THE CAPSULE OF SPINAL GANGLION CELLS

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There are various accounts of the nature of the capsular elements of spinal ganglion cells. De Castro (1931) describes a 'corona' of spindle-shaped satellite cells with branching processes within an external covering of collagenous and reticular fibres. The intracapsular part of the axon, he states, is tortuous and convoluted forming a 'glomerulus'. Ranson's *Anatomy of the Nervous System* (1953) shows a nucleated membranous sheath continuous with the axon neurilemma and satellite cells embedded in the substance of the nerve cell, while Cajal's pictures, reproduced in Maximow & Bloom's text-book (1957) have a discontinuous layer of satellite cells, some of them enclosed by dendritic loops arising from the surface of the nerve cell, separated by a clear space from the cell body. In haemalum- and eosin-stained sections examined with the light microscope there is a 'shrinkage?' space around most of the ganglion cells, limited externally by a capsule of cells amongst which are fibroblasts, while in a number of neurons nuclei indent the cell surface.

In previous reports on the fine structure of spinal ganglion cells (Hossack & Wyburn, 1954; Dawson, Hossack & Wyburn, 1955; Dawson & Wyburn, 1955) mention was made of the capsular elements, but with the thick sections produced at that time by a modified Spenser microtome no detailed interpretation was attempted.

MATERIAL AND METHODS

The spinal ganglia of normal adult rabbits were used. The animals were anaesthetized with Nembutal and the ganglia removed before death, placed in a few drops of fixative on a glass slide, cut into one or two pieces with a sharp razor blade, and quickly transferred to a dark tube containing Zetterquist (1956) 1 % buffered osmic acid.

The fixation was continued for 4 hr. at room temperature, the tissues were washed overnight, then were dehydrated through a graded series of alcohols. One hour in each change was sufficient to ensure preliminary dehydration; final dehydration was effected by a 2 hr. immersion in alcohol which had been re-distilled over calcium hydride.

Following several changes of *n*-butyl methacrylate the tissue was embedded in a mixture of pure *n*-butyl methacrylate containing 5 % methyl methacrylate, and 1 % benzoyl peroxide as catalyst, before being finally embedded in gelatin capsules. Polymerization was carried out by placing the capsules in an oven at 56° C. for 3–6 hr.

Sections of from 500 to 300 Å. were cut with a Cooke and Perkins microtome, mounted on copper mesh grids, and examined in the Philips E.M. 100.

DESCRIPTION

Each ganglion cell is closely and completely invested by a sheath of nucleated cytoplasm, including the area of the 'implantation cone' of the axon. The narrowest part of the sheath—between the nuclei—is $\pm 0{\cdot}5\mu$; and the widest—at the nuclei —is $\pm 3\mu$ (Pl. 1, figs. 1, 2). Only the broader regions, therefore, can normally be seen with the light microscope giving the impression of nuclei indenting the nerve cell surface at intervals. The satellite nuclei more often than not occur in pairs separated by the cell membranes, so that the broad ends of adjacent cells are in contact (Pl. 1, fig. 1). It is more difficult to see the cell boundaries in the internuclear regions (Pl. 2, fig. 4). The satellite sheath thus appears in section as pear-shaped cells with long tapering processes, so arranged that the broad ends and the narrow ends of the cells are adjacent to one another. The satellite cytoplasm tends to be lighter in texture than the nerve cell cytoplasm, largely due to the absence of Nissl's substance, although there are scattered accumulations of small granules very similar to those of the Nissl aggregations, and the mitochondria, like those of the nerve cell, are small, dense and spherical. No endoplasmic reticulum or Golgi substance was seen in the sheath cytoplasm. The satellite sheath is bounded externally by double membranes seen in section as two lines, the outer one a basement membrane. Internally, there is also a double separating membrane between the sheath and the nerve cell—resolved at the lower end of fig. 5 (Pl. 2)—so that the satellite nuclei are not, as has been stated, embedded in nerve cell substance. The boundary between sheath and nerve cell is, however, highly irregular, with folds, loops, involutions, and evaginations of nerve cell substance into the satellite cytoplasm, as though 'streamers' of various sizes and shapes were flowing out from the nerve cell surface. Some of the coiled tubular projections cut in section look like vacuolated mitochondria in the satellite cytoplasm (Pl. 2, fig. 4), and other prominent protrusions might well represent the so-called subcapsular dendrites which have been shown in silver-stained preparations. Although for the most part the cells of spinal ganglia occur singly, each with its own coverings, pairing is not infrequent, and in some of the cell pairs, parts of the adjacent cell surfaces are in immediate contact, with no intervening satellite sheath (Pl. 2, fig. 3). Palay (1957) shows electron micrographs of cell pairs, each cell, however, with a covering of satellite cytoplasm and the two sheaths separated by an extracellular cleft.

Immediately outside the satellite sheath there are Schwann cells with unmyelinated axons in their substance. The Schwann cells are usually in close contact with the satellite sheath, and indeed at light microscopic level it would be difficult, if not impossible, to distinguish one from the other. They are, however, separated from the satellite sheath by two double membranes, their own plasma cell membrane, that of the satellite cells, and the two corresponding basement membranes (Pl. 2, fig. 5). The Schwann cells do not form a continuous enclosing covering, and there are areas of cell surface with only satellite sheath (Pl. 1, fig. 2). The distribution of Schwann cells appears to be determined by the course of the axon of any particular cell which, as shown by the study of silver-stained preparations with the light microscope, varies considerably. The axon may form a 'glomerulus' to one or other side, at the upper or lower pole of the cell, or may 'festoon' the whole cell. Thus the

main axon may be cut transversely, obliquely, and/or longitudinally in any one section. The number of unmyelinated nerve fibres in the Schwann cells varies, the size range extending from fibres of the same diameter as the cell axon (4 μ) to those of 0·2 μ or less (Pl. 5, fig. 9), all of them closed off from the Schwann cell cytoplasm by double membranes. Because of this, the Schwann cell cytoplasm, including in some sections the adjacent satellite cytoplasm, is cut up into irregular patterns by a network of double lines. The larger axons, with their characteristic texture and mitochondria, are obvious, but elsewhere the random criss-cross network of double lines makes it difficult to distinguish between fine nerve fibres and territories of Schwann cell cytoplasm. Other Schwann cells have, in addition to a large axon, what could be small nerve fibres around the periphery, an arrangement not unlike that of the unmyelinated nerve fibres of peripheral nerve, although there are no clearly identifiable mesaxons (Pl. 5, fig. 9). This disorderly reticulation is a constant feature of the pericellular Schwann cells associated with the axon glomerulus and, it is suggested, is caused by a plexus of fine nerve fibres within these Schwann cells which overflows into the adjoining satellite cytoplasm (Pl. 3, figs. 6, 7). External to the Schwann cells there are connective tissue cells and collagenous fibres.

Out of all the material prepared, only one block yielded a few sections through the axon hillock with about 20 μ of cell axon. The chances of obtaining similar sections from other cells were so small that the description of this region has had to be based on the appearances in the sections of this one cell which may not, of course, apply to all other ganglion cells. The sheath of satellite cytoplasm forms a collar at the junction of cell and axon, and thereafter the axon runs within its own Schwann cells. Flanking this first part of the axon and within the substance of the Schwann cells there is an irregular network of double-lined loops breaking up the cytoplasm and enclosing territories which in texture and type of mitochondria resemble the main axon (Pl. 4, fig. 8). Again, this appearance is interpreted to represent a plexus of fine nerve fibres coiled round the commencing cell axon within the Schwann cell cytoplasm, although the possibility of interdigitating cell processes cannot be excluded. The bounding membrane of the axon shown as a double line in fig. 10 (Pl. 5), like that of the parent cell, is irregular, with prolongations of axon substance into Schwann cell cytoplasm. Some of these projections are tubular, pursuing an irregular course, others are short triangular-shaped protrusions (Pl. 5, fig. 10). In the angle between the cell and the axon there is a Schwann cell with the 'pole' of its nucleus and a large nerve fibre. From its size it seems probable that this is a section through the first turn of the axon glomerulus.

DISCUSSION

The inability to distinguish between the Schwann cells accompanying the axon glomerulus and the true satellite cells at light-microscopic level accounts for the confusion arising from the different descriptions of the capsular elements of spinal ganglion cells. The relationship of satellite cells and nerve cell is comparable to that of axon and Schwann cells. The Schwann cells found outside the satellite sheath are associated with the axon glomerulus and are not primarily forming a capsule for the ganglion cells. The significance of the irregular nerve cell surface is not at

all clear. Similar but less elaborate folding of the limiting membranes of contiguous cells has been noted elsewhere, e.g. epithelial cells of the mucous membrane of the large intestine, and is most simply explained as a mechanism for fixing the cells together. Palay (1957) thinks the increased cell surface is necessary for the adequate nutrition of the neuron. Certainly the nearest blood vessels are extracapsular and the required substances have to diffuse through connective tissue elements as well as satellite sheath. Whatever its real meaning, the close fit of the satellite cytoplasm on to the irregular cell contour makes it unlikely that the clear space between the neuron and the capsule, seen in haemalum and eosin specimens, separates the nerve cell from the true satellite cells.

While the large axons in the Schwann cells are about the same diameter as the cell axon at its commencement, there are other axons less than half this size. While it is possible that the thickness of the axon changes within the glomerulus, it may, on the other hand, divide into two or more main branches, to rejoin before its bifurcation into peripheral and central processes.

Ehrlich (1886), Cajal (1890) and Dogiel (1896) have described a pericellular plexus of unmyelinated fibres round spinal ganglion cells, particularly plentiful, they state, in those of the horse, and less so in healthy human ganglia. According to Cajal, the pericellular plexus is prominent in diseased, injured, or transplanted ganglia, probably due to regenerative sprouting, and De Castro (1931) states that the pericellular plexus is extracapsular in position. If it is accepted that the network of double lines in the Schwann cell cytoplasm is due to a fine fibred nerve plexus invaginating into the cell substance, then there is the question of the origin of the feeding fibres of the plexus. Do they arise from the axon prior to its bifurcation? From the cell surface? From neighbouring neurons? Or from a combination of any or all of these sources?

While some of the tubular prolongations of the nerve cell surface could be regarded as dendrites, they were never seen to penetrate the satellite cytoplasm or connect up in any way with the network of the plexus. The irregularity of the axolemma of the proximal axon makes it difficult to be certain that definite branching exists. There is, however, at least one projection which could well be the origin of a branch to the surrounding plexus (Pl. 5, fig. 10). In another section (not shown) there was a short length of axon about 1 μ in diameter in its Schwann cell which divided into three branches at one end. Again, the plexus is most marked within the Schwann cells associated with the axon glomerulus. What evidence there is would thus seem to indicate that the pericellular plexus is formed by branches from the axon prior to its bifurcation. The plexus is not necessarily coiled round the whole nerve cell as shown in the classical Cajal preparations, but follows the course of the axon glomerulus, and so will vary in its relation to the cell surface in the different types of ganglion cell. The question of contributions from other neurons is unlikely to be settled by electron-microscope studies. It has always been assumed that the nerve impulses pass unchanged through the spinal ganglia, although Gasser (1955) has demonstrated that 'the pattern of impulses in the nerve continues in the roots, with conduction velocities of its components reduced to between 50 and 60 % of their nerve values'.

There was no evidence of anything in the nature of a true synapse between the

cell surface and surrounding nerve fibres, but whatever the origin, the existence of a pericellular plexus must have some purpose in terms of neural activity and could for instance have a phasic effect on the mainstream of afferent impulses.

SUMMARY

The capsule of spinal ganglion cells consists of satellite cells closely adherent to the irregular surface of the nerve cell. External to this there are Schwann cells associated with the axon glomerulus, which have a varying relation to the parent nerve cell. The Schwann cells contain a size range of axons, including what is interpreted as a plexus of fine fibres. The 'pericellular' plexus is also seen in the satellite cell, and it is suggested is formed by fine branches from the pre-bifurcation part of the axon.

REFERENCES

CAJAL, S. RAMON Y (1890). *Pequeñas communicaciones anatómicas. Sobre la existencia de terminaciones nerviosas pericelulares en los ganglios raquideanos.* Barcelona.

CAJAL, S. RAMON Y (1906). Die Struktur der sensiblen Ganglien des Menschen und der Tiere. *Ergebn. Anat. EntwGesch.* **16**, 177–215.

CAJAL, S. RAMON Y (1913). Estudios sobre la degeneración y regeneración del sistema nervioso. *Madrid, Moya*, 1, 2.

DAWSON, I. M., HOSSACK, J. & WYBURN, G. M. (1955). Observations on the Nissl's substance, cytoplasmic filaments and the nuclear membrane of spinal ganglion cells. *Proc. Roy. Soc.* B, **144**, 132–142.

DAWSON, I. M. & WYBURN, G. M. (1955). The spinal ganglion cell. *Proc. 2nd Int. Congr. Neuropath., Lond.*, pp. 597–599.

DE CASTRO, F. (1931). In *Cytology and Cellular Pathology of the Nervous System.* Ed. Wm Penfield. New York: Paul B. Hoeber, Inc.

DOGIEL, A. S. (1896). Der Bau der Spinalganglien bei den Säugetieren. *Anat. Anz.* **12**, 140–152.

EHRLICH, P. (1886). Ueber die Methylenblaureaction der lebenden Nervensubstanz. *Dtsch. med. Wschr.* **12**, 49–52.

GASSER, H. S. (1955). Properties of dorsal root unmedullated fibers on the two sides of the ganglion. *J. gen. Physiol.* **38**, 709–728.

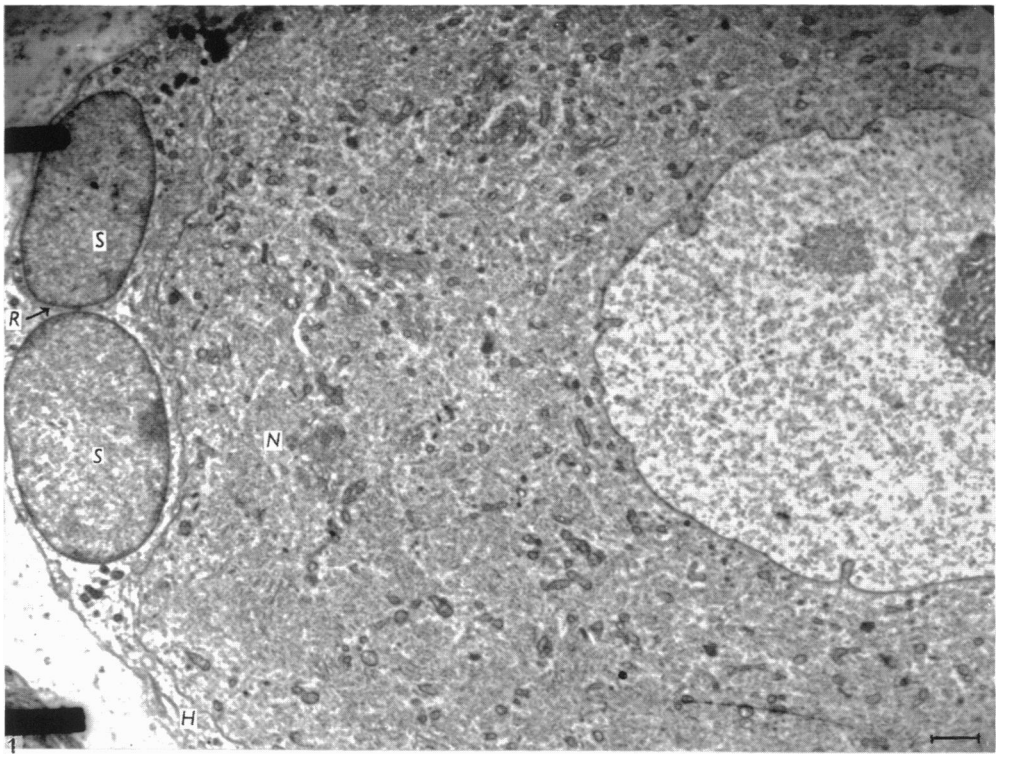
HOSSACK, J. & WYBURN, G. M. (1954). Electron microscope studies of spinal ganglion cells. *Proc. Roy. Soc. Edinb.* B, **65**, 239–250.

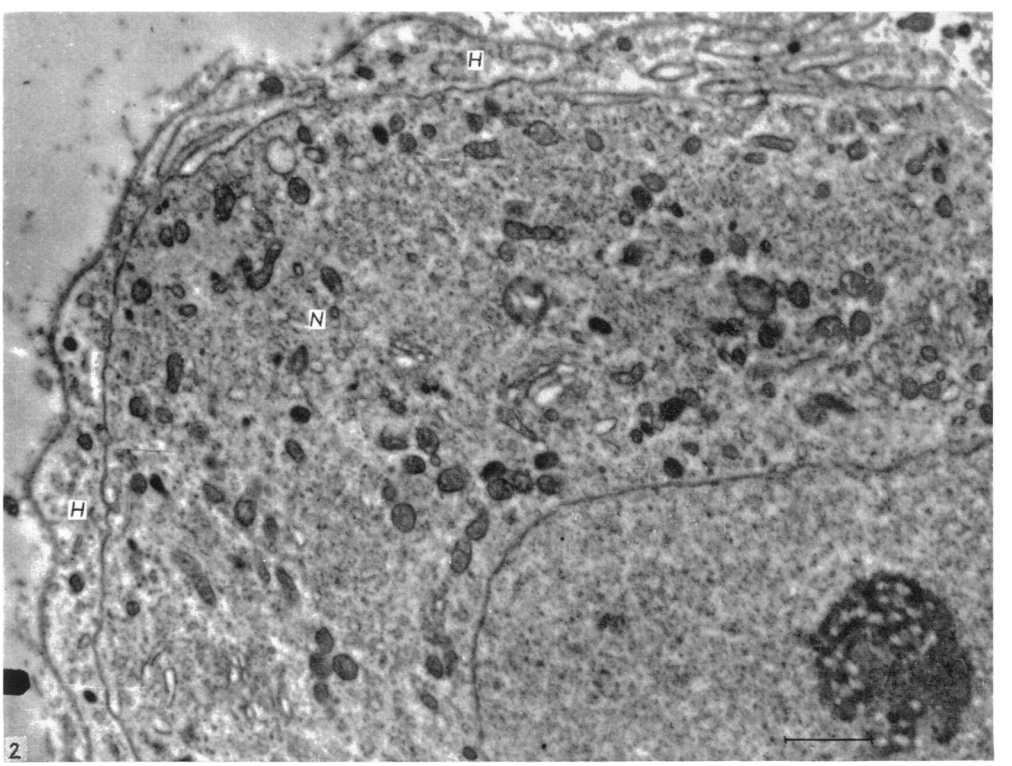
MAXIMOW, A. A. & BLOOM, W. (1957). *A Textbook of Histology*, 7th ed. Philadelphia: W. B. Saunders and Co.

PALAY, S. L. (1957). In *New Research Techniques in Neuroanatomy.* Ed. Wm F. Windle. Springfield, Illinois: C. C. Thomas.

RANSON, S. W. (1953). *The Anatomy of the Nervous System.* Philadelphia: W. B. Saunders and Co.

ZETTERQUIST, H. (1956). *The Ultrastructural Organisation of the Columnar Absorbing Cells of the Mouse Jejunum.* Stockholm: Karolinska Institutet.





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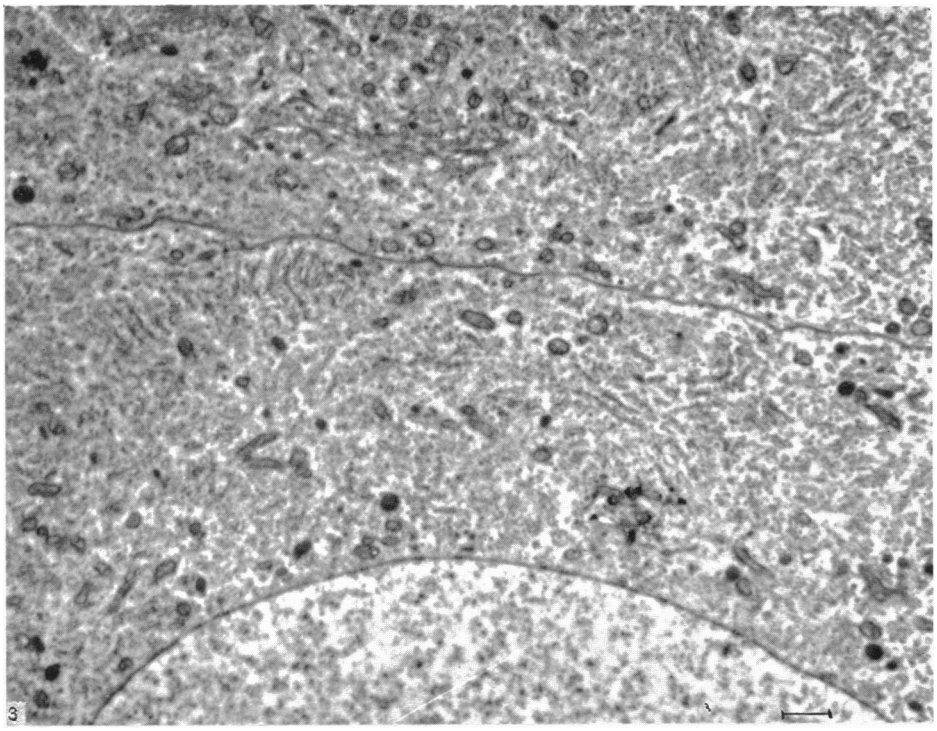
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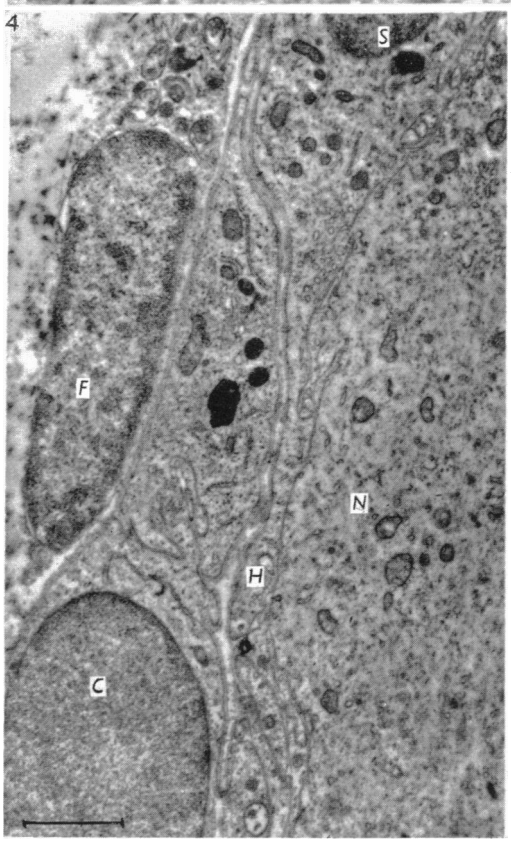
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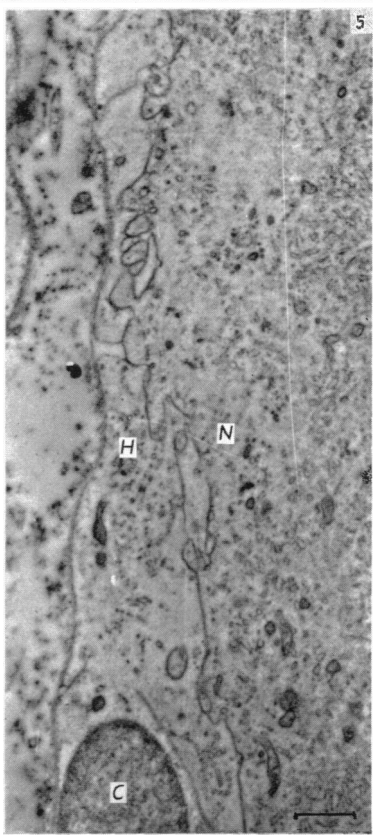
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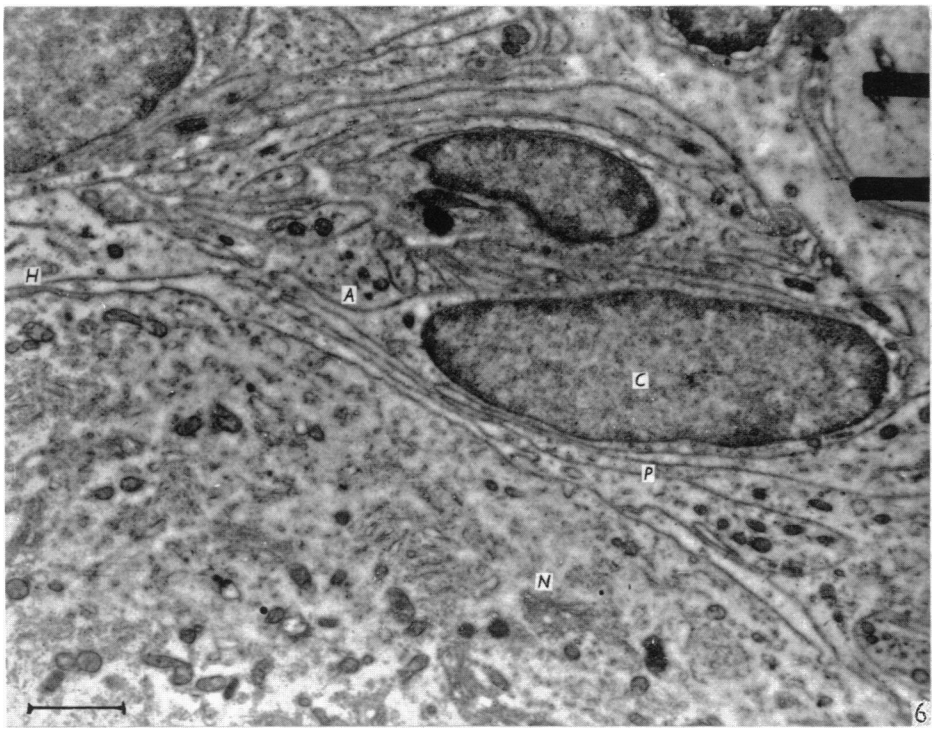
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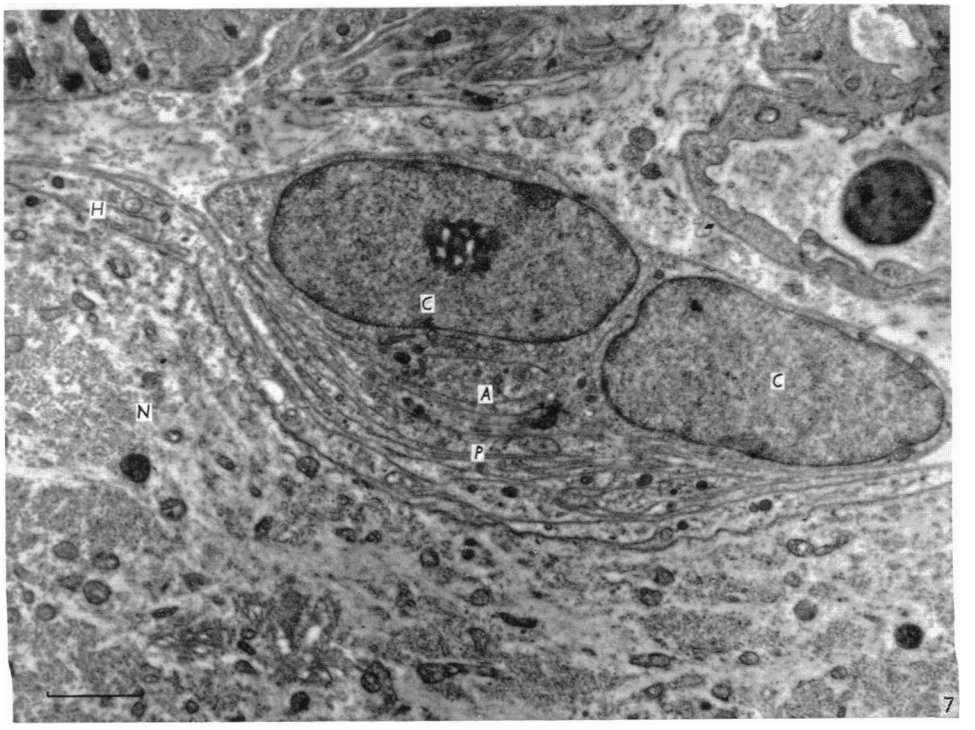
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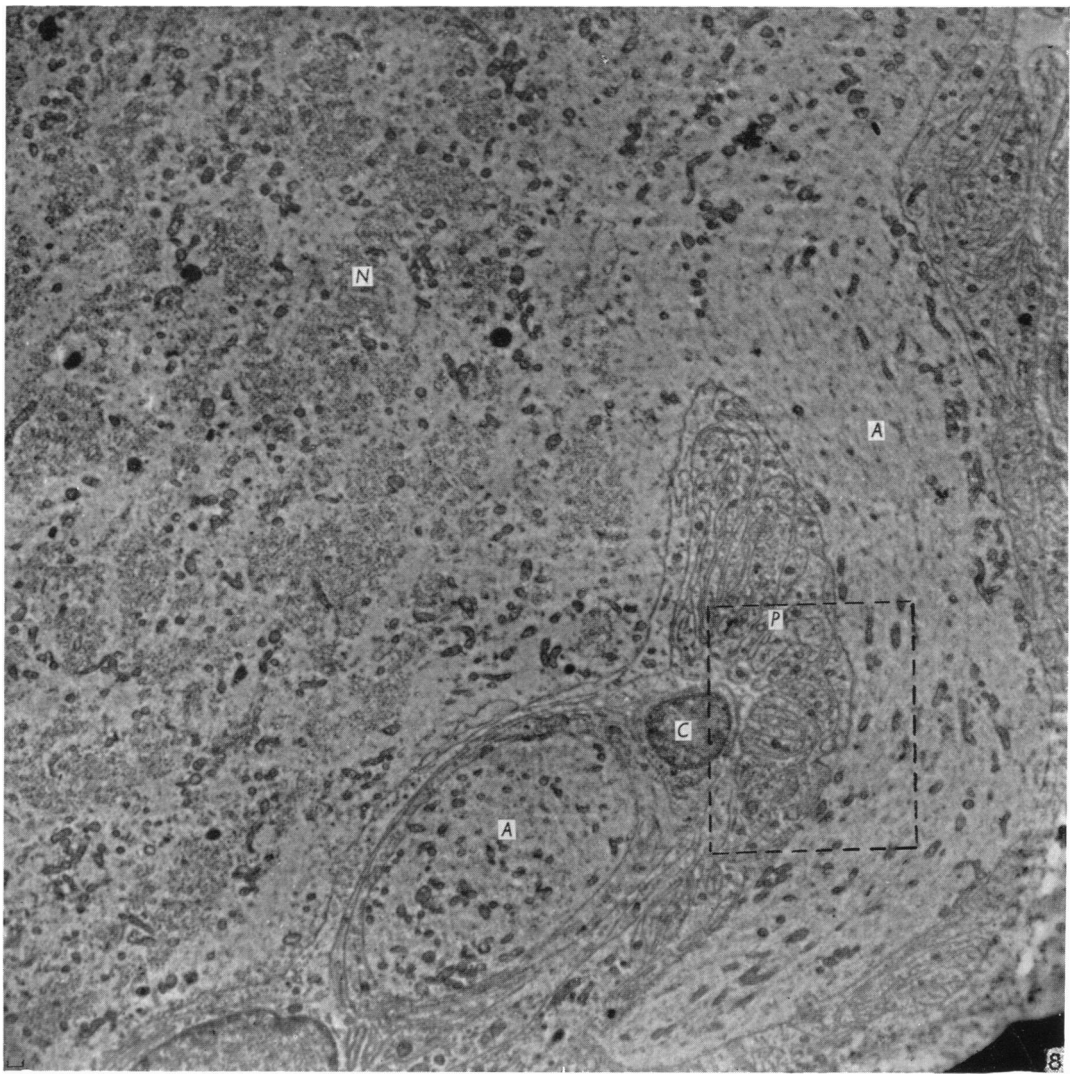

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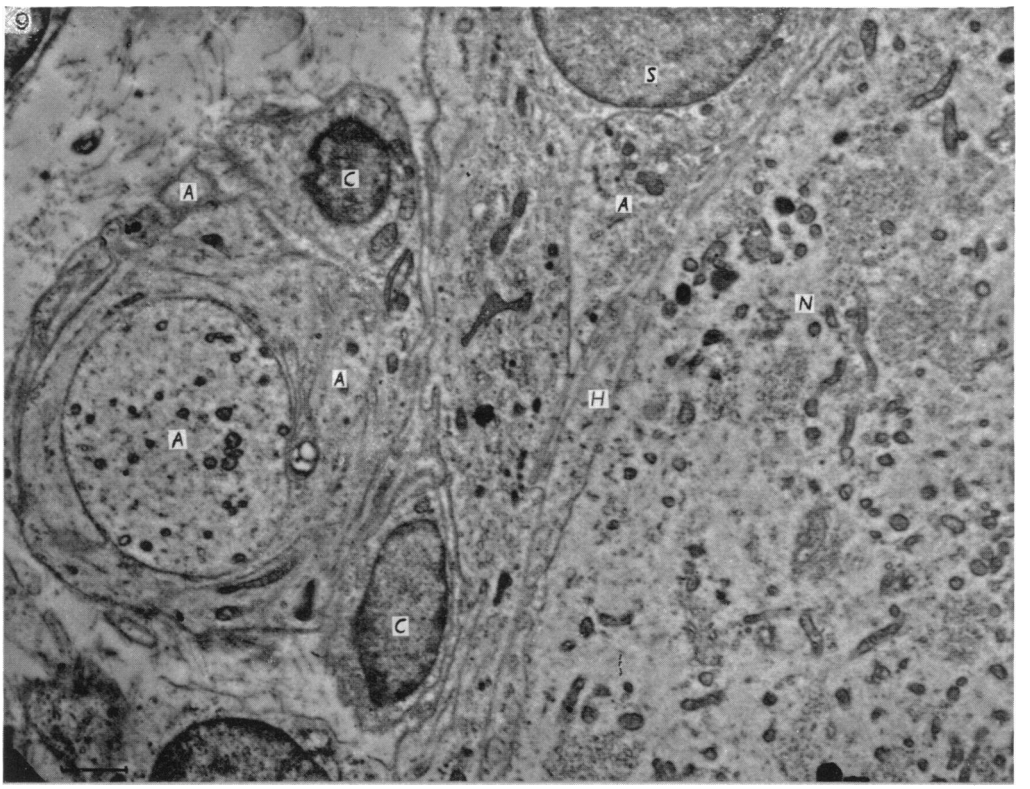
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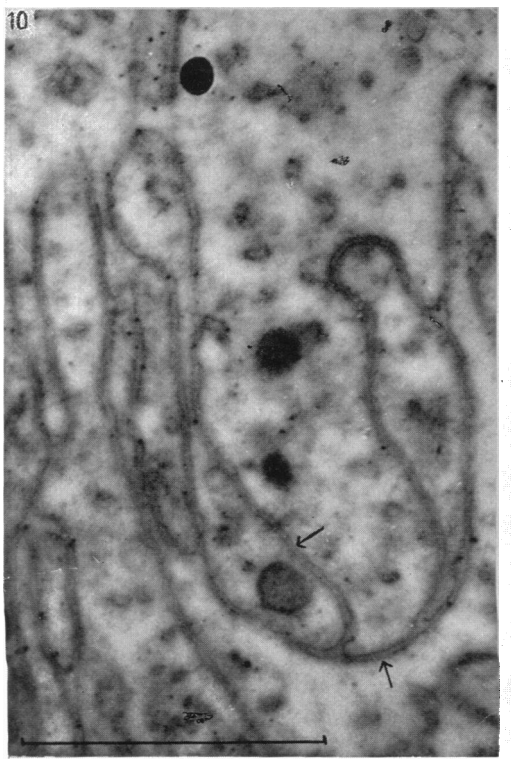
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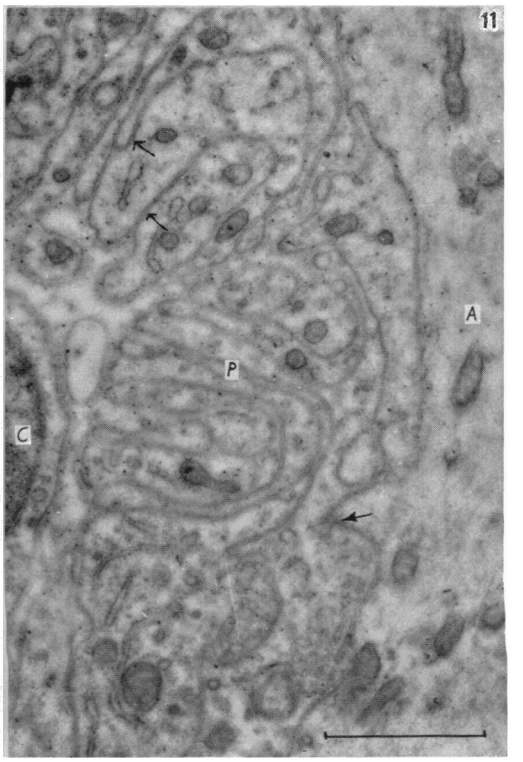
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EXPLANATION OF PLATES

All figures are electron micrographs. *A*, axon; *F*, fibroblasts; *N*, nerve cell; *R*, intercellular membrane; *C*, Schwann cell nucleus; *H*, satellite sheath; *P*, pericellular plexus; *S*, satellite nucleus. The marker in each figure represents the length of one micron.

Plate 1

Fig. 1. Section of nerve cell showing satellite cells with their nuclei.
Fig. 2. Section of nerve cell showing non-nucleated part of satellite sheath and the irregular folding and protrusion of the nerve cell surface.

Plate 2

Fig. 3. A pair of nerve cells with no intervening satellite cytoplasm.
Fig. 4. Section of nerve cell with irregular surface and satellite sheath.
Fig. 5. Section of nerve cell with satellite sheath, Schwann cell, and fibroblast.

Plate 3

Fig. 6. Section of nerve cell with Schwann cells, showing axons and pericellular plexus.
Fig. 7. Section of nerve cell with Schwann cells, showing axons and pericellular plexus.

Plate 4

Fig. 8. Section of nerve cell in region of axon hillock, showing commencing axon surrounded by pericellular plexus.

Plate 5

Fig. 9. Section of nerve cell, satellite sheath, and Schwann cells with a size range of axons.
Fig. 10. Higher magnification of area indicated in fig. 8. Note irregular axolemma and the double lines enclosing pericellular plexus.
Fig. 11. Higher magnification to show the double lines of invaginating axons.