Earlier observations of ours have suggested that, under in vitro conditions resembling those under which platelets function haemostatically in vivo, their activation is promoted by the red cells. Some of the evidence suggested that this is through limited haemolysis with release of ADP. However, newly determined time relationships make this uncertain. Could red cells provide ADP without haemolysis? Or, is their flow properties affect the process more? To analyse the problem, we are determining dependence of red cell deformability on membrane constitution; and release of haemoglobin and adenine nucleotides under different conditions. Ten percent human red cell suspensions in physiological saline flow under constant pressures through 2, 3, 4 and 5 μm micropore filters, the flow rate measured continuously with an electronic balance. Initial flow rates are increased by fluidising agents, e.g. ethanol, and decreased by agents with opposite effect. Our results are consistent with the new hypothesis of S.J. Singer on the mode of action of amphipathic agents, such as chlorpromazine, on red cell membranes.

**ENZYMIC CLEAVAGE OF IN VIVO FORMED MAILLARD-TYPE COMPOUNDS INVOLVED IN HAEMOSTASIS. L. Mester, L. Szabados and M. Mester. Institut de Chimie des Substances Naturelles, Centre National de la Recherche Scientifique, Gif-sur-Yvette-91, France.**

Desoxyfructose derivatives of serotonin (Mester et al., 1975), of haemoglobin (Fückiger and Winterhalter, 1976), of poly-L-lysine (Mester et al., 1975) and of lysine rich histones (Kertesz-Crisba, 1977) are easily formed in vivo by a simple Maillard-type chemical reaction. Some of these compounds interfere with platelet functions (Mester et al., 1976) or contribute to the thickening of the basal membrane of blood vessels (Cerami et al., 1979).

While the chemical synthesis of Maillard-type compounds proceeds readily even in vivo, the chemical cleavage of them needs severer conditions which certainly do not exist in vivo (Gottschalk, 1952). However, a slow liberation of serotonin from desoxyfructose-serotonin is observed in vivo, suggesting the existence of an enzyme system for the cleavage of Maillard-type sugar-amine derivatives. In vivo, using a sheep liver microsomal preparation rich in Cytochrome P450 enzyme, the liberation of serotonin is in linear correlation with the enzyme concentration. The cleavage of desoxyfructose-serotonin is activated by NADPH having its optimum at pH=7.4, excluding definitely the occurrence of a chemical hydrolysis.

Factors interfering with the enzyme system involved in the cleavage of Maillard-type compounds, may also interfere with haemostasis.

**FREE ATP IN BLOOD DURING HEMORRHAGE. M.A.A. Kratzer and G.V.R. Born. Department of Pharmacology, University of London, King's College, Strand, London WC2R 2LS.**

When blood vessels are injured so that they bleed, circulating platelets adhere to the damaged vessel wall and aggregate within the first seconds. The mechanism of the initial platelet aggregation remains uncertain. To investigate the initiation stage of haemostasis, the carotid arteries of rats were punctured with a 100 μm needle and free ATP, as an indicator of CTP, was measured in the emerging blood. This was brought into contact with luciferin-luciferase in a polyethylene tube, internal diameter 0.8 mm. The light produced at the blood/enzyme interface was measured with a sensitive photon-counting device which gave background counts of 1 photoelectron/sec. and could detect < 10⁻¹⁰ M ATP in 2 μl blood.

When an artery was injured, the emerging blood contained about 10⁻⁷ M ATP in a first peak after about 2 sec. After about one min., the ATP concentration rose to a second peak of about 5.10⁻⁶ M. This was decreased by heparin (500 U/kg body weight) or by chlorpromazine (1 mg/kg). The observations suggest that the second peak represents ATP released from platelets. The source of ATP accounting for the first peak remains uncertain; possibly this ATP is released from red cells undergoing high shear stress from the haemodynamic effects of haemorrhage.