

Morphometric changes in the blood and lymphatic channels during inflammation

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Abstract. When the abdominal cavity is inflamed, the lymphatic channel, i.e. the system of lymph vessels and lymph nodes, which play an important role in protecting the body from infections and ensuring proper metabolism, may be affected. Inflammation of the lymphatic channel can lead to its enlargement, soreness and even the formation of purulent foci. The study of morphometric changes in the lymphatic channel during inflammation is an important task for a more detailed understanding of inflammatory processes occurring in the body. The results of our experiments showed that during inflammatory processes of the abdominal cavity there are changes in the blood and lymphatic vessels. Relative volumes of vessels of different types per unit significantly decreased compared to the indicators of the control group. In submucosa and muscularis the morphometry data confirm the fact of arterial vessels spasm, lymphatic vessel reduction and simultaneous persistent venous vessels expansion. In rats with the developed inflammatory process in lymphatic vessels the expansion of lymphatic slits and significant thickening of the wall of large lymphatic vessels was found, which is associated with an increase in the volume of blood plasma, increasing the load on lymphatic vessels as a drainage system of the body.

1 Introduction

The lymphatic channel plays an important role in maintaining immunity and providing drainage of excess fluid from tissues, it is formed from tiny lymphatic capillaries that are in proximity to blood capillaries [1]. The main function is to transport lymph, which contains immune system cells, antibodies and other important components [2]. After filtration, lymph enters the lymphatic vessels, which continue as lymphatic ducts. These join together and eventually flow into the venous system from where lymph returns to the blood and circulates through the body [3]. Any alteration in this system can lead to edema, immune response disorders and other diseases. In abdominal inflammation, the lymphatic channel, i.e. the system of lymphatic vessels and lymph nodes, which play an important role in protecting the body from infections and ensuring proper metabolism, may also be affected [4]. Inflammation of the lymphatic channel can lead to its enlargement, soreness and even the formation of purulent foci [5].

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The study of morphometric changes in the lymphatic channel during inflammation is an important task for a more detailed understanding of pathological processes occurring in the body. For example, increased lymphatic vessel wall thickness or increased lymph node size may indicate the presence of inflammation or fibrous tissue formation [6]. This may provide new insights into abdominal inflammation and its effect on the lymphatic system. The aim of our study is morphometric study of blood and lymphatic vessels in inflammation of abdominal organs.

2 Materials and Methods

2.1 Objects

The experiments were performed on male Wistar rats weighing 250 ± 5 g. All animals were kept in the same conditions at a constant ambient temperature of $23-26^{\circ}\text{C}$ and received the same nutrition. Three groups of rats were formed, 1st group - 10 control rats, 2nd group with inflammation of abdominal organs after 2 days (12 rats), 3rd group with inflammation of abdominal organs after 5 days (12 rats).

All groups of animals were kept under the same feeding and keeping conditions in the vivarium. All experiments performed on animals were conducted in strict accordance with the rules developed and approved by the Local Ethics Committee of the Institute of Genetics and Physiology, Protocol No. 12-314 from November 11, 2022, as well as the rules of bioethics approved by the European Convention for Protection of Vertebrates (Strasbourg, 1986) and the guidelines outlined in the European Union Directive 2010/63/EU of September 22, 2010, entitled "On protection of animals used for scientific purposes".

2.2. Animals and Experimental Design

We chose a method of modeling the inflammation of abdominal cavity organs, which is close to the acute inflammatory process that characterized the end of the acute phase of peritonitis by etiopathogenesis, clinical manifestations and phases of course to that in humans. Acute inflammation of abdominal cavity organs in rats was induced by injection of fecal suspension into the abdominal cavity at the rate of 0.5 ml of 10% solution per 100 g of animal body weight. Animals for the study were taken on the 2nd and 5th day after fecal injection.

2.3. Morphological Research

After narcotization of animals, for morphometric study of blood and lymphatic vessels, their pieces were cut out, from which, after fixation in 10% formalin solution, histological slices of vessels with thickness of 4-5 μm were obtained using Thermo Scientific HM 325 microtome. After standard wiring, paraffin sections were prepared and stained with hematoxylin-eosin [7]. Conducted by measuring the diameter of different parts of the blood and lymphatic channels were counted using morphometric grids [8]. It includes arteries (ART), lymphatic vessels (LV) and veins (VEN). We studied the number of these vessels per unit area (S) and their diameter in dynamics during abdominal inflammation. Digital images of histologic preparations were obtained using LEICA DM 750 microscopes with camera and LEICA Application Suite software. 75 preparations of blood and lymphatic vessels were examined.

2.4 Statistical analysis

Statistical processing of the obtained results was performed in Stat Plus Pro 2009 program (Analyst Soft, Inc.) using Student's t test. Data are presented as arithmetic mean (M) ± error of mean (±m). Differences were considered significant at $p < 0.05$.

3 Results and Discussion

Data of morphometric counting of the number of vessels of different types per unit area, their diameter in the wall of blood and lymphatic vessels. In the early stage of inflammation in the mucosa there is an indistinct dilation of venous vessels. In submucosa and muscularis sheath morphometry data confirm the fact of arterial vessels spasm, capillary blood flow reduction and simultaneous persistent venous vessels dilation (Fig. 1).

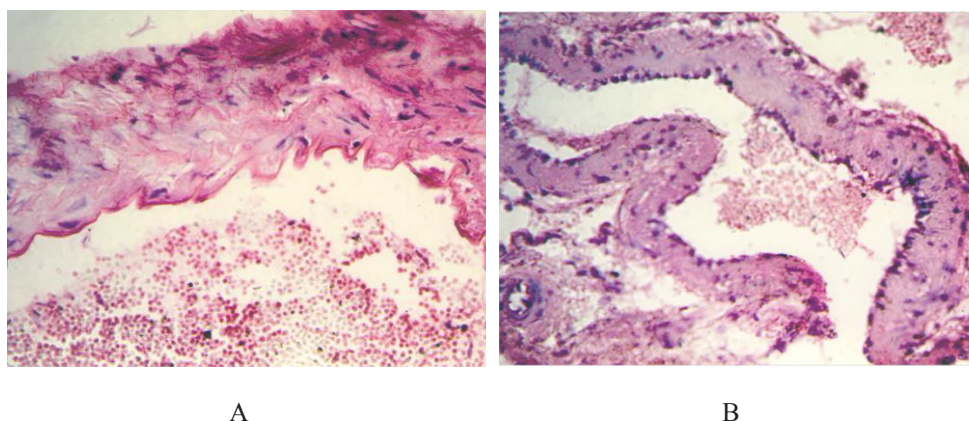


Fig. 1. Histologic structure of blood vessels after inflammation of abdominal cavity organs. Note: A – arterial vessel, B – venous vessel. Note: H&E stain. x 200.

On the 1st day from the beginning of the experiment in the mucous membrane the average diameter of veins significantly increases, while in lymphatic vessels, on the contrary, decreases. On the 2nd day after the abdominal cavity inflammation, paralytic dilatation of capillaries occurs in the mucous sheath. The average diameter of the vein is significantly greater than the similar indicator of the previous terms of the experiment. On the 5th day of the inflammatory period in experimental animals there was an increase in the severity of morphological changes: sharply pronounced diffuse polymorphous cellular infiltration of all membranes with predominance of neutrophils, destruction of a significant part of villi, diffuse hyperemia of vessels, focal diapedesis hemorrhages were observed. Morphological study of lymphatic and blood vessels in control animals revealed that their structure corresponds to that described in the literature [9, 10].

Morphological changes were revealed in lymphatic and blood vessels during abdominal cavity inflammation: atrophy in vessel walls and development of stasis characterized by dilatation of postcapillaries and veins, diapedesis hemorrhages, vagueness of microvessel contours (Fig. 1, B). The wall of large arteries shows thickening and tortuosity of the internal elastic membrane, edema of the subendothelial layer, flattening and focal desquamation of endothelial cells. Desquamation and proliferation of endothelial cells of a large artery, formation of a wall thrombus (Fig. 1, A).

In rats with the developed inflammatory process in lymphatic vessels there was found the expansion of lymphatic slits and significant thickening of the wall of large lymphatic vessels, which is associated with an increase in the volume of blood plasma, increasing the load on

lymphatic vessels as a drainage system of the body. In the thoracic duct, the lumen of lymphatic vessels is dilated (Fig. 2).

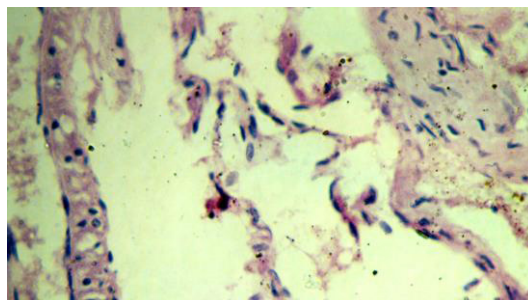


Fig. 2. Histologic structure of lymphatic vessels in abdominal inflammation. Note: H&E stain, x 200.

The picture of morphological changes in the lymphatic and circulatory systems in inflammation of the abdominal cavity indicates profound changes in hematopoietic, immune, barrier-filtration, transport and other functions of these organs. The obtained material shows that inflammation of abdominal cavity organs in experimental animals is accompanied by structural changes in lymphatic and blood vessels.

After inflammatory processes, 24 hours after the beginning of the experiment, there is an indistinct dilation of venous vessels (Ve) in the mucosa sheath. At the same time the diameter of lymphatic vessels (LV) decreases. Relative volumes of vessels of different types per unit S significantly decreased compared to the control group. In submucosa and muscular sheath morphometry data confirm the fact of arterial vessels spasm, reduction of capillary blood flow and simultaneous persistent vein dilation. Examination of relative vessel volumes showed that while the number of artery and lymphatic vessels per unit area decreased slightly, the number of veins increased significantly (Table 1).

Table 1. Mean values of diameters of artery, vein and lymphatic vessels of the abdominal cavity 2nd and 5th day after inflammatory processes, μm^2

Parameters	Number of vessels per unit area, S					
	Ar		Ve		LV	
	2nd day	5th day	2nd day	5th day	2nd day	5th day
Mucosa sheath	5.04±0.11	6.05±0.10	8.76±0.15	8.11±0.3**	12.7±0.4	11.1±0.16**
Submucosa sheath	6.06±0.14**	7.86±0.12**	23.25±0.22	25.84±0.19**	7.9±0.12	7.8±0.15**
Muscular sheath	2.06±0.11	4.09±0.13*	6.94±0.14	9.1±0.16**	4.07±0.14	5.9±0.12**
Parameters	Diameter of vessels					
	Ar		Ve		LV	
	2nd day	5th day	2nd day	5th day	2nd day	5th day
Mucosa sheath	31.56±0.34	39.49±0.6	1.69±0.19	21.48±0.17**	6.58±0.11	7.68±0.06*
Submucosa sheath	33.64±0.37	35.56±0.2**	18.12±0.07	20.89±0.03*	7.44±0.17	8.05±0.12*
Muscular sheath	41.59±0.39	43.24±0.33**	21.07±0.4	22.31±0.23**	6.19±0.3	7.84±0.13*

Note: *P < 0,01; **P < 0,001 - reliability of differences between indicators

On the 5th day after abdominal inflammation, paralytic dilatation of capillaries occurs in the mucous membrane. The average diameter of the vein is significantly greater than the

analogous index of the previous terms of the experiment. Relative volumes of capillaries and vein are increased in comparison with the control group. In the abdominal cavity the average diameter of artery, vein and lymphatic vessels is increased. The indices of relative volumes of different types of vessels per unit area also increase.

Paralytic dilation of capillaries of intestinal villi and crypts is combined with venous dilation, disturbances of rheological properties of blood and lymph in the form of microthrombosis and changes in permeability of vascular walls. The number of vessels per unit area practically does not differ from the indices after 2nd day abdominal inflammation. In comparison with the indicators after 5th day abdominal inflammation there is a decrease in the number of lymphatic vessels in the mucosa and submucosa, which reflects the uneven distribution of vessels.

4 Conclusion

The most significant morphometric features reflecting the dynamics of blood and lymphatic vessels in the inflammation of abdominal cavity organs were confirmed on the basis of the study. The picture of morphological changes in the lymphatic and circulatory systems during abdominal cavity inflammation indicates profound changes in hematopoietic, immune, barrier-filtration, transport and other functions of these organs. In modeling abdominal inflammation, arterial vasospasm is weakened. There is a paralytic dilatation of lymphatic vessels. Sharp dilation of venous vessels, impaired rheological properties of blood and lymph. In inflammation observed microthrombosis and changes in the permeability of vascular walls. Perivascular hemorrhages are determined around the vein. Similar changes are noted in the mesentery. Venous stasis aggravates ischemia. The most pronounced necrobiotic processes are observed in the mucosa. Violation of regional hemodynamics causes hypoxia both in the epithelial cover of intestinal villi and in the endotheliocytes of capillaries. Developing acute erosions are superficial: only the covering epithelium is destroyed and rejected. The occurrence of erosions is associated with the peculiarities of blood supply, which are the periphery of microcirculation.

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