

Poster  
Board  
P3-006

- 0664 PLASMA FIBRINOGEN AND FACTOR XIII DURING INTERMITTENT THROMBOLYTIC THERAPY  
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The precursor components of the fibrin crosslinking system, viz, fibrinogen, and Factor XIII have been studied in plasma from patients undergoing intermittent thrombolytic therapy. Fibrinogen was isolated as fibrin monomer in the presence of EDTA and Trasylol and the level of degradation of the subunits quantitated by densitometric scans after separation on SDS gels. Plasma samples were also clotted in the presence of calcium and the crosslinked subunits separated on SDS gels. Gels were radioiodinated using a chloramine T method, then sliced and counted to determine the amounts of  $\gamma$  dimer and  $\alpha$  polymers. Daily single infusion of streptokinase caused degradation of circulatory clottable fibrinogen such that about 80% contained degraded chains MW 25,000. In crosslinked fibrin prepared from the plasma there was no diminution of the relative amount of  $\gamma$  dimer formed but that of the high molecular weight polymers was greatly reduced. In the interval between infusion of streptokinase (12 hours) the fraction of undegraded fibrinogen returned to near normal which was associated with an increased capacity to form polymers. Three methods were used to determine Factor XIII levels during therapy, viz, dansyl cadaverine incorporation, immunoelectrophoresis ( $\alpha$  and  $\beta$  subunits) and the rate of formation of  $\gamma$  dimers and  $\alpha$  polymers using  $^{125}$ I fibrinogen free of Factor XIII. Over the course of the therapy levels fell by 30% of the pretreatment value using the first two methods, slightly greater losses being observed using the third procedure. The daily fluctuation of plasma levels observed with fibrinogen, pre and post streptokinase infusion did not occur with Factor XIII.

## Megakaryocytes

Level 5 - Red Side (Waterloo Foyer)

Discussion Group 12.00 - 12.45

- P5-025 0665 Separation of Man's and Rabbit's megakaryocytes populations from sternal bone marrow. H. DUPONT, M.A. DUPONT, F. LORIENT, M. BOISSEAU, H. BRICAUD - Unité de recherches Cardiológicas U8 INSERM et Laboratoire d'Hémostasiologie. Avenue du Haut-Lévêque - 33600 - PESSAC - FRANCE.
- In order to assess the role of the megakaryocytes disturbance in several pathological thrombotic situations, we designed a technic for concentration and separation of megakaryocyte population from bone marrow.
- Human bone marrow megakaryocytes were isolated from sternal puncture by two consecutive gradients of isomolar Bovine serum albumine (B.S.A.) solution in HANKS without  $Ca^{++}$  and  $Mg^{++}$  (Ph = 7.4) and density from 1.0024 to 1.0087 gm/ml at 1 g. Starting with suspensions of bone marrow cells containing only 0.04 % megakaryocytes, the proportion of megakaryocytes was enriched 50 times.
- Five populations of Rabbit megakaryocytes were separated by B.S.A. density gradient centrifugation from 1.050 to 1.098 gm/ml at 100 g during 20 mn. The cytophotometric evaluation of megakaryocyte ploidy degree required a novel separation according to the megakaryocyte size by velocity sedimentation in B.S.A. gradient at 1 g. By these technics, the proportion of megakaryocyte was enriched 60 to 100 times in each homogenous class. Then, qualitative and quantitative study were realized by cytophotometric evaluation of DNA-Feulgen at 560 nm and residues arginyls and lysyls in histones at 370 and 592 nm and correlated with ultrastructural features. Thus modifications of megakaryocytopoiesis in atherosclerotic coronary disorder in Man and experimental atherosclerosis in rabbit were recognized.

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