CHANGES IN PRIMARY AFFERENT DEPOLARIZATION
OF SENSORY NEURONES DURING PERIPHERAL NERVE
REGENERATION IN THE CAT

By K. W. HORCH and S. J. W. LISNEY*

From the Department of Physiology, College of Medicine, University of Utah, Salt Lake City, Utah 84108, U.S.A.

SUMMARY

1. Micro-electrode recordings were made from normal and regenerating sural nerve fibres in cats. Increases in the excitability of the central terminals of these fibres after conditioning stimulation of other sural nerve fibres were taken as evidence for primary afferent depolarization.

2. At all recovery times studied the excitability changes seen were significantly less than those seen in control animals. Two factors contributed to the changes in primary afferent depolarization. First, the proportion of fibres that showed no evidence of primary afferent depolarization increased significantly. This proportion became smaller as recovery progressed. Secondly, where primary afferent depolarization was present, the magnitudes of the effects were slightly but significantly decreased compared with control values.

3. Excitability changes of the central terminals of sural nerve fibres were also measured after conditioning stimulation of the ipsilateral, unlesioned accessory sural nerve. One month after sural nerve transection there was a significant increase in the proportion of fibres showing no evidence of excitability changes following accessory sural nerve conditioning stimulation compared with control animals. Thus, the loss of primary afferent depolarization of regenerating sural nerve fibres was neither simply a consequence of desynchronization of the volley of impulses entering the spinal cord after conditioning stimulation of other regenerating sural fibres, nor due to fewer fibres being activated during conditioning stimulation of the lesioned nerves.

4. A possible explanation of these results is that after peripheral nerve crush or transection the central terminals of the damaged fibres retract or atrophy. Then as regeneration of the nerve proceeds, the central terminals of the fibres re-form.

INTRODUCTION

During earlier electrophysiological studies of regenerating nerve fibres it was observed that the incidence of dorsal root reflexes elicited from regenerating nerves was much less than in normal sural nerves (Horch, 1976, 1978). One month after nerve

* M.R.C. Travelling Fellow. Present address: Department of Physiology (Oral Biology), The Medical School, University of Bristol, University Walk, Bristol BS8 1TD.

transection or crush there were no dorsal root reflexes at all; after nerve transection the dorsal root reflexes returned over the course of a year but their frequency of occurrence never equalled that seen in control animals. Crushed sural nerves were not studied beyond one month.

A dorsal root reflex is a centrally generated, antidromically propagated action potential in a sensory nerve fibre and is a consequence of depolarization of the central terminals of that fibre. This primary afferent depolarization (p.a.d.) results from the synchronous action of numerous presynaptic contacts of other neurones on the central terminals of that particular fibre (for a full description see Schmidt, 1971). The loss of dorsal root reflexes in regenerating sural nerve fibres suggests a reduction in the amount of primary afferent depolarization, which might be due to a loss of presynaptic contacts or a reduction in their efficacy. In these cases there would be a loss of both dorsal root reflexes and primary afferent depolarization. There can, however, be primary afferent depolarization of a nerve without this resulting in a dorsal root reflex. This would occur if the depolarization were insufficient to initiate an action potential or if the antidromically propagated spike were blocked at some point. Atrophy of the central portions of primary afferent fibres as a result of nerve lesion could cause these effects.

The present experiments were carried out to see whether there were detectable changes in primary afferent depolarization of individual regenerating sural nerve fibres. A brief account of some of the experiments has been published (Horch & Lisney, 1980a).

METHODS

Experiments were performed on adult cats (2·5–6·5 kg) anaesthetized with sodium pentobarbitone (42 mg/kg I.P.). The left sural nerve was exposed under aseptic conditions and either cut with fine scissors or repeatedly crushed over a length of 2–3 mm with watchmaker's forceps just distal to the popliteal fat pad. Where the sural nerve was transected no attempt was made to approximate the nerve stumps. The skin wound was sutured and procaine penicillin (300,000 units, I.M.) given.

Preparation for acute experiments

Three cats were prepared for each of the following recovery periods: 1, 2, 3 and 6 months after nerve crush and 3, 6, and 9 months after nerve transection. Four animals were examined 1 month after nerve transection, and several unoperated cats were used to provide control data. After the appropriate time had been allowed for nerve regeneration the animals were re-anaesthetized with sodium pentobarbitone and the trachea, left carotid artery and left radial vein cannulated. Supplementary doses of anaesthetic (2·5 mg/kg I.V.) were given as required during the preparative surgery and every 2 hr once the animals had been paralysed with gallamine triethiodide. Rectal temperature was kept close to 37·5 °C using radiant heat and the animals were artificially ventilated to maintain end-tidal CO_2 % between 4·5 and 5·0 %. Systemic arterial blood pressure was monitored and data collected only if the mean blood pressure exceeded 75 mmHg. The left sciatic nerve was dissected free in the sciatic notch, the surrounding skin made into a pool to retain liquid paraffin, and the nerve placed on a pair of Pt–Ir wire electrodes. The sural nerve was exposed between the knee and ankle and an oil pool formed around it. The two branches of the nerve were dissected free distally and each placed on a pair of electrodes. In some experiments the accessory sural nerve was also prepared for electrical stimulation. A site for micro-electrode recording from the sural nerve was prepared just distal to where it joins the sciatic nerve. A lumbar laminectomy was carried out, an oil pool created, and the dura overlying the spinal cord removed to expose the dorsal root entry zones caudal to L4.

Isolation and identification of single sural nerve fibres

A glass micro-electrode (filled with 4 M-NaCl, impedance 20–40 MΩ at 1 kHz) was introduced through an opening in the perineurium and slowly advanced while the sciatic nerve was stimulated with 100 μsec pulses at an intensity which had previously been found to be supramaximal for all the myelinated sural nerve fibres. When a single unit was isolated its response latency to sciatic nerve stimulation at twice threshold was measured and used in calculating the conduction velocity of the fibre. Then the two peripheral branches of the sural nerve were stimulated in turn to find which contained the fibre being studied. Finally, once the testing of excitability changes had been completed (see below), the receptor type of the fibre was determined using the classification of Horch, Tuckett & Burgess (1977). Fibres which could not be activated by gentle mechanical stimulation of the skin were classified as inactive. This group would include fibres blocked distal to the recording site, possibly because they were damaged during the dissection, as well as fibres associated with high threshold mechanoreceptors.

Measurement of excitability changes

To stimulate the central arborizations of sural fibres, six electropolished tungsten wire electrodes, insulated except at their tips, were mounted 2 mm apart in a longitudinal array. This array was inserted into the right side of the cord between L7 and S1 and angled towards the left (experimental) side. It was lowered to a depth of 1·6–1·9 mm from the cord surface (Eccles, Schmidt & Willis, 1963). During cord stimulation the electrodes were connected to give three adjacent pairs; at any particular time one pair was made the cathode while the anode was a metal clip fixed to nearby muscle.

Changes in the excitability of the central portions of individual nerve fibres were measured in terms of the current required to excite them in conditioning-testing trials in which each test stimulus to the spinal cord was preceded by a conditioning stimulus to some other group of nerve fibres. The test stimulus consisted of a train of seven constant current pulses (0·2 msec duration, 0·5 msec between the onset of successive pulses), the first pulse being 70 % of the amplitude of the last and the intermediate ones incrementing at 5 % intervals. In preliminary experiments it was found that the stimulus intensity required to evoke an antidromic response to the last pulse of the train was the same as that required when a single shock (0·2 msec duration) was used, indicating that the earlier pulses of the train had no significant effect on the excitability of the central terminals of the fibres. The steps in testing the excitability of the central portions of a nerve fibre were as follows. First, the antidromic response latency to a single cord stimulus (0·2 msec duration, 1·5 × threshold) was measured. Knowing this latency the amplitude of the train stimulus could be adjusted until the unit responded consistently to the last or next-to-last pulse of the train. This became the test stimulus, and five successive test stimuli (at 1/sec) constituted a control trial. A digital computer measured the response latency to each of the five stimuli and made a permanent record of the maximum, minimum and mean latencies for that trial. In conditioning-testing trials five of the same test stimuli were preceded in each case by a conditioning stimulus. When there was an increase in the excitability of the central portions of the nerve fibre, less current was needed to excite it and this was reflected by a reduction in the response latency because the terminals were excited by an earlier pulse in the train. For example, a reduction in the mean response latency in a conditioned trial of 1·0 msec indicated that 10 % less current was required to excite the central terminals of the unit during that trial. Five seconds were allowed between each alternate control and conditioning-testing trial. By noting the maximum and minimum latencies it was possible to detect false triggers, such as could be produced by stimulus artifacts and indirect responses such as dorsal root reflexes. Trials in which there were false triggers were rejected.

For every fibre the changes in central excitability following conditioning stimulation of the sciatic nerve (below threshold for that unit) and the peripheral sural branch that did not contain the unit (at twice threshold for the maximal myelinated fibre response) were measured at a conditioning-testing interval of 30 msec for each of the three pairs of cord electrodes. A full sequence of conditioning-testing trials was then carried out using the pair of cord electrodes at which the excitability changes were greatest. For many fibres this pair was not necessarily the pair which showed the lowest threshold for stimulation. Various conditioning-testing intervals, alternating with controls, were tested in a random fashion. In some experiments conditioning stimulation of the accessory sural nerve was also carried out.

In four control experiments excitability changes following mechanical stimulation of the skin

were studied. Three moving-coil stimulators (stylus diameter 0·3 mm) were equally spaced along the proximal–distal axis of the sural nerve field. They were adjusted so that the discharge recorded from the sural nerve to a single indentation of the skin (100 Hz haversine, 150 μm displacement) was approximately the same for each stimulator. This stimulus could be expected to excite a variety of cutaneous mechanoreceptors (Horch & Burgess, 1976; Tuckett, Horch & Burgess, 1978).

Quantification of excitability changes

Examples of the excitability changes in the central portion of individual sural nerve fibres following conditioning stimulation of other sural nerve fibres are shown in Fig. 1. From plots such as these, three measurements were made for each fibre. The first was the greatest change in central excitability observed (greatest effect), expressed as percentage of the control central threshold and measured to the nearest 5 %. The second was the conditioning-testing interval at which the greatest effect occurred, taken to the nearest 10 msec. The third was the rate at which the excitability changes died away at longer conditioning-testing intervals. This decay was approximated to an exponential decay, and so measured in terms of the exponential decay constant.

Two-tailed Mann–Whitney U tests and χ^2 tests of contingency tables were used to assess the significance of differences between control and experimental groups.

RESULTS

The object of inserting the array of cord-stimulating electrodes from the contralateral side and to the depth at which excitability changes are greatest (see Methods) was so that there would be a better chance of stimulating the terminal portions of the fibres rather than the more distant parent axons (Schmidt & Willis, 1963; Whitehorn & Burgess, 1973). To test whether this procedure was effective, single unit recordings were made from filaments dissected from the central cut end of one of the peripheral branches of the sural nerve. Changes in excitability of the central terminals of the fibre following conditioning stimulation of the other peripheral branch of the sural nerve were then measured for each of the three pairs of cord electrodes. Collision experiments (see Horrobin, 1966; Lisney & Matthews, 1978) combining stimulation of the fibre in the dorsal columns at L4, the spinal cord and the sciatic nerve were carried out to see whether the cord electrodes were exciting the main axon or one of the axon collaterals. For eight of the nine Aα fibres tested, the pair of cord electrodes which showed the greatest excitability changes was exciting a branch in which there was a significant conduction time (0·2–0·9 msec) to the main axon. For the other unit, the conduction time (0·1 msec) was not considered significant in view of the likely errors in measuring the latencies. Thus by positioning the cord electrodes in the way described, the stimuli usually excited terminal branches rather than the main axons of the sural nerve fibres.

Because the medial branch of the sural nerve was invariably smaller than the lateral one, different numbers of fibres were excited depending upon which was being used for conditioning stimulation; however, this did not appear to affect the results. Indeed, it was found that activity in just a limited number of Aα fibres was sufficient to produce a saturated level of primary afferent depolarization. The electrical conditioning stimuli to the branches of the sural nerve that we used were supramaximal for all the myelinated fibres.

Primary afferent depolarization of normal sural nerve fibres

Complete conditioning-testing sequences were obtained for 156 fibres from seven cats. Sixteen showed no excitability changes with conditioning stimulation of the branch of the sural nerve which they were not in, but eight of these did show an effect following conditioning stimulation of the sciatic nerve at an intensity below threshold for that particular fibre. The results for three fibres that did show excitability changes with sural nerve conditioning stimulation are shown in Fig. 1. From plots such as these the greatest change in central threshold, the conditioning-testing interval at which this occurred, and the decay constant were measured. The distributions of the values obtained are shown in Fig. 2. Fibres with Aδ conduction velocities, whether or not they could be activated by gentle mechanical stimulation of skin, tended to
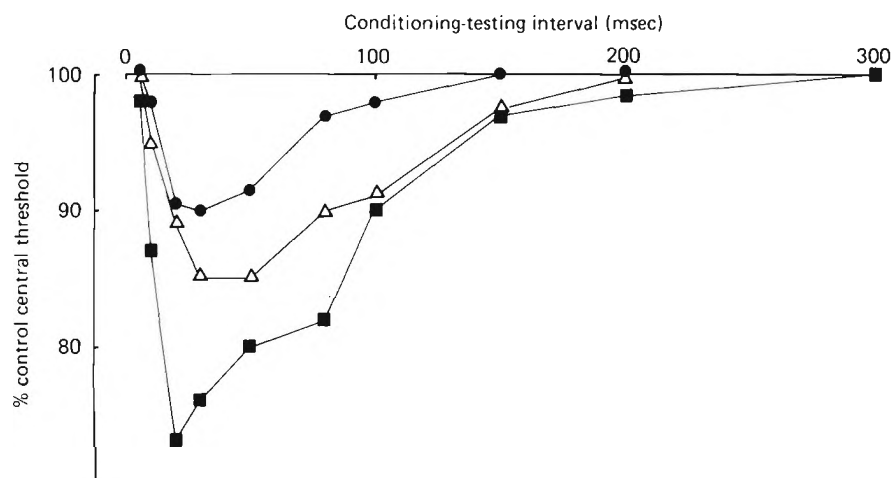
Fig. 1. Changes in excitability of the central terminals of three sural nerve fibres following electrical conditioning stimulation of other sural nerve fibres. The three fibres, one associated with a Type 1 mechanoreceptor ($\triangle$) and the other two with GI hair receptors (● ■), were isolated during the same experiment. The results for the two fibres associated with GI hair receptors show the variation in the magnitude of the effects seen for different fibres of the same receptor type with the same conditioning stimulus.

show smaller excitability changes than fibres conducting at velocities in the Aα range, but there were both Aα and Aδ fibres that showed no excitability changes at all after sural nerve conditioning stimulation. Within the Aα group of fibres there appeared to be no relationship between receptor type and the magnitude of the excitability changes, but because of the small sample sizes involved this could not be tested statistically.

Primary afferent depolarization of regenerating sural nerve fibres

After nerve injury there were marked alterations in primary afferent depolarization of the fibres. The most striking feature was an increase in the number of fibres which showed no evidence of changes in central excitability. For those fibres that did show an effect, there were small, but in some instances significant, differences in the magnitude and time course of the excitability changes.

One month after sural nerve transection virtually none of the 133 fibres tested showed evidence of primary afferent depolarization following sural nerve conditioning stimulation (Fig. 3A). Some 72 % of them did show evidence of an effect after conditioning stimulation of the sciatic nerve below threshold for the particular unit, but the magnitude of the effects were significantly less than in controls. By three months after transection there had been a significant reduction ($P < 0.001$) in the proportion of fibres showing no effect with sural nerve conditioning stimulation and from three to six months there was a further reduction ($P < 0.001$) in this proportion. Between six and nine months the proportion of the population showing no effect did not alter significantly but there was still a greater proportion of fibres showing no effect ($P < 0.001$) compared with unlesioned nerves (compare Figs. 2A and 3A). Where there were measurable changes in central excitability at these four recovery
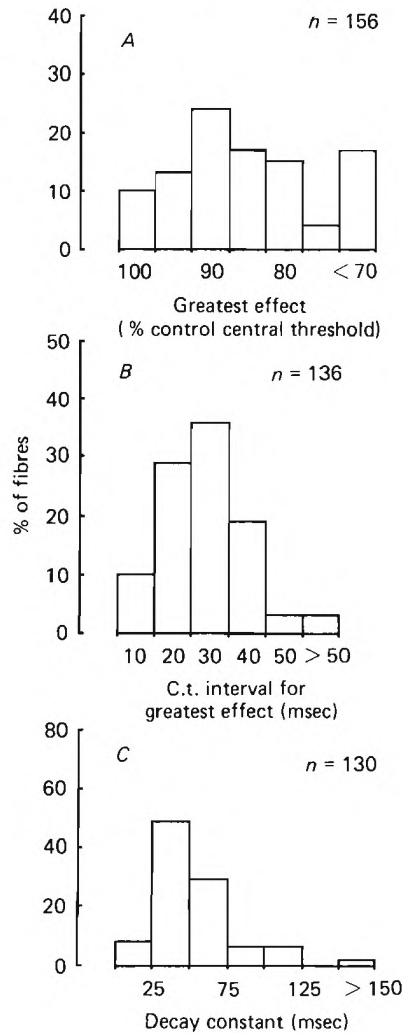
Fig. 2. Histograms showing the distribution of: *A*, the magnitude of the greatest excitability change (greatest effect), *B*, the conditioning-testing (c.t.) interval at which the greatest effect was produced and *C*, the rate at which the excitability changes died away for unlesioned sural nerve fibres following conditioning stimulation of other sural nerve fibres. In each case the number of observations is given.

times, the size of the effects compared with the control group showed no consistent trends.

One month after sural nerve crush all but a few of the fibres tested showed no evidence of primary afferent depolarization with sural nerve conditioning stimulation (Fig. 3*B*). Following sciatic nerve conditioning stimulation below threshold for each particular unit, sixty-four of the 111 fibres showed evidence of an effect, but the size of these effects were less than those seen in control experiments. By two months after crush there had been a significant reduction ($P < 0.001$) in the proportion of fibres in the sample showing no evidence of primary afferent depolarization with sural nerve
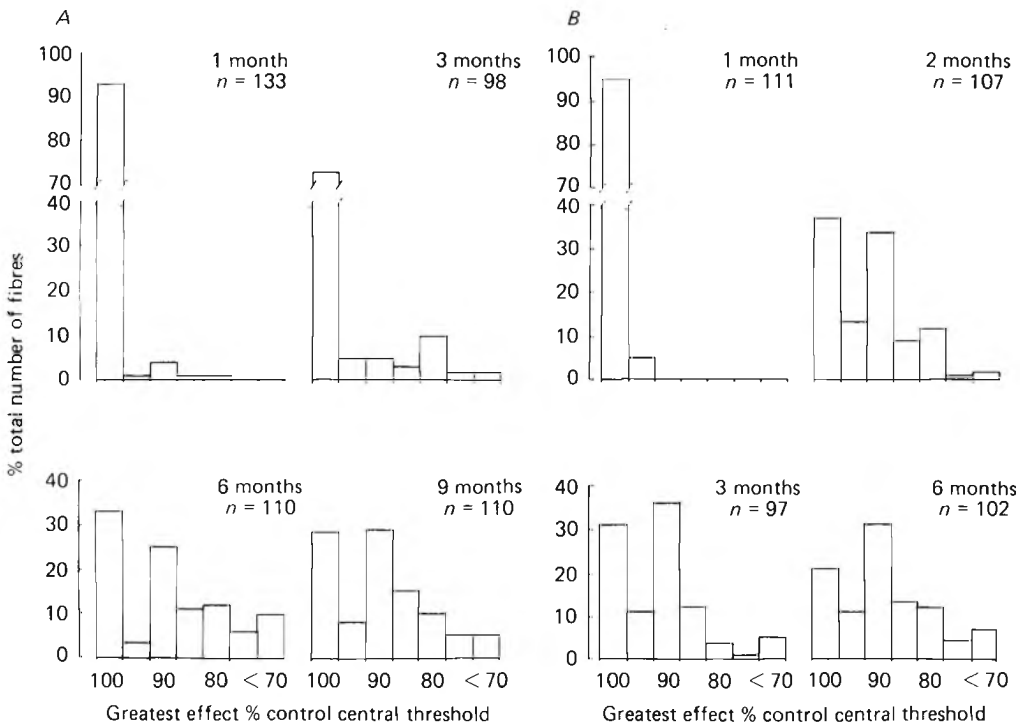
Fig. 3. Histograms showing the distribution of the greatest excitability changes of the central terminals of regenerating sural nerve fibres following conditioning stimulation of other regenerating sural nerve fibres at four recovery times after *A*, nerve transection and *B*, nerve crush. In each case the number of observations is given.

conditioning stimulation. Although it appeared that there were further reductions in the proportion of fibres showing no effect from two to three months and again from three to six months after crush, the differences were not significant. The proportion of the total number of fibres tested at six months that showed no evidence of primary afferent depolarization was significantly greater ($P < 0.002$) than for control sural nerves. As with the transected sural nerves, there were no consistent trends in the differences in size and time course of the effects seen for fibres that did show evidence of primary afferent depolarization.

Whether or not a regenerated fibre showed evidence of primary afferent depolarization seemed independent of its having formed a functional ending in the skin. Some fibres which had established a functional peripheral connexion showed no change in central excitability after sural nerve conditioning stimulation whereas others which could not be activated from the skin showed large excitability changes after conditioning stimulation. While some of the latter may have been blocked distal to the peripheral stimulation site, giving a false impression of having no functional ending in the skin, this was almost certainly not the case for the majority of these fibres.

Primary afferent depolarization with conditioning stimulation of the accessory sural nerve

It is possible that some of the reduction in primary afferent depolarization seen shortly after nerve lesion was because at that time many of the regenerating nerves had not yet crossed the injury site (Horch & Lisney, 1980 b), and so the conditioning stimuli applied to the distal portions of the sural nerve would not have excited as many fibres as in control experiments. Also, the conduction velocities of those fibres that were excited would have been slower than in control nerves (Horch & Lisney, 1980 b), and so the volley entering the spinal cord would have been more temporally dispersed. If either or both of these factors were a major consideration in explaining the loss of primary afferent depolarization of the regenerating nerve fibres, one would expect the primary afferent depolarization seen in these nerves after conditioning stimulation of an undamaged nerve to be of the same order as that seen in normal sural nerve fibres. The accessory sural nerve was chosen for such a test because its area of innervaton is adjacent to that of the sural nerve. These experiments were combined with ones in which changes in central excitability of sural nerve fibres following conditioning stimulation of other sural fibres was being measured so that direct comparisons between the two sets of results could be made.

For normal sural nerve fibres the magnitude and time course of the changes in central excitability seen following accessory sural nerve conditioning stimulation differed significantly from those seen after sural nerve conditioning stimulation (compare Figs. 2 and 4). The magnitudes of the greatest effects were smaller ($P < 0.0001$) with accessory sural nerve stimulation, the conditioning-testing interval at which the greatest effect occurred tended to be greater ($P < 0.05$), and the excitability changes died away more quickly ($P < 0.0001$).

One month after transection of the sural nerve, twenty-one of the 108 fibres tested with accessory sural nerve conditioning stimulation showed evidence of primary afferent depolarization. This was a significant reduction in the proportion of fibres showing an effect compared with control ($P < 0.001$). For the fibres that did show excitability changes, only the decay constants differed significantly from control, the values being smaller ($P < 0.05$). Comparing the results obtained one month after transection for accessory sural and sural nerve conditioning stimulation it was found that with accessory sural nerve stimulation a greater proportion of fibres showed evidence of primary afferent depolarization ($P < 0.01$). Since only nine of 133 fibres showed excitability changes with sural nerve conditioning stimulation, tests for differences in the magnitude or time course of the excitability changes seen with the two conditioning stimuli were not possible. Nine months after sural nerve transection, sixty-one of the ninety-eight fibres tested with accessory sural nerve conditioning stimulation showed evidence of primary afferent depolarization, a proportion not significantly different from that in controls. For the fibres that did show changes in central excitability, the magnitude and time course of the effects were the same as in control animals. Compared with the effects seen with conditioning stimulation of the sural nerve nine months after transection, there was no significant difference in the proportion of fibres showing no effect, but accessory sural nerve conditioning stimulation produced effects which were smaller ($P < 0.01$) and which died away
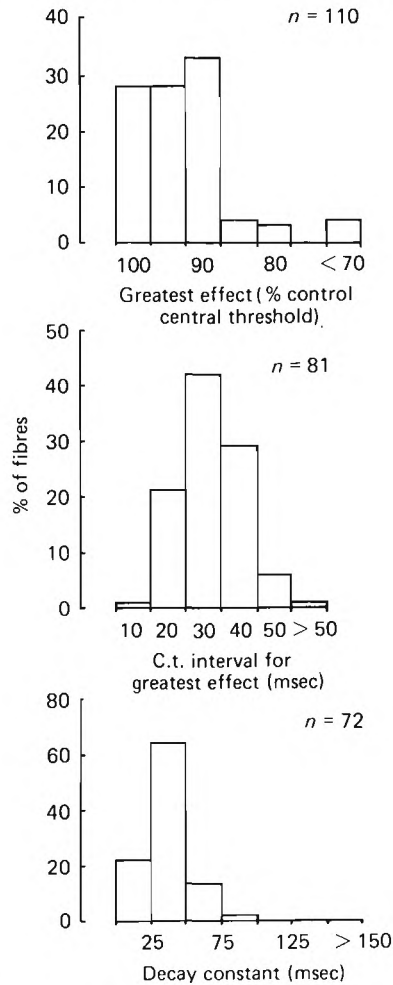
Fig. 4. Histograms showing the distribution of: *A*, the magnitude of the greatest excitability change (greatest effect), *B*, the conditioning-testing (c.t.) interval at which the greatest effect was produced and *C*, the rate at which the excitability changes died away for unlesioned sural nerve fibres following conditioning stimulation of accessory sural nerve fibres. In each case the number of observations is given.

quicker ($P < 0.005$). There was no difference in the conditioning-testing intervals at which the greatest excitability changes were seen.

Primary afferent depolarization following mechanical stimulation of the skin

Results have been reported which suggest that primary afferent depolarization of cutaneous afferents is organized in such a way that stimulation of other afferents with nearby receptive fields produces greater depolarization of the central terminals than does stimulation of afferents with more distant receptive fields (Schmidt, Senges & Zimmermann, 1967; Jänig, Schmidt & Zimmermann, 1968; Whitehorn & Burgess, 1973). We examined this in control animals using mechanical stimulation of three areas of the sural nerve field as conditioning stimuli.

Complete sets of results were obtained for sixty-eight fibres, all of which showed evidence of primary afferent depolarization after conditioning stimulation of the sciatic nerve at an intensity below threshold for the fibre being investigated. All the types of cutaneous mechanoreceptor described by Horch *et al.* (1977) were represented in the sample. Thirteen fibres showed no evidence of primary afferent depolarization following mechanical conditioning stimulation with any of the three stimulators. The fibres in this group tended to inervate tonic receptors, i.e. G2, F2, Type 1. For some of the fibres that did show an effect with mechanical conditioning stimulation the magnitude of the greatest effect fell away with increasing distance between the fibre's receptive field and the site of mechanical stimulation. For other fibres, however, there was no sign of this relationship. For each of the fifty-five fibres that showed excitability changes, the magnitude of the greatest effect measured with each stimulator was plotted against the distance from the centre of the fibre's receptive field to the centre of the contact area of the stimulator stylus. Whether considering the results for each of the four experiments individually or grouping them all together, there was no correlation between the magnitude of the greatest effect and the distance between receptive field and stimulation site. Thus with the procedures described, we were unable to obtain clear-cut evidence that presynaptic inhibition of sural nerve fibres is spatially organized within the confines of the field of innervation of the sural nerve.

DISCUSSION

There is scant information available about primary afferent depolarization of identified, single cutaneous afferents, whether it be following electrical conditioning stimulation of nerves supplying the neighbouring area of skin or following mechanical stimulation of the skin itself. Eccles *et al.* (1963) obtained direct evidence of primary afferent depolarization of cutaneous afferents during experiments in which intracellular recordings were made from the terminal portions of nerves but the number of fibres studied was small. Darian-Smith (1965), Schmidt *et al.* (1967), Jänig *et al.* (1968) and Whitehorn & Burgess (1973) took changes in central excitability of single cutaneous afferents as being indicative of primary afferent depolarization but the number of fibres investigated was not mentioned in any of these papers. The results of the present experiments do not appear to be in conflict with this earlier work but it is not possible to compare the magnitude of the excitability changes seen in the different sets of experiments because of the differences in the techniques used to measure them.

In the present work the magnitude of the greatest effect seen and the duration of the effect varied considerably from fibre to fibre (Fig. 2). It may be that there is a wide variation in the degree of primary afferent depolarization of different fibres in the sural nerve, even between fibres with the same type of receptor ending, but there are other factors arising from the experimental arrangement that could have contributed to this variation. There were some differences between animals, perhaps due to small differences in the positioning of the array of electrodes in the spinal cord. Differences between units in a particular animal may have been because some sural fibres enter the cord via rootlets other than L7 and S1 (Ekholm, 1967), so the cord electrodes might not have been in the best position for demonstrating excitability changes of the terminal portions of these fibres.

The loss of primary afferent depolarization was most pronounced one month after crush or transection of the nerve. This finding is consistent with a recent observation of Devor & Wall (1980) who found that in rats in which the sciatic nerve on one side had been cut and ligated, dorsal root potentials recorded at the L5 level following electrical stimulation of the sciatic nerve were much reduced on the operated side compared with those seen on the other side. Dorsal root potentials are indicative of primary afferent depolarization (for a full discussion see Schmidt, 1971). In the present work primary afferent depolarization reappeared in crushed sural nerve fibres more quickly than in transected ones, but in both the recovery was still incomplete six and nine months, respectively, after the injury.

How can the loss and return of primary afferent depolarization of damaged sural nerve fibres be explained? The experiments in which conditioning stimulation of the unlesioned accessory sural nerve was carried out eliminate the possibility that the loss was purely a consequence of desynchronization of the volley of impulses entering the spinal cord after conditioning stimulation of lesioned sural nerve fibres. These experiments also rule out the possibility that it was simply because fewer fibres were activated during conditioning stimulation of the lesioned nerves. One possible explanation is that after nerve crush or transection the central terminals of the fibres retract or atrophy: then as regeneration proceeds the central terminals re-form. If this is the case the present experiments suggest that establishing a function peripheral ending in the skin is not a prerequisite to regeneration of the central terminals. Some of the fibres at the longer recovery times showed no evidence of primary afferent depolarization but they were excited by skin stimulation whereas there were other fibres that did show primary afferent depolarization but apparently had no peripheral connections. Csillik & Knyihár (1978) have suggested that before the central terminals of injured nerves will re-form, normal patterns of axoplasmic transport must be re-established.

The retraction of primary afferent nerve terminals after injury would contribute to the loss of primary afferent depolarization in two ways. First, there would be a reduction in the input from these nerves to the pool of interneurones responsible for producing primary afferent depolarization. Secondly, there would be a loss of axoaxonic synapses from these interneurones onto primary afferent fibres because of the retraction of the afferent nerve endings. There is histological (Knyihár & Csillik, 1976; Moradian & Rustioni, 1977), histochemical (Csillik & Knyihár, 1975; Jessell, Tsunoo, Kanazawa & Otsuka, 1979; Devor & Claman, 1980) and electrophysiological (Horch, 1976) evidence for degenerative changes in the spinal cord of the central portions of primary afferent fibres after injury of their peripheral portions. Similar changes have been reported for trigeminal sensory nerves, particularly nerves associated with the teeth (Grant & Arvidsson, 1975; Westrum, Canfield & Black, 1976; Anderson, Rosing & Pearl, 1977; Gobel & Binck, 1977; Westrum & Canfield, 1977a, b, 1979). Equally important is the finding that some of the histochemical changes associated with primary afferent endings in the spinal cord were reversed if peripheral nerve regeneration was allowed to take place (Csillik & Knyihár, 1975, 1978).

An alternative factor which could contribute to the loss of primary afferent depolarization would be a dependence of the interneurones responsible for producing

this effect on a trophic influence from the primary afferent fibres. Crush or transection of the primary afferents could interrupt this trophic influence and it would be re-established when the fibres regenerate peripherally. We have no evidence for or against this suggestion, but if such a trophic influence exists the reduction in primary afferent depolarization seen with accessory sural nerve conditioning stimulation could only be explained if there was a shared interneuronal pool. We have not examined the effects of joint sural and accessory sural nerve conditioning stimulation to see whether or not such a pool exists.

The electrophysiological observations reported here and the histochemical results of Csillik & Knyihár (1975, 1978) strongly suggest a loss and then a re-establishment of primary afferent nerve terminal contacts in the spinal cord after peripheral nerve injury. It seems reasonable to propose that during the recovery period, when primary afferent terminals are growing and making new connexions with other neuronal elements in the dorsal horn of the spinal cord, there is a potential for new connexions to be made as well as the re-forming of old ones. One might then imagine that the central nervous system would take advantage of this to compensate for the inappropriate peripheral connexions that are a consequence of nerve transection. Indeed, there is evidence that suggests that the central nervous system is able to take limited advantage of this potential plasticity (Horch & Burgess, 1980).

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