In-silico Prediction of Surface Residue Clusters for Enzyme-Substrate Specificity of highly homologous enzymes

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Introduction

One of the most remarkable properties of enzyme substrate binding is high specificity. Several possible mechanisms have been suggested to illustrate such exquisite substrate specificity, e.g. substrate-binding in the catalytic centers of enzymes, loop-based hinge-motion, and cofactor binding and intra- or inter-molecule (domain-domain) interactions. We believe that there must be sets of specificity-determining residues (i.e. clusters of amino acids whose structural, dynamic and physico-chemical properties directly or indirectly affect interaction and transformation) that enable different enzymes to recognize their unique substrates.

Objective

This study aims to develop a surface patch ranking (SPR) to identify co-evolved surface residue clusters, called Specificity-Determining Residue (SDR) clusters, for substrate specificity determination among homologous enzymes.

Method

Surface patch ranking (SPR) for finding SDR clusters

Residue categorization: For RuBisCo CO2/O2 specificity, we further categorize how an SDR residue within SDR clusters changes among cyanobacteria, green plants and marine non-green algae. The aim is to study when the residues mutate during the RuBisCo evolution and how they correspond to the changes of specificity.

Results

1. The SDR clusters include both highly conserved residues and non-conserved yet complementary residues.

2. Some of the identified SDR clusters, primarily the mono-residue ones, represent residues that are directly involved in enzyme-substrate interactions. Others, mostly the multi-residue ones, represent residues vital for domain-domain and regulator-enzyme interactions.

3. Specificity-determining residues within the SDR clusters of RuBisCos are further categorized. Each category associates uniquely with given RuBisCos groups, their evolutionary history and substrate specificity levels.

Conclusions

1. The SPR algorithm has the ability to identify co-evolved surface residue clusters for substrate specificity determinations.

2. The RSP-based RuBisCo analysis enable us to associate CO2/O2 specificity-determining residues within the identified SDR clusters with the RuBisCo evolutionary history and specificity levels, thus, providing additional information for the selection of residues and SDR clusters for a successful site-directed mutagenesis.

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