

RELEASE OF FIBRINOLYTIC ENZYMES BY MACROPHAGES IN RESPONSE TO SOLUBLE FIBRIN. L.A. Sherman, J. Lee, and C.C. Stewart. Washington Univ. School of Medicine, Dept. of Pathology, Dept. of Radiology (Section of Cancer Biology), and SCOR in Thrombosis, St. Louis, Missouri, U.S.A.

In previous data (J.Exp.Med. 147:76,1977), we have demonstrated that soluble fibrin/fibrinogen complexes are bound to the plasma membrane of guinea pig peritoneal macrophages. This binding is largely irreversible and is not a result of phagocytosis. We have extended our studies to examine the response in vitro of peritoneal macrophages to soluble fibrin/fibrinogen complexes. Unstimulated mouse macrophages were collected by peritoneal lavage and 5-60 µg of soluble fibrin/fibrinogen complexes placed into tissue culture dishes containing the unstimulated cells. Aliquots of the media were collected at 24, 48 and 72 hours. The cell-free media contained increasing amounts both of plasminogen activator and an enzymatic activity which resulted in fibrin and fibrinogen proteolysis independent of the amount of plasminogen present. The major proteolytic activity was due to the non-plasminogen dependent enzyme. Similar enzymes were released from peritoneal macrophages stimulated in vivo. The plasminogen activator enzyme had a low molecular weight comparable to that previously reported by Unkeless et al, with in vivo stimulation. Other coagulation moieties, such as plasmin and α-2 macroglobulin plasmin complexes did not result in release of the macrophage proteolytic enzymes. The results suggest that the previously described release of fibrinolytic enzymes after thioglycolate injections, may also result from the more pathophysiological stimulation by soluble fibrin/fibrinogen complexes. Release of these enzymes from phagocytic cells may be important, not only in blood clearance of soluble fibrin/fibrinogen complexes, but as part of thrombus reabsorption and wound healing.

ALTERATIONS OF PLASMIN ACTIVITY, PLASMINOGEN LEVELS AND ACTIVITY OF ANTI-PLASMINS DURING ENDOTOXIN SHOCK IN DOGS. A.O. Aasen, M.J. Gallimore, K. Ohlsson* and E. Amundsen. Institute for Surgical Research, Rikshospitalet, Oslo, Norway and Surgical Department, Malmö General Hospital, Malmö, Sweden.

Endotoxin shock was induced in dogs by intravenous infusion of a lethal dose of E.coli endotoxin over a period of 3 hours. Typical changes of cardiovascular parameters were found and evidence of an intravascular clotting process was observed. Spontaneous plasmin activity and "immediate" and "time dependent" antiplasmin activities were determined by means of assays utilizing the chromogenic tripeptide derivative S-2251 (Kabi Peptide Research Division, Mölndal, Sweden). Levels of plasminogen, α₂-macroglobulin (α₂-M), and α₁-antitrypsin (α₁-AT) were determined immunochemically. During shock, gradually decreasing values of "immediate" antiplasmin and α₂M were observed. During the late stages of shock "immediate" antiplasmin was found to be reduced by up to 89 per cent and α₂M up to 50 per cent of pre endotoxin infusion values. A less marked lowering of "time dependent" antiplasmin and α₁-AT also occurred during shock. These changes of plasma antiplasmins were accompanied by decreasing values of plasminogen and evidence of plasmin activity. These findings indicate that plasminogen is converted to plasmin during endotoxin shock and emphasize the role of antiplasmins in the pathophysiology of endotoxin shock.

THROMBOLYTIC EFFECT OF A PROTEOLYTIC ENZYME FROM ASPERGILLUS OCHRACEUS, AWEGILLASE^(R). H. Landmann and F. Markwardt. VEB Arzneimittelwerk Dresden and Institute of Pharmacology and Toxicology, Medical Academy Erfurt, GDR.

The mould *Aspergillus ochraceus* produces and liberates into the culture medium a proteolytic enzyme. It was highly purified and obtained in crystalline form by ethanol-calcium precipitation. The enzyme (molecular weight approximately 20,000) hydrolyses typical trypsin and chymotrypsin substrates as well as proteins, such as casein, haemoglobin and fibrin. It is inhibited by blood plasma antiproteases. In animal experiments, the enzyme shows relatively low toxicity as well as antithrombotic and thrombolytic effects, even at doses lower than the plasma antiprotease level. Experimentally produced clotting and deposition thrombi in peripheral veins and arteries as well as in coronary arteries are removed by proteolytic degradation. Based on first clinical studies, the enzyme, Awegillase^(R), is considered a suitable tool for both local and systemic thrombolytic therapy.