Marwan Al-Shawi's Personal Statement

Throughout my career I have studied a phenomenon fundamental to all living cells; the coupling of energy utilization to the transport of ions and molecules across biological membranes. Despite an enormous amount of work in this field, our understanding of transport proteins remains at a simplistic model level. For only a very few proteins is there anything approaching a detailed molecular characterization. This lack understanding represents one of the fundamental questions in biology yet to be resolved. Currently, there is a revival of the field, as the importance of these proteins in disease processes and to the practice of medicine is increasingly recognized. One third of all drugs in current use are targeted to membrane located channels and transporters. The multidrug transporter, P-glycoprotein, which I study in molecular detail, has proven to be very important. This protein is an ATP-driven drug-exporting pump that counteracts chemotherapy in cancer cells and limits the bioavailability of therapeutic drugs in other tissues. Modulation of the drug-exporting activity of P-glycoprotein, through the application of fundamental knowledge, could improve cancer treatments, AIDS treatments and epilepsy treatments, environmental toxicology, and allow for a greater number of oral therapeutic drugs to be developed.

I have aimed to solve scientifically challenging problems. This has necessitated my developing some expertise in the diverse fields of biochemistry, biophysics, physical chemistry, molecular biology, cell biology and computer science. As a graduate student, I investigated the stoichiometric relationships of proton translocation coupled to electron transport in oxidative and photo-phosphorylation. My work demonstrated the presence of directly coupled primary proton pumps operating in the chemosmotic mechanism of energy coupling. Next, as a post-doc, I defined the regulation of the $Ca^{2+}$-ATPase by nucleotides, $Mg^{2+}$ and pH. A lasting contribution to bioenergetics was my identification of the catalytic residues of the ATP-synthase by site directed mutagenesis and detailed kinetic analyses. This identification was performed prior to the availability of the crystal structure. Introduction of application of linear free energy analyses to the study of the membrane transport proteins is another of my important contributions. I was the first to show that P-glycoprotein was in fact a primary transport protein that hydrolyzed ATP at a rate commensurate with drug transport. Together with Drs. Alan Senior and Ina Urbatsch, we were the first to purify and reconstitute active P-glycoprotein and the first to establish a reaction and transport cycle model of P-glycoprotein. To do this, I overexpressed P-glycoprotein to a record level of 32% of CHO plasma membrane proteins. Several pharmaceutical firms that screen for drug inhibitors and modulators of P-glycoprotein currently use the CR1R12 cell line, which I developed. The knowledge that my fundamental research has direct applications to medicine is very rewarding.

At the University of Virginia, I continued my work on the molecular mechanism of the ATP-synthase as a co-Investigator with Dr. Robert Nakamoto. Our collaboration has been very successful and we have published several papers together. We defined a rotational mechanism of energy conservation by the ATP-synthase and have gone on to define the chemical and conformational pathways of coupling. For the last nine years, I have also been the Principle Investigator on an NIH-funded project to establish the "Drug Transport Mechanism of P-glycoprotein". In this project we developed a high-yield human plasma membrane protein expression system in yeast. We have established a new chemical and coupling model of drug transport that address all aspects of P-glycoprotein function and physiology. Additionally we pioneered the application of EPR studies in this field and engineered novel spin-labeled drugs for transport, kinetic and structural studies. We have generated realistic atomic detail structural models of P-glycoprotein in the lipid bilayer. Through molecular dynamics we have investigated the dynamic interaction of drugs with the membrane and protein during the reaction cycle. This 200,000 atom model is one of the biggest systems ever simulated in such detail. Clearly we are in a good position to make important discoveries regarding the nature of drug transport by P-glycoprotein.

A complimentary function that I enjoy, is the mentoring and teaching of students, post-docs and technicians both formally and informally. I will always make time to mentor and illuminate my areas of scientific and technical expertise to any person that seeks such knowledge. When I have contributed to the understanding or career development of a student I derive a great deal of satisfaction.

The structure and function of membrane proteins is currently a targeted strategic research initiative of the NIH and part of their long term vision. I am currently very well positioned to take advantage of these initiatives. Thus I intend to continue making important findings in the structure-function relationships of P-glycoprotein which should have long term benefits to chemotherapy and AIDS treatment. As more resources become available, I will leverage my expertise to study the structure and function of some of the 47 other related human ATP-binding cassette (ABC) transporters. These proteins play vital roles in the cellular transport of nutrients, metabolites, drugs, toxins, xenobiotics, lipids, cholesterol, steroid and hormones. Defects in these transporters cause a myriad of crippling human disease including cystic fibrosis, adrenoleukodystrophy, macular dystrophy, Tangier disease, Dubin Johnson syndrome, sitosterolemia and many others. Detailed structure-function studies of these proteins are urgently needed to provide the scientific basis for ameliorating these tragic defects.

The University of Virginia provides me with an excellent environment to pursue my passions. The availability of marvelous research facilities, the immediate presence of great well-informed and supportive colleagues who are at the forefront of academic medicine, as well as the availability of bright, enthusiastic and highly motivated students and fellows all support and enhance my research and academic missions.