The fine structure of the vessels in the human dental pulp

ERIK DAHL & IVAR A. MJÖR

Department of Anatomy, Dental Faculty, University of Oslo, Norway

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The vessels in the human dental pulp have been studied with light and electron microscopy with emphasis on the distribution and the type of vessels present. All vessels were relatively thin walled. Apart from typical arterioles, venules and capillaries, vessels considered as possible arteriolo-venule anastomoses and lymphatics were also observed. In a discussion on the correlation between structure and functions, attention was focussed on anastomoses, capillary permeability and pulp tissue pressure.

Key-words: Dental pulp; blood vessels; microscopy, electron

Erik Dahl, Department of Anatomy, Dental Faculty, P.O. Box 1052, Blindern, Oslo 3, Norway

The fine structure of the blood vessels in the dental pulp has been described in animals (*Han & Avery*, 1963; *Kukletová*, 1970a) and in human teeth (*Cahen*, 1969; *Eifinger*, 1970; *Harris & Griffin*, 1971). The present paper deals specifically with the organization of the blood supply to the different areas of the pulp tissue, the ultrastructure of the different types of vessels and the relationship between blood vessels and the other structures in the human dental pulp.

MATERIALS AND METHODS

Twenty premolars from children aged 10—13 years were immediately cleaved (*Furseth & Mjör*, 1969), and the pulps were removed and placed in a 2.5 per cent glutaraldehyde solution buffered at neutral pH as described by *Dahl & Mjör* (1973), i.e. post-fixed in 1 per cent osmium tetroxide and embedded in Vestopal W (*Ryter & Kellenberger*, 1958). Ultrathin sections were examined in a Siemens Elmiskop Ia electron microscope. From the same plastic blocks, sections 1 μm were prepared for light microscopy. These sections were stained on a heating stage with an aqueous solution of 0.1 per cent toluidine blue adjusted to pH 8.9 with 0.67 M Na_2HPO_4.

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OBSERVATIONS

Light microscopy

The 1 μm thick toluidine blue stained sections from the central area of the dental pulp reveal a rich vascularization. Different types of vessels were identified, the largest measuring about 140—150 μm in diameter.

The arterioles are easely recognized (Fig. 1) by their well developed tunica media with smooth muscle cells. A regular adventitia is not seen, but in some arterioles a perimuscular area which morphologically differs from the surrounding tissue is encountered (Fig. 2). The venules are characterized by their thin walls, and often irregular lumen. No adventitia is seen in relation to the venules (Figs. 1, 3).

Numerous nerve fibers are seen accompanying the larger arterioles (Fig. 1), but no nerves are found adjacent to the venules. From the centrally located arterioles, branches arborize towards the more peripheral part of the pulp. Capillaries of different sizes are observed throughout the whole pulp, but they seem to be most numerous in the subodontoblastic layer (Fig. 4). Lymphatic vessels cannot be differentiated from blood vessels with light microscopy.

Electron microscopy

Arterioles. The wall of the arterioles of the dental pulp comprises the three usual coaxial coats, viz the tunica intima, tunica media and an ill defined tunica adventitia, which often is difficult to differentiate from the surrounding tissue.

Tunica intima. The vessel lumen is lined by a single layer of endothelium which may appear squamous and sometimes also of a somewhat cuboidal form, resting on a distinct well developed basement membrane (Fig. 5). Remnants of elastic tissue is occasionally encountered.

Tunica media. Up to two layers of smooth muscle cells may be found oriented in a circumferential spiral. The smooth muscle cells are surrounded by a distinct basement membrane, and in all respects consistent with smooth muscle cells in vessels in other organs (Fig. 5).

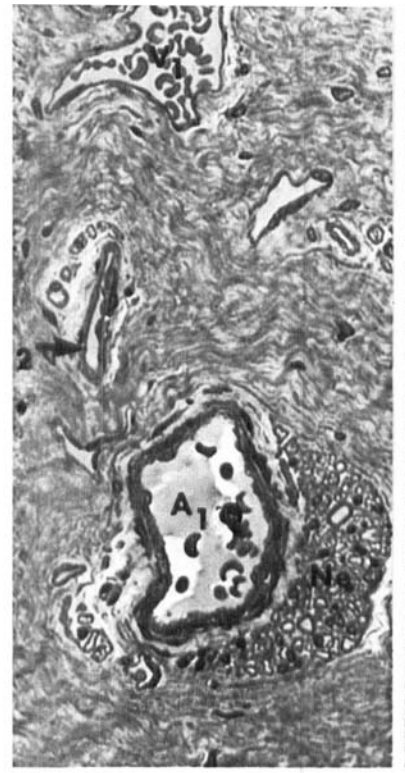
Tunica adventitia. Connective tissue in the close vicinity of the tunica media is composed of ground substance and scattered bundles of collagen fibres, sometimes infiltrated by cytoplasmic processes from fibrocytes (Fig. 5). However, the tunica adventitia does not exhibit any peculiar feature differentiating it from the surrounding connective tissue.

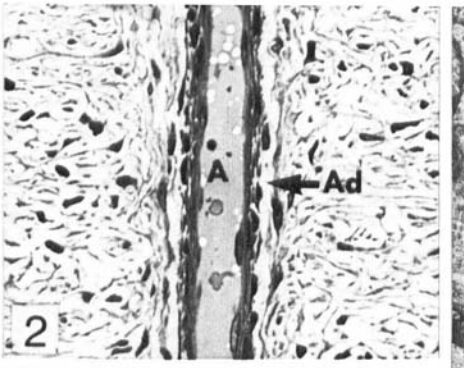
Capillaries. The blood capillaries, approximately 4—8 μm in diameter, consist of endothelial cells with marginal folds which overlap each other, or the intercellular junctions are composed of a complex infolding and interdigitations (Fig. 6). The nucleus sometimes appears lobulated and protrudes into the lumen (Fig. 6). The luminal surface of the endothelium may display numerous cytoplasmic projections, giving it an irregular appearance (Fig. 6). Fenestrations in the endothelial plasma membranes occurs regularly and rather frequently (Figs. 6, 7), the pores being spanned by a thin diaphragm (Fig. 7). A distinct well developed continuous basement membrane are seen surrounding the blood capillaries (Figs. 6, 7, 8).

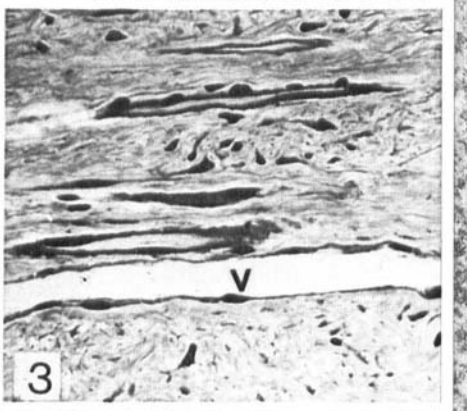
Pericytes are seen ensheathing some of the capillaries (Fig. 8), which appear different both in size and form. In some instances the endothelial lining is extremely thin, and with a rather dilated lumen (Fig. 8).

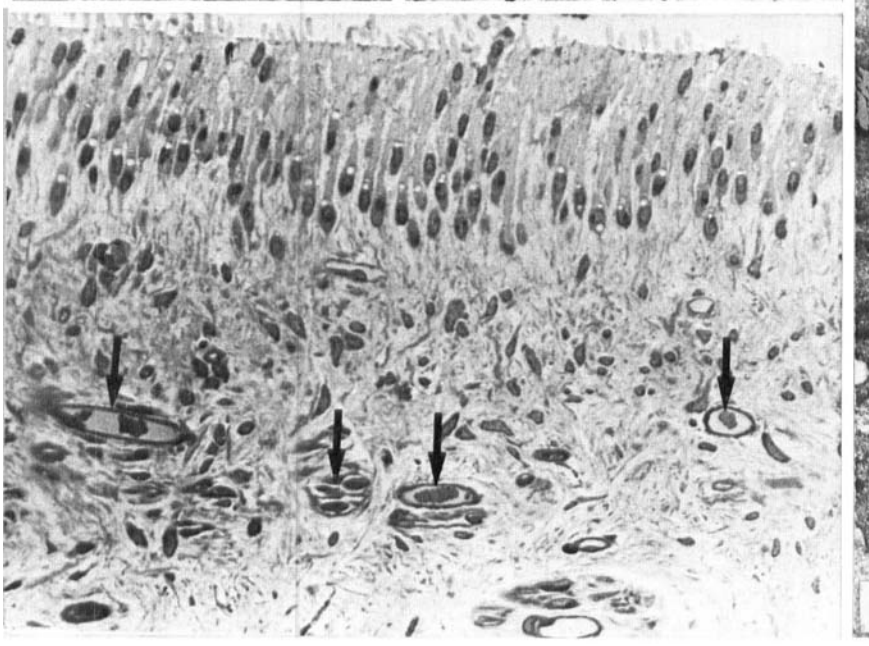
Capillaries deprived of basement membranes or in open communication with the surrounding tissue are not observed. Even though deep intercellular clefts are encountered in some capillaries, an area of zona adherens (or ocludens) is always observed (Fig. 10). Bundles of filaments (Fig. 10) are a regular, characteristic feature of the endothelial cells.

Possible arteriolo-venule anastomoses. A type of vessel which appears quite different from the regular arterioles are oc-









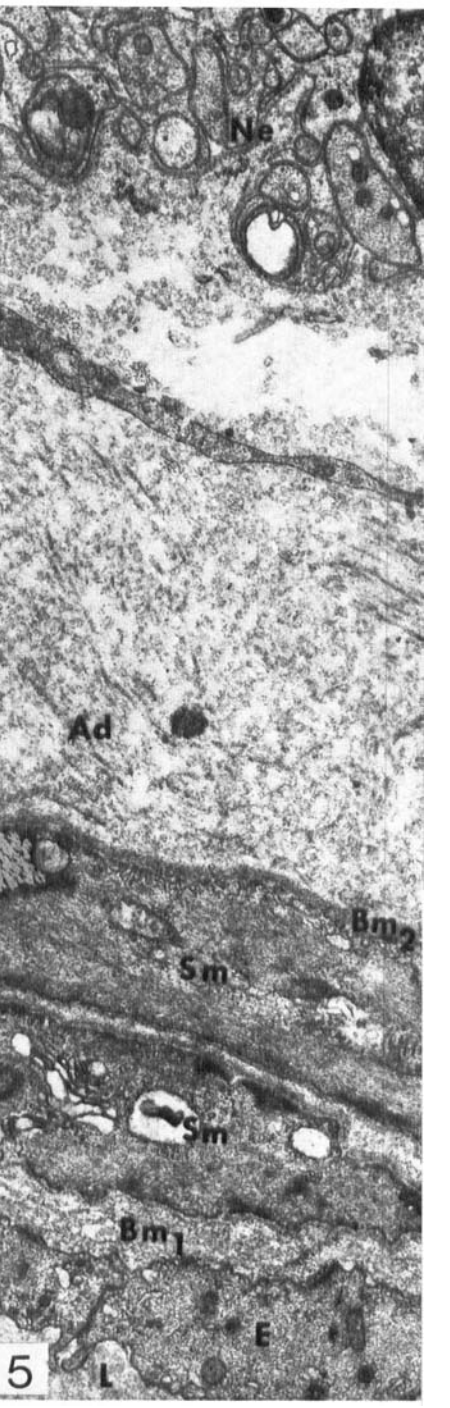


Fig. 1. Light micrograph from the central are of the pulp with a large arteriole (A_1), a venule (V_1) and a small arteriole (A_2). Note the nerve fibers (Ne) adjacent to the arterioles. ×500.
Fig. 2. Longitudinal sections of an arteriole (A) with a well defined muscle layer and a rather ill defined adventitia (Ad). ×300.
Fig. 3. Longitudinal section of a venule (V). Note that the vessel wall is thinner and more delicate than the arteriole in Fig. 2. ×300.
Fig. 4. In the subodontoblastic area only precapillaries and capillaries (arrows) of different sizes are seen. ×300.
Fig. 5. Electron micrograph illustrating details of an arteriole with the vessel lumen (L), endothelial cells (E), smooth muscle layer (Sm) and an ill defined adventitial coat (Ad) which morphologically is similar to the pulp tissue. Note the nerve fibres (Ne) adjacent to the vessel wall. Bm_1, basement membrane adjacent to endothelial cells; Bm_2, basement membrane between muscle layer and adventitia. ×15.000.

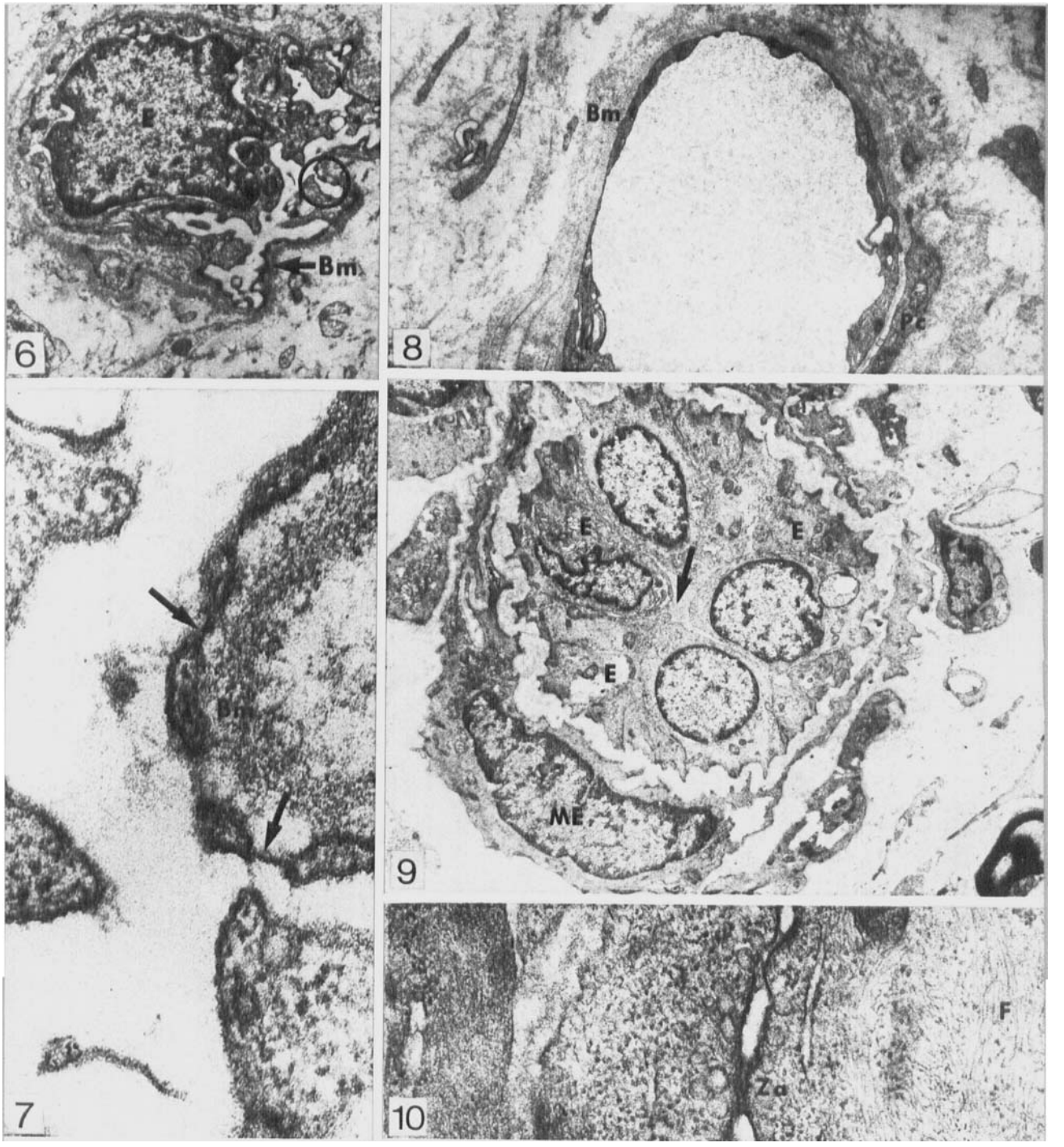


Fig. 6. A small capillary characterized by an irregular lumen, endothelial cell (E) with numerous cell processes projecting into the vessel lumen. Encircled area is shown in Fig. 7. The basement membrane (Bm) is well developed. ×12.000.

Fig. 7. High magnification of the encircled area in Fig. 6, demonstrating typical fenestra which are characteristic for the pulp capillaries (arrows). Bm, basement membrane. ×120.000.

Fig. 8. A thin-walled capillary with a well developed basement membrane (Bm), partly surrounded by a pericyte (Pc). ×9.000.

Fig. 9. Possible arteriolo-venule anastomosis with a small lumen (arrow). Cuboidal endothelial cells (E) projecting into the vessel are surrounded by cells which morphologically may be a kind of myo-endothelial cells (ME). ×6.000.

Fig. 10. Bundles of filaments (F) are regularly observed in the endothelial cells in the different kind of pulp vessels. Za, Zona adherens. ×60.000.

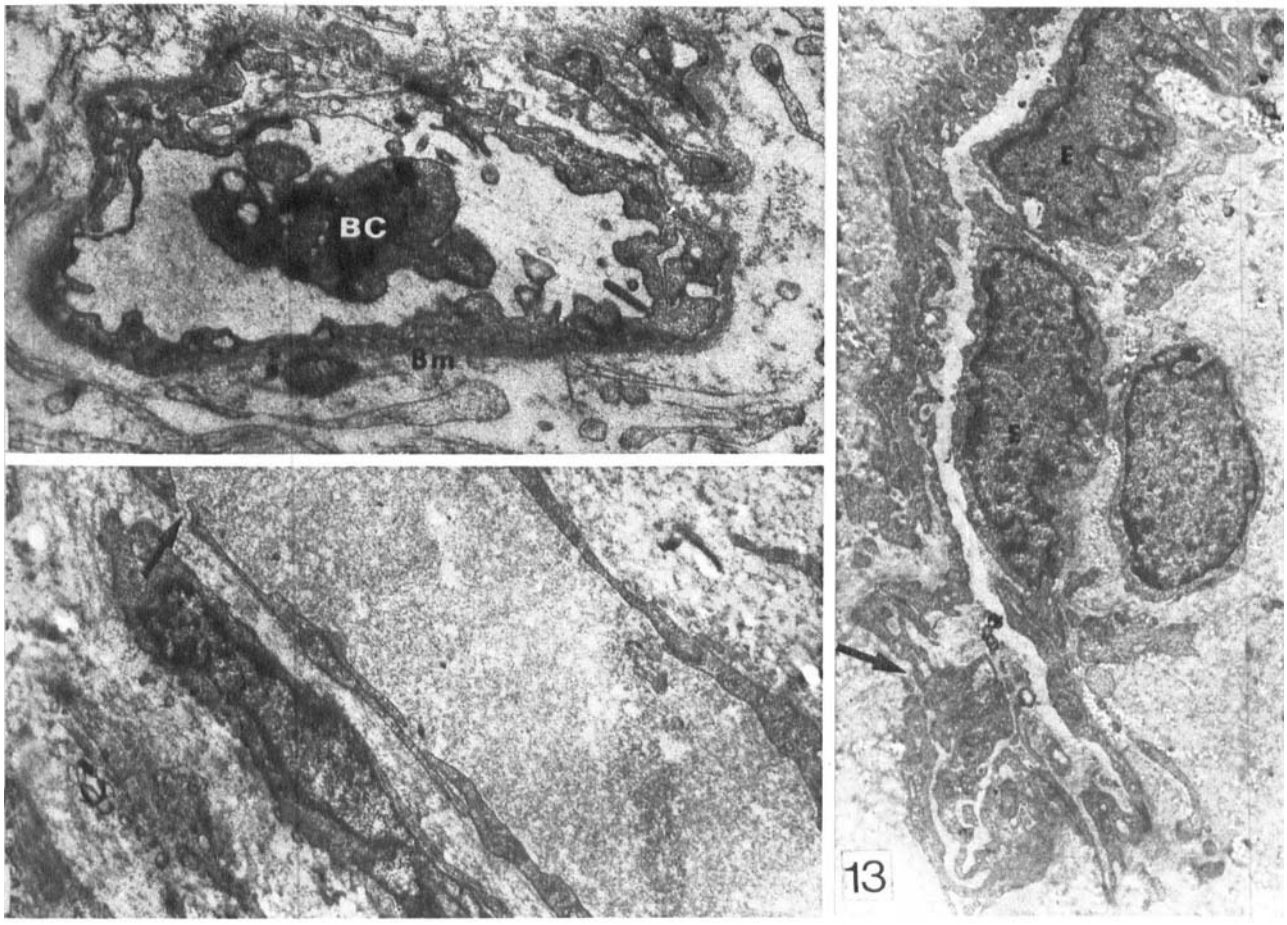


Fig. 11. A small venule characterized by lack of smooth muscle cells in the vessel wall, but with a well developed basement membrane (Bm). Part of a white blood cell (BC) is seen in the vessel lumen. ×15.000.
Fig. 12. A vessel, characterized by discontinued endothelium (arrow) and lack of basement membrane, may be a lymphatic vessel. ×8.000.
Fig. 13. Another possible lymphatic vessel characterized by irregular lumen, lack of basement membrane, irregular endothelial cells (E) with possible discontinuation of the endothelial cells (arrows). ×9.000.

casionally encountered (Fig. 9). They are characterized by cuboidal endothelial cells projecting into the vessel lumen producing a corrugated lumen. The endothelial cells are surrounded by a well developed basal lamina and smooth muscle cells (Fig. 9). No tunica adventitia is observed in these vessels.

Venules. The submicroscopic structure of the venules is to some extent similar to that of the arterioles (Fig. 11). The wall of the vessels is, however, thinner in relation to the size of the lumen. The tunica media is composed of a discontinuous layer of smooth muscle cells, and in many cases it can not be found. In such cases the connective tissue is immediately adjacent to the basement membrane. Fibrocytes or their processes often occur in the close vicinity of the vessels.

Possible lymphatic vessels. In addition to the blood capillaries, arterioles and venules, vessels of a different structure are also found in the dental pulp (Figs. 12, 13). The lumens of the vessels are often small (Fig. 13). A characteristic finding is that the endothelium of the vessel is not in all places continuous (Fig. 12). Some intercellular junctions apparently remain open so that the vessel lumen communicates

directly with the surrounding connective tissue. Furthermore, these vessels lack a fully developed basement membrane (Fig. 13).

DISCUSSION

In the present study the general structure of the dental pulp, the fibroblasts, undifferentiated mesenchymal cells and extracellular material are consistent with previous reports (e.g. *Kukletová*, 1970; *Eifinger*, 1970; *Harris & Griffin*, 1969).

Light microscopic investigations of arterioles in the human dental pulp have revealed that their maximum size is roughly 150 μm (e.g. *Eifinger*, 1970), and the present study support these findings. The arterioles which were found located to the central area were of a similar structure as described in other organs (*Rhodin*, 1967). However, elastic tissue was not found in the intimal layer, nor was any well defined tunica adventitia observed. It should also be stressed, that even though nerve fibres were found accompanying the greater arterioles, no neuro-muscular membranous contacts were observed.

Blood vessels of the capillary types were most numerous in close proximity to the odontoblast layer indicating that this area is well vascularized (*Harris & Griffin*, 1969). *Provenza* (1958) using light microscopy differentiates the pulp vessels as true capillaries, pre-capillaries and metarterioles. *Harris & Griffin* (1971) have classified the capillaries in two different groups, viz. fenestrated capillaries without pericytes and non-fenestrated capillaries. Both fenestrated and non-fenestrated capillaries were regularly observed in the present study. It was not possible therefore to draw any conclusions regarding any possible morphologically differences between the two types. They may in fact simply be sectioned at different levels in their course.

A striking finding was the intracytoplasmic filaments seen in the endothelial cells. The function of these filaments has not been conclusively explained (*Kukletová*, 1970a). From a morphological point of view it is not possible to decide whether they represent tono- or myofilaments. However, as experimental data are still lacking to prove that endothelial cells contracts upon stimulation, the filaments are assumed to represent tonofilaments which strengthen the cell (*Odland*, 1961; *Rhodin*, 1962, *Cecio*, 1967; *Schipp*, 1968; *Kukletová*, 1970a). Precapillary sphincters (*Rhodin*, 1967) were not observed. However, blood vessels which in previous reports have been characterized as arteriovenous anastomoses (*Harris & Griffin*, 1971) have also been observed in the present study.

The possible presence of lymphatic vessels in the dental pulp has been discussed extensively (e.g. *Eifinger*, 1970; *Kukletová*, 1970b). The present study demonstrates vessels similar in structure to what previously have been described as lymph vessels in the human dental pulp (*Riedel, Fromme & Tallen*, 1966) and the calf dental pulp (*Kukletová*, 1970b). On the basis of special features, like irregular lumen, thinnes of the endothelium, absence of red cells in the lumen, absence of basal lamina and a discontinuous endothelial layer, it appears reasonable to suggest that these vessels represent dental pulp lymphatic vessels.

Correlation of structure and function

The dental pulp is supplied by blood through the apical foramen through which usually one small artery is running to-

gether with one or two veins (*Meyer*, 1951; *Sicher & Bhaskar*, 1972). Shortly after entering the pulp tissue, the artery arborizes in several arterioles finally forming a capillary plexus in the coronal pulp (*Kramer*, 1968). *Provenza* (1958) described the peripheral circulation as containing arterio-venous anastomoses connecting an arteriole to a vein. Usually before the capillary circulation is established, an arterio-venous anastomoses connects the afferent arteriole to the efferent vein, i.e. alternate circulation routes are available (*Harris & Griffin*, 1971). The blood supply to the different areas is controlled by the anastomosis depending of the actual functional need.

The importance of the fenestrations of the capillaries is connected to the permeability, and differences in permeability of capillaries are explained by assuming differing ratios of the types of pores (e.g. *Maul*, 1971). In the pulp tissue, a large supply of metabolites to the odontoblasts are needed, which is reflected by the existance of numerous fenestrated capillaries in the subodontoblastic layer. It has often been suggested that the pulp is particularly sensitive to circulatory disturbances because of its encapsulated location (e.g. *Boling & Robinson*, 1938). Interruptions leading to small increases in the pulp pressure may then give collapse of the veins which are thinwalled in the pulp (*Provenza*, 1964; *Pohto*, 1967). However, it should be born in mind that an increase in pulp pressure may be a localized phenomenon (*van Hassel*, 1971).

The magnitude of the pulp tissue pressure *per se* has been claimed to be of importance for the ability of the pulp to sustain circulatory disturbances (*Brown*, 1968). Pulp pressure values of about 30 mm Hg have been claimed to imply a capillary pressure exceeding that found in other tissues (*Brown* et al., 1969), and this makes the pulp an extraordinary organ in some respect, although similar or higher tissue pressures are found in other tissues.

Nishijima, Imanishi & Akai (1965) have demonstrated a flow of extravascular fluid in tissue spaces. These spaces pass straight from the capillaries to the centrally located veins. »The perivascular space» which contains few fibrils, is also claimed to play an important role as a pathway for the lymph stream, and a similar observation has been made by *Isokawa* (1960). In the present study, both thin-walled veins, rather large tissue spaces and also possible lymph vessels have been observed which seems to support previous observations.

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