

ISOLATION AND PROPERTIES OF HUMAN VASCULAR PLASMINOGEN ACTIVATOR. R. Allen, D.S. Pepper,
and J.D. Cash. S.E. Scotland Regional Blood Transfusion Service, Edinburgh, Scotland.

The vascular tree of cadaver legs was washed out with a detergent - glucose mixture and the soluble activator precipitated with 8% PEG 6,000. The redissolved precipitate was chromatographed on 4% agarose where it exhibited an apparent molecular weight of 111 resolved from lipids, factor VIII antigen, albumin and haemoglobin. The active material eluted in the void volume of G-200 Sephadex when using 0.15 M NaCl eluant, but was dissociated and retarded by re-chromatography in 1 M NaCl or lysine. The dissociated, purified enzyme had an apparent molecular weight of 56,000 and was a serine protease. The enzymic activity of the carrier complex was stable for several days at 4°C, but the dissociated enzyme was rapidly inactivated at 4°C.

FVIII: A HOMOLOGOUS SERIES OF COVALENTLY LINKED OLIGOMERS. C.G.Cockburn and Diana M.L.Reay. Department of Haematology, Institute of Child Health, London, England.

Purified human FVIII was electrophoresed through SDS agarose gels of different concentrations and through SDS polyacrylamide (PA) gels of exceptionally large pore size (3.75% monomer, 1% cross linking). The gels were calibrated (a) with the FVIII subunit (b) with dimethyl-suberimidate cross-linked human fibrinogen. After Coomassie-blue staining the PA gels were scanned densitometrically.

The results show that freshly purified FVIII at pH 6.5 exists as a homologous series of oligomers and that a linear relationship obtains between the mobility of oligomers 1-6 and log molecular weight. Another linear relationship exists between the higher oligomers due to their significantly raised mobility. This suggests that the higher oligomers have a compact structure which is still partially ordered after SDS denaturation. A plot of log oligomer molarity vs. log subunit number has a gradient of approximately -1, suddenly decreasing to -5.5 after the hexamer, strongly indicating a decrease in the rate of polymerisation after this point. Oligomers 1-6 comprised 88% of the total FVIII, and oligomers 7-9 a further 6%. FVIII left at room temperature for 24 hours associated into extremely large oligomers.

EXCHANGE BETWEEN INTRA- AND EXTRAVASCULARLY INJECTED RADIOIODINATED FIBRINOGEN. R. Rüegg and P.W. Straub. Hematology Division, Dept. of Medicine, Kantonsspital, University of Zurich, Switzerland.

To test the hypothesis that in certain diseases plasma fibrinogen may be degraded extravascularly and that circulating fibrin/-ogen degradation products (FDP) may stem from extravascularly degraded fibrin/-ogen, 8 patients with pleural or peritoneal effusions (4 transudate, 4 exudate) were given ¹²⁵I-fibrinogen i.v. and simultaneously ¹³¹I-fibrinogen intracavitarily. Measurements of plasma and effusion volumes allowed quantitation of the exchange of fibrinogen and its protein-bound derivatives. Plasma t/2 was shortened in 6/7 patients. At 48 hrs 7.6 ± 3.9% of i.v. injected radioactivity was found in the effusion (18% of it clottable, 24% protein-bound but unclottable), 4.8 ± 2.2% of intracavitarily injected radioactivity was found in plasma (29% clottable and 28% protein-bound unclottable). Immunoreactive serum FDP were elevated in 7/8, plasma euglobulin lysis time normal in all 7/7 patients. Effusion fluids were fibrinolytically active, usually showing higher FDP concentrations than serum. It is concluded that 1) the shortened plasma half-life of fibrinogen is due in part to loss into the effusion, 2) fibrinogen is degraded in effusions and 3) plasma FDP in patients with effusions may stem partly from extravascular fibrinogen proteolysis. The findings suggest cautious interpretation of FDP levels and fibrinogen turnover results in patients with suspected DIC and effusions.