

where A_b is the amount of bone mineral in a cross section of bone and G is the graph area obtained by plotting and measuring the area between the I and I_0 curves on semilogarithmic graph paper, as described previously. C is a constant which depends on the physical dimensions of the graph paper used.

If G is measured at two different energies, the Ca/P ratio for the cross section of bone under observation could be obtained from Eq. 3 and two equations of the form of Eq. 4. This method could also be used to determine the relative amount of fat in tissue.

This technique has been tested on a two-phase material with I^{125} (27.3 kev) and Am^{241} (59.6 kev) as radioactive sources. The materials used were blocks of paraffin and $CaCO_3$, homogeneously mixed in known proportions. The measurements resulted in determination of the percentage, by weight, of $CaCO_3$ present in the blocks, over a wide range of compositions, to an average accuracy of within 3 percent.

The principal errors at the present time are the result of uncertainties in the absorption coefficients and the lack of complete monochromaticity in the photon sources. These errors can be reduced through further study. If the only errors involved were statistical ones, an accuracy to within less than 2 percent could be expected in determining the mineral content of bone by this method.

The radiation exposure per scan with the techniques described is of the order of 0.15 rem with an I^{125} (27.3 kev) source of 5-mc activity. The exposure is about 0.05 rem when a source of Am^{241} (59.6 kev) of 5-mc activity is used. This exposure is limited to a small area of the forearm and should be compared with the maximum permissible dose to the forearms of children of 7.5 rem per year (6, 7).

Note added in proof. We have found a simple method for making "point" sources of I^{125} . Iodine is removed from solutions by an ion-exchange resin, Dowex 1 $\times$ 4, 20–50 mesh (8). Single grains of the resin, allowed to stand for periods of about 48 hours in freshly prepared carrier-free radioactive iodine solutions, will take up 5 mc of iodine. Grain diameters are less than 1 millimeter.

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5. D. E. A. Jones and H. C. Raine, *Brit. J. Radiol.* **22**, 549 (1949). The composition of the Mix-D used in our work is as follows: 55.2 percent (by weight) paraffin, 27.6 percent polyethylene, 12.5 percent MgO , 4.7 percent TiO_2 . It has been experimentally determined that Mix-D of this composition corresponds best with tissue of the forearm with respect to absorption of radiation.
6. *Federal Register* **25**, 104 (17 Nov. 1960).
7. We thank Dr. F. C. Larsen for his interest and advice, and David Knutsen, John Duffy, and Mrs. Susan Steinhart for assistance in making measurements and handling data. This work was supported in part by the Wisconsin Alumni Research Foundation; by the James Picker Foundation, on recommendation of the Committee on Radiology, National Academy of Sciences-National Research Council; and by the National Institutes of Health.
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Aragonite and Calcite as Constituents of Adult Oyster Shells

Abstract. Adult oyster shells are composed mainly of calcite, but there are five small areas of aragonite: the resilium, the two pads at which the adductor muscle is inserted, and the two pads at which Quenstedt's muscles are inserted. Quenstedt's muscles are proposed as a name for the small modified, one-time pedal muscles of unknown function, discovered by Quenstedt.

Nearly all the bivalve mollusks have shells composed of conchiolin and aragonite, the orthorhombic allomorph of calcium carbonate. Calcite, the more stable, rhombohedral allomorph, is not found in their shells. In contrast, the adult shell of the oysters of the family Ostreidae is composed mainly of conchiolin and calcite. Only five small, distinct, well-defined areas of the adult oyster shell are composed of aragonite. These areas are: the resilium between the two valves; the pads, one on each valve, at which the large adductor muscle is inserted; and the pads, one on each valve, at which Quenstedt's muscles are inserted (Fig. 1).

The resilium is one of the three portions of the ligament, which connects the two valves and allows the animal to open or shut its shell. The ligament is made of two kinds of conchiolin and, in the oysters, is divided lengthwise into three portions. The mid-portion is

the resilium and consists of one kind of conchiolin, while both flanking portions consist of another kind. The resilium is strong under compression, which is built up when the animal shuts its shell, but weak under tension; it is fibrous, brown to whitish gray, and semitranslucent and has lighter and darker parallel layers, normal to its fibers. The layers are produced during the intermittent growth of the ligament. The very fine fibers are subparallel but diverge like a fan toward the ventral border of the ligament; they are nearly normal to the ventral or growing border of the ligament, which in life is covered by the epithelium of the ligamentous crest of the animal's soft parts. The resilium fibers are composed of conchiolin and of aragonite; the aragonite fibers are the whitish ones. An x-ray diffraction pattern of many small fragments of the resilium from several oysters (*Crassostrea virginica*), ground to a powder, showed the curve of the mineral aragonite (1).

The large and powerful adductor muscle is inserted on the two valves on special pads, commonly called muscle imprints, which are located on the posterior half of each valve. Each pad is a smooth and very thin calcareous film, glossier and slightly more translucent than the rest of the valve. In *Crassostrea virginica* the pads appear dark purple in color, because the calcitic shell material directly beneath the semitranslucent pads is purple and can be seen through them.

With the aid of an electric engraving tool (Burgess vibro graver) many small fragments were broken from the pads of several valves of *Crassostrea virginica*. With this method it is not possible to obtain a pure sample from the pads alone, because they are too thin, and the calcitic shell material beneath the pads was dug into inadvertently. The x-ray diffraction pattern of the sample showed a curve indicative of calcite and aragonite. In addition, staining with Feigl's solution (2) demonstrated on several different species of oysters that the adductor muscle pads are composed of aragonite.

Quenstedt (3) discovered tiny places of insertion of muscles on the valves of the Early Jurassic oyster *Gryphaea arcuata* Lamarck, 1801, and surmised that they were vestiges of the anterior adductor muscle. Herdman and Boyce (4) were the first to give a histological description of these muscles based on *Crassostrea virginica*. Their work is still

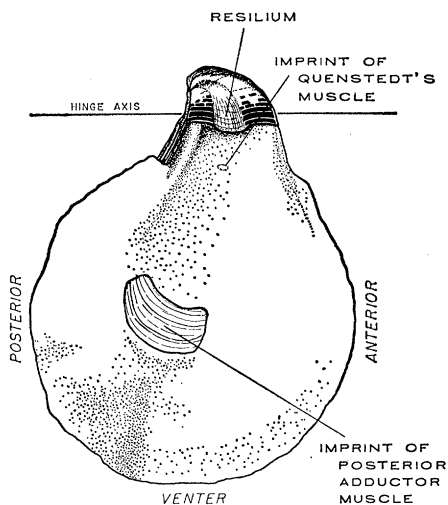


Fig. 1. The left valve of *Crassostrea virginica* (about $\times 0.6$).

the best, although the muscles have been seen or commented on, or investigated by, several other authors (5, 6), some of whom were unaware of the earlier work and claimed to have discovered them. I have found the places of insertion in all fossil and living species and genera available to me, provided the valves were well enough preserved. The conclusion is justified that these muscles, for which the name Quenstedt's muscles is proposed here, are present in all the Ostreidae.

In an average full-grown oyster, the muscles are 1 to 3 mm in diameter at their insertion on the valves, and the places of insertion are 3 to 13 mm from the ventral border of the ligament. The pads on which they are inserted are orbicular, or oval to elongate, and consist of aragonite as can be demonstrated by staining with Feigl's solution.

Each muscle rises from its valve at an angle of about 50 degrees and thins as it extends inward obliquely, in a slightly ventral direction, to pass close by the groove that separates the mantle fold from the outer labial palp. Then it turns to the anterior, splits, and spreads out. Most of its fibrils end in connective tissue between the inner and the outer lamella of the outer demibranch, at the dorsal, or adoral, end of this demibranch. Although the two opposite muscles converge and come close to meeting at the midline, they do not merge, as has been demonstrated by Herdman and Boyce (4) and confirmed by Elsey (6).

These muscles are very probably modified pedal muscles. However, adult oysters have no traces of a foot left and the muscles must serve other func-

tions. No direct observations concerning their function have ever been made, but from their arrangement it has been surmised that they draw the labial palps and the dorsal, or adoral, ends of the gills forward and outward (4) or adjust the position of the ends of the gills (5). In view of the uncertainty concerning their present function and concerning their exact homology with one of the several pairs of pedal muscles in normal Bivalvia, it is best to call them Quenstedt's muscles.

As long as the size of the oyster increases by growth, new pads must be laid down, because the muscles increase in size and shift their position toward the venter. New pads are laid down over old ones, but extend beyond them on the ventral side. In this fashion there develops a continuous stack of such pads, ending in the last pad occupied by the muscle. These stacks, consisting of aragonite, are buried within the calcitic shell material of the valves so that only the last pad remains visible. The stack belonging to the adductor muscle is known as the hypostracum. The one belonging to Quenstedt's muscle is best called a myostracum, a term introduced by Oberling (7) for all muscle pads and their stacks.

The results of investigations presented here are based on the adult shells of the common oyster species *Crassostrea virginica* (Gmelin, 1791), living today on the Gulf and Atlantic shores of North America. Although this species furnished most of the material investigated, countless other species and genera of the family, living and extinct, have been studied. The results given here have been found applicable in every case studied; it is likely that they apply to all adult oysters from the Late Triassic to today, although no Jurassic or Triassic oysters were found well enough preserved for testing.

It is not the intention here to claim priority of discovery of the aragonitic composition of the adult oyster hypostracum. Oberling (8), in his dissertation, dated 7 March 1955, was well aware of this fact. Also, he quoted R. W. Graves as stating in a letter dated 8 June 1953, that K. E. Chave had analyzed one sample of *Crassostrea virginica* and had found its hypostracum to consist entirely of aragonite. Before I began testing for aragonite, my colleague Otto Majewski called my attention to Oberling's work and to the aragonitic composition of the oyster hypostracum, which he had already

tested. These observations were pointed out to P. S. Galtsoff in a letter I wrote on 18 August 1961. Very recently, Heinz A. Lowenstam wrote me that he had used x-rays to prove the aragonitic composition of the hypostracum, because he had been led to expect to find aragonite there when he read the article on aragonite in the resilium of oysters (1).

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Ammonia: Possible Use for Preserving Fish

Abstract. *Eviscerated oil sardines (Sardinella longiceps) that have been treated with ammonia can be stored at a temperature of 25° to 30°C for more than 2 months without deterioration of their nutritive value. There is no measurable residue of ammonia in the final product.*

In many parts of the world, large catches of good edible fish become available with short seasons. When facilities for cold storage and refrigerated transport are inadequate, major parts of the catches, especially in countries like India, are not used effectively. Spoilage is extensive, and the fishermen and traders get comparatively poor returns. In such a fish as the oil sardine (*Sardinella longiceps*), the crudely prepared oil becomes the main product, while the more valuable body tissue, which contains proteins of high quality, is dried in the sun to make products which are not suitable for human consumption.

In the tropics, the spoilage of the fish starts within a few hours after the catch. If these changes could be prevented, the fish could be stored in bulk