

PECULIARITIES OF THE LYMPHATIC VESSELS DISTRIBUTION IN NORMAL AND AFFECTED ORAL MUCOSA

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Abstract: The immunohistochemical staining is employed the D₂-40 antibody in visualizing the lymphatic vessels in prepared tissue sections taken from various parts of the fixed or free (jugular, gingival, papillary, labial) oral mucosa. The lymphatic vessels form rich terminal networks in the superficial structures and in the subepithelium of the oral mucosa. The specific features of the lymphatic networks in both the normal and affected oral mucosa can help the research of the role played by the lymphatic system in a series of mucosal, periodontal or potentially malign conditions.

INTRODUCTION

The lymphatic circulation has been shown to impact the tissue fluids' homeostasis, by facilitating the interstitial transport of immunological-functioning proteins.

The fluids and proteins that leave the blood capillaries are collected from the interstitial spaces by the lymphatic capillaries and re-enter the blood circulation by means of larger-size lymphatic vessels' network. The presence of an edema signals an inflammatory process, in diseases that involve such a process as well. It occurs when the quantity of fluid that is drained by inflamed blood vessels exceeds the draining capacity of the lymphatic vessels. The oral mucosa provides for the exchanges with the “outside” and for resistance to the “outside.”

The functions of the mucosa presuppose the existence of complex vascular layout featuring numerous tiny blood vessels, with anastomoses that are capable of transferring larger or smaller molecules from the blood into the interstitial space. In the case of acute, subacute and chronic inflammatory processes, the result may be severe obstruction and the raise of the permeability of oral mucosa.

Clinical data also testify the considerable role of the lymphatic circulatory system in mucosal microcirculation. However, difficulties encountered in the proper identification of the respective lymphatic vessels have so far made the mapping of these vessels lag behind the one of the blood vessels.

The sustained comparative examination of the normal oral mucosa and microcirculatory disorders accompanying the lymph-node neoplastic dissemination is indicative of the efforts made in order to improve the means of visualizing the distribution of the lymphatic vessels.

PURPOSE

The purpose of the research paper is to immunohistochemically identify (detect) the lymphatic capillary vessels in the buccal mucosa.

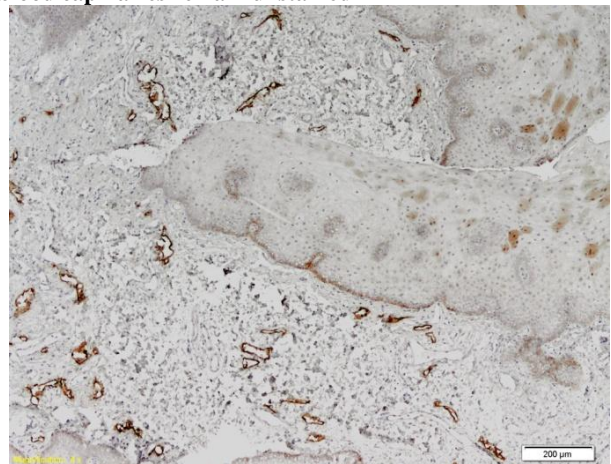
MATERIALS AND METHODS

The tissue specimens were immersed, overnight, in a freshly-prepared fixative of 4% paraformaldehyde in saline

solution, with 7-ph phosphorous buffer (PBS) that would allow for the immunohistochemical analysis, in observance of the standard procedures of detecting the D₂-40 and CD-31 antibodies. This revealed the presence of the lymphatic vessels in the tissues under scrutiny, identified by D₂-40 antigen staining. D₂-40 immunohistochemical staining evinces the presence of the endothelial cells that, from a morphological point of view, may be considered to be the lymphatic vessels.

In normal mucosa, the lymphatic vessels have been superficially visualized in the submucosal layer. A high density of these vessels has been in the surface mucosa and in the subepithelium. There are few such vessels next to the submucosal glands. Their distribution is rather horizontal. The small vessels which feature endothelial cells have positively responded to the D₂-40 marker. They are irregular in shape and have no structure. Their walls are thick and feature red cells in the lumen.(1,2,3)

Figure no. 1. In the mucosa section under scrutiny: in the corium one can observe how podoplanin has stained the endothelium of the lymphatic capillaries. Here and there, the lymphatic capillaries drain into pre-collecting vessels. The blood capillaries remain unstained



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CLINICAL ASPECTS

Figure no. 2. In the mucosa section under scrutiny: renders clear the positive marking of the lymphatic vessels' endothelium and the unmarked blood vessels. The basal stratum of the stratified pavementous epithelium has got poorly marked

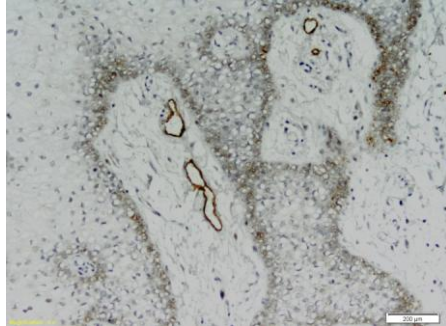


Figure no. 3. In the mucosa section under scrutiny: a more powerful objective lens shows several lymphatic capillaries whose endothelium is intensely positively-marked by podoplanin.

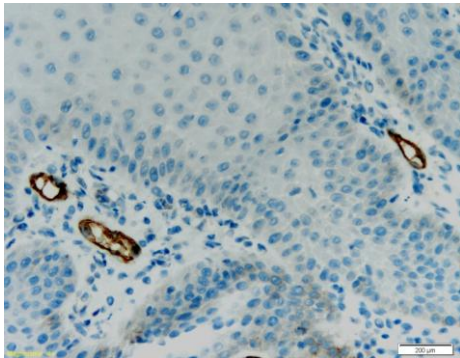
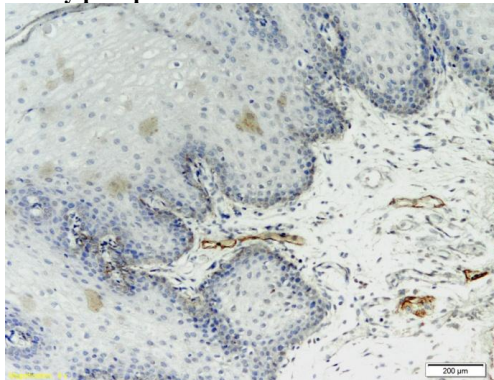


Figure no. 4. In the mucosa section under scrutiny: one can note the positive marking of the corium's lymphatic capillaries by podoplanin



RESULTS AND DISCUSSIONS

In the mucosa tissue specimens collected, the endothelium of the lymphatic capillaries got selectively stained by means of podoplanin (monoclonal D2-40 antibodies). The cutaneous tissue, the gut, and the lungs, i.e., the structures that get in touch with the environment (such as the mucosa, the skin) feature the highest exposure of the lymphatic system. So far, conventional H.E. staining has been unable to discriminate the lymphatic capillaries from the blood ones. However, researchers have resorted to antibodies which specifically colour the lymphatic vessels only and leave the blood vessels unstained. The D2-40 monoclonal immunostatin interacts with a

sialoglyco-protein linked to the MV 40,000 O-linked, which has proved to be a reliable lymphatic-vessel selective marker.

The lymphatic vessels have been identified by means of their positive reaction to D2-40 staining, whereas the blood vessels have not got coloured at all.

This antibody staining has been stable and it has shown the lymphatic vessels' major distribution in the superficial mucosal layer in the form of thin-walled channels that feature a continuous non-muscular-cell endothelial stratum.

The discrimination between the lymphatic capillaries and the blood ones can be also made in vivo by means of injecting the examined tissues with Indian ink (or neoprene latex) in order to facilitate better visualization or to render three-dimensional images of the respective structures.

The research made by Hoseman et al (1998) has shown, at nasal-mucosa levels, the existence of a network of superficial and deep-layer lymphatic vessels in the vicinity of the basal epithelial stratum, which are associated to the local glands.(4) The primordial vessels exhibit an endothelial layer, no smooth muscles and, in general, no valves and collectors. However, the collectors feature a contracting compartment (exhibiting smooth muscles) and are provided with valves.

The basic structure, as well as the origin of lymphatic vessels, can be interpreted in two ways.

The lymphatic vessels are able to collect and transport the interstitial fluid that contains proteins, colloids and, sometimes, metastatic cells. Our own research has evidenced the presence of lymphatic capillaries in the initial lymphatic network at diverse oral mucosal layers.(5,6)

CONCLUSIONS

The immunohistochemical results have revealed the origin of the lymphatic vessels, their structural basis and organization, which can be interpreted in several ways. One of these approaches claims that the lymphatic vessels are directly connected to the interstitial tissue.

The research that we have performed contributes new information on the physio-pathologic processes that occur in the oral-cavity mucosa.

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