

## PROPERTIES OF DORSAL ROOT UNMEDULLATED FIBERS ON THE TWO SIDES OF THE GANGLION

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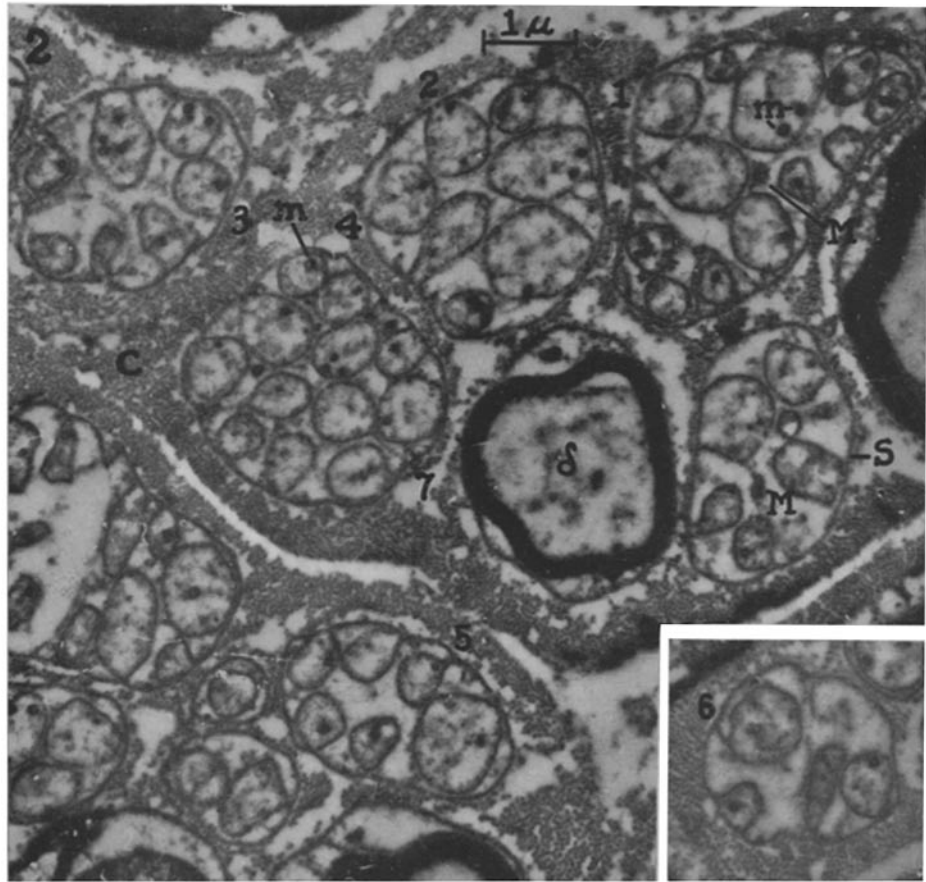
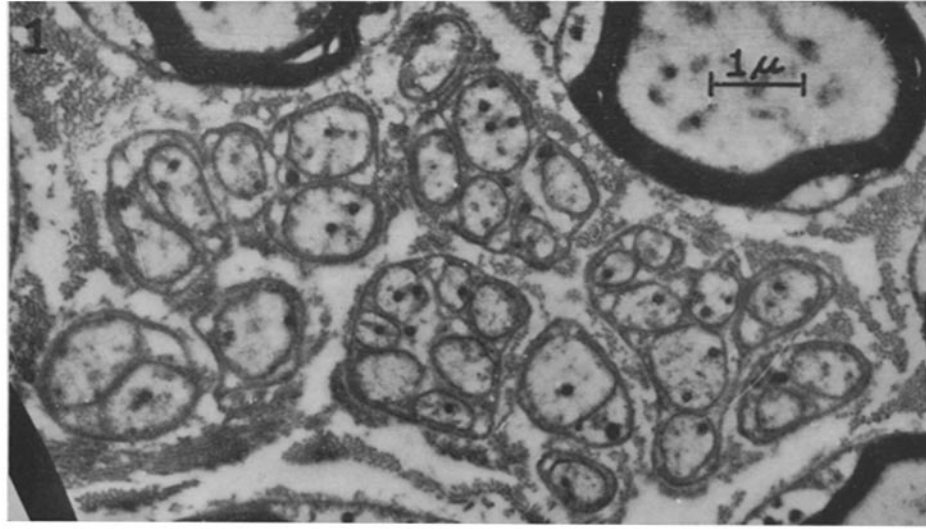
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Among the experiments preliminary to a paper by the author (1950) on conduction in unmedullated fibers arising in dorsal root ganglia, an observation (unpublished) was made, prompted by a figure in a paper by Ranson and Davenport. The figure was a reproduction of a microphotograph of a silver-stained section of a dorsal root ganglion, in which were to be seen some *T*-divisions of the axons. It showed that the centrally directed branch is smaller than the peripherally directed branch, a situation suggesting that conduction would be slower in the root than in the nerve. A few experiments sufficed to establish the validity of the inference; and thereby conduction in the unmedullated fibers in the roots was placed in contrast with that of the medullated fibers. Over fifty years ago Dale found essential correspondence between the diameters of the medullated fibers on the two sides of the ganglion. And, in accord with what now can be predicted from his measurements, the impulses in the medullated fibers course through the roots on their nerve schedule. For the unmedullated fibers, not only was there no known structure with which the velocity could be correlated, there was not even available an adequate technique whereby the structure could be revealed. Consequently the investigation came to a standstill. With the advent of electromicroscopic techniques the impasse came to an end. How the block was removed and the findings about conduction in the roots ensuing upon the removal are described in this paper.

In the paper previously mentioned conduction in the unmedullated fibers in skin nerves was correlated with the sizes of the fibers as measured with light photography of silver-stained preparations. Although every precaution was taken to avoid the errors inherent in an attempt to measure with green light structures with dimensions 2 to  $<1$  times its wave length, when the possibility arose of making the measurements employing the electron wave length, a method so greatly superior presented itself that it was decided to repeat the analysis.

### *Electron Microscope Picture of Unmedullated Fibers*

The silver stain, which has been of great service in the derivation of information about unmedullated fibers, characterized the method of choice at the time



FIGS. 1-2  
710

of departure. It was soon found, however, that the stain was nothing more than a precipitation of silver granules about the fibers (Fig. 3). Furthermore, the fixation method belonging to the technique was not conducive to the production of satisfactory resolution of cytological structures. Hence new methods had to be sought.

*Methods.*—Trials of various fixatives left no doubt about the superiority of osmic acid, a fixative that had already won a position in electron microscopy. How the osmic acid was to be used then remained to be determined. It was noticed that when the fibers were fixed in a buffer at pH 7.4 the pictures did not agree with ones obtained when osmic acid vapor was used. The fibers appeared to be considerably smaller when they were fixed in the buffer than they did when fixed in the vapor. Only when the buffering was adjusted to lower pH values could the dimensions in the vapor be duplicated. Therefrom, the obvious suggestion was that the pH of the fixative was of importance. In the absence of information about the pH of the fibers, the way out of the difficulty appeared to be to devise a method that would subject the fibers to a minimum of alteration. In the technique of handling nerves in oscillograph experiments Krebs's solution equilibrated with 5 per cent CO<sub>2</sub> in oxygen is employed. By first immersing the dissected nerves in the above solution then rapidly transferring them to a like solution containing 1 per cent of osmic acid (which is a neutral substance) the smallest possible shift was to be expected. The procedure, when tried, gave so successful a result that it was adopted as a routine.

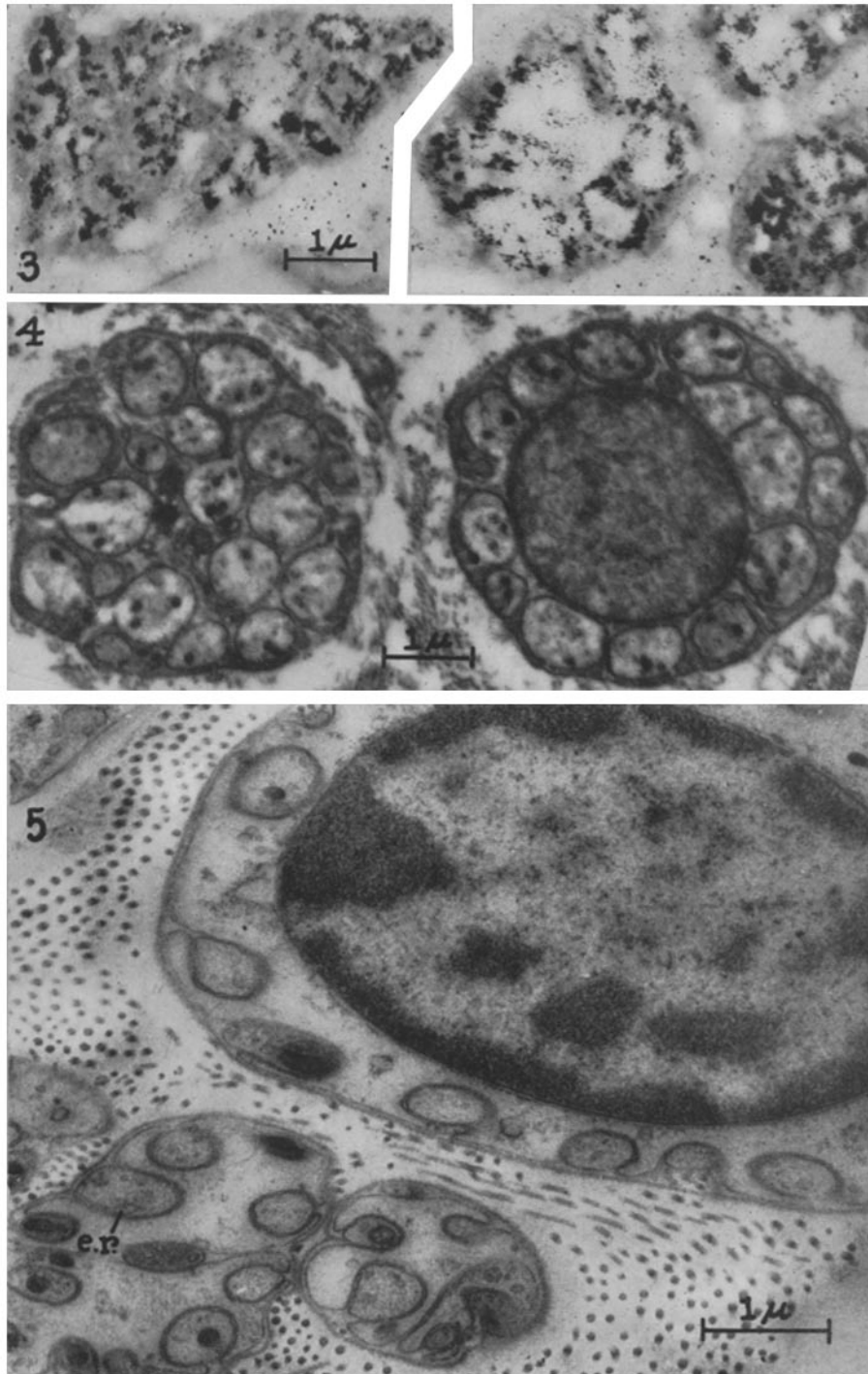
Standard methods for embedding the fixed material in a polymer were employed. The plastic was formed from butylmethacrylate alone, or from the latter combined with 10 or 20 per cent of methylmethacrylate, when greater hardness was desired. Routinely sections were cut at a thickness of 0.1  $\mu$ . But for special purposes both thinner and thicker sections were made. Control of magnification was effected through accompanying pictures of latex particles.

*Description of Fibers and Sheaths.*—In order to understand the pictures shown in the figures familiarity with Nageotte's description of the sheath system of unmyelinated fibers is essential. Characterizing the system is a sequence of tubes of various sizes made up of a syncytium formed by the Schwann cells. These tubes branch and reunite; and at intervals where the tubes are largest the nuclei appear (Fig. 4). Confirmation and extension of Nageotte's description, which has never received the attention that it merits, will be presented in a later section. Figs. 1, 2, and 4 show at 12,000 diameters the appearance of the Schwann tubes in cross-section, and the disposition of the fibers within them.

Only in the smallest tubes does a fiber have a sheath to itself. As the tubes

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FIGS. 1 and 2. Electron microscope pictures of groups of C fibers in the sural nerve of the cat, reproduced at 12,000 diameters. In Fig. 2: *m*, mitochondria in axons; *M*, mitochondria in Schwann axoplasm; *C*, collagen fibers;  $\delta$ , a delta fiber. For the significance of the numbers see text.



FIGS. 3-5  
712

become larger the number of fibers increases to an undetermined upper limit of about fifteen. On the average the sheaths of s.C fibers in visceral nerves contain more fibers than the sheaths of their d.r.C counterparts in skin nerves (Fig. 4). Otherwise the sheathing of the two subdivisions of the C fibers is similar; and the difference between their physiological properties acquires no explanation from their morphology.

In cross-sections of the skin nerves the unmyelinated fibers are always found in groups in association with delta fibers, their closest functional allies. About their sheaths are to be seen the collagen fibers which occupy the space between them (Fig. 2). Each fiber is sharply delineated by an osmiophilic border, and is further placed in contrast with the relatively featureless Schwann cytoplasm about it by the greater density of its own cytoplasm. In the axoplasm there can be identified neurofilaments, endoplasmic reticulum, and mitochondria. Mitochondria can be resolved in all the figures presented in this paper made from material fixed in osmic acid. In cross-section they appear as tiny black rings. Their long axis is in parallel with that of the axons. Endoplasmic reticulum can be seen in Fig. 5.

More and more in the physiology of nerve fibers the significance of considering axons in relation to their sheath system is becoming apparent. In evidence, the inferences deducible from the sheath system of the unmyelinated fibers provide an illustration. The membrane sharply visible on the outside of the Schwann tubes may be taken as the starting point. To this membrane, as can be confirmed through longitudinal sections, the axons are in continuous attachment. On account of the analogy with the mesentery the attachments have been designated mesaxons. In high magnification pictures of very thin sections, at the border of the Schwann sheath two osmiophilic lines are re-

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FIG. 3. Electron microscope picture of C fibers in the saphenous nerve of the cat, stained with silver by the Ranson method. Note the silver precipitation about the fibers. To derive the better known light pictures with this stain, it should be borne in mind that in sections 10 to 20 times as thick (1 to 2  $\mu$ ) the granules about the fibers would form a continuous ring. Without special precautions in microphotography the rings are caused to appear as solid black dots as the result of light diffraction (Gasser, 1950).

FIG. 4. Two Schwann sheaths containing s.C fibers in the hypogastric nerve of the cat. Note the central position of the Schwann nucleus in the sheath at the right.

FIG. 5. Shows the fine structure of the Schwann membrane and its relation to the axons. Cross-sections of Schwann tubes found in the testis of the cat. As the tubes are not surrounded by a lamellated sheath the fibers are probably near their termination. Fixation, osmic acid, buffered to pH 7.5. Magnification,  $\times 17,660$ . The inner of the two black lines at the edge of the sheath is the Schwann plasma membrane. It turns inward to surround the axons. Inside the loops a second black line evinces the axon plasma membrane. *e.r.*, endoplasmic reticulum. (Courtesy of D. W. Fawcett.)

solvable. It is the lamina responsible for the inner of the two lines that turns inward and surrounds the axon. Through the variations in the nature of this envelopment the different forms of the mesaxons seen in the sections can most readily be made understandable.

Numerals placed on Fig. 2 point out a systematic sequence of mesaxon variants. At, 1, the beginning of the series, a considerable sector of the axon circumference is held in contact with the Schwann membrane. Through shortening of this sector, 2, 3, a stage is reached at which the axon meets the membrane only at a point, 4. The two infoldings from the Schwann membrane have now come together; and in the rest of the sequence they form ligaments of increasing lengths, 5, 6, until in the end they are long enough to permit a fiber to occupy a position near the axis of the tube, 7. Much longer mesaxons are to be found in the roots than in the nerves. Where the axons pass a nucleus they are still attached to the membrane, and are outside of the nucleus which occupies an axial position in the tube (Fig. 4).

From the foregoing description it follows that the mesaxon is characterized by two osmiophilic lines caused by the meeting of the two infoldings from the border of the Schwann tube, and their parallel course. After the two lines separate in the encirclement of the axon there are again two lines, the inner one supplied by the axon plasma membrane. The fine structure of the situation is exquisitely shown in a preparation obtained by D. W. Fawcett incidental to a study of the testis. It is reproduced in Fig. 5 through his courtesy. Fixation was in osmic acid buffered to pH 7.5. To which of the two subgroups of the C fibers the twiglets may belong is irrelevant, since, as previously stated, the two subgroups are similarly sheathed. All cytologists consulted agree in designating the outer line about the Schwann sheath as a basement membrane. Thus, the inner line must represent the plasma membrane of the Schwann cells. Through this interpretation, what had been a puzzling problem becomes non-existent. The problem was as to what the conduction mechanism might be if the axon were in the Schwann cell, that is surrounded by a medium in which potassium ions would presumably be predominant. The axon is *not* in the Schwann cell. It is *outside*. The outer surface of the axon plasma membrane is turned toward the outer surface of the Schwann plasma membrane, as infolded.

With the altered description of the C fiber relationships the problem of the biophysical mechanism of C fiber conduction has merely been restated; and it is far from solved. What is the content of the space between the basement membrane and the Schwann plasma membrane? The basement membrane, itself, may well be polarized. Witness the potential change observed by Ottoson *et al.* when a microelectrode is caused to pass through the basement membrane of the skin. Although the Schwann protoplasm may provide an external circuit for conduction, the narrowness of the space between the axon and the semipermeable membrane of the Schwann cell about it can hardly

fail to have an effect on the C potential cycle when it is considered in relation to the ionic movements across the axon membrane during activity, as now being elucidated by Hodgkin and his colleagues. Then, whatever may be the effect of the space relationships, the difference between the two subgroups of C must still be explained on some other basis.

Comment on the effective mechanical protection of the C axons by their sheaths, is hardly necessary. But two comments in comparison of the unmyelinated and myelinated sheaths may be brought forward. Pictures in possession of the author indicate the presence of a basement membrane about the myelinated fibers. And, in Geren's description of the embryology of the myelin, during the first stage before the coiling starts the picture is identical with that of the adult unmyelinated fiber (*vid.* her Fig. 2).

*Consideration of Interaction between the Fibers.*—Since the question is so often asked by those who see for the first time electron microscope pictures of cross-sections of C fibers, as to whether the fibers conduct independently, it was decided to see how good an answer could be obtained to the question. By independence is here meant absence of suprathreshold excitation, one fiber by another; for everyone who has examined the subject, either in nerves of invertebrates or in the medullated fibers of vertebrates, has found that some degree of interaction occurs, even in absence of abnormality beyond that incidental to conservative experimental conditions.<sup>1</sup> The latter is revealed through synchronization of impulses in adjacent fibers, if their conduction velocities in isolation are not far apart, or through small changes in the threshold values of testing shocks that take place in inactive fibers when impulses are passing in the neighborhood. And here the manifestations of interaction reach their limit. For interaction to rise to suprathreshold excitation requires such laboratory artifices as proximity to cut ends, chemical agents, or cathodal polarization, as shown by Hering and many others after him. In order to place C fibers in contrast with A fibers, it would be necessary to show that under normal conditions interaction could attain a suprathreshold magnitude.

At the outset it can be said that there is no positive evidence in favor of such a view. Temporal dispersion of impulses, which implies also spatial dispersion, takes place in the conventional manner. Examples of the compound C action potential at four distances of conduction are shown in Fig. 6. In order to avoid the leading errors introduced by long interpolar distances, constant position of the leads and movement of the stimulating cathode were chosen as constituting the better of two ways of performing the experiment. The records were mounted at even spacings corresponding to equal increments of

<sup>1</sup> *Crab*, Arvanitaki and Fessard, Katz and Schmitt. *Squid*, Arvanitaki. *Frog*, Blair and Erlanger, Marrazzi and Lorente de N6. *Toad*, Otani. Mammalian A fibers spontaneously firing, presumably on account of absence of CO<sub>2</sub>, were found by Adrian to become synchronized by interaction.

conduction distance, and through each one a line was drawn in extension of the starting base line. From numbered positions in the action potential described by Gasser (1950, Table I) dotted lines were drawn to show the temporal positions of the events on the base lines. In this instance they denote velocities of 1.8, 1.2, 1.0, 0.84, and 0.71 m./sec. If straight lines can be drawn through these positions it follows that the conduction, which has taken place, is con-

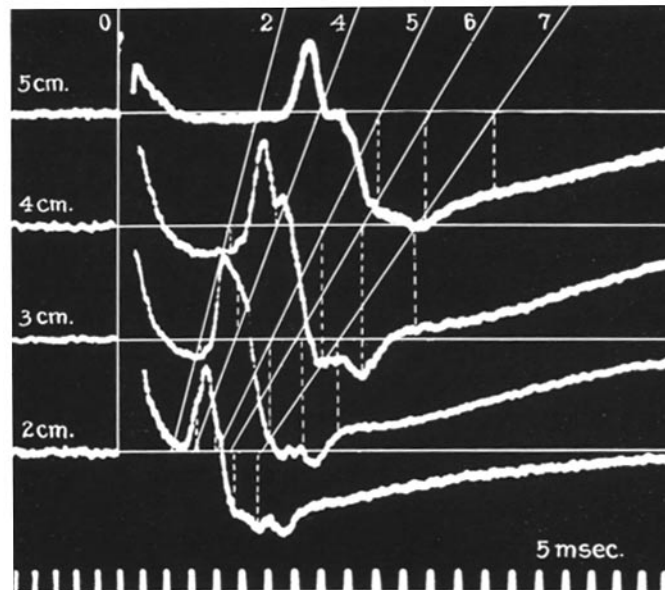


FIG. 6. Action potentials in a saphenous nerve of the cat as recorded at four conduction distances. Leading, d.c. differential; leads at constant separation, 24 mm., distal lead cocaineized. Variation in the conduction distances obtained by moving the stimulating cathode, kept at earth potential. In keeping with the decrease in height of the summed potentials as dispersion increases, the amplification is increased with the conduction distances. 37.5°C. For the significance of the added lines, see text.

sistent with independent linear velocities. That such straight lines can be drawn is shown in the figure.

In a system of fibers of slow conduction the impulses in the faster fibers outrun those in the slower ones within short distances. If any fibers were to stimulate other fibers it would be the faster outrunning fibers stimulating the slower ones ahead of the proper arrival time of their impulses and causing the later elevations to appear too early at the longer distances. There is no evidence for a forward displacement of this kind. But it must be admitted that the findings do not provide compelling proof of its absence. The action potential recording technique is not a sufficiently penetrating analytical device.



It will be noted that owing to the extremely rapid temporal dispersion the last elevation easily seen at 2 cm. has become quite invisible at 5 cm. Other evidence must be brought into the argument.

In assessing the effect of an active fiber upon an adjacent resting fiber one must draw upon such information as is available. Katz and Schmitt investigated two adjacent 30  $\mu$  fibers, held together only by sparse strands of connective tissue, in a preparation dissected from a limb nerve of the crab. Consultation of a microphotograph of a similar pair of fibers in a lobster nerve brought out the fact that the proximity of two such fibers would be considerably less than a fiber diameter. Reduce the scale of the preparation somewhat over thirty times and it becomes a reasonable model for adjacent unmyelinated fibers in a Schwann sheath. Katz and Schmitt found in the resting fiber of the pair that at the peak of the second phase of interaction, when the resting fiber was being maximally depolarized by a passing impulse in the active fiber, the threshold magnitude of a test shock still had 89 per cent of the resting value. The interaction was so low that a large allowance can be made in compensation for the possible effect of the difference between the sheaths of C fibers and crustacean fibers. (For the structure of the sheath of the lobster see Geren and Schmitt.)

The argument does not end, however, with the magnitude of the interaction. Consideration of the longitudinal relationships shows how premature is any inference from the appearance of cross-sections alone. For fibers to interact they must lie in parallel. Owing to the devious course a fiber must take through the numerous branches of the sheath network the relative positions of the individual fibers must be continually changing.

With spike durations estimated at 2 msec. (Gasser, 1950) the wave lengths of the spikes would fall in the range of 4.5 to 1 mm. If it be assumed that the regions of active inflow have 0.4 of these lengths, the depolarizing stages of the interactions would have lengths of 1.8 to 0.4 mm. In relation to these values the rate of branching of the Schwann sheaths assumes considerable interest. Owing to the absence of information on this point, it had to be obtained.

Trials with teasing methods and methods depending on light magnification turned out to be unrewarding, so it became necessary again to resort to the electron microscope. Longitudinal sections yielded some information; but it was so little satisfying that in the end it was decided to embark on the laborious but certain method of building up the sheath from cross-sections. At 0.25  $\mu$  thickness sections could be cut without danger of loss; and they were not too thick for recording when the microscope was operated at 100 kv. In a small funiculus at the surface of the saphenous nerve of the cat, where the fixation was good, a small group of sheaths was selected for examination through serial sections located about 34 levels in the block, the levels being determined by a log of the advancement of the microtome. Prints were made

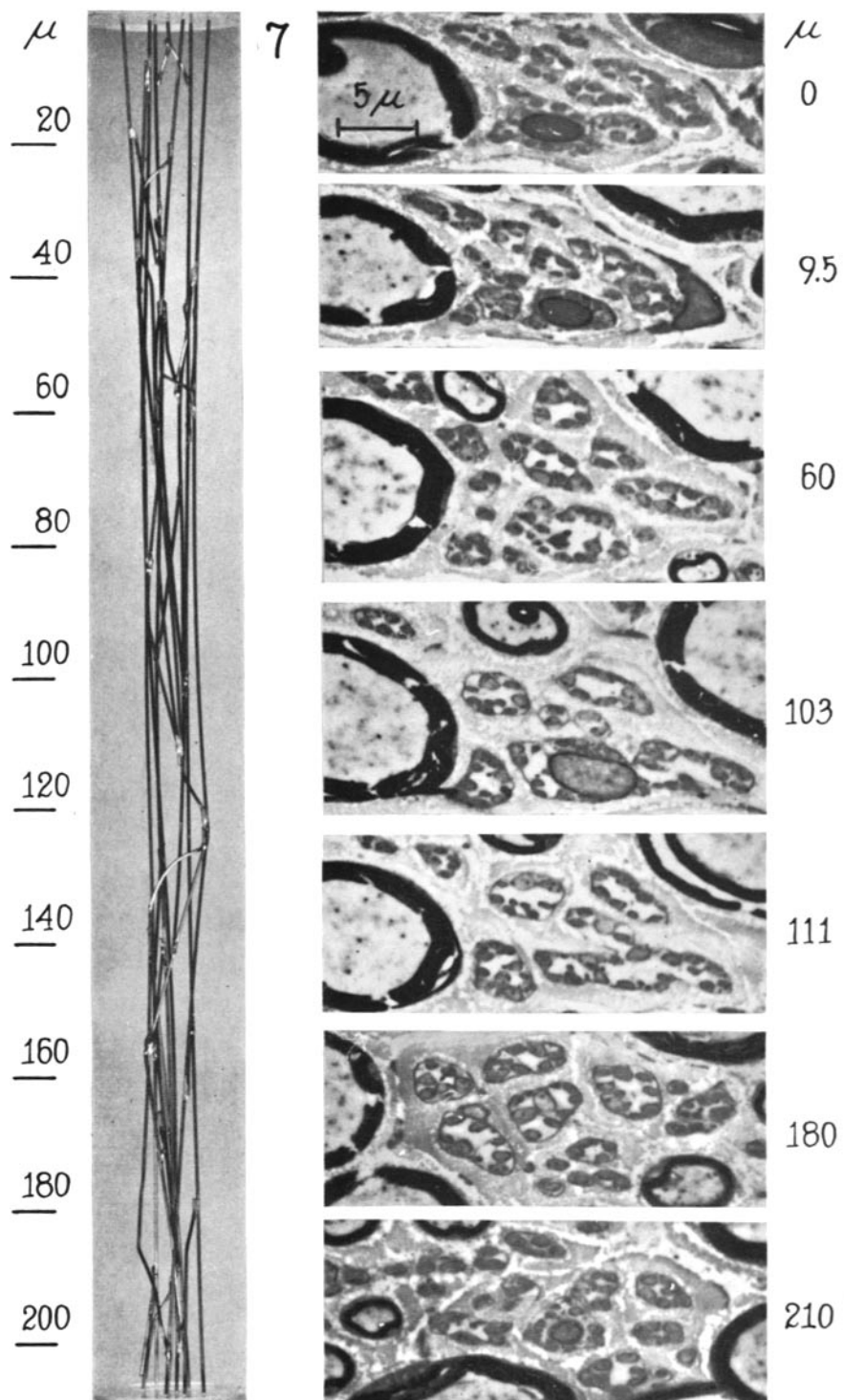


FIG. 7  
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at 3,000 diameters; and from these, in order to get the picture in three dimensions a model was constructed, 3,000 times the natural size, by soldering together pieces of No. 14 B. S. gauge iron wire. At the ends of the model the wires were fitted into a plastic block at positions in the centers of the sheaths as they appeared at the 0 and 210  $\mu$  levels. Between these levels the wires were separated in accord with the sections they represented. Thus, only the axes of the sheaths were shown. The sheath diameters varied between 1+ and 5+  $\mu$ ; and the model is somewhat simplified by omitting most of the branches that contained only one fiber.

A photograph of the model is shown in Fig. 7, together with some samples of the sections used in its construction. It shows the sheath system through which 54 fibers pass over a distance of 210  $\mu$ . With the aid of samples taken at several additional levels the composition of the bundle was followed to a length of 0.5 mm. It kept continuously changing. Between 0.2 and 0.5 mm. interchange began with neighboring groups, particularly the one below and to the right of the one illustrated in the figure. More fibers were gained than were lost (if any were lost); and at the end, at 0.5 mm. there were 79 fibers in the aggregate.

On the model, measurements were made of the lengths of the segments in which the fibers would have to remain in the same sheath. The distribution is shown in Fig. 8. Much to the surprise of the author it was found that the lengths had essentially been confirmed in advance by measurements made on the longitudinal sections. With the wave lengths of the second depolarization stages of interaction, 1.8 to 0.4 mm., there are now to be compared sheath segment lengths ranging from several to 63  $\mu$ . As the sample in the model appears to represent a repeating situation, it follows that, even if some longer segments have been missed and if the fibers do not change their relationships as often as opportunity offers, the lengths through which the fibers remain in parallel are still small in comparison with those of the second stages of interaction. With respect to the third stage of interaction (Katz and Schmitt) the d.r.C fibers would occupy a unique position among nerve fibers on account of the precipitous transition to the large postspike positivities. This stage of hyperpolarization of neighboring inactive fibers should be larger (and longer) than

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FIG. 7. Sheath system through which 54 d.r.C fibers pass in a distance of 210  $\mu$ . At the left is a picture of a model constructed to scale ( $\times 1,100$  in the reproduction). At the right are samples from the sections as they appeared at the levels indicated (reproduced at  $\times 2,160$ ). The model is photographed as though the bottoms of the prints were at the front, and one were looking at it somewhat from the right. To be noted: the divisions, at 9.5  $\mu$ , of the sheath about the nucleus at the lower left of 0  $\mu$ ; the large aggregation of fibers in one sheath in the region of 110  $\mu$ ; and, as between the 180 and 210  $\mu$  levels, the degree of division of the sheaths at the left of 180  $\mu$  and the regrouping of the fibers in the other sheaths.

its analogues. As far as there might be any effect at all during this stage, active d.r.C fibers would tend to depress the excitabilities of adjoining fibers.

No matter how it is examined, allegation of cross-excitation between d.r.C fibers receives little support. Indeed, one can with some assurance assume that they conform to the general rule of isolated conduction. As to mutual effects on velocities, those too must be small, and for the same reasons. If there be any effects during the multiple contacts of a fiber they would be both positive and negative, so that they would tend to cancel one another.

*Measurement of the Fibers.*—The sural nerve was used because it is a slender nerve with all its fibers contained within a single lamellated sheath. In the original plan for measurement of the fibers complete coverage of the nerve was set up as the objective; but in the end attainment of the objective was prevented by one of the features of the standard technique for mounting sections in the electron microscope. The sections are placed on grids; and, as a result, the fibers behind the bars of the grids are lost to view. In an attempt to compensate for the loss low power pictures of all the squares in a suitable

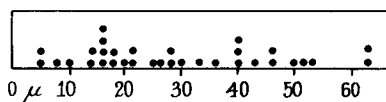


FIG. 8. Lengths of the sheath segments in the model shown in Fig. 7.

section were made, and supplemented by similar sets for nearby sections, with the idea that fibers obscured by the grids in one set might not be covered in another. The pictures from the master set were laid out on an enlarged grid scale, and the missing fibers were sought by matching fields from the supplementary sets. Largely owing to the changing appearance of the sheaths, which militated against identification of the fibers, the procedure was only partially successful. Thus, in the outcome it was estimated that only somewhat over one-half of the fibers in the nerve were accounted for. With the low power map as a key to the fibers, high magnification pictures of the fields identified in the key were then made so that the final prints on which the measurements were made were all at 12,000 diameters. Magnification in the electron microscope was controlled by latex particles.

As in all histological procedures the effect on the fiber dimensions of embedding in plastic for electron micrography had to be taken into the reckoning. Only an approximation of the effect was attempted. Adjacent pieces of sural nerve were embedded in butylmethacrylate polymer and in paraffin. Comparison of the diameters of the sections showed the diameter of the former to be 1.19 times that of the latter. That the comparison from the sections was valid for the fibers was verified by measuring a score of the largest fibers in

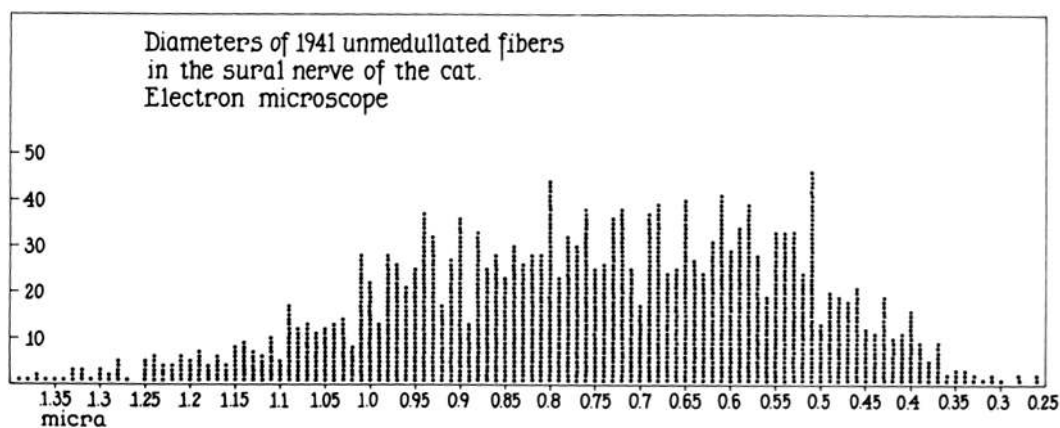


FIG. 9.

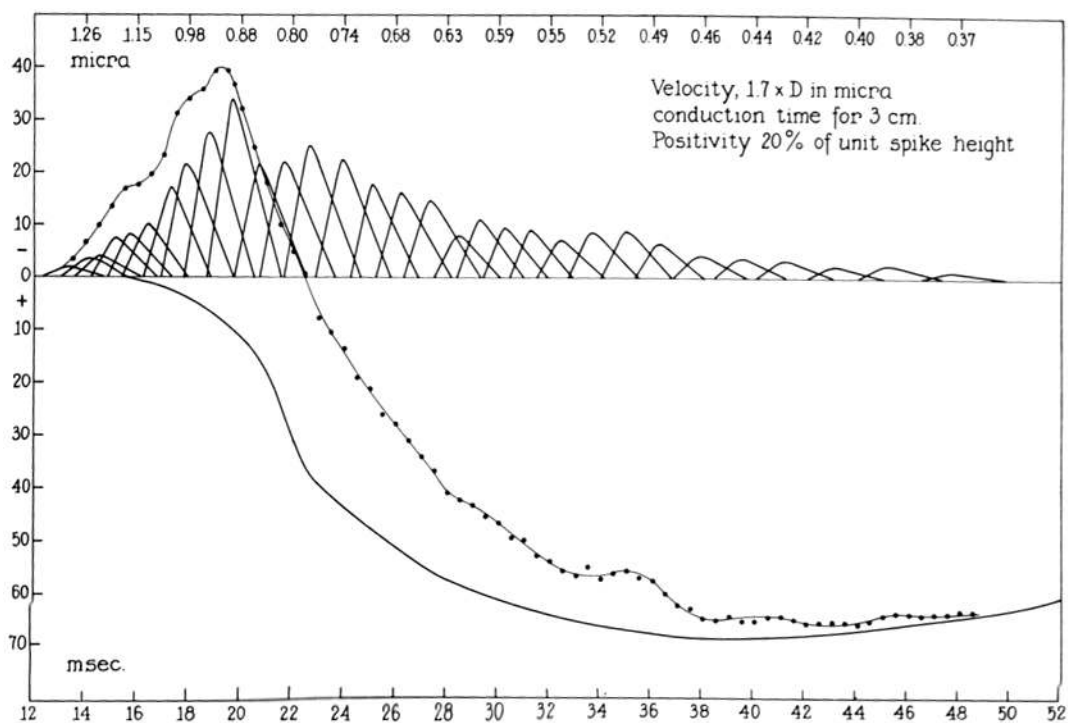


FIG. 10. Construction of a d.r.c. action potential from the diameter measurements presented in Fig. 9. On the upper scale can be read the size ranges assembled for the triangles, and on the lower scale conduction times representing velocities ranging between 2.34 and 0.61 m./sec.

each case and obtaining the ratio of the means of the measurements. In order to be able to bring the new measurements into relationship with the size analysis previously made on silver-stained material, a shrinkage of 10 per cent based on the findings of Hursh, then used, was accredited to the paraffin technique. With a 19 per cent increase over the size in paraffin, the fibers in methacrylate would be swollen by 7 per cent. A correction for this amount of swelling was made.

In an ideal preparation all the fibers would be round. As the ideal is never reached, values for the diameters require calculation of the square roots of products of the two major axes of ellipses. After correction for magnification and swelling, the diameters, so obtained, were plotted at  $0.01\ \mu$  intervals. The representation of the several sizes can be seen in Fig. 9. Through the introduction of accuracy of measurement for fibers smaller than one-half a micron, there is improvement over the previous analyses. Otherwise, no difference of significance appears. The size range is encompassed between 0.3 and  $1.35\ \mu$ .

Construction of the action potential was made according to the specifications previously laid down (Gasser, p. 254), which will not be repeated here. It is presented in Fig. 10. The velocities were precisely accounted for when the diameters in the new measurements were multiplied by a constant. Conduction in these fibers is a *linear* function of the diameter. How far afield one can go with a square root hypothesis can readily be ascertained by anyone who will take the trouble to try it.

#### *Dorsal Roots*

*Histology.*—What a difference in the appearance of the fibers there is between root and nerve can be seen at a glance in Fig. 11, which is presented at a magnification the same as that used in Figs. 1 and 2. Immediately obvious are the large sizes of some of the Schwann sheaths and the fact that, although the sheaths contain many more fibers than they do in the nerves, there is also much more unoccupied space in the Schwann cytoplasm. The fibers are smaller than in the nerves; and, instead of being attached to the membrane by individual mesaxons, often they are held in bunches.

Where and how the reduction in the sizes of the fibers comes about are questions that present themselves. To the first question a definite answer can be given. In sections at the nerve pole of the ganglion the fibers resemble those in Figs. 1 and 2. In sections of the root there is no difference between the ganglion end and the cord end. Clearly the reduction in size must take place in the ganglion. That the reduction takes place by division of the central branch is the most likely hypothesis; but direct proof for it is not forthcoming, although much time was spent in searching for it. In the sections  $0.1\ \mu$  in thickness the small coverage is against the chance of finding the proper locations. The large axons, before the *T*-division, were seen, and many small axons, the

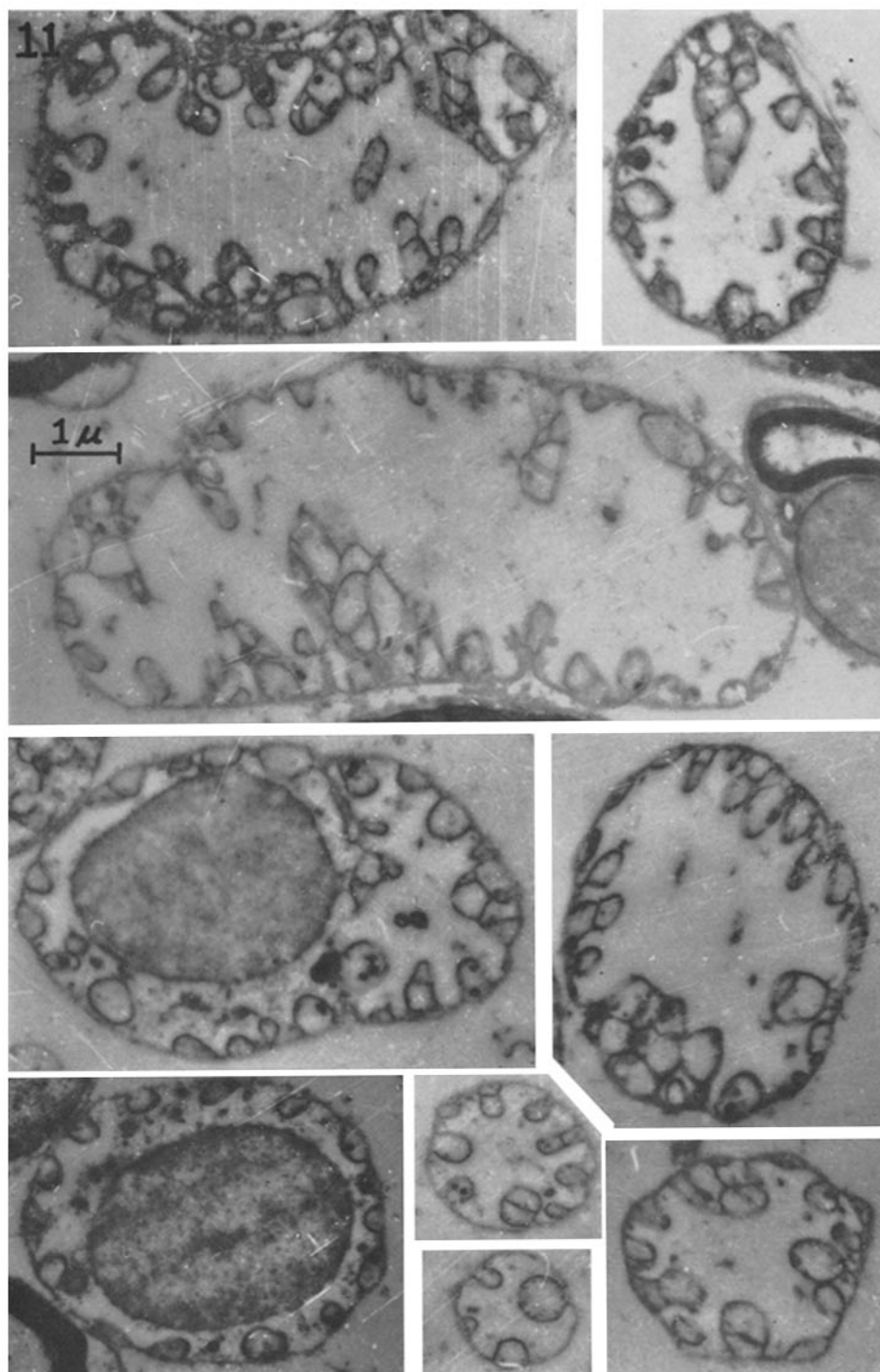


FIG. 11. Schwann sheaths from electron microscope pictures of spinal dorsal roots of the cat. All at the same magnification,  $\times 12,000$ .

latter becoming more numerous as the sections approached the root pole of the ganglion. But splitting of the central branch did not come into view nor, for that matter, the well known *T*-division itself. Thus, it is necessary to turn to indirect evidence for the answer to the second question.

Davenport and Ranson counted the cells in the dorsal root ganglia, the total number of axons in the roots, and the number of medullated axons. If one take their counts for the second sacral root of the cat, the root closest to the ones studied in this paper (L. 7 and S. 1), and allow a ganglion cell

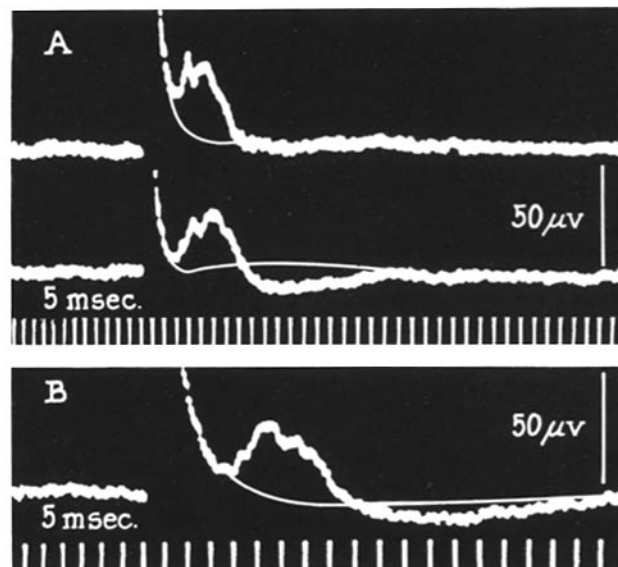


FIG. 12. Action potentials of the C fibers in S1 dorsal roots of the cat. D.C. differential amplification. Stimulating cathode at earth potential. Distal lead cocaineized. A, records at two conduction distances: 17.2 mm. (upper), 20 mm. (lower). Distances between leads: 7 mm. (upper), 5 mm. (lower). 37.6°C. B, another root. Conduction distance, 20 mm. Distance between leads, 8 mm. 37.7°C.

for each myelinated fiber, it works out that there is left one ganglion cell for every 0.7 of an unmyelinated fiber. When one now takes into consideration that what the authors were counting were black dots revealed by the silver stain and that many of the black dots would have been constituted by two to five or more unresolved fiber components, it follows that the number of fibers in the root must have been a multiple, larger than one, of the number of residual cells in the ganglion. There is room for many more fibers than ganglion cells, hence evidence that division must take place.

*C Action Potential in Dorsal Roots.*—Technical difficulties beyond those of



recording C fibers in skin nerves are present in relation to the roots. Selection of a conduction distance optimal for placing the action potential in the most favorable position for recording on the A after-potential is not possible; and the action potential is smaller. Shunting by the large fibers is much less in the skin nerves, in which the d.r.C fibers are concentrated, than in the roots where they are in parallel with the total segmental population of afferent fibers. Also, in line with the small size of the fibers in the roots, temporal dispersion is faster so that the amount of summation of the component units is reduced.

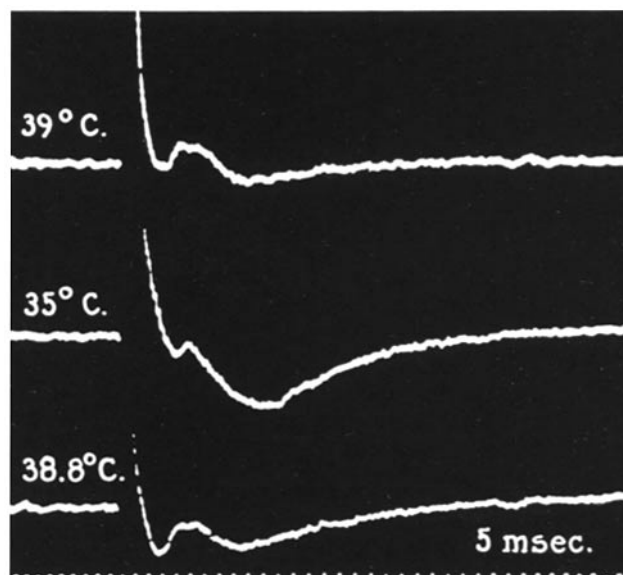


FIG. 13. Effect of temperature on the postspike positivity of the C action potential of a S1 dorsal root of the cat. Conduction distance, 20 mm. Distance between leads, 9 mm. From above downward: start, cooling, recovery.

As a result of all the contributing factors it was found that it was necessary to deal with potentials ranging between 15 and 35 microvolts. This meant amplification well into the noise level and the use of a filter in order to make the records legible. A filter such that there was a 30 per cent reduction in size at 1,000 cycles was employed. No observable alteration of the form of the action potential resulted.

Examples of the action potential are seen in Figs. 12 and 13. As in the peripheral branches the action potential manifests post-spike positivity. In no preparation, however, was it so strikingly displayed, which admits of the interpretation that the differential may be real rather than a result of procedure. Also, as in the peripheral branches, the post-spike potential is tem-

perature-sensitive (Fig. 13). Whatever the significance of the temperature sensitivity of the potential may be in skin nerves, there, at least, the consideration can be brought into the argument that skin nerves function over a range of temperatures, all usually below the temperature of the rectum. In the spinal canal, on the other hand, the temperature must be close to rectal and relatively invariable. Thus, temperature sensitivity in the roots may be

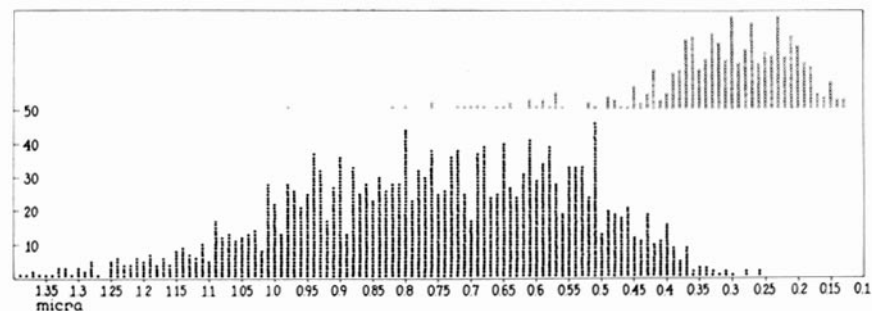


FIG. 14. Sample of the fiber sizes in a cat dorsal root (circles), plotted in comparison with the distribution of fiber sizes in a nerve (dots).

TABLE I

*Velocities at the Starts of the Elevations in the Compound Action Potentials of Nerves and Roots m./sec. and the Ratios between Them*

Nerve	Root	R/N
2.3	1.44	0.63
1.89	1.12	0.59
1.58	0.89	0.57
1.31	0.65	0.50
1.06	0.59	0.56
0.89	0.48	0.54
0.72	—	—

nothing more than a reflection of homogeneity of parts of the neuron, and may be a property upon which there are no physiological demands.

The compound action potential of the root is characterized by a series of elevations in correspondence with those in the nerve. For the regularly identifiable positions in the nerve compound action potential, velocities were given in Table I of the previous paper (Gasser, 1950). Wherever in the present series the velocities in the roots could be determined at corresponding positions, the readings were made. Averages of the readings are placed in Table I along side of the nerve velocities; and in the third column the ratio between the two is calculated. In summary, what the table reveals is that the pattern of im-

pulses in the nerves continues in the roots, with the conduction velocities of its components reduced to between 50 and 60 per cent of their nerve values.

If the guide to the construction of the action potential from the histological picture still holds for very small fibers, then the root action potential should be producible with a set of velocities obtained by multiplying by 1.7 a set of fiber sizes 50 to 60 per cent of those found in the nerve. In order to test the validity of the deduction, measurements were made of the fibers in a collection of prints representing electron microscope sections close to one another at the ganglion end of a root. Enough fibers were measured to give a fair sample of the size range, but *not* enough or under the proper conditions to yield a distribution curve. When plotted in Fig. 14 in comparison with the distribution curve of Fig. 9 it is seen that the expectation is fulfilled. Thus, it appears that the size-velocity relationship for unmyelinated fibers is sufficiently determined so that one can infer the size distribution from the action potential. An analysis, which in this case would be prohibitively laborious, is unnecessary.

#### SUMMARY

As an aid in the interpretation of the physiological properties of unmyelinated nerve fibers, particularly those having their cells of origin in the dorsal root ganglia, more precise information about their morphology has been acquired through employment of the electron microscope.

The appearance of the fibers in the skin nerves is described, with special reference to the structure of their sheaths; and a notation is made about the bearing of the axon-sheath relationship on the biophysical mechanism of conduction (p. 714).

There is no basic difference between the sheath systems of the d.r.C and the s.C fibers.

Attention is called to a point of similarity between the sheaths of unmyelinated and myelinated axons (p. 715).

An assessment was made of the likelihood of interaction between the fibers. In action potentials showing temporal dispersion at several distances, the elevations appeared in their calculated positions. A model of a group of Schwann sheaths was constructed from successive electron microscope sections, showing that the lengths of the sheath branches are short in comparison with the wave lengths of the action potentials. Supported by these and other considerations, the argument is strongly in favor of the conclusion that among d.r.C fibers, as in other fibers, there is no cross-excitation between the axons.

A new analysis of the size distribution of the fibers in a sural nerve was made from electron microscope pictures; and from the measurements the action potential was constructed. The result confirmed the view, previously expressed,

that the velocities of conduction in the fibers can be *precisely* accounted for by multiplying the diameters by a constant.

In the dorsal roots, the striking change that takes place in the appearance of the fibers and their disposition in the Schwann sheaths can be seen in Fig. 11. The axons partake of the special properties of the peripheral branches, which necessitated the creation of the subdivision of d.r.C fibers. But, their diameters are much smaller. At a set of reduced conduction velocities the configuration of the compound action potential in the nerves is repeated in the roots, with the root velocities still conforming to the size-velocity rule derived from nerve axons.

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