Understanding the genetics behind mushroom production

TEAGASC research has characterised the genetic activity in mushroom mycelium when mushrooms are actively being produced. This has identified clusters of key genes that mirror crop productivity, as well as genes with interesting expression profiles.

The mushroom industry in Ireland is acknowledged to be one of the best in the world, growing high-quality *Agaricus bisporus* mushrooms for export. It is the largest horticultural sector in Ireland, with a farm gate value of €119 million in 2019, with over 80 % exported to the UK. The industry has improved productivity through harvester training, efficiencies in crop management and increases in compost and casing quality, to a point where average yields are in the region of 30 kg of top-class fresh mushroom product/m^2 – some of the best yields in the world. Significant nutrition remains in mushroom compost at the end of the normal three-flush cropping cycle, so there is potential to increase substrate utilisation, especially in the third flush; however, more information is needed on what mushroom genes relate directly to crop productivity (**Figure 1**).

Microarray analysis

Microarray analysis of gene expression in the compost over the course of a crop was done using custom-designed Agilent microarrays that are available for *A. bisporus*, and which cover the whole genome. Compost samples were obtained every 48 hours from day 13 of the crop cycle until the end of the third flush harvesting (day 37) – 13 time points in total – for comparison to samples taken on day 11. Labelled cRNA was extracted from the compost samples, hybridised to the microarrays, and gene transcript levels were calculated using the appropriate software. Putative temporal gene expression profiles were then identified by comparing transcript levels at each time point with the level detected at day 11 – a time point before the production of mushrooms had started. Using statistical and bioinformatic analysis, the profiles were grouped into clusters of genes with similar expression profiles.

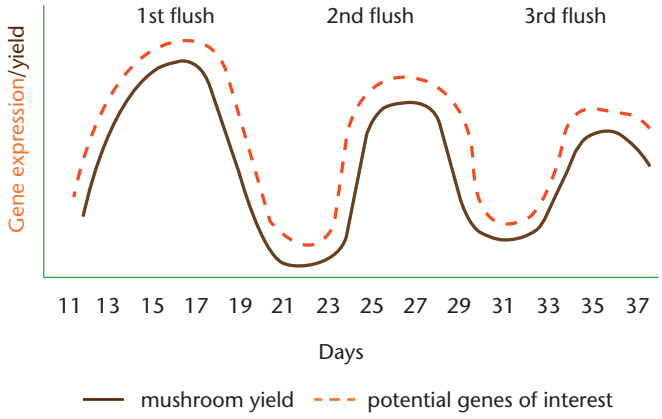

FIGURE 1: Gene expression and yields over the normal three-flush cropping cycle.

Protein expression

Protein expression in compost samples was also measured for seven time points, taken before, during and after the first flush. Proteins were extracted and purified for LC-MS/MS and all of the *A. bisporus* proteins present were identified. Using bioinformatic analysis these proteins were functionally annotated. Fold change calculations were used to identify significantly up- and down-regulated proteins at each time point and compared to the earliest time point to examine differential protein abundances over the first flush cropping period.

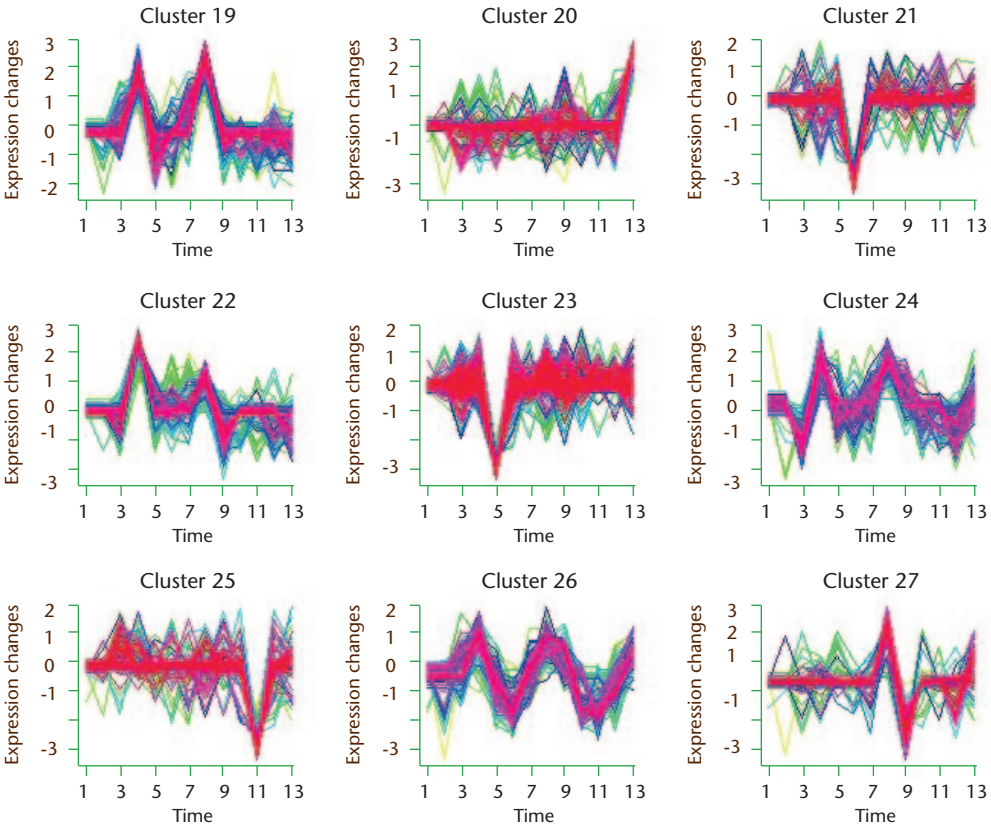

FIGURE 2. Gene expression patterns of A. bisporus *genes.*

Gene clusters and proteins

The gene expression profiles identified by microarray analysis indicated that around 2,000 (20 %) of the *A. bisporus* genes were differentially expressed during the period when mushrooms were being actively produced and these could be aggregated into 40 clusters, each with a distinct expression pattern (see examples in **Figure 2**). Some clusters correlated with the rise and fall of mushroom yield associated with the first, second and third flushes of mushrooms (cluster 26), while some gene clusters were switched on (cluster 20) or off (cluster 19) during the third flush, when mushroom yield was rapidly decreasing. This provides a large bank of data that can be interrogated regarding which genes influence the production of mushrooms. Cluster 26 contained many genes known to be involved in lignocellulose degradation, the main component in mushroom compost. These data have now generated a transcriptomic atlas of *A. bisporus* during mushroom substrate utilisation, which can be referred to with a high degree of confidence. Proteomic analysis identified 558 *A. bisporus* proteins in the period spanning before, during and after the first flush. It was found that the total number of proteins changed similarly to the transcriptome data, but when the gene expression patterns were compared to the proteome data, the proteins took longer to reach their peak.

Opportunity/benefit

This project has increased our understanding of what genes are active or inactive when mushrooms are growing and accessing nutrients from the substrate. This information will enable mushroom breeders and scientists to identify new and interesting genes and pathways that are important for the growth of the mushroom.

Acknowledgements

This work was funded by the Department of Agriculture, Food and the Marine (DAFM) (grant number: 14/S/865). Eoin O'Connor was funded by the Teagasc Walsh Fellowship scheme. Eoin O'Connor and David Fitzpatrick of Maynooth University also contributed to this article.

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