Abstract No.102

Spectroscopic Monitoring of Salt Impurity as an IEF Interfering Issue in Tear Sample Preparation

Neda Saraygord-Afshari, Zaiddodine Pashandi, Hossein Naderi-Manesh, Mostafa Naderi*

Department of Biophysics, Faculty of Biology, Tarbiat Modares University, Tehran, IR
(E-mail: afshari_ne@yahoo.com)

Human tear film which is composed of three different layers (the innermost glycocalix-mucin, an intermediate aqueous-mucin and an outermost lipid layer) is a valuable specimen for clinical studies. Changes in tear proteome content can be an indicator of many ocular and systemic diseases even in the early stages. Unfortunately high salt content and structural complexity of tear film offers a physicochemical complexity to the sample which makes it difficult to analysis by two-dimensional gel electrophoresis (2DE) as the most powerful technique in biomarker discovery.To seek an appropriate sample preparation, essential for obtaining reliable results in tear proteome analysis, basal tears from healthy volunteers were collected by Schirmer strips and Protein extraction was performed by incubation of each strip in 100 µl of 100 mM ammonium bicarbonate at room temperature for half an hour. All samples were pooled and homogenized to avoid intra individual variability. For tear 2DE interfering removal eight different precipitation methods (acetone, acetone/methanol, TCA/acetone, ammonium sulfate (50, 70 and 90%) tri-n-butyl phosphate (TBP) and chloroform/methanol) were used. A UV-Vis monitoring system (NanoDrop 2000c) was then applied to investigate the probable salt contamination after different sample treatment and 2DE was performed to see if incomplete salt removal can affect the final proteome map or not.Herein we demonstrated the presence of ammonium bicarbonate in samples treated with acetone, acetone/methanol and TCA/acetone methods. Contamination of ammonium sulfate was also observed in those treated with ammonium sulfate precipitations. No salt contamination was observed in TBP and chloroform procedures. Finally comparison of the resulted 2DE pattern revealed the fact that although too much salt in the sample disturbs isoelectric focusing, in the case of salty tear proteome trace amount of salt can improve the protein solubility during the onset IEF separation and affects their transfer to the IPG strips.

Keywords: Tear Film, UV-Vis Spectroscopy, 2D PAGE, Salt Impurity.

Abstract No.103

Spectroscopic studies of the interaction of bovine beta-lactaglobulin with a new designed Pd(II) complex

Elnaz Karbasi [1], Adeleh Divsalar [2], Ali Akbar Saboury [1], Hassan Mansouri-Torshizi [3]*

1. Institute of Biochemistry & Biophysics, University of Tehran, IR
2. Dept. of Biological Sciences, Tarbiat Moallem University, Tehran, IR
3. Dept. of Chemistry, University of Sistan and Bluchestan, Zahedan, IR
(E-mail: e.karbasi@ibb.ut.ac.ir)

Beta-lactoglobulin (BLG) is one of the major soluble cow's milk proteins and also one of the main allergens in milk. This protein seems to be resistant and stable to gastric digestion and denaturation. The purpose of this work is to establish whether the treatment of BLG by Pd(II) complex would affect its allergnicity, as well as its structure. Then, the new Pdcomplex [Pd (bpy) (Hex.Gly)]NO_3 was designed and the interaction of this new synthesized complex with bovine milk carrier protein of BLG was investigated using fluorescence at room and physiologic temperatures. The results of fluorescence studies indicated that Pd(II) complex can quench the intrinsinc fluorescence emission of the protein and also may approved that it can bind to the protein and alter the protein structure. The binding parameters of this interaction have calculated using quenching methods at different temperatures. Above results represented that the new synthesized Pd(II) complex can bind to the milk carrier protein of BLG and also can change the tertiary structure of the protein.

Keywords: β-Lactoglobulin, Pd Complex, Fluorescence, Quenching.

Abstract No.104

A Computational Analysis of Interactions of Curcuminoids with Human Serum Albumin

Massoumeh Ighaei [1], Jaber Sardroodi [1], Alireza Rastkarebrahimzadeh [2], Faramarz Mehrnejad [3]*

1. Department of Chemistry, Faculty of Science, Azarbaijan University of Tarbiat Moallem, Tabriz, IR
2. Department of Physics, Faculty of Science, Azarbaijan University of Tarbiat Moallem, Tabriz, IR
3. Department of Cellular and Molecular Biology, Faculty of Science, Azarbaijan University of Tarbiat Moallem, Tabriz, IR
(E-mail: eighaie@yahoo.com)