

# **Supplemental Material**

**Table S1. Plasma concentrations and correlations of sphingolipid species.**

Specie	Concentration ( $\mu\text{g/mL}$ )					Spearman correlation							
	Mean	SD	10th	90th	Fold difference between 90th and 10th percentiles	Cer-16	Cer-20	Cer-22	Cer-24	SM-16	SM-20	SM-22	SM-24
			percentile	percentile									
Cer-16	0.27	0.06	0.20	0.34	1.7	1.00							
Cer-20	0.08	0.03	0.05	0.12	2.3	0.52	1.00						
Cer-22	0.63	0.19	0.41	0.86	2.1	0.60	0.60	1.00					
Cer-24	4.50	1.08	3.25	5.89	1.8	0.59	0.49	0.87	1.00				
SM-16	125.30	19.37	102.37	150.57	1.5	0.45	0.32	0.23	0.29	1.00			
SM-20	17.86	3.57	13.54	22.41	1.7	0.30	0.36	0.46	0.47	0.49	1.00		
SM-22	26.86	5.88	19.78	34.13	1.7	0.34	0.30	0.56	0.52	0.58	0.78	1.00	
SM-24	14.41	3.54	10.31	18.86	1.8	0.29	0.10	0.39	0.48	0.54	0.66	0.90	1.00

Cer denotes ceramide; SM, sphingomyelin

**Table S2. Risk of incident atrial fibrillation per two-fold higher sphingolipid level.**

	Model 1			Model 2			Model 3		
	HR	95% CI	p	HR	95% CI	p	HR	95% CI	p
Cer-16	1.04	(0.86, 1.25)	0.6762	1.01	(0.83, 1.23)	0.8880	1.31	(1.03, 1.66)	0.0274
Cer-20	0.82	(0.72, 0.93)	0.0015	0.78	(0.68, 0.90)	0.0004	0.72	(0.61, 0.84)	<0.0001
Cer-22	0.85	(0.74, 0.99)	0.0312	0.80	(0.68, 0.94)	0.0058	0.71	(0.59, 0.86)	0.0004
Cer-24	0.81	(0.68, 0.96)	0.0173	0.77	(0.64, 0.92)	0.0052	0.67	(0.54, 0.83)	0.0003
SM-16	1.00	(0.76, 1.32)	0.9999	1.12	(0.82, 1.55)	0.4724	1.73	(1.18, 2.55)	0.0051
SM-20	0.62	(0.50, 0.76)	<0.0001	0.64	(0.51, 0.81)	0.0002	0.55	(0.43, 0.71)	<0.0001
SM-22	0.73	(0.60, 0.88)	0.0010	0.74	(0.60, 0.91)	0.0049	0.60	(0.46, 0.77)	0.0001
SM-24	0.80	(0.68, 0.95)	0.0118	0.85	(0.71, 1.02)	0.0840	0.76	(0.61, 0.94)	0.0124

Model 1 includes adjustment for age, sex, race, and study site; Model 2 includes Model 1 adjustment terms and additional adjustment for body mass index, systolic blood pressure, treated hypertension, smoking, alcohol use, high-density lipoprotein, low-density lipoprotein, PR-interval, and prevalent diabetes, heart failure, and history of myocardial infarction; in model 3, Cer-16 and SM-16 include adjustment for Cer-22 and SM-22, respectively; Cer-20, -22, -24, and SM-20, -22, and -24 include adjustment for Cer-16 and SM-16, respectively. Significance was evaluated as  $p < 0.0063$ . HR denotes hazard ratio; CI, confidence interval; Cer, ceramide; SM, sphingomyelin.

**Table S3. Risk of incident atrial fibrillation per two-fold higher sphingolipid, adjusted for additional biomarkers.**

	CRP (n=3,847)			Fibrinogen (n=3,858)			NT-proBNP (n=3,415)			Troponin T (n=3,414)		
	HR	95% CI	p	HR	95% CI	p	HR	95% CI	p	HR	95% CI	p
Cer-16	1.25	(0.98, 1.61)	0.0766	1.29	(1.00, 1.65)	0.0464	1.00	(0.77, 1.31)	0.9810	1.16	(0.89, 1.51)	0.2677
Cer-20	0.72	(0.61, 0.85)	0.0001	0.71	(0.61, 0.84)	<0.0001	0.82	(0.69, 0.97)	0.0246	0.74	(0.62, 0.88)	0.0007
Cer-22	0.69	(0.56, 0.84)	0.0002	0.69	(0.56, 0.84)	0.0002	0.86	(0.69, 1.06)	0.1520	0.70	(0.56, 0.86)	0.0006
Cer-24	0.63	(0.50, 0.79)	0.0001	0.65	(0.51, 0.81)	0.0002	0.82	(0.64, 1.05)	0.1089	0.65	(0.51, 0.83)	0.0005
SM-16	1.94	(1.30, 2.88)	0.0011	1.80	(1.21, 2.69)	0.0037	1.12	(0.72, 1.72)	0.6209	1.62	(1.05, 2.48)	0.0279
SM-20	0.56	(0.43, 0.73)	<0.0001	0.58	(0.45, 0.76)	0.0001	0.79	(0.59, 1.05)	0.1001	0.62	(0.47, 0.82)	0.0008
SM-22	0.56	(0.43, 0.73)	<0.0001	0.60	(0.46, 0.79)	0.0002	0.86	(0.65, 1.15)	0.3167	0.65	(0.49, 0.86)	0.0027
SM-24	0.71	(0.56, 0.89)	0.0030	0.75	(0.59, 0.94)	0.0115	0.96	(0.75, 1.23)	0.7473	0.78	(0.62, 1.00)	0.0471

Each line represents four separate models; one for each biomarker. Models include adjustment for age, sex, race, study site, body mass index, systolic blood pressure, treated hypertension, smoking, alcohol use, high-density lipoprotein, low-density lipoprotein, PR-interval, and prevalent diabetes, heart failure, and history of myocardial infarction; Cer-16 and SM-16 include adjustment for Cer-22 and SM-22, respectively; Cer-20, -22, -24, and SM-20, -22, and -24 include adjustment for Cer-16 and SM-16, respectively. HR denotes hazard ratio; CI, confidence interval, Cer, ceramide; SM, sphingomyelin; CRP, C-reactive protein; NT-proBNP, N-terminal prohormone of brain natriuretic peptide.

**Table S4. Beta coefficients and 95% confidence intervals for interactions terms of sphingolipids with age, sex, BMI, race, and prevalent CHD in Cox regression models of incident atrial fibrillation.**

	Age interaction			Sex interaction			BMI interaction			Race interaction			CHD interaction		
	Beta	95% CI	p	Beta	95% CI	p	Beta	95% CI	p	Beta	95% CI	p	Beta	95% CI	p
Cer-16	0.00	(-0.03, 0.03)	0.96	-0.02	(-0.39, 0.36)	0.92	0.01	(-0.04, 0.05)	0.80	0.1	(-0.51, 0.72)	0.74	-0.23	(-0.66, 0.20)	0.29
Cer-20	0.00	(-0.02, 0.03)	0.75	0.11	(-0.13, 0.36)	0.37	-0.01	(-0.04, 0.01)	0.35	0.12	(-0.23, 0.46)	0.51	0.17	(-0.11, 0.45)	0.24
Cer-22	0.01	(-0.02, 0.04)	0.40	-0.14	(-0.42, 0.14)	0.33	0.00	(-0.03, 0.04)	0.78	0.32	(-0.16, 0.79)	0.19	0.14	(-0.19, 0.46)	0.41
Cer-24	0.01	(-0.02, 0.05)	0.38	-0.10	(-0.45, 0.25)	0.58	0.02	(-0.02, 0.06)	0.36	0.59	(0.00, 1.18)	0.05	0.06	(-0.34, 0.45)	0.78
SM-16	-0.02	(-0.07, 0.03)	0.46	0.62	(0.08, 1.16)	0.03	0.06	(-0.00, 0.12)	0.06	0.71	(-0.07, 1.48)	0.07	-0.22	(-0.80, 0.37)	0.47
SM-20	0.01	(-0.03, 0.05)	0.53	0.45	(0.04, 0.86)	0.03	0.01	(-0.03, 0.06)	0.55	0.26	(-0.32, 0.83)	0.38	0.19	(-0.28, 0.65)	0.43
SM-22	0.02	(-0.02, 0.05)	0.39	0.32	(-0.05, 0.70)	0.09	0.01	(-0.02, 0.05)	0.48	0.4	(-0.11, 0.91)	0.13	-0.12	(-0.54, 0.30)	0.57
SM-24	0.01	(-0.02, 0.05)	0.41	0.20	(-0.13, 0.54)	0.24	0.01	(-0.03, 0.05)	0.58	0.3	(-0.17, 0.78)	0.22	-0.25	(-0.61, 0.12)	0.19

Models includes adjustment for age, sex, race, study site, body mass index, systolic blood pressure, treated hypertension, smoking, alcohol use, HDL, LDL, PR-interval, and prevalent diabetes, heart failure, and history of myocardial infarction; Cer-16 and SM-16 include adjustment for Cer-22 and SM-22, respectively; Cer-20, -22, -24, and SM-20, -22, and -24 include adjustment for Cer-16 and SM-16, respectively. Cer denotes ceramide; SM, sphingomyelin; BMI, body mass index; CHD, coronary heart disease; CI, confidence interval

**Table S5. Risk of incident atrial fibrillation in subset of participants with plasma phospholipid fatty acid measures at 1992-93 (n=3,230), with and without adjustment for plasma phospholipid fatty acids.**

	correlation with corresponding saturated fatty acid*	Primary model			Primary model with adjustment for corresponding phospholipid saturated fatty acid		
		HR	95% CI	p	HR	95% CI	p
Cer-16	0.00	1.47	(1.12, 1.93)	0.005	1.42	(1.09, 1.87)	0.010
Cer-20	0.29	0.67	(0.56, 0.80)	<0.001	0.68	(0.57, 0.82)	<0.001
Cer-22	0.26	0.64	(0.52, 0.80)	<0.001	0.67	(0.54, 0.83)	<0.001
Cer-24	0.21	0.58	(0.46, 0.75)	<0.001	0.60	(0.47, 0.77)	<0.001
SM-16	-0.21	1.78	(1.16, 2.72)	0.008	1.82	(1.19, 2.79)	0.006
SM-20	0.46	0.55	(0.41, 0.73)	<0.001	0.57	(0.42, 0.78)	<0.001
SM-22	0.51	0.58	(0.43, 0.77)	<0.001	0.63	(0.46, 0.85)	0.003
SM-24	0.55	0.74	(0.58, 0.94)	0.015	0.79	(0.61, 1.03)	0.088

Models includes adjustment for age, sex, race, study site, body mass index, systolic blood pressure, treated hypertension, smoking, alcohol use, high-density lipoprotein, low-density lipoprotein, PR-interval, and prevalent diabetes, heart failure, and history of myocardial infarction; Cer-16 and SM-16 include adjustment for Cer-22 and SM-22, respectively; Cer-20, -22, -24, and SM-20, -22, and -24 include adjustment for Cer-16 and SM-16, respectively. HR denotes hazard ratio; CI, confidence interval; Cer, ceramide; SM, sphingomyelin.

\* measured in total phospholipids<sup>16</sup> (for example, correlation of Cer-16 and SM-16 with phospholipid 16:0, palmitic acid)

**Table S6. C-statistics from models of incident atrial fibrillation at 10 years.**

	10 year	
	<u>C-statistic</u>	<u>% improvement</u>
Base model*	<b>0.669</b>	
Cer-16	0.672	0.44%
Cer-20	0.677	1.20%
Cer-22	0.672	0.44%
Cer-24	0.673	0.59%
SM-16	0.674	0.70%
SM-20	0.674	0.77%
SM-22	0.674	0.70%
SM-24	0.670	0.17%

\*Includes adjustment for age, sex, race, study site, body mass index, systolic blood pressure, treated hypertension, smoking, alcohol use, high-density lipoprotein, low-density lipoprotein, PR-interval, and prevalent diabetes, heart failure, and history of myocardial infarction.

Cer denotes ceramide; SM, sphingomyelin