

# **Dysfunctional HDL**

**Hussein Osamah, MD**

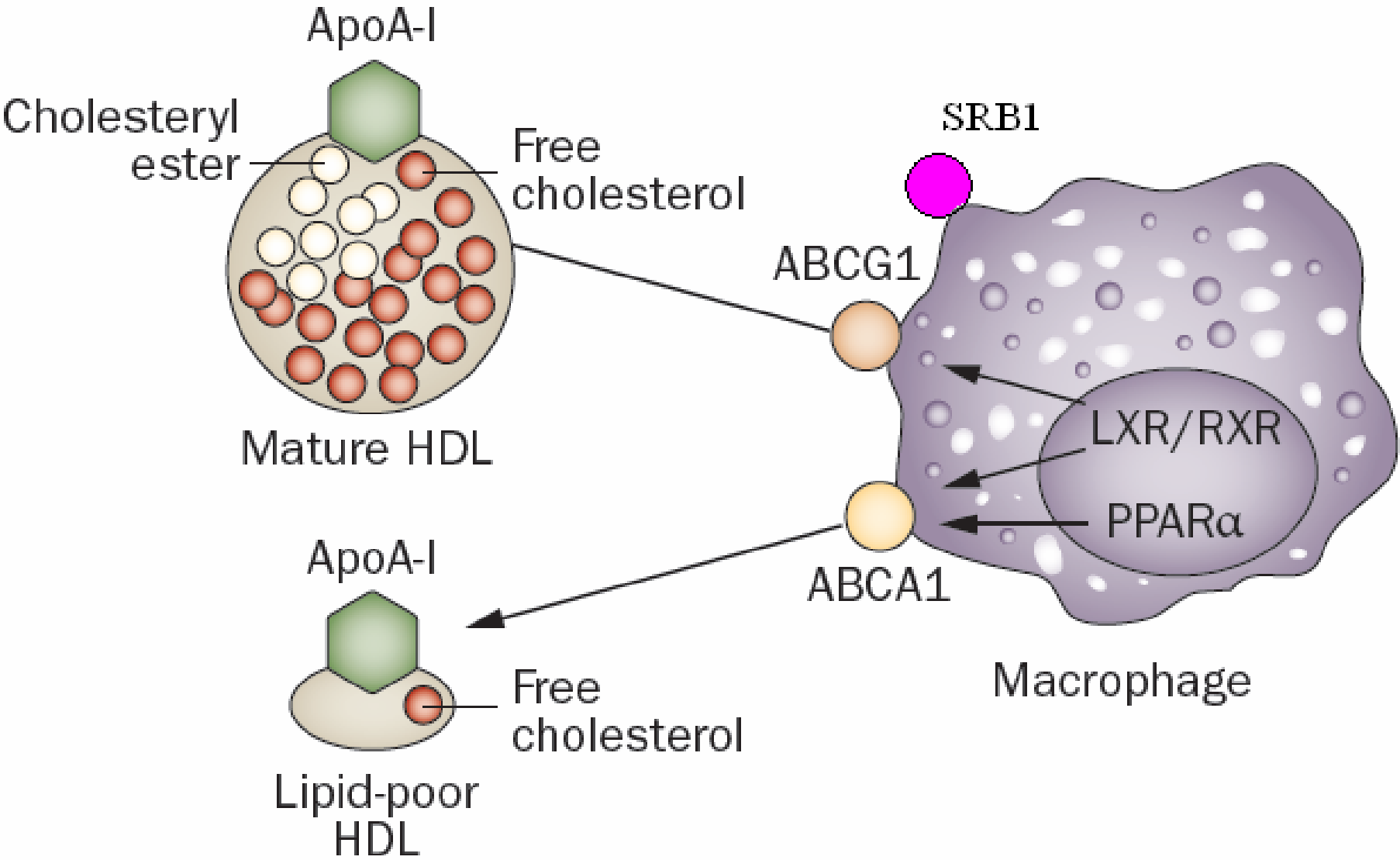
**Internal Medicine A**

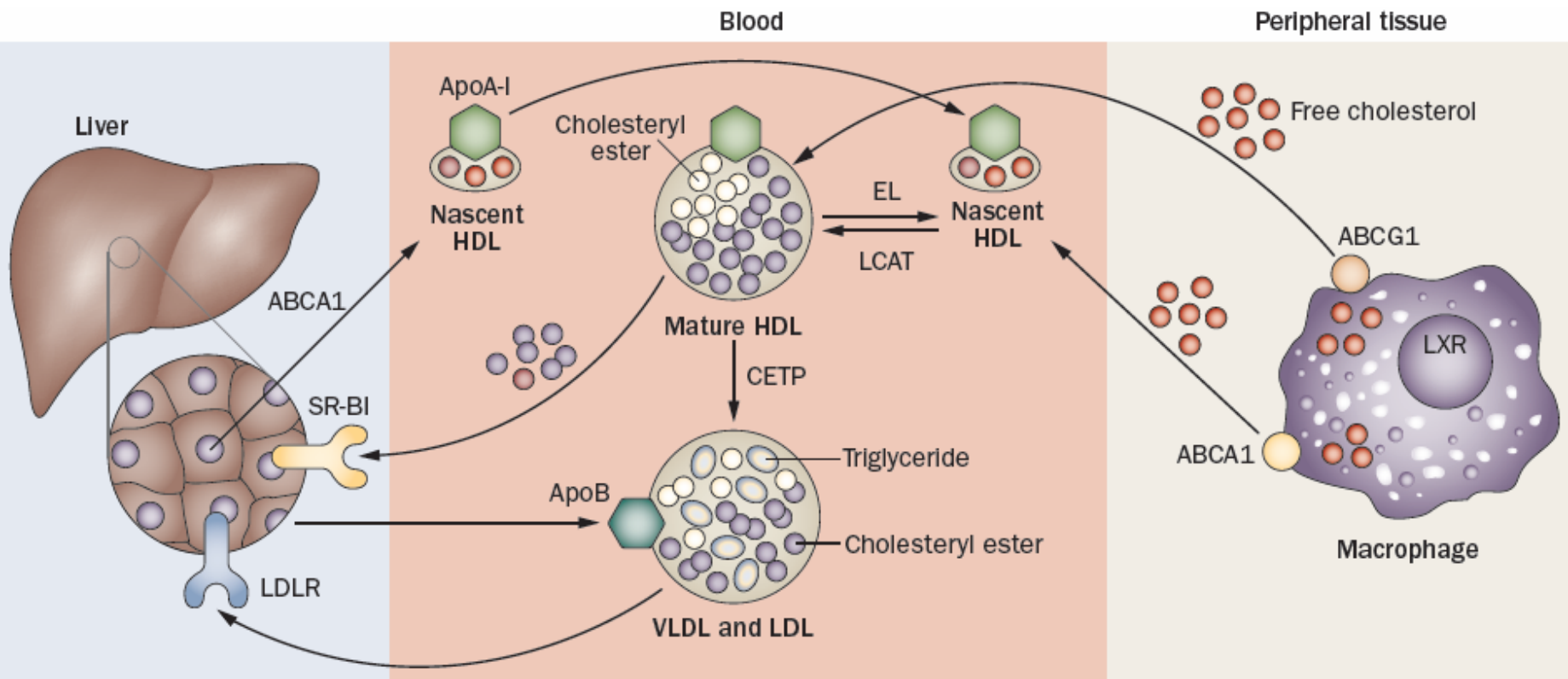
**Ziv Medical Center**

**Faculty of Medicine, Technion**

**Reverse cholesterol transport** includes the removal of cholesterol from cholesterol laden peripheral tissues including lipidated macrophages which are deposited at the site of atherosclerotic lesions, and their delivery to the liver or other cholesterol metabolizing tissues for catabolism.

Bruce C. Annu Rev Nutr 1998;18:297–330.





Duffy D. Nat Rev Cardiol 2009

# **HDL correlates inversely with the incidence of coronary heart disease and atherosclerosis.**

Miller NE. Lancet 1977;1:965–8.

Gordon DJ. Circulation 1989;79:8–15.

Shah PK. Circulation 1992;85:1279–85.

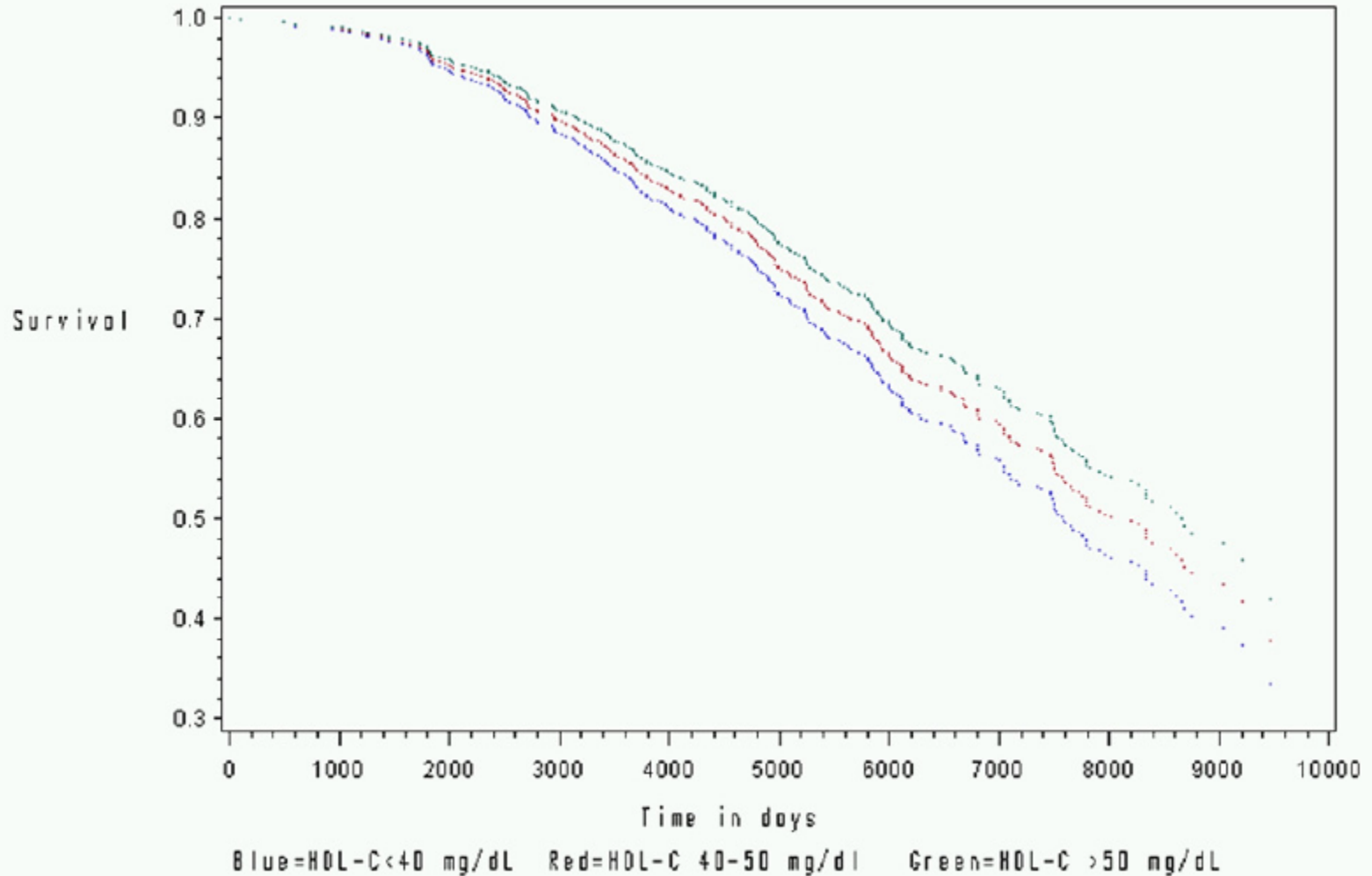
Wilson PW. Framingham Heart Study. Arteriosclerosis 1988;8:737–41.

Forte TM. J Clin Invest 1984;74:1601–13.

# **Studies in humans showed that an increase in plasma HDL levels correlated with slower progression of atherosclerotic lesions and possible stabilization of unstable atherosclerotic plaques.**

Gordon DJ. N Engl J Med 1989;321:1311–6.

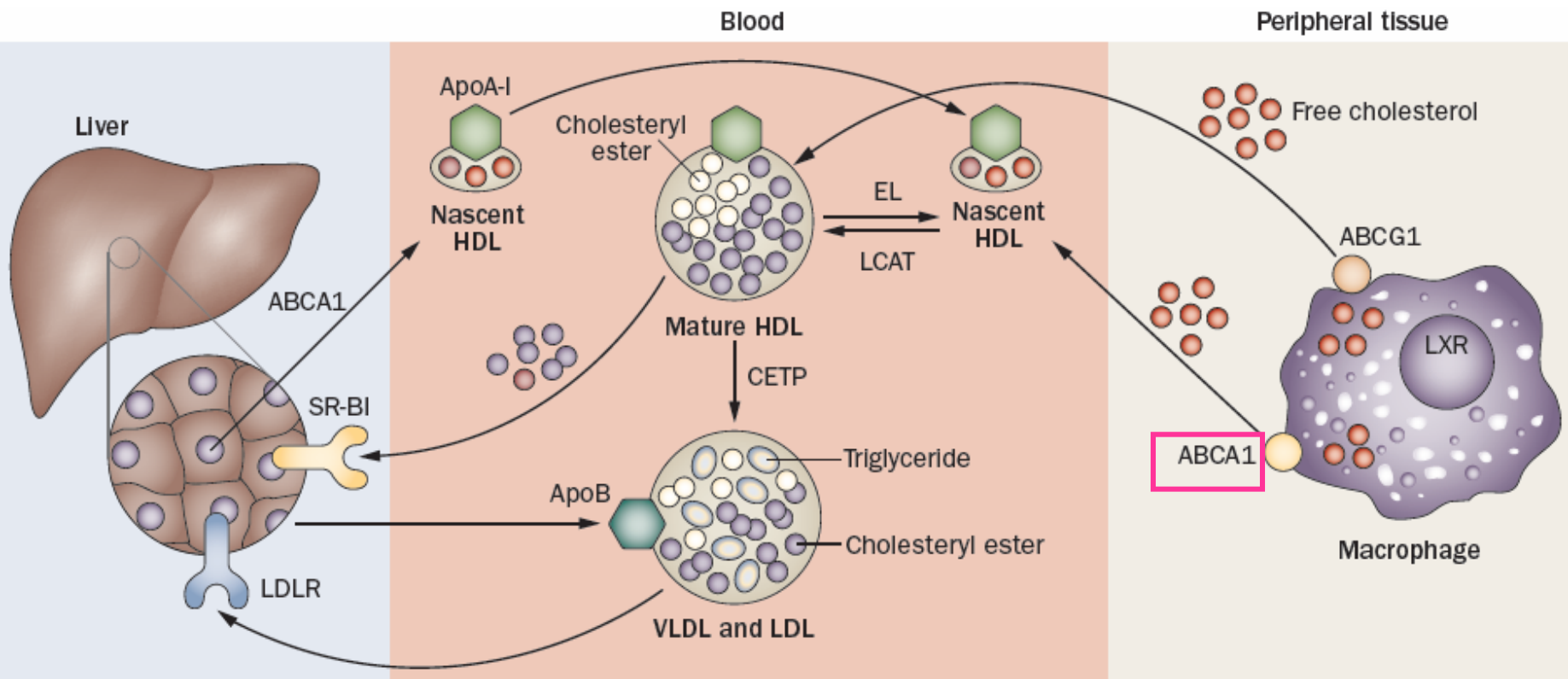
## Time to death, by category of baseline HDL-C



Time to death in 652 men enrolled in the VA NAS by **40 mg/dl (blue line)**, **40 to 50 mg/dl (red line)**, and **≥50 mg/dl (green line)** of initial high-density lipoprotein cholesterol (HDL-C).

Rahilly-Thierney 2011 Am J cardiol on-line.

# **Relation between HDL-C level and atheroprotection**



Duffy D. Nat Rev Cardiol 2009

## **Epidemiological study:**

- 1. Lower plasma levels of HDL cholesterol due to heterozygosity for **loss-of-function mutations in ABCA1**, were not associated with an increased risk of IHD.**

Frikke-Schmidt R. JAMA 2008;299:2524–32.

Roma P. Clin Invest 1993;91:1445–52.

# Epidemiological study:

## 2. **Apolipoprotein A-I Milano** (Arg173→Cys mutation).

**Human carriers of this mutation have very low HDL levels (less than half of normal), but enhanced protection against atherosclerosis.**

**HDL of these subjects has superior atheroprotective properties compared to the HDL of unaffected individuals.**

Frikke-Schmidt R. JAMA 2008;299:2524–32.

Roma P. Clin Invest 1993;91:1445–52.

**3. Low-fat high-carbohydrate intake** in humans that frequently leads to reduced HDL levels, though the atheroprotective potential of this HDL is improved.

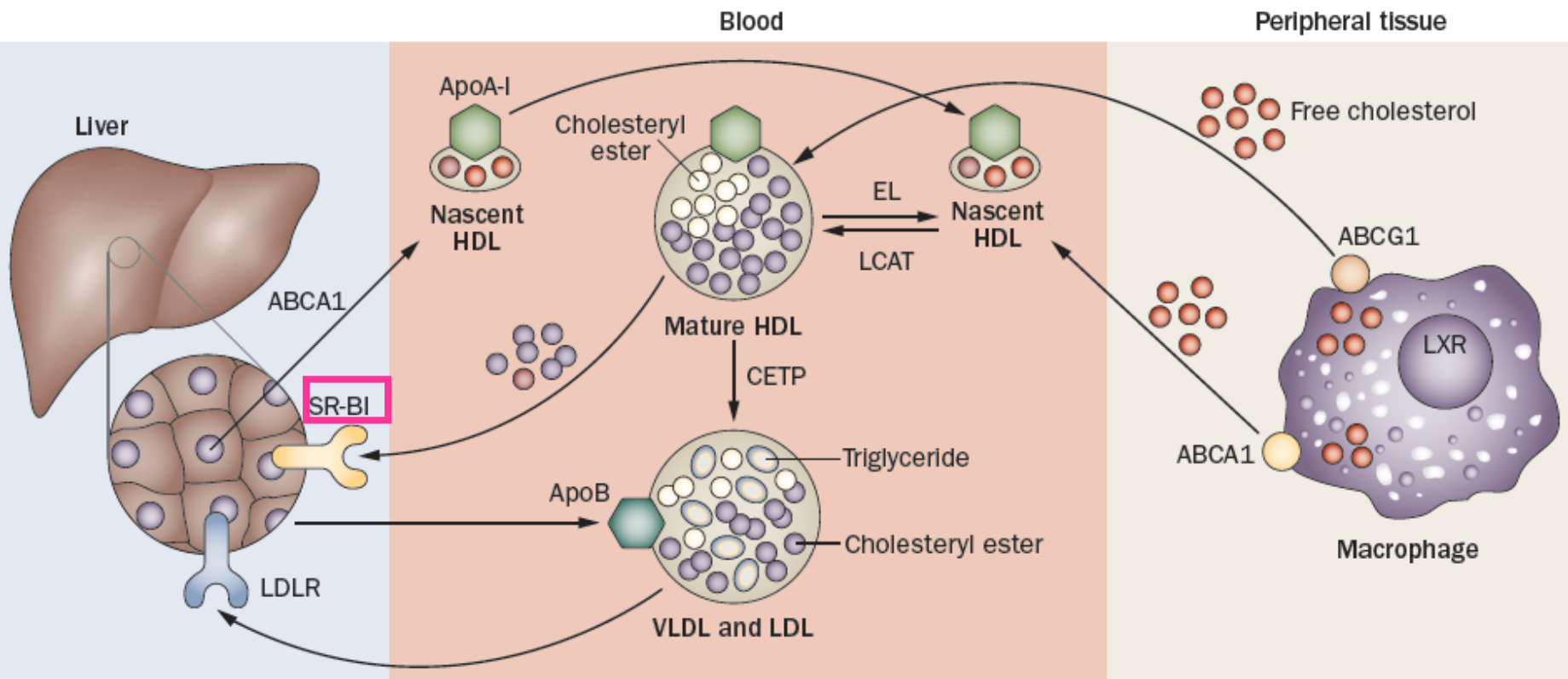
Roberts CK. J Appl Physiol 2006;101:1727–32.

**4. CETP inhibitor torcetrapib** which showed that following treatment with torcetrapib, a substantial elevation of HDL is accompanied by a higher risk for CHD.

Barter PJ. N Engl J Med 2007;357:2109–22.

**In a post hoc analysis of the torcetrapib study by intravascular ultrasound, atheroma regression, rather than progression, was observed in the subjects who achieved the highest levels of HDL-C, suggesting that CETP inhibition does not lead to dysfunctional HDL.**

Nicholls SJ. ILLUSTRATE *Circulation*. 2008;118:2506–2514.



Duffy D. Nat Rev Cardiol 2009

**Increased plasma HDL concentration that results from reduced catabolism due to blockage in the dynamic flow of HDL-lipids from peripheral tissues to the liver may not contribute to atheroprotection.**

**In  $SR-BI^{-/-} \times apoE^{-/-}$  mice:**

**Elevated plasma HDL cholesterol and increased incidents of atherosclerosis compared to  $apoE^{-/-}$  mice.**

Trigatti B. Proc Natl Acad Sci USA 1999;96:9322–7.

# **Overexpression of SR-BI protected mice from atherosclerosis despite a significant reduction in their plasma HDL levels.**

Kozarsky KF. *Arterioscler Thromb Vasc Biol* 2000;20:721–7.

Webb NR. *J Lipid Res* 2002;43:1890–8.

**Humans with hepatic lipase deficiency were found to have increased risk for CHD despite their elevated HDL cholesterol levels.**

Connelly PW. Crit Rev Clin Lab Sci 1998;35:547–72.

# **HDL mediated atheroprotection**

# HDL antioxidant properties

## Antioxidant enzymes on HDL:

1. **Paraoxonase**

2. **Acetyl-hydrolase platelet activation factor**

**prevent the biosynthesis of oxidized LDL in *in vitro* studies.**

Aviram M. Several studies.

NavabM. J Lipid Res 2008;50:S145–9.

## 2. Endothelial function protection

**HDL stimulates endothelial nitric oxide synthase.**

Nofer JR. J Clin Invest 2004;113:569–81.

**17-estradiol associates with HDL in the form of fatty acyl esters that are generated by the plasma enzyme lecithin:cholesterol acyl transferase.**

**HDL associated estradiol increases the atheroprotective potential of HDL by stimulating the enzymatic activity of eNOS in an SR-B1-dependent fashion while HDL from men had minimal activity.**

***In vitro* studies in Fu5AH cell cultures suggested that in addition to SR-B1, the LDLr may also play important role in the uptake of estradiol containing HDL.**

Kanji SS. Endocrinol Metab 1999;84:2481–8.

Gong M. J Clin Invest 2003;111:1579–87.

Badeau RM. Biochim Biophys Acta 2007;1771:1329–34.

# Endothelial function protection

**HDL stimulates SR-B1-dependent endothelial cell migration and the recruitment of endothelial progenitor cells into the intimal layer of the artery at the site of endothelial injury.**

Tso C. Arterioscler Thromb Vasc Biol 2006;26:1144–9.

# 3. HDL anti-inflammatory properties

## 1. Inhibits the expression of pro-inflammatory adhesion molecules

Barter PJ. *Circ Res* 2004;95:764–72.

## 2. HDL blocks the pro-inflammatory activity of C-reactive protein.

Wadham C. *Circulation* 2004;109:2116–22.

## 3. Inhibits production of pro-inflammatory prostaglandins by monocytes at the site of atherosclerotic lesion.

Jambou D. *Biochim Biophys Acta* 1993;1168:115–21.

# **HDL anti-inflammatory properties**

**5. Prevents and/or neutralizes pro-inflammatory effects of oxidized LDL phospholipids on the endothelium.**

Navab M. *Arterioscler Thromb Vasc Biol* 2001;21:481–8.

**6. HDL reduces the LDL-induced MCP-1 production.**

**7. Studies in mice with advanced lesions showed that overexpression of human apoA-I or Acetyl-hydrolase platelet activation factor reduces macrophage adhesion to the vessel wall.**

Theilmeyer G. *FASEB J* 2000;14:2032–9.

## **4. Maintenance of VLDL triglyceride Homeostasis**

**Formation of apoCIII-containing HDL prevented apoCIII-induced hypertriglyceridemia.**

**HDL acts as a buffer or sink which sequesters away apoCIII thus preventing their excess accumulation on VLDL and the generation of atherogenic remnant particles with abnormal apoprotein composition.**

Kypreos KE.Biochemistry 2008;47:10491–502.

## 5. Insulin sensitivity

**HDL stimulates glucose uptake and fatty acid oxidation thus opposing insulin resistance.**

Han R. Diabetologia 2007;50:1960–8.

**Cholesterol efflux from macrophages represents only a small fraction of overall flux through the RCT pathway.**

**It is probably the component that is most relevant to atheroprotection.**

Cuchel M. Circulation 2006;113:2548-55.

## **Cholesterol efflux:**

- 1. Protect macrophages from LDL-induced apoptosis.**
- 2. Enhance endothelial function.**

Terasaka N. HDL protects macrophages from oxidized LDL-induced apoptosis by promoting efflux of 7-ketocholesterol via ABCG1. Proc Natl Acad Sci U S A 2007.

Cui D. Pivotal advance: macrophages become resistant to cholesterol-induced death after phagocytosis of apoptotic cells. J Leukoc Biol 2007.

Terasaka N. ABCG1 and HDL protect against endothelial dysfunction in mice fed a high cholesterol diet. J Clin Invest 2008.

## **HDL<sub>2</sub> plasma levels have shown the greatest hazard reduction for incident CHD.**

Williams PT. Prospective study of CHD vs HDL<sub>2</sub>, HDL<sub>3</sub> and other lipoproteins in GofGofman's Livermore Cohort. Atheroscl 2011.

## **But not in all studies.**

Superko HR. Advanced lipoprotein testing and subfractionation are clinically useful. Circulation 2009;119:2383.

## **Macrophage cholesterol efflux positively correlated with concentration of HDL<sub>2</sub> but not with total plasma HDL concentrations.**

Linsel-Nitschkhe P. Lipids health Dis 2009.

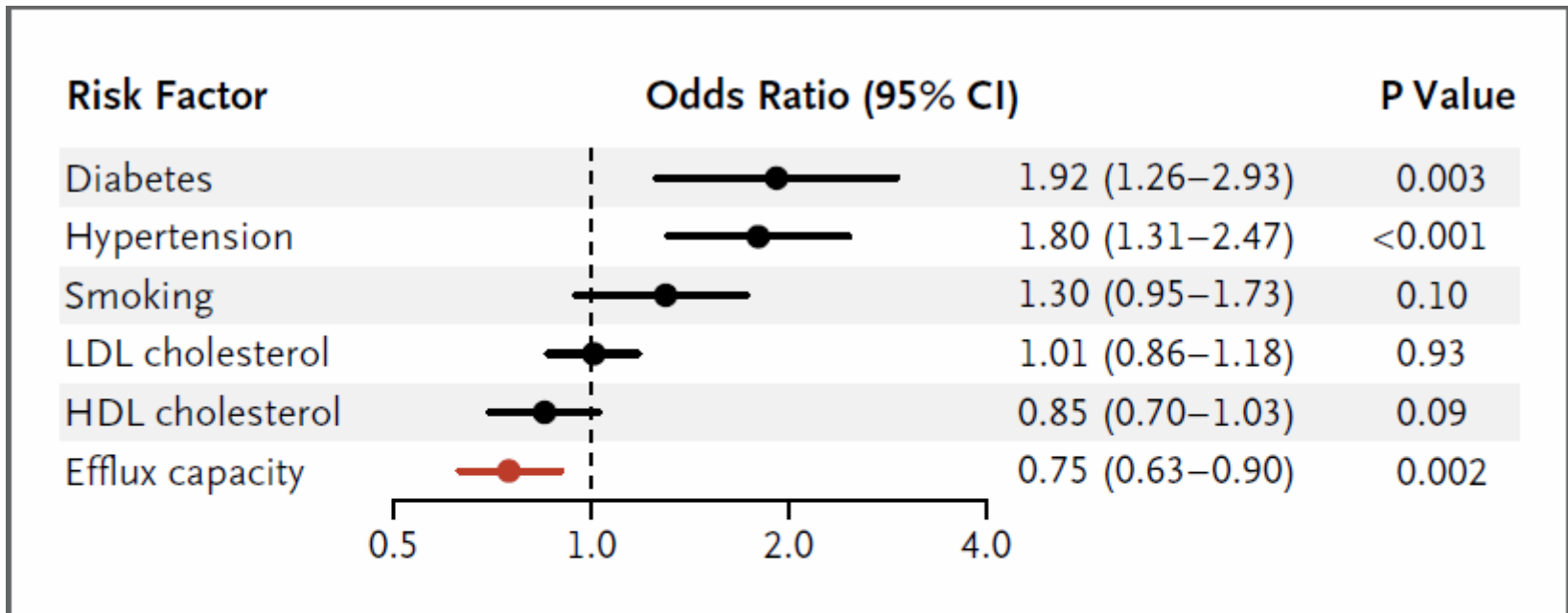
# Cholesterol Efflux

**Table 3. Coronary Artery Disease Status According to Quartile of Efflux Capacity.**

Variable	No. of Patients	Odds Ratio for Coronary Artery Disease (95% CI)*		
		Adjusted for Cardiovascular Risk Factors	Adjusted for Cardiovascular Risk Factors and HDL Cholesterol	Adjusted for Cardiovascular Risk Factors and Apolipoprotein A-I
Quartile 1	198	1.00	1.00	1.00
Quartile 2	198	0.75 (0.48–1.16)	0.79 (0.51–1.24)	0.77 (0.49–1.21)
Quartile 3	198	0.58 (0.37–0.89)	0.64 (0.41–1.00)	0.63 (0.40–0.99)
Quartile 4	199	0.40 (0.25–0.63)	0.48 (0.30–0.78)	0.46 (0.28–0.75)
P value for trend		<0.001	0.002	0.002

\* Cardiovascular risk factors included in the logistic-regression model were age, sex, smoking status, presence or absence of diabetes, presence or absence of hypertension, and low-density lipoprotein cholesterol. HDL denotes high density lipoprotein.

Cholesterol Efflux Capacity, High-Density Lipoprotein Function, and Atherosclerosis Amit V. Khera NEJM 364;127, 2011



**Odds Ratios for Coronary Artery Disease According to Efflux Capacity and Selected Risk Factors.**

The logistic-regression model was also adjusted for age and sex. Odds ratios for continuous variables are per 1-SD increase.

Cholesterol Efflux Capacity, High-Density Lipoprotein Function, and Atherosclerosis Amit V. Khera NEJM 364;127, 2011

## Effect of Pharmacologic Interventions on Cholesterol Efflux Capacity.\*

Pharmacologic Intervention	No. of Patients	Percent Change in Cholesterol Efflux Capacity (95% CI)	P Value	
			vs. Baseline	vs. Placebo
Thiazolidinedione				
Pioglitazone	16	11.3 (1.8 to 20.8)	0.02	0.04
Placebo	23	0.0 (-6.2 to 6.1)	0.99	
Statin				
Pravastatin, 40 mg	23	-0.4 (-6.5 to 5.6)	0.88	0.71
Atorvastatin, 10 mg	26	2.7 (-4.8 to 10.2)	0.47	0.81
Atorvastatin, 80 mg	25	-2.5 (-9.1 to 4.1)	0.45	0.38
Placebo	25	-1.1 (-6.5 to 4.2)	0.66	

Patients treated with pioglitazone received 30 mg per day for 6 weeks, followed by 45 mg per day for an additional 6 weeks. Patients treated with statins received continuous therapy at a fixed dose for 16 weeks.

Cholesterol Efflux Capacity, High-Density Lipoprotein Function, and Atherosclerosis Amit V. Khera NEJM 364;127, 2011

**The levels of HDL cholesterol and apolipoprotein A-I were significant determinants of cholesterol efflux capacity but accounted for less than 40% of the observed variation.**

**An inverse relationship was noted between efflux capacity and CIMT both before and after adjustment for the HDL cholesterol level.**

Cholesterol Efflux Capacity, High-Density Lipoprotein Function, and Atherosclerosis Amit V. Khera NEJM 364;127, 2011

**Efflux capacity was a strong inverse predictor of coronary disease status (adjusted odds ratio for coronary disease per 1-SD increase in efflux capacity, 0.70; 95% confidence interval [CI], 0.59 to 0.83;  $P < 0.001$ ).**

**This relationship was attenuated, but remained significant, after additional adjustment for the HDL cholesterol level (odds ratio per 1-SD increase, 0.75; 95% CI, 0.63 to 0.90;  $P = 0.002$ ) or apolipoprotein A-I level (odds ratio per 1-SD increase, 0.74; 95% CI, 0.61 to 0.89;  $P = 0.002$ ).**

Cholesterol Efflux Capacity, High-Density Lipoprotein Function, and Atherosclerosis Amit V. Khera NEJM 364;127, 2011

**Efflux capacity served as the stronger predictor of both CIMT and CHD status in regression models that included both variables.**

Cholesterol Efflux Capacity, High-Density Lipoprotein Function, and Atherosclerosis Amit V. Khera NEJM 364;127, 2011

# **Increased efflux capacity after therapy with pioglitazone:**

## **1. Enhanced transcription of apolipoprotein A-I?**

van Dam MJ. Association between increased arterial wall thickness and impairment in ABCA1-driven cholesterol efflux: an observational study. *Lancet* 2002.

## **2. Glycation of HDL significantly reduced the HDL-mediated cholesterol efflux from macrophages.**

Matsuki K. Metformin restores impaired HDL-mediated cholesterol efflux due to glycation. *Atherosclerosis* 2009;206:434.

## **3. Poor glycated control in T1DM patients was associated with accelerated oxidation damage to apolipoprotein A-I.**

Jaleel A. *Diabetes* 2010;59:2366.

## **4. HDL from diabetic subjects has evidence of glycated apoAI and apoAII, and this glycated apoAI has altered structure and lipid binding activity.**

Calvo C. *Biochem Biophys Res Commun.* 1988;153:1060 –1067.

Calvo C. *Diabete Metab.*1988;14:264 –269.

**HDL incubated with high glucose results in a reduction in paraoxonase activity and decreased antiinflammatory activity.**

Hedrick CC. *Diabetologia*. 2000;43:312–320.

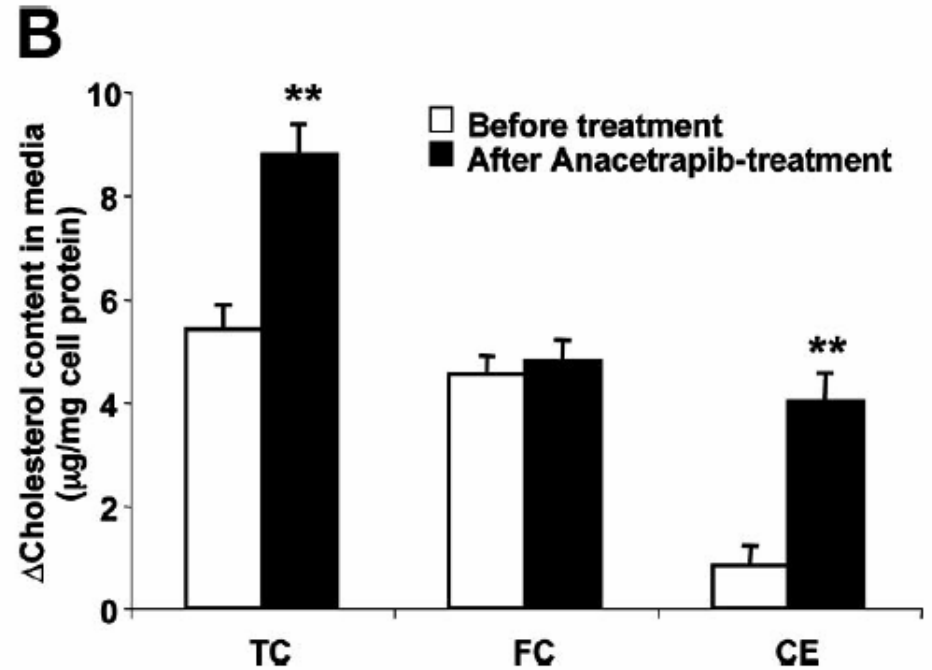
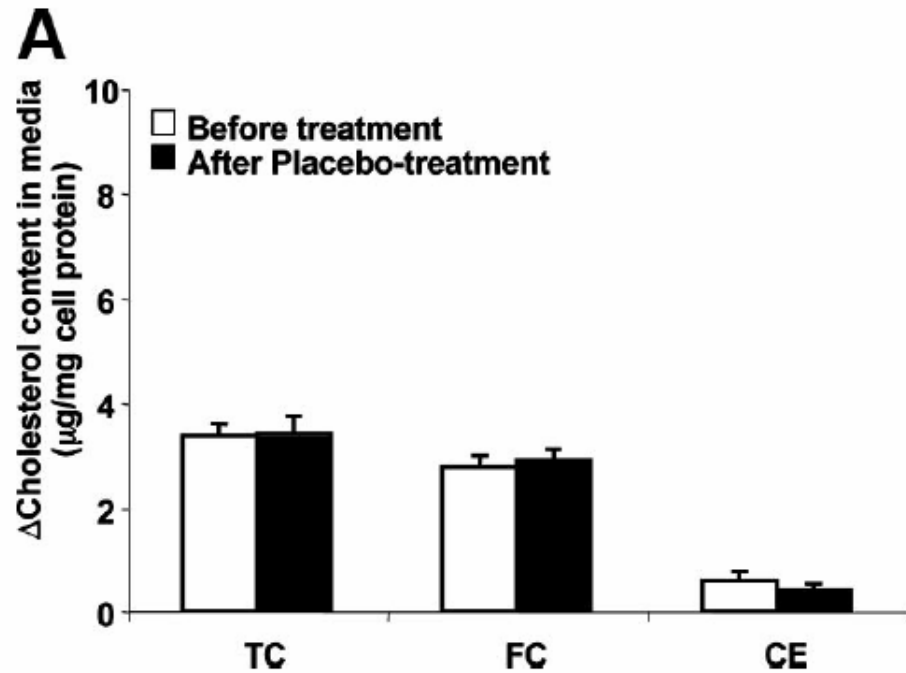
**Lipid peroxides added to human plasma can covalently modify apoAI.**

Szapacs ME. *J Proteome Res*. 2008;7:4237–4246.

**Incubation of HDL with activated neutrophils was shown to render apoAI more negatively charged and decrease its cellular cholesterol acceptor activity.**

Cogny A. *Biochem J*. 1996;314(Pt 1):285–292.

Cogny A. *Eur J Biochem*. 1994;222:965–973.



The increased efflux potential of **anacetrapib**-HDL was more prominent at higher HDL cholesterol concentrations (12 g/mL), which was associated with an increased content of LCAT and apolipoprotein E and completely dependent on the expression of **ABCA1** and **ABCG1**.

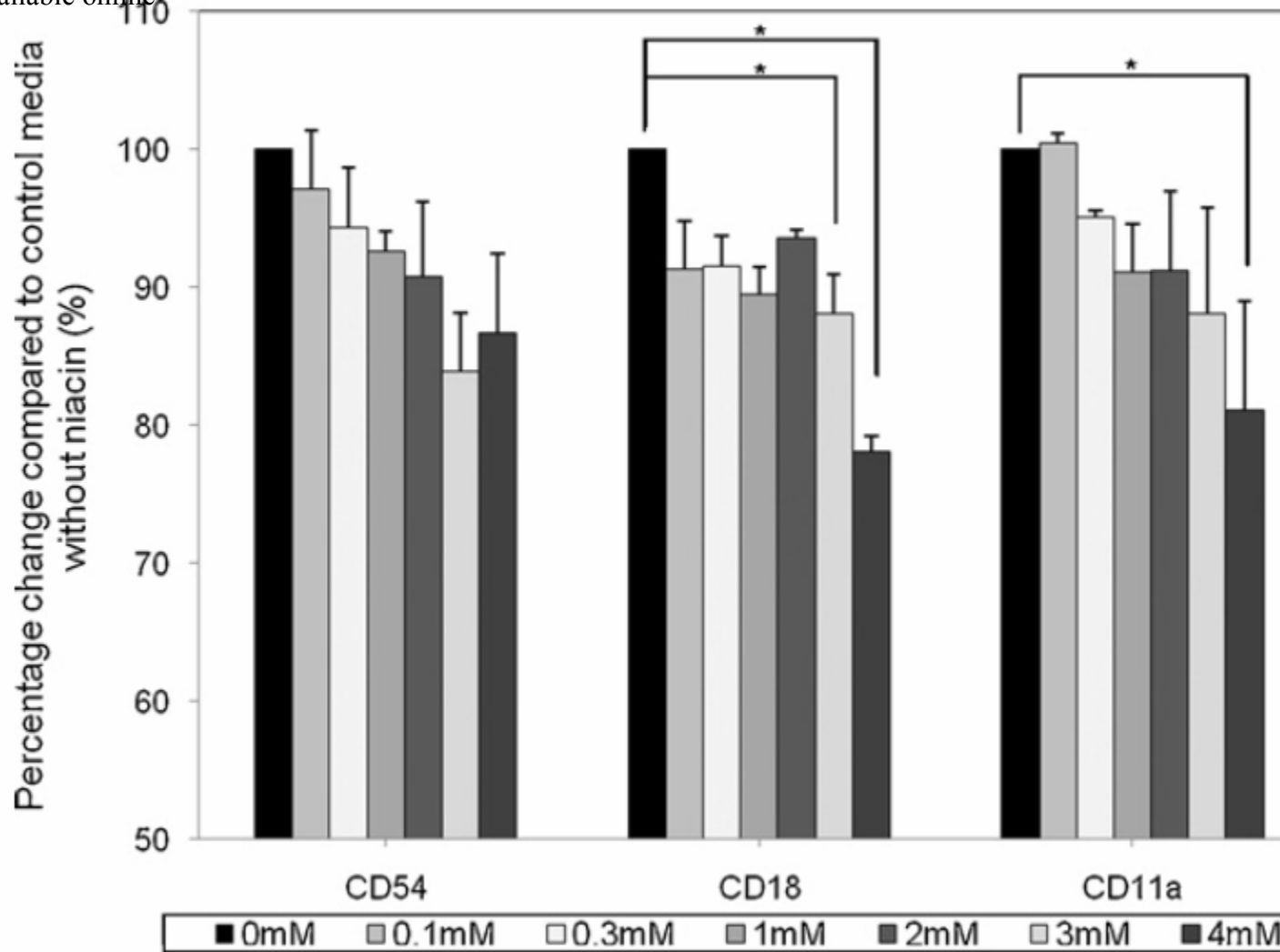
**Potent antiinflammatory effects of HDL were observed at low HDL concentrations (3 to 20 g/mL) and were partly dependent on the expression of ABCA1 and ABCG1.**

Yvan-Charvet L. *Arterioscler Thromb Vasc Biol.* 2010;30:1430-1438

# Niacin results in reduced monocyte adhesion in patients with type 2 diabetes mellitus.

S. Tavintharan Atheroscl 17December2010

Available online



- 39 Figure displays the changes in CD54, CD18 and CD11a expression on the surface of THP-1 monocytes treated with 18 h of niacin from 0 to 4mM

## **Tredaptive)**

**Niacin has:**

**Anti-atherothrombotic effects:**

**Anti-oxidative.**

**Improvement of endothelial dysfunction.**

**Attenuated systemic and arterial inflammation.**

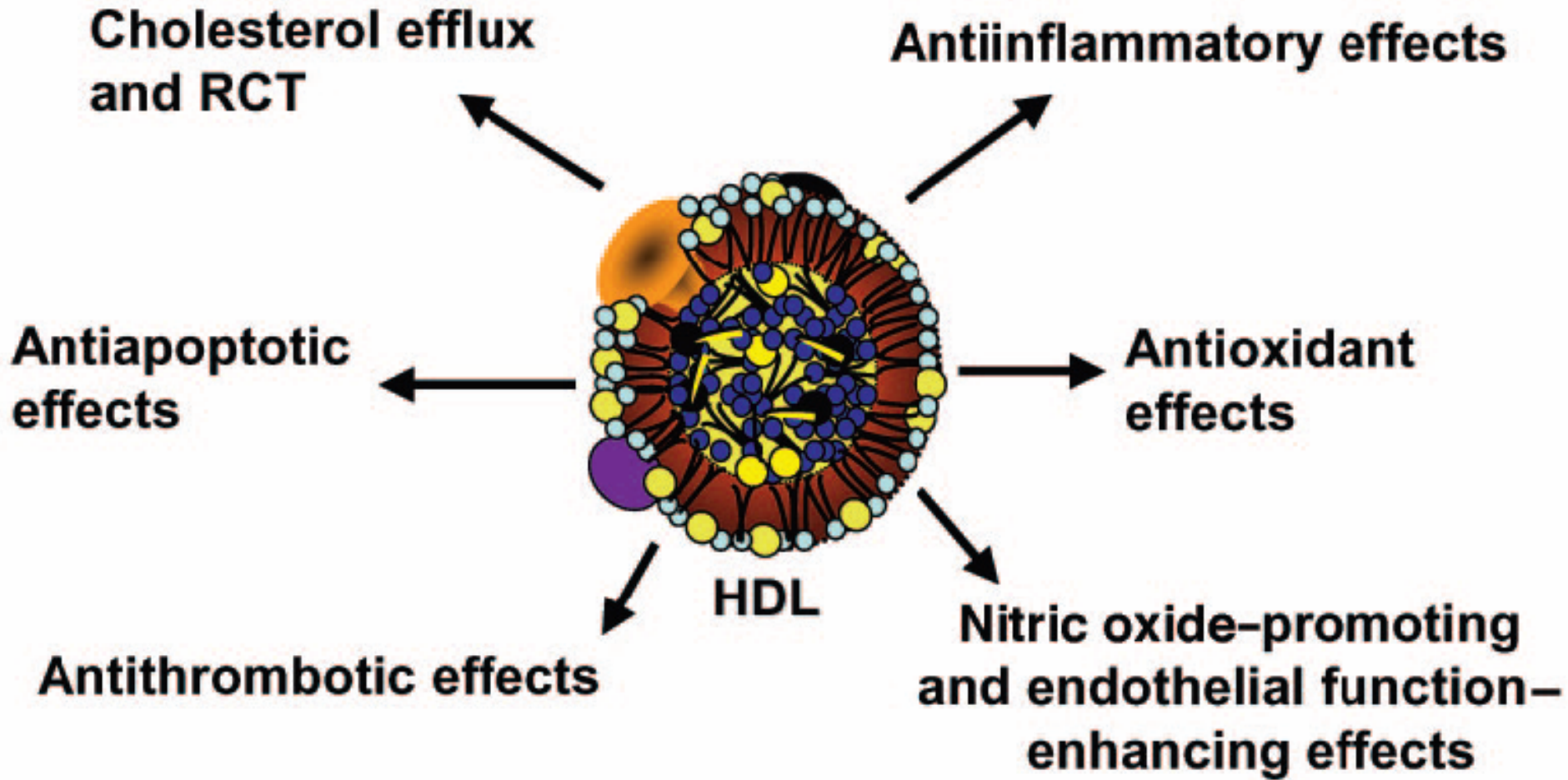
**Enhanced plaque stability.**

**Diminished thrombosis.**

(Rosenson, 2003; Carlson, 2005).

**Extended-release (ER) niacin (Niaspan®)**

**ER niacin with antiflushing agent laropiprant  
(Tredaptive)**

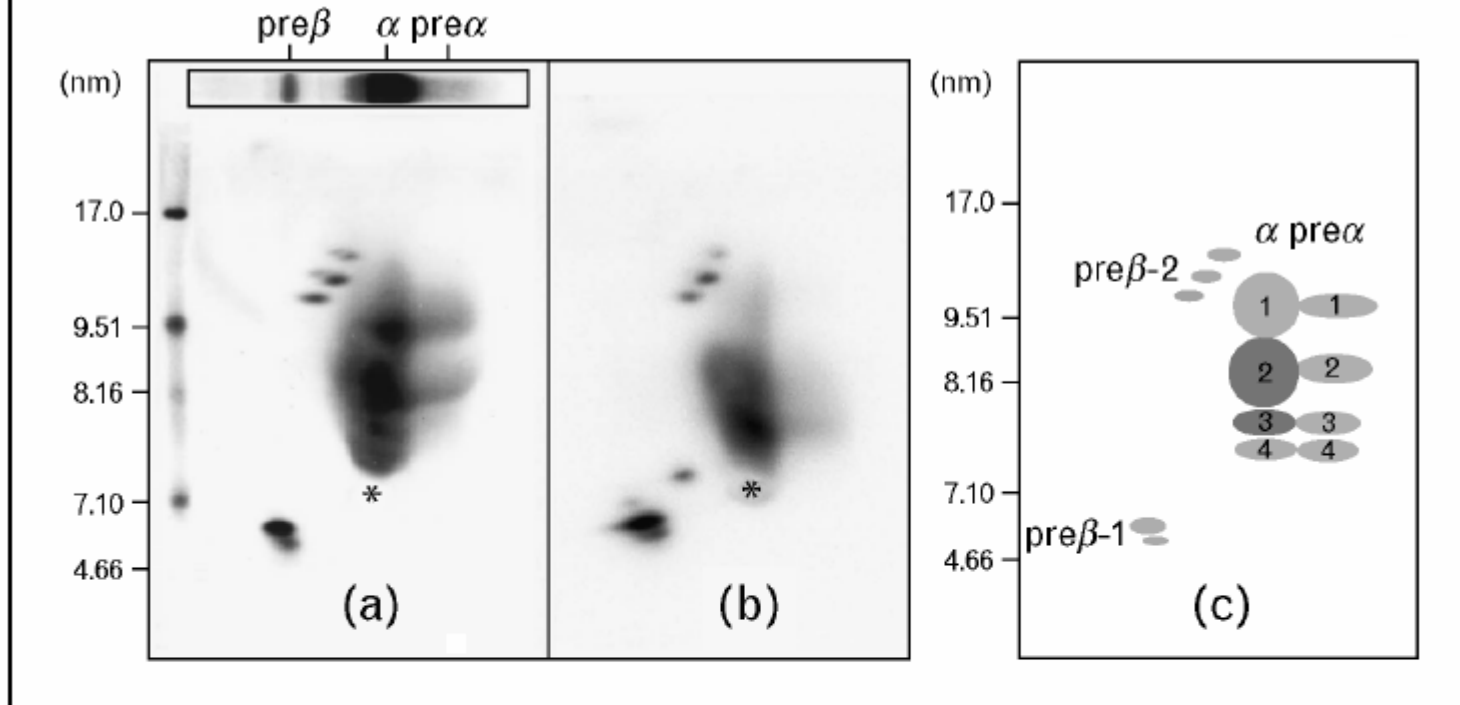


# Diversity of HDL particles

## **Non-denaturing two-dimensional electrophoresis:**

**There is a number of HDL particles with distinct shape, size and composition.**

Asztalos BF, Schaefer EJ. Am J Cardiol 2003;91:12E-7E.



ApoA-I-containing HDL subpopulation profiles of control (a) and CHD (b) subjects with much less  $\alpha$ -1 HDL than the control. Panel (c) is a schematic representation of the individual HDL particles. Size is on the vertical axis going from large to small particles, and charge on the horizontal axis going from pre $\beta$  to  $\alpha$  to pre $\alpha$  HDL particles. Lighter gray represents HDL particles containing apoA-I without apoA-II and darker gray represents HDL particles containing both apoA-I and apoA-II. All  $\alpha$  HDL particles contain C apolipoproteins, while  $\alpha$ -2 HDL contains SAA. ApoA-IV and apoE are on their own separate HDL particles.

**Comparison between patients with CHD and a control group:**

**CHD patients have:**

**Reduced  $\alpha_1$ , pre-  $\alpha_1$ , pre-  $\alpha_2$ , and pre-  $\alpha_3$  HDL subspecies**

**Elevated  $\alpha_3$  subspecies.**

**Administration of statins to patients with CHD increased  $\alpha_1$  and pre-  $\alpha_1$ , suggesting that some of the atheroprotective properties of statins may be mediated by increasing select HDL subpopulations.**

**HDL from many CAD patients was proinflammatory, thus increasing monocyte chemotaxis in response to LDL, unlike the HDL from healthy controls that reduced monocyte chemotaxis.**

Ansell BJ. *Circulation*. 2003;108:2751–2756.

**Each HDL particle carries:**

**ApoA1.**

**May also other apolipoproteins (apoAII, apoAIV, apoE, apoC).**

**HDL is associated with accessory proteins:**

**LCAT.**

**Phospholipid transfer protein.**

**Paraoxonase 1.**

**Myeloperoxidase.**

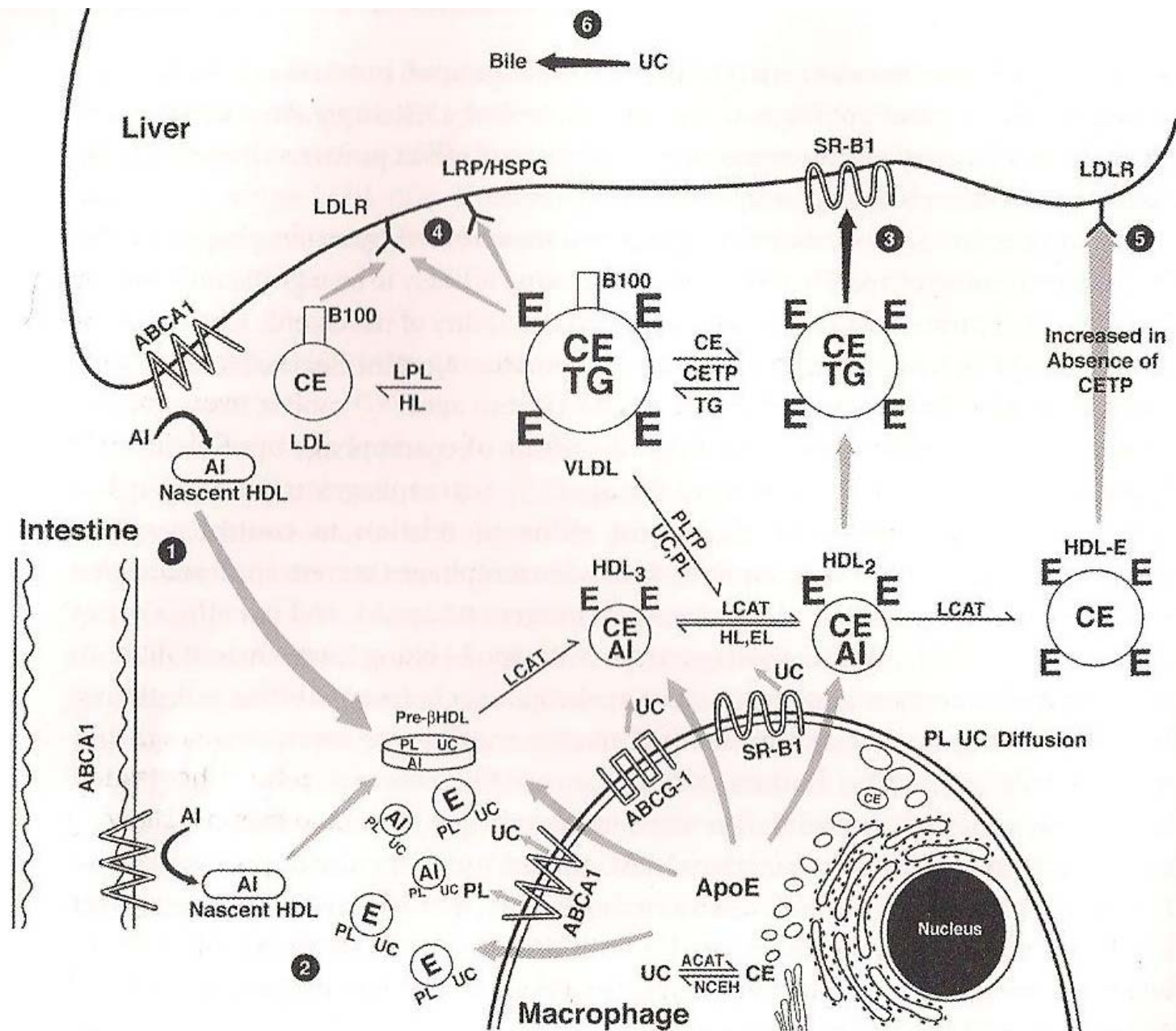
**Serum amyloid A1.**

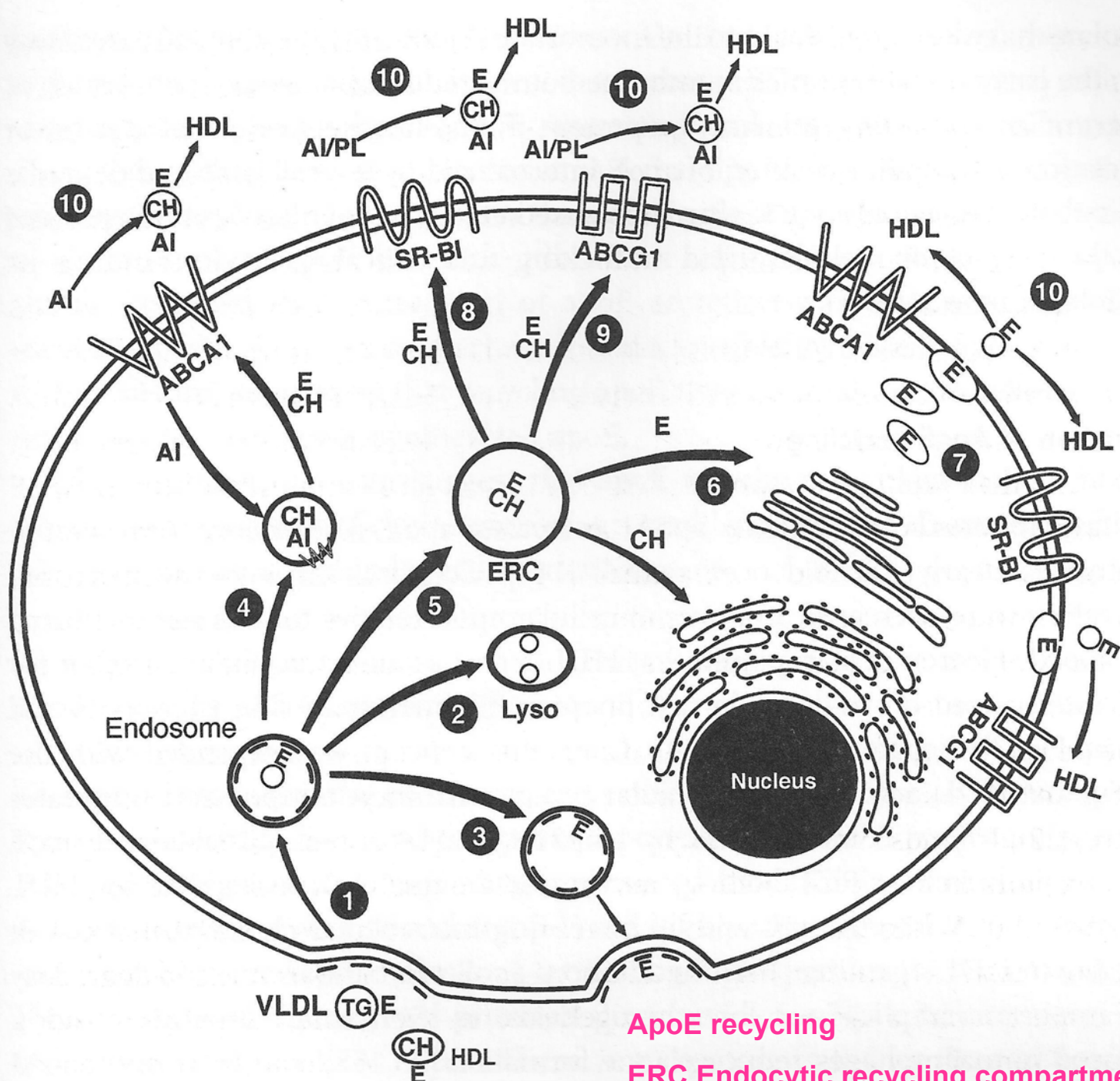
**Platelet-activating factor acetylhydrolase.**

**The HDL proteome is quite large, and each HDL particle cannot accommodate all of the accessory proteins.**

**HDL proteomic study of HDL<sub>3</sub> from different subjects found that the levels of PON1, PON3, and apoL-I correlate with HDLs antioxidant activity.**

Davidson WS. *Arterioscler Thromb Vasc Biol.* 2009;29.





ApoE recycling  
 ERC Endocytic recycling compartment

## Assays for HDL function

**HDL inhibits monocyte chemotaxis induced by LDL using an in vitro reconstituted artery wall model by the coculture of smooth muscle cells and endothelial cells.**

**Cell free antioxidant activity.**

**Inhibition of endothelial cell adhesion molecule expression.**

**Ability of HDL to act as an acceptor of cellular cholesterol.**

Navab M. *J Lipid Res.* 2000;41:1495–1508.

Ansell BJ, *Circulation.* 2003;108:2751–2756.

Cockerill GW. *Arterioscler Thromb Vasc Biol.* 1995;15:1987–1994.

## **Dysfunctional HDL:**

- 1. Metabolic syndrome, Diabetes mellitus.**
- 2. Atherogenic dyslipidemia.**
- 3. Inflammatory diseases**
- 4. Chronic renal disease.**
- 5. Autoimmune disease**
- 6. Women, increased HDL-C, no other cardiovascular risk factors, evident atherosclerosis.**

## **Treatment for dysfunctional HDL:**

- 1. Treat metabolic derangements.**
- 2. Treat inflammatory/autoimmune disease.**
- 3. Niacin.**
- 4. Fibrates?**
- 5. CETP inhibitors?**

# תודה על ההקשבה