Statement of Teaching Interests

My interest in Academic Career is two-fold. First, it brings me satisfaction to help others learn new subjects. Second, teaching helps me to move up to the next level of understanding. Just reading about a subject is never enough; in most situations, one needs to explain it to others. I found teaching to be exciting to me; the academic track is my preferred career choice. I have developed and presented many lectures and seminars. People often comment that my presentation style is accessible and my enthusiasm is contagious. I am also frequently told that I make a good teacher because I am personable and responsive to group dynamics. Based on my past experience, I am confident that I am successful in these endeavors.

Experience. My confidence is reinforced by my extensive teaching experience, involving 6 years at Chemistry Faculty of M.V.Lomonosov Moscow State University, Russia and 6 years at Hunter College of The City University of New York, NY. During 6 years as an Adjunct Lecturer at Hunter College I taught General, Organic, and Physical Chemistry to predominantly minority undergraduate students. During those years I learned how to gain the respect and attention from the students, who are not necessarily interested in the subject. During 6 years as a Jr. Scientist and Scientist (equivalent to a junior faculty) at Moscow State University my job responsibilities involved teaching. I taught selected lectures in the Crystallography course (~300 students/class), and full course of Point and Space Groups Symmetry for advanced undergraduates. I also prepared and taught a new course in Electronic Models for Molecules and Solids.

From my past experience as a teacher, and especially as a student, I learned that the biggest mistake a science instructor can make is to focus on technical details and mathematical formulae. A good teacher first communicates the idea, and only then illustrates it with the formulas if necessary. Also, motivating students and cultivating their interest may be more important than the knowledge conveyed. Curiosity combined with the problem solving skills will help the students much more than the passive knowledge.

I firmly believe that students need a challenge constantly, not only at midterms and finals. A short 10 min pop-quiz needs to be a part of every class. I also believe that animated computer graphics is an excellent tool to capture students' attention, while it can not replace chalk board

completely. I also think it would be advantageous to present the students with the state of modern science by constantly updating the courses with scientific news. I also think it is important to show science in the making. Students should not be just the passive spectators, but the active participants in this process. Literature research assignments may be given to the teams of students, so they will learn teamwork, independent research, making presentations to the class, and engage in critical discussion.

Qualifications. My education and experience make me well qualified to teach a range of graduate and undergraduate classes on Chemistry and Biochemistry, including General, Physical, and Biophysical Chemistry. In addition, I would like to incorporate molecular modeling class as a part of curriculum. Using powerful graphical front-end programs and multifunctional molecular dynamics packages would help student visualize the new concepts. The students could search crystallographic databases of biomolecules, investigate protein aggregation and unfolding, receptor/ligand binding, and the effect of the enzymatic environment on the reactions at the active sites. If the necessary equipment and software are unavailable, I intend to pursue external (NSF) funding for it.

Student research supervision. At Moscow State I supervised three students in their Diploma (equivalent of B.S./M.S.) Research Thesis. One involved literature search, systematization and interpretation of the crystal structures, and two other one involved FORTRAN code modifications for crystal packing and electron density analysis. Both resulted in full-size scientific publications. At Los Alamos National Lab, I conducted recruitment and supervising of two Summer Graduate Student Interns in the group of Theoretical Chemistry and Molecular Physics. I am proud to say that one of my students earned an award for outstanding oral presentation at Championing Scientific Careers Symposium (August 2003). At Hunter College I have had significant experience helping undergraduates with no prior knowledge of Quantum Chemistry to run *ab initio* calculations with *Gaussian 94*. It is very realistic to involve students in productive research in Computational Biophysical Chemistry and Crystallography. I enjoyed it and I am looking forward to the opportunity to involve students once again in my research.

Statement of Research Interests

Inexpensive empirical force field methods are capable of dynamical description of large systems (up to millions of atoms in size), found in Biological and Materials application. However, my early experiences with simulations of molecular crystal structures convinced me that simplified force fields are often insufficient to reproduce many important properties. In search for a better tool I turned to quantum chemistry, and found that Density Functional Theory (DFT) and approximations to it provide sufficiently accurate description of electronic properties and intermolecular interactions. Unfortunately, DFT treatment is feasible for systems with up to hundreds of atoms. Therefore, combining accuracy of DFT with efficiency of the force field in hybrid methods (QM/MM) seems to me unavoidable. DFT, on one hand, leaves some room for improvement, when applications call for better accuracy. On the other hand, careful parameterization for the force field in order to reproduce the details of DFT results is also appropriate in many cases. These methods are the focus of my research interests. Equipped with these powerful tools, I am ready to take on the challenging applications in Computational Nanoscience and Biophysics. My experience with DFT at Hunter College and LANL, combined with bimolecular MD simulations at CCNY makes me fully qualified to obtain external funding and complete the following projects.

Figure 1. Structure of the insulin amyloid fibril based on cryo-electron microscopy data (Ref.3). Potential applications if these fibrils span medicine, nutrition, catalysis, electronics, and structural materials.

Theoretical study on H-bonded aggregation: toward rational control over amyloid fibril formation. Amyloid fibril (Fig. 1) is a form of protein self-assembly of 8-13 nm in diameter and several μm in length, where β-strands are arranged perpendicular to the axis of the fibril, and β-sheets ran parallel to it. *In vivo* their formation had been associated with spider silk[1] and several "diseases of protein misfolding": Alzheimer's disease, Parkinson's disease, and prion diseases, where protein looses its native α-helical structure and aggregates. For this reason amyloid fibrils have attracted enormous interest in recent years. *In vitro* amyloid fibril formation had been observed for a wide range of

proteins and synthetic peptides with potential applications in medicine, nutrition, catalysis, electronics, and structural materials.[2] Formation and macroscopic assembly of these fibrils is sensitive to the pH, salt concentration, buffer composition of the solution, and other factors. This sensitivity holds a promise for much better control over nanofabrication, than that in carbon nanotube field. Amyloid fibrils are also highly accessible for chemical modification, unlike the inert carbon nanotubes.[3]

Before commercial applications of the amyloid fibrils can be fully realized, predictive relationship must be established between the ability to form fibrils, their structure and properties, on one hand, and protein sequence and environmental conditions, on the other hand. Conformational changes between native α-helices and amyloid β-sheets resemble the transitions between crystal polymorphs. In fact, kinetic model for the fibril formation similar to crystallization has been introduced,[4] although it is far from complete. Here I propose to explore this analogy in experimental study on H-bonding aggregation of small molecules, combined with molecular simulations to obtain parameters for this kinetic model.

In combined crystallographic and light scattering study[5] we found kinetically stable nanosized H-bonded aggregates to form and remain in aqueous solution of only one of 6-methyluracil polymorphs. In the solid, the molecules aggregate by cyclic amide H-bonds, similar to ones found in β-sheets. Thus, 6-methyluracil presents an example of a model system. It affords both unambiguous X-ray and *ab initio* calculations of hydrogen bonding. However, the methodology for high-level simulations of H-bonds in aqueous solution needs to be developed. A combination of explicit and implicit solvation models with *ab initio* calculations already gave promising results in description of thermodynamic properties of neat liquids,[6] and simple aqueous ions.[7] These studies demonstrate that convergence is achieved at a relatively small cluster size (5-6 solvent molecules). The results can be used to derive the potential of mean force and to refine the parameters of a classical model. In this project I will extend this approach to describe the formation of hydrogen bonds in amyloid fibrils, and other self-assembling systems. I will use the obtained values for energy barriers and local minima to build a detailed kinetic model. Such a model will assist in control over nanofabrication of amyloid fibrils and in design of therapeutic agents against the protein misfolding diseases mentioned above.

Absolute binding free energies with accurate fragment potentials: applications to rational drug design. An accurate description of specific binding between molecules in solution (molecular recognition) presents the cornerstone of Computational Biophysics, and serves many practical goals, including the rational drug design. Among possible approaches to binding affinity calculations end-point free energy method is the least computationally demanding. Its rigorous thermodynamic formalism had been described by different authors (see Ref. 8 for a review). However, numerical results for many protein/ligand systems were at best only qualitatively correct, and the most successful implementation (Linear Interaction Energy method)[9] was using system-dependent empirical parameters. The methodological study of avidin/biotin complex and its analogs demonstrated,[14] that inaccuracies in the force field is the major cause for incorrect results, while several components of binding free energy can be replaced with inexpensive yet accurate estimates. Specifically, position of the ligands inside the binding site distorted significantly form crystallographic data after both MD run and optimization. Apparently, the upgrade in the quality of the force field is likely to improve calculated free energies.

Effective fragment potentials (EFP)[10] present the next generation of the force fields. Unlike Amber and Charmm force fields, where only electrostatic or total energy is fit to *ab initio* calculations on small molecules, EFP is designed to reproduce each of the different energy components: exchange, electrostatic, polarization and dispersion. This eliminates the possibility of error cancellation and ensures reliable results for molecular orientations which were not included in the training set. EFP had been parameterized for aminoacid residues in full range of conformations,[11] and was shown to give accurate description for protein electrostatics, as indicated by pKa values[12] and spectroscopic properties of short H-bonds[13] in proteins.

Here I propose to apply EFP to calculations of protein/ligand affinities. On the first phase of this project EFP will be applied to the avidin/biotin complex and analogs. Structural stability of the complexes and the ability of EFP to reproduce water/heptane distribution for all involved ligands will be verified. Different approximations for the major components of binding free energies will be tested: protein/ligand interaction and reorganization energies, librational and conformational enthropy of the ligand. If successful, the methodology will be implemented in program *AutoDock*, which was recently modified to account for protein mobility,[14] and its performance will be compared with the best scoring functions available.

Accurate description of pure spin states: applications to enzymatic catalysis and photosensitive proteins. Nitrile hydratase (NHase) is an iron-containing metalloenzyme used in the commercial conversion of acrylonitrile to acrylamide. The mechanism by which this biochemical reaction occurs is unknown, but studies of the model complexes indicate the dramatic role of the protein environment on the spin state of Fe ion and its catalytic activity. Recent progress in QM/MM techniques made it possible to study the effect of the enzyme on chemical reactions at the active center, for instance cytochrome P450 monooxygenase.[15] However, the accuracy of DFT (used as QM part) is not uniform. Both pure and hybrid (with part of HF exchange) DFT methods err significantly for hydrogen abstraction and S_N2 reactions,[16] and fail to even qualitatively describe dissociation of two-center three-electron bonds.[17] Unrestricted DFT formalism provides an improvement in description of the transition state, but it no longer represents a pure singlet and hence distorts the energy profile close to the reaction barrier. The situation becomes even worse when the active site includes high-spin multicenter metal complexes, where *ad-hoc* decontamination corrections must be used.[18] The reason for DFT failures is that local and semi-local correlation functionals do not reliably describe non-local nature of the static electron correlation. Replacement of the exact exchange with local exchange functional introduces the error of the opposite sign. For near equilibrium geometries the correlation is weak, so that the errors cancel and DFT is successful. For transition states the improvements are necessary. Unrestricted formalism captures part of the static correlation and hence improves the performance.

Here I propose to modify Kohn-Sham formalism of DFT and include static correlation in noninteracting system description with different orbitals for different spins, while keeping pure spin state by proper antisymmetrization (spin balanced unrestricted Kohn-Sham method, SBUKS). This scheme eliminates the need in local exchange, and hence recovers correct asymptotic of the functional, improving description of the charge-transfer states. Unlike other multideterminant DFT modifications, this scheme retains attractive computational efficiency. Similar formalism, applied at semiempirical Hartree-Fock level, was successful in predicting the correct order of the spin states of model compounds for NHase active site.[19] My preliminary results show encouraging improvements for bond-breaking and other strongly-correlated systems.[20] In the first phase of this project SBUKS scheme will be implemented in QM/MM modules of CHARMM, and applied to NHase model compounds. The performance will be

compared to the available experimental data to determine the necessary theory level. In the second phase NHase will be studied with modified QM/MM code to investigate the catalytic mechanism. In the third phase of the project SBUKS scheme will be used as reference state for TD-DFT and applied to the ground and excited states potential energy surfaces of photoactive yellow protein, studied recently at CASSCF QM/MM level.[21] I expect accuracy of SBUKS treatment to pass that of CASSCF at significantly lower computational cost. If successful, this project opens new avenues for computational investigations of important processes in Molecular Biology, including light harvesting, photoreceptors, and signaling pathways.

Biomedical use of two-photon absorption: design of nanomaterials for targeted drug delivery and fluorescent labels. Two-photon (TP) fluorescent microscopy employs nonlinear optical process when molecule simultaneously absorbs two photons and emits one. It has a number of advantages over the standard (linear) microscopy, including high three-dimensional resolution (due to quadratic dependence on intensity), and increased penetration depth in tissue with reduced photodamage (by operating with incident light in the visible red-NIR region). These allow for real-time imaging of the living tissue, if effective TP absorbing chromophores are introduced into it.[22] Semiconductor nanocrystals, known as colloidal quantum dots were reported to have largest TP cross-sections (up to ~50,000 GM) which strongly depend on passivating ligands, attached to its surface.[23] Unfortunately, the details about the quantum dot structure remain unknown.

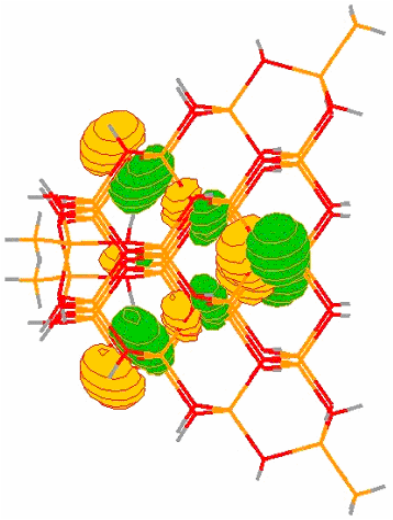

Figure 2. HOMO calculated at DFT (B3PW91/ LANL2mb) level in $Cd_{32}Se_{14}(SeH)_{36}(PH_3)_4$, the cluster modeling colloidal quantum dot of 2 nm in diameter. Electronic structure of quantum dots strongly depends on passivating ligands.

The computer code which I developed at LANL implements time dependent DFT calculation of TP cross-section. It was very successful for large organic chromophores.[24] Here I propose to apply this method to study TP cross-section dependence on the passivating ligands in CdSe quantum dots. The $Cd_{32}Se_{14}(SePh)_{36}(PPh_3)_4$ cluster, which had been unambiguously characterized using X-Ray diffraction[25] (Fig. 2) provides as an excellent starting point to model surface modifications. As a result, more effective fluorescent labels for *in-vivo* two-photon microscopy will be developed.

Photodissociation is another possible consequence of two-photon absorption. In the second part of this project I propose to design a nanocage, which is capable of encapsulating pharmacophore molecule, and photodissociation upon two-photon absorption of IR light. Two-photon absorption induced photochemical drug release was found to have several advantages over regular photodynamic therapy, including deep tissue penetration, tight spatial localization and fewer side effects.[26] Self-assembling nanocage, formed by metal-ligand interactions (Fig.3) was found capable to encapsulate specific molecules.[27] This cage needs to be redesigned to fit the pharmacophore molecule (such as 5-fluorouracil), incorporate two-photon absorbing chromophore (such as stilbene moiety), and photodissociative fragment (such as coumarine photodimer). The design will be assisted by *ab initio* calculations of two-photon absorbtion and photodissociation.

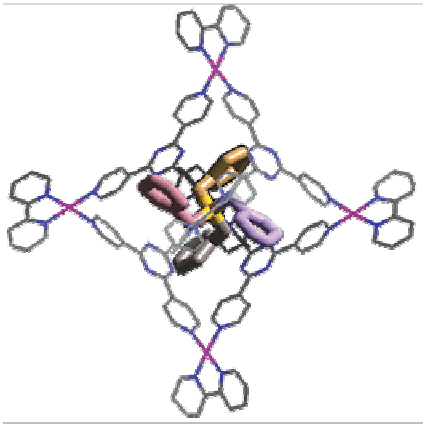

Figure 3. Nanocage with encapsulated guest molecule (ref.30). If redesigned to fit pharmacophore and dissociate in two-photon regime, it may be used for targeted drug delivery

I intend to seek funding for this project from NIH. Bioengineering Consortium (BECON) of NIH identified "Nanosystem design and application: fundamental principles and tools to assemble nanosystems; development of fluorescent probes at the nanometer scale for monitoring biochemical processes on the surface and inside a cell; development of nanoparticles and nanospheres that enable controlled released of therapeutic agents… into targeted cells" as priority area for nanoscience research support (Program ID 04-00157).[28]

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