Using Metaboanalyst – hands-on

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THE UNIVERSITY OF
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Knowledge that will change your world

T argeted
M etabolomics &
P roteomics
L aboratory

Univariate Analysis

    Fold Change Analysis   T-tests   Volcano plot

    One-way Analysis of Variance (ANOVA)

    Correlation Analysis   Pattern Searching

Chemometrics Analysis

    Principal Component Analysis (PCA)

    Partial Least Squares - Discriminant Analysis (PLS-DA)

    Sparse Partial Least Squares - Discriminant Analysis (sPLS-DA)

    Orthogonal Partial Least Squares - Discriminant Analysis (orthoPLS-DA)

Feature Identification

    Significance Analysis of Microarray (and Metabolites) (SAM)

    Empirical Bayesian Analysis of Microarray (and Metabolites) (EBAM)

Cluster Analysis

    Hierarchical Clustering: Dendrogram   Heatmaps

    Partitional Clustering: K-means   Self Organizing Map (SOM)

Classification & Feature Selection
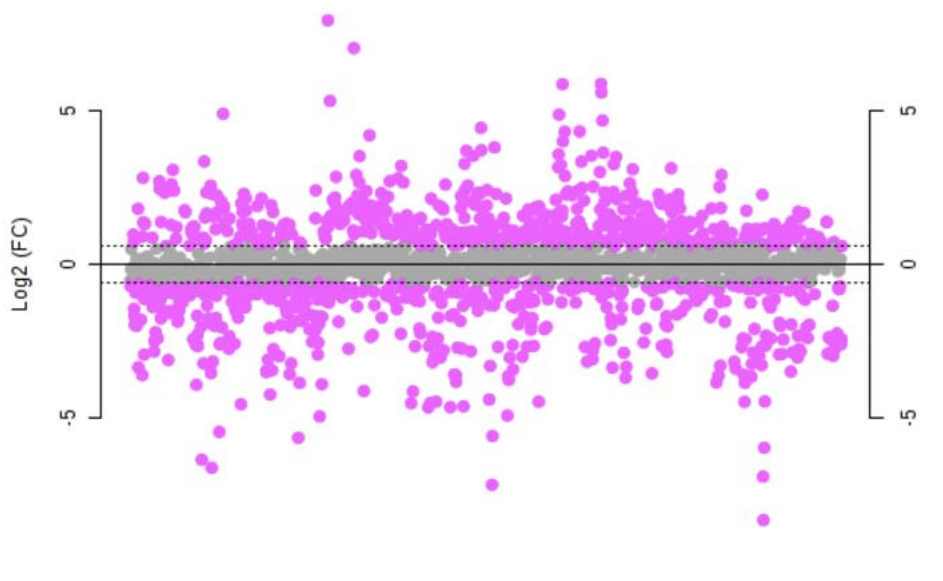
    Random Forest

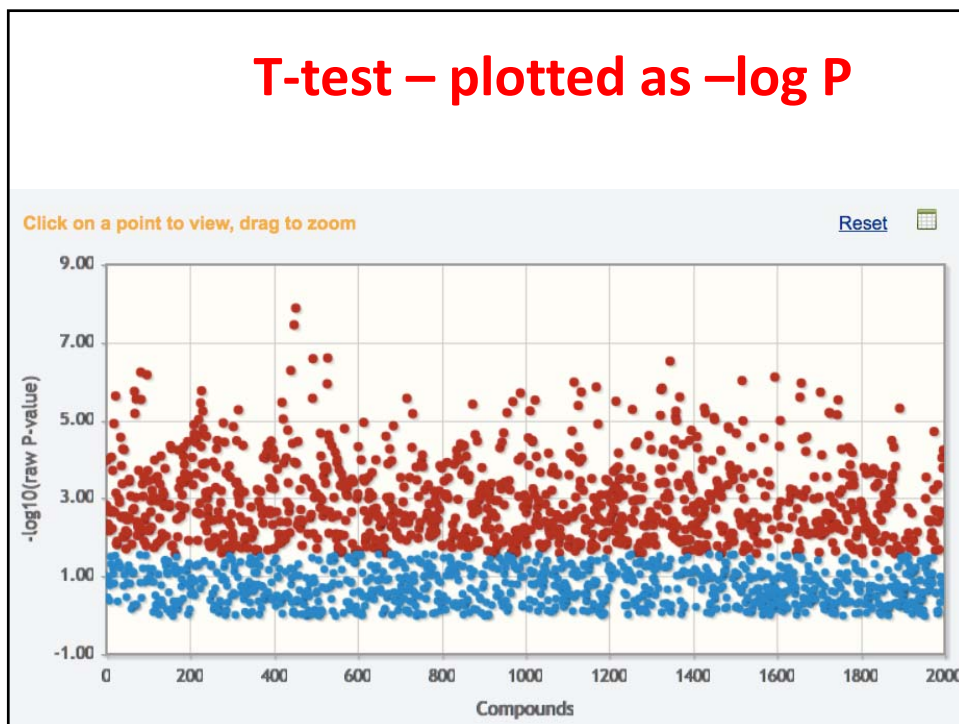   Support Vector Machine (SVM)

Statistical procedures

Univariate analyses
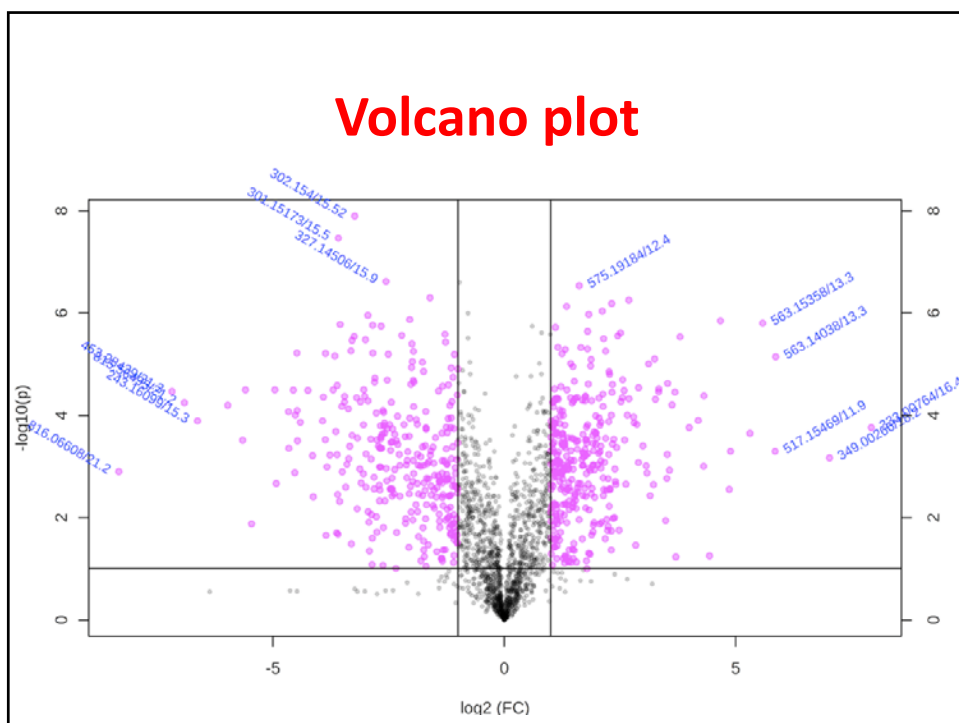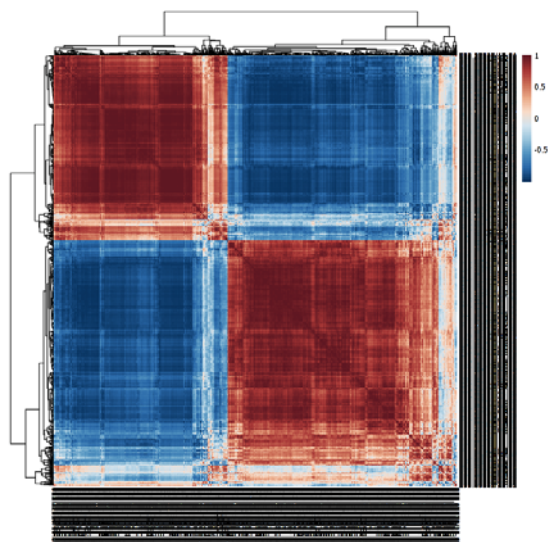
Fold changes <-1.5 >1.5

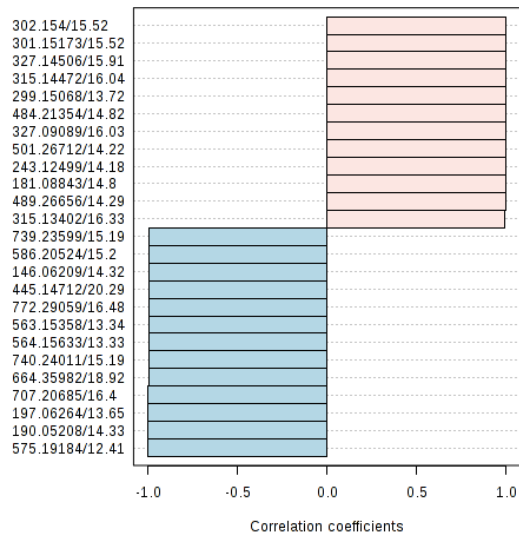# T-test – plotted as –log P
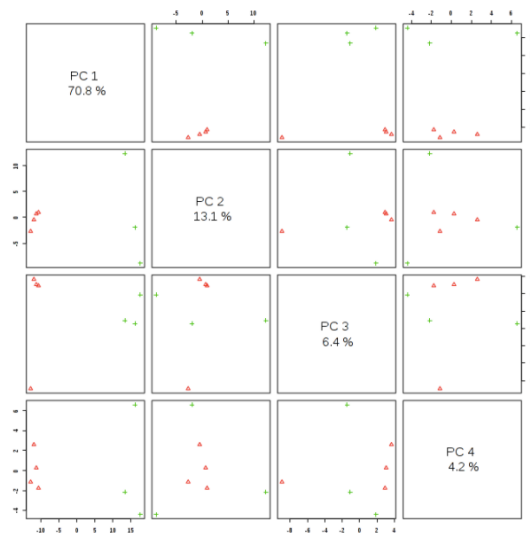

Volcano plot

Correlations plot

Pattern hunter
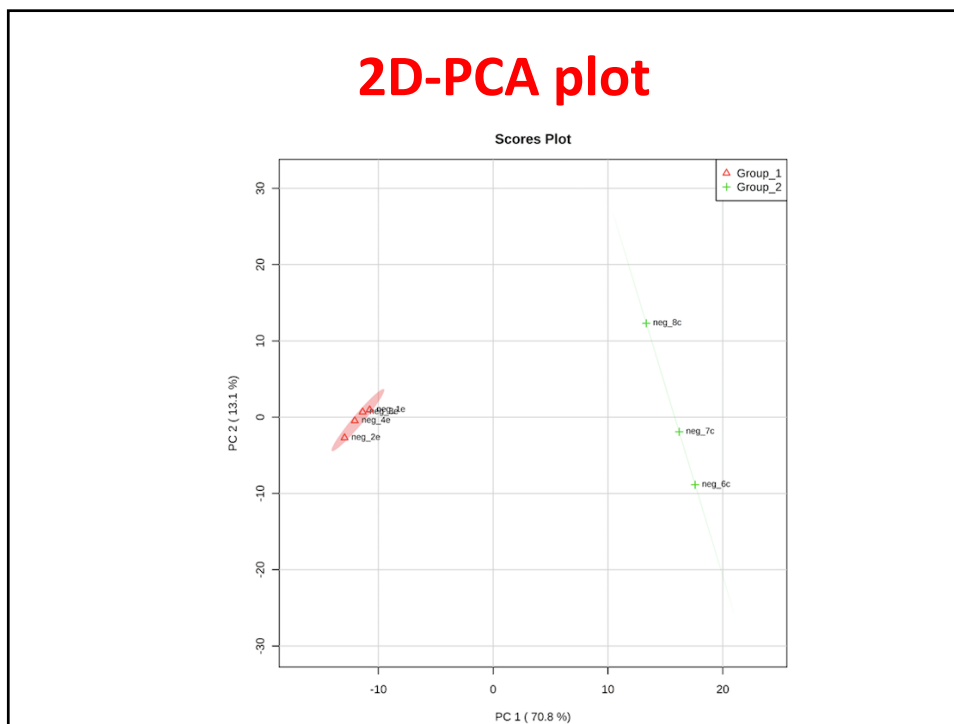
Top 25 peaks (mz/rt) correlated with the 1-2
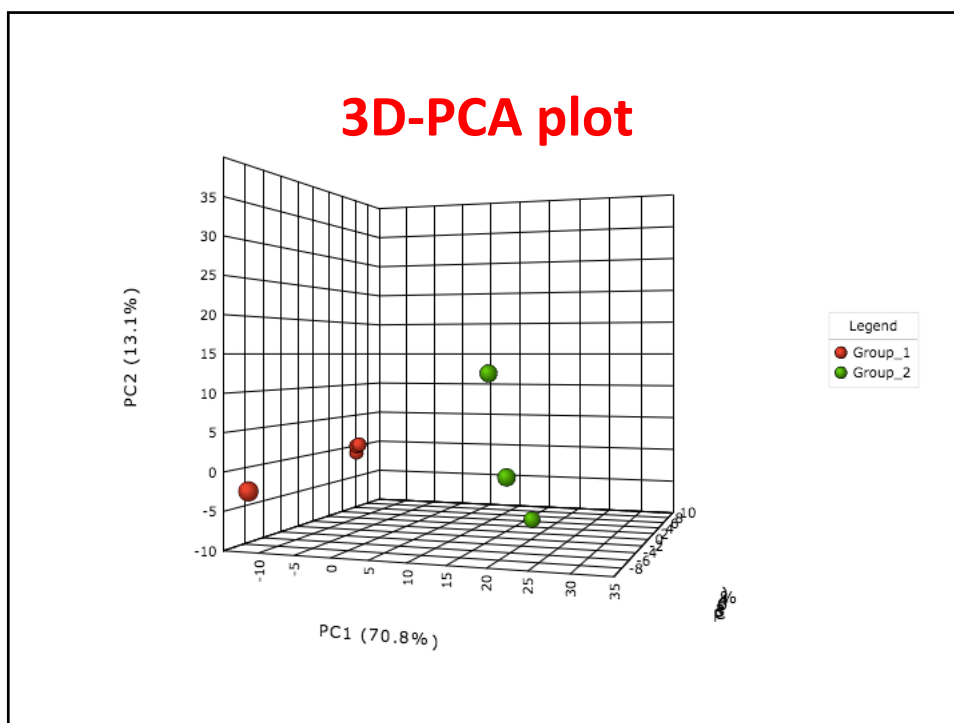
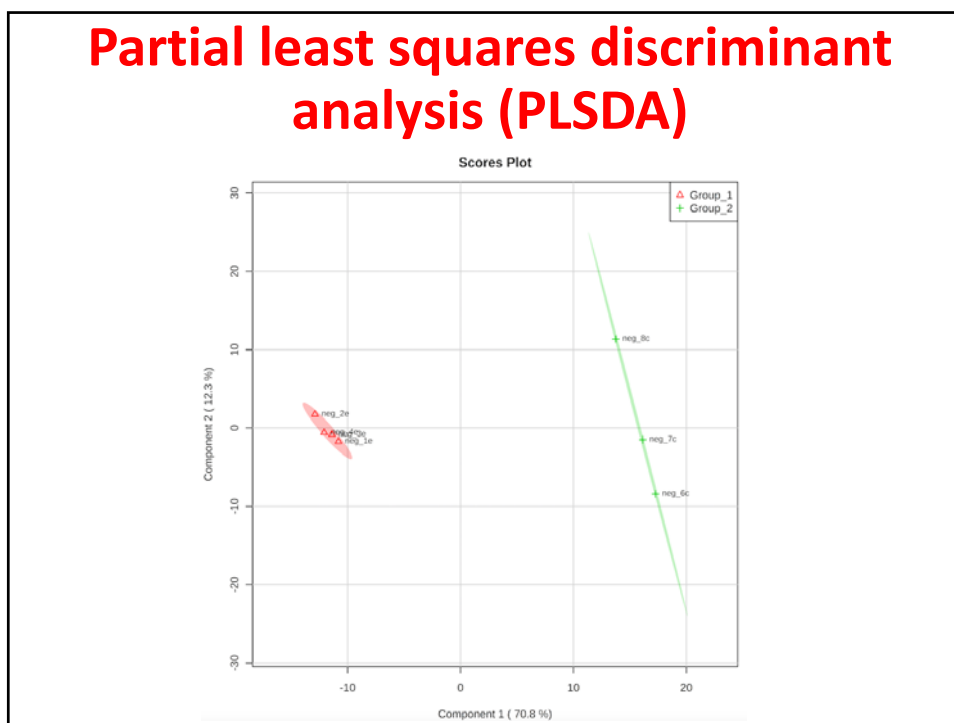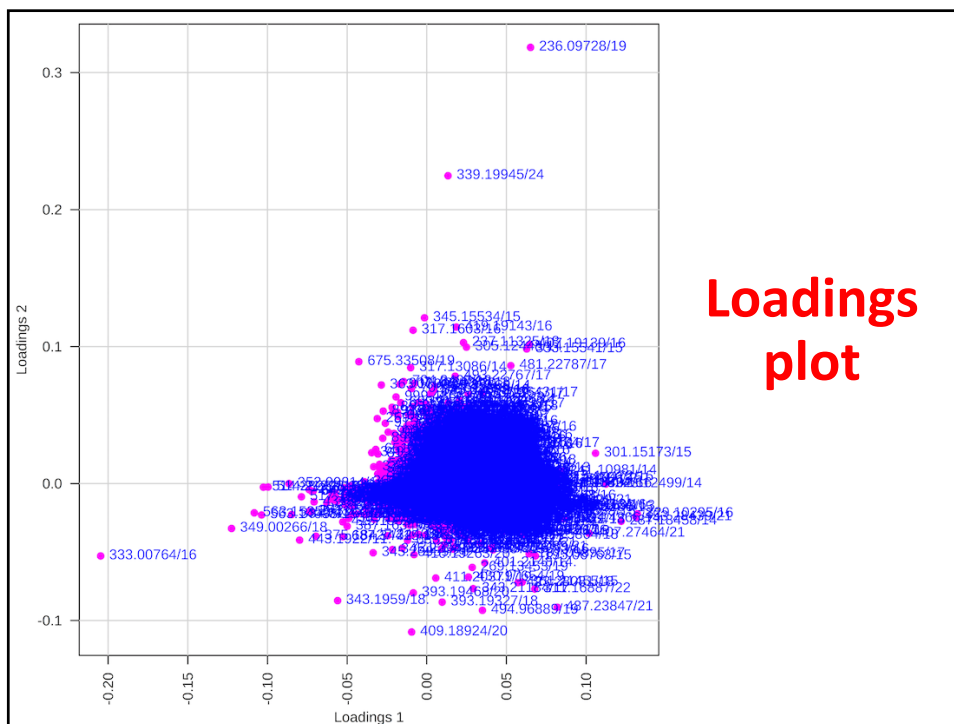# Multivariate analyses

# Principal components analysis (PCA)

2D-PCA plot

3D-PCA plot

Loadings plot

Partial least squares discriminant analysis (PLSDA)

Loadings plot

Variable in projection

Mass Defect

- **On the scale of ^{12}C – 12.0000, the other elements have non-integer atomic weights**
  - ^{1}H = 1.007825   +0.007825
  - ^{14}N = 14.003074   +0.003074
  - ^{16}O = 15.994914   -0.005086
  - ^{31}P = 30.973761   -0.026239
  - ^{32}S = 31.972071   -0.027929
- **H-rich metabolites have strong positive mass defects, whereas those with many oxygens, particularly phosphate and sulfate, have a low or even negative mass defects**

http://physics.nist.gov/cgi-bin/Compositions/stand_alone.pl

Mass defects across sample set

	mzmed	Nominal mass	Delta
1	mzmed	Nominal mass	Delta
2	407.191392	407	0.19139239
3	333.155409	333	0.15540888
4	481.227872	481	0.22787241
5	419.191434	419	0.19143395
6	345.155342	345	0.15534179
7	329.102953	329	0.10295299
8	453.284388	453	0.28438771
9	555.264314	555	0.26431404
10	287.184577	287	0.18457711
11	493.227675	493	0.22767484
12	236.097277	236	0.0972769
13	241.109806	241	0.10980586
14	317.130863	317	0.13086284
15	305.138376	305	0.13837606
16	243.124987	243	0.1249871
17	315.144717	315	0.14471681
18	311.168872	311	0.16887233
19	389.180407	389	0.18040723
20	407.274637	407	0.27463734
21	301.151733	301	0.15173284
22	395.191336	395	0.19133615
23	379.165732	379	0.16573173
24	315.134024	315	0.13402411
25	361.214545	361	0.2145451
26	343.195899	343	0.19589949
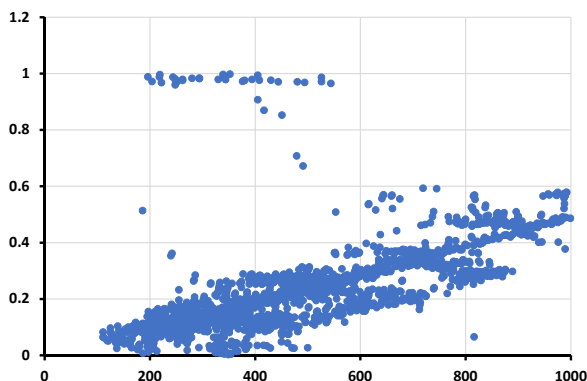
Other multivariate methods

- Sparse PLSDA (sPLSDA)
  -
- Orthogonal PLSDA (oPLSDA)
  -

- For this data set, they were not needed

Heat map – top 25 ions