Supplementary Information for:

Title: Metformin Monotherapy Alters the Human Plasma Lipidome Independent of Clinical Markers of Glycemic Control and Cardiovascular Disease Risk in a Type 2 Diabetes Clinical Cohort

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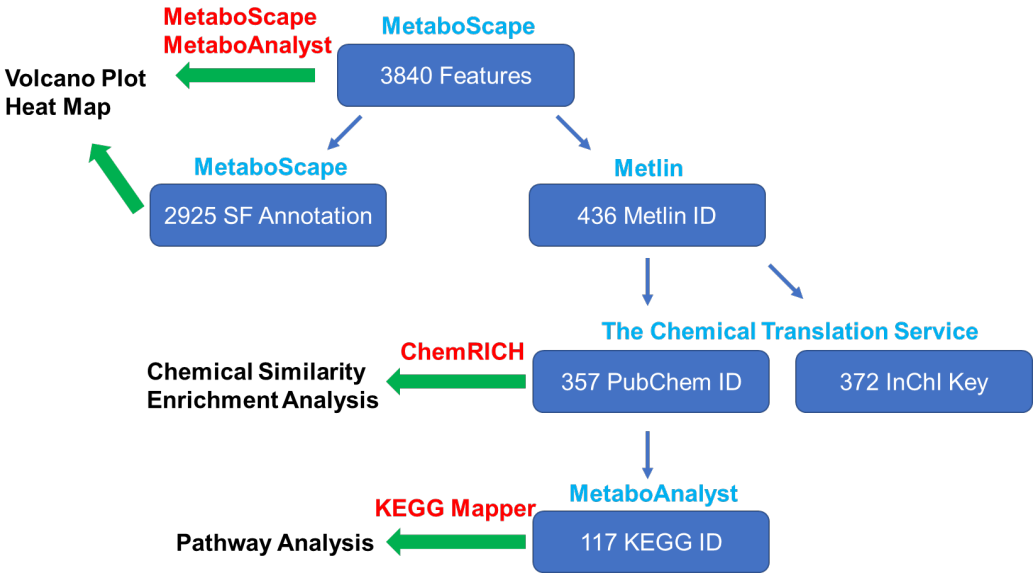

Figure S1: Overview of annotation workflow. Features were identified in MetaboScape. MetaboScape was used to annotate features by chemical formulas, and Metlin was used to annotate features by chemical name. The Chemical Translation Service was used to obtain chemical identifiers, PubChem ids, InChI keys, and KEGG ids of Metlin-annotated features. These chemical identifiers were used for MetaboAnalyst and ChemRich analyses.

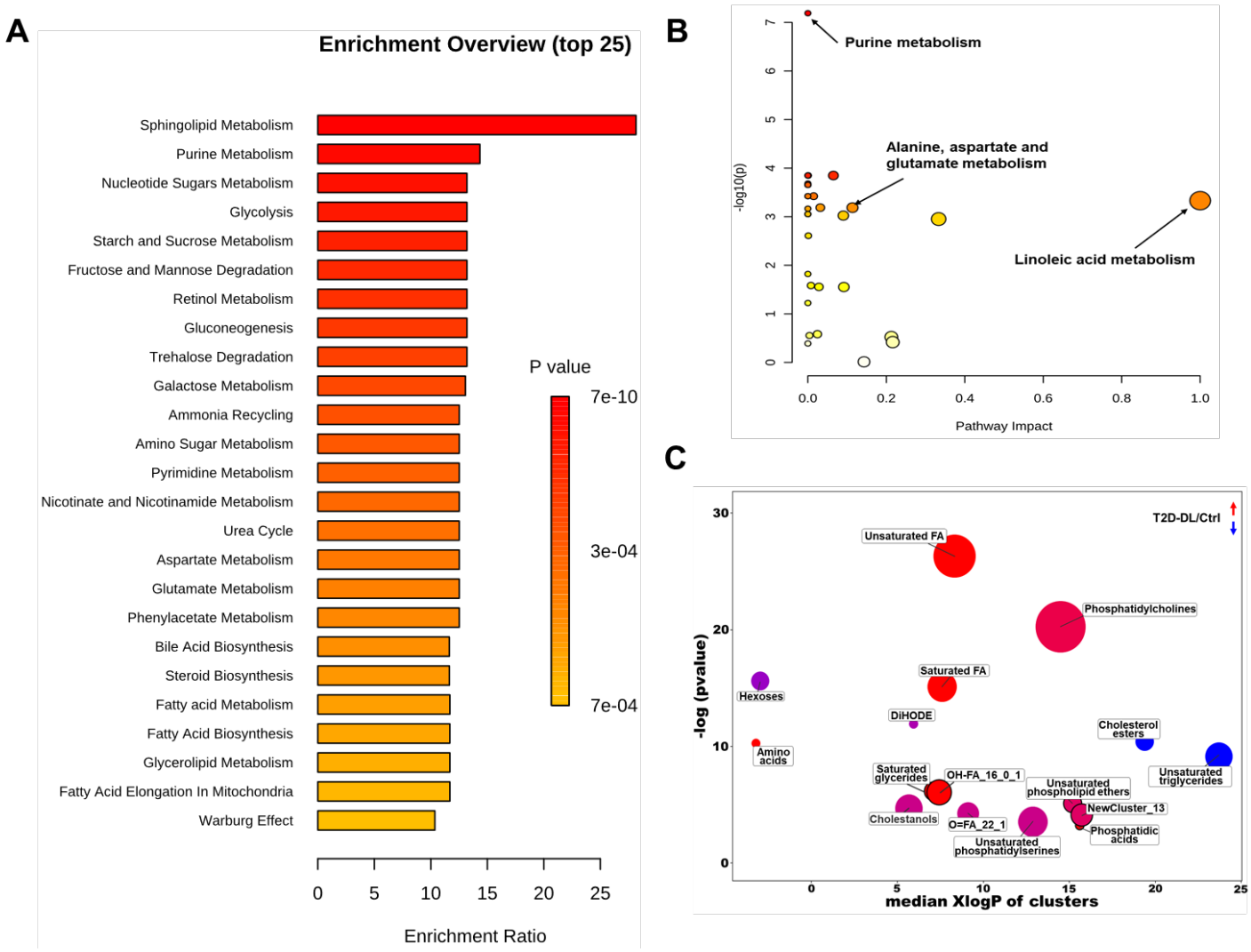

Figure S2. Metabolic differences between diet/lifestyle (T2D-DL) and control groups. **A**. Enrichment analysis of metformin group compared to control. **B**. Pathway analysis of the two groups with a gradient from red (smaller p-value) to yellow (larger p-value). **C**. ChemRich analysis with size of circle correlating to number of statistically significant features (p-value < 0.05) that are altered. Red is upregulated in the diet/lifestyle group, blue downregulated and purple contains upregulated and downregulated metabolites.

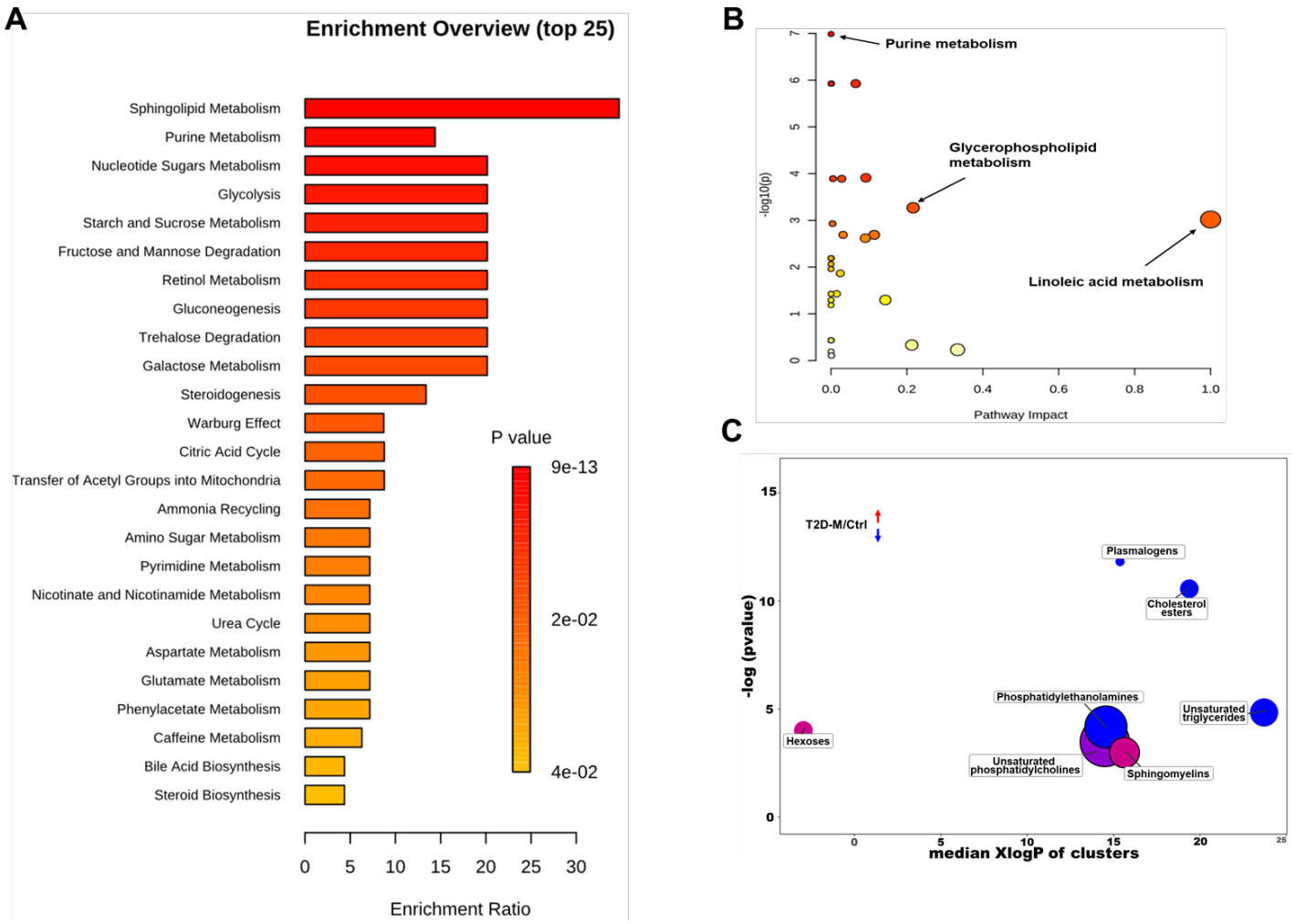

Figure S3. Metabolic differences between metformin (T2D-M) and control groups. **A**. Enrichment analysis of metformin group compared to control. **B**. Pathway analysis of the two groups with a gradient from red (smaller p-value) to yellow (larger p-value). **C**. ChemRich analysis with size of circle correlating to number of statistically significant features (p-value < 0.05) that are altered. Red is upregulated in the metformin group, blue downregulated and purple contains upregulated and downregulated metabolites.

Table S1. Significantly altered metabolites between T2D-DL (Diet and Lifestyle) and nondiabetic.

Class/Pathway and Metabolites	InChI Key	*p*(FDR)	Ratio (T2D-DL/ND)
Amino acids, peptides, and analogues			
Distichonic acid A	SEATYFZPTMHEIW-UHFFFAOYSA-N	0.0324	1.78
Glutamine	ZDXPYRJPNDTMRX-VKHMYHEASA-N	0.0022	1.26
Valine	KZSNJWFQEVHDMF-BYPYZUCNSA-N	0.0057	2.12
Anthracenes			
Palmidin C	VUUFXTUVVIEIMH-UHFFFAOYSA-N	0.0025	1.24
Bile acids, alcohols and derivatives			
27-Nor-5b-cholestane-3a,7a,12a,24,25-pentol	UBCPEZWGUOSYHO-JAPSQKQGSA-N	0.0390	1.81
Castasterone	VYUIKSFYFRVQLF-YLNAYWRASA-N	0.0241	0.65
Nutriacholic acid	DXOCDBGWDZAYRQ-QPVZPPSOSA-N	0.0077	1.96
6α-Hydroxycastasterone	CVXIEYXJQSRIAC-KLUYZAHOSA-N	0.0399	0.70
Carbohydrates and carbohydrate conjugates			
a-L-Fucopyranosyl-(1->2)-b-D-galactopyranosyl-(1->2)-D-xylose	DPTWUYFOEZBIEX-UHFFFAOYSA-N	0.0045	0.76
Dulcitol	FBPFZTCFMRRESA-GUCUJZIJSA-N	0.0025	2.95
Fucose	SHZGCJCMOBCMKK-FPRJBGLDSA-N	0.0011	0.62
Glucoheptonic acid	FATUQANACHZLRT-KMRXSBRUSA-L	0.0060	1.19
Glucose	WQZGKKKJIJFFOK-DVKNGEFBSA-N	0.0018	1.19
Cholestane steroids			
Cholesterol sulfate	BHYOQNUELFTYRT-DPAQBDIFSA-N	0.0124	0.25
Diradylglycerols			
DG (29:1)	XRIOEAXABJXXOB-CCZSOTCQSA-N	0.0399	1.63
Diterpenoids			
Phytenic acid	WDWBNNBRPVEEOD-PFXVRADUSA-N	0.0000	1.74
Estrane steroids			
3,5-Tetrahydronorethindrone	DPDZKJQFRFZZCW-VIVHBNLFSA-N	0.0077	2.98
Fatty acid esters			
Octanoylcarnitine	CXTATJFJDMJMIY-CYBMUJFWSA-N	0.0399	2.20
Palmitoylcarnitine	XOMRRQXKHMYMOC-OAQYLSRUSA-N	0.0014	1.64
Sorbitan palmitate	IYFATESGLOUGBX-RERQMSIWSA-N	0.0135	1.29
Fatty acids and conjugates			
10,13-nonadecadienoic acid	FLYBGKXSHCVONZ-HZJYTTRNSA-N	0.0008	3.21
11,14-Eicosadienoic acid	XSXIVVZCUAHUJO-AVQMFFATSA-N	0.0304	1.36
15-hydroxy-pentacosanoic acid	JIUVLHYELZYWJO-UHFFFAOYSA-N	0.0414	2.22
17-methyl-6-octadecenoic acid	QWCJNFBLSZGETP-CLFYSBASSA-N	0.0044	1.59
2,3-dihydroxy stearic acid	UAZFXPRZXKJSFJ-UHFFFAOYSA-N	0.0101	1.36
2-heptadecylenic acid	GEHPRJRWZDWFBJ-FOCLMDBBSA-N	0.0045	1.49
2-methoxy-hexadecanoic acid	YNBIUHDYZWCBSF-UHFFFAOYSA-N	0.0063	2.50
4,7,10,13,16,19-Docosahexaenoic acid	MBMBGCFOFBJSGT-SFGLVEFQSA-N	0.0221	1.55
8,11-eicosadiynoic acid	AJBBHZDIKZCZAI-UHFFFAOYSA-N	0.0045	1.44

Adrenic Acid	TWSWSIQAPQLDBP-DOFZRALJSA-N	0.0082	1.49
Class/Pathway and Metabolites	**InChI Key**	***p*(FDR)**	**Ratio (T2D-DL/ND)**
Fatty acids and conjugates (continued)			
Clupanodonic acid	YUFFSWGQGVEMMI-JLNKQSITSA-N	0.0064	1.60
Eicosanedioic acid	JJOJFIHJIRWASH-UHFFFAOYSA-N	0.0003	1.81
Eicosatrienoic acid	HOBAELRKJCKHQD-QNEBEIHSSA-N	0.0194	1.44
Hexadecenoic acid	ZVRMGCSSSYZGSM-CCEZHUSRSA-N	0.0124	1.69
Lauric acid	POULHZVOKOAJMA-UHFFFAOYSA-N	0.0021	2.38
Margaric acid	KEMQGTRYUADPNZ-UHFFFAOYSA-N	0.0025	1.33
Myristic acid	TUNFSRHWOTWDNC-UHFFFAOYSA-N	0.0201	1.36
Myristoleic acid	YWWVWXASSLXJHU-WAYWQWQTSA-N	0.0017	1.92
OAHSA	OCHJVQODRYVDAA-YPKPFQOOSA-N	0.0020	1.61
Oleic Acid	ZQPPMHVWECSIRJ-KTKRTIGZSA-N	0.0003	1.63
PAHSA	XXHBLSWAKHZVLN-UHFFFAOYSA-N	0.0204	1.38
Palmitic acid	IPCSVZSSVZVIGE-UHFFFAOYSA-N	0.0033	1.42
Pentadecylic acid	WQEPLUUGTLDZJY-UHFFFAOYSA-N	0.0259	1.31
POHSA	VCXRHEIVUHPWLL-SEYXRHQNSA-N	0.0433	1.36
Ricinoleic acid	WBHHMMIMDMUBKC-QJWNTBNXSA-N	0.0045	1.59
SAHSA	NQJLCZWOOVLQNP-UHFFFAOYSA-N	0.0009	1.54
Stearic acid	QIQXTHQIDYTFRH-UHFFFAOYSA-N	0.0006	1.40
Fatty acyl glycosides			
Melibiitol	PYZZIILDSAJNLZ-QZNPSGCDSA-N	0.0201	0.67
Fatty alcohols			
Artemoin A	KKUONIIRIFHWJC-UHFFFAOYSA-N	0.0115	1.77
Momordol	HDAGCVMZABLHLE-UHFFFAOYSA-N	0.0011	2.53
Glycerophosphates			
PA (44:5)	CAEQPKFDJRNAGN-BMJUURGTSA-N	0.0003	3.28
PA (30:1)	IUGHITQXXGJGCD-AHVPQQLLSA-N	0.0390	1.59
Glycerophosphocholines			
Glycerophosphocholine	SUHOQUVVVLNYQR-MRVPVSSYSA-N	0.0054	2.46
PC (22:6)	LSOWKZULVQWMLY-APPDJCNMSA-N	0.0237	3.03
PC (34:1)	WTJKGGKOPKCXLL-VYOBOKEXSA-N	0.0000	0.14
PC (34:4)	PWFGSGJBCRORHV-DVHMRFIGSA-N	0.0000	0.22
PC (36:4)	IIZPXYDJLKNOIY-JXPKJXOSSA-N	0.0173	1.25
PC (36:5)	KLTHQSWIRFFBRI-CPFPVJFHSA-N	0.0047	1.63
PC (38:6)	IESVDEZGAHUQJU-ZLBXKVHBSA-N	0.0011	1.40
PC (o-14:0)	WNCMKZYTLBIUSK-JOCHJYFZSA-N	0.0353	0.75
PC (o-34:3)	QEWRXEALGMKRGP-MZWPLUPDSA-N	0.0399	1.63
PC (o-36:4)	DUUSFCFZBREELS-SEHARXJXSA-N	0.0274	0.41
PC (o-38:4)	IRWRFKUTKSUFST-MDYGELLQSA-N	0.0477	1.96
PC (o-38:5)	VJNPDLZENXBRLB-MQEDXBOASA-N	0.0000	3.61
PC (o-38:6)	QQQQNYAHSSIZBU-HIQXTUQZSA-N	0.0054	2.15
Glycerophosphoethanolamines			
PE (36:2)	CFANDHZPOSNKNO-UDHSZFGOSA-N	2.87e-10	5.52

PE (44:3)	NTIXPPFPXLYJCT-SWXWTCIYSA-N	0.0481	1.96
Class/Pathway and Metabolites	**InChI Key**	***p*(FDR)**	**Ratio (T2D-DL/ND)**
Glycerophosphoethanolamines (continued)			
PE (p-36:2)	BIYXBOGKCFGRKR-NZWMGPRYSA-N	0.0290	0.33
PE (p-38:1)	UAPYZCDNFGJZSC-QSKLCZHJSA-N	0.0425	1.67
Glycerophosphoserines			
PS (22:1)	UVRWNZBXLMSZFS-IEACRGRWSA-N	0.0257	0.76
PS (o-35:0)	CRNHCTGXEIEELY-RGULYWFUSA-N	0.0355	2.35
PS (o-38:0)	ALHYOYYGOVGUJA-ZFESHMOZSA-N	0.0083	2.32
Hydroxycinnamic acids and derivatives			
Erythrinasinate A	XEOWPOLWKNHXGL-NHQGMKOOSA-N	0.0165	1.55
Hydroxyindoles			
Indoxyl	PCKPVGOLPKLUHR-UHFFFAOYSA-N	0.0292	1.25
Indoles			
Indole-3-ethanol	MBBOMCVGYCRMEA-UHFFFAOYSA-N	0.0116	0.74
Lineolic acids and derivatives			
9-hydroperoxy-10,12-octadecadienoic acid	JGUNZIWGNMQSBM-UINYOVNOSA-N	0.0006	3.02
Dimorphecolic acid (9-HODE)	NPDSHTNEKLQQIJ-SIGMCMEVSA-N	0.0036	0.63
Linoleic acid	OYHQOLUKZRVURQ-HZJYTTRNSA-N	0.0022	1.48
Linolenic acid	DTOSIQBPPRVQHS-PDBXOOCHSA-N	0.0068	1.48
α-kamlolenic acid	YPHQMIRXEFDOQM-ALXQIFAGSA-N	0.0186	0.62
Medium-chain hydroxy acids and derivatives			
3-Hydroxydodecanoic acid	MUCMKTPAZLSKTL-UHFFFAOYSA-N	0.0042	0.60
Monoradylglycerols			
MG (14:0)	TVIMZSOUQXNWHO-UHFFFAOYSA-N	0.0022	1.48
MG (16:0)	QHZLMUACJMDIAE-SFHVURJKSA-N	0.0001	1.51
MG (16:1)	CXUXMSACCLYMBI-FPLPWBNLSA-N	0.0000	1.75
Naphthofurans			
Cafamarine	TXMUQQBIIGAMBQ-UHFFFAOYSA-N	0.0257	0.31
Phosphosphingolipids			
SM (d32:1)	KYICBZWZQPCUMO-PSALXKTOSA-N	0.0328	2.13
SM (d34:1)	RWKUXQNLWDTSLO-GWQJGLRPSA-N	0.0000	1.36
SM (d36:1)	LKQLRGMMMAHREN-YJFXYUILSA-N	0.0000	0.25
SM (d36:2)	NBEADXWAAWCCDG-QDDWGVBQSA-N	0.0000	1.47
Pregnane steroids			
Allopregnanolone	AURFZBICLPNKBZ-SYBPFIFISA-N	0.0022	2.43
Purine ribonucleotides			
Inosine diphosphate	JPXZQMKKFWMMGK-KQYNXXCUSA-N	0.0000	1.98
Steroid esters			
Cholesteryl linoleate	NAACPBBQTFFYQB-LJAITQKLSA-N	0.0007	0.16
CE (20:4)	IMXSFYNMSOULQS-BEDFLICRSA-N	0.0026	0.21
Stilbenes			
Dihydroresveratrol	HITJFUSPLYBJPE-UHFFFAOYSA-N	0.0022	2.54

Class/Pathway and Metabolites	InChI Key	*p*(FDR)	Ratio (T2D-DL/ND)
Sulfones			
2-Propenyl 3-(2-propenylsulfonyl)-1-propenyl disulfide	OBJCYMGLWKMJIK-ALCCZGGFSA-N	0.0031	2.31
Terpene glycosides			
Goshonoside F6	NUDLZKKCTSSWNM-MHWRWJLKSA-N	0.0460	1.86
Tricarboxylic acids and derivatives			
Citric acid	KRKNYBCHXYNGOX-UHFFFAOYSA-N	0.0110	1.44
Triacylglycerols			
TG (52:2)	BHPBNQOWGMVFMA-QGJNQHKMSA-N	0.0489	0.24
TG (52:3)	JOSCSCZRVHLRGE-CQBMRIABSA-N	0.0086	0.31

Table S2. Significantly altered metabolites between T2D-M (metformin-treated group) and nondiabetic.

Class/Pathway and Metabolites	InChI Key	*p*(FDR)	Ratio (T2D-M/ND)
Amines			
Arachidoyl Ethanolamide	AUJVQJHODMISJP-UHFFFAOYSA-N	0.0066	0.41
Amino acids, peptides, and analogues			
Glutamine	ZDXPYRJPNDTMRX-VKHMYHEASA-N	0.0339	1.16
His Asp	MDCTVRUPVLZSPG-BQBZGAKWSA-N	0.0153	1.95
Anthracenes			
Palmidin C	VUUFXTUVVIEIMH-UHFFFAOYSA-N	0.0046	1.25
Bile acids, alcohols and derivatives			
27-Nor-5b-cholestane-3a,7a,12a,24,25-pentol	UBCPEZWGUOSYHO-JAPSQKQGSA-N	0.0026	2.24
Nutriacholic acid	DXOCDBGWDZAYRQ-QPVZPPSOSA-N	0.0054	2.10
Carbohydrates and carbohydrate conjugates			
Fucose	SHZGCJCMOBCMKK-FPRJBGLDSA-N	0.0012	0.66
Glucoheptonic acid	FATUQANACHZLRT-KMRXSBRUSA-L	0.0176	1.16
Glucose	WQZGKKKJIJFFOK-DVKNGEFBSA-N	0.0001	1.22
Cholestane steroids			
Cholesterol	HVYWMOMLDIMFJA-DPAQBDIFSA-N	0.0024	0.61
Cholesterol sulfate	BHYOQNUELFTYRT-DPAQBDIFSA-N	0.0116	0.25
Diacylglycerols			
DG (33:3)	QRWYRQDTDGUNRG-ABCZUFMESA-N	0.0204	1.47
Fatty alcohols			
Momordol	HDAGCVMZABLHLE-UHFFFAOYSA-N	0.0355	1.96
Flavonoid glycosides			
Caohuoside D	IINPFAXRBBMPBJ-BCNKTGMKSA-N	0.0029	2.39
Glycerophosphates			
PA (44:5)	CAEQPKFDJRNAGN-BMJUURGTSA-N	0.0006	5.55
Glycerophosphocholines			
PC (22:6)	LSOWKZULVQWMLY-APPDJCNMSA-N	0.0031	4.34
PC (34:1)	WTJKGGKOPKCXLL-VYOBOKEXSA-N	0.0000	0.03
PC (34:2)	JLPULHDHAOZNQI-ZTIMHPMXSA-N	0.0161	0.69
PC (o-36:4)	DUUSFCFZBREELS-SEHARXJXSA-N	0.0218	0.41
PC (o-38:5)	VJNPDLZENXBRLB-MQEDXBOASA-N	0.0198	2.10
PC (P-38:2)	CRLRRUDVEGPSAN-WRPVVQBASA-N	0.0080	2.54
Glycerophosphoethanolamines			
PE (36:2)	CFANDHZPOSNKNO-UDHSZFGOSA-N	0.0000	5.40
PE (36:4)	SSCDRSKJTAQNNB-DWEQTYCFSA-N	0.0237	0.31
PE (38:4)	ZSWHHKKYMMYPPD-MSZSINMWSA-N	0.0168	0.67
PE (38:6)	MPWUZHVZZKSTPV-MADBQMNMSA-N	0.0176	0.24
PE (40:6)	XYYHNDVKALDFHQ-OXHZBIAZSA-N	0.0355	0.37
PE (o-38:5)	DHHREGPDDQTYPB-UNRGBGOYSA-N	0.0056	0.22
PE (p-36:2)	BIYXBOGKCFGRKR-NZWMGPRYSA-N	0.0155	0.28
PE (p-36:4)	KDMBUUZGCXQNBE-XBICFDGKSA-N	0.0070	0.21
PE (p-38:5)	VWNWYWMBTKYEEL-SXDACRMGSA-N	0.0038	0.22

Class/Pathway and Metabolites	InChI Key	*p*(FDR)	Ratio (T2D-M/ND)
Glycerophosphoethanolamines (continued)			
PE (p-38:6)	WVGALBKSWOUIEZ-XNHMFJFDSA-N	0.0116	0.31
PE (p-40:6)	FIJFPUAJUDAZEY-MNDXXDKYSA-N	0.0194	0.25
Glycerophosphoinositols			
PI (36:4)	KIQYUSYSJTUGFZ-YLSTVYOTSA-N	0.0108	0.22
Glycerophosphoserines			
PS (22:1)	UVRWNZBXLMSZFS-IEACRGRWSA-N	0.0397	0.77
PS (36:1)	AJFWREUFUPEYII-PAHWMLEVSA-N	0.0229	0.35
PS (38:4)	SVOUGFFDROZBJI-DNALCEECSA-N	0.0208	0.35
PS (o-35:0)	CRNHCTGXEIEELY-RGULYWFUSA-N	0.0176	3.64
Lineolic acids and derivatives			
Dimorphecolic acid (9-HODE)	NPDSHTNEKLQQIJ-SIGMCMEVSA-N	0.0128	0.73
Medium-chain hydroxy acids and derivatives			
3-Hydroxydodecanoic acid	MUCMKTPAZLSKTL-UHFFFAOYSA-N	0.0128	0.66
Phosphosphingolipids			
SM (d36:1)	LKQLRGMMMAHREN-YJFXYUILSA-N	0.0000	0.15
SM (d38:0)	UGRZESKDAPEULH-ACEXITHZSA-N	0.0388	1.91
SM (d42:2)	WKZHECFHXLTOLJ-QYKFWSDSSA-N	0.0211	0.56
Purine ribonucleotides			
Adenosine triphosphate	ZKHQWZAMYRWXGA-KQYNXXCUSA-J	0.0321	2.30
Inosine diphosphate	JPXZQMKKFWMMGK-KQYNXXCUSA-N	0.0000	1.94
Steroid esters			
CE(20:4)	IMXSFYNMSOULQS-BEDFLICRSA-N	0.0024	0.23
Cholesteryl linoleate	NAACPBBQTFFYQB-LJAITQKLSA-N	0.0017	0.25
Sulfones			
2-Propenyl 3-(2-propenylsulfonyl)-1-propenyl disulfide	OBJCYMGLWKMJIK-ALCCZGGFSA-N	0.0253	1.97
Tricarboxylic acids and derivatives			
Citric acid	KRKNYBCHXYNGOX-UHFFFAOYSA-N	0.0447	1.37
Triacylcglycerols			
TG (39:0)	WQFIBIGMXQGDCU-LDLOPFEMSA-N	0.0022	0.50
TG (52:2)	BHPBNQOWGMVFMA-QGJNQHKMSA-N	0.0487	0.24
TG (52:3)	JOSCSCZRVHLRGE-CQBMRIABSA-N	0.0046	0.27
Triterpenoids			
Fasciculol C	YRXIDKUVBPMNRA-YTOKZNHUSA-N	0.0056	2.09
Vitamin D and derivatives			
(20R)-24-Hydroxygeminivitamin D3	UAKRHUPVCAWPPT-DBQRCUIGSA-N	0.0116	1.36