Markers of Endothelial Cell Injury in Chronic Glomerulonephritis in Different Stages of Chronic Kidney Disease

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Abstract

We conducted a study to examine to what extent the endothelium cells are injured in glomerulonephritis patients. Forty-five GN patients were divided in three groups; first was GN with normal kidney function, second GN renal impairment and third was GN on haemodialysis. vWF and CRP levels were measured in all three groups and compared to a healthy control group. All patients had biopsy proven GN, non- diabetic and normotesive. Venous samples were collected and laboratory investigations were made. vWF and CRP levels were measured using the ELISA. Our results have proven that endothelial injury is early in the glomerular disease, even before renal impairment occurs. Results from the second group have proven that renal impairment further deteriorates the endothelial cell injury. The hemodialysis group study revealed remarkable findings. First, CRF independently caused endothelial damage. Second, we concluded that haemodialysis improves endothelial function in glomerulonephritis patients but, this remains to be proven with a prospective study.

Keywords: *vWF, CRP, Endothelium, Glomerulonephritis.*

Abbreviations

ACE=angiotensin-converting enzyme.

ADMA=asymmetrical di-methyl arginine

ANCA=antineutrophil cytoplasmic antibodies (p=perinuclear,c=cytoplasmic)

ARIC=Atherosclerosis Risk in Community

BMI= body mass index

CAPD=continuous ambulatory peritoneal dialysis

CHF=chronic heart failure

CRP=C-reactive protein

CVD= cardiovascular disease

ECM= extracellular matrix

EDHF= endothelium-derived hyperpolarising factor

EDRF=endothelium-derived relaxing factor.

ELISA=enzyme linked immuno-sorbort essay

eNOS= endothelial nitric-oxide synthase.

ESRD=end-stage renal disease

ET-1= endothelin-1

GBM=glomerular basement membrane

GFR=glomerular filtration rate

GMP(cGMP)=cyclic guanosine mono-phosphate

GN=glomerulonephritis

GTP=guanosine tri-phosphate

Hb%=hemoglobin concentration

HD=haemodialysis

HDL=high density lipoprotein

HDx=heamodialysis

HIV=human immuno-deficiency virus

HUVECs= human umbilical vein endothelial cells

ICAM1=intercellular adhesion molecule 1

IgA= immunoglobulin A

IgG=immunoglobulin G

IL-6= interleukin-6

IV= intravenous

JAF=junction-associated actin filament system

LDL=low density lipoprotein

L-NAME=nitro-L-arginine methyl ester

L-NMMA= N_G-monomethyl-L-arginine

MAP=mitogen-activated protein

MCP1=macrophage chemotactic protein 1

MCSF=macrophage-colony-stimulating factor

MIA=malnutrition, inflammation and atherosclerosis

NF-B=nuclear factor –B

NO= nitric oxide.

PAI-1 = plasminogen activator inhibitor-1

PDGF=platelet-derived growth factor

PGI_2=prostacyclin

PPAR= peroxisome proliferator-activated receptor

TBRI= Theodor Bilharz Research Institute

TFPI= tissue factor pathway inhibitor

TNF-∞=tumour necrosis factor-alpha

t-PA=tissue-type plasminogen activator

USA= United States of America

VCAM-1= vascular cell adhesion molecule

VSMCs = vascular smooth muscle cells

vWf= von Willebrand factor

5-HT= sertonin

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Introduction

Despite significant progress in renal replacement therapy, the, mortality from cardiovascular disease (CVD) in patients with chronic renal failure (CRF) is many times higher than in the general population. The traditional risk factors are frequently present in CRF patients. However, based upon conventional risk factor analysis, these factors do not fully explain the extraordinary increase in morbidity and mortality in CVD among patients with CRF. Accumulating evidence suggests that CRF is associated with impaired endothelial cell function. In recent years, the role of endothelial dysfunction (ED) and excessive oxidative stress (OS) in the development of CVD has been highlighted **(Annuk etal, 2003).**

Chronic renal failure (CRF) is associated with an increased risk of ischaemic heart disease (IHD), but the mechanisms responsible are controversial. Endothelial dysfunction is unrelated to Low density lipoprotein (LDL) oxidation, suggesting that LDL oxidation might not be a major cause of vascular ED in CRF. In contrast vascular ED was more severe in CRF than in angina patients and is associated with increased acute- phase proteins and plasma cytokines, demonstrating a chronic inflammatory state. These observations may explain the VED and increased IHD risk of patients with CRF **(Bolton etal, 2001)**

Cardiovascular diseases (CVD), being mostly a clinical manifestation of atherosclerosis. It is now generally accepted that the first step in atherosclerosis is endothelial dysfunction. **(Pawlak etal, 2005)**

Hypertension in CRF is characterized by biochemical evidence of marked endothelial dysfunction, apparently more pronounced than in patients with essential hypertension (EH). Amplified endothelial activation in CRF probably contributes to the high rate of atherosclerotic complications in CRF. **(Cottone etal, 2002)**

It is probable that disturbances in fibrinolytic activity and endothelial dysfunction may play a role in vascular complications such as stroke or ischemic heart disease in CRF patients. **(Malyszko etal, 2003)**

von Willebrand factor (vWF), a procoagulant glycoprotein derived from endothelial cells and platelets, is involved in platelet adhesion and aggregation at sites of vascular injury, and serves as a carrier for coagulation factor VIII .Raised levels of plasma soluble vWF antigen (vWF:Ag) is an established index of endothelial cell activation and/or dysfunction in patients with atherosclerotic CVD, as well as in those at increased risk of CVD. Recently, large epidemiological studies have proved that high vWF; Ag concentration is an independent predictor of CVD-related ischaemic events in the general population. **(Borawski etal, 2001)**

C- reactive protein (CRP) is significantly increased in ESRD patients as compared with the normal controls. Chronic inflammation occurs in patients with ESRD, which might be aggravated by hemodialysis. Activation of the monocytes might be involved in the pathogenesis of inflammation in uremia. **(Liang etal, 2003)**

Glomerulonephritis

Definition

The term glomerulonephritis encompasses a range of immune-mediated disorders that cause inflammation within the glomerulus and other compartments of the kidney. Studies with animal models have shown the crucial interaction between bone-marrow-derived inflammatory cells and cells intrinsic to the kidney that is both fundamental and unique to the pathogenesis of glomerulonephritis. The mechanisms of interaction between these cells and the mediators of their coordinated response to inflammation are being elucidated. Despite these pathophysiological advances, treatments for glomerulonephritis remain non-specific, hazardous, and only partly successful. Glomerulonephritis therefore remains a common cause of end-stage kidney failure worldwide. Molecule-specific approaches offer hope for more effective and safer treatments in the future **(Briganti etal, 2006).**

Glomerulonephritis is a subject of confusion among health-care workers. Much of the confusion stems from the nomenclature, which attempts to encapsulate the aetiology, pathology, and clinical presentation. The aetiological description refers to primary (aetiology unknown, generally thought to be a manifestation of autoimmunity) or secondary (associated with one of several autoimmune, infectious, malignant, or metabolic diseases) forms of glomerulonephritis. The pathological description addresses glomerular involvement, cell involvement, and changes in non-cellular components of the glomerulus (panel 1). **(Kerr etal, 2001)**

Epidemiology

Many cases of glomerulonephritis result in mild, asymptomatic illness that is not recognised by the patient, is not brought to medical attention, and remains undiagnosed. The incidence and prevalence of such mild episodes of glomerulonephritis are unknown but could be substantial. Population-based screening studies have shown that evidence of kidney damage—proteinuria, haematuria, low calculated glomerular filtration rate, or a combination of these features—is present in 16% of adults in Australia and a similar proportion in the USA. **(Coresh etal, 2003).**

Although diabetic and hypertensive nephropathies are the major contributors, glomerulonephritis is likely to be the cause in a substantial proportion. The epidemiology of more serious and clinically apparent episodes of glomerulonephritis is better defined.

Accurate classification requires a histological diagnosis; consequently, the reported prevalence might vary according to local indications for renal biopsy. A study of all renal biopsies done in the Australian state of Victoria during 1995 and 1997 found that 21·5 people per 100 000 population per year underwent renal biopsy, yielding a yearly incidence of biopsy-proven glomerulonephritis of 12·3 per 100 000. The most commonly diagnosed types of glomerulonephritis in adults were IgA nephropathy, focal and segmental glomerulosclerosis, and vasculitis; those most commonly diagnosed in children were minimal change disease, focal and segmental glomerulosclerosis, lupus nephritis, and IgA nephropathy. A population based study spanning 1976–2002, which included 898 biopsy-proven cases of glomerulonephritis among 412,735 inhabitants of a region of

western France, documented similar incidence, prevalence, and pattern of disease and an increase in the incidence of crescentic glomerulonephritis over time **(Simon etal, 2004).**

Most patients with glomerulonephritis incur chronic kidney disease, with the attendant risks of premature cardiovascular disease and progressive kidney failure. The burden of glomerulonephritis is most clinically apparent in its contribution to end-stage kidney failure, which necessitates dialysis or transplantation. Glomerulonephritis is recognised as the second commonest cause of end-stage renal failure worldwide. In Australia, glomerulonephritis is the most common cause, leading to 27% of cases of end-stage renal failure in 2001; in the USA, however, glomerulonephritis ranks third behind diabetes and hypertension. The propensity for each type of glomerulonephritis to cause end-stage renal failure varies, but the types most frequently identified on biopsy are also those most likely to result in kidney failure. In Australia during 2001, the most frequently occurring types of biopsy-proven glomerulonephritis causing end-stage renal failure were IgA nephropathy (27%), focal sclerosing (14%), rapidly progressive/ crescentic glomerulonephritis (10%, including vasculitis and antiglomerular-basement-membrane disease), membranous (5%), and lupus nephritis (4%). **(McDonald and Russ 2002)**

Predisposing Factors

Predisposition to glomerulonephritis might also be hereditary, as is the case with Alport's syndrome, in which mutations in the *COL4A5* (X-linked), *COL4A3*, or *COL4A4* (both autosomal recessive) genes result in defective synthesis of collagen type IV with consequent dysfunction and inflammation of the glomerular basement membrane. **(King etal, 2002)**

Glomerulonephritis is more common in the male than in the female population. Lupus nephritis is the major exception; the frequency is two or more times higher among the female than among the male population. However, since the female to male ratio of systemic lupus erythematosus is roughly ten, yet that of nephritis is only two to three, male patients with lupus seem more likely to develop nephritis than female patients **(Sarnak etal, 2003).**

Defined populations are clearly at increased risk of glomerulonephritis. Children in less developed countries, and those in impoverished sections of more developed countries, such as Australian Aboriginals, are at increased risk of infection-associated glomerulonephritis after streptococcal skin or throat infections. This disease is now rare in more developed countries. Individuals with chronic infections with viruses, particularly hepatitis B or C virus and HIV, are at increased risk of several types of immune-mediated glomerulonephritis. **(Collins etal, 2003)**

Classification of Glomerulonephritis

Classification according to the clinical presentation is the simplest and most effective tool for the clinician (panel 2). Although each clinical presentation can be associated with several distinct types of glomerulonephritis defined by aetiology and pathology, the approach to diagnosis and management is best directed by the clinical presentation. Rapidly progressive glomerulonephritis is a medical emergency, demanding urgent histological diagnosis and treatment to prevent renal failure. Nephritic and nephrotic presentations generally indicate urgent renal biopsy to guide management and provide prognostic information.

Chronic glomerulonephritis is also an indication for diagnostic biopsy and for the initiation of therapy to slow progression of renal impairment and limit the consequences of renal failure. Asymptomatic urinary abnormalities require a less urgent approach— diagnostic tests including renal biopsy in some cases, or observation over time in others. **(Chadban etal, 2003)**