

STUDIES ON THE MYONEURAL PHYSIOLOGY OF ECHINODERMATA

III. SPONTANEOUS ACTIVITY OF THE PHARYNGEAL RETRACTOR MUSCLE OF *CUCUMARIA*

By W. POPLÉ

Department of Zoology, University of Natal, Durban

AND D. W. EWER

Department of Zoology, Rhodes University, Grahamstown

(Received 18 March 1958)

Spontaneous activity is a well attested characteristic of holothurian muscle (Hill, 1926; du Buy, 1936*a, b*; Wells 1942). Some preparations show regular rhythmical contractions and have been used to test pharmacological reagents (Lutz, 1930; Wyman & Lutz, 1930; Iriye & Dille, 1941), while in other preparations the spontaneous activity is so variable that this is not considered possible (Riesser, 1933; Bacq, 1939; Ambache & Sawaya, 1953; Welsh, 1954). For certain preparations there has been no record of spontaneous activity (Morin, 1931; Fischer, 1938; Steinbach, 1940; Galambos, 1941; Prosser, Curtis & Travis, 1951; Prosser, 1954; van Weel, 1955). The variations between different preparations suggest that spontaneous activity is not an inevitable property of holothurian muscle, and in fact Tao (1927) and Buddington (1937) have shown that such activity depends, in part, on the presence of intact nervous tissue. Buddington (1937) studied the cloacal breathing of *Thyone*, and showed that the characteristic patterns of the spontaneous contractions of isolated muscles of the cloacal complex relate to the inherent functions of each muscle in the behaviour of the animal.

The isolated pharyngeal retractor muscle of *Cucumaria sykkion* (Lampert) also shows contractions which are considered 'spontaneous' because they occur in the absence of any apparent external stimulation (Pople & Ewer, 1954). These contractions are of the same form and duration as those which pull in the tentacular crown of the entire animal. The experiments reported below show that these contractions are neurogenic in origin and may be reflexly elicited, but that their typical pattern depends upon the presence of a 'centre' within the ganglionic mass which forms the retractor motor complex.

ANATOMY OF THE PREPARATION

The following description is limited to those structures which have a direct influence on the activity of the retractor muscle and is not a complete account of the mechanisms associated with the pharyngeal complex of *Cucumaria*. When

C. sykion is feeding the crown of tentacles is extended, the retractor muscles (Fig. 1 *a*, *r*) are stretched and the pharyngeal portions of the longitudinal muscles of the body wall (*l*) are contracted. Withdrawal of the tentacles into the animal is brought about by a slow contraction of the retractor muscles which results in a folding in of the pharyngeal mass. A swift, partial withdrawal can also occur; this is brought about by an initial quick contraction of the retractor muscles and may or may not be followed by a slow, complete contraction of the muscle. When withdrawn the

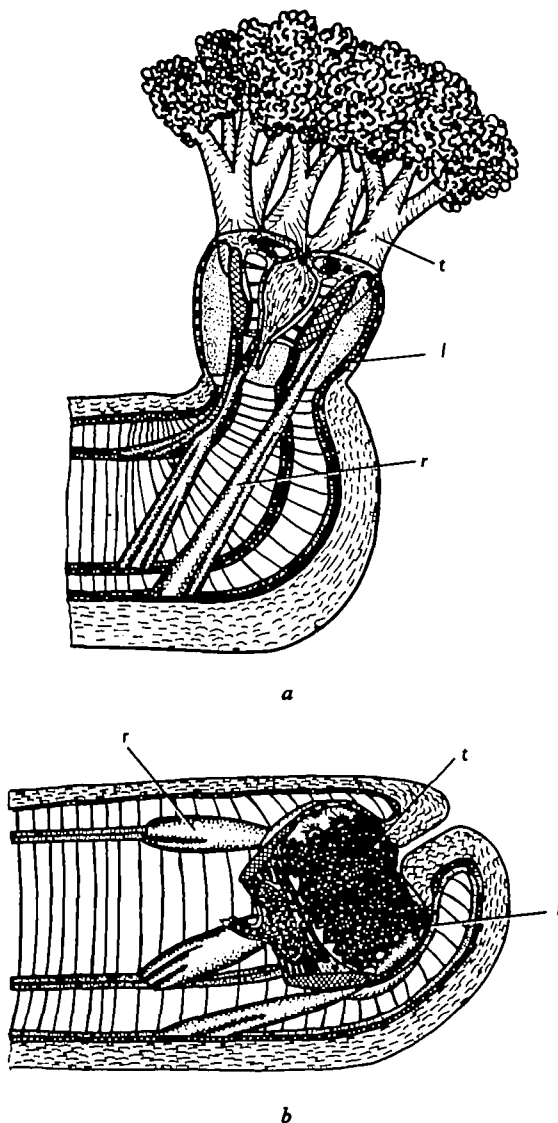


Fig. 1. *C. sykion*. Diagrammatic sections of the pharyngeal region to show relations of retractor muscles (*a*) with tentacles expanded, and (*b*) with tentacles retracted. *l*, Longitudinal muscle; *r*, retractor muscle; *t*, tentacle.

tentacles are compressed within the pharyngeal mass (Fig. 1*b, t*), the retractor muscles have contracted to a shorter length and the pharyngeal portions of the longitudinal muscles are stretched.

The standard preparation used in this investigation consisted of the mid-ventral ossicle with its attached retractor and longitudinal muscles, the two associated tentacles together with the portion of buccal membrane between them and the nervous supply associated with these structures. A diagrammatic representation of the afferent nerves to the retractor motor complex in this preparation is shown in Fig. 2. It has been assumed that severing the retractor nerve in the region 'a-a'' de-afferents the motor complex and that cauterization into the base of the retractor muscle, at the position of the motor complex, denervates the muscle. It has not proved possible to separate the radial nerve from the overlying longitudinal muscle, so that every reference to either radial nerve or longitudinal muscle applies to that structure in association with the other.

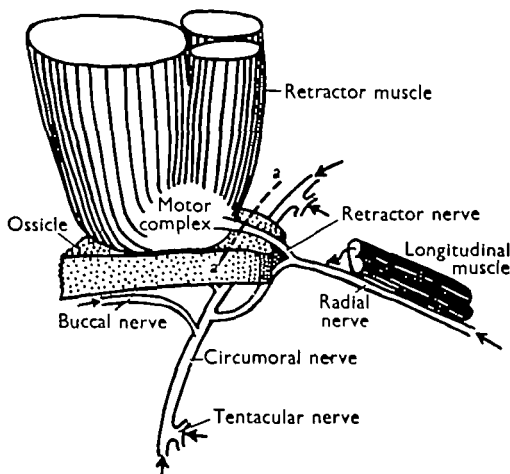


Fig. 2

Fig. 2. *C. sykkion*. Diagram of the motor complex at the base of a pharyngeal retractor muscle and its associated nerve supply. Arrows indicate the directions of afferent input to the motor complex. a-a', For explanation see text.

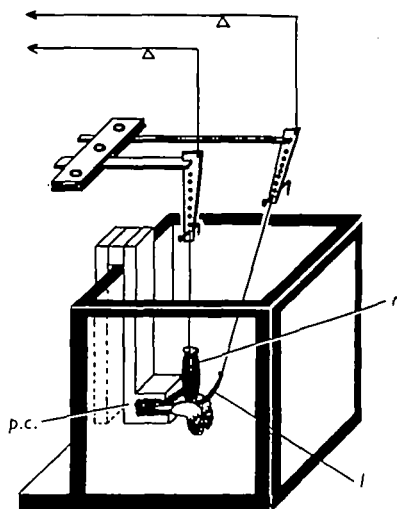


Fig. 3

Fig. 3. Diagram of a standard preparation of retractor muscle, *r*, and longitudinal muscle, *l*, set up in a Perspex trough to show the method adopted to apply constant stretches to the two muscles. p.c., Perspex clamp holding ossicle.

METHODS

The methods used were in general the same as those described in earlier papers in this series (Pople & Ewer, 1954, 1955), but modifications were made in the technique of holding the preparation. The most successful way was to grip the ossicle in a piece of foam plastic which was then squeezed into a slot in a vertical Perspex clamp (Fig. 3). This ensured that there was no pressure on either the tentacles or buccal membrane. To be able to stretch either the longitudinal or the retractor

muscle to a controlled length, terylene threads from the free ends of the muscles were tied to pins. Each pin was then slipped through one of a series of holes in a strip of celluloid fixed to a flat spring. By this means different stretches could be applied to either muscle. The contractions of the muscles, which bend the springs, were recorded in the usual manner.

The work of Millott (1954) has emphasized that echinoderm nerves are sensitive to direct stimulation by light. As will presently be seen, this is true for retractor-muscle preparations of *Cucumaria*. For this reason all experiments were conducted in a light-tight dark room; a heavily shaded neon light was used for short periods when it was necessary to check the progress of an experiment. When needed, a general low-intensity light stimulus was provided by a 40 W. fluorescent strip-lamp having an intensity at the preparation of 4–6 lumens/sq.m. A number of animals were dissected under a dim light known to be below the threshold intensity of light stimulation for the preparation; this was, however, discontinued because it was found that, after the first 2 hr., the behaviour of such preparations was no different from those dissected under normal illumination.

SPONTANEITY AND THE CHARACTERISTICS OF THE SLOW CONTRACTION

The most common 'spontaneous' contraction, which has occurred in more than 300 preparations used in this investigation, is a slow, complete shortening of the whole retractor muscle. These contractions may be considered 'spontaneous' because they can occur in the apparent absence of any immediate external stimulation. However, the expression 'spontaneous' has become associated with a number of different causal mechanisms: thus there may be 'internal causation' as in a pace-maker mechanism, 'unspecific causation' where there is general, low-intensity, afferent stimulation and 'unknown causation' with the implication that the activity is not explicable in terms of known reflex mechanisms. To avoid any of these implications the term 'slow contraction' rather than 'spontaneous contraction' will be used as far as possible.

The typical slow retractor contraction has two main characteristics which distinguish it from the quick (Q) and the delayed (D) responses previously described for this muscle (Pople & Ewer, 1954, 1955). First, the rate of development of tension (0.05 ± 0.03 g./sec.) is at least ten times slower than that of the Q and D responses, the contraction time at 20–25° C. being 50–120 sec. in contrast to 2–13 sec. for the Q and D responses. The tension developed by the slow contraction is maintained for 2–4 min. and is followed by an even slower relaxation; the whole event (Fig. 4) takes about 10 min. as compared with 1 min. for a Q and D response. Secondly, in a typical slow contraction, there is always a complete shortening and a maximal tension development, whereas the magnitudes of the Q and D responses are determined by the intensity of the stimulus which evokes them. This complete shortening is shown by the muscle when it is subjected to a stretch of a few mm. and to a tension of a fraction of a gram as much as when it is under the maximal stretch and tension that the muscle can withstand without damage.

At the commencement of a slow contraction the muscle shortens at a gradually increasing rate which gives a smooth sigmoid form to the resulting curve. This characteristic clearly distinguishes both the slow and D responses from the Q response which always commences contraction at its maximal rate and so is recorded as an abrupt step. In cases where the slow contraction is released by a specific stimulus it is possible to assess the latent period of the response. Owing to the gradual development of the contraction very exact measurement is not possible, but, by recording at high speed, latency values varying from 8 to 100 sec. have been found. The latent period of the D response is 3–4 sec. and is thus both considerably shorter and more constant than that of the slow contraction.

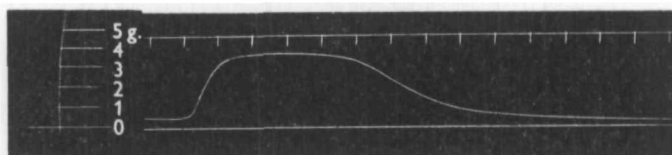


Fig. 4. *C. sykon*. A typical slow contraction of the pharyngeal retractor muscle. Temperature 23° C.; time marker, 1 min.

As well as being distinguished from the Q and D responses by its form, the slow contraction differs also in the way in which it is released. The Q and D responses are only obtained from the retractor muscle when the radial nerve is abruptly stimulated, either electrically or mechanically. Slow contractions can result from a number of other methods of stimulation which are described below, but they also arise in preparations which, left in the dark at a constant temperature and in conditions which exclude mechanical stimulation, are apparently unstimulated.

ORIGIN OF THE SLOW CONTRACTIONS

In order to locate the mechanism which controls the slow contraction, two types of preparations were used.

(a) *The denervated retractor muscle.* If the motor complex of one muscle of a five-muscle preparation is destroyed by cautery, that muscle will produce no further slow contractions even though the remaining four muscles continue to make slow contractions for 48 hr. Cautery of the complex does not destroy the muscle fibres for they will still respond to direct electrical stimulation. Again, if a preparation is treated with 10^{-8} curare (D-tubocurarine chloride, Burroughs Wellcome) the slow contractions cease, but they return after the curare has been washed out of the muscle. The curarized muscle will respond to direct electrical stimulation.

These results suggest that the slow contractions are neurogenic rather than myogenic in origin.

(b) *The de-afferented retractor muscle.* If the retractor nerve of a muscle showing rhythmical slow contractions is severed, the muscle ceases its activity. The ability to produce a slow contraction has not, however, been destroyed completely, for, if

the preparation is left for several hours, it may produce occasional slow contractions. It is possible also to release a slow contraction from such a preparation by a number of unspecific stimuli; for example, by general low-intensity stimulation by light, by draining and replacing the sea water around the preparation, which presumably produces a mechanical stimulation, and also by altering the ionic composition of the sea water.

From these results we may tentatively conclude that the mechanism producing the slow contraction lies in the motor complex, but that the normal occurrence of the contractions is dependent on afferent stimulation.

INHIBITION OF SLOW CONTRACTIONS

A preparation from which the tentacles and buccal membrane have been carefully removed still possesses a considerable afferent supply to the retractor motor complex from the radial and circumoral nerves. If such a preparation is left in the dark the retractor muscle will produce slow contractions for several hours, provided that the longitudinal muscle is not stretched. If, however, the longitudinal muscle is stretched the slow contractions of the retractor muscle cease. When the stretch on the longitudinal muscle is released, the slow contractions of the retractor return (Fig. 5). That stretching the longitudinal muscle inhibits the retractor slow

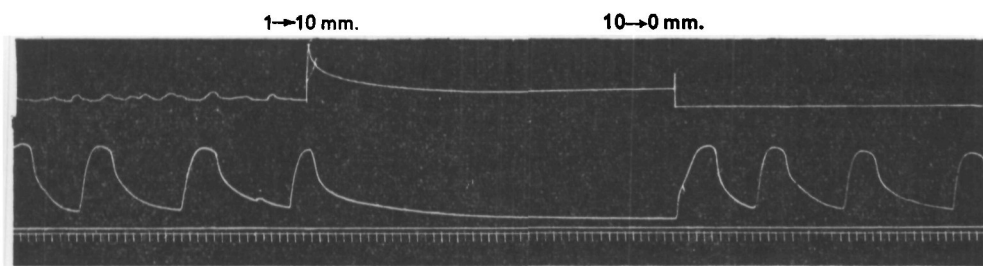


Fig. 5. *C. sykion*. Retractor and longitudinal muscle preparation without tentacles. Upper trace: longitudinal muscle; lower trace: retractor muscle. At the commencement of the excerpt the longitudinal muscle is stretched 1 mm. After 30 min. it is stretched to 10 mm. and after a further 45 min. all stretch on the longitudinal muscle is released. The sudden increase in tension in the retractor muscle as stretch is released is an artifact arising from the handling of the preparation. Time marker, 1 min.

contractions is not unexpected as these two muscles are functional antagonists. But it is also necessary to recognize that the occurrence and form of the slow contractions depend on a balance between excitatory and inhibitory inputs, for, if the longitudinal muscle is held at an intermediate stretch, patterns of partially inhibited slow contractions are obtained. These changed patterns will continue for periods longer than 12 hr. provided that the stretch on the longitudinal muscle is maintained and the changes from one pattern to another are always completely reversible.

The stages in the development of this inhibition are illustrated in Fig. 6. It will be seen that, for a stretch of 1 mm. or less, no change occurs, but that with greater stretches on the longitudinal muscle the pattern of the retractor contractions is altered. These changes are noteworthy. First, the frequency of the contractions is diminished; secondly, the duration of the contractions is prolonged; thirdly, the tension developed may no longer be maximal and finally the tension developed may vary markedly during the course of a contraction. It will be seen that at different stretches there is a close correlation between the duration of the contraction and the duration of the period between contractions. The uneven tension development, shown, for example, in Fig. 6 for a stretch of 3 mm., may perhaps be the expression of a varying balance of inhibitory and excitatory influences.

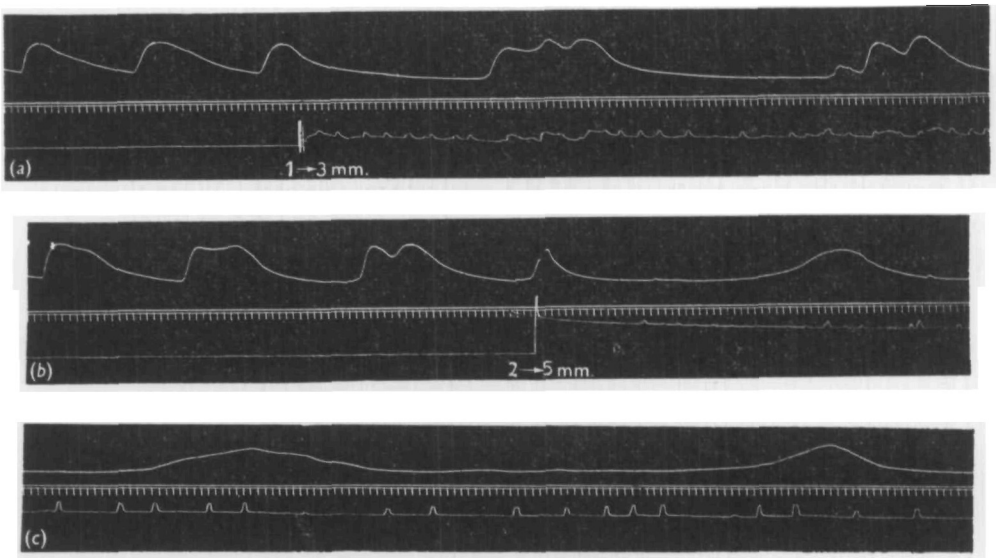


Fig. 6. *C. sykkion*. Retractor and longitudinal muscle preparation without tentacles. Upper trace: retractor muscle; lower trace: longitudinal muscle. Time marker 1 min. (a) Stretch on longitudinal muscle increased from 1 to 3 mm.; (b) stretch on longitudinal muscle increased from 2 to 5 mm.; (c) stretch on longitudinal muscle 6 mm. throughout. All extracts are from the same preparation.

Evidence for the total inhibition of an event as uncertain as these slow contractions must inevitably be negative and therefore unsatisfactory. But it is also possible to release slow contractions reflexly by stimulation of the tentacles. The reflex response thus produced contains a further element—a twitch—whose presence is witness to the occurrence of effective stimulation. For this purpose a preparation was used of the mid-ventral pharyngeal structures with the two tentacles attached on either side of the ossicle. Such a preparation shows not only slow contractions but also discrete twitches (Fig. 7a). During these twitches the muscle contracts at the maximal rate found during a slow contraction, but they show neither the initial gradual increase in tension nor the maintenance of tension characteristic of a

slow contraction. Unlike the typical slow contraction, the twitch does not usually involve more than 30 % shortening of the muscle. As the twitches only occur in preparations which have tentacles and buccal membrane intact, they are probably associated with excitation from the sensory tissues which these structures possess. The twitches can occur in the apparent absence of external stimulation, but are more frequent when a preparation is first set up and then become smaller and cease after a few hours. The magnitude of these twitches is variable, unlike the typical slow response.

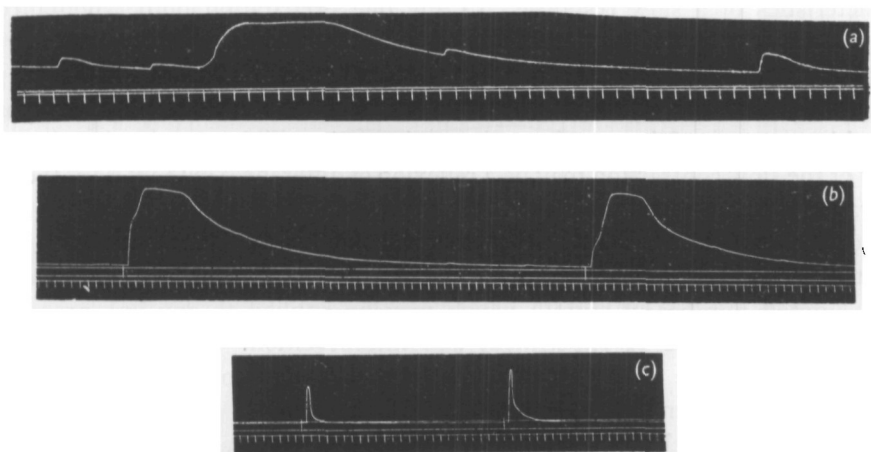


Fig. 7. *C. sykon*. Retractor and longitudinal muscle preparation with tentacles. Responses of the retractor muscle. Time marker, 1 min. (a) Spontaneous twitches and slow contractions with no stretch on the longitudinal muscle; (b) longitudinal muscle not stretched: responses to touching tentacle with a glass rod, first gently and then very gently; (c) longitudinal muscle stretched to 4 mm.: responses to touching tentacle first gently and then with violent rubbing.

If a tentacle is gently touched with a glass rod the retractor muscle will produce a twitch and, provided that the longitudinal muscle is not in a stretched position, this twitch will be followed by a slow contraction (Fig. 7*b*). If the tentacle is violently rubbed a number of times, the size of the twitch may be larger, but there is no difference in the size of the slow contraction produced. If the longitudinal muscle is stretched and the tentacle is touched or rubbed, then the retractor muscle will give a twitch, but it will not be followed by a slow contraction (Fig. 7*c*). Thus it can be seen that, although a gentle touch on the tentacle is sufficient to release a slow contraction, yet if the motor complex be in a state of inhibition due to the stretching of the longitudinal muscle, then even violent stimulation of the tentacles will not produce a slow contraction. It thus seems legitimate to conclude that stretching of the longitudinal muscle produces a real inhibition of 'spontaneous' slow contractions.

SLOW CONTRACTIONS RELEASED BY ELECTRICAL STIMULATION

Preparations can be obtained in which, for long periods, slow contractions do not occur 'spontaneously'. Two of the commonest used were: (a) a single retractor-muscle preparation with as little buccal sensory tissue as possible and with the longitudinal muscle stretched; and (b) a double retractor-muscle preparation pinned down with moderate pressure upon the tentacles.

In such preparations electrical stimulation of the radial nerve may be followed by a contraction of the retractor muscle which has, in general, the quantitative characteristics of a slow response. In any one preparation both 'spontaneous' and electrically released slow contractions are identical, but there is considerable variation in the quantitative details of a slow contraction from preparation to preparation. This variability is found, moreover, neither with the Q and D responses nor with responses of the muscle to direct stimulation. This suggests that the variability of the slow contraction may be a reflexion of differences in the state of the slow-contraction mechanism within the motor complex; this state will be largely determined by the amount of intact afferent tissue, both excitatory and inhibitory, which inevitably varies from preparation to preparation. This variability and the relatively limited number of stimuli which may be applied to any one preparation makes for difficulties of experiment and interpretation. The results which are reported below have been obtained from a sufficient number of preparations to warrant their acceptance.

A single 2 msec. electrical stimulus applied to the radial nerve can produce a typical slow contraction. The threshold for this slow contraction is always greater than that for both Q and D responses (Fig. 8), and thus it has not been possible by electrical stimulation to release a slow contraction alone. If single stimuli at a fixed voltage, just above threshold for the release of a slow contraction, are given at intervals during the life of a preparation, although the Q and D responses appear regularly, the occurrence or non-occurrence of the slow contraction is to a large extent unpredictable (Fig. 9). However, the longer the time interval since one slow contraction, the greater the chance that a threshold stimulus will release another. This effect is shown in Table 1, which summarizes the results from four preparations tested in this manner. This gradual lowering of threshold for discharge demonstrates a variation in the excitability of the slow-contraction mechanism with time. That this variation is a regular process in any one preparation may be shown by the use of single, regularly spaced test shocks above threshold voltage. In such experiments it is found that the time interval between slow contractions is almost constant in any one preparation, although with different preparations this period may vary from 20 min. to 1 hr.

It was considered possible that regular repetitive stimulation of the radial nerve might produce a more rapid development of the excitability of the centre within the motor complex and result in more frequently elicited slow contractions. To study this question series of stimuli were applied to the radial nerves of different preparations at frequencies varying from $2\frac{1}{2}$ /sec. to 1/10 min. and at various

intensities. After numerous fruitless experiments it became clear that it is not possible to 'prime' the slow-contraction system by way of the pathways in the radial nerve associated with the Q and D responses. Indeed, batteries of stimuli are no more effective than single stimuli in releasing slow contractions.

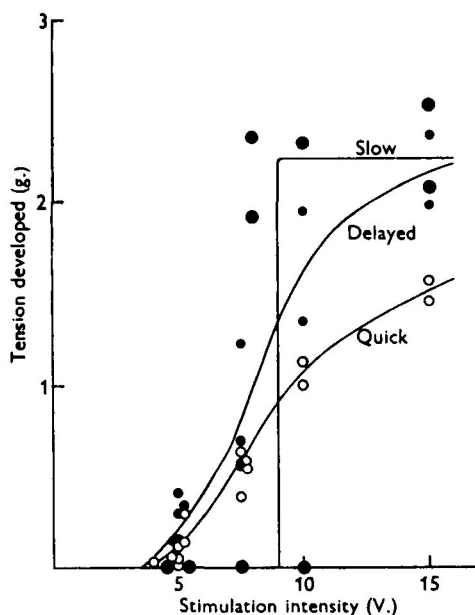


Fig. 8. Graph showing the relationship between the intensity of stimulation and the tension developed by the retractor muscle in response to single stimuli of 2 msec. duration. The Q, D and slow responses are shown separately. Small open circles: quick response; small filled circles: delayed response; large filled circles: slow response.

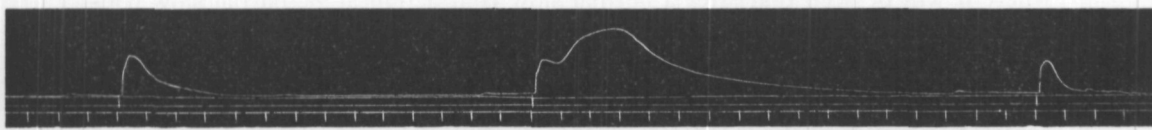


Fig. 9. *C. sykon*. Response of retractor muscle to three single 1 V. stimuli applied to the radial nerve. In all three cases a Q and D response are given, but a slow contraction follows only in the second case. Time marker, 1 min.

Table 1. *Effect of time since a slow contraction on the probability of releasing another by electrical stimulation at threshold*

Time since previous slow contraction (min.)	15-30	30-60	60 and more
Threshold stimuli producing a slow contraction	6	9	9
Threshold stimuli not producing a slow contraction	13	11	3
Percentage effective stimuli	32	45	75

Although the electrically released slow contraction usually has an 'all-or-nothing' character, yet partial or incomplete contractions did occur with rather less than 10% of the stimuli given in experiments studying conditions around threshold. Such partial contractions could not, however, be produced by careful grading of the intensity of the threshold stimuli. They reflect conditions within the motor complex and not the intensity of the releasing stimulus applied to the radial nerve. Similar partial contractions may be obtained if a preparation is stimulated at a voltage above threshold, provided the longitudinal muscle is stretched and, if the stretch be sufficient, the response may be completely inhibited. If, now, the intensity of the stimulus applied to the radial nerve is increased, a slow contraction may again be elicited. This parallels the condition found with 'spontaneous activity' of the retractor muscle, the slow contraction of which could be partially or totally inhibited by stretching the longitudinal muscle, and emphasizes that the activity of the afferent supply to the motor complex is as important in determining the character of the response released by stimulation of the radial nerve as it is for 'spontaneous' slow contractions.

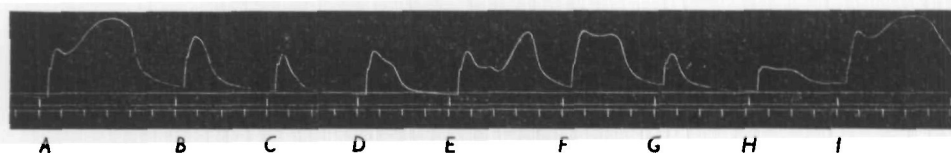


Fig. 10. *C. sykkion*. Response of retractor muscle to single 1 V. stimuli applied to the radial nerve at 4 min. intervals. Time marker, 1 min.

Without an apparent change in the background of afferent activity, partial slow contractions may also be elicited electrically in preparations from which typical slow contractions can likewise be obtained. Such contractions are shown in Fig. 10, where it will be seen that after a normal slow contraction at *A*, partial slow contractions were released at *D*, *E* and *F*, to be followed later by further partial contraction at *H* and a normal slow contraction at *I*. These results give the impression that, in certain circumstances, stimuli from the radial nerve may force the slow contraction centre of the motor complex to discharge prematurely.

While it has not been possible to show any consistent influence of stimulation of the radial nerve upon the character of the slow contraction, there are marked effects of the slow contraction upon the relative magnitudes of the Q and D responses. Preparations which show no 'spontaneous' activity and in which the threshold for an electrically elicited slow contraction is very high will produce Q and D responses of constant size to series of stimuli which lie below the threshold for the slow response. Sometimes there is a decrease in the size of the Q response with time, but this appears to be an accommodation effect at the point of stimulation. However, in a preparation giving slow responses, the Q and D responses preceding and following the slow contraction are modified. The commonest modification, seen in preparations making only an occasional slow contraction, is an increase in

size of the D response relative to the Q when the stimulus immediately precedes the slow contraction and the opposite effect 10 min. after the slow contraction. In other preparations, in which there are frequently occurring slow contractions, there

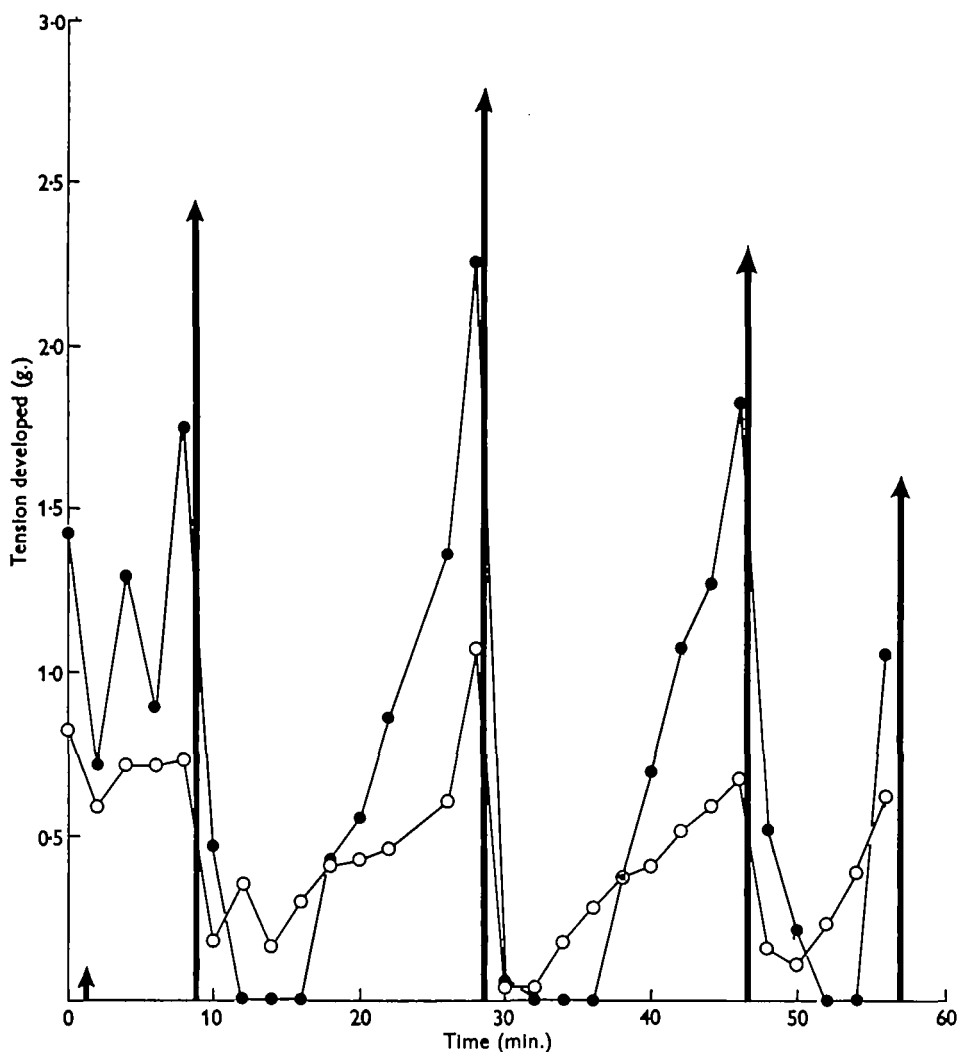


Fig. 11. Graph representing the tensions of the Q and D responses of the retractor muscle to a series of test shocks of 2 V. intensity and 2 msec. duration applied to the radial nerve at 2 min. intervals during a period of spontaneous activity. Open circles: quick response; full circles: delayed response. Arrows show time of onset of slow contractions: their height indicates the maximal tension developed by each contraction.

are increases in the absolute size of the Q and D responses until the slow contraction occurs and then there is a marked decrease following the contraction (Fig. 11). The D response is affected to a greater extent than the Q.

If the radial nerve is stimulated during a slow contraction, only minute Q and D responses will be elicited. This is a mechanical effect as the muscle has developed almost maximal tension due to the slow contraction. It is well known that histologically all fibres of this muscle are parallel, and one would therefore expect that, should the Q and D responses and the slow contraction use different final efferent pathways, simple algebraic summation of tensions would result. As this is not the case, it seems likely that all three responses have indeed common final pathways to the muscle.

The effects recorded with electrical stimulation of the radial nerve support the picture obtained from the earlier results, that the slow contraction mechanism is controlled by a system within the motor complex of the retractor muscle and that in this it differs markedly from the Q and D contractions which are immediate reflex responses arising from stimulation of the radial nerve, albeit the detailed characteristics of these responses may be modified by influences within the ganglionic mass of the motor complex. As the two systems probably have a common final path in the ribbon axons to the muscle fibres they must overlap. This overlap probably lies within the motor complex where intrinsic neurones are known to exist; these neurones have earlier been invoked to explain the characteristics of the D response (Pople & Ewer, 1954).

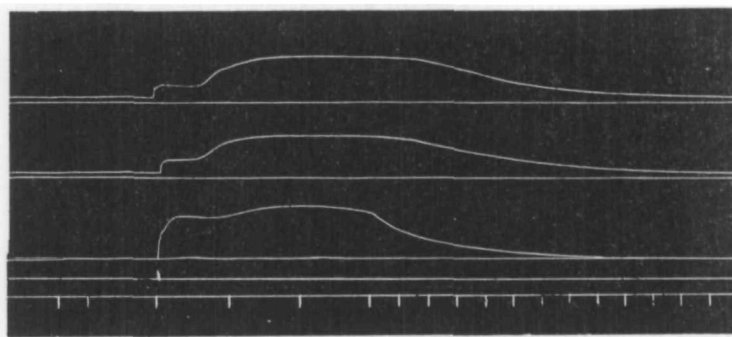


Fig. 12. *C. sykkion*. Extract from a preparation of five retractor muscles. Upper trace: right dorsal muscle; middle trace: right ventral muscle; lower trace: mid-ventral muscle. A single 2 msec. stimulus of 8 V. applied to the mid-ventral radial nerve at the time shown by the upper signal marker. Time marker, 1 min.

CONDUCTION AROUND THE CIRCUMORAL NERVE

It has been shown (Pople & Ewer, 1955) that, following stimulation of a radial nerve, impulses are transmitted to the retractor muscles of other radii. Except for a decrease in the tension developed the Q and D responses of a para-radial muscle are similar to those of the muscle whose radial nerve is stimulated. Slow contractions can also be elicited from all the muscles of a five-retractor-muscle preparation by the stimulation of any one radial nerve (Fig. 12). These slow contractions are of the typical 'all-or-nothing' character described above, and do not show the decrement around the ring found in the Q and D responses.

When a radial nerve is stimulated a slow contraction is not always obtained from all the muscles of a five-muscle preparation, although they all show Q and D responses. With subsequent stimulation it will not be the same muscles which fail to give a slow contraction. This failure of a muscle to produce a slow contraction is not due to a dying away of excitation around the ring, for the muscles furthest from the stimulated radius may produce slow contractions while those nearer do not. Thus the release by electrical stimulation of slow contractions around the ring does not depend upon the slow contractions of a neighbouring muscle nor merely on the impulses arriving in the Q and D system.

Table 2. *Times of commencement of slow contractions in different muscles of a five-muscle preparation relative to the occurrence of the 'leader' of each series*

(All times given in minutes. L indicates leading muscle. Arrows indicate possible path of excitation).

Time of occurrence of 'leader' contraction'	Interval between successive leader contractions	Pharyngeal retractor muscles				
		Left dorsal	Left ventral	Mid-ventral	Right dorsal	Right ventral
3.0	—	1.2	← L →	1.1	→ 1.1	→ 2.6
14.7	11.7	4.5	L	5.4	→ 3.2	→ —
31.5	16.8	4.1	L	—	→ 1.8	→ 1.9
47.8	16.3	17.5	← 16.8	← L →	→ 10.5	→ 16.4
95.8	48.0	7.5	L	8.6	→ 6.8	→ 6.0
151.3	55.5	→ 0.6	→ 0.6	—	→ 2.1	← L
176.7	25.4	1.8	← 0.9	← L →	→ 0.5	→ 0.6
206.7	30.0	—	—	3.6	← 1.7	← L
258.8	52.1	→ 1.4	→ 1.9	3.1	← 2.3	← L
320.1	61.3	L	→ 1.6	—	→ 5.0	→ 5.3
346.7	26.6	2.1	← 1.2	← L →	→ 1.3	→ 4.4
365.8	19.1	1.8	—	L	23.6	2.7
413.3	47.5	→ 0.6	→ 2.9	1.1	← 1.1	← L

This behaviour is paralleled in the activity of an unstimulated five-muscle preparation. When such a preparation is first set up the contractions of the five muscles appear to be independent and at random, but after about 3 hr. the contractions become less frequent and tend to occur in all the muscles together (Fig. 13). In Table 2 the times of commencement of spontaneous slow contractions are shown relative to the moment when the first muscle of a series contracted. It will be seen that usually, although not invariably, the muscles on either side of this 'leader' contract first and are then followed by the distal ones. This is similar to the mode of conduction of the Q and D responses round the circumoral nerve ring (Pople & Ewer, 1955). In ten preparations carefully studied there was no evidence for dominance by any one radius in this spontaneous activity.

A further study of Table 2 shows that the time interval between the occurrence of the slow contraction of the leader and that of either para-radial muscle is long and variable. The shortest interval that has been observed is 12 sec. and the longest several minutes. It seems unlikely that this represents the conduction time of impulses passing round the ring from the leading radius, and must rather be

regarded as a latency in the slow-contraction mechanism of the motor complex of the muscle showing the contraction.

The mode of circumoral propagation implies that some pathway exists which ensures that the discharge of the slow-contraction mechanism of one radius will trigger the discharge of those of other radii, provided their mechanisms are fully excitable. It would be difficult to explain the leader phenomenon in unstimulated preparations without this assumption. Further evidence in favour of this is obtained from a consideration of the size of the Q and D responses accompanying the electrical release of slow contractions in a five-muscle preparation. The Q and D responses associated with slow contractions in radii para-radial to that of the stimulated radial nerve may be smaller than those released by stimuli threshold for a slow contraction, when the para-radial nerves are themselves stimulated. Nevertheless the slow contractions are maximal and do not show the circumoral gradation of the Q and D responses. Moreover, in these conditions, particular retractor muscles may fail to give slow contractions, although the more distal ones do, confirming that the releasing impulses from the leading radius pass independently to all other radii.

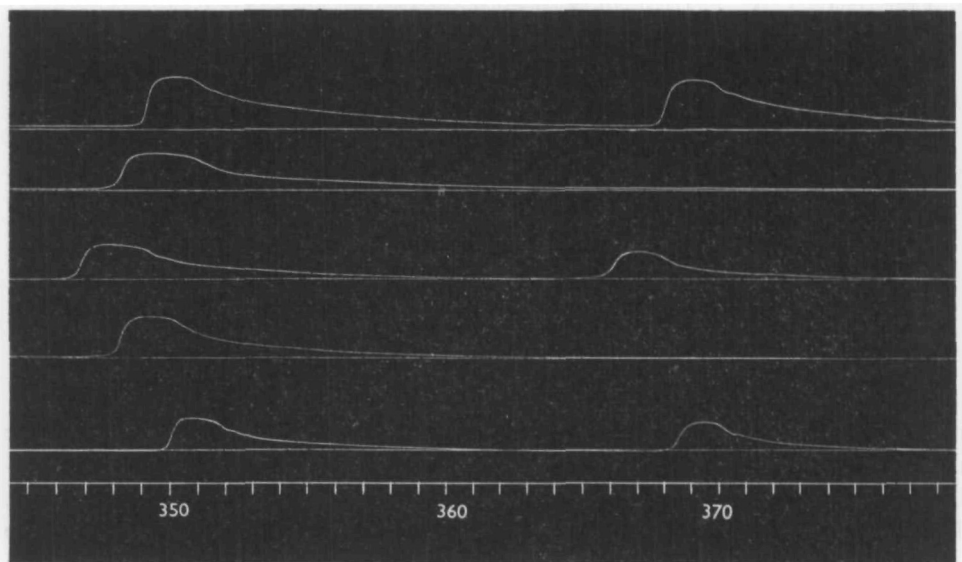


Fig. 13. *C. sykion*. Extract from a preparation of five retractor muscles showing spontaneous activity. Figures indicate time in minutes from setting up preparation. Time marker, 1 min.

DISCUSSION

Batham & Pantin (1954) and Ross (1957) have described the properties of the slow contractions of sea anemones. These show a number of striking similarities to the slow contractions described above, but there are also important differences. In both sea anemones and *Cucumaria* these slow contractions are obtained in response to electrical stimulation and to various unspecific stimuli such as light or touch; both display a long and variable latency; in both cases the slow contractions differ

from quicker contractions which are produced by the same muscle; in both the contractions are smooth and sigmoid in nature and they have very similar time relations: these latter similarities probably reflect the fact that in both cases the slow contractions will work normally against the pressure of a hydrostatic skeleton. But there are many important differences. In sea anemones the thresholds of quick and slow responses to electrical stimulation are the same, in *Cucumaria* they are markedly different; in sea anemones a number of electrical stimuli are required to elicit a slow contraction, in *Cucumaria* a single stimulus may be sufficient; in sea anemones the magnitude of the slow response may be graded by the number of electrical stimuli applied, in *Cucumaria* it is typically all-or-nothing; in sea anemones the maximal tension developed by the slow contraction is only about half that developed by the quick, in *Cucumaria* the slow contraction normally develops maximal tension; in sea anemones the slow contraction may alone be obtained in response to electrical stimulation, in *Cucumaria* it is always accompanied, in these conditions, by Q and D responses.

Batham & Pantin (1954) consider that the slow contraction of a sea anemone has its genesis peripherally, arising from a 'state of excitation in the muscle which, if it reaches a sufficient intensity, is followed after a considerable delay by a smooth contraction'. In *Cucumaria* our evidence suggests that the slow contraction has its genesis within the motor complex of the retractor muscle and not at the muscle or the myo-neural junctions. Moreover, although typically an all-or-nothing response, the slow contraction of *Cucumaria* may be graded, but this gradation appears also to be a function of the co-ordinating activity of the motor complex and not, in any simple manner, determined by the stimulus applied. The slow response is a fragment of the behaviour pattern associated with withdrawal of the tentacular crown, and its partial or complete inhibition by stretching of the pharyngeal portion of a longitudinal muscle may be correlated with the fact that these muscles are stretched when the tentacles are withdrawn and further activity by the retractor muscles is not required.

We have already suggested, in a consideration of the Q and D responses, that the motor complex of the retractor muscle is capable of modifying sensory input in the manner of the central nervous system. The present results emphasize that the motor complex may have an integrative role as well. It is possible at this stage to offer only a formal and hypothetical explanation of how this is effected and of the implications of the phenomena we have observed. It may be assumed that within the motor complex of the retractor muscle there is an arrangement of neurones capable of producing the programme of slow contraction; we may speak of this as the pattern centre (Fig. 14), and visualize it as normally in an excited state. This excitation will only be released by sensory input facilitating its passage through a 'releasing mechanism' or 'gate'. Also it is postulated that there is an inhibitory centre which is excited by the discharge of the pattern centre. The inhibitory centre has, further, certain characteristics such that its excitation only gradually dies away. It also acts upon the gate, but its effects spread beyond this and are responsible for the variations in magnitude of the Q and D responses before and after a slow con-

traction (Fig. 11). Such a system, given a steady input of sensory impulses, would produce rhythmic patterned responses. A marked fall in sensory input would result in occasional, random slow contractions, such as are observed in preparations where the motor complex is isolated. At the same time any sudden increase in sensory input might result in a full, patterned response provided the level of inhibition was low enough. But incomplete responses, reflecting some balance of excitation and inhibition, can be released by electrical excitation (Fig. 10). Further,

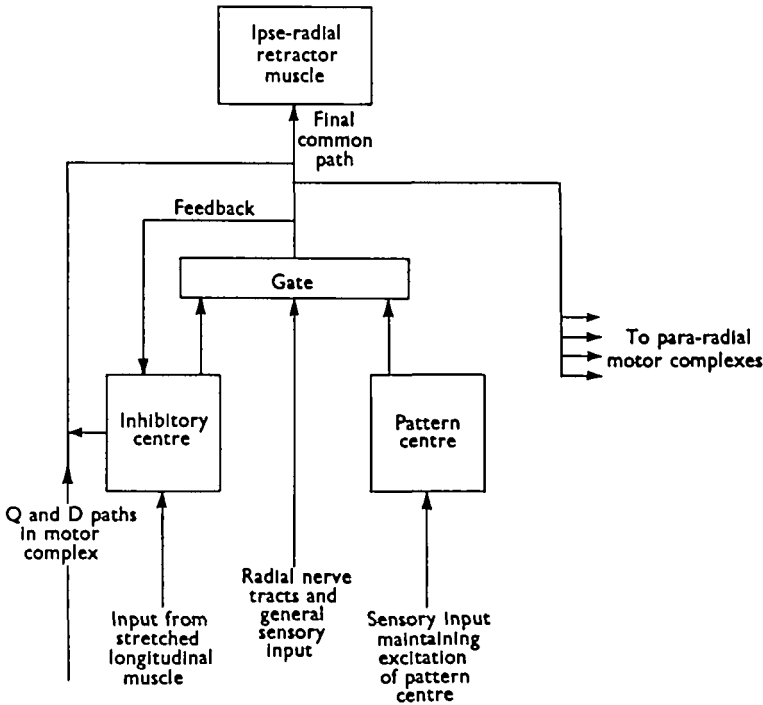


Fig. 14. Schematic representation of possible relations within the motor complex of a retractor muscle.

impulses arising as a result of stretching the longitudinal muscle can enhance the excitation of the inhibitory centre so that all activity is prevented (Fig. 5). When the resultant inhibition is only partial the duration of the slow contractions is prolonged, suggesting inadequate feed-back to inhibit the response rapidly. At the same time the frequency with which these contractions occur falls (Fig. 6), which may be due to the local and longitudinal inhibitions summing; the level of local inhibition must now fall lower than normally before excitation impinging upon the gate will allow discharge of the pattern centre. It is, however, clear that many different and more elaborate 'schemes' to explain the present observations may be developed.

The major role of the retractor muscle of *Cucumaria* is that of drawing in the tentacular crown. This is a complex pattern of activity involving several muscles

and, so far, we have examined only one element of this pattern. This slow contraction appears, however, to be a 'fixed action pattern' and shows some of the properties characteristic of endogenous movements (Lorenz, 1950) of higher forms such as fishes and birds. Since it can occur 'spontaneously', it is not a simple reflex response to external stimuli but can be regarded as showing 'internal motivation' after the manner of an instinctive movement. Spontaneous activity of a retractor-muscle preparation may be regarded, albeit crudely, as 'vacuum activity'. A phenomenon resembling the dissipation of action specific energy following the performance of a consummatory act is also shown. This can be seen in a preparation in which the general level of excitation is low, as judged by the lack of spontaneous activity; in such a preparation activity in response to external stimulation is followed by a refractory period. These slow contractions thus show on the motor side parallels with phenomena attributed to the accumulation and discharge of 'action specific energy' in instinctive patterns of higher forms. On the sensory side, however, the difference is vast. In *Cucumaria* completely unspecific stimulation, such as light or touch, will elicit the response. This is indeed hardly remarkable, for the complex patterns of releaser stimuli associated with innate responses in higher forms can only occur when there are the exteroceptors capable of pattern discrimination and such are absent in *Cucumaria*. Nevertheless, the similarities of motor behaviour which we have emphasized suggest that there may be here, in simple form, the type of unit upon which more complex innate patterns are built; that the activity of such units is capable of a relatively simple formal explanation which introduces no novel principle of nervous activity and more especially, need not involve the idea of the slow accumulation of excitation or 'nervous energy', and that further study of this and similar activities, especially in sedentary animals where perceptual complications do not enter, may well assist our comprehension of the more elaborate activities of more active animals.

SUMMARY

1. In the apparent absence of any immediate external stimulation the retractor muscles of *Cucumaria* show slow contractions which take 1-2 min. to develop fully. These contractions develop the maximal possible tension and are of an all-or-nothing character.
2. The contractions are neurogenic in origin, ceasing if the retractor motor complex is destroyed and being only very rarely shown by a preparation in which the motor complex is de-afferented.
3. The slow contractions may be partially or fully inhibited by stretching the longitudinal muscle.
4. Slow contractions may be released by electrical stimulation of the radial nerve. The threshold of stimulation is higher than that for the previously described 'quick' and 'delayed' responses.
5. The slow contractions released by electrical stimulation are normally maximal, but partial slow contractions may be released in certain conditions. Slow contractions are not more frequently released by repetitive stimulation of the radial nerve.

6. The occurrence of a slow contraction modifies the characteristics of the quick and delayed responses.

7. When the activity of all five retractor muscles is recorded, co-ordinated slow responses may be observed; in such responses excitation appears to spread from one radius and to be conducted independently to the other four.

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