



# Impaired cholesterol efflux capacity may help explain development of early coronary artery disease in subjects with very high HDL-C

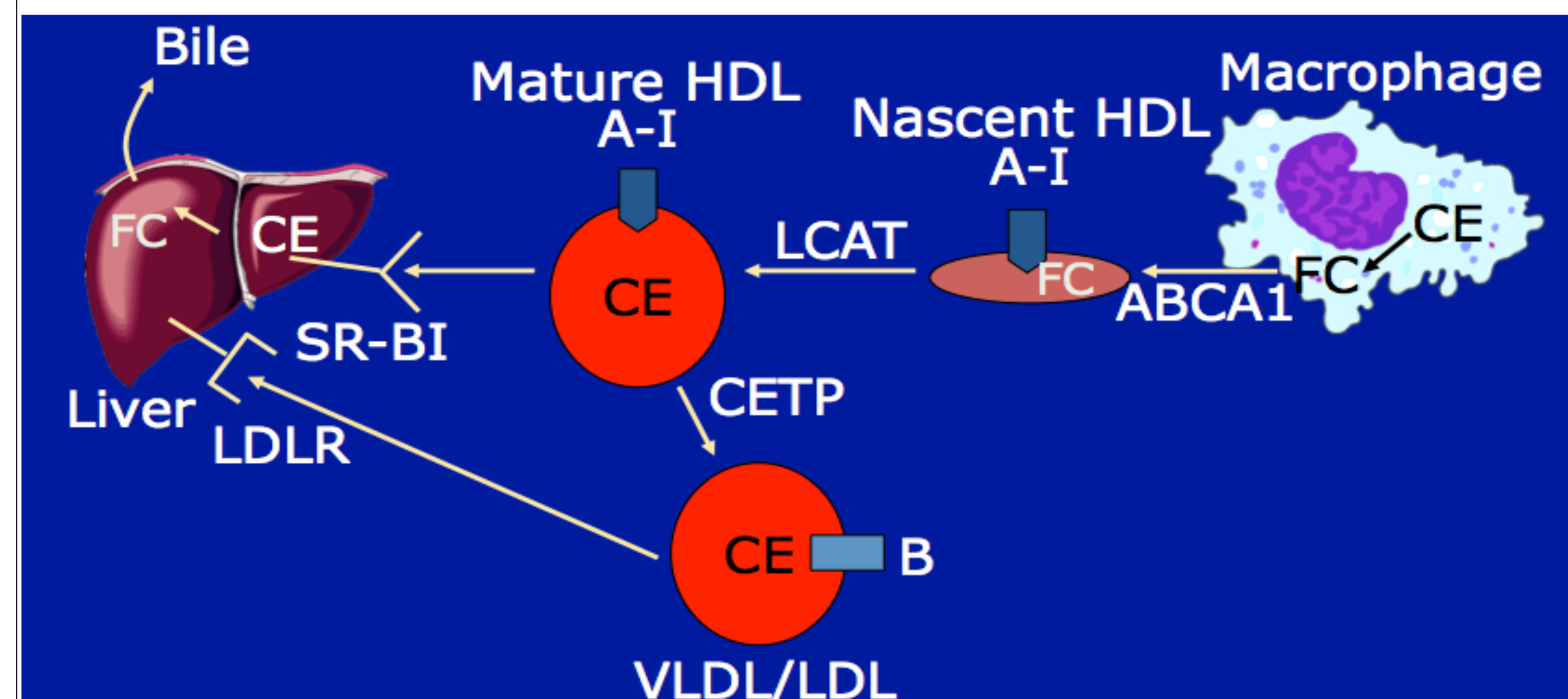
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## INTRODUCTION

In epidemiologic studies, plasma levels of HDL cholesterol (HDL-C) are strongly and inversely associated with coronary artery disease (CAD), and high HDL-C is generally associated with reduced risk and apparent 'protection' from CAD.



**Figure 1: Reverse cholesterol transport** is the process by which HDL removes cholesterol from lipid laden macrophages and transports it to the liver to be excreted from the body.

We have identified a number of individuals with extremely high HDL-C who have a paradoxical phenotype of early CAD.

We hypothesized that such individuals may have HDL with altered structure and function, and systematically compared these cases to matched controls with similarly high HDL-C but without CAD.

## METHODS

### Study design:

- 55 subjects with HDL-C above the 90<sup>th</sup> percentile, CAD, and no major risk factors for coronary disease.
- Controls selected per case, each matched for age, race, gender, and HDL level.

### Plasma cholesterol efflux capacity<sup>1</sup>:

- J774 cells labeled with <sup>3</sup>H cholesterol and incubated with plasma from each subject
- The ability of HDL to act as an acceptor was measured in the presence or absence of ABCA1 upregulation by cAMP.

### Lecithin-cholesterol acyltransferase (LCAT) activity:

- <sup>3</sup>H free cholesterol was exchanged into plasma and the ability to be converted to cholesterol ester by LCAT was measured.

**HDL particle size:** HDL particle size and distribution was measured by NMR by Liposcience

**PLTP activity:** Plasma PLTP activity was measured using the Kamiya PLTP activity assay kit.

### Statistical analysis:

- A logistic regression was done using HDL classified case/control status as the dependent variable. Race was self-reported.

## RESULTS

### Characteristics of Cases and Controls

	Cases	Controls
N	55	120
Age	64±11	69±12
% female	36.4	40
% Caucasian	96.4	93.9
% African American	3.6	6.1
Mean age of onset of CAD	60	-
Males	60	-
Females	60	-
%Diabetes	0	0
%smokers	47.3	47.0
Triglycerides	80 ± 34	85 ± 38
Total Cholesterol	201 ± 47 <sup>++</sup>	228 ± 37
HDL-C	86 ± 21	86 ± 20
LDL-C	97 ± 38 <sup>++</sup>	125 ± 33
Total phospholipid	253 ± 55 <sup>+</sup>	274 ± 52
HDL Phospholipid	92 ± 37 <sup>+</sup>	109 ± 43
HDL Triglyceride	13 ± 9 <sup>+</sup>	11 ± 4
ApoA-I	195 ± 42	194 ± 40
ApoB	77 ± 21 <sup>++</sup>	89 ± 19
Lp(a)*	22.5	13.7
ApoA-II	43 ± 11	40 ± 15
ApoC-III	15 ± 5	13 ± 5
ApoE	5 ± 2	6 ± 2 <sup>+</sup>

**Table 1: Characteristics of cases and controls** (mean ± st. dev). Lipid and apolipoprotein parameters are mg/dl. <sup>+</sup> p <0.05 compared to cases, <sup>++</sup> p <0.001 \*Median value reported

### ABCA1- mediated cholesterol efflux is reduced in subjects with high HDL-C and CAD

	Basal (-cAMP)	Activated (+cAMP)	ABCA1-specific
Cases	1.36 ± 0.28	1.96 ± 0.39 <sup>+</sup>	0.60 ± 0.24 <sup>+</sup>
Controls	1.40 ± 0.30	2.11 ± 0.43	0.71 ± 0.32
P	0.44	0.040	0.033

**Table 2: Cholesterol efflux capacity** (%efflux/ 2hr/ 1% plasma)

### Efflux to HDL-C is reduced in subjects with high HDL-C and CAD

	Basal -cAMP	Activated +cAMP
Cases	0.017 ± 0.003	0.023 ± 0.005 <sup>+</sup>
Controls	0.017 ± 0.003	0.025 ± 0.006
P	0.73	0.029

**Table 3: Cholesterol efflux capacity** (%efflux/ 2hr/ 1% plasma)

### Plasma LCAT activity, HDL particle size, and plasma PLTP activity were not different between cases and controls

	LCAT activity	Large HDL particles	Medium HDL particles	Small HDL particles	PLTP activity
Cases	153.73 ± 55.23	12.41 ± 4.57	11.16 ± 6.27	16.78 ± 6.20	0.49 ± 0.18
Controls	169.28 ± 56.13	12.53 ± 4.04	10.67 ± 7.44	16.13 ± 6.50	0.51 ± 0.20
P	0.3	0.87	0.68	0.55	0.46

**Table 4:** Means and standard deviations of plasma LCAT activity (nmol esterified/ hr/ mL), HDL particle size (μmol/L), and plasma PLTP activity (nmol/ml/min).

## CONCLUSIONS

- HDL-PL levels were lower in cases than controls.
- Total and ABCA1- mediated plasma cholesterol efflux capacity is reduced in cases with high HDL and CAD. This finding may help explain the development of early coronary disease in the cases.
- There were no differences in HDL particle size, LCAT activity, or PLTP activity between cases and controls.

Changes in HDL particle composition, specifically reduced HDL phospholipid composition, may help explain the reduction in cholesterol efflux capacity in cases with coronary disease.

## REFERENCES AND ACKNOWLEDGEMENTS

<sup>1</sup>Khera, A.V., et al., *Cholesterol efflux capacity, high-density lipoprotein function, and atherosclerosis*. N Engl J Med, 2011. 364(2): p. 127-35.

This work was supported by a grant from the Doris Duke Charitable Foundation to The University of Pennsylvania to fund Clinical Research Fellow Anandita Agarwala.