The Sortilin Pathway – A New Target for Reducing LDL and CVD Risk

Ronald M. Krauss
Children's Hospital Oakland Research Institute
UCSF and UC Berkeley

Disclosures

AFFILIATION/FINANCIAL INTERESTS	CORPORATE ORGANIZATION
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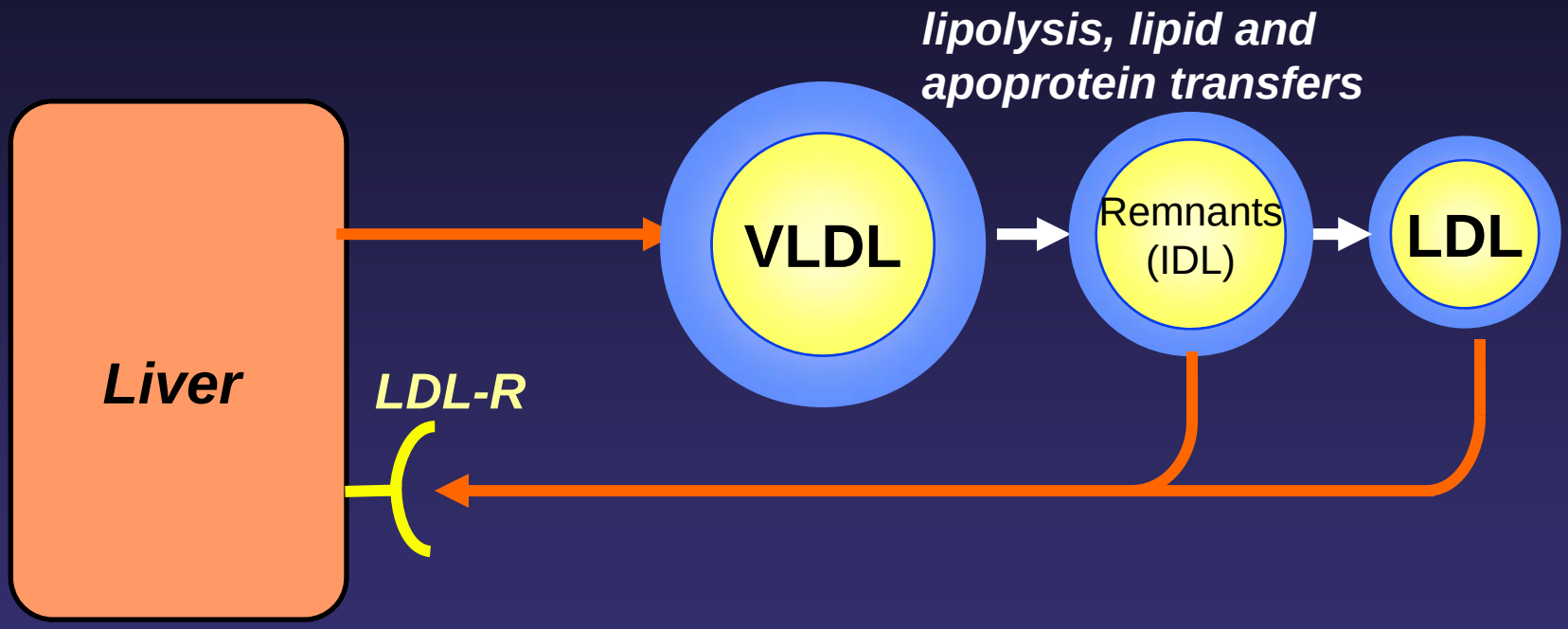
LDL Metabolism

LDL Metabolism – Traditional Model



Eisenberg et al., Biochim Biophys Acta 326: 361-377, 1973

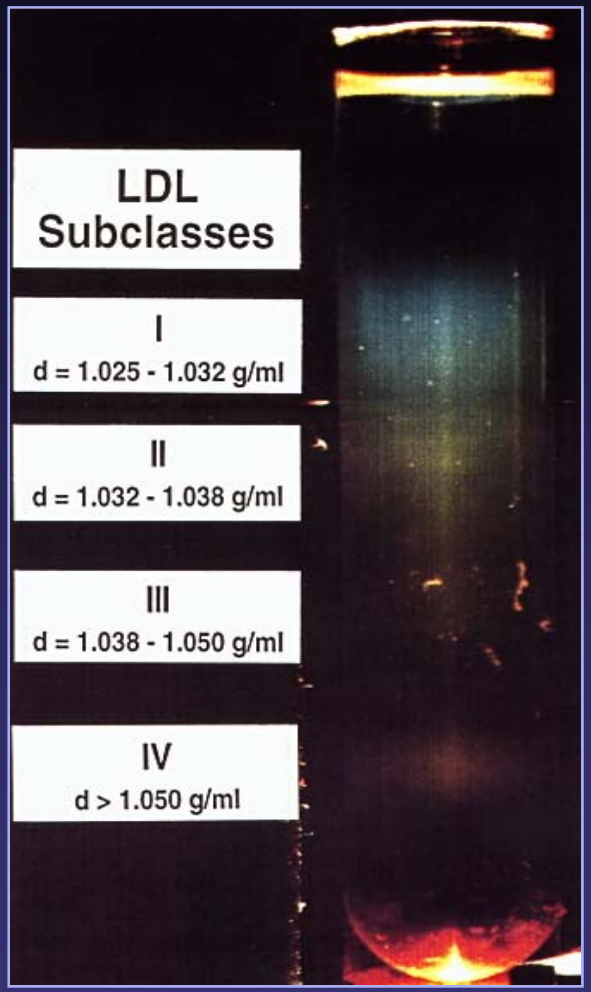
But LDL comprises multiple discrete subclasses

Large

Medium

Small

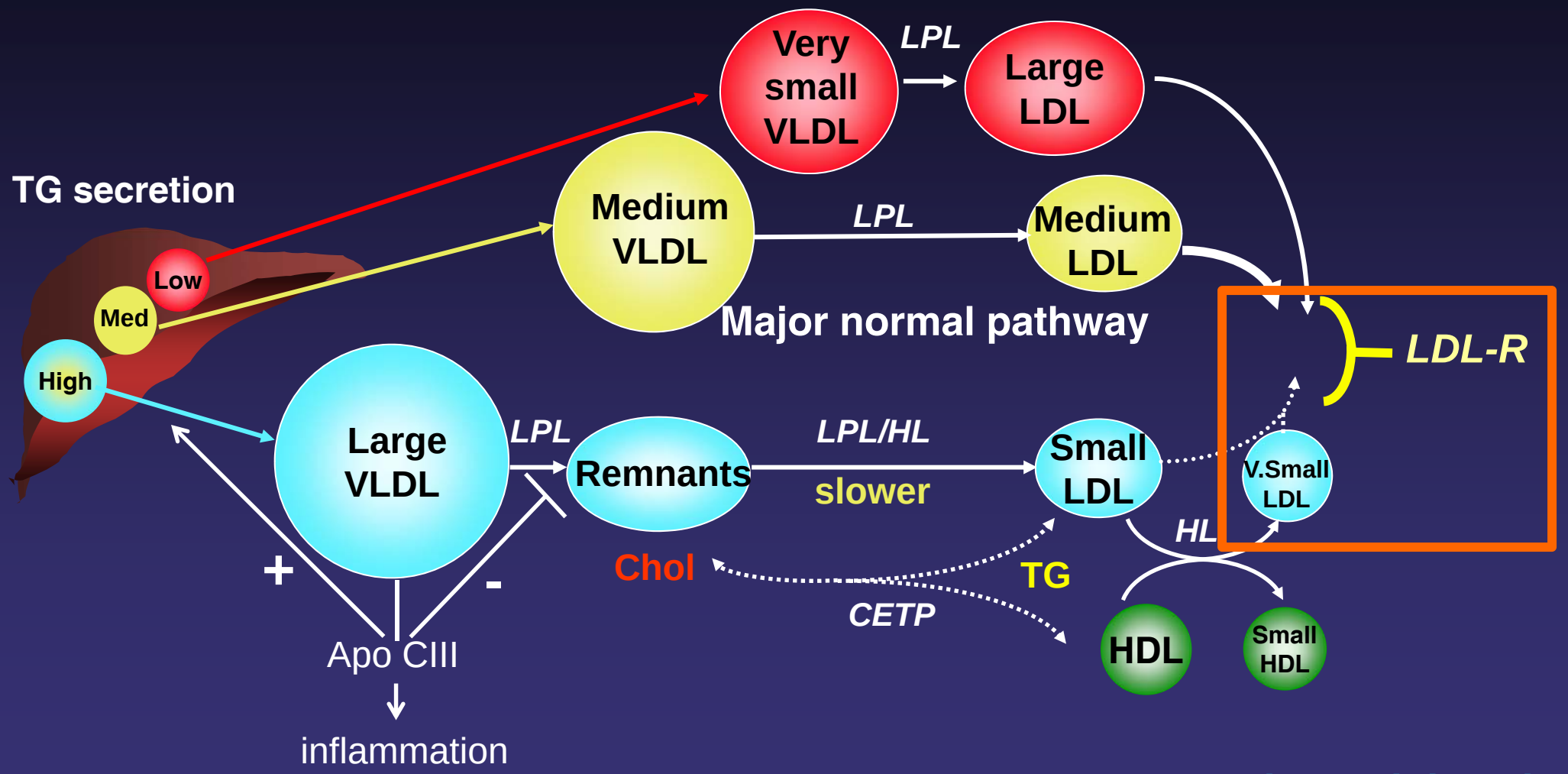
Very small



More buoyant = larger particle diameter, higher lipid content

Krauss and Burke, JLR 1982

Model for pathways that give rise to major LDL subclasses



Adapted from Berneis and Krauss, JLR 43:1155, 2002

Relatively low hepatic uptake of small/very small LDL via the LDL receptor is consistent with evidence that >10-15% of LDL particles are not cleared by this receptor.

Spady et al. PNAS 80:3499, 1983

Small & Very Small LDL and Atherogenesis

On-treatment small/very small LDL associated with angiographic progression of coronary disease – HATS

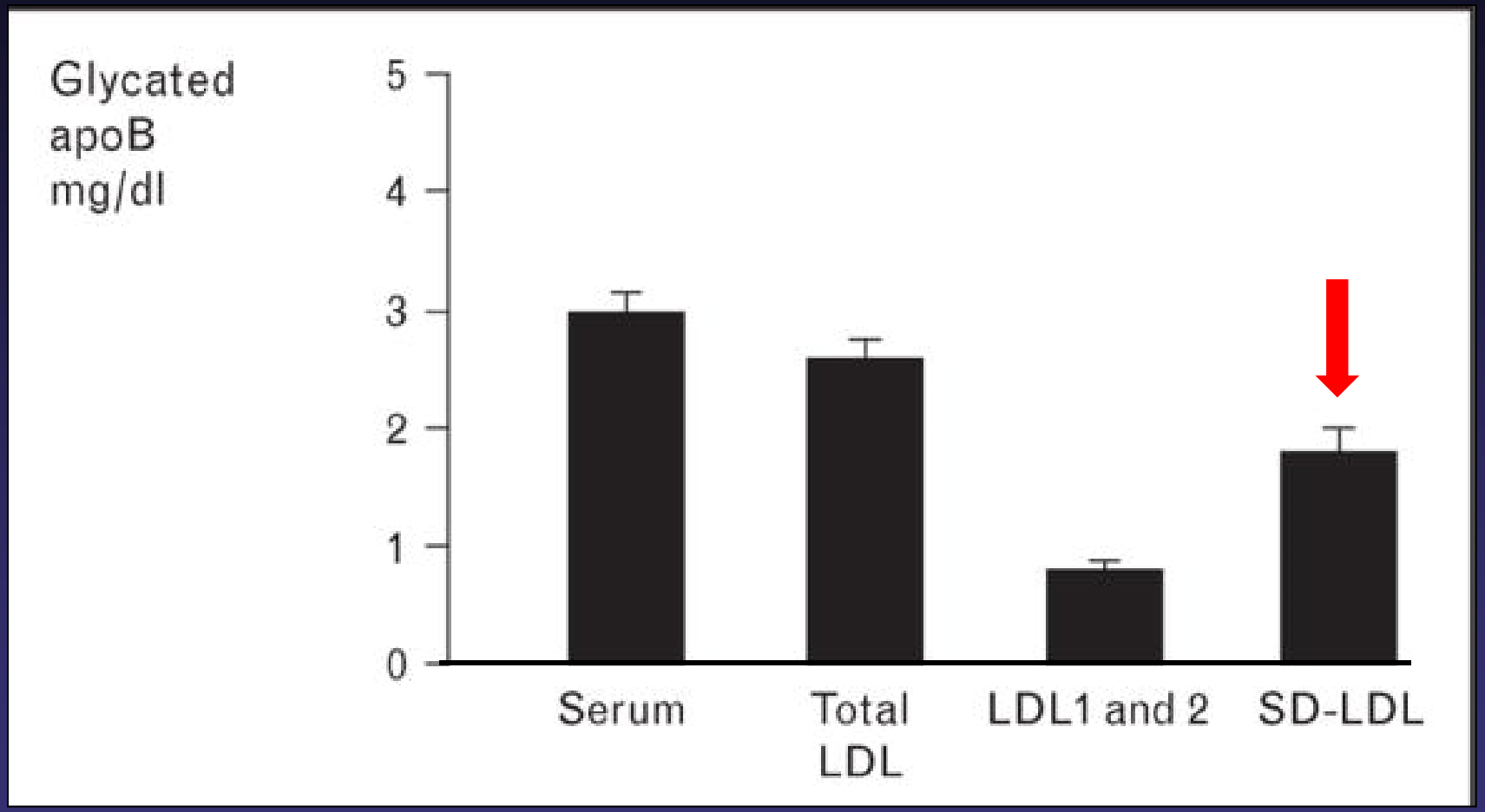
	p adjusted for lipids	p adjusted for apoB or TC/HDL
Ion Mobility		
Large VLDL	0.02	0.05
Medium VLDL		0.03
Small LDL 3a	0.04	0.03
Very small LDL 3b	0.02	0.03
GGE (LDL only)		
Very small LDL 3b	0.00001	0.00001
Very small LDL 4a	0.04	0.04
NMR		
Small LDL	0.03	0.03
VAP		
LDL 4		0.04 (adj. TC/HDL)

Williams et al., Atherosclerosis Jan 2014 epub

Why are smaller LDL particles more predictive of CHD than larger LDL?

- Reduced LDL receptor binding – longer plasma residence time
- Greater arterial proteoglycan binding
- Greater oxidative susceptibility
- Association with other risk biomarkers:
  - Reduced HDL, increased remnants
  - Insulin resistance
- Atherogenic components
  - Oxidized lipids
  - Glycated apoB
  - ApoC-III

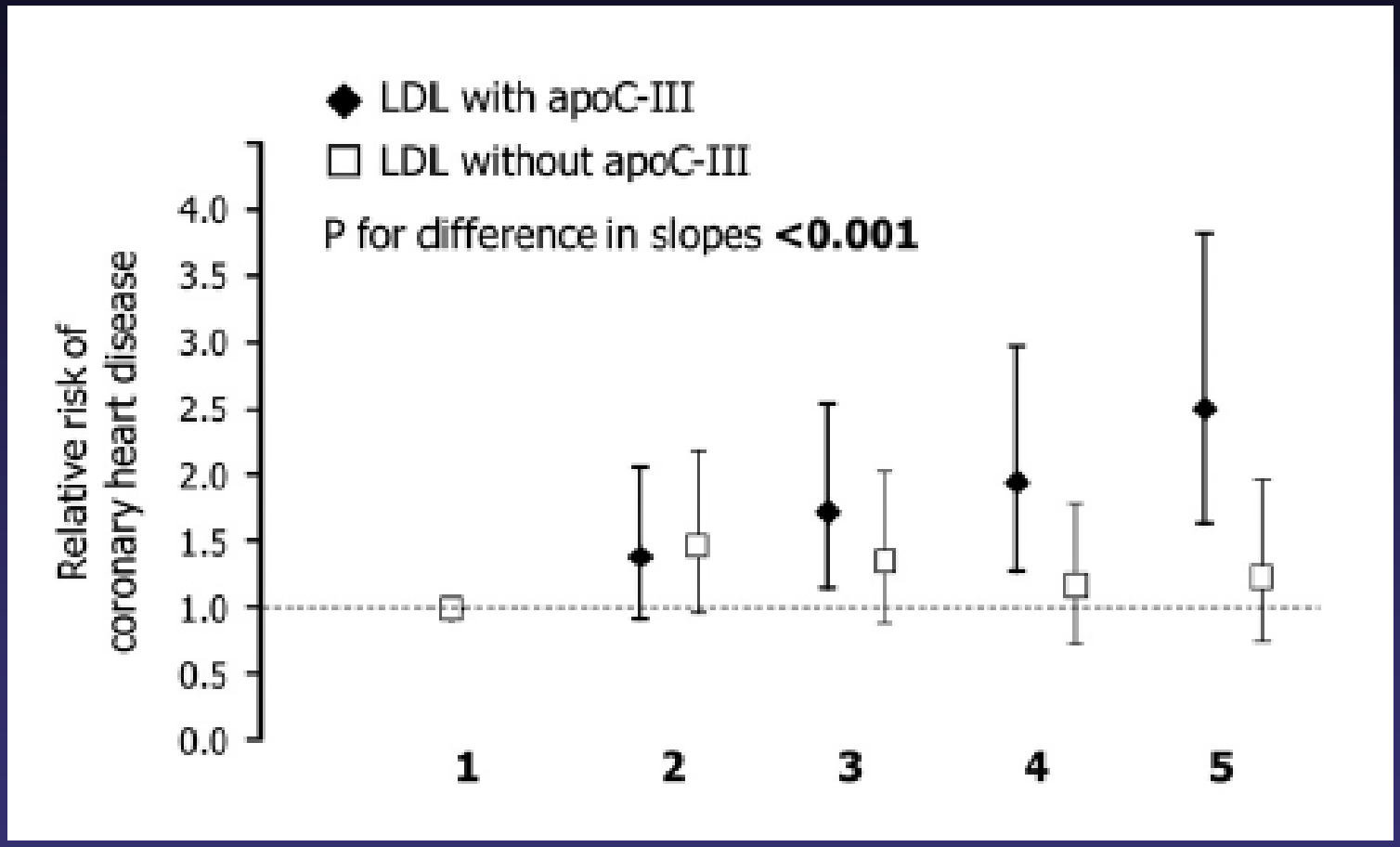
Glycated apoB is enriched in small, dense LDL particles in non-diabetic individuals



Small, dense LDL fractions are also preferentially glycated *in vitro*

Soran and Durrington Curr. Opin. Lipidol. 22:254, 2011

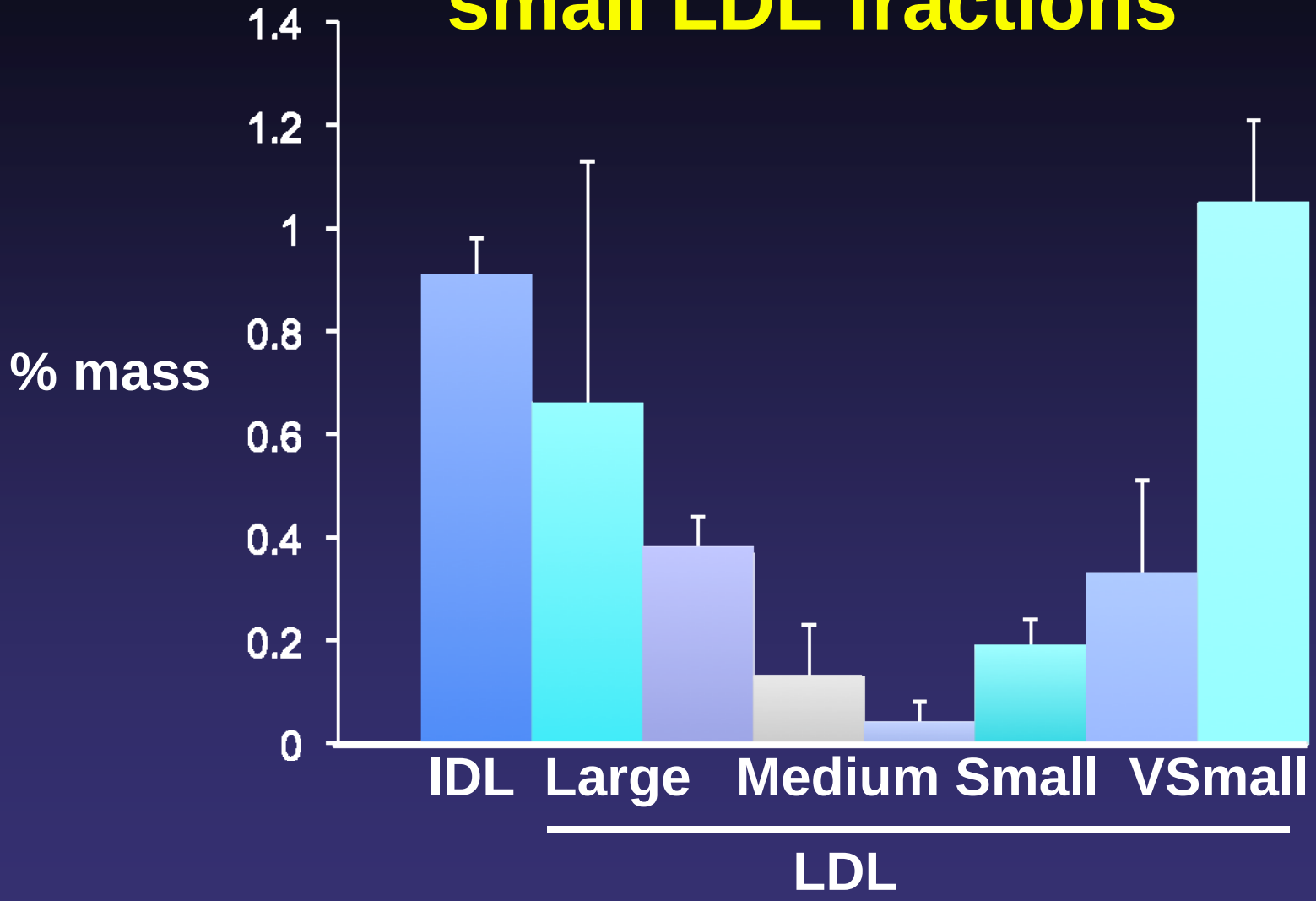
Risk for developing coronary heart disease is associated with LDL containing apo-CIII



With apoC-III	n=	240	270	297	318	351
Without apoC-III	n=	265	295	317	288	311

Mendivil et al., Circulation 124:2065, 2011

ApoCIII is enriched in IDL and very small LDL fractions



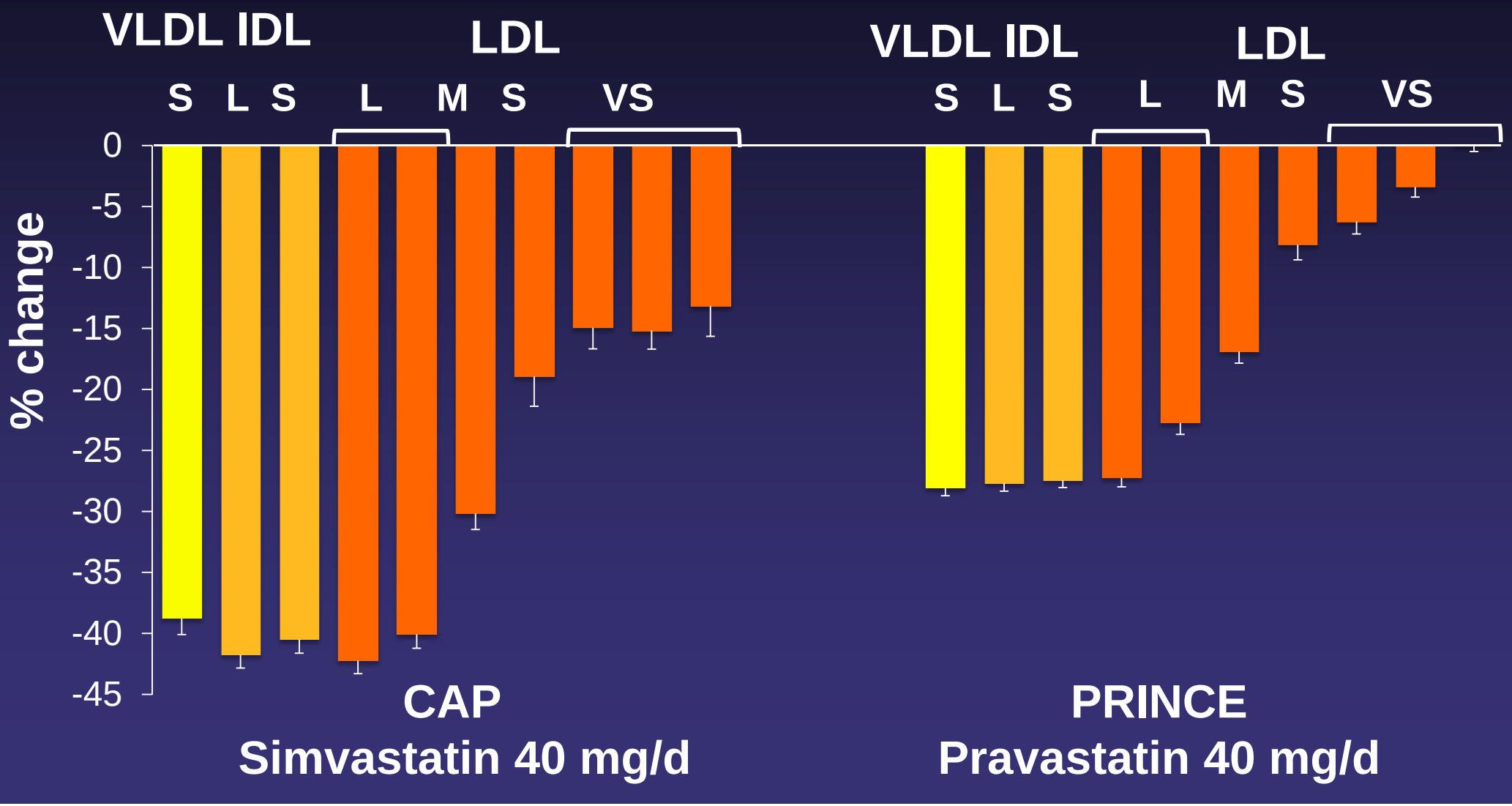
Consistent with findings in type 2 DM; Hiukka et al. Diabetes, 2009

Data from Krauss et al., J. Lipid Res 53: 540, 2012

- **Statins act to reduce LDL in large part by increasing receptor-mediated uptake.**

- **How effective are statins for reducing plasma levels of small and very small LDL particles?**

Lesser reductions in plasma levels of small and very small LDL cholesterol with statin treatment



- **Statins act to reduce LDL in large part by increasing receptor-mediated uptake.**

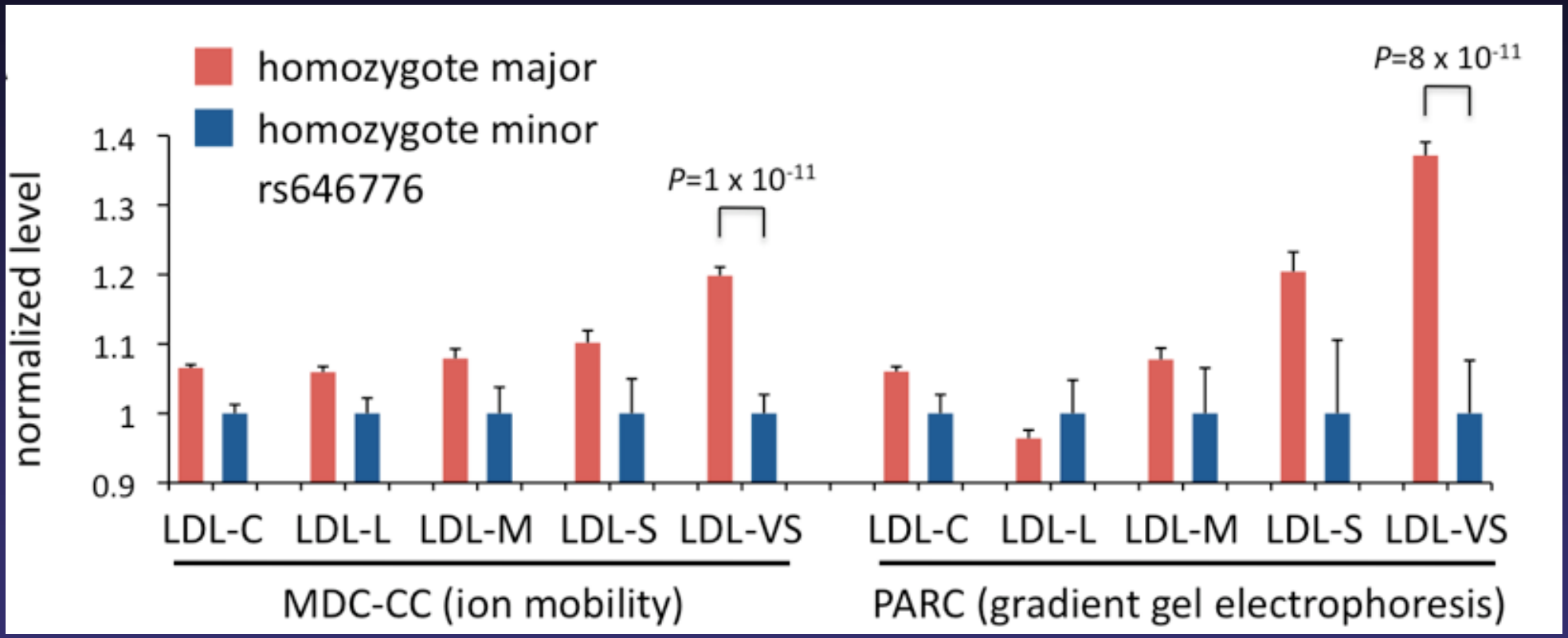
- **Is there a mechanism other than LDL receptor uptake for reducing plasma levels of small and very small LDL particles?**

Top Lipid-Associated SNPs Also Associated with CAD

Trait	Locus	SNP (rs)	n Lipid	p Lipid	n CAD	p CAD
LDL-C	*LDLR*	6511720	170, 607	3.9 x 10-262	86,870	1.2 x 10-7
	SORT1	629301	142,643	5.4 x 10-241	82,222	6.1 x 10-10
	APOB	1367117	173,007	9.5 x 10-183	79,823	2.3 x 10-2
	APOE	4420638	93,103	1.5 x 10-178	36,066	2.1 x 10-4
TRIG	*APOA1/A5*	964184	90,991	6.6 x 10-224	110,492	4.8 x 10-11
	LPL	12678919	177,749	1.8 x 10-199	110,065	4.7 x 10-5
	TRIB1	2954029	177,729	1.0 x 10-107	81,977	2.8 x 10-5
HDL-C	*CETP*	3764261	177,533	1.4 x 10-769	83,626	2.2 x 10-3

Global Lipids Genetics Consortium. Nat Genet 45:1274, 2013

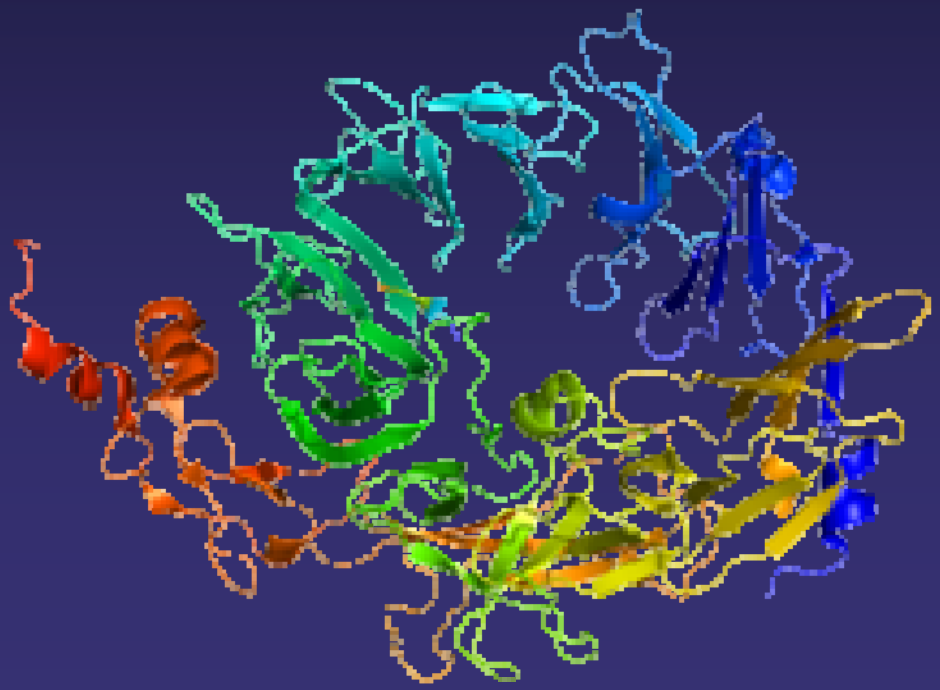
SORT1 SNP is preferentially associated with an increase of very small LDL particles in two independent studies



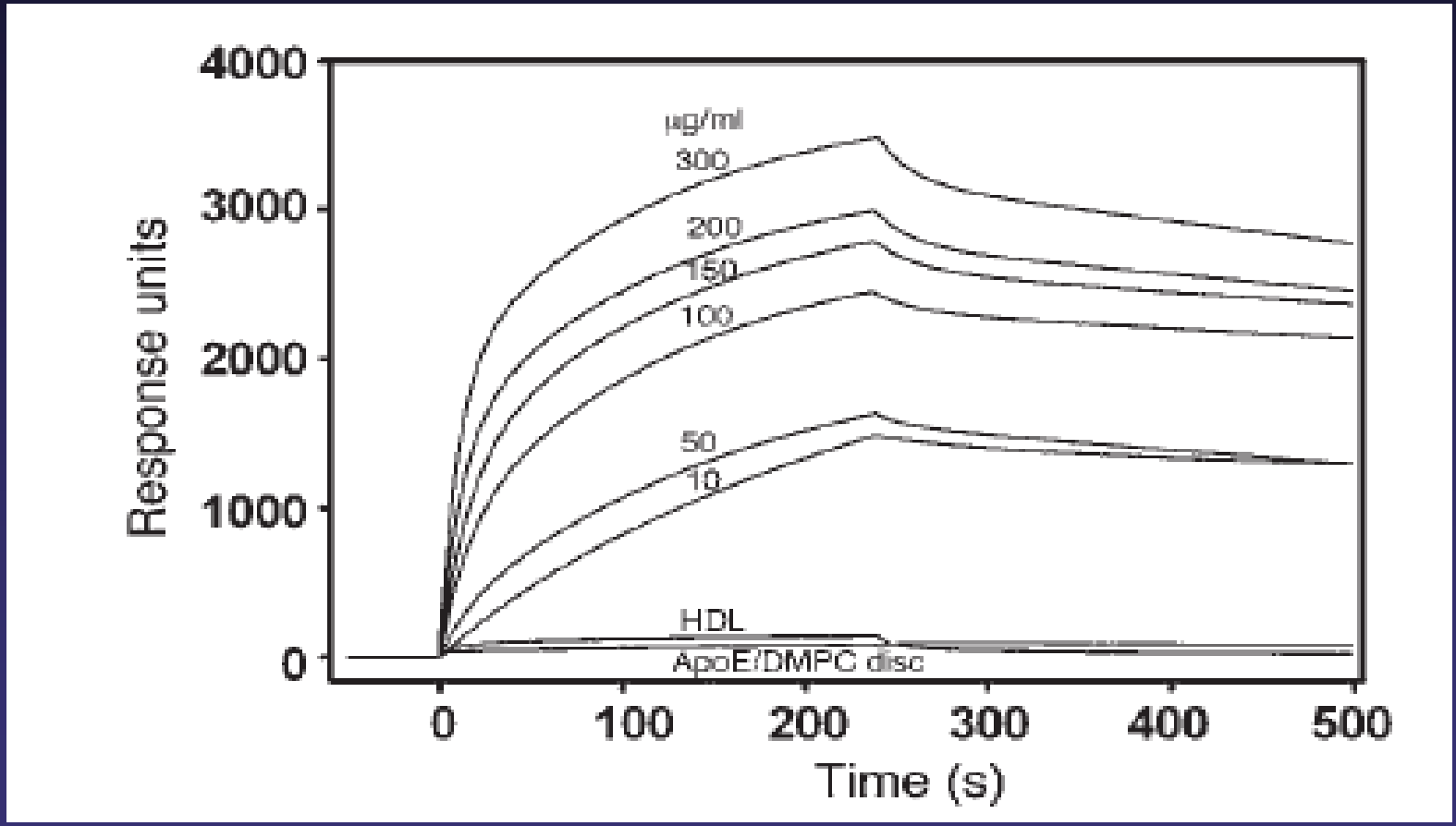
rs 646776 is at the *CELSR2/PSRC1/SORT1* locus on Chr1p; in LD with SNP that regulates expression of these genes

Musunuru et al., Nature 466:714, 2010

Sortilin: a trans-Golgi network transmembrane protein that binds a number of unrelated ligands that participate in a wide range of cellular processes.

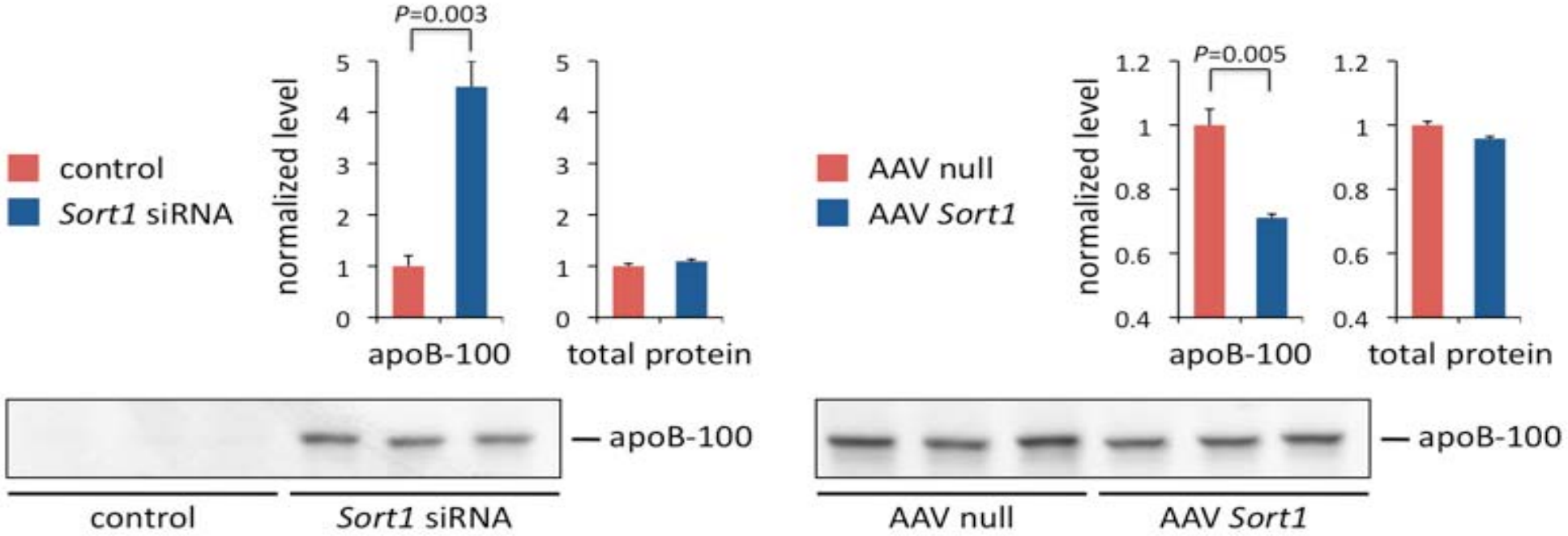


Sortilin binds with high affinity to apoB



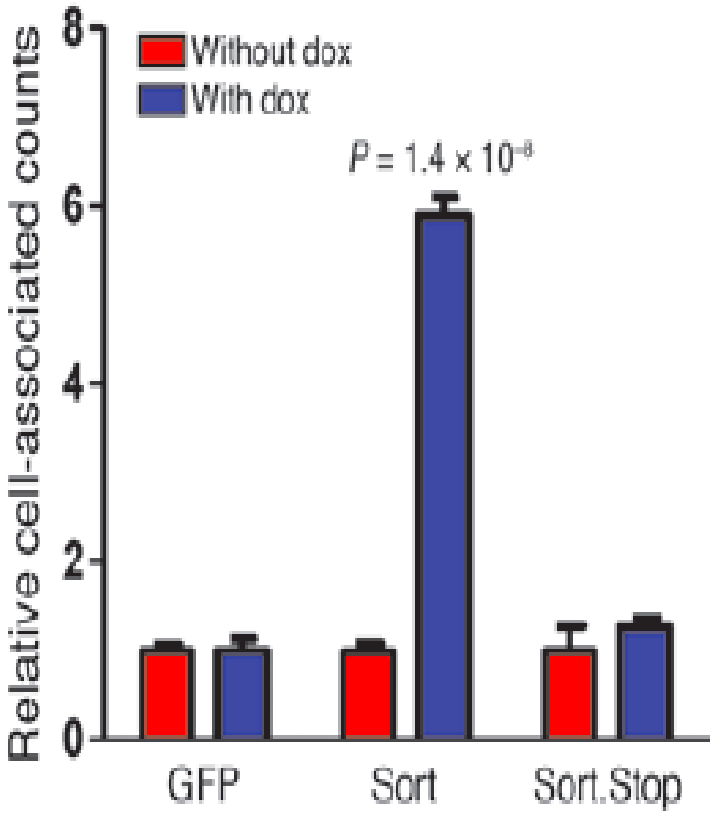
Strong et al., J Clin Invest 122:2807, 2012

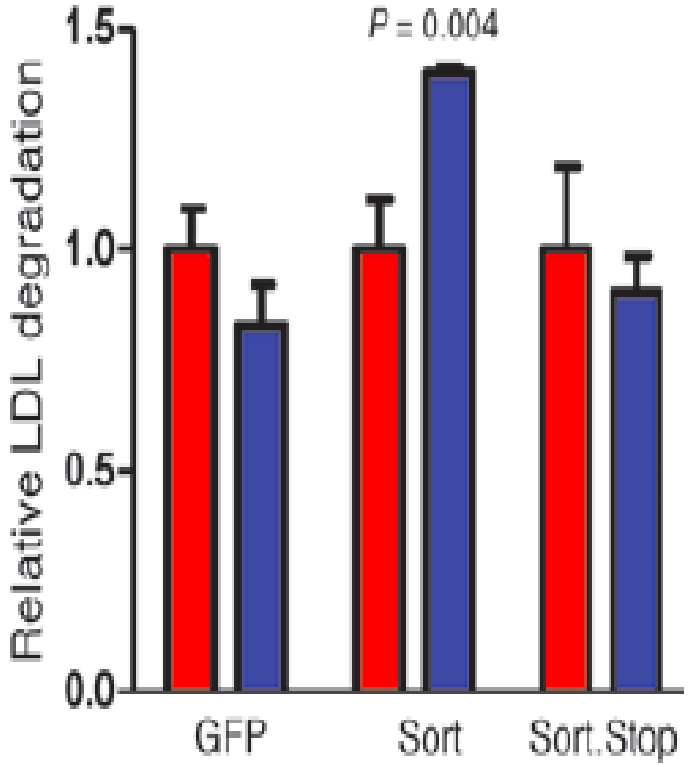
Hepatocyte apoB-100 secretion is increased by sort1 knockdown and decreased by sort1 overexpression



Musunuru et al., Nature 466:714, 2010

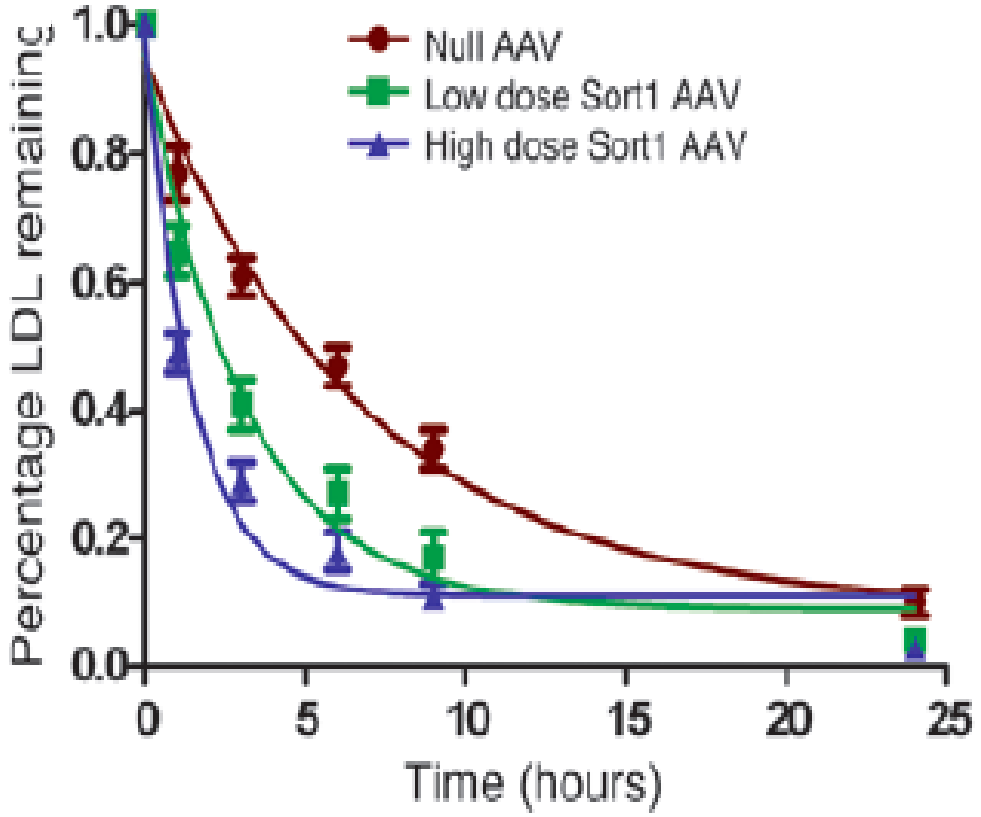
Increased sortilin expression promotes LDL uptake and lysosomal catabolism in vitro

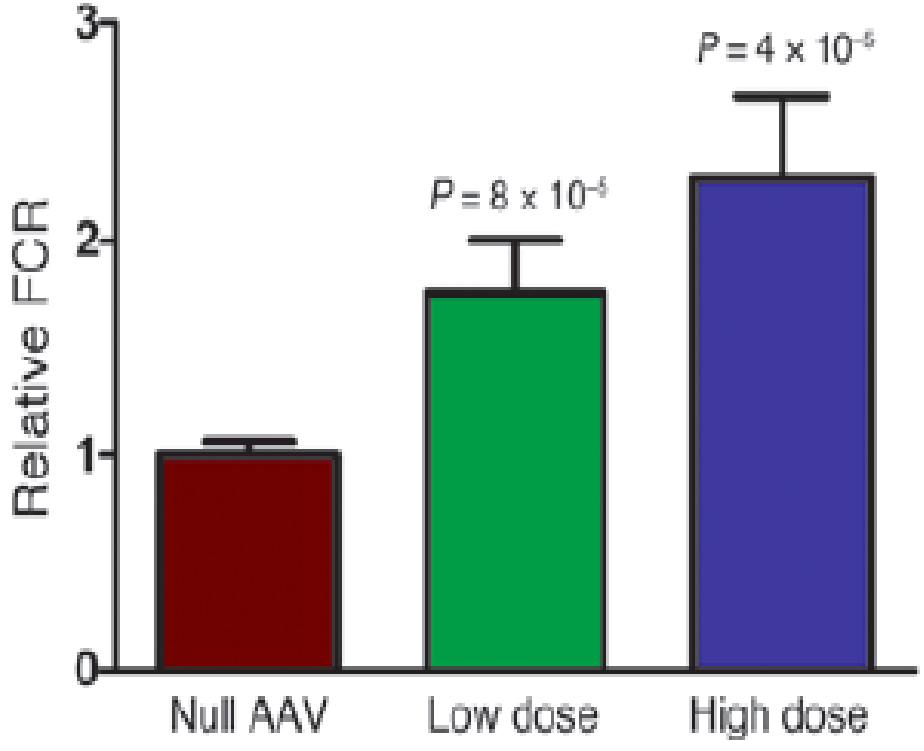




Strong et al., J Clin Invest 122:2807, 2012

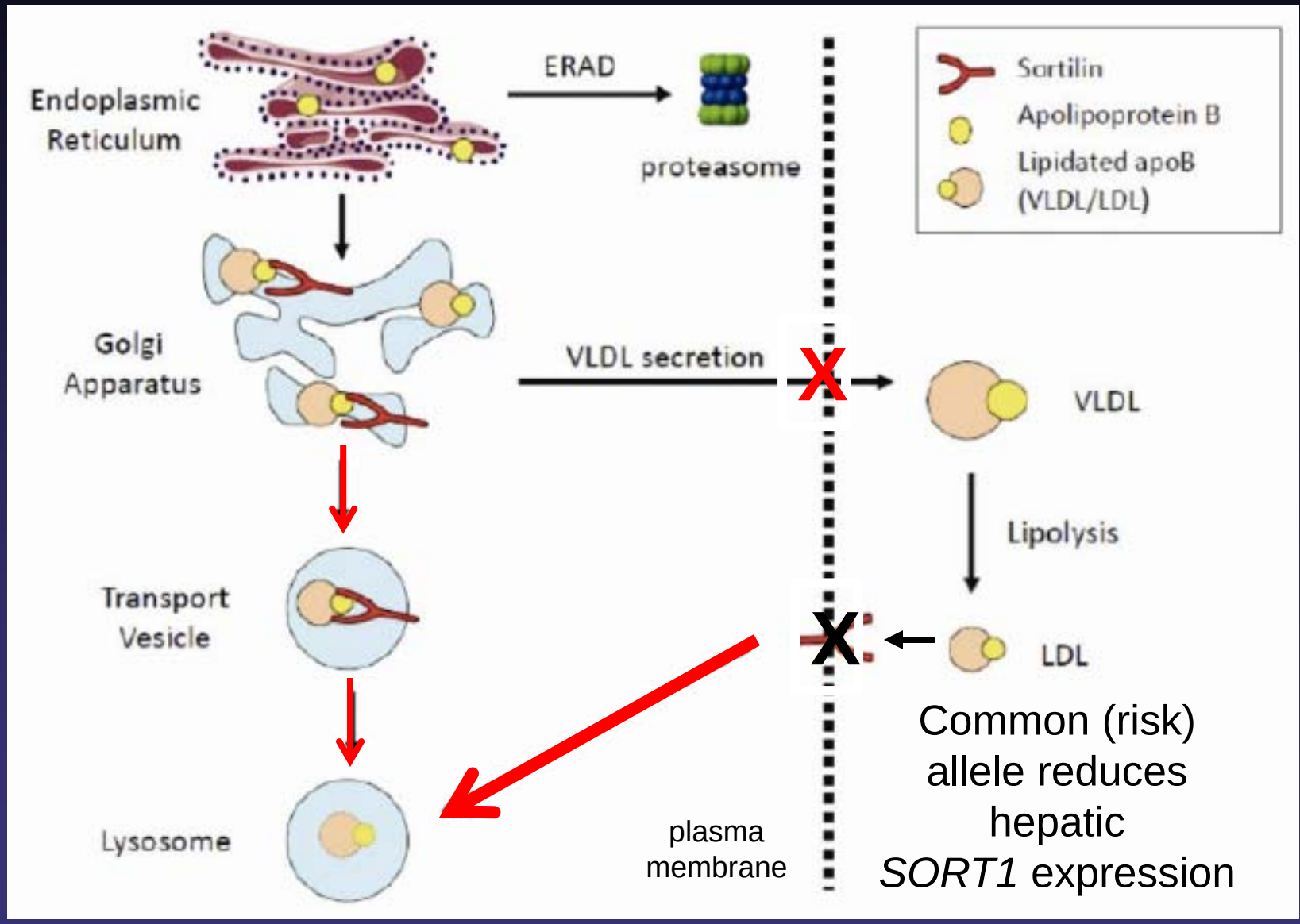
Sortilin expression promotes LDL uptake in mice *in vivo*





Strong et al., J Clin Invest 122:2807, 2012

Summary: Sortilin, the product of *SORT1*, reduces hepatic apoB secretion and increases hepatic LDL uptake



Strong et al., JCI 122:2807, 2012

And - SORT1, which can increase LDL uptake via sortilin, may reduce LDLR- mediated LDL uptake by increasing PCSK9 secretion

Cell Metabolism

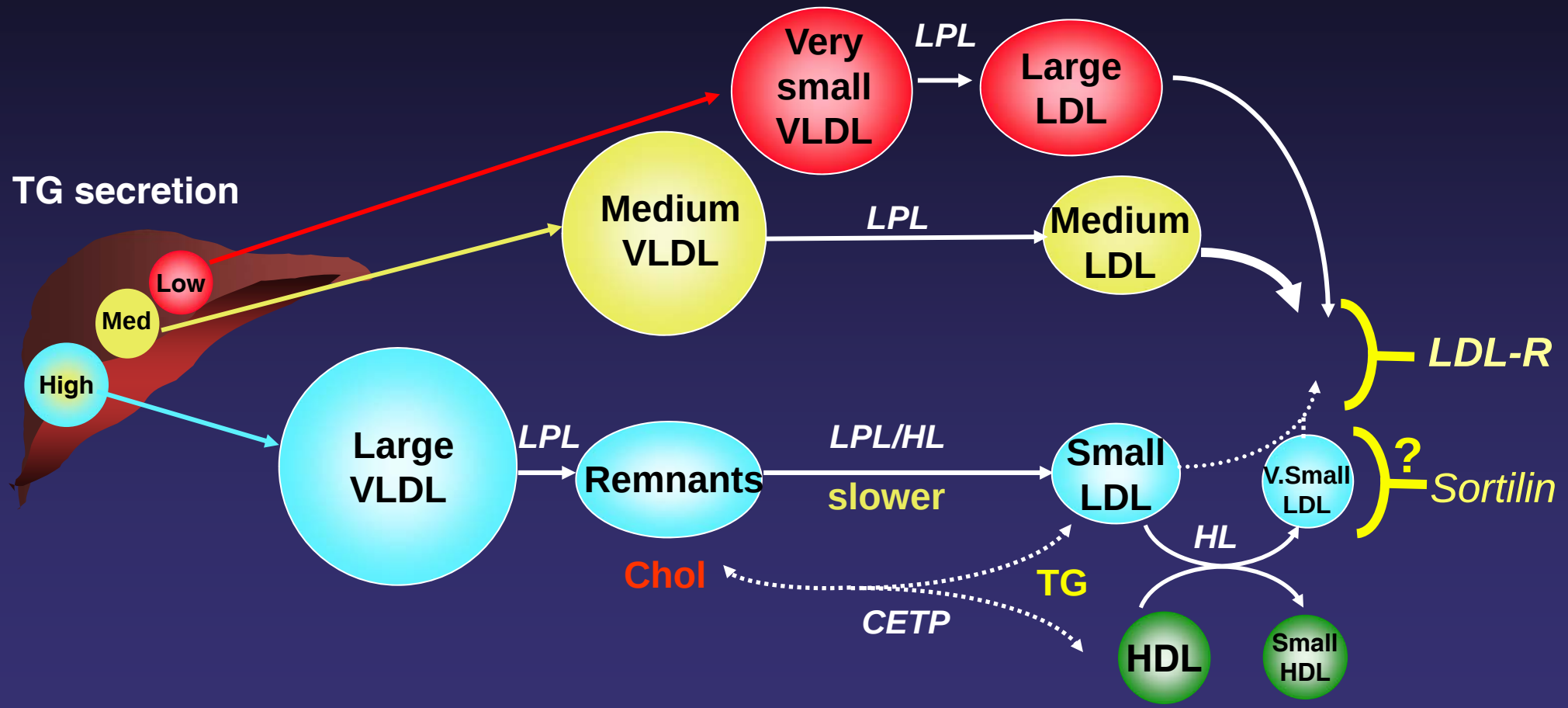
Volume 19, Issue 2, 4 February 2014, Pages 310–318

Short Article

The Hypercholesterolemia-Risk Gene *SORT1* Facilitates PCSK9 Secretion

Camilla Gustafsen[1], Mads Kjolby[1, 2, 5], Mette Nyegaard[3], Manuel Mattheisen[4], Jesper Lundhede[1], Henriette Buttenschøn[3, 6], Ole Mors[6, 7], Jacob F. Bentzon[5], Peder Madsen[1], Anders Nykjaer[1, 2], Simon Glerup[1, 2],

Two receptor clearance mechanisms for LDL particles



Adapted from Berneis and Krauss, JLR 43:1155, 2002

Mendelian randomization indicates that LDL-C and TG, but not HDL-C are causative biomarkers for CAD

Predictor	Covariate	β	s.e.m.	P
LDL-C	–	0.41	0.039	4×10^{-20}
	$\beta_{\text{HDL-C}}$	0.38	0.039	9×10^{-19}
	$\beta_{\text{triglycerides}}$	0.40	0.034	1×10^{-23}
	$\beta_{\text{HDL-C}}$, $\beta_{\text{triglycerides}}$	0.38	0.034	2×10^{-22}
HDL-C	–	–0.18	0.052	0.0006
	$\beta_{\text{LDL-C}}$	–0.12	0.041	0.005
	$\beta_{\text{triglycerides}}$	–0.09	0.048	0.057
	$\beta_{\text{LDL-C}}$, $\beta_{\text{triglycerides}}$	–0.04	0.037	0.35
TG	–	0.44	0.074	2×10^{-8}
	$\beta_{\text{LDL-C}}$	0.42	0.057	5×10^{-12}
	$\beta_{\text{HDL-C}}$	0.36	0.074	3×10^{-6}
	$\beta_{\text{LDL-C}}$, $\beta_{\text{HDL-C}}$	0.36	0.057	1×10^{-9}

Do et al., Nat Genet 45:1345, 2013

Correlations between standard lipid measures and LDL particle fractions in subjects with insulin resistance (n=155)

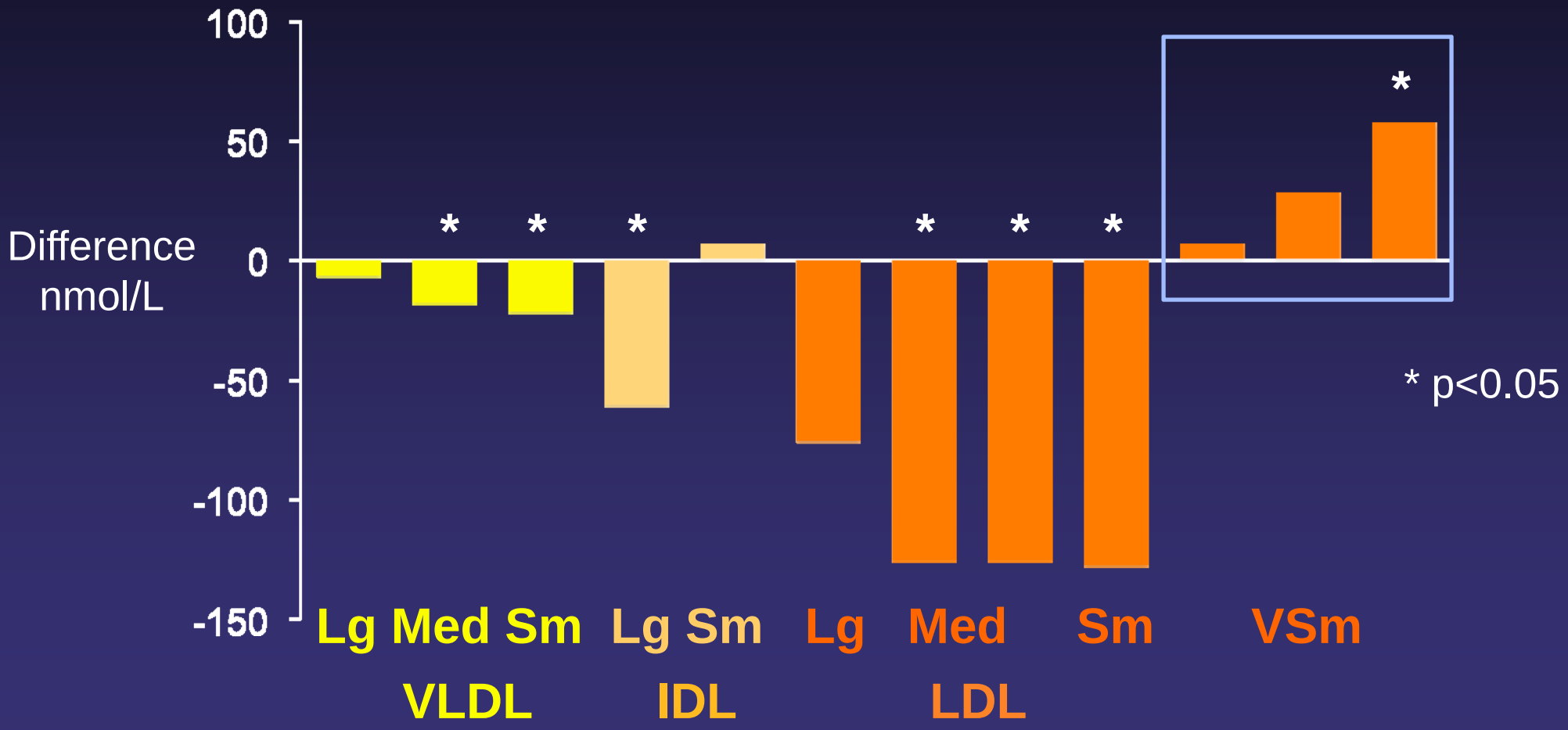
LDL Particles	LDL-C	TG	ApoB	Non-HDL-C
Total	0.73	0.31	0.73	0.77
Large	0.51	-0.33	0.12	0.29
Medium	0.64	0.20	0.61	0.65
Small	0.44	0.49	0.67	0.61
Very Small	0.19	0.65	0.55	0.45

A common TG-raising APOA5 allele is associated with increase in very small LDL 4

	Healthy men with *APOA5* haplotypes: **1/1* (n=300)	**1/3* (n=49)	p
Trig	97.8±61.5	130.9±73.7	0.0005
HDL-Chol	49.9±12.3	47.5±12.7	ns
LDL-Chol	126.4±33.4	138.4±33.5	0.020
LDL 1	118.9±40.9	117.0±51.5	ns
LDL 2	103.0±37.5	109.5±34.9	ns
LDL 3	46.0±31.8	59.1±44.4	0.012
LDL 4	9.5±9.0	15.7±17.8	0.0002

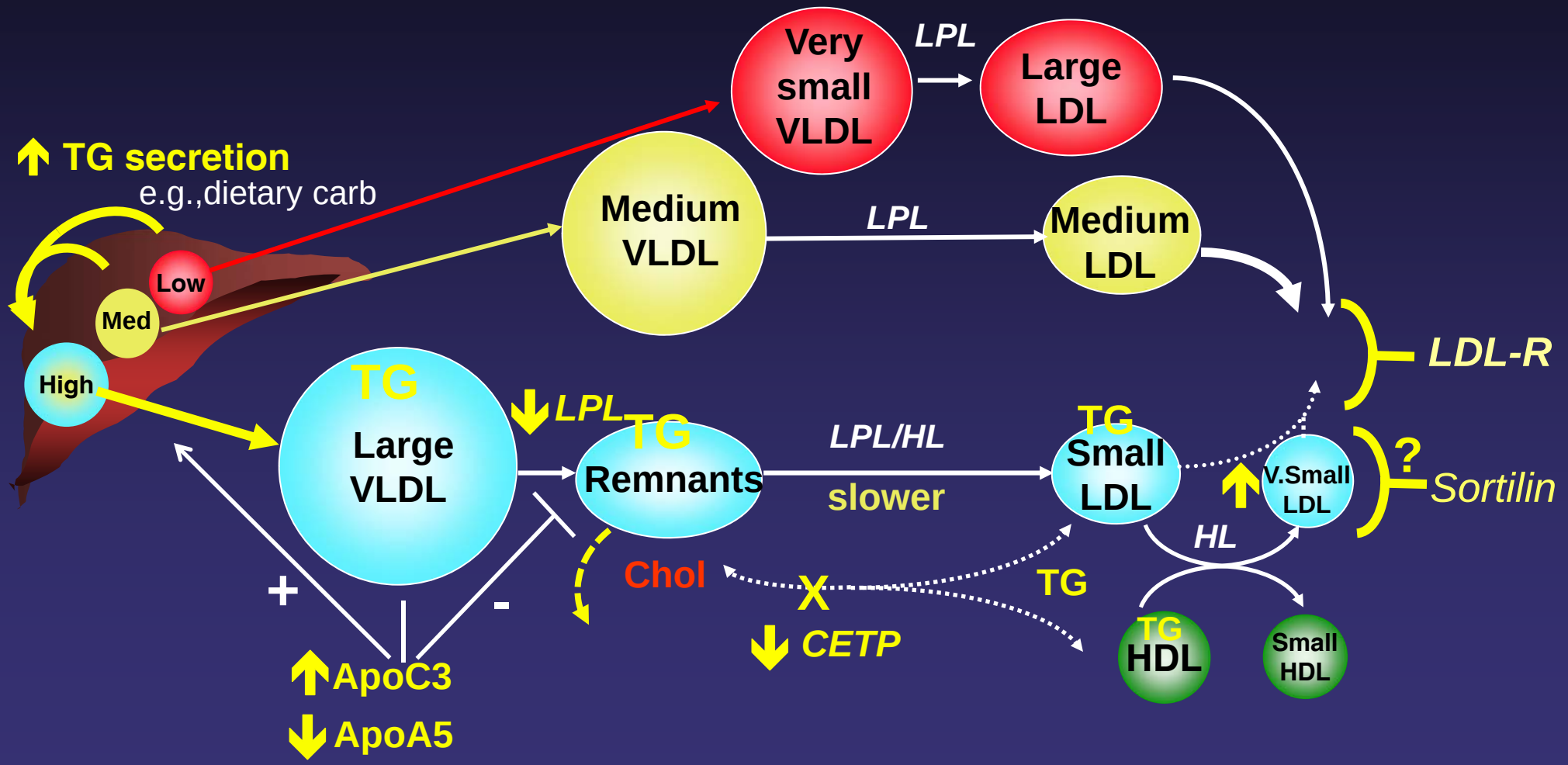
Krauss, ATVB 25:2265, 2005

CETP inhibition (anacetrapib 150 mg/d vs. placebo) which causes TG-enrichment of apoB-containing particles, increases very small LDL



Krauss et al., J. Lipid Res 53: 540, 2012

Multiple mechanisms for TG enrichment of precursors increase production of small and very small LDL particles



Adapted from Berneis and Krauss, JLR 43:1155, 2002

Conclusions
Sortilin – a new target for reduction of atherosclerosis risk?

- Very small LDL particles, which are strongly related to CHD risk, are produced from TG-rich precursors, are cleared poorly by LDL receptors, and are relatively resistant to statin lowering.
- Sortilin genetic variation is strongly related to both levels of very small LDL and CHD risk.
- Sortilin binds apoB and acts both to reduce hepatic apoB secretion by lysosomal catabolism, and to increase LDL clearance by acting as a cell surface receptor.
- Regulation of sortilin is as yet poorly understood, but may offer a new therapeutic approach to lower CHD risk.