Review Article

Apolipoprotein J: A New Predictor and Therapeutic Target in Cardiovascular Disease?

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Abstract

Objective: To review the functional mechanism of apolipoprotein J (apoJ) in the process of atherosclerosis and the feasibility of apoJ as a therapeutic endpoint.

Data Sources: Relevant articles published in English from 1983 to present were selected from PubMed. The terms of "atherosclerosis, apolipoprotein J, clusterin (CLU), oxidative stress, and inflammation" were used for searching.

Study Selection: Articles studying the role of apoJ with atherosclerosis and restenosis after injury were reviewed. Articles focusing on the intrinsic determinants of atherosclerosis were selected. The exclusion criteria of articles were that the studies on immunologic vasculitis.

Results: ApoJ, involved in numerous physiological process important for lipid transportation and vascular smooth muscle cell differentiation, including apoptotic cell death, cell-cycle regulation, cell adhesion, tissue remodeling, immune system regulation, and oxidative stress, plays a role in the development of clinical atherosclerosis. In the process of relieving atherosclerosis, apoJ can promote cholesterol and phospholipid export from macrophage-foam cells, and exhibit cytoprotective and anti-inflammatory actions by interacting with lots of known inflammatory proteins which may predict the onset of clinical cardiovascular events and may actually play a causal role in mediating atherosclerotic disease such as C-reactive protein, paraoxonase, and leptin. As known as CLU, apoJ has been identified to play central roles in the process of vascular smooth cells migration, adhesion, and proliferation, which can contribute significantly to restenosis after vascular injury.

Conclusions: Intense effort and substantial progress have been made to identify the apoJ that relieves atherosclerosis and vascular restenosis after percutaneous coronary intervention. More work is needed to elucidate the exact mechanisms of and the interrelationship between the actions of apoJ and to successfully achieve regression of atherosclerosis by regarding it as a therapeutic endpoint.

Key words: Apolipoprotein J; Atherosclerosis; Coronary Artery Disease; Inflammation; Restenosis

Introduction

Coronary artery disease, also known as atherosclerotic heart disease,[1] is not only the most common type of heart disease and cause of heart attacks. Recently, treatment for coronary heart disease (CHD) with medication (including statin therapy to reduce low-density lipoprotein-cholesterol [LDL-C] levels significantly), percutaneous coronary intervention (PCI) (angioplasty) or coronary artery bypass surgery improved greatly,[2] despite all this, it was as of 2012, the most common cause of death in the world, and a major cause of hospital admissions.[3] Thus, better understanding of the pathogenetic mechanisms driving atherosclerosis in patients will ultimately be a key point to develop novel therapeutic modalities to futher reduce CHD risk.

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Epidemiological studies have identified an inverse association with the concentration of high-density lipoprotein-cholesterol (HDL-C) and the incidence of CHD in human prospective population studies. There is also circumstantial evidence from human intervention studies and direct evidence from animal intervention studies that HDLs protect against the development of atherosclerosis. A number of therapeutic strategies are being developed to

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target HDL-C in an attempt to halt the progression or induce regression of atherosclerosis and reduce cardiovascular events.[4] Several lifestyle and pharmacological interventions have the capacity to raise the level of HDL-C although it is not known whether all are equally protective. HDLs have several documented functions independent of "reverse cholesterol transport" (RCT), which is thought to be one of the most important mechanisms by which HDL protects against atherosclerosis, including inhibiting lipid oxidation, impairing leukocyte adhesion and monocyte activation, promoting nitric oxide production and flow-induced vasodilation, preventing endothelial cell damage and death, and inhibiting activation of platelets, and coagulation cascade[5] although the precise mechanism by which they prevent atherosclerosis remains uncertain. Nor is it known whether the cardioprotective properties of HDL are specific to one or more of the many HDL subpopulations that comprise the HDL fraction in human plasma. Recent study found that HDL-proteome remodeling plays a role for altered functional properties of HDL leading to altered vascular effects of HDL in coronary disease.[6] Therefore, detailed compositional assessment of HDL, including both lipid and protein components, coupled with sophisticated measures of HDL function, may yield mass-based biomarkers that can serve as proxies for function and that can then be applied with much greater ease and precision to large population and clinical trials. Apolipoprotein J (apoJ), also known as clusterin (CLU), which is tightly associated with both lipid and apoAl in HDLs in blood, has been proposed as such biomarker through numerous researches. This article aims to review the feasibility of apoJ as a therapeutic endpoint and to discuss what we still need to do in the future.

Characteristics

ApoJ, also known as testosterone repressed prostate message-2, sulfated glycoprotien-2, and Sp-40 and CLU, is a chaperone protein which may have either an intracellular or extracellular function. Its gene encoding a 449 amino acid protein is located on chromosome 8p21-p12.[7] As a secreted protein, it has a 22 amino acid signal peptide. The mature protein, which is a 75,000–80,000 secretory glycoprotein, undergoes a proteolytic cleavage, producing α- and β-chains, which are linked as a heterodimer by five disulfide bonds.[8] As an apolipoprotein, apoJ is found in a subset of dense HDL particles containing apoA-I and paraoxonase (PON)[9] and is found in most physiological fluids, including human plasma, urine, breast milk, semen, and cerebrospinal.[10] The wide distribution and sequence conservation of CLU suggest that this protein performs functions of fundamental biological importance. It is involved in numerous physiological process important for lipid transportation and vascular smooth muscle cell (VSMC) differentiation, including carcinogenesis and tumor growth, apoptotic cell death, cell-cycle regulation, DNA repair, cell adhesion, tissue remodeling, membrane recycling, immune system regulation, and oxidative stress.[11] In particular, apoJ plays a significant role in inflammation and immune responses through molecular interactions with complement factors, immunoglobulins, transforming growth factor-b, phosphorylated IkBa, and activated Bax.[12]

Apolipoprotein J and Atherosclerosis

Multiple factors play a role in the development of clinical atherosclerosis, including lipids, inflammation, physical sheer forces, and aging. This review is concerned with the role of apoJ in the each process of atherosclerosis.

Lipid transportation

In plasma, apoJ forms HDL particles with apoA-I and apoE and may play an important role in RCT from peripheral tissues to the liver,[8] which is thought to be the main role of HDL to protect against atherosclerosis. ApoJ was detected in the intima as well as the media in the early stage of atherosclerosis such as aortas with diffuse intimal thickening or fatty streaks, rather than in normal aortic walls, that is, those without any atherosclerotic lesions or diffuse, intimal thickening.[13] Because of the presence of apoJ in human and the function of apoJ-HDL particles, it has been hypothesized that apoJ has a protective role in atherosclerosis. Later confirmed this, apoJ can promote cholesterol and phospholipid export from macrophage-foam cells which constitute the hallmark cell type of atherosclerotic lesions.[14]

Inflammation and oxidative stress

Atherosclerosis is an inflammatory disease.[15] As a result of oxidative stress, inflammation is also a key factor in all aspects of coronary disease including the initiation and progression of atherosclerotic plaque, plaque rupture, and atherothrombosis, including in those with normal cholesterol levels and in those being treated with "statins" and antiplatelet agents.[16] ApoJ is a protein biosensor of oxidative stress and inflammation, which is upregulated in many pathological processes including atherosclerosis.[17] A few studies enter into apoJ and carotid atherosclerosis,[18] while apolipoprotein's expression on human carotid tissue and its association with parameters related to the disease development has been examined, which indicates that apoJ is association with oxidative and cellular stress.[19] There is mounting evidence that the chronic increase of proteins that participate in the acute inflammatory response may not only predict the onset of clinical cardiovascular events but may actually play a causal role in mediating atherosclerotic disease, such as C-reactive protein (CRP), PON, and leptin.

Apolipoprotein J and C-reactive protein

CRP was the first acute-phase protein identified and thus is the best studied marker of inflammation in humans. In certain studies, CRP was a more powerful predictor of cardiovascular risk than traditional risk factors such as LDL.[20] Its production increases during chronic vascular inflammatory by various stimuli, such as oxidative stress, shear stress, or infection.[21] For example, in the patients with metabolic syndrome, which greatly increases the risk of clinically significant atherosclerosis, CRP levels are also increased. Similarly, plasma apoJ levels were higher

in subjects with metabolic syndrome than in those without metabolic syndrome and showed an upward trend with increased metabolic syndrome component numbers.[22] This suggests that apoJ may increase the risk of clinical atherosclerosis.[23] It is possible therefore that the plasma apoJ level associated with plasma CRP level may be a potential biomarker of cardiovascular atherosclerosis disease.

Apolipoprotein J and paraoxonase 1

In vitro studies suggest that PON1, of which activity and/or protein levels are inhibited during the acute-phase response in animals[24] and also in humans,[25] is found in a subset of dense HDL particles containing apoA-I and apoJ and inhibits lipid peroxidation or degrades biologically active LDL oxidation,[26] which is thought to be one important mechanism for converting the lipoprotein to a form that promotes the formation of lipid-laden macrophages, the cellular hallmark of the early atherosclerotic lesion.[27] A deficiency of PON1 enhanced atherosclerosis in hypercholesterolemic mice and was associated with an increase in oxidized phospholipids.[28] In animals, apoJ levels increase in situations in which PON1 levels decrease. For example, the apoJ/PON1 ratio is increased by feeding mice an atherogenic diet, by injecting mildly oxidized LDL into mice that are susceptible to atherosclerosis, or by inducing inflammation in rabbits, and the ratio of apoJ/PON is higher in individuals at risk of future clinical cardiovascular disease.[29] These results suggest that apoJ may have a deleterious effect on the antioxidation activity of PON1 and could have beneficial effects on the cardiovascular system.

Apolipoprotein J and leptin

Leptin has pleiotropic actions including regulation of vascular function, platelet aggregation, and angiogenic effects, which suggests that leptin may play a role in the development of cardiovascular atherosclerotic diseases.[30] *In vitro* studies indicate that raised serum apoJ and decreased leptin concentrations are associated with significant coronary arterystenosis and may therefore serve as markers of CHD.[31]

There has been a lot of research into the other biomarkers of inflammation associated with CHD, such as the peroxisome proliferator-activated receptor gamma, homocysteine, and adiponectin, etc. This results of a large number of studies have speculated that the serum levels and expression of apoJ play an important role in the pathogenesis of atherosclerosis, but so far the exact role and the molecular mechanism of action of apoJ in atherosclerosis have not been determined. Therefore, there is a lot of work to do in future, and in particular, the in-depth study of the correlation of apoJ and biochemical markers which has been identified, associated with pathogenesis of CHD will be an important breakthrough direction.

Apolipoprotein J and Restenosis

The long-term efficacy of PCI, a useful technique for treating patients with coronary atherosclerosis, is limited by vascular restenosis, which occurs in up to 8.9% of patients undergoing this procedure with drug-eluting stents.[32] Restenosis after PCI is a consequence of balloon-induced smooth muscle cell (SMC) migration, proliferation, matrix production, and remodeling. Numerous attempts to prevent restenosis after PCI have met with very limited success. This failure reflects the complexity of the pathophysiological process of restenosis. Our understanding of the cellular and molecular mechanisms of restenosis has made it difficult to identify appropriate cellular or molecular targets for therapy of restenosis after PCI. But is known VSMCs play a key role promoting intimal thickening and constrictive remodeling in the process of restenosis, and apoJ is a potent inhibitor of VSMC migration, adhesion, and proliferation. Its genetic targets are linked to cell senescence and differentiation.[33]

Recently, on the contrary, a study reported that reduced CLU expression reduced the proliferation of VSMCs and induced G1 arrest via p53 and p21.[34] Likewise, Hamada *et al*. found that the atherosclerotic lesion of D-KO mice was significantly smaller than that of apoE-KO mice, suggested that apoJ is an atherogenic factor;[35] however, they also deduced that apoJ deficiency did not reduce atherosclerosis via lipoprotein metabolism, which was due largely to that there were no significant differences in total cholesterol, HDL cholesterol, and triglyceride levels between apoE-KO and D-KO. So why would apoJ in restenosis after injury actually do the reverse? Let us try to deduce what could have contributed to the problem in this review.

ApoJ, known as CLU, plays a dual role as a tumor suppressor and a tumor promoter. This diverse set of functions can be attributed to the existence of two alternatively spliced forms, secretory clusterin (sCLU) and nuclear clusterin.[36] The sCLU form is cytoprotective, for example, Kim *et al*. reported that sCLU inhibited the proliferation and migration of VSMCs.[37] Another researchers could further illustrated in their report, in which, they found that atRA-induced proliferative arrest and apoptosis of intimal SMCs are associated to a shift of CLU isoforms, in particular sCLU reduction, which is associated with proliferative arrest and re-differentiation, whereas Bax-related apoptosis is associated with n-CLU overexpression.[38] In view of the above, the role of CLU and its isoforms in SMC behavior is complex and chronologically regulated in response to microenvironmental changes. Further studies, therefore, are needed to verify if the modulation of CLU isoforms may represent a target in the pharmacological control of human vascular diseases, including restenosis.

Conclusions

ApoJ, as a multifunctional protein, which has been identified to be involved in all stages of atherosclerosis, may be an effective and sensitive prognostic indicator and biomarkers of atherosclerosis lesions and cardiovascular disease. Currently, there have been a lot of research into the exact role of apoJ in atherosclerosis such as mechanisms of expression of apoJ induced by a series of signal transduction pathways in the acute progressive phase of type 2 diabetes

and CHD (for example, activation of ROS system, protein glycosylation, apoptosis of VSMCs and oxidation of LDL, etc.), the intracellular sites of biosynthesis and exact biological effects of apoJ (for example, the role of pro-apoptotic or anti-apoptotic pathways, the regulation of cholesterol transportation and the internalization process of apoJ binding to its specific receptors, etc.), but none of them are definite. Therefore, additional evidence are required to corroborate preliminary associations among the physiological mechanism of apoJ, mass-based proxies of function, and atherosclerotic risk to firmly establish reliable measures and ultimately facilitate comprehensive assessment of apoJ-directed drugs in development.

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Conflicts of interest

There are no conflicts of interest.

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