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Restorative processes of the vascular bed of the brain in the progressive course of ischemic stroke

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ABSTRACT

The objective is to study morphometry of the restoration processes of the cerebral vascular bed in the progressive course of ischemic stroke. We studied the structure of the vessels and parenchyma of the brain of patients who died of ischemic stroke in the angio-neurological department of the National Hospital under the Ministry of Health of the Kyrgyz Republic. With a progressive course of ischemic stroke, brain vessel lesions are characterized by peculiar changes in the shape and contents of the vessels, different from the vessels surrounding the ischemic area, and tissues of the opposite hemisphere. The study showed that reparative processes appear early and are expressed by a change in the number of capillaries, angiogenesis and recalibration of existing vessels. With a progressive ischemic stroke, dystrophic processes prevail over the compensatory-adaptive reactions of the central nervous system, while the latter are insufficient to restore lost functions.

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INTRODUCTION

Worldwide, stroke is the second leading cause of death (Go *et al.*, 2014). Ischemic strokes are the most common, the rest is hemorrhagic that has cerebral and subarachnoid (Patel and White, 2011) .

Acute ischemic stroke is classified into five stages

1) large artery atherosclerosis

2) cardio-embolism

3) small vessel occlusion

4) stroke of other determined etiology

5) stroke of undetermined etiology (Adams *et al.*, 1993).

In recent decades, questions related to elucidating the features of the pathogenesis of cerebral infarction, which were formulated as the most relevant at the moment, have attracted significant attention from researchers. Numerous recent reports

have been devoted to studying the vascular mechanisms of the occurrence and development of cerebral infarction, as well as structural changes in the vascular system. However, they mainly describe large superficial and intracerebral vessels, while the pathomorphological reactions of the vessels of the microvasculature are less studied (Arnao *et al.*, 2016; Lesser *et al.*, 2008).

Until now, the questions of the correspondence of precise pathological shifts to specific periods of the course and development of ischemic brain damage, especially the changes in the vascular bed depending on the clinical nature of ischemic stroke, as well as reparative and compensatory-adaptive processes of the vascular system for this type of brain pathology remain inadequately examined.

Objective

To study morphometry of the restoration processes of the cerebral vascular bed in the progressive course of ischemic stroke.

MATERIALS AND METHODS

The work was performed on sectional material using light-optical and morphometric methods. We studied the structure of the vessels and parenchyma of the brain of patients who died of ischemic stroke in the angio-neurological department of the National Hospital under the Ministry of Health of the Kyrgyz Republic. The study comprises of 18 sectional cases. The autopsy of deceased patients was carried out within 6-12 hours after stating dead. Atherosclerosis of the precerebral and (or) intracerebral vessels, hypertension, and a combination of these diseases was the initial pathology for cerebral infarction. Intracranial atherosclerotic stenosis of the major arteries (intracranial internal carotid artery, middle cerebral artery, vertebral artery, and basilar artery) is the most common proximate mechanism of ischemic stroke worldwide. (Gorelick *et al.*, 2008). The studied groups were selected in such a way that they presented different periods (from 12 hours to 24 days) of the course of cerebral infarction: from the moment of occurrence to the time of death. Brain tissues of people of the same age and gender who died in an accident served as control.

The brain areas of the affected area, from adjacent areas, as well as from the corresponding zones of the opposite hemisphere, were examined. The brain tissue was fixed in buffered formalin (according was dehydrated in ascending alcohols with enlightenment in xylene, and was poured into paraffin wax). Sections for light-optical and morphometric studies, 5–7 μm thick, were stained with hematoxylin and eosin, van Gieson's picrofuchsin, Weigert fuchsin, Schiff-iodic acid reagent, and Nissl cresyl violet.

In this study, the distribution density of microvessels, their diameters, and the cross-sectional area of the vascular bed were morphometrically studied. The degree of venous congestion was determined by calculating the total area of the standard number of veins (at least 20 for each observation).

RESULTS AND DISCUSSION

The most typical for the early (13-24 hours) period of cerebral infarction development changed in the shape and contents of small vessels. In the focus of ischemia, erythrocytic, fibrin-free platelet clots with granulocytes were expressed, obstructing the capillaries, arterioles and venules; a large number of capillaries containing plasma without blood cells; a sharp expansion of some capillaries with thinning of the walls, and an equally severe narrowing of others. Two variants of changes were equally common: concentric contraction with a thickening of the endothelial cells and flattening with a simultaneous thinning of the latter. The same process around neurons was expressed to a much lesser extent. By the end of the first day, a pronounced leukocyte reaction, leukocyte thrombi with an admixture of other formed elements, erythropoiesis, destruction of red blood cells in some erythrocyte thrombi with the formation of hyaline thrombi were determined. In some capillaries, phenomenon of necrobiosis and wall necrosis were detected.

Morphometric studies showed a decrease in the number of capillaries in the affected area on the 1st day by 23.1% against the control value, the diameter of vessel lumen of the microvasculature decreased by 8.6% and their cross-sectional area by 1.8 times. In this case, the lumen of the veins increased by 7.1%, which led to an increase of 14.7% in the total area of the standard number of veins Table 1. In the initial period of the cerebral infarction, the structure of the brain substance is still preserved. Necrobiotic changes were detected primarily in nerve cells. Among the latter, cells with ischemic and homogenizing changes were found; along with simple cytolysis and acute swelling, neurons with hyperchromatic and severe modifications were found. In a study, all these are acute and occur with rapidly developing insufficient blood supply (Castillo and Noya, 1999).

In the periphery of the affected zones to the cerebral cortex, as well as in the areas of the unaffected opposite hemisphere, histological changes corresponding to the characteristics of each particular case were revealed, depending on the degree

Table 1: Morphometric indicators of the vascular bed in the affected area (M±m)

Indicators	Dates of study				
	Control	1^{st} Day	5^{th} Day	14^{th} Day	24^{th} Day
Number of capillaries (per 1 μm^2)	284,9±29,3	219,1±16,5*	277,4±36,4*	132,9±14,0*	132,9±13,7*
Diameter of capillaries (μm^2)	9,4±1,2	8,6±0,5*	9,7±0,7*	11,5±0,8*	8,7±0,7*
Sectional area of capillaries (μm^2)	69,6±18,3	58,4±4,1	78,7±12,8*	103,4±10,4*	56,8±9,8
The total cross-sectional area of the veins (x103 μm^2)	32,9±0,8	37,2±0,3*	42,4±0,5*	34,4±0,8*	39,0±1,5*

* -indicators significantly differ from control at p <0.05

Table 2: Morphometric indicators of the vascular bed in the peripheral zone (M±m)

Indicators	Dates of study				
	Control	1^{st} Day	5^{th} Day	14^{th} Day	24^{th} Day
Number of capillaries (per 1 μm^2)	284,9±29,3	171,4±10,5*	246,8±13,0*	143,5±15,5*	132,9±14,0*
Diameter of capillaries (μm^2)	9,4±1,2	12,5±1,5*	8,6±0,3*	8,3±0,9*	8.7±0,8*
Sectional area of capillaries (μm^2)	69,6±18,3*	122,7±9,8*	60,5±4,9*	54,1±14,2*	65,8±9,7*
The total cross-sectional area of the veins (x103 μm^2)	32,9±0,8	53,7±1,8*	26,9±1,0*	46,5±1,4*	48,8±1,8*

* -indicators significantly differ from control at p <0.05

Table 3: Morphometric indices of the vascular bed in the contralateral hemisphere (M±m)

Indicators	Dates of study				
	Control	1^{st}Day	5^{th} Day	14^{th} Day	24^{th} Day
Number of capillaries (per 1 μm^2)	284,9±29,3	170,5±10,4*	439,5±45,5*	188,9±17,5*	207,9±9,7*
Diameter of capillaries (μm^2)	9,4±1,2	10,5±0,4*	11,3±1,9	7,7±0,4*	10,3±1,6*
Sectional area of capillaries (μm^2)	69,6±18,3	96,1±6,7	134,4±43,2	50,3±5,4*	125,8±36,6*
The total cross-sectional area of the veins (x10^3 μm^2)	32,9±0,8	42,9±2,6*	34,7±2,1	37,1±2,5*	44,4±1,7*

* -indicators significantly differ from control at p <0.05

of postmortem autolysis, the duration and severity of the agonal period, the duration of the resuscitation period. The capillary distribution density in the peripheral zone decreased by 39.8%, and the lumen and cross-sectional area of the vessels of the microvasculature were 32.8% and 26.3% respectively, which is higher than the control group. Veins lumen is widened, in combination with the revealed intra-arterial plethora, periarterial oedema, plasma impregnation of perivascular spaces, testified to the development of acute venous stasis. They were confirmed by the total area of veins, increasing to 53.7 $\pm$ 0.6 x 103 μm " (Ovbiagele *et al.*, 2013).

In the medulla of the cortex, opposite to the pathological focus of the hemisphere, pronounced pericellular and perivascular oedema, small foci of liquefaction of the primary substance were determined. In many small and medium arteries, changes in the vascular walls characteristic of hypertension were detected: thickening and sclerosis of the wall, hyperelastosis in combination with stratification of the elastic membrane. In individual vessels, mild endothelial proliferation was observed. In the microvasculature, an expansion of the microvessel lumen was detected morphometrically by 11.6% against the control, which was 22.1% higher than the index in the lesion, the cross-sectional area of the vessels increased 1.4 and 1.6 times, respectively. Moreover, the numerical density of the functioning vessels was lower than the control by 40.2%. The lumen of the veins is narrowed in comparison with the peripheral areas but had a larger diameter than the same vessels of the focus. The total area of the veins reached 42.9 $\pm$ 2.6 $\times$ 103 μm^2.

On the 5th day of the course of cerebral infarction, numerous granular debris appeared in the necrotic area, which was adjacent to the walls of the proliferating capillaries. Around the vessels with restored blood flow, uneven oedema persisted, a productive focal reaction of endothelial cells was found in their walls. Autolytic changes developed in previously necrotic vessels, weakly contoured shadows of arterioles were revealed, the contents of collapsed capillaries looked like narrow ribbons, squeezed by perivascular oedema. Morphometrically, an increase in numerical density, expansion of the lumen of capillaries, and a growth in the total cross-sectional area of veins were determined during this period Table 1. The brain tissue in the focus of necrosis was a structureless crumbling mass.

In the peripheral zones to the affected area, blood hypoperfusion and capillary collapse were determined. Significant perivascular oedema was observed around all the vessels. The focal proliferation of microvasculature was determined. The numerical density of capillaries in this zone was higher than the previous term. A marked constrictor reaction of both microvessels and more massive vascular formations were noted, which led to a decrease in the cross-sectional area of the bloodstream Table 2. In the parenchyma of the brain, annular perivascular necrosis, liquefaction, and pericellular oedema were observed. Deep ischemic changes or complete karyocytolysis of neurocytes developed. Glial cells with pyknotic nuclei, granularly clumpy inclusions in the cytoplasm and gliocyte karyocytolysis phenomenon were determined.

Generalized stasis was determined in the microcirculatory vessels of the contralateral unaffected hemisphere on the 5th day of the development of ischemic infarction with the following characteristics: diapedesis of red blood cells, plasmorrhagia. The vessels of larger caliber characterized by thickening, swelling of the endothelial cells, hyalinosis or plasma impregnation of the vessels, narrowing of the lumen of individual vessels due to fibrosis of the entire wall. The numerical density of capillaries was 1.5 times higher than the control indicator, a significant increase in their gaps was observed, up to 11.3 $\pm$ 1.9 μm, which led to an increase in the cross-sectional area by 1.9 times. There were a persistent plethora of veins. However, morphometric indicators slightly exceeded the control value Table 3.

Numerous perivascular necrosis, liquefaction zones, and small foci necrosis were revealed in the brain parenchyma. To a large extent, perivascular and pericellular oedema was expressed. Attention was drawn to the presence of a significant number of hyperchromic neurocytes with intensively basophilic cytoplasm and a pycnotic nucleus. There were individual neurocytes with ischemic and homogenizing changes encapsulated by glial cells. Later, such neurons are likely to undergo neuronophagy (Seners and Baron, 2018).

Dystrophic changes represent morphological changes in neuroglia, necrobiosis and destruction of part of gliocytes, mainly located away from neurocytes and blood vessels, by hypertrophy of preserved glial cells.

Large cerebral infarcts, by the end of the second week of their development, necrosis of proliferating capillaries including arterioles and veins, were found in the epicentre of the foci. The decay zones of the capillary walls were located not only in the centre of the lesion but also on its periphery, that is, in each lesion it is mosaic. The same was true for vascular proliferation zones. On the 14th day of the

development of ischemic infarction in many vessels of the brain substance, thickening, stratification and collagenization of the vascular walls, obliteration and recanalization of the lumens were observed. Around the altered vessels, there were extensive perivascular oedema, tissue cribrosity. The number of vessels of the microvasculature in the lesion decreased by 2.1 times compared with the control, a dilatory reaction of capillaries was observed, the average cross-sectional area of which increased by 1.5 times.

The lumen of the veins is narrowed, the indicator of their total area testified to the weak development or absence of venous stasis in this area of the brain Table 1. The substance of the brain in the lesion was sharply edematous, cirrhotic, wedge-shaped foci of incomplete necrosis were detected at the stage of resorption of decay products with the presence of signs brain substance organization. Healthy nerve cells in this zone were not detected at all. Only individual ischemic neurocytes remained visible. Focal hyperplasia and hypertrophy of glial elements, often wearing a perivascular character, attracted attention. In the peripheral zones to the focus, after two weeks of the development of ischemic infarction, decayed vessels were determined in large numbers. In the capillaries, generalized stasis, erythrocyte diapedesis, and plasmorrhagia were observed. In vessels of a larger caliber, thickening, swelling of endothelial cells, hyalinosis or plasma impregnation of the walls of the vessels, narrowing of the lumen of individual vessels due to fibrosis of the entire wall were detected. The numerical density of the functioning capillaries significantly decreased and amounted to 50.4% of the control indicator, the diameters and cross-sectional area of the vessels decreased by 11.8% and 22.2%, respectively.

A marked plethora of veins was noted, the total area of which increased to 46.5 $\pm$ 1.4 x 103 μm 2. Focal incomplete necrosis around collapsed blood vessels, small foci of complete necrosis around thrombosed and obliterated blood vessels were determined in the substance of the brain. The number of neurocytes is reduced, in a large number of ischemic neurons, the residual bodies of dead nerve cells were observed. At the same time, in some areas, hypertrophy of neurons and their nuclei was observed. Focal hypertrophy of the nuclei of glial cells, the proliferation of gliocytes, most pronounced near ischemic altered neurons, an increase in satellite up to 6-8 gliocytes per neuron were detected.

In the contralateral hemisphere, similarly to the peripheral zones, collapsed bloodless capillaries were revealed, the lumens of many of them were obstructed by the swelling nuclei of endotheliocytes. Small vessels had thickened vascular walls, often with their hyalinosis. In large vessels, fibrosis of the walls, cholesterol deposits, around which proliferation of fibrocyte-like cells was detected. The numerical density of the vessels of the microvasculature was 66.3% of the control. Moreover, the constrictor reaction was more pronounced: their diameter and cross-sectional area were reduced by 11.8% and 27.7%, respectively. There was a plethora of vessels of the venous link, an increase in the total area of the veins Table 3. Microscopic foci of brain tissue necrosis were found in the cortex. Mild diffuse hyperplasia of glial elements was observed.

Up to 24 days after the development of ischemic stroke, the affected area after the first stroke was represented by a gliomesodermal scar with a large number of collagen fibres. The tissues surrounding the scar and the contralateral territories contained a significant amount of vessels with wall changes characteristic of hypertension in the sclerotic stage. A sharp decrease in the lumen of the vessels due to thickening of intima, hypertrophy and sclerosis of the muscle layer was noted. Preservation of reduced blood supply in the outbreak and peripheral zone was expressed in a decrease in the numerical density of capillaries, lumen and cross-sectional area of blood vessels. In the contralateral hemisphere, there was a tendency to increase the numerical density of blood vessels, the dilatory reaction of capillaries, signs of venous congestion were revealed Table 1,Table 2 and Table 3. RCTs have shown conclusively that patients treated in dedicated stroke units fare better than those treated in general wards without the same ready access to personnel trained and experienced in managing stroke (Indredavik *et al.*, 1991; Jørgensen *et al.*, 1995). The spectrum of stroke syndrome in these studies included both ischemic and hemorrhagic stroke, ranging from mild (transient ischemic attack and minor stroke) to more severe (Zhu *et al.*, 2009).

Small focal zones of complete and incomplete necrosis were determined in the substance of the brain. The marked focal proliferation of gliocytes, satellite. Focal neuronal prolapse was observed, a significant number of shadow cells. Along with this, giant pyramidal neurons were detected, the area of which was increased 5-8 times compared with ordinary neurocytes. The processes in these cells were significantly thickened, enlarged in the volume of the nucleus and nucleoli, and a clumpy basophilic substance was determined in the cytoplasm. The hypertrophy of these cells undoubtedly reflects compensatory-adaptive processes that provide increased functional load in conditions of a

sharply reduced number of neurocytes (Seners and Baron, 2018).

Thus, the study showed that the early stages of the development of ischemic brain damage are characterized by peculiar changes in the shape and contents of the vessels of the microvasculature, which differ from the areas surrounding the ischemic focus, and tissues of the opposite hemisphere. With various forms of progression of ischemic brain damage, the reaction of the microvasculature has its own characteristics. Besides, a peculiar dynamics of changes in the numerical density, lumen and cross-sectional area of the vascular bed is revealed. The total cross-sectional area of the standard number of veins can be used as a diagnostic indicator of the degree of development of venous stasis, detected in varying degrees of severity at all observation times.

It was revealed that with ischemic stroke, reparative processes appear early and are expressed in a change in the number of capillaries, angiogenesis and recalibration of existing vessels. The slower development of ischemic stroke allows the compensatory mechanisms detected by morphological methods to appear earlier. In ischemic stroke, dystrophic processes prevail over compensatory-adaptive reactions of the central nervous system, while the latter is insufficient to restore lost functions.

CONCLUSION

Besides, the study showed that in response to the occurrence of a vascular lesion, destructive changes occur in the unaffected hemisphere of the brain. After the elimination of the acute period of the stroke, the processes of adaptive reactions aimed at restoring impaired functions begin to activate. They are based on reparative and compensatory-recovery processes (hypertrophy of neurons, glial element hyperplasia, proliferation and recalibration of blood vessels, etc.). However, these processes are not always sufficient and correlate with the clinical course of the disease.

Conflict of Interest

None.

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REFERENCES

Adams, H. P., Bendixen, B. H., Kappelle, L. J., Biller, J., Love, B. B., Gordon, D. L., Marsh, E. E. 1993. Classification of subtype of acute ischemic stroke. Definitions for use in a multicenter clinical trial. TOAST. Trial of Org 10172 in Acute Stroke Treatment. *Stroke*, 24(1):35–41.

Arnao, V., Acciarresi, M., Cittadini, E., Caso, V. 2016. Stroke incidence, prevalence and mortality in women worldwide. *International Journal of Stroke*, 11(3):287–301.

Castillo, J., Noya, M. 1999. Mechanisms of progression of cerebral infarction. *Neurologia*, 14(2):2–12.

Go, A. S., Mozaffarian, D., Roger, V. L., Benjamin, E. J., Berry, J. D., Blaha, M. J., Dai, S., Ford, E. S., Fox, C. S., Franco, S., Fullerton, H. J., Gillespie, C., Hailpern, S. M., Heit, J. A., Howard, V. J., Huffman, M. D., Judd, S. E., Kissela, B. M., Kittner, S. J., Lackland, D. T. 2014. *Heart disease and stroke statistics-2014 update: a report from the American Heart Association*, 129:28–292.

Gorelick, P. B., Wong, K. S., Bae, H. J., Pandey, D. K. 2008. Large Artery Intracranial Occlusive Disease: A Large Worldwide burden but a relatively neglected frontier. *Stroke*, 39(8):2396–2399.

Indredavik, B., Bakke, F., Solberg, R., Rokseth, R., Haaheim, L. L., Holme, I. 1991. Benefit of a stroke unit: a randomized controlled trial. *Stroke*, 22(8):1026–1031.

Jørgensen, H. S., Nakayama, H., Raaschou, H. O., Larsen, K., Hübbe, P., Olsen, T. S. 1995. The Effect of a Stroke Unit: Reductions in Mortality, Discharge Rate to Nursing Home, Length of Hospital Stay, and Cost. *Stroke*, 26(7):1178–1182.

Lesser, T., Venth, S., Lesser, K. 2008. Progressive Stroke with occlusion of the common carotid artery-An indication for revascularization. *Zentralbl Chir*, 133(04):374–375.

Ovbiagele, B., Goldstein, L. B., Higashida, R. T., Howard, V. J., Johnston, S. C., Khavjou, O. A., Lackland, D. T., Lichtman, J. H., Mohl, S., Sacco, R. L., Saver, J. L., and, J. G. T. 2013. Forecasting the Future of Stroke in the United States: A Policy Statement From the American Heart Association and American Stroke Association. *Stroke*, 44(8):2361–2375.

Patel, R. A., White, C. J. 2011. Acute ischemic stroke treatment: State of the art. *Vascular Medicine*, 16(1):19–28.

Seners, P., Baron, J.-C. 2018. Revisiting 'progressive stroke': incidence, predictors, pathophysiology, and management of unexplained early neurological deterioration following acute ischemic stroke. *Journal of Neurology*, 265(1):216–225.

Zhu, H. F., Newcommon, N. N., *et al.* 2009. Impact of a Stroke Unit on Length of Hospital Stay and In-Hospital Case Fatality. *Stroke*, 40(1):18–23.