

# The role of dysfunctional HDL in atherosclerosis

Mohamad Navab,<sup>1,\*</sup> Srinivasa T. Reddy,\* Brian J. Van Lenten,\* G. M. Anantharamaiah,<sup>†</sup> and Alan M. Fogelman\*

Department of Medicine,\* David Geffen School of Medicine at UCLA, Los Angeles, CA 90095-1679; and Atherosclerosis Research Unit,<sup>†</sup> Department of Medicine, University of Alabama at Birmingham, Birmingham, AL 35294

**Abstract** This review focuses on HDL function in modulating LDL oxidation and LDL-induced inflammation. Dysfunctional HDL has been identified in animal models and humans with chronic inflammatory diseases including atherosclerosis. The loss of antiinflammatory function correlated with a loss of function in reverse cholesterol transport. In animal models and perhaps in humans, dysfunctional HDL can be improved by apoA-I mimetic peptides that bind oxidized lipids with high affinity.—Navab, M., S. T. Reddy, B. J. Van Lenten, G. M. Anantharamaiah, and A. M. Fogelman. The role of dysfunctional HDL in atherosclerosis. *J. Lipid Res.* 2009. S145–S149.

**Supplementary key words** inflammation • high density lipoprotein • LDL • mimetic peptides • L-4F • D-4F

## THE ROLE OF HDL IN MODULATING LDL OXIDATION

Chisolm and colleagues (1) were the first to demonstrate that HDL protects against LDL-induced cytotoxicity. HDL was also found to prevent LDL oxidation by cultured endothelial cells (2–4). Mackness et al. (5) reported that an HDL-associated enzyme, paraoxonase (PON), was capable of preventing the accumulation of lipid hydroperoxides in LDL. We reported that cultures of human aortic endothelial cells and smooth muscle cells could oxidize LDL even in the presence of antioxidant containing serum inducing the cells to produce monocyte chemoattractant-1 (MCP-1) (6). HDL from normal subjects prevented cell-mediated oxidation of LDL and prevented the induction of MCP-1 by LDL (6). Watson et al. demonstrated that normal HDL contained platelet activating factor acetylhydrolase (7) and paraoxonase (8), which could prevent or destroy the formation of the LDL-derived oxidized phospholipids. Van Lenten et al. (9) reported that the activity of these

HDL-associated enzymes was reduced in rabbits and humans during the course of an acute phase reaction, resulting in HDL that increased cell-mediated LDL oxidation and increased MCP-1 production.

We reported (10) that apolipoprotein (apo)A-I and an apoA-I peptide mimetic removed seeding molecules from human LDL, rendering the LDL resistant to oxidation by human artery wall cells. The apoA-I-associated seeding molecules included hydroperoxyoctadecadienoic acid (HPODE) and hydroperoxyicosatetraenoic acid (HPETE). LDL from mice genetically susceptible to fatty streak lesion formation was highly susceptible to oxidation and was rendered resistant to oxidation after incubation with apoA-I in vitro. Injection of apoA-I, but not apoAII or murine serum albumin, into mice rendered their LDL resistant to cell-mediated oxidation. Infusion of apoA-I into humans also rendered their LDL resistant to cell-mediated oxidation. It was concluded that 1) oxidation of LDL by artery wall cells requires seeding molecules that include HPODE and HPETE; 2) LDL from mice genetically susceptible to atherosclerosis is more readily oxidized by artery wall cells; and 3) normal HDL and its components can remove or inhibit the activity of lipids in freshly isolated LDL, which are required for oxidation (10).

We also demonstrated (11) that treatment of human artery wall cells with apoA-I, but not apoA-II, or treatment with an apoA-I peptide mimetic, or with normal HDL, or paraoxonase, rendered the cells unable to oxidize LDL. Human endothelial cells were found to contain 12-lipoxygenase (12-LO) protein. Transfection of the cells with antisense to 12-LO (but not sense) eliminated the 12-LO protein and prevented LDL-induced MCP-1. Addition of 13(*S*)-HPODE or 15(*S*)-HPETE dramatically enhanced the nonenzymatic oxidation of both 1-palmitoyl-2-arachidonoyl-*sn*-glycero-3-

*This work was supported in part by US Public Health Service grants HL-30568 and HL-34343 and the Laubisch, Castera, and M.K. Grey Funds at UCLA. M.N., S.T.R., G.M.A. and A.M.F. are principals in Bruin Pharma and A.M.F. is an officer in Bruin Pharma.*

*Manuscript received 8 October 2008 and in revised form 23 October 2008.*

*Published, JLR Papers in Press, October 27, 2008.  
DOI 10.1194/jlr.R800036-JLR200*

Abbreviations: apo, apolipoprotein; CHD, coronary heart disease; HII, HDL-inflammatory index; HPETE, hydroperoxyicosatetraenoic acid; HPODE, hydroperoxyoctadecadienoic acid; 12-LO, 12-lipoxygenase; MCP-1, monocyte chemoattractant-1; PAPC, 1-palmitoyl-2-arachidonoyl-*sn*-glycero-3-phosphocholine; PON, paraoxonase.

<sup>1</sup>To whom correspondence should be addressed.

e-mail: mnavab@mednet.ucla.edu

phosphocholine (PAPC) and cholesteryl linoleate. On a molar basis 13(S)-HPODE and 15(S)-HPETE were approximately two orders of magnitude greater in potency than hydrogen peroxide in causing the formation of oxidized phospholipids from PAPC. Purified paraoxonase inhibited the biologic activity of these oxidized phospholipids. HDL from 10 of 10 normolipidemic patients with coronary artery disease failed to inhibit LDL oxidation and failed to inhibit MCP-1 production, whereas HDL from 10 of 10 age- and sex-matched control subjects did. It was concluded that 1) mildly oxidized LDL is formed in three steps, one of which involves 12-LO and each of which can be inhibited by normal HDL, and 2) HDL from at least some coronary artery disease patients with normal blood lipid levels is defective both in its ability to prevent LDL oxidation and in its ability to inhibit the biologic activity of oxidized PAPC (11).

#### USING A CELL BASED ASSAY TO SELECT APOA-I MIMETIC PEPTIDES

Garber et al. (12) reported that HDL from mice fed an atherogenic diet and treated with vehicle or treated with injections of mouse apoA-I had dysfunctional HDL in the cell based assay described above (9–11). In contrast, mice treated with an apo A-I mimetic peptide (5F) had anti-inflammatory HDL and a reduction in atherosclerotic lesion area. Thus, this assay appeared to have the potential to predict the response of mice to diet induced atherosclerosis.

This cell based assay was also used by Datta et al. (13) to select peptides for testing in mouse models. They studied a series of peptides, all of which bound nonoxidized lipids with equal high affinity. They found that the prototypic class A amphipathic helical peptide 2F and the peptides 3F3, 3F14, and 7F were relatively ineffective in preventing LDL-induced MCP-1 production in the cell based assay. In contrast, peptides 4F, 5F, and 6F were highly effective. Whereas the peptide 5F inhibited lesion formation in mice (12), the prototypic peptide 2F did not (13, 14). To eliminate the possibility that variation in the number of phenylalanine residues on the hydrophobic face was the major determinant of activity, peptides with three phenylalanines on the hydrophobic face were compared for their ability to bind nonoxidized lipids and were also compared in the cell based assay (15). The peptide 3F-2, but not 3F3 or 3F14, was found to be effective in the cell based assay although the binding affinity for nonoxidized lipids was found to be similar for all of the peptides (15).

Subsequently, Handattu et al. (16) demonstrated that 3F-2 reduced atherosclerotic lesions in apoE null mice, whereas 3F14 did not.

#### HDL AS A MEANS OF PREDICTING RISK AND ASSESSING RESPONSE TO TREATMENT

We reported (17) that HDL from 27 patients with documented coronary heart disease (CHD) failed to inhibit LDL-induced MCP-1 production, whereas HDL taken

from 31 age- and gender-matched healthy volunteers did (17). Ansell et al. (18) studied a group of 26 patients with CHD or CHD equivalents. Blood was taken from these individuals prior to and after six weeks of treatment with simvastatin. HDL was isolated from these individuals and studied in the cell based assay (18). The values obtained with LDL plus the test HDL were divided by the values obtained with LDL alone to give the HDL-inflammatory index (HII). Thus, LDL alone would give an HII of 1.0. Proinflammatory HDL was defined by an HII value  $\geq 1.0$  and antiinflammatory HDL was defined by an HII value  $< 1.0$ . Prior to treatment, the HII values for the CHD patients were  $1.38 \pm 0.91$  versus the controls whose HII values were  $0.38 \pm 0.14$ . After six weeks of simvastatin treatment, the CHD patients' HII values declined to  $1.08 \pm 0.71$ .

We reported (19) that inbred strains of mice that are susceptible to atherosclerosis all had proinflammatory HDL as assessed by the cell based assay, whereas inbred strains of mice that are resistant to atherosclerosis all had antiinflammatory HDL. We also reported that HDL from human subjects with proinflammatory HDL as assessed by the cell based assay was less effective in promoting cholesterol efflux from cholesterol loaded human monocyte-macrophages in vitro than HDL from human subjects with antiinflammatory HDL (19). Cruz et al. (20) reported similar findings (i.e., proinflammatory HDL in patients with leprosy was less effective in promoting cholesterol efflux from macrophages). Cruz et al. (20) also reported that proinflammatory HDL from leprosy patients was much less effective in promoting the conversion of monocytes into CD1b+ dendritic cells.

In cholesterol-fed rabbits, Van Lenten et al. (21) observed that HDL-cholesterol levels did not predict response to treatment with apoA-I mimetic peptides. In contrast, HII values significantly predicted lesion area. Lesion area was also predicted by serum amyloid A levels. Moreover, serum amyloid A levels and HII values were significantly correlated (21).

Bloedon et al. (22) found that 75% of patients with CHD or CHD equivalents, all of whom were on a statin, had proinflammatory HDL as determined with the cell based assay. Administration of a single oral dose of an apoA-I mimetic peptide resulted in low plasma levels ( $\sim 4$  nM) but was associated with a significant improvement in HII values compared with placebo (22).

#### HDL INFLAMMATORY PROPERTIES IN CHRONIC DISEASES THAT ARE ASSOCIATED WITH AN INCREASED RISK OF ATHEROSCLEROSIS OR WITH VASCULAR DYSFUNCTION

Women with systemic lupus erythematosus have a 7–50-fold increased risk of CHD. McMahon et al. (23) found that 44.7% of women with systemic lupus erythematosus and 20.1% of women with rheumatoid arthritis had proinflammatory HDL despite normal HDL-cholesterol levels. In contrast only 4.1% of healthy women had pro-inflammatory HDL (23).

Charles-Schoeman et al. (24) reported that administration of 80 mg/day of atorvastatin to patients with rheumatoid arthritis resulted in a significant improvement in HDL inflammatory properties whereas those receiving placebo had a significant worsening after 12 weeks of treatment.

Roberts et al. (25) reported that the HII improved in obese men with features of the metabolic syndrome despite a fall in HDL-cholesterol levels following a three week residential program which included diet and daily aerobic exercise.

Opole et al. (26) found that HDL inflammatory properties significantly improved following LDL apheresis in patients with familial hypercholesterolemia despite a decrease in HDL-cholesterol.

Dodani et al. (27) reported that the HII was correlated with carotid intima-media thickness in South Asian immigrants. Kalantar-Zadeh et al. (28) randomly selected 189 patients with chronic kidney disease who were on hemodialysis and followed them prospectively for 30 months. Subjects with proinflammatory HDL had significantly more comorbid conditions and a worse quality of life as determined by SF36 questionnaires. Thirty months after determination of the HII, patients with proinflammatory HDL had a significantly worse outcome as determined by Kaplan-Meier curves adjusted for case-mix variables as compared with patients with antiinflammatory HDL (28). There was no significant difference in total cholesterol levels, LDL-cholesterol levels, HDL-cholesterol levels, or triglyceride levels between the subjects with proinflammatory HDL and those with antiinflammatory HDL (28).

A study by van Leuven et al. (29) reported that patients with Crohn's disease had dysfunctional HDL and increased carotid intima-media thickness.

Weihrauch et al. (30) reported that HDL from patients with scleroderma and from mice with many features of human scleroderma was proinflammatory. In the mouse model, treatment with an apoA-I mimetic peptide improved the antiinflammatory properties of the HDL and also improved a number of the scleroderma-like features in these mice (30).

### HDL FUNCTION VERSUS HDL-CHOLESTEROL LEVELS

The studies reviewed above show that HDL inflammatory properties and the ability of HDL to promote cholesterol efflux from macrophages or to promote macrophage conversion to dendritic cells are correlated. Additionally, these studies show that HDL inflammatory properties are correlated with serum amyloid A levels and atherosclerotic lesion area in cholesterol-fed rabbits. However, none of these properties correlated with HDL-cholesterol levels in either animals or humans. Bhattacharyya et al. (31) reported that patients with CHD had significantly lower levels of HDL-cholesterol than normal controls but there was considerable overlap. The *PON1* genotype predicted a dose-dependent association with *PON1* activity and with levels of systemic indices of oxidative stress measured as

hydroxyecosatetraenoic and hydroxyoctadecadienoic acids and 8-isoPGF2 $\alpha$  levels. The adjusted hazard ratios for major adverse cardiac events between the highest and lowest *PON1* activity quartiles were independent predictors of events by multivariate analysis (31).

Wu et al. (32) reported that in the structure of apoA-I, tyrosine 166 is a preferred target for site-specific oxidative modification and this site directly interacts with and activates LCAT. Nicholls, Zheng, and Hazen (33) reported that myeloperoxidase preferentially associates with HDL, causing oxidative damage to apoA-I, which results in a loss in the ability of HDL to promote cholesterol efflux. Vaisar et al. (34) used shotgun proteomics to demonstrate that HDL in patients with CHD is enriched in apoE and in acute phase proteins. Subsequently, Green et al. (35) demonstrated that treatment with a statin and niacin decreased the apoE content of HDL and restored the proteome of HDL to resemble more closely that of healthy subjects.

We reported (36) that injection of oxidized phospholipids into atherosclerosis susceptible C57BL/6J mice, but not in atherosclerosis resistant C3H/HeJ mice, resulted in a decrease in *PON1* activity. Forte et al. (37) reported that mouse models of atherosclerosis have elevated plasma levels of oxidized phospholipids, which result in decreased activity of LCAT, *PON1*, and platelet-activating factor acetylhydrolase compared with controls.

Persegol et al. (38) reported that HDL from subjects with abdominal obesity was defective compared with normal HDL in counteracting the inhibitory effect of oxidized LDL on vascular relaxation.

### UNDERSTANDING THE ANTI-INFLAMMATORY PROPERTIES OF HDL THROUGH APOA-I MIMETIC PEPTIDES

Our group reported (39) that administration of an apoA-I mimetic peptide increased *PON1* activity in apoE null mice, improved the antiinflammatory properties of the HDL in these mice, and improved reverse cholesterol transport from macrophages. The maximal plasma concentration of the apoA-I mimetic peptide in these mice was  $\sim$ 130 nM. In the studies of Bloedon et al. (22), the maximal plasma concentration was  $\sim$ 4 nM. The concentration of apoA-I in the plasma of the mice (39) and in the plasma

TABLE 1. Affinity of phospholipids for L-4F and human apoA-I

Lipids	KD (nM)	
	L-4F	ApoA-I
PAPC	192,821 $\pm$ 56,505	99,871 $\pm$ 14,114
PGPC	7.2 $\pm$ 4.5	4,206 $\pm$ 4,228
POVPC	3.6 $\pm$ 2.6	30,852 $\pm$ 28,314
PEIPC	0.01 $\pm$ 0.01	50,720 $\pm$ 5,721
KODiA-PC	1.58 $\pm$ 3.5	20,960 $\pm$ 11,680

*Abbreviations:* PGPC, 1-palmitoyl-2-glutaroyl-*sn*-glycero-3-phosphorylcholine; POVPC, 1-palmitoyl-2-(5-oxovaleroyl)-*sn*-glycero-3-phosphorylcholine; PEIPC, 1-palmitoyl-2-(5,6-epoxyisoprostane E2)-*sn*-glycero-3-phosphorylcholine; KODiA-PC, 1-palmitoyl-2-(5-keto-6-octene-diyl)-*sn*-glycero-phosphatidylcholine. Adapted from Table 1 in reference 40.

TABLE 2. Affinity of nonoxidized and oxidized fatty acids for L-4F and apoA-I

Ligands	KD (nM)	
	L-4F	ApoA-I
<i>Nonoxidized Fatty Acids</i>		
Arachidonic acid	15.0 ± 1.3	10.5 ± 1.5
Linoleic acid	11.73 ± 3.5	6.55 ± 2.53
<i>Oxidized Fatty Acids</i>		
5(S)-HPETE	91.7 ± 15.6	1,582,000 ± 349,648
12(S)-HPETE	98.3 ± 34.2	822,600 ± 266,325
15(S)-HPETE	51.8 ± 11	1,046,400 ± 160,782
13(S)-HPODE	16.8 ± 4.3	1,230,800 ± 569,295
12(S)-HETE	23.4 ± 7	849,600 ± 327,395
15(S)-HETE	21 ± 6.8	1,289,400 ± 139,245
9(S)-HODE	25.8 ± 10.6	1,312,200 ± 534,323
13(S)-HODE	31.8 ± 3.7	1,803,400 ± 279,731

*Abbreviations:* HPETE, hydroperoxyicosatetraenoic acid; HPODE, hydroperoxyoctadecadienoic acid, HETE, hydroxyicosatetraenoic acid; HODE, hydroxyoctadecadienoic acid. Adapted from Table 2 in reference 40.

of the humans (22) was ~35 μM. How could such small concentrations of an apoA-I mimetic peptide change the antiinflammatory properties of HDL when the concentration of apoA-I in the same plasma was many orders of magnitude greater? This question was addressed by Van Lenten et al. (40). Physiologic concentrations (~35 μM) of human apoA-I did not inhibit LDL-induced MCP-1 production in human aortic endothelial cell cultures, but adding nanomolar concentrations of 4F significantly reduced this inflammatory response. Indeed, as little as 4.3 nM of 4F was significantly antiinflammatory in this assay and 0.43 μM was as effective as normal antiinflammatory HDL (40). In measuring binding affinity,  $KD = Kd/Ka$  where  $Kd$  is the dissociation rate constant and  $Ka$  is the association rate constant. The larger the  $KD$  is, the weaker the binding. Van Lenten et al. (40) determined by surface plasmon resonance that there was no difference in the binding affinity of any lipid tested for binding to D-4F or L-4F (40). As shown in Table 1 [adapted from Table 1 in Van Lenten et al. (40)] there was no significant difference in the binding affinity of the non-oxidized phospholipid PAPC for L-4F compared with full length human apoA-I. However, when PAPC was oxidized, the binding affinity of the resulting oxidized phospholipids for L-4F was many orders of magnitude greater than for human apoA-I (Table 1). Indeed, the binding affinity of PEIPC for L-4F was 5 million-fold greater than for full length human apoA-I.

As shown in Table 2 [adapted from Table 2 in Van Lenten et al. (40)] L-4F and human apoA-I bound nonoxidized fatty acids with equal high affinity. However, when arachidonic acid was oxidized to produce various fatty acid hydroperoxides (HPETE) or alcohols (HETE) or when linoleic acid was oxidized to produce various fatty acid hydroperoxides (HPODE) or alcohols (HODE), only L-4F continued to bind the oxidized lipids with high affinity. The remarkable difference in the binding affinity of oxidized lipids for 4F compared with human apoA-I provides an explanation as to how such small concentrations of an apoA-I mimetic peptide could change the antiinflammatory properties of HDL when the concentration of apoA-I

in the same plasma was many orders of magnitude greater. Because the 4F peptide renders dysfunctional HDL functional, these data also are consistent with previous reports (36, 37) that oxidized lipids can render HDL dysfunctional.

## REFERENCES

- Hessler, J. R., A. L. Robertson, and G. M. Chisolm. 1979. LDL-induced cytotoxicity and its inhibition by HDL in human vascular smooth muscle and endothelial cells in culture. *Atherosclerosis*. **32**: 213–229.
- van Hinsbergh, V. W., M. Scheffer, L. Havekes, and H. J. Kempen. 1986. Role of endothelial cells and their products in the modification of low-density lipoproteins. *Biochim. Biophys. Acta*. **878**: 49–64.
- Ohta, T., K. Takata, S. Horiuchi, Y. Morino, and I. Matsuda. 1989. Protective effect of lipoproteins containing A-I on Cu<sup>2+</sup> catalyzed oxidation of human low density lipoprotein. *FEBS Lett*. **257**: 435–438.
- Parthasarathy, S., J. Barnett, and L. G. Fong. 1990. High density lipoprotein inhibits the oxidative modification of low-density lipoprotein. *Biochim. Biophys. Acta*. **1044**: 275–283.
- Mackness, M. I., S. Arrol, and P. N. Durrington. 1991. Paraonase prevents accumulation of lipoperoxides in low-density lipoprotein. *FEBS Lett*. **286**: 152–154.
- Navab, M., S. S. Imes, S. Y. Hama, G. P. Hough, L. A. Ross, R. W. Bork, A. J. Valente, J. A. Berliner, D. C. Drinkwater, H. Laks, et al. 1991. Monocyte transmigration induced by modification of low density lipoprotein in cocultures of human aortic wall cells is due to induction of monocyte chemotactic protein 1 synthesis and is abolished by high density lipoprotein. *J. Clin. Invest.* **88**: 2039–2046.
- Watson, A. D., M. Navab, S. Y. Hama, A. Sevanian, S. M. Prescott, D. M. Stafforini, T. M. McIntyre, B. N. La Du, A. M. Fogelman, and J. A. Berliner. 1995. Effect of platelet activating factor acetylhydrolase on the formation of minimally oxidized-low density lipoprotein. *J. Clin. Invest.* **95**: 774–782.
- Watson, A. D., J. A. Berliner, S. Y. Hama, B. N. La Du, K. F. Faull, A. M. Fogelman, and M. Navab. 1995. Protective effect of HDL associated paraonase-inhibition of the biological activity of minimally oxidized low density lipoprotein. *J. Clin. Invest.* **96**: 2882–2891.
- Van Lenten, B. J., S. Y. Hama, F. C. de Beer, D. M. Stafforini, T. M. McIntyre, S. M. Prescott, B. N. La Du, A. M. Fogelman, and M. Navab. 1995. Anti-inflammatory HDL becomes pro-inflammatory during the acute phase response. *J. Clin. Invest.* **96**: 2758–2767.
- Navab, M., S. Y. Hama, C. J. Cooke, G. M. Anantharamaiah, M. Chadha, L. Jin, G. Subbanagounder, K. F. Faull, S. T. Reddy, N. E. Miller, et al. 2000. Normal high density lipoprotein inhibits three steps in the formation of mildly oxidized low density lipoprotein: step 1. *J. Lipid Res.* **41**: 1481–1494.
- Navab, M., S. Y. Hama, G. M. Anantharamaiah, K. Hassan, G. P. Hough, A. D. Watson, S. T. Reddy, A. Sevanian, G. C. Fonarow, and A. M. Fogelman. 2000. Normal high density lipoprotein inhibits three steps in the formation of mildly oxidized low density lipoprotein: steps 2 and 3. *J. Lipid Res.* **41**: 1495–1508.
- Garber, D. W., G. Datta, M. Chaddha, M. N. Palgunachari, S. Y. Hama, M. Navab, A. M. Fogelman, J. P. Segrest, and G. M. Anantharamaiah. 2001. A new synthetic class A amphipathic peptide analogue protects mice from diet-induced atherosclerosis. *J. Lipid Res.* **42**: 545–552.
- Datta, G., M. Chaddha, S. Hama, M. Navab, A. M. Fogelman, D. W. Garber, V. K. Mishra, R. M. Eband, R. F. Eband, S. Lund-Katz, et al. 2001. Effects of increasing hydrophobicity on the physical-chemical and biological properties of a class A amphipathic helical peptide. *J. Lipid Res.* **42**: 1096–1104.
- Navab, M., G. M. Anantharamaiah, S. Hama, D. W. Garber, M. Chaddha, G. Hough, R. Lallone, and A. M. Fogelman. 2002. Oral administration of an apo A-I mimetic peptide synthesized from D-amino acids dramatically reduces atherosclerosis in mice independent of plasma cholesterol. *Circulation*. **105**: 290–292.
- Datta, G., R. F. Eband, R. M. Eband, M. Chaddha, M. A. Kirksey, D. W. Garber, S. Lund-Katz, M. C. Phillips, S. Hama, M. Navab, et al. 2004. Aromatic residue position on the nonpolar face of class A amphipathic helical peptides determines biological activity. *J. Biol. Chem.* **279**: 26509–26517.
- Handattu, S. P., D. W. Garber, D. C. Horn, D. W. Hughes, B. Berno, A. D. Bain, V. K. Mishra, M. N. Palgunachari, G. Datta, G. M.

- Anantharamaiah, and R. M. Eband. 2007. ApoA-I mimetic peptides with differing ability to inhibit atherosclerosis also exhibit differences in their interactions with membrane bilayers. *J. Biol. Chem.* **282**: 1980–1988.
17. Navab, M., S. Y. Hama, G. P. Hough, G. Subbanagounder, S. T. Reddy, and A. M. Fogelman. 2001. A cell-free assay for detecting HDL that is dysfunctional in preventing the formation of or inactivating oxidized phospholipids. *J. Lipid Res.* **42**: 1308–1317.
  18. Ansell, B. J., M. Navab, S. Hama, N. Kamranpour, G. Fonarow, G. Hough, S. Rahmani, R. Mottahedeh, R. Dave, S. T. Reddy, and A. M. Fogelman. 2003. Inflammatory/anti-inflammatory properties of high-density lipoprotein distinguish patients from control subjects better than high-density lipoprotein cholesterol levels and are favorably affected by simvastatin treatment. *Circulation.* **108**: 2751–2756.
  19. Navab, M., G. M. Anantharamaiah, S. T. Reddy, B. J. Van Lenten, B. J. Ansell, S. Hama, G. Hough, E. Bachini, V. R. Grijalva, A. C. Wagner, et al. 2005. The double jeopardy of HDL. *Ann. Med.* **37**: 173–178.
  20. Cruz, D., A. D. Watson, C. S. Miller, D. Montoya, M-T. Ochoa, P. A. Sieling, M. A. Gutierrez, M. Navab, S. T. Reddy, J. L. Witztum, et al. 2008. Host-derived oxidized phospholipids and HDL regulate innate immunity in human leprosy. *J. Clin. Invest.* **118**: 2917–2928.
  21. Van Lenten, B. J., A. C. Wagner, M. Navab, G. M. Anantharamaiah, S. Hama, S. T. Reddy, and A. M. Fogelman. 2007. Lipoprotein inflammatory properties and serum amyloid A levels but not cholesterol levels predict lesion area in cholesterol-fed rabbits. *J. Lipid Res.* **48**: 2344–2353.
  22. Bloedon, L. T., R. Dunbar, D. Duff, P. Pinell-Salles, R. Norris, B. J. DeGroot, R. Movva, M. Navab, A. M. Fogelman, and D. J. Rader. 2008. Safety, pharmacokinetics, and pharmacodynamics of oral apoA-I mimetic peptide D-4F in high-risk cardiovascular patients. *J. Lipid Res.* **49**: 1344–1352.
  23. McMahon, M., J. Grossman, J. FitzGerald, E. Dahlin-Lee, D. J. Wallace, B. Y. Thong, H. Badsha, K. Kalunian, C. Charles, M. Navab, et al. 2006. Proinflammatory high-density lipoprotein as a biomarker for atherosclerosis in patients with systemic lupus erythematosus and rheumatoid arthritis. *Arthritis Rheum.* **54**: 2541–2549.
  24. Charles-Schoeman, C., D. Khanna, D. E. Furst, M. McMahon, S. T. Reddy, A. M. Fogelman, H. E. Paulus, G. S. Park, T. Gong, and B. J. Ansell. 2007. Effects of high-dose atorvastatin on anti-inflammatory properties of high density lipoprotein in patients with rheumatoid arthritis: a pilot study. *J. Rheumatol.* **34**: 1459–1464.
  25. Roberts, C. K., C. Ng, S. Hama, A. J. Eliseo, and R. J. Barnard. 2006. Effect of a short-term diet and exercise intervention on inflammatory/anti-inflammatory properties of HDL in overweight/obese men with cardiovascular risk factors. *J. Appl. Physiol.* **101**: 1727–1732.
  26. Opole, I. O., J. M. Belmont, A. Kumar, and P. M. Moriarty. 2007. Effect of low-density lipoprotein apheresis on inflammatory and noninflammatory high-density lipoprotein cholesterol. *Am. J. Cardiol.* **100**: 1416–1418.
  27. Dodani, S., R. Kaur, S. Reddy, G. L. Reed, M. Navab, and V. George. 2008. Can dysfunctional HDL explain high coronary artery disease risk in South Asians? *Int. J. Cardiol.* **129**: 125–132.
  28. Kalantar-Zadeh, K., J. D. Kopple, N. Kamranpour, A. M. Fogelman, and M. Navab. 2007. HDL-inflammatory index correlates with poor outcome in hemodialysis patients. *Kidney Int.* **72**: 1149–1156.
  29. van Leuven, S. I., R. Hezemans, J. H. Levels, S. Snoek, P. C. Stokkers, G. K. Hovingh, J. J. Kastelein, E. S. Stroes, E. de Groot, and D. W. Hommes. 2007. Enhanced atherogenesis and altered high density lipoprotein in patients with Crohn's disease. *J. Lipid Res.* **48**: 2640–2646.
  30. Wehrauch, D., H. Xu, Y. Shi, J. Wang, J. Brien, D. W. Jones, S. Kaul, R. A. Komorowski, M. E. Csuka, K. T. Oldham, et al. 2007. Effects of D-4F on vasodilation, oxidative stress, angiostatin, myocardial inflammation, and angiogenic potential in tight-skin mice. *Am. J. Physiol. Heart Circ. Physiol.* **293**: H1432–H1441.
  31. Bhattacharyya, T., S. J. Nicholls, E. J. Topol, R. Zhang, X. Yang, D. Schmitt, X. Fu, M. Shao, D. M. Brennan, S. G. Ellis, et al. 2008. Relationship of paraoxonase 1 (PON1) gene polymorphisms and functional activity with systemic oxidative stress and cardiovascular risk. *JAMA.* **299**: 1265–1276 (PubMed).
  32. Wu, Z., M. A. Wagner, L. Zheng, J. S. Parks, J. M. Shy 3rd, J. D. Smith, V. Gogonea, and S. L. Hazen. 2007. The refined structure of nascent HDL reveals a key functional domain for particle maturation and dysfunction. *Nat. Struct. Mol. Biol.* **14**: 861–868.
  33. Nicholls, S. J., L. Zheng, and S. L. Hazen. 2005. Formation of dysfunctional high-density lipoprotein by myeloperoxidase. *Trends Cardiovasc. Med.* **15**: 212–219.
  34. Vaisar, T., S. Pennathur, P. S. Green, S. A. Gharib, A. N. Hoofnagle, M. C. Cheung, J. Byun, S. Vuletic, S. Kassim, P. Singh, et al. 2007. Shotgun proteomics implicates protease inhibition and complement activation in the antiinflammatory properties of HDL. *J. Clin. Invest.* **117**: 746–756.
  35. Green, P. S., T. Vaisar, S. Pennathur, J. J. Kulstad, A. B. Moore, S. Marcovina, J. Brunzell, R. H. Knopp, X. Q. Zhao, and J. W. Heinecke. 2008. Combined statin and niacin therapy remodels the high-density lipoprotein proteome. *Circulation.* **118**: 1259–1267.
  36. Navab, M., S. Hama-Levy, B. J. Van Lenten, G. C. Fonarow, C. J. Cardinez, L. W. Castellani, M-L. Brennan, A. J. Lusis, and A. M. Fogelman. 1997. Mildly oxidized LDL induces an increased apolipoprotein J/paraoxonase ratio. *J. Clin. Invest.* **99**: 2005–2019.
  37. Forte, T. M., G. Subbanagounder, J. A. Berliner, P. J. Blanche, A. O. Clermont, Z. Jia, M. N. Oda, R. M. Krauss, and J. K. Bielicki. 2002. Altered activities of anti-atherogenic enzymes LCAT, paraoxonase, and platelet-activating factor acetylhydrolase in atherosclerosis-susceptible mice. *J. Lipid Res.* **43**: 477–485.
  38. Persegol, L., B. Verges, P. Gambert, and L. Duveillard. 2007. Inability of HDL from abdominally obese subjects to counteract the inhibitory effect of oxidized LDL on vasorelaxation. *J. Lipid Res.* **48**: 1396–1401.
  39. Navab, M., G. M. Anantharamaiah, S. T. Reddy, S. Hama, G. Hough, V. R. Grijalva, A. C. Wagner, J. S. Frank, G. Datta, D. Garber, and A. M. Fogelman. 2004. Oral D-4F causes formation of pre- $\beta$  high-density lipoprotein and improves high-density lipoprotein-mediated cholesterol efflux and reverse cholesterol transport from macrophages in apolipoprotein E-null mice. *Circulation.* **109**: 3215–3220.
  40. Van Lenten, B. J., A. C. Wagner, C. L. Jung, P. Ruchala, A. J. Waring, R. I. Lehrer, A. D. Watson, S. Hama, M. Navab, G. M. Anantharamaiah, et al. 2008. Anti-inflammatory apoA-I mimetic peptides bind oxidized lipids with much higher affinity than human apoA-I. *J. Lipid Res.* **49**: 2302–2311.