Protein Science (2003), 12:398-400.
Published by Cold Spring Harbor Laboratory

Protein Science
A PUBLICATION OF THE PROTEIN SOCIETY

NEWS FROM THE PROTEIN SOCIETY
February 2003

Meeting Report

CARL W. ANDERSON[1], JUAN CALVETE[2], JAY W. FOX[3], AND ETTORE APPELLA[4,5]

[1]Carl W. Anderson, Biology Department, 50 Bell Avenue, Brookhaven National Laboratory, Upton, NY 11973 USA.
[2]Juan J. Calvete, Instituto de Biomedicina, C.S.I.C., Jaime Roig, 11, E_46010 Valencia, Spain.
[3]Jay W. Fox, Department of Microbiology, University of Virginia Health Systems, P.O .Box 800734, Charlottesville, VA 22908_0734 USA.
[4]Chemical Immunology Section, Laboratory of Cell Biology, NCI, National Institutes of Health, Bethesda, MD 20892 USA.
[5]President International Association of Protein Structure Analysis and Proteomics

Methods in Protein Structural Analysis Conference (MPSA2002), the 14th conference in this series, was held in the elegant city of Valencia, Spain September 8-12, 2002 and was attended by 200 researchers from more than two dozen countries. The MPSA conferences began in 1974 with a small workshop in Boston, MA organized by Richard A. Laursen, Boston University, for the purpose of exchanging information on the then newly developed instruments and chemistry for sequencing proteins. MPSA conferences are held every two years and have usually alternated between Europe and the U.S. They are now sponsored by the International Association of Protein Structure Analysis and Proteomics (IAPSAP, http://www.med.virginia.edu/medicine/basic_sci/micr/fox/iapsap/iapsap.html), a not-for-profit organization established in 1999 to promote the discovery and exchange of new methods and techniques for the analysis of protein structure (Appella et al., 2001). Although the scope of MPSA conferences has expanded as new techniques for protein analysis have been developed, the focus of the conferences has remained on sharing cutting edge techniques for protein analysis. The rationale for MPSA2002 was to explore new experimental and computational approaches for protein structural analysis that extend from primary sequence analysis through advanced techniques for imaging protein machines in living and near life-like circumstances.

The conference opened in the elegant, neoclassical Paraninfo of the Old University of Valencia with a Plenary Lecture, "The Atomic Structure of the 30S Ribosomal Subunit: Insights into the Decoding Mechanism", by Venki Ramakrishnan, Medical Research Council, Cambridge, U.K. After reviewing the recently derived structure of this small, megadalton-sized subunit of the bacterial ribosome, which is responsible for binding mRNA and the anticodon stem-loop of tRNA, Dr. Ramakrishnan went on to explain insights into the decoding mechanism that his structural work revealed, how the structure contributes to the fidelity of translation, and a rationale for the wobble hypothesis and why degeneracy in the genetic code is tolerated at the third position (Ramakrishnan, 2002). He also explained in structural terms how aminoglycoside antibiotics affect the fidelity of translation.

Subsequent sessions were held at the Colegio de Médicos, a modern facility not far from the ultra-modern Ciudad de las Artes y las Ciencias (Arts and Science Park) located in the old river bed that partially surrounds the old city of Valencia. Udo Heinemann, Max-Delbrück Centrum für Molekulare Medizin, Berlin, described efforts of the Berlin-based "Protein Structure Factory", which is an international effort committed to developing high throughput methods for protein expression, sample preparation, and data acquisition for structure determination by both X-ray diffraction and NMR. The Factory concentrates on structures of human proteins and reports a ~70 percent success rate at expressing human proteins in *E. coli* using one of two expression systems. Carol V. Robinson, who recently moved to Cambridge

from the Oxford Center for Molecular Sciences, described studies on the very large, multi-subunit protein complexes using a modified Micromass Q-TOF instrument; these included the GroEL-GroES-ATP bacterial chaperone complex, other multi-subunit plant and animal chaperones, as well as the spliceosome. Bonnie A. Wallace, Birbeck College, University of London, then described how synchrotron radiation circular dichroism spectroscopy could be used to determine protein secondary structure, analyze protein folds, and measure folding kinetics. UV synchrotrons permit measurements to 160 nm, which substantially increases the information content of the spectrum compared to conventional CD instruments.

Janet Thornton, EMBL European Bioinformatics Institute (EBI), Hixton, U.K., asked the questions: How did function evolve? How reliable is sequence analysis in the transfer of information to query sequences? Proteins with less than 40 percent sequence identity have more than a 50 percent chance of performing a different function. Sheena E. Radford, University of Leeds, U.K., described mechanisms of protein folding and misfolding *in vitro* and *in vivo* derived from her studies of colicins and other small immunity proteins. Folding intermediates are important! Alfonso Valencia, Centro de Biotecnologia, C.S.I.C., Madrid, then described the development of software tools for massive sequence analysis by his Protein Design Group in collaboration with the EMBL bioinformatics institute. Returning to the theme of protein synthesis, this time from the practical side, Shigeyuki Yakoyama, RIKEN Yokohama Institute, Japan, described cell-free systems for large-scale protein synthesis derived from bacteria, wheat germ, or rabbit reticulocytes. These systems, programmed with either DNA and RNA polymerase or mRNA, are capable of producing in excess of a mg protein per ml per hr and may operate up to 10 hr. Thus, it has become possible to make proteins for structural analysis using such systems.

The second full day began with Chris P. Ponting, MRC Functional Genetics Unit, Oxford University, U.K., who described genome cartography through domain annotation, emphasizing the relationships between the mouse and human genomes. In contrast to humans, mice have dozens of domain and protein families that have expanded and are rapidly evolving. Bruce J. Mayer, University of Connecticut Health Center, USA, then described methods to manipulate the global tyrosine phosphorylation state of cultured cells. Using a battery of different SH2 (tyrosine-phosphate binding) domains, one is able to profile or fingerprint the cell's phosphorylation state. He also described a method, termed FIT (functional interaction trap), to determine the downstream consequences of protein phosphorylations. Manfred Nimtz, GFB-Gesellschaft für Biotechnologische Forschung, Germany, discussed the use of MS techniques in structural characterization of glycoproteins, such as those from the human filarial parasite that causes river blindness, while Martin R. Larsen, University of Southern Denmark, Odense, described the characterization of modified proteins by gel electrophoresis and mass spectrometry, concentrating on sample preparation using micro-columns packed with a variety of loadings including pencil graphite.

Wednesday's session on Advanced Technologies began with a talk by Volker Doetsch, University of California San Francisco, USA, who described how new ^{15}N and ^{13}C labeling techniques could be used in high resolution macromolecular NMR spectroscopy inside living cells to determine how proteins, such as bacterial NmerA or human calmodulin, switch from an active to an inactive conformation. John S. Vogel, Center for Accelerator Mass Spectrometry, Livermore, USA, then described how accelerator-based mass spectrometry, coupled with conventional high resolution protein separation techniques, could be used to overcome the inefficiencies of measuring long-lived isotopes, such as tritium or carbon-14, to quantify with subattomole sensitivity protein interactions with small molecules such as toxins, vitamins, and natural biochemicals at an accuracy of 1.5%. Francesc X. Avilés, Institut de Biotechnologia i de Biomedicina, Universitat Autònoma de Barcelona, Spain, described a technique they call "Intensity-Fading MALDI-TOF MS", which uses non-specific proteinases, to explore secondary structure elements of proteins and the effects of mutations on protein stability.

Wolfgang Baumeister, Max-Planck-Institut für Biochemie, Martinsreid, Germany, described the use of cryo-electron microscopy tomography to investigate molecular machines in whole cells embedded in vitrified ice in a close-to-life state. With a resolution of about 4 nm, complexes in the range of 0.5 to 1 MDa can be identified with good fidelity. José Maria Valpuesta, Centro Nacional de Biotecnologia, C.S.I.C., Madrid, Spain, described how the cytosolic chaperonin CCT recognizes its specific substrates using specific combinations of subunits. CCT is involved in the folding of critical structural components such as actin and tubulin. Rolf Apweller, EBI, Hixton, U.K., described recent developments at the EBI Sequence Retrieval System (SRS) server including the Integrated Resource of Protein Domains and Functional Sites (InterPro). New funding from the U.S. NIH should allow improved functional and automatic annotation of entries in the SWISS-PROT and TrEMBL databases. Manfred J. Sippl, University of Salzburg, Austria, went on to describe the role of structure prediction in genomics, the assessment of fold recognition from recent CASP competitions, and the use of ProSup, a tool for refined protein structure alignment.

For the final sessions, participants returned to the Paraninfo in the Old University to hear Bernard Jacq, Laboratoire de Génétique et Physiologie du Développement, IBDM-CNRS, Marseille, France) return

to the question: What is protein function? The meaning and understanding of the term varies depending on the functional level considered. Jacq proposed a new method for protein function comparison, PRODISTIN, based on the identiy of interacting proteins and showed how this concept could be applied to *Helicobacter pylori* and *Sacromyces cerevisiae*. Marc Van Regenmortel, Ecole Supérieure de Biotechnologie de Strasbourg, Illkirch, France, reminded us that there is no single causal link between structure and function and that at the molecular level, primary protein activity is usually binding activity, which can be investigated to determine and predict quantitative structure-activity relationships (QSAR) in response to chemical environment and protein sequence using the BIACORE. Gitte Neubauer, Cellzome AG, Heidelberg, Germany, then described the use of tandem-affinity purification coupled to MALDI-TOF mass spectrometry and bioinformatics to show how shared components of protein complexes of *S. cerevisiae* revealed networks underlying biological processes. Finally, Edouard C. Nice, The Ludwig Institute for Cancer Research, Melbourne, Australia, described the use of multiple biophysical techniques to reveal structure-function relationships among the EGF/EGFR family and the use of affinity-based optical biosensors as microaffinity purification platforms for proteomics analyses.

In the space available, we have only provided a taste of the exciting paellas of information that were offered to participants over the four days of scientific presentations. The MPSA2002 program and abstracts are available on the meeting website (http://www.mpsa2002.ibv.csic.es/); the scientific presentations were videotaped, and the meeting organizers plan to make selected presentations available through the internet.

To commemorate its origins and honor Pehr Edman, in 1988, MPSA began awarding at each meeting the Edman Award to individuals whose efforts had significantly advanced the field (Wittmann-Liebold, 1989). This years Edman Awardees were Hans Jörnvall, Karolinska Institute, Stockholm, Sweden, for contributions to protein chemistry, structure and function through the development of techniques associated with the use of Edman chemistry and structure-function studies on alcohol dehydrogenases, and Emeritus Professor Kenneth Walsh, University of Washington, Seattle, Washington, USA, for his contributions to the development of mass spectrometry for characterizing the chemical structure of proteins and their posttranslational modifications and for concepts contributing to the evolution of protein structure and function. In 2000, IAPSAP added a Distinguished Postdoctoral/Young Investigator Award. This year's Distinguished Young Investigator Award went to Dorothee Kern, Department of Biochemistry, Brandeis University, USA, for studies on the dynamic processes that trigger protein function related to enzyme catalysis, ligand binding and signaling events. Both the Edman Award and the Distinguished Young Investigator prizes were sponsored by PE Applied Biosystems.

During the five day meeting participants also enjoyed the many attractions of Valencia, including the Ciudad de las Artes y las Ciencias, the Central Market (one of the largest indoor fresh produce markets), the Cathedral and the IVAM (Modern Art Museum), in nearly perfect, late-summer weather. Indeed, the meeting banquet, which featured a half-dozen different paellas, was held outdoors in the Botanical Garden. MPSA2002 closed with the announcement of the venue for MPSA2004, which will be held in Seattle, Washington, USA, August 29 through September 2, 2004.

References

Appella, E., J. W. Fox, and C. W. Anderson. 2001. Meeting Report. Methods in Protein Structural Analysis Conference (MPSA), Charlottesville, Virginia, September 16-20, 2000. Protein Science **10**, 459-461.

Ramakrishnan, V. 2002. Ribosome structure and the mechanism of translation. Cell **108**, 557-572.

Wittmann-Liebold, B. (ed). 1989. *Methods in Protein Sequence Analysis*, Springer-Verlag, Berlin, pp 575.