

P034 Residues Important for the Function of eIF2B?
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eIF2B plays a crucial role in regenerating active eIF2-GTP after each round of translation initiation and is a target for controlling translation. The core nucleotide exchange function of this complex is localised to a short region of its largest subunit eIF2B ϵ . This region, the catalytic domain, is necessary and sufficient for GEF activity *in vivo* and *in vitro*. By targeting highly-conserved residues within the catalytic centre for site-directed mutagenesis to alanine, we were able to identify two residues which are fundamental to the activity of eIF2B in yeast. Glu569, is essential for eIF2B activity and cell viability. However, by supporting growth through over-expressing eIF2 and the *IMT4* gene, we find that eIF2B ϵ ^{E569A} expresses well and interacts with its substrate eIF2 *in vitro*. Another eIF2B ϵ mutation, L568A, displays slow growth at 15°C also producing a *gcd* phenotype. eIF2B ϵ mutants T552I and S576N, are also cold sensitive, while mutations away from the catalytic domain are not, suggesting a common function of the catalytic domain residues. Further investigations into protein-protein interactions suggest this function is to stabilise eIF2 binding. To provide a greater insight into the mechanism of nucleotide exchange, we are now identifying where the catalytic centre interacts with eIF2.