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Organic Phosphate Compounds of Erythrocytes from Individuals with Cirrhosis of the Liver.* (29787)

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Comparatively little information is available concerning the metabolism of the red cells of cirrhotic individuals. Pitcher and Williams(1) noted that the red cells of patients with various types of liver diseases, including cirrhosis, had low levels of reduced glutathione despite the presence of normal activities of glucose-6-phosphate dehydrogenase and 6-phosphogluconate dehydrogenase. Bock *et al*(2) reported that the aldolase, triose isomerase, phosphoglyceraldehyde dehydrogenase, lactic acid dehydrogenase, and methemoglobin reductase activities were significantly increased in the washed red cells of cirrhotic patients. Italian investigators(3) noted that the concentration of ATP† was markedly decreased, ADP was significantly decreased, while AMP was nearly normal. The decrease in ATP appeared to be proportional to the severity of the disease syndrome.

The present investigation was undertaken to determine the concentrations of a number of phosphorylated compounds in the washed red cells of patients with cirrhosis of the liver.

Materials and methods. The following diagnoses were made on the 21 patients studied: Laennec's cirrhosis (12), post-necrotic cirrhosis(4), cirrhosis associated with hemochromatosis (2), cardiac cirrhosis (2), and acute yellow atrophy associated with serum hepatitis (L.B). Healthy young adults served as controls. In all cases, blood was drawn during the fasting state into acid-citrate-dextrose solution. The bloods of 5 patients were collected and stored in glass-stoppered tubes to determine the effect of storage at 4°C. The preparation of the erythrocytes, extraction with trichloroacetic acid, and the anion exchange analysis of the phosphorylated compounds(4) have been described(5). The phosphate concentrations are expressed as μmoles/g Hb since phosphorylated compounds have been shown to form complexes with hemoglobin(6).

Results and discussion. The analytical data for P_{total}, P_{org}, P_i, DPG, ATP, ADP, AMP, HDP, and IMP of freshly drawn erythrocytes from control and cirrhotic subjects are plotted in Fig. 1 and the statistical evaluation is presented in Table I. The statistically significant increase in P_{org} concentration

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† Abbreviations used in this paper are: P_{org}, organic phosphate; P_i, inorganic phosphate; P_{total}, total phosphate; DPG, 2,3-diphosphoglycerate; ATP, ADP, AMP, adenosine tri-, di-, monophosphate; HDP, hexose diphosphate; IMP, inosine monophosphate.

TABLE I. Effect of Storage at 4°C on Erythrocyte Acid Soluble Phosphates from Control and Cirrhotic Subjects. All values are expressed as μmoles P/g Hb.

Compound	Days storage	Controls Avg ± S.E.	Controls Min-max	Cirrhotics Avg ± S.E.	Cirrhotics Min–max	P*
P_{org}	0	47.6 ± 1.3 (12)	38.8 – 55.3	56.8 ± 2.2 (22)	35.2 – 85.0	<.01
	14	22.6 ± 1.3 (4)	20.1 – 26.5	30.8 ± 4.3 (7)	14.8 – 49.7	N.S.
	21	16.7 ± .7 (5)	14.8 – 19.1	19.3 ± 3.7 (5)	10.4 – 34.3	N.S.
P_i	0	3.3 ± .4 (12)	1.0 – 4.5	3.4 ± .3 (21)	1.5 – 5.8	N.S.
	14	30.8 ± 1.0 (4)	28.3 – 33.1	23.0 ± 1.8 (8)	17.4 – 33.1	<.01
	21	32.1 ± 1.0 (5)	26.3 – 35.8	34.2 ± 2.8 (5)	26.0 – 40.7	N.S.
DPG	0	26.8 ± 1.0 (12)	21.0 – 32.2	35.4 ± 1.2 (22)	26.2 – 50.9	<.001
	14	4.8 ± .9 (4)	2.3 – 6.4	13.8 ± 3.8 (7)	4.1 – 32.5	<.05
	21	.7 ± .1 (5)	.4 – 1.0	4.7 ± 1.0 (5)	.1 – 18.7	<.05
HDP	0	2.1 ± .2 (12)	.9 – 3.3	1.7 ± .2 (22)	.7 – 4.7	N.S.
	14	2.7 ± .5 (4)	1.7 – 4.2	2.9 ± .3 (5)	2.3 – 4.0	N.S.
	21	3.3 ± .8 (5)	.9 – 5.2	1.7 ± .4 (5)	.1 – 2.9	N.S.
IMP	0	.31 ± .16 (7)	.10 – .65	.20 ± .04 (5)	.11 – .35	N.S.
	14	.35 ± .04 (4)	.21 – .42	.54 ± .03 (5)	.46 – .63	<.01
	21	.50 ± .08 (5)	.40 – .90	.56 ± .07 (5)	.33 – .83	N.S.
ATP	0	9.8 ± .4 (12)	7.7 – 12.3	8.4 ± .6 (22)	4.1 – 14.6	N.S.
	14	7.9 ± .6 (4)	6.8 – 9.6	7.1 ± 1.0 (7)	2.7 – 11.1	N.S.
	21	5.8 ± .2 (5)	5.1 – 6.4	3.8 ± .6 (5)	1.6 – 5.6	<.02
ADP	0	1.9 ± .2 (12)	1.1 – 3.3	1.9 ± .2 (22)	.7 – 3.2	N.S.
	14	2.5 ± .1 (4)	2.0 – 2.8	1.4 ± .2 (7)	.2 – 2.1	<.01
	21	2.6 ± .1 (5)	2.2 – 3.0	1.5 ± .2 (5)	.9 – 2.1	<.01
AMP	0	.42 ± .06 (8)	.10 – .68	.26 ± .04 (12)	.06 – .52	<.05
	14	.55 ± .08 (4)	.34 – .74	.38 ± .05 (5)	.23 – .48	N.S.
	21	.92 ± .08 (5)	.71 – 1.17	.81 ± .12 (5)	.46 – 1.09	N.S.
ΣA†	0	4.5 ± .2 (8)	3.6 – 5.2	3.6 ± .4 (5)	1.9 – 4.2	N.S.
	14	4.6 ± .3 (4)	3.9 – 5.4	3.4 ± .5 (5)	1.6 – 5.2	N.S.
	21	4.2 ± .2 (5)	3.7 – 4.8	2.9 ± .3 (5)	1.5 – 3.7	<.01

* Student's "t" test was used to determine the P value.
† Sum of adenyl compounds (ATP, ADP, and AMP) expressed as μmoles adenine/g Hb.
Figures in parentheses represent No. of subjects. N.S., not significant; values for P >.05.

for the red cells of the cirrhotics is due chiefly to the increase in DPG. A small but significant decrease is noted in the AMP values. In the patient with acute yellow atrophy, the concentrations of red cell P_{org}, DPG, and HDP are elevated.

The effect of storing the bloods of cirrhotic patients for 2 and 3 weeks at 4°C on the concentrations of phosphorylated compounds in red cells is summarized in Table I. The DPG values of these red cells remain significantly elevated during storage; the IMP concentration is increased after 14 days. The values for ADP are decreased after 14 and 21 days and ATP is decreased after 21 days. The sum of the adenyl compounds is significantly less in the cells of the cirrhotics after 21 days of storage.

Effect of serum P_i concentration. Plasma P_i concentrations of 15 freshly drawn "cirrhotic" bloods are plotted against the respective red cell P_{org}, DPG, ATP, and P_i concentrations (Fig. 2). Inspection of these data shows that most of the P_{org} and DPG values are elevated within the control P_i range, while most of the ATP values fall in the lower half or below the control limits. Practically all the red cell P_i values are normal. In a previous report(5), which dealt with the analysis of phosphorylated compounds in the red cells of uremic patients, the P_{org} and DPG values were also elevated in bloods with normal plasma P_i concentrations. In these experiments, the P_{org}, ATP, and P_i values increased as the plasma P_i values increased, but a number of DPG values were within or below the normal range.

The significance of the elevated DPG and

the comparatively low ATP values in the "cirrhotic" red cells, particularly during storage of blood in the cold, is not clear. A partial inhibition in the phosphoglyceric acid kinase, for example, could result in the increase of DPG and the slightly lowered ATP concentrations. It is difficult to assess the possible influence of abnormal plasma components on the metabolism of the red cells.

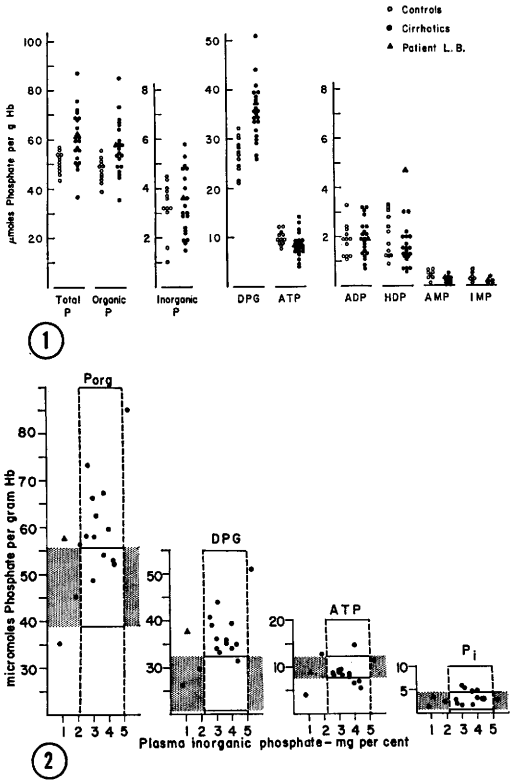

FIG. 1. Partition of acid soluble phosphate compounds in the erythrocytes from controls and from cirrhotics.

FIG. 2. Relation of plasma P_i to red cell phosphate compounds. The control ranges for the red cells and the plasma P_i are shown by the shaded area and by the dotted lines, respectively.

Summary. The concentrations of phosphorylated compounds of freshly drawn washed red cells of cirrhotic patients were determined. A significant increase in the values of P_{org} and DPG was observed in these cells and the remaining compounds, including ATP, were not appreciably affected. After storage of the bloods of cirrhotic patients for 14 and 21 days, the DPG levels were higher and the ADP levels were lower than those of the respective controls.

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Independence of Luteinizing Hormone and Adrenocorticotrophin Secretion in the Rat. (29788)

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Stresses of various sorts can decrease the weights of gonads and accessory sex organs in the rat(1). This has led to the view that stress may inhibit gonadotrophin secretion. Recently, Moore reported that sympatholytic and parasympatholytic drugs which blocked ovulation in rats also evoked adrenocorticotrophin (ACTH) discharge(2). When the rats became tolerant to the ACTH-releasing properties of these agents, they no longer blocked ovulation. Consequently he postu-

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