Abstract No.205

Spectrofluorimetric Study of the Interaction of Daunomycin Antibiotic with Histone H1

*Seyed Jalal Zargar**

Department of Cell and Molecular Biology, School of Biology, College of Science, University of Tehran, Tehran, IR
(E-mail: zargar@khayam.ut.ac.ir)

Histone H1 is a very lysine rich histone fraction of chromatin which appears to play an important role in the compaction and stability of the structure of chromatin . Daunomycin is an antitumor anthracycline antibiotic wildly used as a chemotherapentic agent for the treatment of various cancers.In the present study, we have investigated the interaction of daunomycin with the purified calf thymus histone H1 in solution using fluorescence spectroscopy technique in 20 mM phosphate buffer , pH=7.0 and 1 mM EDTA at room temperature. The results show that daunomycin decreases the fluorescence emission of histone H1 at 325 nm and induces hypochromicity at the emission spectrum of this protein. Also the fluorescence emission of daunomycin is increased at 595 nm and hyperchromicity is induced at the emission spectrum of this drug after incubating with histone H1.Increasing ionic strength elevates this effect. The results suggest that H1 can be considered as a target for daunomycin action at the chromatin level.

Keywords: Chromatin, Histone H1, Daunomycin, Fluorescence Spectroscopy.

Abstract No.206

Role of Hydrogen Bond Network Around the p-hydroxybenzylringof Coelenterazine in Mnemiopsinbioluminescence Activity by Site-directed Mutagenesis

Zohreh Jahani[1], Maryam Molakarimi [1], Reza H. Sajedi[2], Saman Hosseinkhani[2], Majid Taghdir [2]*

1. Dept. of Biology, Faculty of Sciences, University of Guilan, Rasht, IR
2. Dept. of Biochemistry, Faculty of Biological Sciences, Tarbiat Modares University, Tehran, IR
(E-mail: Maryam_molakarimi@yahoo.com)

Photoproteins are Ca2+-regulated proteins that emit light. So far, the most investigations of photoproteins have been done on coelenterate photoproteins such as aequorin and obelin and there is no information about the mechanism of cetenophorephotoroteins and architecture of its coelentrazine binding site. In native aequorin and obelinphotoproteins, exists a water molecule (W1) in the binding pocketthat makes hydrogen bonds with the phenolic OH of the 2-ρ-hydroxybenzyl, the Oγ-atom of Thr166, and the carbonyl oxygen of Ile105. The main role of W1 is considered to be the stabilization of coelenterazine moiety. In the mnemiopsin, W1 that exists in the binding pocket of native aequorin and obelin is missing and the hydroxyl group of Ser128 establishes a hydrogen bond with 2-p-hydroxybenzyl one hand and the other to with Ile124 and thus possibly creating a network of hydrogen bondsin this position.In the well-known photoproteins, Ile105 and Thr166 are essential in the formation this network and their corresponding residues are Ile124 and Val183 in mnemiopsin. Therefore, V183T and S128G substitutions based on hydrogen bond network around the ring of coelentrazine were performed. On the contrary, the V183T and S128G mutants showed 75% and 19% activity respectively when compared to wild type variant.V183T mutant was active in higher concentrations of calcium and its maximum wavelength of bioluminescence spectrum had about 20 nm blue shift relative to the wild type (470 nm vs. 490 nm). Other characteristics of the mutant did not show any changes. CD and fluorescence spectroscopyandmolecular modelingstudiesshowed some structural changes in the mutants. Based on homology modeling studies, thishydrogen bond network has been removedin S128G mutant, while in V183T mutant, an additional hydrogen bond was observed. The results indicated that the hydrogen bond network around the ρ-hydroxybenzyl ring in position C2 of coelenterazineis important in the bioluminescence activityof photoproteins.

Keywords: Site-directed mutagenesis, Photoprotein, Mnemiopsin, Coelenterazine.

Abstract No.207

Stabilization of Glucose Oxidase Upon Immobilization on Albumin Amyloid Nano-fibers

Sara Farahi [1], Mehran Habibi-Rezaei [1], Ali Akbar Moosavi-Movahedi [2]*

1. School of Biology, College of Science, University of Tehran, Tehran, IR
2. Institutes of Biochemistry and Biophysics, University of Tehran, IR
(E-mail: mhabibi@khayam.ut.ac.ir)

Glucose oxidase (GOD, EC 1.1.3.4) is an oxidoreductase with numerous biomedical and industrial applications. It catalyzes oxidation of glucose to gluconic acid. Molecular oxygen accepts electrons to

produce hydrogen peroxide upon the recation by GOD. Hydrogen peroxide is produces gradually and does its antimicrobial activity in effective and gentle manner. GOD also is routinely used to evaluate glucose in physiological fluid included with medical diagnosing kits. Measurement of glucose concentration is important for determination of blood glucose levels in diabetic patients. GOD has been successfully immobilized onto various scaffolds to improve its stability. In this study we used covalent binding strategy for immobilization of GOD on glycation induced biocompatible amyloid nanofibers of bovine serum albumin in which glutaraldehyde was used as a cross-linker. The optimum concentration of the enzyme for immobilization was determined at 160 µg GOD per mg amyloid nano-fibers. . The kinetic parameters of the free and immobilized GOD were also determined. Despite on a decreasing of the catalytic performance (kcat/Km) of the enzyme upon immobilization, the covalently bound GOD on amyloid nano-fibers retained almost 40-70% of its activity after incubation at temperatures 25, 45 and 65ºC but a major decrease in the activity was occurred for free enzyme under the same conditions. Hence, presented activity in a broad range of temperature most importantly at 40°C< in immobilized form compared to the free enzyme are explained. Interestingly, comparison between pH profiles of the free and immobilized enzymes indicates on increasing pH sensitivity of the GOD activity in the pH range lower than 6 and the optimum pH shift from 5 to 6 upon immobilization was observed.

Keywords: Catalytic Nano-Fibers, Amyloid, Glucose oxidase, Glycation, Immobilization.

Abstract No.208

Salt-induced Bundle Formation of F-actin Using a Detailed Model

*Sarah Mohammadinejad** [1], *Ramin Golestanian* [2], *Hossein Fazli* [1]

1. Department of Physics, Institute for Advanced Studies in Basic Sciences (IASBS), Zanjan, IR
2. Rudolf Peierls Centre for Theoretical Physics, University of Oxford, Oxford, OX1 3NP, UK, GB
(E-mail: sarah@iasbs.ac.ir)

Actin filaments assemble into networks or bundles helped by linker proteins, depending on their different roles in cell function. Similar patterns have also been observed experimentally in solutions of F-actin rods with multivalent salt. In bundle structure, F-actins place close to each other in parallel formation. So despite their same high negative charge, why do they attract each other?Here we aim to represent a model which contains enough structural details of F-actins to study the mechanism of this attraction. For this purpose, we apply the 4-sphere model for the structure of G-actin. In this model, G-actin is composed of four subdomains, one of which carries the most charge of G-actin (sd1). Using this 4-sphere model, we consider real structure and size of F-actin in addition to bending and twist rigidity and helical charge distribution of F-actins.Applying MD simulation to a group of these F-actins, we observe that they attract each and form a hexagonal lattice of the same lattice size as the experiment results. Also it is observed at equilibrium that counter-ions tend to assemble midline between two neighbor F-actins. This distribution of counter-ions around F-actins and how sd1s arrange on F-actins will shed light on understanding the mechanism.Accurate look at equilibrium details of arrangement of sd1 of two neighbor F-actins and plotting the force on them along F-actins, it is concluded that at equilibrium, F-actins will rotate and twist until they expose their sd1s close to each other. Because of their high negative charge, they will attract a cloud of counter-ions in a region between themselves. This cationic cloud will locally attract the two F-actins in its side and it is effectively seen that the two F-actins attract each other.

Keywords: F-actin, Bundle, Counter-ion, Multivalent salt, 4-sphere model, MD simulation.

Abstract No.209

Amyloid Peptide Nanofibrils: from Structure to their Lipid Peroxidative Effects

*Maryam Ferdousi** [1], *Mehran Habibi-Rezaei* [1], *Saeed Balalaei* [2], *Ali Akbar Moosavi-Movahedi* [3]

1. School of Biology, University College of Sciences, University of Tehran, Tehran, IR
2. Department of Chemistry - K.N.Toosi University of Technology, IR
3. Institute of Biochemistry & Biophysics (IBB), University of Tehran, IR
(E-mail: maryam.ferdousi@gmail.com)

Alzheimer's disease is a neurodegenerative disorder which is the most common cause of senile dementia. Amyloid beta (Aβ) is an amphiphilic peptide of 39 to 43 amino acids which is produced from a transmembrane precursor by a proteolytic cleavage, furthermore it is the main component of the neuritic plaques of the Alzheimer's disease.In this research some of the structural, physicochemical, and cytotoxic properties of the Aβ (25-35) and one of its modified, cys-amyloid beta (25-35), which has an additional cysteine residue on its N-terminal, have been investigated. The objective of this research is to