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THE ULTRASTRUCTURE OF ODONTOBLASTS IN PERFUSION FIXED, DEMINERALISED INCISORS OF ADULT RATS

by

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INTRODUCTION

Several electron microscopic studies of the early stages of dentinogenesis have been published (*Watson & Avery,* 1954; *Nylén & Scott,* 1958; *Lenz,* 1959; *Noble et al.,* 1962). These studies have provided valuable information concerning the cytodifferentiation of odontoblasts and their fibrillogenetic function. Concerning the finer morphology of odontoblasts, however, many questions remain unanswered, because of the limited level of optical resolution previously available. A more detailed cytological description of odontoblasts has recently been published by *Frank* (1966), and provides evidence for the existence of nerve fibers in human dentine.

The present study was performed on fully differentiated odontoblasts from rat incisors. This report presents a description of organelles and inclusions not previously observed in similar material. Fixation of odontoblasts for electron microscopy presents technical problems. Perfusion fixation with glutaraldehyde solves some of these, but limits the choice of material to experimental animals. Rats are well suited to the technique, since a subpredentinal capillary plexus is present in the incisors, in close relationship to the odontoblasts (*Adams,* 1959). Satisfactory perfusion fixation allows demineralisation of the teeth prior to post fixation

in osmium tetroxide. Good preservation of the odontoblasts can be obtained by these means.

MATERIAL AND METHODS

Female albino rats with an average body weight of 250 g were anaesthetised with barbiturate by intraperitoneal injection. The perfusion medium consisted of 2 % glutaraldehyde in a cacodylate buffer at pH 7.4, with 3 % dextran and 3 % glucose (*Rostgaard & Rehnke,* 1966). A hospital type infusion set was used at a pressure equivalent to 4 feet of water. The thorax was opened under artificial respiration and the heart and ascending aorta exposed. The left ventricle was opened, and a blunt infusion cannula passed through the ventricle and into the aorta, where it was fixed in position with a ligature. The right atrium was opened to allow of drainage. Two hundred ml of perfusion medium were infused over a period of 20 minutes.

The mandible was then dissected free and divided in half through the symphysis. The half mandibles were demineralised in a 5 % solution of EDTA in 0.2 molar glucose adjusted to a pH of 7.4. Demineralisation was performed with the tissue suspended in gauze bags, under continuous stirring and exchange of the solution (*Belanger et al.,* 1965). Thereafter the specimens were washed overnight in 0.2 molar tris maleate buffer at pH 7.4. The demineralised incisors were then cut into small blocks by means of parallel sections, at right angles to the long axis of the tooth and 1 mm apart, prior to post fixation in 1 % osmium tetroxide (*Sjöstrand,* 1956) for 2 hours. These tissue specimens were then dehydrated through graded ethanols and embedded in epon (*Luft,* 1961) in flat gun powder wads (LKB Products) from which it could later be cut free and reorientated, allowing of sectioning in the desired plane. Polymerisation was performed at 60°C for 24 hours.

Sections for light microscopical orientation were cut at 1—2 microns and stained with 1 % paraphenylenediamine (*Estable Puig et al.,* 1965) or with toluidine blue. For electron microscopy, thin sections were cut out with glass knives on an LKB-Ultrotome at a feeding of 600—800 Å per section. Sections were stained with both uranyl acetate (*Watson,* 1958) and lead citrate (*Reynolds,* 1963), prior to examination in a Siemens Elmiskop I at 60 Kv,

using a 400 microns aperture in the double condenser and a 50 microns objective aperture.

RESULTS

The following description is imited to the mature odontoblasts from the middle third of the mandibular incisors. These cells are fully differentiated and active in dentinogenesis. The relationship between the odontoblastic processes and dentine is not described, as the preservation of the distal parts of the processes is not adequate.

Under the light microscope, the odontoblasts appear as tall, slender cells arranged in a manner reminiscent of pseudo-stratified cylindrical epithelium (Fig. 1). The cells are polarised, the nucleus being situated near the pulpal end of the cell, and the remaining cell components are asymmetrically distributed. During the course of differentiation the length of the odontoblasts increases considerably, and the mature cells are approximately 50 microns long. The cell breadth at the level of the nucleus is about 6 microns. Distal to the nucleus the breadth varies considerably, the cell frequently being considerably narrowed in the vicinity of the sub-predentinal capillary plexus. This plexus is very well developed in the rat, the vessels being regularly situated about 10 microns from the predentine and the same distance from each other. The capillaries are usually considerably dilated and empty of blood cells as a result of perfusion. Numerous anastomoses unite the sub-predentinal plexus with a less regular and more loosely meshed subodontoblastic plexus. In the area corresponding to the cell free zone of Weil, of human pulp, numerous fibroblasts are frequently seen in rat material. There may in fact be more fibroblasts in this peripheral region of the pulp than in the central zone.

The odontoblast cell body and the odontoblastic process are described separately below. By definition, the boundary between these two components passes through the dentinal extremity of the cell's terminal bars.

The ultrastructure of the cell body

The *plasma membrane* is fairly straight on the lateral and pulpal cell surfaces. Laterally small cytoplasmic processes are occa-

sionally seen, but interdigitations with the plasma membrane of neighbouring cells are infrequent. The cell contact between adjacent odontoblasts is established by means of three different types of surface modifications. The most frequent are *desmosomes*, which are found in large numbers at various sites (fig. 6). These consist of a thickening of the respective plasma membranes with a slight condensation of the underlying cytoplasmic matrix, but without intracytoplasmic filaments. The intercellular space is about 200 Å in width and shows no structure. Sometimes, however, a slight condensation of the ground substance is seen.

In addition *tight junctions* of various length are observed. They are characterized by a very parallel course of the adjacent plasma membranes. The space between these is about 100 Å and is filled by a structureless substance of high density which may indicate a fusion of the two membranes. It has been impossible to obtain resolution of the unit membrane lamina, presumably because the EDTA chelation has had a modifying effect upon these. Desmosomes and tight junctions are also seen along the pulpal surface of the odontoblasts, connecting them with fibroblasts.

At the predentinal border, odontoblasts are attached to each other by structures of the type usually described as *terminal bars* (Fig. 10). These consist of a thickening of the plasma membranes and a considerable condensation of the subjacent cytoplasmic matrix, which appears as a dense plaque. Next to the plaque a massive bundle of filaments runs around the cell as a cuff, concentric with the cell surface and perpendicular to the long axis of the cell. The intercellular space has the same appearance as described under desmosomes. The *filament cuff* has been observed in the absence of the terminal bars, in cases where a broader intercellular space is occupied with Korff fibers. The filament cuff has also occasionally been observed to extend along the plasma membrane into the odontoblastic process.

The *nucleus* of the odontoblast is, as previously mentioned, situated toward the pulpal pole of the cell. It is ellipsoid in shape with a long diameter of about 8 microns. The general features of nuclear content, nucleoli and nuclear envelope do not deviate from the norm for similar cells fixed in glutaraldehyde and osmium tetroxide (Fig. 2).

In sections through the long axis of the nucleus a characteristic

inclusion has frequently been seen (Figs. 2, 3 and 4). This consists of 6—7 filaments placed parallel with each other. The diameter of the filaments is about 75 Å and they are placed about 200 Å from each other (Fig. 4). The total diameter of the inclusion is 1000—1500 Å and the greatest observed length is about 5 microns (Fig. 3). Typically, the inclusion is placed in the nucleus' long axis, frequently in relation to one of the poles but not in direct contact with the nuclear membrane. The filaments often appear to thin out at the ends of the inclusion. More than two inclusions per nucleus have not been observed (Fig. 2). Similar inclusions to those described above have in rare instances also been seen in fibroblasts from the corresponding part of the pulp.

The odontoblasts have a well developed and highly organised rough-surfaced *endoplasmic reticulum* (ER), the membrane system of which is studded with ribosomes (Figs. 6 and 7). In the pulpal end of the cell a few ER-cisterns are seen, and these appear to be concentrically placed with respect to the nucleus. Most of the ER, however, is found on the dentinal side of the nucleus and is arranged in long, parallel cisterns, which often communicate with each other. The distance between the individual cistern membranes is about 400—500 Å. The attached ribosomes have a diameter of about 150—200 Å, and are extraordinarily tightly packed, being placed not more than a few hundred Å from each other. As a characteristic of cells actively engaged in protein synthesis, marked dilatations of the ER-cisterns are often found. The latter occur more frequently near the nucleus than in the rest of the cell. The largest observed dilated cistern measured 2×5 microns. The content of the cistern is finely granulated or homogeneous and has a higher density than the cytoplasmic matrix.

Not infrequently, however, a *linear structure* showing periodicity is observed in dilated ER-cisterns (Fig. 7). The structure is composed of dots arranged in pairs of parallel rows. The dots of two corresponding rows are out of phase with each other and accordingly create a zig-zag appearance, reminiscent of a helix or a paracrystalline structure. The individual dots have a diameter of 50—75 Å. The distance between individual dots of a single row is 200 Å, and the distance between two rows of a pair is also 200 Å.

A moderate number of *free ribosomes* are seen in the cytoplasmic matrix in the cell body. They have the same diameter as the above mentioned membrane bound ribosomes and are frequently associated in groups of 3—7.

Mitochondria are found in all regions of the cytoplasm (Figs. 5 and 7). Sometimes they appear to be more frequent in the distal region of the cell body. The mitochondria are elongated and have a regular contour. They are often placed with their long axis parallel to the ER-cisterns and in close relationship with these. Very regular and closely packed transverse cristae extend almost the entire breadth of the mitochondria. The mitochondrial matrix has medium density but lacks granules.

A well developed *Golgi apparatus,* comprising the usual components: smooth paired membranes, vacuoles and vesicles, occupies the supranuclear region of the odontoblast (Figs. 6 and 8). The vacuoles have a variable diameter, often about 0.5 microns, and they appear to be formed by a partial dilatation of the smooth paired membranes. The content of the vacuoles is finely granulated and has a low density. The vesicles are especially numerous and have a diameter of about 500 Å. Their content often has a somewhat higher density than that of the vacuoles.

In the same vicinity of the cell a variable quantity (1—10) of *round bodies* are seen. They have an average diameter of 0.5 microns and are reminiscent of lysosomes. They are membrane bound and have a very variable content. Most frequently it consists of a homogeneous, very dense matrix (Figs. 6 and 8). Sometimes the content consists of droplets of variable size and density (Fig. 16). Exceptionally, large lysosome-like bodies, containing complicated membrane systems arranged in so called myelin figures, are seen (Fig. 17).

Also in the same area of the cell are found a few *multivesicular bodies* (Figs. 6 and 8). They contain uniform, light vesicles with a diameter of 500 Å, embedded in a dense matrix. The diameter of the multivesicular bodies is variable, frequently about 0.6 microns.

Finally, in the Golgi zone, very dense *rod shaped bodies,* round in cross section and with a breadth of 0.1 microns, are observed. Their length is very inconstant. They also occur in large numbers in the odontoblastic process.

In topographic relationship to the Golgi zone a pair of *centrioles* is found (Fig. 6). Frequently one of the centrioles appears to represent the basal body of a *cilium,* that reaches out to the intercellular space through a narrow invagination of the plasma membrane (Figs. 8, 9 and 6). Cilia have not been observed in other situations, and not more than one cilium per odontoblast has been found. The cilium has the typical 9+0 structure of stereocilia.

Microtubules are found in large numbers in the odontoblast (Figs. 5 and 6). They are circular in transverse section, with an outer dense wall and a less dense interior. They measure 200—250 Å in diameter and their length is undetermined, but in favourable sections they can be followed for a distance of several microns. Their presence in the central portion of the cell body is to a certain extent masked by other organelles, but in the marginal region of the cell they appear very distinct and run an almost straight course parallel with the cell's long axis (Fig. 5).

At the periphery of the cell a number of *coated vesicles* may be seen (Figs. 6 and 8). However, since the majority of these structures are found in the odontoblastic processes, they will be described in detail in the appropriate section.

The ultrastructure of the odontoblastic process

The process is a direct continuation of the odontoblast, as both the plasma membrane and the cytoplasm extend from the cell body into the process. However, many of the organelles described as being present in the cell body are lacking in the process. Thus no ribosomes or ER are found, and *mitochondria* are extremely rare, though they have been observed in the process both at the predentinal and dentinal level (Fig. 15).

Numerous *microtubules* are seen in the processes and their small side branches (Figs. 11, 12, 13 and 14). They have the same appearance and dimensions as those seen in the cell body. The microtubules occur evenly distributed over the entire cross section of the process and almost always have a straight course, parallel with the long axis of the process. They are always unbranched and do not appear to have any connection with other organelles of the process.

A number of *filaments* are also seen in the process (Figs. 12

and 14). These are 50—75 Å in diameter and have an orientation and distribution similar to that of the microtubules.

Another characteristic organelle in the process is the *rod shaped dense body,* already mentioned in connection with the Golgi zone (Figs. 11 and 13). In the process they have a more regular size with a length of about 0.5 microns and a diameter of about 0.2 microns. The great majority are rod shaped, although occasionally kidney shapes or round forms are seen. In favourable sections they are seen to be surrounded by a membrane, which is separated from the content by an electron lucid zone measuring 150 Å. The contents are very dense and usually homogeneous.

The margins of the process show numerous *coated vesicles.* These are about 1500 Å in diameter, and are characterised by small spines, about 200 Å in length, arranged perpendicularly on the cytoplasmic side of the vesicle's surface (Figs. 11 and 13). The coated vesicles have a floccular content, of rather low density, most frequently distributed near the margin. Since the vesiclar membrane is often seen to be continuous with the plasma membrane, it seems likely, that the vesicles develop from invaginations, which subsequently become pinched off from the plasma membrane (Fig. 13). An alternative interpretation is that the vesicles empty their content on the cell surface.

Finally, many *microvesicles* of the type seen in the Golgi apparatus, are also observed in the process (Fig. 12). They are usually situated near the plasma membrane. The vesicles are often found in groups.

The number of the above mentioned organelles decreases considerably at the level of the predentine-dentine junction. In the peripheral region of the process the only organelle constantly present, is the microtubule.

The extracellular space

The extracellular space between the cell bodies of the odontoblasts varies considerably in breadth. It is at a minimum at the level of the nucleus and greatest distal to the subpredentinal capillary plexus. The ground substance is amorphous or finely granulated and contains very few collagen fibrils (Fig. 6). The capillaries are surrounded by a basal membrane. Nerve fibers have not been observed.

The matrix of the predentine consists of a network of collagen fibrils with typical periodicity, embedded in an amorphous ground substance of very slight density (Fig. 11). The vast majority of the fibrils have a course which runs at right angles to the odontoblastic process. A fibril-free zone, about 2—3 microns in breadth, is usually present in the region adjacent to the pulp. The finest fibrils have a diameter of about 200 Å. The diameter increases to 600—700 Å peripherally in the predentine. Betweeen the collagen fibrils numerous side branches of the odontoblastic processes are seen (Fig. 11). The side branches have the same organelle content as the main processes and do not appear to have any modifying effect upon the orientation of the surrounding collagen fibrils.

DISCUSSION

The results of the present study confirm previous observations concerning the appearance and situation within the cell of the classical cell organelles: nucleus, mitochondria, ER and Golgi apparatus (*Watson & Avery,* 1954; *Nylen & Scott,* 1958; *Lenz,* 1959; *Noble et al.,* 1962; *Frank,* 1966). However, perfusion fixation with glutaraldehyde gives improved preservation and it is thus possible to observe organelles and inclusions which have not previously been described in odontoblasts.

After the introduction of glutaraldehyde, microtubules have been observed in many cells of various origins (*Behnke,* 1964; *De-Thé,* 1964; *Sandborn et al.,* 1964). Virtually nothing is known about their function. It has been suggested that they may play a role in cytoplasmic streaming (*Ledbetter & Porter,* 1963) or act as a cyto-skeleton (*Porter et al.,* 1964; *Behnke,* 1965). Their characteristic orientation within the odontoblast, and especially in the process, where they are reminiscent of the neurotubules within axons, make the above mentioned suggestions feasible. They may function both in providing structural support to the odontoblastic process during dentinogenesis and in directing intracytoplasmic flow.

A body of unknown identity has previously been described in the Golgi zone of the odontoblast (*Nylen & Scott,* 1958). In the present study lysosome-like bodies are seen in far greater numbers and not only in the Golgi zone, though they are most numerous here. Studies are in preparation concerning their cyto-

chemical nature. Estimated on morphological basis alone, they are supposed to belong to the group of lysosomes classified as "storage granules" (*de Duve,* 1963), since they rarely contain autophagocytosed organelles.

The genesis of cilia in fibroblasts from the small intestine from rats and chickens have been described by *Sorokin* (1962). Recently, cilia have been observed in fibroblasts in the dental pulp of the Guinea pig (*Han et al.,* 1965). They occur also in rat fibroblasts in the present material. In addition, the odontoblast appears to have retained the ability to develop a single, atypically placed cilium. The cilium is not supposed to have any mechanical or sensory function. It probably represents an evolutionary reminent.

The uniform perpendicular orientation of the predentinal collagen fibrils in relation to the odontoblastic processes and the increasing diameter of the fibrils in pulpo-dentinal direction, indicates that the odontoblasts both influence the spatial arrangement of the fibrils during their formation and secrete tropocollagen. Korff fibers, traditionally presumed to be of significance in early dentinogenesis (*Nylen & Scott,* 1958) are rarely observed in the present material. In agreement with this observation, the rough surfaced endoplasmic reticulum of the odontoblast is very highly differentiated, with a large number of dilated cisterns. The paracrystalline material which often appears in these may represent some precursor of collagen precipitated by fixation. The synthesis of collagen by odontoblasts has been demonstrated by means of autoradiography at the light microscopical level by *Caneiro & Leblond* (1959). They found label within the Golgi zone at 30 minutes after injection of tritiated glycine and in the predentinal matrix at 4 hours. The manner in which tropocollagen is transported through the cell to the process, and secreted from the latter, can only be elucidated by autoradiography at the electron microscopic level. However, the presence of numerous microvesicles in the marginal region of the predentinal part of the process, and the similarity of these vesicles with those seen in the Golgi apparatus, suggests a possible mechanism. Vacuoles of the order of size 0.4—2 microns, described by *Frank* (1966) and suggested as secretion vacuoles in odontoblasts from human material, have not been observed in the present study.

The function of the coated vesicles in the odontoblastic process is unknown. They are not supposed to be concerned with the secretion of collagen, since they are also found in large numbers in cells without any collagenetic function, for example in the papillary layer of the enamel organ (personal observation). It is more probable that they play a "micropinocytotic" role, removing fluid and organic substance from the dentinal matrix prior to calcification. Evidence exists which indicates that the coated vesicles in other cells are associated with protein transport across the plasma membrane into cells (*Roth & Porter,* 1962, 1964).

Histochemical studies are also needed to elucidate the function of the rod shaped dense bodies. They appear to be formed in the Golgi zone by accumulation of dense droplets within vacuoles, and can be followed through the odontoblast to the process at the level of the predentine. As they are not found more peripherally in the process it is tempting to suspect that they contain precursors of the amorphous, dentinal matrix. How such a substance could leave the process is not evident from the electron micrographs.

A nuclear inclusion of exactly similar appearance to that seen in odontoblasts in the present study has been described in nerve cells from the olfactory bulb, the cerebellum and cerebral cortex in rabbits, by *Siegesmund et al.* (1964). These authors have reviewed the literature and found that such intra-nuclear structures had been described by several older anatomists, but only in certain species. They were referred to as the "rodlets of Roncoroni". The significance of the rodlets is totally unknown, and it is difficult to imagine what connection may exist between their occurrence in such functionally different cell types as odontoblasts and neurones.

No nerve fibres have been observed in relation to the odontoblasts, and this is good agreement with earlier light microscopic observations, which describe nerve fibres as ending more centrally in the pulp without communication with the odontoblasts in rodent incisors (*Kazimieras et al.,* 1962).

SUMMARY

An electron microscopical study has been performed upon mature odontoblasts in perfusion fixed, demineralised incisors from

adult rats. The preservation technique which has been adopted has made it possible to demonstrate several organelles and inclusions not previously described in odontoblasts.

The odontoblasts are long, polarised cells attached to each other by desmosomes, tight junctions and terminal bars. The large nucleus, situated at the pulpal pole, contains parallel filaments, forming rodlets of a type previously only observed in certain neurones. Dilated cisterns of the highly organised ER frequently contain an inclusion with a paracrystalline pattern. In the supranuclear region of the cell body a well developed Golgi apparatus is present; and in relationship to this is seen a pair of centrioles, one of the centrioles constituting the basal body of a stereocilium. The cell body contains in addition mitochondria, ribosomes, rod shaped dense bodies, various lysosome-like bodies and a few multivesicular bodies. At the margin of the cell coated vesicles and numerous microtubules are seen. An intracellular filament cuff is present at the border between the cell body and the odontoblastic process.

The process itself contains rod shaped dense bodies, microvesicles, coated vesicles, filaments and numerous microtubules in its predentinal portion. Mitochondria are only rarely seen.

The possible significance of some of the organelles is discussed.

RÉSUMÉ

L'ULTRASTRUCTURE DES ODONTOBLASTES DES INCISIVES DE RATS ADULTES FIXÉES A LA PERFUSION ET DÉMINERALIEES

Une étude au microscope électronique d'odontoblastes à maturité dans des incisives de rats adultes fixées par perfusion et déminéralisées a permis d'observer, à cause de la technique de préservation, plusieurs organelles et inclusions non décrites jusqu'ici dans les odontoblastes.

Les odontoblastes sont des cellules longues, polarisées, dont la cohésion intercellulaire est assurée par des desmosomes, des "tight junctions" et des bandelettes obturantes. Le grand noyau placé vers le tissu pulpaire, contient des filaments parallèles formant des batonnets d'un type que l'on n'a jusqu'à présent observé que dans certains neurones. L'ergastoplasme nettement organisé contient des cavités qui presèntent souvent des inclusions à structure paracristalline. Dans la partie supranucléaire de la cellule

se trouve un appareil de Golgi bien developpé. En relation avec celui-ci on voit une paire de centrioles, dont l'une forme le corps basal d'un stéréocil. La cellule contient en plus des mitochondries, des ribosomes, des corps denses en forme de batonnets, différents corps qui ressemblent aux lysosomes et de plus quelques corps multivésiculaires. Au bord de la cellule, on peut voir des "coated vesicles" et de nombreux microtubules. Une manchette intracellulaire de filaments se trouve à la limite entre la cellule et le prolongement odontoblastique.

Le prolongement à son tour contient dans sa partie qui se trouve dans la prédentine des corps denses en forme de batonnets, des "microvesicles", des "coated vesicles", des filaments et de nombreux microtubules. Des mitochondries sont rarement observées.

La signification possible de quelques-unes de ces organelles est discutée.

ZUSAMMENFASSUNG

DIE ULTRASTRUKTUR DER ODONTOBLASTEN VON DEMINERALISIERTEN, PERFUSIONSFIXIERTEN INCISIVEN ERWACHSENER RATTEN

Es wurde eine elektronmikroskopische Untersuchung ausdifferentierte Odontoblasten in perfusionsfixierten, demineralisierten Incisiven erwachsener Ratten vorgenommen. Die Präservationstechnik, die angewandt wurde, ermöglichte es verschiedene Organellen und Inklusionen zu demonstrieren, die nicht früher in Odontoblasten beschrieben worden sind.

Die Odontoblasten sind lange, polarisierte Zellen, die miteinander durch Desmosomen, "tight junctions" und Schlussleisten verbunden sind. Der grosse Nucleus, im pulpalen Pol gelegen, enthält parallele Filamente, die Stäbchen eines Types bilden, die früher nur in einige Neuronen beobachtet wurden. Dilatierte Cisternen des hochentwickelten ER enthalten häufig eine Inklusion mit einem parakrystallinen Muster. In der supranuclearen Region des Zellkörpers findet sich ein gutentwickelter Golgi Apparat und in Relation zu diesem ist ein Centriolenpaar zu sehen; eine dieser Centriolen ergibt den Basalkörper eines Stereocilium. Der Zellkörper enthält darüber Mitochondrien, Ribosomen, stäbchenförmige dichte Körper, verschiedene lysosom-ähnliche Körper und einige wenige multivesiculäre Körper. Am Zellrand sind

"coated vesicles" und zahlreiche Microtubuli zu sehen. Eine intracelluläre Filament-manchette befindet sich an der Grenze zwischen dem Zellkörper und dem Odontoblasten-Prozessus.

Der Prozessus enthält in seinem prädentinalen Teil stäbchenförmige dichte Körper, Mikrovesiklen, "coated vesicles", Filamente und zahlreiche Mikrotubuli. Mitochondrien sind nur selten zu sehen.

Die eventuelle Bedeutung einiger dieser Organellen wird erörtert.

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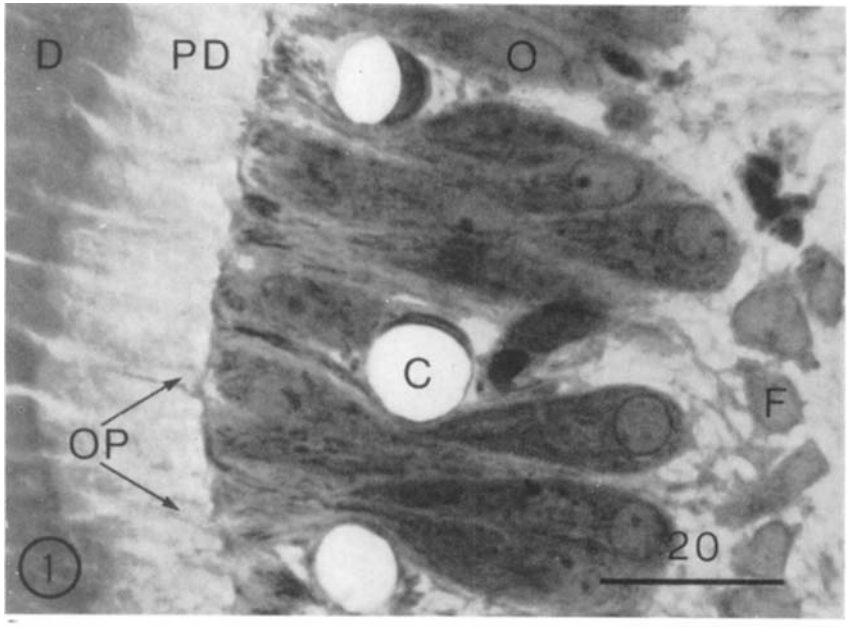
Fig. 1: Light micrograph of dentine (D), predentine (PD) and odontoblasts (O), showing subpredentinal capillaries (C). OP=odontoblastic processes. F=fibroblast. ×1,200

Fig. 2: Survey electron micrograph of an odontoblast nucleus (N) containing nuclear inclusions (NI). ×12,500


D
PD
O
C
F
OP
20
1

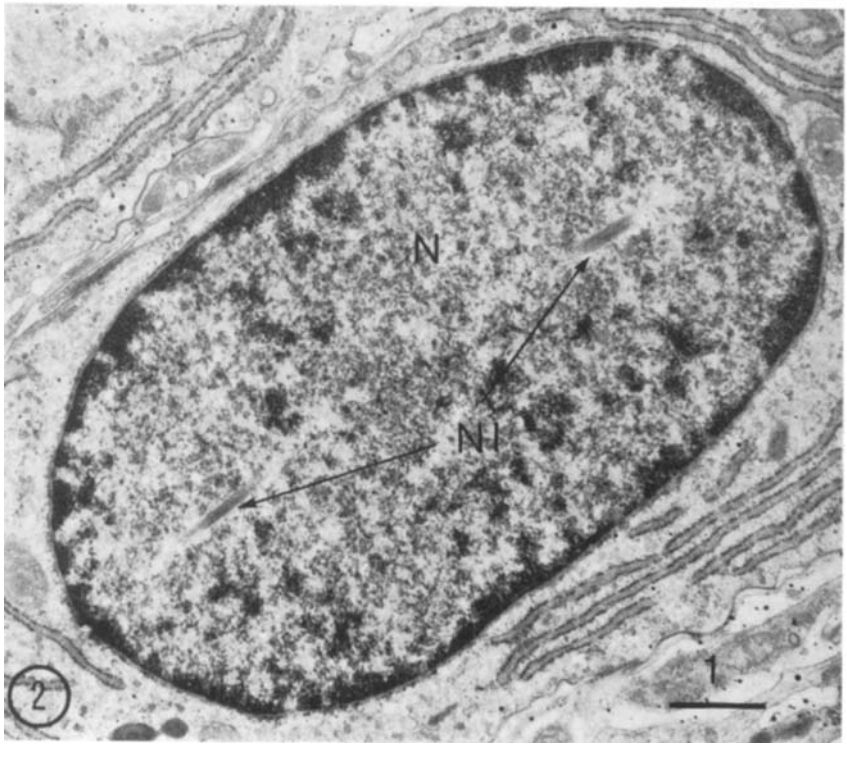
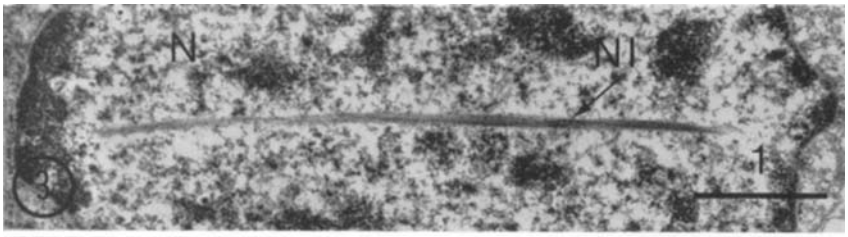
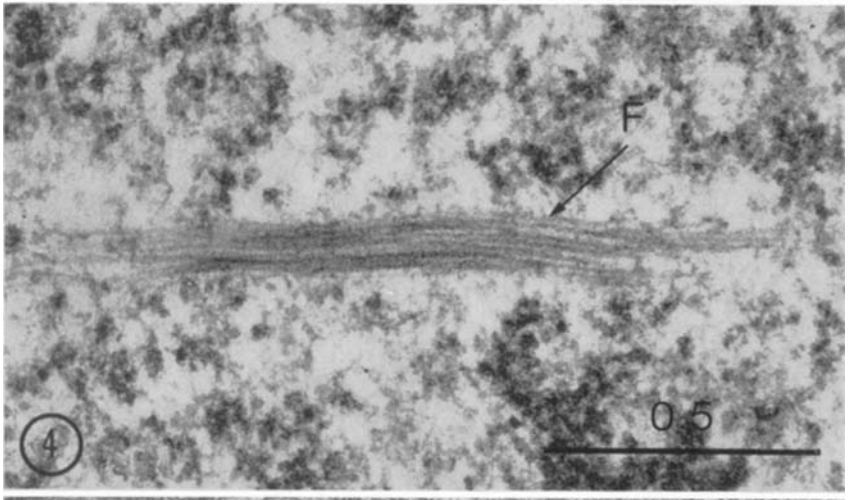

N
NI
1
2

Fig. 3: Longitudinal section through an odontoblast nucleus (N) showing a nuclear inclusion (NI) extending through the entire length of the nucleus. ×17,000

Fig. 4: Higher magnification of the nuclear inclusion, which is seen to consist of several, parallel filaments (F). ×72,500

Fig. 5: Longitudinal section through the marginal region of an odontoblast, showing numerous microtubules (MT) running parallel to the long axis of the cell. M=mitochondrion. ER=endoplasmic reticulum. ×32,000


N
NI
1
3


F
0·5
4

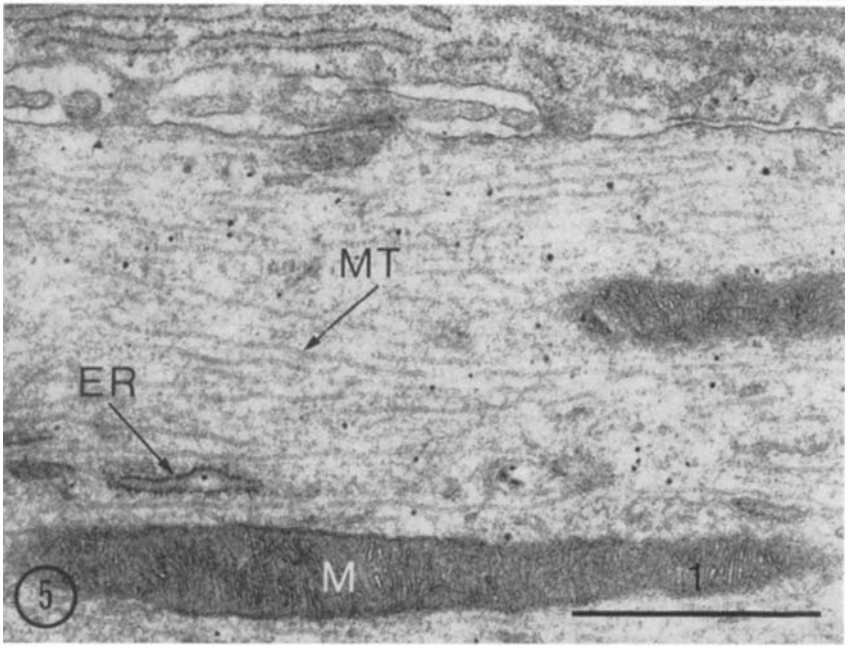

MT
ER
M
1
5

Fig. 6: Longitudinal section through odontoblasts, showing Golgi apparatus (G) and endoplasmic reticulum (ER) arranged in parallel cisterns. N=nucleus. MB=multivesicular body. L=lysosome-like body. RB=rod shaped dense body. MT=microtubules. CE=centriole. CI=cilium. C=collagen fibril. CV=coated vesicle opening on to the cell surface. ×21,000

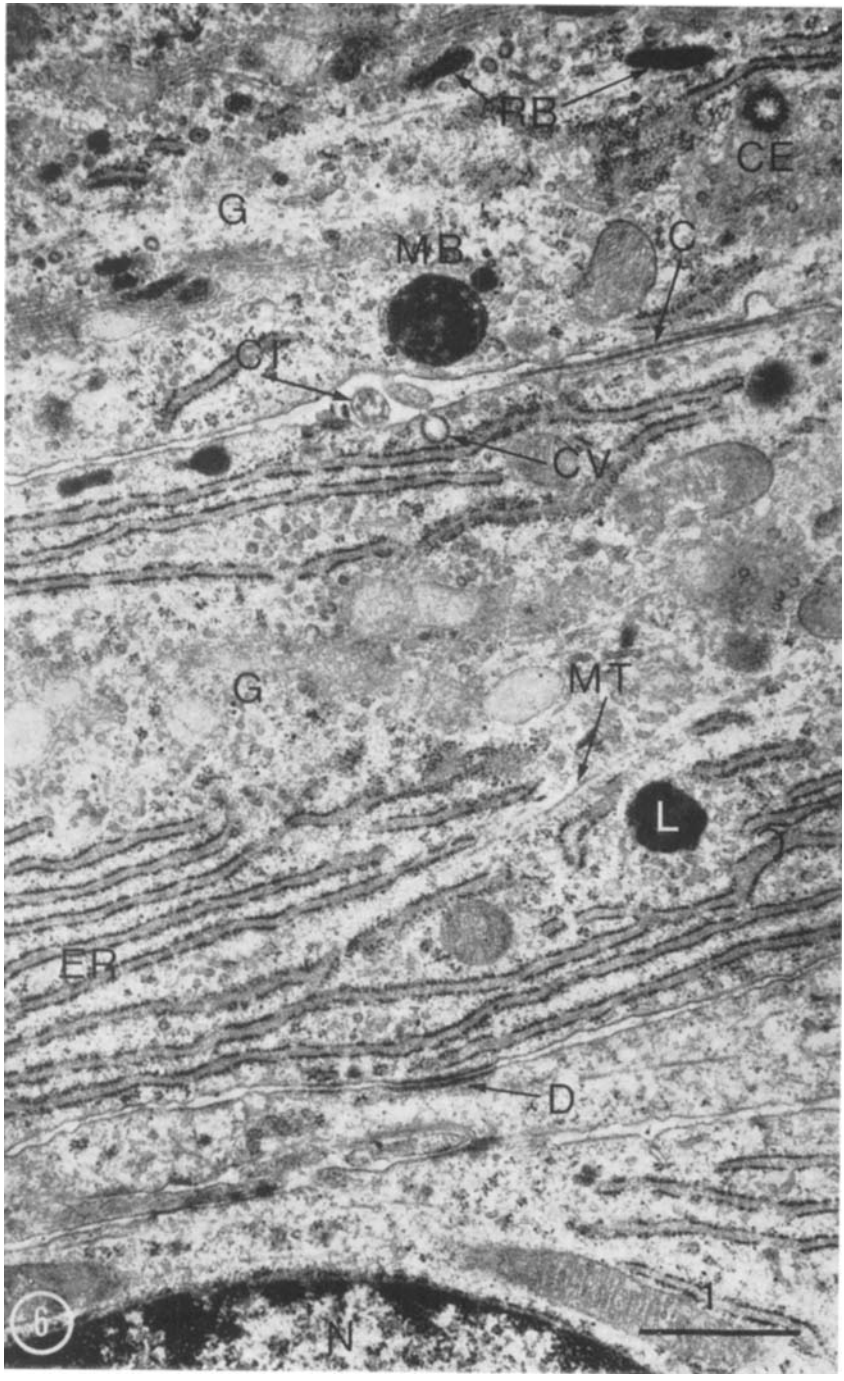
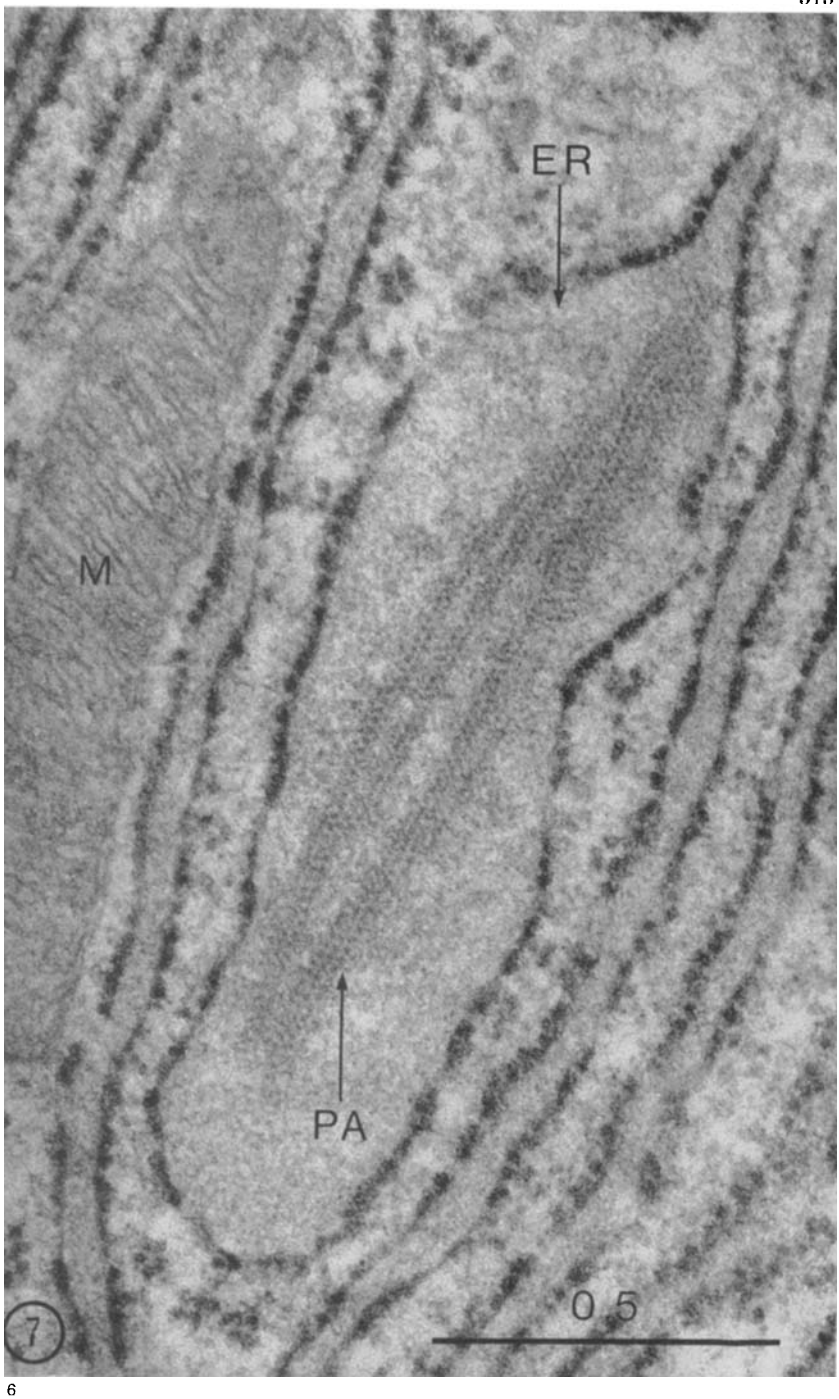

RB
CE
G
MB
C
CT
CV
G
MT
L
ER
D
N
1

Fig. 7: Dilated cistern of endoplasmic reticulum (ER) containing a paracrystalline material (PA). M=mitochondrion. ×89,000



6

Fig. 8: Longitudinal section through odontoblasts, showing cilia (CI) arising from the Golgi zone (G). MB=multivesicular body. L=lysosome-like body. ER=endoplasmic reticulum. CV=coated vesicles opening on to the cell surface. P=plasma membrane. ×24,000

Fig. 9: Longitudinal section through a cilium (CI) and the invaginated plasma membrane (P). B=basal body. ×25,500

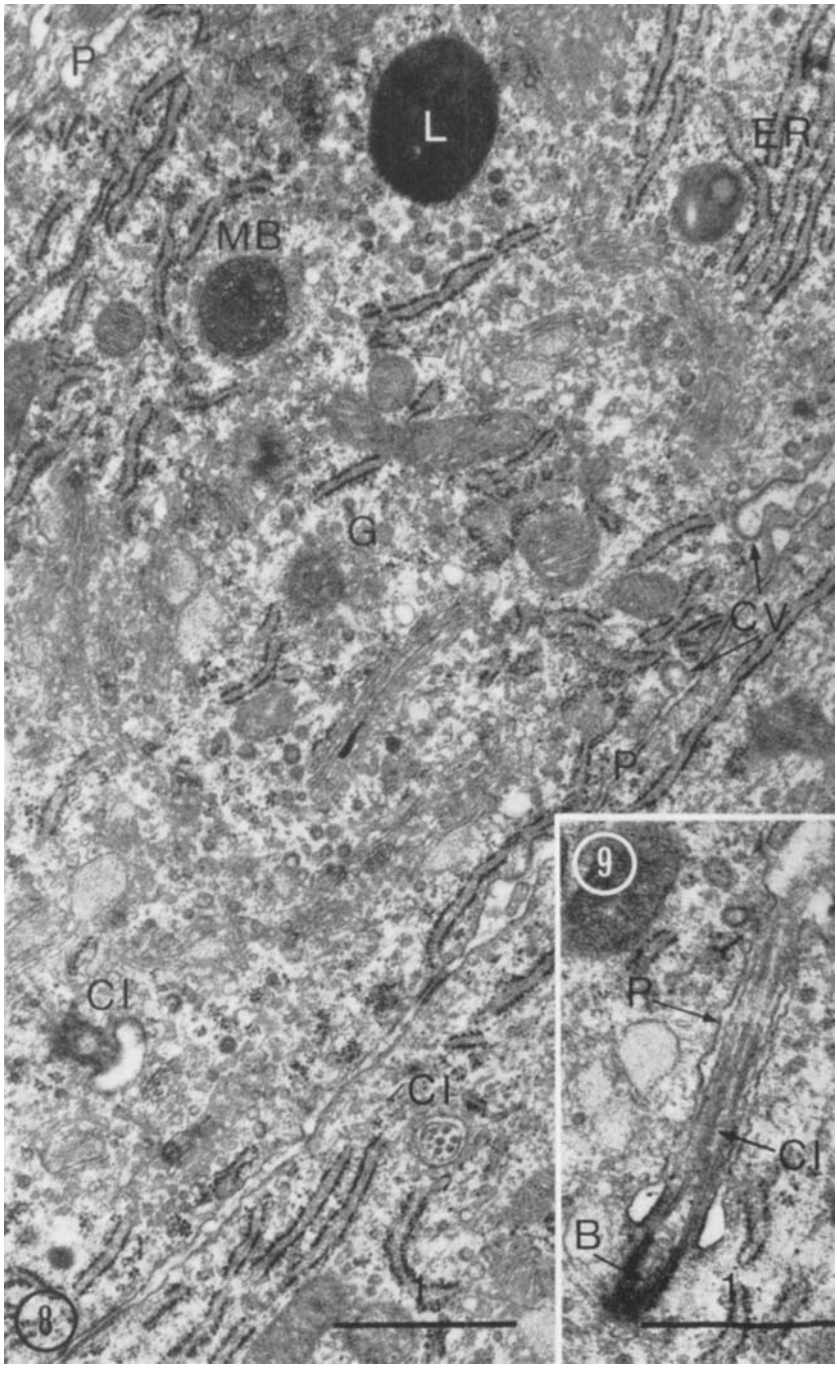

P
L
ER
MB
G
CV
P
CI
CI
8
9
R
CI
B
1

Fig. 10: Longitudinal section through odontoblast, showing terminal bars (TB) connecting the cells at the predentinal border. The adjacent cytoplasmic matrix contains numerous filaments forming a continuous cuff (FC). OP=odontoblastic process. ×37,000

Fig. 11: Longitudinal section through an odontoblastic process (OP), with side branch (SB) in the predentine. Among the numerous collagen fibrils (C) are seen transverse sectioned side branches (arrows). MT=microtubules. RB=rod shaped dense bodies. CV=coated vesicles opening on to the cell surface. ×32,500

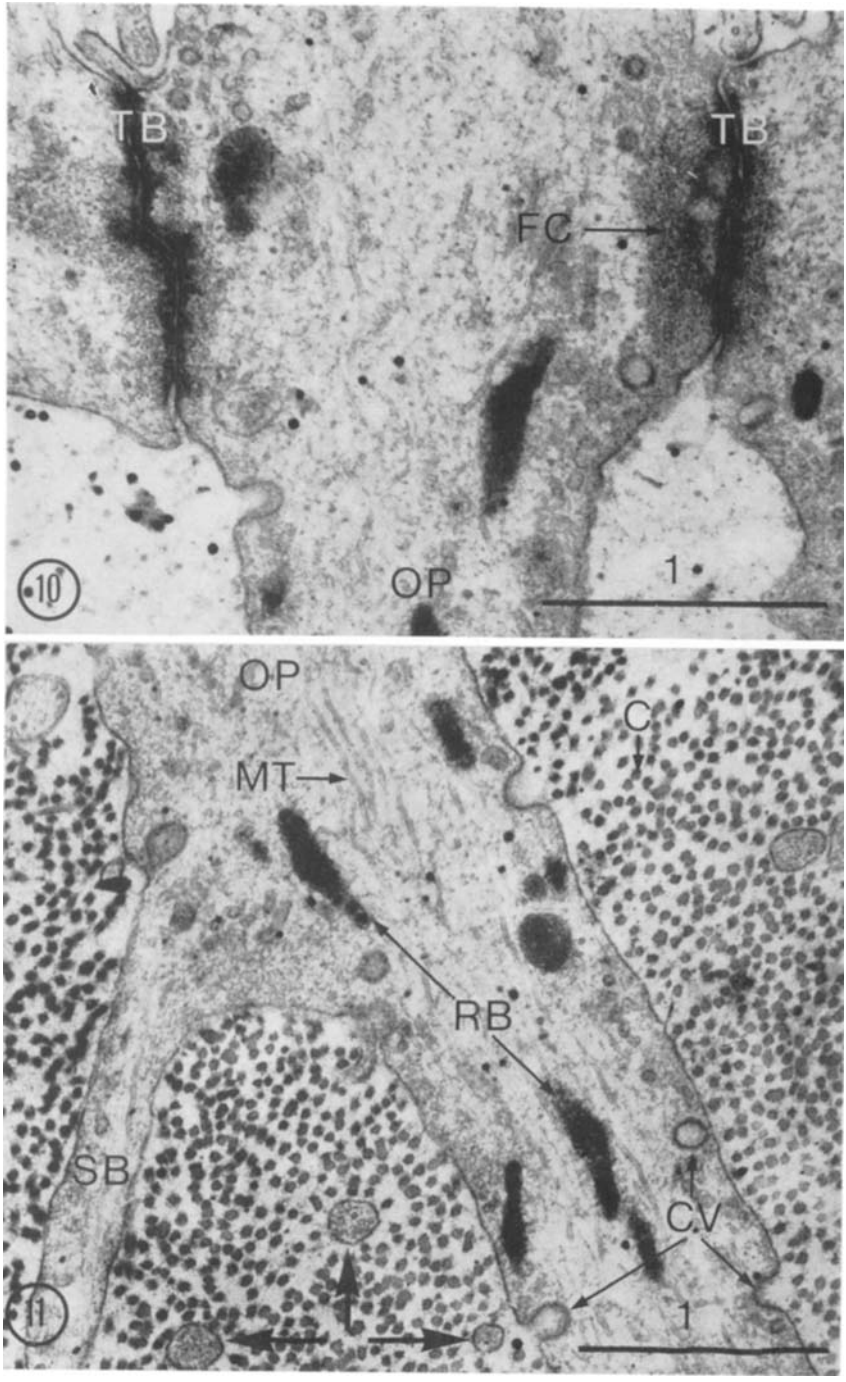

TB
TB
FC
10
OP
1
OP
C
MT
RB
SB
C.V
11
1

Fig. 12 and 13: Longitudinal section through odontoblastic processes in the predentine. MT=microtubules. MV=microvesicles. RB=rod shaped dense bodies. CV=coated vesicles, two of which open on to the cell surface. Filaments (arrow) ×43,000 and 47,000

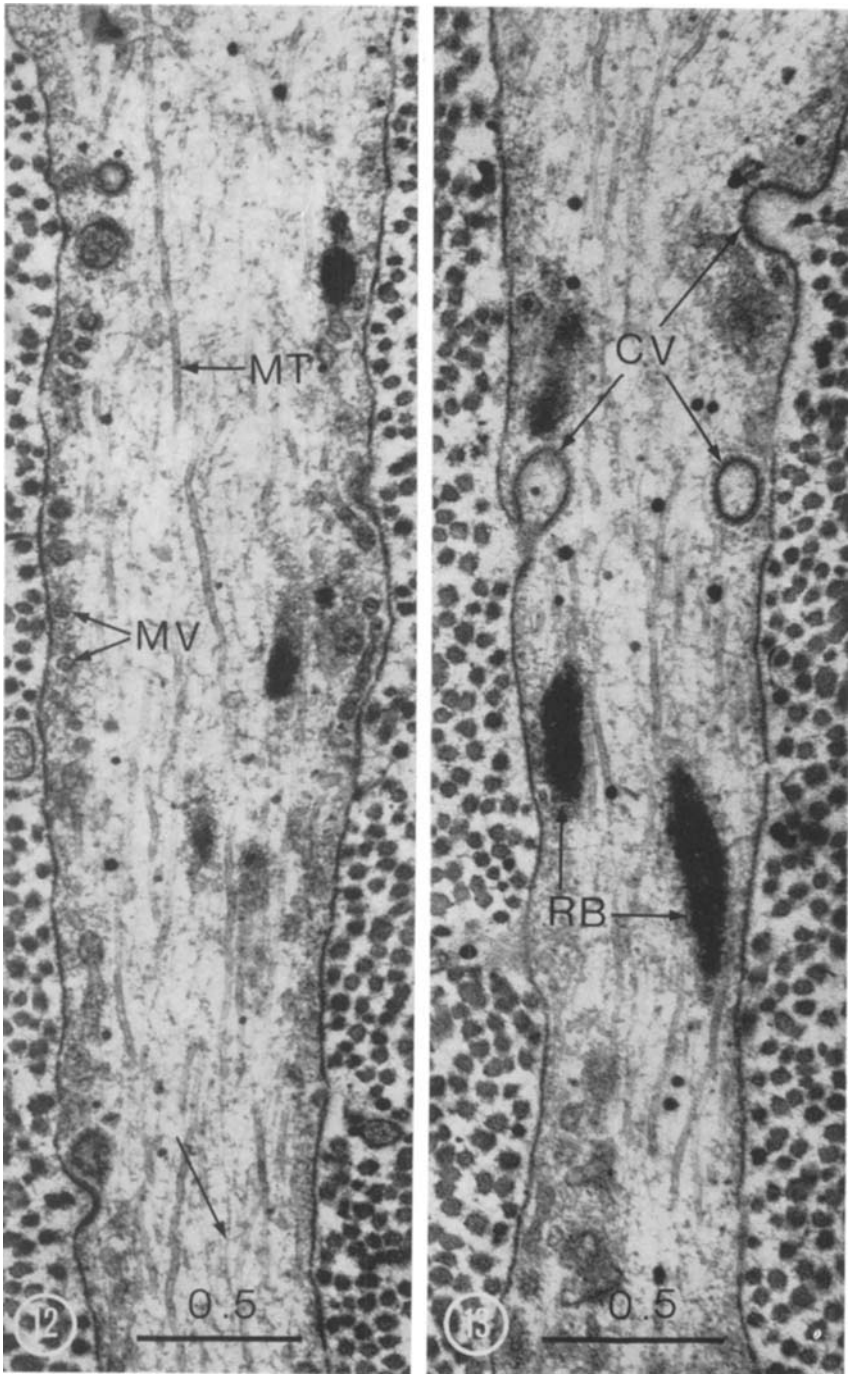
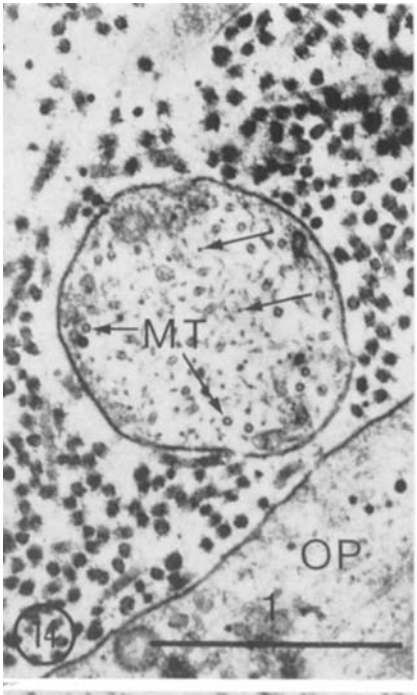

MT
MV
0.5
CV
RB
0.5

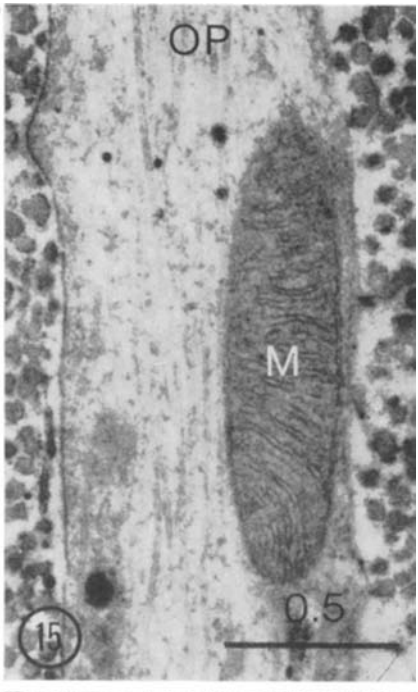
Fig. 14: Transverse section through a side process containing microtubules (MT) and filaments (arrows). Part of a longitudinally sectioned odontoblastic process (OP) is seen. ×33,000

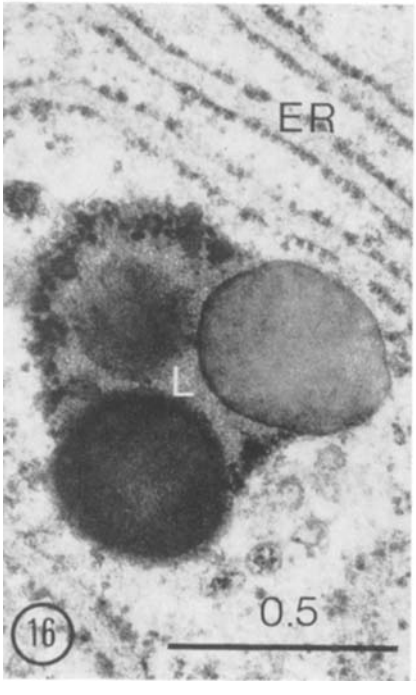
Fig. 15: Longitudinal section through an odontoblastic process (OP) in the predentine, containing a mitochondrion (M). ×45,500

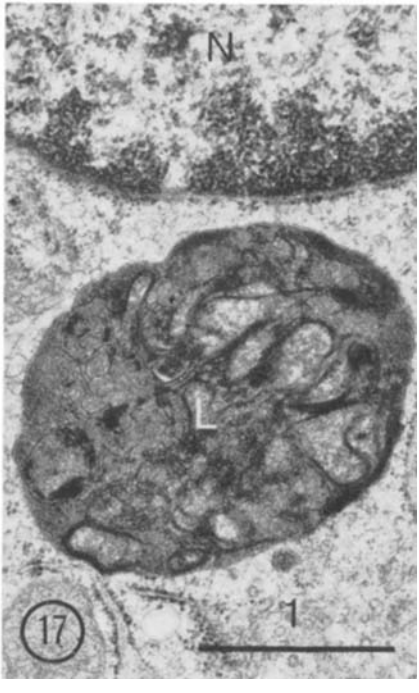
Fig. 16: Lysosome-like body (L) containing droplets of different density. ER=endoplasmic reticulum. ×59,500

Fig. 17: Lysosome-like body (L) containing membrane fragments. N= nucleus. ×25,500


MT
OP
1
14


OP
M
0.5
15


ER
L
0.5
16


N
L
1
17