neurosecretory cells from their axons, but the brain and the perikaryon of the cells are left untouched.

The results of experiment iii may account for our failure to restore the metabolism to normal by implantation of medial neurosecretory cells and should caution against the premature postulation of the involvement of a hormone.

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7. A remote possibility remains that the results are due to ablation of cells so close to the medial neurosecretory cells that our present surgical techniques cannot differentiate between them.
8. Supported by NIH grants (AI-05054, AI-03112). The assistance of J. E. Guira and D. G. Evans is gratefully acknowledged.

10 March 1965

Growth Rate of Giant Clam *Tridacna gigas* at Bikini Atoll as Revealed by Radioautography

Abstract. *At Bikini Atoll, radioactivity from strontium-90 deposited in the growing shell of a giant clam, presumably during the testing of nuclear weapons in 1956 and 1958, produced unmistakable lines on radioautographs made from transverse sections of the shell. The regular banding seen in the sections is interpreted as annular in nature. One annulus precedes the 1956 layer of radioactivity, two intervene in 1958, and six follow to the time of collection, so that this clam (length, 52 centimeters) was in its 9th year of life.*

Written records of the giant clam *Tridacna gigas* Linné have existed for centuries. Considered remarkable at first simply because of its large size (2 m in greatest length and several hundred kilograms in weight), the clam was later (*1*) found to contain symbiotic algae within its tissues. Yonge proposed that the clam attained such size by "farming" the zooxanthellae within the greatly expanded tissues of the siphons, and by utilizing the photosynthetic products in nutrition (*2–4*). However, in spite of an almost universal curiosity about the age of these giants of the coral reefs, no studies on their rate of growth have been reported. The rate of growth of *T. gigas* over a period of several years can now be estimated by reading the annulations in the shell and relating them to marks introduced by radioactivity from nuclear detonations.

Although the ability of *T. gigas* to concentrate Co^{60} in its soft parts has been emphasized (*5*), little is known concerning the uptake of radionuclides by the shell, which is shown here to contain Sr^{90}. To elucidate the pattern of deposition of nuclides in the shell after nuclear detonations, one valve of a specimen 52 cm in length (*6*) from Bikini Atoll was transversely sectioned with a 51-cm circular diamond saw. Figure 1 shows a section, 6 mm in thickness, from the region immediately anterior to the umbo. Figure 2 shows a radioautograph resulting from exposing the section to "No Screen" x-ray film for a period of 3 months. Two lines each about 2 mm wide, representing layers of radioactive material, appeared on the film. Other sections farther from the umbo also showed these marks. Records (*7*) reveal that tests of nuclear devices were conducted at Bikini Atoll only in 1946, 1954, 1956, and 1958. It is reasonable to attribute the layers of radioactivity to the two most recent test series. The 1956 Redwing series at Bikini extended from 20 May through 20 July, and the 1958 Hardtack series from 11 May through 22 July.

The positions of the layers containing radioactivity were determined by superimposing the radioautograph on the shell section, and are shown as stippled lines in Fig. 1, top. This view by transmitted light accentuates the conspicuous alternating dark, relatively opaque, layers, as contrasted with the lighter, more translucent bands, clearly indicating apparent years of age. Up to the 1956 line the clam was in its first year of life. Two years intervene between the two stippled lines, to 1958, and then six more years to the inner surface of the shell representing 1964, so that the clam was in its 9th year. The 1956 line corresponds to a shell length of about 10 cm, and the 1958 line, to about 24 cm.

It is of special interest that a tropical organism living in water with a mean monthly temperature varying less than 3°C (*8*) throughout the year should display distinct annulations. Seasonally varying environmental factors other than temperature, such as winds, currents, weather, light, and the abundance of planktonic food, could influence growth. At Bikini Atoll the relatively constant winter trade winds from the east are frequently interrupted in summer by other winds, particularly from the south (*9*), and surface currents would be similarly influenced.

Spawning is probably of a seasonal nature and thus may influence shell growth. Yonge (*2*) cites the spawning of the closely related genus *Hippopus* in January of the Australian summer and gives 30°C as the minimum temperature for spawning of the giant clam (*3*). Wada (*10*) reported that *Tridacna* collected in the Palau Islands in April, May, and June of 1938, 1940, and 1941 frequently discharged sperm and eggs when brought into the laboratory, although he said nothing of those collected in other seasons.

During growth, new shell material is added exclusively on the inside. Although the mantle is attached only at the pallial sinus, it contacts and deposits new material (aragonite) upon the entire inner surface of the shell. The extrapallial portion of the shell, distal to the pallial sinus and comprising about half of the total inner surface, is prismatic, while the central basal part of the shell is nacreous. In macroscopic views of sections (Fig. 1, bottom) the distal, prismatic part is relatively opaque and shows only faint layering; the central, nacreous part is more translucent and distinctly layered. The two areas are clearly demarcated by a boundary layer leading from the basal edge of the existing pallial sinus obliquely through the shell toward the umbo at the base (Fig. 1, *P*).

Figure 3 shows a low-power photomicrograph obtained by using crossed polaroid discs of a thin (15 to 20 μ) shell section at the position indicated by the dashed lines of Fig. 1 (top). Although Fig. 3 shows the outer border of the shell at the upper left, it does not extend to the inner border. The prismatic outer layer occupies the first and most of the second column of photographs down to the sloping light area which is the pallial layer marked *P*, while the rest of column 2 and all of columns 3 and 4 consist of nacre. The prismatic layer is composed of vertical columns about 45 μ in thickness disposed normally to the outer surface of the shell. The nacreous layers below are more irregular, with only slight or localized indications of vertical striae, but with both primary, coarse layering and fine striations oriented approximately parallel to the inner shell

surface. The degree of separation of the fine striations seems to affect the differentiation of the dark and light annular bands revealing age; the striations are finer and more closely packed (about 15 μ between centers) in the dark, relatively opaque areas of Fig. 1 than in the lighter, more translucent regions (25 μ). Probably the light areas are deposited during seasons of warmest water temperature, 29° to 30°C, which occur at Bikini Atoll from August to October, inclusive (*8*). Possibly a cool summer accounts for the lack of a distinct light area on the inside of the shell in Fig. 1 even though it was collected on 22 August.

Tridacna gigas grows fast compared to other molluscs. Wilbur and Jodrey (*11*) estimated from uptake of Ca^{45} that the shell of the oyster, a relatively fast-growing mollusc, increased in weight about 1 g per 70 cm^2 of surface per month. The annual increment in thickness of the giant clam shell under consideration was approximately 1 cm. With a shell density of 2.75 this would yield

$$(1.0)\,(70)\,(2.75)/12 = 16$$

grams per 70 cm^2 per month, or 16 times the growth rate of the oyster. Annular bands on the only other specimen (*12*) of *T. gigas* sectioned to date in this laboratory indicate growth approximately 1.5 times as fast, to a length of 55 cm in only 6 years. Such rapid growth, where length increased by 5 to 8 cm per year, or by 50 cm or more in from 6 to 9 years, probably gives the giant clam the distinction of being the fastest growing of bivalves.

In order to identify the radionuclides responsibile for the lines on the radioautographs, a strip about 7 mm wide along the 1958 line of radioactivity was bandsawed and chipped from the shell section shown in Fig. 1, top. The broken pieces of this excised strip, weighing 14 g, yielded 8 net counts per minute in a low-level, anticoincidence gross beta counter of 0.4 geometry with a background of 0.8 count per minute. There was no detectable peak above background even after 3900 minutes of counting on a 256-channel gamma spectrometer with a 7.6-cm sodium iodide crystal detector. The material was finally analyzed for Sr^{90}-Y^{90}, considered the most likely radionuclides

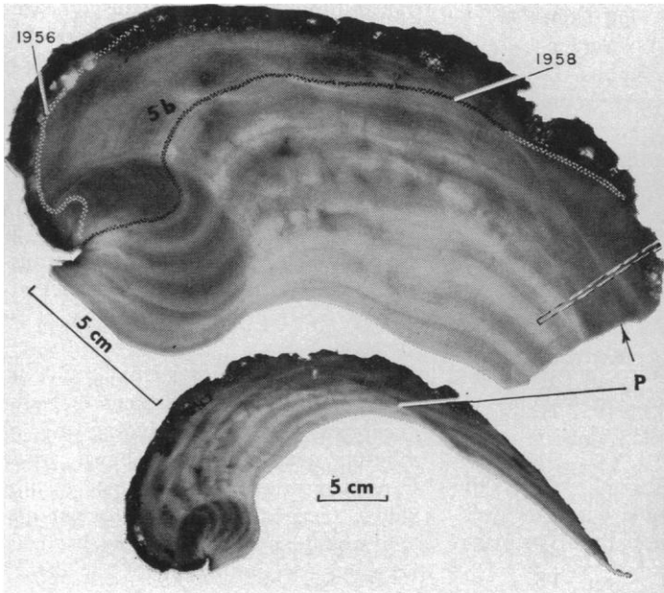

Fig. 1 (above). Transverse section, 6 mm in thickness, of *Tridacna gigas* shell near the umbo. (Top) Basal portion to a scale 2.5 times the lower figure, showing by stippled lines the positions of the layers of radioactivity traced from the radioautograph (*P*, indicates the pallial mark). (Bottom) Entire section.

Fig. 2 (above right). Radioautograph of basal portion of section of shell of *Tridacna gigas* shown in Fig. 1. Retouching was limited to the dotted lines indicating the outlines of shell and eroded areas. The two light undulating lines originating near the umbo resulted from the beta activity of Sr^{90} presumed to have been deposited in 1956 and 1958, respectively. Radioactivity is also evident in the umbonal cleft and elsewhere on the outer surface as well as in the eroded spaces. The white spot left of center resulted from Ca^{45} mixed with the ink used to label the section *5b* (see Fig. 1).

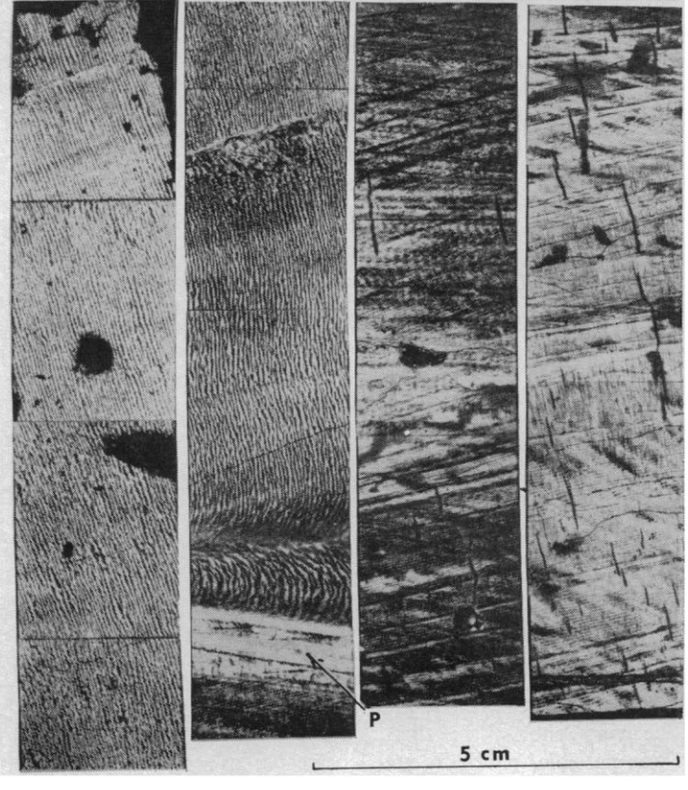

Fig. 3 (right). Composite photomicrograph of thin transverse section ground to a thickness of 15 to 20 μ. Removed from the position indicated by dashes on the right side of Fig 1, (top). Length and width of section, 41 by 2 mm. The continuous strip is divided for convenience into four columns starting with the outside of the shell at the upper left (*P* indicates the pallial mark).

because of the absence of gamma activity and because of the association of Sr with Ca. Beta-counting of the Y^{90} daughter of the Sr^{90} gave 16 disintegrations per minute per gram of shell material, and the decay rate was appropriate for Y^{90}. Thus, although there were undoubtedly other radionuclides in the shell shortly after the detonations, physical decay left detectable amounts of only Sr^{90}.

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13. Work done under contract AT(45-1)1385 with the AEC. I thank also Mr. Egil Oas for preparing the thick and thin sections of the shell, Miss Lorna Matson for the radioautographs, and Dr. Grant Gross for identification by x-ray diffraction of aragonite in the shell.

7 June 1965

Ultrasonic Scanning of Biologic Tissue by a New Technique

Abstract. *The size of the ultrasonic beam and beam dispersion severely limit resolution by two-dimensional scanning systems. Resolution and tissue penetration are improved by using a highly focused, ultrasonic transducer array in conjunction with an electronic timing system for the selection of particular echo information.*

Pulsed ultrasonic energy has been used in echo-ranging systems in biologic tissue for years; Gordon has published a comprehensive review of techniques used in this field (*1*). Several ultrasonic scanning systems have been devised which generate a two-dimensional image corresponding to the cross-section of the acoustic interfaces in tissue. Unfortunately, present scanning systems do not have fine resolution, yielding data that are difficult to interpret. Our current work to improve resolution may provide a new and versatile tool for research and diagnosis.

Ultrasonic scanning systems that generate a two-dimensional image have hitherto produced the image by displaying the echoes received after an ultrasonic pulse had been transmitted along a line corresponding to the path of propagation of the pulse in the subject tissue. The transducer was then moved about the subject in some manner, and the two-dimensional image was generated by summation of these individual line elements. Because the duration of the ultrasonic pulse may be very short (less than 1 μsec), it was possible to obtain excellent resolution in the depth direction or along the path of propagation of the ultrasonic beam. However, since the transducers required were operating primarily in the Fresnel region, the extent of the ultrasonic beam in directions normal to the path of propagation prevented good resolution in lateral directions. A focused transducer could be used to improve resolution in lateral directions at depths near the focal length of the transducer (*2*), but this improvement entailed the sacrifice of resolution at distances removed from the focal region.

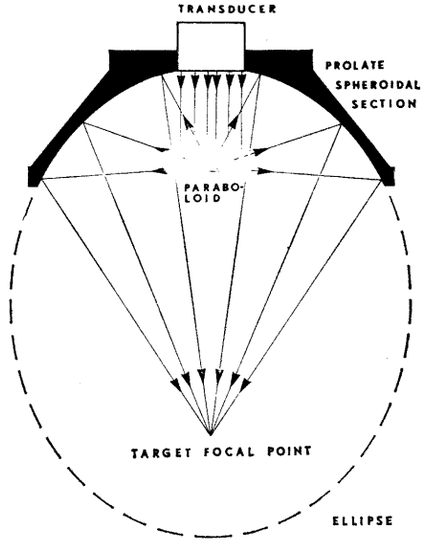

Fig. 1. Diagrammatic cross-sectional view of a multireflector, ultrasonic transducer array.

A technique has been developed that utilizes only the echoes that return from the region of minimum beam size, that is, the focal point of a focused transducer. This system generates a two-dimensional image by scanning the transducer in two dimensions and generating the image point by point. Echoes returning from the focal point are selected on the basis of a known, discrete time of return to the transmitting transducer. Thus, one restriction on the focusing system for the transducer array is that the length of the path of propagation from any point on the transducer surface to the target area must be constant. Fortunately the velocity of ultrasonic propagation in tissue is so close to its velocity in water that water can be used as the coupling medium between the transducer array and the tissue without affecting the focusing characteristics of the array.

Several possible systems could be employed for such a focused transducer-array system, including use of multiple transducers or of a single transducer with a concave face to provide a focused beam. The former system would be limited by the probable necessity for summation of the detected video signals rather than use of the video signals themselves; a difficulty inherent in the latter system would be development of a transducer large enough to produce a large solid angle of incidence convergent on the target area. A large solid angle is desirable so that target interfaces can be detected even when situated behind a strong reflecting surface or when oriented otherwise than normal to the transducer. The use of a sonic lens, which would cause the beam to converge by refraction, has the inherent limitation that the duration of the tone burst is lengthened by the reverberations within the lens itself. Because resolution by the system in a depth direction is limited by the timing accuracy and by the duration of the transmitted pulse, any reverberations within the focusing system must be avoided. These considerations then dic-