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Low amperage direct electric current inactivates HSV-1 & AdV-5

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The purpose of this research was to investigate the effect of constant direct electric current (DC) on the viral infectivity (Herpes Simplex Virus type 1 & Adeno Virus type 5) without using antiviral agents or other chemical disinfectants in vitro. It was found that direct current (200 μA in 10 minutes) fully inhibited viral infectivity. Probably due to electrolysis and electro-osmotic processes affecting the genome organization and capsid architecture, the viral particles lost their ability to infect Vero cells. The applied current density in the above experiments was 20 μA/mm2 delivered from platinum electrodes. The effect of DC on the viral infectivity was assessed by determination of the 50% tissue culture infective dose (TCID50). In conclusion, such a selective tool can be used to establish a proved technique for decontaminating biological fluids (like blood) from viruses.

Keywords: Herpes Simplex Virus Type 1 (HSV-1), Adeno virus Type 5 (AdV-5), Vero cells, viral inactivation, direct electric current

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P-10-844-1

Molecular dynamics simulation on the chemical denaturation of a polypeptide

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Denaturation of poly-glutamate at alkaline pH and different temperatures in the absence and presence of different concentrations of guanidine chloride was studied by molecular dynamics (MD) implemented in Gromacs 3.2.1. This was done by inserting one molecule of polypeptide and different number of sodium, chloride and guanidine ions in the box having 4.5×4.5×4.5 nm3 dimension at the wide range of time (up to 450 ns). Physico-chemical parameters, such as, radial distribution function, radius of gyration, circular dichroism at 222 nm (CD222), solvent accessible surface area (SASA) was obtained. Analyzing the related trajectories showed that the polypeptide was stabilized at low concentration and was unfolded at high concentration of denaturant. In addition, guanidine ions affect the peptide denaturation both directly and indirectly. Sodium ion decreases the denaturation while chloride and guanidine ions increase it because sodium ion decreases the number of water near the polypeptide and other ions enhance them. This is due to difference in hydration of ions, so that chloride and guanidine ions are related to weakly hydrated ions while sodium relates to strongly hydrated ions.

Keywords: secondary structure, transition state, salting in, chemical denaturation, molecular dynamics

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P-10-452-1

A thermodynamic study of CN- ion interaction with Jack bean urease

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The binding thermodynamic of cyanide ion with Jack bean urease at 300 and 310K at pH=7 was investigated by Isothermal Titration Calorimetry (ITC). The extended solvation model was used to reproduce binding parameters of CN^-+JBU interaction over the whole range of CN^- concentrations. The binding parameters recovered from the solvation model were attributed to the cyanide ion interaction. It was found that cyanide ion acted as a noncooperative inhibitor of urease, and there is a set of 12 identical and independent binding sites for CN^- ions. The dissociation equilibrium constants are 749.99 and 892.10μM at 300 and 310K, respectively. The molar enthalpies of binding are -13.60kJmol-1 and -13.20kJmol-1 at 300 and 310K, respectively.

Keywords: Jack bean urease, cyanide ion, isothermal titration calorimtry, binding parameters

O-11-884-1

Irreversible thermal denaturation of human carbonic anhydrase

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Carbonic Anhydrase (CA; carbonate hydro-lyase, EC 4.2.1.1)), a zinc containing enzyme, catalyzes reversible hydration of carbon dioxide. This metalloenzyme is found in animals, photosynthesizing organisms, and in some nonphotosynthetic bacteria. Thermal denaturation of the protein was studied by enzymatic activity, circular dichroism, and differential scanning calorimetry. The results showed that CA thermal denaturation is an irreversible process. Fitting DSC data and consideration of the validity of two-state model realized to conclude CA undergoes irreversible thermal denaturation in one step. Using this model activation energy, T* parameter (the absolute temperature at which the rate constant of denaturation is equal to 1 min−1), and total enthalpy of CA denaturation were calculated.

Keywords: carbonic anhydrase, DSC, thermal denaturation, irreversibility

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Effects of temperatures on interaction of human serum albumin with new designed platinum complex

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Human serum albumin (HSA) is the most abundant serum protein in the body of all vertebrates, forming about 60% of the mass of plasma proteins, with a typical concentration of 5 g/100 mL in the bloodstream. HSA has a high affinity to an extraordinarily diverse range of materials, such as drugs, metabolites, fatty acids and metal ions. In present study, we have investigated the effect of temperature (27, 37 42 and 47˚C) on the structure of HSA in the absence and presence of Pt (II) complex using fluorescence and UV-Visible spectroscopy. A strong fluorescence quenching interaction of Pt(II) complex with HSA was observed at different temperatures. The binding parameters were evaluated by fluorescence quenching method. The thermodynamic parameters, including $\Delta H°$, $\Delta S°$ and $\Delta G°$ were calculated by fluorescence quenching method. Thermal denaturation study of HSA in the absence and presence of Pt (II) complex represented that thermal stability of protein significantly decreased in the presence of Pt(II) complex. Since the conformational changes in the carrier protein of HSA due to drug (the designed ligand) binding may have deleterious effect, therefore provide useful information to design better metal anticancer complexes with lower side effects in the future.

Keywords: HSA, platinum complex, quenching, thermodynamic parameters

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P-11-397-1

Investigation of the interaction between silver nanoparticles and Doxorubicin

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Breast cancer, which affects an important percentage of human beings, occurs when abnormal cells grow out of control in one or both breasts. They can invade nearby tissues and form a mass, called a malignant tumor. Anthracyclines, particularly Doxorubicin are widely used antibiotics for medical treatments of breast cancer. Their bioactivity is due essentially to the ability to bind DNA and to the redox activity of the anthraquinone group. In this research, we have studied the interaction between silver nanoparticl and Doxorubicin using UV-Visible spectroscopy and new solvation method at 37˚C. Our new solvation model was used to reproduce the thermodynamic parameters of the interaction between anticancer drug of doxorubicin and silver nanoparticle. From this model we have determined the binding constant and enthalpies of this interaction. The interaction of doxorubicin with varying silver nanoparticle concentration represented a set of two binding sites.

Keywords: breast cancer, Doxorubicin, silver nanoparticles, thermodynamic parameters

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O-11-368-1

Electromagnetic fields of 940 MHz induced conformational change of β-Thalassemia hemoglobin

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This study is to investigate the conformational change of β-thalassemia hemoglobin upon irradiation by mobile phone radiofrequency. Hemoglobin is important carrier protein in human body. Patients with β-thalassemia symptoms, suffer from anaemia resulting from the defective synthesis of the β–globin chains of hemoglobin. The effect of EMFs on the structure of β-thalassemia hemoglobin has never been studied before. In this research the effect of microwave fields in 940 MHz frequency band (which belongs to mobile phone radiofrequency) on structure of β-Thalassemia were investigated. Hemoglobin was purified from fresh human blood. Structural changes were studied with UV/vis, circular dichroism and fluorescence spectroscopy. Secondary and tertiary structures of hemoglobin were changed upon the effect of EMFs of mobile phone. α-helix content of the protein ,Tm (melting point) and aggregation property of hemoglobin were altered upon above effect. The results of this study showed that exposure of β-thalassemia hemoglobin to 940 MHz mobile phone radiofrequency caused conformational change and this effect depends on the field

intensity and time of exposure. The structural change of hemoglobin was irreversible.

Keywords: electromagnetic fields, mobile phone, β-thalassemia hemoglobin, circular dichroism, fluorescence

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P-11-217-2

Loop/coil conformations responsible for higher thermostability of alcohol dehydrogenases

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Protein stabilization is a major challenge, constituting different strategies for achieving stabilized proteins to use in bioreactor systems. Commercially available biocatalysts in bioreactor systems for reduction of ketones and aldehydes include alcohol dehydrogenases (ADHs). We have studied thermal, conformational and kinetic stabilities of the three ADHs from different sources yeast (YADH), horse liver (HLADH) and thermophilic Thermoanaerobacter brockii (TBADH) by the use of experimental methods as well as computational bioinformatics analysis. Results indicated higher structural stability and kinetic activity for the both TBADH and HLADH than YADH. In the case that YADH was completely inactivated the residual activities of HLADH and TBADH were 70% and 100%, respectively. Results indicates enzymatic activation energy (Ea) of HLADH and TBADH was higher than YADH because of higher structural rigidity. Higher coil (turn and loop) percentage and score value of the similarity in TBADH and HLADH than in YADH related to higher thermostability of the mentioned enzymes. Multiple alignments by ClustalW showed the conservation of higher number of proline (Pro) residues (two folds) between HLADH/TBADH. All together, structural analyses have proposed loop/coil conformations and especially hot loops by Pro residues, responsible for extreme and higher thermostability of TBADH/HLADHs, respectively.

Keywords: multimeric proteins, ADHs, thermal inactivation, thermostability, proline

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O-11-946-1

Agregation and disagregation of insulin by different concentration of sodium dodecyl sulphate: A spectroscopic study

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More than 20 different diseases are caused at least partially by abnormal protein aggregation, which may result from mutations and physical or chemical changes of cellular environment. Its prevention or even moderate inhibition has been mostly experimental. Therefore, achieving a better understanding of effective parameters on chaperone ability of protein is critical in finding a solution to those devastating diseases. Insulin aggregation (88 uM) performed chemically in the presence of 20 mM dithiotritol (DTT) in 10 mM sodium phosphate buffer, pH 7.0 in a presence of different concentration of SDS. The aggregation of insulin was monitored at 360 nm in a Cary 100 UV-vis spectrophotometer equipped with Le Peltier temperature controller. The ANS fluorescence emission was scanned in a Cary-Eclipse spectrophotometer maintained at a constant temperature and excitation wavelength of 387 nm. The results of UV-visible and fluorescence spectrophotometeric measurements indicated that the SDS in final concentrations of up to 40 μM reduced insulin aggregation, due to the binding of the hydrophobic tail of SDS to the exposed hydrophobic sites of insulin and then strongly preventing aggregation of insulin. But SDS in final concentrations of lower than 5 μM enhanced insulin aggregation. At this concentration hydrophobic interaction between the hydrophobic tail of SDS and insulin is negligible but due to hydrophobic salt effect of nanomolar concentration of SDS enhanced exposure hydrophobic site of insulin and enhanced aggregation of insulin.

Keywords: chaperone-like properties, insulin, sodium dodecyl sulfate, hydrophobic tail, negative head groups, SDS, aggregation, enhancer, reducer, hydrophobic salt effect

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P-11-452-2

A thermodynamic study of Nickle ion interaction with bovine carbonic anhydrase II molecule

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A Thermodynamic study on the interaction of bovine carbonic anhydrase II (CAII) with nickle ions was studied by using isothermal titration calorimetry (ITC) at 27°C in Tris buffer solution at pH=7.5. The enthalpies of Ni2++CAII interaction are reported and analysed in terms of the new solvation theory. It was indicated that there are three identical and non-cooperative sites for Ni2+. The binding of a nickle ion is exothermic with dissociation equilibrium constants of 81.306 and 99.126 μM at 27°C and 37°C respectively. The binding of nickle ions can cause some changes in the stability of the enzyme at low and high Ni2+ concentrations.

Keywords: bovine carbonic anhydrase, nickle ion, isothermal titration calorimetry

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Estimation of binding parameter of paracetamol with normal and glycated Human Serum Albumin

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This study was designed to examine the interaction of Paracetamol (PC) with human serum albumin (HSA) in normal and glycated form under similar physiological condition. Human serum albumin is the most abundant protein which can bind with most components for deliver to target organs. In diabetic persons it has been glycated which changes the ability of protein. Electrochemical method and UV-VIS spectroscopy were used to obtain the binding parameters in aqueous solution. Molecular docking study prepared more detail about the location, forces and residues that are incorporate in the interactions. At pH=7.4 in acetate buffer on the surface of glassy carbon electrode,

paracetamol showed one oxidation and reduction peak. After the addition of HSAs into the paracetamol solution a supramolecular complex was formed in the mixed solution. The result showed that binding constant for HSA-PC is bigger than GHSA-PC. This means in diabetic persons the free drug concentration (in blood not in conjugation with protein) is higher than the healthy people. The binding constant and binding ratio for HSA and GHSA was 1.0×104 and 0.8×104 and the number of binding is 2:1 for HSA-PC and 1:1 for GHSA-PC. The results were confirmed by UV-VIS spectroscopy. Molecular docking study suggested that the primary binding site of paracetamol is located in subdomain IIIA. It is considered that paracetamol binds to this site IIIA mainly by a hydrophobic and hydrogen interaction between the drug and the residues in proteins.

Keywords: paracetamol, continuous cyclic voltammetry, glycated human serum albumin, binding parameters, docking

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O-11-985-1

QSAR studies on the anti-Alzheimer drugs

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Genetic, pathological and biochemical evidence implicate aggregation of beta-amyloid (Ab) peptide in the pathogenesis of Alzheimer's disease. Current marketed therapies for Alzheimer's disease are palliative and only modestly effective. A disease modifying drug would be a major therapeutic breakthrough towards the treatment of this disease. On the other hand quantitative structure-activity relationship (QSAR) method is widely applied to prediction of biological activity. In this studies over 60 samples taken from literature and after structural optimization by Heperchem software, up to 1500 descriptors were calculated by Dragon software. Then, variable selection and multiple linear regression were performed by SPSS package. Results showed that correlation between predicted and experimental IC50 is reasonable and also hydrophobicity and steric factor have significant effect on protein aggregation.

Keywords: amyloid aggregation, QSAR, hydrophobicity, principal component analysis

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P-10-1009-2

Interaction of an asymmetric cationic porphyrin with ionic and nonionic surfactants (SDS and TX-100)

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Porphyrins play an important role in many biological processes, such as oxygen transport, photosynthesis, catalysis. Furthermore, the natural and synthetic porphyrins have been exploited for several clinical applications and act as therapeutic drugs and targeting agent in photodynamic therapy and tumor localization. The mechanism of biological effect of drug porphyrins may involve their penetration through membranes and binding to cellular organelles. Binding of porphyrins to the simplest model for biomimic membranes (micelle, vesicle, etc.) has been of great interest due to possibility of understanding these biological processes. In this work, preparation and characterization of a cationic water-soluble porphyrin,5-(hexadecyl pyridinum-4-yl)-10,15,20-tris(methylpyridinum-4-yl)porphyrin (MHxTM), and its Cu(II) complex have been investigated. Also spectral properties for these porphyrins (absorption spectra, fluorescence spectra and resonance light-scattering (RLS)) were studied in SDS and TX-100 micelle solution. The results revealed that MHxTM forms aggregates, including ordered H-aggregate structure, induced by association with surfactant monomers below the SDS critical micelle concentration (CMC), and forms micellized monomer upon the CMC. Also, the results of emission experiments showed that more than 80% of MHxTM is located in amphiphilic region of TX-100 micellar solution.

Keywords: porphyrin, water-soluble, aggregate, micelle

P-11-123-3

Aggregation of denatured β-Lactoglobulin at different pH and the effect of α-crystallin to prevent it

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β-Lactoglobulin is a major whey protein of milk, which at physiological pH values exists as a dimer. Aggregation of β-lactoglobulin occurs mainly via intermolecular disulphide bond exchange. Each monomer has a molecular weight of 18.3 kDa, contains 162 amino acid residues, two disulphide bonds (C106-C119 and C66-C160) and one free cysteine residue (C121). Upon heating, β-lactoglobulin dissociates into monomers, leading to exposure of hydrophobic groups and the free sulfhydryl group.The aggregation of the heated protein at different pH was investigated by visible absorption spectroscopy and electrophoresis. Upon heating, β-lactoglobulin aggregated which increased with increasing pH. The presence of DTT led to more rapid aggregation and precipitation of β-lactoglobulin. α-Crystallin prevented the aggregation of heat-stressed β-lactoglobulin and was a more efficient chaperone at higher pH values. In the presence of DTT, however, α-crystallin was a less efficient chaperone due to faster aggregation of heated and reduced β-lactoglobulin.

Keywords: β-lactoglobulin, aggregation, DTT
