detection by using SOD/Cys/Au, Cu^{2+}/Cys/Au and Cys/Au electrodes. The Cu/Cys/Au electrode shows quasi reversible peaks with formal potential of 29 mV versus Ag/AgCl at scan rate 50 mVs^{-1} as same as SOD/Cys/Au electrode. The ampromrtic response for was monitored at an electrode potential 250 mV at pH 7.4 phosphate buffer and 500 rpm. In addition, the linear detection range and detection limit of superoxide anion radical at Cu^{2+}/Cys/Au electrode were 3.4-254.2 and 2.3 µM respectively. Comparison between voltammograms of different electrodes revealed that current intensity was increasing by the order of Cu^{2+}/Cys/Au > SOD/Cys/Au > Cys/Au electrodes. This increasing order was also seen for the amprometric response. The experimental results revealed that Cu^{2+}, either as coordinated with Cys or as SOD redox center, plays a critical role in electrochemical response on the Cys/Au electrode. It seems that in Cu^{2+}/Cys/Au electrode, Cu^{2+} coordinate with amine and carboxyl groups of Cys and form a complex. Thus, Cu^{2+}/Cys/Au electrode shows better superoxide dismutase activity than SOD/Cys/Au electrode, since Cu^{2+} in the metal active-site of SOD is structurally located deep in a channel and direct electron transfer between enzyme and the electrode is difficult.

Keywords: Superoxide Dismutase, Superoxide Detection, Cysteine, Electrochemistry.

Abstract No.256

Evaluation of the Smac Based Peptides and Protein in Antagonizing XIAP as Anticancer Agents

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XIAP prevents apoptosis through inhibition of caspase-9 by the BIR-3 and caspases -3 and -7 through BIR2 domain. SMAC which is released from the mitochondria competes with caspases in binding to XIAP unleashing caspase activity and causing cell death. SMAC peptides and protein were used to investigate their ability in relieving the executioner caspase activities inhibited by both the BIR1-2 domains and the BIR1-2-3 domains of XIAP. Furthermore, the potency of these peptides was compared to the Smac protein in antagonizing XIAP. AKPD, ANPR, SGVD, AVPI peptides and the SMAC protein were preincubated with the IAP domains and the activity of caspases was studied in the presence of these mixtures. Moreover, the ability of these peptides in preventing the interaction of BIR1-2 domain with the large and the small subunits of executioner caspases was studied. It was observed that the Smac protein by far is the most potent agent in reversing caspase inhibition. In addition, Caspase-3 inhibition by XIAP domains was more sensitive to SMAC peptides than that of caspase-7. Finally, while, BIR1-2 inhibited caspase-3 was very sensitive to SMAC interference, BIR1-2 inhibited caspase-7 responded to SMAC antagonism very weakly. These results indicate that under conditions of extensive XIAP cleavage and involvement of caspase-7 as the driving force for execution of apoptosis, Smac, and by extension Smac based anticancer agents, cannot be effective in inducing cell death.

Keywords: Apoptosis, XIAP, Executioner Caspases, Smac Peptides and Protein.

Abstract No.257

Comparison of Wild Type and Double Mutated Aequorin Variants from Luminescence and Kinetic Aspects

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The photoprotein aequorin is a small calcium-dependent bioluminescent protein which emits blue light by an intramolecular reaction. The emission properties, stability and decay kinetics of this reporter protein can be changed by directed mutagenesis of key residues. In the present work, three double mutants including variants of Y82F/W86F, Y82F/D153G, and W86F/D153G are prepared. With respect to our results, it seems that presence of W86F mutation shifts the emission to shorter wavelengths, while the Y82F mutation results in shift of emission to longer wavelengths. Furthermore, analysis of the variants for light half-life showed decreased t1/2 for the two mutants of Y82F/D153G and W86F/D153G. Conversely, the Y82F/W86F variant displayed a 2-fold increase of light half-life compared to wild type aequorin. Finally, comparative thermostability analyses of double mutants showed higher stability only for Y82F/D153G variant while the single W86F mutant reached the highest stability against thermal treatment. Our results suggest that replacement of few residues in the active site or binding pocket of aequorin affects its luminescence and kinetic properties and promises the feasibility of new reporter production with limited substitutions.

Keywords: Aequorin, Site Directed Mutagenesis, Luminescence Properties, Double Mutants.

Abstract No.258

Protective Effects of 6 Genotypes of Walnuts Against free Radical-Mediated Protein Oxidation

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The role of oxidative protein damages in the pathophysiology of human diseases is currently a topic of considerable interest as oxidized proteins has been implicated in a wide spectrum of clinical disorders. In this study, the antioxidant activity of 6 genotypes of walnuts, were investigated employing various established in vitro systems including ferric reducing ability (FRAP), 1,1-diphenyl-2-picrylhydrazyl (DPPH), and inhibitory effect on protein oxidation as well as the inhibition of Fe2+/ascorbate induced lipid peroxidation in human plama samples. Total phenolic content (TPC) and total flavonoid content (TFC) of the samples were also determined by a colorimetric method. The addition of Fe2+/ascorbate to the plasma samples significantly increased the of protein oxidation by loss of protein-bound sulphydryl (P-SH) groups and increased lipid peroxidation (LPO) The plant extracts showed inhibitory effects against P-SH oxidation, and LPO to varying degrees. Based on this study, the protective effects of walnuts extract could be due to its TPC. In that respect, free radical induced protein oxidation was suppressed significantly by the addition of walnut over a range of concentration. These results clearly demonstrated that in the shells of walnut have higher antioxidant activities than the hulls of walnut. KH501, KH403, KH509 genotype with the highest phenolic content in its shells has more antioxidant activity against protein oxidation.

Keywords: Protein Oxidation, Juglans Regia, Antioxidant Capacity, Lipid Peroxidation.

Abstract No.259

The Fibrillation Study of β-Lactoglobulin Upon Incubation with Aflatoxin M1

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Aflatoxin M1(AFM1) appears in milk as a direct result of the ingestion of food contaminated with aflatoxin B1 by cattle. The role of milk in human nutrition is well-known. The formation of AFM1 occurs in liver and it is secreted into the milk, which is cytotoxic and genotoxic. AFM1 is bound to milk proteins. As a result of the binding affinity of AFM1 for milk proteins, the toxin is distributed unevenly between whey and curd. The purpose of this report is to study the effect of AFM1 on β-lactoglobulin (β-Lg) fibrillation. Regards to this proposal that AFM1 enters to whey proteins (especially, β-Lg), supposed it would interact with this protein and affects on β-Lg fibrillation. β-Lg solution with concentrations of 1(W/V%) at pH 2 was prepared and interacted with different concentration of AFM1. After heating of the solutions at 85 °C for 24 h, the fibrillation of them were investigated. Strange results showed that AFM1 reduces the intensity of fluorescence of fibrils. By increasing the concentration of AFM1, the intensity of fluorescence of fibrils was decreased. This means AFM1 as a toxin reduces the fibrillation of β-lactoglobulin.

Keywords: Aflatoxin M1, β-Lactoglobulin, Fibrillation, Milk proteins.

Abstract No.260

Fibrillar Protein Aggregation May be Detrimental Via Different Oxidative Routes: Relevance to the Etiology of Amyloid-Related Neurodegenerative Disorders Using the Experimental-Based Evidences

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The exact mechanism of cell death in neurodegenerative diseases remains obscure, but the aberrant assembly of proteins into fibrillar