Cell Chemical Biology, Volume 25

Supplemental Information

The O-GIcNAc Transferase Intellectual Disability

Mutation L254F Distorts the TPR Helix

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Supplemental figure 1: *in vitro* characterisation of OGT_{L254F}. Related to Figure 1B

(A) Michaelis-Menten kinetics of OGT wild type and mutant against RB2 peptide. Xand y-axis intercepts of the dashed lines mark the Michaelis constant (K_M) and maximum velocity (V_{max}) of the reaction. Error bars represent standard deviation of three replicates. (B) Western blot analyses showing OGT activity against de-O-GlcNAcylated HEK-293 cell lysate. Results represent six biological replicates and orange boxes depict the area that was used for quantification. *Cp*OGA: *Clostridium perfringens*; L: L254F; W: wild type.

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Supplemental figure 2: Analysis of the intra-TPR repeat distance. Related to Figures 2B and 2C

Molecular dynamics simulation data investigating the intra TPR distance observed in $sTPR_{WT}$ and $sTPR_{L254F}$ are shown on left and right panels, respectively. sTPR: simplified TPR; TPR: tetratricopeptide repeat.



Supplemental figure 3: Analysis of the angle B-A'-B'. Related to Figures 2B and 2C

Molecular dynamics simulation data investigating the B-A'-B' angle observed in $sTPR_{WT}$ and $sTPR_{L254F}$ are shown on left and right panels, respectively. sTPR: simplified TPR; TPR: tetratricopeptide repeat.