

NEUROMUSCULAR PHYSIOLOGY OF THE
LONGITUDINAL MUSCLE OF THE EARTHWORM,
LUMBRICUS TERRESTRIS

III. MAPPING OF MOTOR FIELDS

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INTRODUCTION

The body-wall musculature of annelids is not organized into distinct bundles of muscle fibres as in many arthropods and chordates. Instead, the body muscle is composed of cylindrical sheets of muscle fibres surrounding the body cavity. These muscle sheets extend continuously along the length of the worm and are organized into concentric layers of antagonistic muscle, an outer circular layer and an inner longitudinal layer. This organization makes it convenient to describe the pattern of innervation of the body-wall muscle in terms of motor fields or territories.

Prosser (1935) first studied motor fields in earthworm longitudinal muscle by viewing the spatial limits of muscle contractions in response to nerve stimulation. Based on his observations he suggested that the motor fields for one segmental nerve may extend into adjacent segments, thus providing the possibility for overlapping of motor fields.

In the following experiments we have attempted to define the boundaries of territories or fields of innervation for motor axons innervating the longitudinal muscle. Two approaches were taken, one involving the mapping of external electrical responses of the muscle in response to segmental nerve stimulation and the other involving intracellular recording of muscle responses to stimulation of segmental nerves.

MATERIALS AND METHODS

Procedures for maintaining animals, methods for electrical stimulation and recording, and an appropriate physiological saline are described in previous papers (Drewes & Pax, 1973 *a, b*).

The nerve-muscle preparation consisted of a flat strip of body wall 15-20 segments in length as shown in Fig. 2. The segmental nerves were severed close to their central connexions with the ventral nerve cord. The nerve cord along with the remaining undissected portion of the worm were then discarded. Following stimulation of the segmental nerve stumps the electrical responses of the longitudinal muscle were recorded and mapped over the surface of the muscle.

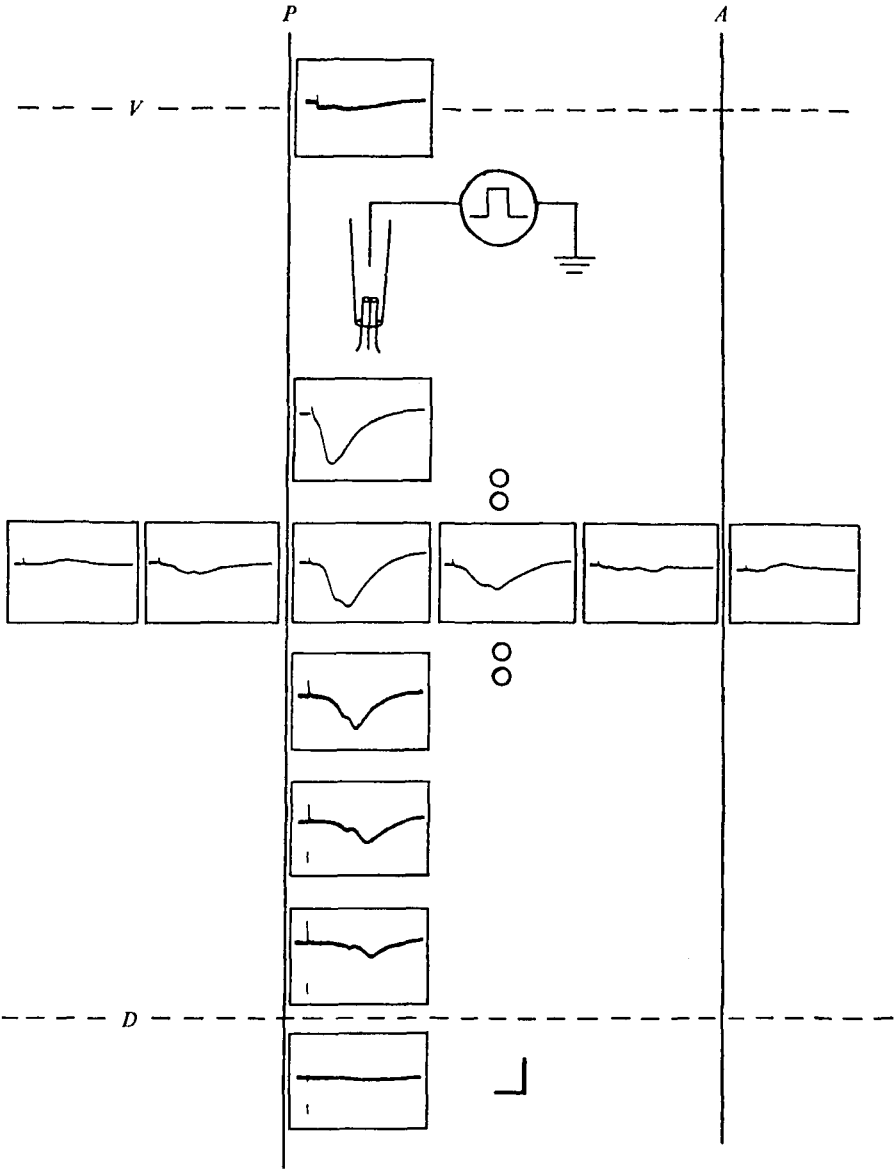


Fig. 1. Mapping of external electrical responses of the longitudinal muscle to single, supra-maximal stimuli to SNII-III. Responses recorded in a transverse line through the nerve are negative in sign and diminish in amplitude with increasing distances from the site of stimulation. Also as the distance between stimulating and recording electrodes is increased two distinct phases are apparent in the muscle response, the first phase being mediated by the fast excitatory axon and the second by the slow excitatory axon. At the dorsal midline (*D*) and the ventral midline (*V*) the amplitude of potentials is negligible. When mapped in a longitudinal direction positive potentials are recorded in adjacent segments. *A*, Anterior segmental boundary; *P*, posterior segmental boundary. Voltage scale: 1.0 mV. Time scale: 10 msec.

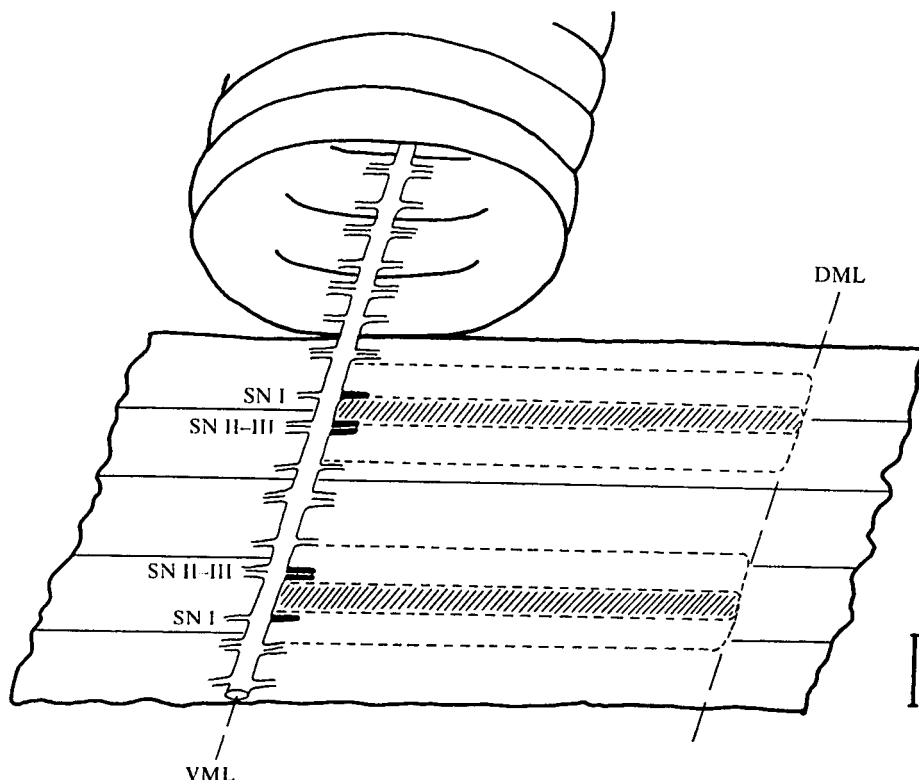


Fig. 2. Overlapping motor fields of adjacent segmental nerves. The body wall of the earthworm is laid open showing the longitudinal muscle layer and the segmental nerves arising from the ventral nerve cord. The boundaries of motor fields for the longitudinal muscle, indicated by dashed lines, were determined by mapping external muscle potentials. Fields for adjacent nerves in the same segment overlap one another, the area of overlap being indicated by the lower shaded area. Also fields for adjacent nerves in neighbouring segments overlap one another, the area of overlap being indicated by the upper shaded area. DML, dorsal midline; VML, ventral midline. Scale: approximately 1 mm.

RESULTS

External electrical recordings

External electrical responses to segmental nerve stimulation were recorded from numerous sites on the surface of the longitudinal muscle in six preparations. For each nerve approximately 200 responses to single stimuli were mapped from 42 different positions on the surface of the muscle.

Some typical responses to stimulation of SNII-III are shown in Fig. 1. Recordings were first made at various sites along a transverse line passing through the segmental nerve. In this way a transverse profile of the external muscle potentials was obtained. As the recording electrode was positioned farther away from the nerve, two negative phases in the response became increasingly apparent, each phase becoming smaller and having a longer onset latency as the distance from the nerve was increased. In all cases these two phases corresponded in time course and threshold to the electrical responses mediated by the fast and slow axons in the segmental nerve (Drewes & Pax, 1973*b*).

At the dorsal and ventral midlines fast and slow responses were barely recordable and at points beyond the midlines no responses were observed.

Electrical responses were also recorded at various points along a longitudinal coordinate corresponding to a line passing midway between the two pairs of setae in each segment (Fig. 1). Thus a longitudinal profile of external potentials was obtained. Analysis of these potentials was difficult since the form of the potentials was often complex. The complexity appears to be due to a reversal in the sign of the potential, a reversal from negative to positive being seen if the electrode is moved anteriorly or posteriorly to recording sites farther away from the transverse line passing through the segmental nerve.

From the previous study it appears that the electrical events in muscle fibres are primarily local, non-propagated postsynaptic potentials (Drewes & Pax, 1973*b*). Thus the presence of a negative external potential most probably indicates the presence of a current sink whose origin is the postsynaptic excitation of muscle. Positive potentials may indicate current sources for the postsynaptic currents, and it may be assumed that no postsynaptic current (i.e. innervation) occurs in these areas. Records taken from the area of transition between positive and negative potentials are more difficult to interpret. The negligible amplitude of potentials recorded from these areas indicates there is little or no net current flow. It is likely that these areas probably represent the spatial limits for the occurrence of synaptic currents and for innervation.

Using this interpretation of electrical events it is possible to draw rough boundaries for the motor fields of each segmental nerve as shown in Fig. 2. In this figure the longitudinal boundaries of the field were drawn so as to just include all points where a transition between negative and positive potentials occurred. The transverse boundaries were drawn to include all points where negative potentials were detected, but to exclude points where no potential was detected. By these criteria each of the motor fields consists of a narrow strip extending from dorsal to ventral midlines. In all cases these boundaries appear to apply to innervation by both fast and slow axons since negative potentials, when recorded, were always in the form of two separable responses. Since the field of innervation for each segmental nerve includes about one lateral half of a segment there appears to be an overlap of the fields of adjacent nerves. Such overlap is both intrasegmental and intersegmental.

The apparent overlap of motor fields of adjacent segmental nerves may be explained in two ways. First, there may be coincidental and overlapping excitatory innervation of muscle fibres from different segmental nerves. This explanation seems likely from a physical standpoint, since Hanson (1957) has shown that longitudinal muscle fibres may be two or three times longer than a single segment. An alternative explanation is that branches of motor axons from one nerve may interdigitate with branches from adjacent segmental nerves, thus giving only the appearance of overlapping motor fields.

Intracellular recordings

In the following experiments we have used intracellular recordings to study the possibility of coincidental and overlapping innervation of muscle fibres by different segmental nerves. To study the possibility of overlapping innervation between segmental nerves in the same segment (intrasegmental overlap), recordings were made approximately midway between the septal boundaries of a segment. To study overlap

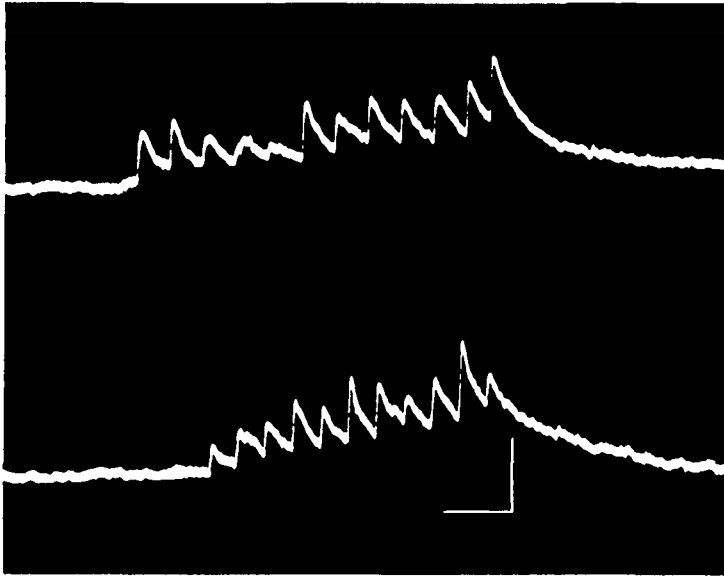


Fig. 3. Intracellular activity in a longitudinal muscle fibre following supramaximal repetitive stimulation of segmental nerves in the same segment. In the upper record stimulation of SNI at 5 Hz results in a series of summating postsynaptic potentials. In the lower record repetitive stimulation of SNII-III at 5 Hz also results in a series of summating potentials in the same muscle fibre, indicating that this fibre is innervated by two different segmental nerves in the same segment. Voltage scale: 5 mV. Time scale: 0.5 sec.

between segmental nerves of different segments (intersegmental overlap), recordings were made near the septal boundary between adjacent segments. Segmental nerves were stimulated individually using supramaximal stimulus intensities and a low frequency of stimulation (5 Hz).

Four preparations were examined and similar results were obtained from each preparation. In each preparation muscle fibres were commonly found which received innervation from both SNI and SNII-III in the same segment as shown in Fig. 3. The response to stimulation of SNI at 5 Hz (upper trace) consists of a series of excitatory postsynaptic potentials ranging from 1 to 4 mV in amplitude. A slight summation of the potentials is apparent. The response of the same muscle fibre to stimulation of SNII-III (lower trace) consists of a similar series of excitatory postsynaptic potentials ranging from 2 to 5 mV in amplitude. In addition to summation of the potentials there is also a slight facilitation.

Similar results were obtained when testing for intersegmental overlap of motor fields; that is, excitatory postsynaptic potentials were recorded from muscle fibres following stimulation of either of two adjacent segmental nerves in two neighbouring segments. These experiments lend further support to the idea of overlap of motor fields for adjacent segmental nerves.

DISCUSSION

The results of the present study support the observations made by Prosser (1935), that in the earthworm longitudinal muscle the motor fields of adjacent segmental nerves overlap one another. Such overlap occurs between adjacent nerves in the same segment as well as between adjacent nerves of two neighbouring segments.

Motor fields have been described in only one other annelid (Stuart, 1970). For the leech Stuart described the motor fields for the six excitatory axons innervating the longitudinal muscle. It was shown that the motor fields for each axon in a segment were different. One pair of excitatory axons, arising from two large longitudinal motor neurones, innervates the left and right halves of a segment and functions to bring about a uniform shortening of the animal. Such a response may occur when the animal withdraws from a noxious stimulus. In contrast, the motor fields of the other five pair of axons innervating the longitudinal muscle are much smaller, subdividing the body wall into small strips or patches of muscle. Stuart suggests that contraction of any one of these strips would cause a bending of the body. The co-ordinated action of these small motor fields may account for the complex bending and twisting movements of the animal.

The motor fields in the earthworm longitudinal muscle are somewhat different from those of the leech. In the earthworm motor fields are large, fields of both fast and slow axons extending from dorsal to ventral midlines. The arrangement of these motor fields would probably not permit a wide variety of twisting and bending movements, such as one sees in the leech. However, this arrangement would appear to be well suited to locomotor activities requiring uniform and symmetrical contractions of the segmental musculature. Examples of such movements would be the peristaltic forward locomotion of the worm (Gray & Lissman, 1938; Chapman, 1950; Seymour, 1969, 1971), or the rapid escape response (Rushton, 1946; Roberts, 1962*a, b*). The only prerequisite for such responses is the synchronous output to right and left sides of the animal. The absence of output from either side would provide the possibility of turning movements.

SUMMARY

1. The motor fields of individual segmental nerves have been mapped electrophysiologically in the longitudinal muscle of the earthworm, *Lumbricus terrestris*.
2. The anterior pair of segmental nerves (SNI) innervates approximately the anterior two-thirds of its segment and a small portion of the segment just anterior to it.
3. The posterior pair of segmental nerves (SNII-III) innervates approximately the posterior two-thirds of its segment and a small portion of the segment just posterior to it.
4. Adjacent nerves (both intrasegmental and intersegmental) have partially overlapping motor fields; that is, adjacent segmental nerves innervate some of the same longitudinal muscle fibres.

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