Knowledge that will change your world

Data sets at the Metabolomics Workbench

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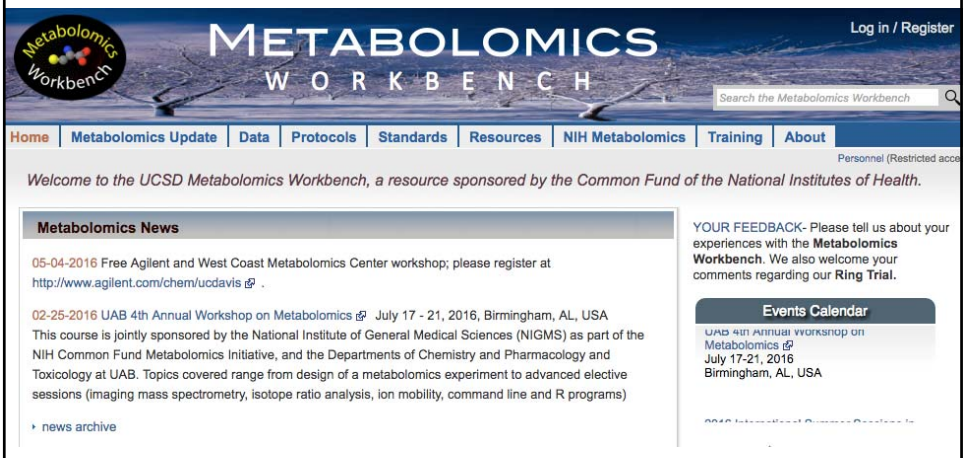
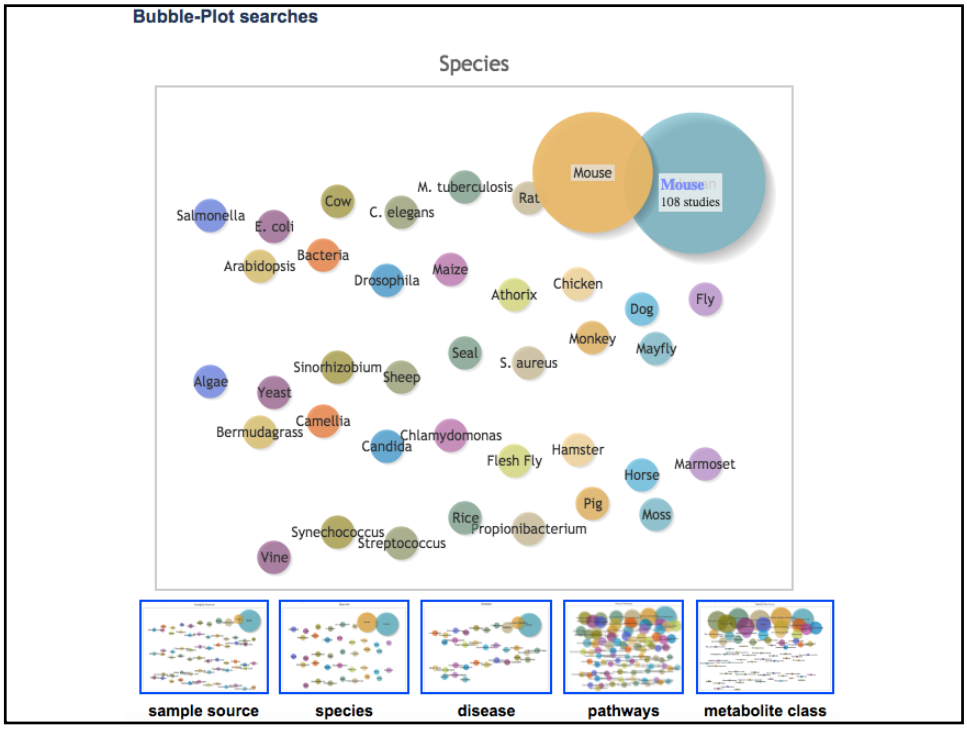
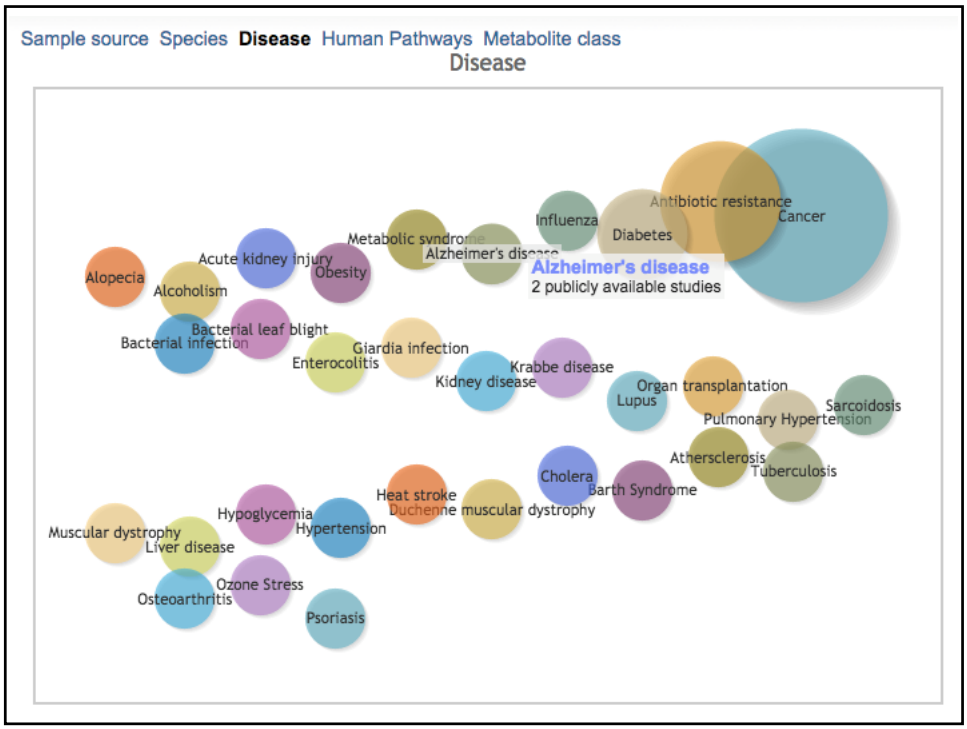
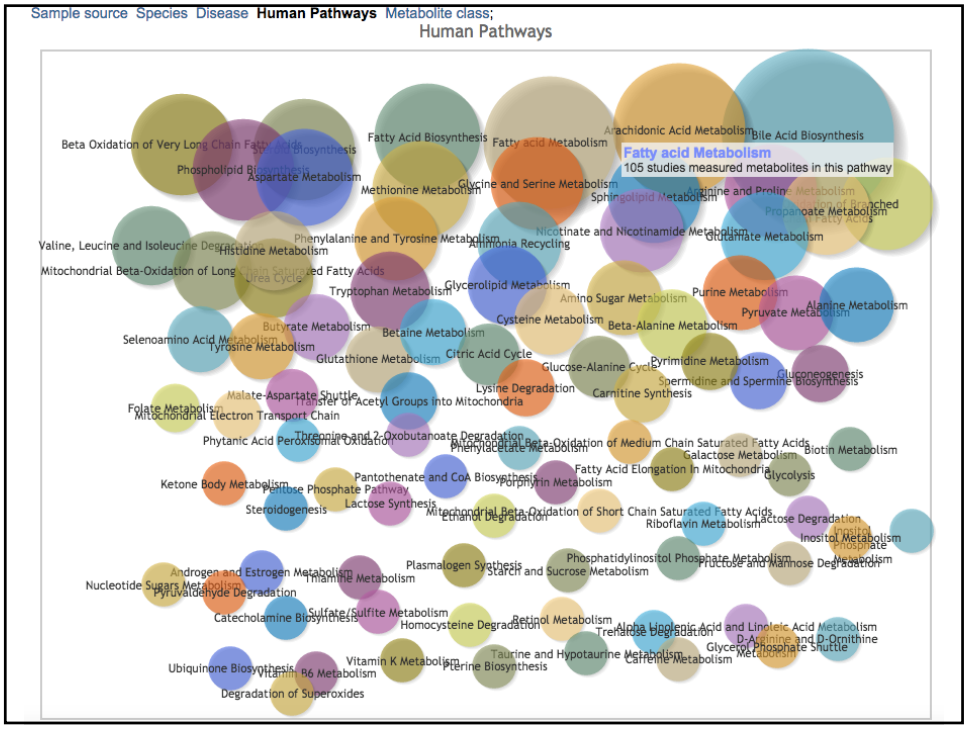
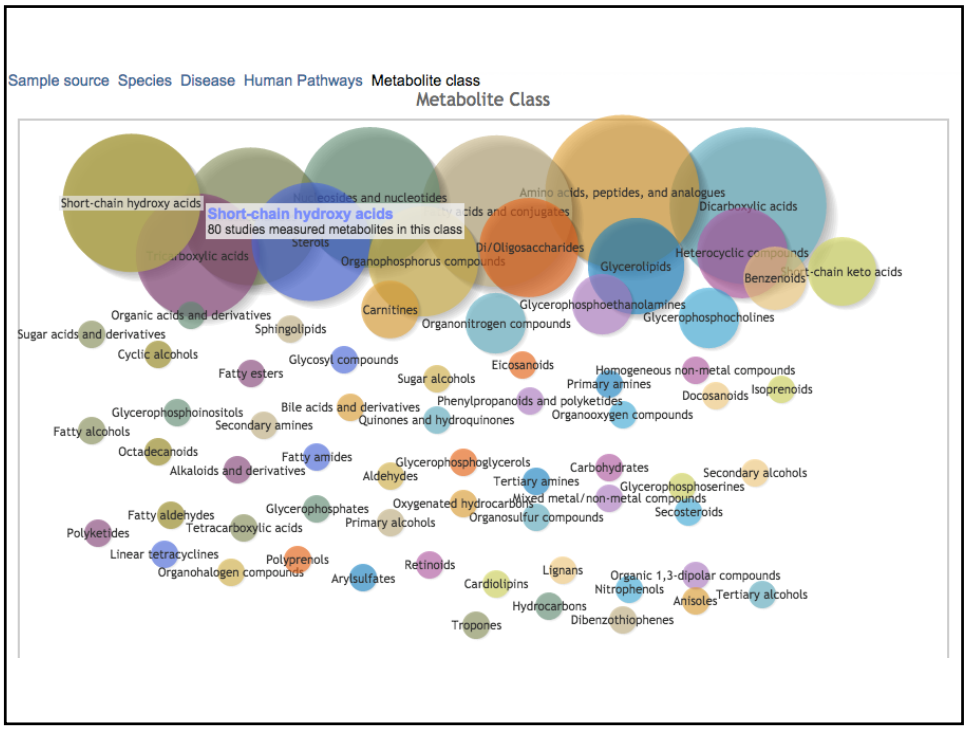
http://www.metabolomicsworkbench.org/




Bubble-Plot searches
Species
Mouse
Mouse
108 studies
M. tuberculosis
Rat
Cow
C. elegans
Salmonella
E. coli
Bacteria
Arabidopsis
Drosophila
Maize
Athorix
Chicken
Dog
Fly
Monkey
Mayfly
Seal
S. aureus
Sinorhizobium
Sheep
Algae
Yeast
Camellia
Bermudagrass
Chlamydomonas
Candida
Flesh Fly
Hamster
Horse
Marmoset
Pig
Moss
Rice
Propionibacterium
Synechococcus
Streptococcus
Vine
sample source
species
disease
pathways
metabolite class


Sample source Species Disease Human Pathways Metabolite class
Disease
Antibiotic resistance
Cancer
Influenza
Diabetes
Metabolic syndrome
Alzheimer's disease
Alzheimer's disease
2 publicly available studies
Acute kidney injury
Obesity
Alopecia
Alcoholism
Bacterial leaf blight
Bacterial infection
Giardia infection
Enterocolitis
Krabbe disease
Kidney disease
Organ transplantation
Lupus
Sarcoidosis
Pulmonary Hypertension
Atherosclerosis
Tuberculosis
Cholera
Barth Syndrome
Heat stroke
Duchenne muscular dystrophy
Hypoglycemia
Hypertension
Muscular dystrophy
Liver disease
Ozone Stress
Osteoarthritis
Psoriasis


Sample source Species Disease Human Pathways Metabolite class;
Human Pathways
Fatty acid Metabolism
105 studies measured metabolites in this pathway
Arachidonic Acid Metabolism
Bile Acid Biosynthesis
Fatty Acid Biosynthesis
Beta Oxidation of Very Long Chain Fatty Acids
Phospholipid Biosynthesis
Aspartate Metabolism
Methionine Metabolism
Glycine and Serine Metabolism
Sphingolipid Metabolism
Propanoate Metabolism
Fatty Acids
Valine, Leucine and Isoleucine Degradation
Phenylalanine and Tyrosine Metabolism
Histidine Metabolism
Ammonia Recycling
Nicotinate and Nicotinamide Metabolism
Glutamate Metabolism
Mitochondrial Beta-Oxidation of Long Chain Saturated Fatty Acids
Urea Cycle
Tryptophan Metabolism
Glycerolipid Metabolism
Amino Sugar Metabolism
Purine Metabolism
Alanine Metabolism
Pyruvate Metabolism
Cysteine Metabolism
Beta-Alanine Metabolism
Butyrate Metabolism
Betaine Metabolism
Selenoamino Acid Metabolism
Tyrosine Metabolism
Glutathione Metabolism
Citric Acid Cycle
Glucose-Alanine Cycle
Pyrimidine Metabolism
Gluconeogenesis
Spermidine and Spermine Biosynthesis
Lysine Degradation
Carnitine Synthesis
Malate-Aspartate Shuttle
Transfer of Acetyl Groups into Mitochondria
Folate Metabolism
Mitochondrial Electron Transport Chain
Threonine and 2-Oxobutanoate Degradation
Phytanic Acid Peroxisomal Oxidation
Phenylacetate Metabolism
Mitochondrial Beta-Oxidation of Medium Chain Saturated Fatty Acids
Biotin Metabolism
Galactose Metabolism
Fatty Acid Elongation in Mitochondria
Glycolysis
Ketone Body Metabolism
Pentose Phosphate Pathway
Pantothenate and CoA Biosynthesis
Porphyrin Metabolism
Steroidogenesis
Lactose Synthesis
Ethanol Degradation
Mitochondrial Beta-Oxidation of Short Chain Saturated Fatty Acids
Riboflavin Metabolism
Lactose Degradation
Inositol Metabolism
Androgen and Estrogen Metabolism
Thiamine Metabolism
Plasmalogen Synthesis
Starch and Sucrose Metabolism
Phosphatidylinositol Phosphate Metabolism
Fructose and Mannose Degradation
Nucleotide Sugars Metabolism
Pyruvaldehyde Degradation
Sulfate/Sulfite Metabolism
Catecholamine Biosynthesis
Homocysteine Degradation
Retinol Metabolism
Alpha Linolenic Acid and Linoleic Acid Metabolism
Tretinoin Degradation
D-Arginine and D-Ornithine Metabolism
Glycerol Phosphate Shuttle
Carnitine Metabolism
Taurine and Hypotaurine Metabolism
Ubiquinone Biosynthesis
Vitamin K Metabolism
Vitamin B6 Metabolism
Pterine Biosynthesis
Degradation of Superoxides


Sample source Species Disease Human Pathways Metabolite class
Metabolite Class
Short-chain hydroxy acids
80 studies measured metabolites in this class
Amino acids, peptides, and analogues
Dicarboxylic acids
Organophosphorus compounds
Di/Oligosaccharides
Glycerolipids
Heterocyclic compounds
Benzenoids
Short-chain keto acids
Carnitines
Glycerophosphoethanolamines
Glycerophosphocholines
Organonitrogen compounds
Organic acids and derivatives
Sugar acids and derivatives
Sphingolipids
Cyclic alcohols
Glycosyl compounds
Eicosanoids
Homogeneous non-metal compounds
Fatty esters
Sugar alcohols
Primary amines
Docosanoids
Isoprenoids
Phenylpropanoids and polyketides
Glycerophosphoinositols
Bile acids and derivatives
Secondary amines
Quinones and hydroquinones
Organooxygen compounds
Fatty alcohols
Octadecanoids
Fatty amides
Alkaloids and derivatives
Glycerophosphoglycerols
Aldehydes
Carbohydrates
Tertiary amines
Secondary alcohols
Glycerophosphoserines
Mixed metal/non-metal compounds
Fatty aldehydes
Glycerophosphates
Oxygenated hydrocarbons
Secosteroids
Tetracarboxylic acids
Primary alcohols
Organosulfur compounds
Polyketides
Linear tetracyclines
Polyprenols
Organohalogen compounds
Arylsulfates
Retinoids
Cardiolipins
Lignans
Organic 1,3-dipolar compounds
Nitrophenols
Anisoles
Tertiary alcohols
Hydrocarbons
Tropones
Dibenzothiophenes

Browse and Search Data and Metadata

Browse and search data and metadata from experimental projects and studies, and search the Metabolite Database, which contains metabolite structures and annotations.

Browse

- Summary of all projects (groups of studies)
- Summary of all studies
- Bubble plots of studies by disease, sample source, species, pathway and metabolite class

Search

Experimental Projects / Studies

- Data/metadata in experimental projects/studies

Metabolites

- Metabolite data/metadata in experimental studies and Metabolite Database
- Untargeted MS data with an m/z value

Download data

You have selected to download the following file: **ST000003.zip (5.3G)**

The download file contains the following type of raw data files: **.xml, .bin, .xsd, .cd,.cG, .mzxml,.sd,.sp**

Please review the following information before you **continue to download**:

- Some of the compressed files available for download are quite large. You might encounter problems during their download through the browsers. These files are also accessible for download through an anonymous FTP server, **ftp://www.metabolomicsworkbench.org**. A variety of free and commercial standalone file transfer clients exist to download large data files directly from the FTP servers: FileZilla , WinSCP , download managers , etc.
- All the ZIP files available for download are generated using Deflate compression method. An integrity check is also performed on each ZIP file to make sure the file is not corrupted. You should be able to extract data from these files using Microsoft Windows Explorer. In case you encounter problems extracting data form these files, consider using 7-zip to perform an integrity check and to extract data from the files.
- The download ZIP files contain Microsoft Excel (XLS and XLSM) files. You may not be able to open the ZIP files using Windows Explorer after downloading these files. In order to open these files in Windows, you may need to **unblock** these files: Right click to select **Properties** and then clicking **Unblock** button. Alternatively you may need to adjust the EXCEL TRUST Center settings: Click on File > Options. Select Trust Center > Trust center settings

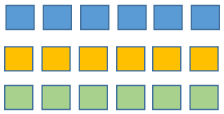
Downloading the ST00003 file

- **5.3 GB**
- **Took about 6 hours to download**
- **When unzipped, it contained three Excel files and the raw data files**

Metadata template

Acquisition parameters

Normalized data metabolites



Raw data files

Metadata 1

B	C	D
	Project Identifier	Project ID (will be added)
	Project Title*	Metabolomic analysis of mouse embryonic fibroblasts, embryonic stem cells, and induced pluripotent stem cells
	Project Type	
	Project Summary	
	Institute*	University of California - Davis
	Department*	Genome Center
	Laboratory	Fiehn Laboratory
	Funding source	California Institute for Regenerative Medicine grant F
	Principal Investigator Last Name*	Fiehn
	Principal Investigator First Name*	Oliver
	Address	451 Health Sci Drive, Davis, CA 95616
	Email*	ofiehn@ucdavis.edu
	Phone	+1-530-754-8258
	* = Required fields	

Metadata 2

Study Identifier	Study ID (will be added)
Study Title*	Metabolomic analysis of mouse embryonic fibroblasts, embryonic stem cells, and induced pluripotent stem cells
Study type* (timecourse,drug dosage,etc.)	Cell Type Comparison
Study Summary (Description/Design)*	mouse embryonic fibroblasts, embryonic stem cells, and induced pluripotent stem cells were compared via metabolomic analysis
Institute*	University of California - Davis
Department*	Genome Center
Laboratory	Fiehn Laboratory
Study contact person: Last Name*	Meissen
Study contact person: First Name*	John
Address	451 Health Sci Drive, Davis, CA 95616
Email*	jkmeissen@ucdavis.edu
Phone	+1-530-752-9922
Submission Date (MM-DD-YYYY)*	01-16-2013
Number of Groups*	3
Total Number of subjects/patients/samples*	18
Study Comments	

Metadata 3

A	B	C	D
Use a separate column for each factor		Sample Identifier	Factor1
(See ExampleofStudyDesign tab for example)		Local Sample ID	Cell Line
		iPSC-T1R1	iPSC
		iPSC-T1R2	iPSC
		iPSC-T1R3	iPSC
		iPSC-T2R1	iPSC
		iPSC-T2R2	iPSC
		iPSC-T2R3	iPSC
		m15-T1R1	embryonic fibroblast
		m15-T1R2	embryonic fibroblast
		m15-T1R3	embryonic fibroblast
		m15-T2R1	embryonic fibroblast
		m15-T2R2	embryonic fibroblast
		m15-T2R3	embryonic fibroblast
		mESC-T1R1	embryonic stem cell
		mESC-T1R2	embryonic stem cell
		mESC-T1R3	embryonic stem cell
		mESC-T2R1	embryonic stem cell
		mESC-T2R2	embryonic stem cell
		mESC-T2R3	embryonic stem cell

Metadata 4

C	D	E	F
Subject Identifier	Subject ID (will be added)		
Subject Type (Human, Aminal, Plant, Cells)*	Animal		
Subject Species*	*M. musculus*		
Number of Replicates*	1		
Taxonomy ID			
Genotype/Strain			
Age or Age Range	Embryo		
Weight or Weight range			
Height or Height range			
Gender			
HUMAN:Race			
HUMAN:Ethnicity			
HUMAN:Trial Type			
HUMAN:Lifestyle Factors			
HUMAN:Medications			
HUMAN:Prescription/OTC			
HUMAN:Smoking Status			
HUMAN:Alcohol/Drug Use			
HUMAN:Nutrition			
HUMAN:Inclusion Criteria			
HUMAN:Exclusion Criteria			
ANIMAL:Animal Supplier			
ANIMAL:Housing			
ANIMAL:Light cycle			
ANIMAL:Feed			
ANIMAL:Water			
ANIMAL:Inclusion Criteria			
CELL:Biosource or Supplier			
CELL:Strain Details			
CELL:Primary or immortalized			
CELL:Passage number			
Subject Comments			

* = Required fields

Notes:

Use a separate column for each subject type (group).

Indicate the Number of Replicates for each subject type. Each subject/patient/sample will be assigned a SUBJECT_ID (e.g. SU000001, SU000002, SU000003 etc.)

Metadata 5

C	D
Treatment Type Identifier	Treatment Type ID (will be added)
Treatment Summary	
Treatment Protocol ID	
Treatment Protocol Filename	
Treatment Protocol Comments	
Treatment (Abiotic, Biotic, Intervention)	
Treatment Compound	
Treatment Route	
Treatment Dose	
Treatment Dose Volume	
Treatment Dose Duration	
Treatment Vehicle	
ANIMAL:Veterinary Treatments	
ANIMAL:Anesthesia	
ANIMAL:Acclimation Duration	
ANIMAL:Fasting	
ANIMAL:EndPoints Euthanasia	
ANIMAL:EndPoints Tissue Collection List	
ANIMAL:EndPoints Tissue Processing Method	
ANIMAL:EndPoints Clinical Signs	
HUMAN:Fasting	
HUMAN:EndPoints Clinical Signs	
CELL:Storage	

Metadata 6

C	D
Collection Identifier	Collection ID (will be added)
Collection Summary	
Collection Protocol ID	
Collection Protocol Filename	
Collection Protocol Comments	
Sample Type (Blood, Urine, Saliva, Tissue, Cell, other)*	
Collection Method	
Collection Location	
Collection Frequency	
Collection Duration	
Time of Collection (Relative to dose)	
Volume/Amount Collected	
Storage Conditions	
Collection Vials	
Storage Vials	
Collection Tube Temp	
Additives	
BLOOD Serum or Plasma	
TISSUE/CELL Identification	
TISSUE/CELL Quantity Taken	
* = Required fields	

Metadata 7

C	D
Sample Prep Identifier	Sample Prep ID (will be added)
Sample Prep Summary	Cold Methanol Extraction
Sample Prep Protocol ID	
Sample Prep Protocol Filename	
Sample Prep Protocol Comments	
Processing Method (Lyophilization, Homogenization, Lysis, etc)	Homogenization and Solvent Removal w/ Speed Vac
Processing Storage conditions	On Ice
Extraction Method	3:1 MeOH/H2O
Extract Concentration/Dilution	1,000,000 Cells/mL
Extract Enrichment (SPE, Desalting, etc)	
Extract Cleanup	
Extract Storage	-80C
Sample Resuspension	100 uL of 9:1 ACN/H2O with 5mM NH4OAc + 0.2% Acetic Acid
Sample Derivatization	
Sample Spiking (Internal Standards, Retention Standards, etc)	
Sample Prep Protocol Reference/ID	
Organ	
Organ Specification	
Cell Type	embryonic fibroblasts, embronic stem cells, induced pluripotent stem cells
Subcellular Location	

Metadata 8

B	C	D
	Chromatography Identifier	Chromatography ID (will be added)
	Chromatography Summary	Untargeted HILIC Method
	Methods File ID	
	Methods Filename	
	Chromatography Comments	
	Instrument Name	Agilent 1200 Series
	Column Name	Waters Acquity 1.7um BEH HILIC 2.1x150 mm
	Column Pressure	300 Bar (Max)
	Column Temperature	40
	Flow Gradient	0-4 min - 0% B, 4-12 min - Gradient to 45% B, 12-20 min - 45% B
	Flow Rate	0.25 mL/min
	Injection Temperature	4 C
	Internal Standard	
	Internal Standard MT	
	Retention Index	
	Retention Time	
	Sample Injection	5 uL
	Sampling Cone	
	Solvent A	H2O + 5mM NH4OAc + 0.2% Acetic Acid
	Solvent B	9:1 Acetonitrile:H2O + 5mM NH4OAc + 0.2% Acetic Acid
	Analytical Time	20 min
	Capillary Voltage	
	Migration Time	
	Oven Temperature	

Metadata 9

C	D	E
Analysis Identifier	Analysis ID (will be added)	
Laboratory Name	Fiehn Laboratory	
Operator Name	John Meissen	
Analysis Type (MS, NMR, other)	MS	
Instrument Name	Agilent 6530 QTOF	
Software/Version	B02	
Instrument Parameters File		
Analysis Protocol ID		
Analysis Comments		
Acqusition Date	02/26/11 + 05/16/11	
Acqusition Time	20 min	
Acquisition ID		
Acquisition Parameters File		
Acquired (Raw) File		
Processing Parameters File		
Processed File		
Randomization Order		
Detector Type		
Data Format		

Notes:
Use a separate column for each analysis set, e.g positive ion-mode and negative ion-mode MS)

Metadata 10

B	C	D
	MS Identifier	MS ID (will be added)
	Analysis Identifier	Analysis ID (will be added)
	Instrument Type	Agilent 6530 QTOF
	MS Type	MS and MS/MS
	MS Comments	
	Ion Mode	Positive
	Capillary Temperature	
	Capillary Voltage	
	Collision Energy	Ramped - 0.02eV/m/z + 10 eV
	Collision Gas	N2
	Dry Gas Flow	Neb - 10 L/min, Sheath 9 L/min
	Dry Gas Temp	Neb/Sheath - 350C
	Fragment Voltage	
	Fragmentation Method	
	Gas Pressure	35 PSI
	Helium Flow	
	Ion Source Temperature	
	Ion Spray Voltage	
	Ionization	Electrospray Ionization
	Ionization Energy	
	Ionization Potential	
	Mass Accuracy	<1.5 mDa
	Precursor Type	
	Reagent Gas	
	Source Temperature	

LC-MS parameters - overall

Acquisition Method Report

Agilent Technologies

Acquisition Method Info

Method Name	JKM_HILICWatersBEH1p7.m
Method Path	D:\MassHunter\Data\Ben\HILIC2\mESC1_0015.d\AcqData\JKM_HILICWatersBEH1p7.m
Method Description	Default Method

Device List

h-ALS-SL+
1260 BinPump
Bin Pump2
Column-SL
1260 DAD
MS Q-TOF

TOF/Q-TOF Mass Spectrometer

Component Name	MS Q-TOF	**Component Model**	G6510A
Ion Source	X-ESI	**Tune File**	AutoTune.tun
Stop Mode	NoLimit	**Stop Time**	30.00
Can wait for temp.	Enable	**Fast Polarity**	N/A
MS1CentroidDataAbsThreshold	200	**MS1CentroidDataRELThreshold**	0.010
MS2CentroidDataAbsThreshold	5	**MS2CentroidDataRELThreshold**	0.010

LC-MS parameters – MS conditions

Time Segments

Time Segment #	Start Time	Diverter Valve State	Storage Mode	Ion Mode
1	0.0	MS	Both	ESI+Agilent Jet Stream

Time Segment 1

Acquisition Mode AutoMS2

MS Min Range	100
MS Max Range	1700
MS Scan Rate	4.00
MS/MS Min Range	50
MS/MS Max Range	1700
MS/MS Scan Rate	4.00
Isolation Width MS/MS	Medium (~4 amu)

Ramped Collision Energy

Slope	2.0000
Offset	10.0000

LC-MS parameters – excluded ions

Mass	Delta Mass	Charge	Type	Retention Time	Delta Retention	Isolation Width	Collision Energy
121.05	20.000000	1	Exclude	0.00		Medium (~4	
124.02	20.000000	1	Exclude	0.00		Medium (~4	
129.10	20.000000	1	Exclude	0.00		Medium (~4	
140.00	20.000000	1	Exclude	0.00		Medium (~4	
155.97	20.000000	1	Exclude	0.00		Medium (~4	
157.99	20.000000	1	Exclude	0.00		Medium (~4	
159.00	20.000000	1	Exclude	0.00		Medium (~4	
179.03	20.000000	1	Exclude	0.00		Medium (~4	
180.98	20.000000	1	Exclude	0.00		Medium (~4	
181.03	20.000000	1	Exclude	0.00		Medium (~4	
199.00	20.000000	1	Exclude	0.00		Medium (~4	
200.02	20.000000	1	Exclude	0.00		Medium (~4	
256.96	20.000000	1	Exclude	0.00		Medium (~4	
262.98	20.000000	1	Exclude	0.00		Medium (~4	
297.99	20.000000	1	Exclude	0.00		Medium (~4	
338.97	20.000000	1	Exclude	0.00		Medium (~4	
354.28	20.000000	1	Exclude	0.00		Medium (~4	
533.88	20.000000	1	Exclude	0.00		Medium (~4	
550.63	20.000000	1	Exclude	0.00		Medium (~4	
721.51	20.000000	1	Exclude	0.00		Medium (~4	
922.01	20.000000	1	Exclude	0.00		Medium (~4	

Ramped Collision Energy

Slope	Offset
2	10

LC-MS parameters 4

Precursor Selection

Max Precursors Per Cycle	1
Threshold (Abs)	1000
Threshold (Rel)	0.000
Precursor abundance based scan speed	No
Active exclusion enabled	Yes
Active exclusion excluded after (spectra)	3
Active exclusion released after (min)	0.50
Sort precursors	By abundance only

Static Exclusion Ranges

Charge State
2
1
Unk

Source Parameters

Parameter	Value
Gas Temp (°C)	350
Gas Flow (l/min)	10.0
Nebulizer (psi)	35
SheathGasTemp	350
SheathGasFlow	9.0

LC-MS parameters 5

Scan Segment 1

Scan Source Parameters

Parameter	Value
VCap	3500
Nozzle Voltage (V)	500
Fragmentor	125
Skimmer1	65.0
OctopoleRFPeak	750

ReferenceMasses

Ref Mass Enabled	Enabled
Use Bottle A RefNebulizer	False
Ref Nebulizer	

AutoRecalibration

Average Scans	1
Detection Window	100
Min Height	500

Reference Masses
<Positive>
121.05087300
922.00979800

Chromatograms

Chrom Type	Label	Offset	Y-Range
TIC	TIC	15	10000000

LC-MS parameters – pump 1

Name	1260 BinPump	Model	G1312B
Ordinal #	1	Options	SSV
Stop Time (min)	20	Post Time (min)	20
Flow (ml/min)	0.25	Pressure Min (bar)	0
Pressure Max (bar)	575	Max Flow Gradient (ml/min)	100
Solvent A		Solvent B	
Solvent Ratio A	0	Solvent Ratio B	100
Solvent Type A1		Solvent Type B1	
Solvent Type A2		Solvent Type B2	
Compress. A (*10-6/bar)	100	Compress. B (*10-6/bar)	115
Stroke A (µl)	Auto	Stroke B (µl)	Auto
Stroke Syncronization			
Contact 1	Off		
Contact 2	Off		
Contact 3	Off		
Contact 4	Off		

Pump Time Table

Time	Flow	Pressure	Solv Ratio B
0	0.25	575	100
4	0.25	575	100
12	0.25	575	45
20	0.25	575	45

LC-MS parameters – pump 2

Name	Bin Pump2	Model	G1312A
Ordinal #	2	Options	
Stop Time (min)	No Limit	Post Time (min)	Off
Flow (ml/min)	2	Pressure Min (bar)	0
Pressure Max (bar)	400	Max Flow Gradient (ml/min)	100
Solvent A		Solvent B	
Solvent Ratio A	100	Solvent Ratio B	0
Solvent Type A1		Solvent Type B1	
Solvent Type A2		Solvent Type B2	
Compress. A (*10-6/bar)	100	Compress. B (*10-6/bar)	115
Stroke A (µl)	Auto	Stroke B (µl)	Auto
Stroke Syncronization			
Contact 1	Off		
Contact 2	Off		
Contact 3	Off		
Contact 4	Off		

LC-MS parameters – diode array

Name	1260 DAD	Model	G1315C
Ordinal #	1	Options	
Stop Time (min)	As Pump	Post Time (min)	Off
Delay Time (min)	0		
Store Spectra	Spectrum Data	Threshold	10
Pre-Run Balance	Yes	Post-Run Balance	No
Balance Mode	1	Margin for -ve absorbance	100
Peak Width2	GT 0.1 min (2.0s)	Slit	4
Output Zero Offset1 (%)	8	Output Zero Offset2 (%)	5
Ouput Attenuation1 (mAU)	1000	Output Attenuation2 (mAU)	1000
UV Lamp	Yes	Vis Lamp	No
From (nm)	190	To (nm)	800
Step (nm)	2		
Contact 1	Off		
Contact 2	Off		
Contact 3	Off		
Contact 4	Off		

LC-MS parameters – diode array

Channel	Sample WL (nm)	Sample BW (nm)	Ref. WL (nm)	Ref. BW (nm)	Ref. On
A	250	4	0	0	Off
B	254	16	0	0	Off
C	210	8	0	0	Off
D	230	16	0	0	Off
E	280	4	0	0	Off
F	280	16	0	0	Off
G	280	16	0	0	Off
H	280	16	0	0	Off

LC-MS parameters - sampler

Name	h-ALS-SL+	**Model**
Ordinal #	1	**Options**
Stop time (min)	As Pump	**Post Time (min)**
Injection Type	Needle Wash	**Injection Volume (µl)**
Overlap Time (min)	Disable Overlapped Injection	**Draw Position (mm)**
Draw Position Detection	1	**Draw Speed (µl/min)**
Eject Speed (µl/min)	200	**Flush Out Factor**
Automatic Delay Volume Reduction	No	**Equilibration Time (sec)**
Wash Vessel	N/A	**Wash Location**
Wash Time (sec)	5	**Wash Cycles**
Contact 1	Off	
Contact 2	Off	
Contact 3	Off	
Contact 4	Off	

LC-MS parameters - temperature

Name	Column-SL	**Model**
Ordinal #	1	**Options #**
Stop time (min)	As Pump	**Post Time (min)**
Left Temp. (°C)	40	**Right Temp. (°C)**
Left Ready (°C)	When Temp Within Set Point +/- 0.8	**Right Ready (°C)**
Valve Position	1	
Contact 1	Off	
Contact 2	Off	
Contact 3	Off	
Contact 4	Off	

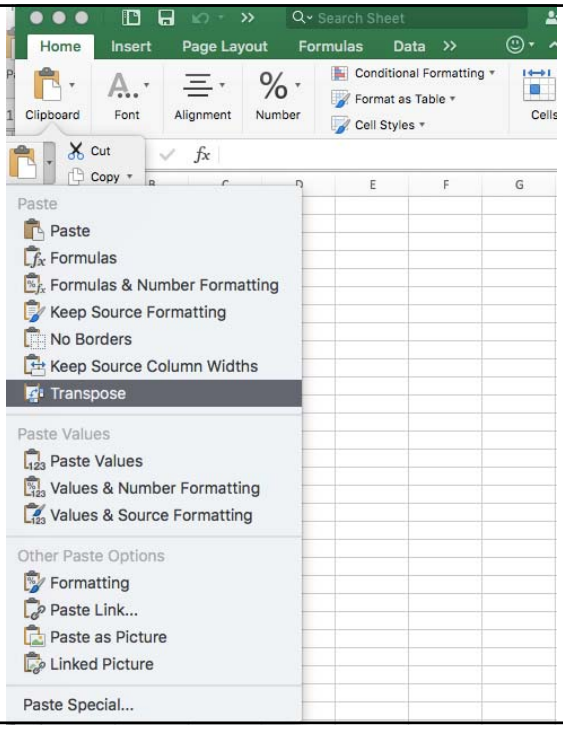
Normalized data set

1	MS1 m/z	118.0863	126.0219	132.0768	162.1125	204.123	220.118	232.1543	268.104	300.2897
2	Theoritical m/z	118.0861	126.0221	132.0767	162.1121	204.1229	220.1179	232.1529	268.1039	300.29
3	mDa Error	-0.16363	0.151183	-0.06158	-0.4	-0.1	-0.05769	-1.4432	-0.13896	0.285535
4	ppm Error	-1.38571	1.199655	-0.46624	-2.46743	-0.4899	-0.26209	-6.21658	-0.51831	0.950864
5	Retention Time/Index	11.404	4.761	12.156	13.483	13.117	4.856	12.52	3.1354	4.282
6	Method	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos
7	ID-Run	iPSC-1	iPSC-1	iPSC-1	iPSC-1	iPSC-1	iPSC-1	m15-1	iPSC-1	mESC-1
8	Database	Metlin	Metlin	Metlin	Metlin	Metlin	Metlin	Metlin	NIST	Metlin
9	Name	Betaine	Taurine	Creatine	Carnitine	Acetylcarni	Pantotheni	Butyryl car	Adenosine	Sphingosin
10	Molecular Formula	C5H11NO2	C2H7NO3S	C4H9N3O2	C7H15NO3	C9H17NO4	C9H17NO5	C11H21NO	C10H13N5	C18H37NO
11	PubChem	247	1123	586	10917	7045767	6613	213144	60961	5280335
12	PubChem sn1									
13	PubChem sn2									
14	KEGG	C00719	C00245	C00300	C00318	C02571	C00864	C02862	C00212	C00319
15	BinBase/mzMine ID	4807	10837	12971	4173	7568	9080	3017	10956	10321
16	iPSC-T1R1	144301	459870	465781	43557	23803	10585	2875		32290
17	iPSC-T1R2	146452	405380	456427	49718	25064	9483	3040	1607	24231
18	iPSC-T1R3	159604	459803	494302	58249	28873	11310	3573	322	33665
19	iPSC-T2R1	54013	111315	39520	16884	10445	8283	2267	10701	30264
20	iPSC-T2R2	55288	103051	36568	12434	13572	7752	1956	10157	28371
21	iPSC-T2R3	50295	132147	48141	17086	13467	8117	1746	9497	25047
22	m15-T1R1	104109	521094	298459	82978	58040	11909	4748	2558	40221
23	m15-T1R2	116864	345155	204896	62559	60942	15376	7944	3616	65597
24	m15-T1R3	123262	550930	373895	104781	78014	14255	6328	4270	36550
25	m15-T2R1	18045	155927	30721	15399	14093	17510	6241	2526	26985
26	m15-T2R2	25772	176304	40126	15273	17948	19908	7011	2647	26934
27	m15-T2R3	29957	196313	42868	18291	17926	18640	7777	2671	26344
28	mESC-T1R1	102812	621628	863781	89799	59787	17575	8756		29871
29	mESC-T1R2	97364	625820	823663	91218	58049	18093	8075		35639
30	mESC-T1R3	117925	710433	903878	95964	60402	19692	8320	4575	39898
31	mESC-T2R1	9071	46063	15248	6057	5318	5397	2620	9162	47069
32	mESC-T2R2	9486	76412	27482	9526	5925	4725	1843	7470	45466
33	mESC-T2R3	9903	59713	21785	8107	5779	5208	2156	7624	45168

Transposing data in Excel

A	B	C	D	E	F	G	H	I	J	K	L	M	N	O
MS1 m/z	118.0863	126.0219	132.0768	162.1125	204.123	220.118	232.1543	268.104	300.2897	468.3085	480.309	494.3243	496.3399	522.3556
Theoritical m/z	118.0861	126.0221	132.0767	162.1121	204.1229	220.1179	232.1529	268.1039	300.29	468.309	480.309	494.3247	496.3403	522.356
mDa Error	-0.16363	0.151183	-0.06158	-0.4	-0.1	-0.05769	-1.4432	-0.13896	0.285535	0.5	0	0.362	0.41	0.36
ppm Error	-1.38571	1.199655	-0.46624	-2.46743	-0.4899	-0.26209	-6.21658	-0.51831	0.950864	1.067671	0	0.732312	0.826046	0.689185
Retention Time/Index	11.404	4.761	12.156	13.483	13.117	4.856	12.52	3.1354	4.282	11.53302	7.686333	11.45467	11.27573	11.20805
Method	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos	HILIC Pos
ID-Run	iPSC-1	iPSC-1	iPSC-1	iPSC-1	iPSC-1	iPSC-1	m15-1	iPSC-1	mESC-1	iPSC-1	iPSC-1	m15-1	iPSC-1	iPSC-1
Database	Metlin	Metlin	Metlin	Metlin	Metlin	Metlin	Metlin	NIST	Metlin	LipidBlastP	LipidBlastP	LipidBlastP	LipidBlastP	LipidBlastP
Name	Betaine	Taurine	Creatine	Carnitine	Acetylcarni	Pantotheni	Butyryl car	Adenosine	Sphingosin	lysoPC 14:0	lysoPE 18:1	lysoPC 16:1	lysoPC 16:0	lysoPC 18:1
Molecular Formula	C5H11NO2	C2H7NO3S	C4H9N3O2	C7H15NO3	C9H17NO4	C9H17NO5	C11H21NO	C10H13N5	C18H37NO	C22H46NO	C23H46NO	C24H48NO	C24H50NO	C26H52NO
PubChem	247	1123	586	10917	7045767	6613	213144	60961	5280335	460604	9547071	24779461	460602	16081932
PubChem sn1										14:0	18:1	16:1	16:0	18:1
PubChem sn2														
KEGG	C00719	C00245	C00300	C00318	C02571	C00864	C02862	C00212	C00319	C04230	C04438	C04230	C04230	C04230
BinBase/mzMine ID	4807	10837	12971	4173	7568	9080	3017	10956	10321	10801	11750	8607	9595	9089

- Highlight all the data on the MS1 m/z values row
- Open a new Excel file



In the new Excel file, click on A column, cell 1 and select "Transpose"

	A
1	MS1 m/z
2	118.086264
3	126.021949
4	132.076762
5	162.1125
6	204.123
7	220.117958
8	232.154343
9	268.104039
10	300.289714
11	468.3085
12	480.309
13	494.324288
14	496.3399
15	522.3556
16	524.3707
17	675.54413
18	690.5065
19	700.527308
20	701.5592
21	702.5433
22	703.5753
23	706.5388
24	716.5226
25	718.538
26	724.5289
27	728.558849
28	729.59106
29	730.5387
30	732.5552

Excel file cleaned up to remove color and lines and to give each *m/z* value four decimal places

Now do the same for the retention time

	A
1	MS1 m/z
2	118.0863
3	126.0219
4	132.0768
5	162.1125
6	204.1230
7	220.1180
8	232.1543
9	268.1040
10	300.2897
11	468.3085
12	480.3090
13	494.3243
14	496.3399
15	522.3556
16	524.3707
17	675.5441
18	690.5065
19	700.5273
20	701.5592
21	702.5433
22	703.5753
23	706.5388
24	716.5226
25	718.5380
26	724.5289
27	728.5588
28	729.5911
29	730.5387
30	732.5552

	A	B
1	m/z	Rt
2	118.0863	11.40
3	126.0219	4.76
4	132.0768	12.16
5	162.1125	13.48
6	204.1230	13.12
7	220.1180	4.86
8	232.1543	12.52
9	268.1040	3.14
10	300.2897	4.28
11	468.3085	11.53
12	480.3090	7.69
13	494.3243	11.45
14	496.3399	11.28
15	522.3556	11.21
16	524.3707	11.30
17	675.5441	11.09
18	690.5065	4.35
19	700.5273	4.07
20	701.5592	11.03
21	702.5433	4.14
22	703.5753	10.92
23	706.5388	9.70
24	716.5226	4.26
25	718.5380	4.21
26	724.5289	3.81
27	728.5588	4.06
28	729.5911	10.91
29	730.5387	9.23
30	732.5552	9.65

- **Added the retention time**

Now we deal with the areas of each metabolite

Make another new Excel file

Highlight and copy the metabolite peak areas

iPSC-T1R1	144301	459870	465781	43557	23803	10585	2875		32290	1771	2066	2212
iPSC-T1R2	146452	405380	456427	49718	25064	9483	3040	1607	24231	1438	1706	2663
iPSC-T1R3	159604	459803	494302	58249	28873	11310	3573	322	33665	1827	2133	3297
iPSC-T2R1	54013	111315	39520	16884	10445	8283	2267	10701	30264	4500	5902	14685
iPSC-T2R2	55288	103051	36568	12434	13572	7752	1956	10157	28371	2501	5823	8383
iPSC-T2R3	50295	132147	48141	17086	13467	8117	1746	9497	25047	3434	6020	11877
m15-T1R1	104109	521094	298459	82978	58040	11909	4748	2558	40221	3261	18040	12788
m15-T1R2	116864	345155	204896	62559	60942	15376	7944	3616	65597	6972	25374	17977
m15-T1R3	123262	550930	373895	104781	78014	14255	6328	4270	36550	5103	22821	13343
m15-T2R1	18045	155927	30721	15399	14093	17510	6241	2526	26985	5683	8912	13782
m15-T2R2	25772	176304	40126	15273	17948	19908	7011	2647	26934	5496	16891	14454
m15-T2R3	29957	196313	42868	18291	17926	18640	7777	2671	26344	5604	8557	15070
mESC-T1R1	102812	621628	863781	89799	59787	17575	8756		29871	3304	2420	4214
mESC-T1R2	97364	625820	823663	91218	58049	18093	8075		35639	2519	2808	4256
mESC-T1R3	117925	710433	903878	95964	60402	19692	8320	4575	39898	3752	2950	5938
mESC-T2R1	9071	46063	15248	6057	5318	5397	2620	9162	47069	6486	4769	19408
mESC-T2R2	9486	76412	27482	9526	5925	4725	1843	7470	45466	6668	3810	15451
mESC-T2R3	9903	59713	21785	8107	5779	5208	2156	7624	45168	6925	4485	18290

Go to the new file

Transposed peak areas

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R
1	iPSC-T1R1	iPSC-T1R2	iPSC-T1R3	iPSC-T2R1	iPSC-T2R2	iPSC-T2R3	m15-T1R1	m15-T1R2	m15-T1R3	m15-T2R1	m15-T2R2	m15-T2R3	mESC-T1R1	mESC-T1R2	mESC-T1R3	mESC-T2R1	mESC-T2R2	mESC-T2R3
2	144301	146452	159604	54013	55288	50295	104109	116864	123262	18045	25772	29957	102812	97364	117925	9071	9486	9903
3	459870	405380	459803	111315	103051	132147	521094	345155	550930	155927	176304	196313	621628	625820	710433	46063	76412	59713
4	465781	456427	494302	39520	36568	48141	298459	204896	373895	30721	40126	42868	863781	823663	903878	15248	27482	21785
5	43557	49718	58249	16884	12434	17086	82978	62559	104781	15399	15273	18291	89799	91218	95964	6057	9526	8107
6	23803	25064	28873	10445	13572	13467	58040	60942	78014	14093	17948	17926	59787	58049	60402	5318	5925	5779
7	10585	9483	11310	8283	7752	8117	11909	15376	14255	17510	19908	18640	17575	18093	19692	5397	4725	5208
8	2875	3040	3573	2267	1956	1746	4748	7944	6328	6241	7011	7777	8756	8075	8320	2620	1843	2156
9		1607	322	10701	10157	9487	2558	3616	4270	2526	2647	2671			4575	9162	7470	7624
10	32290	24231	33665	30264	28371	25047	40221	65597	36550	26985	26934	26344	29871	35639	39898	47069	45466	45168
11	1771	1438	1827	4500	2501	3434	3261	6972	5103	5683	5496	5604	3304	2519	3752	6486	6668	6925
12	2066	1706	2133	5902	5823	6020	18040	25374	22821	8912	16891	8557	2420	2808	2950	4769	3810	4485
13	2212	2663	3297	14685	8383	11877	12788	17977	13343	13782	14454	15070	4214	4256	5938	19408	15451	18290
14	66055	62401	64752	106424	79105	89241	134478	225115	174603	152838	172888	154558	141401	142072	141988	164749	161136	162089
15	17191	17167	17290	73596	42390	60631	118673	143841	114514	76747	81848	82498	32849	30477	36011	90418	84608	88738
16	108693	103626	113683	108808	93930	93300	145222	204910	168732	127989	146755	129175	236806	223285	221103	172712	162789	159881
17	29929	26473	29008	23607	20798	21648	36131	28548	37382	28841	23577	28006	42783	42586	42049	29250	33489	31004
18	18084	16551	17398	29767	30644	30625	30566	18441	26919	42641	40923	41504	13586	15850	15377	24357	23999	22331
19	13998	15807	15077	25040	25195	26792	62225	97571	61288	52257	48671	51941	14737	15808	17302	26367	25717	22931
20	84947	79009	86398	71358	62355	62814	80863	75156	94941	106423	108826	104474	139881	142519	132411	98125	96742	91781
21	52490	59667	55648	71131	75987	79332	192965	81285	178041	110242	93827	114506	43789	46463	43294	58192	69277	54190
22	1144931	1094742	1193675	477444	460519	472838	1443859	849973	1367595	860919	836003	823144	1306052	1339528	1334168	436096	733519	640063
23	759439	718293	801996	568769	592863	573382	839920	421555	894680	978676	928944	945656	757892	1205728	1054442	834070	781688	797497
24	42601	36654	41327	81300	82061	81425	97897	66697	90292	105311	111872	106257	29825	33665	30506	58031	50796	49593
25	120745	120621	125354	166662	174231	179203	192310	89865	177485	161255	143471	164831	72027	76156	73158	112985	121816	104639
26	321736	340582	322255	355237	400188	383111	170502	122885	161795	278712	284182	280979	357660	382831	385383	488145	429478	405398
27	38096	43811	40994	59224	64305	65478	184483	85926	183727	88323	86692	98501	33222	33332	33042	45930	46476	40032
28	16647	15772	15824	14530	13107	13800	22316	14807	23600	15367	13721	15054	20411	19992	17685	13698	15953	13081
29	53805	73243	58201	89078	101109	101431	64893	86538	41694	142324	153366	137326	144965	53367	55667	85413	95926	101333
30	679077	1025469	803417	902646	1071880	1068033	801055	907726	528219	1451514	1484609	1413636	1521451	570753	734753	1017944	1131446	1173389

Now we can select (one column at a time) the peak area data and add it to the *m/z* and Rt data

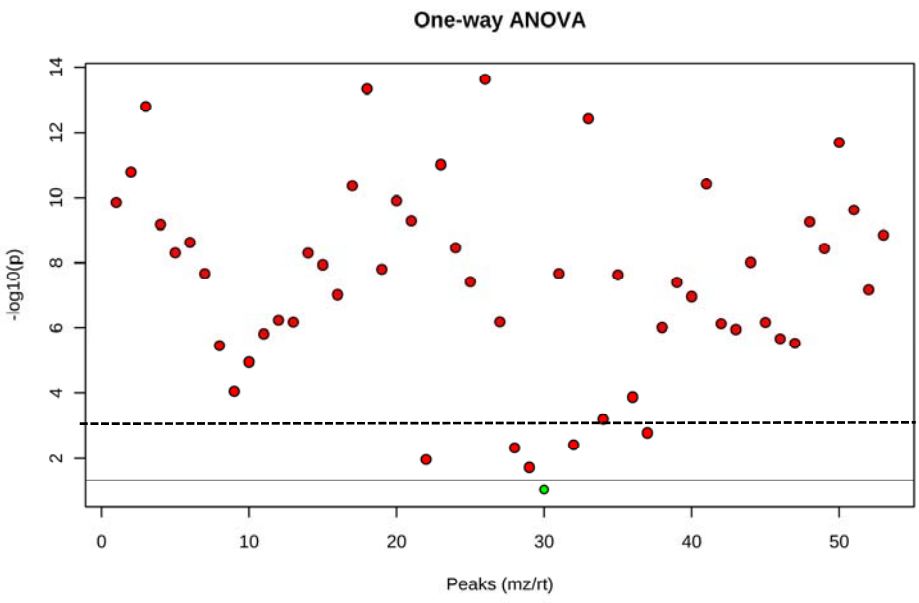
	A	B	C
1	m/z	Rt	iPSC-T1R1
2	118.0863	11.40	144301
3	126.0219	4.76	459870
4	132.0768	12.16	465781
5	162.1125	13.48	43557
6	204.1230	13.12	23803
7	220.1180	4.86	10585
8	232.1543	12.52	2875
9	268.1040	3.14	
10	300.2897	4.28	32290
11	468.3085	11.53	1771
12	480.3090	7.69	2066
13	494.3243	11.45	2212
14	496.3399	11.28	66055
15	522.3556	11.21	17191
16	524.3707	11.30	108693
17	675.5441	11.09	29929
18	690.5065	4.35	18084
19	700.5273	4.07	13998
20	701.5592	11.03	84947
21	702.5433	4.14	52490
22	703.5753	10.92	1144931
23	706.5388	9.70	759439
24	716.5226	4.26	42601
25	718.5380	4.21	120745
26	724.5289	3.81	321736
27	728.5588	4.06	38096
28	729.5911	10.91	16647
29	730.5387	9.23	52805
30	732.5552	9.65	679077

Save as a .csv file

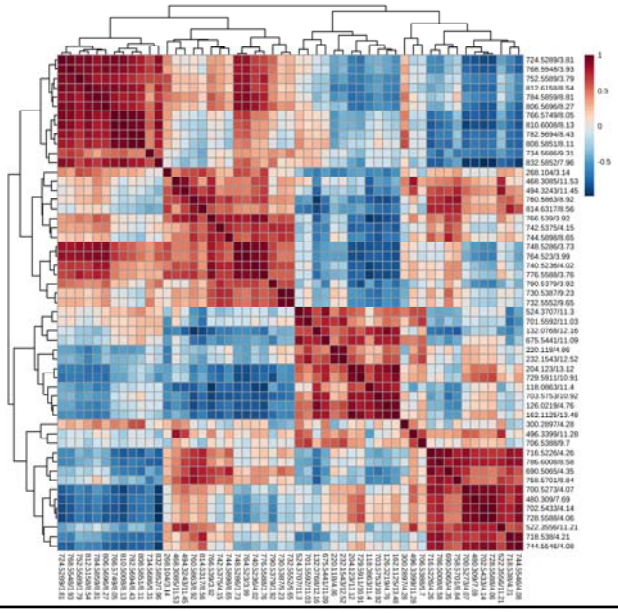
Repeat this for each biological sample

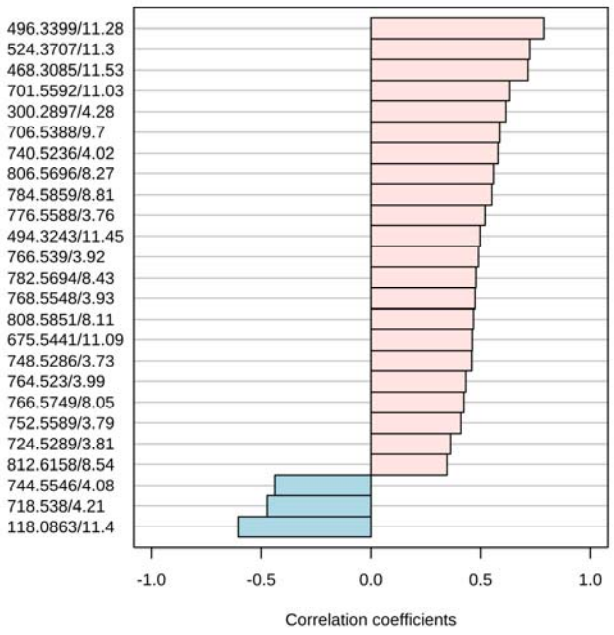
Combine files for each sample type into a folder

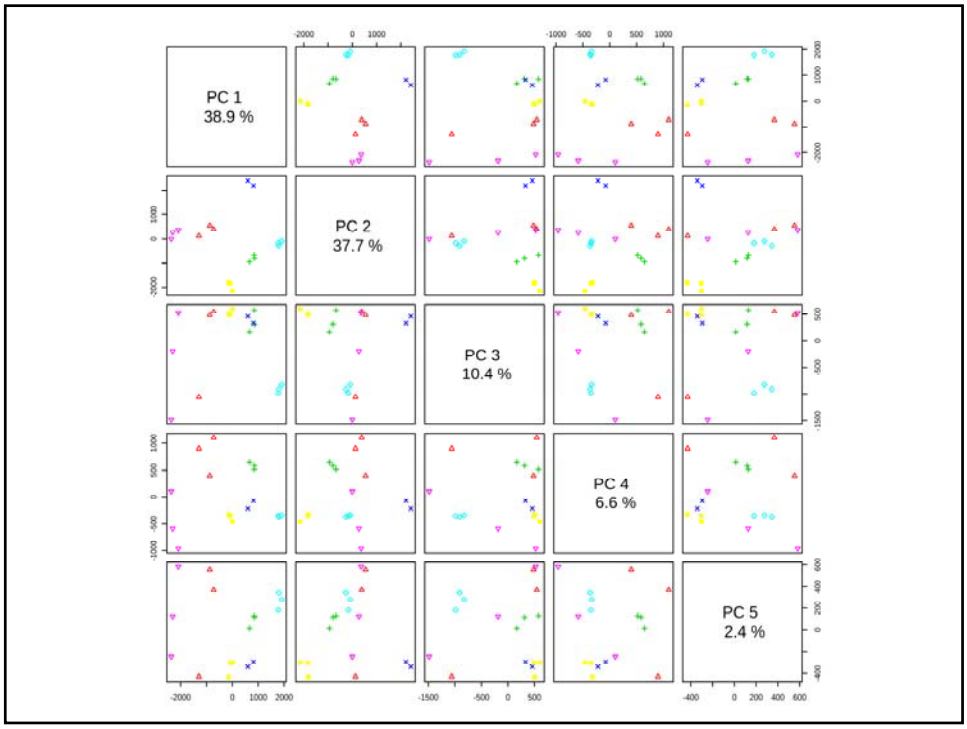
Combine folders into a .zip file and submit to MetaboAnalyst

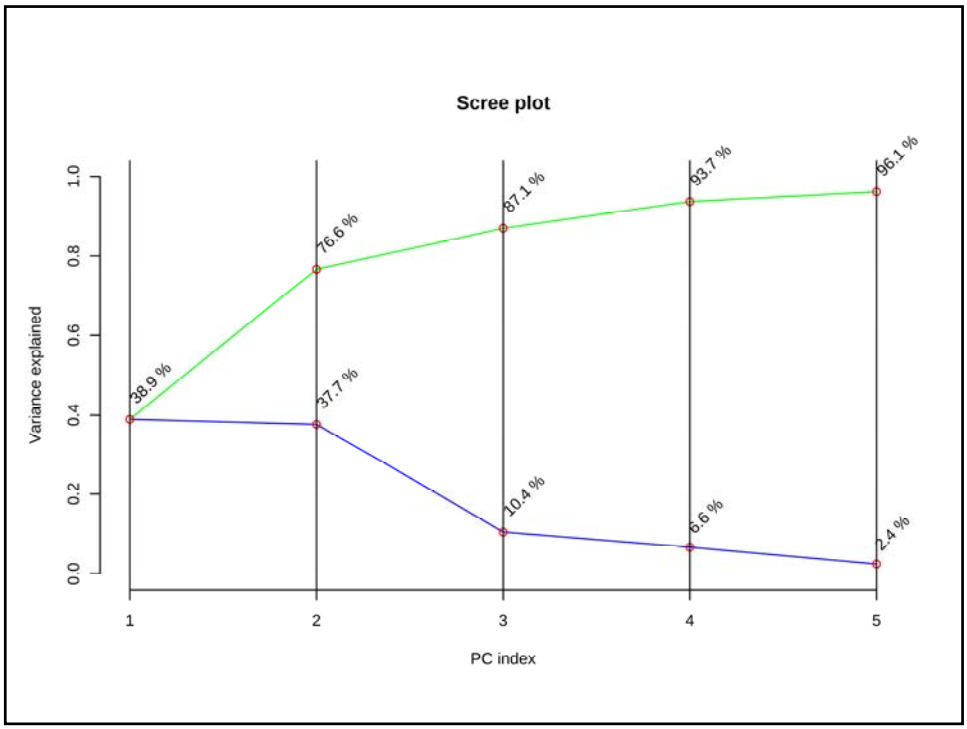


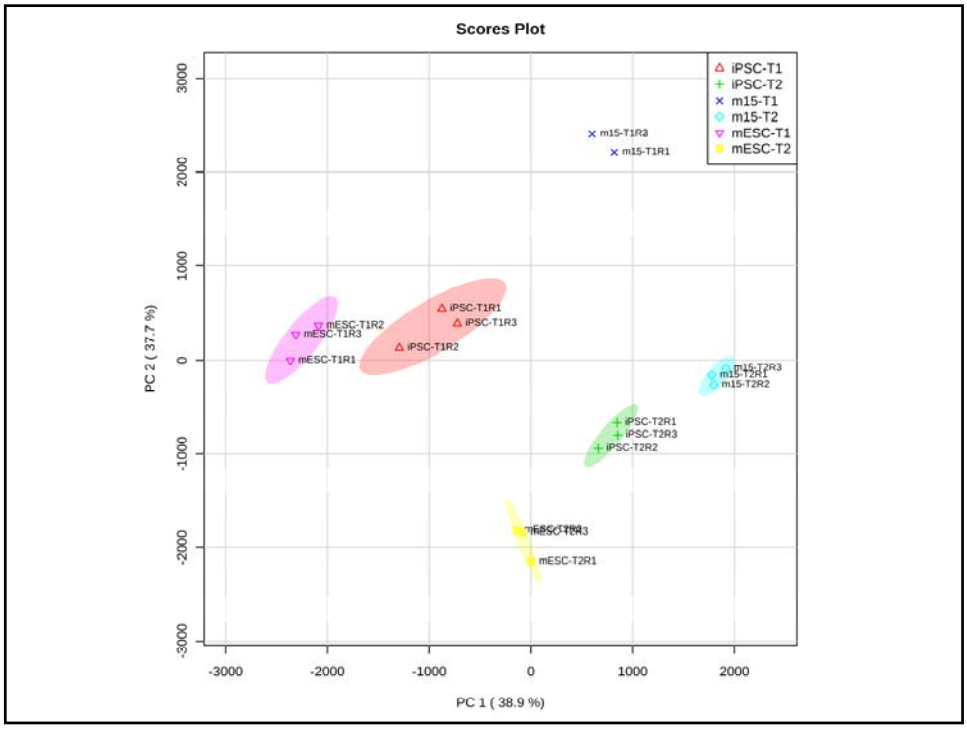
Correlation analysis

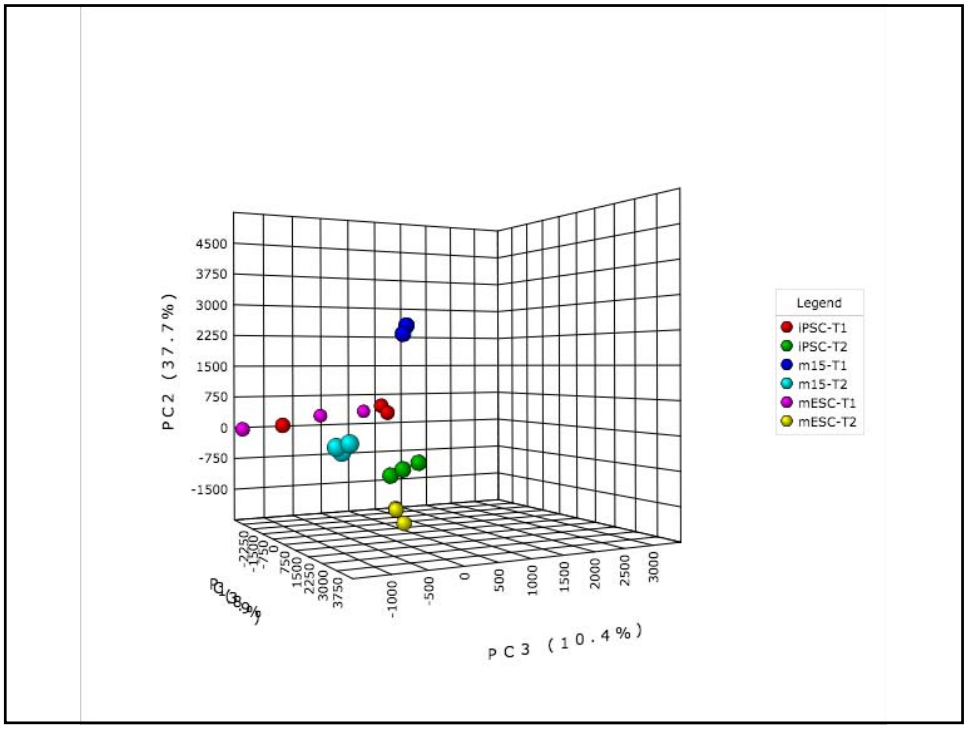


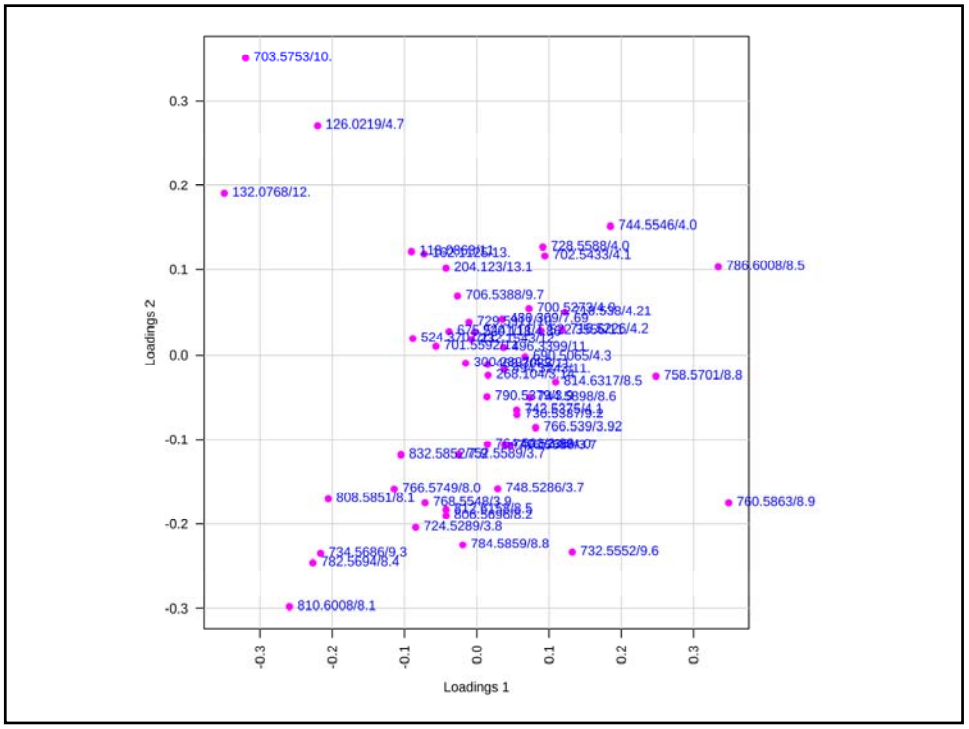


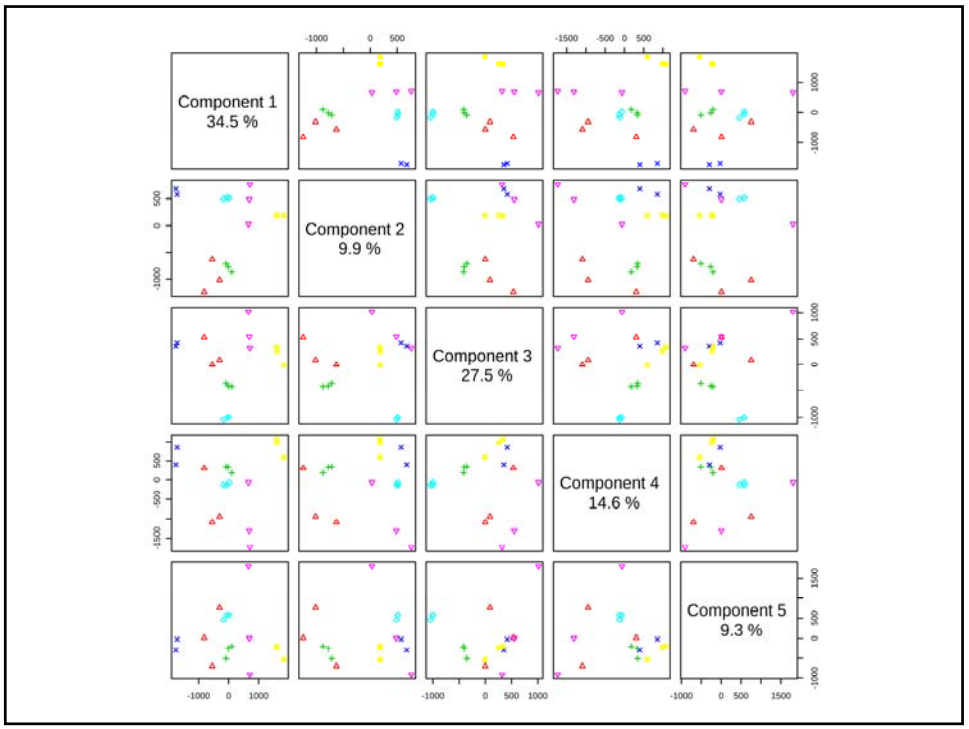

PC 1
38.9 %
PC 2
37.7 %
PC 3
10.4 %
PC 4
6.6 %
PC 5
2.4 %


Scree plot
Variance explained
PC index
38.9 %
76.6 %
87.1 %
93.7 %
96.1 %
37.7 %
10.4 %
6.6 %
2.4 %

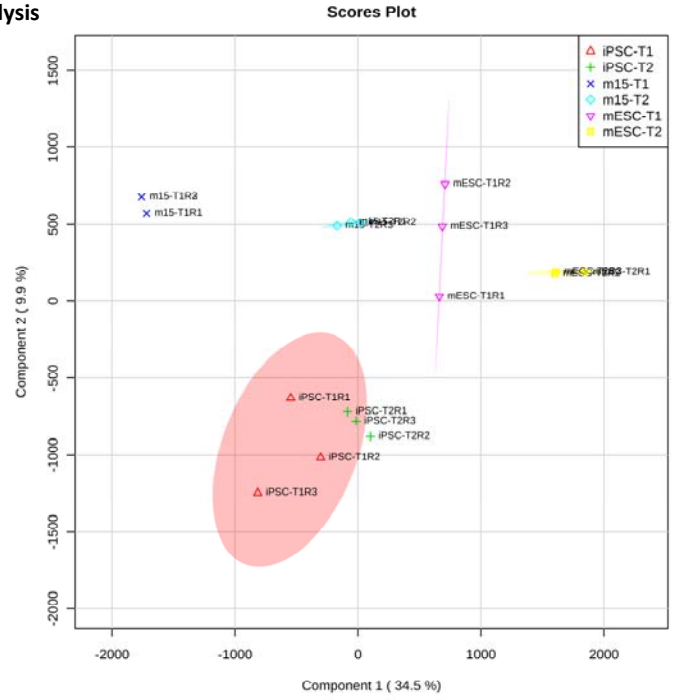

Scores Plot
iPSC-T1
iPSC-T2
m15-T1
m15-T2
mESC-T1
mESC-T2
m15-T1R3
m15-T1R1
iPSC-T1R1
iPSC-T1R3
iPSC-T1R2
mESC-T1R2
mESC-T1R3
mESC-T1R1
m15-T2R3
m15-T2R1
m15-T2R2
iPSC-T2R1
iPSC-T2R3
iPSC-T2R2
mESC-T2R1
PC 2 (37.7 %)
PC 1 (38.9 %)


Legend
iPSC-T1
iPSC-T2
m15-T1
m15-T2
mESC-T1
mESC-T2
PC 2 (37.7 %)
PC 3 (10.4 %)

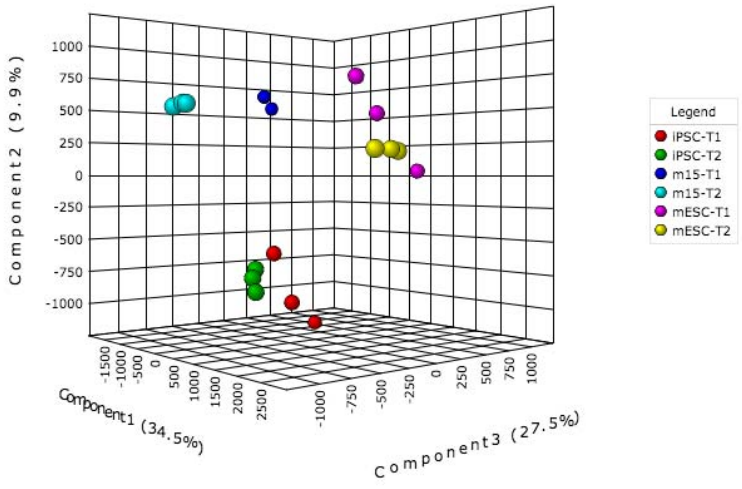

703.5753/10.
126.0219/4.7
132.0768/12.
744.5546/4.0
728.5588/4.0
702.5433/4.1
204.123/13.1
786.6008/8.5
706.5388/9.7
758.5701/8.8
766.539/3.92
766.5749/8.0
748.5286/3.7
808.5851/8.1
760.5863/8.9
724.5289/3.8
784.5859/8.8
732.5552/9.6
734.5686/9.3
782.5694/8.4
810.6008/8.1
Loadings 1
Loadings 2

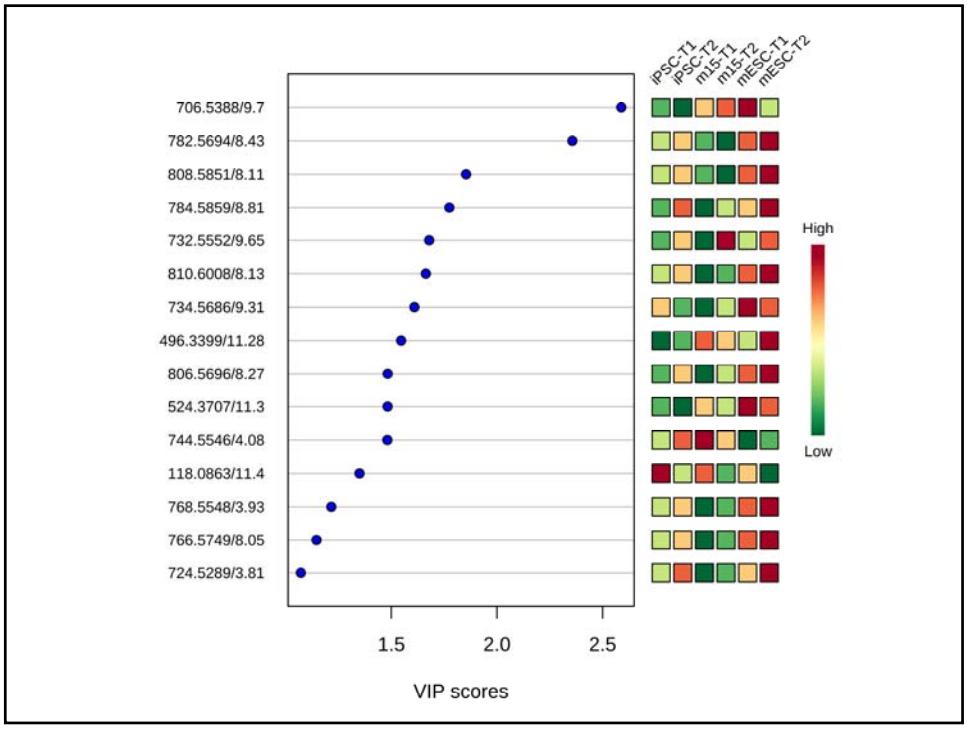

Component 1
34.5 %
Component 2
9.9 %
Component 3
27.5 %
Component 4
14.6 %
Component 5
9.3 %

PLS-DA analysis

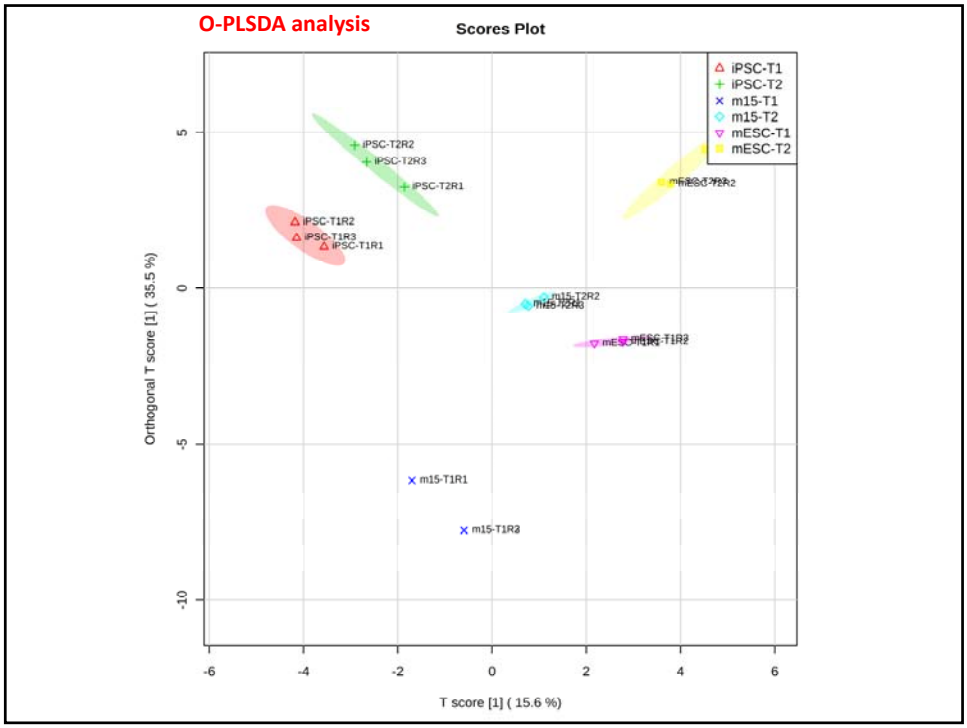


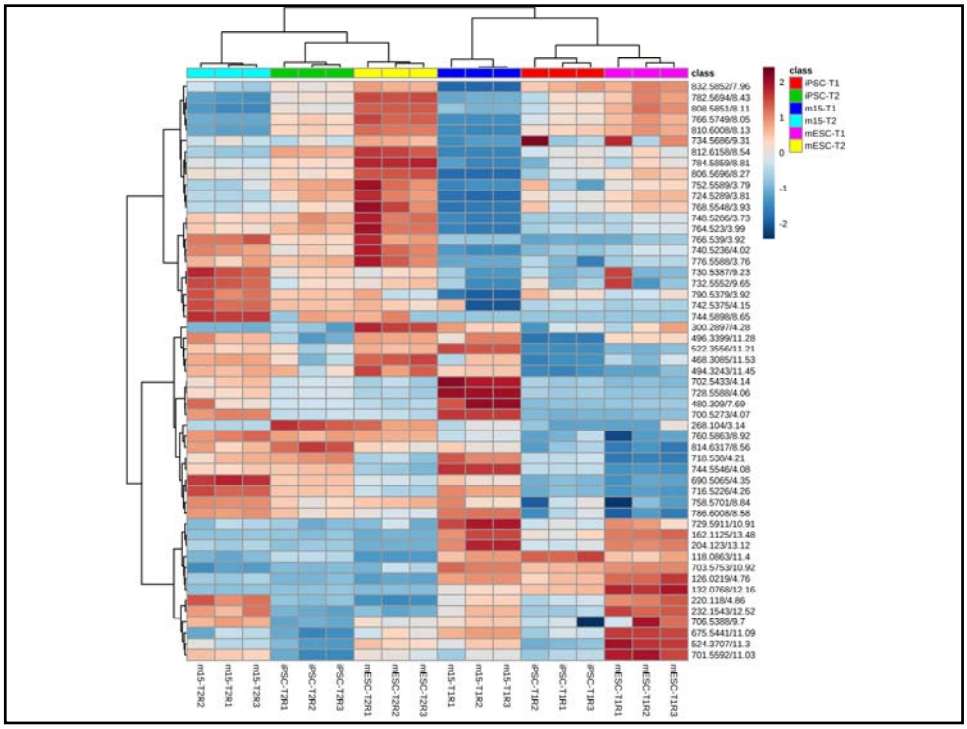
3D-PLSDA analysis





O-PLSDA analysis




class
iPSC-T1
iPSC-T2
m15-T1
m15-T2
mESC-T1
mESC-T2
2
1
0
-1
-2
832.5852/7.96
782.5694/8.43
808.5851/8.11
766.5749/8.05
810.6008/8.13
734.5686/9.31
812.6158/8.54
784.5869/8.81
806.5696/8.27
752.5589/3.79
724.5289/3.81
768.5548/3.93
740.5206/3.73
764.523/3.99
766.539/3.92
740.5236/4.02
776.5588/3.76
730.5387/9.23
732.5552/9.65
790.5379/3.92
742.5375/4.15
744.5898/8.65
300.2897/4.29
496.3399/11.28
522.3556/11.21
468.3085/11.53
494.3243/11.45
702.5433/4.14
728.5588/4.06
480.309/7.69
700.5273/4.07
268.104/3.14
760.5863/8.92
814.6317/8.56
718.539/4.21
744.5546/4.08
690.5065/4.35
716.5226/4.26
758.5701/8.84
786.6008/8.58
729.5911/10.91
162.1125/13.48
204.123/13.12
118.0863/11.4
703.5753/10.92
126.0219/4.76
132.0768/12.16
220.118/4.86
232.1543/12.52
706.5388/9.7
675.5441/11.09
624.3707/11.3
701.5592/11.03
m15-T2R2
m15-T2R1
m15-T2R3
iPSC-T2R1
iPSC-T2R2
iPSC-T2R3
mESC-T2R1
mESC-T2R2
mESC-T2R3
m15-T1R1
m15-T1R2
m15-T1R3
iPSC-T1R2
iPSC-T1R1
iPSC-T1R3
mESC-T1R1
mESC-T1R2
mESC-T1R3