MULTIPLE INNERVATION OF THE PACINIAN CORPUSCLE OF SLOW LORIS—AN ULTRUSTRUCTURAL STUDY

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SYNOPSIS

Five freshly dissected digital Pacinian corpuscles from the hand of slow loris examined electron microscopically showed that two of them were innervated by more than one nerve fibre. In one corpuscle, the accessory fibre had many myelin rings but possessed scanty axoplasm. It did not possess an unmyelinated terminal segment.

In the other corpuscle, the myelinated accessory fibre divided into two myelinated branches which were found within the outer core. These two accessory fibres subsequently lost their myelin sheaths but their unmyelinated terminals did not bear any lateral processes. The accessory fibres were accompanied by a pair of vascular capillaries which persisted as far as their unmyelinated terminal segments.

There is general concensus of opinion that Pacinian corpuscles are innervated, each by a single centrally situated nerve fibre. This view is based chiefly on the morphology of the corpuscles in the cat's mesentery. Innervation of some digital Pacinian corpuscles by multiple fibres has been recently described by Kanagasuntheram and Krishnamurti (1969) and Kanagasuntheram, Krishnamurti and Ahmed (1970) in the foetal and adult slow loris (*Nycticebus coucang*). However, these findings have been reported after light microscopy. The present account is therefore an attempt to deal with some variations occurring in the pattern of innervation of the digital Pacinian corpuscles of slow loris at the ultrastructural level.

MATERIAL AND METHODS

Five freshly dissected digital Pacinian corpuscles from the hand of slow loris were fixed in phosphate buffered glutaraldehyde and post-fixed in Dalton's chrome-osmium fixative. After dehydration, the material was embedded in Araldite and ultra-thin sections were then cut in a Porter-Blum ultramicrotome. Sections taken at approximately regular intervals were stained with uranyl acetate and lead citrate and examined in an Hitachi HS-8 electron microscope. Moreover, occasional 0.5 µ. thick sections were prepared from the material, stained with methylene blue and examined under the light microscope.

OBSERVATIONS

Of the five corpuscles examined, two had more than one nerve fibre within them. In one of these (PC 1), the main nerve fibre entered the corpuscle at one pole while the accessory fibre made its entry almost at the opposite pole. The latter was myelinated but its axoplasm which was scanty contained only a few mitochondria and was surrounded by a large number of myelin rings (Fig. 1). This myelin invaginated the main axon terminal which was situated within the inner core of the corpuscle (Fig. 2). Moreover, the accessory fibre which was devoid of the unmyelinated terminal segment was accompanied by a pair of capillaries (Fig. 3). The myelinated segment of the main nerve fibre innervating the corpuscle was also accompanied by a pair of blood vessels.

In the second corpuscle (PC 2) which was much larger than any of the others belonging to the present series, two myelinated fibres, the main and accessory one, were seen near its proximal pole (Fig. 4). However, the site of entry of the accessory fibre could not be determined as the relevant sections here were inadvertently lost. The accessory fibre, after traversing a third of the length of the corpuscle, divided into two (Fig. 5) and these two fibres which were myelinated were located within the outer core (Fig. 6). The accessory fibres extended for about half the length of the corpuscle and subsequently, after losing their myelin sheaths, became surrounded by cytoplasmic processes that bore some resemblance to those belonging to the inner core (Figs. 7 and 8). No radial clefts were seen in the inner core sur-

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Fig. 1. Electron micrograph showing heavily myelinated accessory fibre in PC 1 with scanty axoplasm and a few mitochondria. \times 22,000.



Fig. 2. Electron micrograph showing myelinated accessory fibre (arrow) invaginating the main axon terminal of PC 1. \times 10,800.



Fig. 3. Electron micrograph showing the accessory fibre (arrow) accompanied by a pair of blood vessels in PC 1. \times 4.200.



Fig. 4. Light micrograph showing the main fibre (bottom arrow) and the accessory fibre (top arrow) near the proximal pole of PC 2. \times 510.



Fig. 5. Light micrograph showing the main and accessory fibres in PC 2. Note the accessory fibre has now divided into two branches (arrows). \times 510.



Fig. 6. Electron micrograph showing the two branches of the accessory fibre shown in Fig. 5. Note that the two accessory fibres are myelinated and are accompanied by only one pair of blood vessels. $\times 4$ 200.



Fig. 7.



Fig. 8.

Figs. 7 and 8. Electron micrograph showing cytoplasmic processes encircling the terminal segments of the two accessory fibres in PC 2. Note the absence of radial clefts. \times 22,000, \times 18,000 respectively.



Fig. 10. Electron micrograph showing a small process (arrow) arising from the main nerve fibre and entering the radial cleft of the inner core in PC 2. \times 18,000.

rounding the accessory fibres. Moreover, the terminal portions of the accessory fibres did not have the lateral processes described in the main terminal by Kanagasuntheram. Krishnamurti and Wong. The accessory fibres contained only a few mitochondria and practically no small vesicles. Both accessory fibres terminated at different levels inside the corpuscle well proximal to the termination of the main nerve fibre.

The main nerve fibre of this large corpuscle (PC 2) which was accompanied by a pair of blood vessels lost its myelin sheath after traversing about half the length of the corpuscle. Thereafter, the unmyelinated fibre was surrounded by the lamellae of the inner core which had well developed radial clefts through which small projections emerged from the main nerve fibre (Fig. 10). Subsequently, two subsidiary fibres. presumably branches from the main nerve, were seen within the lamellae of the inner core (Fig. 9). These fibres contained both mitochondria and neurofilaments but their precise mode of origin and termination was not determined as the relevant sections were lost. The distal half of the unmyelinated nerve fibre showed lateral processes thus confirming our previous observations.



Fig. 9. Electron micrograph showing the main fibre and its subsidiary branches within the inner core in PC 2. \times 8,400.

The blood vessels accompanying the main nerve fibre terminated where the myelinated segment ended. On the other hand, the blood vessels following the accessory fibres persisted even after these two fibres had lost their myelin sheaths and extended as far as the distal end of the unmyelinated segment (Fig. 11). Although there were two accessory fibres, only one pair of vessels were seen accompanying them.

No dense core vesicles were seen in any of the nerve fibres in the five corpuscles examined.



Fig. 11. Electron micrograph showing the unmyelinated segment of an accessory fibre (arrow) in PC 2 accompanied by a pair of blood vessels. \neq 4,200.

DISCUSSION

Accessory Fibres

The presence of accessory fibres within the Pacinian corpuscle was described by several investigators (Takashi, Saki and Uzizima, 1955; Takashi, 1957; Kanagasuntheram and Krishnamurti, 1969; Santini, 1969 and Kanagasuntheram et al, 1970). Such findings were viewed with suspicion owing to the limitations and uncertainties of light microscopy. The present findings at ultrastructural level have confirmed the view that there could be more than a single nerve fibre innervating a corpuscle although, the functional significance of such accessory fibres remains uncertain. For example, the heavily myelinated accessory fibre with scanty axoplasm and which indented the main terminal in PC 1 does not possess an unmyelinated segment. It is therefore possible that this accessory fibre is possibly undergoing degeneration and hence non-functional. However, in PC 2, the accessory myelinated fibre which later subdivided and finally terminated as two unmyelinated endings may have had some functional significance. These terminals may possibly have served as additional channels for transmission of neural impulses. In addition to these accessory fibres, two other smaller subsidiary fibres were found close to the main nerve fibre. Similar fine fibres, arising from the main fibre, have been described by Cauna and Mannan (1958). It is also noteworthy that neither the accessory fibres nor the subsidiary fibres contained any dense core (noradrenergic) vesicles and consequently, these fibres cannot be equated with the noraThe presence of several nerve terminals inside PC 2 brings this particular end-organ on par with the Meissner's corpuscle which is innervated by more than a single fibre. Thus this particular corpuscle is an exception to the general concept that the Pacinian corpuscle is supplied by a single unbranched nerve fibre which serves as a private line to the CNS (Cauna and Mannan, 1961).

Radial Clefts of the Inner Core

Pease and Quilliam (1957); Quillam (1966) and Nishi, Oura and Pallie (1969) believe that the radial clefts in the inner core of the Pacinian corpuscle may serve as the route whereby metabolites reach the active nerve ending from the vascular capillaries supplying the corpuscle. The presence of some nerve processes within the radial clefts lends additional support to such a concept since these processes are regarded by Kanagasuntheram *et al* as the possible excitatory sites whenever a mechanical stimulus is applied to the corpuscle. Moreover, the subsidiary fibres found within the inner core in PC 2 may have proceeded through the radial clefts in order to reach their final destination.

Vascular Capillaries of the Corpuscle

The findings of Pease and Quilliam (1957) that the vascular capillaries extend as far as the myelinated segment of the central fibre supplying the corpuscle are confirmed by the present observations. However, the passage of the capillaries in relation to the unmyelinated segments of the accessory fibres in PC 2 may be regarded as an interesting exception. Moreover, although the myelinated accessory fibre in PC 2 subdivided first into two myelinated branches before continuing as the unmyelinated terminals, the vascular capillaries did not undergo further subdivision. It may therefore be postulated that the number of vessels inside a corpuscle is determined by the number of myelinated fibres entering the hilar region of the corpuscle. Thus, near the hilar region there were two myelinated fibres (main and accessory) and hence only two pairs of vascular capillaries.

Lastly, the presence of accessory fibres within a corpuscle could have a developmental origin in that fusion of adjacent corpuscles could occur as suggested by Cauna and Mannan (1959). If this view is accepted, then PC 2 is merely a twin corpuscle whereas in PC 1, the blindly terminating myclinated fibre might have got included into the corpuscle which was developing in response to a normal stimulus from another nerve fibre lying in close proximity to the under-developed accessory fibre. Finally, it may be concluded that variations in the pattern of innervation of the Pacinian corpuscles are not unusual and these may possibly explain the different physiological patterns of activity exhibited by the corpuscle.

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REFERENCES

- 1. Cauna, N. and Mannan, G.: "The structure of the human digital Pacinian corpuscles (corpuscula lamellosa) and its functional significance." J. Anat., 92, 1, 1958.
- 2. Cauna, N. and Mannan, G.: "Development and postnatal changes of digital Pacinian corpuscles (corpuscula lamellosa) in the human hand." J. Anat., 93, 271, 1959.
- 3. Cauna, N. and Mannan, G.: "Organisation and development of the preterminal nerve pattern in the palmar tissues of man." J. Comp. Neur., 117, 309, 1961.
- 4. Kanagasuntheram, R. and Krishnamurti, A.: "Nerve endings in the skin of the foetal hand of slow loris." Folia Primat., 10, 263, 1969.
- Kanagasuntheram, R., Krishnamurti, A. and Ahmed, M. Mumtazuddin: "Nerve terminations in the digital skin of the hand in some lorisoids." Folia Primat., 12, 42, 1970.
- 6. Kanagasuntheram, R., Krishnamurti, A. and Wong, W. C.: "The digital Pacinian corpuscles in slow loris; observation on the lateral processes of the terminal nerve fibre." Acta Anat. (In press), 1970.
- 7. Nishi, K., Oura, C. and Pallie, W.: "Fine structure of Pacinian corpuscles in the mesentery of the cat." J. Cell Biol., 43, 539, 1969.
- 8. Pease, D. C. and Quilliam, T. A.: "Electron microscopy of the Pacinian corpuscle." J. Biophysic. Biochem. Cytol., 3, 331, 1957.
- 9. Quilliam, T. A.: "Unit design and array patterns in receptor organs." Touch, heat and pain. London: J. A. Churchill, 1966.
- Santini, M.: "New fibres of sympathetic nature in the inner core region of Pacinian corpuscles." Brain Research, 16, 535, 1969.
- 11. Takashi, M.: "On the development of the complex pattern of Pacinian corpuscles distributed in the human retroperitoneum." Anat. Rec., 128, 665, 1957.
- 12. Takashi, M., Saki, I. and Uzizima, H.: "On the terminal neural apparatus detectable in the retroperitoneum of man—a complex pattern of Pacinian corpuscle." Anat. Rec., 122, 17, 1955.