ESTIMATION OF FIBRONECTIN AND ITS RELATION TO INFECTION IN PATIENTS WITH NEPHROTIC SYNDROME

THESIS

Submitted for Partial Fulfilment of

Master Degree in Internal Medicine

By

Dr. **TAREK YOUSSEF SAAD**

M.B.,B.Ch.

Supervised by

Prof. Dr. **BADAWI LABIB MAHMOUD**

Professor of Internal Medicine

Ain Shams University

Dr. **HANY ALI REFAAT**

Assistant Professor of

Internal Medicine

Ain Shams University

Dr. **MONA MOHAMED RAFIK**

Assistant Professor of

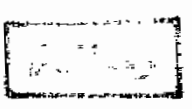
Clinical Pathology

Ain Shams University

Dr. **KHALED HUSSEIN ABOU-SEIF**

Lecturer of Internal Medicine

Ain Shams University



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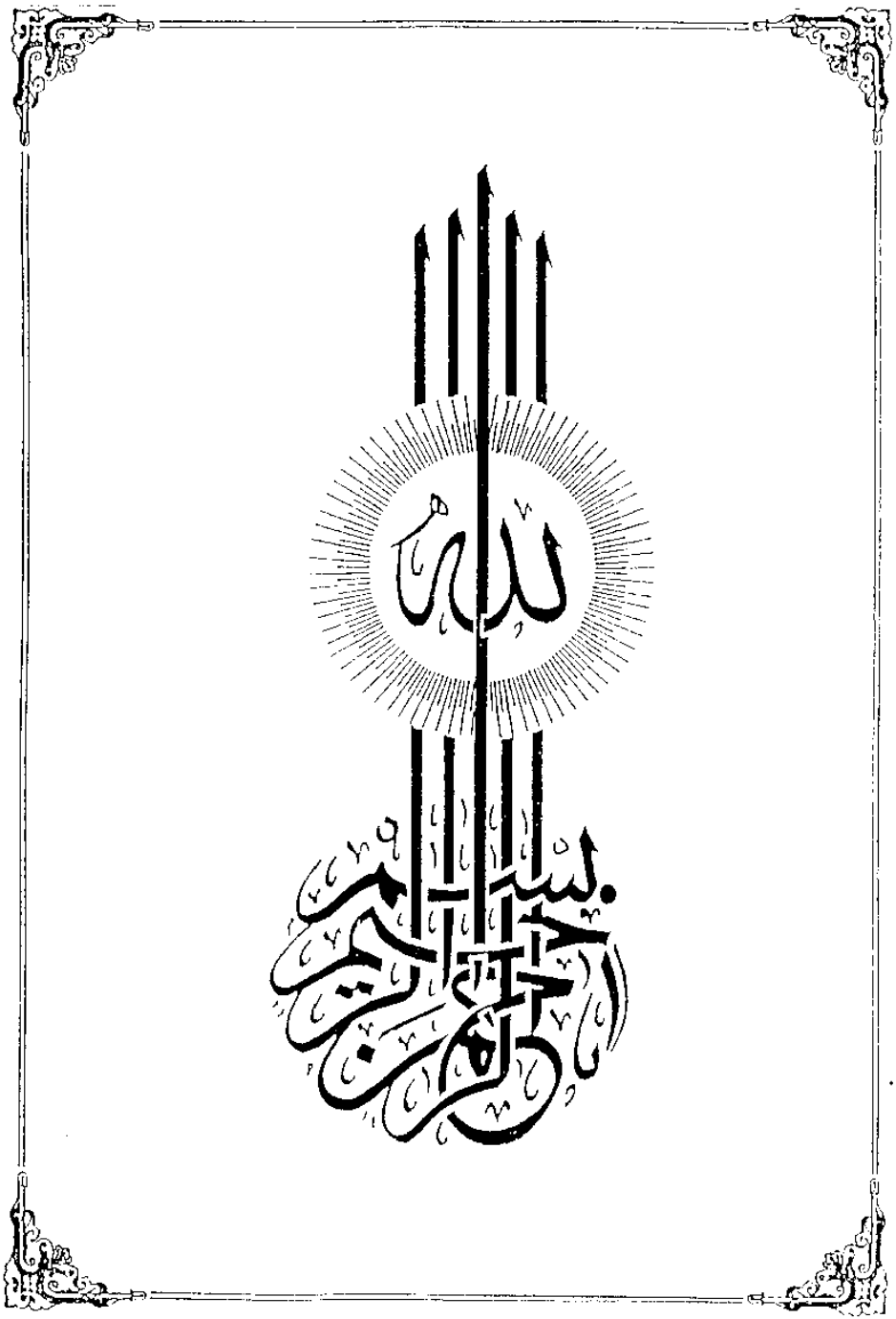

LIST OF ABBREVIATIONS

Ab	: Antibody.
AIDS	: Acquired immuno-deficiency syndrome.
ARC	: AIDS-related complex.
Arg	: Arginine.
Asp	: Aspartate.
BM	: Basement membrane.
C3	: Third component of complement.
CAPD	: Continuous ambulatory peritoneal dialysis.
CIG	: Cold-insoluble globulin.
CRP	: C-reactive protein.
CSF	: Cerebrospinal fluid.
DIC	: Disseminated intravascular coagulopathy.
dl	: Deciliter.
DNA	: Deoxy-ribonucleic acid.
ECM	: Extra-cellular matrix.
E. coli	: Escherichia coli.
EDTA	: Ethylene diamine tetra-acetate.
ELISA	: Enzyme-linked immuno-sorbent assay.
EM	: Electron microscope.
ESRD	: End-stage renal disease.
F	: Female.
Fig.	: Figure.
FN	: Fibronectin.
GBM	: Glomerular basement membrane.
GBS	: Group B streptococci.
Gly	: Glycine.
gm	: Gram.
GN	: Glomerulonephritis.
HCMV	: Human cytomegalo virus.
HD	: Haemo-dialysis.
hr.	: Hour.
HSPG	: Heparan sulfate proteoglycan.
HSV	: Herpes simplex virus.
IBM-PC	: IBM-personal computer.

IF	: Immuno-fluorescent.
Ig	: Immuno-globulin.
KDa	: Kilodalton (one dalton is the mass of one hydrogen atom).
Kg	: Kilogram.
L.	: Liter.
LETS	: Large external transformation-sensitive protein
LM	: Light microscope.
M	: Male.
m^2	: Square meter.
McAb	: Monoclonal antibody.
MCNS	: Minimal-change nephrotic syndrome.
mg	: Milligram.
ml	: Milliliter.
mm	: Millimeter.
MN	: Membranous nephropathy.
MPGN	: Membrano-proliferative glomerulo-nephritis.
nm	: Nanometer.
No.	: Number.
NS	: Nephrotic syndrome.
PAN membrane	: Polyacrilo-nitrate membrane.
PMNL	: Polymorphonuclear leucocytes.
RES	: Reticulo-endothelial system.
RNA	: Ribo-nucleic acid.
SDS-PAGE	: Sodium dodecyle sulphate polyacrylamide gel electrophoresis.
Ser	: Serine.
Staph. aureus	: Staphylococcus aureus.
Strept. pyogenes	: Streptococcus pyogenes.
TLC	: Total leucocytic count.
ul	: Microliter.
UTI	: Urinary tract infection.
vs.	: Versus.

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INTRODUCTION AND AIM OF THE WORK

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Within the complex architecture of a multi-cellular organism most normal cells keep their places by anchoring themselves to basement membranes and connective tissues made up largely of a fibrous mesh of proteins and other substances. In the adult body, few cell types routinely move through this extra-cellular matrix (ECM) and during embryonic development and wound healing certain cells migrate extensively. The movement is highly organized and most of the cells reach their destinations unerringly. How is this organization of cells, both fixed and dynamic, maintained ? Part of the answer lies in a variety of large glycoproteins that bind cells to the ECM. The best understood of those anchroing and organizing molecules are the versatile proteins known as fibronectins (Hynes, 1986). FN is clearly one of the glues in our body that provides important links to diverse kinds of molecules (Polin, 1990).

In 1969, Saba and Diluzio suggested that depletion of a plasma protein could be responsible for reticulo-endothelial system dysfunction. This protein, opsonic surface-binding glycoprotein, was identical to cold-insoluble globulin (CIG) or FN. FN is a glycoprotein of high molecular weight, 440 kilodaltons, present in plasma and other extracellular body

fluids and is important in adhesion of cell to cell and cell to substratum (Yamada and Olden, 1978), motility, and phagocytosis (Yamada et al., 1978). It participates in wound healing, blood coagulation and in immunologic clearance of injured tissue and of complement- or antibody-coated material such as bacteria, fungi, and effete cells. Both cell-associated and plasma FN play a key role in maintenance of microvascular integrity and vascular permeability (Polin, 1990). These biological activities are related to the specific binding of FN to a series of macromolecules including collagen, glycosaminoglycans, fibrin, and bacteria (e.g. Staph. aureus and Streptococci), (Engvall and Ruoslahti, 1977; Stathakis and Mosesson, 1977; Kuusela et al., 1985).

FN is increased in the expanded mesangium of a variety of glomerular disorders (Ikeya et al., 1985), and it has a key role in the initiation of formation of immune complexes in situ in some forms of glomerulonephritis (Lake et al., 1985).

The aim of this work is to study plasma and urinary levels of FN in patients with nephrotic syndrome with and without infection and their correlatoin with plasma and urinary proteins, kidney functions, and with total leukocytic count.

REVIEW OF LITERATURE

Chapter 1

FIBRONECTIN

FIBRONECTIN

DEFINITION :

The term fibronectin (FN) describes a family of structurally- and immunologically-related high molecular weight glycoproteins that are present on many cell surfaces, in extracellular fluids, in connective tissues, and in most basement membranes (Mosesson and Amrani, 1980; Cosio and Bakaletz, 1986).

This term was derived from the latin roots "fibra" meaning fiber and "nectere" meaning to link, bind, or connect (Ruoslahti et al., 1981; Hynes, 1986). It was created to emphasize the property of the protein to bind to fibrous proteins like collagen (Engvall et al., 1978), and fibrin (Ruoslahti and Vaheri, 1975).

FN molecules serve as cables and connectors. They can assemble into fibrils, bind to cells, and link cells to other kinds of fibrils in the extracellular matrix (ECM). FN, also, bears an important but still poorly understood relation to the internal organization of cells and its adhesive characters make it a crucial component of blood clots and of pathways followed by migrating cells (Hynes, 1986).

Two forms of FN have been recognized, a soluble form in

plasma and other body fluids like cerebrospinal fluid and amniotic fluid, and an insoluble form which is present on the surface of a range of different cells including hepatocytes (Owens and Cimino, 1982), endothelial cells (Jaffe and Mosher, 1978), fibroblasts, lipocytes (Stenman and Vaheri, 1978), and platelets (Plow et al., 1979), in extracellular spaces of connective tissue, and as a component of basement membrane (Mosesson and Amrani, 1980; Ruoslahti et al., 1981).

Plasma (soluble) FN, a 450 KDa heterodimer, is a major plasma protein (300-400 mg/L.), whereas cellular (insoluble) FN is mainly present as a polymer in the connective tissue matrix and on cell surfaces (Christiansen et al., 1988). Both forms are immunologically similar but differ in their solubility at neutral pH (Polin et al., 1989).

The relationship between plasma, and cell surface FN has not been well established, and it seems likely that a dynamic equilibrium exists between circulating, and cell-associated FN that a variation in plasma concentration may reflect important changes at the tissue level (Mosher, 1984).

Discovery and nomenclature :

Plasma FN was first described in 1948 by Morrison et al. as a major concomitant of fibrinogen. It was initially named "cold-insoluble globulin of plasma" (CIG). Physico-chemical analysis reported by Edsall et al. (1955) led to the suggestion that CIG was a modified dimer of fibrinogen. In 1957,