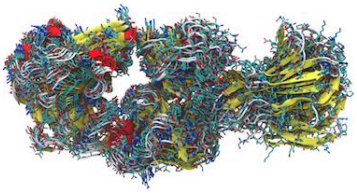

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Improvement of the homodimer interface energy of a β-glycosidase

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Abstract:

β-glycosidases are responsible to catalyze the hydrolysis of β-O-glycoside bond from glycoconjugates releasing a monosaccharide as product. Owing to the enormous variety of substrates present in the nature, β-glycosidases are very diverse in substrate specificity and sequence. β-glycosidases are grouped as Glycosidase Hydrolase family 1 (GH1). Structurally, these enzymes adopt the classical TIM barrel tertiary structure ($(\beta/\alpha)_8$ barrel), which is responsible to shelter the catalytic residues in the top of the barrel. Here, we choose the β-glycosidase from an organism called *Spodoptera frugiperda* (Sfβgly) to study the relationship of its catalytic and structural properties. We found out by gel filtration, that Sfβgly coexists as a dimer and monomer in solution. After separation followed by kinetic assay of each fraction, different catalytic efficiency was observed. It was found the dimer presents a remarkably higher catalytic efficiency against synthetic substrates. The interface of the homodimer consists of four hydrogen-bonds, about 688 $Å^2$ of buried surface area by hydrophobic residues and exhibits a theoretical solvation free energy ($\Delta^i G$) of about −17 kcal mol^{-1}. In addition, mutations have been made at the interface to infer about the critical residues responsible for the dimer stabilization. Now, in an attempt to preserve its superior catalytic rate in solution, we are interested in modifying the dimer interface to increase its stability. Hence Sfβgly would be only in the dimeric state. Thus, we envisage to use Rosetta program to find the lowest free energy of the dimer interface by re-design the sequence of non-critical residues and backbone optimization. Once the models are finished, we will then produce this Sfβgly construction as recombinant protein and experimentally test it for the strictly formation of homodimers.

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