Effect of Chronic Venesection on Plasma Fibronectin

Thesis
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By

Somaya Mohamed Elhamy
M.B.B.Ch.,

Supervisors

Prof. Dr. Tarif Hamza Sallam
Prof. of Clinical Pathology
Faculty of Medicine
Ain Shams University

Prof. Dr. Aleya Sadek Mostafa
Ass. Prof. of Clinical Pathology
Faculty of Medicine
Ain Shams University

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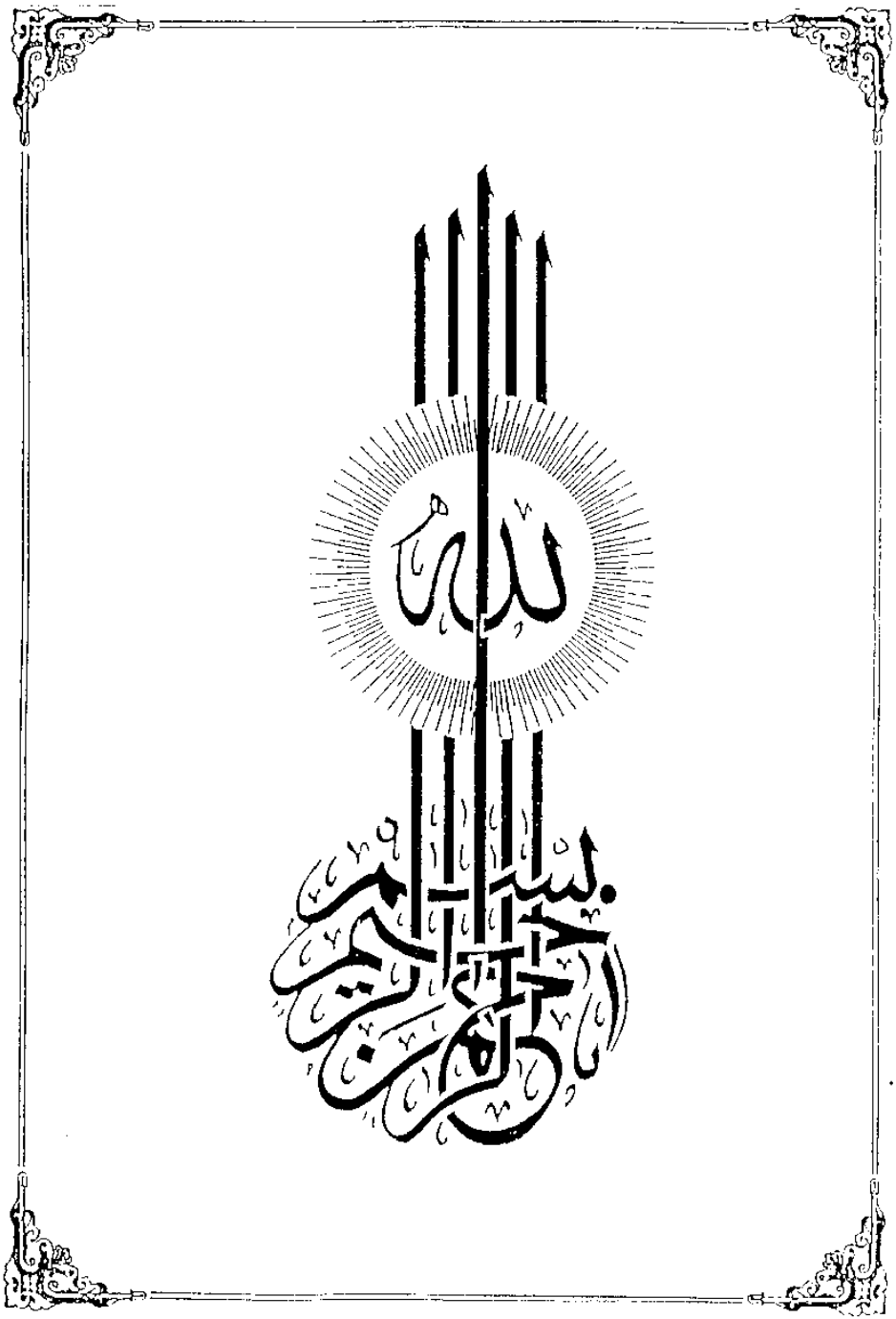
بسم الله الرحمن الرحيم

سُبْحَانَكَ لَا عِلْمَ لَنَا إِلَّا مَا عَلَّمْتَنَا

إِنَّكَ أَنْتَ الْعَلِيمُ الْحَكِيمُ



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INTRODUCTION

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Fibronectin is a large glycoprotein which is found in blood and other body fluids. It has been known by many other names, including cold insoluble globulin, α_2 surface binding opsonic protein, antigelatin factor, large external transformation sensitive protein, and, cell surface protein (Mosher, 1983). It is also found in α granules of platelets and platelets contain about 0.5% of the blood content of fibronectin , which is released when platelets are stimulated with thrombin or collagen.

In addition to its interaction with collagen, fibrin and heparin fibronectin interacts with other components of the haemostatic and fibrinolytic system. Fibronectin is a substrate for thrombin, plasmin and factor XIII a (Mosher et al., 1975). It enhances activation of plasminogen by urokinase (Iwanaga et al., 1978).

Plasma fibronectin is a major opsonin for cells of the mononuclear phagocytic systems, facilitating the clearance of particulate materials from the circulation.

Plasma fibronectin is decreased in patients

following major surgery or major trauma and in severly ill patient with evidence of D.I.C and acute leukaemia, the marked increase in fibronectin is found in deep dermis of scleroderma skin, parallel to the increase in collagen.

Repeated vensection results in loss of many important materials from the collected blood, on the contrary compansatory increase in other materials according to the relative rate of loss. Many changes in the haematological and immunological findings were reported in relation to repeated vensection, the results in these reports point to a probable change in plasma fibronectin being an important haematological and immunological factor.

So, the aim of this work is to determine the plasma fibronectine level in fifteen subjects exposed to repeated blood donation and also in fifteen normal control using single radial immunodiffusion method.

REVIEW OF LITERATURE

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Structure of Fibronectin

Structural studies of fibronectin derived from cell surfaces, tissue culture medium or extracellular fluids pointed to the presence of molecular heterogencity (Chen et al., 1977). Despite this, the outline of basic molecular architecture can be shown (Fig. 1).

Plasma fibronectin molecules have a molecular weight of 450,000 Daltons and composed of two similar but non identical polypeptide chains with estimated molecular weights of 220,000 and 230,000 (Mosher, 1975). The two chains are disulfide linked at the carboxyl end of the molecule (Balian et al., 1979). Intrachain disulfide bridges are clustered in the terminal thirds of each chain, most of them located at the amino terminal end (Iwanaga et al., 1978).

There is also a significant number of free sulfhydryl groups (Pearlestin et al., 1980).

Structural Model of Plasma fibronectin:

Most circulating molecules are composed of two more or less identical chains of approximate molecular weight 220,000 Daltons, each linked near the cooH -

terminal end by disulfide bridging (S-S). The NH_2 terminal is designated pyrrolidine carboxylic acid (PCA). The general location of interchain disulfide bridging is also depicted as are the approximate locations of sulfhydryl groups (SH), carhohydrate groups (CHO), and collagen-binding, fibrinagen binding, cell binding, cross linking and heparin binding sites. The dashed arrow indicate a region in the dimeric molecule which is cleaved during the course of hydrolysis by several proteolytic enzymes.

Several minor fibronectin components of smaller size than the two chain structure have been identified in plasma molecules and most of them range in size from 235,000 to 146,000 Daltons. It may be derived by catabolic processes from large parent molecules (Chen et al., 1976), and it is not yet known whether the degradative processes, they appear to reflect, occur intravascularly (Mossesson, 1980). Plasma fibronectin contains also 5% carbohydrate (Fukuda and Hakamari, 1979). All oligosaccharide units are linked to the peptide backbone by asparagine residues at 4-6 sites along the middle portion of the peptide chain (Yamada et al., 1977). The major sugars in

plasma molecules are mannose, galactose, N-acetyl glucosamine and sialic acid (Fukuda and Hakomori, 1979). It was suggested that carbohydrate fraction may serve to increase fibronectin resistance to proteolysis (Fukuda and Hakomori, 1979).

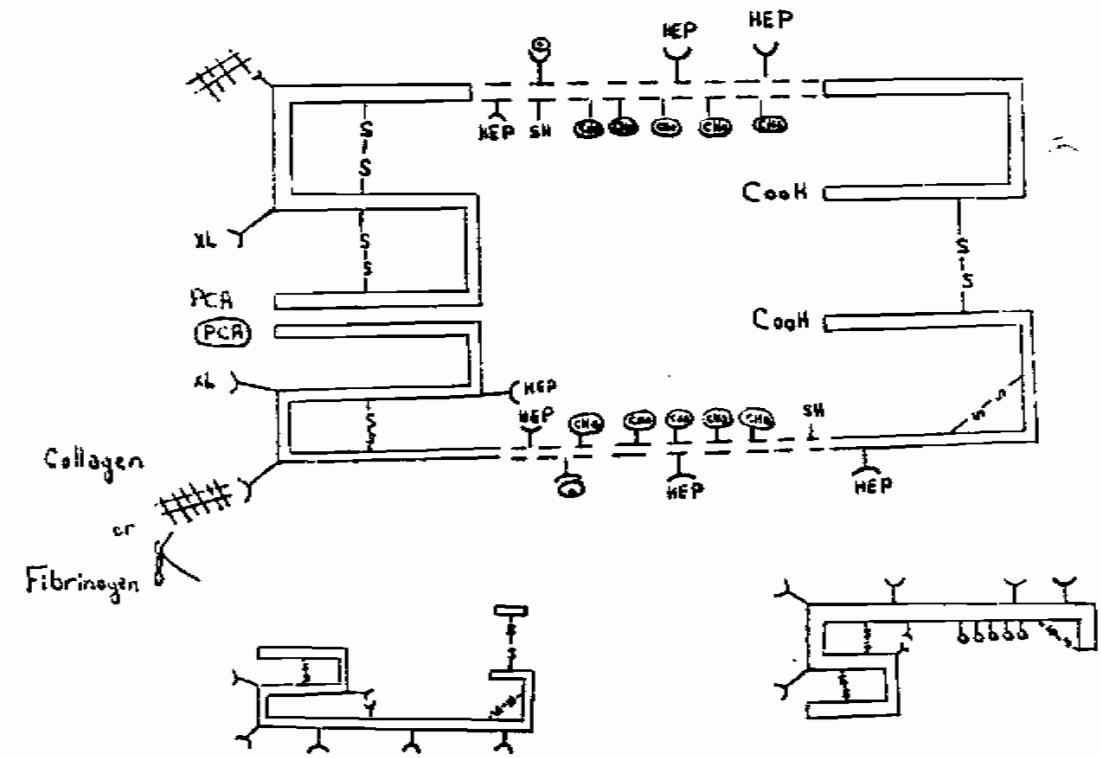

Fig. (1) Structural Model of Plasma fibronectin Mol. wt. 450,000

SYNTHESIS AND KINETIC

Although many cell types have the capacity to synthesize fibronectin evidence suggests that most, if not all, circulating fibronectin is produced by hepatocyte (Gonzlez et al., 1982).

Radiolabelled plasma fibronectin is cleared from the circulation in a complex manner after intravenous injection. Half life of it 24 to 72 hours to be estimated. The fates of fibronectin leaving the circulation are obscure. Immunofluorescence and tissue extraction studies, however, indicate that some of protein is deposited in extracellular tissue matrices (Sherman and Lee, 1982).

Fibronectin either in soluble or matrix form is sensitive to neutral tissue proteinases. Destruction of fibronectin may be critically involved after mast cell activation and in inflammatory conditions in vivo (Alan et al., 1981).

Physical Properties of Fibronectin

Plasma fibronectin is a large glycopratein having the ability to bind to collagenous constituents of connective tissue, to the intracellular macromolecules and DNA, to the plasma protein fibrinogen and amyloid component and to the surface of bacteria and eukaryotic cells (Mossesson and Amrani, 1980). These affinities provide a mechanism for its postulated roles in wound healing and in reticuloendothelial clearance of circulating particles, clot remnants and cellular debris (Saba and Jaffe, 1980).

Fibronectin levels are known to decrease following dissemenated intravascular coagulation (Mosher and Williams, 1978), blunt trauma, burn injury, surgery and particles induced reticuloendothelial blockage. Depletion of fibronectin in animals by injection of antifibronectin antisera results in diminished ability to survive trauma or bacterial sepsis (Lanser and Saba, 1982). Decreased levels of fibronectin have been correlated with depression of phagocytic clearance capacity by the reticuloendothelial system (Saba, 1970).

In animals intravenous adminsteration of purified

homologous fibronectin after surgery has been reported to reverse post operative phagacytic depression (Saba, 1980). So, in human plasma cryoprecipitate obtained from blood banks is used as a rick source of fibronectin in attemps to restore R.E.S. function and prevent organ failure in septic burn, surgery and trauma (Saba, 1978).

The adminstration of purified fibronectin would avoid overloading the patient with unneeded plasma proteins such as fibrinogen and would render the results less ambiguous.

Biological activities of the fibronectin

Fibronectin is a B-globulin with a sedimentation coefficient of about 13 and a molecular mass of 440 kilodaltons. It is composed of 2-2.2 x 10^5 dalton subunits. Fibronectin conc. in normal serum 20% to 50% less than in plasma. Concentration of fibrinogen in plasma is 2400 Ug/ml., of which 100% is incorporated into the clot, that of fibronectin which incerporated into the clot is 35% and that of α_2 plamin inhibitor in plasma of which 24% is incorporated into the clot.