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MECHANISMS FOR DIFFERENTIAL BLOCK AMONG SINGLE MYELINATED AND NON-MYELINATED AXONS BY PROCAINE

BY DONALD N. FRANZ AND ROGER S. PERRY

From the Department of Pharmacology, University of Utah, Salt Lake City, Utah, U.S.A.

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SUMMARY

1. The differential sensitivity of saphenous nerve fibres in the cat to block by procaine HCl was re-examined by recording identifiable unit action potentials from small nerve filaments.

2. Small myelinated axons were blocked more quickly than large myelinated axons, but this differential effect could not be accounted for by differences in anaesthetic concentration requirements.

3. The onset of block in non-myelinated axons was slower than or equal to that of small myelinated axons depending on anaesthetic concentration.

4. Absolute differential block of non-myelinated and small myelinated axons was obtained by limiting the length of axons exposed to procaine to 2 mm.

5. Differential rates of blocking among myelinated axons appear to depend on differences in the length of axons that must be exposed to blocking concentrations of procaine and to arise from the irregular distribution of such concentrations within an exposed nerve.

INTRODUCTION

Previous investigations of the differential block of nerve fibres by local anaesthetics indicate that small axons are generally more susceptible to block than are larger axons (Gasser & Erlanger, 1929; Heinbecker, Bishop & O'Leary, 1934; Matthews & Rushworth, 1957; Nathan & Sears, 1961). In seeking an explanation for differential effects of local anaesthetics according to fibre size, Nathan & Sears (1961, 1962) showed that absolute differential block of nerve fibres in spinal roots could be produced not only by critically low concentrations of procaine but also by solutions critically deficient in sodium. They therefore concluded that differential blocking was due to differences in critical or minimal concentrations (C_m) necessary to block fibres of various sizes and proposed that such differences must

reflect differences in safety factor for conduction between fibres of different sizes. In contrast to theories based on differences in surface to volume relationships that do not adequately account for observations that the order of recovery is reversed, the theory proposed by Nathan & Sears satisfies the order of both blocking and recovery.

All the previous observations including those of Nathan & Sears (1961) were based on the disappearance of certain elevations from compound action potentials recorded from whole nerve trunks or spinal root filaments. Such elevations represent the sum of numerous individual action potentials, and the details of behaviour of individual axons cannot be observed. However, in any condition in which total block is preceded by progressive slowing of conduction velocity, the loss or the reduction of certain elevations can occur not only by blocking but also by slowing conduction in individual axons at different rates. In the latter case, late components of original elevations may disappear into the base line before their axons are actually blocked (cf. Fig. 8 in Paintal, 1965). In the present study such uncertainties were avoided by recording from small bundles or filaments of axons that contained only a few active fibres, thereby permitting continuous observation of the all-or-none conduction in individual axons at all times (Franz & Iggo, 1968). This discrete information is not available when recording from larger populations of axons.

Our studies indicate that differential block of peripheral nerve by local anaesthetics is attributable to differences in the critical length of axons that must be blocked rather than to differences in minimal concentrations necessary to block axons of different sizes. A preliminary report of some of this work has been presented (Perry & Franz, 1970).

METHODS

Experiments were conducted on adult cats anaesthetized with sodium pentobarbitone, 35–40 mg/kg, intraperitoneally, supplemented by smaller intravenous doses as required. Deep body temperature was maintained near 37° C by a thermostatically controlled external heater. The saphenous nerve was exposed between the knee and groin and prepared for stimulation and recording from fine nerve strands as shown in Fig. 1 and described previously (Iggo, 1955, 1957; Chambers & Franz, 1968). The nerve was crushed with a ligature distal to the recording site. Conduction distances were between 62 and 85 mm.

Midway between stimulating and recording sites, a segment of nerve trunk was dissected away from surrounding tissues and carefully desheathed under microscopic control (15–30 ×) with the aid of microscalpels. This procedure exposed individual, bare fascicles of axons as shown in the enlarged diagram of Fig. 1 except that the perineurium was left intact. Care was taken to avoid tension, rotation or constriction of the nerve or interruption of its blood supply. The exposed section of nerve was passed through either of two types of plexiglas bathing chambers (Figs. 1 and 7.4) by means of a slit just above the floor of the chamber. The base of the chamber was

then sealed water-tight by applying a liquid solution of 4% agar in normal saline (Andersen, Eccles & Loyning, 1964) around the open slit (Figs. 1 and 7A). The liquid agar was applied at a temperature of 40–43° C and it rapidly congealed upon cooling a few degrees. The top of the open chamber extended above the level of mineral oil which covered the rest of the exposed nerve and tissue. Mammalian-Locke solutions alone or containing various concentrations of procaine hydrochloride (0·015–0·5%) at 37° C were washed in and suctioned off by tubes leading into the chamber. The Mammalian-Locke solution had the following composition (mM): NaCl, 154; 5·6; $CaCl_2$, 2·2; $NaHCO_3$, 1·8; glucose, 5·0. The pH of the solutions was between 7·0 and 7·2. Bathing solutions could be completely exchanged within 30 sec as tested by a dye washout. Prolonged exposure of the nerve to a dye solution also indicated that penetration of the dye along the nerve outside the inner limits of the chamber was less than 0·5 mm. The cylindrical chamber (diameter, 20 mm) shown in Fig. 1 was used for all experiments except those in which short lengths of nerve were exposed to procaine (Fig. 7A). Chambers were filled to a depth of about 10 mm.

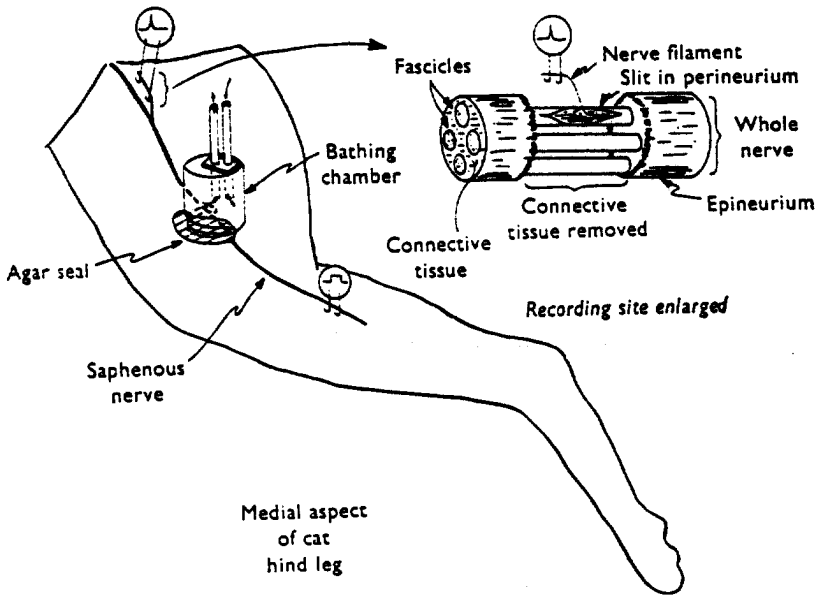

Fig. 1. Diagram of experimental arrangement and enlarged diagram of recording site. The nerve segment passing through the bathing chamber was cleaned of connective tissue as shown in the enlargement, but the perineurium was not slit. Cat's foot and bathing chamber were clamped rigidly to prevent movement.

The procedure for dissecting filaments for recording is illustrated in Fig. 1. After dissecting connective tissue away from the fascicles, a longitudinal slit was made in the perineurium of one fascicle to expose the enclosed axons. A fine filament of axons was then dissected away from the rest and placed on the recording electrodes.

The number of active fibres in a filament was determined by stimulating at frequencies of 10–20/sec while raising the intensity so slowly that unit action potentials were added one at a time (see Brown & Iggo, 1967, Fig. 1, for an example).

If the number of active fibres was too great to distinguish individual units, usually greater than 15, the number was further reduced by splitting the filament. Obtaining filaments with a satisfactory distribution of myelinated axons was not difficult. However, selection of strands which also contained a satisfactory number of non-myelinated axons was more difficult; often there was none or there were too many. During experimental trials the nerve was stimulated once every 4 or 5 sec at supramaximal intensity and responses were displayed on an oscilloscope (Tektronix 502A or 565) and photographed. A lighted timer (Reflexor, Bioelectric) projected on to the oscilloscope screen provided an accurate record of elapsed time (sec) of procaine exposure on each photographic record. Measurements of conduction velocities and blocking times were made from enlarged projections of the film.

The identification of myelinated and non-myelinated axons was based on measurements of conduction velocity in the absence of procaine at 37° C, and 2·5 m/sec was chosen as the dividing line (Gasser, 1950; Iggo, 1958). Figs. 5 and 6 illustrate typical records of strands containing both myelinated and non-myelinated axons. The potentials generated by myelinated axons were clearly separated from those of non-myelinated axons. Calculations showed that the slowest myelinated axons examined had conduction velocities greater than 5 m/sec whereas non-myelinated axons conducted at less than 1·2 m/sec. Unit potentials too small to be clearly distinguished from 'noise' were not included in measurements; this restriction was especially important for the non-myelinated axons whose potentials were usually small. Axons were considered blocked at the time when their unit potentials first disappeared permanently from the trace.

RESULTS

Conduction velocity

The velocities at which impulses were conducted in individual axons through the section of nerve bathed in procaine slowed progressively before complete block. This slowing caused a dispersion (i.e. a differential increase in latency) of the unit potentials recorded from individual axons in any strand so that each could be readily identified and followed until its sudden disappearance from the trace. Calculations of average conduction velocity through the chamber indicated that minimal conduction velocities just prior to complete block varied over a wide range. Some of the largest myelinated axons reached 10% of their normal conduction velocity before block whereas some of the small myelinated axons were blocked after slowing to only 70–80% of normal. Fig. 2 shows that minimal conduction velocities were clearly related to fibre size. The largest axons reached much lower average conduction velocities than did the smallest axons before their conduction was blocked. Values for intermediate-sized axons formed a continuum between the two extremes.

In addition to the reductions in conduction velocity prior to block, the maximum frequency of impulses that axons could conduct was also decreased, impairment at any time being more severe in small than in large axons.

Differential block of myelinated axons

The technique of recording from fine nerve filaments permitted the simultaneous determination of blocking times for up to fifteen individual axons of different sizes. The series of potentials in Fig. 3A was recorded from a filament containing four alpha and three delta fibres and illustrate the type of data obtained in these experiments. The first trace shows a control response and the other traces are responses at various times after exposure to 0·5% procaine solution. The small late hump on the delta

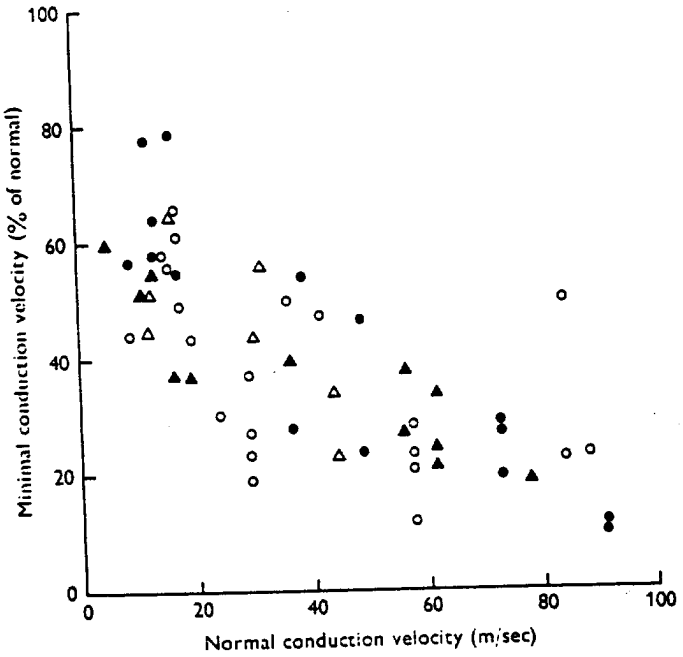

Fig. 2. Relationship between normal conduction velocities of fifty-five myelinated axons at 37° C and minimal, proportional conduction velocities recorded just before block by procaine HCl, 0·5%. Minimal values were calculated on the basis of average slowing in the 20 mm nerve segment contained within the bathing chamber. Different symbols represent four different cats from which inclusive data were obtained from seven filaments.

peak suddenly disappeared at 53 sec, indicating block of that axon. The second delta fibre was blocked 10 sec later, and the remaining delta fibre was blocked at 74 sec. Note the increases in latency compared to the control record. By 112 sec conduction in the slowest alpha fibre was noticeably slowed and it was blocked within 5 sec. The sudden reduction in peak height between 170 and 175 sec indicates that a second alpha fibre was

blocked. Block of the remaining two alpha fibres is shown in the last two records. In Fig. 3*B* the conduction velocities of these seven axons are plotted according to their respective blocking times, and the order of block according to fibre size is clear.

Fig. 4 shows the blocking times of 174 myelinated axons according to their conduction velocities determined as shown in Fig. 3. Some of the

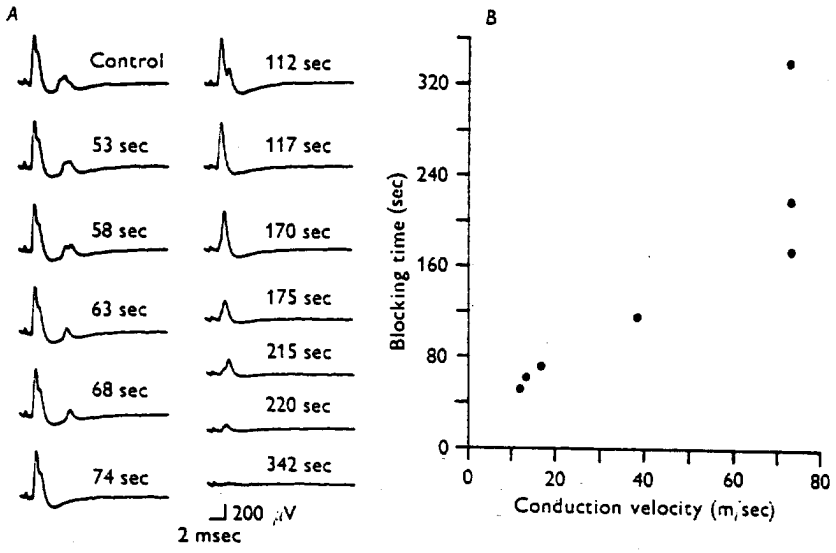

Fig. 3. Differential rate of blocking of myelinated axons by 0·5% procaine HCl. Sequential records during block of four alpha and three delta fibres are shown in *A*. Time (sec) after start of exposure to procaine is indicated above each trace; initial small deflexions indicate stimulus artifacts. Conduction distance, 73 mm. Blocking times of the axons in *A* are plotted in *B* according to their conduction velocities.

variability seen here was probably due to differences in time for diffusion of the drug to the axons. Axons buried deep in a fascicle are less accessible to the drug than those lying close to the surface (de Jong, 1970) and would, therefore, require more time for block. Such differences in accessibility account for observations that the order of block in some filaments was not strictly according to fibre size, a few larger axons being blocked before all the smaller axons. Likewise, axons of the same size in any filament were seldom blocked at the same time (Fig. 3). However, the order of block in several filaments in which blocking tests were repeated after complete recovery was the same. Another source of variability resulted from pooling the data from different preparations, since the times required for complete block among different preparations were not strictly uniform, presumably

due to variable amounts of remaining epineurium that impedes penetration of the drug. Despite these sources of variability, the tendency for small axons to be blocked by procaine before large axons is readily apparent.

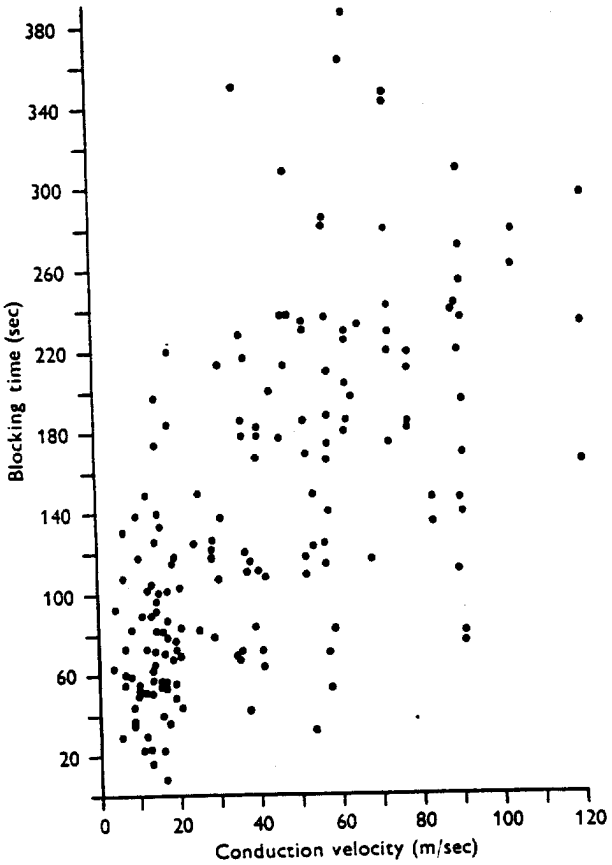

Fig. 4. Blocking times of myelinated axons exposed to 0·5 % procaine HCl. The individual blocking times of 174 myelinated axons in twenty-two filaments from four cats are plotted according to their normal conduction velocities. Exposure length, 20 mm.

The order of recovery upon washing was virtually the reverse of the order of blocking. Those axons that were blocked last recovered first and those blocked first recovered last. Complete recovery of all axons to their control latency after the start of intermittent washout usually required 30–45 min.

Myelinated vs. non-myelinated axons

Comparisons of blocking times between myelinated and non-myelinated axons exposed to 0·2% procaine solution were made in five experiments. In general, non-myelinated and small myelinated axons were blocked at about the same time, but in two experiments some non-myelinated axons were still conducting after all the delta and some alpha fibres had been blocked. In Fig. 5*A* in which seven myelinated and six non-myelinated axons recorded from the same filament are compared, the latter persisted until the three delta and two of the four alpha fibres were blocked; two of the non-myelinated axons survived blockade of all but one alpha fibre (70 sec) but were blocked at 74 sec.

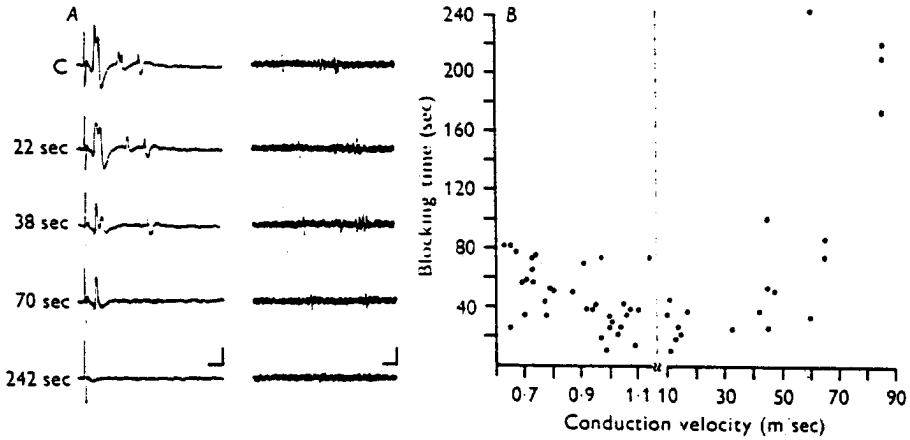

Fig. 5. Differential rate of blocking of myelinated and non-myelinated axons by 0·2% procaine HCl. *A* shows sequential block of axons in a nerve filament containing seven myelinated axons (left, normal conduction velocities, 11·3–60 m/sec) and six distinguishable non-myelinated axons (right, normal conduction velocities, 0·92–1·14 m/sec). Right trace starts 40 msec after end of left trace. Time (sec) after introduction of procaine is indicated on the left. Conduction distance, 80 mm; exposure length, 20 mm. Calibrations: left, 2 msec, 50 μV; right, 5 msec, 25 μV. Graph in *B* shows the blocking times for twenty myelinated and thirty-four non-myelinated axons according to their normal conduction velocities. These data were obtained in two preparations from five filaments containing both types of axon.

The blocking times of myelinated and non-myelinated axons determined from five filaments containing both are compared in Fig. 5*B*. These experiments indicated that non-myelinated axons were no more susceptible to block than were the delta and some of the smaller alpha fibres. Again, the differential rate of blocking among myelinated axons according to size was quite evident.

Threshold concentrations of procaine

In contrast to the observations of Nathan & Sears on spinal roots (1961), we could not find a concentration of procaine that would selectively block only the smaller myelinated axons. Solutions containing 0·015% procaine

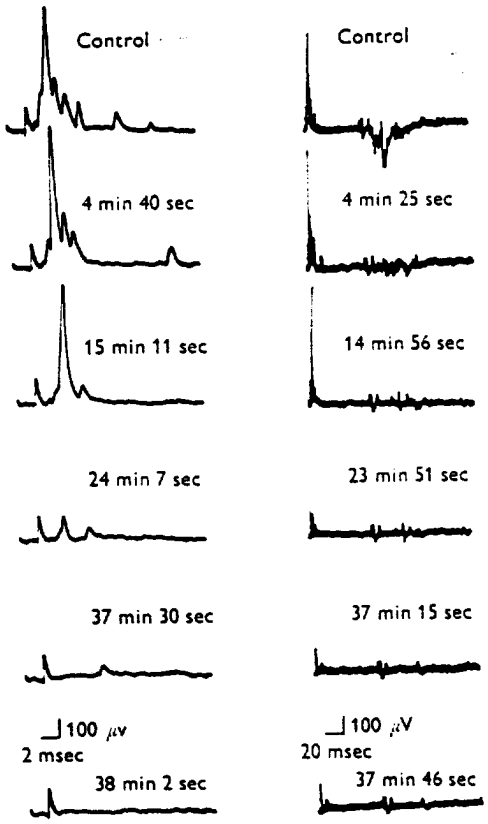

Fig. 6. Sequential block of myelinated (left) and non-myelinated (right) axons by a minimum blocking concentration of procaine HCl (0·02%). Time in minutes and seconds is indicated above each trace. The oscilloscope time base was alternated between sweeps (generated at 15 sec intervals) to record the detail of both groups of axons. Bottom traces show the survival of four non-myelinated axons after all myelinated axons were blocked. Conduction distance, 68 mm; exposure length, 20 mm; initial deflexions indicate stimulus artifacts.

did not block most myelinated axons within one hour whereas 0·02% solutions blocked all of them in the same order as seen with higher concentrations, but much longer exposure times were required. However, during experiments with low concentrations of procaine it was found that

some non-myelinated axons continued to conduct after all the myelinated axons were blocked. The series of traces in Fig. 6 is taken from an experiment in which the nerve was exposed to a 0·02% solution of procaine. A variety of nine myelinated axons and a large, undetermined number of non-myelinated axons were present in this filament. By 15 min many of both types of axon had been blocked. By 24 min more non-myelinated than myelinated axons were still conducting. The last trace shows the persistence of four non-myelinated axons after the remaining myelinated axons were blocked.

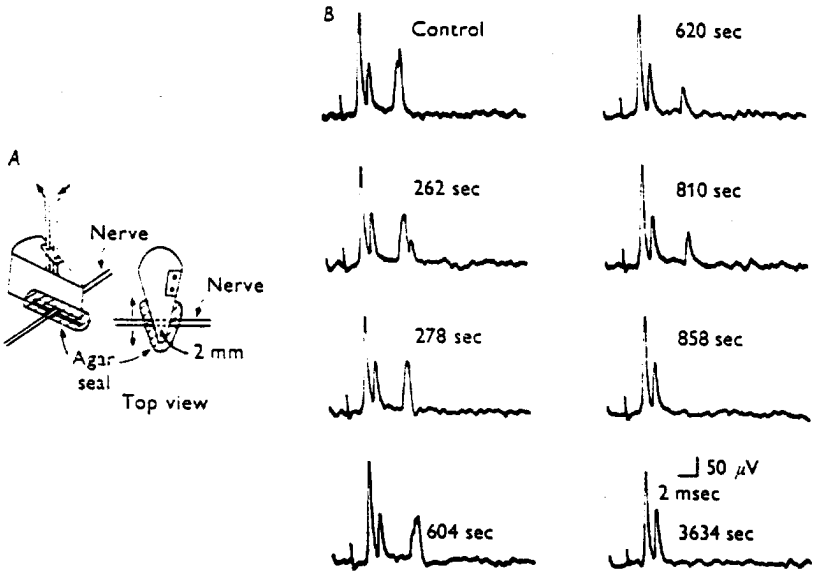

Fig. 7. *A*, diagrams of modified, wedge-shaped bathing chamber with which it was possible to arrange the exposure length between 1 and 10 mm.

B, absolute differential block of myelinated axons by 0·2% procaine HCl. Sequential records show progressive block of four delta fibres by 858 sec; three alpha fibres (two in first peak, one in second peak) were not blocked after 1 hr. Conduction distance, 85 mm; exposure length, 2 mm as shown in *A*.

While testing sub-blocking concentrations of procaine it was noted that block could be induced if the temperature of the procaine solution in the chamber was lowered by 5–10° C. Chamber solutions could be rapidly cooled to this extent by leaving the suction on after the inflow had been shut off. This practice was avoided in other testing, but it demonstrated the additive effect of local anaesthetic and cooling on nerve conduction.

Blocking short lengths of nerve

Since an absolute differential block between large and small myelinated fibres could not be achieved by varying the concentration of procaine, the possibility that differences in critical length of axons exposed to blocking concentrations of procaine might explain the order of block according to fibre size was tested.

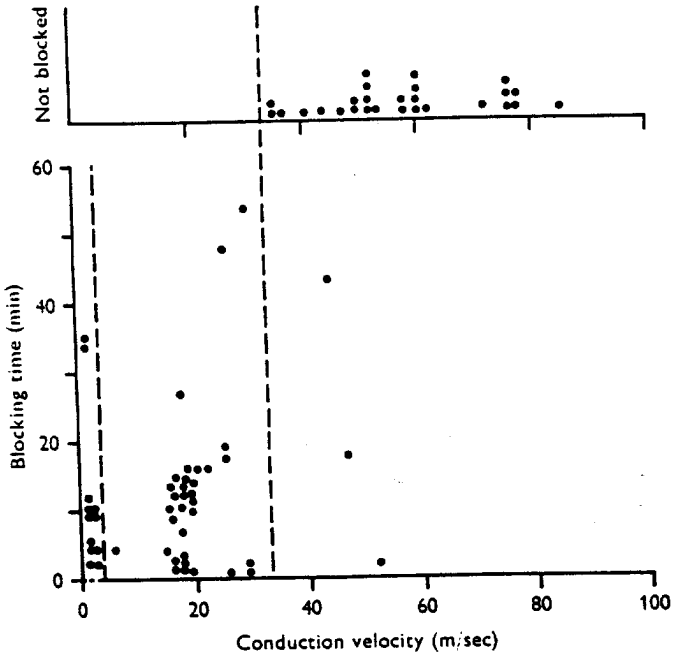

Fig. 8. Differential block of myelinated axons during exposure of 2 mm of saphenous nerve to 0·1 or 0·2 % procaine HCl. Axons that were not blocked within 60 min are compared with those that were blocked. The data include thirty alpha (> 35 m/sec) and thirty-three delta (< 30 m/sec) axons recorded from eight filaments in three cats; blocking times of twelve non-myelinated axons from two of the filaments in one cat are also plotted at the far left. No systematic differences between the 0·1 and 0·2 % procaine solutions were found in these experiments.

The length of nerve exposed to procaine was varied between 2 and 10 mm in different experiments by using a modified bathing chamber as shown in Fig. 7*A*. When 4 mm or more of nerve was exposed, it was not possible to produce an absolute differential block of small myelinated axons. In fact, all axons were blocked within 12 min when 4 mm of nerve was exposed to a 0·2 % solution. However, when only 2 mm of nerve was

exposed to a 0·1 or 0·2 % solution of procaine for more than 1 hr, all of the delta fibres could be blocked without blocking the larger myelinated fibres. The series of potentials in Fig. 7*B* from one such experiment show the successive block of conduction in four delta fibres within 858 sec. The last trace shows that the three alpha fibres were still functional after 1 hr. Results of eight experiments (including those in Fig. 7*B*) in which only 2 mm of nerve was exposed to procaine are shown in Fig. 8. In these experiments, only filaments containing clearly separated alpha and delta populations of axons were tested in order to avoid axons with intermediate conduction velocities that might have partially obscured the absolute difference between the two populations. Despite this precaution three of the smaller alpha axons were also blocked within the one-hour time limit (see Discussion). Non-myelinated axons were blocked at about the same time as the delta fibres in these experiments.

DISCUSSION

The technique of recording from small bundles of axons provides a distinct improvement over the conventional method of recording from nerve trunks. By monitoring conduction in a small number of individual axons, their blocking times can be accurately determined regardless of prior slowing. Measuring the reduction in peak heights of compound action potentials does not allow for this slowing of conduction and lacks sensitivity for accurate determination of conduction failure.

In general, however, our results agree with previous conclusions that smaller myelinated axons are blocked by local anaesthetics before large axons. The non-myelinated axons were about equally as sensitive to local anaesthetic block as were the small myelinated axons except when threshold concentrations of procaine were used. As noted previously by Nathan & Sears (1961), some non-myelinated axons were more resistant to block by procaine than were the smaller myelinated axons. In the present experiments, some non-myelinated axons were even more resistant than large myelinated axons when exposed to threshold concentrations (Fig. 6). The greater resistance of some non-myelinated axons to block by low concentrations of procaine or by sodium deficiency was attributed by Nathan & Sears (1961, 1962) to the difference in mode of conduction for myelinated and non-myelinated fibres. In non-myelinated fibres only the adjacent area along an axon must be activated for propagation of action potentials whereas in myelinated axons the action currents generated at one node must be sufficient to depolarize the next node to threshold. These comparable results suggest that the safety factor for conduction may be somewhat greater in non-myelinated than in myelinated axons. A similar

explanation was tendered to account for the greater resistance of non-myelinated axons to block by low temperatures (Paintal, 1967; Franz & Iggo, 1968).

Despite repeated attempts to produce an absolute differential block of small myelinated axons by varying the concentration of procaine, no such concentration was found: either all axons were blocked or few were blocked within a 0·005% difference in concentration. Our experiments indicate that 0·02% procaine is just above threshold for blocking all the saphenous myelinated fibres within 1 hr.

An absolute differential block between alpha and delta fibres was obtained only in experiments in which the length of nerve exposed to procaine was limited to 2 mm. This observation is readily explained by differences in their respective internodal lengths. The estimated internodal length of the delta fibres tested in these experiments varies between 0·3 and 0·7 mm (12–30 m/sec): that for the alpha fibres varies between 0·8 and 1·4 mm (35–85 m/sec) (Hursh, 1939; Coppin & Jack, 1972). If three successive nodes must be inactivated to block conduction completely (Tasaki, 1953), it follows that a 2 mm exposure would be sufficient to block three or more nodes of delta fibres, but the alpha fibres would escape total block because only one or two nodes would usually be exposed to the anaesthetic. However, it is possible for three successive nodes of axons with internodal lengths of 0·8–1·0 mm to be contained within the 2 mm, which may account for the occasional block of small alpha fibres that occurred in these experiments (Fig. 8). The necessity for inactivating a sufficient number of nodes to insure block of the largest myelinated fibres was appreciated by previous investigators in exposing 8 mm or more of nerve to the local anaesthetic (Gasser & Erlanger, 1929; Heinbecker *et al.* 1934; Matthews & Rushworth, 1957; Nathan & Sears, 1961; de Jong, 1970).

The foregoing considerations provide a tenable explanation for the sequence of block from small to large myelinated axons, that is, the differential *rate* of blocking by local anaesthetics. The importance of exposure length for conduction block suggests that a minimal blocking concentration would reach the shorter *critical length* of small axons before reaching the longer *critical length* of large axons, and the small axons would be blocked first.

Although the perineurium and perineural epithelium are known to present the main barriers to penetration of local anaesthetics and other substances into nerve fascicles (Shantha & Bourne, 1968; de Jong, 1970), discrete information regarding intimate diffusion around individual axons within fascicles is not available. In view of the tight packing of axons, Schwann cells and myelin, and endoneural connective tissue within nerve fascicles, the extracellular space available for rapid and uniform distribu-

tion of foreign substances is necessarily limited (Sunderland, 1965). Indeed, brief exposure to radioactively labelled, extracellular fluid indicators has demonstrated sparse and uneven labelling around densely packed myelinated nerve fibres compared to adjacent connective tissue (Brown, Stumpf & Roth, 1969). An irregular distribution of extracellular markers throughout the endoneurium was also evident in immature mouse nerve exposed to protein tracers for 24 hr (Kristensson, 1970; Kristensson & Olson, 1971). Therefore, a uniform distribution of local anaesthetic molecules around all axons within the early minutes of exposure to even suprathreshold concentrations would appear unlikely.

Direct evidence for an irregular distribution of intrafascicular procaine was furnished in our experiments with the 20 mm chamber by comparing the degree of average conduction slowing in individual axons just prior to block. In contrast to the nearly uniform slowing of conduction velocity in myelinated axons during exposure to cold in similar experiments (Paintal, 1965; Franz & Iggo, 1968), the degree of slowing among axons exposed to procaine was not uniform (Fig. 2). In the cold block studies of Franz & Iggo, minimum conduction velocities of a sample of thirteen myelinated axons just prior to block ranged between 1·4 and 4·6% of control values (mean, 2·5%). However, just before block by procaine, minimum values from a representative sample of fifty-five axons varied between 10 and 80%. Inasmuch as the underlying mechanisms responsible for conduction slowing and ultimate block both by cold and by local anaesthetic are probably very similar, the much higher average conduction velocities and their wide variability just before block by procaine must reflect large regional differences in the distribution and concentration of the anaesthetic within an exposed nerve fascicle. This conclusion is strongly supported by the data presented in Fig. 2 which shows a definite tendency for small axons to be blocked at relative conduction velocities that were much higher than those for large axons. Since these data are based on *average* conduction velocities along 20 mm of exposed nerve, the systematic variation according to axon size indicates that the anaesthetic was not uniformly distributed along the total length of each axon, especially the small ones, at the time of block. The tendency for larger axons to reach lower average conduction velocities before blocking reflects the longer time required for effective concentrations of local anaesthetic to reach the greater proportion of their total length necessary for block. These considerations are presented diagrammatically in Fig. 9.

Our observations that the order of axon recovery upon removal of the anaesthetic was virtually the reverse of the original order of blocking are consistent with clinical experience and other experimental reports. These observations may be explained by the likelihood that anaesthetics would

recede primarily along the same intrafascicular routes by which they were distributed upon entry (Fig. 9*D*). Alternatively since those axons blocked early are exposed to higher concentrations of the anaesthetic for a longer period of time, more time may be required to reduce the concentration of strongly bound anaesthetic molecules to subthreshold levels. Theories of differential block based on surface to volume relationships (de Jong, 1970) have been challenged for their failure to account for the order of recovery observed (Ritchie, Cohen & Dripps, 1970). The present proposal for a mechanism of differential block satisfactorily accounts for the sequence of both blocking and recovery.

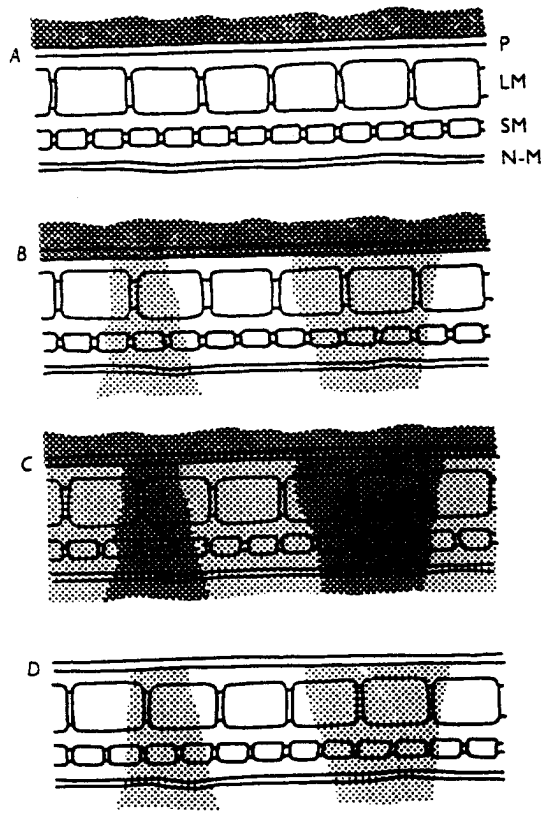

Fig. 9. Diagrammatic representation of proposed mechanism for differential rate of block by local anaesthetics. Section of peripheral nerve shows perineurium (P), large (LM) and small (SM) myelinated axons, and a non-myelinated axon (N-M). Intensity of shading designates anaesthetic concentrations as threshold or suprathreshold. *A*, initial exposure; *B*, differential block; *C*, total block; and *D*, partial recovery. Conduction block is assumed to occur only when three adjacent nodes of an axon are exposed to blocking concentrations of the local anaesthetic.

Our failure to produce absolute differential block between large and small myelinated axons by adjusting the concentration of procaine differs from the results of Nathan & Sears (1961). However, their experiments were performed on spinal roots where the barriers to diffusion of the anaesthetic throughout the nerve are much less than in peripheral nerve with intact perineurium, and uniform distribution of the anaesthetic in spinal roots would be achieved more quickly. This fundamental difference is reflected by the longer blocking times encountered in the present study and may account for our inability to demonstrate a relationship between fibre size and minimal anaesthetic concentration requirements. Nevertheless, our results on the differential *rate* of block are in full agreement with those of Nathan & Sears.

Based on their ability to produce absolute differential blocking of small myelinated fibres in spinal roots by both procaine and sodium-deficient solutions, Nathan & Sears (1961, 1962) proposed the theory that differential blocking must indicate differences in safety factor for conduction between large and small diameter fibres. However, the evidence for differences in safety factor among myelinated axons is not supported by recent findings on cold block of peripheral nerve. Although earlier studies had reported that small peripheral fibres were more readily blocked by cold than large fibres (Lundberg, 1948; Douglas & Malcolm, 1955), more recent studies, in which the present technique of recording unit action potentials was employed, demonstrated that all myelinated axons were blocked at about the same temperature regardless of size (Paintal, 1965; Franz & Iggo, 1968). Since cold block is not restricted by diffusion barriers but occurs by ionic mechanisms that are probably similar to anaesthetic block, the latter cold block experiments indicate that the safety factor for conduction in myelinated axons of all sizes is essentially the same. Our failure to demonstrate absolute differential blocking of peripheral nerve by a one-hour exposure to low concentrations of procaine is consistent with this conclusion. Therefore, we have proposed an alternate theory of differential blockade by local anaesthetics (above) based on differences in the *critical length* of axons of different sizes that must be exposed to blocking concentrations. The possibility that differential dispersion of individual action potentials from small myelinated axons, which is evident in our Figures, could have influenced the observations of Nathan & Sears (1961, 1962) on differential blocking of spinal roots by critical concentrations of procaine or sodium cannot be resolved by the present study.

In addition to differences in onset of complete block among axons, Wedensky or transition block undoubtedly contributes greatly to differential effects observed clinically. Before complete block, the maximum frequency of impulses that axons can carry is progressively reduced

according to their normal conduction velocity. Since many sensory receptors respond to mild or intense stimulation by generating high frequencies of impulses, considerable sensory impairment would be anticipated in the absence of complete block. Such considerations may also partially account for differential block between sensory and motor functions. Although, size for size, sensory axons are apparently no more sensitive to local anaesthetics than are motor axons (Matthews & Rushworth, 1957; D. N. Franz & L. Mabey, unpublished), motor fibres normally function at frequencies well below 30 Hz (Freyschuss & Knutsson, 1971; Tan, 1971) whereas many sensory fibres normally carry much higher frequencies. Consequently, low frequency motor function could persist after higher frequency sensory function has been severely impaired by transition block.

An additional and probably more important factor accounting for differential block of sensory and motor fibres relates directly to their relative sizes. Except for the large Type I proprioceptive afferent fibres, most other afferent fibres are smaller than skeletal motor fibres (Boyd & Davey, 1968) and would therefore be blocked first and recover last.

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