

MUSCULAR ACTIVITY OF THE MANTLE OF *SEPIA* AND *LOLIGO* (CEPHALOPODA) DURING RESPIRATORY MOVEMENTS AND JETTING, AND ITS PHYSIOLOGICAL INTERPRETATION

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SUMMARY

1. The action of the mantle of *Sepia* and *Loligo* has been monitored under nearly natural conditions. Respiratory movements are confined to the anterior mantle whilst during jet cycles the circular muscles contract powerfully throughout the mantle.

2. Contraction of circular muscle results in thickening of the mantle and expulsion of water from the mantle cavity. Activity of radial muscles causes the mantle to become thinner and to expand in surface area so as to inhale water.

3. During such movements these two groups of muscles antagonize each other directly without the participation of a discrete skeleton.

INTRODUCTION

Recent studies on jet propulsion of *Sepia* and *Loligo* (Trueman & Packard, 1968; Packard, 1969; Johnson, Soden & Trueman, 1972) have considered the dynamics of this locomotory mechanism whilst Ward (1972) and Ward & Wainwright (1972) have described some of the changes that occur in the musculature of the mantle during respiration and jetting. In this study we have attempted to observe and monitor the action of the mantle of whole animals in as natural condition as possible and to consider how the events of a jet cycle are brought about.

A jet cycle consists of the expulsion of water from the mantle cavity by contraction of circular muscle fibres (the compression or power stroke) from a resting condition followed by recovery of the mantle musculature and filling of the cavity (the return or recovery stroke). A jet cycle may be preceded by hyperinflation of the mantle cavity prior to the production of a jet of maximal pressure. Our knowledge of the neurophysiology of the mantle muscles still rests on the researches of Prosser & Young (1937), Young (1938) and Wilson (1960) who used isolated preparations of mantle muscle and stellate ganglion of squids. The present investigation is discussed in the light of these previous researches and some of the outstanding problems are emphasized.

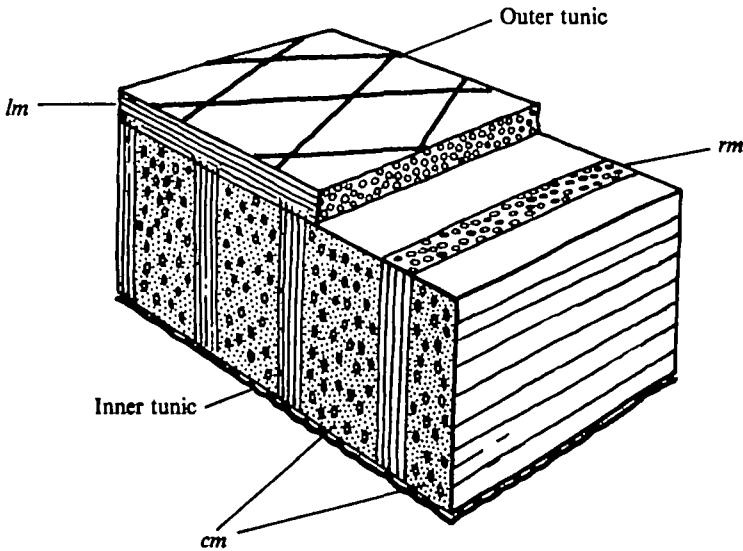


Fig. 1. Diagrammatic section of a portion of the squid mantle to show outer and inner tunics of helically arranged collagen fibres and muscle layers. *cm*, circular muscles; *lm*, longitudinal muscles; *rm*, radial muscles (after Ward and Wainwright, 1972).

Before considering the experimental observations it is necessary to outline the structure of the mantle (Fig. 1) (see Ward & Wainwright, 1972). The main bulk of the mantle consists of bundles of circular muscle fibres which run around the mantle cavity in the plane of the body's transverse axis. They are traversed by sheets of radial muscle fibres passing through the thickness of the mantle. External to these is a thin layer of longitudinal muscle, particularly apparent in *Sepia* near the mantle margin, and an inner and outer tunic of helically arranged collagen fibres (Ward & Wainwright, 1972) running at an angle of 60° to each other and to the transverse axis. The collagen fibres are less well developed in our sections of *Sepia officinalis* than described for *Loligo* (Ward & Wainwright, 1972).

We refer to two main types of movement produced by these structures: (i) those of respiration (rhythmic) and (ii) those of locomotion (characterized by a steep pressure pulse and jetting).

MATERIALS AND METHODS

Recordings of mantle movements were made on sub-adult *Loligo vulgaris* Lamarck and adult *Sepia officinalis* (L.) at the Stazione Zoologica, Naples. Ciné film of the latter species was also taken at the Marine Laboratory, Plymouth, to determine changes in mantle dimensions out of water.

The procedure for filming was to place the cuttlefish in air, ventral side uppermost, having removed the skin to expose the mantle muscle during narcosis with urethane. This surface was marked with an irregular pattern of dark points by dusting lightly with fine crystals of silver nitrate. Film was taken during the jet cycle of both ventral and lateral aspects simultaneously by use of a mirror.

In recording the activity of the mantle we sought to use the animal in as natural a condition as possible, rather than an isolated nerve-muscle preparation. We inserted

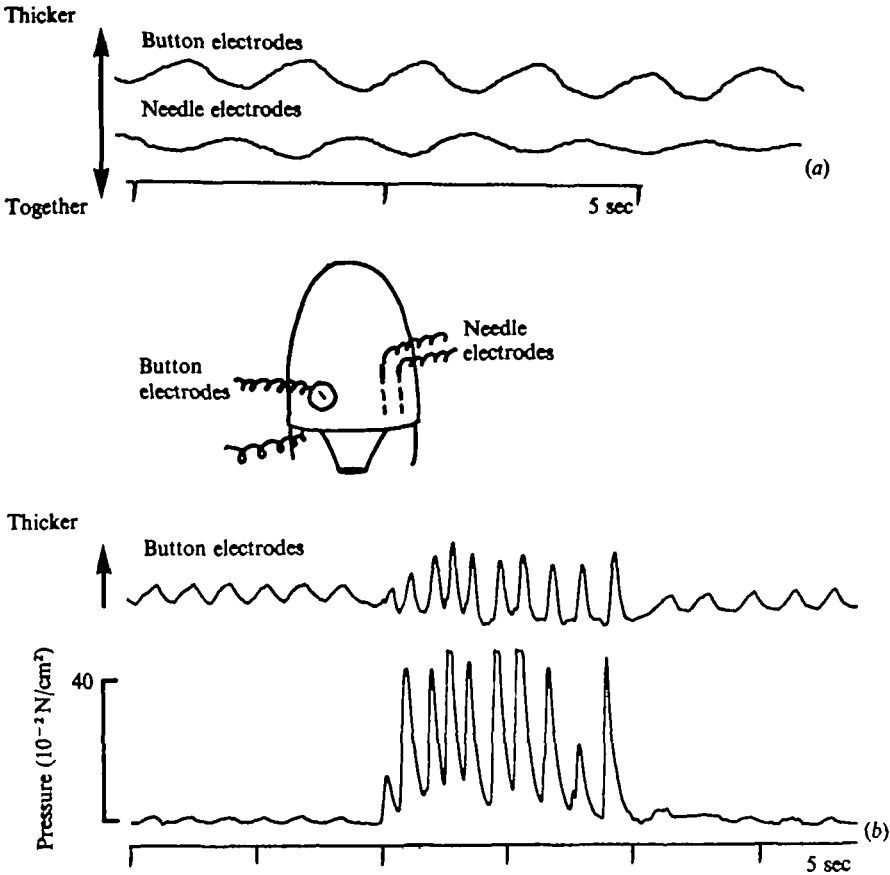


Fig. 2. Recordings of changes in dimension of the mantle of *Sepia* (a) and *Loligo* (b). (a) Changes in thickness (button electrodes) and circumference (needle electrodes) of the mantle during respiratory movements, electrodes being attached to the ventral surface of the mantle as shown diagrammatically. (b) Changes in thickness of the mantle and mantle cavity pressure (below) during respiration and jetting.

a pair of electrodes in an appropriate part of the mantle and a small high frequency current ($2 \mu\text{A}$, 25 K Hz) was passed between them using an 'impedance pneumograph' (Narco Instruments Inc.). Changes in the separation of the electrodes affected the impedance recorded. The electrodes were of two kinds: 'needle' electrodes (inserted parallel to each other antero-posteriorly along the outer surface of the mantle), to record extension or contraction in the circular direction, and 'button' electrodes (one placed on the outer and one on the inner mantle surface, sewn into position through the mantle), to record changes in thickness (Fig. 2a). These electrodes were connected by fine screened cable to the impedance pneumograph, while the animal was held in a small net suspended in a large tank of sea water. The squid and cuttlefish could not get out of the net, but were able to respire and jet quite normally. An approximate calibration of the needle electrodes was achieved by comparing the deflexions obtained experimentally with manual movement of the needles over known distances. Pressure was monitored with the same technique as used previously (Trueman & Packard, 1968).

EXPERIMENTAL RESULTS

(a) Respiratory movements

Respiration was monitored by electrodes placed in the anterior region of the mantle of *Sepia* (Fig. 2*a*). The decrease in impedance recorded from the needle electrodes on contraction of the circular muscles corresponded to an increase from the button electrodes, due to simultaneous thickening of the mantle. The movement of the needle electrodes represented a shortening of the circular muscle by about 1%.

This contraction generates a water pressure on respiration of about 2×10^{-2} N/cm² and, compared with the jet cycle, the exhalation of only a small volume of water from the mantle cavity.

(b) The jet cycle

Higher pressures are developed in the jet cycle than during respiration and in *Loligo* (Fig. 2*b*), as in *Sepia*, the mantle thickens to a greater extent. The contraction of *Septa* circular muscle is approximately 10% when producing a pressure pulse of only 15×10^{-2} N/cm². Insertion of needle electrodes on the outer mantle surface, as in Fig. 3, allows the contraction of different parts of the mantle to be compared. A respiratory rhythm can be recorded from the anterior mantle, but not from the posterior part (Fig. 3*a*). During the jet cycle, however, contractions of the posterior part of the mantle correspond with high pressure pulses (Fig. 3*b*). It is difficult to interpret the fluctuations recorded simultaneously from the anterior mantle, but they certainly show some correspondence with pressure pulses.

Inspection of cine film of the jet cycle showed contraction of the mantle muscles from the resting condition in common with the records discussed above. In addition, a more expanded or hyperinflated state of the active mantle was observed, apparently resulting from thinning of the mantle and the consequent extension of the circular muscles. This feature has also been recorded during normal jet swimming by means of needle electrodes (Fig. 3*c*). It most commonly occurs in the more anterior part of the mantle with the inhalation of additional water immediately prior to a jet cycle. It is likely that maximal inflation of the whole of the mantle produces a jet of maximum volume and the greatest possible jet thrust. In *Loligo* additional thinning of the mantle can be observed to occur during successive jet cycles for the base of the trace from the button electrodes is then depressed in comparison to the level recorded during respiration (Fig. 2*b*). The mantle muscles may, thus, be in one of three alternate phases: first, a resting condition, to which it returns when circular muscles relax; secondly, contraction of the circular fibres with thickening of the mantle; and, thirdly, maximal extension of the circular fibres associated with thinning of the mantle.

(c) Dimensional changes of the mantle

It has been demonstrated above that the mantle of *Sepia* and *Loligo* reduces its circumference by contraction of the circular muscle fibres and becomes thicker during respiratory exhalation and jetting. Similar observations were made on *Eledone cirrhosa* (Lamarck). Insertion of a pair of needle electrodes in the mantle of *Sepia*, one in the

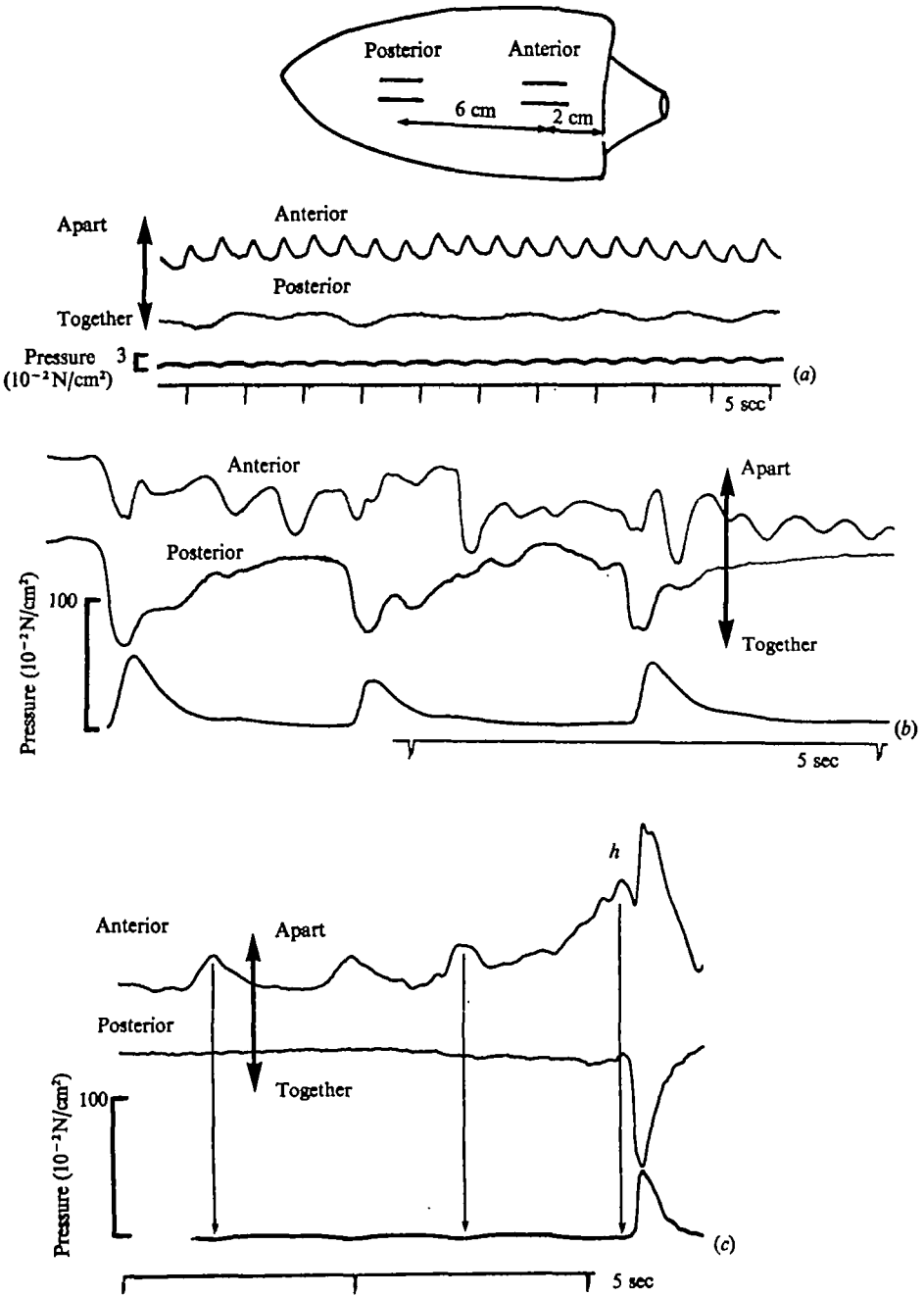


Fig. 3. Contraction of circular muscles of anterior and posterior regions of the mantle of *Sepia* recorded by needle electrodes (as in diagram) and associated mantle cavity pressure traces. (a) Respiration; (b) jetting; (c) respiration followed by jetting. Note inflation (*h*) of anterior mantle above resting volume immediately prior to the jet pulse.

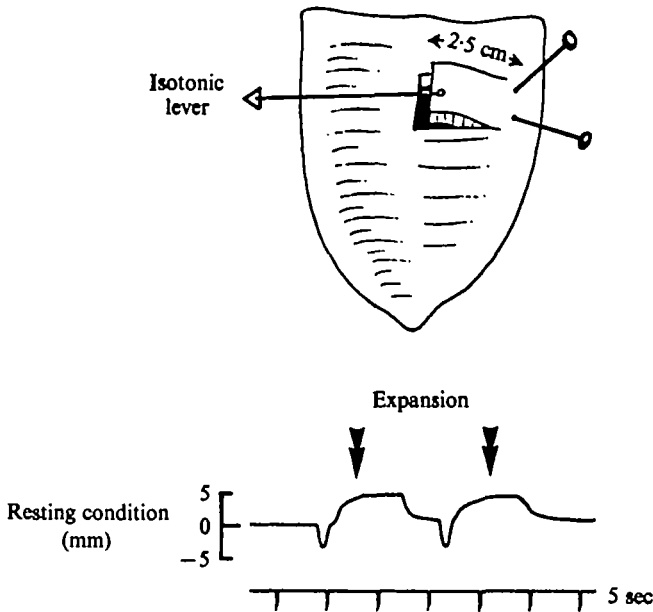


Fig. 4. Radial muscle contraction. Records of the changes in width (circular direction) of a flap of mantle muscle (arranged as shown diagrammatically) during hyperinflation (i.e. expansion of the mantle cavity beyond resting volume). The deflexions below baseline record contraction of circular fibres; prolonged upward deflexions, with subsequent return to baseline, record the activity of radial muscle fibres. They cause the mantle to thin, and to extend. Nerve supply to flap intact.

anterior and one in the posterior mid-ventral region, showed no change in the length of the mantle during respiratory movements and that more powerful contractions also had very little effect on its length.

Ciné film of the ventral aspect of the mantle during a jet cycle was analysed to observe dimensional changes in more detail, but no attempt was made to compensate for changes in the curvature of the mantle. The length of the mantle in any one region either remains constant or slightly increases during the development of the power stroke, a length increase of about 5% appearing only along the midline of the muscle. At the same time, width is reduced by nearly 20%.

These changes have been further demonstrated using an intact *Sepia* while jetting in air, by cutting a flap of the mantle so as to retain its innervation from the stellate ganglion. The preparation was fixed by pins through its distal margin in order to record changes in its length by means of an isotonic lever. The contraction and expansion of the mantle could thus be directly monitored during the jet cycle (Fig. 4). Recordings showed the resting condition of the circular muscles, their contraction at jetting and their sustained expansion from the resting length by a similar amount to the contraction.

After the expansion seen in Fig. 4 the mantle returns to its resting condition and subsequently contracts to produce a movement similar to the power stroke. This is followed by a second active expansion of the mantle and a return to the resting condition without subsequent jetting. Both relaxation phases have a quasi-exponential course lasting about 2.5 sec similar to the relaxation of tension in a catch muscle.

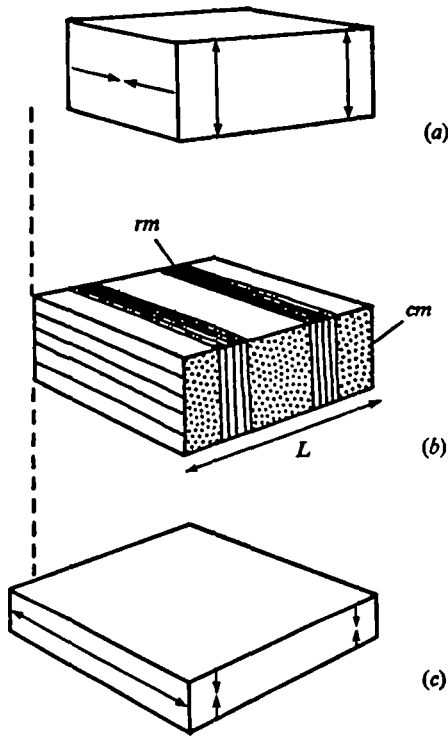


Fig. 5. Diagrammatic representation of the changes in shape of a block of mantle muscle in which one dimension (L) remains constant. (a) thickening as a result of contraction of circular fibres (cm) and passive sketch of the radials (rm); (b) resting condition; (c) thinning by contraction of radial fibres and passive sketch of the circulars. Resting dimension indicated by broken line, contraction and extension of muscles by arrows.

following rapid stretch (Rüegg, 1971). While we have no unequivocal records of the mantle flap preparation showing jetting from the stretched condition without initial return to the resting condition this may be shown in Fig. 3c where the anterior part of the mantle is dilated immediately prior to jetting.

Since the length of the whole mantle is almost constant during the jet cycle these changes in dimension can only be explained in terms of the circular and radial muscles and of the collagen fibres described by Ward and Wainwright (1972). The return of the mantle to the resting condition (Fig. 4) could be due to active muscular contraction, to passive relaxation brought about by the collagenous layer or to a combination of these two factors (see Discussion). Further investigations are required before this can be elucidated, but the experiment with the muscle flap (supported only along one edge and at one point opposite) is clear evidence that the mantle muscle can expand beyond the resting condition without external forces (hydrostatic or other). This prejet expansion of the mantle must be achieved by contraction of the radial muscle fibres; it leads to the thinning of the mantle block and stretching of the circular fibres beyond their resting length.

The changes caused by the two sets of muscles at constant mantle length are envisaged in a muscle block (Fig. 5). The mantle may be held at constant length by

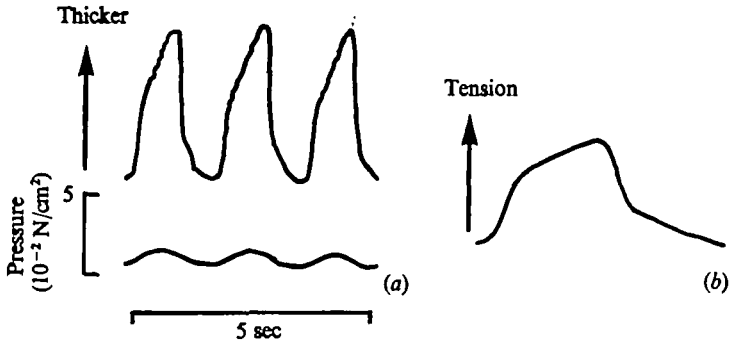


Fig. 6. Recordings of *a*, the contraction-relaxation of the circular muscles of the mantle of *Sepia*, in respiration (pressure trace below) obtained by use of button electrodes; and *b*, development of tension in the squid mantle due to stimulation of many small fibres of a stellar nerve, responses being due to shocks at 25/sec for $\frac{1}{4}$ sec (from Wilson, 1960).

two mechanisms: isometric tension developed in the longitudinal muscle and the presence of a trellis of collagen fibres. Although blood freely permeates the mantle the antagonism between the groups of muscle fibres must be largely due to their direct action upon each other rather than to their action being affected by the presence of a constant volume of body fluid, such as has been classically described in the fluid-muscle systems of worms and anemones (Chapman, 1958). The effectiveness of muscle antagonism in the cephalopod mantle leads us to suggest that the use of a body cavity containing fluid may only be necessary for force transference over relatively large distances or where a major change of shape occurs.

DISCUSSION

Data has been presented on three main kinds of activity: respiratory movements of the mantle, jetting and the expansion of the mantle which causes water to be inhaled. Respiration occurs rhythmically, with small pressure fluctuations within the mantle cavity produced by contractions in only the anterior half of the mantle. These are probably caused by small nerve fibre activity (Wilson, 1960). The generation of a high pressure pulse used in locomotion involves the whole mantle, the pressure being initiated in the posterior region by a giant fibre response. Transition from breathing to jetting is accompanied by a transition from small-nerve-fibre type of activity to giant fibre responses and a shortening in duration of the pressure pulse (see the records in Fig. 2*b* of Trueman & Packard, 1968).

These findings accord with the electrophysiological results obtained using isolated preparations of mantle muscle and stellate ganglion of squid (Prosser & Young, 1937; Young, 1938; Wilson, 1960). For instance, the area of mantle innervated by fine nerve fibres is very much smaller than that innervated by giant fibres and responses to electrical activity in individual nerve fibres can, in principle, be confined to small areas of the mantle; the tension and pressure curves obtained during quiet breathing in *Sepia* have the same shape as the tension curve obtained from repetitive stimulation of small nerve fibres in the isolated nerve-muscle preparation (Fig. 6) (Wilson, 1960).

The area innervated by giant fibres is large and not confined to one section of the

mantle; their motor units are segments radiating outwards from the stellate ganglion and include both anterior and posterior parts of the mantle musculature (Young, 1938). All of these giant fibres (= 3rd order giant axons) are innervated by one pre-synaptic 2nd order giant axon. A single shock to a 3rd giant axon leads to an all-or-none (non-facilitating, non-summing) response of the whole of the mantle musculature innervated by that fibre. The duration of the mechanical response (circa 0.3 sec), following a single shock, is similar to the duration of the developed tension and about twice as long as the pressure pulses, in our records of maximal pressure. The rise time of the mechanical response, during giant fibre stimulation, is of similar duration to the rise time to peak tension in our records and to maximum swimming velocity from photographic analysis of the squids escape response (Packard, 1969, 1972).

It is not certain that radial muscle fibres are actively contracting during the return stroke of the mantle muscle to resting length; the collagen network could be partially responsible for this although contraction of the radial fibres must cause hyperinflation of the mantle. Any electrical activity of the radial muscle sheets during jetting appears to have been swamped by the electrical activity of the circular muscles observed by Ward (1972). Simultaneous electrical activity in the nerves supplying the two sets of muscles during the power stroke (with the circulars over-riding the weaker radials) would still allow the radials to be effective antagonists to the circulars, if they have a longer relaxation time course than the circulars. This would be achieved by the presence of a larger tonic (or catch) component in these muscles. The prolonged return to baseline, after pre-jet expansion and before the onset of jetting (Fig. 4), provides some evidence for this. Alternatively, the return to resting length at the end of the power stroke could be analagous to the elastic recoil of the vertebrate heart during diastole, with the passively stretched radials providing the 'elastic' component, without the intervention of nerve activity (Rüegg, 1971).

The obliquely striated muscle fibres of cephalopods have still to be studied in the same detail as those in the body wall of nematodes and annelid worms (see Rosenbluth, 1967; Knapp & Mill, 1971). If phasic and tonic components in oblique fibres also occur in cephalopods, then such fibres would be particularly well suited to the requirements of a locomotory system that uses agonists and antagonists within a single block of muscle.

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