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CEREBRAL MICROCIRCULATION DISORDERS IN EXPERIMENTAL CIRCULATORY HYPOXIA

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In experimental studies concerning the damaging effect of oxygen insufficiency on the central nervous system the assay of its blood supply is one of the most important problems. Systemic hypoxia, regardless of its origin leads to generalized hemodynamic disorders, which are also reflected on the state of the nervous system, despite the autonomic mechanism controlling brain circulation (Kowada et al., 1968; Mchedlishvili, 1973).

The effects of systemic blood pressure reduction have their pathological manifestations in the central nervous system, the character and topography of which depend on the regional vascularisation and metabolic properties of particular areas of the brain (Vogt, Vogt, 1922; Scholz, 1963; Ridge, 1967; Lampert, 1961; Lindenberg, 1963; Zülch, 1955).

Even more important is the assay of blood supply to the brain in those types of experiments in which ischemic factors are the fundamental elements of the models employed. A great number of very sophisticated methods have been elaborated for measurements of blood supply and flow in the brain (Lassen, Ingvar, 1961; Walz et al., 1972; Mchedlishvili et al., 1972). All these methods offering very precise quantitative data, give only to a limited extent the insight into the topography of blood flow disturbances. Morphological, pathological and pathophysiological studies of the cerebral blood vessels in the experimental model of circulatory hypoxia have been described in a number of publications originating from the Institute of Physiology of the Georgian Academy of Sciences (Mchedlishvili et al., 1965; 1971; Mchedlishvili, Baramidze, 1971; Mchedlishvili, 1972). The majority of these studies concerned, however, the leptomeningeal vessels and those penetrating the cerebral cortex. Therefore, it seemed of interest to evaluate the state of the blood vessels network in the brain hemispheres in conditions of circulatory hypoxia.

MATERIAL AND METHODS

The studies were carried out on adult rabbits of both sexes, weighing from 2.5 to 3.5 kg, in which circulatory hypoxia was evoked according to the method described by Mchedlishvili (1972). For morphological studies animals were sacrificed in groups at the following time intervals: group 1 — at the 10th minute of ischemia; group 2 — at time "0", what mean the end of 1 5minutes ischemia. This group consisted two variants: a) without blood reinjection, b) with blood reinjection.

The other groups consisted of animals sacrificed 15 min., 2, 6, 12, 24 and 48 hrs following ischemia. Exsanguination was controlled by blood pressure measurements, indicating that during ischemia systemic blood pressure was reduced to 20 mm Hg. The reinjection of the blood led to the return of b.h. to the normal level or slightly below the norm. Each group consisted of 3 experimental and 2 control animals. The control animals were submitted to the same surgical procedure as the experimental ones, except exsanguination. The third group consisted of 3 normal animals which were not submitted to any experimental procedures.

The brains were fixed in neutral formalin, divided into blocks horizontally at the level of the optic chiasm, infundibulum and interpeduncular fossa. Frozen sections, 40 μ thick were stained according to Pickworth's method.

RESULTS

In the brain of normal rabbits the cortical vascular system consisted of a dense network of vessels, varying in caliber and lumen width, arranged in typical layers corresponding to the neuronal stratifications. A varying diameter of vessels representing veins, arteries and capillaries was a striking feature (Fig. 1). Radial arrangement of cortical arteries, and long, wide veins perforating all the neuronal layers was characteristic for all neocortical areas. Vascularisation of the white matter was less dense, and dominated by large veins, showing typical radial arrangement (Fig. 1, 2). The vascular system of the striatum, much more abundant than that of the white matter did not show any characteristic organization (Fig. 2). Abundant vasculature of the hippocampal area, on the contrary, was characterized by typical architectonics, dominated by two vascular plexuses corresponding to nerve cells layers and radially oriented vessels localized between them. Large wide veins were situated in the most central part of Ammon's horn (Fig. 3).

In animals sacrificed at the 10th minute of oligemia a remarkable reduction of blood content in the vascular bed dominated the cortical picture. The typical stratification of cortical angioarchitecture was no more

visible. Visualized was predominantly the network of capillary vessels with only some penetrating arteries (Fig. 4). The rarefaction of the vascular net indicated that, besides reduction of the blood content within the vessels, a great part of them did not contain any elements stainable by the method employed. In the white matter large veins were filled with a considerable amount of erythrocytes. The same situation as in the cortex was seen in Ammon's horn and in the basal ganglia (Fig. 5). However, even at that time some areas of the cerebral cortex showed a relatively better blood supply (Fig .6). These were predominantly the gyri situated in these parts of the brain, which were vascularized by branches of posterior cerebral arteries. It should be noted, that even here the blood supply was diminished to a great extent, as compared with that of normal, intact animals. The small areas of reduced vascularization ,intermingled with those of relatively better blood supply, were very characteristic for these brain parts.

At time "0" without blood reinjection, the morphological picture showed no significant differences from that observed at the 10th minute of oligemia. The reduction of blood content in the vascular bed all over the brain seemed even more significant.

The blood reinjection brought about a significant amelioration of blood supply on the side with no permanent ligation of the carotid artery, and a less pronounced improvement on the side of the ligated one. It is worth mentioning that time "0" following blood reinjection was the only time with an evident difference in blood supply to both hemispheres, this being most significant in the Ammon's horn structures.

At the 15th min. following blood reinjection the most striking feature to be noted was the widespread cerebral hyperemia of high degree, involving practically all structures of the brain hemispheres (Figs. 7, 8). The vessels of all types and calibers were remarkably widened, however, some regional differences were evident. In the white matter distended large veins dominated the morphological picture. In the hippocampal gyrus against the background of venous hyperemia, poor filling of arterial plexus in the layer of bipyramidal nerve cells was seen. Uneven distribution of blood content within the vessels of the same area and caliber was also a significant feature.

Two hours after oligemia, all structures of the cerebral hemispheres were still hyperemic, but in the majority of animals widenning of venous vessels prevailed over that of arterial ones. Widespread patches of vessels with reduced erythrocyte content were very characteristic; this being dominant in the cerebral cortex (Fig. 9). In the 6th hr the number of these areas of reduced blood supply against the background of

hyperemic tissue was even slightly increased (Fig. 10). The opposite situation was observed at the 12th hr of the experiment. The blood supply to the majority of cortical areas seemed to be significantly reduced as compared both with the immediately preceding stage of observations (6 hrs) and with normal animals. This phenomenon was entirely limited to cortical structures (Fig. 11); the basal ganglia blood supply remained at the level observed in animals sacrificed at the 6th hr.

Normalization of the blood supply started 24 hrs following the oligemic episode. However, even at that time, patchy foci of reduced blood supply were present within the cerebral cortex (Fig. 12). They predominated in the areas of the frontal lobes. Full normalization of the morphological picture of the cerebral vascular network was observed in animals sacrificed 48 hrs following oligemia.

Despite the common pattern of blood supply abnormalities described above, significant differences in their intensity and distribution were noted. In all experimental animals the pathological changes were

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Fig. 1. Cerebral cortex with aboundant vascular network; perpendicularly arranged radial arteries are visible. Less rich vascular net of the white matter (right corner) is dominated by large veins. Pickworth's meth. × 60.

Ryc. 1. Kora mózgu charakteryzująca się bogatą siecią naczyniową; widoczne prostopadle przebiegające tętnice promieniowe. Sieć naczyń w istocie białej uboższa, dominują w niej duże naczynia żylne. Met. Pickwortha. Pow. 60 ×.

Fig. 2. Vascular network of basal ganglia and internal capsule. Note the difference in the character and amount of vessels between white and grey matter formations. Pickworth's meth. × 60.

Ryc. 2. Sieć naczyniowa jąder podstawy i torebki wewnętrznej. Zwraca uwagę różnica charakteru naczyń i ich bogactwa w formacjach szarych i białej. Met. Pickwortha. Pow. 60 ×.

Fig. 3. Vascular network of normal Ammon's horn of the rabbit brain, with a characteristic arrangement dependent of its cellular stratification. Pickworth's meth. × 60.

Ryc. 3. Sieć naczyniowa rogu Amona, z charakterystycznym układem uwarunkowanym warstwową budową tej struktury anatomicznej. Met. Pickwortha. Pow. 60 ×.

Fig. 4. Poor erythrocyte filling of the cortical vascular network with complete loss of its normal arrangement; 10th min of ischemia. Pickworth's meth. × 60.

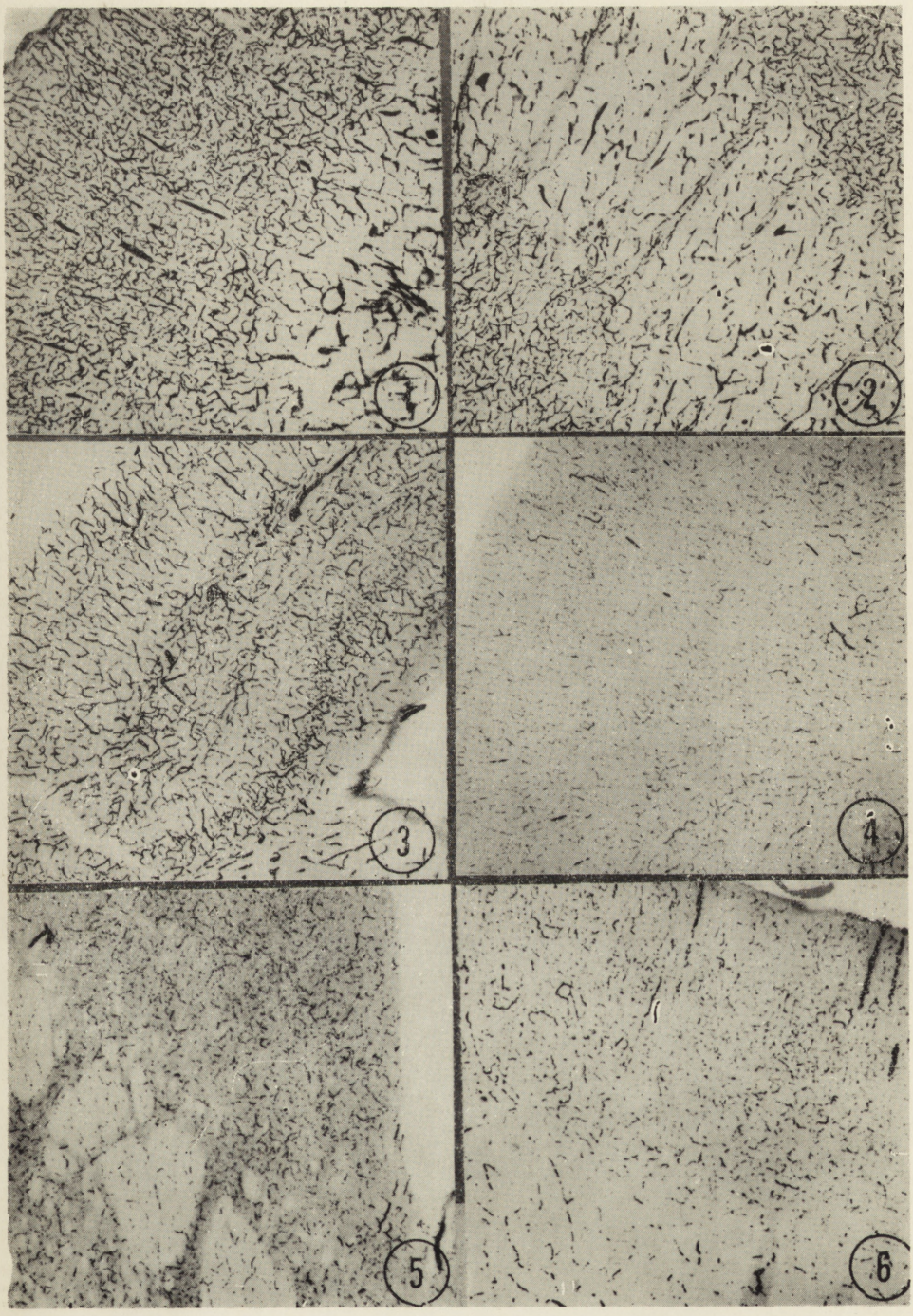
Ryc. 4. Skąpe wypełnienie sieci naczyniowej kory mózgu. Zatarty prawidłowy obraz unaczynienia kory; 10-min. niedokrwienie. Met. Pickwortha. Pow. 60 ×.

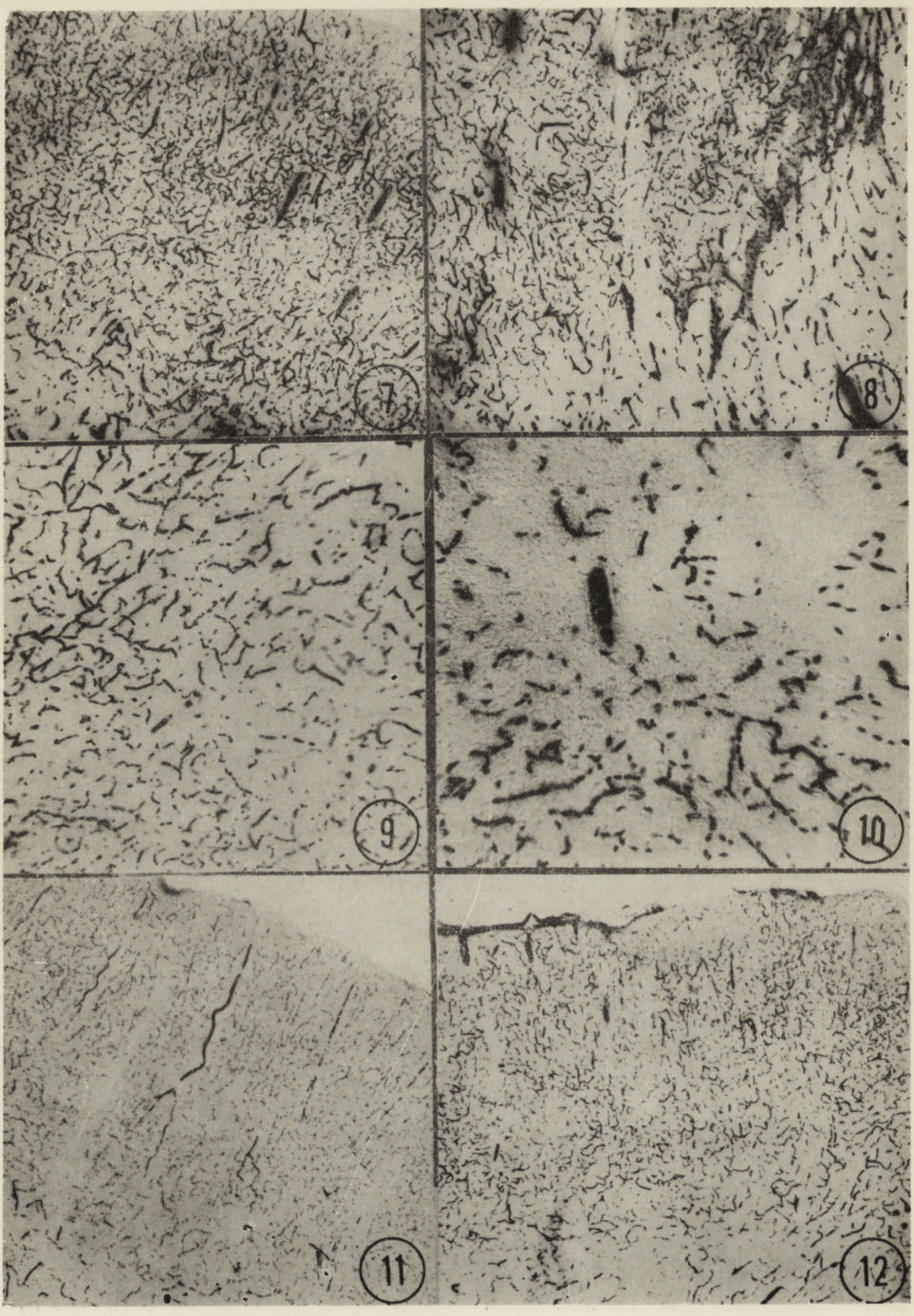
Fig. 5. Poverty of the visualized vascular network of basal ganglia and internal capsule; 10th min of brain ischemia. Pickworth's meth. × 60.

Ryc. 5. Zubożenie uwidaczniającej się sieci naczyniowej jąder podstawy i torebki wewnętrznej w 10 minucie niedokrwienia. Met. Pickwortha. Pow. 60 ×.

Fig. 6. Relatively better visualized vascular network of the cerebral cortex in the area of posterior cerebral artery vascularization; 10th min of brain ischemia (compare with *Fig. 4).* Pickworth's meth. × 60.

Ryc. 6. Stosunkowo lepiej uwidoczniona sieć naczyń krwionośnych kory mózgu w obszarze unaczynienia przez tętnicę mózgu tylną; 10 min. niedokrwienia (por. *ryc. 4).* Met. Pickwortha. Pow. 60 ×.





most significant in what is called borderline zones between areas vascularized by different large cerebral arteries, this being most pronounced in the frontal cortical areas and the thalamus.

The control animals did not show any essential differences in the morphological picture of the brain vascular network as compared with normal animals, which were not submitted to any experimental procedure.

DISCUSSION

Our morphological observations, concerning the period of generalized oligemia and early stages of postischemic recovery show full concomitance with Kapuściński's results (1973) dealing with cerebral blood flow in the same experimental model of circulatory hypoxia. The significant brain ischemia involving all structures of cerebral hemispheres, during systemic oligemia (leading to blood pressure reduction to the level of 20 mm Hg), corresponds well with the essential blood flow reduction observed at the same period of the experiment. However, at the morphological level, even at this time, the cerebral cortex situated in the areas of posterior cerebral artery vascularization, in spite of reduced

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Fig. 7. Postischemic hyperemia of the cerebral cortex at the 15th min following blood retransfusion. Pickworth's meth. × 60.

Ryc. 7. Przekrwienie kory mózgu w 15 min. po niedokrwieniu. Met. Pickwortha. Pow. 60 ×.

Fig. 8. Hyperemia of basal ganglia and internal capsule at 15th min following brain ischemia. Pickworth's meth. × 60.

Ryc. 8. Przekrwienie jąder podstawy i torebki wewnętrznej w 15 min. po niedokrwieniu. Met. Pickwortha. Pow. 60 ×.

Fig. 9. Uneven filling of the cortical blood vessels at the 2nd hour following blood retransfusion. Pickworth's meth. × 100.

Ryc. 9. Nierównomierne wypełnienie naczyń kory mózgu w 2 godz. po niedokrwieniu. Met. Pickwortha. Pow. 100 ×.

Fig. 10. Area of poor filling of cortical vessels, laying on the background of engorged vascular network, 6 hrs following blood retransfusion. Piskworth's meth. × 160.

Ryc. 10. Pole ubogiego wypełnienia sieci naczyń kory mózgu położone na tle jej przekrwienia w 6 godz. po niedokrwieniu. Met. Pickwortha. Pow. 160 ×.

Fig. 11. Poor filling of the cortical vascular network at the 12th hr following blood retransfusion. Pickworth's meth. × 60.

Ryc. 11. Skąpe wypełnienie sieci naczyń w korze mózgu w 12 godz. po niedokrwieniu. Met. Pickwortha. Pow. 60 ×.

Fig. 12. Cortical vascular network at the 24th hr following blood retransfusion. Note uneven filling of blood vessels (compare *Fig. 1*). Pickworth's meth. × 60.

Ryc. 12. Sieć naczyń kory mózgu w 24 godz. po niedokrwieniu. Zwraca uwagę utrzymujące się nierównomierne wypełnienie sieci naczyniowej (patrz *ryc. 1*). Med. Pickwortha. Pow. 60 ×.

blood supply shows significantly better irrigation than that vascularized by anterior and middle cerebral arteries. This might be indicative of a relatively good efficiency of the vertebral arterial system in the case of bilateral ligation of carotid arteries in rabbits (Kapuściński, unpublished data). Reinjection of blood into the arterial system, with return of the blood pressure to normal level brings about an immediate amelioration of blood supply to the brain hemispheres, the phenomenon being for a short period of time limited to the side of the non ligated carotid artery. The short-lasting deterioration of the blood supply to the hemisphere on the ligated side seems to be a phenomenon quite different in nature from the no-reflow phenomenon, described by Ames et al. (1968).

Cerebral ischemia is followed by considerable hyperemia, involving all types of blood vessels. This again corresponds to a significant cerebral blood flow increase, found by Kapuściński (1973) in the period following immediatelly generalized oligemia and lasting from the 15th to 90th minute of the recovery phase. These observations support also the data presented by Mchedlishvili et al. (1974). The prevalence of nervous tissue engorgement, observed at the 2nd and 6th hours following ischemia as compared with the relative decrease of cerebral blood flow in the 2nd hour of the postischemic period, noticed by Kapuściński (1974) might indicate that passive hyperemia and venous stasis are a phenomenon following active hyperemia.

The disturbances of cerebral microcirculation during later phases of the recovery period form a separate and very important problem from the standpoint of the pathogenesis of morphological, histochemical and biochemical changes occurring in the brain following circulatory hypoxia (Albrecht, 1974; Sikorska et al., 1974; Zelman, 1974). They consist in generalized ischemic features of the cerebral cortex, following the period of brain hyperemia (being seemingly reflex in their nature) and in the occurrence of patchy areas of reduced blood supply in the cerebral cortex during the whole period of hyperemia and normalization. Their presence is concordant with the observations made by Kowada et al. (1968), who noted the presence of cerebral circulation disturbances in the period following other types of hypoxic and/or ischemic accidents. The nature of focal circulation abnormalities is unknown. It seems, however, that they may be somehow related with structural and functional abnormalities, concerning pial and cortical arteries, found by Mchedlishvili et Baramidze (1971) in circulatory hypoxia. The focal character of blood-brain barrier disturbances and their localization confined to the cerebral cortex (Gadamski, Szumańska, 1974) are also suggestive of a close relation of this phenomenon with the above described abnor-

malities in cortical circulation. Their occurrence in the postischemic period in the presence of full normalization of the systemic blood pressure indicates that the structural abnormalities found in the brain tissues depend not only on the reduced blood supply to the brain during systemic oligemia, but also on supply disturbances taking place during the whole period of recovery. They are also strongly suggestive of abnormalities in the autonomic regulation of brain circulation, which are not visualized by global isotope measurements.

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ZABURZENIA MIKROKRĄŻENIA MÓZGOWEGO W HIPOKSJI KRĄŻENIOWEJ

Streszczenie

Przy pomocy techniki benzydynowej Pickwortha oceniono stan sieci naczyniowej mózgu w hipoksji krążeniowej, zarówno w okresie ogólnoustrojowej oligemii jak i w czasie 48 godzin po epizodzie niedokrwiennym.

Stwierdzono, że w okresie wykrwawienia zwierzęcia, związanego ze spadkiem ciśnienia układowego do wartości 20 mm Hg — dochodzi do wybitnie nasilonych cech niedokrwienia mózgu, wyraźniejszych w obszarze unaczynienia tętnic przedniej i środkowej mózgu i mniej nasilonych w polach zaopatrywanych przez tętnicę mózgu tylną.

Retransfuzja krwi prowadząca do wzrostu ciśnienia układowego do wartości zbliżonych do normy wywołała znacznego stopnia przekrwienie wszystkich struktur półkul mózgowych, utrzymując się do 6 godziny po zabiegu doświadczalnym, z tym jednak, że w okresie drugiej i szóstej godziny dominował obraz przekrwienia żylnego tkanki. Na tle uogólnionego przekrwienia mózgu, początkowo tętniczego a następnie żylnego w korze półkul występowały przez cały okres zdrowienia, aż do 24 godziny rozsiane ogniska upośledzonego ukrwienia tkanki. W 12 godzinie po zabiegu stwierdzono ponownie uogólnione niedokrwienie kory. Zapoczątkowana w 24 godzinie normalizacja ukrwienia mózgu następowała po 48 godzinach od zabiegu.

Zwrócono uwagę na występowanie „późnych" zaburzeń krążenia mózgowego w okresie zdrowienia poniedokrwiennego, podkreślając w szczególności występowanie rozsianych, plackowatych ognisk niedokrwienia kory. Zaburzenia ukrwienia w okresie zdrowienia mogą, obok krótkotrwałego epizodu niedokrwiennego odgrywać istotną rolę w patogenezie metabolicznych i morfologicznych uszkodzeń tkanki.

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НАРУШЕНИЯ МОЗГОВОЙ МИКРОЦИРКУЛЯЦИИ ПРИ ЦИРКУЛЯЦИОННОЙ ГИПОКСИИ

Резюме

С помощью бензидиновой техники Пикворта оценивали состояние сосудистой сети мозга при циркуляционной гипоксии как в период олигемии всего организма, так и через 48 часов после момента ишемии.

Было установлено, что в период обескровливания животного, связанного с падением системного давления до величины 20 мм ртутного столба, доходит до исключительно сильно выраженной ишемии мозга, особенно в области васкуляризации передней и средней артерии мозга и менее интенсивной в областях, снабжаемых задней артерией мозга.

Ретрансфузия крови, ведущая к увеличению системного давления до величин, близких к норме, вызывала значительную гиперемию всех структур мозговых полушарий, удерживающуюся до 6-го часа после экспериментальной процедуры, причем в период второго и шестого часа преобладала картина венозной гиперемии ткани. На фоне общей гиперемии мозга, сначала артериальной, а затем венозной, в коре полушарий в течение всего периода возвращения к норме, вплоть до 24 часов, выступали разбросанные очаги нарушенного кровоснабжения ткани. Через 12 часов после процедуры снова наблюдалась общая ишемия коры. Начавшаяся на 24-ом часу нормализация кровоснабжения мозга наступала через 48 часов после процедуры.

Было обращено внимание на проявление „поздних" нарушений мозгового кровообращения в период постишемической нормализации, при этом особенно подчеркивается появление разбросанных мелких очагов ишемии коры. Нарушения кровоснабжения в период возвращения к норме могут, наряду с кратковременным моментом ишемии, играть важную роль в патогенезе метаболических и морфологических нарушений ткани.

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