Poster Presentation

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Exploration of strategies to altering thermal properties of industrial enzymes

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Endoproteases and exopeptidases occupy a pivotal position with respect to their commercial applications in food (e.g. as additives in whey protein processing) and, as additives in detergent, textile and a number of other industries. Food processing at low temperatures by cold-active enzymes has many advantages as it minimises undesirable chemical reactions as well as the risk of microbial contamination. Cold-active enzymes were found to display higher specific activity and catalytic efficiency resulting in lower quantities of enzyme required and significantly shortened processing times. On the other hand, industrial hydrolysis typically occur at elevated temperatures due to the faster reaction rates, increased substrate solubility and thermophilic biocatalysts are required to maintain reactions at very high temperatures. The aim of our work is to exploit structure-function relationships of extremophilic enzymes that give rise to novel industrially useful proteases. We are using the high-throughput capability of the Oxford Protein Purification Facility (OPPF) to study a number of structural modifications leading to protein extremophilic functional behaviour. Several strategies to effectively alter the thermal properties of commercial serine endoproteases and aminopeptidases are being tested including; i) site directed mutagenesis targeted to reduce quantity of prolines, salt bridges, S-S bridges, and hydrophobic clusters, and ii) iterative saturation mutagenesis relying on residues with low B-factors (local rigidity) according to available 3D structures are currently being implemented. Our recent results reveal the potential for an emerging universal mechanism to modify the thermostability of any given enzyme.

Keywords: Thermostability, Endoprotease, Extremophilic