

P058 Put3p phosphorylation modulates proline-independent activation of *PUT1*

M. K. Leverentz & R. J. Reece

University of Manchester

For an organism to prosper it must sense and adapt to a changing environment. In many cases this means utilising sub-optimal nutrient sources. *Saccharomyces cerevisiae* can utilise a wide variety of nitrogen sources. These range from those of high quality, e.g., ammonia and glutamine, to those of low quality, e.g., γ -amino butyric acid (GABA) and proline. Put3p is a DNA-binding transcriptional activator that is responsible for the expression of the *PUT1* and *PUT2* genes (collectively termed the *PUT* genes), which encode enzymes for the utilization of proline as a nitrogen source. Put3p acts as a direct proline sensor, binding to proline when the amino acid is at high intracellular concentrations and inducing the transcription of the *PUT* genes approximately 20-fold. However, the activity of the *PUT* promoters also respond to low quality nitrogen sources other than proline, such as GABA and urea, with 2-4 fold increases in activity. This suggests that when nitrogen quality decreases the cell anticipates the reliance on proline by increasing the levels of Put1p and Put2p, even though proline may not yet be present. We have identified several sites of phosphorylation within Put3p and have shown that two of these post-translational modifications have a modulating effect on the magnitude of the proline-independent, but not proline-dependent, activation of the *PUT1* gene.