Correlation of Plasma Homocysteine Level and Vascular Access Thrombosis in Hemodialysis Patients

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List of Abbreviations

Ab	Antibody
ACA	Anticardiolipin antibodies
ACE	Angiotensin-Converting Enzyme
ADP	Adenosine diphosphate
Alb	Albumin
AVF	Arterio-Venous Fistula
bHcy	Bound Hcy
Ca	Calcium
CBS	Cystathionine B-synthase
CE-LIF	Capillary electrophoresis with laser-induced fluorescence detection
CH.GN.	Chronic glomerulonephritis
Chr.Pyeloneph.	Chronic Pyelonephritis
Cr.	Creatine
CRP	C-reactive protein
CVD	Cardio Vacsular Disease
DM	Diabetes Mellitus
EC	Electrochemical detection
EIA	Enzyme immunoassay
ESRD	End stage renal disease
fHcy	Free Hcy
GFR	Glomerular Filtration Rate
Hb	Hemoglobin
HCT	Hematocrit
HCV	Hepatitis C virus
Hcy	Homocysteine
Hcy-SR	Hcy-mixed disulfide
HDL	High density lipoprotein
HTN	Hypertension
IL	Interleukin
ISHD	Ischemic heart disease
LA	Lupus anticoagulant

LC	Liquid chromatography
LDL	Low density lipoprotein
Lp	Lipoprotein
MTHFR	Methylenetetrahydrofolate reductase
NO	Nitric Oxide
NTD	Neural Tube Defects
Obst.Uropathy	Obstructive uropathy
PAI-١	Plasminogen activator inhibitor type ١
PDGF	platelet-derived growth factor
PKD	Polycystic kidney disease
PLT	Platelets
PO_4	Phosphorus
TG	triglycerides
tHcy	Total Hcy
TNF	Tumor necrosis factor
t-PA	Tissue plasminogen activator
UV	Ultraviolet
vWF	von Willebrand factor

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INTRODUCTION

Homocysteine is a sulfar containing amino acid that result from demethylation of methionine (١).

Recently it has attracted considerable interest as it may, by several mechanisms mediate premature atherosclerosis and cardiovascular diseases (٢)

Vascular access integrity remains the Achilles heel of modern hemodialysis (٣). Without an adequate vascular access; hemodialysis efficiency is reduced, which results in increased mortality and morbidity (٤). Thrombosis is the primary cause of access failure in polytetrafluroethyelene grafts and native arteriovenous fistulas (٥). The access dysfunction due to thrombosis is the most common cause of hospitalization among maintenance hemodialysis patients (٦).

Serum total homocysteine usually increases in dialysis patients with end-stage renal disease (٧). The cause of hyperhomocysteinemia in CRF (chronic renal failure) is still under intensive scrutiny (٨). Hyperhomocysteinemia may still represent one of many factors in uremia, which contributes to increased cardiovascular risk (٩). Recently vascular access thrombosis in dialysis patients is claimed to be associated with hyperhomocysteinemia (١٠).

AIM OF THE WORK

The aim of this study is to evaluate the role of hyperhomocysteinemia as a risk factor for vascular access thrombosis in hemodialysis patients.

HOMOCYSTEINE

Introduction:

Homocysteine (Hcy) is a sulphur containing amino acid that consists of various forms: a protein-bound fraction (٧٠–٨٠٪), a free oxidized form (٢٠–٣٠٪) and a free reduced form (~١٪), which recently has attracted considerable interest as it may promote vascular disease as well as its plasma level elevation is associated with risk assessment and diagnosis of other clinical conditions.(١١)

Biochemistry:

General Metabolism

Hcy is an endogenous sulfur-containing amino acid intermediate of the essential amino acid methionine and is not obtained from the diet. An overview of the metabolic pathway is presented in Figure ٢. Methionine enters the one-carbon metabolic cycle either through the dietary consumption of methionine-containing protein or through endogenous protein breakdown. It is then converted intracellularly to *S*-adenosylmethionine, which functions as a universal methyl donor for a variety of important acceptors,

including nucleic acids, neurotransmitters, hormones, and phospholipids. *S*-Adenosylhomocysteine, a byproduct of these reactions, is hydrolyzed to form Hcy and adenosine. This reaction actually favors the production of *S*-adenosylhomocysteine, although the normally rapid egress or metabolism of intracellular Hcy and adenosine allows this reaction to continue forward (١٢). Hcy then follows one of the following two metabolic pathways:

(١) remethylation to methionine by methionine synthase using vitamin B١٢ (cobalamin) as a cofactor and ٥-methyltetrahydrofolate as a substrate; or, alternatively, by betaine/Hcy methyltransferase in the presence of betaine (in human subjects, the latter reaction is mainly confined to the liver and kidney);

(٢) transsulfuration to cystathionine by cystathionine _-synthase, in an irreversible vitamin B٦ (pyridoxal-٥-phosphate)-dependent reaction; cystathionine is then degraded by cystathionase to _-ketobutyrate, ammonium, and cysteine.

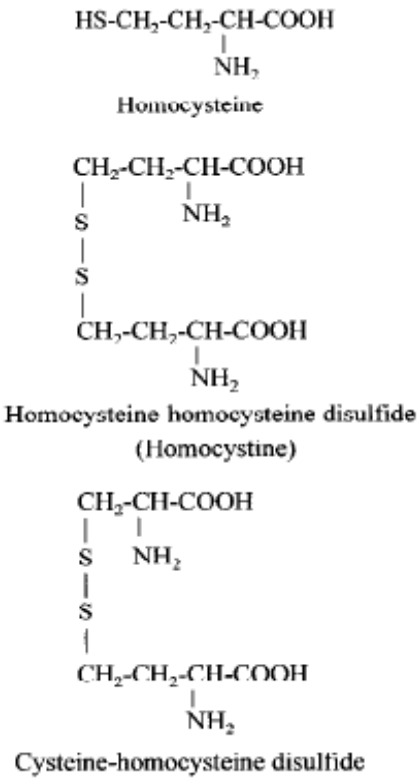

Figure 1. Molecular species of homocysteine.

Physiology:

Protein Binding

In normal subjects, approximately ٧٥٪ of total plasma Hcy is bound via a disulfide bond, to protein, primarily albumin, [bound Hcy (bHcy)], while the remaining ٢٥٪ exists in a free unbound form [free Hcy (fHcy)] (١١)

Hcy Flux

Hcy production occurs in all cells as a consequence of the normal methylation process. The Hcy volume of distribution in healthy subjects was observed to be approximately ٠,٤ L/kg, similar to that in subjects with severe renal insufficiency (١٣). Intracellular Hcy levels rise with enhanced intracellular Hcy production and/or inhibition of intracellular metabolism. To maintain low intracellular levels of this putatively cytotoxic substance, Hcy that is not metabolized within the cell is exported to the plasma compartment (١٤). Calculations based on steady-state kinetics in healthy adult humans estimate that ١,٢ mmol of Hcy, or approximately ٥ to ١٠٪ of the total daily cellular production, is delivered daily to the plasma compartment (١٥). Because Hcy is constantly produced and exported by cells, it must also be constantly cleared for plasma levels to remain within ١٠٪ of baseline values, as they do in healthy human subjects (١٦).

Normal Kidneys and Hcy Metabolism

The kidney seems to be just as capable of filtering and metabolizing Hcy as it does other amino acids. Hcy has a molecular mass of ١٣٥ D (١٧), which is well within the

filtration range of normal glomeruli. Assuming plasma fHcy concentration of ٣ uM and a normal GFR of ١٢٥ ml/min, the daily amount of filtered Hcy would be approximately ٠,٥ mmol. As with other amino acids, there is abundant evidence that filtered Hcy is avidly reabsorbed and only minimally excreted (٦ umol/d, or ١٪) in the urine (١٨).

Tubular uptake mechanisms specific for Hcy have been identified. Kinetic studies of minced rat renal cortical tissue identified low-*K*m/high-affinity and high-*K*m/low-affinity homocystine uptake systems, the former shared with cystine and the dibasic amino acids arginine, ornithine, and lysine (١٩). This finding is supported by studies in which rats and human subjects exhibited dramatically increased urinary homocystine excretion after intravenous boluses of arginine and lysine or aminoisobutyric acid (٢٠), an inhibitor of lysine, arginine, and ornithine tubular reabsorption.

Human kidneys contain the necessary Hcy-metabolizing enzymes, transsulfuration (cystathionine _-synthase and cystathionase) and remethylation (methionine synthase) enzymes in significant amounts (٢١). Compared with liver, the kidney contains more betaine.

Hcy methyltransferase and less cystathionase and methionine synthase (١٤). However, cystathionine _-synthase gene expression has been documented in the kidney (٢٢).