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CONDUCTION ALONG THE VENTRAL NERVE CORD OF A HEMICHORDATE WORM

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INTRODUCTION

Although there is general agreement that hemichordates are more closely related to echinoderms and chordates than to any other groups of animals, their nervous systems appear to be no more highly organized than those of the coelenterates. Nerve fibres in enteropneust hemichordates form an intra-epithelial net, spreading extensively over the surface of the body and along the anterior part of the digestive tract. Hess (1937) and Silen (1950) found an internal nervous system, but the claim has been disputed by Bullock (1945) and Knight–Jones (1952). A few fibres have been seen to cross the basal lamina as individual strands, presumably to innervate muscle fibres. The only modification of this diffuse plexus is a concentration of nerve fibres to form dorsal, ventral, collar and, in some species, proboscis nerve cords. Bullock (1945) states that the cords, particularly the internal collar cord, appear to be primarily conduction tracts with little neuropile and cannot be considered to be homologous with chordate central nervous systems. Further evidence for the relatively unsophisticated nervous organization in these animals is the absence of sense organs, the presence of more sense cells than interneurones, and the lack of complexity in behaviour (Bullock, 1940).

Studies of the functional organization of the nervous system are few and based on deductions gained from analyses of muscular or ciliary activity (Bullock, 1940; Knight–Jones, 1952) or of luminescent responses (Baxter & Pickens, 1964). This paper presents the first recordings of muscle and nerve potentials from enteropneusts and decribes conduction and integration along the ventral cord.

MATERIALS AND METHODS

Worms were collected from subtidal sand flats in Kaneohe Bay, Oahu, Hawaii. Although potentials can be recorded from fragments of worms, the nerve cords in these fragments are often damaged. Accordingly, all experiments were performed on whole animals or on sections cut from whole animals. For recording from the ventral cord, the proboscis was removed and a slit was made along the mid-dorsal line from the posterior tip of the trunk up to and across the collar. This eliminated the dorsal cord as a longitudinal conduction pathway and the proboscis as a trigger for peristaltic waves. The close proximity of nerve fibres to muscle precluded freeing the ventral cord of other tissue by dissection. The worms were pinned to a pan of wax, ventral side up, and covered with sea water.

Suction electrodes with minimum tip diameters of 200 μm were attached to the cord.

Tips of smaller size were difficult to keep free of mucus. Leads from silver wires inserted into the sea-water-filled electrodes were connected to AC amplifiers and a dual-beam oscilloscope. Techniques for recording with suction electrodes are described by Josephson (1965) who, with Hoffman *et al.* (1959), discusses some of the difficulties and artifacts normally encountered. Enteropneusts are fragile worms, but suction electrodes can be employed routinely provided low negative pressures are used. Experiments early in this study showed that excessive suction was as effective as a cut across the cord in permanently blocking conduction. The consistent responses to electrical stimuli in all subsequent experiments indicate that damage caused by suction was minimal or non-existent. Single or multiple shocks were delivered through another suction electrode of slightly greater diameter.

Because enteropneusts show great changes in length, it is difficult to define resting length and, consequently, to give exact values for any properties, such as conduction velocity and decay of spike size, that are a function of length. For the most part, worms chosen for these experiments were 4–5 mm in diameter across the collar and 10–12 cm long when they were lying undisturbed in a dish of sea water at 25 °C. The diameter of the cords in such worms was about 200 μm, or approximately that of the tips of the electrodes.

RESULTS

The origin of potentials recorded from the ventral cord

When stimulating and recording electrodes are in the ventral cord, a weak shock will elicit one or more pulses, 20–50 μV in height. Although muscle fibres underlie the cord and the latter is embedded in epithelial tissue, these pulses are considered to originate from nerve fibres for the following reasons. First, weak shocks evoke pulses but no movement of tissue around stimulating or recording electrodes. Secondly, pulses are recorded only when the electrode is on the cord; if it is only $\frac{1}{2}$ mm lateral to the cord, no potentials are seen. Thirdly, pulses could originate from the epithelial tissue in which the cords are found, but this is discounted, in spite of evidence that epithelial tissue acts as a conducting pathway in hydrozoan coelenterates (Mackie, 1965; Mackie & Passano, 1968; Josephson & Macklin, 1969), because pulses cannot be recorded lateral to the cord from epithelial tissue similar in gross appearance to cord epithelium. Fourthly, conduction of small pulses is blocked by a cut across the cord or by an adventitious break in the cord even though adjacent epithelial and muscular tissues are undamaged. Fifthly, stronger shocks evoke larger pulses from the cord and body wall. In all probability, these are muscle potentials because they occur just prior to contraction and not during relaxation, disappear rapidly with repetitive stimulation or in the presence of excess magnesium ion, and are of longer duration, appear after a longer latent period, and have higher thresholds than the small pulses.

Conduction velocity along the ventral cord

The shapes of the small pulses vary with the distance between stimulating and recording sites. However, by measuring the latency between the first pulses recorded from two widely separated electrodes, velocities along the fastest conduction pathways can be determined. In a worm that is neither severely contracted nor completely relaxed, the conduction velocity along the anterior ventral cord is the same in both

directions (Fig. 1) and approximately twice that along any part of the posterior cord (Fig. 4D) except the most posterior tip of the trunk where velocity falls off sharply. For example, in 11 worms, whose resting lengths differed by no more than 15%, average conduction velocities along the anterior, proximal posterior, and distal cord

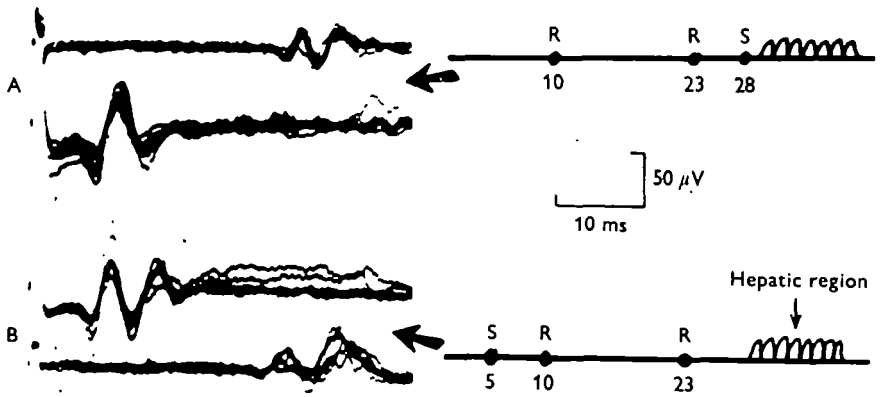

Fig. 1. Conduction velocity along the anterior ventral cord toward (A) and away (B) from the collar. The line drawings to the right of the superimposed oscilloscope traces show the positions of the stimulating and recording electrodes in relation to the hepatic region. Numbers indicate the distance in millimeters from the posterior edge of the collar. Eight shocks were given for each position of the stimulating electrode, but only responses to the last six shocks were recorded.

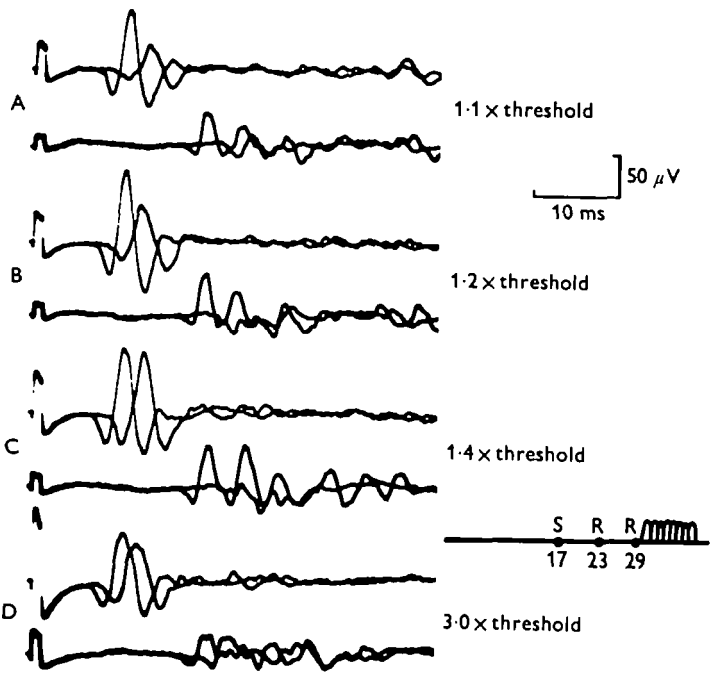

Fig. 2. Facilitation of conduction velocity and size. Responses recorded with two electrodes as shock strength were raised from 1·1 to 3·0 times threshold. Superimposed traces in each line show pulses evoked by paired shocks 1 s apart. The second response appears after a shorter latency in every case, even when there is no increase in the size of the second response (C) or when a strong shock causes a reduction in the size of both responses (D). Facilitation of size is evident at lower stimulus intensities (A, B).

were 83 (range 110–71), 44 (range 64–32), and 17 (39–11) cm/s, respectively. The hepatic region, marked on the dorsal surface by sacculations, is a transition area between the anterior and posterior ventral cord. Conduction velocities along the cord in this region are not uniform, but range between 95 and 38 cm/s. Conduction velocities of the second, third and fourth pulses in the anterior cords of the same 11 worms ranged from 40 to 60 cm/s.

When the second of a pair of shocks is given to the ventral cord within 3 s of the first, the latency between stimulus and response decreases (Fig. 2). This is interpreted as a change in conduction velocity along a fibre tract as a result of the passage of an impulse. Condition velocity is not facilitated by repeated subthreshold stimuli until

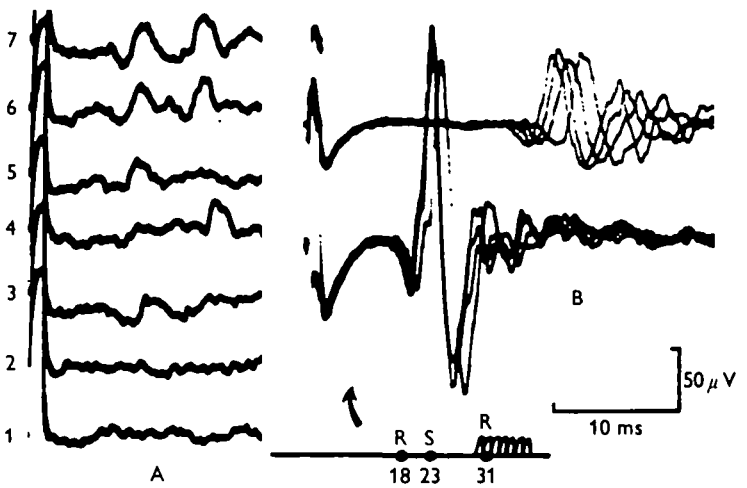

Fig. 3. Effects of repetitive stimulation on facilitation of conduction velocity. (A) Responses to seven, just sub-threshold stimuli, applied 2 s apart, are shown sequentially from bottom to top. There is a decrease in the latency of the first spike but no appreciable increase in its size. Stimulating and recording electrodes were only 4 mm apart, so, as expected from results obtained with supra-threshold stimuli, e.g. as in (B), the decrease in latency is evident after the first pulse appears but not after the second or subsequent pulses. (B) Superimposed traces of responses to four shocks recorded with two electrodes. The third and fourth responses appear after a shorter latency than the second at the distal but not at the proximal recording site. This is partly due to the recording site's greater distance from the stimulating electrode, and partly due to its location in the hepatic region, an area of greater lability.

the first spike appears (Fig. 3 A). In most cases the increase in velocity is between 10 and 20 %, but increases as high as 28 % have been seen. The percentage increase is not a function of stimulus strength above threshold (Fig. 2).

If more than two suprathreshold shocks are given, the third and fourth produce smaller increases in velocity (Fig. 3 B). Facilitation of conduction velocity is not limited to one region but occurs in all parts of the cord, whether impulses are travelling toward or away from the collar. This phenomenon has been shown to occur in nerve fibres of several other invertebrates and vertebrates (Bullock, 1951), percentage increases in velocity being in the same range as in *Ptychodera*. However, the maximum interval between shocks which produces facilitation in the worm is 3 s, whereas facilitation is no longer present in the others when stimuli are over 200 ms apart.

The size and shape of the pulses

When the recording electrode is close to the stimulating electrode, a just supra-threshold shock evokes a single pulse whose chronaxie for excitation is 0·65–0·8 ms. The briefest pulses recorded were 4 ms in duration, but this value reflects some distortion of the pulse by the capacity-coupled amplifier. With small increases in stimulus strength or duration the potential gradually increases in height until its maximum size is reached at a shock strength of 1·3 times threshold. The maximum pulse is larger in the anterior than in the posterior ventral cord, but in both is 3 to 4 times the size of the potential evoked by a just supra-threshold shock. As the recording electrode is

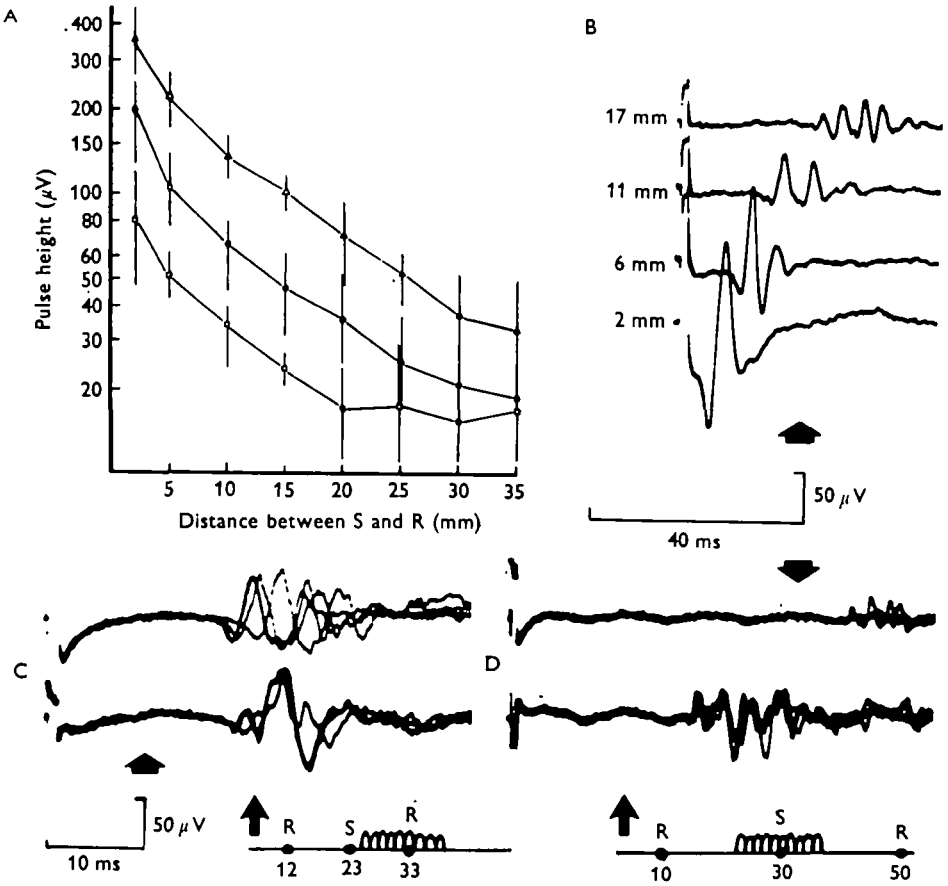

Fig. 4. Pulse height as a function of distance between stimulating and recording electrodes. (A) The range of values for the size of the first or only pulse at each distance is indicated by the vertical line through the mean. Curves are for (top) the anterior ventral cord of four large worms, (middle) the anterior ventral cord of ten average worms, and (bottom) the posterior ventral cord of these same ten worms. At 30 and 35 mm, the ranges of values for all three cords overlap, and from 25 to 35 mm, the ranges of values for the bottom curve extend down to 8 μV. (B) Typical traces obtained by moving the recording electrode different distances from the stimulating electrode. (C, D) Comparative differences in conduction velocity and decay of pulse height in anterior and posterior cords. Superimposed traces of responses to four shocks given 1 s apart are shown, with upper traces recorded from the posterior electrodes. Note differences in electrode distances and time bases in (C) and (D).

moved posteriorly away from the stimulating electrode, the difference in size decreases until a point is reached where the maximum size is also the threshold size; in other words, a large graded potential is broken up into several small, all-or-none spikes. Consequently, pulses can always be recorded at the posterior tip of the worm when the ventral cord is stimulated near the collar, provided the cord is not refractory in the hepatic region. This is an interesting finding in that the decay of spike amplitude in another simple nerve cord, the radial nerve of sea urchins, is complete and responses are not recorded beyond a distance of 6 cm (Sandeman, 1965). The decay in size of the first pulse with distance along the cord of *Ptychodera* is logarithmic, the same in both directions in any one section of the cord, and nearly linear after the first 5–10 mm (Fig. 4). The non-linearity at shorter distances is due to the splitting of this first compound pulse into two major components.

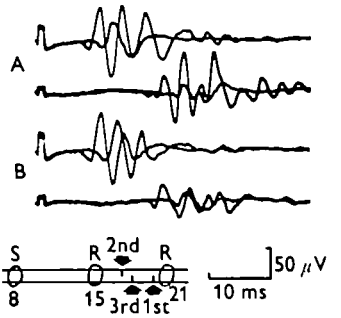

Fig. 5. Interruption of conduction pathways by closely spaced transverse cuts. The diagram shows the position of the electrodes on the cord with arrows pointing to three cuts, each transecting half the cord. The responses to paired shocks 1s apart are superimposed (A, B). The pulses in the lower pairs of traces in (A, B) are larger than expected when compared pulses in the upper traces, e.g. see Fig. 4, because they were recorded with an electrode with a small tip. Nevertheless, the responses to the second shock appear earlier and are larger in all four pairs of traces. The responses recorded after the second cut are shown in (A) and after the third cut in (B). Note that in (B) the three pulses recorded at the distal electrode appear with the same latency as in (A) but are reduced in size. Responses recorded from the intact cord are no different than those shown in (A). Shock strengths were 1·1 (A) and 1·2 (B) times threshold.

If stimulus intensity is below that which evokes a maximum spike, the response to the second of a pair of shocks is greater than to the first (Fig. 2A, B). This facilitation of size is probably due to the recruitment of previously inactive fibres. The maximum interval for facilitation is 2 s, and yet, below this limit it has been impossible to show a consistent effect of the length of the interval between paired stimuli on the size of the nerve potential, in contrast to the marked effect of interval on the muscle potential (Fig. 10A). It is difficult to determine whether or not there is a refractory period. A second shock 50 ms after the first evokes a normal response. Paired shocks less than 20 ms apart evoke a single compound potential that is more complex than the response to a single shock.

Facilitation of conduction velocity and of size must occur at different sites, because the latency between the second stimulus and the response is shorter no matter what the size of the first response (Fig. 2A, C), and because the maximum effective interval between paired stimuli is 3 s for facilitation of conduction velocity and 2 s for facilitation of size (Fig. 3A).

The components of the compound potential appear to be distributed uniformly throughout the cord. If a stimulating electrode is placed so that it covers only half the cord, the evoked potential is smaller, but there is still facilitation of spike size, number and conduction velocity. When a cut is made across the same half of the cord on which a stimulating electrode is located, no change occurs, provided the cut is several millimeters from stimulating or recording electrodes. Similarly, when a cut is made across each half of the cord between stimulating and recording electrodes centered on the cord, there is no change in conduction velocity or reduction in the maximum size of the compound potential (Fig. 5A), unless the cuts are 1 mm apart, in which case, the size of the pulses distal to the cuts are smaller or conduction is blocked completely (Fig. 5B). Thus, it appears that excitation passing along the cord can cross from one side to the other if the bridge of tissue connecting the two halves is not too narrow.

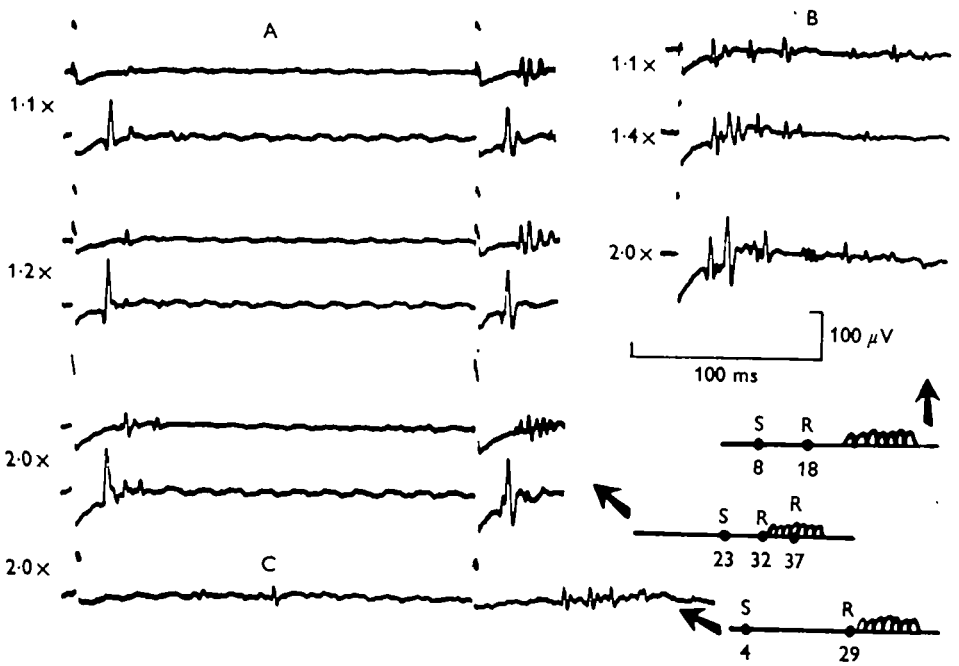

Fig. 6. Increases in the number of pulses as a result of facilitation (A, C), repetitive firing (B, C), and the activation of fibres of higher threshold (A, B). Numbers to the left of the traces indicate shock strength above threshold. Repetitive firing to a weak shock (B) is unusual.

Number of potentials

Fractionation of the single potential occurs as the recording electrode is moved away from the stimulating owing to the temporal dispersion of pulses making up the compound pulse and, undoubtedly, to the dropping out of pulses travelling short distances. When the recording electrode is 5–10 mm from the stimulating electrode, two pulses appear (Fig. 4B). The second has a higher threshold. Further along the cord the first decreases in size and the second breaks up into several pulses. If the number of pulses is indicative of the minimum number of tracts or large fibres, then there are at least four of them in the anterior cord (Fig. 4B). The only consistent dif-

ferences between the smaller pulses is in their conduction velocities; thresholds and amplitudes are quite variable.

In addition, extra pulses are evoked when the strength or duration of a stimulus is increased, typically from fibres of lower conduction velocity (Fig. 6A, B). There is a limit to the number of additional fibres that can be recruited, however, in that the number of pulses decreases when shock strength is greater than three times the threshold (Fig. 2D).

Iterative firing also adds pulses. Although not predictable this activity is most frequently observed following strong or repeated shocks (Fig. 6C). On rare occasions it occurs in fresh preparations when shock strength is low (Fig. 6B).

The number of pulses also increases as a result of facilitation; that is, the responses to the second and third shocks in a series are greater than to the first (Fig. 6A, C). The maximum effective interval between stimuli is 500 ms, but below this limit there is no apparent effect of the length of interval on the number of pulses evoked in response to the second shock.

Even though additional pulses can be evoked under a variety of conditions, it has been impossible so far to identify individual spikes in different preparations on the basis of their relative size or the behaviour they elicit.

Integration along the cord

Lability is a property of all parts of the cord in that pulses are not conducted very far if the cord is subjected to excessive stimulation. Nevertheless, the dropping out of pulses in the hepatic region under normal conditions appears to be the result of integrative processes and not of lability. While the output from the hepatic area may be less than the input, the reverse is often true (Fig. 7). It seems fairly obvious that the latter cannot be due to the temporal dispersion of spikes that make up the compound potential because of the variability in number and latency of pulses in the posterior cord.

In fresh preparations the response to a single stimulus given to the ventral cord near the collar is 'through-conducted' to the posterior tip of the trunk, even though there is almost always a difference between the numbers of pulses in the anterior and posterior cord. On the other hand, conduction toward the anterior end of the worm across the hepatic region ordinarily does not occur, unless the stimulating electrode is close to the hepatic region on the posterior cord. In a few instances, however, spikes arising spontaneously at the posterior tip are transmitted all the way to the collar.

Local muscle potentials

If a shock of one and a half times the strength required to evoke the initial nerve spike is applied to the cord, a large potential, 25–40 ms in duration, appears after a short delay (Fig. 8A). Because the potential can be recorded in nearly every instance only within 6 mm of the stimulating electrode (one rare exception is shown in Fig. 9) and precedes localized contraction around this electrode, it will be called the local muscle potential. It is considered to be an actively evoked potential, and not one that is conducted passively from the point of stimulation, for the following reasons. First, a cut made across the nerve cord with a fine needle does not appear to damage underlying muscle fibres, and yet a potential cannot be recorded beyond this cut. Secondly,

the potential disappears with repetitive stimulation or soon after an anaesthetic is applied (Figs. 8B, 10), suggesting that there is failure of transmission at neuromuscular junctions. Thirdly, if the stimulating electrode is 1 mm lateral to the cord, a shock three times stronger than an effective stimulus to the cord does not evoke this potential. The size of the potential is a function of the distance between stimulating and re-

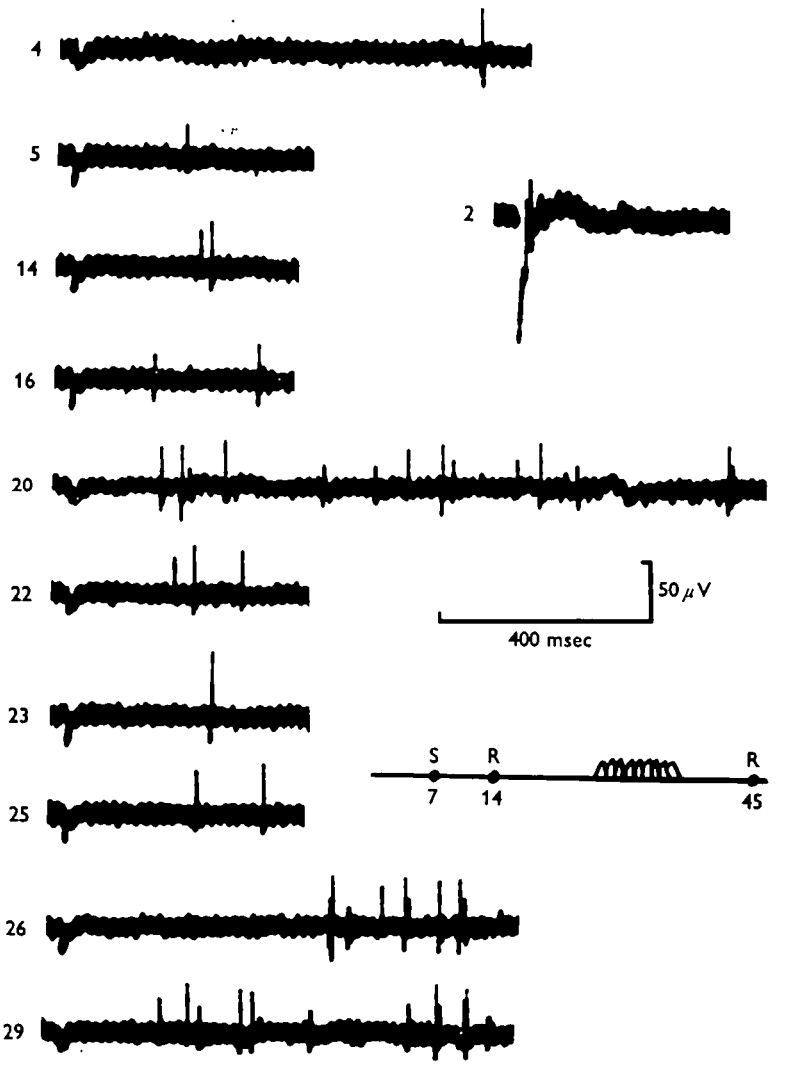

Fig. 7. Variability of spike number and size in the posterior ventral cord in response to repetitive stimulation. Each of the 30 shocks that were given 2 s apart evoked pulses in the anterior cord that were similar to those recorded at the anterior electrode in response to the second shock (2), in which a stimulus artifact is followed by two nerve spikes and a longer-lasting muscle potential. Pulses were not recorded posterior to the hepatic region until the 4th shock (4) and even then each stimulus did not evoke a response. All pulses appearing in the posterior cord during repetitive stimulation are shown on the left side of the figure except those after the 17th and 28th shocks. These latter are omitted because they consisted of single pulses with the same height and latency as the first spike after the 16th shock. Pulses appearing in the last halves of traces 20, 26 and 29 could conceivably originate distal to the posterior recording electrode and be passing toward the hepatic region, as has been seen to occur in other preparations.

cording electrodes (Fig. 8), the number and strength of the stimuli (Fig. 9), and the interval between paired or multiple shocks (Fig. 10A). Manipulations of these same stimulus parameters can determine the number of nerve spikes (Figs. 4, 6) and yet a casual relationship between the number of nerve spikes and the size of the muscle potential has not been established. In most instances when the recording electrode is 5 mm from the stimulating the shock applied must be strong enough to evoke two nerve pulses before a local muscle potential will appear. However, these pulses are considered to arise from separate tracts and, indeed, the tract which gives rise to the second pulse with the higher threshold may be the only one of the two that carries

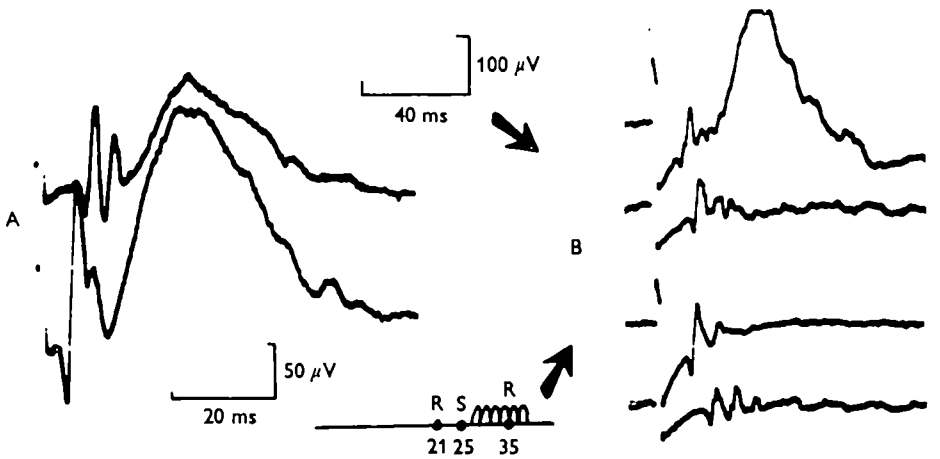

Fig. 8. The local muscle potential. (A) Size of the potential when the recording electrode is 2 mm (lower trace) or 5 mm (upper trace) from the stimulating. The muscle potential is eliminated (B) when the worm is bathed in a solution of five parts sea water and one part magnesium chloride isosmotic with sea water. Nerve spikes and a muscle potential were evoked by a single shock 1 min after immersion (upper traces), but the muscle potential did not appear after a test shock 10 min later (lower traces). The nerve cord was still responsive 50 min later.

excitation to the muscle. In any event, the same muscle potential that is evoked by a single shock will not appear until after the second of two weak shocks is given. In contrast to what is shown in Fig. 6A, there may be no increase in the number of nerve pulses after the second weak shock (Fig. 10B), just what would be expected if facilitation is required at neuromuscular junctions. Responses to a single, strong shock supposedly meet this requirement because they contain pulses from fibres that fire repetitively. However, repetitive firing in *Ptychodera* has never been shown to precede a local muscle potential.

Conduction in other parts of the nervous system

The anterior metasome nerve ring is the conduction pathway from collar cord to ventral cord (Bullock, 1940; Knight-Jones, 1952). If the collar cord is exposed by dissection and stimulated, spikes are evoked in the ventral cord. Minimum conduction velocity along these connectives is estimated at 60 cm/s or about the same as that along the ventral cord. It has not been possible to record from the nerve ring itself, however, nor does stimulation of the region in which the nerve ring is found evoke spikes in the ventral cord, with the following exception. When the stimulating electrode is $\frac{1}{2}$ mm

lateral to the ventral cord near the collar, a stimulus of ten times threshold strength for the cord will evoke a ventral cord pulse that is smaller, has a different shape, and a longer latency than the typical cord pulse. Conduction velocity through this pathway, which is not found elsewhere along the trunk, is about one-twentieth that through the cord, or 4 cm/s. This pathway may be through the nerve net.

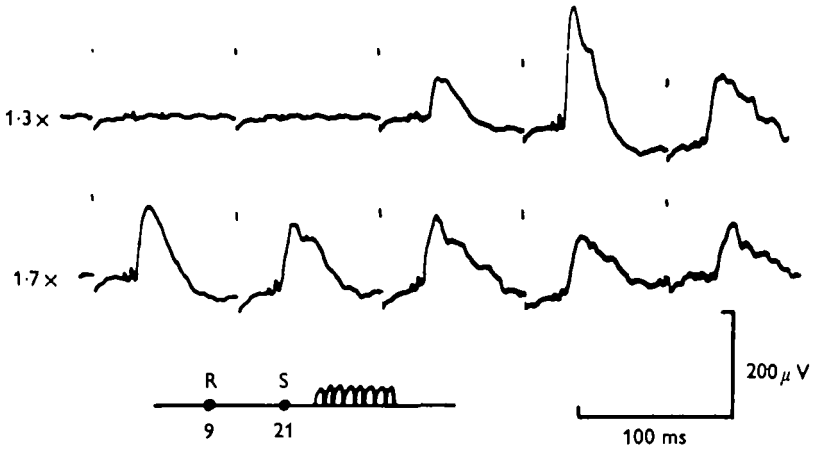

Fig. 9. The influence of the strength and number of shocks on the size of the muscle potential. Facilitation of size is evident at 1·3 times threshold, but maximum size of response can be obtained after the first shock if stimulus strength is increased. Shock strengths are based on the intensity of stimulus required to evoke the initial nerve pulse.

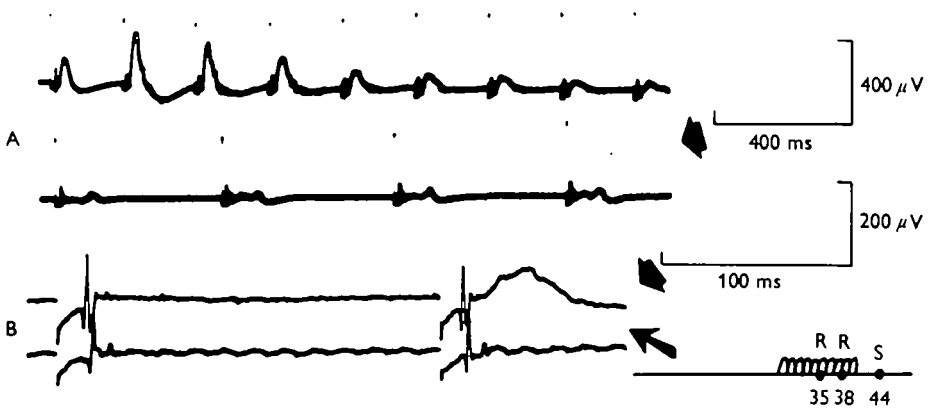

Fig. 10. Size of the muscle potential as a function of the interval between repetitive stimuli. (A) Repetitive stimulation at two different frequencies, with a pair of nerve spikes and a longer-lasting muscle potential following each shock. Facilitation and anti-facilitation of size occur when shocks are 200 ms apart but not when they are 500 ms apart. (B) An expanded portion of a similar recording when weaker shocks are given. A muscle potential appears after the second shock even though there is no change in the number of nerve spikes.

Nerve spikes can be evoked from the dorsal cord and are conducted at about the same rates as they are in the ventral cord. However, in spite of the morphological connexions between dorsal and ventral cords via the nerve ring and trunk plexus, at no point along the body are spikes conducted from one cord to the other. On the other hand, spikes can be recorded in the collar cord when the dorsal cord is stimulated.

DISCUSSION

The small pulses described above must originate from nervous rather than non-nervous tissue because they cannot be recorded lateral to the ventral cord and are not conducted beyond transverse cuts. The source of these pulses within the cord is difficult to determine. There are no distinct giant fibres in the Ptychoderidae (Bullock, 1944) and recent electron microscope studies reveal that most fibres are less than one-third of a micron in diameter (Pickens & Ferris, 1969). Although pulses may represent activity in single, large axons, it is postulated that small fibres lying close together are triggered synchronously through ephaptic interaction to produce potentials of sufficient size to be recorded externally. In either case, it is difficult to see how signals could pass down the cord without being modified by activity in adjacent tracts, since the fibres are unmyelinated and there are no glial cells to insulate neurones from each other (Bullock, 1945).

Nevertheless, a single compound pulse breaks up into at least four components as it travels along the anterior ventral cord (Fig. 4). These pulses arise from neurones which appear to be uniformly distributed throughout the cord because cuts across part of the cord reduce pulse amplitudes yet fail to eliminate any component completely. An alternate explanation is that the four pulses arise as a result of repetitive firing in the same tract but are not of the same height because of the variable number of parallel fibres in the tract that are recruited during the passage of each pulse. However, this is discounted because when repetitive firing is seen, pulses are of the same height. Furthermore, the interval between each of the four pulses increases as they are conducted down the cord, which would mean that de-facilitation of conduction velocity takes place when, in fact, facilitation is more likely to occur. In the posterior ventral cord there must be three or more conduction pathways because, first, a compound pulse evoked by shocking the posterior cord splits up into three components as it is conducted along the cord and, secondly, spikes of at least three different sizes appear when the anterior cord is stimulated and excitation crosses the hepatic region (Fig. 7). In an earlier paper Baxter & Pickens (1964) concluded that three conduction pathways have to be present in order to explain the spread of luminescence in *Ptychodera*. For two types, the startle and disjunct luminescence, conduction velocities along the cord are within the range of values given in this paper for the fastest tracts. A third type of luminescence, characterized by its extensive spread, sweeps along the worm at 3–4 cm/s, or at the velocity of propagation of peristaltic waves. Applied shocks evoke no nerve spikes that travel at such a slow rate, however, and any activity of nervous origin which may occur during a peristaltic wave is masked by spike-like muscle potentials. Horridge (1968) has suggested that nervous systems became more complex when nerve nets were collapsed to form tracts of parrallel fibres. Differences in function between two sets of fibres in the same net may have given rise to two conducting systems. Two classes of fibres are thought to be present in the radial nerve cord of a sea urchin (Sandeman, 1965), but there is no clear indication whether in *Ptychodera*, an animal which is functionally if not phylogenetically closely related, the several conducting pathways are labelled lines or non-specific but parallel channels for information. In either case, the hemichordate cord differs from the radial nerve of sea urchins in that the former has through-conduction tracts, pulses arising from it are conducted four to

five times more rapidly, there is facilitation of pulse size, and the refractory period is short.

In addition to several conducting pathways, the ventral cord has an integrative area in the hepatic region. Information passing through this region is almost modified in pulse rate and number (Fig. 7). However, it is a very labile part of the cord, so that sometimes pulses are through-conducted in response to a single shock, whereas at other times the facilitory effect of several shocks is required for spread of excitation. On the other hand, transmission of pulses toward the collar is generally blocked in the hepatic region no matter how many shocks are given to the posterior trunk. Behavioural correlates of these responses can be obtained in whole animals when weak mechanical stimuli are applied to either one end of the worm or to the other (Pickens, 1970).

Facilitation occurs when successive increments of response are greater than the preceding ones and is demonstrated by employing paired or repeated stimuli (Bullock & Horridge, 1965). Using these criteria it has been shown that in *Ptychodera* there is facilitation of conduction velocity, number of spikes, size of the local muscle potential, and, under conditions mentioned in the last paragraph, spread of excitation. Facilitation of the size of compound nerve spikes described earlier is not a separate category, but is facilitation of number on a compressed time scale. A strong shock will evoke as large a pulse or as many pulses as a pair of weak shocks, presumably because fibres of higher threshold are recruited and others are made to fire repetitively. Facilitation of conduction velocity, on the other hand, is evident only if a pair of stimuli are applied; that is, conduction velocity cannot be increased by giving a single stronger shock. Repetitive discharge to single shocks has been postulated as a mechanism for satisfying the requirement for facilitation at neuromuscular junctions (Pantin, 1935; Baxter & Pickens, 1964). However, iterative firing in the cord of *Ptychodera* occurs in response to a shock which is stronger ($2 \times$ threshold—Fig. 6) than that which is necessary to evoke a local muscle potential ($1{\cdot}5 \times$ threshold—Fig. 9), and usually is associated with gross movements such as startle responses and peristaltic waves. Consequently, if the same muscle fibres and motor neurones are involved in gross and local movements, facilitation may be a requirement at synapses within the cord in addition to being a requirement at junctions between nerve and muscle. In any event, repeated stimuli will have a tendency, through the combined action of these facilitory effects, to produce the burst of closely spaced nerve pulses that would satisfy a requirement for neuromuscular facilitation if it is present in *Ptychodera*.

SUMMARY

1. Compound and all-or-none pulses have been recorded from the ventral nerve cord of a hemichordate worm.

2. Compound pulses are composed of at least four smaller pulses, suggesting that four conduction pathways may be present in the cord.

3. Conduction velocities in the anterior ventral cord are as high as 1·1 m/s but fall to about half this value in the posterior cord. In both parts of the cord the passage of the first pulse facilitates the passage of the second so that conduction velocities are increased by 10 to 20%.

4. Paired or multiple stimuli produce repetitive firing and facilitation of size and number of pulses in the cord.

5. Although strong or repeated shocks evoke local, graded muscle potentials, no correlation has been found between the size of these potentials and the number of pulses in the cord.

6. Integration occurs in the hepatic region of the cord. It is the most labile to through-conduction, output from this region does not have a one-to-one relation to input, and pulses originating in the posterior cord and travelling toward the collar are usually blocked in this area.

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