water and molecular oxygen. It has one of the highest turnover numbers known ($4\times107\ s^{-1}$). Catalase has numerous industrial and medical applications. It is widely used in the food industry in order to remove the excess hydrogen peroxide from the milk and in the food wrappers to retard oxidation, it is also used in the contact lenses and in the textile manufacturing to ensure that the final product is peroxide free. In industry, it is highly desirable to increase the rate of enzymatic reaction via controlled increase in the temperature. In this report, xylitol as a compatible osmolyte increases the thermal stability of bovine liver catalase through destabilization of the denatured state. The increase in the thermal stability is augmented, in comparison to the native, by an increase in the activity at the room temperature. This increase in enzymatic activity is further enhanced by the intrinsic rate increase caused by temperature which might be of particular interest in industrial applications.

Keywords: Catalase, Xylitol, Compatible osmolyte, Functional stability, Thermal stability.

Abstract No.145

Investigation on Stability and Immobilization of Enzyme-Conjugated Models Based on Different Composition of Chitosan

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The objective of this paper replies to this question that if new chitosan carriers in film and microsphere form could be suitable support to immobilize an enzyme model.The study was conducted in two different stages. In the first stage, suitable carriers were preparated and then its physicochemical effects were investigated. In the second phase, the enzyme immobilization was studied. Briefly, some sample of the carriers in film (FG6) and microsphere (MG6) form were placed in test tubes containing solution of RNAase at pH 7.2. Finally samples were influenced on mixture of RNA and DNA present in agarose gel and rubbed in electrophoresis. To qualitatively evaluate whether RNAs were immoblized the supports, each of four samples were analyzed by native agarose gel electrophoresis stained with RNA. The results acquired from the RNAs immoblization were represented. Qualitative study on the enzyme immobilization in incubated films and microsphers was investigated.The results reflect that visually detectable amounts of RNA are sufficiently stable in the samples a, b, c and d. Similar amounts of stained RNA were observed in the samples of a and b respectively, in 4 and 25 °C. Although in the sample of d has reduced level of RNA. Therefore, the samples c and d depicted changed electrophoretic mobilities, including enzymatic degradation of trapped RNA at the bottom of the sample in 25 °C. It implies that RNAas immobilization was not appropriate as a result of inadequate stabilty. Regarding to obtained results, this investigation has obviously suggested that enzyme immobilization characteristics of chitosan were dependent on the physical form as film or microsphere.

Keywords: Physicochemical Properties, Enzyme Emmoblization, Conjugated Compound.

Abstract No.146

Kinetically Study on Mezalazine Conjugated Biomacromolecules as Bioactive Model and its Application in Enzyme Delivery

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The objective of this paper reply to this question that if an enzyme could release potentially from the prepared biomacromolecules. So prior to evaluate the release pattern of an enzyme from the films or microspheres of a biomacromolecule, it is important to study release kinetics of a bioactive molecules from the carrier (Mezalazine). In brief the following stages were carried out as order, preparation of cross-linked chitosan films and microspheres, conjugation of drug model on films and microspheres, study of in vitro release kinetics of drug-conjugatedcarriers. From these experimental data, it appears that biomacromolecules films and microspheres have shown better controlled release of mezalazine in FG18M5, MG18M5 and especially in MG12M5since their release pattern are near to zero order kinetics. By

cross linking in the network of chitosan in the film and microsphere forms, it seems that the macromolecules probably become more rigid and thus decrease the release of drug. Therefore increase on degree of cross linking affected the release rate. Although fast release of drug from chitosan film, as explained above, was attributed to low degree of cross linking in its network than that of chitosan microsphere (Figure). Figure.In vitro release from chitosan carriers in film and microsphere forms as a function of time at pH 7.4The films and microspheres prepared at 12% w/w degree of cross-linking have shown better features in comparison to those prepared with low and high degree of cross-linking (6 and 18% w/w, respectively). The films and microspheres prepared with all degree of cross-linking (6, 12 and 18% w/w) showed burst-release of mezalazine within a first step of drug release. Then mezalazine was released in a controlled manner in second stage of drug release during 9 h.

Keywords: Release Kinetics, Enzyme Delivery, Bioactive Model.

Abstract No.147

Evaluating of Tyrosinase Mechanism by it's Inhibition and Activation

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The mechanism of mushroom tyrosinase (MT) is complicated because of occurrence of two different enzyme forms (Em and Eo) simultaneously. In this study the inhibition kinetics of a new synthesized ligand (derivative of aniline) in two different forms of the enzyme (Em & Eo) were investigated separately by using Double reciprocal Lineweaver-Burk plots. In both forms the ligand inhibits the enzyme competitively but the results revealed that the oxidation of diphenol by Eo is about 1.3 times faster than oxidation by Em and the tendency of the ligand to bind Em is about 1.57 times more than binding to Eo. These results describe why the low concentration of ligand (2.5 × 10-5 M) activates creolase activity and decreases lag phase and also activates and inhibits the catecholase activity at below and at saturated concentration of substrate, respectively. Also circular dichroism (CD) and fluorescence experiments confirm the binding of ligand to the enzyme. Docking results show that ligand makes 3 hydrogen bonds, 2 with one histidine residue in the active site and one with proline residue.

Keywords: Mushroom Tyrosinase, Inhibition, Activation, Mechanism.

Abstract No.148

Site-directed Mutagenesis of Mnemiopsin: Implication of one of Conserved Residues in Bioluminescence

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Bioluminescence helps for communication, attraction, repulsion, or camouflage in marine life. Photoproteins are the primary reactants of the light-emitting reactions of various bioluminescent organisms and mostly found in marine organisms including coelenterates and ctenophores. Although many photoproteins in nature were discovered but have been very few studied in detail, except for coelenterate photoproteins. In this group, biochemical aspects of photoproteins aequorin and obelin have been well investigated and their crystal structures have been solved. With the help of protein engineering, they have found variety of analytical and biological applications. In contrast, little efforts have been made on cetenophore photoproteins such as mnemiopsin. Recently, cDNA encoding mnemiopsin photoprotein of mnemiopsis leidyiwas cloned in our lab. In this study, site directed mutagenesis using the quick change method was used to make variants of mnemiopsin displaying different functional and structural properties. Since our goal was determination of the role of one conserved residue in coelenterazine binding cavity in mnemiopsin bioluminescence, mutations were introduced at this position (position 39). This residue is among highly conserved residues in coelenterazine binding cavity in each type of photoproteins. For investigation of side chain size and charge effect on luminescence activity, we replaced Arg 39 by Lys and neutral and negatively charged residues, Met and Glu, respectively. The results indicate that except for R39K mutant, that increases luminescence intensity more than eight fold, compared to wild type, R39M and R39E mutants lost luminescence activity completely. The most notable properties of mutant R39K was its higher activity, slow rate of luminescence decay and broad pH activity profile compared to wild type. These properties make it highly attractive for both basic studies and applications. The results of structural studies