

Design of Robust Malate Dehydrogenases by Assembly of Motifs of Halophilic and Thermophilic Enzyme Based on Interaction Network

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Abstract

Robust oxidoreductases in non-aqueous system is promising to bridge the gap between research and industrial application. Malate dehydrogenases (MDHs) were select as model enzyme for redesign based on motif assembly study. Computationally inserted the robust motifs which response to salt concentration into the selected scaffold, and resulted novel MDHs. Top-scoring MDH with structure compatibility and dynamic harmony is expressed for experimental verification. Result indicated the MDH03 with enhanced thermostability, extended pH adaptation and ionic liquid tolerance. The activity of MDH03 was 1.78-fold of parent MDH in present of [EMIM]BF₄. Further study of amino acid residues interaction network explain the robustness of MDH03 based on high-density salt bridges. Research also indicated the hydrophobic contacts and pi-pi contacts of interfacial interactions of motifs play key role for activity and stability of MDH03. This work promote an approach to design robust dehydrogenase with high ionic liquids tolerance and the further application in biosensors.

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