commentary

HDL and innate immunity: a tale of two apolipoproteins[1]

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In addition to the well-recognized transport function of lipoproteins, a large body of evidence has demonstrated that lipoproteins also play an important role in host defense as part of the innate immune system (for review, see Ref. 1). One of the key defensive functions is the ability of HDL and other lipoproteins to bind endotoxin (lipopolysaccharide, LPS) and other bacterial products and neutralize their toxic effects. In this issue of the *Journal of Lipid Research*, Wang et al. (2) provide insights into the structural requirements for LPS neutralization by apolipoprotein A-I.

The helical structure of apolipoprotein A-I is based on eight similar 22 amino acid and two 11 amino acid tandem repeats, but the areas required for HDL formation and function can now be attributed to specific regions based on studies of specific mutations (3). The central region (amino acids 144–186) activates LCAT and contributes to HDL maturation and stability. The N- (44–65) and C- (220–241) terminal repeats are necessary to initiate lipid binding, form nascent HDL, and remove cholesterol from macrophages. A larger portion of the C-terminal region (190–243) is critical for phospholipid binding and promoting cholesterol efflux. Naturally occurring mutations of cysteines in apolipoprotein A-I, such as A-I_{Milano} and A-I_{Paris}, are associated with protection against atherosclerosis even when HDL cholesterol levels are decreased (4).

The paper by Wang et al. (2) addresses what regions of apolipoprotein A-I are required for neutralization of LPS by substituting other amino acids for specific cysteine residues. Serine substitution of one cysteine (228) in the C-terminal domain dramatically reduced the ability of HDL to neutralize LPS, while another C-terminal substitution (cysteine 195), proximal to the last 22 residue repeat, had little effect. Midregion substitutions (cysteines 107, 129, and 173) also had little effect. On the other hand, substitution in the first N-terminal repeat (cysteine 52) and especially the next region (cysteine 74) formed HDL that was more effective at neutralizing LPS and protecting from LPS induced lung injury.

Thus, Wang et al. (2) have shown that, as with cholesterol and phospholipid metabolism, specific regions of apolipoprotein A-I are essential for LPS neutralization. Furthermore, the regions involved in LPS neutralization are different than those involved in cholesterol and phospholipid metabolism. For example, these authors have previously shown (5) that substitution for cysteine residues 129 and 195 impaired lipid binding, while substitutions at 173 and 195 impaired the ability of HDL to promote cholesterol efflux. In contrast, a substitution at 107 had an increased capacity to promote cholesterol efflux. As noted above, the substitutions at 52 and 74 enhanced the ability of HDL to neutralize LPS, yet these substitutions had no effect on HDL structure or the ability of HDL to remove cholesterol from macrophages. Consequently, the increase in protection from LPS makes the 52 and 74 substitutions "super" apolipoproteins A-Is for host defense, with little downside in adversely affecting reverse cholesterol transport and increasing the risk of atherosclerosis. It remains to be seen whether similar naturally occurring mutations occur in humans and whether such mutations in apolipoprotein A-I will have a beneficial effect during gram-negative infections. Additionally, studies should examine whether similar modifications of apolipoprotein A-I will also enhance the neutralization of other toxic bacterial products, such as lipoteichoic acid.

Infections activate Toll-like receptors stimulating the secretion of cytokines, which have profound effects on lipid and lipoprotein metabolism (for review, see Ref. 1). The changes in lipid and lipoprotein metabolism are part of the acute phase response (APR), a pattern best known for increases in serum proteins (6). Positive APR proteins are those whose circulating levels increase during the APR while negative APR proteins decrease. Two positive APR proteins, C-reactive protein and serum amyloid A, bind to lipoproteins and hence can be considered to be apolipoproteins. The increases in serum proteins during the APR are transcriptionally mediated and usually driven by activation of transcription at NF-κB and NF-IL-6 response elements (6). However, many of the changes in lipid and lipoprotein metabolism are part of the negative

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APR, driven by decreases in transcription. Furthermore, the changes in lipid metabolism often occur indirectly through decreases in certain pathways shunting substrate into other pathways. For example, the increase in serum triglyceride levels that characterizes the APR is in part mediated by a decrease in fatty acid storage and oxidation in adipose tissue and muscle, which, coupled with increased lipolysis, increases the flux of fatty acids to the liver. Hepatic fatty acid oxidation is also decreased, directing the fatty acids into triglyceride synthesis, resulting in increased VLDL formation and secretion (1). These changes are mediated by decreases in nuclear hormone receptors including RXR, PPAR-α, and PPAR-γ, as well as decreases in coactivators, such as PGC-1α and β (1, 7).

Similar to apolipoprotein A-I, apolipoprotein E also plays an important role in host defense, neutralizing LPS, protecting from *Klebsiella pneumoniae* and *Listeria monocytogenes* infection, inhibiting malaria sporozoite invasion, and modulating T-cell activation (1, 8–10). Studies have shown that apolipoprotein E-deficient mice demonstrate increased toxicity to LPS administration or experimental *Klebsiella* or *Candida albicans* infections. Despite the importance of apolipoprotein E, the regulation of apolipoprotein E during the APR was confusing. Apolipoprotein E mRNA levels decrease in liver and in peripheral cells such as macrophages like a classic negative APR protein, but apolipoprotein E levels in the circulation are maintained or even increased in sepsis and HIV infection, consistent with apolipoprotein E being a positive APR protein (1, 11). In this issue of the *Journal of Lipid Research*, Li, Thompson, and Kitchens (12) clarify the mechanisms underlying the regulation of apolipoprotein E during infection. In mice, apolipoprotein E is mostly associated with HDL, yet Li, Thompson, and Kitchens (12) demonstrate that its clearance is mediated principally by hepatic LDL receptors. They show that during the APR, LDL receptor levels decrease in the liver, leading to a decrease in apolipoprotein E clearance, resulting in increased circulating apolipoprotein E. Thus, despite the decrease in apolipoprotein E mRNA levels in multiple tissues during the APR, serum apolipoprotein E levels actually increase and thereby could play an important role in host defense.

In contrast to the infection-induced decrease in hepatic LDL receptor levels, Li, Thompson, and Kitchens (12) further show that LDL receptors increase on macrophages, which would facilitate the delivery of lipids to the cells at the front line of host defense. This up-regulation of LDL receptors joins a growing list of changes that occur in the macrophage, which would promote lipid storage and foam cell formation (1, 13). Such changes may be beneficial from the perspective of host defense, as the macrophage utilizes neutral lipids to kill invading microorganisms (14). However, if such changes are chronic, it is likely that these changes will increase the risk of atherosclerosis.

From our modern perspective, we are most concerned with atherosclerosis, but we should admire the multiple mechanisms by which lipoproteins and apolipoproteins are active in fighting infection and inflammation. Perhaps we can learn to harness these evolutionarily old pathways to treat infectious diseases without worsening or even decreasing atherosclerosis.

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