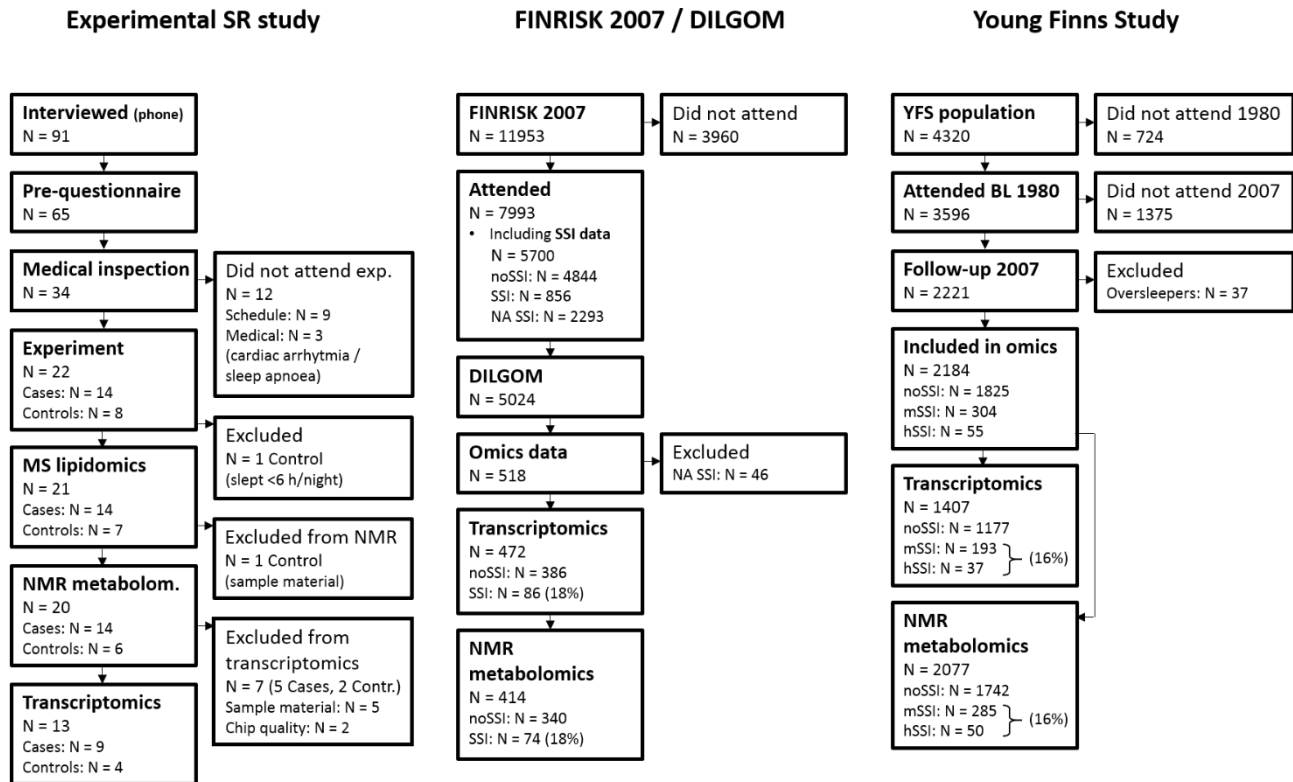


SUPPLEMENTARY INFORMATION

Prolonged sleep restriction induces changes in pathways involved in cholesterol metabolism and inflammatory responses

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Supplementary figures



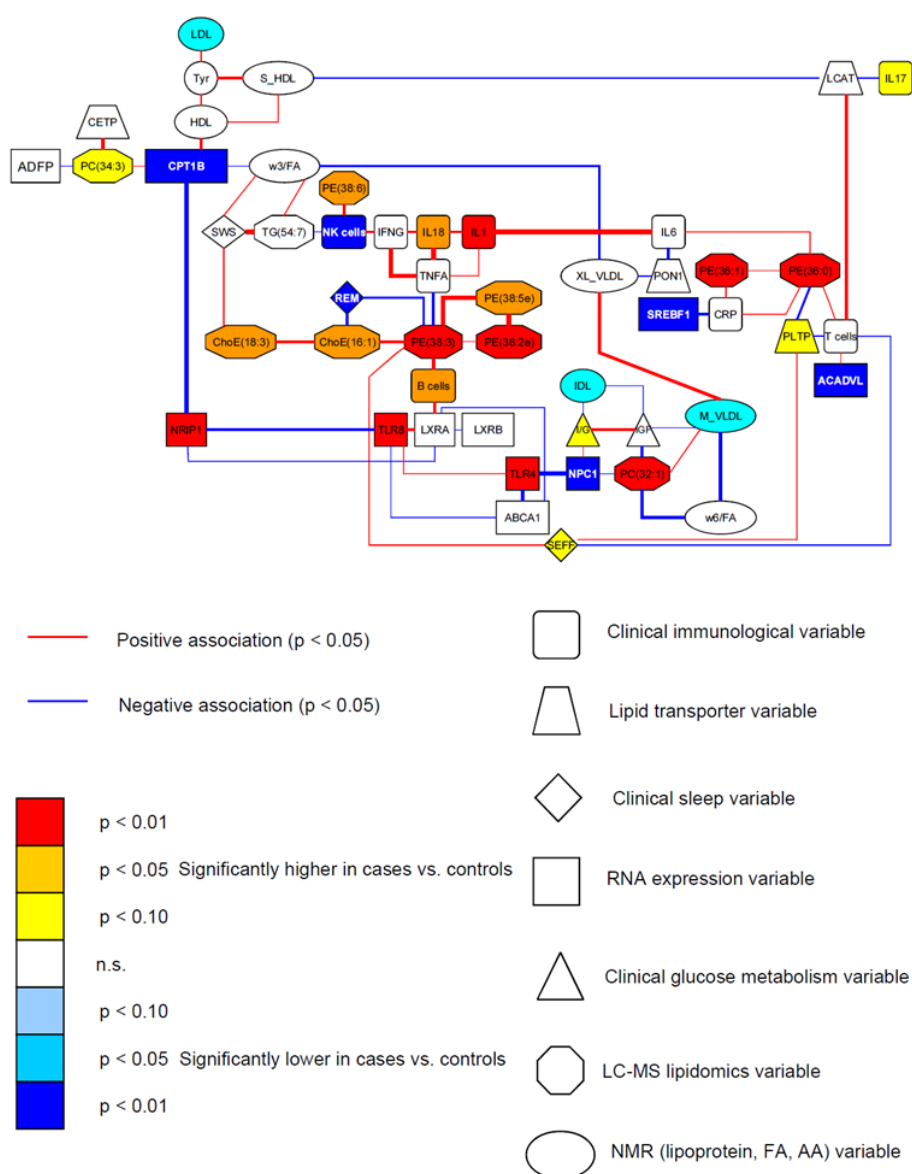
Supplementary Figure S1. Flow of the analyses.

For the experimental sleep restriction (SR) study, sleep of healthy young males was restricted to 4 h/night for 5 nights in controlled laboratory conditions and vigilance states were monitored with continuous EEG recording. In the epidemiological sample Dietary Lifestyle and Genetic determinants of Obesity and Metabolic Syndrome (DILGOM) and the replication sample Cardiovascular Risk in Young Finns Study, real-life subjective sleep insufficiency (SSI) was evaluated using questionnaires. Gene expression was analysed from peripheral blood mononuclear cells (PBMC) or whole blood using microarrays. Serum lipoprotein and lipid profiles were assessed with NMR metabolomics for all three samples and mass spectrometry (MS) for the SR study.

Pathways (N=9):	lipid homeostasis	cholesterol transport	sterol transport	sterol homeostasis	cholesterol homeostasis	lipid localization	cholesterol efflux	lipid transport	steroid metabolic process
GO pathway ID:	GO:0055088	GO:0030301	GO:0015918	GO:0055092	GO:0042632	GO:0010876	GO:0033344	GO:0006869	GO:0008202
GO P value:	8.8E-4	4.8E-2	4.8E-2	5.2E-2	5.2E-2	6.1E-2	6.5E-2	1.0E-1	2.9E-1
Genes (N=15)									
ABCG1									
NPC1									
CAV1									
NPC1L1									
GOT1									
ACACA									
ASGR2									
HEXA									
PPARA									
LPA									
PLTP									
LEPR									
UGT1A									
CYP27B1									
TSPO									

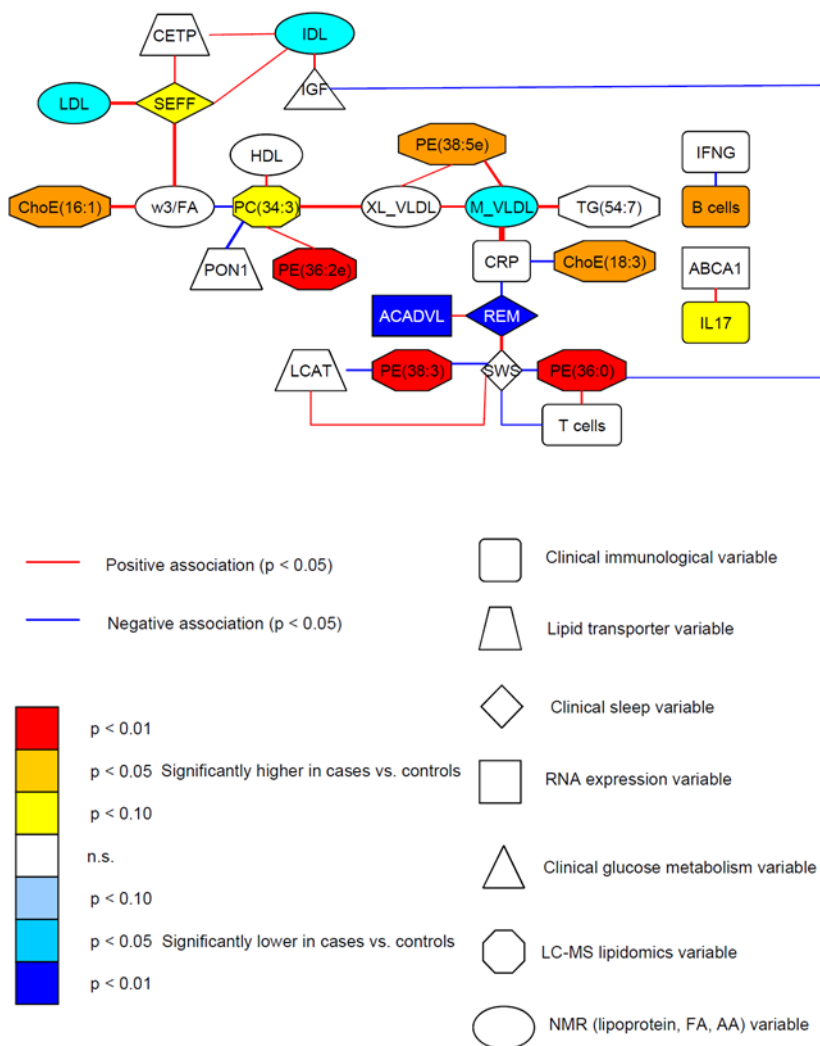
Supplementary Figure S2. "Lipid cluster" pathways and genes.

The 9 Gene Ontology (GO) Biological Processes and 15 contributing genes in the "Lipid cluster" that was found down-regulated in DILGOM subjects with subjective sleep insufficiency (SSI). (Green = the gene is part of this GO pathway.)



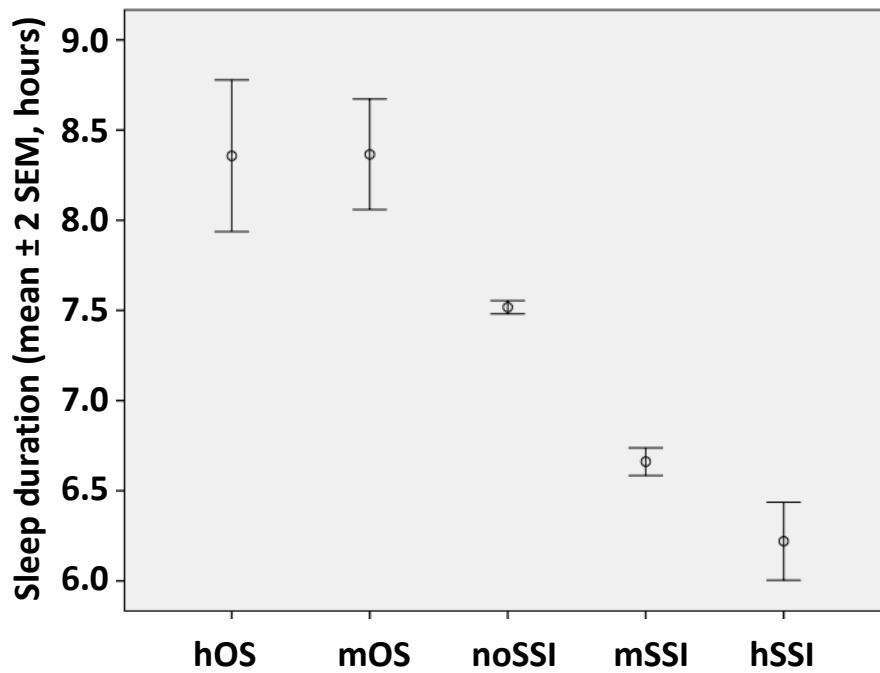
Supplementary Figure S3. Dependency network analysis using baseline-normalized variables for the experimental sleep restriction (SR).

Node shapes represent different type of variables such as clinical immunological variables, RNA expression variables, LC-MS lipidomic variables, and NMR lipoprotein variables among others (see **Supplementary Table S5**). Node colour corresponds to significance and direction of regulation comparing the experimental SR group (cases) with the control group.



Supplementary Figure S4. Dependency network analysis of the control group in the experimental sleep restriction (SR) study.

Please see **Supplementary Figure S3** and **Supplementary Table S5** for more information.



Supplementary Figure S5. Correlation of sleep insufficiency with sleep duration.

Self-reported sleep duration in subjects with different levels of subjective sleep insufficiency (SSI) or “oversleep” (OS) in the YFS sample (N=2110). Sleep duration was strongly associated to SSI ($P=4.9E-93$ and $\beta=-0.5$ h, in our linear model adjusted for sex and age, N=2077 without the OS groups). mOS = moderate oversleep (N=26), heavy oversleep (N=7), noSSI = no subjective sleep insufficiency (N=1742), mSSI = moderate SSI (N=285), hSSI = heavy SSI (N=50).

Supplementary tables

Supplementary Table S1. Characteristics of the experimental and epidemiological samples.

In the experimental sleep restriction (Exp SR) study, sleep of healthy young males (cases, N=14) was restricted to 4 h per night for 5 nights in laboratory conditions, whereas the control group (controls, N=7) had 8 hours time in bed. The epidemiological subsample of DILGOM included 18% subjects with subjective sleep insufficiency (SSI). In the Young Finns Study (YFS) replication sample, 16% had moderate (m) or heavy (h) SSI, estimated using self-reported sleep need and length.

Mean($\pm$ s.d.) values for sleep length, age, and body mass index (BMI) in each group are presented. NoSSI = subjects with no or only mild SSI. M = males, F = females.

Exp SR (N=21)	N (% M/F)	Sleep (h:min/d)	Age (years)	BMI (kg/m ²)
Controls	7 (100/0)	7:26($\pm$ 0:17)	23.0($\pm$ 3.0)	23.7($\pm$ 2.5)
Cases	14 (100/0)	3:54($\pm$ 0:05)	23.3($\pm$ 1.7)	23.5($\pm$ 2.7)
All	21 (100/0)	5:04($\pm$ 1:43)	23.2($\pm$ 2.2)	23.6($\pm$ 2.3)

DILGOM (N=472)	N (% M/F)	Sleep (h:min/d)	Age (years)	BMI (kg/m ²)
noSSI	386 (48/52)	7:44($\pm$ 1:13)	52.5($\pm$ 13.9)	26.4($\pm$ 4.9)
SSI	86 (37/63)	6:41($\pm$ 0:51)	49.6($\pm$ 12.5)	25.8($\pm$ 3.9)
All	472 (46/54)	7:31($\pm$ 1:13)	51.9($\pm$ 13.8)	26.3($\pm$ 4.7)

YFS (N=2077)	N (% M/F)	Sleep (h:min/d)	Age (years)	BMI (kg/m ²)
noSSI	1742 (54/46)	7:31($\pm$ 0:46)	37.8($\pm$ 5.0)	25.9($\pm$ 4.7)
mSSI	285 (61/39)	6:40($\pm$ 0:39)	37.0($\pm$ 4.9)	26.5($\pm$ 4.8)
hSSI	50 (58/42)	6:13($\pm$ 0:46)	38.1($\pm$ 5.1)	27.4($\pm$ 5.3)
All	2077 (55/45)	7:23($\pm$ 0:50)	37.7($\pm$ 5.0)	26.0($\pm$ 4.7)

Supplementary Table S2. Down-regulated gene clusters in epidemiological subjective sleep insufficiency (SSI).

Clusters of Gene Ontology (GO) pathways enriched among the transcripts with lower expression among DILGOM subjects with SSI ($P < 0.05$). The GO Biological Processes contributing to each cluster are listed. Genes for cluster 5 ("Lipid cluster") are shown in more detail in **Fig. S2**.

Cluster 1		Enrichment Score: 1.60
Gene Ontology annotations:	Genes N	P Value
angiogenesis	9	1.7E-2
blood vessel morphogenesis	11	1.9E-2
vasculature development	12	2.5E-2
blood vessel development	11	4.7E-2
Total	12	0.025

Cluster 2		Enrichment Score: 1.52
Gene Ontology annotations:	Genes N	P Value
regulation of response to external stimulus	9	2.5E-2
regulation of response to extracellular stimulus	3	3.3E-2
regulation of response to nutrient levels	3	3.3E-2
Total	9	0.030

Cluster 3		Enrichment Score: 1.47
Gene Ontology annotations:	Genes N	P Value
positive regulation of transcription, DNA-dependent	22	2.2E-3
positive regulation of RNA metabolic process	22	2.5E-3
positive regulation of gene expression	24	5.3E-3
positive regulation of transcription	23	7.4E-3
positive regulation of nucleobase, nucleoside, nucleotide and nucleic acid metabolic process	24	1.2E-2
positive regulation of nitrogen compound metabolic process	24	1.7E-2
positive regulation of macromolecule biosynthetic process	24	2.0E-2
positive regulation of biosynthetic process	25	2.1E-2
positive regulation of cellular biosynthetic process	24	3.2E-2
positive regulation of transcription from RNA polymerase II promoter	15	3.7E-2
positive regulation of macromolecule metabolic process	28	4.2E-2
regulation of RNA metabolic process	47	1.8E-1
regulation of transcription, DNA-dependent	45	2.3E-1
regulation of transcription from RNA polymerase II promoter	20	2.6E-1
regulation of transcription	62	3.3E-1
transcription	43	7.9E-1
Total	69	0.034

Cluster 4		Enrichment Score: 1.45
Gene Ontology annotations:	Genes N	P Value
metal ion transport	20	7.8E-3
ion transport	28	1.2E-2

cation transport	21	2.2E-2
calcium ion transport	8	3.9E-2
di-, tri-valent inorganic cation transport	8	9.7E-2
monovalent inorganic cation transport	10	2.7E-1
Total	28	0.035

Cluster 5 ("Lipid cluster")		Enrichment Score: 1.35
Gene Ontology annotations:	Genes N	P Value
lipid homeostasis	7	8.8E-4
cholesterol transport	4	4.8E-2
sterol transport	4	4.8E-2
sterol homeostasis	4	5.2E-2
cholesterol homeostasis	4	5.2E-2
lipid localization	8	6.1E-2
cholesterol efflux	3	6.5E-2
lipid transport	7	1.0E-1
steroid metabolic process	7	2.9E-1
Total	15	0.045

Supplementary Table S3. Serum metabolites in experimental sleep restriction (SR) and epidemiological subjective sleep insufficiency (SSI).

Concentrations of lipoprotein particles (VLDL, IDL, LDL, and HDL), apolipoproteins, cholesterol, fatty acids, triglycerides, phospholipids, amino acids, and small energy metabolites were measured with NMR metabolomics. Experimental SR was compared to baseline (BL) (P from paired t tests, FC = fold change). In the epidemiological sample, the concentrations were correlated to SSI adjusting for age and sex. (P values < 0.05 marked with blue. Green/Red = lower/higher expression among subjects after SR or with SSI.)

NMR metabolomics			Experimental SR			Epidemiological SSI		
Metabolite		Cluster	SR vs BL			SSI + Sex + Age		
Name	Short	(n=21)	P	P correct ed	FC	SSI P	SSI β	β Unit
Concentration of chylomicrons and extremely large VLDL particles	XXL_VLDL_P	10	0.588		1.08	0.841	1.4E-09	mmol/l
Phospholipids in chylomicrons and extremely large VLDL	XXL_VLDL_PL	-	0.346		1.36	0.772	4.9E-05	mmol/l
Triglycerides in chylomicrons and extremely large VLDL	XXL_VLDL_TG	-	0.144		1.40	0.753	3.1E-04	mmol/l
Total lipids in chylomicrons and extremely large VLDL	XXL_VLDL_L	-	0.153		1.46	0.711	5.3E-04	mmol/l
Concentration of very large VLDL particles	XL_VLDL_P	-	0.748		1.08	0.569	2.5E-08	mmol/l
Phospholipids in very large VLDL	XL_VLDL_PL	-	0.367		1.29	0.684	3.0E-04	mmol/l
Triglycerides in very large VLDL	XL_VLDL_TG	10	0.495		1.15	0.599	1.5E-03	mmol/l
Total lipids in very large VLDL	XL_VLDL_L	10	0.433		1.21	0.613	2.2E-03	mmol/l
Concentration of large VLDL particles	L_VLDL_P	11	0.640		0.94	0.477	2.0E-07	mmol/l
Total cholesterol in large VLDL	L_VLDL_C	11	0.665		0.92	0.477	2.6E-03	mmol/l
Free cholesterol in large VLDL	L_VLDL_FC	12	0.901		0.98	0.527	1.2E-03	mmol/l
Phospholipids in large VLDL	L_VLDL_PL	11	0.574		0.93	0.497	2.1E-03	mmol/l
Triglycerides in large VLDL	L_VLDL_TG	11	0.714		0.95	0.468	7.7E-03	mmol/l
Cholesterol esters in large VLDL	L_VLDL_CE	11	0.379		0.86	0.424	1.4E-03	mmol/l
Total lipids in large VLDL	L_VLDL_L	11	0.730		0.95	0.479	1.2E-02	mmol/l
Concentration of medium VLDL particles	M_VLDL_P	1	0.054		0.83	0.280	8.1E-07	mmol/l
Total cholesterol in medium VLDL	M_VLDL_C	3	0.046	0.971	0.82	0.321	6.0E-03	mmol/l
Free cholesterol in medium VLDL	M_VLDL_FC	1	0.072		0.80	0.299	3.2E-03	mmol/l
Phospholipids in medium VLDL	M_VLDL_PL	1	0.026	0.536	0.81	0.265	5.3E-03	mmol/l
Triglycerides in medium VLDL	M_VLDL_TG	1	0.064		0.83	0.281	1.6E-02	mmol/l
Cholesterol esters in medium VLDL	M_VLDL_CE	3	0.031	0.645	0.84	0.350	2.8E-03	mmol/l
Total lipids in medium VLDL	M_VLDL_L	1	0.051		0.83	0.284	2.7E-02	mmol/l
Concentration of small VLDL particles	S_VLDL_P	1	5.3E-04	0.011	0.78	0.188	1.1E-06	mmol/l

Total cholesterol in small VLDL	S_VLDL_C	3	1.9E-04	0.004	0.78	0.289	5.7E-03	mmol/l
Free cholesterol in small VLDL	S_VLDL_FC	3	4.9E-04	0.010	0.75	0.210	2.8E-03	mmol/l
Phospholipids in small VLDL	S_VLDL_PL	3	3.5E-04	0.007	0.78	0.198	4.3E-03	mmol/l
Triglycerides in small VLDL	S_VLDL_TG	1	0.001	0.029	0.79	0.179	1.1E-02	mmol/l
Total lipids in small VLDL	S_VLDL_L	3	3.7E-04	0.008	0.78	0.199	2.1E-02	mmol/l
Concentration of very small VLDL particles	XS_VLDL_P	4	0.012	0.261	0.87	0.233	7.7E-07	mmol/l
Phospholipids in very small VLDL	XS_VLDL_PL	4	0.008	0.172	0.87	0.421	2.2E-03	mmol/l
Triglycerides in very small VLDL	XS_VLDL_TG	5	9.0E-04	0.019	0.80	0.141	3.9E-03	mmol/l
Total lipids in very small VLDL	XS_VLDL_L	4	0.033	0.703	0.88	0.290	9.4E-03	mmol/l
Concentration of IDL particles	IDL_P	4	0.005	0.106	0.91	0.433	1.2E-06	mmol/l
Free cholesterol in IDL	IDL_FC	4	0.018	0.372	0.92	0.562	2.2E-03	mmol/l
Phospholipids in IDL	IDL_PL	4	0.006	0.132	0.91	0.460	3.6E-03	mmol/l
Total lipids in IDL	IDL_L	4	0.008	0.175	0.92	0.484	1.3E-02	mmol/l
Concentration of large LDL particles	L_LDL_P	6	6.6E-05	0.001	0.90	0.393	2.2E-06	mmol/l
Total cholesterol in large LDL	L_LDL_C	6	4.3E-05	8.9E-04	0.90	0.420	1.4E-02	mmol/l
Free cholesterol in large LDL	L_LDL_FC	4	3.9E-04	0.008	0.92	0.419	3.5E-03	mmol/l
Phospholipids in large LDL	L_LDL_PL	6	9.1E-04	0.019	0.92	0.348	4.5E-03	mmol/l
Cholesterol esters in large LDL	L_LDL_CE	6	3.3E-05	6.9E-04	0.89	0.409	1.1E-02	mmol/l
Total lipids in large LDL	L_LDL_L	6	4.4E-05	9.2E-04	0.90	0.402	2.0E-02	mmol/l
Concentration of medium LDL particles	M_LDL_P	7	1.2E-05	2.5E-04	0.89	0.323	2.1E-06	mmol/l
Total cholesterol in medium LDL	M_LDL_C	7	5.5E-06	1.1E-04	0.88	0.319	1.1E-02	mmol/l
Phospholipids in medium LDL	M_LDL_PL	5	1.3E-04	0.003	0.92	0.275	3.1E-03	mmol/l
Cholesterol esters in medium LDL	M_LDL_CE	7	7.2E-06	1.5E-04	0.86	0.326	8.5E-03	mmol/l
Total lipids in medium LDL	M_LDL_L	7	7.3E-06	1.5E-04	0.89	0.319	1.5E-02	mmol/l
Concentration of small LDL particles	S_LDL_P	7	3.1E-04	0.006	0.90	0.375	2.2E-06	mmol/l
Total cholesterol in small LDL	S_LDL_C	7	4.5E-04	0.009	0.88	0.344	6.5E-03	mmol/l
Total lipids in small LDL	S_LDL_L	7	0.002	0.032	0.90	0.351	9.0E-03	mmol/l
Concentration of very large HDL particles	XL_HDL_P	13	0.087		1.22	0.122	-2.2E-05	mmol/l
Total cholesterol in very large HDL	XL_HDL_C	-	0.372		1.18	0.252	-8.8E-03	mmol/l
Free cholesterol in very large HDL	XL_HDL_FC	-	0.057		1.41	0.190	-2.9E-03	mmol/l
Phospholipids in very large HDL	XL_HDL_PL	13	0.015	0.311	1.28	0.086	-1.5E-02	mmol/l
Triglycerides in very large HDL	XL_HDL_TG	5	0.483		0.97	0.481	-2.4E-04	mmol/l
Cholesterol esters in very large HDL	XL_HDL_CE	-	0.422		1.14	0.242	-6.2E-03	mmol/l

Total lipids in very large HDL	XL_HDL_L	13	0.125		1.22	0.143	-2.4E-02	mmol/l
Concentration of large HDL particles	L_HDL_P	14	0.012	0.251	1.13	0.048	-5.9E-05	mmol/l
Total cholesterol in large HDL	L_HDL_C	14	1.7E-04	0.004	1.29	0.055	-2.6E-02	mmol/l
Free cholesterol in large HDL	L_HDL_FC	13	0.004	0.087	1.22	0.092	-5.4E-03	mmol/l
Phospholipids in large HDL	L_HDL_PL	14	0.078		1.09	0.052	-2.0E-02	mmol/l
Cholesterol esters in large HDL	L_HDL_CE	14	1.4E-04	0.003	1.30	0.051	-2.0E-02	mmol/l
Total lipids in large HDL	L_HDL_L	14	0.003	0.058	1.16	0.050	-4.7E-02	mmol/l
Concentration of medium HDL particles	M_HDL_P	16	0.882		1.01	0.291	-2.5E-05	mmol/l
Total cholesterol in medium HDL	M_HDL_C	16	0.206		1.07	0.172	-9.9E-03	mmol/l
Free cholesterol in medium HDL	M_HDL_FC	16	0.940		1.00	0.319	-1.5E-03	mmol/l
Phospholipids in medium HDL	M_HDL_PL	16	0.937		1.00	0.321	-5.6E-03	mmol/l
Cholesterol esters in medium HDL	M_HDL_CE	16	0.127		1.08	0.148	-8.4E-03	mmol/l
Total lipids in medium HDL	M_HDL_L	16	0.654		1.02	0.251	-1.5E-02	mmol/l
Concentration of small HDL particles	S_HDL_P	17	0.083		0.96	0.703	1.2E-05	mmol/l
Triglycerides in small HDL	S_HDL_TG	2	0.002	0.038	0.82	0.185	1.3E-03	mmol/l
Total lipids in small HDL	S_HDL_L	17	0.044	0.930	0.94	0.646	4.3E-03	mmol/l
Serum total cholesterol	Serum_C	6	0.004	0.091	0.94	0.808	1.5E-02	mmol/l
Total cholesterol in IDL	IDL_C	4	0.044	0.922	0.94	0.549	7.0E-03	mmol/l
Total cholesterol in IDL (Lipido)	IDL_C_eFR	5	6.3E-04	0.013	0.83	0.379	5.3E-03	mmol/l
Total cholesterol in LDL	LDL_C	6	1.6E-05	3.4E-04	0.89	0.373	3.1E-02	mmol/l
Total cholesterol in LDL (Lipido)	LDL_C_eFR	4	0.003	0.053	0.93	0.759	1.3E-02	mmol/l
Total cholesterol in HDL	HDL_C	14	0.161		1.04	0.096	-4.1E-02	mmol/l
Total cholesterol in HDL2 (Lipido)	HDL2_C	14	0.078		1.06	0.098	-4.2E-02	mmol/l
Total cholesterol in HDL3 (Lipido)	HDL3_C	17	0.756		0.99	0.727	1.1E-03	mmol/l
Esterified cholesterol	EstC	6	0.013	0.270	0.94	0.822	1.0E-02	mmol/l
Free cholesterol	FreeC	7	0.003	0.068	0.92	0.878	-2.7E-03	mmol/l
Serum total triglycerides	Serum_TG	1	0.011	0.234	0.85	0.319	4.1E-02	mmol/l
Triglycerides in VLDL	VLDL_TG	1	0.044	0.933	0.84	0.278	4.0E-02	mmol/l
Triglycerides in VLDL (Lipido)	VLDL_TG_eFR	1	0.013	0.272	0.81	0.237	3.7E-02	mmol/l
Triglycerides in IDL	IDL_TG	5	0.006	0.119	0.85	0.342	2.2E-03	mmol/l
Total fatty acids	TotFA	12	0.030	0.631	0.92	0.949	-1.1E-02	mmol/l
monounsaturated fatty acids; 16:1, 18:1	MUFA	12	0.003	0.070	0.86	0.914	-7.5E-03	mmol/l
omega-3 fatty acids	FAw3	18	0.433		1.04	0.973	-3.6E-04	mmol/l

ratio of omega-3 fatty acids to total fatty acids	FAw3toFA	18	0.092		1.11	0.852	1.4E-02	
omega-6 fatty acids	FAw6	3	2.6E-04	0.005	0.89	0.960	-2.5E-03	mmol/l
ratio of omega-6 fatty acids to total fatty acids	FAw6toFA	21	0.094		0.96	0.944	1.7E-02	
omega-7, omega-9 and saturated fatty acids	FAw79S	12	0.150		0.93	0.948	-8.2E-03	mmol/l
ratio of omega-7, omega-9 and saturated fatty acids to total fatty acids	FAw79StoFA	10	0.270		1.01	0.898	-3.1E-02	
18:2, linoleic acid	LA	3	2.0E-05	4.3E-04	0.87	0.877	6.3E-03	mmol/l
other polyunsaturated fatty acids than 18:2	otPUFA	3	0.470		0.98	0.941	-3.1E-03	
22:6, docosahexaenoic acid	DHA	18	0.171		0.91	0.119	-8.5E-03	mmol/l
Mean diameter for VLDL particles	VLDL_D	10	0.349		1.01	0.932	-7.9E-03	nm
Mean diameter for LDL particles	LDL_D	15	0.535		1.00	0.955	-6.6E-04	nm
Mean diameter for HDL particles	HDL_D	14	5.6E-04	0.012	1.01	0.056	-3.1E-02	nm
Apolipoprotein A-I (Lipido)	ApoA1	17	0.974		1.00	0.252	-1.8E-02	g/l
Apolipoprotein B (Lipido)	ApoB	5	1.1E-04	0.002	0.89	0.494	9.6E-03	g/l
Apolipoprotein B by apolipoprotein A-I (Lipido)	ApoBtoApoA1	5	2.8E-04	0.006	0.89	0.236	1.0E-02	
Total phosphoglycerides	TotPG	12	0.664		0.98	0.219	-1.7E-02	mmol/l
phosphatidylcholine and other cholines	PC	12	0.532		0.98	0.183	-3.7E-02	mmol/l
sphingomyelins	SM	3	0.374		0.94	0.360	3.9E-03	mmol/l
ratio of triglycerides to phosphoglycerides	TGtoPG	2	0.011	0.230	0.86	0.504	2.1E-02	
Double bond protons of mobile lipids	MobCH	2	0.002	0.046	0.91	0.667	1.8E-02	
CH2 groups of mobile lipids	MobCH2	1	0.044	0.926	0.88	0.818	1.9E-01	
CH3 groups of mobile lipids	MobCH3	1	0.020	0.427	0.92	0.972	5.5E-03	
average number of methylene groups in a fatty acid chain	CH2inFA	10	0.538		1.00	0.319	-1.3E-02	
average number of methylene groups per a double bond	CH2toDB	10	0.229		1.02	0.386	-3.5E-02	
average number of double bonds in a fatty acid chain	DBinFA	21	0.189		0.98	0.544	3.4E-03	
ratio of bisallylic groups to double bonds	BISToDB	21	0.040	0.835	1.02	0.855	3.8E-04	
ratio of bisallylic groups to total fatty acids	BISToFA	21	0.889		1.00	0.684	2.3E-03	
description of average fatty acid chain length, not actual carbon number	FALen	18	0.389		1.00	0.937	-1.2E-03	
3-hydroxybutyrate	bOHBut	20	0.020	0.422	0.86	0.074	-1.3E-02	mmol/l
Acetate	Ace	20	0.068		0.85	0.531	-3.3E-03	mmol/l
Acetoacetate	AcAce	2	0.083		0.74	0.267	-2.4E-03	mmol/l
Citrate	Cit	15	0.354		1.09	0.228	-1.5E-03	μmol/l
Creatinine	Crea	19	0.956		1.01	0.121	2.0E-03	mmol/l
Glucose	Glc	9	0.067		0.96	0.905	6.7E-03	mmol/l

Glycoproteins	Gp	8	0.917		1.00	0.944	-1.1E-03	mmol/l
Lactate	Lac	8	0.040	0.844	1.15	0.048	-3.8E-02	mmol/l
Pyruvate	Pyr	8	0.665		1.02	0.156	-2.2E-03	mmol/l
Urea	Urea	19	0.755		1.02	0.570	1.1E-03	mmol/l
Alanine	Ala	8	0.164		1.06	0.374	-3.5E-03	mmol/l
Glutamine	Gln	15	0.417		0.98	0.916	4.7E-04	mmol/l
Histidine	His	15	0.402		1.06	0.626	-2.6E-04	mmol/l
Isoleucine	Ile	9	0.791		0.99	0.694	3.2E-04	mmol/l
Leucine	Leu	9	0.158		1.07	0.878	-1.3E-04	mmol/l
Phenylalanine	Phe	9	0.813		1.01	0.722	-2.5E-04	mmol/l
Tyrosine	Tyr	9	0.868		0.99	0.443	-5.3E-04	mmol/l
Valine	Val	9	0.885		0.99	0.878	-3.4E-04	mmol/l

Supplementary Table S4. Changes in serum lipids after experimental sleep restriction (SR).

Baseline-normalised values for phospholipids (phosphatidyl ethanolamides, PE, and phosphatidyl cholines, PC), triglycerides (TG), and cholesterol esters (ChoE) detected by mass spectrometry (MS). 10 PE, 3 PC, 2 ChoE, and one TG species increased in SR ($P < 0.05$).

MS lipid					Experimental SR		
ID	m/z	RT	Name	N	P value	Cases mean	Controls mean
1139	748.5942	375	PE(36:0)	52	0.0004	1.58	1.00
176	746.5722	351	PE(36:1)	61	0.001	1.24	0.85
78	732.5556	336	PC(32:1)	61	0.001	1.66	0.96
247	752.5552	381	PE(38:5e)	61	0.006	1.02	0.55
230	770.6054	381	PE(38:3)	61	0.006	1.00	0.72
932	730.5777	411	PE(36:2e)	42	0.008	1.02	0.65
588	640.6041	515	ChoE(16:1)	61	0.009	1.34	0.82
1206	704.5345	305	unknown	37	0.013	1.85	1.15
325	764.5381	375	PE(38:6)	60	0.015	1.02	0.91
1365	770.6094	358	PE(38:3)	41	0.021	0.97	0.74
278	664.6038	495	ChoE(18:3)	61	0.022	1.52	0.96
190	894.7589	474	TG(54:7)	61	0.024	1.23	0.81
679	846.5296	317	unknown	59	0.025	1.53	1.09
93	756.5545	317	PC(34:3)	61	0.037	1.59	1.21
1050	752.5242	279	unknown	29	0.040	1.67	1.21
53	768.5898	351	PE(38:4)	61	0.041	0.93	0.75
525	728.5599	389	PE(36:3e)	61	0.042	1.00	0.70
79	766.5728	345	PC(p16:0/20:4)	61	0.045	1.02	0.83
116	746.6041	383	PE(36:1)	61	0.048	0.88	0.74
581	857.7637	505	unknown	56	0.048	0.71	0.91

Supplementary Table S5. Variables included in the experimental sleep restriction study dependency network analysis.

Results of the analysis are depicted in **Supplementary Fig. S3 and S4.**

Method / Data set	Variable	Abbreviation in the network
Clinical Glucose Metab.	Insulin/glucose ratio	Insulin/glucose
Clinical Glucose Metab.	Insulin-like growth factor	IGF
Clinical Immunological	C-reactive protein	CRP
Clinical Immunological	T cells	T cells
Clinical Immunological	B cells	B cells
Clinical Immunological	NK cells	NK cells
Clinical Immunological	Interleukin 1	IL1
Clinical Immunological	Interleukin 6	IL6
Clinical Immunological	Interleukin 17	IL17
Clinical Immunological	Interleukin 18	IL18
Clinical Immunological	Tumor necrosis factor alpha	TNFA
Clinical Immunological	Interferon gamma	IFNG
EEG Sleep	Sleep efficiency	SEFF
EEG Sleep	REM sleep	REM
EEG Sleep	Slow wave sleep	SWS
LC-MS Lipidomics	Phosphatidylethanolamine (32:1)	PE(32:1)
LC-MS Lipidomics	Phosphatidylethanolamine (36:0)	PE(36:0)
LC-MS Lipidomics	Phosphatidylethanolamine (36:1)	PE(36:1)
LC-MS Lipidomics	Phosphatidylethanolamine (38:6)	PE(38:6)
LC-MS Lipidomics	Phosphatidylethanolamine (38:3)	PE(38:3)
LC-MS Lipidomics	Ether phosphatidylethanolamine (38:5e)	PE(38:5e)
LC-MS Lipidomics	Ether phosphatidylethanolamine (36:2e)	PE(36:2e)
LC-MS Lipidomics	Cholesterol ester (16:1)	ChoE(16:1)
LC-MS Lipidomics	Cholesterol ester (18:3)	ChoE(18:3)
LC-MS Lipidomics	Triglyceride (54:7)	TG(54:7)
Lipid Transporter	Cholesterol ester transfer protein	CETP
Lipid Transporter	Phospholipid transfer protein	PLTP
Enzyme Activity	Lecithin-cholesterol acyltransferase	LCAT
Enzyme Activity	Paraoxonase 1	PON1
RNA Expression	ATP-binding cassette transporter A1	ABCA1
RNA Expression	Niemann-Pick disease C1	NPC1
RNA Expression	Nuclear receptor-interacting protein 1	NRIP1
RNA Expression	Carnitine palmitoyltransferase 1B	CPT1B
RNA Expression	Toll-like receptor 4	TLR4
RNA Expression	Toll-like receptor 8	TLR8
RNA Expression	Acyl-CoA dehydrogenase, very long chain	ACADVL
RNA Expression	Adipose differentiation-related protein	ADFP
RNA Expression	Sterol regul. elem. bind. transcr. factor 1	SREBF1
RNA Expression	NR1H2	LXRB

RNA Expression	NR1H3	LXRA
NMR Lipoprotein	Extra-large Very low density lipoproteins (sum concentration incl. XXL_VLDL and XL_VLDL particle concentrations)	XL_VLDL
NMR Lipoprotein	Medium Very low density lipoproteins (incl. L_VLDL, M_VLDL, S_VLDL)	M_VLDL
NMR Lipoprotein	Intermediate density lipoproteins (incl. XS_VLDL, IDL)	IDL
NMR Lipoprotein	Low density lipoproteins (incl. L_LDL, M_LDL, S_LDL)	LDL
NMR Lipoprotein	High density lipoproteins (incl. XL_HDL, L_HDL, M_HDL)	HDL
NMR Lipoprotein	Small High density lipoproteins	S_HDL
NMR Fatty acid	Omega-3 ratio to total fatty acids	w3/FA
NMR Fatty acid	Omega-6 ratio to total fatty acids	w6/FA
NMR Amino acid	Tyrosine	Tyr