Abstract No.16

The Effect of Temperature and pH on Porcine Pancreatic Alpha-Amylase

Mahmood Najafian[1], Zahra Najafian[2], Azadeh Ebrahim-Habibi[2], Amir Arasteh[3]*

1. Dept. of Biology, Jahrom Branch, Islamic Azad university, Jahrom, IR
2. Endocrinology and Metabolism Reseach center, Tehran University of Medical Sciences, Tehran, IR
3. Dept. of Biology, Faculty of Science,Islamic Azad University, Rasht, IR
(E-mail: mn.najafian@yahoo.com)

Alpha-amylase (EC 3.2.1.1) is the most important enzyme in starch digestion. This enzyme is distributed all over various organisms. In this study the effect of temperature and pH were tested on activity toward porcine pancreatic alpha-amylase (PPA).Bernfeld method was used for the enzyme assay. To determine the optimum temperature, amylase activity was measured at different temperatures. For thermal stability, the enzyme solution was kept at different temperatures for 60 min and then immediately cooled and the residual activity was measured. To determine the optimum pH, amylase activity was measured at different pH values. For pH stability, the enzyme solution at a desired pH, was kept at 4ºC for 24 h, and then activity was measured. The optimum temperature of the enzyme was 40.7ºC. The rates of thermal inactivation, the enzyme retained 100, 80 and 30% activity for 120 min when heated to 35,45 and 55ºC respectively. At 65 and 75ºC the enzyme lost activity after heating for 120 and 60min respectively. The optimum pH of the enzyme was 7.6. The enzyme was stable over a wide pH range. The enzyme was not stable below pH 4.5 or above pH 10.0. The present study show that more than 50% of activity is between 15 and 55ºC. The enzyme also exhibited high activity in basic or near neutral conditions.

Keywords: Alpha-amylase, Temperature, Stability, Activity.

Abstract No.17

Structure-Function Relationship of B-Lactoglobulin in the Presence of Dodecyltrimethyl Ammonium Bromide

Asghar Taheri-Kafrani[1], Abdol-Khalegh Bordbar[1], Thomas Haertle[2], Elaheh Asgari-Mobarakeh[1]*

1. Department of Biotechnology, Faculty of Advanced Science and Technology, University of Isfahan, IR
2. Fonctions et Interactions des Protéines, BIA, Inst.Ntl Res. Agronomique, Nantes, FR
(E-mail: a.taheri@ast.ui.ac.ir)

Bovine β-Lactoglobulin (β-LG) present in milks has been found "in vivo" in complexes with lipids such as butyric and oleic acids. To elucidate the still unknown structure-function relationship in this protein, the structural changes of β-lactoglobulin in the presence of cationic surfactant such as dodecyltrimethyl ammonium bromide (DTAB) have been investigated using various experimental techniques such as UV-Vis spectrophotometry, fluorimetry, isothermal titration calorimetry (ITC) and circular dichroism (CD). Subsequently, the retinol binding by β-LG has been investigated in the presence of various amounts of this surfactant as its extrinsic functional binding fluorophore. Comparison of the results allowed to determine the binding of retinol by β-LG in the presence of DTAB. The results of UV-Vis and fluorescence studies showed a red shift in wavelength and an increase in absorbance and enhancement in the intensity of the quantum yield of protein during its interaction with DTAB. The results of UV-Vis indicated the cooperative character of binding at pH 2.0. The results of fluorescence studies showed that the binding strength of β-LG/DTAB complex increases with the increase of the pH. CD results showed the shifts in positions of the major minima and change in magnitude of ellipticity and subsequently signified two significant changes in structure of β-LG between 10 to 30 and 50 to 100 molar ratio of [DTAB]/[β-LG]. ITC measurements indicated the endothermic nature of β-LG/DTAB interactions at pH 6.7 and the exothermic nature of β-LG/DTAB interactions at pH 8.0. The analysis of the binding data demonstrates the absence of significant changes in retinol binding properties of β-LG in the presence of various amounts of this surfactant. This implies that surfactant binding does not change the conformation of β-LG in the regions defining retinol-binding site nor interferes with retinol binding by a competition for the same binding site(s).

Keywords: β-Lactoglobulin, Isothermal Titration Calorimetry, Circular Dichroism, Interaction, DTAB.

Abstract No.18

The Anticancer Activity and DNA/HSA Binding Properties of Benzo [H] Quinoline (Bhq) Pt (II) Complexes

Roghayeh Mohammadi[1], Reza Yousefi[1], Zahra Pouryasin[2], Sadaf Aghevlian[2], Hamidreza Samouei[3], Mehdi Rashidi[3], Sayed Masoud Nabavizadeh[3], Reza Faghihi[4]*

1. Dept. of Biology, College of Sciences, Shiraz University, Shiraz, IR