Some Aspects of Functional Morphology of Extracellular Matrix in Lymph Capillary Endothelium

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Argyrophilic portion of endothelial glycocalyx in lymph capillaries from the central tendon of rabbit diaphragm was examined. It is shown that optical density along the perimeter of endotheliocyte is unequal and depends on the cell shape (scalloped, fusiform, or irregular). Aseptic inflammation is characterized by a marked increase in high-density zones and expansion of argyrophilic glycocalyx. It is proposed to consider argyrophilic glycocalyx as an independent structural and functional unit of the endothelium.

Key Words: *lymph capillary; endothelium; argyrophilic glycocalyx*

We found no publications on selective argyrophilia of the glycocalyx in cell-to-cell contacts within the endothelial layer of capillaries and vessels. Taking into account the high resorbtion activity of cell-to-cell contacts of lymph capillary endothelium, which is probably responsible for the glycocalyx argyrophilia, we explored the effect of some factors on the reactivity of argyrophilic zones.

MATERIALS AND METHODS

Central diaphragmatic tendons from rabbits weighing 2.5-3 kg were studied. The biomaterial was obtained from 30 control and 24 experimental animals (central tendon twice stitched with surgical silk under ether narcosis). The animals were decapitated 30 min—20 days after the start of the experiment. Analogous material was obtained from cattle killed at a slaughter house by draining the blood after electrical shock. The samples were impregnated 10-60 min after sacrifice. Lymph capillary endothelium was visualized by the method of Ranvier, the specimens being fixed in 10% neutral formalin. Cell boundaries were studied using a computer-assisted image analysis system on lymph capillary (postcapillary) wall area of 11861.72 μ^2 (pleural surface).

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RESULTS

In our previous studies [1] we distinguished 3 types of endotheliocytes with different shapes: scalloped, fusiform, or irregular (Fig. 1, *a*). In the present study we investigated these cell types. It was shown that these cells have different length of cell boundaries. Scalloped endotheliocytes had the longest perimeter: 341.9—166.8 μm (227.5±1.4 μm). Fusiform cells had a lower length of cell boundaries, which varied from 145.9 to 264.8 μm (211.5±0.98 μm). The minimum length of cell boundaries was observed in irregular endotheliocytes (138.8±1.15 μm); this can be attributed to its active locomotor rearrangement [3]. The difference between minimum and maximum lengths of cell boundaries was the most pronounced in irregular endotheliocytes (75.1 and 191.8 μm). If the perimeter of scalloped cells is taken as 100%, the perimeter of fusiform and irregular endotheliocytes constitutes 92.94 and 61.0%, respectively. From these data we concluded that different cell types have different resorption capacity. It can be assumed that

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Fig. 1. Endothelium of lymph capillaries of the central diaphragmatic tendons from a control rabbit (*a*) and on day 2 of aseptic inflammation (*b*). Cell boundaries are shown with arrows. Impregnation by the method of Ranvier, fixation with 10% formalin, ×200.

cells with most lengthy boundaries, primarily, scalloped cells have higher adsorption capacity.

Endotheliocytes had different impregnation capacity along their perimeters. This phenomenon is responsible for different optical density of argyrophilic glycocalyx around the cells. For instance, a scalloped endotheliocyte is surrounded by glycocalyx with an optical density ranging from 2.19 to 16.46%, i.e., differing 7.5-fold. Analogous relationships were noted in fusiform cells. Cell-to-cell contacts are presented primarily by low-density glycocalyx, particularly irregular endotheliocyte boundaries. In these cells, the total length of the low-density glycocalyx zones surpassed that of zones with heavy and moderate impregnation more than 16- and 1.5-fold, respectively. The total length of zones with moderate impregnation is practically the same in different types of endotheliocytes, being slightly higher in irregular cells than in other cell types. Generally, the total length of these zones is comparable to that of dense zones: these are 5-fold longer in scalloped cells and 5.4- and 10-fold longer in fusiform and irregilar cells, respectively.

These data suggest that first, resorption capacity is different in different endotheliocyte zones. The most intense resorption occurs in high-density zones, which under normal conditions constitute only minimal part of cell boundaries. Second, resorption capacity is to a great extent determined by endotheliocyte shape and structure of local cell-to-cell contacts [4,5]. Third, irregular endotheliocytes exhibit lower functional activity than fusiform and especially scalloped cells.

These suggestions were confirmed by aseptic inflammation experiments. It should be noted that the number of scalloped endotheliocytes increases and they become a predominant cell type. Argyrophilic component of different endotheliocytes responded to inflammation in the same manner. The most prominent feature is the increase in the high-density cell contacts: 4-fold in scalloped cells, and 3.5- and 3.8-fold in fusiform and irregular cells, respectively. Zones with moderate density increased to a lesser extent (2-fold in scalloped cells and by 30 and 20% in fusiform and irregular cells). These changes are accompanied by shortening of low-density zones, particularly in scalloped cells (2-fold).

Under the chosen experimental conditions, the total area of cell-to-cell contacts increased due to their thickening. The most pronounced changes were observed in fusiform cells (30% widened). In control animals, the total area of cell-to-cell boundaries of fusiform cells constituted 16.7% of a 11861-μm^2 standard area, while 6 h postoperation this parameter increased to 23.1%. In scalloped and irregular cells, these changes were less pronounced (from 19.4 to 20.3% and from 20.1 to 22.1%, respectively). Unequal extension of argyrophilic zones in different cell types under conditions of aseptic inflammation may reflect their reserve capacity. Argyrophilic component in capillaries with scalloped endotheliocytes is evidently the most specialized for resorption function and therefore is less altered. Fusiform endotheliocytes lining lymph vessels fulfill the sorbtion function and participate in lymph transport, which becomes more intense in inflammation. This may account for explains accumulation of argyrophilic glycocalyx around the fusiform cells. It should be noted that these changes were observed only in the peripheral zone of inflammation, while in the stitched area cell boundaries look like thin weakly impregnated lines (Fig. 1, *b*).

Different impregnation pattern of cell boundaries in lymph capillaries of healthy animals and its changes under pathological conditions attest to high functional lability of the endothelium. This is confirmed by the fact that cell boundaries cannot be visualized immediately after death (for instance, when animals are killed by bloodletting after electrical shock). Impregnation of lymph capillaries was attained only 20-40 min after death. There are data on the reaction of cell boundaries induced by various experimental interventions [6,7] and pathologies [2].

Taking into account the specific structure and reaction of extracellular argyrophilic glycocalyx in lymph capillary endothelium, we think it reasonable to consider this component as an independent structural and functional unit distinct from the remaining nonargyrophilic part of endothelium.

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