 600 to 700 A long. Occasionally longer (about 1200 A) and frequently shorter (down to 200 A) particles were seen. Because the lengths of the particles may be several times the thickness of the section, it is apparent that the significance of these values (especially the smaller ones) is open to question until a method can be devised to demonstrate that the particle lies in, and not oblique to, the plane of the section. The volume occupied by the mineral component, from considerations of ash weights and densities of the organic and mineral components, is approximately 40 percent, a figure which is generally compatible with the appearance of these electron micrographs. The remainder of the volume is composed of areas of lower electron density in which the periodic banding that is typical of collagen fibers could sometimes be observed.

More recently it has been possible to section unembedded cortical bone, thus allowing examination shortly after sacrifice of the animal and without previous chemical treatment. Preliminary studies of such sections have shown structures indistinguishable from those described above for sections embedded in methacrylate.

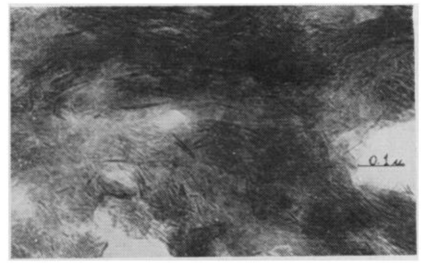

Fig. 1. Electron micrograph of a section of undecalcified cortical bone from the mid-shaft of the femur of an adult dog. The section shown is parallel to the long axis of the bone.

These electron micrographs are interpreted to show that the crystallites of bone are rod- or needle-shaped structures and not hexagonal platelets as reported by Robinson *et al.* (*2*). Fernandez-Moran and Engstrom (*6*) reported evidence of fine structure within these rod-shaped particles. However, our studies so far have failed to support this observation. Currently we are investigating the relationship of these crystals to the organic fibers of bone.

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References and Notes

1. C. Wolpers, *Grenzgebiete Med.* 2, 527 (1949).
2. R. A. Robinson, *J. Bone and Joint Surg.* 34A, 389 (1952); R. A. Robinson and M. L. Watson, *Anat. Record* 114, 383 (1952); ———, *Trans. Macy Conf. Metabolic Interrelations* 5, 72 (1953); ———, *Ann. N.Y. Acad. Sci.* 60, 596 (1955).
3. W. Schwarz and G. Pahlke, *Z. Zellforsch. u. mikroskop. Anat.* 38, 475 (1953).
4. J. B. Finean and A. Engstrom, *Biochim. et Biophys. Acta* 11, 178 (1953); ———, *ibid.* 23, 202 (1957).
5. H. Fernandez-Moran and A. Engstrom, *Nature* 178, 494 (1956).
6. ———, *Biochim. et Biophys. Acta* 23, 260 (1957).
7. This work was performed under the auspices of the U.S. Atomic Energy Commission.
8. L. A. Woodruff and W. P. Norris, *Stain Technol.* 30, 179 (1955).
9. The diamond knife was made available through the kindness of H. Fernandez-Moran.
10. The technical assistance of O. T. Minick of the Electron Microscope Laboratory is gratefully acknowledged.

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