EFFECT OF ADRENALIN ON ALKALINE PHOSPHATASE ACTIVITY AND COAGULABILITY OF THE BLOOD

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The effect of adrenalin on alkaline phosphatase activity and coagulability of the blood was studied in experiments on 36 rabbits. Injection of adrenalin hydrochloride increased alkaline phosphatase activity in the blood serum and plasma and this is accompanied by the development of a hypercoagulemic reaction. Adrenalin does not directly activate alkaline phosphatase. Preliminary injection of acetylsalicylic acid in a dose causing acetylation of the cell membranes prevents the increase in alkaline phosphatase activity in the plasma produced by adrenalin but does not block the hypercoagulemic reaction.

The secretion of adrenalin by the adrenals is one of the most important mechanisms for hormonal regulation of the coagulability of the blood in emergency [8]. Increased production of adrenalin produces hypercoagulation in man and some mammals. On the other hand, evidence has recently been obtained that alkaline phosphatase participates both in blood coagulation and in fibrinolysis [2, 3, 10].

It was therefore decided to study whether correlation exists between the hypercoagulemic reaction and changes in alkaline phosphatase activity after injection of adrenalin.

EXPERIMENTAL METHOD

Four series of experiments were carried out on 36 rabbits in which the clotting time of the blood was determined in S.Ts. Bazaron's apparatus with automatic temperature regulation [5], and the alkaline phosphatase activity of the serum and plasma was determined [1] before, immediately, and 5, 15, 30, and 60 min after injection of adrenalin. Adrenalin hydrochloride was injected into the marginal vein of the ear over a period of 3 min in a dose of 0.5 ml of a 0.1% solution per kg body weight. Blood for testing was collected with graduated silicone-treated cannulas from the femoral artery. To obtain plasma, 1 ml of blood was immediately poured into a cold test tube with 1 mg dry heparin, mixed thoroughly, and centrifuged for 15 min at 3000 rpm.

The numerical results were subjected to statistical analysis [9].

EXPERIMENTAL RESULTS AND DISCUSSION

In the experiments of series I the clotting time of the blood in six rabbits immediately after injection of adrenalin was reduced from 1681 ± 57.2 (initial) to 1071 ± 46.5 sec ($P < 0.001$). The maximal reaction was observed after 5 min, after which the clotting time gradually returned to normal.

Immediately after injection of adrenalin in all cases there was some increase in the serum alkaline phosphatase activity. The activity of this enzyme 5 min after injection of adrenalin reached 9.04 ± 2.46 units (3.64 ± 1.07 units before injection; $P = 0.05$). The alkaline phosphatase activity still remained high 15 min later (mean 7.49 ± 1.33 units; $P < 0.001$).

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During the action of adrenalin the serum alkaline phosphatase activity thus increased parallel with the increase in coagulability of the blood.

Since alkaline phosphatase is present in the blood cells, the increased activity of the enzyme in the serum could be produced by destruction of the cells as a result of the more intensive clotting of the blood and retraction of the clot. In the next series of experiments on 11 rabbits clotting of the blood was accordingly prevented by the addition of heparin. Stabilization of the blood by the addition of sodium oxalate or citrate could not be used because these anticoagulants are inhibitors of alkaline phosphatase.

In these experiments, like the preceding group, a statistically significant ($P < 0.001$) acceleration of blood clotting was observed after the injection of adrenalin. The alkaline phosphatase activity in the plasma showed a parallel increase: immediately after the injection of adrenalin the activity of this enzyme was raised on the average by 45.4% ($P < 0.002$); 5 min later it was increased by 46.7% ($P < 0.05$).

No significant change in alkaline phosphatase activity was found in the heparinized plasma of eight control rabbits receiving an intravenous injection of physiological saline in the same volume as the adrenalin solution.

To discover whether adrenalin itself activates alkaline phosphatase in the serum and plasma, in seven experiments in vitro adrenalin was added to serum and plasma in a final concentration of 2 μg/ml (corresponding approximately to the concentration of this hormone in experiments in vivo). In no case was an increase in the activity of the enzyme found. Consequently, the increase in alkaline phosphatase activity in the experiments in vivo was connected with enzyme reaching the plasma from outside.

A possible explanation could be that under the influence of adrenalin alkaline phosphatase is liberated from platelets, leukocytes, erythrocytes, and endothelial cells of the blood vessels.

Experiments have shown [11, 12] that the liberation of procoagulants from platelets by the action of adrenalin can be prevented by acetylation of their membranes. The possibility cannot be ruled out that acetylation of the membranes may inhibit the analogous reactions of other cells in contact with the blood plasma. In the next series of experiments 11 rabbits were therefore given acetylsalicylic acid by mouth in a dose of 325 mg five times at intervals of 12 h. The last dose was given 2 h before the experiment. The original alkaline phosphatase activity in the plasma of these animals was found to be reduced on the average by 59.6% ($P < 0.01$) compared with the control. After injection of adrenalin the clotting time of the blood as a rule was reduced (as in the preceding series of experiments), but no significant changes in alkaline phosphatase activity were found throughout the experiment.

In the light of data showing the role of tissue thromboplastin [6] and alkaline phosphatase [4] in the membrane function of cells, the increased coagulability of the blood in rabbits receiving acetylsalicylic acid could be due either to liberation of membrane material possessing low phosphatase activity or to the direct activation of factor XII by adrenalin [7]. Another possibility is that the increase in alkaline phosphatase activity and the acceleration of blood clotting are two independent processes developing under the influence of adrenalin.

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