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INTRAERYTHROCYTIC CHANGES OF A METABOLIC REGULATOR, FRUCTOSE 1,6- DIPHOSPHATE (FDP) IN VITRO

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Fructose 1,6-diphosphate (FDP), a metabolic regulator of the glycolytic pathway in the red blood cells is known to have the important roles in regulating cellular function and metabolism. Intraerythrocytic changes of FDP, subjected to various environmental conditions were followed *in vitro*, using a modified enzymatic method of Beutler (1980).

Both human and rat blood samples were taken into the heparinized plastic tubes, plasma was removed by centrifugation and red cells were washed with isotonic phosphate buffer before incubated at 40C in diferrent media for 1 and 24 hours. Packed red cells were hemolysed with perchloric acid, followed by K2CO3 neutralization before the FDP determinations.

At physiological pH (7.4), the presence of glucose significantly elevated intraerythrocytic levels of FDP after incubated for 24 hours. In the contrary, citrate which has been used as anticoagulant in clinical blood sampling and storage was found to lower the levels of FDP when incubated with the red cells. The suppressive effect of citrate on intraerythrocytic FDP was pH dependent; i.e. acidic citrate (ACD, pH 4.8) was more effective in lowering the level of FDP than the less acidic citrate. The change in pH per se, as varying the pH (from 7.4, 6.4, 5.5 to 4.8) in the phosphate buffer, used for red cell incubation caused no change to intraerythrocytic FDP under this incubating condition.

The levels of intraerythrocytic FDP in the Diabetic (DM) and Thalassemia (THAL) patients were variably changed presumably by the pathological states. It could be due to the exposure of red cells to relatively high blood glucose in the diabetic patients that the intraerythrocytic levels of FDP were significantly elavated before the *in vitro* incubation, and then followed the normal pattern of FDP levels in blood from healthy volunteers, after incubated with the assigned medium for both 1 and 24 hours. This indicates that the glycolytic regulation of the DM's red cell is not defective. Levels of intraerythrocytic FDP in thalassemia were much elevated at pre- and post-1 hour incubations, while the levels of intraerythrocytic FDP after 24 hour-incubation of the splenectomized (SP) and non-splenectomized (NS) were in contrast different. The FDP level of the SP-THAL dropped down below but that of the NP-THAL continously elevated beyond their respective pre-incubation levels suggests that some glycolytic regulatory defects occur to the thalassemic red cells.