

Lymphatic vessels in inflammed human dental pulp

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SUMMARY

Investigation has been performed on both the light and electron microscopic characteristics of the lymphatic vessels present in the dental pulp of human teeth which have been affected by serious carious lesions. These conditions provoke a severe inflammatory response resulting in structural and functional modifications of the tissue; increase of the tissue pressure is followed by the need for a more intensive lymphatic drainage. In the inflammed pulps, dilated lymphatic vessels with distended walls and »open junctions« between endothelial cells are detectable. On the other hand they lack certain endothelial structures which characterize the morphology of these vessels under normal conditions. In the pulpal regions affected by fibrotic proliferation shrank vessels with irregular profiles are present. From these observations it is possible to obtain other information on the mechanisms regulating the lymphatic drainage in different structural and functional conditions of the interstitium.

KEY WORDS:

Dental pulp, inflammation, lymphatic vessels.

RÉSUMÉ

La recherche a eu pour but d'examiner au microscope optique et au microscope électronique les caractéristiques morphologiques des vaisseaux lymphatiques présents dans le tissu pulpaire de dents humaines atteintes de graves lésions dues aux caries. En de pareilles conditions, des processus inflammatoires s'installent dans le tissu pulpaire d'une telle intensité qu'ils provoquent des modifications structurelles et fonctionnelles dans le tissu avec une augmentation de la tension tissulaire et donc un besoin majeur de drainage de la part du système lymphatique. Dans les pulpes enflammées, on a pu démontrer la présence de vaisseaux lymphatiques dilatés dans la paroi desquels on trouve des «jonctions ouvertes» alors, qu'il manque certaines des structures endothéliales qui caractérisent ces vaisseaux dans les conditions normales.

Dans les zones de tissu pulpaire avec dégénérescence fibreuse, on trouve des vaisseaux lymphatiques aux parois contractées et irrégulières.

Par ces observations, il a été possible de trouver d'autres informations sur les mécanismes qui règlent le drainage lymphatique dans différentes conditions structurelles et fonctionnelles de l'interstitium.

MOTS-CLÉS:

Pulpe dentaire, inflammation, vaisseau lymphatiques

INTRODUCTION

Carious invasion or other pathogeneous stimuli provoke an inflammatory response in the pulpal tissue wich in their turn result in an increase of blood

capillary permeability and of tissue pressure (van Hassel and Brown, 1969; Van Hassel, 1973). Moreover an invasion of inflammatory cells, i.e. lymphocytes, plasmacells, neutrophils and macrophages also occurs (Bernick, 1977b). A

widespread and persisting inflammatory state can produce a severe disturbance of the pulpal hemodynamics (Kim, 1985; Heyeraas, 1985, 1989). Swelling of the pulpal tissue caused by increasing interstitial pressure in the inflammatory state is prevented by the rigid walls of the dental chamber. So an insufficient drainage of the increased interstitial fluid may produce a failure of the pulpal viability and also cause pulpal death (Van Hassel, 1973; Bernick, 1977b). For these reasons it's evident that an efficient drainage of the pulpal tissue is called for.

It is well-known the importance of the lymphatic vessels in the removal of excess fluid, macromolecules, cellular particles, bacteria and so on from the interstitium. In fact it is a network of lymphatic capillaries and collecting vessels that drains the dental pulp. Their arrangement and morphological characteristics have been investigated by using light and electron microscopes (Kukletova, 1970; Bernick, 1977a; Marchetti and Piacentini, 1990; Bishop and Malothra, 1990).

The morphological characteristics of the lymphatic vessels of the dental pulp have also been studied during inflammatory states caused by carious lesions of the dentine (Bernick, 1977b). Under these conditions dilated lymphatic vessels with well distended walls were described but only using light microscopy.

Previously we have documented in other organs the different ultrastructural characteristics of the endothelial wall of the lymphatic vessels in normal and in oedematous conditions of the tissue (Marchetti et al. 1978). They represent structural adaptations occurring in the lymphatic microvasculature due to functional changes in the interstitial tissue conditions.

We described the morphological characteristics of the lymphatic vessel network draining healthy human dental pulp in a previous paper (Marchetti and Piacentini, 1990). Now the aim of this paper is to investigate the microscopic and the ultrastructural morphological modifications that the lymphatic system can undergo owing to an inflammatory state caused by carious invasion.

MATERIAL AND METHODS

Investigations have been performed on molar teeth which were severely damaged by caries in middle age subjects (45-55 years). After extraction, the teeth were longitudinally cleaved and immersed in a Karnovsky fixative at half strength, for 30 minutes at

4°C. Then the pulps were gently removed and reimmersed in the same paraformaldehyde-glutaraldehyde fixative solution for 3 hours. They were post-fixed in a solution 1,33% of OsO₄ in a collidine buffer for 1 hour, dehydrated and embedded in epoxy resin.

Semithin sections were performed to investigate at light microscopy the pulpal tissue conditions and to distinguish the lymph from the blood vessels. The ultrastructural features of the tissue and the characteristics of the lymphatic vessels were examined on ultrathin sections which were routinely contrasted.

RESULTS

One or more inflamed regions were detectable with the light microscopy in each pulp taken from teeth which had been affected by severe carious processes. These inflammatory lesions had different extension and were characterized by large amount of cellular infiltrate, i.e. lymphocytes and macrophages but also plasmacells and neutrophils. Sometimes coarse bundles of fibres lie between the cellular infiltration making a more or less thick network in the extracellular matrix (Fig. 1). In some pulps, large and numerous pulp stones appeared. Blood or lymphatic vessels were not found in the core of the inflammatory lesions. On the contrary in the periferal regions numerous vessels were easily detectable in a looser matrix where the cellular infiltrates were less numerous. The lymphatic vessels were distinguished from the blood vessels by their large size, thin walls and the absence of pericytes. The majority of the vessels appeared to be dilated because they were characterized by rather distended and less irregular walls (Fig. 2).

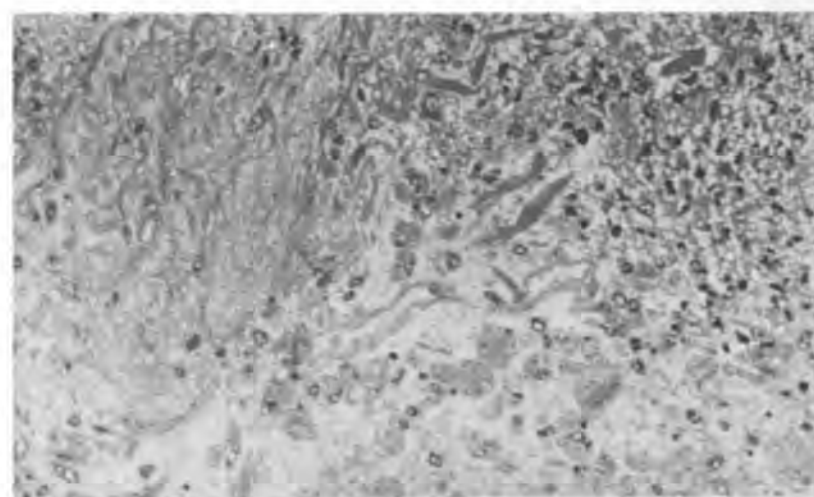


Fig. 1: Une région inflammée d'une pulpe dentaire avec une remarquable infiltration cellulaire. 350 X
An inflammatory region of a dental pulp with conspicuous cellular infiltrates. 350 X

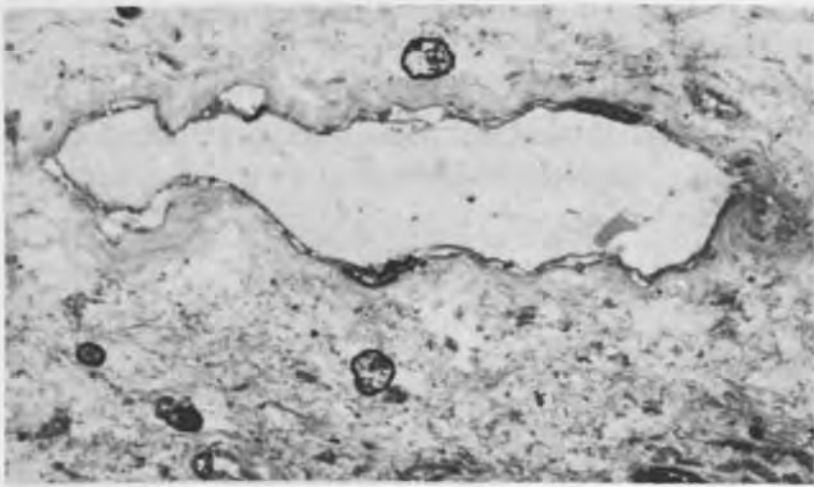


Fig. 2: Un vaisseau lymphatique dilaté avec une paroi mince et distendue qui se trouve dans la matrice autour de la partie centrale enflammée.

1300 X

A dilated lymphatic vessel with a thin and distended wall lies in the loose connective matrix which is peripheral to the inflammatory core.

1300 X

Nevertheless in pulps affected by chronic pulpitis with an increase of the fibrous component, lymphatic vessels with very indented walls and irregular shape were present. They seem to be restrained by the dense bundles of collagen fibres (Fig. 3).

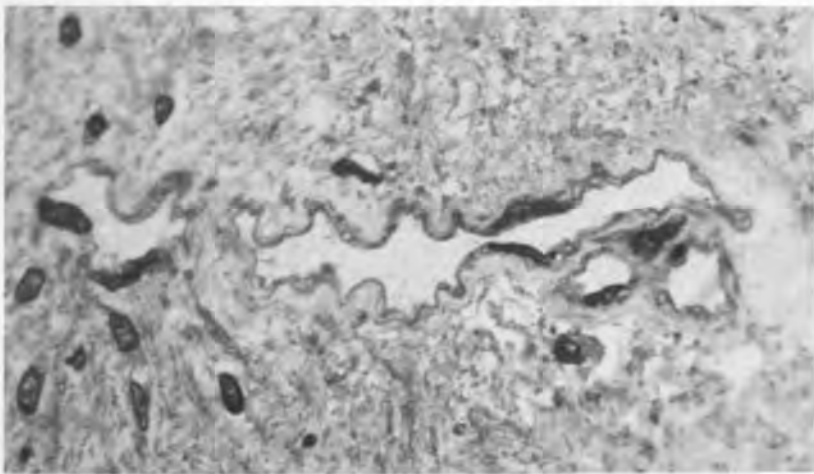


Fig. 3: Un vaisseau lymphatique contracté avec un contour très irrégulier est entouré par des tas de fibrilles dans une zone fibreuse de la pulpe.

1300 X

A contracted lymphatic vessel with a very irregular profile is surrounded by bundles of collagen fibrils in a fibrotic region of the pulp.

1300 X

At ultrastructural level in the inflamed pulpal areas large macrophagic cells with wide vacuoles were concentrated. Their vacuoles were clear and homogeneous or filled by granular or dense material

and sometimes by cristalline micro-structures (Fig. 4). In addition to these cells, lymphocytes and some plasmacells also appeared. Fusiform or irregularly shaped pulpal cells with an extended RER were identified among the inflammatory infiltrates.

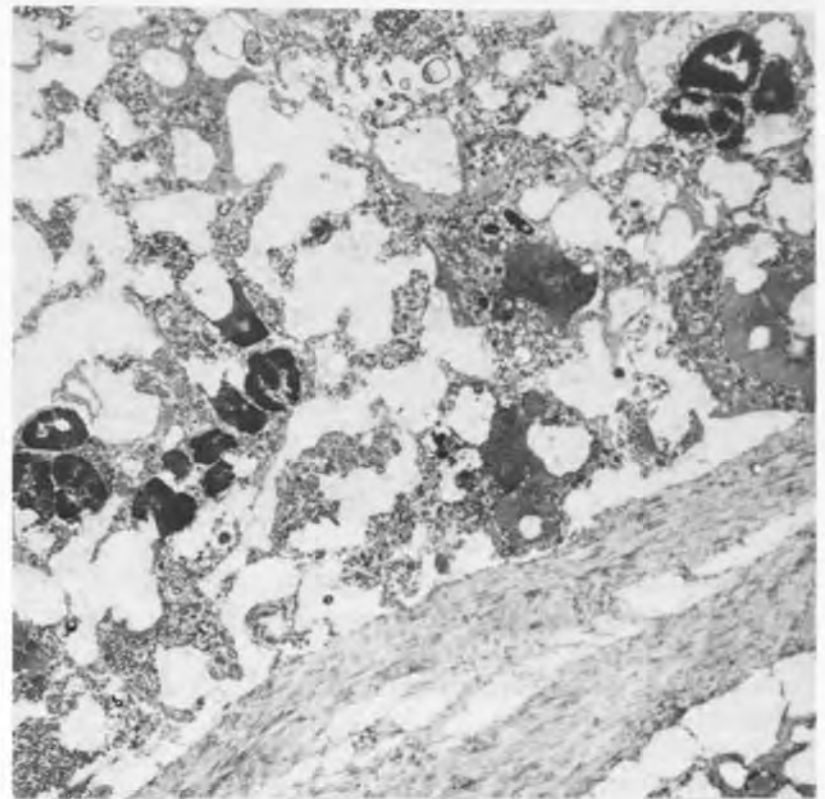


Fig. 4: Infiltration de macrophages et des faisceaux de fibrilles dans la pulpe dentaire enflammée.

3150 X

Macrophagic cellular infiltrates and bundles of collagen fibrils in inflamed dental pulp.

3150 X

Dilated lymphatic vessels displayed a thin distended wall, a rather regular profile without the luminal and abluminal endothelial projections that characterize the morphological properties of normal lymphatic vessels. The junctions between endothelial cells are generally not very intricate: end to end contacts or overlappings between endothelial edges were prevalent.

The main characteristics of these vessels was the presence of «open junctions» between the endothelial cells. These were frequently detectable in the vessel profile as focal gaps of the intercellular connections causing holes of 0,3-0,5 μ in the endothelial wall (Figs. 5-6).

The intraparietal endothelial channels, that are usual morphological features in the walls of normal lymphatic vessels, are absent but sometimes it was possible to observe endothelial cells partly overlapping delimiting sacs which were widely opened towards

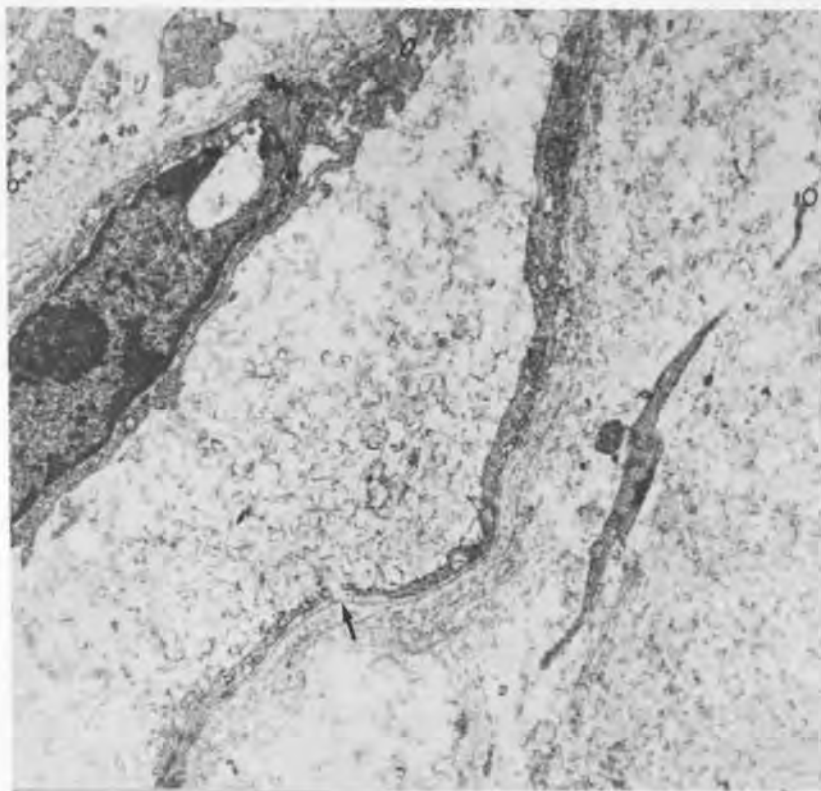


Fig. 5: Jonction ouverte (flèche) entre deux cellules endothéliales dans la paroi d'un vaisseau lymphatique dilaté. 12150 X

Open junction (arrow) between two endothelial cells in the wall of a dilated lymphatic vessel. 12150 X

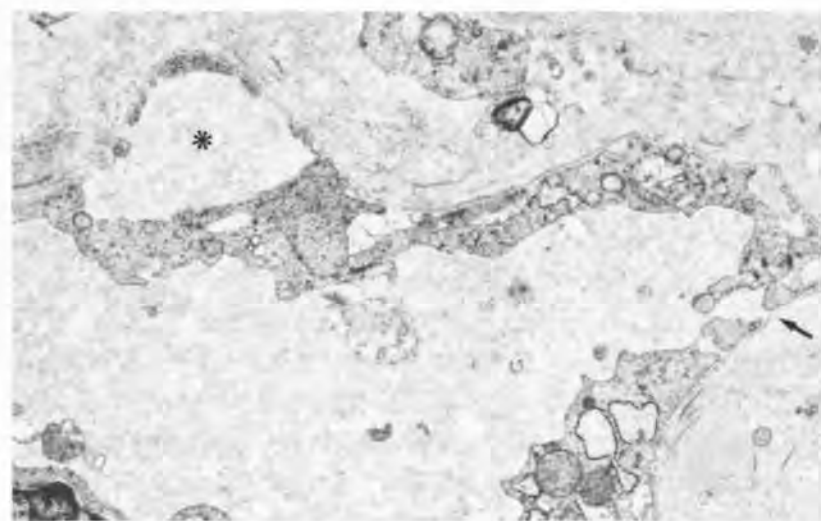


Fig. 6: Un restant d'un couloir intrapariétal en partie bordé à l'extérieur par les extrémités d'une cellule endothéliale. Le contenu (*) semblable au contenu granuleux du vaisseau manque de filaments et de fibrilles. Une jonction ouverte (flèche) est présente. 12150 X

A residue of an intraparietal channel partially bordered on the outside by edges of endothelial cells. The content () similar to the granular content of the vessel lacks filaments and fibrils. One open junction (arrow) is also present. 12150 X*

the extracellular matrix. These features probably represent modifications of the intraparietal channels in the adaptation of the endothelial cells to the distension of the vessel wall (Fig. 6).

The endothelial cells were almost totally lacking in micropinocytotic vesicles on both sides of the endothelial wall. Inside they were very rich in ribosomes and RER cisternae; thin filaments formed a diffuse cytoplasmic network.

A basement membrane which was thicker than normal was associated to the external profile of the endothelial wall. Anchoring filaments were scanty while several collagen fibrils formed a network which was closely related to the bundles of fibres of the extracellular matrix (Fig. 5-6).

The vessels that at light microscope didn't display distended walls at ultrastructural level confirmed they were not dilated. Indeed they lacked open junctions and the endothelial cells were connected by different and sometimes intricate adjoining structures. Also these vessels showed an absence of intraparietal channels and micropinocytotic vesicles whereas the RER was considerable in the cytoplasm of the endothelial cells. The basement membrane was thicker and sometimes duplicated and the network of fibrils was particularly dense.

In a pulp severely damaged by fibrotic degeneration, a very conspicuous number of Weibel-Palade bodies was present in some large lymphatic vessels. They were the prominent component of the endothelial cell cytoplasm.

DISCUSSION

Because of its structural characteristics dental pulp is particularly susceptible to interstitial fluid pressure variations due to its low compliance (Kim, 1985; Heyeraas, 1989). An abundant network of lymphatic vessels drain the extracellular matrix and contribute in maintaining the physiological pressure of the tissue. Our observations have been performed on dental pulps affected by extended and seriously inflammatory lesions because of the carious processes in the dentine.

In this tissue we have observed numerous dilated lymphatic vessels with distended walls among cellular infiltrates and fibre bundles. They are characterized by the presence of open junctions, gaps between contiguous endothelial cells, and by the lack of intraparietal channels and micropinocytotic vesicles that, on the contrary, are typical morphological features in normal pulpal lymphatic vessels (Marchetti and Piacentini, 1990). Intercellular adjoining structures, intraparietal channels and micropinocytotic vesicles are the structures of the endothelial wall which allow the passage of fluids

and various molecules from the interstitial spaces towards the vessel lumen in normal tissue conditions (Marchetti et al., 1987). With inflamed and oedematous states the increase of the interstitial fluid pressure needs a more intensive and quick drainage of the interstitium: the open junctions supply this functional need (Marchetti et al., 1988). The reduction in number of intricate intercellular junctions and the disappearance of intraparietal channels are explained by the distension of the endothelial cells because of the expansion of the vessel lumen. On the other hand we cannot justify the absence of micropinocytotic vesicles in the endothelial cells when open junctions are present between them.

In the pulpal regions where a fibrotic dense extracellular matrix surrounds and compresses the lymphatic vessels, the endothelial walls lack intraparietal channels and micropinocytotic vesicles as well as open junctions. Therefore these vessels do not seem to display any functional activity. Nevertheless this may be a temporary condition and the presence of numerous ribosomes and RER cisternae confirms the persisting functional activity of cytoplasmic structures of the endothelial cells.

The presence of a large number of Weibel-Palade bodies in a pulp affected by fibrotic pulpitis is probably related to degenerative processes of the endothelial wall as has already been demonstrated in obliterating blood vessels (Kagawa and Fujimoto, 1987).

From our observations it is evident that lymphatic system contribute to the drainage of the dental pulp tissue in normal and inflammatory states. Different morphological structures perform this function: the micropinocytotic vesicles, the intercellular adjoining areas and the intraparietal channels in the normal pulpal tissue and especially the open junctions in the inflamed regions. They represent structural adaptations of the endothelial walls to different conditions of the extracellular matrix.

BIBLIOGRAPHIE

- Bernik, S.** — Lymphatic vessels of the human dental pulp. *J. Dent. Res.*, 56: 70-77, 1977a.
- Bernik, S.** — Morphological changes to lymphatic vessels in pulpal inflammation. *J. Dent. Res.*, 56: 841-849, 1977b.
- Bishop, M.A., Malhotra, M.** — An investigation of lymphatic vessels in the feline dental pulp. *Am. J. Anat.*, 187: 247-253, 1990.
- Heyeraas, K.J.** — Pulpal, microvascular and tissue pressure. *J. Dent. Res.*, 64: 585-589, 1985.
- Heyeraas, K.J.** — Pulpal hemodynamics and interstitial fluid pressure: balance of transmucosal fluid transport. *J. Endodontics*, 15: 468-471, 1989.
- Kagawa, H., Fujimoto, S.** — Electron microscopic and immunocytochemical analyses of Weibel-Palade bodies in the human umbilical vein during pregnancy. *Cell. Tiss. Res.*, 219: 557-563, 1987.
- Karnovsky, M.J.** — A formaldehyde fixative of high osmolarity for use in electron microscopy. *J. Cell. Biol.*, 27: 137a-138a, 1965.
- Kim, S.** — Regulation of pulpal blood flow. *J. Dent. Res.*, 64: 590-596, 1985.
- Kukletova, M.** — An electron microscopic study of the lymphatic vessels in the dental pulp in the calf. *Arch. Oral. Biol.*, 15: 1117-1124, 1970.
- Marchetti, C., Poggi, P., Calligaro, A., Casasco, A.** — Ultrastructural study of the adjoining areas between the endothelial cells of the rabbit heart lymph vessels. *J. Submicrosc. Cytol.*, 19: 465-469, 1987.
- Marchetti, C., Poggi, E., Calligaro, A., Arbustini, E.** — Lymphatic system in human dilated cardiomyopathy. *J. Submicrosc. Cytol. Pathol.*, 20: 701-708, 1988.
- Marchetti, C., Piacentini, C.** — Examen au microscope photonique et au microscope électronique des capillaires lymphatiques de la pulpe dentaire humaine. *Bull. du GIRSO*, 33: 19-23, 1990.
- Van Hassel, H. J. and Brown, A. C.** — Effect of temperature changes on intrapulpal pressure and hydraulic permeability. *Arch. Oral Biol.*, 14: 301-315, 1969.
- Van Hassel, H.J.** — Physiology of the human dental pulp. In: *The biology of the human dental pulp*, Siskin, ed. Mosby C.V.Co. pp. 16-24, 1973.
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