

Supplementary Materials

Figure S1. Partial Least Square Discriminant Analysis (PLS-DA) scores plot for male (+) and female (Δ) plasma sample with XN treatments. Ellipse shows the 95% confidence region.

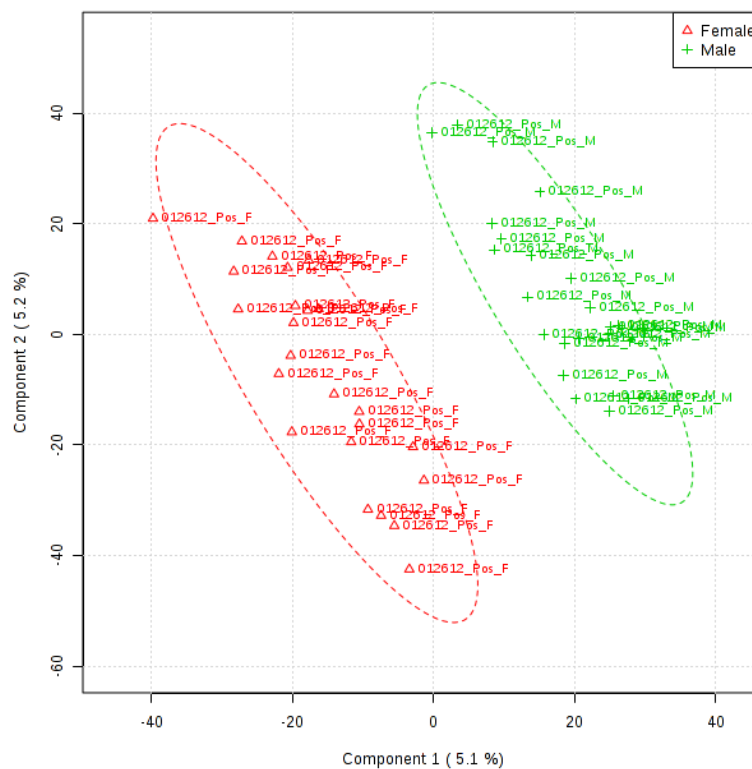


Figure S2. PLS-DA scores plot for control (Δ) and different dose groups low- x; medium- ◇; high-+ (plasma, male). Ellipse shows the 95% confidence region.

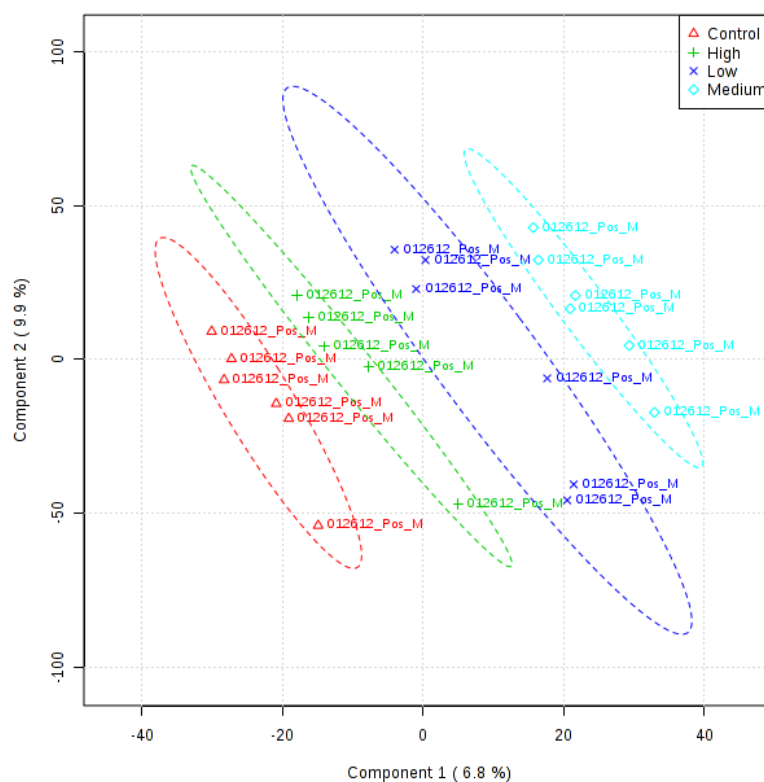


Figure S3. Volcano plots generated for control vs. high dose group (plasma, male) in negative ionization mode (left) and positive ionization mode (right). Each point represents a feature plotted as a function of fold-change (Log_2 (fold change), x -axis) and statistical significance ($-\text{Log}_{10}$ (p -value), y -axis). Vertical dotted lines represent fold changes of ± 1.5 . The horizontal dotted line corresponds to the minimum p -value of selected metabolites (p -value < 0.05). The pink dots represent selected putative markers p -value < 0.05 and fold change < 1.5 .

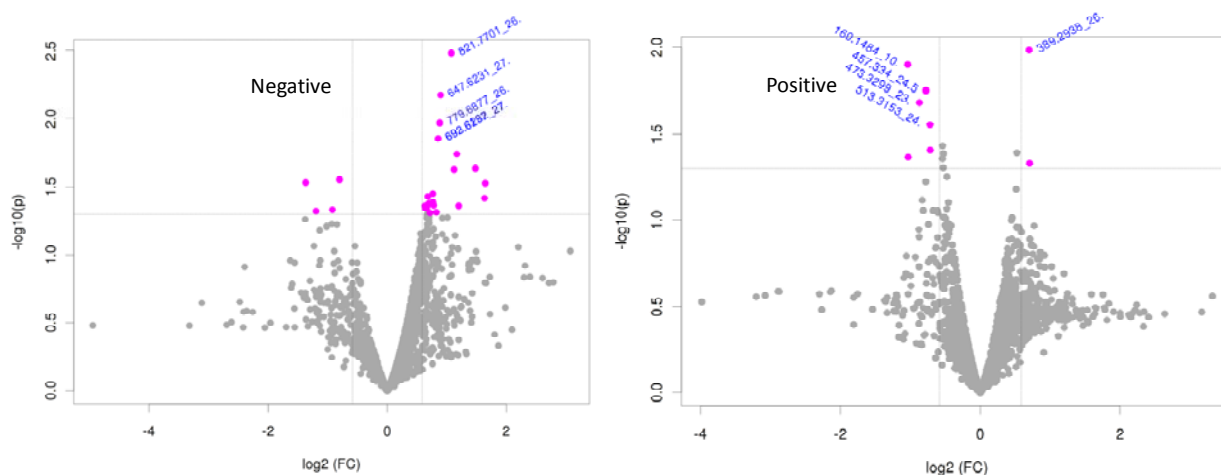


Table S1. Plasma differential metabolites obtained from Zucker (fa/fa) rats with (high dose) and without (control) xanthohumol treatment.

Metabolites	Mode	m/z	Molecular Formula	RT	Fold Change (T/C)	p-Value	Class
3-Methoxy-4-hydroxyphenylglycolaldehyde	Negative	181.0520	C ₉ H ₁₀ O ₄	12.40	3.3346	0.0349	Tyrosine metabolism
5-Hydroxyindoleacetate	Negative	190.0500	C ₁₀ H ₉ NO ₃	12.01	0.6377	0.0250	Tryptophan metabolism
Guanidinoethyl methyl phosphate	Negative	196.0479	C ₄ H ₁₂ N ₃ O ₄ P	19.96	0.4838	0.0224	Enzymatic rxn
Hexadecanedioic acid	Negative	285.2064	C ₁₆ H ₃₀ O ₄	24.66	1.5300	0.0392	Fatty Acyls [FA]
dUMP	Negative	307.0341	C ₉ H ₁₃ N ₂ O ₈ P	5.89	1.1287	0.0368	Pyrimidine metabolism
Octadecanedioic acid	Negative	313.2373	C ₁₈ H ₃₄ O ₄	25.22	1.6276	0.0262	Fatty Acyls [FA]
4-O-beta-D-Glucosyl-4-hydroxycinnamate	Negative	325.0953	C ₁₅ H ₁₈ O ₈	18.46	0.3965	0.0497	Enzymatic rxn in UDP-glucose
2,3-dinor Thromboxane B1	Negative	343.2117	C ₁₈ H ₃₂ O ₆	24.56	1.3326	0.0302	major metabolite of thromboxane B2
Sphingosine-1-phosphate	Negative	378.2398	C ₁₈ H ₃₈ NO ₅ P	24.27	1.2891	0.0477	Sphingolipids [SP]
PA(18:2/0:0)	Negative	433.2356	C ₂₁ H ₃₉ O ₇ P	25.31	0.6159	0.0276	Glycerophospholipids [GP]
PA(18:1/0:0)	Negative	435.2538	C ₂₁ H ₄₁ O ₇ P	25.07	1.5474	0.0498	Glycerophospholipids [GP]
PA(20:4/0:0)	Negative	457.2345	C ₂₃ H ₃₉ O ₇ P	27.14	1.1679	0.0133	Glycerophospholipids [GP]
PG(18:1/0:0)	Negative	509.2872	C ₂₄ H ₄₇ O ₉ P	24.61	1.2307	0.0500	Glycerophospholipids [GP]
24-methylcholest-22-en-3,4,5,6,8,14,15,25,28-nonol	Negative	527.3246	C ₂₈ H ₄₈ O ₉	25.63	0.6111	0.0167	Sterol Lipids [ST]
PI(18:3/0:0)	Negative	593.2734	C ₂₇ H ₄₇ O ₁₂ P	24.81	1.8090	0.0357	Glycerophospholipids [GP]
PA(O-16:0/18:3)	Negative	655.4662	C ₃₇ H ₆₉ O ₇ P	27.18	1.4419	0.0405	Glycerophospholipids [GP]
PA(O-16:0/18:2)	Negative	657.4854	C ₃₇ H ₇₁ O ₇ P	27.12	1.2782	0.0255	Glycerophospholipids [GP]
Anthenoside A	Negative	678.4571	C ₃₈ H ₆₅ NO ₉	26.38	0.6100	0.0302	Sterol Lipids [ST]
PS(18:4/22:6)	Negative	826.4635	C ₄₆ H ₇₀ NO ₁₀ P	26.70	0.8954	0.0489	Glycerophospholipids [GP]
PI(P-18:0/22:4)	Negative	897.5830	C ₄₉ H ₈₇ O ₁₂ P	24.80	1.6147	0.0472	Glycerophospholipids [GP]

Table S1. Cont.

Metabolites	Mode	m/z	Molecular Formula	RT	Fold Change (T/C)	p-Value	Class
11-amino-undecanoic acid	Positive	202.1817	C ₁₁ H ₂₃ NO ₂	27.09	0.8197	0.0480	Fatty Acyls [FA]
2-hydroxy-tridecanoic acid	Positive	231.1952	C ₁₃ H ₂₆ O ₃	26.98	0.8538	0.0329	Fatty Acids
Lauroyl-EA	Positive	244.2278	C ₁₄ H ₂₉ NO ₂	26.99	0.8831	0.0411	Fatty Acyls [FA]
Uridine	Positive	245.0775	C ₉ H ₁₂ N ₂ O ₆	13.55	1.0666	0.0216	Pyrimidine metabolism
N-(L-Arginino)succinate	Positive	291.1280	C ₁₀ H ₁₈ N ₄ O ₆	24.51	0.7727	0.0161	Alanine, aspartate and glutamate metabolism
(22)-1,22,25-trihydroxy-26,27-dimethyl-23,23,24,24-tetradehydrovitamin D3	Positive	457.3340	C ₂₉ H ₄₄ O ₄	24.57	1.5379	0.0389	Sterol Lipids [ST]
3-dehydroecdysone	Positive	463.3062	C ₂₇ H ₄₂ O ₆	26.35	0.6908	0.0224	Cholesterol
Cucurbitacin H	Positive	535.3276	C ₃₀ H ₄₆ O ₈	26.38	0.8705	0.0419	Sterol Lipids [ST]
PC(18:0/20:4(12OH[S]))	Positive	826.6013	C ₄₆ H ₈₄ NO ₉ P	26.36	0.6701	0.0154	Glycerophospholipids [GP]
PE(22:4/22:6)	Positive	840.5483	C ₄₉ H ₇₈ NO ₈ P	25.57	0.6860	0.0443	Glycerophospholipids [GP]
PS(20:1/22:6)	Positive	862.5590	C ₄₈ H ₈₀ NO ₁₀ P	25.57	0.6783	0.0364	Glycerophospholipids [GP]
TG(20:2/20:3/20:3)	Positive	959.7993	C ₆₃ H ₁₀₆ O ₆	22.39	0.8216	0.0375	Glycerolipids [GL]
PE(26:2/26:2)	Positive	964.7657	C ₅₇ H ₁₀₆ NO ₈ P	25.31	0.7242	0.0459	Glycerophospholipids [GP]

T/C- Treated vs Control, *m/z*- mass to charge ratio, PA-acylglycero phosphate, PC- phospho choline, PE-phospho ethonolamine, PG-phospho glycerol, PI-phospho inositol, PS-phospho serine, EA-ethanol amine, FA-fatty acid