

# Muscle receptors in cephalopods

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[Plate 7]

Multipolar nerve cells with the characteristics of muscle receptors have been shown in the arms of *Octopus vulgaris*. The dendrites of these cells branch out into the muscle fibres and their axons make their way to small, intramuscular ganglion centres (ganglion of the sucker and intramuscular nerve cords), in which they seem to end. Multipolar nerve cells with characteristics similar to those of the cells described in *Octopus* have also been shown in the lip of *Sepia officinalis*. Such evidence permits one to think that these structures are more frequent in the cephalopods than has been suspected hitherto and it confirms the presence of a system of proprioceptors.

## INTRODUCTION

Muscle receptors are well-known structures in the nervous system of many animals, among invertebrates they have been described chiefly in insects and crustaceans. Information about these receptors is very scanty for cephalopods and other molluscs, in spite of the frequent use of these animals in research into the nervous system. In their well-known work on nervous degeneration in cephalopods Sereni & Young (1932) demonstrated the presence of multipolar nerve cells in the musculature of the mantle of *Octopus vulgaris*. More recently Alexandrowicz (1960) described a 'muscle receptor organ' in the mantle of *Eledone cirrhosa* in which a limited number (about 50) of multipolar nerve cells are present, not unlike those shown by Sereni & Young (1932), and with the morphological characteristics of muscle receptors.\* It has been demonstrated physiologically that cephalopods may co-ordinate their movements by means of proprioceptive information. Gray (1960) found phasic receptor units responding to mechanical stimuli in the muscle layer of the mantle of *O. vulgaris*. Boycott & Young (1950) have shown that *O. vulgaris*, deprived experimentally of its supraoesophageal lobes, if pulled by one arm, adheres to the bottom of the tank with the opposite arms and often aligns itself along the direction of the pull. Moreover, it is difficult to explain the complicated movements of the arms of the cephalopod when it captures its prey, and also its well-defined postural behaviour during rest, without postulating the presence of deep receptors, able to send a sufficient amount of information from the periphery at least to local reflex centres. Wells (1961, 1963) found that *O. vulgaris* cannot discriminate between (or cannot learn to react in a different way to) two objects of different weight, and he interpreted this as proving that the proprioceptive information does not reach the supraoesophageal centres.

\* In a somewhat unsatisfactory paper, properly criticized by Alexandrowicz (1960), Mikailoff (1921) described multipolar nerve cells in the muscles of cephalopods. In spite of the unacceptable statements of this author, some of the cells illustrated are probably muscle receptors.



I have observed that if the eight arms of an octopus are separated from the brain and from the mantle containing the viscera, they are still able to make co-ordinated movements. Arms prepared in this way can move from one point to another with movements similar to those of an intact animal. If a single point on one of the arms of such an eight arm preparation is mechanically stimulated, the nearest arms turn towards this point. An arm that has been isolated reacts to being stretched by contracting energetically and the contraction lasts as long as the pull is maintained. All these observations point to the presence of deep receptors, able to send information to the peripheral ganglia as to the state of tension in the muscles and, further, demonstrate the possibility that co-ordinated movements may take place in these animals without the participation of the learning centres.

Lack of more detailed morphological knowledge justifies a re-examination of the problem of muscle receptors in cephalopods. This paper reports on the results of observations of the arms of the octopus and of the lip of the cuttlefish.

#### MATERIAL AND METHODS

The material was collected at the Zoological Station at Naples during the summer of 1962. It consisted of specimens of *Octopus vulgaris* and *Sepia officinalis* in various stages of development from birth to maturity. The fragments were fixed, immediately after the death of the animal, in Bouin or in 10 % neutral formol in sea water. Many staining techniques were used but the most suitable were found to be Heidenhain's haematoxylin and Mallory for general histology of the tissues. The methods of Bielschowsky-Gros and Cajal modified by Young (1939) provided the best preparations for the study of the nervous tissue.

#### RESULTS

##### *The arms of the octopus*

##### *(a) The muscular system*

The muscles in the arms of the octopus are arranged in three systems: (1) the intrinsic muscles of the arm, (2) the intrinsic muscles of the sucker, (3) the acetabulo-brachial muscles (these muscles unite those of 1 and 2). The intrinsic muscles of the arm are grouped according to their orientation into: (i) longitudinal, (ii) transverse, and (iii) oblique systems (see figure 1).

##### *(b) Proprioceptor cells on the small nerve cords in the intrinsic muscles of the arm*

In four small canals situated in the angles of a square, lying among the intrinsic muscles of the arm, there are four small nerve cords running without interruption from the base of the arm to its tip. Guérin (1908) first described these cords as being motor centres for the surrounding muscle systems. Martoja & May (1956), using Cajal's method, confirmed Guérin's observations. They also showed anastomotic nerve bundles laterally connecting the internal to the external nerve cord. The diameter of these nerve centres is from 100 to 200  $\mu\text{m}$  according to the size of the animal, and is smaller near the tip of the arm. The canal that contains each nerve cord consists, on the ventral and dorsal sides, of trabecular bundles of



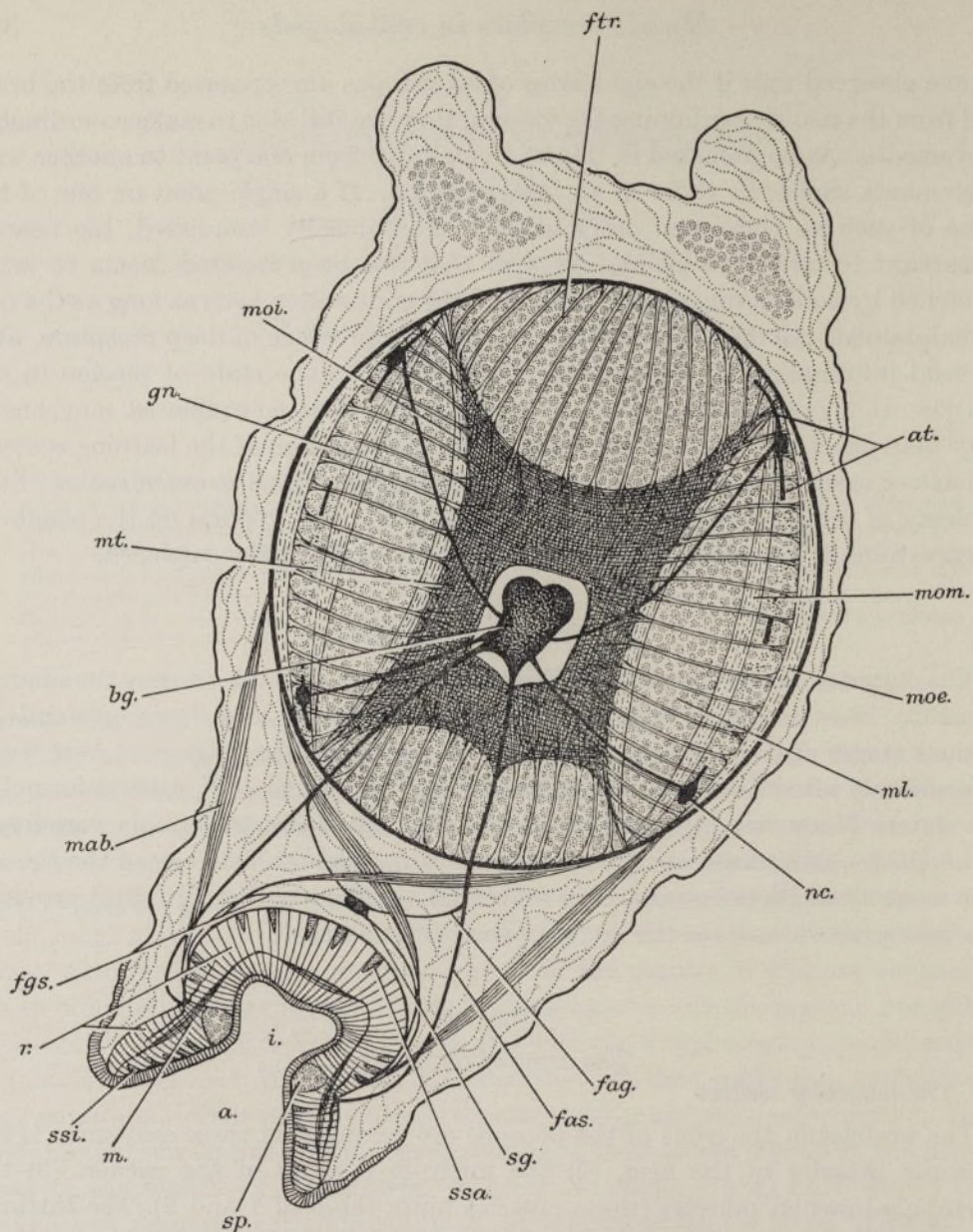


FIGURE 1. Schematic drawing of a cross-section of the arm of *Octopus*.

- |      |  |      |  |
|------|--|------|--|
| a.   | acetabulum   | mt.  | transverse muscles                       |
| at.  | anastomosis between intramuscular nerve cords                      | nc.  | intramuscular nerve cords                |
| bg.  | brachial ganglion (axial nerve cord)                               | r.   | radial muscles                           |
| fag. | nerve bundle connecting the sucker's nerves to the ganglion        | sg.  | sucker's ganglion                        |
| fas. | nerve bundle connecting the brachial ganglion to the sucker        | sp.  | principal sphincter                      |
| fgs. | nerve bundle connecting the sucker's ganglion to the sucker itself | ssa. | secondary sphincter of the acetabulum    |
| ftr. | trabecular bundles of the transverse muscle system                 | ssi. | secondary sphincters of the infundibulum |
| gn.  | nerve bundle connecting the bg. with the nc.                       |      |  |
| i.   | infundibulum   |      |  |
| m.   | meridial muscles   |      |  |
| mab. | acetabulo-brachial muscles   |      |  |
| ml.  | longitudinal muscles   |      |  |
| moe. | oblique external muscles   |      |  |
| moi. | oblique internal muscles   |      |  |
| mom. | oblique median muscles   |      |  |



transverse muscles and, medio-laterally, of oblique muscles (see figures 1 and 3). Little bundles of nerve fibres of varying composition originate from the intra-muscular nerve cords. A few, the smaller ones (10 to 30 fibres each), are distributed to the intrinsic musculature of the arm, whence, branching out, they end on the muscle fibres. The fibres of these small bundles, which originate in the small pear-shaped cells of the nerve cord (see below) probably have a motor function. Other

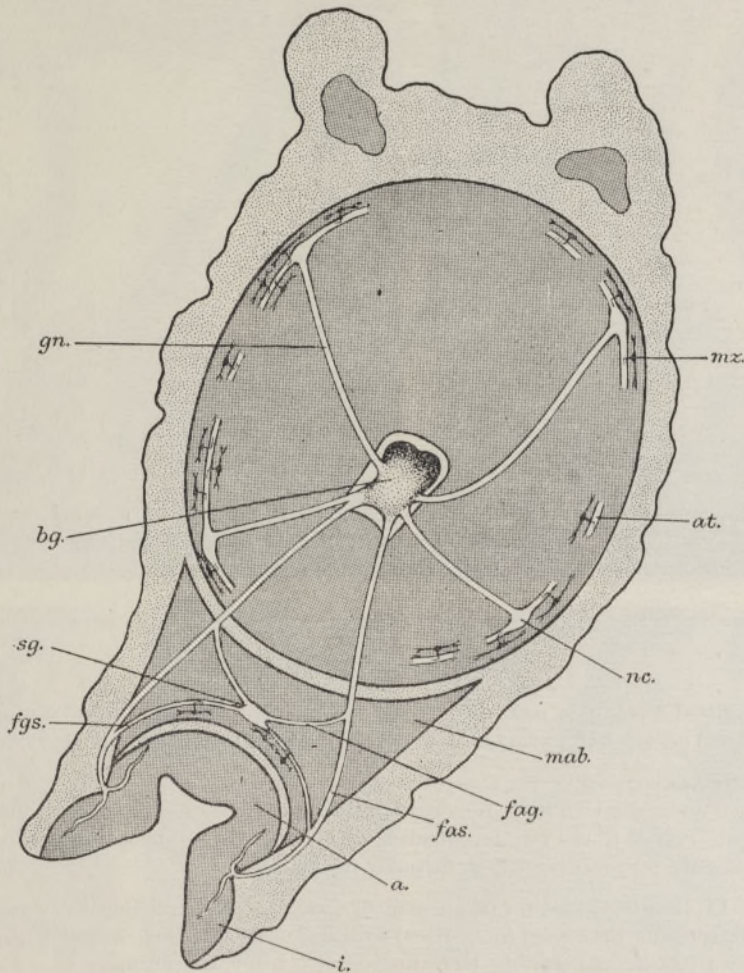


FIGURE 2. A simplified schematic drawing of a cross-section of the arm of *Octopus*. The three principal muscle systems of the arm are shown with the nerve centres and their connexions. Multipolar nerve cells are schematically represented (*mz.*). For the abbreviations see figure 1.

bundles (30 to 60 fibres each) connect the nerve cords with the axial nerve of the arm (figures 1, 2, and figure 7, plate 7), and are regularly spaced about 200 to 400  $\mu\text{m}$  apart. These values, like those first referred to, apply to an octopus of 250 g body weight.

The structure of the nerve cords is similar to that of other ganglia of these invertebrates. The central portion of the cord consists of a network of nerve fibres forming the neuropil. The nerve cells lie around this in an uninterrupted layer



(figure 3). The dimensions of these cells vary from 10 to 15  $\mu\text{m}$  for the smallest to 40 to 50  $\mu\text{m}$  diameter for the largest. The smallest are the most frequent (about 80 %) while the largest are scattered singly along the course of the nerve cord. The

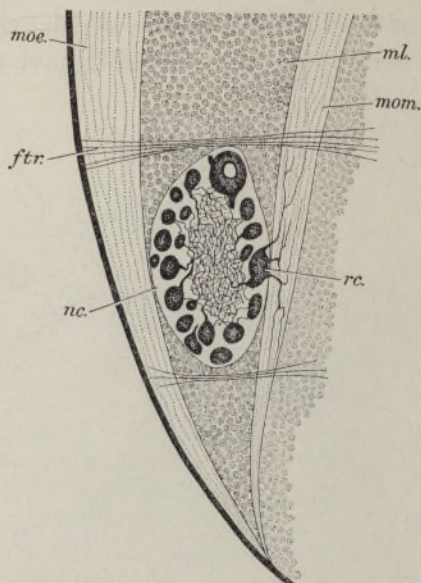


FIGURE 3. Diagram of one intramuscular nerve cord in the arm of *Octopus*. *rc.*, receptor cell with its dendritic branches in the oblique muscle. Other monopolar elements are presumably motoneurons. For other abbreviations see figure 1.

#### DESCRIPTION OF PLATE 7

FIGURE 7. Sagittal section of part of an arm of *Octopus*. Method Cajal-Young. Two nerve bundles (*gn.*) connect the brachial ganglion (*bg.*) with the intramuscular nerve cord (*nc.*).

FIGURE 8. Sagittal section of an arm of *Octopus*. Method Cajal-Young. An anastomotic tract which connects two intramuscular nerve cords is shown in the oblique musculature of the arm. Nerve fibres run longitudinally and no true neuropil is demonstrable. The arrows indicate polymorphous, multipolar nerve cells.

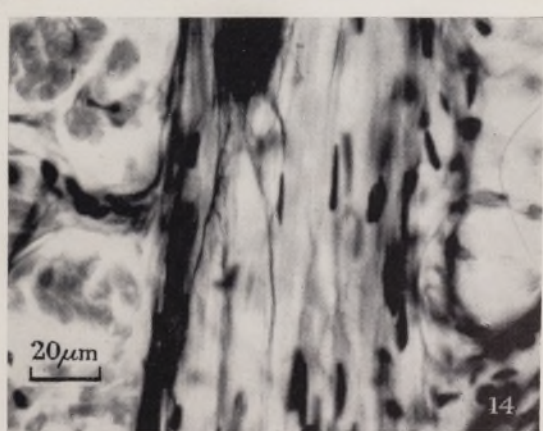
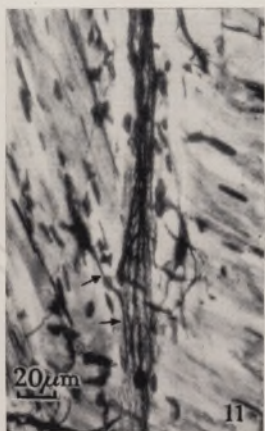
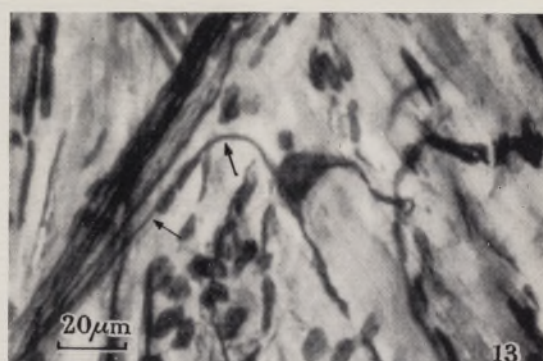
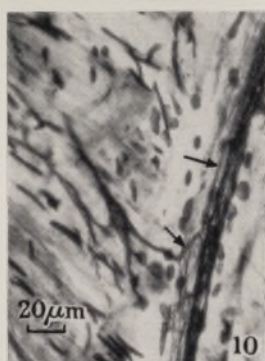
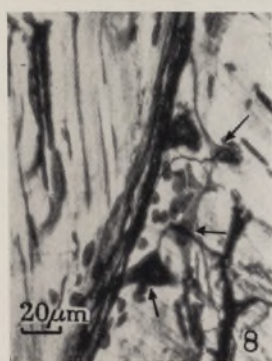
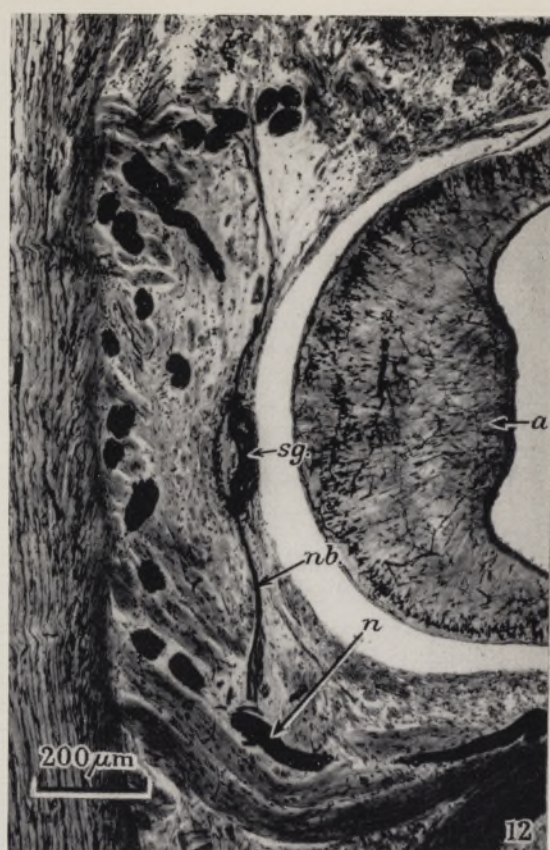
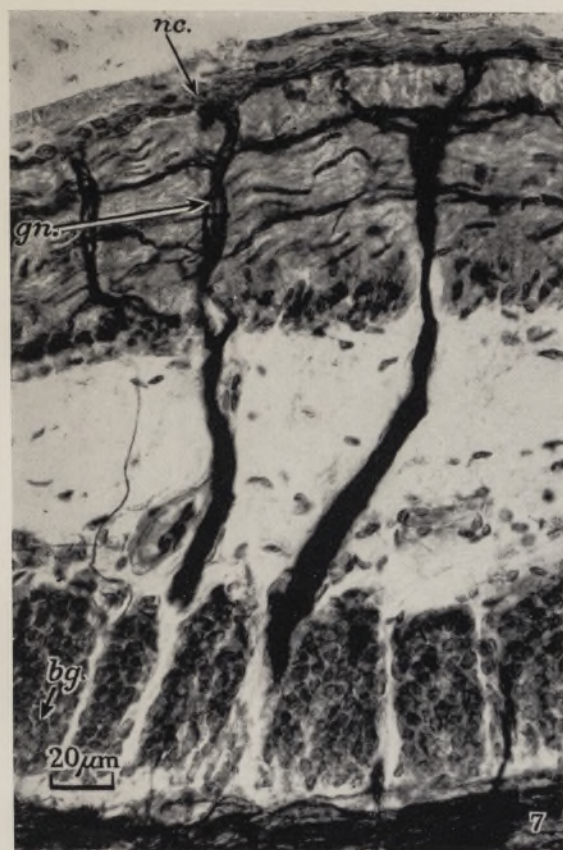
FIGURES 9 to 11. Sagittal section of an arm of *Octopus*. Method Cajal-Young. Multipolar nerve cells sending processes (dendrites) to the muscle bundles. Some of these cells show one process (the axon) joining the anastomotic tract and running in a straight course towards the ganglion centres. The arrows indicate the axon.

FIGURE 12. Sagittal section of an arm of *Octopus*. Method Cajal-Young. The section, passing through the axis of one sucker shows the acetabulum (*a*) of the sucker and the sucker's ganglion (*sg.*). Two nerve bundles (*nb.*) join this ganglion to the nerves running from the brachial ganglion to the sucker (*n*). The multipolar nerve cells lie close to the *nb.* and around the sucker's ganglion.

FIGURE 13. Sagittal section of an arm of *Octopus*. Method Cajal-Young. One multipolar nerve cell sends two processes (dendrites) to the muscles. A third process (the axon) can be followed joining one anastomotic tract. The arrows indicate the axon.

FIGURE 14. Longitudinal muscle bundle in the lip of *Sepia*. Method Bielschowsky-Gros. Detail of one multipolar nerve cell showing its dendritic processes on one muscle bundle. Note the absence of any sheath surrounding the cell and the processes which seem to be in free contact with the muscle fibres.







smallest cells are ovoid and unipolar and send their axons into the intrinsic muscles of the arm. Some of the larger cells are also unipolar but others are much more unusual, multipolar and irregular in shape. They are situated on the outside of the cellular stratum and three or more branches may start from the cell body. One of these (presumably the axon) runs into the neuropil while others branch and enter the surrounding musculature (see figures 2 and 3). These seem to be dendrites of muscle receptors similar to those previously reported by Sereni & Young (1932) and by Alexandrowicz (1960) in the mantle of *Octopus* and *Eledone*. These ganglia also contain bipolar, tapered cells the processes of which run longitudinally in the neuropil. The function of these cells is not yet known.

(c) *Proprioceptor cells on the bundles that unite the small nerve cords.*

Serial sections of the arm, stained with Cajal's method, show that the four intramuscular nerve cords are interconnected by means of oblique fibre bundles (about 30 to 40 fibres in one bundle) (see figures 1 and 2 and figures 8 to 11, plate 7). According to Martoja & May (1956) these bundles are regularly arranged on the left and right sides of the arms, but similar bundles can also be observed, although less regularly, extending from the anterior (or posterior) left to the anterior (or posterior) right nerve cord. The fibres of these bundles do not form a neuropil and between the fibres or at some distance from them there are nerve cells of various sizes and shapes (see figure 2 and figures 8 to 11, plate 7). Some of them, tapered, are like those previously described in the cords. Others are multipolar and quite irregular in shape and have dendrites that branch out into the muscle bundles, while the axon joins up with the fibres which, in a straight line, form the anastomosis. The multipolar nerve cells project their ramifications from the cell body to a distance of at least 100  $\mu\text{m}$  and often more. Observation of them is easier when the sections are cut in the same plane as the muscle layers in which the cell branches out. It is then possible sometimes to observe the cell and its finest ramifications in a single section. Each one in fact branches out into a single muscle system, keeping its ramifications in a single plane. The oblique external and median muscles (figure 1, *moe.* and *mom.*) as well as some peripheral bundle of the longitudinal muscles which lie close to the anastomotic tracts are the only regions in which multipolar nerve cells were found. The axons of these cells can be recognized by having a smaller diameter than that of the dendrites (2 to 4  $\mu\text{m}$  the axon and 5 to 10  $\mu\text{m}$  the dendrites) and by staining deeply with silver methods. The axon starts from a more lightly stained axon hillock. The dendrites are sometimes flattened and for long stretches have the same structure as the cell body. The irregularity of this and the wide base of origin of the dendrites often make it arbitrary to indicate a precise limit between the two structures. Cajal's method shows the fibrillar structure in these cells and the network is continued into the ramifications of the dendrites. When after repeated division these dendrites narrow, they become more visible in the silver preparations for their conspicuous black colouring. They seem to end on the muscle fibres in knobs or small enlargements but the limitation of the light microscope leaves the precise nature of these contacts at present unresolved.



*(d) Proprioceptor cells in acetabulo-brachial muscles*

Four cone-shaped systems of muscle fibres join each sucker to the arm and constitute together the acetabulo-brachial system. Each one of these cones inserts its base into the equator of the acetabulum of the sucker, while the apex inserts itself into the sheath that enfolds the intrinsic musculature of the arm. The muscle bundles of the four cones interlace, forming a complex network in the peduncle of the sucker (figure 1, and figure 12, plate 7). They perform the tasks of maintaining relations between the arm and the sucker and of orientating the latter. They enter, moreover, directly into function during the mechanism of adhesion, contracting simultaneously with the radial muscles during the aspirating phase of the sucker.

Earlier authors (Niemiec 1885; Guérin 1908; Ebersbach 1915; Graziadei 1959, 1960a, 1961, 1962) have described a small ganglion in the peduncle of the sucker of various cephalopods, called the ganglion of the sucker. According to Guérin, this ganglion in *Octopus* is composed of motor cells, which send their axons to the intrinsic musculature of the sucker. In the octopus the ganglion is situated in the axis of the peduncle of each sucker, immediately under the muscular cup of the acetabulum, from which it is separated by a stratum of loose connective tissue (figure 12, plate 7). The surface turned toward the arm is in touch with the interlaced acetabulo-brachial muscles which support it. The ganglion is in the shape of a biconvex lens with the two hemispherical surfaces turned toward the arm and the sucker. The diameter varies according to the diameter of the sucker, from 150  $\mu$ m for suckers of 1 to 2 mm to 500  $\mu$ m for suckers of 5 to 7 mm. The nerves that connect the brachial ganglia with each sucker (roughly 25 to 30) send a few bundles of fibres to the ganglion. In their turn, 20 to 30 thin bundles of fibres are sent forth from the ganglion and penetrate the sucker along the infundibulo-acetabular track. Along these latter nerves, or at some distance from them and fastened on the muscle bundles, are multipolar nerve cells similar to those described above in the intrinsic musculature of the arm. The dendrites of these cells branch out on contact with the acetabulo-brachial muscles, while the axon, joining itself with the nerve fibres composing the little bundles which radiate from the ganglion through the sucker, makes for the ganglion, where it probably ends. The number of the cells is difficult to estimate because of the uncertainty of the silver methods but there are approximately 50 cells around each ganglion. Some of these cells are even found on the external surface of the ganglion itself and are distinguished from the ganglion cells by the dendritic ramifications, which connect with the muscle bundles from which the ganglion hangs.

*(a) Muscular system**The lip of Sepia*

The lip of the cuttlefish is a circular fold which defines the mouth in front and lies at the bottom of the funnel formed by the bases of the eight sessile arms. The walls of this fold are lined with a columnar epithelium with numerous sensory cells intercalated (Graziadei 1960b). The internal structure of the lip consists of bundles of muscle fibres which can be divided into three principal systems according to



their direction: (1) circular or sphincter muscles, (2) longitudinal muscles, and (3) transverse or radial muscles (see figures 4 and 5). These three systems are interlaced but are separated by a considerable amount of connective tissue.

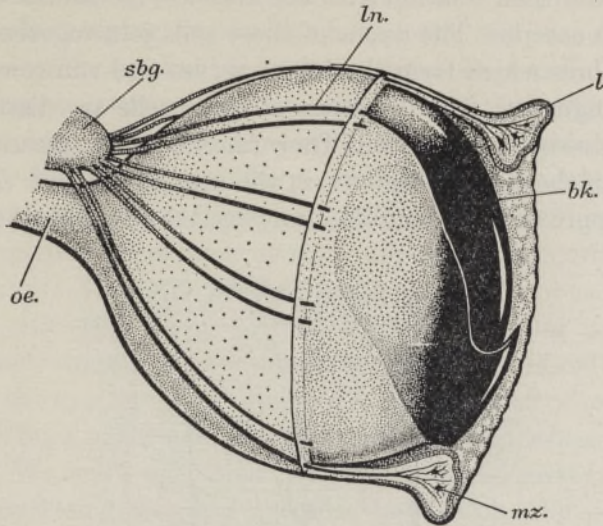


FIGURE 4. Schematic reconstruction of the mouth of *Sepia*. The axon of the multipolar nerve cells (*mz.*) joins up with the fibres which form the labial nerves (*ln.*), and runs towards the superior buccal ganglion (*sbg.*). *oe.*, oesophagus; *l.*, lip; *bk.*, beak.

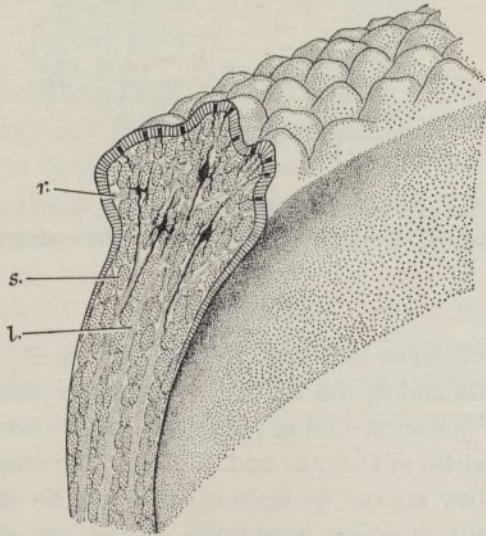


FIGURE 5. Diagram showing a portion of the lip of *Sepia*. The free edge of the lip is complicated by the presence of numerous papillae lined with a columnar epithelium. In this epithelium there are numerous sensory cells. The muscular pattern of the lip is formed by the three muscular systems: *l.*, longitudinal; *r.*, radial; *s.*, sphincter. There are numerous multipolar nerve cells among these bundles (in black).

#### (b) Proprioceptor cells in the muscles of the lip

The innervation of the lip is provided by the labial nerves, which originate in the superior buccal ganglion which contains the motor neurons for the muscles



(figure 4). Many of the afferent fibres originate from the sensory cells previously indicated (Graziadei 1960*b*). But it has also been possible to show multipolar nerve cells in the muscle bundles (figure 6, and figure 14, plate 7) which, in their morphological character and their relation with the muscles, are similar to those described in the arms of the octopus. The axons of these cells join together on a centripetal course with the fibres which form the labial nerves and run towards the superior buccal ganglion (figure 4). The dendrites of these cells are distributed round the cell body in a radius of 50 to 100  $\mu\text{m}$ . Their functional significance is, presumably, the same as that of the similar elements in the arm of *Octopus*. Their number has been estimated approximately and does not exceed a thousand cells for the entire lip.

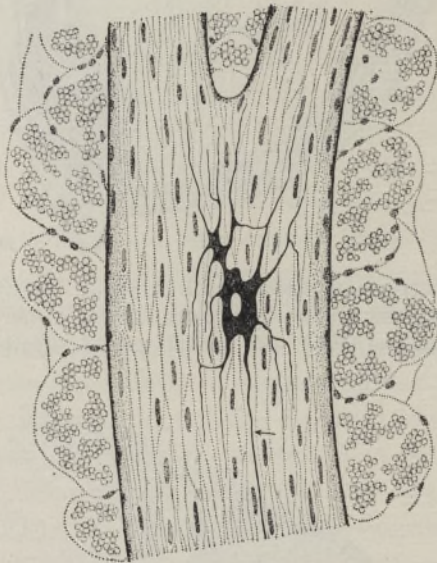


FIGURE 6. Drawing of a muscle bundle of the lip of *Sepia*, with one sensory nerve cell and its many irregular dendritic processes. The arrow shows the axon.

#### DISCUSSION

The demonstration of nerve cells with the characteristics of muscle receptors in the arms of the octopus and in the lip of the cuttlefish confirms and extends the earlier observations of Sereni & Young (1932) and of Alexandrowicz (1960), which were limited to the mantle of *Octopus* and *Eledone*. The receptors observed in the arms of *Octopus*, as they appear in light microscopy, lie directly on the muscle bundles, free from any capsular apparatus. They are situated around small ganglionic centres (the intramuscular nerve cords and the ganglion of the sucker). Until now these ganglia have been considered exclusively as motor centres, owing to the presence in them of neurons which send their axons into the muscles, and whose shape is typical of the motor centres of these invertebrates. The present observations confirm the motor nature of many of the cells of these centres but show also that receptor elements lie inside and around them and have morphological characteristics consistent with the function of muscle receptors. In fact, these neurons in cephalopods, like the well-known stretch receptors in crustaceans



(Alexandrowicz 1951; Kuffler 1954; Eyzaguirre & Kuffler 1955; Florey & Florey 1955) and in insects (Slifer & Finlayson 1956; Finlayson & Lowenstein 1958), have the cell body in the periphery sending dendrites into the muscular elements and the axon towards the ganglia. There are considerable morphological differences between the muscle receptor organs in crustaceans and insects and those now described in cephalopods, as would be expected from the presence of an exoskeleton as well as the different structure of the muscles and of their efferent nerve supply. Indeed, the morphology of the muscle receptor organs is variable among crustaceans themselves (Florey & Florey 1955). The present observations have failed to demonstrate any differentiated muscle fibres belonging to the receptor apparatus, or any efferent neural control system such as those previously described in crustaceans (Alexandrowicz 1951; Florey & Florey 1955). In the octopus's arms as well as in the lip of the cuttlefish, these polymorphous, multipolar nerve cells may be regarded as relatively simple receptor structures giving the animal a rough indication of the stresses in the arm muscles. Moreover, the presence of muscle receptors in some muscular systems of the arm but their absence from others is not well understood at present. We know the arms can perform their movement only by integrating all the interlacing muscular systems, but it is hardly possible that a few muscle systems provided with stretch receptors send an adequate amount of information for the performance of co-ordinated movements of the organ as a whole. As the technical difficulties in demonstrating these receptors are formidable, it is not possible at present to exclude that they have a wider distribution than is here described.

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