BIOPHYSICAL CHEMISTRY LAB

ANNUAL REPORT 2008

Institute of Biochemistry and Biophysics
Tehran, Iran

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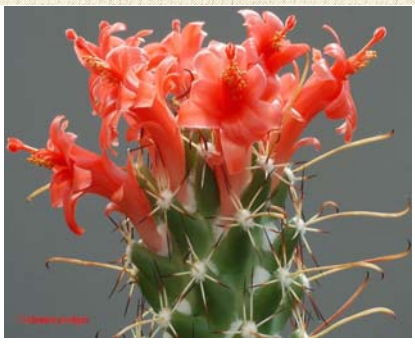
CONTENTS



FOREWORD

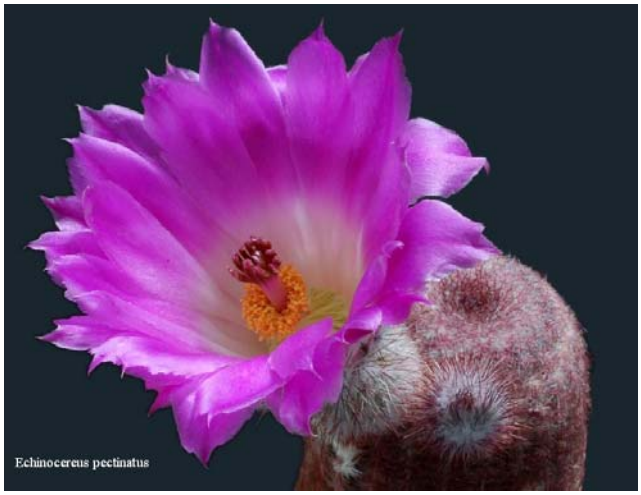
The Laboratory of Biophysical Chemistry (LBC) was established in 1986 in the Institute of Biochemistry and Biophysics (IBB) and now functions as the main base and the mother of Biophysical Chemistry in Iran. This laboratory is well-known in the national and international level. It is also famous worldwide in the research area of Thermodynamics of Protein Denaturation and Protein-Ligand Interaction.

This laboratory enjoys from advanced facilities and is equipped with advanced apparatuses for the research on Biothermodynamics and Biomacromolecular Interactions. LBC is equipped with Nano and Micro Differential Scanning Calorimeters (DSC); Nano and Micro Isothermal Titration Calorimeters (ITC); modern Circular Dichroism (CD) Spectropolarometry, Sensitive Densitometer and Tensiometer; Fluorescence and Uv-vis Spectrophotometers, Microviscometers and Biochemical and Biophysical methods as well as and Computational facilities.

LBC is an appropriate place for the promotion of the research and science in the field of Biochemistry,Nanobiophysics, Biotechnology and Biophysical Chemistry. LBC is a suitable laboratory for training PhD students and postdoctorate researchers, associate researchers and sabbatical leaves for faculty members at national and international levels.

Up to the present time, 35 Ph.D and 53 Master students have developed their theses in this laboratory and graduated from the university. Faculty members, postdoctorates and students and foreign research associates using the facilities of this laboratory which have published hundreds of full research articles in international prestigious journals. LBC is an appropriate laboratory for supporting and promoting the research of scientists and researchers at national and international levels.

Biophysical Chemistry Lab



SUPERVISORS

Ali A. Moosavi-Movahedi
Professor of Biophysics
Protein Thermodynamics

Ali A. Saboury
Professor of Biophysics
Protein- Ligand Binding

Lab Colleagues

Professor M. Shamsipur Razi University Department of Chemistry	**Professor A. Shafiei** University of Tehran Faculty of Pharmacy, Medical Sciences
Professor. P. Norouzi University of Tehran College of Science, Department of Chemistry	**Professor M. R. Ganjali** University of Tehran College of Science, Department of Chemistry
Professor. B. Moshiri University of Tehran Dept. of Electrical and computational enginering	**Dr. K. Nazari** Research Institute of Petroleum Industry, Tehran, Iran
Dr. S. Safarian University of Tehran College of Science, Department of Biology	**Dr. M. Habibi-Rezaei** University of Tehran College of Science, Department of Biology
Dr. G. Ataei Medical Science Shahid Beheshti University Department of Premedical Science	**Dr. M. Amanlou** Medical Science University of Tehran Department of Pharmacy
Dr. A. Niasari University of Tehran Department of veterinary	**Dr. H. Ghourchian** University of Tehran Institute of Biochemistry and Biophysics (I.B.B)

Dr. M. Amani
Medical University of Ardebil

INTERNATIONAL COLLABORATION

★Professor G. Floris:

Department of Applied Science in Biosystem, University of Cagliari, Cagliari, Italy

★Professor T. Haertle

National Research Institute of Agronomique, 44316 Nantes Cedex 03, France

★ Professor F. Ahmad

Director, Centre for Interdiscipinary Research in Basic Sciences, Jamia Millia Islamia (A Central University), Jamia Nagar, NEW DELHI - 110 025, India

★Professor G. Hakimelahi

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★Dr. N. Sheibani

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Miss. M. Amini

A. Divsalar

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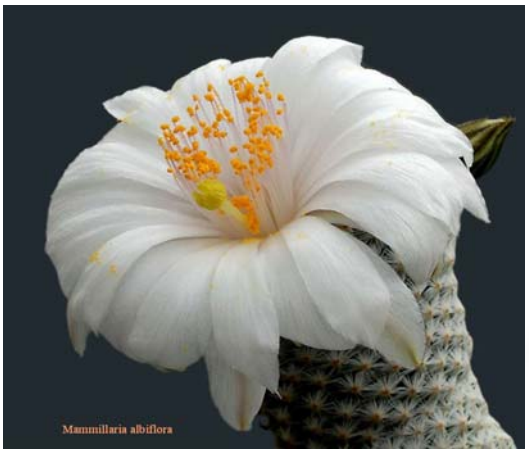
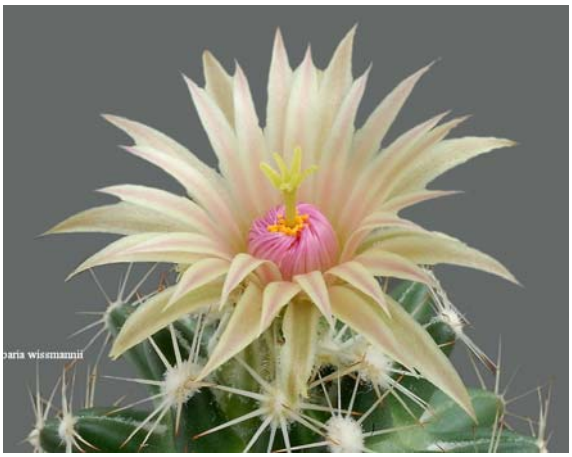
POSTDOCTS



R. Yousefi



STUDENTS (Year 2008)

Doctor of Philosophy (Ph.D)

- S. J. Mousavy
- M. Atri
- J. Badraghi
- S. Bagheri
- M. Salami
- S. Zolghadr Jahromi
- M. Alijanianzadeh
- H. Sepasi-Tehrani
- H. Hadi-Alijanvand
- A. Sharifizadeh
- A. Barzegar
- K. Mahnam

Master of Science (MSc)

- A. Hekmat
- E. Sharifi
- E. Amin
- Y. Sefidbakht
- M. J. Bagheri-Arabi
- Y. Sefidbakht
- F. Farivar

Alumni (Year 2008)

Name	*Topic of Thesis*

Ph.D:

K. Mahnam	Thermodynamics and kinetics studies and computer calculation of adenosine deaminase : Modification by Woodward reagent K and determination of the effective factors in protein modification
A. Barzegar	Study of the physicochemical, electrochemical properties and nanostructural peptide of alcohol dehydrogenase (ADH)

MS.C

A. Hekmat	Role of pH and temperature in the structure and function of coline oxidase
Y. Sefidbakht	Artificial nanozyme design based on HRP as a model in aqoues media
E. Sharifi	Effect of trehalose and cyclodextrin on structural change of nanofibril formation during glycation of human serum albumin
F. Farivar	Kinetic study of histidine-heme in reverse nanomicelles of sodium dodecyl sulfate in organic media

Completed Projects (2008)

1- Structural elucidation on amine oxidase in the presence and absence of different substrates and denaturants. Joint Project with Professor G. Floris, Italy

2- Interaction of adenosine deaminase with inhibitors and substrate analogs. International joint project, Institute of Biochemistry and Biophysics, University of Tehran;Institute of Biochemistry, National Academy of Science, Yeravan, Armenia; Department of Chemical Sciences, University of Camerino, Camerino, Italy

3- The stablization and activation of enzyme via metal ions: industrial, medical and bioenvironmental application. Interchange joint project between Institute of Biochemistery and Biophysics, University of Tehran and Department of Chemistry, Razi University.

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Rebutia pallida

PUBLICATIONS 2008

1- G. Rezaei Behbehani, A. A. Saboury and E. Taleshi, "A direct calorimetric determination of denaturation enthalpy for lysozyme in sodium dodecyl sulfate", *Colloids and Surfaces* B: Biointerfaces 61 (2008), 224-228.

2- A. Barzegar, A. A. Moosavi-Movahedi, S. Rezaei-Zarchi, A. A. Saboury, M. R. Ganjali, P. Norouzi, G. H. Hakimelahi and Fu-Yuan Tsai, "The mechanisms underlying the effect of α-cyclodextrin on the aggregation and stability of alcohol dehydrogenase ", *Biotechnology and Applied Biochemistry*, 49 (2008), 203-211.

3- A. Bayandori-Moghaddam, M. R. Ganjali, R. Dinarvand, T. Razavi, A. A. Saboury, A. A. Moosavi-Movahedi and P. Norouzi, "Direct electrochemistry of cytochrome c on electrodeposited nickel oxide nanoparticles", *Journal of Electroanalytical Chemistry* 614 (2008), 83-92.

4- A. Bayandori-Moghaddam, M. R. Ganjali, R. Dinarvand, S. Ahadi and A. A. Saboury, "Myoglobin immobilization on electrodeposited nanometer-scale nickel oxide particles and direct voltammetry", *Biophysical Chemistry* 134 (2008), 25-33.

5- M. Hassanisadi, A. Barzegar, R. Yousefi, M. Dalgalarrondo, J.-M. Chobert, T. Haertle, A. A. Saboury, A. A. Moosavi-Movahedi, "Chemometric study of the aggregation of alcohol dehydrogenase and its suppression by β-caseins: a mechanistic perspective", *Analytica Chimica Acta* 613 (2008), 40-47.

6- A. Barzegar, R. Yousefi, A. Sharifzadeh, M. Dalgalarrondo, J.-M. Chobert, M. R. Ganjali, P. Norouzi, M. R. Ehsani, A. Niasari-Naslaji, A. A. Saboury, T. Haertle and A.A. Moosavi-Movahedi, "Chaperone activities of bovine and camel β-caseins: Importance of their surface hydrophobicity in protection against alcohol dehydrogenase aggregation", *International Journal of Biological Macromolecules* 42 (2008), 392-399.

7- G. Rezaei Behbehani, A. A. Saboury and E. Taleshi, "A comparative study on direct calorimetric determination of denaturation enthalpy for lysozyme in sodium dodecyl sulfate and dodecyltrimethylammonium bromide", *Journal of Solution Chemistry* 37 (2008), 619-629.

8- G. Rezaei-Behbehani, A. A. Saboury and A. F. Bagheri, "A thermodynamic study on the binding of cobalt ion with myelin basic protein", *Bulletin of the Korean Chemical Society*, 29 (2008), 736-740.

9- G. Rezaei Behbehani, A. A. Saboury and E. Taleshi, "Determination of partial unfolding enthalpy for lysozyme upon interaction with dodecyltrimethylammonium bromide using an extended salvation model", *Journal of Molecular Recognition* 21 (2008) 132-135.

10- F. Valiyev, F-Y. Tsai, A. A. Saboury, H.-J. Liu, A. A. Moosavi-Movahedi, and G. H. Hakimelahi, "Design, synthesis, and antiviral activity of novel phosphoramidates", *Journal of the Iranian Chemical Society* 5 (2008), 228-237.

11- A.A. Moosavi-Movahedi, F. Semsarha, H. Heli, K. Nazari, H. Ghourchian, J. Hong, G. H. Hakimelahi, A.A. Saboury and Y. Sefidbakht, "Micellar histidinate hematin complex as an artificial peroxidase enzyme model: Voltammetric and spectroscopic investigations", *Colloids and Surfaces A: Physicochemical and Engineering Aspects* 320 (2008), 213-221.

12- S. Rezaei-Zarchi, A. A. Saboury, A. Barzegar, S. Ahmadian, A. Bayandori-Moghaddam and A. Javed, " Nano-composition of riboflavin-nafion functional film and its application in biosensing", *Journal of Biosciences* 33 (2008), 279-287.

13- M. Amani, R. Yousefi, A. A. Moosavi-Movahedi, F. Pintus, A. Mura, G. Floris, B. I. Kurganov and A. A. Saboury "Structural changes and aggregation process of Cu/Containing amine oxidase in the presence of 2,2,2´-trifluoroethanol", *Protein and Peptide Letters* 15 (2008), 521-527.

14- G. Rezaei-Behbehani, A. A. Saboury, A. F. Bagheri and A. Abedini, "Application of an extended solvation theory to study on the binding of magnesium ion with myelin basic protein", *Journal of Thermal Analysis and Calorimetry* 93 (2008), 479-483.

15- D. Ajloo, E. Taghizadeh, A. A. Saboury, E. Bazyari and K. Mahnam, "Effects of surfactant, salt and solvent on the structure and activity of adenosine deaminase: molecular dynamic and spectrophotometric studies", *International Journal of Biological Macromolecules* 43 (2008), 151-158.

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17- K. Mahnam, A. A. Moosavi-Movahedi, H. Bahrami, G. H. Hakimelahi, G. Ataie, S. Jalili, A. A Saboury, F. Ahmad, S. Safarian and M. Amanlou "Efficient factors in protein modification: Adenosine deaminase esterification by Woodward reagent K", *Journal of the Iranian Chemical Society* 5 (2008), 464-465.

18- A. Bayandori-Moghaddam, M. R. Ganjali, A. A. Saboury, A. A. Moosavi-Movahedi and P. Norouzi, Electrodeposition of nickel oxide nanoparticles on glassy carbon surfaces: application to the direct electron transfer of tyrosinase", *Journal of Applied Electrochemistry* 38 (2008), 1233-1239.

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22- S. Rezaei-Zarchi, A. A. Saboury and A. Javed, "Electrochemical study of horseradish peroxidase using the nanosilver-modified graphite electrode and its application to hydrogen peroxide biosensor", *Journal of New Materials for Electrochemical Systems* 11 (2008), 199-203.

23- A. Asadi, A. A. Saboury A.A. Moosavi-Movahedi, A. Divsalar and M. N. Sarbolouki, "Interaction of bovine serum albumin with some novel PEG-containing diblock copolymers", *International Journal of Biological Macromolecules* 43 (2008), 262-270.

24- M. Salami, R. Yousefi, M. R. Ehsani, M. Dalgalarrondo, J.-M. Chobert, T. Haertlé, S. H. Razavi, A.A. Saboury, A. Niasari-Naslaji, A. A. Moosavi-Movahedi, "Kinetic characterization of camel and bovine milk proteins hydrolysis using pancreatic enzymes", *International Dairy Journal* 18 (2008), 1097-1102.

25- H. Ramshini, N. Rezaei-Ghaleh, A. Ebrahim-Habibi, A. A. Saboury, and M. Nemat-Gorgani, "Thermally induced changes in the structure and activity of yeast hexokinase B", *Biophysical Chemistry*, 137 (2008), 88-94.

26- A. A. Saboury and M. Alijanianzadeh, "Ethyl xanthate and propyl xanthate as activators and inhibitors of mushroom tyrosinase in different concentrations", *Journal of the Chinese Chemical Society* 55 (2008), 937-942.

27- H. Mansouri-Torshizi, M. Islami-Moghaddam, A. Divsalar, A. A. Saboury, "2,2'-Bipyridinebutyldithiocarbamatoplatinum(II) and palladium(II) complexes: synthesis, characterization, cytotoxicity and rich DNA-binding studies", *Bioorganic & Medicinal Chemistry* 16 (2008), 9616-9625.

28- A. Mohammadi, A. Bayandori-Moghaddam1, Rassoul Dinarvand, J. Badraghi, F. Atyabi, A. A. Saboury, "Bioelectrocatalysis of methyldopa by adsorbed tyrosinase on the surface of modified glassy carbon with carbon nanotubes", International", *Journal of Electrochemical Science* 3 (2008), 1248 – 1257.

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30- G. Rezaei-Behbehani, A. A. Saboury and N. Gheibi, "A new approach for Thermodynamic Study on binding some metal ions with human growth hormone", *Journal of Solution Chemistry*, 37 (2008), 1645-1655.

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32- G. Rezaei-Behbehani, A. A. Saboury and A. Divsalar, " Thermodynamic study of the binding of calcium and magnesium ions with myelin basic protein using the extended solvation theory" *Acta Biochimica et Biophysica Sinica* 40 (2008), 964-969.

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34- K. Mahnam, H. Bahrami, A. A. Moosavi-Movahedi, A. A. Saboury, M. Iranmanesh, G. H. Hakimelahi M. N. Soltani Rad and A. Khalafi-Nezhad, "A theoretical Iinvestigation of mechanism of the adenosine deaminase modification: Reaction of glutamate residue with Woodward reagent K", *Journal of Theoretical and Computational Chemistry* 7 (2008), 1121-1145.

35- N. Rezaei-Ghaleh, H. Ramshini, A. Ebrahim-Habibi, A. A. Moosavi-Movahedi and M. Nemat-Gorgani "Thermal aggregation of α-chymotrypsin: Role of hydrophobic and electrostatic interactions"Biophysical Chemistry132(1),23-32 (2008)

36- H.Heli, M. Amani, A.A.Moosavi-Movahedi, A. Jabbari, G. Floris and A. Mura "Electroactive centers in euphorbia latex and lentil seedling amine oxidases" Bioscience, Biotechnology and Biochemistry 72(1), 29-36 (2008)

37- M. Hajjizadeh, A. Jabbari, H. Heli, A.A. Moosavi-Movahedi, A. Shafiee and K. Karimian " Electrocatalytic oxidation and determination of deferasirox and deferiprone on a nickel oxyhydroxide-modified electrode" Analytical Biochemistry 373(2),337-348 (2008)

38- H. Yadegari, A. Jabbari, H. Heli, A.A. Moosavi-Movahedi, K. Karimian and A. Khodadadi"Electrocatalytic oxidation of deferiprone and its determination on a carbon nanotube modified glassy carbon electrode" Electrochimica Acta 53,2907-2916 (2008)

39- M. Houshmand, A. Jabbari, H. Heli, M. Hajjizadeh, A. A. Moosavi-Movahedi "Electrocatalytic oxidation of aspirin and acetaminophen on a cobalt hydroxide nanoparticles modified glassy carbon electrode" J. Solid State Electrochemistry 12, 1117-1128 (2008)

40- M. Mojtahedi, H. Parasta, M. Jalali-Heravi , J. Chamani, F.C. Chilaka and A.A. Moosavi-Movahedi "Comparison between two different hemichromes of hemoglobins (HbA and HbS) induced by n-dodecyl trimethylammonium bromide: Chemometric study" Colloids and Surfaces B: Biointerfaces 63(2), 183-191 (2008)

41- H. Yadegari , A. Jabbari , H. Heli , A. A. Moosavi-Movahedi , K. Karimian "Electro-oxidation and determination of deferiprone on a glassy carbon electrode" Chemia Analityczna 53(1), 5-16 (2008)

42- H. Yadegari , A. Jabbari , H. Heli , A. A. Moosavi-Movahedi and S. Majidi "Electrochemistry of deferiprone as an orally active iron chelator and HIV-1 replication inhibitor" J Brazilian Chem. Soc. 19(5), 1017-1022 (2008)

43- R. Singh, T.A. Dar, S. Ahmad, A.A. Moosavi-Movahedi and F. Ahmad "A new method for determining the constant-pressure heat capacity change associated with the protein denaturation induced by guanidinium chloride (or urea)" Biophysical Chemistry 133, 81-89 (2008)

44- M.R. Ganjali, P. Norouzi, R. Dinarvand, R. Farrokhi and A.A. Moosavi-Movahedi " Development of fast Fourier transformations with continuous cyclic voltammetry at an Au microelectrode and its application for the sub nano-molar monitoring of methyl morphine trace amounts"Materials Science and Engineering 28(8), 1311-1318 (2008).

45- M.R. Khazaei, M. Habibi-Rezaei, F. Karimzadeh, A.A. Moosavi-Movahedi, A. Sarrafnejad, F. Sabouni and M. Bakhti "Microglial cell death induced by glycated bovine serum albumin: nitric oxide involvement" J. Biochemistry (Tokyo) 144, 197-206 (2008)

46- M. Hajjizadeh , A. Jabbari, H. Heli , A. A. Moosavi-Movahedi "Electro-oxidation and determination of mefenamic acid and indomethacin on a copper electrode" Chemia Analityczna 53,429-444 (2008)

47- A. Barzegar, A. A. Moosavi-Movahedi, K. Mahnam, H. Bahrami, N. Sheibani "Molecular Dynamic Simulations of Nanomechanic Chaperone Peptide and Effects of in silico His Mutations on Nanostructured Function" Journal of Peptide Science 14, 1173-1182 (2008).

48- A. Mahmoudi, K. Nazari, M. khosraneh, V. Kelay and A.A. Moosavi-Movahedi "Can amino acids protect horseradish peroxidase against its suicide-peroxide substrate?" Enzyme and Microbial Technology 43, 329-335 (2008)

49- H. Rahaman, K. A. Khan, I. Hassan, M. Wahid, S. B. Singh, T. P. Singh, A. A. Moosavi-Movahedi, F. Ahmad "Sequence and stability of the goat cytochrome c" Biophysical Chemistry 138, 23-28 (2008)

50- B. Mohajerani, M. Soleymani-Jamarani, K. Nazari, A. Mahmoudi, A.A. Moosavi-Movahedi "Microperoxidase-11-NH2-FSM16 Biocatalyst: AHeterogeneous Enzyme Model for Peroxidative Reactions" Journal of Molecular Catalysis A: Chemical 296, 28-35 (2008)

51- N. K. Poddar , Z.A. Ansari , R.K. B. Singh , A.A. Moosavi-Movahedi , Faizan Ahmad "Effect of monomeric and oligomeric sugar osmolytes on ΔGD, the Gibbs energy of stabilization of the protein at different pH values: Is the sum effect of monosaccharide individually additive in a mixture?" Biophysical Chemistry 138, 120-129 (2008)

52- M. S. Atri, A. A. Saboury, R. Yousefi, M. Dalgalarrondo, J.-M. Chobert, T. Haertle and A. A. Moosavi-Movahedi, "Comparative studies on heat stability and structure of bovine and camel α-lactalbumin", *Protein and Peptide Letters*, submitted (2008).

ABSTRACTS/

International

1- A.A. Saboury and S. Amiri , "Thermodynamics of nickel ion binding to human growth hormone ", *52th Annual Meeting of Biophysical Society*, Long Beach, California, USA (2-6 Feb. 2008). *Biophysical Journal* 94 (2008), 1073-Pos/B49.

2- A. Barzegar, A. A. Moosavi-Movahedi, K. Mahnam, H. Bagheri, A.A. Saboury, D. Ntentopoulou and M. R. Ganjali, "Experimental and computational docking study of alcohol dehydrogenase stabilization by α-CyD", *52th Annual Meeting of Biophysical Society*, Long Beach, California, USA (2-6 Feb. 2008).
Biophysical Journal 94 (2008), 1031-Pos/B7.

3- M. S. Atri, A.A. Saboury, R. Yousefi and A. A. Moosavi-Movahedi, "A comparative study on the structure and thermal stability of camel and bovine alpha-lactalbumin", *52th Annual Meeting of Biophysical Society*, Long Beach, California, USA (2-6 Feb. 2008).
Biophysical Journal 94 (2008), 2732-Pos/B35.

4- A. Divsalar and A.A. Saboury, "Changing of function and structure of milk carrier Protein β-lactoglobulin-A upon interaction with new anti-cancer compounds Pd(II) Complexes", *52th Annual Meeting of Biophysical Society*, Long Beach, California, USA (2-6 Feb. 2008).
Biophysical Journal 94 (2008), 2754-Pos/B57.

5- M. J. Bagheri, A. Divsalar, A. A. Saboury, and H. Mansouri-Torshizi, "Effects of new anti-cancer compound (2,2'-bipyridine butylcinato Pd (II) nitrate) on the structure of blood carrier protein (human serum albumin)", *33rd FEBS Congress & 11th IUBMB Conference :* Biochemistry of Cell Regulation, Athens, Greece (28 June-3 July 2008). *The FEBS Journal* 275 (2008), 158.

6- A. Hekmat A. A. Saboury, A. Divsalar and H. Ghourchian, "Effects of pH on the structure of choline oxidase from Alcaligenes species", *33rd FEBS Congress & 11th IUBMB Conference :* Biochemistry of Cell Regulation, Athens, Greece (28 June-3 July 2008).
The FEBS Journal 275 (2008), 166.

7- H. Ramshini, N. Rezaei-Ghaleh, A. Ebrahim-Habibi, M. Sabbaghian, A. Ghasemi, A. A. Saboury and M. Nemat-Gorgani, "Formation of small soluble aggregates may be responsible for irreversible thermal denaturation of yeast hexokinase B", *33rd*

FEBS Congress & 11th IUBMB Conference : Biochemistry of Cell Regulation, Athens, Greece (28 June-3 July 2008).
The FEBS Journal 275 (2008), 195.

8- S. Zolghadri, A. A. Saboury, A. Divsalar, S. Rezaei, and A. Golestani, "Probing the interaction of silver nanoparticle and hemoglobin at different temperature", *33rd FEBS Congress & 11th IUBMB Conference :* Biochemistry of Cell Regulation, Athens, Greece (28 June-3 July 2008).
The FEBS Journal 275 (2008), 234.

9- A. Asadi, A. Divsalar, A. A. Saboury and M. N. Sarbolouki, "Energetics of serum albumin interaction with PEG containing diblock copolymerse", *33rd FEBS Congress & 11th IUBMB Conference :* Biochemistry of Cell Regulation, Athens, Greece (28 June-3 July 2008).
The FEBS Journal 275 (2008), 390.

10- A. Divsalar and A. A. Saboury, "Effects of new Pd(II) complexes on the allergenicity and proteolysis of bovine □-lactoglobulin", *33rd FEBS Congress & 11th IUBMB Conference :* Biochemistry of Cell Regulation, Athens, Greece (28 June-3 July 2008).
The FEBS Journal 275 (2008), 446.

11- S. Rezaei-Zarch, A. A. Saboury and A. Javed, "Bromide-modified silver electrode for electrochemical recognition of metalloproteins", *First Regional Symposium on Bioelectrochemistry*, University of Tehran, Tehran, Iran (13-15 October 2008).

12- A. Javed, S. Rezaei-Zarch and A. A. Saboury, "Electrochemical behaviour of redox proteins immobolized on nano-composition of riboflavin-nafion functional film modified gold electrode", *First Regional Symposium on Bioelectrochemistry*, University of Tehran, Tehran, Iran (13-15 October 2008).

13- A. Hekmat, A. A. Saboury and H. Ghourchian, "The effects of hydrogen ions on the activity and structure of choline oxidase", *First Regional Symposium on Bioelectrochemistry*, University of Tehran, Tehran, Iran (13-15 October 2008).

14- S. Rezaei-Zarch, A. A. Saboury and A. Javed, "Horseradish peroxidase immobilization on electrodeposited nanometer-scale silver particles and direct voltammetry", *First Regional Symposium on Bioelectrochemistry*, University of Tehran, Tehran, Iran (13-15 October 2008).

15- A. Hekmat, A. A. Saboury, H.Ghourchian "Biotechnological application of choline oxidase, a glycine betaine synthesis enzyme, to inhibite human pathogens at hyperosmotic infection site" *13th International Biotechnology Symposium*, (12-17 October 2008), Dalian, China.
Journal of Biotechnology 136S (2008) S543

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21- Y.Sefidbakht, H.A. Rafieepour, F.Farivar, K.Nazari and A.A.Moosavi-Movahedi "MP-11/NH2-FSM16 nanozyme as an artificial peroxidase: kinetic and voltametric studies" First Regional Symposium on Bioelectrochemistry, University of Tehran, Tehran, Iran, Oct 13-15, 2008

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The mechanisms underlying the effect of α-cyclodextrin on the aggregation and stability of alcohol dehydrogenase

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High concentrations of proteins and enzymes have to be stored for extended periods of time. Under such conditions, at least three major factors contribute to aggregation and loss of protein function: hydrophobicity, propensity to form non-native β-sheet structure and net charge of the polypeptide chain. Here we evaluate these thermal aggregation factors for horse liver ADH (alcohol dehydrogenase) and the effect of α-CyD (α-cyclodextrin) on the ADH aggregation, by using fluorescence, CD, UV–visible spectrophotometry, the DLS (dynamic light scattering) technique and the enzymatic activity assay. In addition, we propose the relative importance of the hydrophobic effect on the ADH aggregation. Although ADH readily forms aggregates at higher temperatures, α-CyD effectively diminishes this phenomenon. This reduction can be attributed to the prevention of the appearance of larger-size aggregated molecules and also to the higher homogeneity of the small nuclei under the α-CyD effect. The observed re-aggregation upon the addition of α-CyD/phenylalanine can be attributed to the competition binding of phenylalanine to the internal hydrophobic cavity of α-CyD. This signifies that aromatic amino acids are important regional components of the residual structure that may form nuclei for aggregation. The results of dynode voltage changes indicate that the thermal unfolding of ADH is accompanied by protein aggregation, which subsequently leads to irreversible thermal unfolding. Moreover, α-CyD causes thermal stabilization and delays the onset of secondary structural unfolding and aggregation by approx. 10°C and the midpoint ('melting') temperatures (T_m) by more than 5°C. Furthermore, α-CyD diminishes the deactivation of the enzyme, decreasing the deactivation constant by more than 50%, and clearly reveals the stabilization of the enzyme not only structurally but also kinetically at higher temperatures.

Introduction

Proteins are inherently unstable in aqueous solution and degrade by a variety of routes, the most common route being aggregation. Aggregation is the assembly of non-native protein conformations into multimeric states, often leading to phase separation and precipitation. Oligomeric proteins are more sensitive to high temperatures and tend to dissociate in the form of inactive aggregates. The problem of aggregation is especially grave in the pharmaceutical industry and in biotechnology, where it may be necessary to handle and store therapeutic proteins at high concentrations and temperatures and for long periods of time [1,2]. Thermal processes (e.g. sterilization, pasteurization and blanching) are important in a number of contexts in the food and pharmaceutical industries; however, enzymes tend to form inactive aggregates at high temperatures. Hence thermal aggregation, inactivation and stabilization of enzymes and proteins have received much attention [3,4]. The overproduction of proteins of biotechnological interest in bacterial cells often results in the aggregation of the expressed protein in inclusion bodies. In humans, this process can lead to the occurrence of several disorders, such as Alzheimer's disease, cystic fibrosis and diseases involving prions. This phenomenon has considerable relevance in cell biology, medicine and biotechnology [5–7]. Understanding the molecular mechanism underlying protein aggregation is of prime importance, not only in biotechnology, but also in the health-related industries. The biophysical features underlying protein aggregation are not completely understood.

Various techniques have been developed to prevent the formation of protein aggregates [2,5,6,8]. One of the

Key words: aggregation, alcohol dehydrogenase (ADH), dynode voltage, hydrophobic effect, irreversible thermal unfolding, α-cyclodextrin.

Abbreviations used: ADH, alcohol dehydrogenase; α-CyD, α-cyclodextrin; DLS, dynamic light scattering.

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Direct electrochemistry of cytochrome *c* on electrodeposited nickel oxide nanoparticles

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Abstract

The convergence of biotechnology and nanotechnology has led to the development of hybrid nanomaterials that incorporate the highly selective catalytic and recognition properties of biomaterials. The conjugation of nanoparticles (NPs) and other nanoobjects (e.g. nanorodes, nanowires and nanotubes) with biomolecules is an attractive area of research within nanobiotechnology. Here, this study focused on the design of cytochrome *c*/nickel oxide nanoparticles/glassy carbon electrode, prepared by the electrochemical deposition of the nickel oxide nanoparticles (NiO NPs) on the glassy carbon (GC) electrode surface and the self-immobilization of cytochrome *c* on the surface of the electrodeposited nickel oxide nanoparticles. The existence of different geometrical shapes of the NiO NPs was exhibited using the scanning electron microscopy (SEM) and atomic force microscopy (AFM). These geometrical structures could lead to the better immobilization of proteins on their surfaces. The resulting electrode displayed an excellent behavior for the redox of the cytochrome *c*. Also, the resulting heme protein exhibited a direct electrical contact with the electrode as a result of the structural alignment of the heme protein on the nanometer-scale nickel oxide surfaces. In line with our investigation, non-physiological electrochemical responses were analyzed for this electrode.

Keywords: Cytochrome *c*; Bioelectrochemistry; Atomic force microscopy; Nanoparticle; Nanotechnology; Nickel oxide

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Chemometric study of the aggregation of alcohol dehydrogenase and its suppression by β-caseins: A mechanistic perspective

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ABSTRACT

Molecular chaperones interact preferentially with certain aggregation-prone intermediates of target protein molecules. An estimation of the chaperone activity based on suppression of aggregation is required to be mechanistically understood. In this study, the multivariate curve resolution chemometric technique was applied on horse alcohol dehydrogenase (ADH) UV-spectra under thermal stress, to obtain the required information about the number and change in concentrations of the species involved. Chemometric analysis of UV-absorption spectra of horse ADH under thermal stress, led to the existence of three different molecular species including native (N), aggregation-prone intermediate (I) and final aggregate (A) species. Appearance and buildup of two molecular species I and A were connected to the disappearance of N-species. In the presence of β-caseins (BCN), however, a new complex between I and BCN (I–BCN) was formed. Meanwhile, by accretion of concentration of I–BCN complex, the light scattering intensity diminished. The data presented in this study clearly demonstrate that the interaction of BCN as a chaperone molecule with I-species takes place in a temperature-dependent manner and leads to a reversible I–BCN complex. In the absence of chaperones, I-state is subsequently converted to the final aggregate species. In the presence of BCN, this molecular species could be converted to the final aggregate state and/or form the I–BCN complex.

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Chaperone activities of bovine and camel β-caseins: Importance of their surface hydrophobicity in protection against alcohol dehydrogenase aggregation

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Abstract

β-Casein (β-CN) showing properties of intrinsically unstructured proteins (IUP) displays many similarities with molecular chaperones and shows anti-aggregation activity *in vitro*. Chaperone activities of bovine and camel β-CN were studied using alcohol dehydrogenase (ADH) as a substrate. To obtain an adequate relevant information about the chaperone capacities of studied caseins, three different physical parameters including chaperone constant (k_c, μM^{-1}), thermal aggregation constant (k_T, $^{\circ}C^{-1}$) and aggregation rate constant (k_t, min^{-1}) were measured. Bovine β-CN displays greater chaperone activity than camel β-CN. Fluorescence studies of 8-anilino-1-naphthalenesulfonic acid (ANS) binding demonstrated that bovine β-CN is doted with larger effective hydrophobic surfaces at all studied temperatures than camel β-CN. Greater relative hydrophobicity of bovine β-CN than camel β-CN may be a factor responsible for stronger interactions of bovine β-CN with the aggregation-prone pre denatured molecular species of the substrate ADH, which resulted in greater chaperone activity of bovine β-CN.

Keywords: Intrinsically unstructured proteins (IUP); β-Casein; Aggregation; Alcohol dehydrogenase (ADH); Chaperone; Hydrophobicity

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Microglial Cell Death Induced by Glycated Bovine Serum Albumin: Nitric Oxide Involvement

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Nonenzymatic glycation results in the formation of advanced glycation end products (AGEs) through a nonenzymatic multistep reaction of reducing sugars with proteins. AGEs have been suspected to be involved in the pathogenesis of several chronic clinical neurodegenerative complications including Alzheimer's disease, which is characterized with the activation of microglial cells in neuritic plaques. To find out the consequence of this activation on microglial cells, we treated the cultured microglial cells with different glycation levels of Bovine Serum Albumin (BSA) which were prepared *in vitro*. Extent of glycation of protein has been characterized during 16 weeks of incubation with glucose. Treatment of microglial cells with various levels of glycated albumin induced nitric oxide (NO) production and consequently cell death. We also tried to find out the mode of death in AGE-activated microglial cells. Altogether, our results suggest that AGE treatment causes microglia to undergo NO-mediated apoptotic and necrotic cell death in short term and long term, respectively. NO production is a consequence of iNOS expression in a JNK dependent RAGE signalling after activation of RAGE by AGE–BSA.

Key words: advanced glycation end products, apoptosis, glycation, microglia, nitric oxide.

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Efficient Factors in Protein Modification: Adenosine Deaminase Esterification by Woodward Reagent K

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Chemical modification of Adenosine Deaminase (ADA) with N-ethyl-5-phenyl isoxazoliom-3-sulfonate (Woodward's reagent K) (WR-K) was studied using experimental and theoretical techniques. Reaction concentration ranges were 0.8-6 mM WR-K at pH 7.8 and 27 °C. It was observed that the maximum number of moles of esterified residues per mol of enzyme ($\bar{\nu}$) in this concentration ranges is 4. However, esterification of ADA does not affect the activity of ADA, suggesting that the active site residues are not esterified. Similar results were obtained when the active site was blocked with 0.1 mM erythro-9-(2-hydroxy-3-nonyl) adenine (EHNA), followed by esterification, as measured by enol ester formation using absorbance at 340 nm. A theoretical approach was employed to study the modification process using molecular dynamic simulation, MM and QM/MM minimization. A full ASA empirical model and B3LYP method were used to evaluate the relative stability of some species which may arise from the reaction of ADA with WR-K. Theoretical results have shown that five residues (Glu 244, Glu 121, Glu 337, Asp 127, Asp 338) can be possible cases for modification in reaction 1:1 between ADA and WR-K at $\bar{\nu} = 1$. Glu 121 was possible initially modified in this process. Besides, it is specified that atomic accessible surface area cannot be an appropriate criterion in determination of primary sites which are modified by WR-K. Ultimately, it is clarified that among effective factors in modification of enzyme surface such as atomic accessible surface, stability of modified segment and local residues changes of ADA, latter factor plays a basic role in this process from kinetics and thermodynamics point of view.

Keywords: Adenosine deaminase, Woodward's reagent K, QM/MM minimization, Molecular dynamic, Free energy of hydration

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Detergency effects of nanofibrillar amyloid formation on glycation of human serum albumin

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ABSTRACT

The prolonged glycation of human serum albumin (HSA) results in significant changes in its structure. The identity of these structural changes and the influence of carbohydrates on these changes require further study. Here, we evaluated structural changes and amyloid formation of HSA upon incubation with Glc, Fru, or Rib. Fluorescence spectrophotometry, surface tension analysis, and transmission electron microscopy (TEM) were utilized to evaluate the structures of glycated HSA. The physicochemical properties including excess free energy, protein adsorption at the air–water interface, critical aggregation concentration (CAC), and surface activity indicated an increase in hydrophobicity and partial unfolding of HSA structure upon glycation. Thus, it appears that AGE products can act as detergents. Incubation of HSA with these sugars after 20 wks induced significant amyloid nanofibril formation. Together these results indicate that prolonged glycation of HSA is associated with a transition from helical structure to β-sheet (amyloid formation).

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Kinetic characterization of hydrolysis of camel and bovine milk proteins by pancreatic enzymes

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ABSTRACT

As differences in protein composition between camel and bovine milk may influence their digestibility, hydrolysis of milk proteins of both species using two pancreatic serine proteases (trypsin and chymotrypsin) was studied. Caseins (CNs) were more rapidly hydrolyzed than whey proteins (WPs) because of their greater flexibility and open structures. The extent of hydrolysis by chymotrypsin of CNs in both species was significantly higher than that of hydrolysis by trypsin, which is due mainly to the greater number of potential hydrolytic sites present in the primary structures of CNs targeted by chymotrypsin than by trypsin. The extent of hydrolysis of camel WPs by each protease separately or by their mixture was less than that of bovine WPs, which can be explained by the greater stability of camel WPs compared with that of bovine WPs. The kinetic parameters (K_m and k_{cat}) widely varied for different substrates, which results from differences in binding affinities and substrate turnovers, respectively.

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2,2′-Bipyridinebutyldithiocarbamatoplatinum(II) and palladium(II) complexes: Synthesis, characterization, cytotoxicity, and rich DNA-binding studies

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ABSTRACT

Butyldithiocarbamate sodium salt (Bu-dtcNa) and its two complexes, $[M(bpy)(Bu\text{-}dtc)]NO_3$ (M = Pt(II) or Pd(II) and bpy = 2,2′-bipyridine), have been synthesized and characterized on the basis of elemental analysis, molar conductivities, IR, ^1H NMR, and UV–vis spectra. In these complexes, the dithiocarbamato ligand coordinates to Pt(II) or Pd(II) center as bidentate with two sulfur atoms. These complexes show 50% cytotoxic concentration (Cc_{50}) values against chronic myelogenous leukemia cell line, K562, much lower than that of cisplatin. The interaction of these complexes with calf thymus DNA was extensively investigated by a variety of spectroscopic techniques. These studies showed that both complexes presumably intercalate in DNA. UV–vis studies imply that they cooperatively bind with DNA and unexpectedly denature the DNA at very low concentrations (~100 μL). Palladium complex breaks the DNA into two unequal fragments and binds stronger to the lighter fragment than to the heavier one. In the interaction studies between the Pt(II) and Pd(II) complexes with DNA, several binding and thermodynamic parameters have been determined, which may provide deeper insights into the mechanism of action of these types of complexes with nucleic acids.

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Electroactive Centers in *Euphorbia* Latex and Lentil Seedling Amine Oxidases

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The electrochemical behavior of redox centers in the active site of amine oxidases from lentil seedlings and *Euphorbia characias* latex was investigated using a mercury film electrode. Tyrosine-derived 6-hydroxydopa quinone (TPQ) and copper ions in the active site are redox centers of these amine oxidases. The enzymes undergo two reduction processes at negative potentials related to the reduction of the TPQ cofactor to the corresponding hydroquinones and the reduction of copper ions, (Cu(II) → Cu(I)). Copper depleted enzymes, prepared by reduction with dithionite followed by dialysis against cyanide, undergo only one reduction process. Nyquist diagrams, recorded at potentials corresponding to the reduction of cofactors as dc-offset, represent charge transfer impedance followed by a Warburg-type line at low frequencies, indicating the occurrence of a diffusion controlled process in the rate-limiting step of the reduction process.

Key words: amine oxidase; lentil seedlings; *Euphorbia* latex; topa quinone; mercury film electrode