ORIGINAL ARTICLES

Post-transcriptional nature of uremia-induced downregulation of hepatic apolipoprotein A-I production

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Chronic kidney disease is associated with premature death from cardiovascular disease, which is, in part, driven by high density lipoprotein deficiency and dysfunction. One of the main causes of high density lipoprotein deficiency in chronic kidney disease is diminished plasma apolipoprotein (Apo)A-I level. Plasma ApoA-I is reduced in dialysis patients and hepatic ApoA-I messenger RNA (mRNA) is decreased in the uremic rats. This study explored the mechanism of uremia-induced downregulation of ApoA-I. Human hepatoma derived cells were incubated in media containing whole plasma or plasma subfractionation from normal subjects and patients with end stage renal disease pre- and posthemodialysis. Cells and culture media were isolated to measure ApoA-I protein and mRNA. ApoA-I promoter activity was measured using transfection with a luciferase promoter construct containing the –2096 to +293 segment of ApoA-I gene. Finally, effect of uremic and control plasma was assessed on ApoA-I RNA stability. Exposure to uremic plasma significantly reduced ApoA-I mRNA expression and ApoA-I protein production. These effects were reversed by replacing uremic plasma with normal plasma. Although no difference in ApoA-I promoter activity was found between cells exposed to uremic and normal plasma, uremic plasma significantly reduced ApoA-I RNA stability. Experiments using plasma subfractions revealed that the inhibitory effect of uremic plasma on ApoA-I mRNA expression resides in fractions containing molecules larger but not smaller than 30 kd. The pre- and postdialysis plasma exerted an equally potent inhibitory effect on ApoA-I mRNA abundance. Uremia lowers ApoA-I production by reducing its RNA stability. The inhibitory effect of uremic milieu on ApoA-I mRNA expression is mediated by non-dialyzable molecule(s) larger than 30 kd. (Translational Research 2013;161:477–485)

Abbreviations: Apo = apolipoprotein; CKD = chronic kidney disease; ESRD = end-stage renal disease; HDL = high density lipoprotein; mRNA = messenger RNA; qPCR = quantitative polymerase chain reaction

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AT A GLANCE COMMENTARY

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Background

Chronic kidney disease (CKD) is associated with premature death from cardiovascular disease which is, in part, driven by high density lipoprotein deficiency and dysfunction. high density lipoprotein deficiency in CKD is primarily caused by diminished plasma apolipoprotein (Apo)A-I level. Plasma ApoA-I is reduced in dialysis patients and hepatic ApoA-I messenger RNA is decreased in the uremic rats. This study explored the mechanism of uremia-induced downregulation of ApoA-I.

Translational Significance

Given the significant burden of atherosclerotic cardiovascular disease in CKD, understanding the mechanisms responsible for uremic dyslipidemia including downregulation of ApoA-I will be crucial to developing effective therapeutic options.

Approximately 9% of the United States population have chronic kidney disease (CKD), translating into 20 million adults.[1] Out of this staggering number, approximately 400,000 patients have advanced (stage 5) CKD requiring maintenance dialysis.[1] Five- year survival for patients with stage 5 CKD is about 35%, a mortality rate that is worse than that associated with many cancers.[2,3] Almost one-half of these deaths are attributed to premature cardiovascular disease.[2,3] Furthermore, there is an independent and graded association between reduced creatinine clearance and the risk of cardiovascular events, hospitalization, and death.[4] Cardiovascular disease in CKD is associated with and, in part, due to profound dysregulation of lipoprotein metabolism, which leads to a highly pro-atherogenic plasma lipid profile. This is marked by impaired clearance and accumulation of oxidation-prone intermediate density lipoprotein particles, chylomicron remnants, and small dense low density lipoprotein coupled with high density lipoprotein (HDL) deficiency and dysfunction.[5,6] The CKD-induced HDL abnormalities include reduction of plasma HDL cholesterol concentration and impaired maturation, antioxidant, anti-inflammatory and reverse cholesterol transport capacities of HDL.[7-10] Several factors contribute to HDL deficiency and dysfunction including reduced plasma levels of apolipoprotein (Apo) A- I and lecithin cholesterolacyltransferase, oxidative modification of HDL limiting its binding to the gateway of cholesterol efflux, and upregulation of acyl-CoA cholesterolacyltransferase impeding the release of free cholesterol from lipid laden cells in the artery wall.[9-15] Together, accumulation of oxidation prone, highly atherogenic and proinflammatory lipoprotein remnants and HDL deficiency and dysfunction opposed to the traditional hypercholesterolemia and elevated low density lipoprotein cholesterol are the primary features of CKD-induced dyslipidemia.[16-20] These lipid abnormalities most likely make a significant contribution to the atherogenic diathesis in this population. Accordingly, interventions aimed at alleviating inflammation and oxidative stress and enhancing HDL-mediated reverse cholesterol transport may confer greater cardiovascular protection in CKD population than the cholesterol-lowering therapies.

The critical role of HDL as the vehicle of reverse cholesterol transport and its effectiveness in prevention and impeding atherosclerosis have been demonstrated in many clinical studies.[21-24] The biosynthesis and maturation of HDL is a complex process involving production and release of its major apolipoprotein components, extraction of phospholipids and cholesterol from the target cells, and exchange of its lipid and apoprotein cargo with Apo B-containing lipoproteins in the circulation. The assembly of these different components leads to the generation of mature HDL particles. The major apolipoprotein of HDL is ApoA-I, which constitutes approximately 70% of the HDL protein content.[25,26] ApoA-I is synthesized in the liver and intestine. Deletion of the ApoA-I gene results in a significant reduction of HDL and increased risk of atherosclerosis.[27-29] Whereas its overexpression significantly increases serum HDL levels and inhibits progression of atherosclerosis and even causes regression of existing atherosclerotic plaques.[30-33]

Several studies in patients with CKD and end-stage renal disease (ESRD) have demonstrated the association of CKD with ApoA-I deficiency.[34-37] However, the available data on the underlying mechanisms of CKD-induced ApoA-I deficiency are limited. The present study was performed to determine the mechanisms by which uremia leads to downregulation of ApoA-I gene expression.

MATERIAL AND METHODS

Pooled plasma preparations. Pre- and postdialysis plasma samples from a group of 50 patients with ESRD were collected in heparinized tubes and pooled. Patients receiving 3-hydroxy-3-methyl-glutaryl-CoA reductase inhibitors and other lipid-altering medications and those with viral and bacterial infections or intercurrent acute