Demonstration of Metabolomics for Evaluation of Environmental Toxicants

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Introduction

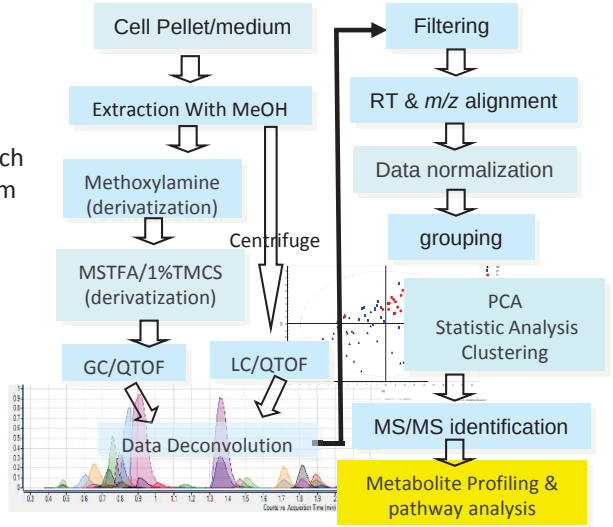
It has been observed that low level exposure to arsenite (75ppb) is sufficient to induce aerobic-glycolysis in cultured human primary lung epithelial cells (BEAS-2B) [1]. With the help of modern analytical instrument, especially high resolution mass spectrometry (QTOF) , we are trying to do both global and target driven analysis of cellular metabolites to further explore where and how arsenite is reacting within the cells.

Experimental Details

Cell Exposure

	Group (V)	Group (VAs)	Group (Sh)	Group (ShAs)
Cells	Lung epithelial cell BEAS-2B			
Knockdown	-	-	H1F-1A	H1F-1A
Dose		1uM Arsenite		1uM Arsenite
Exposure		4 weeks		4 weeks
Replicates	4	4	4	4
Cell counting	Y	Y	Y	Y

Research Diagram

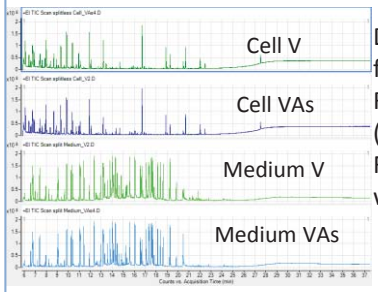


Instrumentation

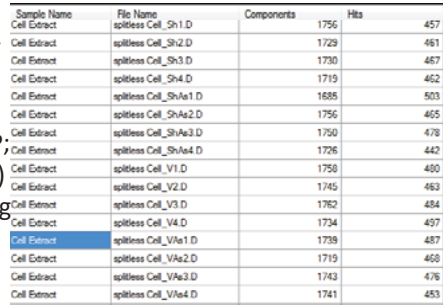
Agilent 7200 GC-QTOF

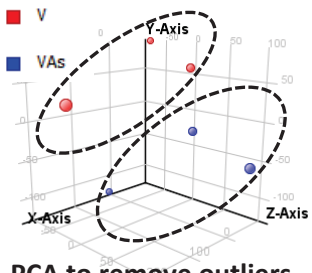
Agilent 6540 LC-QTOF

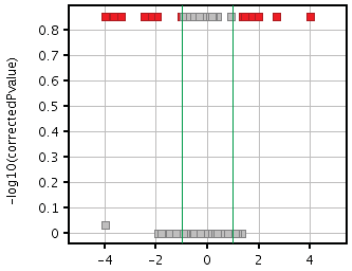
Results and Discussion

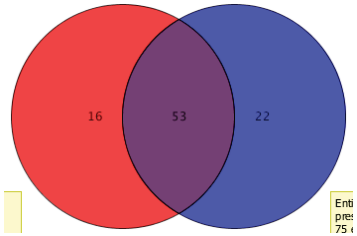


Deconvolution (GC) or molecular feature extraction (LC) first; RT (0.1min) and mass alignment (15ppm) were conducted in MPP; Fiehn, NIST (GC) and METLIN (LC) were used for database screening









PCA to remove outliers **FC>2.0, Moderated T-test (p<0.05)** **Venn Diagram (67% presence)**

Metabolites List

Metabolites	Regulation	RT	Mass
L-(+) lactic acid	down	6.779667	190.085
tyrosine 2	down	17.8205	218.1104
L-valine	down	9.086	144.1205
3-phosphoglyceric acid	down	16.411	317.0345
alpha-glucosamine 1-phosphate	down	16.7002	56.03719
citrulline	up	16.638	157.0868
tyrosine	up	17.8205	218.1104
L-alanine	up	7.439	116.0896
L-leucine	up	8.254	86.09671

Further biological pathway validation

References

[1] Zhao, F., Severson, P., Pacheco, S., Futscher, B.W., Klimecki, W.T., 2013. Arsenic exposure induces the Warburg effect in cultured human cells. Toxicology and applied pharmacology.

Acknowledgements

Agilent Technologies

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