Abstract: The heme group has fundamental role in activation of hemo-enzymes such as horseradish peroxidase (HRP). An alternative approach to the preparation of robust biocatalysts consists in the encapsulation of metalloporphyrins into water-soluble hydrophobic pocket that mimic the polypeptide envelope, which protects the catalytic center of natural enzymes. Artificial enzymes could biomimetically constructed from native protein as host for prosthetic active site to simulate the catalytic functions exhibited by natural enzymes.<br />
In this report, to gain more quantitative insights to study of the structure of caseoperoxidase, the calorimetric (ITC) binding of heme-imidazole to camel β-casein-SDS (3.48 µM/0.96 mM). According to two binding set program fitting of the binding curve (heat of each injection as a function of the heme-imidazole/CF-casein-SDS molar ratio), indicates the enthalpy (ΔH) of the first binding set is \(-9.30 \times 10^5\) cal/mol, and the ΔH of the second binding set is \(-4.54 \times 10^5\) cal/mol. Both binding sets were associated with an exothermic net ΔH, indicating that the long-range electrostatic interactions between the charged regions of the heme-imidazole complexes (especially iron (III)) and the ionic regions of the protein/SDS were strong. The heats of each binding set comprised the sum of both electrostatic and hydrophobic interactions. Therefore, the first binding set with higher binding constant (Kb), associated with a more exothermic enthalpy, corresponded to the electrostatic interactions of the heme-imidazole complex more closely to the charged regions of the protein/SDS. Whereas the lower exothermic ΔH, match to the second binding set, was associated with the more hydrophobic interactions involved in accommodating the heme-imidazole complexes with the hydrophobic regions of protein/SDS as an apoprotein. The maximum number of heme_imidazole complexes that could be incorporated into each apo-enzyme (N) in the first and second binding set is about 4.6. Thus, the constructed biomacromolecule with multiple active sites is a multi-enzyme system.<br />
<br />
Keywords: Caseoperoxidase, Biomimetic, Artificial enzyme, Horseradish peroxidase (HRP), Camel β-casein, Sodium Dodecyl Sulfate (SDS), Imidazole, Heme, ITC

Presentation: Poster