Supplementary Information

**Title:** Obesity, adipokines, and C-peptide are associated with distinct plasma phospholipid profiles in adult males, an untargeted lipidomic approach.

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**Supplementary Table 1: Chemicals and solvents, chromatographic and mass spectrometric analysis parameters, and identification and correction of time of injection effect.**

**Chemicals and solvents**
High performance liquid chromatography-grade (HPLC) solvents were used for the extraction and resuspension of all samples analyzed. The following solvents were used in the analysis: HPLC-grade methanol (Lot#: 000118827, J.T. Baker, Phillipsburg, NJ), HPLC-grade chloroform (Lot#: 55296, Omnisolv, Charlotte, NC), HPLC-grade water (Lot#: 0000063121, J.T. Baker), HPLC-grade isopropanol (Lot#: SHBC6903V, Sigma-Aldrich, St. Louis, MO), and HPLC-grade acetonitrile (Lot #: 53324, EMD chemicals, Gibbstown, NJ).

Internal standards for each lipid class were selected based on commercial availability at time of analysis. The internal standards used in this study are as follows: PE(16:0-d₃₁/18:1) [Product#:110921; Lot#: LM160D31-181PE-33], PG-d₅(16:0/18:1) [Product#: 110919; Lot#:LM160-181PGD5-10], PS(16:0-d₃₁/18:1) [Product#: 110922; Lot#: LM160D31-181PS-25], LysoPE(13:0) [Product#: 110696; Lot#: LM130LPE-10], PC(24:0/24:0) [Product#: 110929; Lot#: LM240PC-28], Cer(d18:1/25:0) [Product#: LM-2225; Lot#: LM5-139D] (Avanti Polar Lipids, Alabaster, AL), and PC(8:0/8:0) [Product#: 10009874; Lot#181547-14] (Cayman Chemical, Ann Arbor, MI). The antioxidant butylated hydroxytoluene (BHT) (Sigma-Aldrich) was present in all solvents at a concentration of 0.1%.

**Internal standards and extraction solution**
A mixture of internal standard (IS) containing PE(16:0-d₃₁/18:1), PG-d₅(16:0/18:1), PS(16:0-d₃₁/18:1), LysoPE(13:0), PC(24:0/24:0), Cer(d18:1/25:0) (Avanti Polar Lipids), and PC(8:0/8:0) (Cayman Chemical) was prepared. This IS mixture was added to an extraction solution composed of HPLC-grade chloroform:methanol (2:1 v/v, Omnisolv, J.T. Baker) with 0.1% BHT (100 µg/µL, Sigma-Aldrich). The final concentration of IS in the extraction solution was 5 ng/µL for all PLs and 0.2 ng/µL for Cer.

**UPLC MS/MS Analysis and Parameters**
The instrument used in plasma lipid separation was a Waters ACQUITY UPLC using ACQUITY UPLC CSH C18 1.7 µm 2.1x100mm column (Waters, Milford, MA). The column was washed prior to analysis for 1.5 h with 50% dichloromethane and 50% 2-propanol, followed by 1.5 h with 100% 2-propanol. The UPLC method used in this analysis performed was based on the Waters application note: Issac et al. Lipid Separation using UPLC with Charged Surface Hybrid Technology. The UPLC method was modified to shorten the run time to 15 min.
**UPLC Parameters**

| Parameter             | Setting                                                                 |
|-----------------------|-------------------------------------------------------------------------|
| Run Time:             | 15.00 min                                                               |
| Solvent A:            | Acetonitrile/water (60:40) + 10 mM ammonium formate                      |
| Solvent B:            | Isopropanol/acetonitrile (90:10) + 10mM ammonium formate                |
| Column Temperature:   | 55.0 °C                                                                 |
| Injection Volume (µL):| 10.00                                                                   |
| Equilibration Time:   | 0.1 min                                                                 |

**UPLC Gradient Table**

| Time (min) | Flow Rate | %A | %B | Curve |
|------------|-----------|----|----|-------|
| Initial    | 0.400     | 60 | 40 | Initial |
| 1.50       | 0.400     | 57 | 43 | 6      |
| 1.60       | 0.400     | 50 | 50 | 1      |
| 9.00       | 0.400     | 57 | 43 | 6      |
| 9.08       | 0.400     | 30 | 70 | 1      |
| 13.50      | 0.400     | 1  | 99 | 6      |
| 13.60      | 0.400     | 60 | 40 | 6      |
| 15.00      | 0.400     | 60 | 40 | 1      |

The MS/MS analysis of plasma lipids was performed using a Waters Xevo G2-XS quadrupole time-of-flight mass spectrometer (Waters). Data acquisition was performed using MS^E in continuum mode with lock mass application. Source parameters are as follows:

**Mass Spectrometer Source and Optics Settings**

| Parameter            | Setting            |
|----------------------|--------------------|
| Analyzer             | Sensitivity mode   |
| Capillary (kV)       | 1                  |
| Sampling Cone (kV)   | 10                 |
| Source Temperature (°C) | 110              |
| Source Offset        | 80                 |
| Desolvation Temperature (°C) | 350              |
| Cone Gas Flow (L/Hr) | 25                 |
| LM Resolution        | 4.7                |
| HM Resolution        | 15                 |

Three MS functions were used to obtain MS/MS spectra and correct for mass drift: Function1 was used to obtain a parent ion spectra of lipids; Function 2 was used to obtain a fragmentation spectra of parent lipid ions; and Function 3 was used to measure leucine enkephalin as lock mass for mass correction.

**Mass Analyzer Settings and Collison Energies**

**Function 1: TOF Parent Ion Function**

| Parameter                 | Setting |
|---------------------------|---------|
| Survey Start Time (min)   | 0.0     |
| Survey End Time (min)     | 15.0    |
**Supplementary Information**

| Survey Ion Mode | ES Mode |
|-----------------|---------|
| Survey Polarity | Negative|

**[PARENT MS SURVEY]**

| Survey Start Mass | 60 |
| Survey End Mass   | 1800 |
| Parent Survey High CE (V) | 30.0 |
| Parent Survey Low CE (V) | 10.0 |

**Function 2: TOF Fragmentation Function**

| Survey Start Time (min) | 0.0 |
| Survey End Time (min)   | 15.0 |
| Survey Ion Mode         | ES Mode |
| Survey Polarity         | Negative |

**[PARENT MS SURVEY]**

| Survey Start Mass | 60.0 |
| Survey End Mass   | 1800.0 |
| Parent Survey High CE (V) | 30.0 |
| Parent Survey Low CE (V) | 10.0 |

**Function 3: TOF Lockmass Function**

| Survey Start Time (min) | 0.0 |
| Survey End Time (min)   | 15.0 |
| Survey Ion Mode         | ES Mode |
| Survey Polarity         | Negative |

**Progenesis QI Data Importation**

Raw UPLC-MS/MS data files were imported into Progenesis QI for data alignment and processing. The following adducts were selected based on the sample matrix (i.e., chloride), mobile phases (i.e., formate), and contaminants (i.e., trifluoroacetate and nitrate) that may have been present in either the samples or the mass analyzer at time of analysis. The following adducts were included for deconvolution of M, 2M, and 3M \( m/z \)s: M-H, M+formate-H, M-methylformate-H, M+Cl-H, M+trifluoroacetate-H, M-methyltrifluoroacetate-H, and M+nitrate-H. After peaking picking and deconvolution by Progenesis QI, a total of 4802 \( m/z \)s were identified. The data was first exported to EZ-Info v3.0 (Umetrics, San Jose, CA), then to Excel (Microsoft, Redmond, WA) for relative mass defect filtering.
**Time of Injection Effect**

After initial data processing of mass defect filtered dataset, we investigated batch and time of injection effects in the data. The variable time of injection was created by multiplying the samples file number by 15 mins, thus, yielding the samples time of injection relative to the to the first injection of the analysis. We observed a time of injection effect after plotting principal components (Pc) 1 and 2. There were distinct clusters of samples based on their time of injection (mins) during the mass spectrometric analysis. In addition, the Pc 1 and 2 score were significantly associated with time of injection in regression analyses.

![Principal components derived from patient (n=126) lipidomic profiles. Number listed next to samples indicate the samples time of injection in mins relative to the first injection of analysis. Samples injected earlier in the analysis clustered tightly together, while samples injected later in the analysis drifted.](image1)

Principal components derived from patient (n=126) lipidomic profiles. Number listed next to samples indicate the samples time of injection in mins relative to the first injection of analysis. Samples injected earlier in the analysis clustered tightly together, while samples injected later in the analysis drifted.

Next, to further examine the time of injection effect, we plotted the range of intensities for each plasma lipid in the 126 samples.

![Time of injection vs. Metabolite Intensity](image2)

Figure displays time of injection and the range of plasma lipid intensities for each sample (n=126) analyzed. As time of injection increased, so did the range of metabolite intensities,
indicating ion suppression throughout analysis. We determined the time of injection effect was due to ion suppression by trifluoroacetate contamination in the mass analyzer. Since a majority of plasma phospholipids are PCs, we normalized our entire data matrix by the IS PC(8:0/8:0). IS normalization of the data matrix removed the time of injection effect on plasma lipids.

Principal components derived from patient (n=126) IS normalized lipidomic profiles. Number listed next to samples indicate the samples time of injection in minutes. Normalization of the data matrix with IS PC(8:0/8:0) removed the time of injection effect. Next, to further examine if normalizing the data matrix removed the time of injection effect, we plotted the range of intensities for each plasma lipid in the 126 samples

Above figure displays time of injection and the range of plasma lipid intensities for each sample (n=126) analyzed. After IS normalization of the data matrix, the time of injection effect was removed.
Supplementary Table 2: Table of lipids significantly associated with BMI with FDR p-values <0.05. Beta coefficients, p-values, Benjamini-Hochberg FDR p-values, and Bonferroni p-values listed.

| Primary ID     | Beta   | p-value     | FDR p-value | Bonferroni p-value |
|----------------|--------|-------------|-------------|--------------------|
| X1.17_564.3289 | -5.25  | 1.48E-10    | 2.59E-07    | 2.59E-07           |
| X1.42_566.3497 | -10.78 | 1.30E-06    | 1.14E-03    | 2.28E-03           |
| X1.21_476.2768 | -14.67 | 4.03E-06    | 2.35E-03    | 7.04E-03           |
| X1.51_592.3513 | -115.61| 1.06E-05    | 3.92E-03    | 1.86E-02           |
| X1.03_562.3132 | -44.59 | 1.12E-05    | 3.92E-03    | 1.96E-02           |
| X5.23_826.5592 | -7.70  | 1.47E-05    | 4.28E-03    | 2.57E-02           |
| X7.15_786.5626 | -6.28  | 1.95E-05    | 4.87E-03    | 3.41E-02           |
| X1.59_554.3446 | -23.44 | 4.62E-05    | 1.01E-02    |                    |
| X4.18_794.5050 | -32.49 | 5.40E-05    | 1.05E-02    |                    |
| X1.49_478.2927 | -17.91 | 8.28E-05    | 1.44E-02    |                    |
| X2.63_393.2768 | 14.21  | 2.19E-04    | 3.39E-02    |                    |
| X9.42_814.5927 | -11.67 | 2.33E-04    | 3.39E-02    |                    |
| X1.08_612.3280 | -13.31 | 2.56E-04    | 3.44E-02    |                    |
| X8.93_856.6036 | 3.98   | 2.76E-04    | 3.45E-02    |                    |
| X6.50_828.5729 | -1.80  | 3.35E-04    | 3.90E-02    |                    |
| X3.52_421.3076 | 85.75  | 4.46E-04    | 4.34E-02    |                    |
| X1.22_544.2648 | -36.54 | 4.50E-04    | 4.34E-02    |                    |
| X2.89_493.3353 | -33.94 | 4.72E-04    | 4.34E-02    |                    |
| X5.27_1031.496 | -56.39 | 4.80E-04    | 4.34E-02    |                    |
| X4.36_825.5432 | -29.25 | 5.16E-04    | 4.34E-02    |                    |
| X2.72_639.5552 | 11.27  | 5.42E-04    | 4.34E-02    |                    |
| X5.43_1068.6601 | -28.22 | 5.51E-04    | 4.34E-02    |                    |
| X8.90_880.6027 | 7.18   | 5.96E-04    | 4.34E-02    |                    |
| X5.24_1108.4552 | -21.48 | 5.97E-04    | 4.34E-02    |                    |
| X5.24_952.4993 | -16.66 | 6.86E-04    | 4.78E-02    |                    |
Supplementary Table 3: Table of lipids significantly associated with waist circumference with FDR p-values < 0.05. Beta coefficients, p-values, Benjamini-Hochberg FDR p-values, and Bonferroni p-values listed.

| Primary ID     | Beta  | p-value   | FDR p-value | Bonferroni p-value |
|----------------|-------|-----------|-------------|--------------------|
| X1.17_564.3289 | -5.36 | 2.02E-08  | 3.52E-05    | 3.52E-05           |
| X7.15_786.5626 | -8.14 | 9.95E-07  | 8.68E-04    | 1.74E-03           |
| X1.42_566.3497 | -11.92| 3.06E-06  | 1.78E-03    | 5.35E-03           |
| X9.42_814.5927 | -15.22| 2.29E-05  | 8.85E-03    | 3.99E-02           |
| X1.51_592.3513 | -125.77| 2.99E-05  | 8.85E-03    |                    |
| X8.93_856.6036 | 5.17  | 3.04E-05  | 8.85E-03    |                    |
| X1.91_594.3757 | -67.46| 4.25E-05  | 1.06E-02    |                    |
| X11.11_1012.7635| -44.18| 7.56E-05  | 1.58E-02    |                    |
| X5.90_810.5600 | -17.97| 8.15E-05  | 1.58E-02    |                    |
| X1.03_562.3132 | -45.07| 1.19E-04  | 1.81E-02    |                    |
| X8.85_788.5789 | -13.63| 1.24E-04  | 1.81E-02    |                    |
| X8.61_766.5363 | 4.13  | 1.31E-04  | 1.81E-02    |                    |
| X5.43_1068.6601| -35.46| 1.35E-04  | 1.81E-02    |                    |
| X5.23_826.5592 | -7.76 | 1.53E-04  | 1.91E-02    |                    |
| X10.33_871.6904| -44.98| 1.67E-04  | 1.92E-02    |                    |
| X6.50_828.5729 | -2.15 | 1.82E-04  | 1.92E-02    |                    |
| X7.60_814.5916 | -16.59| 1.87E-04  | 1.92E-02    |                    |
| X10.16_816.6098| -32.01| 2.03E-04  | 1.97E-02    |                    |
| X1.59_554.3446 | -24.37| 2.32E-04  | 2.12E-02    |                    |
| X7.32_864.6056 | -24.59| 2.72E-04  | 2.12E-02    |                    |
| X0.99_586.3136 | -24.35| 2.77E-04  | 2.12E-02    |                    |
| X4.18_794.5050 | -33.65| 2.78E-04  | 2.12E-02    |                    |
| X9.01_1128.5388| 41.17 | 2.79E-04  | 2.12E-02    |                    |
| X8.90_880.6027 | 8.58  | 3.18E-04  | 2.31E-02    |                    |
| X3.52_421.3076 | 98.52 | 4.15E-04  | 2.90E-02    |                    |
| X9.14_790.5935 | -10.65| 4.36E-04  | 2.93E-02    |                    |
| X10.95_650.6069| 24.14 | 4.62E-04  | 2.99E-02    |                    |
| X10.11_1381.848| -42.51| 4.80E-04  | 2.99E-02    |                    |
| X1.08_612.3280 | -14.34| 5.93E-04  | 3.55E-02    |                    |
| X6.26_834.5625 | -16.98| 6.11E-04  | 3.55E-02    |                    |
| X10.16_826.6359| -30.64| 7.08E-04  | 3.98E-02    |                    |
| X7.02_772.5264 | -10.17| 7.36E-04  | 4.02E-02    |                    |
| X1.21_476.2768 | -12.46| 7.90E-04  | 4.18E-02    |                    |
| X7.47_788.5783 | -9.13 | 9.57E-04  | 4.91E-02    |                    |
| X8.61_1038.473 | 32.37 | 9.91E-04  | 4.94E-02    |                    |
Supplementary Table 4: Table of lipids significantly associated with serum leptin with FDR p-values < 0.05. Beta coefficients, p-values, Benjamini-Hochberg FDR p-values, and Bonferroni p-values listed.

| Primary ID     | Beta | p-value     | FDR p-value | Bonferroni p-value |
|----------------|------|-------------|-------------|--------------------|
| X1.17_564.3289 | -0.92| 2.85E-08    | 4.98E-05    | 4.98E-05           |
| X8.93_856.6036 | 1.02 | 1.65E-06    | 1.36E-03    | 2.88E-03           |
| X1.42_566.3497 | -2.10| 2.34E-06    | 1.36E-03    | 4.08E-03           |
| X1.51_592.3513 | -22.38| 1.92E-05  | 8.39E-03    | 3.36E-02           |
| X1.08_612.3280 | -3.03| 2.50E-05    | 8.74E-03    | 4.37E-02           |
| X1.03_562.3132 | -8.33| 4.10E-05    | 1.19E-02    |                   |
| X1.41_452.2762 | -6.39| 4.85E-05    | 1.19E-02    |                   |
| X0.99_586.3136 | -4.66| 5.80E-05    | 1.19E-02    |                   |
| X9.01_1128.5388| 7.86 | 6.13E-05    | 1.19E-02    |                   |
| X7.15_786.5626 | -1.17| 7.19E-05    | 1.25E-02    |                   |
| X10.33_871.6904| -8.00| 1.19E-04    | 1.89E-02    |                   |
| X5.43_1068.6601| -6.19| 1.31E-04    | 1.90E-02    |                   |
| X1.91_594.3757 | -10.83| 1.75E-04  | 2.22E-02    |                   |
| X5.90_810.5600 | -2.99| 1.78E-04    | 2.22E-02    |                   |
| X8.90_880.6027 | 1.52 | 2.36E-04    | 2.75E-02    |                   |
| X1.21_476.2768 | -2.35| 2.60E-04    | 2.84E-02    |                   |
| X10.53_854.6676 | -2.10| 4.62E-04    | 4.70E-02    |                   |
| X7.32_864.6056 | -4.11| 4.89E-04    | 4.70E-02    |                   |
| X9.66_768.5494 | 3.00 | 5.11E-04    | 4.70E-02    |                   |
| X8.61_766.5363 | 0.65 | 5.56E-04    | 4.85E-02    |                   |
Supplementary Table 5: Table of lipids significantly associated with serum total adiponectin with FDR p-values <0.05. Beta coefficients, p-values, Benjamini-Hochberg FDR p-values, and Bonferroni p-values listed.

| Primary ID     | Beta | p-value  | FDR p-value | Bonferroni p-value |
|----------------|------|----------|-------------|--------------------|
| X1.17_564.3289 | 0.39 | 1.14E-07 | 1.98E-04    | 1.98E-04           |
| X7.15_786.5626 | 0.61 | 1.93E-06 | 1.31E-03    | 3.36E-03           |
| X1.42_566.3497 | 0.93 | 2.26E-06 | 1.31E-03    | 3.94E-03           |
| X4.18_794.5050 | 3.17 | 5.94E-06 | 2.59E-03    | 1.04E-02           |
| X1.51_592.3513 | 10.15| 1.05E-05 | 3.66E-03    | 1.83E-02           |
| X8.62_886.4824 | -4.53| 3.83E-05 | 1.11E-02    | 1.12E-02           |
| X6.50_828.5729 | 0.18 | 4.49E-05 | 1.12E-02    | 1.12E-02           |
| X5.23_826.5592 | 0.63 | 6.07E-05 | 1.32E-02    | 1.32E-02           |
| X1.03_562.3132 | 3.49 | 1.04E-04 | 2.01E-02    | 2.01E-02           |
| X1.21_476.2768 | 1.09 | 1.21E-04 | 2.11E-02    | 2.11E-02           |
| X8.61_912.4667 | -2.37| 1.58E-04 | 2.50E-02    | 2.50E-02           |
| X4.99_859.5280 | 1.27 | 1.94E-04 | 2.71E-02    | 2.71E-02           |
| X1.43_702.3160 | 2.36 | 2.08E-04 | 2.71E-02    | 2.71E-02           |
| X1.49_478.2927 | 1.48 | 2.18E-04 | 2.71E-02    | 2.71E-02           |
| X1.16_700.3013 | 1.79 | 2.50E-04 | 2.91E-02    | 2.91E-02           |
| X5.23_1098.4881| 1.59 | 2.73E-04 | 2.97E-02    | 2.97E-02           |
| X8.44_716.5227 | -1.42| 3.36E-04 | 3.26E-02    | 3.26E-02           |
| X5.22_1030.5052| 2.71 | 3.52E-04 | 3.26E-02    | 3.26E-02           |
| X8.61_980.4565 | -2.94| 3.55E-04 | 3.26E-02    | 3.26E-02           |
| X8.61_896.4927 | -2.09| 4.81E-04 | 4.19E-02    | 4.19E-02           |
| X1.22_544.2648 | 3.15 | 5.66E-04 | 4.70E-02    | 4.70E-02           |
| X7.47_788.5783 | 0.73 | 6.02E-04 | 4.77E-02    | 4.77E-02           |
Supplementary Table 6: Table of lipids significantly associated with serum C-peptide with FDR p-values <0.05. Beta coefficients, p-values, Benjamini-Hochberg FDR p-values, and Bonferroni p-values listed.

| Primary ID    | Beta | p-value  | FDR p-value | Bonferroni p-value |
|---------------|------|----------|-------------|--------------------|
| X10.09_880.5267 | 2.95 | 8.88E-06 | 6.25E-03    | 1.55E-02           |
| X10.09_744.5522 | 1.04 | 1.01E-05 | 6.25E-03    | 1.76E-02           |
| X9.66_768.5494 | 2.14 | 1.07E-05 | 6.25E-03    | 1.87E-02           |
| X7.95_790.5381 | 1.01 | 5.18E-05 | 1.68E-02    |                    |
| X8.44_716.5227 | 2.05 | 5.61E-05 | 1.68E-02    |                    |
| X8.61_1038.473 | 3.89 | 5.76E-05 | 1.68E-02    |                    |
| X1.42_566.3497 | -1.00| 1.10E-04 | 2.61E-02    |                    |
| X8.61_766.5363 | 0.41 | 1.23E-04 | 2.61E-02    |                    |
| X1.17_564.3289 | -0.38| 1.34E-04 | 2.61E-02    |                    |
| X7.15_786.5626 | -0.62| 2.51E-04 | 4.37E-02    |                    |
| X8.93_856.6036 | 0.45 | 2.83E-04 | 4.49E-02    |                    |
Supplementary Table 7: Table of primary IDs with Radii >0.1 from singular value decomposition biplot.

| Primary ID        | Vector 1 | Vector 2 | Radius |
|-------------------|----------|----------|--------|
| X12.43_934.7918   | -0.19    | 0.13     | 0.24   |
| X8.17_830.6155    | 0.03     | 0.22     | 0.22   |
| X1.51_592.3513    | 0.07     | 0.20     | 0.22   |
| X12.14_850.7682   | 0.20     | -0.05    | 0.20   |
| X8.24_880.6145    | -0.08    | -0.18    | 0.20   |
| X3.52_421.3076    | -0.09    | -0.13    | 0.16   |
| X1.35_669.3989    | 0.16     | 0.02     | 0.16   |
| X10.21_1048.6206  | -0.08    | 0.12     | 0.15   |
| X3.45_269.1316    | 0.13     | 0.00     | 0.13   |
| X5.27_1031.496    | -0.05    | 0.12     | 0.13   |
| X12.88_922.7355   | 0.12     | -0.05    | 0.13   |
| X8.28_814.6742    | -0.05    | -0.12    | 0.13   |
| X5.23_964.5107    | -0.05    | 0.11     | 0.12   |
| X1.57_466.3290    | 0.12     | -0.02    | 0.12   |
| X13.87_920.717    | 0.03     | -0.11    | 0.12   |
| X1.2_488.2839     | -0.01    | 0.11     | 0.12   |
| X9.03_842.5446    | 0.02     | 0.11     | 0.11   |
| X14.35_1062.754   | -0.08    | -0.08    | 0.11   |
| X10.38_1017.6967  | 0.10     | 0.06     | 0.11   |
| X9.96_1034.5762   | -0.11    | 0.00     | 0.11   |
| X0.99_963.3915    | -0.10    | -0.05    | 0.11   |
| X1.93_419.3020    | 0.09     | -0.05    | 0.10   |
| X8.87_1060.5423   | -0.10    | -0.02    | 0.10   |
| X1.91_594.3757    | 0.03     | 0.10     | 0.10   |
| X3.52_459.3291    | 0.08     | -0.06    | 0.10   |
Supplementary Table 8. Beta coefficients and Bonferroni corrected p-values calculated from the regression of principal component 4 scores (n=126) with 1,745 plasma lipid metabolites.

### Positive Betas

| Metabolite      | Beta | Bonferroni p-value |
|----------------|------|--------------------|
| X13.10_920.7410| 3.54 | 3.63E-05           |
| X8.23_950.6408 | 3.53 | 4.02E-04           |
| X13.07_988.7940| 3.36 | 2.29E-02           |
| X9.96_1034.5762| 2.46 | 2.48E-06           |
| X14.31_881.7973| 2.38 | 3.74E-04           |
| X13.11_919.7343| 2.26 | 5.00E-04           |
| X14.38_880.7349| 2.19 | 4.18E-05           |
| X14.33_869.3325| 2.05 | 1.64E-08           |
| X14.35_1062.754 | 1.99 | 4.74E-05           |
| X2.65_255.2331  | 1.98 | 1.59E-06           |

### Negative Betas

| Metabolite      | Beta | Bonferroni p-value |
|----------------|------|--------------------|
| X9.03_842.5446 | -1.76| 6.52E-05           |
| X11.20_1012.7670| -1.25| 7.79E-08           |
| X1.91_594.3757  | -1.17| 1.56E-03           |
| X8.67_864.6048  | -1.00| 2.63E-02           |
| X10.11_1381.848 | -0.92| 1.83E-04           |
| X10.33_871.6904 | -0.73| 4.82E-02           |
| X10.19_802.5695 | -0.71| 9.17E-08           |
| X4.37_881.5140  | -0.69| 1.06E-07           |
| X9.45_1261.8069 | -0.66| 4.26E-04           |
| X0.99_586.3136  | -0.65| 4.09E-09           |
Supplementary Table 9: Tables displaying MS/MS spectrum of primary ids (i.e., retention time \( m/z \)) generated by Progenesis QI that were significantly associated with responses, but were not listed in lipidomics databases and libraries.

| Primary ID: X1.35_669.3989 | Measured m/z (Da) | % base peak |
|-----------------------------|-------------------|-------------|
|                             | 174.9469          | 29.32       |
|                             | 193.1514          | 5.68        |
|                             | 218.0910          | 9.22        |
|                             | 231.1625          | 25.90       |
|                             | 257.1484          | 27.50       |
|                             | 275.1544          | 48.08       |
|                             | 277.1760          | 9.09        |
|                             | 395.2210          | 100.00      |
|                             | 396.2443          | 15.57       |
|                             | 669.3997          | 52.81       |
|                             | 670.4040          | 16.69       |
|                             | 671.3848          | 6.04        |

| Primary ID: X4.18_794.5050 | Measured m/z (Da) | % base peak |
|-----------------------------|-------------------|-------------|
|                             | 540.2896          | 1.21        |
|                             | 568.2869          | 0.92        |
|                             | 794.5059          | 100.00      |
|                             | 795.5094          | 46.56       |
|                             | 796.5105          | 18.17       |
|                             | 797.5401          | 3.34        |

| Primary ID: X5.43_1068.6601 | Measured m/z (Da) | % base peak |
|-----------------------------|-------------------|-------------|
|                             | 221.0662          | 4.70        |
|                             | 536.5089          | 2.45        |
|                             | 698.5816          | 7.19        |
|                             | 1022.6674         | 100.00      |
|                             | 1023.6949         | 61.47       |
|                             | 1024.6909         | 21.31       |
|                             | 1025.7070         | 2.86        |
|                             | 1068.7111         | 3.03        |
Primary ID: 10.11_1381.8480

| Measured m/z (Da) | % base peak |
|------------------|-------------|
| 755.6339         | 2.65        |
| 1335.8094        | 100.00      |
| 1336.8085        | 82.18       |
| 1337.8382        | 31.18       |
| 1381.8666        | 8.81        |
| 1382.8512        | 14.69       |

All MS/MS data were performed on samples with the most abundant parent ion(s) of interest, respectively, using the same chromatography column, solvent gradient, and mass analyzer setting. However, the collision energy used for parent ions MS/MS was increased to a ramp of 20-80V. Spectrum lists display all m/zs with >250 counts, from MS/MS of parent ions.
| Primary ID | Common name | Molecular Ion (ESI-neg) | Theoretical m/z (Da) | Mass error (Da) |
|-----------|-------------|------------------------|---------------------|----------------|
| X0.99_586.3136 | LPC(20:5) | [M+formate] | 586.3151 | 0.0015 |
| X1.03_562.3132 | LPC(18:3) | [M+formate] | 562.3151 | 0.0019 |
| X1.08_612.3280 | LPC(22:6) | [M+formate] | 612.3307 | 0.0027 |
| X1.17_564.3289 | LPC(18:2) | [M+formate] | 564.3307 | 0.0018 |
| X1.21_476.2768 | LPE(18:2) | [M-H] | 476.2783 | 0.0015 |
| X1.22_544.2648 | LPS(20:4) | [M-H] | 544.2681 | 0.0033 |
| X1.35_669.3989 | Unknown | - | | |
| X1.42_566.3497 | LPC(18:1) | [M+formate] | 566.3464 | 0.0033 |
| X1.49_478.2927 | LPE(18:1) | [M-H] | 478.2939 | 0.0012 |
| X1.51_592.3513 | LPC(20:2) | [M+formate] | 592.362 | 0.0107 |
| X1.59_554.3446 | LPC(17:0) | [M+formate] | 554.3464 | 0.0018 |
| X1.91_594.3757 | LPC(20:1) | [M+formate] | 594.3777 | 0.002 |
| X2.65_255.2331 | Palmitic acid | [M-H] | 255.233 | 0.0001 |
| X3.52_421.3076 | TLTI | - | | |
| X4.18_794.5050 | Unknown | - | | |
| X4.37_881.5140 | PI(16:0_22:6) | [M-H] | 881.5186 | 0.0046 |
| X5.23_826.5592 | PC(18:2_18:2) | [M+formate] | 826.5604 | 0.0012 |
| X5.43_1068.6601 | Unknown | - | | |
| X5.90_810.5600 | PC(O-16:0_20:5) | [M+formate] | 810.5617 | 0.0022 |
| X6.50_828.5729 | PC(18:1_18:2) | [M+formate] | 828.576 | 0.0031 |
| X7.15_786.5626 | PC(O-16:1_18:2) | [M+formate] | 786.5655 | 0.0029 |
| X7.32_864.6056 | PC(O-18:1_22:5) | [M+formate] | 864.6073 | 0.0037 |
| X7.47_788.5783 | PC(O-16:0_18:2) | [M+formate] | 788.5804 | 0.0023 |
| X8.17_830.6155 | PC(18:0_18:2) | [M+formate] | 830.5917 | 0.0238 |
| X8.23_950.6408 | TLTI | - | | |
| X8.24_880.6145 | PC(20:1_20:4) | [M+formate] | 880.6159 | 0.0024 |
| X8.44_716.5227 | PE(16:0_18:1) | [M-H] | 716.5236 | 0.0009 |
| X8.61_1038.473 | TLTI | - | | |
| X8.61_766.5363 | PE(18:0_20:4) | [M-H] | 766.5392 | 0.0029 |
| X8.67_864.6048 | PC(O-18:0_22:6) | [M+formate] | 864.6124 | 0.0076 |
| X8.90_880.6027 | PC(18:0_22:5) | [M+formate] | 880.6039 | 0.0027 |
| X8.93_856.6036 | PC(18:0_20:3) | [M+formate] | 856.6073 | 0.0037 |
| X9.01_1128.5388 | TLTI | - | | |
| X9.03_842.5446 | TLTI | - | | |
| X9.42_814.5927 | PC(O-18:1_18:2) | [M+formate] | 814.5968 | 0.0041 |
| X9.45_1261.8069 | TLTI | - | | |
| X9.66_768.5494 | PE(18:0_20:3) | [M-H] | 768.5549 | 0.0055 |
| X9.96_1034.5762 | TLTI | - | | |
| X10.11_1381.8480 | Unknown | - | | |
| X10.19_802.5695 | TLTI | - | | |
| X10.33_871.6904 | SM(d19:1/24:1) | [M+formate| 871.691 | 0.0006 |
|----------------|----------------|----------------|---------|-------|
| X11.20_1012.7670 | TLTI | - |
| X12.14_850.7682 | TLTI | - |
| X12.43_934.7918 | TLTI | - |
| X13.07_988.7940 | TLTI | - |
| X13.10_920.7410 | TLTI | - |
| X13.11_919.7343 | TLTI | - |
| X14.31_881.7973 | TLTI | - |
| X14.33_869.3325 | TLTI | - |
| X14.35_1062.754 | TLTI | - |
| X14.38_880.7349 | TLTI | - |
### Supplementary Information

#### Supplementary Table 11: Relationship between untargeted PL FA chains and the % of geometric and positional FA isomers in plasma PL

| Common name* | Palmitic<sup>a</sup> C16:0 | Stearic<sup>a</sup> C18:0 | Oleic<sup>a</sup> c-C18:1 | Elaidic<sup>a</sup> t-C18:1 | Eicosenoic<sup>a</sup> C20:1 | Linoleic<sup>a</sup> c-C18:2 | Linolealidic<sup>a</sup> t-C18:2 | ALA<sup>a</sup> C18:3 | Eicosadienoic<sup>a</sup> b C20:2 | DGLA<sup>a</sup> C20:3 | ARA<sup>a</sup> C20:4 | EPA<sup>a</sup> C20:5 | DPA<sup>ω-3b</sup> C22:5 | DPA<sup>ω-6b</sup> C22:6 | DHA<sup>a</sup> C22:6 | Nervonic<sup>a</sup> b C24:1 |
|-------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| LPC(20:5)   | -               | -               | -               | -               | -               | -               | -               | 0.28            | -               | -               | -               | -               | -               | -               | 0.61            | -               | -               |
| LPC(18:3)   | -               | -               | -               | -               | -               | 0.55            | -0.03           | -               | -               | -               | -               | -               | -               | -               | 0.60            | -               | -               |
| LPC(22:6)   | -               | -               | -               | -               | 0.53            | 0.05            | -0.01           | -               | -               | -               | -               | -               | -               | 0.39            | -               | -               |
| LPC(18:2)   | -               | -               | -               | -               | 0.19            | 0.18            | -               | -               | -               | -               | -               | -               | -               | -               | -               | -               |
| LPC(17:0)   | -               | -               | -               | -               | -               | -               | -               | -               | -               | -               | -               | -               | -               | -               | -               | -               |
| LPC(18:1)   | -               | 0.34            | 0.06            | -               | 0.53            | -0.17           | -               | -               | -               | -               | -               | -               | -               | 0.24            | -               | -               |
| LPC(20:2)   | -               | -               | -               | -               | 0.77            | 0.54            | -0.20           | -               | -               | -               | -               | -               | -               | -               | 0.48            | -               | -               |
| LPC(20:0)   | -               | 0.43            | 0.28            | -               | 0.26            | -0.18           | -               | -               | -               | -               | -               | -               | -               | -               | 0.24            | -               | -               |
| LPC(18:1,18:2) | 0.34      | 0.20            | 0.04            | 0.12            | 0.34            | 0.24            | 0.05            | 0.37            | -               | -               | -               | -               | -               | -               | 0.56            | -               | -               |
| LPC(18:1,18:2) | 0.34      | 0.20            | 0.04            | 0.12            | 0.34            | 0.24            | 0.05            | 0.37            | -               | -               | -               | -               | -               | -               | 0.56            | -               | -               |
| LPC(18:1,18:2) | 0.34      | 0.20            | 0.04            | 0.12            | 0.34            | 0.24            | 0.05            | 0.37            | -               | -               | -               | -               | -               | -               | 0.56            | -               | -               |
| LPC(18:1,18:2) | 0.34      | 0.20            | 0.04            | 0.12            | 0.34            | 0.24            | 0.05            | 0.37            | -               | -               | -               | -               | -               | -               | 0.56            | -               | -               |

Spearman correlations were performed to determine the relationship between the abundance of each plasma PL significantly associated with the responses in our study and the % of FA isomers in plasma PL from our previous study. For instance, LPC(18:2)
and LPE(18:2) were significantly correlated with the % of LA (ω-6, C18:2Δ9Z,12Z), whereas LPC(18:2) and LPE(18:2) were not correlated with the % of trans-isomer linoelaidic acid (ω-6, C18:2Δ9E,12E). Numeric values represent the Spearman correlation coefficients and are bolded if p<0.05. The plasma analyzed from both data sets were collected at the same time point from the same patients.

a The significant PL associated with the responses that were structurally characterized in our study. The experimental methodology employed was the UPLC-ESI-MS^E analysis of crude lipid extracts outlined in this manuscript.

b The geometric and positional FA isomers that were targeted in our previous study (Pickens et al. PLEFA. 2015.). The experimental methodology employed was the FAME analysis of isolated plasma PL by GC-FI.
Supplementary Information

Supplementary Table 12: R code used in statistical analyses presented in the manuscript.

```r
################ Loading Data ################
load("human.data.rda") # Object H. Identifier data of 126 study participants, includes: age, smoking, BMI, WC, # serum markers
dim(H) # 126 rows
load("IS mean imputed.rda") # Data matrix of internal standard normalized mean imputed lipidomic data
dim(IS.mean) # 126 rows and 1745 lipids
all.equal.character(rownames(H), rownames(IS.mean)) #The identifier data for each patient is aligned with their # lipidomic data

################ Scaling of Data ################
##### Pareto Scaling of Internal Standard Normalize Mean imputed Data #####
install.packages("MetabolAnalyze")
library(MetabolAnalyze) # Package used for pareto scaling.
   # https://cran.r-project.org/web/packages/MetabolAnalyze/index.html
pareto.IS = scaling(as.matrix(scale(IS.mean, center=T, scale=F)), type = "pareto") # Center and pareto scale the internal standard normalized mean imputed data.
dim(pareto.IS) # 126 rows and 1745 lipids
all.equal.character(rownames(H), rownames(pareto.IS)) # The identifier data for each patient is aligned with their # scaled lipidomic data

################ ANOVA and Post Hoc ################
##### Kruskal Wallis (KW) one-way ANOVA was conducted across BMI categories along # with Dunn's test for multiple comparison #####

### KW one-way ANOVA ###
kwpv=numeric() # Numeric vector for indexing KW ANOVA p-values
## Loop for KW ANOVA of age, BMI, WC, and serum adipokines, cytokines, and C-peptide # across BMI category

for (i in 1:ncol(H)){
   kwpv[i]=kruskal.test(x=H[,i], g=HSBMI.cat)$p.value # Indexing the p-value from each ANOVA
}
kwbh=p.adjust(kwpv, method = "BH") # Adjusting KW p-values according to Benjamini-Hochberg

##### Dunn non-parametric test for multiple comparison #####
library(PMCMR)
tmp=matrix(NA, ncol = 4, nrow = ncol(H))
colnames(tmp)=c("var","lean-overwt","lean-obese", "overwt-obese")
tmp[,1]=colnames(H)

for (i in 1:ncol(H)){
tmpindex=posthoc.kruskal.dunn.test(x=H[,i], g=HSBMI.cat, p.adjust.method = "BH")
tmpgetp=get.pvalues(tmpindex)
```
tmp[i,2]=tmpgetp[[1]]
tmp[i,3]=tmpgetp[[2]]
tmp[i,4]=tmpgetp[[3]]

#############################################################

Bayesian Analysis of Traits

#### Reproducing Kernel Hilbert Spaces (RKHS) Regressions ####
install.packages("BGLR")
library(BGLR)
# For more information on using BGLR refer to https://github.com/gdlc/BGLR-

### Creating a directory to store file outputs from BGLR ###
dir.create("BMI"); setwd("BMI")
getwd() # Verify working directory

### Setting response, lipidomic data, iterations, and burn in ###
X=pareto.IS # 1,745 pareto scaled lipidomic data
y=H$BMI # Response: body mass index values for each respective patient
nIter=200000 # Long Markov Chain of 200,000 iterations
burnIn=50000 # Number of iterations to discard for burn in

## Computing the metabolomic similarity matrix ##
L=sum(apply(X, FUN=var, MARGIN=2))
G=tcrossprod(X)/L
# The G matrix is an nxn matrix of distances to measure similarities
# between participants with respect to their lipid profiles

## RKHS model parameters ##
# ETA #
ETA.FixMet=list(Met=list(K=G, model="RKHS"), # The G matrix represents the lipidome,
# and the RKHS kernel is specified
Fix=list(~HSage+factor(H$smoking), model="FIXED")) # The fixed effects of the model are age and smoking

## RKHS regression ##
fmGBLUP=BGLR(y=y,ETA=ETA.FixMet, nIter=nIter, burnln=burnln, saveAt="GBLUP_")
# RKHS model is: BMI = fixed effects + lipidomic data

### Variance of Best Linear Unbiased Predictor (BLUP) and variance of error ###
# inference was done based on one of every 5 samples of the last 150,000
# therefore, since the burn in was 50,000 we need to remove the first 10,000 samples
list.files()
VarU=scan("GBLUP_ETA_Met_varU.dat") #load in variance of the lipidome BLUP model (varU). Total of 30,000
# lines.
VarU=VarU[-c(1:10000)] # remove the burn in iterations from varU
VarE=scan("GBLUP_varE.dat") # load in variance of the error (varE). Total of 30,000 lines.
VarE=VarE[-c(1:10000)] # remove the burn in iterations varE

## Calculating percent of the inter-individual differences in response variables that can be
# attributed to lipidome profiles ##
tmp=(VarU/(VarU+VarE))*100 # Calculate % varU as sum of varU + varE
round(quantile(tmp, probs = c(0.025, 0.975)),0) # Determine 95% confidence intervals

#### The other responses were run similarly by creating individual directories for each response and modifying
# the y<- for each individual response  #######

##############################
### Single Lipid Regressions ###
##############################

### Setting the model response and independent variables ###
X=pareto.IS # Data set
XX=H$BMI # Response, dependent variable

### Creating numeric vectors for indexing estimated effect and model p-value ###
BMI.mypv=numeric(ncol(X)) # Numeric variable for indexing each p-value for each lipid
BMI.myB=numeric(ncol(X)) # Beta coefficient for each lipid

### Loop of single marker regressions of pareto scaled data ###
for (i in 1:ncol(X)){ # For each column of data matrix
  tmp=summary(lm(XX~H$age + H$smoking + X[,i])) # list of summary of each linear model
  BMI.mypv[i]=tmp$coefficients[5,4] # Indexing p-value
  BMI.myB[i]=tmp$coefficients[5,1] # Indexing beta coefficient
}

### P-value adjustments ###
##  Bonferroni pvalue correction  ##
p.bon=p.adjust(BMI.mypv, method='bonferroni')

## Benjamini-Hochberg pvalue correction ##
p.BH=p.adjust(BMI.mypv, method='BH')

### Creating a dataframe of regression results ###
Complete.Regression.Report=data.frame('Metabolite'=colnames(X), BMI.stats.reportBMI.myB, BMI.mypv, 'BMI.BH'=p.BH, 'BMI.bon'=p.bon) # Compile all Response beta coefficients, p-values, and corrected p-values

#### The other responses were run similarly by modifying the response input (i.e., either BMI, WC, leptin, etc) # and uniquely indexing model outputs and compiling their results into the file Complete.Regression.Report #####

####################################################################
### Singular Value Decomposition ###
####################################################################

load("beta.report.rda")
# Beta.Report is a dataframe with 1745 rows and 10 columns
# Column 1 contains the name of each of the 1745 lipids
# Columns 2-10 contain the 1745 estimated effects for each respective response (i.e., BMI, WC, 
# leptin, etc)

# Singular value decompositions (SVD) is defined as
# A= U*D*V
# where U is an m x m matrix, D is an m x n, and V is an n x n matrix

### Singular value decomposition of traits ###
pca.beta=princomp(Beta.Report[,2:ncol(Beta.Report)])
lambda=pca.beta$sdev*sqrt(pca.beta$n.obs)
score=t(t(pca.beta$scores)/lambda)
variables=t(t(pca.beta$sloadings)*lambda)
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v.1=variables
v.1[8,1]=v.1[8,1]/25 # scaling MCP-1 PC1 score by a factor of 25

### Setting response and computing principal components (PCs) ###
XX=HSBM # Response, dependent variable to be used in regressions
Z=crossprod(pareto.IS) # Computing an nxn matrix of lipidomic data
ZZ=eigen(Z) # Computing Eigen values and Eigen vectors of nxn matrix of lipidomic data
EV=(ZZ$values/sum(ZZ$values))*100 # Calculating the Eigen value % of total variation

### Creating vectors to index pvalue and beta coefficient
PCA.mypv=numeric(ncol(Z[,1:125])) # p-value for each metabolite
PCA.myB=numeric(ncol(Z[,1:125])) # Beta coefficient for each metabolite

### Loop of BMI regressed on each PC individually ###
for (i in 1:125) {
  tmp=summary(lm(XX~H$age + H$smoking + ZZ$vectors[,i])) # List of outputs from linear model
  PCA.mypv[i]=tmp$coefficients[4,4] # Indexing the p-value from linear model
  PCA.myB[i]=tmp$coefficients[4,1] # Indexing the beta coefficients from linear model
}

### P-value adjustments ###
# Bonferroni pvalue correction
PCA.pbon=p.adjust(PCA.mypv, method='bonferroni')

# Benjamini-Hochberg pvalue correction
PCA.pBH=p.adjust(PCA.mypv, method='BH')

### Setting model response and creating vectors for indexing ###
Y=ZZ$vectors[,4] # Response, dependent variable is PC4 loadings
b=numeric() # Numeric vector for indexing beta coefficients
pval=numeric() # Numeric vector for indexing p-values

### Loop of PC4 scores regressed on each lipid individually ###
for (i in 1:ncol(pareto.IS)) {
  tmp=summary(lm(Y~pareto.IS[,i])) # List of summary of each linear model
  b[i]=tmp$coefficients[2,1] # Indexing beta coefficients
  pval[i]=tmp$coefficients[2,4] # Indexing p-values
}

### P-value adjustments ###
# Bonferroni pvalue correction
p.bon=p.adjust(pval, method='bonferroni')

# Benjamini-Hochberg pvalue correction
p.BH=p.adjust(pval, method='BH')

all.equal(rownames(H),rownames(pareto.IS))

X=data.frame(pareto.IS)

# X0.99_586.3136 LPC(20:5)
cor.test(X$X0.99_586.3136, H$EPA, method="spearman")
summary(lm(log(X$X0.99_586.3136)~log(H$EPA) + H$BMI))

# X1.03_562.3132 LPC(18:3)
cor.test(X$X1.03_562.3132, H$ALinolenic, method="spearman")
summary(lm(log(X$X1.03_562.3132)~log(H$ALinolenic) + H$BMI))

# X1.08_612.3132 LPC(18:2)
cor.test(X$X1.08_612.3132, H$Linoleic, method="spearman")
cor.test(X$X1.08_612.3132, H$Linoelaidic, method="spearman")
summary(lm(log(X$X1.08_612.3132)~log(H$Linoleic) + H$BMI))
summary(lm(log(X$X1.08_612.3132)~log(H$Linoelaidic) + H$BMI))

# X1.17_564.3289 LPE(18:2)
cor.test(X$X1.17_564.3289, H$Linoleic, method="spearman")
cor.test(X$X1.17_564.3289, H$Linoelaidic, method="spearman")
summary(lm(log(X$X1.17_564.3289)~log(H$Linoleic) + H$BMI))
summary(lm(log(X$X1.17_564.3289)~log(H$Linoelaidic) + H$BMI))

# X1.21_476.2768 LPE(18:1)
cor.test(X$X1.21_476.2768, H$Linoleic, method="spearman")
cor.test(X$X1.21_476.2768, H$Linoelaidic, method="spearman")
summary(lm(log(X$X1.21_476.2768)~log(H$Linoleic) + H$BMI))
summary(lm(log(X$X1.21_476.2768)~log(H$Linoelaidic) + H$BMI))

# X1.22_544.2648 LPS(20:4)
cor.test(X$X1.22_544.2648, H$OA, method="spearman")
summary(lm(log(X$X1.22_544.2648)~log(H$OA) + H$BMI))

# X1.42_566.3497 LPC(18:1)
cor.test(X$X1.42_566.3497, H$Oleic, method="spearman")
cor.test(X$X1.42_566.3497, H$Elaidic, method="spearman")
summary(lm(log(X$X1.42_566.3497)~log(H$Oleic) + H$BMI))
summary(lm(log(X$X1.42_566.3497)~log(H$Elaidic) + H$BMI))

# X1.49_478.2927 LPE(18:1)
cor.test(X$X1.49_478.2927, H$Oleic, method="spearman")
cor.test(X$X1.49_478.2927, H$Elaidic, method="spearman")
summary(lm(log(X$X1.49_478.2927)~log(H$Oleic) + H$BMI))
summary(lm(log(X$X1.49_478.2927)~log(H$Elaidic) + H$BMI))

# X1.51_592.3513 LPC(20:2)
cor.test(X$X1.51_592.3513, H$Eicosadienoic, method="spearman")
summary(lm(log(X$X1.51_592.3513)~log(H$Eicosadienoic) + H$BMI))
Supplementary Information

# X1.59_554.3446 LPC(17:0)
# We didn't target this

# X1.91_594.3757 LPC(20:1)
cor.test(X$X1.91_594.3757, HSEicosenoic, method="spearman")
summary(lm(log(X$X1.91_594.3757) ~ log(HSEicosenoic) + HSBMI))

# X2.65_255.2331 Palmitic acid
cor.test(X$X2.65_255.2331, HSPalmitic, method="spearman")
summary(lm(log(X$X2.65_255.2331) ~ log(HSPalmitic) + HSBMI))

# X4.37_881.5140 PI(16:0_22:6)
cor.test(X$X4.37_881.514, HSPalmitic, method="spearman")
cor.test(X$X4.37_881.514, HSDHA, method="spearman")
summary(lm(log(X$X4.37_881.514) ~ log(HSPalmitic) + HSBMI))
summary(lm(log(X$X4.37_881.514) ~ log(HSDHA) + HSBMI))

# X5.23_826.5592 PC(18:2_18:2)
cor.test(X$X5.23_826.5592, HSLinoleic, method="spearman")
cor.test(X$X5.23_826.5592, HLSinoelaidic, method="spearman")
summary(lm(log(X$X5.23_826.5592) ~ log(HSLinoleic) + HSBMI))
summary(lm(log(X$X5.23_826.5592) ~ log(HSinoelaidic) + HSBMI))

# X5.90_810.5600 PC(O-16:0_20:5)
cor.test(X$X5.9_810.56, HSEPA, method="spearman")
summary(lm(log(X$X5.9_810.56) ~ log(HSEPA) + HSBMI))

# X6.50_828.5729 PC(18:1_18:2)
cor.test(X$X6.5_828.5729, HSOleic, method="spearman")
cor.test(X$X6.5_828.5729, HSElaIdic, method="spearman")
cor.test(X$X6.5_828.5729, HLSinoelaidic, method="spearman")
summary(lm(log(X$X6.5_828.5729) ~ log(HSOleic) + HSBMI))
summary(lm(log(X$X6.5_828.5729) ~ log(HSElaIdic) + HSBMI))
summary(lm(log(X$X6.5_828.5729) ~ log(HSinoelaidic) + HSBMI))

# X7.15_786.5626 PC(O-16:1_18:2)
cor.test(X$X7.15_786.5626, HLSinoelaidic, method="spearman")
summary(lm(log(X$X7.15_786.5626) ~ log(HSinoelaidic) + HSBMI))

# X7.32_864.6056 PC(O-18:1_22:5)
cor.test(X$X7.32_864.6056, HSDPA3, method="spearman")
cor.test(X$X7.32_864.6056, HSDPA6, method="spearman")
summary(lm(log(X$X7.32_864.6056) ~ log(HSDPA3) + HSBMI))
summary(lm(log(X$X7.32_864.6056) ~ log(HSDPA6) + HSBMI))

# X7.47_788.5783 PC(O-16:0_18:2)
cor.test(X$X7.47_788.5783, HLSinoelaidic, method="spearman")
summary(lm(log(X$X7.47_788.5783) ~ log(HSinoelaidic) + HSBMI))
cor.test(X$X8.17_830.6155, H$Stearic, method="spearman")
cor.test(X$X8.17_830.6155, H$Linoleic, method="spearman")
cor.test(X$X8.17_830.6155, H$Linoleaidic, method="spearman")
summary(lm(log(X$X8.17_830.6155) ~ log(H$Stearic) + H$BMI))
summary(lm(log(X$X8.17_830.6155) ~ log(H$Linoleic) + H$BMI))
summary(lm(log(X$X8.17_830.6155) ~ log(H$Linoleaidic) + H$BMI))

# X8.24_880.6145 PC(20:1_20:4)
cor.test(X$X8.24_880.6145, H$Eicosenoic, method="spearman")
cor.test(X$X8.24_880.6145, H$AA, method="spearman")
cor.test(X$X8.24_880.6145, H$SAA, method="spearman")
summary(lm(log(X$X8.24_880.6145) ~ log(H$Eicosenoic) + H$BMI))
summary(lm(log(X$X8.24_880.6145) ~ log(H$AA) + H$BMI))
summary(lm(log(X$X8.24_880.6145) ~ log(H$SAA) + H$BMI))

# X8.44_716.5227 PE(16:0_18:1)
cor.test(X$X8.44_716.5227, H$Palmitic, method="spearman")
cor.test(X$X8.44_716.5227, H$OA, method="spearman")
cor.test(X$X8.44_716.5227, H$Stearic, method="spearman")
cor.test(X$X8.44_716.5227, H$Elaidic, method="spearman")
summary(lm(log(X$X8.44_716.5227) ~ log(H$Palmitic) + H$BMI))
summary(lm(log(X$X8.44_716.5227) ~ log(H$OA) + H$BMI))
summary(lm(log(X$X8.44_716.5227) ~ log(H$Stearic) + H$BMI))
summary(lm(log(X$X8.44_716.5227) ~ log(H$Elaidic) + H$BMI))

# X8.61_766.5363 PE(18:0_20:3)
cor.test(X$X8.61_766.5363, H$Stearic, method="spearman")
cor.test(X$X8.61_766.5363, H$Linoleic, method="spearman")
cor.test(X$X8.61_766.5363, H$SAA, method="spearman")
summary(lm(log(X$X8.61_766.5363) ~ log(H$Stearic) + H$BMI))
summary(lm(log(X$X8.61_766.5363) ~ log(H$Linoleic) + H$BMI))
summary(lm(log(X$X8.61_766.5363) ~ log(H$SAA) + H$BMI))

# X8.67_864.6048 PC(O-18:0_22:6)
cor.test(X$X8.67_864.6048, H$DHA, method="spearman")
summary(lm(log(X$X8.67_864.6048) ~ log(H$DHA) + H$BMI))

# X8.90_880.6027 PC(18:0_22:5)
cor.test(X$X8.90_880.6027, H$Stearic, method="spearman")
cor.test(X$X8.90_880.6027, H$DPA3, method="spearman")
cor.test(X$X8.90_880.6027, H$DPA6, method="spearman")
summary(lm(log(X$X8.90_880.6027) ~ log(H$Stearic) + H$BMI))
summary(lm(log(X$X8.90_880.6027) ~ log(H$DPA3) + H$BMI))
summary(lm(log(X$X8.90_880.6027) ~ log(H$DPA6) + H$BMI))

# X8.93_856.6036 PC(18:0_20:3)
cor.test(X$X8.93_856.6036, H$Stearic, method="spearman")
cor.test(X$X8.93_856.6036, H$DGLA, method="spearman")
summary(lm(log(X$X8.93_856.6036) ~ log(H$Stearic) + H$BMI))
summary(lm(log(X$X8.93_856.6036) ~ log(H$DGLA) + H$BMI))

# X9.42_814.5927 PC(O-18:1_18:2)
cor.test(X$X9.42_814.5927, H$Linoleaidic, method="spearman")
cor.test(X$X9.42_814.5927, H$Linoleic, method="spearman")
summary(lm(log(X$X9.42_814.5927) ~ log(H$Linoleaidic) + H$BMI))
summary(lm(log(X$X9.42_814.5927) ~ log(H$Linoleic) + H$BMI))

# X9.66_768.5494 PE(18:0_20:3)
cor.test(X$X9.66_768.5494, H$Stearic, method="spearman")
cor.test(X$X9.66_768.5494, H$DGLA, method="spearman")
summary(lm(log(X$X9.66_768.5494) ~ log(H$Stearic) + H$BMI))
summary(lm(log(X$X9.66_768.5494) ~ log(H$DGLA) + H$BMI))

Supplementary Information

9.42_814.5927
9.66_768.5494
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# X10.33_871.6904 SM(d19:1/24:1)
cor.test(X$X10.33_871.6904, H$Nervonic, method="spearman")
summary(lm(log(X$X10.33_871.6904)~ log(H$Nervonic) +H$BMI))