Supplementary Material (ESI) for Molecular BioSystems

Glycomics meets lipidomics – Associations of N-glycans with classical lipids, glycerophospholipids, and sphingolipids in three European populations

[Online Supporting Information]

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Figure S1. Associations between N-glycan traits and classical lipids (TC, LDL-C, HDL-C, TG) in VIS (Croatia), NSPHS (Sweden), and ORCADES (Scotland). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.

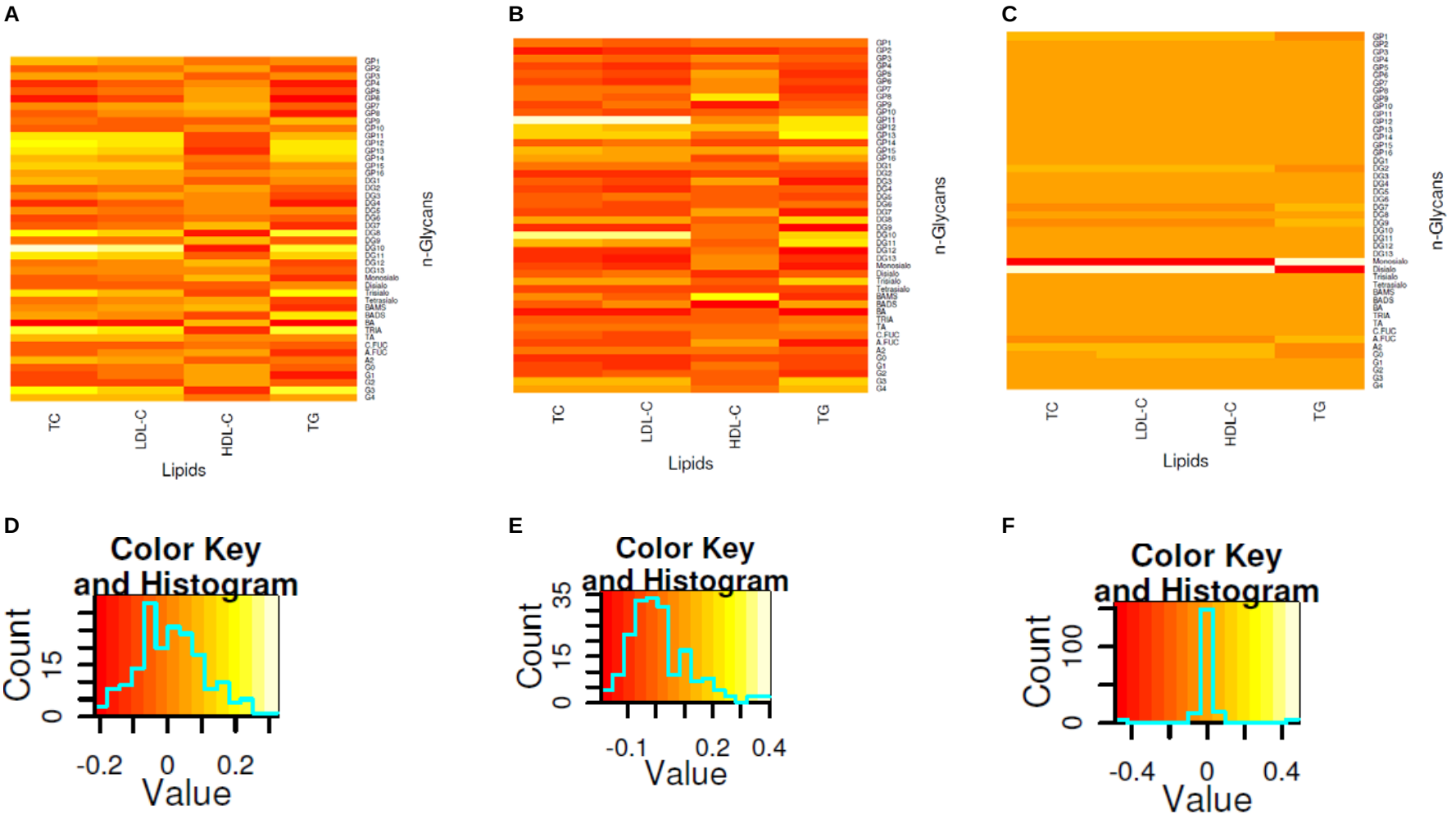


Figure S2. Associations between glycan species and glycerophospholipids in the VIS cohort (Croatia). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.

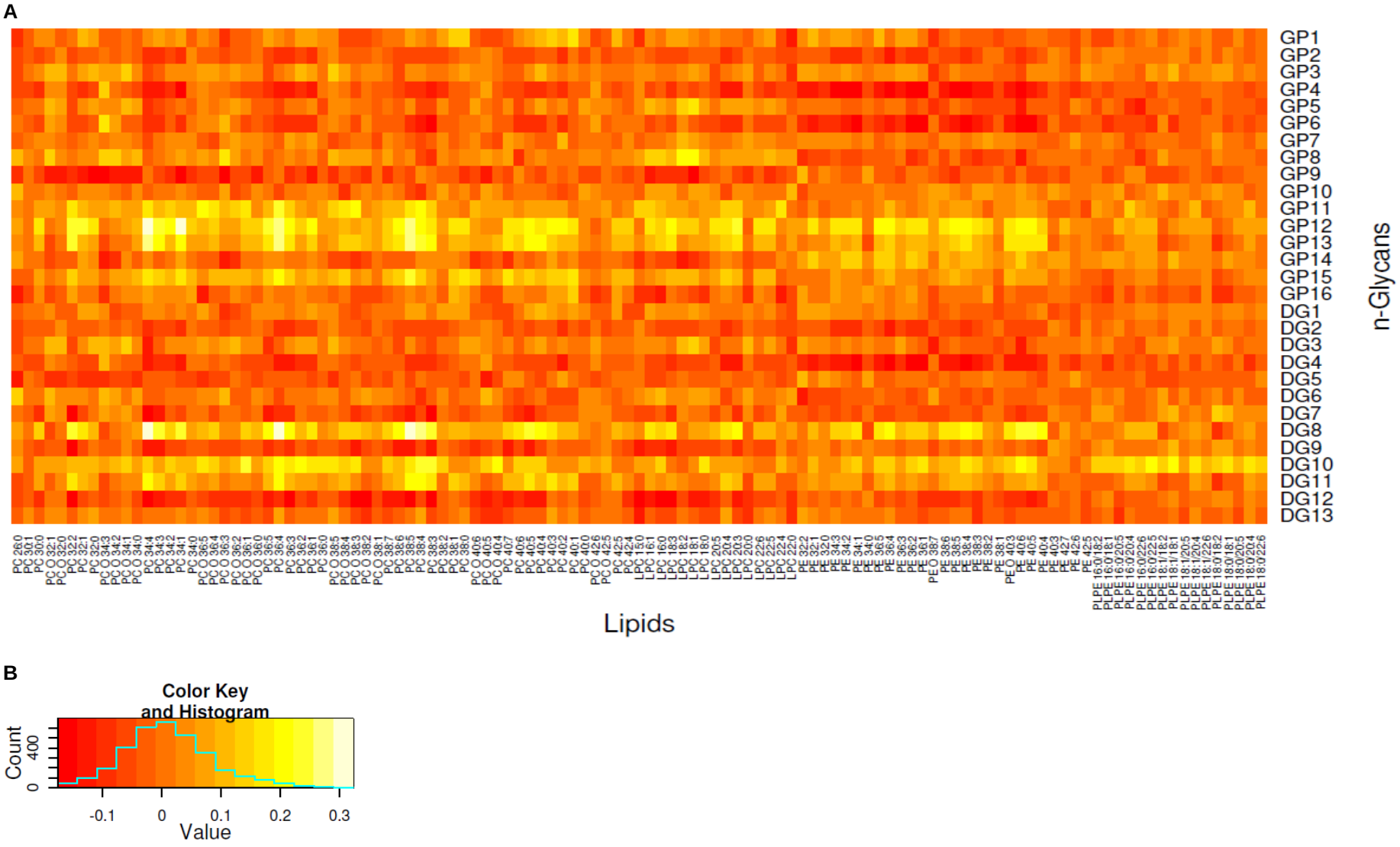


Figure S3. Associations between glycan species and sphingolipids species in the VIS cohort (Croatia). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.

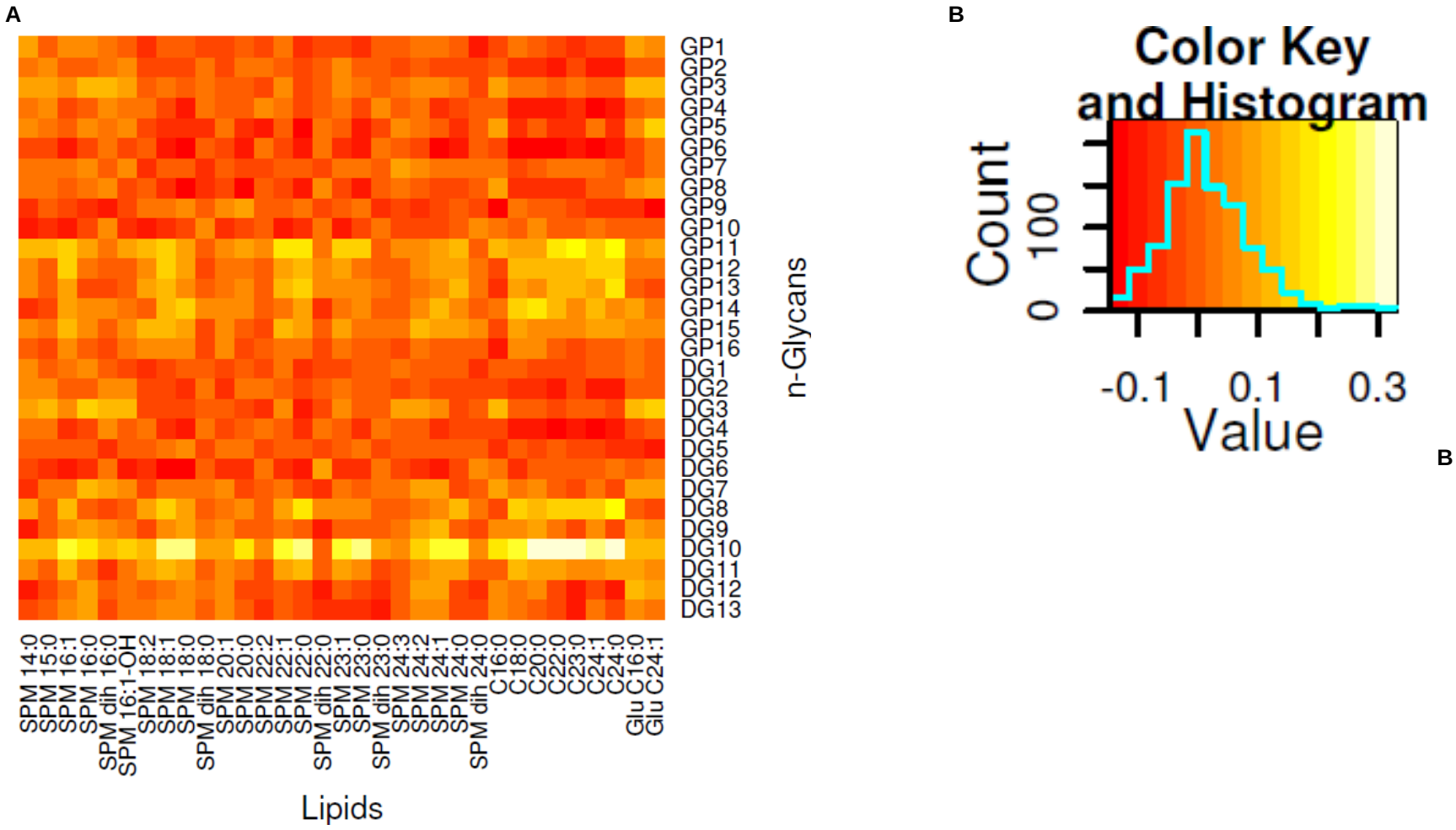
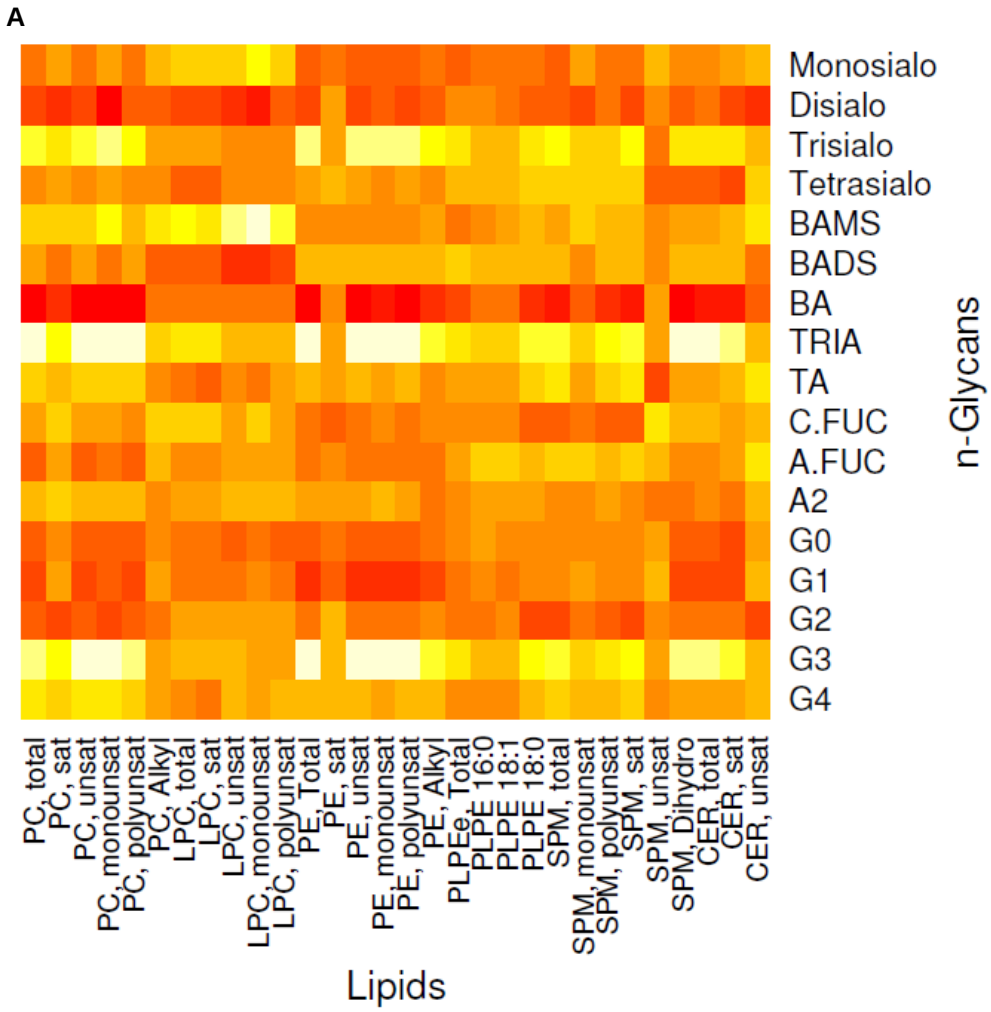


Figure S4. Associations between glycan summary scores and lipid summary scores in the VIS cohort (Croatia). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.



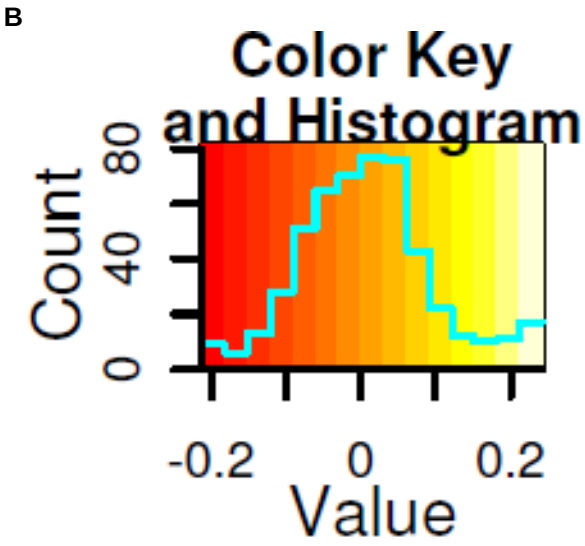
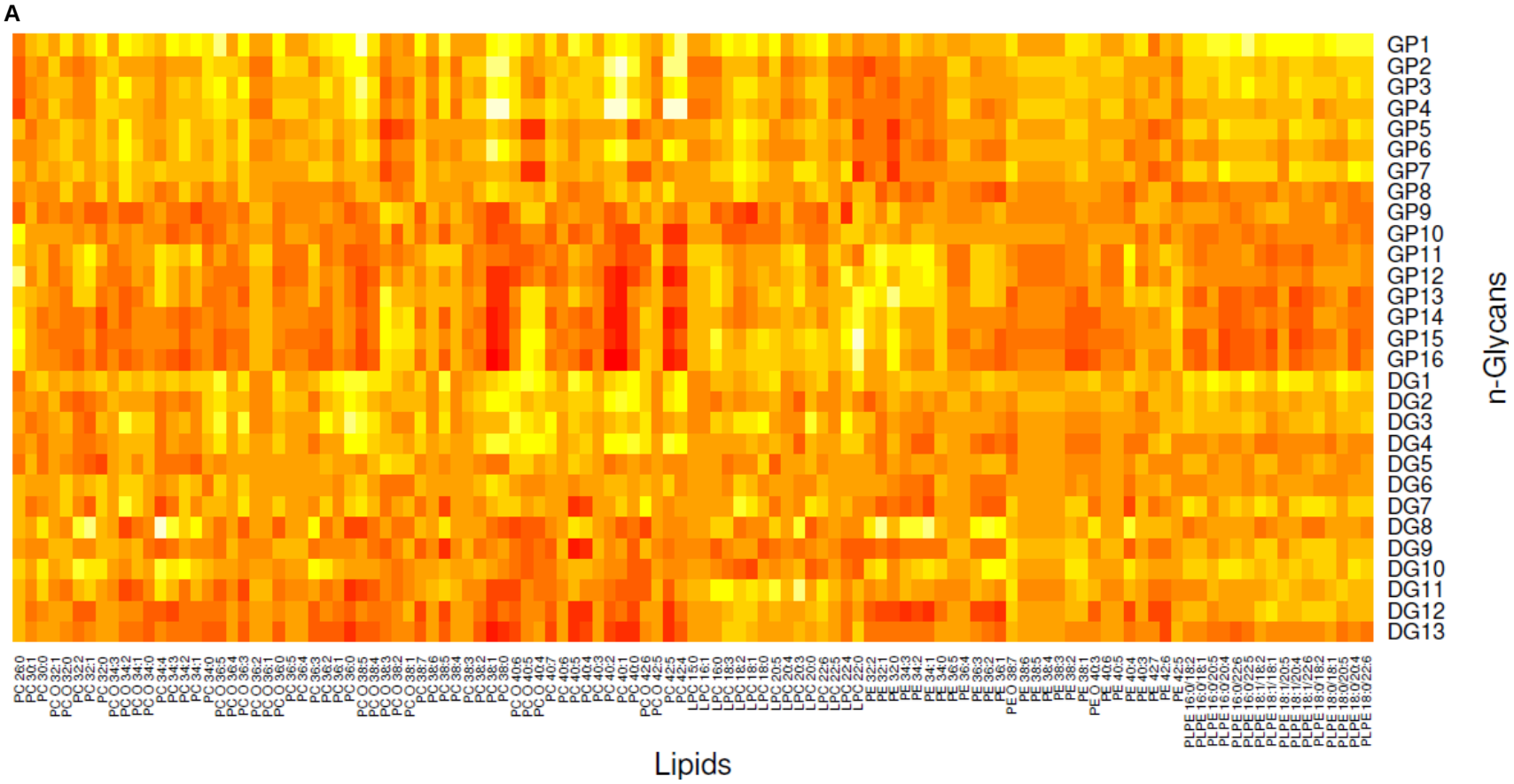


Figure S5. Associations between glycan species and glycerophospholipids in the ORCADES cohort (Scotland). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.



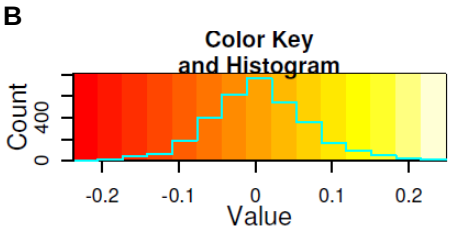


Figure S6. Associations between glycan species and sphingolipids species in the ORCADES cohort (Scotland). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.

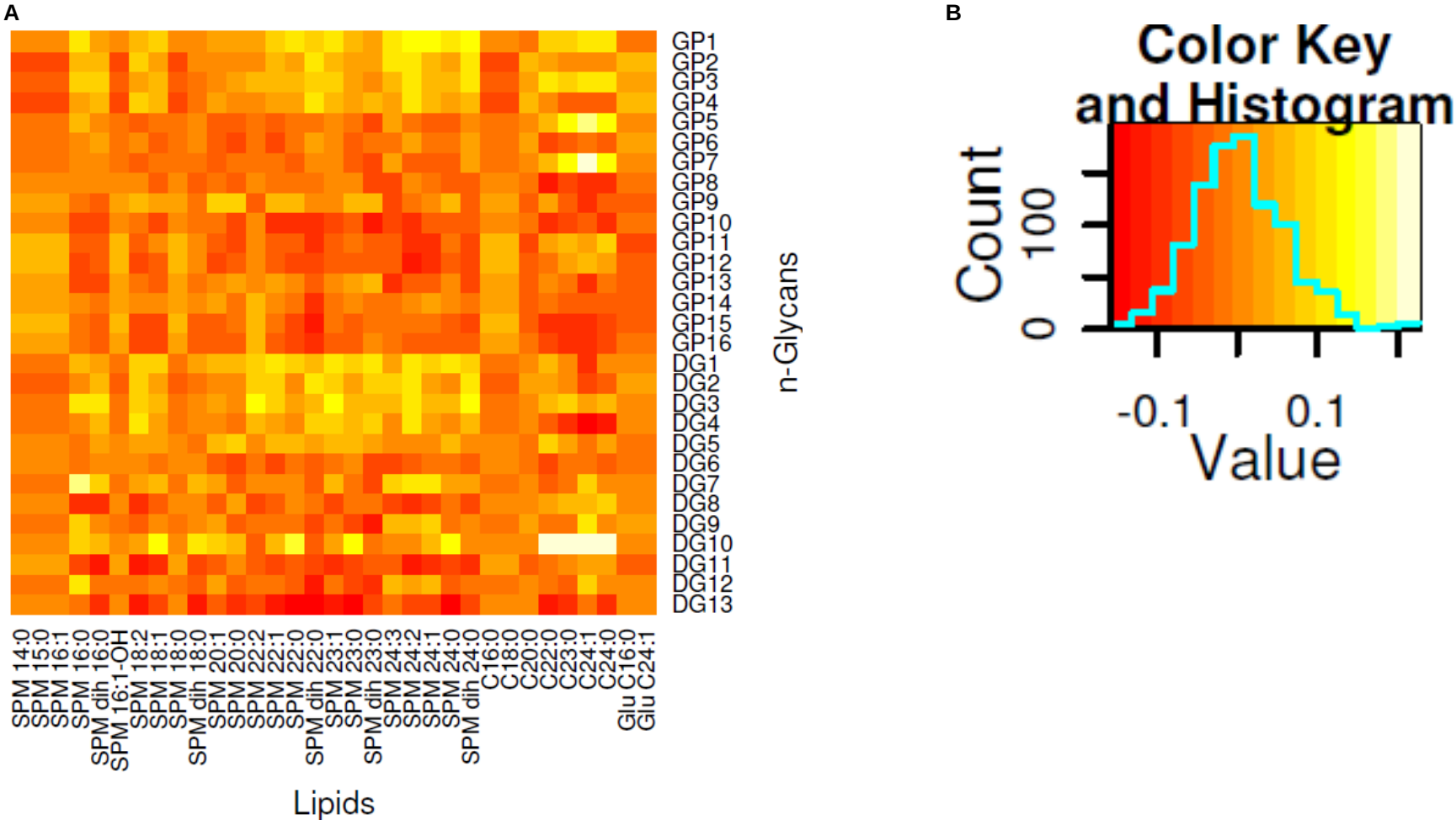


Figure S7. Associations between summary glycan scores and summary lipid scores in the ORCADES cohort (Scotland). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.

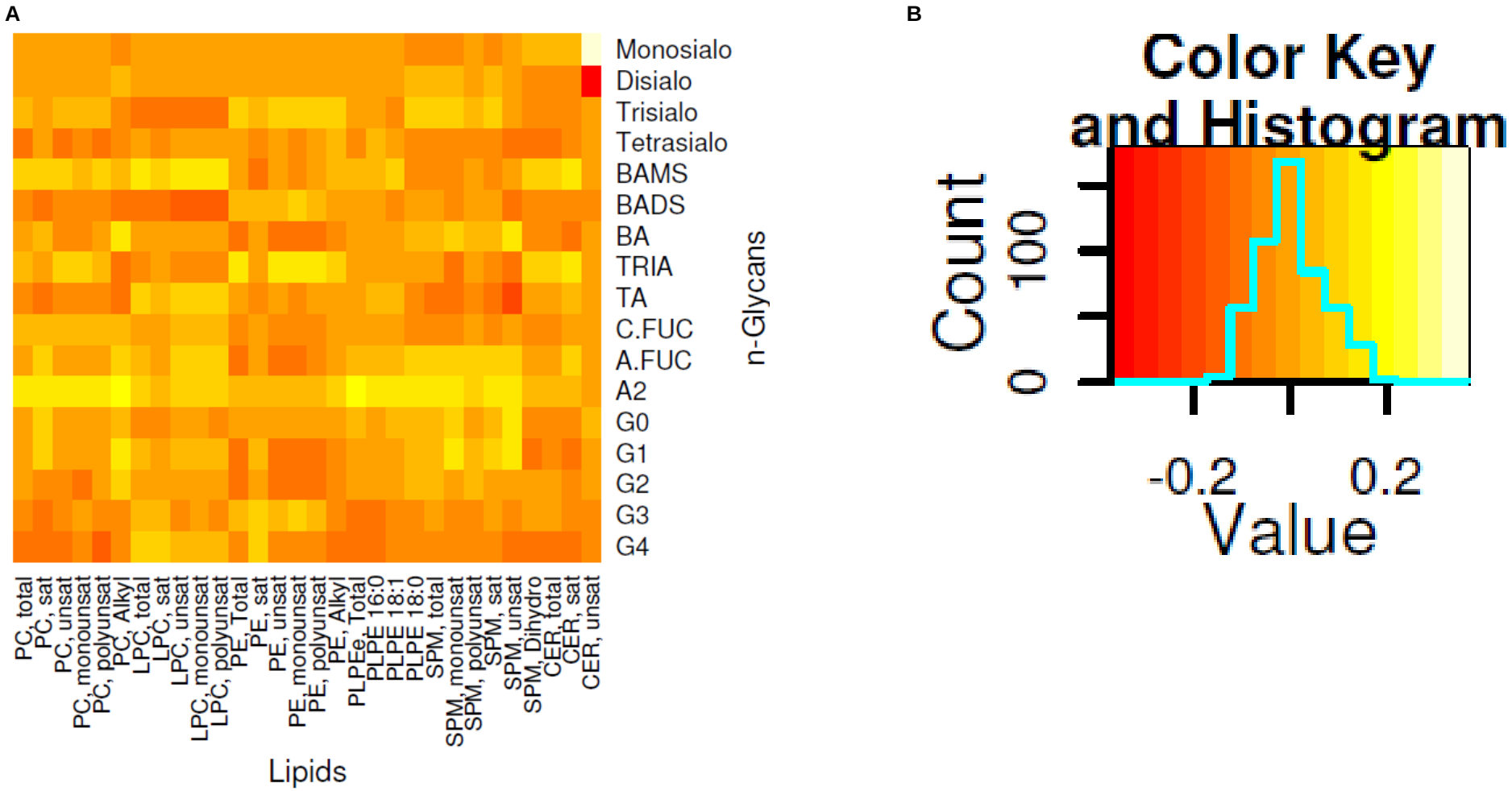
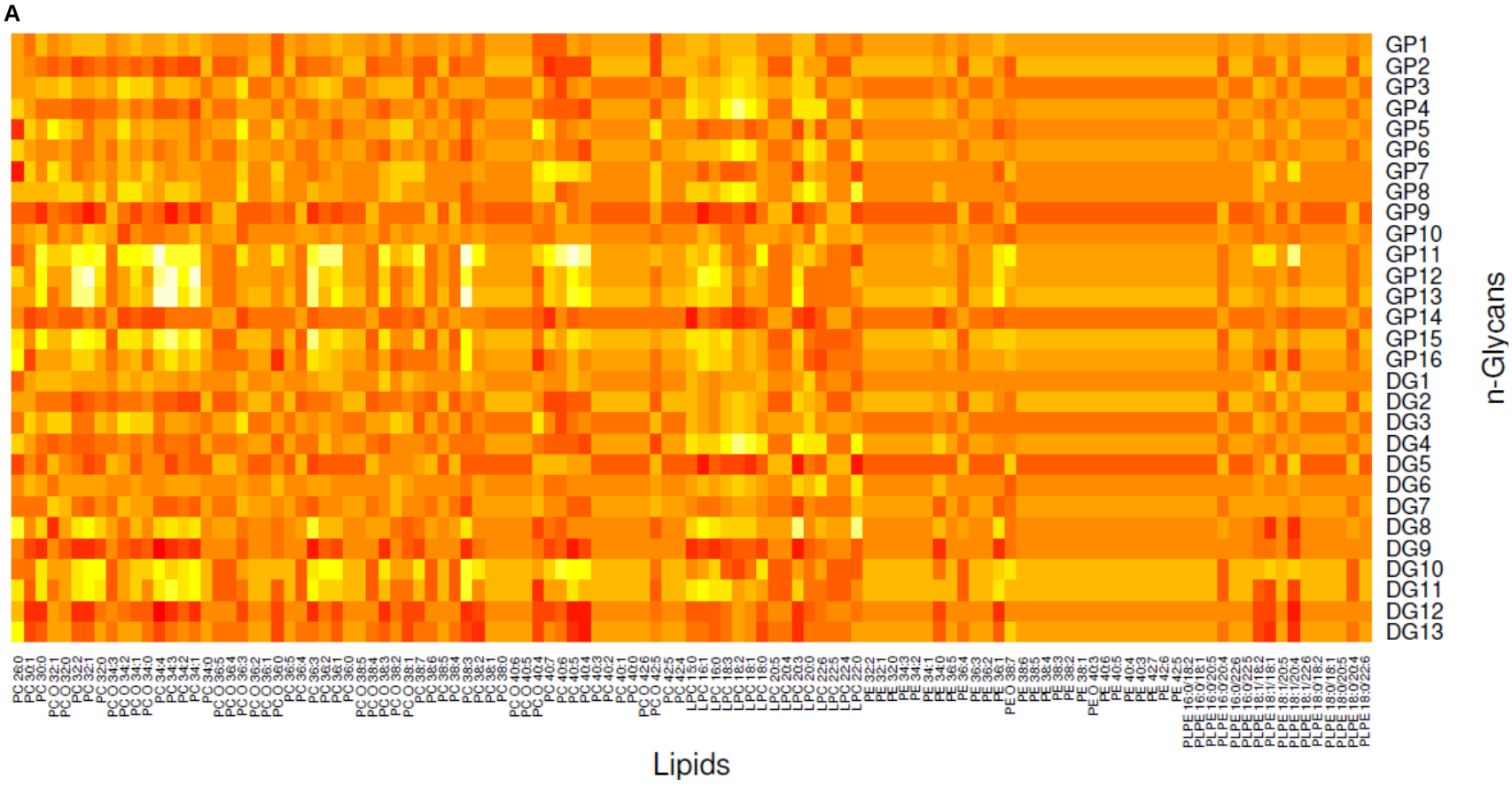


Figure S8. Associations between glycan species and glycerophospholipids in the NSPHS cohort (Sweden). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.



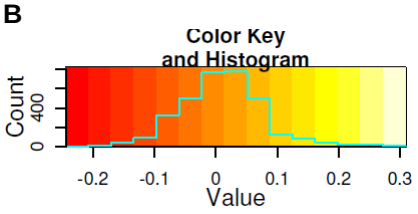


Figure S9. Associations between glycan species and sphingolipids species in the NSPHS cohort (Sweden). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*.

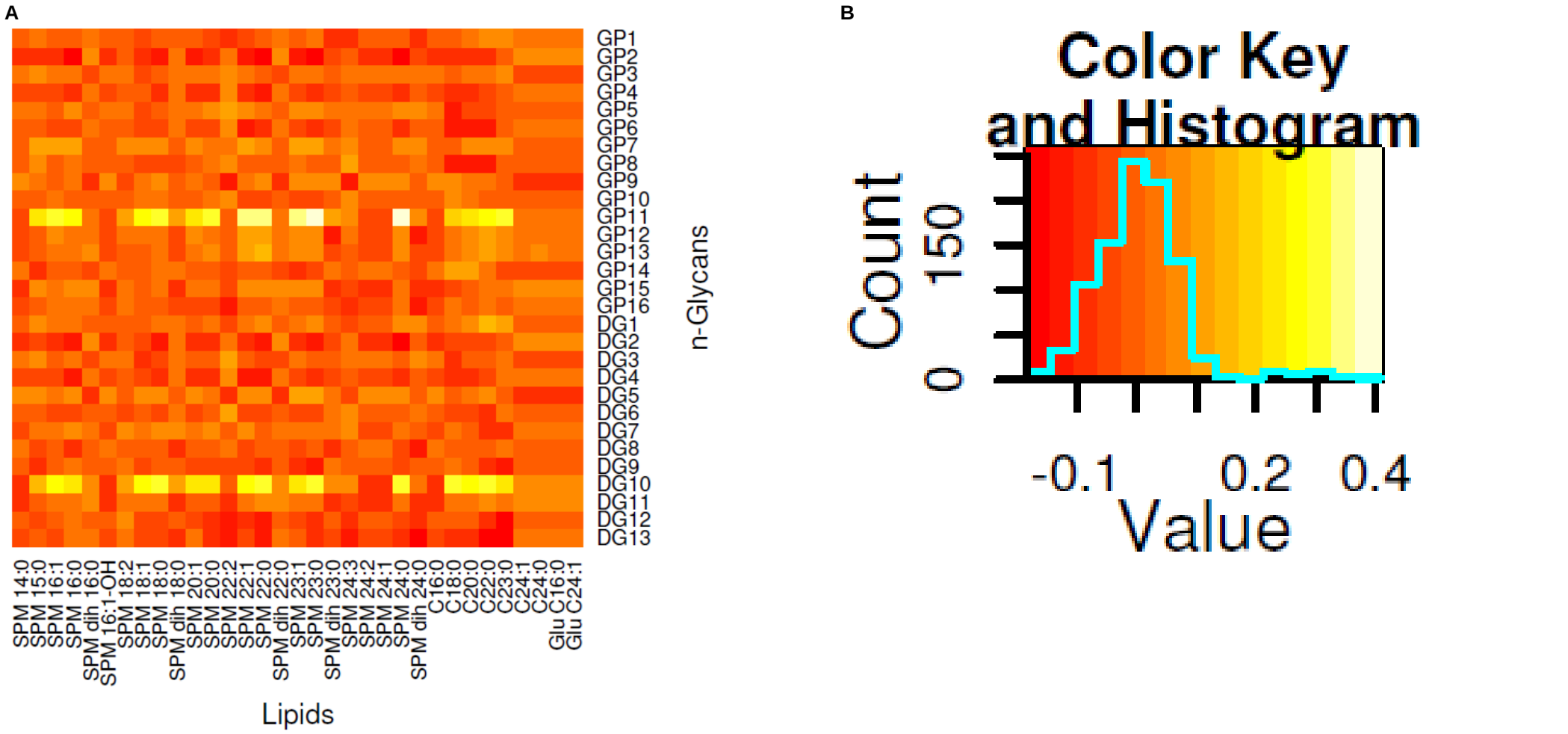
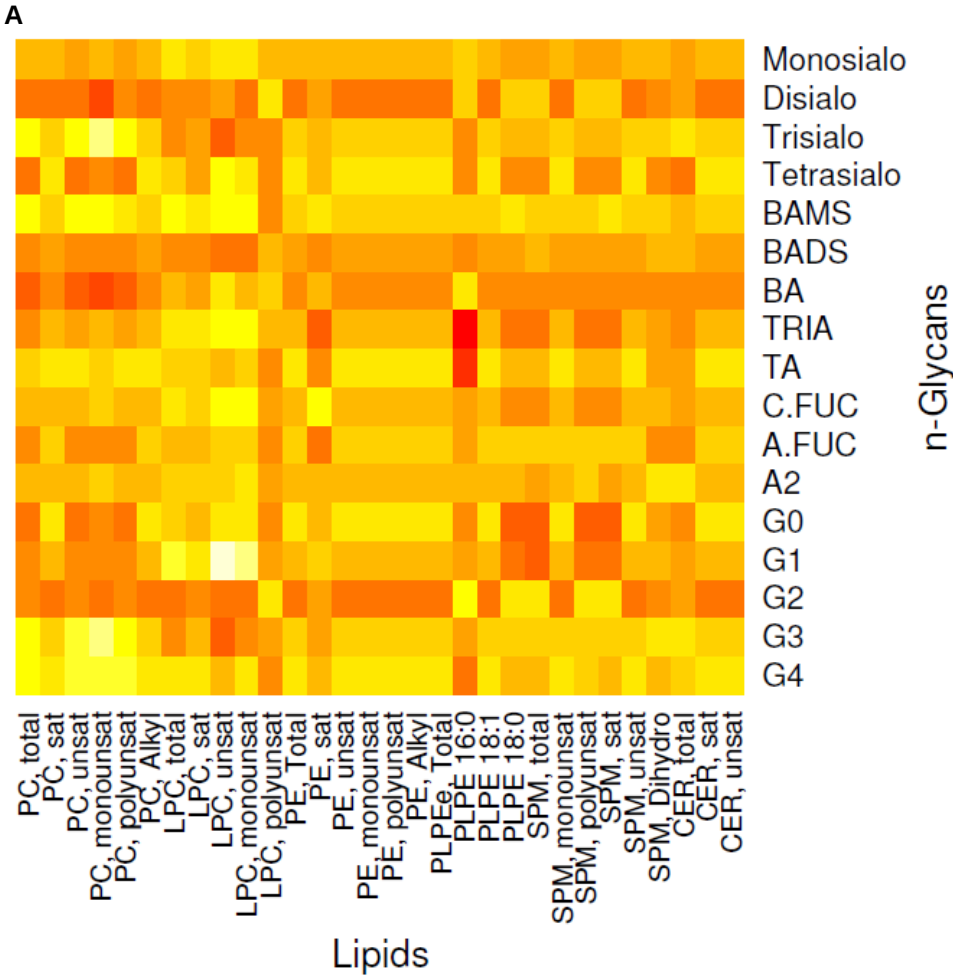


Figure S10. Associations between glycan summary scores and lipid summary scores in the NSPHS cohort (Sweden). A) Heatmap of Pearson correlation coefficients. B) Colorkey and histogram of the Pearson correlation coefficients *r*. See Tables S1 and S2 for glycan and lipid abbreviations.



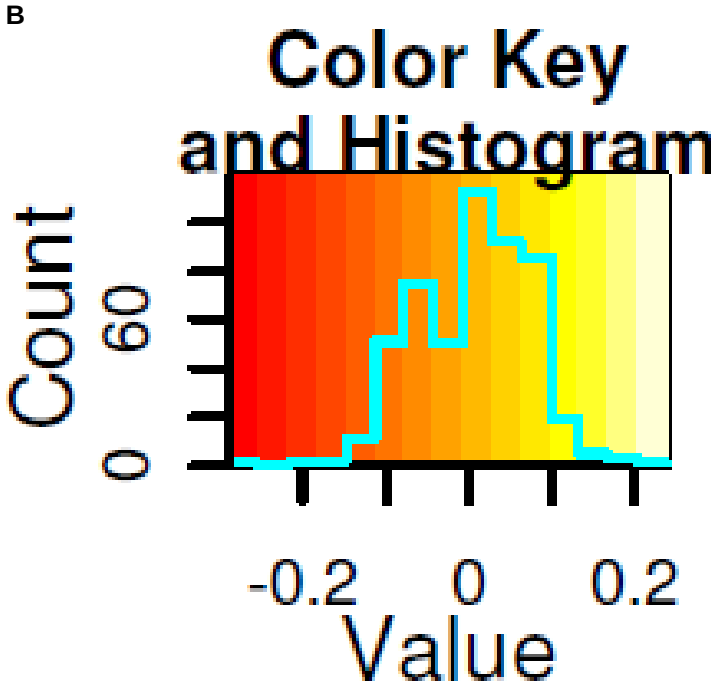


Table S1. Glycan structures present in different HPLC peaks.

Peak	Structure	Peak	Structure	Peak	Structure	Peak	Structure
GP1	A2	GP7	FA2BG2 M7D3 A2G2S(3)1 A2G2S(6)1 M7D1	GP12	A2F1G2S2 A3G3S(3,3)2 A3G3S(3,6)2 A3G3S(6,6)2 A3BG3S(3,3)2 A3BG3S(3,6)2 A3BG3S(6,6)2	DG1	A2
GP2	A2B A1G1 FA2					DG2	A2B A1G1 FA2
GP3	M5 FA2B A2[6]G1 A2[6]BG1	GP8	A2BG2S(3)1 A2BG2S(6)1 M5A1G1S1 FA2G2S(3)1 FA2G2S(6)1 A3G3 FA2BG2S(3)1 FA2BG2S(6)1			DG3	M5 FA2B A2[6]G1 A2[6]BG1
GP4	A2[3]G1 A2[3]BG1 M4A1G1 FA2[6]G1 FA2[6]BG1 A1[6]G1S(3)1 A1[6]G1S(6)1 FA2[3]G1 FA2[3]BG1 M6D1, D2 A1[3]G1S(3)1 A1[3]G1S(6)1	GP9	A2F1G2S(3)1 A2F1G2S(6)1 M8D2, D3 A2G2S(3,3)2 A2G2S(3,6)2 A2G2S(6,6)2 M8D1,D3	GP13	A3G3F1S2 FA3G3S(3,3)2 FA3G3S(3,6)2 FA3G3S(6,6)2 FA3BG3S(3,3)2 FA3BG3S(3,6)2 FA3BG3S(6,6)2 A3G3S(3,3,6)3 A3G3S(3,6,6)3 A3G3S(6,6,6)3	DG4	M4A1G1 A2[3]G1 A2[3]BG1 FA2[6]BG1 FA2[3]G1 FA2[3]BG1
						DG5	M6D1, D2 M6D3 A2G2 A2BG2
GP5	M6D3 A2[6]G1S(3)1 A2[6]G1S(6)1 A2G2 A2[3]G1S(3)1 A2[3]G1S(6)1 A2BG2	GP10	A2BG2S(3,3)2 A2BG2S(3,6)2 A2BG2S(6,6)2 A3BG3S(3)1 A3BG3S(6)1 FA2G2S(3,3)2 FA2G2S(3,6)2 FA2G2S(6,6)2	GP14	A3F1G3S(3,3,6)3 FA3F1G3S(6,6,6)3 A4G4S(6,6)2 A3F1G3S(3,6,6)3 A3F1G3S(6,6,6)3 A4G4S(6,6,6)3 A4F1G4S2 A4G4S3	DG6	FA2G2 M5A1G1 FA2BG2
						DG7	M7D3 A2F1G2 M7D1
GP6	FA2[6]G1S(3)1 FA2[6]G1S(6)1 FA2[6]BG1S(3)1 FA2[6]BG1S(6)1 M4A1G1S1 FA2G2 FA2[3]G1S(3)1 FA2[3]G1S(6)1 A2BG1S1 FA2[3]BG1S(3)1 FA2[3]BG1S(6)1	GP11	FA2BG2S(3,3)2 FA2BG2S(3,6)2 FA2BG2S(6,6)2 M9	GP15	A4G4S4 A4F1G4S3	DG8	A3G3 A2F2G2 FA3G3 M8D2, D3 M8D1,D3
				GP16	A4G4S(6,6,6,6)4 A4G4S(3,6,6,6)4 A4BG4S4 FA4G4S4 A4F1G4S4 A4G4LacS4 A4F2G4S4 FA4F1G4S4	DG9	FA3BG3 A3F1G3
						DG10	M9 FA3F1G3
						DG11	A4G4 A4BG4 A3F2G3 FA4G4
						DG12	A4F1G4
						DG13	A4G4Lac A4F2G4 FA4F1G4

Note: Structure abbreviations: all N-glycans have two core GlcNAcs; F at the start of the abbreviation indicates a core fucose α1-6 linked to the inner GlcNAc; Mx, number (x) of mannose on core GlcNAcs; D1 indicates that the α1-2 mannose is on the Manα1-6Manα1-6 arm, D2 on the Manα1-3Manα1-6 arm, D3 on the Manα1-3 arm of M6 and on the Manα1-2Manα1-3 arm of M7 and M8; Ax, number of antenna (GlcNAc) on trimannosyl core; A2, biantennary with both GlcNAcs as β1-2 linked; A3, triantennary with a GlcNAc linked β1-2 to both mannose and the third GlcNAc linked β1-4 to the α1-3 linked mannose; A4, GlcNAcs linked as A3 with additional GlcNAc β1-6 linked to α1-6 mannose; B, bisecting GlcNAc linked β1-4 to β1-3 mannose; Gx, number (x) of β1-4 linked galactose on antenna; [3]G1 and [6]G1 indicates that the galactose is on the antenna of the α1-3 or α1-6 mannose; F(x), number (x) of fucose linked α1-3 to antenna GlcNAc; Lac(x), number (x) of lactosamine (Galβ1-4GlcNAc) extensions; Sx, number (x) of sialic acids linked to galactose; the numbers 3 or 6 or in parentheses after S indicate whether the sialic acid is in an α2-3 or α2-6 linkage. If there is no linkage number, the exact link is unknown.

Table S2. Overview over the examined N-glycan traits.

Abbreviation	Name	Category	Formula
GP1 .. GP6	Glycan Peak 1 .. Glycan Peak 6	Core glycans	Calculated from HILIC profile
GP7 .. GP8	Glycan Peak 7 .. Glycan Peak 8	Monosialylated biantennary core glycans	Calculated from HILIC profile
GP9 .. GP11	Glycan Peak 9 .. Glycan Peak 11	Disialylated biantennary core glycans	Calculated from HILIC profile
GP12 .. GP14	Glycan Peak 12 .. Glycan Peak 14	Trigalactosylated core glycans	Calculated from HILIC profile
GP15 .. GP16	Glycan Peak 15 .. Glycan Peak 16	Tetragalactosylated core glycans	Calculated from HILIC profile
DG1 .. DG7	Disialylated Glycan 1 .. Disialylated Glycan 7	Biantennary core core glycans	Calculated from HILIC after sialidase treatment
DG8 .. DG10	Disialylated Glycan 8 .. Disialylated Glycan 10	Triantennary core glycans	Calculated from HILIC after sialidase treatment
DG11 .. DG13	Disialylated Glycan 1 .. Disialylated Glycan 13	Tetraantennary core glycans	Calculated from HILIC after sialidase treatment
Monosialo	Monosialylated Glycans	Summary Score	Calculated from WAX profile
Disialo	Disialylated Glycans	Summary Score	Calculated from WAX profile
Trisialo	Trisialylated Glycans	Summary Score	Calculated from WAX profile
Tetrasialo	Tetrasialylated Glycans	Summary Score	Calculated from WAX profile
C-FUC	Core fucosylated glycans	Summary Score	= DG6/(DG5+DG6)*100
A-FUC	Antennary fucosylated glycans	Summary Score	= DG7/(DG5+DG7)*100
A2	Biantennary nongalactosylated glycan	Summary Score	= (GP1+DG1)/2
BA	Biantennary glycans	Summary Score	= DG1 + DG2 + DG3 + DG4 + DG5 + DG6 + DG7
BAMS	Monosialylated biantennary glycans	Summary Score	= (GP7 + GP8) / (DG5 + DG6 + DG7) * 100
BADS	Disialylated biantennary glycans	Summary Score	= (GP9 + GP10 + GP11) / (DG5 + DG6 + DG7)*100
TRIA	Triantennary glycans	Summary Score	= DG8 + DG9 + DG10
TA	Tetraantennary glycans	Summary Score	= DG11 + DG12 + DG13
G0	Nongalactosylated glycans	Summary Score	= DG1 + DG2
G1	Monogalactosylated glycans	Summary Score	= DG3 + DG4
G2	Digalactosylated glycans	Summary Score	= DG5 + DG6 + DG7
G3	Trigalactosylated glycans	Summary Score	= GP12 + GP13 + GP14
G4	Tetragalactosylated glycans	Summary Score	= GP15 + GP16

Table S3. Overview over of examined lipid traits.

Abbreviation	Common Name	Lipid Maps			
		Category	**Class**	**Subclass**	**ID**
TC	Total Cholesterol	[ST] Sterol Lipids	[ST01] Sterols	[ST0101] Cholesterol and derivatives	[LMST01010001]
LDL-C	Low-Density Lipoprotein Cholesterol	[ST] Sterol Lipids	[ST01] Sterols	[ST0101] Cholesterol and derivatives	[LMST01010001]
HDL-C	High-Density Lipoprotein Cholesterol	[ST] Sterol Lipids	[ST01] Sterols	[ST0101] Cholesterol and derivatives	[LMST01010001]
TG	Triglycerides	[GL] Glycerolipids	[GL03] Triadylglycerols	[GL0301] Triacylglycerols	
GP	Glycerophospholipid	[GP] Glycerophospholipids			
PC	Phosphatidylcholine	[GP] Glycerophospholipids	[GP01] Glycerophosphocholines		
LPC	Lysophosphatidylcholine	[GP] Glycerophospholipids	[GP01] Glycerophosphocholines	[GP0105] Monoacylglycerophosphocholines	
PE	Phosphatidylethanolamine	[GP] Glycerophospholipids	[GP02] Glycerophosphoethanolamine		
PEPL	Phosphatidylethanolamine-based plasmalogen	[GP] Glycerophospholipids	[GP02] Glycerophosphoethanolamine	[GP0203] 1Z-alkenyl,2-acylglycero-phosphoethanolamine	
SP	Sphingolipid	[SP] Sphingolipids			
SM	Sphingomyelin	[SP] Sphingolipids	[SP03] Phosphosphingolipids	[SP0301] Ceramide phosphocholines	
CER	Ceramide	[SP] Sphingolipids	[SP02] Ceramides		
GlcCER	Glucosylceramide/ Glucocerebroside	[SP] Sphingolipids	[SP05] Neutral glycosphingolipids	[SP0501] Simple GLC series	

Abbreviations which differ from the LipidMaps classification were used in the text to improve readability. Abbreviations are in order of presentation.