

Can metabolomics identify unintended effects in transgenic potato tubers?

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Introduction

It is imperative that the public's concerns regarding the safety and value of genetically modified foods be addressed, if these are to be commercially successful. One issue under debate is whether genetic modification is accompanied by unexpected metabolic perturbations. This study aims to assess the capability of GC/MS and LC/MS approaches at screening for such unintended effects and establishing "substantial equivalence". The data described is from a replicated field trial designed to investigate the effects of transgenesis, empty vector insertion, and the tissue culture procedures within the common cultivar, Desiree.

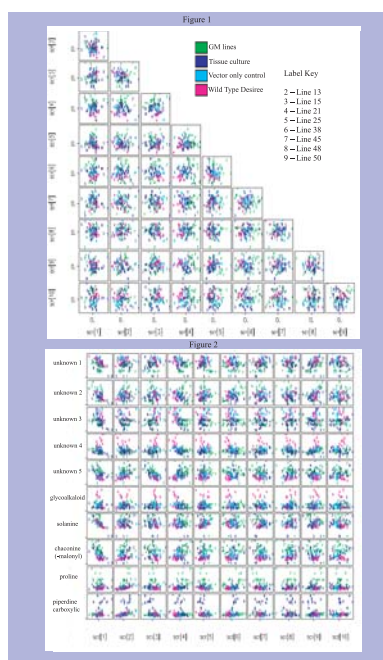
Plant Materials

The sub-set chosen for this study (8 lines, 96 individual samples) were as follows:

Line Number	Type	Treatment
50	Desiree Wild Type	None
38	Desiree TCI	Tissue culture grown
15	W2GBSSV04	Transformed with 'empty' pBIN19 vector
13	W2GBSS47	Potato GBSS promoter, putative pullulan (W2) gene and CAMV polyA tail
25	MAL2V1	Transformed with 'empty' pBIN19 vector
21	MAL15A	X2 35S promoter, anti-sense Mal1 gene and CAMV polyA tail
48	SAMPATVC1	Transformed with 'empty' pBIN19 vector
45	SAMPATIE	Patatin promoter, sense SAMDC gene and nos polyA tail

Samples taken from each line comprise of tubers grown in 4 separate plots in a full replicated field trial (2001 season). Samples from each plot were taken in triplicate, resulting in a total of 12 samples for each line.

Results

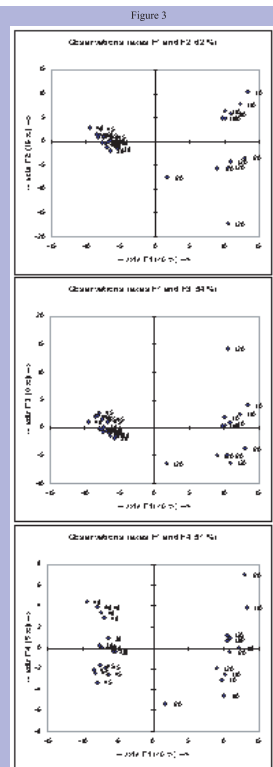


Visual display of results is produced by Principal Components Analysis (PCA) using Genstat and ExcelSTAT.

Score plots showed separation of treatments (Figure 1).

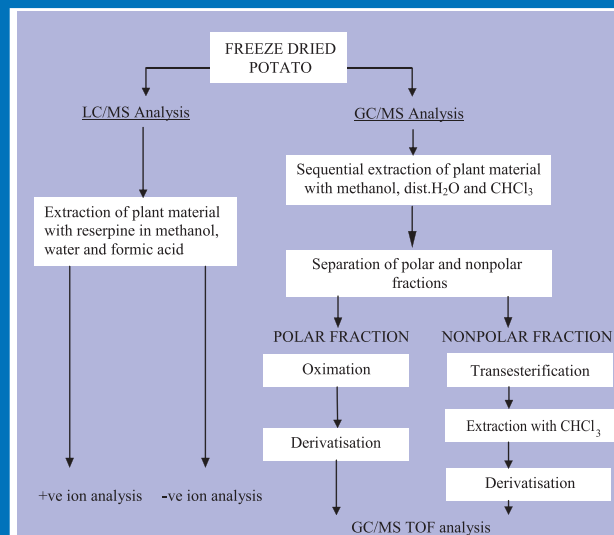
Different treatments had varying effects on individual compounds (Figure 2).

PCA analysis of the LC/MS data (including Pentland Javelin samples) showed a greater variation was present between the two land races than between the plants of different treatments (Figure 3).



Key:
Line 110: Desiree wild type
Line 120: Pentland Javelin
Line 38: Desiree TCI (tissue culture control)
Line 45: SAMPAT IE (transgenic)
Line 48: SAMPAT VC1 (empty vector control)

Methods and data reduction



Data reduction has been carried out using Xcalibur™ and employing a number of user-created processing methods. In addition, AMDIS has aided the visualisation of near-co-eluting components. From the 750 components identified by our methods, we used 178 in this analysis.

Conclusions

- An untargeted (metabolomics) approach is capable of rapidly providing significantly large data on large sample numbers.
- Traditional multi-variate approaches are able to identify variation within large sample sets and begin to address "substantial equivalence" in transgenic materials.
- Data indicate numerous perturbations that result from stable transgenesis, yet also show comparable quantitative perturbations from insertion of empty vectors as well as *in vitro* culture.
- Preliminary data would indicate that the scale of variation observed by treatment (*in vivo* manipulation, empty vector or transgene insertion) is less than that present within cultivars and indeed, subspecies of *Solanum tuberosum* i.e. natural variation was greater than that following genetic modification.

Acknowledgements

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