POLISH MEDICAL JOURNAL, Vol. XI, No. 2/1972
(Translated from Diagnostyka Laboratoryjna, Vol. VI, No. 3/1971)

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FUNCTION OF THE LIVER AND SOME BIOCHEMICAL INDICES DURING 3-HOUR EXTRACORPOREAL PERFUSION IN A PATIENT WITH ACUTE LIVER FAILURE *

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Some biochemical indices established during extra-corporeal perfusion of pig liver with blood of the patient with acute liver failure were described. The liver failure was a complication of viral hepatitis. There were abnormalities during perfusion in acid-base equilibrium manifested by noncompensated respiratory alkalosis with hypopotassemia.

The serum bilirubin level decreased while pyruvate and lactate concentrations increased. The serum alanine-aminotransferase activity was lower than in normals. The serum aspartate aminotransferase activity was higher and it appeared that this increase was caused by mitochondrial fraction. This suggestion was confirmed by the observation that in this condition the glutamate dehydrogenase activity (typical mitochondrial enzyme) was also elevated. During perfusion the increase of glucose uptake by the patient was observed. The serum glucose concentration should be then continuously controlled.

A number of laboratory investigations were performed in a patient with acute liver failure, connected to a pig liver, during 3-hour perfusion. The present paper contains the results of these investigations. The course and the method of perfusion are described elsewhere.

MATERIAL AND METHODS

In a patient J. R., aged 40, hospitalized at the Department of Infectious Diseases, viral hepatitis was complicated with acute liver failure. The patient was therefore transferred to the Department I of Surgery in order to perform perfusion of the liver. At the time of admission the patient was unconscious,

* This work was partially supported by a grant from Department VI of the Polish Academy of Sciences.

did not react to voice and orders. He had symptoms of jaundice, his liver protruding 1·5—2 finger breadths below the right costal arch. The patient was twice connected to a heterogenous liver, for 3 hours on the first day.

During perfusion the following determinations were made — transaminases (AspAT, AspAT-IIR, AlAT), lactic dehydrogenase (LDH), glutamic dehydrogenase (GLDH), alkaline phosphatase (AP), bilirubin level, sodium, potassium

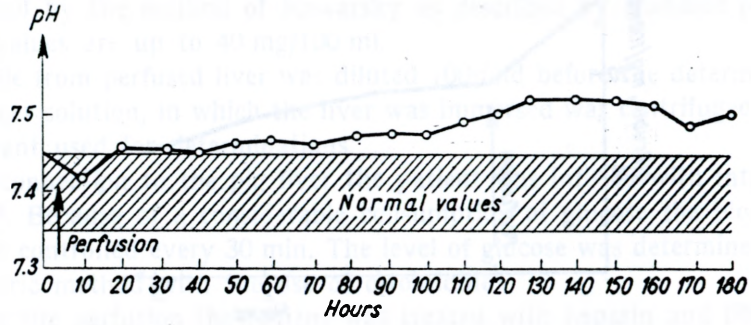

Fig. 1. pH of the arterial blood in a patient with liver necrosis during 3-hour perfusion.

Table I

Blood concentration of electrolytes in the patient with liver necrosis during 3-hour perfusion

Time in hours	Na^+ mEq/l	K^+ mEq/l	Cl^- mEq/l
8[00]	131	4·3	98
Beginning of the perfusion	140	3·5	95
After 1 hour	138	3·7	97
After 2 hours	139	3·6	97
After 3 hours	148	3·4	98

and chloride levels, partial pressure of carbon dioxide (pCO_2), concentration of bicarbonates (HCO_3^-) and concentration of hydrogen ions in blood (pH). Moreover, the level of pyruvic acid and lactic acid, ammonia and urea was determined. Determinations were made in the bile excreted by the connected liver and in the Ringer solution, in which the pig liver was immersed. The bile excreted was collected every hour into a graduated cylinder. The volume and concentration of bilirubin, activity of AspAT, AlAT, and AP were determined in each portion. The level of bilirubin, activities of AspAT, AlAT, LDH, GLDH and AP were determined in the Ringer solution.

The activity of transaminase was determined by the colorimetric method of Umbreit et al. (10). The normal values were accepted as 40 u/ml for AspAT

and 30 u/ml for AlAT. The fraction of AspAT-IIR was determined after precipitation with Rivanol according to the technique described by *Krawczyński et al.* (6). Dehydrogenase activity was measured by the spectrophotometric method of Bergmeyer (1), using a Unicam SP-500 spectrophotometer. The normal values for LDH were accepted as 195 mIU and 2 mIU for GLDH. Alkaline phosphatase was determined by means of sodium p-nitro-phenyl-

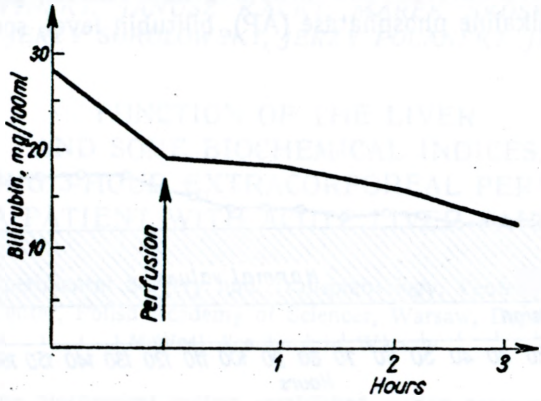

Fig. 2. Serum bilirubin level in the patient with liver necrosis during 3-hour perfusion

Table II

Activity of certain enzymes in the serum of patient with liver necrosis during 3-hour perfusion

Time in hours	AspAT	AspAT-IIR	AlAT	GLDH	LDH	AP
8^{00}	1260	400	2400	18·4	2480	76
Beginning of perfusion	1100	380	2800	24·2	4200	68
After 1 hour	1670	800	1000	24·0	6840	71
After 2 hours	1940	1420	400	29·1	8400	64
After 3 hours	2500	1400	400	30·1	4200	58

phosphate as a substrate (2). The activity of this enzyme in the serum of a healthy subject does not exceed 43 IU. The level of bilirubine was determined by the method of Malloy and Evelyn (8). The normal values reached a level up to 1·2 mg/100 ml of serum. The electrolytes were determined by the flame photometric method using a Zeiss photometer. The normal values for sodium concentration in serum were 132—143, for potassium 3·6—5·6 and for chlorides 96—106 mEq/l. Partial pressure of carbon dioxide and blood pH were determined in the Astrup apparatus. The concentration of bicarbonates was read

from a nomogram. These determinations were made in arterial blood. The normal values for pCO_2 were accepted as 40 mm Hg, for pH — values from 7·35 to 7·45, and for bicarbonate concentration 24—27 mEw/l. Pyruvic acid was determined by the method of Friedemann and Hougen (3), the normal values being up to 1·2 mg/100 ml. Lactic acid was measured by the colorimetric method of Homolka (4), the upper normal value being up to 29 mg/100 ml. Ammonia was estimated by the diffusion method described by *Jonany* and *Reinier* (5), according to which the normal values do not exceed 40 γ/100 ml. Urea was determined by the method of Kowarsky as described by *Homolka* (4) — the normal values are up to 40 mg/100 ml.

The bile from perfused liver was diluted 100-fold before the determinations. The Ringer solution, in which the liver was immersed was centrifuged and the supernatant used for determinations.

After connection to the pig liver the patient was infused with intravenous glucose *. Because of a considerable consumption of glucose the blood sugar level was controlled every 30 min. The level of glucose was determined by the colorimetric method with the use of o-toluidine.

During the perfusion the patient was treated with heparin and fibrinolytic inhibitors. The disturbances in blood clotting occurring during perfusion are described elsewhere (7).

RESULTS AND DISCUSSION

The process of perfusion was accompanied by a lowering of hydrogen ions concentration in blood (pH of blood became more alkaline). One hour after connection of the liver respiratory alkalosis was observed, which manifested itself as initial slight decrease in pCO_2 and increase of pH up to 7·47. The concentration of bicarbonates during this period remained in the normal range. After 2 hours of perfusion, pCO_2 decreased further with simultaneous increase in concentration of bicarbonates. The changes in hydrogen ions concentration in blood during extracorporeal perfusion are presented in Fig. 1.

The alkalosis was accompanied by a relatively low level of potassium with a slight increase in the level of sodium and unchanged chloride concentration (Table I).

The level of blood ammonia during 3-hour perfusion fluctuated within a normal range of 20—40 γ/100 ml.

The level of bilirubin in the blood of the patient during perfusion is illustrated in Fig. 2. After connection with a liver the level of bilirubin in blood decreased gradually from 28·3 mg/100 ml to 12·4 mg/100 ml, that is by 43% of the initial value.

* The rate of infusion was regulated according to the level of glucose in blood. To avoid hypoglycaemia the level of blood glucose was kept around 200 mg/100 ml.

The activity of enzymes in the blood of the patient increased, particularly during the first hour (Table II).

At the end of perfusion the activity of AlAT, LDH and AP decreased, the activity of AspAT, AspAT-IIR and GLDH being unchanged. The ratio of AspAT: AlAT, that is the index of Ritis, was 0·37 at the moment of connection, 1·67 one hour, 4·85 two hours and 6·25 three hours later. An increase in index of Ritis was caused by a constant increase in AspAT activity, and on the other hand by a decrease of AlAT activity in the serum of patient. It is seen from Fig. 3, that an increase of total AspAT activity was accompanied by an elevation of the activity of AspAT-IIR fraction.

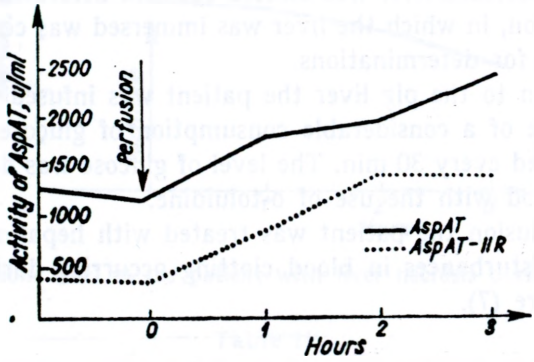

Fig. 3. Serum activity of AspAT and AspAT-IIR in the patient with liver necrosis during 3-hour perfusion.

The level of pyruvic and lactic acid was elevated during the first hour of perfusion and amounted to 2·1 mg and 41 mg per 100 ml respectively, and the level of these acids after 3 hours of perfusion was 3·2 mg and 52 mg/100 ml respectively.

During the period of perfusion the patient was intravenously infused with 1·5 l of 5% glucose. Despite a constant rate of infusion the changes in the sugar level were quite considerable, this being shown in determinations made every 30 min. The course of glycemia during perfusion is shown in Fig. 4.

During the perfusion the pig liver excreted about 57 ml of dark brown bile. The first portion of bile after one hour was quite clear (pH of the bile was 8·7). The level of bilirubin in the bile was considerably elevated and still increased during perfusion. Excretion of bile and the level of bilirubin is presented in Fig. 5.

The activity of excreted enzymes in separate portions of the bile is described in the form of a graph in Fig. 6.

It is seen that the enzymatic activity of bile decreased along with time of perfusion.

In Ringer's solution, in which the liver was immersed during the perfusion, the level of bilirubin was 3·8 mg/100 ml. The activity of AspAT, AlAT, LDH and GLDH is presented in Table III.

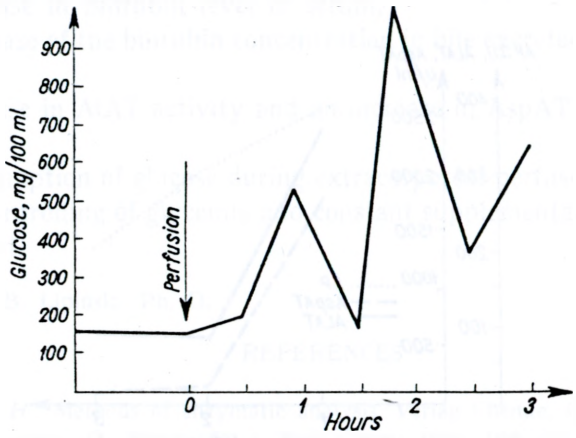

Fig. 4. Serum glucose level in the patient with liver necrosis during 3-hour perfusion.

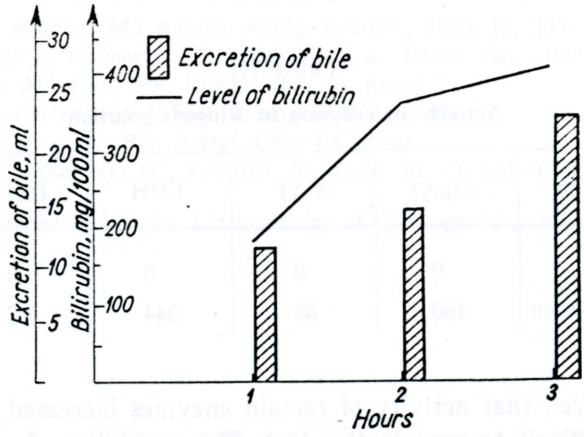

Fig. 5. Excretion of bile and the bile bilirubin level in the patient with liver necrosis during 3-hour perfusion.

The perfusion was performed with the same patient on the next day and the results obtained were similar to those described in the present paper.

During the second perfusion the alkalosis was even more pronounced, and a decrease of bilirubin and increase in AspAT higher than during the first perfusion. The patient showed considerable improvement after the second perfusion. The blood level of ammonia was normal despite occurrence of coma

before and during perfusion. In this case the disturbances in ammonia production did not seem to play a significant role.

The alkalosis progressing during perfusion can be caused by abnormal inactivation of estrogens and progestrone, which stimulate the respiratory center, thus resulting in increased loss of CO_2 and more alkaline pH.

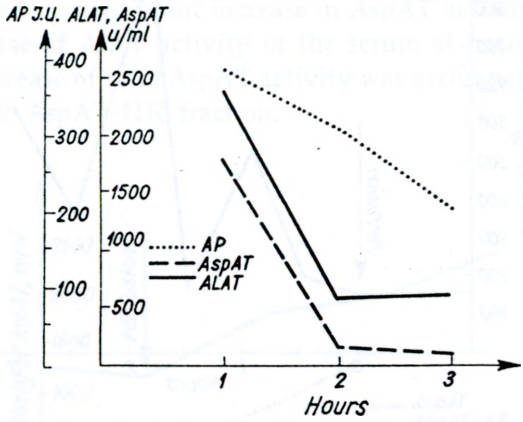

Fig. 6. Activity of certain enzymes in bile in the patient with liver necrosis during extracorporeal perfusion.

Table III

Activity of enzymes in Ringer's solution

Ringer's solution	AspAT	AlAT	LDH	DLDH	AP
Before perfusion	0	0	0	0	0
3 hours after perfusion	180	68	344	8·7	0

It was observed that activity of certain enzymes increased during the perfusion. It is difficult to explain this fact. The possibility of a release of such a great quantity of enzymes during the perfusion with liver was not confirmed in the experiments with animals. On the other hand, the activity of alanine transaminase decreased during perfusion to 16% of the initial values and the aspartate transaminase increased, thus resulting in high values of the index of the Ritis. This great increase in aspartate transaminase activity is caused by an increase in AspAT-IIR fraction, which is a mitochondrial enzyme. In the second hour of perfusion AspAT-IIR fraction amounted to 73% of the total AspAT activity

CONCLUSIONS

The following observations were made during 3-hour perfusion:

1) unbalanced respiratory alkalosis,

2) a decrease in bilirubin level in serum,

3) an increase of the bilirubin concentration in bile excreted by a heterologous liver,

4) a decrease in AlAT activity and an increase in AspAT, Asp, AT-IIR and GLDH.

High consumption of glucose during extracorporeal perfusion made necessary a constant controlling of glycemia and constant supplementation of blood sugar in the patient.

Translated by B. Lipiński, Ph. D.

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