Microscopic changes of the pancreas vessels in the dynamics after experimental thermal injury

Zykova N. P.
I. Horbachovsky Ternopil National Medical University, Ternopil, Ukraine

The problem of burns is relevant in the world and in Ukraine. Thermal injury is not limited to local tissue changes, but causes significant morphofunctional disorders in the organs and systems of the affected organism, the result of this factor is a violation of blood supply to organs, including the pancreas. The aim of this work was to establish the histological condition of the vessels of the pancreas after experimental thermal injury. The experiments were performed on 40 adult white rats. Burns were applied under thiopental-sodium anesthesia with copper plates at a temperature of 96-97° on the epilated surface of the skin of the back of animals for 15 seconds (the lesion area was 18-20 % of the body surface). Decapitation of animals was performed on 1, 7, 14 and 21 days of the experiment. Pieces of the pancreas were taken for histological examination and processed according to generally accepted methods. Histological sections 5-6 μm thick were stained with hematoxylin-eosin, impregnated with silver by the Gordon-Sweets method, as well as staining by the tricolor method of MSB (ORB). Histological specimens were examined and documented using an SEO SCAN light microscope and a Vision CCD Camera. In the early stages of the experiment revealed a violation of the blood supply to the body. The lumens of most arteries were found to be dilated, and the wall thickness increased due to hypertrophy of the smooth muscle cells of the media. Leukocyte infiltration was determined in the perivascular spaces. The lumens of the small and middle veins were dilated and filled with blood, with parietal thrombi in some of them. Capillaries are characterized by a moderate expansion of their lumens, stasis. There was swelling of the paravalvular spaces. Significant destructive-degenerative, inflammatory and sclerotic changes were observed in the later stages of the experiment. The walls of the vessels were moderately swollen. Smooth myocytes of the arterial media were hypertrophied, the cytoplasm was vacuolated, and contained pyknotically altered nuclei. Adventitia of vessels, especially veins, was swollen and fibrous. Occasionally there were violations of the wall of the components of the microcirculatory tract, which was accompanied by hemorrhage. There are single thrombosed vessels, spasmed arterioles, hemocapillaries with dilated lumens. The greatest changes, especially on the 21st day, were the adventitial membrane with the growth of collagen and reticular fibers. The blood supply to the vessels was moderate. Vessels of small diameter and hemomicrocirculatory tract were also significantly altered, their wall is deformed, thin, homogeneous. The lumens are dilated, with the formation of blood clots. Thus, in the early stages of the experiment there are adaptive-compensatory processes and initial destructive changes in the walls of the vessels of the organ; manifested by stasis, thrombosis, edema, deformation, infiltration of the wall and perivascular space. In the late period, significant destructive-degenerative changes in the structure of the walls of the vessels of the pancreas, accompanied by hemorrhage, inflammation, sclerosis of the wall and edema of the adventitia. These changes lead to a deterioration of blood supply and, consequently, a decrease in the functional activity of the body.

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burns leads to the recognition that the problem of thermal burns remains relevant today [9, 10, 21, 27, 28, 34, 38]. As a result, after a burn, the development of a burn disease is possible, which is accompanied by damage to all internal organs and systems of the body [1, 5, 7, 24]. Burn disease develops when 15-20 % of the body surface is damaged, and its severity is determined by the area and depth of damage to the skin. Burn disease has a complex multicomponent pathogenesis, characterized by hypovolemia, circulatory disorders, especially microcirculation, severe intoxication and infection of the body, including the pancreas [3, 4, 11, 17, 18, 20, 23, 25, 26, 35].

The aim of the study was to establish histological changes in the components of the circulatory system of the pancreas in the dynamics of experimental thermal trauma.

Materials and methods
The experiments were performed on 40 adult white rats. The burn injury was reproduced according to the methodology developed at the Department of Histology and Embryology of I. Horbachevsky Ternopil National Medical University Ministry of Health of Ukraine. The burn was applied under thiopental-sodium anesthesia with copper plates heated in boiled water to a temperature of 97-100°C on the epilated surface of the skin of the animal's back for 15 seconds. Pieces of the pancreas were taken for microscopic examination on days 1, 7, 14, and 21 of the experiment. Pieces of the pancreas of experimental animals were placed in certain clamps depending on the method of study [8, 13].

The pieces were fixed in 10 % formalin solution, then the pieces were dehydrated in alcohols of increasing concentration in a machine for histological treatment of AT-4 tissues, poured into paraffin blocks. Sections obtained on a sled microtome MS-2 with a thickness of 4-5 μm were stained with hematoxylin and eosin. Impregnated with silver by the Gordon-Sweets method to detect basement membranes, reticular and immature collagen fibers, as well as staining by the tricolor method MSB (ORB) - an elective staining method for intravascular coagulation syndrome [8, 24]. Histological specimens were studied using an SEO CCAN light microscope and photo-documented using a Vision CCD Camera with an image output system from histological specimens.

Results
Conducted microscopic studies showed that on the 1st day of the experiment there was a violation of the blood supply to the pancreas. There was an expansion of the lumen of blood vessels, their fullness and thickening of the walls. The lumens of the large and middle arteries are moderately blood-filled and dilated. Swelling of all wall membranes was detected, but to the greatest extent, wall thickness increased due to hypertrophy of smooth muscle cells of the media (Fig. 1).

The lumens of the huge and middle veins were also dilated and filled with blood, there was swelling of the perivascular spaces, there was a slight leukocyte infiltration of the adventitia. Arterioles had a narrowed, blood-filled lumen, a slight thickening of the wall. Due to the spasm of the arteriole wall, their capacity was reduced. In the hemocapillaries, dilation of the lumens and their sharp blood supply, existing stasis and sludge effect of erythrocytes were observed. The endothelium looked swollen, as a result of which its protrusion into the lumen of the vessels was observed.

After 7 days, an increase in alternative changes of vessels, which had an adaptive-compensatory character, was detected. There was a partial deformation of the wall of arteries of small and medium diameter, an increase in the thickness of the media due to hypertrophy and reduction.
of smooth myocytes, hyperchromic nuclei, edematous, moderate leukocyte infiltration. Perivascular edema and disorganization of connective tissue elements of adventitia were also observed. The lumens of the arteries remained blood-filled (Fig. 2).

The lumen of the venous vessels remained dilated and blood-filled, and their wall was characterized by destruction of the muscular membrane, edema of the perivascular spaces, leukocyte infiltration, to the greatest extent, the adventitial membrane. Among the arterial and venous units, vessels with the phenomenon of dystonia were observed, which was manifested by alternation of thickened and thinned sections of the vessel wall and deformation of their lumen. The vessels of a hemomicrocirculatory channel which were characterized by the stagnant phenomena in a gleam, thickening, hypostasis and destruction of a wall underwent the greatest changes, parietal thrombi were observed. However, vessels with thinned walls and sagging lumens were found. The lumens of most hemocapillaries are excessively dilated and blood-filled, perivascular edema was detected. Capillaries with obliterated lumen were noted in some places. Uneven protrusion of endothelial cell nuclei into the vascular lumen was observed, the basement membrane was indistinctly contoured.

Microscopically on the 14th day there was a significant remodeling of the vascular wall with increasing destructive changes, compared with the previous term. Most arteries (regardless of diameter) had a blood-filled deformed lumen and were characterized by moderate edema of the wall - thickening of the middle membrane due to smooth muscle elements and the initial signs of adventitial sclerosis, associated with increased activity of fibroblasts producing fibrous components of intercellular spaces. Smooth myocytes of the arterial media are hypertrophied, the cytoplasm is vacuolated, swollen, enlightened, and contains pyknotically altered nuclei. The intima of the vessels was also destroyed, the protrusion of endothelial cell nuclei into the lumen of the vessels was determined, sometimes their desquamation. The inner elastic membrane of the intima was swollen, thickened, in some areas homogeneous, blurred, fuzzy. Focal extravasal hemorrhages with violation of vascular integrity were determined (Fig. 3).

The venous part of the vascular bed at this time was characterized by moderate wall edema, compared with previous terms, thinning or hypertrophy of the media, sclerosis of the adventitia, which was manifested by the growth of collagen and reticular fibers. Large extravascular clusters of leukocytes were detected. Most hemocapillaries had large, blood-filled lumens with destructively altered erythrocytes, thrombosis, histoleukocyte infiltration of the paravasal spaces. The cells of the lumenal areas of the plasmalemma are swollen with fuzzy contours, the nucleus is elongated. In some places there was a violation of the vascular wall of the hemomicrocirculatory tract, which was accompanied by hemorrhages.

At day 21, histological changes were characterized by significant destructive-degenerative, inflammatory and sclerotic changes in the structural components of the walls of arterial and venous vessels. The blood supply to the vessels is moderate, their wall is destructively changed, which is manifested by thinning, thickening, leukocyte infiltration and significant sclerosis of the intima, media and, to the greatest extent, adventitia with complete obliteration of the lumen. It has been established that vessels of large and medium diameter are characterized by moderate blood supply, thickening of their wall due to leukocyte infiltration, hypertrophy of the muscular membrane, complete sclerosis, mainly adventitia. Vessels of small diameter with hypertrophied media, complete sclerosis of the wall and obliteration of the lumen (Fig. 4).

Fig. 3. Microscopic changes of vessels on the 14th day after burn injury. A. Artery of large diameter. 1 - blood-filled lumen, 2 - deformation and destruction of the wall, 3 - edema and infiltration of the adventitia. Staining by MSB (JAG). x200. B. Artery of large diameter. 1 - collagen and reticular fibers of the vessel wall, 2 - shaped elements in the lumen, 3 - violation of the tortuosity of the inner elastic membrane. Gordon-Sweets staining. х200.
Most vessels of the microcirculatory tract were characterized by moderate blood supply, edema of smooth muscle tissue, with partial atrophy of myocytes, complete sclerosis of the intima, media and adventitia, accompanied by the growth of collagen and reticular fibers. The lumen of some hemocapillaries are dilated, blood-filled with the formation of blood clots, but most capillaries had a narrow lumen, their wall is deformed, thin, homogeneous. Detachment of the endothelium from a homogeneous, indistinctly contoured, thickened basement membrane was observed. Perivascular leukocyte infiltration, sometimes diapedetic hemorrhages, with formation of inflammatory infiltrates are found.

**Discussion**

Structural changes of the pancreas, in particular, the vascular bed, as a reaction to the pathological process in the body, regardless of its origin, attracts attention, mainly due to the involvement of the gland in endocrine and exocrine function [3, 4]. At disturbance of blood circulation, the morphological changes which are followed by hypoxic changes which become rather defining in functional ability of organ grow. Vessels of the hemomicrocirculatory tract play an important role in the regulation of blood flow and outflow from the organ, tissue perfusion [7, 37, 39].

A significant amount of scientific work is devoted to the study of microscopic changes in the pancreas, both organs and its individual components, in particular the bloodstream. The structural organization of blood vessels corresponded to the data described in the literature on the peculiarities of their structure in white rats, other animals and humans [6, 17, 25].

According to the results of the study of the vascular bed of the pancreas in the simulation of hypertension [32] found that age-related changes in the arterial link of rats in normal and in the simulation of trauma, showed an interdependence between rat age and trauma, manifested in decreased vascular lumen, endothelial damage, remodeling of the walls, which leads to a violation of vascular capacity. Other authors [21] studied the state of the bloodstream during dehydration of the body, and showed that destructive changes in the microcirculatory tract do not depend on the duration of the experiment, but progress depending on the severity of dehydration. The results of studies in hypothermia of the pancreas [2, 15, 29, 30, 33], showed that superficial and shallow hypothermia did not cause significant morphological changes in the bloodstream, but with increasing duration of hypothermia caused more significant changes. Capillaries and veins are sharply filled with swollen walls, dilated lumen, the formation of a "sludge" effect. Most of these data are consistent with the results of our research. According to the results of research, the effect of heavy metals [14, 16, 36] on the gland in 30-, 60-, and 90-day terms was observed by dilation and edema of blood vessel walls and their overflow with blood, intense lymphohistocytic inflammatory infiltration, growth of connective tissue elements forming "fields" of sclerosis, which is identified with the results of our research. The study of the effect of narcotic substances [19, 22, 31] on the vessels of the pancreas had a similar character, which was manifested by edema, infiltration and profound destructive changes in blood vessels.

Thus, the results of our own research and data from the scientific literature indicate that due to the action of various exogenous factors and stressors on the body of experimental animals develop changes in the arterial and venous vascular bed and vascular links of the hemomicrocirculatory tract.

Further studies are planned to determine the structural changes in the components of the blood vessels of the pancreas after severe thermal trauma to the skin and under conditions of correction.

**Conclusion**

Histological examinations of pancreatic vessels after experimental thermal trauma revealed that in the early stages...
of the experiment there are adaptive-compensatory processes and initial destructive changes in the walls of vascular organs, manifested by stasis, thrombosis, edema, deformity, infiltration of the wall and perivascular space. In the late period, significant destructive-degenerative changes in the structure of the walls of the vessels of the pancreas, accompanied by hemorrhage, inflammation, sclerosis of the wall and edema of the adventitia are found. These changes lead to a deterioration of blood supply and, consequently, a decrease in the functional activity of the organ.

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