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The ultrastructure of the sensory end-organs (baroreceptors) in the atrial endocardium of young mini-pigs

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INTRODUCTION

Two main types of nervous ramification have been observed in the atrial endocardium: (*a*) an anastomosing network of thin fibres dispersed throughout the endocardium and (*b*) circumscribed, arborizing nerve endings arising from thicker myelinated fibres. The latter terminations are embedded in an aggregate of cells, and form with these a characteristic, well-defined end-organ. The end-organs are most frequent around the atriovenous junctions, but are also found scattered throughout the atrial endocardium (Smirnow, 1895; Dogiel, 1898; Lawrentjew, 1929; Holmes, 1957; Chabarowa, 1959; Miller & Kasahara, 1964; Floyd, Linden & Saunders, 1972). Physiological experiments (Coleridge, Hemingway, Holmes & Linden, 1957; Coleridge, Coleridge & Kidd, 1964; Kappagoda, Linden & Snow, 1972) in conjunction with morphological investigations (Coleridge *et al.* 1957) have provided firm evidence that these end-organs are pressure-sensitive mechanoreceptors.

Morphological investigations of the endocardial end-organs have been largely confined to methylene blue stained and metal-impregnated whole mount specimens, and the detailed structure of the end-organs is not known. In the present work, the structure of a number of endocardial receptor organs has been studied by light and electron microscopy in a large series of sections. The end-organs were found to contain cells resembling Schwann cells, and a nerve-terminal apparatus with numerous small mitochondria and glycogen granules, the latter being frequently associated with complexes of smooth endoplasmic reticulum. Also characteristic was the frequent occurrence of cyto-segresomes and lipofuscin granules, and there were indications that the end-organs may receive thin, possibly efferent, nerves in addition to the thick afferent fibres.

MATERIALS AND METHODS

Two mini-pigs (male and female, aged $6\frac{1}{2}$ and 7 weeks, weighing 7 and 8 kg respectively) were employed in the investigation. The animals were anaesthetized by intraperitoneal injection of Nembutal (70 mg/kg body weight) and pressure-ventilated through a tracheal cannula. After bilateral thoracotomy, the aorta and vena cava were clamped simultaneously just above the diaphragm and a cannula was tied in retrograde position in the thoracic aorta. The heart was arrested in

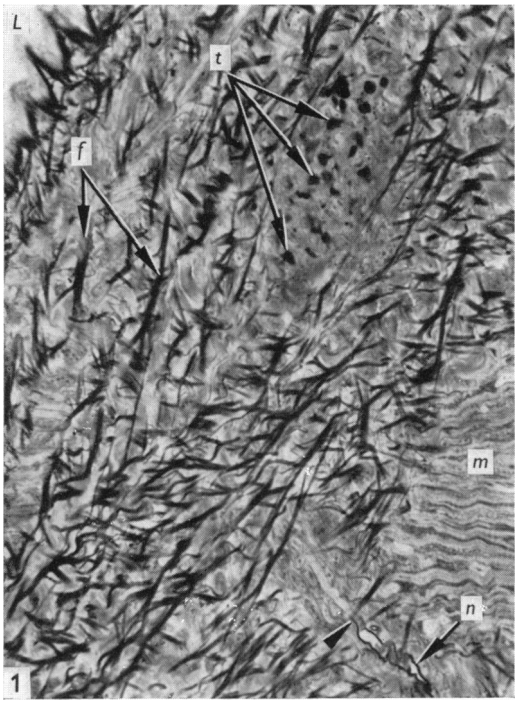
diastole by injecting approximately 10 ml KCl solution (150 mmol/l) into the left auricle and fixed immediately afterwards by perfusion through the aortic cannula and by infusion of fixative through two cannulae inserted in the chambers of the left and right ventricles. The left auricle and the inferior vena cava were cut open to drain off the fixative, and the neck vessels were clamped to minimize the loss of fixative through the cephalic vessels. The modified Karnovsky (1965) fixative consisted of 1% formaldehyde (prepared by depolymerization of paraformaldehyde) and 2·5% glutaraldehyde (TAAB) in 0·1 M phosphate buffer; its total osmolality was approximately 830 mOsm, and its pH, 7·3. An initial flow through the aortic cannula of about 200 ml/minute was maintained for about 1 minute and then reduced. After perfusion for 25 minutes, the heart was removed and immersed in cold fixative. Preliminary examination of whole mount specimens from other pigs had indicated the regions where end-organs were most numerous. From such regions – especially the orifices of the inferior pulmonary veins and an area in the anteromedial wall of the right atrium adjoining the ascending aorta – large pieces of tissue were removed and fixed for a further 5 hours by immersion in the above fixative at 4 °C. These tissue pieces were divided into a number of small blocks, which were post-fixed for 2 hours at room temperature in a 2% solution of OsO_4 in 0·1 M phosphate buffer, pH 7·3. The blocks were then dehydrated in ethanol and embedded in Epon 812. Most of them were oriented with the plane of sectioning parallel to the endocardial surface, but a few were cut perpendicular to this. The end-organs were located by inspecting 2 μm sections, which had been stained with toluidine blue or paraphenylene diamine, under the light microscope as they were cut. End-organs were found in about one quarter of the blocks. When an end-organ had been located in this way, series of ultrathin sections alternating with 1 μm sections were prepared with a diamond knife on an LKB Ultrotome. A total of 12 end-organs were examined in this manner. Most of the sections destined for electron microscopy were cut with the microtome set to 30 nm, the remainder at 70 nm. The sections were stained with uranyl acetate followed by lead citrate (Reynolds, 1963) or bismuth subnitrate (Riva, 1974) and examined in a Hitachi HS-7S electron microscope. A few grids were examined in a Phillips 300 electron microscope equipped with a tilt stage.

Fig. 1. Oblique section through the endocardium at the entrance of the right inferior pulmonary vein into the left atrium, revealing a compact end-organ. The sensory nerve terminals (*t*) are densely stained by paraphenylene diamine. A myelinated nerve fibre (*n*) emerges from the myocardium (*m*) and leaves the myelin sheath at arrow head. The continuity of the nerve fibre with the end-organ was established in the EM sections. *f*, densely stained elastic fibrils; *L*, atrial lumen. × 425.

Fig. 2. Section parallel to the surface of the endocardium in the roof of the left atrium showing an end-organ with abundant collagen fibres. *c*, irregularly shaped cells of the end-organ located among bundles of collagen fibrils; *sm*, strand of smooth muscle cells. Toluidine blue stain. × 425.

Fig. 3. Conglomeration of two end-organs (e_1 and e_2) located close to the myocardium (*m*) in the roof of the left atrium. A somewhat concentric arrangement of the cells is seen in the periphery of the organs. In the EM sections the nerve supplying the organs (shown in Fig. 5) was found in the space adjoining the myocardium. Toluidine blue stain. × 425.

Fig. 4. Section cut tangential to the endocardium in the anteromedial wall of the right atrium at the entrance of the superior vena cava, revealing an end-organ supplied by a thick nerve fibre (*n*). Nerve terminals (*t*) are seen scattered between the cells of the end-organ, the nuclei of which are clearly seen (*c*). *L*, atrial lumen. Toluidine blue stain. × 1050.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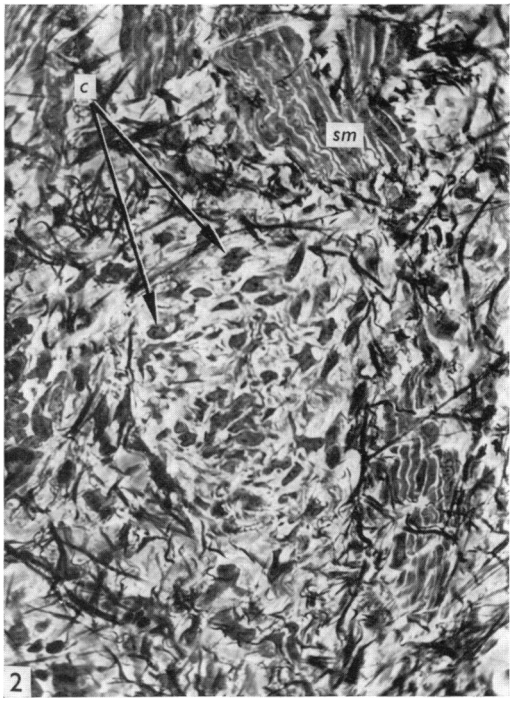
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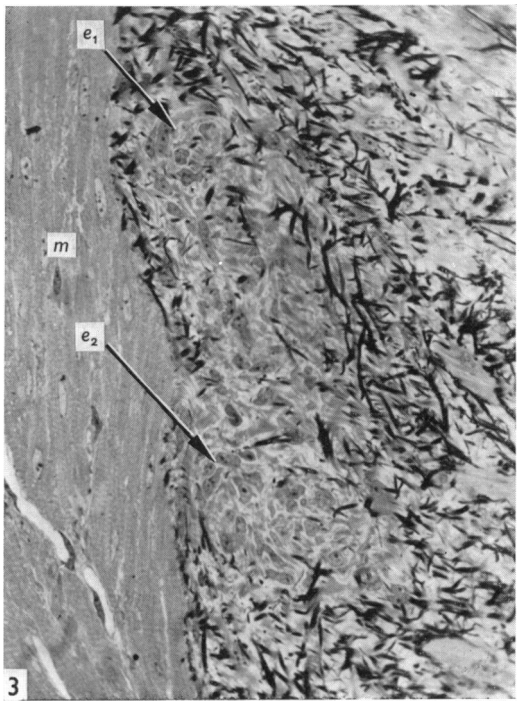
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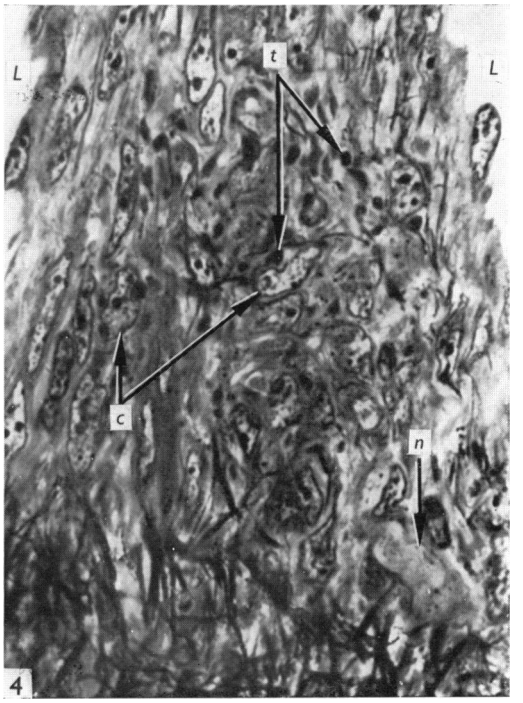
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OBSERVATIONS

Light microscopy

An individual end-organ appeared under the light microscope as a well-defined, flattened group of cells lying at a varying depth in the endocardium (Figs. 1–4). It measured 50–200 μm in length (parallel to the surface of the endocardium) and 10–20 μm in thickness. An organ sometimes produced a low, flat endocardial prominence, and at other times it could be situated close to the underlying myocardium (Fig. 3). A 4–9 μm thick nerve fibre entered the organ at its margin (Fig. 4) or from its deep side. The nerve fibre sometimes was seen to branch and supply several organs lying close together or conglomerations of such (Fig. 3). The last portion of the nerve before it entered the end-organ was always unmyelinated: myelination began at a varying, often considerable, distance from the organ (Fig. 1). Between the cells of the end-organ the terminal branches of the nerve could be distinguished, their ends being especially distinct with paraphenylene diamine staining (Fig. 1).

Electron microscopy

General structure

Two characteristic components entered into the structure of the end-organs: (1) the branches and terminals of the thick nerve fibres and (2) an aggregation of cells surrounding the nerve branches up to the terminals, which were completely or partly free. These cells, in their relation to the nervous structures and in their general morphological features, closely resemble Schwann cells, and so they will be called such in the following account. The end-organs also contained varying numbers of collagenous and elastic fibrils. Some organs were very compact and contained only a few connective tissue fibrils (Figs. 1, 8), whilst others were intersected by numerous bundles of fibrils (Figs. 2, 9). In these more loosely constructed organs a few fibroblasts were also present which had no particular relation to the nervous structures. No special orientation of fibrils, distinguishing them from those of the endocardium, was observed in the end-organs.

The organ had no capsule, but the peripheral components tended locally to be arranged concentrically with respect to its surface. None of the end-organs was found to be vascularized. Smooth muscle cells were often observed in the endocardium, but without special relation to the end-organs (Fig. 2). Apart from

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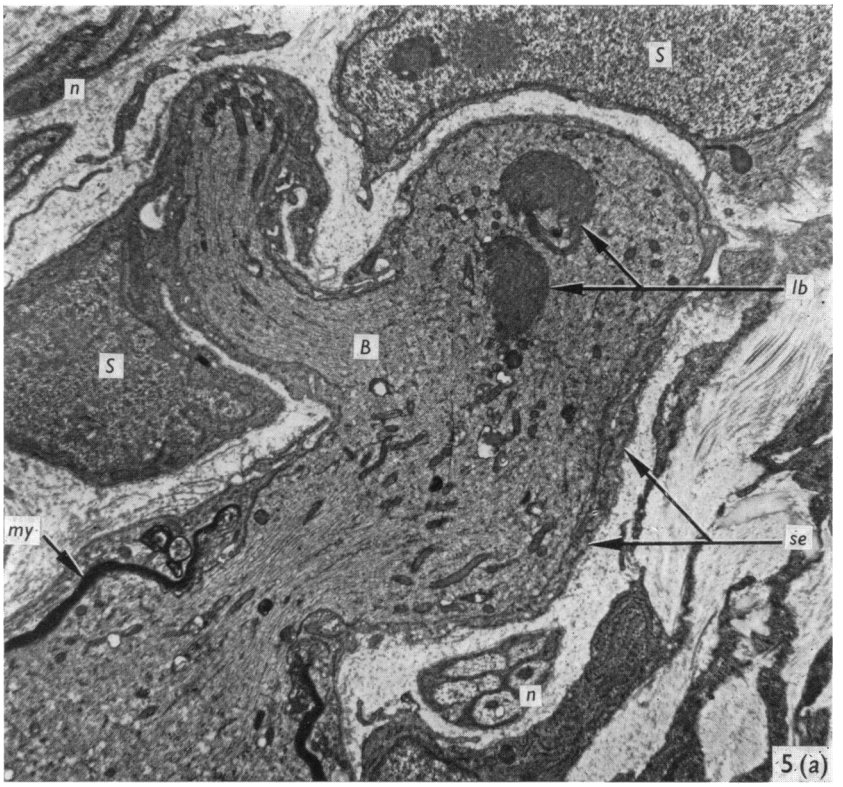
Fig. 5. (*a*) Low magnification showing the termination of the myelin sheath (*my*) of the thick nerve fibre supplying the end-organs shown in Fig. 3. Branching occurs at *B*. After the termination of the myelin sheath the nerve is surrounded by Schwann cell extensions (*se*). Small nerves (*n*) accompany the thick fibre. *S*, nuclei of Schwann cells. The thick fibre contains two laminated bodies (*lb*) which are shown, at higher magnification (Fig. 5*c*) in a neighbouring section, to be composed of concentrically arranged cisternae and a moderately electron-dense material (arrows). × 7100.

Fig. 5. (*b*) Detail of Fig. 5 showing the termination of the myelin sheath by a 'half node of Ranvier'. Small desmosomes (*d*) connect the cells across the nodal cleft. A fine periodic structure is seen between the membranes of the nerve fibre and the myelin-forming cell (inset). Dense particles (*p*) have accumulated in the cytoplasmic pockets of the Schwann cell. The nerve fibre contains numerous microtubules (*mt*) and some lipofuscin-like dense bodies (*li*). × 19500.



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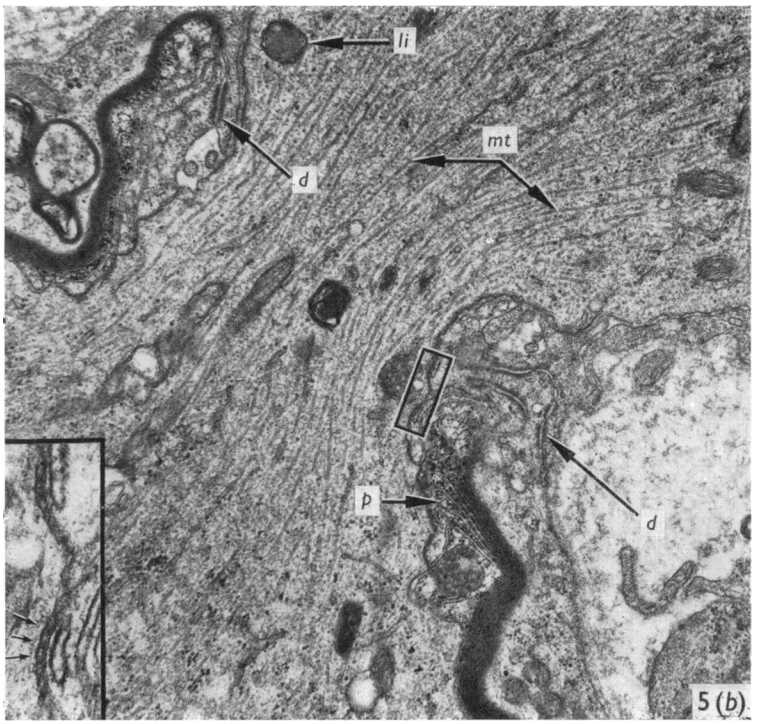

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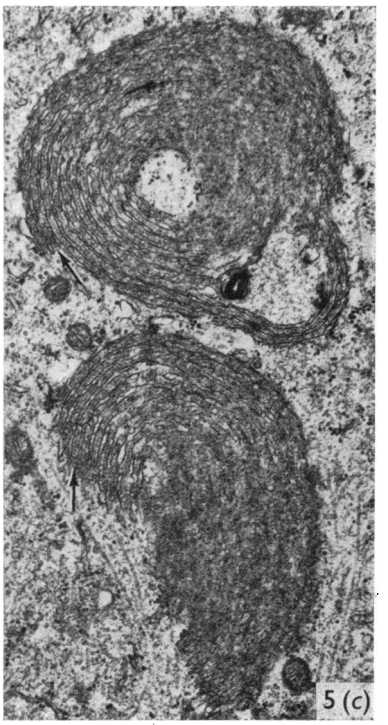

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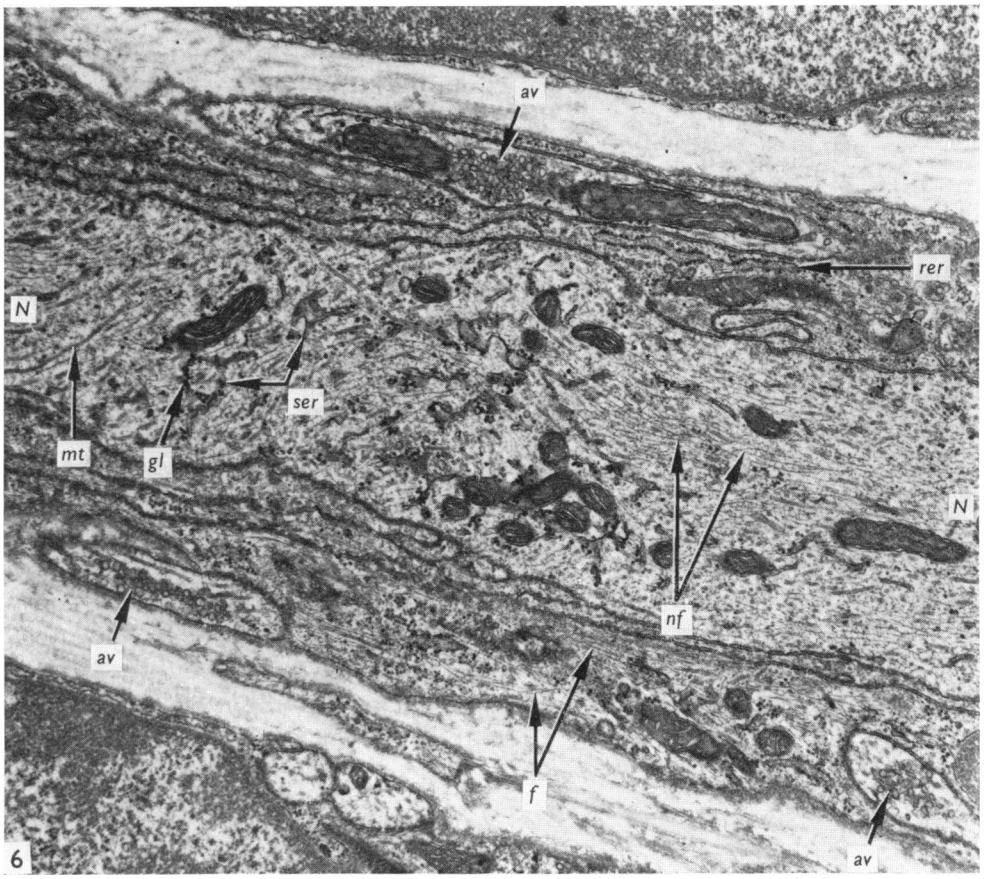


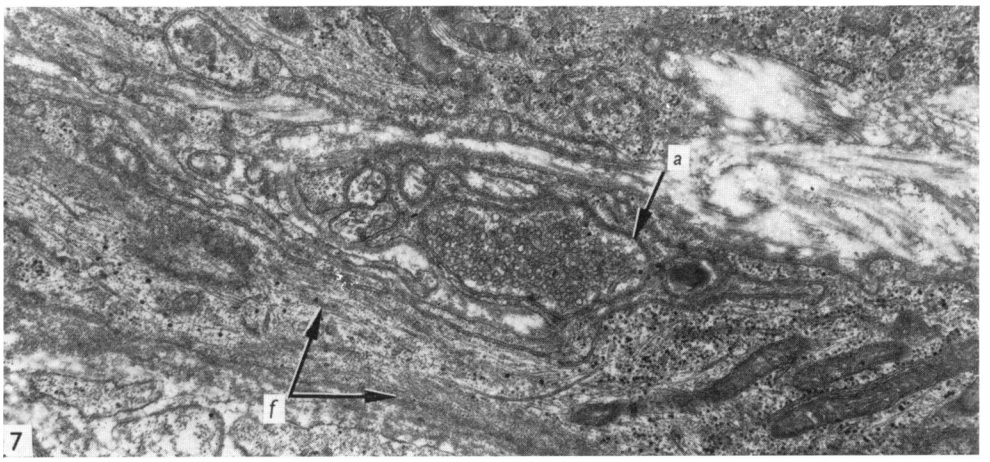


Fig. 6. Longitudinal section of a thick nerve fibre (*N–N*) surrounded by Schwann cell extensions shortly before the entrance into the end-organ. The thick nerve fibre contains mitochondria, fragments of smooth endoplasmic reticulum (*ser*), microtubules (*mt*), filaments (*nf*), and glycogen particles (*gl*). Thin axon profiles containing clusters of small agranular vesicles (*av*) are located in the Schwann cells surrounding the thick fibre. *rer*, rough endoplasmic reticulum; *f*, filaments of Schwann cells. × 20000.

Fig. 7. An axon profile (*a*) containing an accumulation of small agranular vesicles located among Schwann cells containing bundles of filaments (*f*) in the periphery of an end-organ. × 19000.

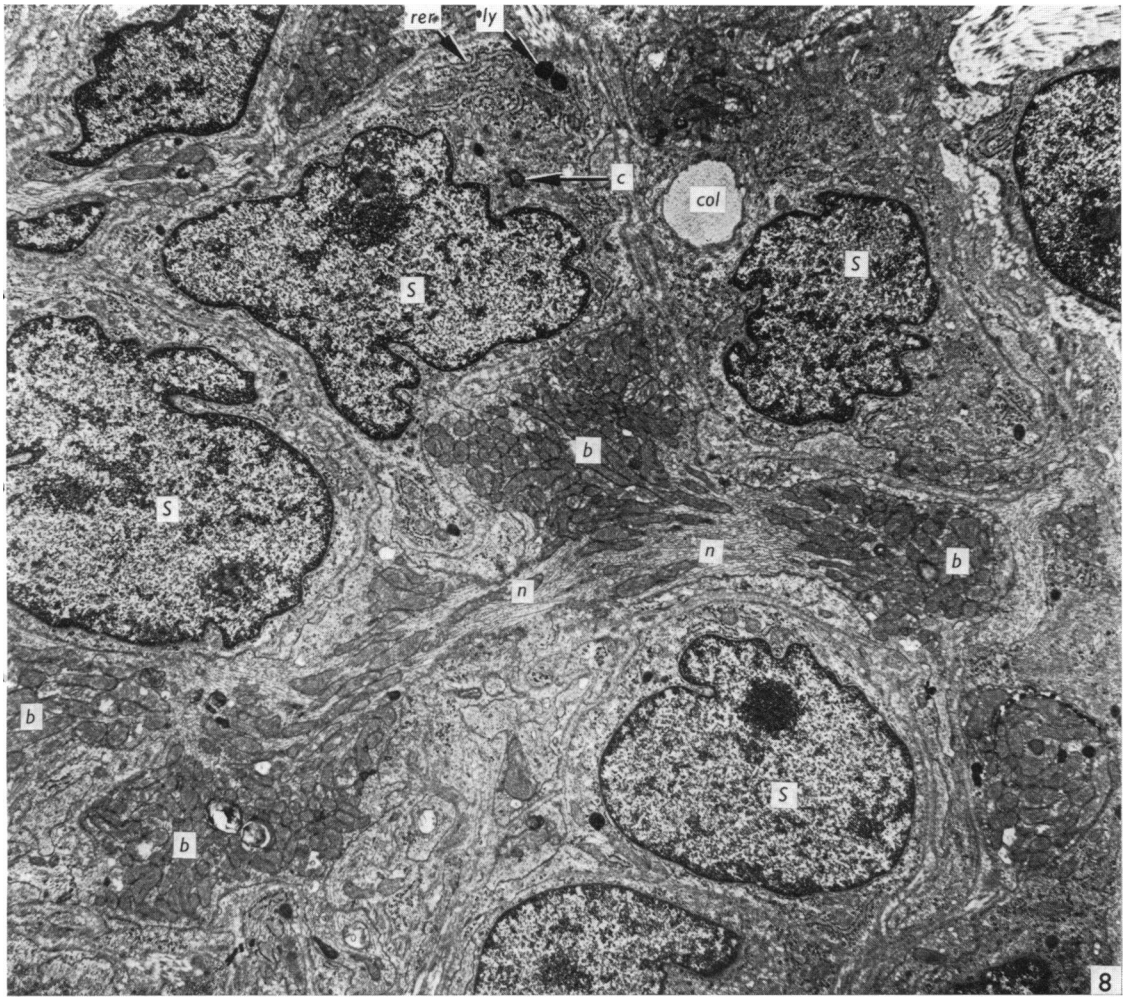


Fig. 8. Low power magnification from the centre of the compact end-organ shown in Fig. 1. A nerve fibre (*n*) is giving off buds or branches (*b*) containing numerous mitochondria. *S*, nuclei of Schwann cells of the end-organ; *rer*, rough endoplasmic reticulum; *c*, centriole; *ly*, lysosome-like body of a Schwann cell; *col*, collagen. Bismuth subnitrate stain. × 6800.

variations in the content of connective tissue fibrils, no fundamental differences were found between the end-organs examined.

The nervous component

The last portion of the thick fibre, just before it entered the end-organ, was always found to be unmyelinated, being surrounded by only a few layers of Schwann cell extensions (Fig. 6). At some distance from the organ the nerve was often furnished with a myelin sheath, ending in a 'half' node of Ranvier, with small desmosome-like junctions with the Schwann cells around the unmyelinated continuation of the fibre, and with specialized junctions between the axolemma and the myelin-

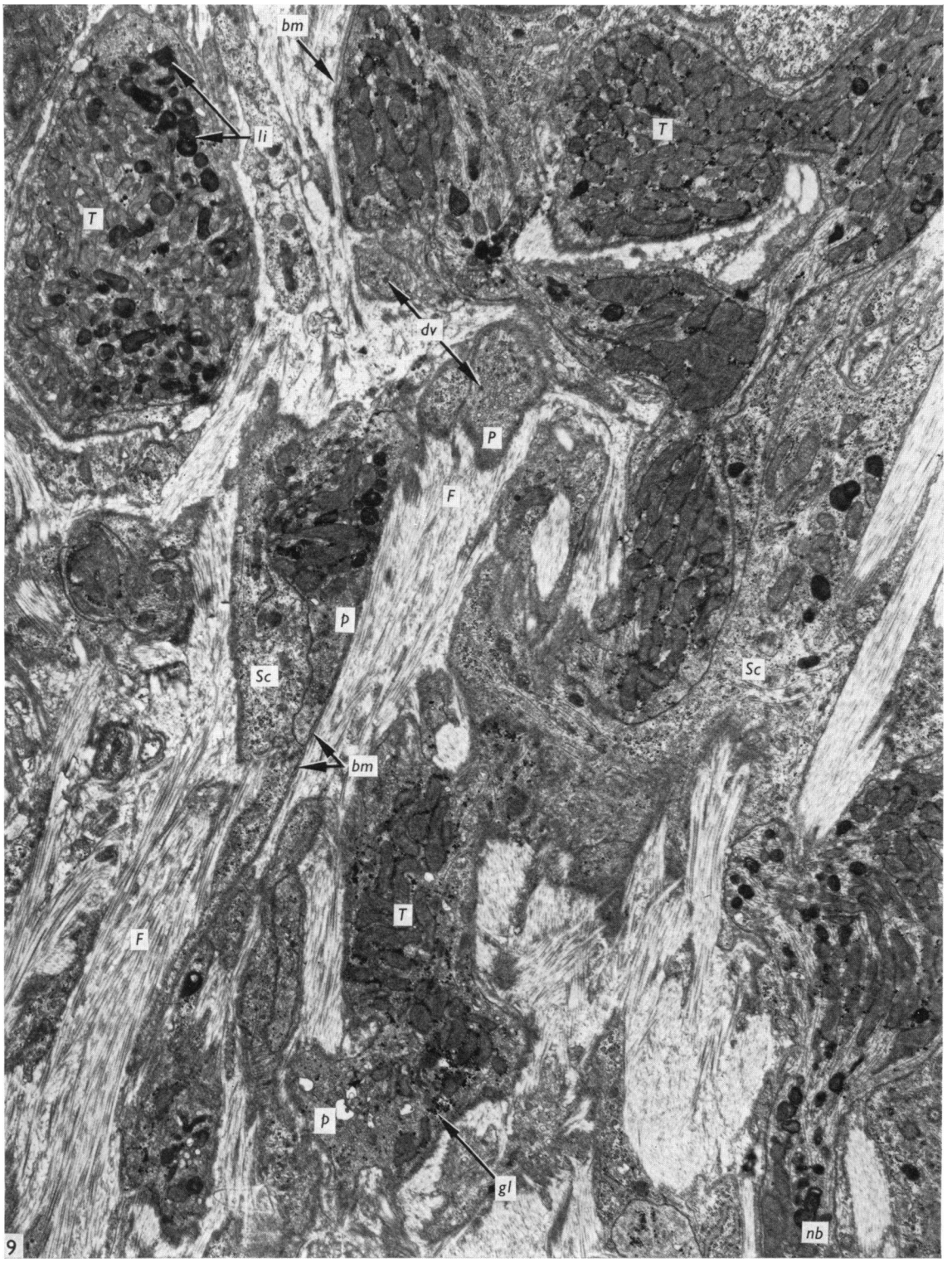
forming Schwann cell in the paranodal region (Figs. 5*a*, *b*). The nerve fibre contained microtubules, neurofilaments and oblong or round mitochondria (Figs. 5*a*, 6). Cisternae of smooth endoplasmic reticulum (SER) with a moderately electron-dense content were found throughout the nerve fibre and were often associated with accumulations of glycogen particles (Figs. 6, 13). The SER usually occurred in small irregular profiles, but sometimes the cisternae were arranged in more organized structures (Fig. 15), which will be described later. Granular endoplasmic reticulum was not seen, nor were free ribosomes, but the large number of glycogen granules made it difficult to be sure. Electron-dense, often laminated bodies resembling lipofuscin granules (Fig. 5*a*), were frequently found, as well as a few, scattered, small, clear vesicles.

The thick nerve fibres were regularly attended by a number of thin fibres containing accumulations of uniform, small, agranular vesicles. At some distance from the end-organ, the thin fibres were seen as small independent nerves running alongside the thick fibre (Fig. 5). Near the end-organ, thin fibres of this kind were found in a number of cases in the Schwann cell sheath around the thick fibre (Fig. 6), which they followed to the point where it entered the end-organ. Small nerve profiles containing similar vesicular accumulations were found scattered in the end-organs, mainly in their peripheral areas (Fig. 7). It was not clear whether these nerves were, or were not, in continuity with the thin nerve fibres outside the organ.

Upon entering the end-organ the thick nerve fibre began to give off lateral branches. With repeated branching a large number of terminals arose, which ramified throughout the organ (Figs. 8, 9).

The most obvious morphological feature of the terminals was their large content of mitochondria of different shapes and sizes. The smallest diameter of the mitochondrial profiles was relatively constant (around 0·2 μm) but the lengths varied considerably, though profiles longer than 1 μm were seldom seen. Cristae were foliate as a rule and their orientation varied (Figs. 9, 16, 17). In the cytoplasm between the mitochondria, only a few microtubules (Figs. 10, 21) and a few filaments were seen. The number of microtubules seemed to fall abruptly at the junction of the nerve branch and the terminal swelling (Figs. 9, 14). A varying number of small, agranular, rather clear vesicles were scattered within the terminals (Figs. 9, 10). Their size varied considerably (30–100 nm) and small particles were often seen on their inner surface. Near the ends of the terminals there were often protrusions of mitochondrion-free cytoplasm, which sometimes contained many such vesicles (Figs. 9, 10). There was an occasional occurrence of solitary, large, uniform (70–80 nm) dense-cored vesicles (Figs. 9, 11, 21). Glycogen granules were found in nearly all terminals (Fig. 9), sometimes in very large numbers (Figs. 12, 14). Granules were about 22·5 nm in diameter. With section thicknesses around

Fig. 9. Survey of an area of an end-organ with abundant terminals (*T*) partially surrounded by cytoplasm of Schwann cells (*Sc*). At lower right a pre-terminal nerve branch (*nb*) is seen. A great part of the surface of the terminals is in direct contact with the connective tissue fibrils (*F*). The basement membrane covering of the terminals (*bm*) is seen to be continuous with that of the Schwann cells. The terminals contain numerous mitochondria, a varying number of lipofuscin-like bodies (*li*), glycogen granules (*gl*), and small clear vesicles, which are especially numerous in the protrusions (*P*) arising from the terminals. In addition, a few large dense-cored vesicles (*dv*) are seen. ×9600.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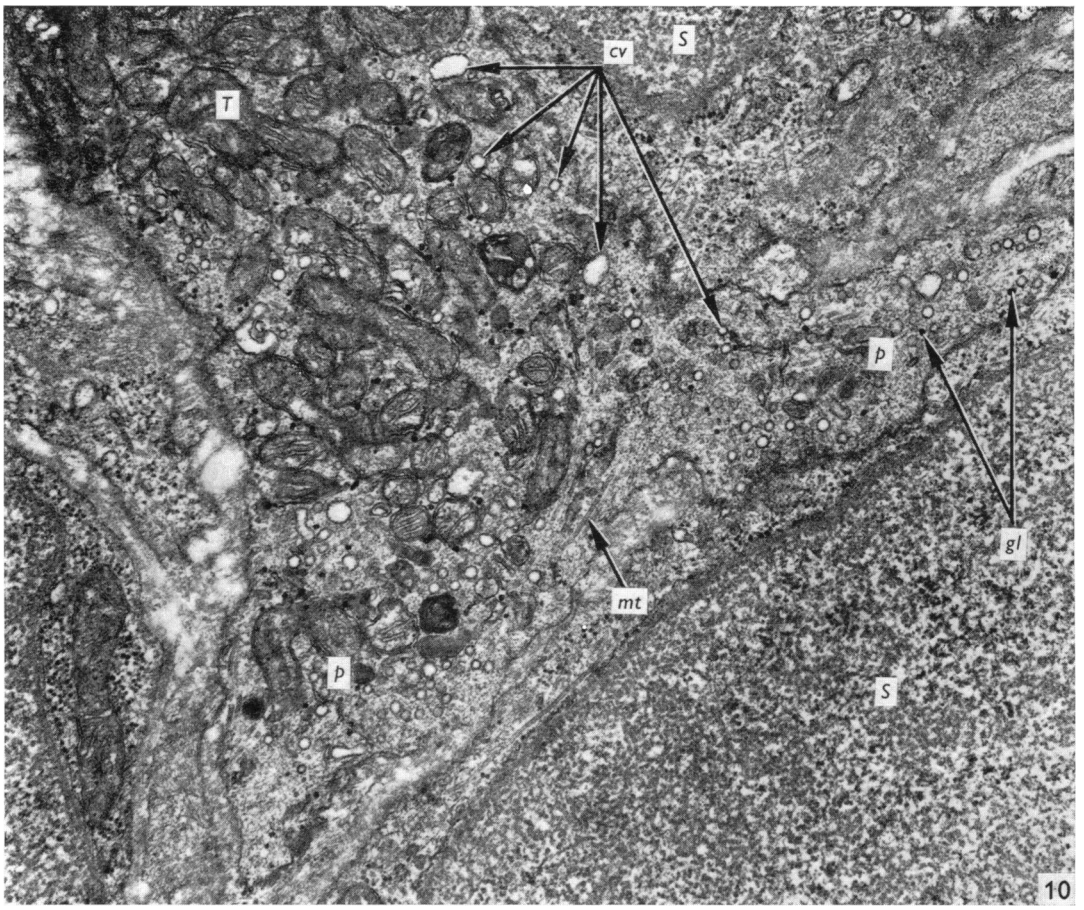


Fig. 10. A terminal (*T*) with protrusions (*p*) containing clear vesicles (*cv*) of varying size and form. Particulate material is seen adhering to the inner surface of several of the vesicles. *mt*, microtubule; *gl*, glycogen particles; *S*, nuclei of Schwann cells. ×25700.

30 nm, the majority of granules were only lightly stained (Figs. 11, 17), probably because the glycogen had been eluted from the sections during processing. With greater section thicknesses (around 70 nm), glycogen granules were often seen in small rosette-like accumulations (Fig. 12). Accumulations of glycogen granules were often found in relation to SER cisterns containing a moderately electron-dense material. The cisterns usually occurred singly or in small irregular groups (Fig. 13). There seemed, however, to be a gradual transition to organized glycogen–cistern complexes, where the cisterns were arranged in regular parallel or concentric systems with numerous glycogen granules in between (Figs. 14, 15). Communications between the cisterns were often seen in such complexes (Fig. 15). These organized glycogen–cistern complexes were observed both in the terminals (Fig. 14) and in the thick fibre at its junction with the end-organ (Fig. 15). In most sections the complexes were seen as regular rows of glycogen particles separated by bands of

homogeneous material, with a poorly-resolved membranous boundary (Fig. 14*a*). However, when sections of this kind were tilted in the electron microscope, the membranes could be clearly demonstrated (Fig. 15). Large complexes were sometimes observed in connection with areas of almost homogeneous, moderately dense structure (Fig. 16). Tilting showed these areas to be built up of numerous, closely packed, small cisterns without intervening glycogen particles (Fig. 17). Highly organized complexes were found in four of the twelve end-organs investigated; complexes with lower organization were found in them all.

A broad spectrum of cyto-segresomes and lipofuscin-like bodies, which seemed to represent different stages of a degradation process, were found widely distributed both in the terminals and in the branches (Fig. 9) and main stem of the nerve. It was the exception to find terminals without such bodies. The earliest stages were represented by vacuoles containing cell structures of normal appearance – as a rule mitochondria and small fragments of SER, occasionally also glycogen particles (Figs. 12, 18, 21). The boundary of such a vacuole could sometimes be resolved into two closely applied membranes. In later stages, the contents of the vacuoles were sequestrated, their density increased, and they were transformed into electron-dense, often partly concentric, laminated inclusions (Figs. 18, 21). The cyto-segresomes seemed, especially in the early stages, to be capable of fusing to form conglomerates. Older cyto-segresomes were often enclosed in new ones (Fig. 19). The number of cyto-segresomes varied from terminal to terminal (Fig. 9); sometimes it appeared as if complete terminals were disintegrating (Figs. 18, 19). Terminals of this kind could often be found adjacent to terminals with only a few inclusions. A special search through several hundred sections provided no conclusive evidence that the inclusions were extruded from the terminals. Besides these cyto-segresomes there were, occasionally, large (2 μm) bodies composed of numerous regularly arranged and closely applied concentric membranes or cisterns. These bodies could be related to the homogeneous-looking laminated bodies sometimes observed in connection with accumulations of glycogen particles and may perhaps be the remnants of glycogen cistern complexes.

The thick nerve fibre and its branches were embedded in the Schwann cells of the end-organ, but separated from them by a uniform gap of about 20 nm (Figs. 9, 11). A few desmosomes were seen between the nerves and the Schwann cells. Large parts of the surfaces of the terminals were not covered by Schwann cells, and some terminals were quite free (Figs. 9, 12, 14). Such naked surfaces were equipped with a basement membrane (Figs. 9, 16, 21), which was continuous with the basement membrane on the Schwann cells. In places the nerve terminals had no basement membrane, particularly where they lay close to connective tissue fibrils.

The cells of the end-organ

The Schwann cells of the end-organ were irregular in shape, with processes (Figs. 8, 9, 14). They were covered by a basement membrane which appeared to be missing for short distances here and there. The basement membrane did not extend into the narrow (20 nm) gap between Schwann cells and nerves. The nucleus was often irregular, with deep indentations (Figs. 8, 14). The cytoplasm was rather sparse and contained only a few mitochondria. Apart from rare desmosome-like

junctions with the neural structures, no plasma membrane specializations were found. Cisterns of granular endoplasmic reticulum with a moderately electron-dense content were scattered throughout the cytoplasm (Figs. 8, 14). Free ribosomes were numerous and were often arranged in polyribosomes (Fig. 11). A Golgi complex (sometimes several) was seen in most cells (Fig. 11). Centrioles were also found regularly (Figs. 8, 11), and a solitary cilium was sometimes seen. Microtubules and thin filaments were seen in all cells (Figs. 10, 11), the filaments sometimes collected into small, loose bundles. Electron-dense, homogeneous and membrane-bound bodies resembling lysosomes were frequently seen in the cells (Fig. 8).

At the periphery of the organs the Schwann cells were sometimes elongated, and contained more regularly arranged bundles of thin filaments (Fig. 7).

In the less compact end-organs, which contained larger amounts of connective tissue fibrils, a few fibroblasts with long processes were found which had no particular relationship with the nervous structures.

DISCUSSION

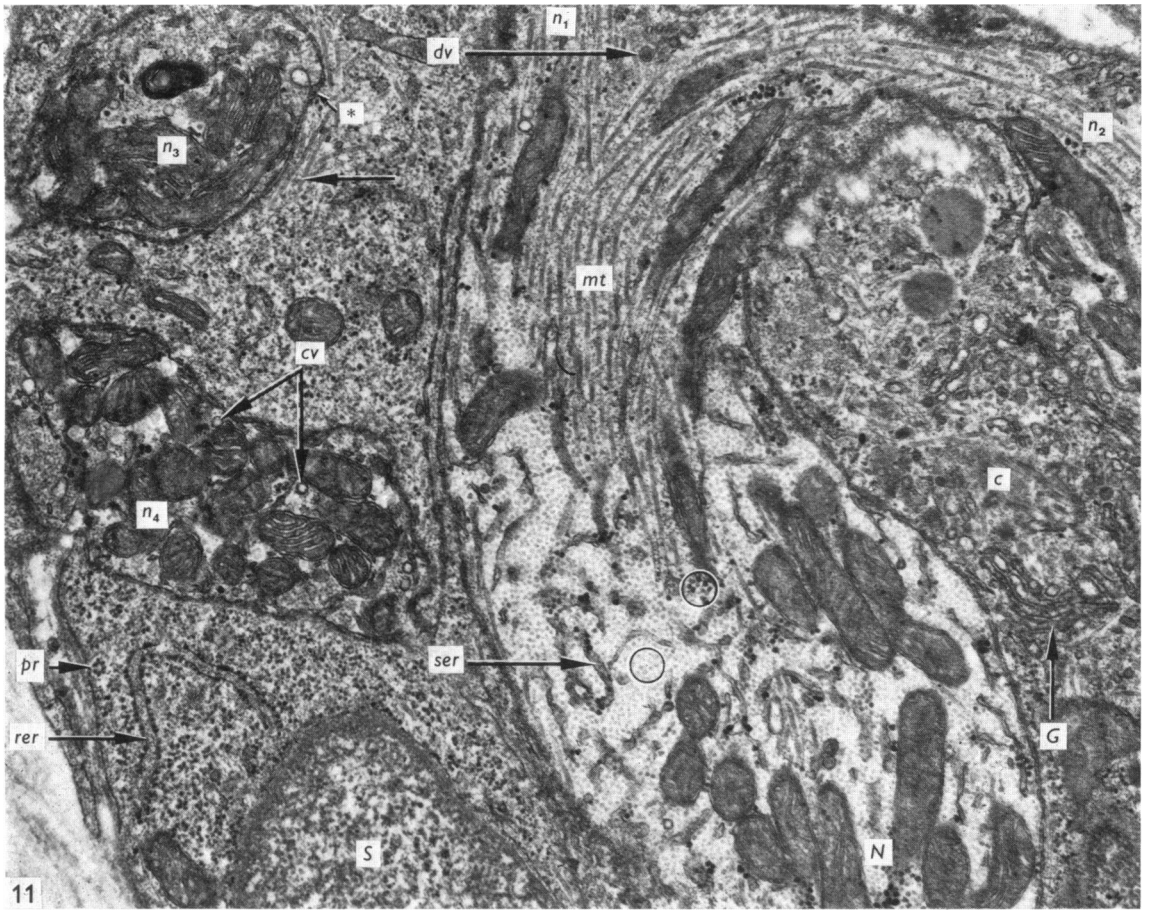
In the present investigation, electron microscopy has confirmed that the atrial endocardium contains unencapsulated nervous end-organs. The presence of these organs has long been recognized with the light microscope (Smirnow, 1895; Dogiel, 1898; Holmes, 1957; Miller & Kasahara, 1964). Their disposition in the atria corresponds to the areas where physiological experiments have revealed the presence of distinct receptors, sensitive to punctate pressure stimuli (Coleridge *et al.* 1957; Coleridge *et al.* 1964; Kappagoda *et al.* 1972) and it is now generally accepted that the circumscribed unencapsulated nerve formations represent end-organs of the thick, rapid conducting, afferent vagus fibres discharging regularly in time with the cardiac rhythm.

The main cells of the end-organs seemed to be Schwann cells structurally modified to ensheath a dense nervous arborization, and appeared similar to the 'spezifischen Terminalzellen' (Knoche & Schmitt, 1964) described around the sensory nerves of the carotid sinus. No specialization of the cells, which might suggest a primary receptor role for these cells, could be demonstrated. They were like Schwann cells also in being separated from the nervous structures by a uniform gap. There were no structures of a synaptic nature, such as vesicular accumulations in parts of the

Fig. 11. A large nerve branch (*N*) surrounded by Schwann cells of an end-organ is dividing into two branches (n_1 and n_2). The large branch contains numerous glycogen granules (circles) most of which appear only faintly stained in this thin (30 nm) section. The glycogen granules surround cisterns of smooth endoplasmic reticulum (*ser*) containing a moderately dense material. Two other nerve profiles (r_3 and n_4) are seen. The Schwann cell cytoplasm contains numerous ribosomes which in some places form polyribosomes (*pr*) or are attached to rough endoplasmic reticulum (*rer*). *c*, centriole surrounded by a large Golgi complex (*G*); *S*, nucleus of Schwann cell; *mt*, microtubules in nerve and in Schwann cell (unlabelled arrow); *dv*, dense-cored vesicle; *cv*, clear vesicles. A structure resembling a newly formed pinocytotic vesicle is seen at asterisk. × 26800.

Fig. 12. 70 nm section demonstrating rosette arrangement of glycogen particles (*gl*) in a small terminal. Lipofuscin-like dense bodies (*li*) containing glycogen particles are seen in a neighbouring terminal. × 25200.

Fig. 13. Section of the thick nerve fibre near the entrance into an end-organ showing irregular complexes of SER cisternae and glycogen particles (*gcc*). *mvb*, multivesicular body. × 22500.



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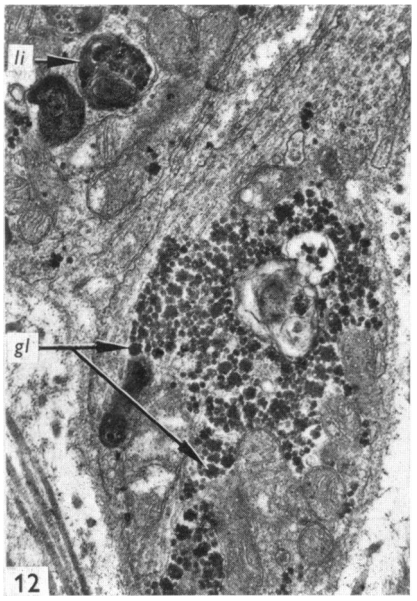

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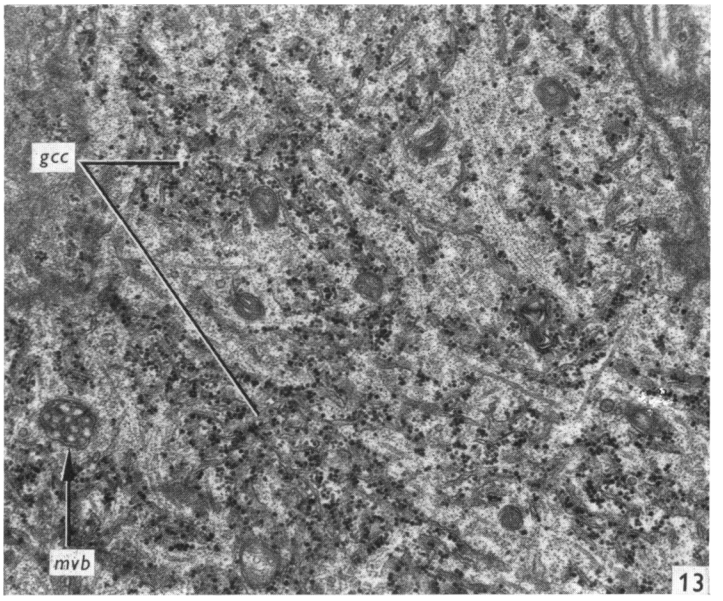

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cells close to the nerves, or nexus junctions. The occasional small desmosomes were not considered to have any significance in this respect.

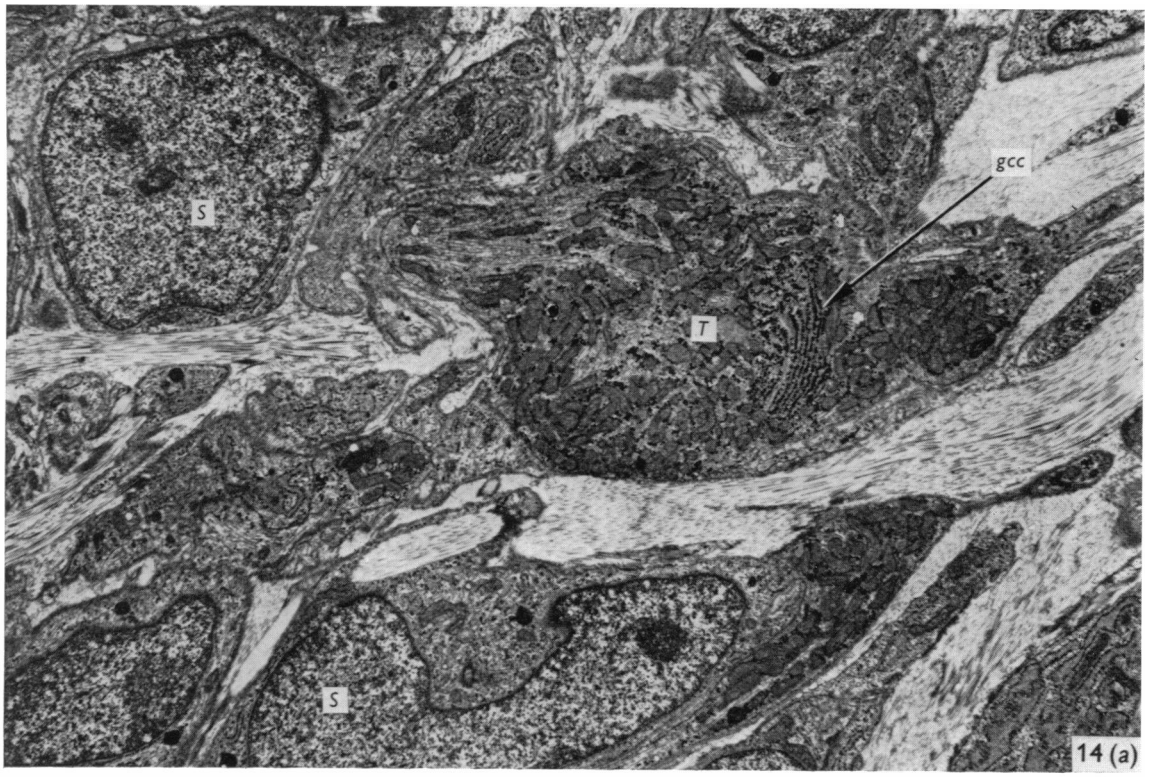
It must be assumed that the mitochondrion-rich terminal arborizations of the end-organ belong to its afferent nervous receptor. It is interesting that the end-organ also receives fine nerve fibres which are possibly autonomic efferent nerves. Their detailed character could not be determined with the aldehyde fixation employed in this investigation (Richardson, 1966). Similar axons of unknown destination have been observed around the afferent fibre to Golgi tendon organs (Schoultz & Swett, 1972).

Afferent nerve terminals in baroreceptors contain a profusion of mitochondria. They share this characteristic with nerve endings in numerous specialized mechanoreceptors, for example, muscle spindles (Merrillees, 1960; Crowe & Ragab, 1970; Landon, 1972), Golgi tendon organs (Schoultz & Swett, 1972), Pacini bodies (Pease & Quilliam, 1957; Nishi, Oura & Pallie, 1969; Spencer & Schaumburg, 1973), Meissner bodies (Cauna & Ross, 1960) and those in the wall of the carotid sinus (Knoche & Schmitt, 1964; Rees, 1967; Chiba, 1972). Nerve endings profuse in mitochondria have also been described as occurring sporadically in tissues containing autonomic nerves, for example, vessel walls (Hagen & Wittkowski, 1969; Burnstock, Gannon & Iwayama, 1970), the vas deferens (Merrillees, 1968), the capsule of the suprarenal gland (Unsicker, 1973) and the myocardium (Chiba & Yamauchi, 1970). It has in some cases been possible to trace such nerve fibres back to myelinated fibres (Hagen & Wittkowski, 1969; Burnstock *et al.* 1970), suggesting that they may be sensory nerves. The present findings support the view that large accumulations of mitochondria are a characteristic feature of a variety of sensory nerve terminals.

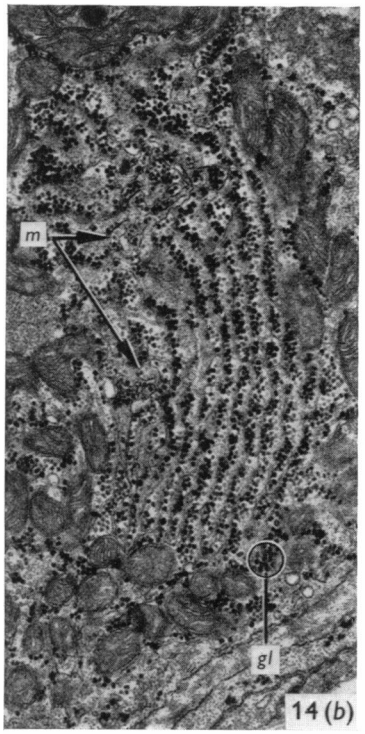
Glycogen granules were a regular occurrence in some of the above-mentioned sensory nerve endings, e.g. in muscle spindles and in the wall of the carotid sinus. An interesting find in the present study was the regular occurrence of glycogen particles associated with cisterns of SER, arranged in regular complexes. These structures bore a striking similarity to the glycogen membrane complexes which have been observed in spinal ganglion cells (Pannese, 1969), and in the paraboloid of certain photoreceptor cells. The smooth cisterns of the paraboloid have been described as deriving from granular endoplasmic reticulum (Yamada, 1960; Petit, 1968). These observations support the idea of a close connection between glycogen metabolism and endoplasmic reticulum such as has been observed, for example, in liver cells (Vrensen & Kuyper, 1969; Vrensen, 1970; Cardell, 1971). Granular endoplasmic reticulum was not found in the nervous structures of the baroreceptors, but in one instance, bodies comprising cisterns like those in the glycogen-cistern complexes of the end-organs were observed in the thick afferent fibre outside the

Fig. 14. (*a*) Low power view (× 6700) of a large terminal (*T*) containing a regular parallel glycogen-cistern complex (*gcc*), shown in detail in (*b*) (× 22 500). *gl*, glycogen particles arranged in rows between cisterns, the membranous boundaries of which are poorly resolved in this projection. At the upper and left margin of the complex, irregularly contoured membranes (*m*) are seen. S, nucleus of Schwann cell.

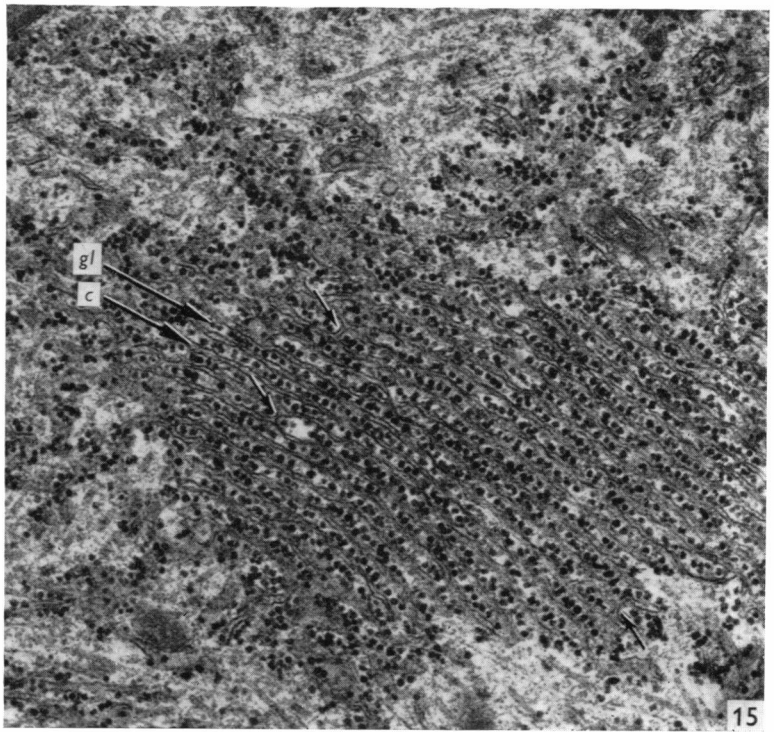
Fig. 15. Glycogen-cistern complex located in the thick nerve fibre at the entrance into an end-organ. The section has been tilted to demonstrate the architecture of the cisterns (*c*) which contain a moderately electron-dense material and in several places intercommunicate (small arrows). Densely stained glycogen particles (*gl*) are arranged in rows between the cisterns. × 36 000.



14 (*a*)



14 (*b*)



15

end-organ (Figs. 5*a*, 5*b*). It is possible that these bodies represent inactive, not-yet-glycogenic complexes undergoing transport to the end-organ. The significance of the regular presence of large amounts of glycogen in the mitochondrion-rich nerve terminals of the baroreceptors is unknown.

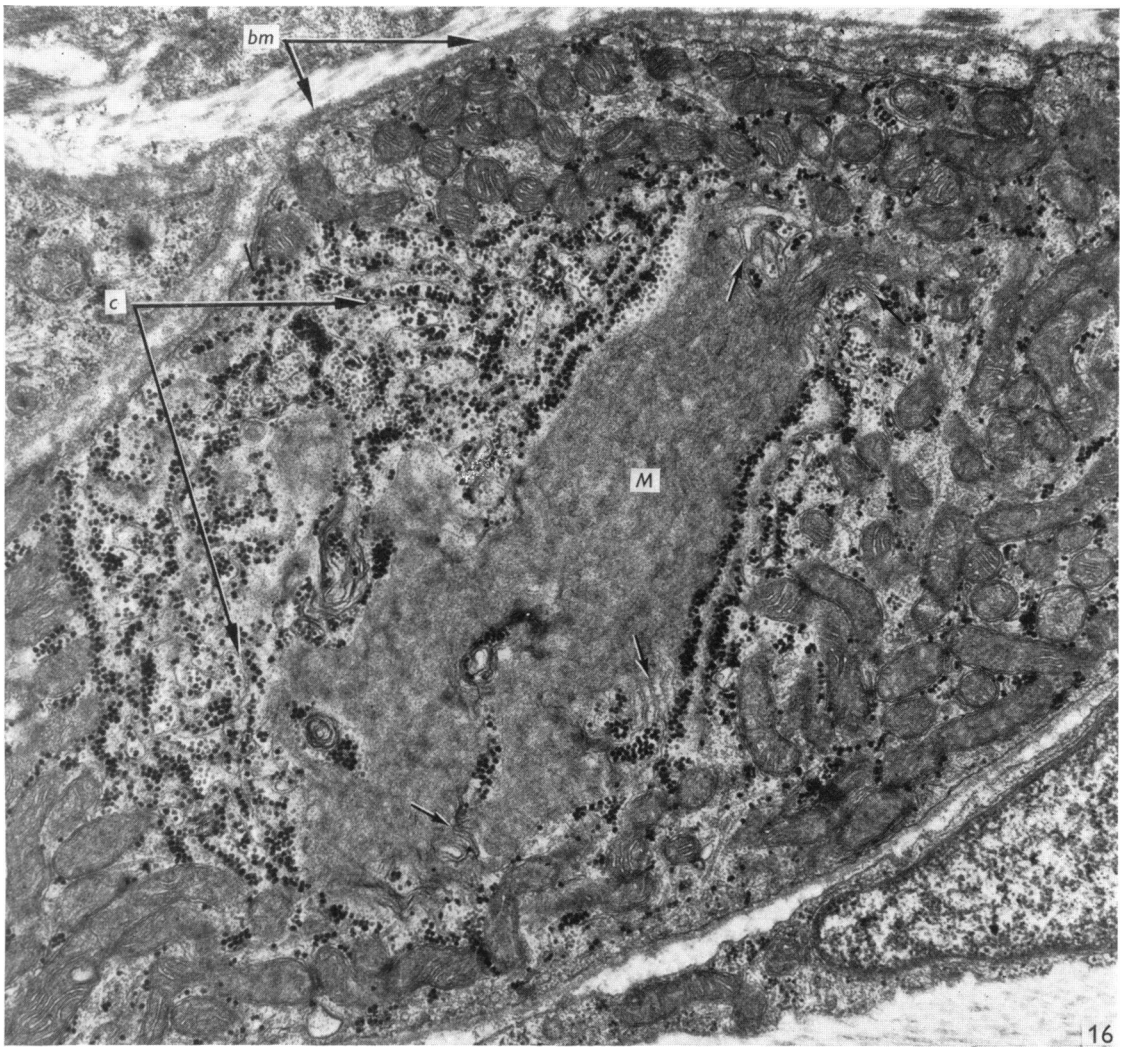
The afferent terminals regularly contained a number of small vesicles. They were often particularly numerous in the mitochondrion-free cytoplasmic protrusions on the surface of the terminals. These vesicles were of varying size, their contents were electron-lucent and their inner surface was often lined with small particles. They thus seemed to differ from the types of vesicles seen in efferent axons. They bore a considerable resemblance to pinocytotic vesicles, but definite indications of pinocytotic activity were not observed in the terminals. Similar vesicles have often been described in sensory terminals, for instance in Golgi tendon organs (Schoultz & Swett, 1972), and in Pacini bodies (Spencer & Schaumburg, 1973).

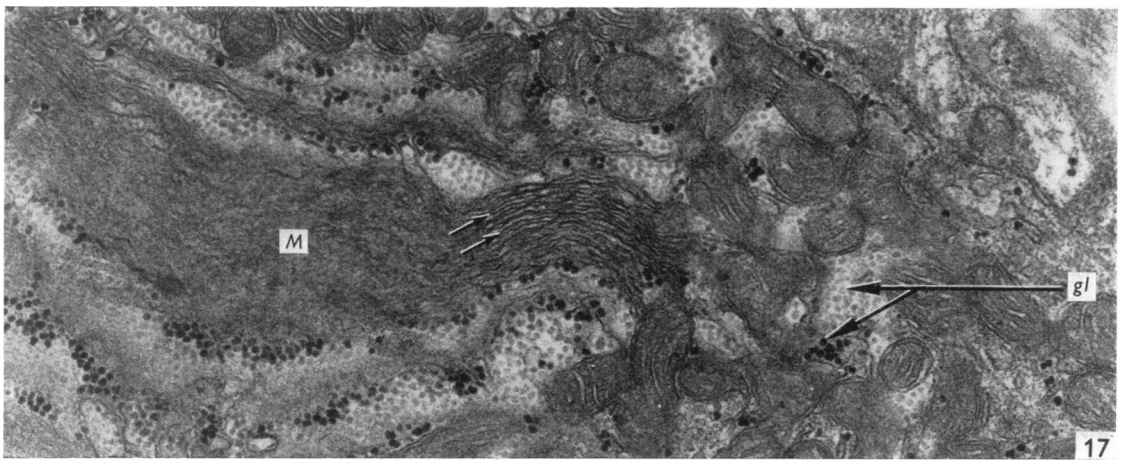
A broad spectrum of cyto-segresomes and lipofuscin-like inclusions was dispersed throughout the terminals of the end-organs. This indicated that a lively breakdown of the cytoplasmic components of the terminals was taking place, leading to the accumulation of lipofuscin-like structures. Lipofuscin inclusions are known to accumulate with age in many neurons (Samorajski, Ordy & Keefe, 1965; Brunk & Ericsson, 1972) but it seems remarkable that they should be so extensive in the end-organs of the very young (7 weeks) animals employed in the present investigation. Similar electron-dense, often laminated, bodies have been described frequently in sensory nerve endings, and it has been suggested (Cauna, 1959) that the neural material of certain receptors undergoes a continuous process of loss and replacement, for instance in the Meissner bodies of the skin (Cauna & Ross, 1960). The present findings strongly support this view. The bi-directional, fast axoplasmic flow, including mitochondria and other structural elements, which occurs in motor neurons, and probably also in sensory nerves (see Wuerker & Kirkpatrick, 1972, for review), should provide a continuous source of new mitochondria for the terminals. It may be that the retrograde component of this flow carries away the lipofuscin-like bodies, since these were often encountered in addition to the mitochondria in the thick afferent fibre.

The high level of cytochrome oxidase activity in the mitochondria of other sensory nerve endings indicates that they are metabolically very active (Hanker, Dixon & Moore, 1973). According to a prevailing hypothesis concerning mechanoreceptor function (Goldman, 1965), mechanical deformation first increases the permeability of the nerve membrane. The restoring processes connected with this may require large amounts of energy and be associated with a rapid turnover of mitochondria. This should apply especially to the baroreceptors of the heart, which fire in every heart cycle. On the other hand it is possible that the turnover is not particularly high, but that slow elimination results in the accumulation of degenerate mitochondria.

Fig. 16. Large terminal containing an irregular body of almost homogeneous structure (*M*) located in an area containing numerous glycogen granules. The body is in continuity with membranous structures in several places (small arrows). Irregular cisternae (*c*) are seen among the surrounding glycogen particles. *bm*, basement membrane of terminal. × 24800.

Fig. 17. Tilting of a body (*M*) similar to that of Fig. 15 suggests a composition of closely apposed narrow cisternae (small arrows). Densely and lightly stained glycogen particles (*gl*) are seen in addition to mitochondria in the surroundings. × 36500.


bm
c
M
16


M
gl
17

SUMMARY

Light and electron microscopical studies on pig hearts have confirmed the presence in the right and the left atrial endocardium of distinct circumscribed, unencapsulated end-organs (baroreceptors) associated with 4–9 μm thick nerve fibres. The myelin sheath terminates before the entrance of the nerve into the end-organ. The regular presence of thin axon profiles containing clusters of small agranular vesicles, both around the thick nerve fibre and in the periphery of the end-organs, suggests a double innervation. The thick fibre arborizes inside an aggregate of Schwann-like cells to form a large number of terminals which are considered to be the mechano-sensitive receptors of the organ. In addition to the nervous structures and the Schwann-like cells the organs contain a varying number of connective tissue fibrils. The terminals are covered by a basement membrane but are partly or completely devoid of Schwann cell covering. Most of the space inside the terminals is occupied by tightly packed mitochondria. Glycogen granules are regularly present, sometimes abundant. SER occurs widely, frequently forming complexes with glycogen granules. These complexes may be highly organized as parallel or concentric structures, suggesting a close connection between SER and glycogen metabolism in the terminals. The terminals are regularly furnished with small protrusions containing numerous 30–100 nm clear vesicles which resemble pinocytotic vesicles. In addition, a few 80 nm dense-cored vesicles are found. The occurrence of numerous cyto-segresomes and lipofuscin-like bodies suggests a lively turnover of organelles in the terminals.

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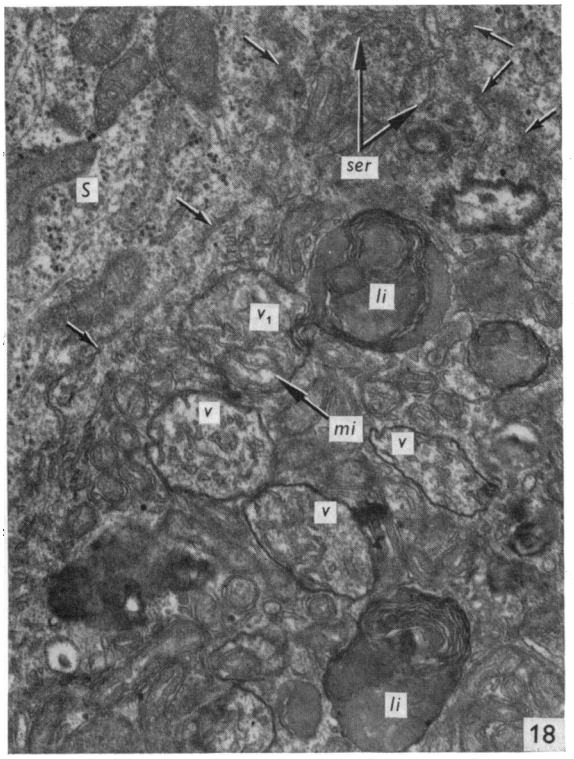
Figs. 18–21. Demonstrate terminals showing various degrees of autophagic activity.

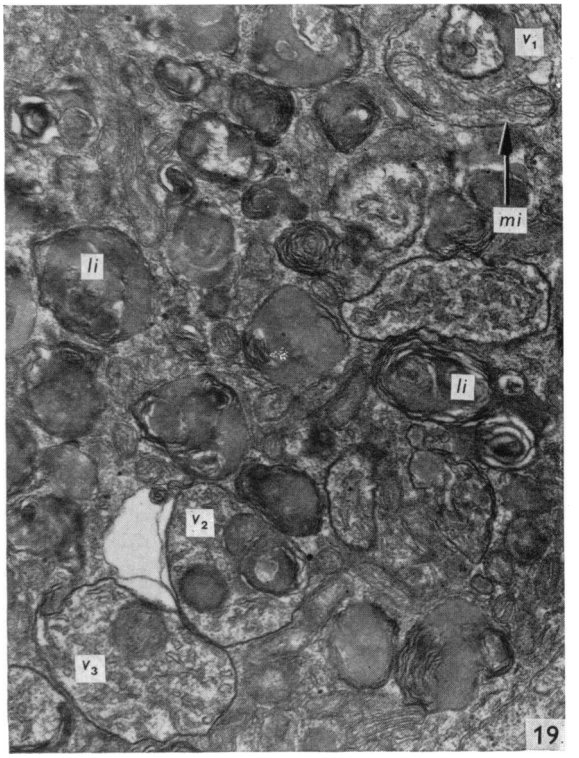
Fig. 18. Part of a terminal adjacent to a Schwann cell (*S*, borders marked with small arrows). Vacuoles (*v*) are seen containing small fragments of SER, the vacuole v_1 in addition contains a partly sequestered mitochondrion (*mi*). *li*, lipofuscin-like bodies; *ser*, SER in the cytoplasm of the terminal. × 27200.

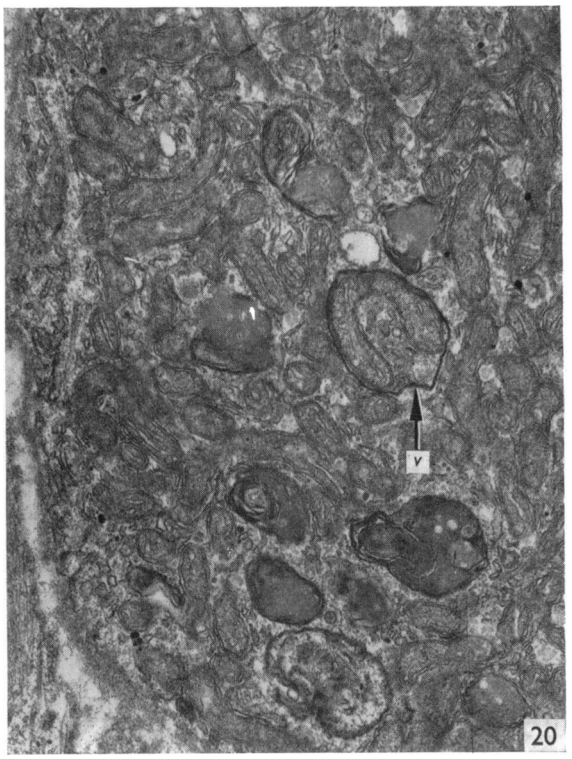
Fig. 19. Part of a terminal which appears degenerate. Several lipofuscin-like bodies (*li*) can be seen among vacuoles, some of which (v_2 and v_3) contain lipofuscin-like bodies. The vacuole v_1 contains a mitochondrion (*mi*). × 24600.

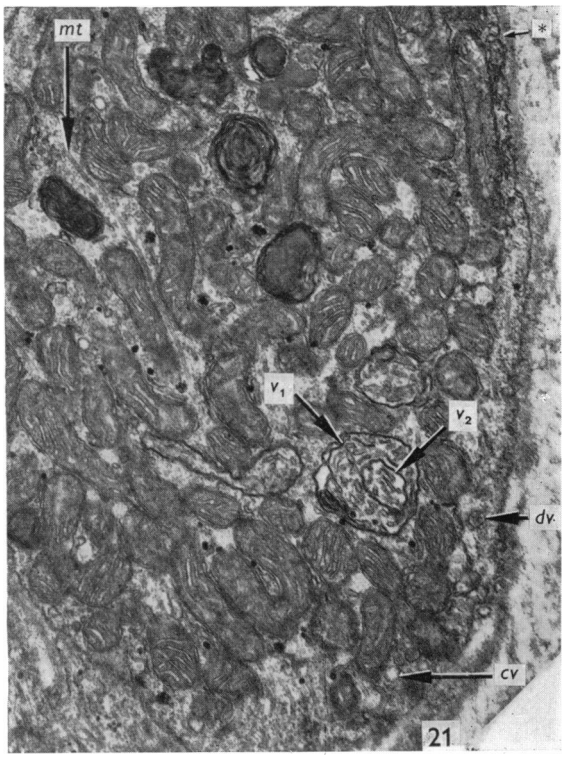
Fig. 20. Part of a terminal showing a vacuole (*v*) containing an almost normally structured mitochondrion. × 28400.

Fig. 21. Part of a terminal whose basement membrane covering is seen to the right. v_1, vacuole which apparently has formed around another vacuole v_2; *mt*, microtubule; *cv*, clear vesicle; *dv*, dense-cored vesicle. A structure resembling a pinocytotic vesicle during formation is seen at asterisk. × 25900.


S
ser
li
v_1
v
mi
v
v
li
18


v_1
mi
li
li
v_2
v_3
19


v
20


mt
*
v_1
v_2
dv
cv
21

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