SUPPLEMENTAL MATERIAL

Supplementary Methods

In order to maintain the case-control (matched) study design and to avoid loss of sample size, we conducted subgroup analysis by adding appropriate interaction terms to the models. Subgroup-specific linear regression parameters and odds ratios were calculated using combinations of main effect and interaction terms. Inclusion of interaction terms in conditional logistic regression models allowed us to calculate subgroup-specific odds ratios. Because cases and controls were not matched on treatment allocation to rosuvastatin versus placebo, the main effect for treatment along with interaction term was included. For example, in order to calculate the effect of the efflux capacity among participants on-statin, we ran the following regression model:

$\text{Logit(p)} = \beta_0 + \beta_1(efflux) + \beta_2(trt01) + \beta_3(efflux * trt01)$, where efflux is efflux capacity and trt01 is a dichotomous treatment variable.

Then among treated participants effect of efflux is calculated as: $(\beta_1 + \beta_3)efflux$, while among non-treated participants effect of efflux is calculated as: $(\beta_1)efflux$.
Odds ratios are then $OR_{efflux|trt=1} = exp(\beta_1 + \beta_3)$ and $OR_{efflux|trt=0} = exp(\beta_1)$

The standard error of $\beta_1 + \beta_3$ is computed from the variance-covariance matrix of the estimated regression coefficients as follows:

$$se(\beta_1 + \beta_3) = \sqrt{var(\beta_1) + var(\beta_3) + 2 \cdot cov(\beta_1 \beta_3)}$$

$$= \sqrt{[\,1 \;\; 1\,] \times \begin{bmatrix} var(\beta_1) & cov(\beta_1 \beta_3) \\ cov(\beta_3 \beta_1) & var(\beta_3) \end{bmatrix} \times \begin{bmatrix} 1 \\ 1 \end{bmatrix}}$$

The confidence interval for $OR_{efflux|trt=1}$ was calculated based on this standard error estimate.

Supplementary Table 1: Number of each component of the composite endpoint among cases.

	JUPITER Trial Primary Endpoint	Primary Endpoint + All-Cause Mortality	Sensitivity Analysis MI, Stroke, CVD Death
N Current Study / N in Overall JUPITER Trial (%)	314/393 (80%)	525/736 (71%)	182/240 (76%)
Myocardial Infarction	87	87	87
Stroke	72	72	72
Unstable Angina Hospitalization	34	34	
Arterial Revascularization	180	180	
Cardiovascular Death	60	60	55
Death from Any Cause		271	

Due the possibility of multiple events in a given individual, the sum of composite endpoints may exceed the total number of cases. MI – myocardial infarction. CVD – cardiovascular disease.

Supplementary Table 2. Spearman Correlation Coefficients of Baseline Cholesterol Efflux Capacity with Clinical Variables and Biomarkers.

	N	Correlation Coefficient	P-Value
Age	1050	0.09	0.002
BMI	1043	-0.1	0.001
Systolic Blood Pressure	1048	0.06	0.051
hsCRP (log)	1050	-0.09	0.003
Phospholipase A2 Activity	798	-0.21	< 0.0001
Non-HDL Cholesterol	1050	0.19	< 0.0001
LDL Cholesterol	1050	0.15	< 0.0001
HDL Cholesterol	1050	0.39	< 0.0001
Triglycerides (log)	1050	0.13	< 0.0001
Apolipoprotein B	1046	0.18	< 0.0001
Apolipoprotein A-I	1046	0.48	< 0.0001
HDL Particle Number	996	0.39	< 0.0001
LDL Particle Number	996	0.10	0.002
Glucose	1049	-0.01	0.67
Glycated hemoglobin	1040	-0.02	0.44

Supplementary Table 3. Association between Baseline Cholesterol Efflux Capacity and Incident Cardiovascular Events According to Clinical Subgroups.

	N	OR/SD Increment	95% Confidence Interval	P-Value	P-Value for Interaction
Sex					
Male	456	0.96	0.75 - 1.21	0.70	0.23
Female	172	0.71	0.47 - 1.09	0.12	
Treatment Group					
Placebo	379	0.88	0.68 - 1.14	0.34	0.89
Rosuvastatin	249	0.9	0.68 - 1.21	0.49	
Race					
White	564	0.86	0.7 - 1.07	0.18	0.39
Non-white	64	1.08	0.64 - 1.83	0.76	
Current Smoker					
No	532	0.91	0.73 - 1.13	0.4	0.57
Yes	96	0.79	0.5 - 1.25	0.32	
FH of Premature CHD					
No	534	0.86	0.69 - 1.09	0.22	0.54
Yes	94	1.01	0.64 - 1.59	0.97	
Metabolic Syndrome					
No	372	0.9	0.69 - 1.18	0.45	0.88
Yes	252	0.88	0.65 - 1.19	0.4	
hs-CRP					
< Median	311	0.92	0.69 - 1.23	0.57	0.76
≥ Median	317	0.87	0.67 - 1.13	0.3	
Non-HDL Cholesterol					
< Median	313	0.89	0.68 - 1.16	0.38	0.98
≥ Median	315	0.89	0.67 - 1.19	0.44	
LDL Cholesterol					
< Median	310	0.89	0.69 - 1.15	0.38	0.96
≥ Median	318	0.89	0.67 - 1.17	0.39	
Apolipoprotein B					
< Median	307	0.91	0.69 - 1.18	0.47	0.83
≥ Median	318	0.94	0.7 - 1.28	0.72	
LDL Particle Number					
< Median	295	0.99	0.75 - 1.3	0.93	0.59
≥ Median	296	0.89	0.65 - 1.21	0.46	
HDL Cholesterol					
< Median	306	0.85	0.63 - 1.14	0.28	0.59
≥ Median	322	0.94	0.71 - 1.24	0.65	
Apolipoprotein A-I					
< Median	310	0.98	0.72 - 1.33	0.89	0.83

≥ Median	315	1.02	0.76 - 1.36	0.90	
HDL Particle Number					
< Median	294	1.16	0.81 – 1.66	0.41	0.57
≥ Median	297	1.02	0.75 – 1.38	0.91	

Odds ratios were adjusted for age, race, treatment group, smoking status, systolic blood pressure, body-mass index, fasting glucose, low-density lipoprotein cholesterol level, log-transformed triglyceride level, and family history of premature coronary artery disease. The stratification variable (e.g. smoking status for current smokers versus nonsmokers) was excluded from list of covariate adjustments for each respective analysis. OR – Odds Ratio.

Supplementary Table 4. Efficacy of Rosuvastatin Therapy According to Baseline Cholesterol Efflux Capacity.

	N (N Rosuvastatin / N Placebo)	Efficacy of Rosuvastatin (OR)	95% Confidence Interval	P-Value	P-Value for Interaction
Cholesterol Efflux Capacity					
Tertile 1	210 (74 / 136)	0.60	0.31 - 1.16	0.13	
Tertile 2	209 (82 / 127)	0.72	0.40 - 1.30	0.27	0.93
Tertile 3	209 (93 / 116)	0.67	0.30 - 1.23	0.20	

Odds ratios for rosuvastatin (versus placebo) in predicting the JUPITER trial primary endpoint were adjusted for age, race, treatment group, smoking status, systolic blood pressure, body-mass index, fasting glucose, low-density lipoprotein cholesterol level, log-transformed triglyceride level, and family history of premature coronary artery disease in conditional logistic regression model. Case control pairs were matched on sex, age at baseline and time of the event. OR – Odds Ratio.

Supplementary Table 5: Spearman Correlations between Change in HDL-related Biomarkers

	Δ HDL Cholesterol	Δ Apolipoprotein A-I	Δ HDL Particle Number	Δ Cholesterol Efflux Capacity
Δ HDL Cholesterol	--			
Δ Apolipoprotein A-I	0.46 P < 0.001 N=840	--		
Δ HDL Particle Number	0.36 P < 0.001 N=739	0.51 P < 0.001 N=733	--	
Δ Cholesterol Efflux Capacity	0.26 P < 0.001 N=614	0.36 P < 0.001 N=608	0.20 P < 0.001 N=557	--

Supplementary Table 6: Association between 12-Month Change in HDL-related Biomarkers and Risk of Incident Cardiovascular Events.

	N (N Cases / N Controls)	Magnitude of 10% Change	Odds Ratio per 10% Change (95%CI), P Value
Δ HDL Cholesterol	458 (229/229)	5.1 mg/dl	0.91 (0.8 - 1.04) P=0.17
Δ Apolipoprotein A-I	452 (226/226)	16.1 mg/dl	0.90 (0.77 - 1.06) P=0.2
Δ HDL Particle Number	396 (198/198)	3.2 µmol/l	0.97 (0.84 - 1.12) P=0.64
Δ Cholesterol Efflux Capacity	462 (231/231)	1.6%	0.96 (0.88 - 1.04) P=0.33

Odds ratios for the JUPITER trial primary endpoint were assessed per standard deviation increment change and adjusted for age, race, treatment group, smoking status, systolic blood pressure, body-mass index, fasting glucose, low-density lipoprotein cholesterol level, log-transformed triglyceride level, and family history of premature coronary artery disease

Supplementary Table 7. Baseline Characteristics in On-statin Analysis of Cholesterol Efflux Capacity and Incident Cardiovascular Disease

	Primary Endpoint Controls (N=98)	Primary Endpoint Cases (N=80)	Primary Endpoint + Total Mortality Controls (N=136)	Primary Endpoint + Total Mortality Cases N=112)
Age, years	70 (64-74)	70 (65-76)	70 (64-74.2)	70 (65-76)
Female Sex	23 (23%)	20 (25%)	33 (24%)	31 (28%)
White Race	94 (96%)	73 (91%)	128 (94%)	100 (89%)
Body-Mass Index, kg/m^2	29 (26-31)	27.8 (25-31)	29.3 (26-32)	27 (24-31)*
Systolic Blood Pressure, mm Hg	137 (124-144)	134 (124-145)	136 (126-144)	132 (125-144)
Current smoker	13 (13%)	19 (24%)	19 (14%)	28 (25%)*
FH of Premature CHD[†]	14 (14%)	10 (12%)	21 (15%)	14 (12%)
Metabolic syndrome	40 (42%)	32 (41%)	54 (40%)	40 (36%)
On-Statin hsCRP, mg/l	4 (3-6)	5 (3-9)*	4 (3-7)	5 (3-9)*
Lp-PLA2 Activity, nmol/min/mL	197 (170-224)	192 (163-229)	199 (172-226)	194 (160-225)
Fasting Glucose, mg/dl	95 (90-100)	94.5 (90-103)	95 (90-101)	95 (90-103)
On-Statin Lipids, mg/dL				
LDL Cholesterol	53 (42-67)	61 (44-81)	52 (41-66)	59 (43-84)*
HDL Cholesterol	56 (43-66)	50 (43-60)	54 (49-66)	50 (43-60)
Triglycerides	96 (75-141)	99.5 (73-127)	94 (70-139)	104 (76-134)
Apolipoproteins				
Apolipoprotein B, mg/dl	65 (58-74)	74 (58-87)*	65 (56-74)	73.5 (58-88)*
Apolipoprotein A-I, mg/dl	166 (147-196)	159 (145-182)	165 (144-193)	159 (143-177)
HDL Particle Number, µmol/l	34 (31-39)	31 (28-35)*	34 (31-39)	30 (27-35)*
Cholesterol Efflux Capacity, %	14 (12-17)	13 (11-16)*	13 (11-17)	13 (11-16)

Values represent n(%) or median (25-75%). FH – Family History - CHD – Coronary Heart Disease - hsCRP – high-sensitivity C-Reactive Protein - Lp-PLA2 – lipoprotein-associated phospholipase A_2 - LDL – low-density lipoprotein - HDL – high-density lipoprotein.

* indicates Chi-square or Wilcoxon rank-sum test p-value < 0.05

[†] Family history of premature coronary disease was defined as diagnosis of the disease in a male first-degree relative before the age of 55 y or in a female first-degree relative before the age of 65 y.

Supplementary Table 8. Association between On-Statin Cholesterol Efflux Capacity and Incident Cardiovascular Events According to Clinical Subgroups

	N	OR/SD Increment	95% Confidence Interval	P-Value	P-Value for Interaction
Sex					
Male	135	0.66	0.42 - 1.05	0.08	0.24
Female	43	0.37	0.16 - 0.86	0.02	
Current Smoker					
No	146	0.6	0.39 - 0.93	0.02	0.77
Yes	32	0.69	0.32 - 1.48	0.34	
FH of Premature CHD					
No	154	0.64	0.42 - 0.98	0.04	0.9
Yes	24	0.6	0.23 - 1.56	0.3	
Metabolic Syndrome					
No	103	0.56	0.34 - 0.91	0.02	0.53
Yes	72	0.71	0.39 - 1.28	0.25	
hs-CRP					
< Median	89	0.56	0.33 - 0.96	0.03	0.61
≥ Median	89	0.67	0.4 - 1.12	0.13	
Non-HDL Cholesterol					
< Median	88	0.59	0.34 - 1.03	0.06	0.81
≥ Median	90	0.55	0.32 - 0.93	0.02	
LDL Cholesterol*					
< Median	155	0.56	0.37 - 0.85	0.01	0.92
≥ Median	22	0.6	0.17 – 2.06	0.41	
HDL Cholesterol					
< Median	88	0.53	0.28 - 0.98	0.04	0.53
≥ Median	90	0.67	0.4 - 1.12	0.13	
Apolipoprotein B#					
< Median	151	0.66	0.44 - 0.98	0.04	0.88
≥ Median	24	0.6	0.19 - 1.88	0.38	
Apolipoprotein A-I					
< Median	89	0.65	0.36 - 1.18	0.16	0.78
≥ Median	89	0.58	0.34 – 1.00	0.05	

LDL Particle Number					
< Median	79	0.51	0.28 - 0.92	0.03	0.6
≥ Median	80	0.62	0.36 - 1.08	0.09	
HDL Particle Number					
< Median	79	0.64	0.32 - 1.27	0.2	0.64
≥ Median	80	0.79	0.44 - 1.41	0.43	

Odds ratios for the JUPITER trial primary endpoint were adjusted for age, race, smoking status, systolic blood pressure, body-mass index, fasting glucose, on-treatment low-density lipoprotein cholesterol levels, on-treatment log-transformed triglyceride levels, and family history of premature coronary artery disease. OR – Odds Ratio.

* To avoid multicollinearity, threshold was calculated using both cases and controls.

Supplementary Table 9. Associations between Baseline HDL-Related Phenotypes and Incident Cardiovascular Events

	Tertile One	Tertile Two	Tertile Three	P-Trend
HDL Cholesterol				
Range, mg/dl	23-43	44-57	58-98	
N Cases / N Controls	121/113	113/96	76/104	
Adjusted OR (95%CI)	Reference	1.27 (0.83-1.95)	0.9 (0.53-1.50)	0.80
P-Value	--	0.26	0.68	
Apolipoprotein A-I				
Range, mg/dl	89-146	147-175	176-261	
N Cases / N Controls	114/101	124/103	72/107	
Adjusted OR (95%CI)	Reference	0.94 (0.61-1.44)	0.58 (0.34-1.0)	0.06
P-Value	--	0.77	0.05	
HDL Particle Number				
Range, µmol/l	17-28.6	28.7-34.2	34.3-53.2	
N Cases / N Controls	110/84	104/94	89/109	
Adjusted OR (95%CI)	Reference	0.87 (0.55-1.38)	0.59 (0.36-0.97)	0.03
P-Value	--	0.54	0.04	

Odds ratio adjusted for age, , race, treatment group, smoking status, systolic blood pressure, body-mass index, fasting glucose, low- density lipoprotein cholesterol level, log-transformed triglyceride level, and family history of premature coronary artery disease. OR – Odds Ratio. P-trend – P-value for linear trend across tertiles.

Supplementary Table 10. Associations between On-statin HDL-Related Phenotypes and Incident Cardiovascular Events

	Tertile One	Tertile Two	Tertile Three	P-Trend
HDL Cholesterol				
Range, mg/dl	22-44	45-61	62-118	
N Cases / N Controls	114/99	87/106	47/99	
Adjusted OR (95%CI)	Reference	0.92 (0.42-2.02)	0.37 (0.16-0.86)	<0.0001
P-Value	--	0.84	0.02	
Apolipoprotein A-I				
Range, mg/dl	97-150	151-179	180-288	
N Cases / N Controls	106/108	85/90	52/106	
Adjusted OR (95%CI)	Reference	1.38 (0.63-3.05)	0.74 (0.24-2.26)	0.07
P-Value	--	0.42	0.60	
HDL Particle Number				
Range, µmol/l	20.8-30.3	30.4-35.7	35.8-53.1	
N Cases / N Controls	108/87	68/94	44/89	
Adjusted OR (95%CI)	Reference	0.27 (0.11-0.70)	0.17 (0.07-0.44)	0.41
P-Value	--	0.01	<0.0001	

Odds ratio adjusted for the JUPITER trial primary endpoint adjusted for age, race, treatment group, smoking status, systolic blood pressure, body-mass index, fasting glucose, on-statin low-density lipoprotein cholesterol level, on-statin log-transformed triglyceride level, and family history of premature coronary artery disease. OR – Odds Ratio. P-trend – P-value for linear trend across tertiles.