4. V. A. Levtov and D. I. Parolla, in: Problems in Regulation of the Regional Circulation [in Russian], Leningrad (1969), pp. 96-105.
5. Yu. A. Medvedev, in: Problems in Pathological Anatomy [in Russian], Leningrad (1970), pp. 203-218.
6. V. I. Osipov, Artificial Hypotension [in Russian], Moscow (1967).
7. H. S. Bennett, Am. J. Anat., 67, 151 (1940).
8. S. Davies, Q. J. Microscop. Sci., 80, 81 (1937).
9. L. Nicander, Acta Anat. Suppl., No. 16 (1952).

MICROSPECTROPHOTOMETRIC CHARACTERISTICS OF RENAL ENZYMES IN ACUTE CIRCULATORY FAILURE

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Activity of lactate, succinate, glucose-6-phosphate, β-hydroxybutyrate, NADH, and NADPH dehydrogenases, and of alkaline and acid phosphatases in the kidneys was investigated in intact cats, during hexobarbital anesthesia, after hypotension produced by injection of a ganglion blocker (Arfonad) for 5 h, 24 h after the end of artificial hypotension, and after massive blood loss. All parts of the nephron responded by differential changes of enzyme activity. A sharp decrease in the role of the principal pathway of oxidation (the Krebs cycle) in energy metabolism took place, and under the conditions of circulatory hypoxia the glycolytic pathway and pentose shunt became predominant.

KEY WORDS: acute vascular failure; kidneys; enzyme activity.

Acute systemic circulatory failure (collapse), of varied origin, frequently leads to acute renal insufficiency [4, 6], which is based on circulatory hypoxia of the kidney and, it has been suggested, a disturbance of the function of energy-forming systems, especially of oxidoreductases and hydrolases [1-4, 6].

In this investigation the dynamics of activity of various oxidoreductases and hydrolases in the kidney was studied in the early stages of acute circulatory failure and after blood loss.

EXPERIMENTAL METHOD

Oxidative and hydrolytic enzymes of the kidneys were investigated in 50 male cats divided into four groups: 1) intact (8), 2) anesthetized (10), 3) killed immediately after artificial hypotension for 5 h, and 24 h after its end (25), and 4) killed after hypotension for 4 h, caused by blood loss (6). The general anesthetic, a 10% solution of hexobarbital, was injected intraperitoneally (0.1 g/kg). Artificial hypotension was produced in the anesthetized animals by intravenous drip injection of a 0.1% solution of Arfonad or by repeated bleeding from the femoral artery. The blood pressure fell to 50% of its initial level, at which it was maintained throughout the experiments.

To assess the dynamics of activity of oxidoreductases and hydrolases, a series of eight enzymes was chosen for testing: lactate dehydrogenase (LD), the most important enzyme of glycolysis; glucose-6-phosphate dehydrogenase (G6PD), an indicator of the intensity of glucose oxidation in the pentose shunt; succinate dehydrogenase (SD), a histochemical indicator of the Krebs cycle; β-hydroxybutyrate dehydrogenase (β-HBD),

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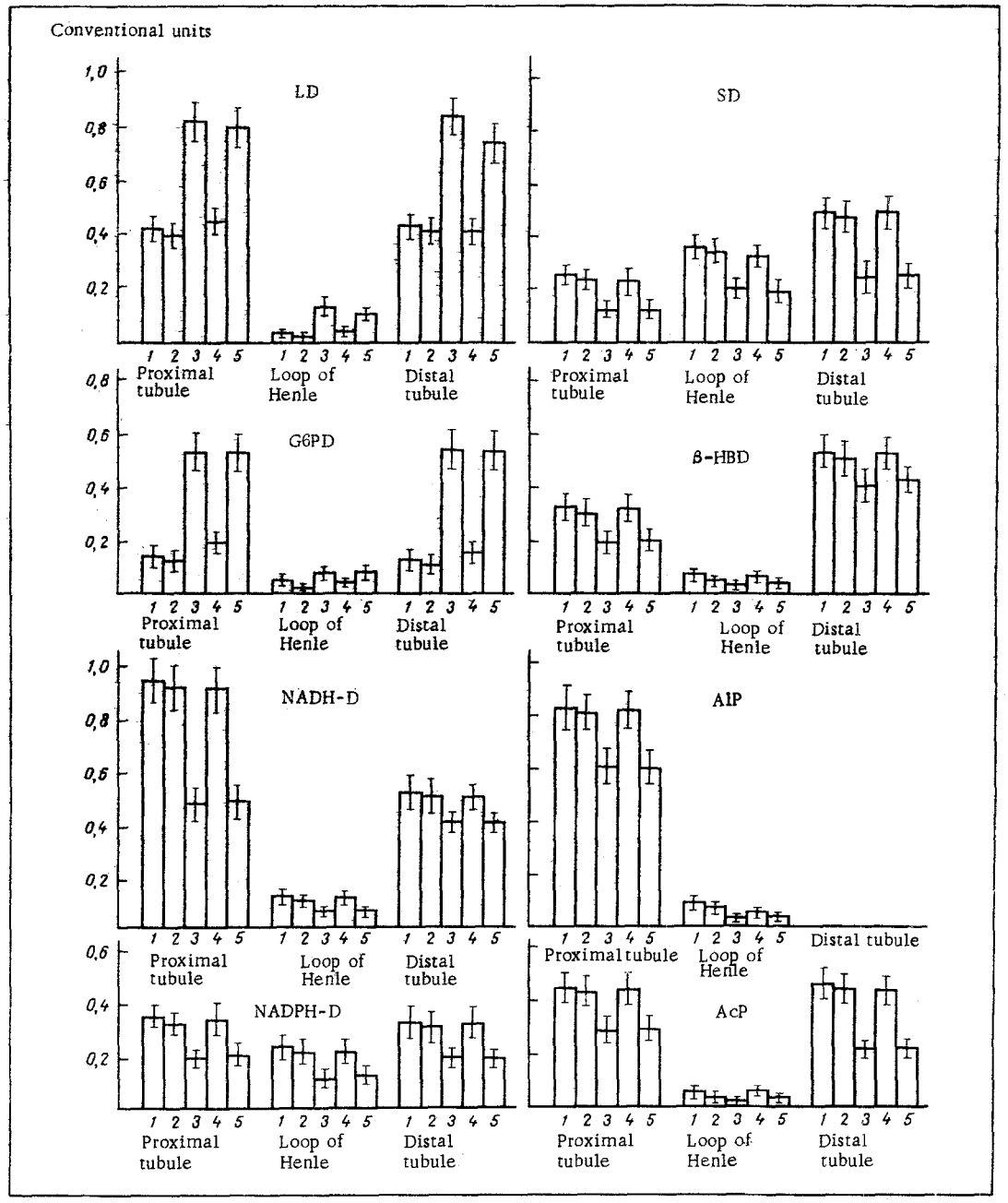

Fig. 1. Changes in activity of oxidoreductases and hydrolases in epithelium of different parts of the cat nephron during artificial hypotension, 24 h after its end, and after acute blood loss: 1) intact animals; 2) control animals (hexobarbital anesthesia); 3) after hypotension for 5 h (injection of Arfonad); 4) 24 h after end of hypotension; 5) after acute blood loss.

concerned in the oxidation of fatty acids; dehydrogenases of the reduced forms of NAD (NADH-D) and NADP (NADPH-D), indicators of the general activity of systems connected with NAD and NADP and also carrying electrons to the cytochrome system; alkaline phosphatase (AlP), hydrolyzing phosphoric acid esters and connected with ATP formation; and acid phosphatase (AcP), the most important representative of the lysosomal enzymes. Activities of LD, G6PD, β-HBD, SD, NADH-D, NADPH-D, AlP and AcP were determined. Histochemical reactions were carried out on frozen sections. To extract the lipid inclusions the kidney sections were treated before incubation with cold acetone [5]. The comparative quantitative determination of enzyme activity in the sections was carried out with the MUF-5 instrument.

EXPERIMENTAL RESULTS

Microspectrophotometric investigation of the renal enzymes (Fig. 1) showed that the nephron of intact animals has a characteristic tissue enzyme profile. In the epithelium of the proximal portions, the enzyme with higher activity than all the others was NADPH-D: The activity of AlP was 17%, AcP 55%, LD 57%, NADPH-D 64%, β-HBD 67%, SD 74%, and G6PD 85% lower, respectively, than the NADH-D activity. In the epithelium of the distal portions, NADH-D activity was 44% lower than in the proximal portions, β-HBD activity was at the same level, whereas SD activity was 10%, AcP 17%, NADPH-D 37%, and G6PD 87%, respectively, lower than NADH-D activity. The AlP activity could not be detected in the distal tubules. In the descending thin portion of the loop of Henle comparatively low activity of most of the above enzymes was discovered. The SD activity, which was the highest in this situation, was 63% lower than the NADH-D activity in the proximal tubules, and in the loop of Henle the NADH-D activity was 33%, NADPH-D 66%, AlP 88%, β-HBD 83%, AcP 86%, G6PD 92%, and LD activity 93% lower than the SD activity.

Hexobarbital anesthesia had no significant effect on the tissue enzyme profile of the kidneys of the intact cats. Changes in the activity of the overwhelming majority of the above enzymes were not statistically significant ($P > 0.05$).

The essential feature of the dynamics of the tissue enzyme profile of the nephron after artificial hypotonia for 5 h was the changes in the activity of oxidoreductases and hydrolases in its different parts. In the proximal tubules LD activity was increased by 110% and G6PD activity by 320%, whereas SD activity was reduced by 52%, β-HBD by 37%, NADH-D by 47%, NADPH-D by 61%, AlP by 33%, and AcP activity by 29%, respectively. In the distal tubules LD activity also was increased by 100% and G6PD activity by 390%, whereas SD activity was reduced by 47%, β-HBD by 17%, NADH-D by 23%, NADPH-D by 44%, and AcP by 17%. In the loop of Henle LD activity was increased by 590% and G6PD activity by 220%, whereas SD activity was reduced by 41%, β-HBD by 47%, NADH-D by 37%, NADPH-D by 47%, AlP by 67%, and AcP by 23%, respectively.

Similar changes in enzyme activity also were observed after blood loss. In the proximal tubules LD activity was increased by 100% and G6PD activity by 320%, whereas SD activity was reduced by 52%, β-HBD by 33%, NADH-D by 47%, NADPH-D by 41%, AlP by 23%, and AcP by 33%, respectively. In the distal tubules LD activity was increased by 80% and G6PD by 370%, whereas SD activity was reduced by 47%, β-HBD by 17%, NADH-D by 23%, NADPH-D by 41%, and AcP by 52%, respectively. In the loop of Henle LD activity was increased by 470% and G6PD by 220%, whereas SD activity was reduced by 41%, β-HBD by 58%, NADH-D by 41%, NADPH-D by 44%, AlP by 60%, and AcP by 9%, respectively.

The normal tissue enzyme profile of the kidneys was restored 24 h after the end of artificial hypotension. Compared with their levels during artificial hypotension, activity of LD in the proximal tubules was reduced by 44% and of G6PD by 64%, whereas SD activity was increased by 100%, β-HBD by 60%, NADH-D by 90%, NADPH-D by 70%, AlP by 30%, and AcP by 40%. In the distal tubules LD activity also was reduced by 52% and G6PD by 72%, whereas SD activity was increased by 100%, β-HBD by 30%, NADH-D by 30%, NADPH-D by 80%, and AcP by 110%. In the loops of Henle LD activity was reduced by 71% and G6PD by 55%, whereas SD activity was increased by 60%, β-HBD by 100%, NADH-D by 60%, NADPH-D by 90%, AlP by 180%, and AcP by 30%.

In the loops of Henle the activity of all enzymes except AcP was changed under comparable conditions: The changes were significant ($P < 0.001$) during artificial hypotension, blood loss, and hexobarbital anesthesia, as well as the changes between artificial hypotension and 24 h after its end.

The results confirm the general views expressed by Serov et al. [7-9] and Avtandilov et al. [1, 2] regarding the compensatory character of the changes in the enzyme systems of the nephron as a result of the various procedures. Quantitative studies of the activity of oxidoreductases and hydrolases during acute vascular insufficiency revealed regular changes in the enzyme homeostasis of the kidneys. They are clear evidence of a sharp decrease in the importance of the principal oxidation pathway in the Krebs cycle in the provision of energy and predominance of the glycolytic pathway and the pentose shunt during circulatory hypoxia. This fact can evidently be regarded as the response of the tissue enzymes to hypoxia and as a manifestation of compensatory and adaptive reactions at the subcellular level in the enzyme systems of the kidneys.

The changes in activity of the oxidoreductases and hydrolases described above can demonstrate a very important general pathological rule: Tissue enzyme mechanisms compensate for disturbed metabolism. On the other hand, the prolonged action of acute vascular insufficiency leads to tissue enzyme changes [2] in the kidney tissue, expressed as a reorganization of the intracellular metabolism of the renal epithelium to the glycolytic and pentose pathways, the energy value of which is extremely low.

LITERATURE CITED

1. G. G. Avtandilov, A. A. Sumbatov, I. S. Kruglova, et al., Vestn. Akad. Med. Nauk SSSR, No. 10, 64 (1971).
2. G. G. Avtandilov, I. S. Kruglova, and K. D. Dzhalalov, Arkh. Patol., No. 6, 46 (1972).
3. A. K. Ageev, Histochemistry of Alkaline and Acid Phosphatases in Man in Health and Disease [in Russian], Leningrad (1969).
4. V. L. Byalik, A. F. Kiseleva, V. M. Kamenko, et al., Arkh. Patol., No. 10, 16 (1970).
5. Yu. K. Eletskii, Arkh. Patol., No. 11, 68 (1965).
6. N. F. Kan'shina, Arkh. Patol., No. 10, 3 (1970).
7. V. V. Serov and A. G. Ufimtsev, Byull. Éksp. Biol. Med., No. 5, 113 (1967).
8. V. V. Serov, Vestn. Akad. Med. Nauk SSSR, No. 3, 32 (1970).
9. V. V. Serov, in: Proceedings of the Fourth International Congress of Histochemistry and Cytochemistry, Kyoto (1972).
10. R. Hess, D. G. Scarpelli, and A. G. E. Pearse, Nature, 181, 1531 (1952).

CHANGES IN PHAGOCYTES AND LYMPHOCYTES IN OBSTRUCTIVE PNEUMONIA

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A cytochemical investigation was made of the state of the phagocytes and lymphocytes exposed to the action of a foreign body under septic and aseptic conditions. One of the main mechanisms of the development of an inflammatory-destructive process in the lungs is by local and systemic changes in the ratio between the numbers of phagocytes and lymphocytes, disturbance of the permeability of their lysosomal membranes, and an increase in the number of destroyed macrophages, neutrophils, and lymphocytes, all of which can be regarded as indices of cellular regression.

KEY WORDS: pneumonia; regression; macrophages; leukocytes.

Investigations by Anichkov et al. [1-3] have shown that after partial obstruction of a bronchus (by suturing or introduction of a foreign body) progressive inflammatory-destructive changes develop in the lungs. Their development is associated with activation of the autogenous flora and the arrival of an exogenous flora in the respiratory tract. Macrophages and lymphocytes of the blood have been studied by the writer during this process [6, 8, 9].

EXPERIMENTAL METHOD

A length of nylon thread 6 cm long and 0.6 mm in diameter was introduced into the trachea of 32 rabbits. The distal end of this length of thread lay at the bifurcation of the trachea. Four healthy rabbits acted as the control. The experimental animals (four at each time) were killed 1 and 3 days, 1 and 2 weeks, and 1, 2, 3, and 6 months after introduction of the thread. To assess the action of the foreign body on the phagocytes and lymphocytes under aseptic conditions, a length of sterile nylon thread 1 cm long and 0.2 mm in diameter was introduced intraperitoneally into 43 albino mice. Five animals served as the control group. The experimental

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