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ESCAPE FROM RECURRING TACTILE STIMULATION IN *BRANCHIOMMA VESICULOSUM*

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The modification of behaviour by experience occurs throughout the Animal Kingdom. This is one of the fundamental facts of animal behaviour and has been the central preoccupation of a vast literature on animal learning, but it cannot be said that there is more than the slightest knowledge of the physiological changes which occur in animals during even the simplest learning process.

The discovery of phenomena such as subsynaptic depression, neuromuscular facilitation and fatigue, and post-tetanic potentiation help us to appreciate the potentially plastic properties of nervous material. But the evolution of an understanding of how learning does in fact occur demands the physiological analysis of behaviour patterns which are subject to systematic variation through experience in the life of the intact animal. Particularly appealing examples of such cases are the escape reflexes of various invertebrate groups; these reflexes almost invariably wane under repeated stimulation, and since they must be rapid, there is hope that they will also be neurologically simple.

There has been much work on certain aspects of these reflexes because of their frequent mediation by systems of strikingly large nerve cells which in their own right have been of great interest to neuroanatomists and physiologists. Physiological investigations of the plastic qualities of the reflexes in which these cells play a role have for the most part been incidental to general examinations of the systems' functional anatomy. However, in a number of species synapses with labile transmission properties apparently responsible for behavioural waning have been found (or inferred) and shown to be situated primarily afferent to (*Myxicola*: Nicol, 1948*b*; Roberts, 1962*c*. Crayfish:* Wiersma, 1947, 1961. Squid and cuttlefish:* Young, 1938, 1939; Wells, 1962), primarily efferent to (Cockroach: Roeder, 1948. Dragonfly larva:* Fielden, 1960), or on both sides of (Earthworm: Roberts, 1962*a*. *Nereis* and *Harmothoë*: Horridge, 1962) large, rapidly conducting neurons.

In the sabellid worms such as *Branchiomma* (Nicol, 1950), *Eudistylia* (Nicol, 1948*c*), and *Myxicola* (Nicol, 1948*d*) the escape reflex is a rapid withdrawal of the worm into its tube in response to vibration or to tactile or visual stimulation of the worm's protruded crown. In *Branchiomma* rapid waning of the response to shadows (Nicol, 1950) exemplifies the sort of behaviourally significant variation in excitability which seems to be typical of giant fibre reflexes. However, *Branchiomma*'s response to tactile stimulation, the most urgent warning of an approaching predator, has seemed to be an exception, for the worm has been said to escape repeatedly and without fail (Nicol,

* Probable on the basis of the references cited but not conclusively demonstrated.

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1950) from tactile stimulation of its branchial crown, and it has been claimed (Yerkes, 1906; Hargitt, 1909) that such stimulation following shadows can increase the excitability of the unstable shadow-escape reflex in a related form. These relationships make a comparison of the tactile and visual modes of input into a common giant-fibre system of considerable interest. The shadow-escape reflex has been studied in some detail by Nicol (1950). The present paper is concerned with *Branchiomma*'s escape responses to recurring tactile stimuli.

METHODS

Members of the species *Branchiomma vesiculosum* (Montagu) were collected from the Salcombe Estuary and kept until needed under circulation at the Plymouth Marine Laboratory. Worms sent to Cambridge from Plymouth seemed to survive and react satisfactorily in the Cambridge Zoological Laboratory's marine aquarium for many months, but some attempt was made to use the worms within a month or so of their delivery.

The behavioural observations (Figs. 1–3) described in § I were made at Plymouth. Twenty-three tubes known to contain worms were planted in individual, partly sand-filled jars which were kept under continual sea water circulation; water temperature varied between 16·5 and 17·5° C.

Uniform tactile stimulation was achieved by brushing a worm's crown with a horizontal glass fibre mounted on the end of a long, counter-weighted lever damped by a dashpot. The fibre started its excursion from a constant location just lateral to the base of the branchial crown and moved upward and medially at an angle of about 25° to the vertical with a speed of about 10 cm./sec.

The stability of a worm's responsiveness to this stimulus was tested by repeatedly applying it until the worm had entirely failed to respond on three (not necessarily consecutive) occasions; this criterion of reflex failure was chosen for reasons of experimental convenience. Stimulation followed 30 sec. after the worm's recovery from the previous stimulation which ranged from 4 sec. to over 18 min. but required a median of 20 sec. in fresh animals. Every worm was tested in this way once when 'fresh', particular care having been taken to disturb it in no way for 12 hr. prior to the test, and once again after a rest period to examine extent of recovery over time. Two worms which failed to emerge from their tubes had to be discarded. Judgement of the occurrence, completeness, speed, and latency of reflex withdrawal was purely subjective; judgements concerning speed and latency should be considered subject to substantial error.

Objective records of reflex activity were obtained from worms kept in stirred, aerated, room-temperature (17–20° C.) sea water at Cambridge. Giant axon (and unavoidably some muscle) potentials were recorded from pairs of bare 75μ silver wire electrodes threaded into *Branchiomma* tubes on alternate sides 1–2 cm. below the mouth; the potentials amplified by a Tektronix 122 preamplifier ranged from 10 to 50 μV. in the highly conducting medium of sea water. It should be noted that potentials cease to be recorded when a worm withdraws below the site of the electrodes. A photo-cell in the path of a light beam partially interrupted by the protruded branchial crown gave a record of crown withdrawal. In most of these observations

a tactile stimulus was supplied by a low-inertia electromagnetic prodder which established a maintained contact with the base of the branchial crown during a 1 mm. excursion which required only a few milliseconds for its completion; this gave a fairly discrete stimulus with respect to which latencies could be measured.

In some experiments *Branchiomma* was removed from its tube, pinned ventral side up on cork, and partly or completely covered with continuously aerated sea water. Electrical recording and stimulation were accomplished through a pair of platinum wires resting lightly on the worm's ventral surface. Movement was recorded by attaching the caudal end of a free posterior length of the worm to the light isotonic lever of a transducer whose output voltage was proportional to the lever deflexion. Square pulses of 0·25 msec. duration delivered through a radio-frequency isolation unit were used for electrical stimulation. In some experiments the giant-fibre reflex was elicited by touching the ventral surface of the thorax with a blunt glass fibre. For the experiment of Fig. 9 the fibre was made thin and flexible so that not more than 0·4 g. could be exerted, and the preparations were always rested for 30 min. before the start of an experiment.

For experiments on the giant motor axons worms were anaesthetized in a mixture of sea water and isotonic magnesium chloride, decapitated (to prevent repetitive giant-fibre discharge; see below), slit dorsally, pinned out to display the internal faces of the dorsal and ventral longitudinal muscles, and allowed to recover from anaesthesia in sea water; the undamaged gut was left *in situ*. Giant motor axon potentials were recorded through a 25 μ platinum wire fused into a glass capillary which was lowered on to the longitudinal muscle of the abdomen in the region of distribution of the giant motor axons; placement of the electrodes is critical. Stimulation by 0·1 msec. shocks was via a similar or slightly larger cathode. The positions of the stimulating anode and earthed neutral recording electrode in the sea-water bath were adjusted to reduce artifact.

Material for microscopic examination was fixed in Bouin, embedded by Peterfi's celloidin-paraffin method, and stained with Mallory triple stain. Material kindly loaned me by Dr J. A. C. Nicol, which had been fixed in sea water Bouin and stained with Palmgren's silver, showed the intersegmental giant fibre's branches into the neuropile especially clearly.

RESULTS

I. *Escape from recurring contact*

Gently brushing a fresh worm's protruded branchial crown causes a rapid and complete withdrawal of the worm into its tube. When the worm's crown again emerges, it can again be stimulated to withdraw, but as the cycle of stimulation, withdrawal, and re-emergence is repeated a stronger stimulus becomes necessary to prevent the waning of the response.

The waning of the response to a constant stimulus shows a rather uniform course of symptoms leading to complete failure to react in any visible way to the stimulus, but proceeding at a rate which varies widely among individuals. Generally, delayed withdrawal replaces immediate withdrawal, slow withdrawal replaces fast withdrawal, rapid re-emergence replaces slow re-emergence, and incomplete withdrawal replaces complete withdrawal. The development of these trends is illustrated in Fig. 1. Failure

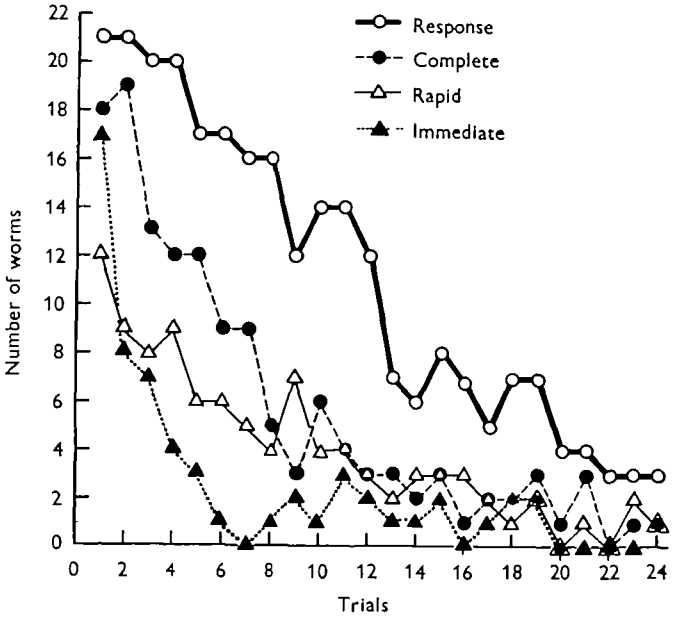

Fig. 1. The number of worms displaying various types of response to successive uniform tactile stimulations of the branchial crown.

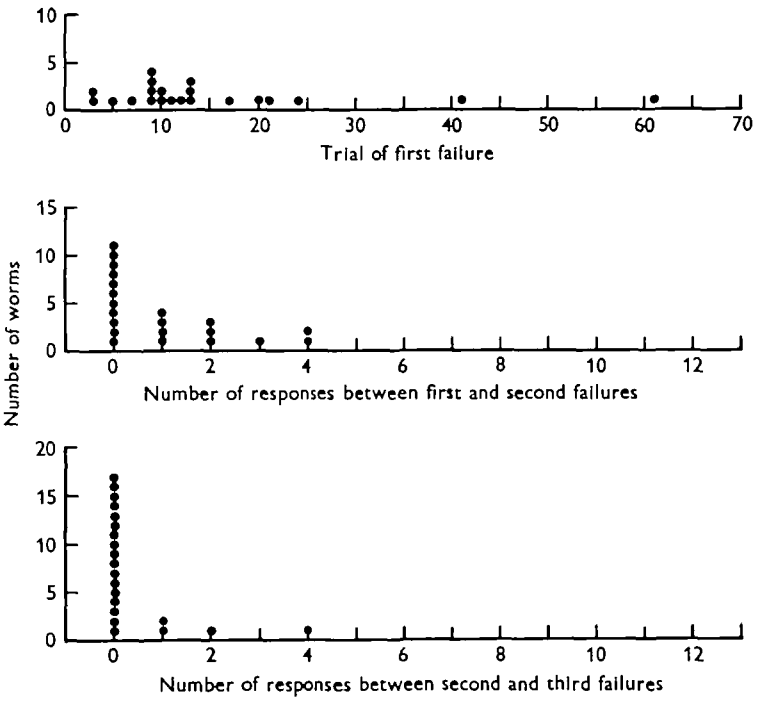

Fig. 2. The distribution of trials on which the first three complete failures to respond occurred in the series of Fig. 1. Note that the three failures tend to come consecutively.

to respond develops precipitously; once a worm has failed to respond to a stimulus, it is unlikely to respond to a subsequent one (Fig. 2). Recovery from this state of inexcitability is slow (Fig. 3).

We shall proceed to more detailed comments on the causes and implications of these behavioural events.

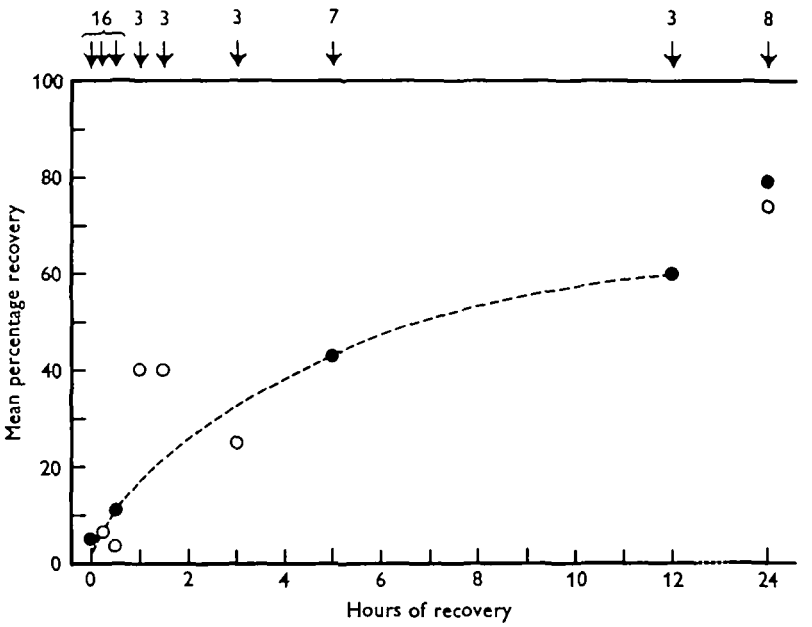

Fig. 3. Recovery of responsiveness after failure of the escape reflex following the series of Fig. 1. The number of responses made before the occurrence of three failures to respond was measured when each worm was fresh and again after a recovery period. The ratio of the two measurements (× 100) is the definition of percentage recovery. The numbers of worms tested are noted above the graph. Filled circles are based on observations from Plymouth as described under 'Methods'. Open circles, taken under less rigorous conditions at Cambridge, should be given less weight.

II. *The giant-axon system*

A pair of conspicuous giant nerve fibres run the length of *Branchiomma* above the other elements of the ventral nerve cord (Nicol, 1948*a*; Fig. 4). Electrical shocks applied to a worm's ventral surface excite the axons to produce all-half (see below)-or-none action potentials which propagate the extent of the worm at about 11 m./sec. and cause almost simultaneous contraction of the entire longitudinal musculature. The axons can follow such stimulation indefinitely at 40 shocks/sec., briefly at much higher frequencies, and are excitable by high voltages within 1–2 msec. after a previous shock. They are not a significantly weak link, in the reflex arc to be described.

By carefully adjusting the stimulating voltage near threshold the amplitude of the action potential can be halved; presumably one member of the pair of giant axons has ceased to fire. If stimulation rates are not excessive, spikes thus set up in one fibre are followed in 0·5 msec. or more by a second action potential also half full-size. These late potentials often occur while the directly excited fibre is refractory and sometimes

travel toward rather than away from the stimulating electrodes which therefore could not have initiated them; it is presumed that transmission from the directly excited to the originally silent axon has occurred (as shown for a number of other species by Bullock, 1953). Transfer of single spikes readily occurs in abdomen and thorax, but the processes involved are labile; cross-over in headless worms usually fails at frequencies less than 10/sec. Inter-fibre transmission in the head, however, is stable at frequencies in excess of 40/sec.*

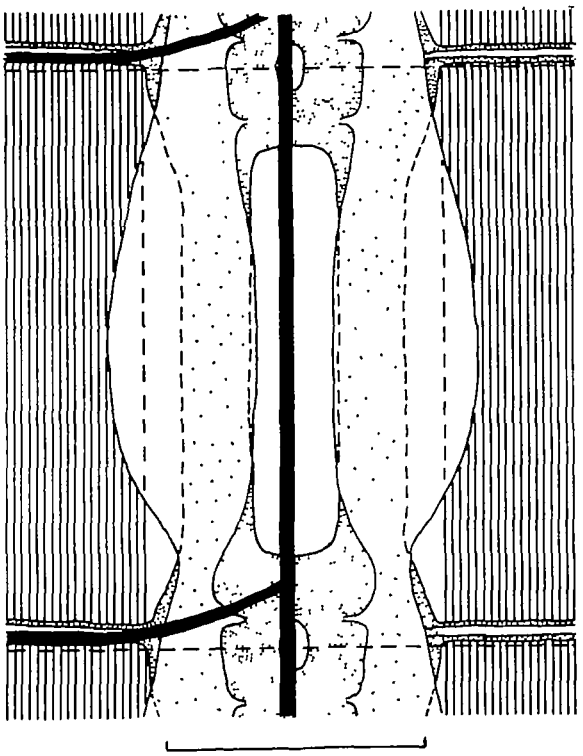

Fig. 4. Dorsal view of the nervous system in an abdominal segment of *Branchiomma vesiculosum*. The ventral longitudinal blood vessel and its segmental branches are shown in solid black on the left side. The giant fibres (clear) are shown running above the remainder of the ventral nerve cord (stippled) into whose ganglionic swellings they send ventromedially directed branches. A peripheral nerve which arises from the cord dorsally in the posterior part of each segment carries the giant motor axon; the nerve is shown running laterally over the dorsal face of the ventral longitudinal muscle (striped). No conclusions about the precise relationship of the giant motor axons to the giant fibres should be drawn from the diagram. Segmental boundaries are indicated by horizontal dashed lines. The diagram is schematic but approximately to scale; scale 400 μ.

* I have noted, with several other workers (Bullock, 1953; Kao & Grundfest, 1957; Kao, 1960; Wilson, 1961), the occurrence of multiple giant-fibre discharge in response to single direct shocks. It is my experience that: (1) such discharge generally descends from the head in one or both giant axons and is abolished by decapitation, (2) is not restricted to fresh worms and is not (generally) responsible for multiple responses to sensory stimulation though it occurs at a similar frequency, and (3) can be induced by 5 per sec. stimulation of the giant fibres for several minutes. In four worms an average of 4·8 min. of stimulation was required to produce multiple discharges. If stimulation was terminated after the first multiple discharge, only an average of 37·5, 13·5, and 11·6 sec. of stimulation was required to reinstate it at successive 30 sec. intervals. In one extensively tested worm this persisting 'facilitatory' state decayed with a half-time of at least 8 min.

The structural basis for cross-over is uncertain except in the head region where the axons communicate by contact though not fusion as they decussate in the supra-oesophageal ganglion (Nicol, personal communication). Though the axons send obvious branches ventromedially into the neuropile segmentally (Fig. 4), these become fine and are lost before their fates can be determined. If cross-over is due to a non-chemically mediated synapse or anastomosis, as appears to be the case in the analogous earthworm (Wilson, 1961) and crayfish (Watanabe & Grundfest, 1961) giant-fibre systems, the ease with which it may be fatigued in *Branchiomma* and other polychaetes (Bullock, 1953) is of considerable interest for an understanding of synaptic

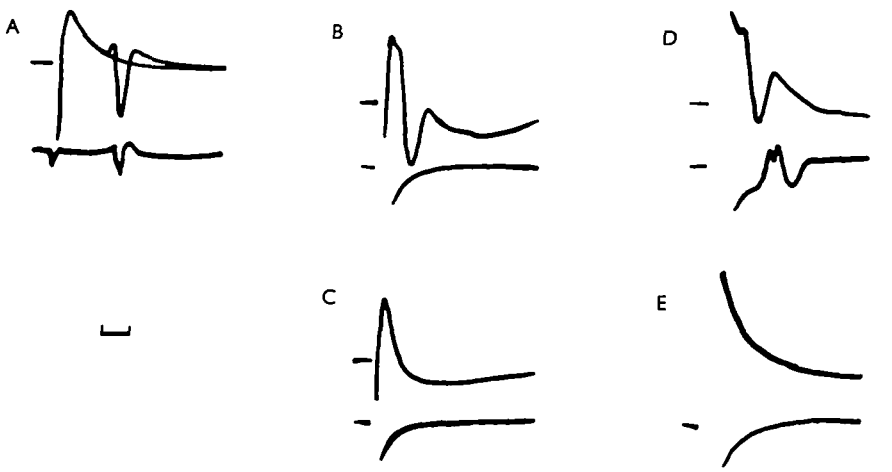

Fig. 5. Activity of the giant motor axon. Upper trace: a fine electrode on the dorsal longitudinal muscle of the abdomen in the region of distribution of a giant motor axon. Lower trace: electrodes recording activity of the intersegmental giants. A: an action potential which was sent down an intersegmental giant fibre 40 times/sec. passed first the segment of the upper trace and then that of the lower trace electrodes. This multiple sweep photograph was taken when the giant motor-axon potential became intermittent after 4 min. of perfect following (calibration 2 msec.); the muscle potential had completely fatigued. B–E: a different animal. B: the giant motor axon was stimulated directly in its course over the ventral longitudinal muscle; note that no potential is propagated into the intersegmental giant. D: when the ipsilateral intersegmental giant was stimulated, the action potential invaded the giant motor axon as it passed through the segment of the upper trace electrode and continued on to reach the lower trace electrodes; this demonstrates that all relevant conduction pathways were intact. C and E: stimulation just below threshold to show the stimulus artifacts in B and D respectively. Calibration 1 msec. tracings from oscillograph records.

lability. However, the occurrence of polarized fatigue of cross-over and of a 0·3–0·5 msec. apparent synaptic delay in *Eudistylia polymorpha* (Hagiwara, Morita & Naka, 1964) suggests that the giant axons of the *Sabellinae* may in fact associate in the body segments via polarized, chemically mediated synapses.

Out-of-phase spikes in the two giants are on occasion seen to result from tactile stimulation of the thoracic region (Fig. 7, e.g. D and H) which suggests that cross-over between the giant axons does in fact play a role in normal reflex functioning.

Sagittal sections of *Branchiomma* display in cross-section a nerve containing a large (10–30 μ) axon in the caudal part of each segment. This nerve (Fig. 4) passes across the internal surface of the ventral longitudinal muscle, mounts to the dorsal muscle

laterally, and runs medially on its internal surface. A fine platinum wire fused into a glass capillary picks up large action potentials travelling at about 2 m./sec. in the region of distribution of these axons whenever the ipsilateral (longitudinally running) giant fibre fires. Similar action potentials which are produced by shocks delivered to this region through fine electrodes are associated with twitches apparently restricted to the ipsilateral musculature of the stimulated segment and the one posterior to it. The anatomical relation of these giant motor axons to the longitudinally running, intersegmental giant fibres is uncertain; the most that I can say is that they come into fairly close promixity. That they are continuous with each other as Nicol (1951)

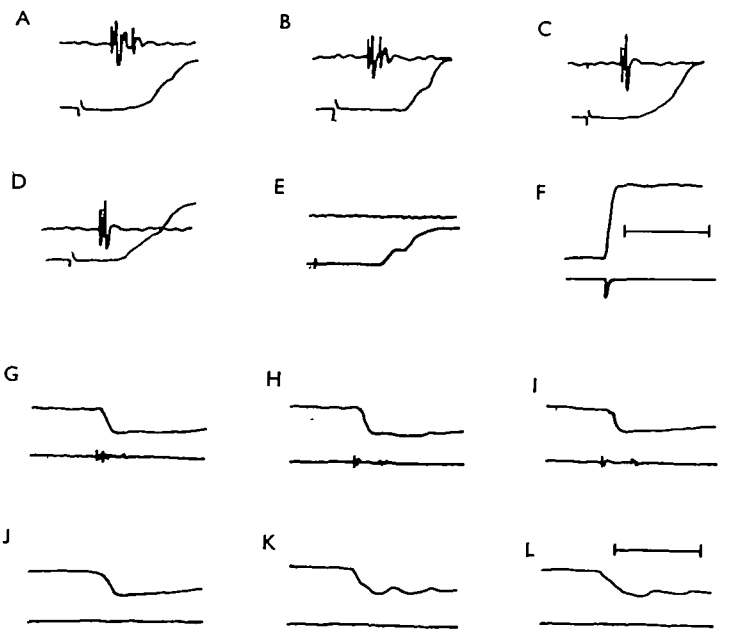

Fig. 6. Responses to touching the branchial crown. In records A–E the upper trace records giant fibre and muscle activity picked up by electrodes in the wall of the worm's tube; the lower trace records movement of the worm's crown preceded by stimulus artifact. Records G–L were taken from a different worm; the order of the traces is inverted; there is no stimulus artifact because the stimuli were applied by hand. A–C were complete withdrawals; D and E incomplete withdrawals; F is a photoelectric record of the prodder's 1 mm. excursion (upper trace). Calibration 50 msec. except in E where it is 250 msec. Tracings from oscillograph records.

stated on the basis of anatomical evidence seems unlikely, since action potentials set up in the giant motor axon do not invade the longitudinal giants (Fig. 5B–E). But other synaptic properties are not prominent. Transmission from longitudinal to motor giants is one-to-one and can be sustained for several minutes at 40 spikes/sec.; when after some minutes it does become intermittent (Fig. 5A), it may be due to conduction block in the motor axon, which has sometimes been seen to develop at these frequencies. A spike set up directly in a giant motor axon near its origin arrives at a fixed distal recording electrode 0·2–0·4 msec. before a spike set up about 1 mm. away in the ipsilateral intersegmental giant; one does not know how much time to attribute to nerve conduction, but transmission delay is clearly very short, perhaps too short

to allow for a chemically mediated synapse. In the present context the giant-fibre system may be considered as a functional unit, since action potentials are relayed from longitudinal to motor giants rapidly and reliably. But it should be remembered that the longitudinal giants may feed other motor pathways and the giant motor axons may be fed by other interneurones in a much less reliable manner.

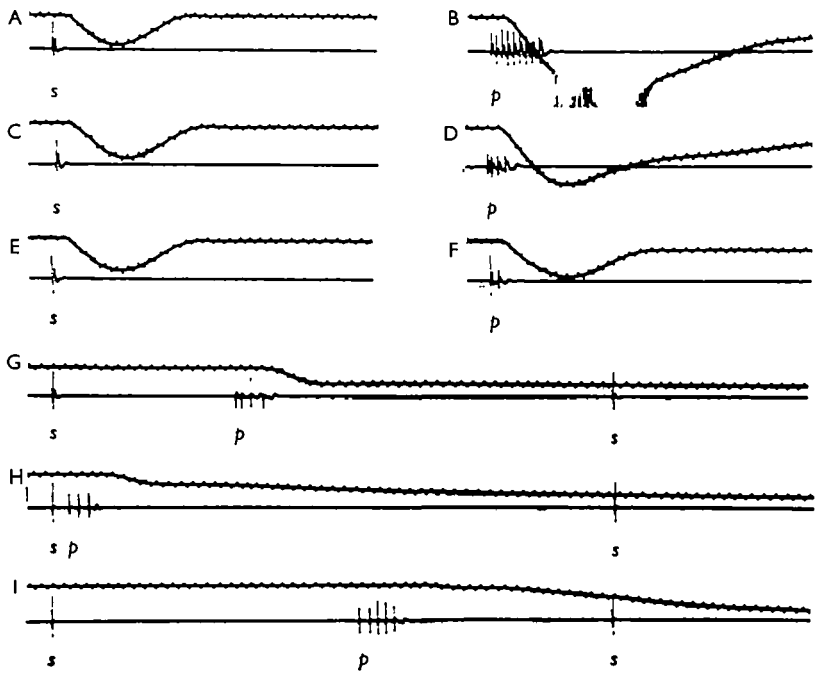

Fig. 7. Responses to direct giant-fibre stimulation and to prodding the thoracic body wall. Upper trace: contractions of posterior abdomen recorded by a light isotonic lever; 50 cycle/sec. periodic wave-form superimposed. Lower trace: giant fibre and muscle electrical activity. The records were taken sequentially from a single worm; an electrical shock to the giant axons is indicated by *s* and a prod by *p*. Records A–F were obtained at 2·5 min. intervals. Records G–I show responses to prods delivered during the 5th, 10th, and 20th minutes of continuous 1 per sec. shocks. Note that asynchrony of action potentials in the two axons is manifested at this slow film speed by small, thick spikes. Spikes in A–F retouched.

III. *The activation of the giant-fibre system*

Stimuli capable of causing rapid end-to-end shortenings of *Branchiomma* (shadows, vibration, touch of the crown, touch of the naked body) are all capable of exciting the giant nerve cells. However, their ability to do so is decidely transient. It varies according to the mode and strength of stimulus applied.

The immediate and smartly executed escape from contact shown by a fresh worm follows upon the start of a train of action potentials (at about 100/sec.) in its giant fibres (Figs. 6A and 7B). Such repetitive action potentials characteristic of fresh *Branchiomma* are not generally seen in *Myxicola infundibulum* (also of the family Sabellidae) which rarely gives more than a single impulse (Roberts, 1962*b*) but are common in *Nereis virens* (Horridge, 1959) and *Lumbricus terrestris* (Roberts, 1962*a*).

As stimuli are repeated, the giant fibres respond with a shorter train of action

potentials (Fig. 6A–C) and finally not at all. As is suggested by the occurrence of 'slow' withdrawals in behavioural records, escape from contact may occur without the accompaniment of giant fibre activity (Fig. 6E, J–L). Though usually relatively slow, such reactions on occasion approach the velocities of giant-fibre withdrawals (Fig. 6J), but their latencies are always greater than 40 msec., which is much longer than the shortest latencies of the giant-fibre reflex (Fig. 10). Thus, the adaptive value of this giant-fibre system may be more closely related to its potential speed of activation than to its ability to produce synchronous muscular contraction.

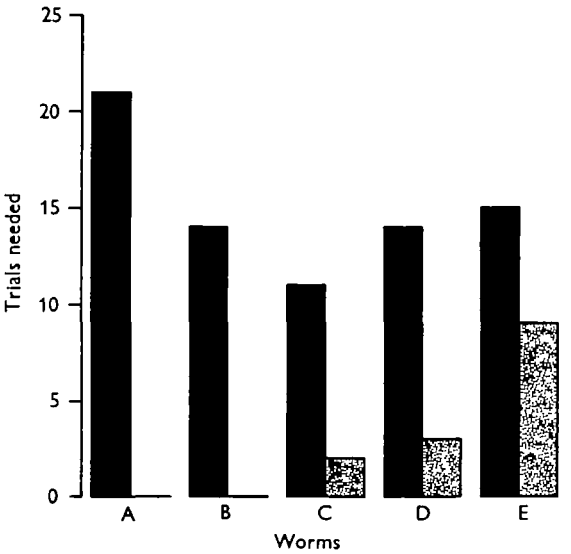

Fig. 8. Intertentacular transfer of reflex failure. For each worm the number of stimulations preceding two successive incomplete withdrawals is shown for stimuli applied first on one side (left bar) and then on the other side (right bar) of the branchial crown.

IV. *Some properties of the afferent-giant pathways*

Both central and peripheral factors might affect the reflex excitability of the giant nerve-cell system. Without records of the electrical activity of sensory axons of all sizes, which I have been unable to obtain, it is almost impossible to evaluate the extent of sensory-ending accommodation to repeated stimuli. However, the presence of transfer of reflex failure from a repeatedly stimulated to a previously unstimulated sensory field suffices to demonstrate the occurrence of transmission failure in a central portion of the reflex arc.*

Worms were repeatedly stimulated to withdraw by carefully bringing a glass rod into contact with the filaments of only one side of the branchial crown. Sufficient trials were given to bring the worms to a state where two successive stimuli failed to cause complete withdrawal; four more stimuli were given and the stimulation was then transferred to the other side, trials being continued until the worm again failed to give complete responses on two successive trials. Fig. 8, which presents the results of this experiment, appears to show that reflex failure can transfer to a previously

* Transfer might alternatively result from hormonal or centrifugal control of receptor sensitivity.

unstimulated sensory field. However, since it is difficult to be sure that the effective stimulus for withdrawal is not vibration or movement of the crown rather than *local* contact, and since the 'unstimulated' sensory field may in fact be stimulated by brushing against the wall of the tube, the experiment is not definitive.

These problems were circumvented by carrying out a similar experiment with the ventral thoracic body wall as a sensory field for the reflex. In six worms the second thoracic segment was prodded 45 times* at half minute intervals and the excitability of the reflex then tested by prodding the seventh segment; in six other worms the

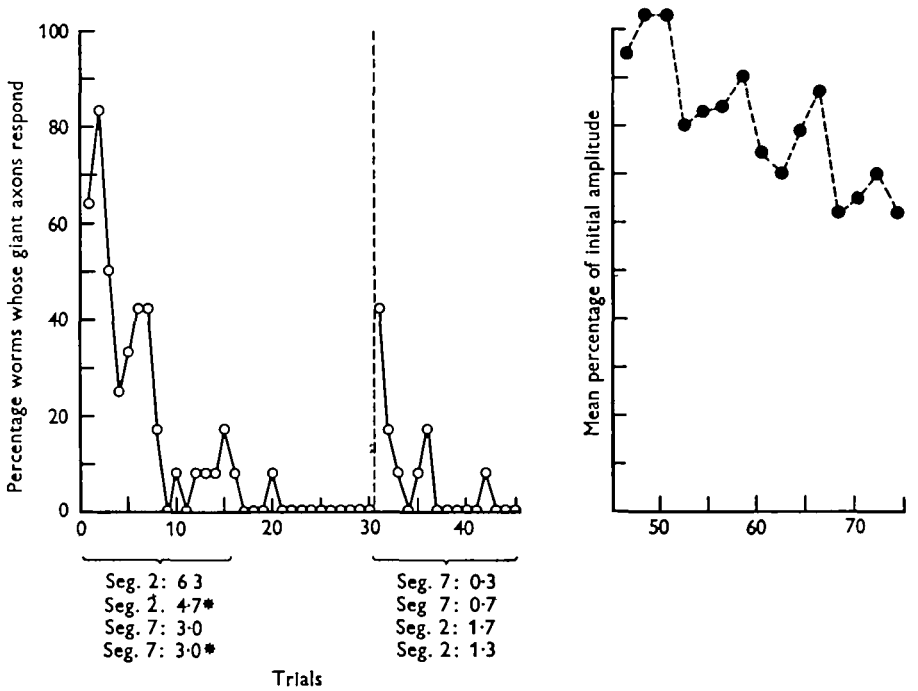

Fig. 9. Giant-fibre responses to repetitive prodding, and longitudinal muscle contractions in response to repetitive giant-fibre action. Trials 1–30: twelve worms were prodded at a given segment twice per minute; the giant-fibre reflex fails. Trials 31–45: the site of stimulation was changed to test for intersegmental transfer of reflex failure; the reflex partially recovers but fails more rapidly. Trials 46–75: the giant axons of nine of the worms were stimulated electrically twice per minute; rapid muscle twitches always occur but gradually decrease in amplitude; each point is the mean for two trials. Below the graph are given the average number of responses in each subgroup of three worms to the first fifteen prods of the first- and second-tested segments respectively; the asterisk indicates that the start of the experiment was delayed (see text).

reverse procedure was employed to control for gradients of sensitivity along the length of the worms. To control for the possibility of decay of the preparation with time, half of the worms were set up for kymograph and giant fibre recording 30 min. (the duration of the experimental procedures) before starting the experimental routine. The

* An attempt was made to limit the first 30 stimulations to one side of the segment so that transfer of reflex failure to the other side of the segment could be tested during the next 15 trials. But because it was technically difficult to limit the side of stimulation, the transfer of failure which occurred is of doubtful significance and only the first 30 of these trials have been plotted in Fig. 9.

results, plotted in Fig. 9, show clear but not total intersegmental transfer of reflex failure to a previously unstimulated sensory field. Every worm tested gave fewer responses to the 15 prods of the second tested segment than to the first 15 prods of the originally stimulated segment ($P < 0{\cdot}01$). The occurrence of such transfer demonstrates that a large component of reflex failure is due to central nervous events but does not rule out the possibility that sensory accommodation occurs as well.

Direct stimulation of the giant fibres does not appear to reduce their responsiveness

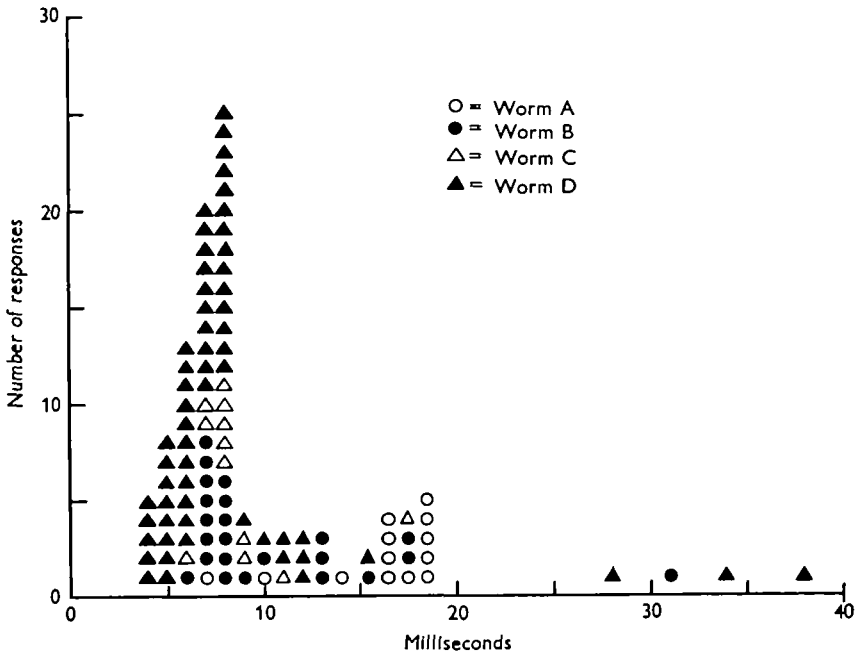

Fig. 10. Distribution of latencies of the giant-fibre reflex. The time required for touch of the branchial crown near its base to cause action potentials in the giant fibres was measured in four worms. The delays shown were derived from the observed latencies by subtracting 1 msec. (worm A) and 2 msec. (worms B–D) of giant fibre conduction time from the observed latencies. Two hundred and twelve records were taken at intervals of 15 or more minutes; the latencies of those responses which occurred during the 40 msec. oscilloscope sweep are shown.

to prods of the body wall (Fig. 7G–I). Thus, reflex failure is not attributable to a self-induced decrease of excitability by the giant-fibre system, nor is the information responsible for intersegmental (or presumably intertentacular) transfer of failure carried by these neurons.

These facts may be simply accounted for by postulating interneurons which collect from multiple sensory fields and transmit labilely to the giant neurons, but of course many other schemes can be imagined (note especially the possible relevance of Retzius's and Horridge's 'C' fibres; Horridge, 1963).

The latency of the giant-fibre reflex should be related to the number of synapses in the afferent-to-giant pathway and should therefore bear on the existence of the postulated interneuron(s). However, since there is no information on conduction time in ganglionic nerve, the shortest reflex latencies (Fig. 10) are not long enough to reject the possibility of a monosynaptic pathway.

V. *The activation of muscle by the giant axons*

A giant-fibre action potential invading a segment of *Branchiomma* is followed in about 2 msec. by a muscle potential which is in turn followed by a mechanical contraction. A light isotonic lever records the mechanical contraction at about 21 msec. after the peak of an electrically elicited spike in a worm removed from its tube. A photocell recorded withdrawal of a worm's crown from 10 to 42 msec. after the first of a train of synaptically evoked spikes; this variability in delay is presumably at least partly a function of the disposition of the animal's length within its tube and of the position of the photocell.

This sequence of events is relatively stable. Although direct stimulation of the giant fibres twice per minute yields gradually declining amplitude of muscular contraction (Fig. 9), stimulation received in the natural environment probably does not make such insistent demands on the motor apparatus. If the thoracic body wall is prodded at that frequency, the giant-fibre response rapidly fails (Fig. 9) and thus protects post-giant fibre structures from further stimulation (at least via the intersegmental giants). Whether strong stimulation of the branchial crown could maintain giant-fibre action at a frequency of 2 per min. is problematical, because such stimulation would ordinarily cause the worm to withdraw into its tube for more than 30 sec. Thus it would seem doubtful that failure between the giant-fibre system and longitudinal musculature ever has more than a slight modulatory effect on the 'strength' of the escape reflex.

The isotonic contraction to single giant-fibre spikes is submaximal, and the shortenings due to successive spikes of a train summate (Fig. 7B, D and F; Nicol, 1951). This summation is necessary inasmuch as single giant-fibre spikes may cause rapid but incomplete and therefore biologically inadequate withdrawals of a worm into its tube (Fig. 6D). Apparently abortive withdrawals do not occur in *Myxicola*, which therefore has no need for multiple spikes (Roberts, 1962*c*).

DISCUSSION

As Nicol (1950) pointed out, tactile and visual stimulation differ profoundly in their ability to excite the giant-fibre system of *Branchiomma*. The worm can almost always be induced to escape from tactile stimulation if it is made sufficiently strong, but this response like other rapid escape responses wanes reversibly under repeated stimulation of fixed intensity. As in other cases that have been studied the waning is largely a central nervous phenomenon, due neither to sensory accommodation (alone) nor to muscular fatigue (Roberts, 1962*a*; Horridge, 1959; Roberts, 1959; Roeder, 1948; Fielden, 1960) and presumably serves to allow the animal to use its musculature for activities other than escape in the prolonged presence of turbulent but non-malevolent conditions. These uniform labile properties are plausible if not necessary attributes of a successful escape reflex and consequently cannot be used to suggest the homology of the responsible structures. Indeed, it is considered very likely that giant-fibre systems have evolved independently on many occasions in the various phyla, and extreme morphological variations have made it impossible to argue for the widespread homology of these systems even among families of polychaetes (Nicol, 1948*a*).

Investigations of synaptic lability in giant-fibre systems have disclosed differences in functional organization which may be accidental variations resulting from independent evolution, but which may ultimately be recognized as significant adaptations to special problems of survival. *Lumbricus terrestris*, *Nereis virens*, *Myxicola infundibulum* and *Branchiomma vesiculosum* all use their giant-fibre reflexes for withdrawing into burrows or tubes. In all of them transmission from sensory nerves to intersegmental giant axons is labile and fails after some repetitions of the stimulus; the significance, if any, of this uniformity is unknown, but it contrasts with the extreme between-species differences which are found in the transmission between intersegmental giant axons and motor nerves. In *Lumbricus* (Roberts, 1962*b*) action potentials are transmitted very labilely from giant axons via largely unknown structures to motor axons which are not notably large. In the errant polychaete *Nereis* (Horridge, 1959) transmission between intersegmental giants and the musculature is less labile (insofar as comparisons between different experimental conditions can be made) and is believed to occur via an axo-axonic contact between intersegmental giants and relatively large 'giant-like' motor axons. In the sedentary polychaete *Branchiomma* transmission between intersegmental and motor giants is very stable but has some synaptic properties (at least polarity), while in *Myxicola* (Nicol, 1948*b*) the giant motor axons are in fact branches of the intersegmental giant fibre. These illustrations suggest speculation on associations between stability of intersegmental giant-to-motor-axon transmission, mode of life (sedentary or errant), motor axon diameter, and familial relationship. These factors are too well confounded to allow the statement of any hypotheses, but it is going to be interesting to examine giant-motor transmission in *Eudistylia* which has a systematic position (Hartman, 1938), gross nervous morphology, and tubicolous habit very similar to that of *Branchiomma* but which does not appear to have notably large motor axons (Nicol, 1948*c*).

Such description of functional organization is justified in itself, but it is to be hoped that it is also a preliminary to analyses of the mechanisms responsible for the occurrence of reflex failure. There are in principle many ways that such failure might be accomplished; for example, transmitter release from presynaptic fibres might decrease, post-synaptic membranes might become desensitized, conduction blocks might develop in exceedingly fine axon branches, pre- or post-synaptic structures might be inhibited, tonic facilitatory activity might be reduced. There is of course no reason to believe that but a single mechanism operates in any one case, and independent evolution promises the possibility of different mechanisms in different animal groups.

SUMMARY

1. *Branchiomma*'s rapid escape from tactile stimuli is mediated by the pair of giant nerve axons which run the length of the body above the ventral nerve cord.

2. The giant neurons are connected by very stable, polarized junctions to giant motor axons.

3. The giant-fibre escape reflex fails if tactile stimuli are repeated; a non-giant system which continues to cause slower escape eventually fails also.

4. Recovery from reflex failure is slow.

5. The failure of the rapid escape reflex occurs prior to the giant fibre. It is not primarily due to sensory ending accommodation. It cannot be caused by direct stimulation of the giant fibres.

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