STRUCTURAL-METABOLIC ASPECTS OF THE DIAGNOSIS OF ACUTE HEPATIC FAILURE

O. D. Mishnev, N. A. Sergeeva, A. I. Shchegolev, and A. P. Raksha

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Acute hepatic failure is an important cause of death in modern clinical medicine and is frequently combined with functional impairment of other organs [2, 8, 10]. The postmortem diagnosis of hepatic failure is associated with certain known difficulties. The initial clinical manifestations of hepatic failure may not be reflected in marked structural changes at the light-optical level. Their morphological diagnosis becomes more realistic when quantitative enzyme histochemical methods are used. However, postmortem autolytic changes developing rapidly in the liver significantly restrict the use of structural-metabolic tests [4, 13]. The most promising method for the diagnosis of acute hepatic failure is evidently that of early autopsy, when the combined use of morphological and functional methods, together with clinical data, becomes possible [6, 12].

The aim of the present investigation was to study the possibility of using the method of early autopsy on dying patients for the morphological and functional verification of the development of hepatic failure.

METHODS

Altogether 18 cases of early autopsy were studied. In five patients, according to the results of clinical—anatomical analysis, the immediate cause of death was acute posthemorrhagic anemia resulting from rupture of an aneurysm of the abdominal aorta, in five patients there was failure of several organs as a result of toxemia in patients with peritonitis (four) and with gangrene of the lower limb (four). In all cases, the clinical laboratory data showed signs of hepatic failure (hypoalbuminemia, hyperbilirubinemia, raised blood enzyme levels). The control group consisted of five early autopsies conducted on medicolegal grounds: in three of them a diagnosis of sudden cardiac death was made, in two death was due to fatal trauma. The patients' ages ranged from 52 to 68 years. Autopsy was carried out 45-90 min after death. Pieces excised from the right lobe of the liver were frozen and kept in liquid nitrogen, after which simultaneous enzyme histochemical tests were carried out on frozen sections in order to detect activity of the following dehydrogenases: succinate (SDH), lactate (LDH), glutamate (GDH), β-hydroxybutyrate (BDH), and glucose-6-phosphate (G6PDH), and also of acid (AcP) and alkaline (AlP) phosphatase [11]. Quantitative evaluation of enzyme activity was carried out by means of a "Microvideomat" television image analyzer (Opton, Germany), controlled by a Wang 720c computer, using a specially devised program of photometric analysis of histologic preparations [3]. Paraffin sections were stained with hematoxylin and eosin. In a parallel series, on an Impact-400 biochemical analyzer (Corning, USA), concentrations of albumin, total protein, cholesterol, glucose, lactate, and nonprotein nitrogen, and also of activity of AlP, LDH, γ-glutamyl transpeptidase (GTP), and alanine (ALT) and aspartate transaminases (AST) in liver tissue homogenates were determined. The numerical results were subjected to statistical analysis by computer.

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TABLE 1. Enzyme Histochemical Characteristics of Hepatocytes in Toxemia and Blood Loss (M ± m, conventional units)

Enzyme	Group		
	control	toxemia	blood loss
SDH	512±21	228±17	281±19
LDH	711±26	599±18	648±19*
GDH	567±22	209±15	262±16
BDH	561±21	329±17	433±17
G6PDH	480±25	188±19	261±21
AcP	124±12	303±26	264±23
AIP	117±11	323±28	229±19

Note. Here and in Table 2, all values except those marked by an asterisk differ significantly from the control ($p < 0.05$).

TABLE 2. Biochemical Characteristics of Liver Tissue Homogenates in Toxemia and Acute Blood Loss (M ± m, per gram tissue

Parameter (μmoles/liter)	Group		
	control	toxemia	blood loss
Total protein	36,0±2,4	14,3±1,8	28,1±2,1
Albumin	16,4±1,1	7,6±1,2	12,0±1,5
Nonprotein nitrogen	67,5±6,0	51,7±4,8	68,9±5,8*
Cholesterol	378,4±32,5	199,9±20,1	102,0±12,4
Glucose	2383,5±284,4	877,0±92,4	892,1±84,5
Lactate	34,9±2,7	31,8±2,6*	21,1±1,8
LDH	58969±6324	175895±18921	64447±6189*
AIP	129,9±14,8	9617,5±832,4	3312,0±236,5
ALT	564,5±62,3	9983,3±884,2	15993,2±1442,8
AST	117,3±15,6	1316,3±128,4	1170,6±121,6
GTP	4393,6±452,7	2249,2±311,6	4043,4±412,6*

RESULTS

In the control group, microscopic study of the liver revealed slight inequality of congestion of the sinusoids, a moderate degree of granular degeneration, and solitary monocellular foci of necrosis in the perivenular areas of the acini. Besides the above-mentioned changes, dilatation and congestion of vessels of the portal tract and sinusoids, aggregation of erythrocytes with microthromboses and widening of the Disse's spaces around the sinusoids, often containing blood cells, and more marked degenerative changes in the hepatocytes in the perivenular and intermediate regions, amounting in some cases to triangular and periacinar necrosis, were observed in the liver of patients dying with evidence of marked toxemia. In patients dying from acute blood loss, anemia of the vessels of the portal tract and sinusoids was observed in the liver, with monocellular and solitary centrilobular areas of necrosis.

More informative data were obtained by quantitative enzyme-histochemical study of the liver tissue, which demonstrated the intensity of metabolic processes in the cells (Table 1). In the liver of patients dying with acute hepatic failure, developing as a result of severe toxemia of varied genesis and acute blood loss, a decrease in activity of the oxidoreductases studied and an increase in activity of the hydrolytic enzymes were observed, in agreement with data in the literature [1, 7]. However, the degree of change in their activity varied. Activity of SDH, the central enzyme of the Krebs' cycle, in toxemia was 55.5% below the control level ($p < 0.05$) and in blood loss 45.1% lower ($p < 0.05$. Activity of G6PDH, the marker enzyme of the pentose phosphate shunt for glucose oxidation, was 60.8 and 45.6% lower than the control, respectively ($p < 0.05$). The decrease in activity of BDH, which characterizes oxidation of fatty acids, by 41.2 and 22.8%, respectively, evidently plays an essential role in the development of degenerative changes in the hepatocytes. Activity of the hydrolytic enzymes in toxemia was increased more than in blood loss. The increase in AcP activity was evidence of activation of the lysosomal-vacuolar system, and the increase in AlP activity reflected increased permeability of the plasma membranes.

It can be concluded from analysis of the changes in enzyme activity observed that in toxemia the disturbance of hepatocyte metabolism was more severe than in acute blood loss, and this was confirmed by the results of morphologic investigation. This is evidently connected with the fact that circulatory hypoxia, leading to inhibition of oxidation-reduction processes in the cells, plays an important role in the pathogenesis of liver damage in toxic conditions, due to gangrene of the lower limb and to peritonitis. The inhibitory action of toxic substances on the energy metabolism of the hepatocytes, demonstrated in our cases by reduction in activity of LDH, an indicator of anaerobic glycolysis, by 15.8%, also is very important.

Biochemical tests on the homogenates also revealed significant disturbances of metabolism in the liver tissue (Table 2). Comparison of the parameters in blood loss and toxemia showed that the changes were similar in type,

but greater deviations from the control values were observed in the latter group, on enzyme-histochemical investigation. Reduction of the content of total protein (by 60.3 and 21.9%), albumin (by 53.7 and 26.8%), and cholesterol (by 47.2 and 73.0% in toxemia and blood loss, respectively) is evidence of inhibition of synthetic processes in the hepatocytes, which is regarded as one feature of hepatic failure [9] and is confirmed by the enzyme-histochemical data showing weakening of GDH activity. The lower glucose concentration in toxemia (by 63.2%) and blood loss (by 62.6%) compared with the control ($p < 0.05$) is in agreement with our previous findings, showing exhaustion of the glycogen reserves in the hepatocytes and the consequent development of hyperglycemia in patients dying as a result of multiple organ failure [6]. In association with the disturbance of glycogen resynthesis from lactic acid, a decrease was observed in the lactate concentration in the liver tissue (by 8.9 and 39.5% in toxemia and blood loss) leading to the development of hyperlactacidemia, which is regarded as the basic factor in the development of the irreversible state in shock [5].

The development of raised blood enzyme levels, the chief clinical marker of the development of hepatocellular failure in extremal states, can be explained by the marked increase in activity of the enzymes studied by enzyme-histochemical and biochemical investigations, and by injuries to hepatocytes revealed microscopically.

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