ing to their interpretation of the electron micrographs which accompany their paper, both types of filaments are relatively short in comparison with the length of the fiber; they lie parallel to the fiber axis and are grouped with separate arrays which alternate with each other and appear to be cross-linked by means of transverse projections which belong to the thick filaments. The existence of the projections does not seem to be firmly established, and their connections with the two types of filaments require corroboration. It is obvious from the electron micrographs published by Philpott, Kahlbrock, and Szent-Györgyi, (1960) that filaments are randomly distributed throughout the cross-sectioned area of the fibril.

In the relaxed state the muscle cells are stretched and on longitudinal sections of either part of the adductor appear to be arranged in parallel lines separated in places by connective tissue (fig. 145).

A contracted adductor muscle is strikingly different in appearance from one which is relaxed. Most of the muscle fibers are folded and the entire organ has a herringbone appearance (fig. 146). The uniform thickness of the folded fibers indicates that their actual length is not shortened by the contraction; the fibers are compressed to occupy a shorter distance between the valves. Folding implies the existence of a force that acts parallel to the longitudinal axis of the fibers. The question arises as to the nature of the force that produces this effect. In an attempt to answer

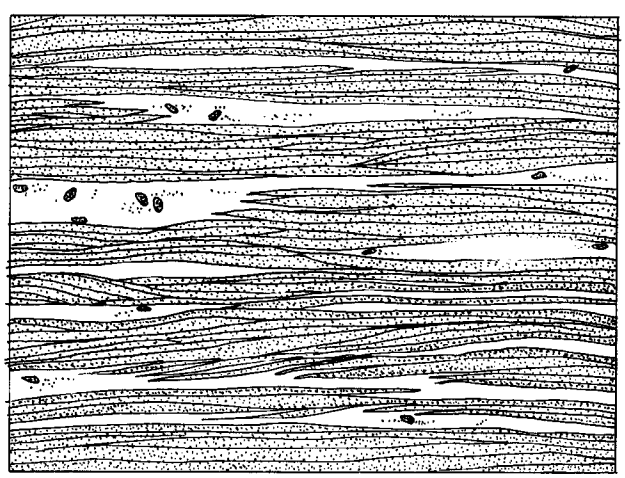


FIGURE 145.—Longitudinal section through a completely relaxed translucent part of the adductor muscle. Bouin, hematoxylin-eosin.

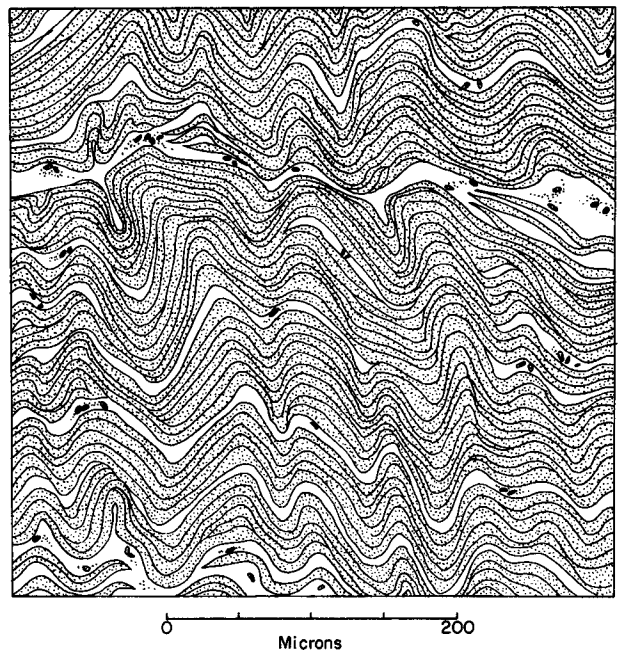


FIGURE 146.—Longitudinal section of the white part of the adductor muscle of *C. virginica* preserved in Bouin with formalin solution. Muscle is in a highly contracted state. Hematoxylin-eosin. Camera lucida drawing.

this question I examined a series of sections of muscles preserved at various degrees of contraction. Oysters were stimulated to close their valves and were preserved in that state by using a strong and rapidly acting fixative applied through an opening cut in a portion of the shell. In such preparations contracted muscle fibers were found only in the area near the attachment to the valves. In the two photomicrographs (fig. 147) the contracted fibers, nearest to the valve (left side), are short, thick, and deeply stained with eosin. The fibers to the right in the same preparation are narrow and folded.

In a partially closed oyster the contracted fibers may be scattered between the folded fibers throughout the entire cross-sectional area. This, condition, shown in figure 148, is drawn from preparations preserved in osmic acid and stained with iron hematoxylin. The contracted fibers appear as isolated dark bodies scattered throughout the moderately folded fibers. It may be deduced from the histological picture that only a small number of muscle fibers are in a contracted state. In order to explain the folding of the noncontracted portion of the adductor it is necessary to assume that a rigidity develops in the contracted fibers in two places—near their contact

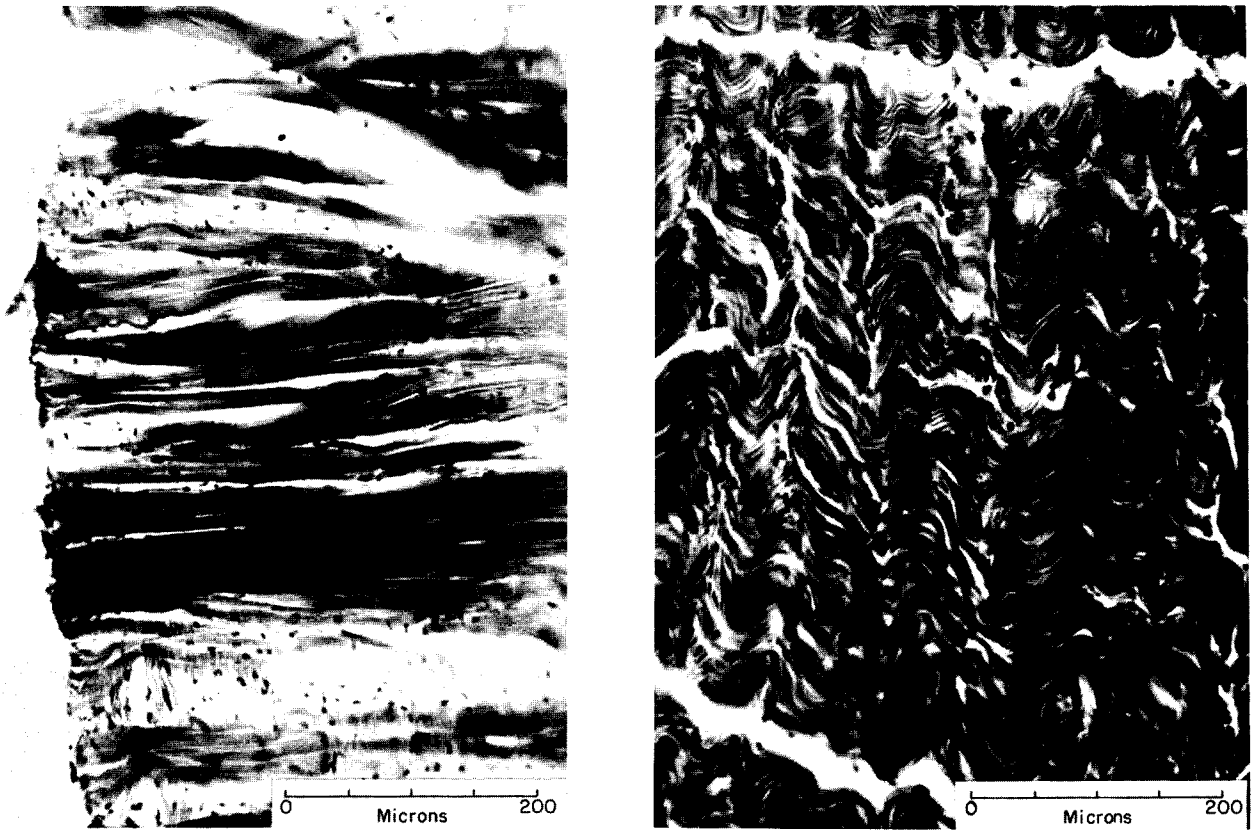


FIGURE 147.—Two photomicrographs of a longitudinal section of the translucent part of the adductor muscle near the valve (the left side of the photograph). The muscle was preserved in a contracted state in Bouin with formalin solution. Note the thick, short contracted fibers on the left and the beginning of folding at the right edge of it. Contracted fibers are deeply stained with eosin. The photomicrograph on the right shows folded fibers a short distance away from the area of the same section shown at left.

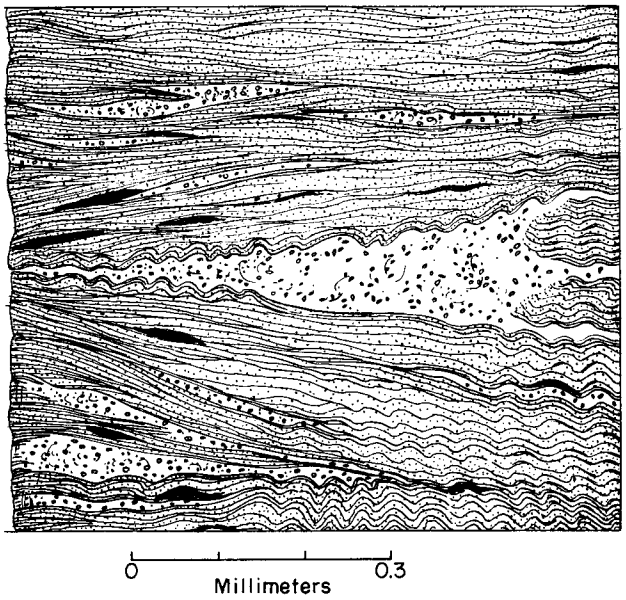


FIGURE 148.—Longitudinal section of partially contracted translucent portion of the adductor. Contracted fibers appear as black spindles. Osmium fixation. Iron hematoxylin.

with the folded fibers and at their anchorage in the connective tissue. Under this condition the contracted portions will bring the valves together and compress the noncontracted fibers into folds. This gives the oyster a considerable degree of flexibility in controlling the degree of opening of the valves.

Observations by Bandmann and Reichel (1955) on *Pinna nobilis* deal with similar conditions. In the smooth muscle of this mollusk plastic lengthening is combined with an orientation of the fiber structure without any changes of its elastic properties. The reverse process (disorientation) takes place during contraction, which is accompanied by an increase in dynamic stiffness. The authors attribute plastic and contractile length alterations to two different mechanisms: change in orientation and change in molecular shape within the contractile elements.

No observations have been made in the living oyster of the contractions of small bundles of fibers that run parallel to the surface of the valves.

These fibers, which are at right angles to the main fibers extending from one valve to the other, are found near the attachment of the adductor to the valves (fig. 149). Their position suggests that they act as braces by bringing together and tightening the principal bundles.

FIGURE 149.—Longitudinal section of a piece of partially relaxed muscle near the attachment to the valve (right side). Note band of muscles at right angle to the main fibers. Kahle, hematoxylin-eosin.

## ATTACHMENT TO SHELL

The adductor muscle of *C. virginica* is fastened so strongly to the shell that when the valves are forced apart the muscle breaks in the middle instead of tearing from the shell. The adhesion sometimes withstands a pulling force of 10 kg. (22 pounds). On the other hand, the connection between the muscle and the shell can be weakened or completely destroyed by applying heat to the shell over the area of the muscle scar. This connection is smooth and glossy.

Brück (1914) found that in the shells of *Anodonta* and *Cyclas* the muscles are fastened by means of a specialized layer of cells which he called holding or adhesive epithelium ("haft epithelium").

Hubendick (1958) used both electron and light microscopy to demonstrate the presence of adhesive epithelium in the areas of attachment of the muscles of the fresh-water snail *Acroloxus lacustris* (Maxwell). The surface of the cells has a dense brush border of minute microvilli which are transversed by very thin cytoplasmic fibrils originating in the base of the cell. The epithelial cells are fastened to the underlying connective tissue by the evaginations which extend into the base of the cells. Since the muscles used by Hubendick were fixed in osmic acid, which resulted in their detachment from the shell, the electron micrographs published in his paper do not show the actual connection between the microvilli and shell material. The shell surface over the area of the attachment has, however, small depressions into which fit the tops of the microvilli. It is, therefore, likely that in *Acroloxus* the adhesion of the muscle is accomplished in this manner.

The holding epithelium of *C. virginica* can be seen on transverse sections of decalcified shell and muscle preparations. Individual cell boundaries are indistinct, but the position of each cell is clearly marked by a large round nucleus (fig. 150). Fine strands resembling those described by Hubendick originate in the base of the cells and terminate at their surfaces. They are not visible at low power but can be seen under oil immersion. The holding epithelium of the oyster is a modification of the surface epithelium of the mantle; the transition from one type to another can be seen in the areas adjacent to the muscle attachment (fig. 151). The holding epithelium of *C. virginica* secretes an organic film of about 2 $\mu$ in thickness that consists of adhesive material by which the muscle fibers are attached to the shell. The chemical nature of this film was not determined, but staining properties suggested the presence of collagen. Since it is known that under proper conditions collagen is digested by collagenase, I made a series of experiments at Woods Hole to determine the effect of this enzyme on the attachment of muscles. Small amounts of phosphate buffer solution (pH 8.4) containing 1 mg. of collagenase per ml. were injected into adductor muscles through holes

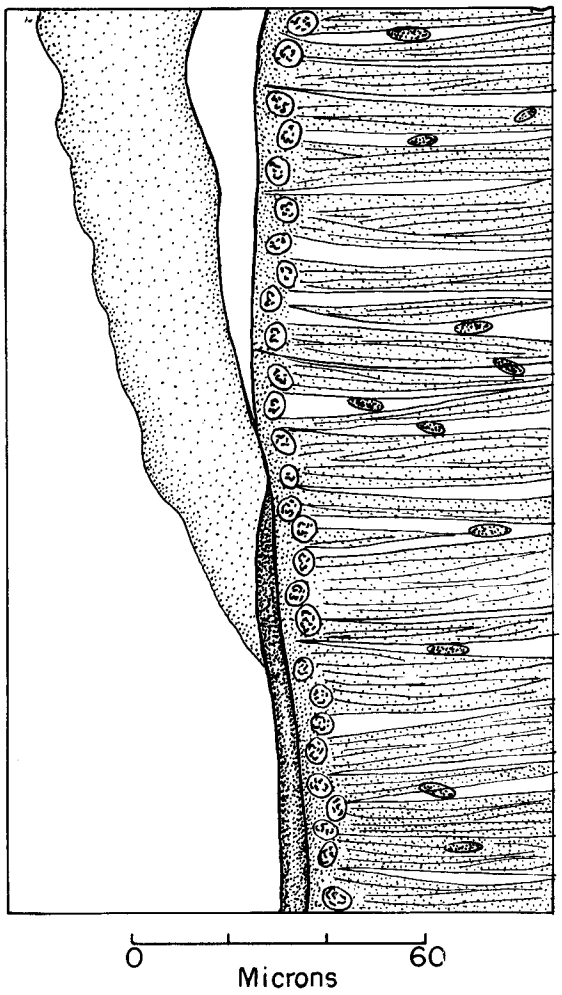


FIGURE 150.—Longitudinal section of the adductor muscle of *C. virginica* where the muscle fibers are attached to the shell. Note the holding epithelium and the cement layer which in the upper part of the illustration is separated from the epithelium and turned over. The decalcified shell is out of the field of view. The cement film is partially detached at the upper half of the preparation and twisted exposing its surface facing the shell; the width of the upper part of the film corresponds to the thickness of the section. Kahle. Hematoxylin-eosin.

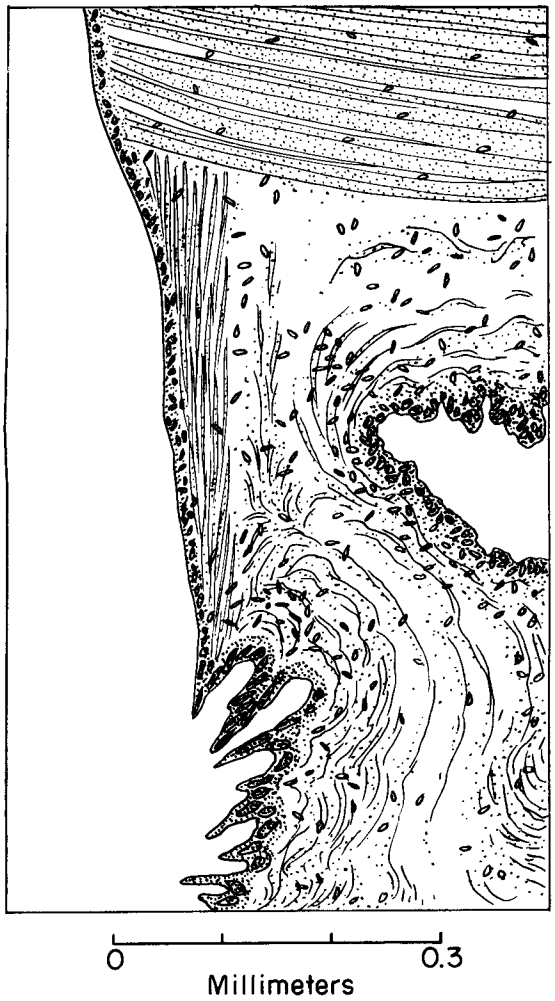


FIGURE 151.—Cross section of the visceral mass of *C. virginica* near the adductor muscle. Notice the gradual change of typical mantle epithelium (left side) into holding epithelium covering the adductor muscle. The shell is not shown. Kahle, hematoxylin-eosin.

drilled in the valves. In another set of experiments the muscles of oysters with the shells attached to them were immersed in the solution of collagenase and were kept at a temperature of 24° to 25° C. for 24 to 48 hours. Solutions of trypsin and phosphate buffer alone, without collagenase, were used for control experiments. In all cases the muscles treated with collagenase became detached within 36 hours. In the controls they remained attached to the shells (fig. 152).

## CHEMICAL COMPOSITION OF THE ADDUCTOR MUSCLE

The chemistry of the adductor muscle of oysters has received less attention than that of the muscles of clams, scallops, and sea mussels. Probably the differences in the chemical composition of the muscles of various marine lamellibranchs are not of fundamental nature, although the proportion of various components may vary greatly between the species and even within mollusks of the same species living in different environments. Older reviews dealing with the comparative physiology of the adductor muscle make no distinction be-