

**BACKGROUND AND REFLEX DISCHARGE OF SYMPATHETIC  
PREGANGLIONIC NEURONES IN THE SPINAL CAT**

BY W. S. BEACHAM AND E. R. PERL

*From the Department of Physiology, University of Utah,  
Salt Lake City, Utah, U.S.A.**(Received 6 January 1964)*

The initial observations showing that sympathetically innervated effector organs of spinal mammals may respond to afferent stimulation were made many years ago, but there continues to be uncertainty as to the magnitude and importance of spinal sympathetic reflexes. Stimulation of the central end of a hind-limb nerve in a chronic spinal dog was reported by Sherrington (1906) to produce a large rise in blood pressure. In acutely prepared spinal cats substantial increases of systemic blood pressure also have been described to result from mechanical stimulation of the viscera (Downman & McSwiney, 1946; Mukherjee, 1957) or from nerve excitation after strychnine administration (Langley, 1924). In contrast, other studies have concluded that following acute or chronic spinal section, the change in sympathetic activity evoked by stimulation of somatic nerves is either abolished (Alexander, 1946; Schaefer, 1960) or relatively small (Brooks, 1933). Well-defined, spatially-restricted vasomotor reactions, however, have been produced by thermal or noxious stimulation of the skin innervated by segments below the level of acute spinal transection (Langley, 1924; Sahs & Fulton, 1940; Kuntz, 1945; Richins & Brizzee, 1949). At least some of such more localized responses to stimulation are dependent upon integrity of the sympathetic motor supply (Kuntz & Hazelwood, 1940). Finally, there is work indicating that part of the sympathetic outflow is tonically active independent of supraspinal and afferent input, and that this tonic background is modified in association with changes in the state of the preparation (Brooks, 1935; Alexander, 1945; but cf. Koizumi & Suda, 1963).

Thus, there are considerable data suggesting that the sympathetic system retains some capacity for reflex or other functional adjustment in the absence of suprasegmental centres. Nevertheless, the majority of past and current experimental studies have not emphasized possible contributions from spinal mechanisms. Furthermore, the nature of spinal cord processes mediating sympathetic reactions is obscure.

The present experiments were carried out to examine certain features of the spinal cord organization concerned with the sympathetic efferent activity. It seemed advantageous for the initial steps of such an analysis to eliminate the complications of transmission in the sympathetic ganglia and action of effector organs. Accordingly, recordings were made from axons of preganglionic neurones in several sympathetic rami communicantes of decapitated cats. The results demonstrate that in such preparations tonic background discharge of preganglionic neurones does occur, that tonic discharge can be modified by afferent input, and that single volleys of afferent impulses initiate reflex discharge of preganglionic neurones by what probably are polysynaptic pathways.

#### METHODS

In adult cats the spinal cord was cut through at C1 and the circulation to the head occluded under ether anaesthesia. Anaesthesia was discontinued and the preparation was maintained by positive-pressure artificial respiration through a tracheal cannula. CO<sub>2</sub> in the tidal air was monitored by an infra-red detector (Beckman LB-1). The respiration was adjusted so as to keep the end-tidal CO<sub>2</sub> concentration close to normal levels (5-6%). Rectal temperatures were maintained between 36° and 38° C by external heat. In many experiments, arterial pressures were monitored from a carotid cannula. 'Flaxedil' (gallamine triethiodide, American Cyanamid) was injected intravenously to keep the preparation paralysed during recording.

The preparation was routinely placed with the left side up. In most experiments, a laminectomy exposed the dorso-lateral spinal cord of the upper thoracic or upper lumbar levels. The thoracic sympathetic rami entering the stellate ganglion were exposed in the retropleural space after detachment of the overlying muscles by removal of the heads of the first two or three ribs and the lateral vertebral processes. The rami joining the lumbar sympathetic ganglia (at L1 to L4) were reached retroperitoneally between the aorta and the vertebral column after removal of the lateral processes. All exposed tissues were covered with mineral oil equilibrated with 95% O<sub>2</sub> and 5% CO<sub>2</sub> in a pool made from flaps of skin and muscle.

Dissections of the sympathetic rami and the sympathetic trunk were carried out with the aid of a binocular dissecting microscope at magnifications of 6-25×. The rami communicantes entering the ganglia were identified by their penetration of its capsule and by their juncture with the appropriate spinal nerve. The rami were sectioned at the ganglia and recordings were made from the central end with fine platinum-iridium leads. The gross appearance of preganglionic and post-ganglionic communicating branches was sometimes not distinctive enough to permit certain separation, and in some segments both were found within a common sheath. In these cases, the final test of whether a ramus contained preganglionic fibres was based on the presence of persistent background activity and a reflexly evoked discharge (see Results). Rami with clearly post-ganglionic distribution were silent after a brief injury discharge following separation from the ganglion. With experience, the majority of preganglionic rami could be identified on the basis of appearance and location.

Single volleys were initiated by electrical pulses (0.1 msec in duration) delivered by pairs of platinum electrodes to dorsal root fibres or peripheral nerves. The dorsal root subdivisions (rootlets) were routinely short (5-12 mm) in the upper thoracic or lumbar regions. Escape of the stimulus directly to the spinal cord was tested by placing a cathodal stimulating electrode directly upon the dorsal root entrance zone with the anode on indifferent tissue

(adjacent bone or muscle) and the maximum stimulus used in the present study did not evoke responses in the rami communicantes unless the stimulating anode was on dorsal root fibres. In a number of experiments, the dorsal roots were left intact and spinal nerves of an upper thoracic or a lumbar segment were stimulated 1 or 2 cm distal to the separation of the sympathetic rami communicantes. Additionally, the left sciatic nerve at the level of the hamstring branch and/or nerves of the brachial plexus were isolated for stimulation.

At the end of a series of observations, a routine control was made for electrotonic spread of recorded activity from nearby somatic nerve trunks by either crushing or damaging the central end of the preganglionic ramus near to its junction with the spinal nerve. The potentials attributed to preganglionic axons regularly disappeared after these procedures. Occasionally, somatic nerves containing motor fibres passed close to the sympathetic ganglia or trunk and were partially enmeshed in the connective tissue surrounding the sympathetic structures (Downman & Hazarika, 1962). These were distinguished from sympathetic nerves by the following observations: they did not enter and diffuse within the sympathetic ganglia and the reflexly evoked activity recorded in them was large in amplitude and short in latency compared to that seen in sympathetic nerves.

The discharge recorded from preganglionic bundles was usually small in amplitude, often being little higher than the inherent noise of the amplifying system. It was necessary to desheath and frequently to split the preganglionic ramus to increase the signal-to-noise ratio. The latter procedure also allowed study of the discharge of individual elements. The band-pass of the amplifying system was also adjusted to improve the signal-to-noise ratio for each recording condition (ordinarily 8 c/s to 10 kc/sec). After amplification, the activity was displayed and photographed from a two-channel oscilloscope or stored on magnetic tape using an FM analogue system. In some cases, the latencies between stimulus and response of a single unit were measured with a digital time-interval meter (Perl, 1962).

At least 5 and as much as 18 hr elapsed between spinal transection and the reported observations. No systematic change in reflex and background discharge was observed over this period which could be attributed to time after spinal section, although the magnitude of preganglionic activity and nature of background discharge varied with factors related to the general condition of the preparation such as blood pressure and local circulation of the spinal cord (cf. Mukherjee, 1957). Results as described were obtained in preparations with mean blood pressures over 50 mm Hg (usual value was 70–90 mm 5–6 hr after spinal section) and with end-tidal CO<sub>2</sub> levels between 4 and 6%. Often, the preparations deteriorated rather than improved later in the experiment, a fact which may be related to the extensive surgical procedures used for exposure of the neural tissue.

## RESULTS

### *Background activity and its alteration by sensory stimulation*

The rami which were clearly preganglionic regularly contained some fibres showing active background discharge. Background discharge of individual elements manifested itself in several ways. Rarely, the discharge frequency was relatively constant in the manner illustrated in Fig. 1A. Regular frequencies from under 1 to 50 impulses/sec were observed in different preparations. A regular preganglionic discharge during anoxia has been described for anaesthetized, intact cats (Iggo & Vogt, 1960). It is possible that the present observations were related to anoxia even though respiration was carefully controlled. For example, there may have been anoxia localized to spinal cord structures; however, the rela-

tively regular discharge rates were maintained for long periods (many hours for the unit of Fig. 1A) and could be altered (see below).

Most commonly, background activity was irregular with no obvious correlation either to the cardiac or respiratory cycles (Fig. 1B). The repetition rate of the irregular type of discharge also varied greatly, from less than 1 impulse/sec to over 60/sec. Analysis of the irregular type of background discharge indicated that cyclic increases in discharge frequency were often present. In Fig. 2, the instantaneous frequency (1/interval) of an individual element from the L2 ramus was plotted against time. The highest discharge frequencies of this element (intervals of 1/12 to 1/25 sec) repeated at periods of roughly 25 sec and did not have a clear relation to the cardiac or respiratory cycle. In a number of cases, the cyclic increases in frequency of discharge apparently involved several units active in near synchrony at relatively long time intervals. As is also evident from Fig. 2, less regular variations in discharge frequency occurred.



Fig. 1. Background discharge of preganglionic fibres. A: Recording from filament of T4 preganglionic ramus. Discharge as illustrated maintained for over 30 min with no change in frequency in the absence of specific stimulation. Firm pressure to each of the four feet was followed by increase in discharge frequency. Time bar equivalent to 1 sec. End-tidal CO<sub>2</sub>, 5%. Rectal temp. 37° C. Ten and one-half hours after spinal transection. B: Recording from filament dissected from combined T3 and T4 preganglionic ramus. Pinch to the left hind paw was followed by complete cessation of background discharge for several sec. Time bar equivalent to 2 sec. End-tidal CO<sub>2</sub>, 4.8%. Rectal temp. 36.5° C. Eleven hours after spinal transection.

In many preparations, both regular and irregular preganglionic background activity could be changed by afferent excitation; however, a large number of tonically active units did not respond to any stimulus tried. In addition to nerve volleys, cutaneous stimuli effective in altering background discharge were pinch, pin prick, or intense pressure, all judged to be of noxious intensity; skin temperature changes; and non-noxious pressure or mechanical transients. Both increases and decreases in background

activity were seen to follow these manipulations. Spatial localization for the effective stimulation was regularly found, although frequently the localization was not discrete. Sometimes, stimulation of one body region resulted in increased discharge frequency while stimulation at a distant point, such as another limb, resulted in depression of background discharge of the same preganglionic axon. Examples of responses from one small preganglionic bundle are shown in Fig. 3. The stimulus used was firm pressure; ipsilateral forepaw (Fig. 3*A*) inhibiting of one element; contralateral hind paw (Fig. 3*B*) exciting another unit; and ipsilateral hind paw (Fig. 3*C*) also exciting the same element as discharged in Fig. 3*B*. Crossed effects such as that depicted in Fig. 3*B* were found, but often were absent in units responding to ipsilateral stimulation.

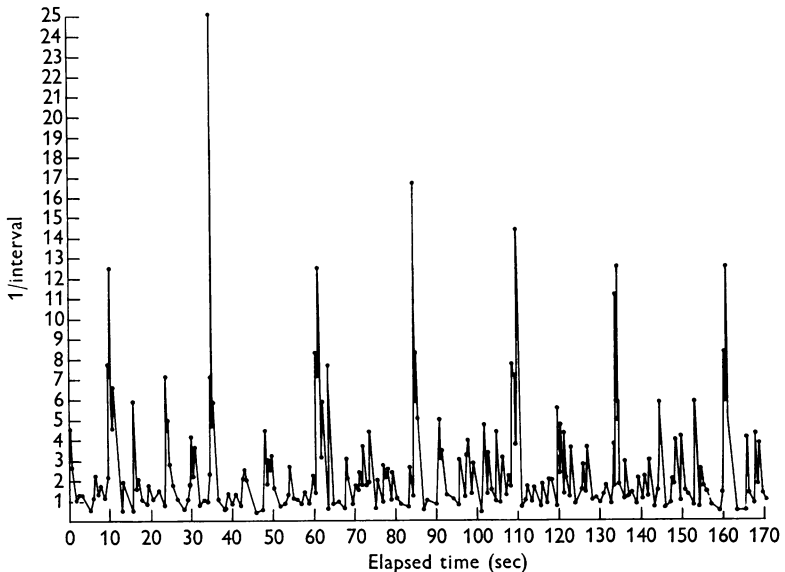


Fig. 2. Background discharge of a L2 preganglionic fibre. Graph constructed by measuring interval between successive discharges of single unit and then plotting this as instantaneous frequency ( $1/\text{interval}$  in sec) against elapsed time from an arbitrary beginning. Cyclic increases in frequency with an approximate 25 sec period were recorded for some minutes longer than shown in this figure. Respiratory cycle was 5 sec and the heart rate was approximately 140/min. End-tidal  $\text{CO}_2$ , 4.2%. Rectal temp. 37.5° C. Twelve hours after spinal transection.

Response to skin temperature change in another experiment is illustrated in Fig. 4; discharge frequency of the unit shown (from T1) decreased in association with cooling of the left upper forelimb skin. Rewarming the skin resulted in a resumption of a higher discharge frequency for this unit. In other elements, increased activity followed either sudden heating or

cooling. Frequently, a given preganglionic neurone responded to several types of stimuli such as deep pressure and sudden temperature change. This was the case for those illustrated in Figs. 3 and 4. In contrast, alterations of activity of other units followed only one kind of stimulus although stimulus specificity may have been a feature for a particular region of the body, elsewhere other types being effective.

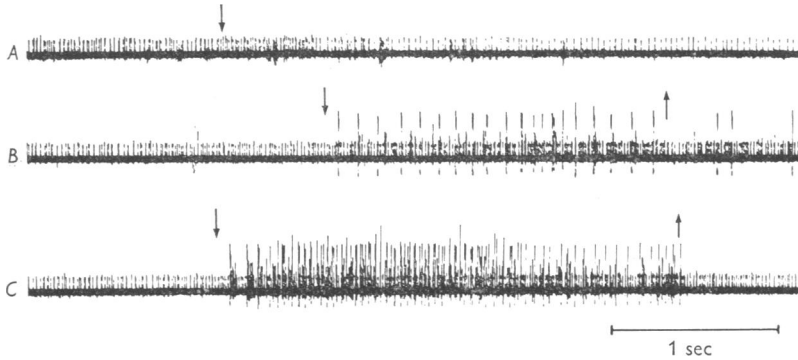


Fig. 3. Response of left T1 preganglionic fibres to firm squeezing of the paws. *A*: At the arrow the left forepaw was squeezed and the stimulus was maintained for the duration of the tracing. *B*: At the first arrow the right hind paw was squeezed, the stimulus ceasing at the second arrow. *C*: At the first arrow the left hind paw was squeezed, the stimulus ceasing at the second arrow. End-tidal CO<sub>2</sub>, 4.2%. Rectal temp. 37.6°C. Ten hours after spinal transection.

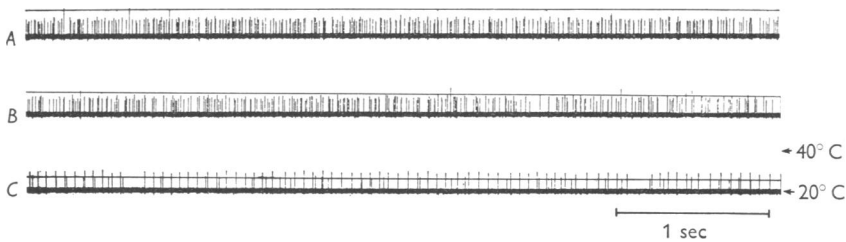


Fig. 4. Response of left T1 preganglionic fibres to cooling of the skin of the left forelimb. The upper trace of each record is the temperature of the left forelimb near the shoulder measured by a small thermistor inserted subcutaneously. *A*: Control. *B*: Just before this record ether vapour was sprayed on the skin over an area 3 cm in diameter centred about the thermistor location. *C*: Begins about 5 sec after the end of *B*. End-tidal CO<sub>2</sub>, 4%. Rectal temp. 37.6°C. Thirteen hours after spinal transection.

*Discharges evoked by electrical stimulation of afferent fibres*

*Reflex discharge to segmentally organized afferent sources.* A single volley of impulses in afferent fibres of a dorsal root or spinal nerve usually evoked a reflex discharge in the preganglionic ramus of the same segment. The response when recorded from an undivided preganglionic ramus varied

from a few units to a sufficient number to give a summated wave with irregular contours. Figure 5 shows a discharge in the L2 ramus which was evoked by stimulation of L2 dorsal rootlets. As the electrical stimulus was progressively increased, the size of the response also increased and the latency decreased. A reflex of this kind was seen at each spinal level studied: T1, T2, T3, T4, L1, L2, L3, L4. In the T1 preganglionic outflow, the reflex response was limited to a few elements, while for every other ramus, in one preparation or another, a greater number of fibres took part in the response.

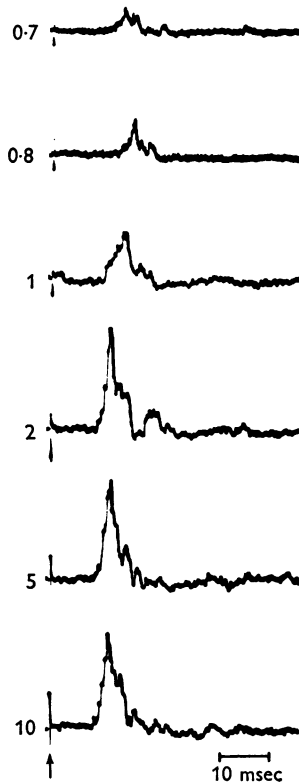


Fig. 5. Reflex response of preganglionic fibres in L2 ramus to single volleys of graded size in L2 dorsal rootlets. The time of the stimulus is indicated by an arrow below the lowest record. The figures to the left of each record give, in arbitrary potentiometric units, the intensity of the electrical stimulus (duration 0.1 msec) initiating dorsal root volleys. End-tidal  $\text{CO}_2$ , 4.8%. Rectal temp.  $37.3^\circ\text{C}$ . Eleven hours after spinal transection.

The reflex evoked by single volleys was not confined to the segment of dorsal root entry. Stimulation of afferent fibres entering nearby segments, also elicited a reflex in one preganglionic ramus. An illustration of this

appears in Fig. 6, constructed from recordings obtained from a T2 preganglionic ramus. Figure 6*A*, *B* and *C* represent the responses to progressively increased intensity of dorsal root stimulation. In some pathways of Fig. 6 (DRT1) the reflex substantially decreased in latency with increasing stimulus intensity, while stimulation of fibres entering other levels did not result in as marked a latency shift. A similar experiment is illustrated for the outflow at the lumbar level in Fig. 7. Dorsal rootlets of segments L3 to L5 reflexly excited some neurones of the L3 ramus. Once again, the reflex size varied with stimulus strength. Unfortunately, the relative size of the reflex discharge could not be used to judge the effective-

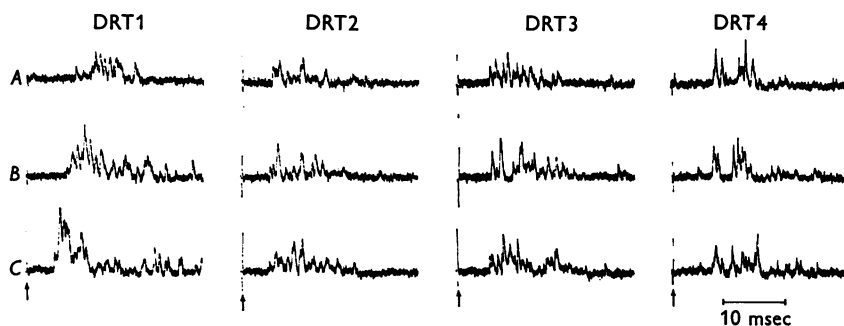


Fig. 6. Response of preganglionic fibres in T2 ramus to single dorsal root volleys. Each vertical row of records illustrates responses evoked by single-shock stimulation of filaments from the indicated segment. Stimuli intensity (of 0.1 msec pulses) progressively increased between *A* and *C* over an approximately 1–5 range. Stimulus time indicated by arrows. End-tidal  $\text{CO}_2$ , 4.4%. Rectal temp. 37.5° C. Eleven and one-half hours after spinal transection.

ness of afferent fibres from different segments. It was usually impossible to stimulate simultaneously all dorsal root fibres from one segment because of their short length and, therefore, frequently a larger proportion of afferent fibres were excited from one segment than from others. When stimuli were applied to the segmental spinal nerves, the largest reflex discharge in a preganglionic ramus was evoked from the afferent fibres of the same or the adjacent segment. Afferent fibres from more distantly located segmental nerves tended to produce a smaller discharge. Some correlation between the size of the reflex discharge and the number of fibres in the afferent nerve seemed to exist; larger nerves frequently gave rise to a larger reflex even if they entered the segment adjacent to the exit level of the ramus. Some preganglionic fibres with an active background discharge did not participate in the reflex initiated by dorsal root volleys but other tonically active fibres took part in the response.



On occasion, volleys of impulses from the dorsal roots or a spinal nerve would decrease the tonic discharge of a fibre. More direct evidence for inhibition of the reflexly evoked discharge by afferent fibres entering the same or adjacent segments was obtained in other experiments: these will be reported in a separate communication (Beacham & Perl, 1964).

Since afferent fibres from several segments could evoke a discharge in one preganglionic bundle, it was of interest to see whether or not the same elements participated in the reflex evoked by afferent axons entering the spinal cord at different levels. In Fig. 8*A*, stimulation of T1 afferent fibres evoked a response consisting of three or four units in the T1 preganglionic ramus. One unitary discharge was prominent because of its relatively large amplitude. The same large amplitude action potential appears after stimulation of T2 (Fig. 8*B*) and T3 (Fig. 8*C*) dorsal roots. However, a preganglionic axon whose action potential was of intermediate size responded regularly to T1 dorsal root stimuli (Fig. 8*A*) but did not participate in the reflex evoked from T2 or T3 afferent fibres. Excitatory convergence of afferent fibres from different segments expressed in this manner was frequently seen, but was clearly absent in many cases.

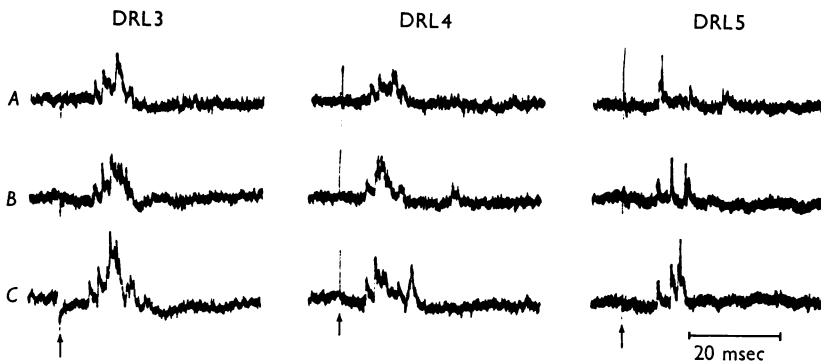


Fig. 7. Response of preganglionic fibres in L3 ramus evoked by single dorsal root volleys. Constructed as in Fig. 6 except that stimulus intensities were increased between *A* and *C* over a 1–10 range. End-tidal  $\text{CO}_2$ , 5%. Rectal temp.  $37.5^\circ\text{C}$ . Nine hours after spinal transection.

*Reflex latency.* The latency of the reflex evoked by stimulation of dorsal root fibres varied considerably from segment to segment and from preparation to preparation. Table 1 lists a representative sample of latencies determined from maximal responses of small populations of preganglionic fibres. The entries in the table are segregated according to the segment of origin of the preganglionic outflow and the entering segment of the dorsal root fibres. The reflex latency ranged from a minimum of 3.0 to over 15 msec. Since the distance from stimulating cathode to the spinal cord entrance

zone was always short (4–6 mm) and efferent pathways in the various experiments were approximately the same length (13–23 mm), large differences in latency were apparently the product of variation in the central mechanism. Note that the latencies were not necessarily shortest for the reflexes evoked by afferent fibres entering the same segment as the preganglionic ramus. The minimum central time was estimated for the shortest latencies observed (3.0–3.5 msec). A representative calculation follows for the reflex initiated by stimulation of the T1 dorsal root and

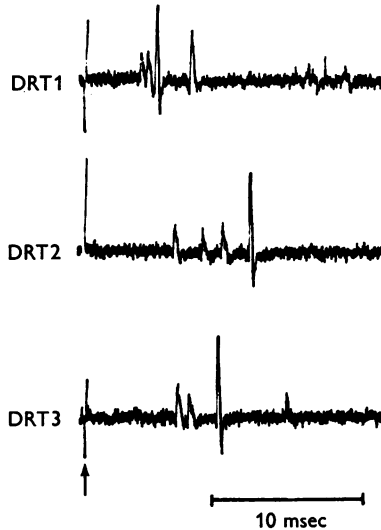


Fig. 8. Response of a small group of preganglionic fibres in T1 ramus evoked by dorsal root volley from the indicated segments. Stimulus intensities were supra-maximal for the preganglionic discharge. Rectal temp. 37.5° C. Twelve and one-half hours after spinal transection.

recorded from T2 preganglionic fibres (experiment *a'*, Table 1). The afferent conduction distance in this experiment was 5 mm. Other evidence suggests that the largest afferent fibres for such reflex discharges conduct between 30 and 40 m/sec (Beacham & Perl, 1964) so that about 0.25 msec of the reflex latency was occupied by conduction into the spinal cord. The distance from the site of recording on the preganglionic ramus to the ventral root exit from the spinal cord was 21 mm. The diameter of the most rapidly conducting preganglionic axons participating in this reflex is unknown; however, a figure of at least 15 m/sec would seem appropriate (Bishop & Heinbecker, 1930). Using this value, about 1.4 msec can be attributed to efferent conduction. Thus, on the information available, the time contributed by afferent and efferent conduction was 1.7 msec or less, leaving a central time of at least 1.3 msec from the 3.0 msec latency.

TABLE 1. Latency in msec of small population reflex discharges evoked by single volleys of impulses in the indicated dorsal roots (DR). Afferent conduction distance was short (4–6 mm) and stimulation intensity was maximal for reflex response. Efferent conduction distance varied from 13 to 23 mm. Each horizontal row contains results from one experiment and each value is the average of ten measurements. *a* and *a'* are from the same experiment. *b* and *b'* are from one experiment, but represent recordings made from different divisions of the preganglionic ramus

| Preganglionic<br>ramus | Source of afferent volleys |       |       |       |
|------------------------|----------------------------|-------|-------|-------|
|                        | DRT 1                      | DRT 2 | DRT 3 | DRT 4 |
| T1 <i>a</i>            | 5.2                        | 4.0   | 5.2   | 7.2   |
|                        | 4.1                        | 8.1   | 8.8   | —     |
|                        | 12.4                       | 13.4  | 15.5  | —     |
| T2 <i>a'</i>           | —                          | 7.7   | —     | —     |
|                        | 3.0                        | 3.4   | 3.8   | 4.5   |
| L2                     | DRL2                       | DRL3  | DRL4  | DRL5  |
|                        | 10.6                       | —     | —     | —     |
| L3 <i>b</i>            | 8.7                        | 9.8   | 10.5  | 11.3  |
|                        | —                          | 6.2   | 5.6   | 7.4   |
|                        | —                          | 7.4   | 6.6   | —     |
| <i>b'</i>              | 7.5                        | 6.4   | 5.7   | —     |

As in other reflex systems, increasing the intensity of the electrical stimulus to an afferent nerve shortened the latency of the reflex discharge. This point is illustrated in Figs. 5 and 6. When the reflex discharge consisted of only a few preganglionic fibres or if the preganglionic ramus was split so that the discharge of a few fibres was studied, the variation in reflex latency could be observed for individual elements. Figure 9 gives the latency of the response of a T1 preganglionic neurone for successive reflexes evoked by stimulation of the T1 dorsal rootlets (*A* and *B*) or the T2 dorsal rootlets (*C* and *D*). The reflex latency is shown on the ordinate with successive tests, at 2 sec intervals, plotted on the abscissa. With weak stimuli (*A* and *C*) considerable variation was observed in the reflex latency, but with more intense stimuli (*B* and *D*), the latency shortened and the variation in latency decreased. The decrease in latency variation was more marked for the reflex evoked by T2 dorsal root than for the one initiated by stimulation of the T1 root. Even with strong stimuli, which initiated large volleys, the variation in reflex latency of the highly responsive (firing index of 100) unit illustrated in Fig. 9 was considerable (0.9 msec). Furthermore, as previously noted in Table 1, the shortest latency did not occur in response to volleys from homonomous afferent fibres (cf. Fig. 9*B* and Fig. 9*D*).

*Reflexes evoked by electrical excitation of limb nerves*

Stimulation of the sciatic nerve was commonly found to evoke discharge of preganglionic neurones in the lumbar outflow, and rarely in the thoracic rami. An example of a typical reflex discharge in a lumbar ramus in

response to sciatic volleys is shown in Fig. 10. The numbers to the left of each record in Fig. 10 represent multiples for the stimulus threshold value for a dorsal root response. Characteristically, only one or a few units of a segmental preganglionic bundle responded to sciatic stimulation. A unit

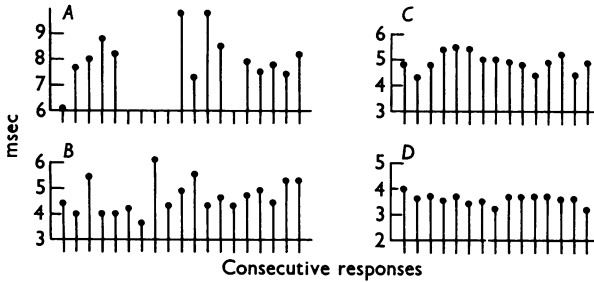


Fig. 9. Latency of reflex discharge of a T1 preganglionic fibre evoked by single T1 (*A* and *B*) and T2 (*C* and *D*) dorsal root volleys. Dorsal root stimuli delivered at 2 sec intervals (indicated on abscissa). Latency indicated in msec on ordinate. Consecutive responses plotted. Missing points in *A* indicate no response. Stimulus intensity for *B* and *D* was 50 times that in *A* and *C*. End-tidal CO<sub>2</sub>, 4%. Rectal temp. 37.5° C. Eight hours after spinal transection.

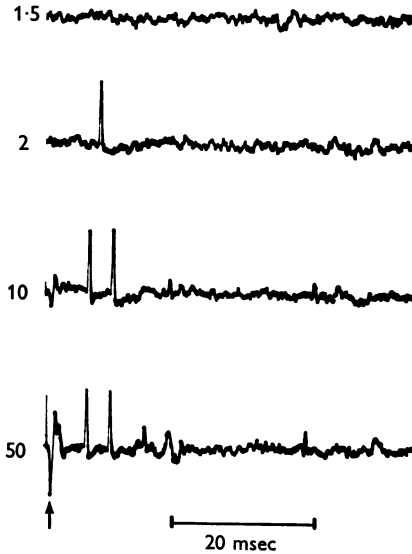


Fig. 10. Response of L3 preganglionic fibres evoked by single volleys in ipsilateral sciatic afferent fibres (initiated distal to hamstring branch). Figures at left of each record give the multiple of threshold for a dorsal cord response at L6 segment. The arrow indicates electrotonic recording of the afferent volley. After crushing the central end of the preganglionic ramus only this component of the evoked response remained. End-tidal CO<sub>2</sub>, 5.4%. Rectal temp. 37.5° C. Eight hours after spinal transection.

responding to sciatic volleys often was not excited by maximal stimulation of the segmental lumbar nerves, despite the fact that such nerves evoked reflex discharge of many other units in the same ramus. The reflex initiated from the sciatic nerve was relatively short in latency, at the minimum being only slightly longer (1 msec) than the shortest latencies observed from stimulation of dorsal roots. This short latency prompted study of the variations in latency for a single element of the same type as was described for units responding to dorsal root stimulation. As in the case of the reflex evoked by dorsal root afferent fibres, a large variation in reflex latency occurred after relatively small volleys but both the reflex latency and the variation decreased markedly as the strength of the electrical stimulus to the sciatic nerve was increased.

Stimulation of the ipsilateral nerves of the brachial plexus evoked a reflex discharge similar in some ways to that elicited from the sciatic nerve. The discharge to volleys in the forelimb nerves appeared in the T2 and T3 preganglionic rami and rarely involved more than a few units.

It was often possible to evoke responses of those preganglionic neurones, which were excited by electrical stimulation of the sciatic or brachial plexus nerves, by light mechanical stimulation of a restricted portion of the respective limb. Cutting the appropriate nerve abolished the response to mechanical stimulation although the discharge could still be initiated by electrical excitation of the central end of the cut nerve.

#### DISCUSSION

The present observations strongly support the concept that elementary reflexes for the sympathetic outflow exist within the spinal cord. The absence of such reflexes in the recordings from neural elements by Alexander (1946) and by Schaefer and his co-workers (Schaefer, 1960), may have been the result of failure to sample the entire sympathetic outflow. For example, these workers examined discharges in nerves to the heart, but the reflexes initiated by single volleys in thoracic afferent fibres of the type shown in Fig. 6 are usually not distributed to the cardiac nerves (Fernandez de Molina & Perl, unpublished). Alternatively, apparent lack of spinal sympathetic reflex response may have resulted from depression of activity due to spinal shock since both Alexander (1946) and Schaefer (1960) made observations relatively soon after spinal transection.

Both differences and similarities appear when the background discharge in the sympathetic outflow is compared to that existing in the skeletal muscle motor supply. In an acutely prepared spinal cat, limb extensor motoneurones rarely discharge in the absence of specific sensory stimulation while some limb flexor motoneurones have low frequency background

activity (Hunt, 1951; Perl, 1962). In contrast, the fusimotor (gamma-efferent) supply to the same skeletal muscles usually contains elements which show tonic discharge at frequencies of 10–60 sec (Hunt, 1951; Hunt & Paintal, 1958). Even though tonic discharge occurs in fusimotor neurones, a large proportion of those studied in acute spinal preparations are neither active nor respond to a variety of stimuli (Hunt & Paintal, 1958). Thus, the regular appearance of background activity in a fraction of the sympathetic preganglionic elements would seem to be closer to that occurring in fusimotor neurones than in the extrafusal motor supply.

Visceral responses in spinal animals have been shown to depend partially upon unknown blood-borne substances (Sahs & Fulton, 1940) or upon the adrenal release of sympathetic hormone (Brooks, 1933). The contribution of circulating humoral substances to the directly recorded preganglionic responses must certainly have been negligible. This is not the case in some of the earlier work on autonomic effector organ reactions in spinal preparations capable of either reflex contraction of skeletal muscle (Brooks, 1933; Downman & McSwiney, 1946) or of releasing adrenaline (Sherrington, 1906; Langley, 1924). How these factors might have influenced the observed responses is difficult to estimate, but cannot be ignored. While the effector destination of the reflex preganglionic discharges is purely speculative without additional evidence, it seems unlikely, on anatomical grounds, that upper thoracic preganglionic rami should end in the adrenal medulla (Hollinshead, 1936).

Direct (monosynaptic) connexions to intermedio-lateral horn cells from primary afferent fibres have been suggested although experimental studies establishing this are not quoted (Ingram, 1960). At least one anatomical study specifically denies the existence of afferent monosynaptic connexions to the sympathetic outflow (Szentágothai, 1948). In regard to this point, the reflex latency for preganglionic response to dorsal root stimulation may be contrasted with the values obtained for the monosynaptic excitation of skeletal motoneurones. After subtraction of afferent and efferent conduction times, Renshaw (1940) derived a central value of 0.5–0.8 msec for the most rapid dorsal to ventral root reflex in sacral segments. The minimum central time in the dorsal root to preganglionic arc was 1.3 msec, considerably longer than found for the two-neurone somatic reflex. Another difference between the somatic monosynaptic response and the simplest reflex to preganglionic neurones is the reflex variation on successive trials. The maximum variation in a reflex with monosynaptic timing to highly responsive flexor motoneurones was about 1 msec for the smallest volleys and shortened to approximately 0.1 msec for large volleys (Perl, 1962). Even with the strongest stimuli employed (20–50 times threshold), the variation in reflex latencies for a highly responsive preganglionic unit was

in the vicinity of 1 msec. Finally, the shortest latencies for dorsal root to preganglionic reflex frequently occurred between afferent fibres entering one segment and the preganglionic ramus of another segment. Once more, this is in contrast to the usual segmental organization of the two-neurone arc for skeletal motoneurons. These considerations lead to the conclusion that the present experiments give no evidence for monosynaptic connexions between dorsal root afferent fibres and preganglionic neurones. On the other hand, it is conceivable that under different circumstances such as the use of powerful facilitation, still shorter latency reflexes to preganglionic neurones could be demonstrated. Attempts to produce this form of facilitation of preganglionic neurones have been complicated by the existence of inhibitory effects from many afferent nerves (Beacham & Perl, 1964). It is also possible that synaptic or intermedullary conduction time for sympathetic reflexes is longer than that for the comparable somatic mechanisms. Thus, the possibility still exists that there are latent monosynaptic reflex connexions to sympathetic preganglionic neurones. However, the short latencies make it clear that spinal reflexes exciting the sympathetic outflow can have a relatively simple pathway.

The present study emphasizes that both convergence and specificity exists in the organization of the spinal sympathetic outflow. Furthermore, the outflow was found to be fractionated. Some preganglionic neurones exhibit background discharge and cannot be excited by stimulation of afferent fibres or receptors, while others respond to dorsal root volleys and do not have tonic activity. Greater or lesser specificity of adequate stimuli was found. These evidences for differences and specificity do not seem to be consistent with a common concept, derived from Cannon's work (Cannon, 1929), that the sympathetic outflow usually responds in a massive fashion and largely has importance in emergency or stressful situations. On the other hand, they are in harmony with current suggestions that many portions of the sympathetic system can be individually brought into action (Bard, 1960).

#### SUMMARY

1. In unanaesthetized, acutely decapitate cats, the activity of sympathetic preganglionic neurones was recorded from fibres in the segmental rami communicantes.

2. Some preganglionic fibres at each of the levels studied (T1, T2, T3, T4, L1, L2, L3, L4) exhibited background discharge. This background discharge often could be modified by noxious, thermal and mechanical stimuli. Both excitatory and inhibitory changes followed adequate stimulation. Occasionally, the afferent stimulation producing a reflex change in background discharge was specific in type and effective only in a localized

region. In other cases, several forms of stimuli provoked reflex changes from a large portion of the body.

3. A reflex discharge of preganglionic neurones was evoked by single electrical shocks delivered to dorsal roots, spinal nerves and limb nerves. The reflex was not strictly segmental in distribution, involved a few to many units, and often could be graded by gradation of the afferent volley. On the basis of reflex latency, variation in reflex latency and segmental distribution, it was concluded that these experiments did not give evidence for monosynaptic connexions to preganglionic neurones.

4. The argument was advanced that the present demonstration of localized and graded sympathetic discharges is in harmony with the concept that the sympathetic system is partly organized for discrete response.

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