

INHIBITION OF FIBRINOLYSIS, ASSOCIATED WITH ONCOGENIC TRANSFORMATION, BY NORMAL AND ANTIPLASMIN-DEPLETED HUMAN PLASMA. D.Collen, A.Billiau, J.Edy and P. De Somer. Lab. of Blood Coagulation, Dept. of Medical Research, and Lab. of Virology, Dept. of Human Biology, University of Leuven, Belgium.

When mixed cultures of mouse fibroblasts and mouse fibroblasts transformed with Kirsten murine sarcoma virus (Arch.ges.Virusforsch. 43, 345, 1973) were grown in petri dishes and overlayed with plasminogen-enriched casein, they showed foci of caseinolysis induced by plasminogen activator secreted by the transformed cells. Caseinolysis was inhibited by the addition of human plasma or bovine pancreatic trypsin inhibitor (BPTI) to the overlay, 1 ml of plasma being equivalent to  $67 \pm 18$  (mean  $\pm$  S.E.), kallikrein inhibitor (KI) units of BPTI.

The culture fluid from a human melanoma line (gift of Dr. G.Barlow, Abbott Labs, Ill.), induced lysis of a fibrin clot, 1 ml of culture fluid being equivalent to 25 CTA units of urokinase. Fibrinolysis was inhibited by addition of human plasma or BPTI, 1 ml of plasma being equivalent to  $94 \pm 34$  KI units of BPTI.

Specific removal of antiplasmin, the fast-reacting plasmin inhibitor from plasma, by immunoadsorption, completely abolished its inhibitory activity on caseinolysis and on fibrinolysis. This depleted plasma still contained normal levels of the known plasma protease inhibitors. It is concluded that antiplasmin is the only protein in human plasma capable of inhibiting the fibrinolysis associated with oncogenic transformation or neoplasia. Whether this effect is exclusively due to inhibition of formed plasmin or also to interference with plasminogen activation remains unsettled.

ON THE REACTION OF HUMAN PLASMIN WITH THE PRIMARY INHIBITOR OF PLASMIN FROM HUMAN PLASMA. Ulla Christensen. Chemical Laboratory IV, University of Copenhagen, Copenhagen, Denmark.

The interaction of human plasmin with the recently discovered primary inhibitor of plasmin from human plasma has been investigated. The inhibitor is an  $\alpha_2$ -glycoprotein with a molecular weight of 60000. Pure preparations of plasmin and the inhibitor were incubated for a specific time, after which residual plasmin was determined from measurements of the rate of consumption of  $\alpha$ -N-benzyloxycarbonyl-L-arginine ethyl ester. Experiments were made over ranges of both plasmin and inhibitor concentrations and for a variety of incubation times. The results show that the reaction consists of at least two steps: rapid, reversible formation of an enzyme-inhibitor complex with the dissociation constant,  $K = 3\text{nM}$ , followed by a slow, irreversible transition with a rate constant,  $k = 6.5 \cdot 10^{-3}\text{s}^{-1}$ . Amino acids with antifibrinolytic effect inhibit the formation of the first complex, but not its further transition.