Intermolecular Associations in 2D and 3D

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Introductory remarks

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Abstract

An overview is given of a stimulating Meeting held at the University of Nottingham in June 2003 focusing on molecular interactions occurring in membranes or '2D' and those occurring in aqueous solution or '3D'. It was held jointly between the Biochemical Society and the British Biophysical Society. The 80 or so delegates who attended benefitted from an exciting exchange of ideas between researchers from a wide spectrum of backgrounds. It is hoped the collection of papers which follow this Introductory paper will provide a useful summary of the state of the art and help stimulate collaboration across the wide range of disciplines represented.

Interactions between molecules underpin the whole of biological science, both in '2D', as in membrane systems, and in '3D' or aqueous systems. Our ability to understand these interactions continues to increase rapidly as new techniques, such as atomic force microscopy and single particle methodologies, are developed, alongside exciting advances involving such long-established techniques as tracer sedimentation equilibrium and solid-state NMR as just two examples. To illustrate this, a two-volume series was recently published describing the practical aspects of technologies for studying molecular interactions in proteins [1,2]. A meeting focusing on the advances in techniques and applications to both systems appeared very timely for both establishing the state of the art and keeping pace with these exciting developments. Cross-fertilization of ideas and approaches was also a desired theme of the meeting, and it was thought productive to mix 'membrane people' with 'solution people'.

On the 19th and 20th June 2003 a Joint Meeting was held between the Biochemical Society – under their Focused Meeting series – and the British Biophysical Society, at the University of Nottingham, with generous support from Wyatt Technology UK, Biacore and Pro2Kem Ltd. About 80 delegates from the U.K., continental Europe, the U.S.A. and Australia used a series of seminal presentations from experts in the field together with around 25 poster presentations to promote a stimulating exchange of ideas between researchers studying interactions in membranes and aqueous solution. One speaker argued his presentation stretched across both areas in equal proportions and requested the use of '2.5D' in his title! The meeting fell into three related sections: 'Structure and Spectroscopic Probes', 'Hydrodynamic Probes' and 'Application to Specific Systems', and we have followed the same division here in the following collection of papers arising from the meeting:

In the first section on 'Structure and Spectroscopic Probes', R. Palmer and H. Niwa provide a review of the advances in crystallographic approaches to study protein–ligand interactions: although the vast majority of structures refer to proteins soluble in aqueous soluble systems, the problems in getting membrane proteins to crystallize are steadily being overcome. R. Kroemer then highlights the advances in molecular modelling techniques. The modelling methods covered ranged from binding site analysis to statistical treatment of sets of ligands. Considerable effort was being put into high-throughput docking and scoring based on energy calculations. The power of modelling methods is strongly affected by the quality of the experimental data they have to relate to: P. Derrick and co-workers follow with a contribution reviewing how the related methods of electrospray ionization and mass spectrometry are helping provide such information. P. O'Shea then introduces the added problems presented by studies with membranes. Whilst studies of

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intermolecular interactions with membranes might seem to involve a reduction of dimensionality (hence the title of this Meeting) and so give an impression that the number of degrees of freedom are reduced to simplify interpretations, they bring a whole host of added complexities, as indicated by the speakers. With this in mind, he shows how the membrane structure, the membrane electrostatics and local molecular dipolar properties have an important bearing on how molecules interact both on and within membranes. This is followed by the paper from E. Gratton and co-workers, who show that FCS (fluorescence correlation spectroscopy) offers a powerful means to study molecular interactions within cells, as well as with membranes. L. Salomé and co-workers then describe their studies on the 2D diffusion of proteins in membranes that pointed to long-range intermolecular forces dominating some of their diffusional properties. J. Clarkson and I.D. Campbell then eloquently describe how NMR spectroscopy continues to amaze with the variety of developments and possible applications towards studies of protein–ligand interactions.

The following three papers then review some of the major advances in hydrodynamic and calorimetric probes. Although of lower resolution than the spectroscopic and imaging probes, they do refer to solution media (aqueous and non-aqueous). D. Winzor gives a personal view on the development of chromatographic procedures for determining reaction stoichiometries and equilibrium constants in both protein self-association and ligand binding by the biosensor variant of affinity chromatography. G. Rivas and A. Minton then provide a review of sedimentation equilibrium methods – focussing on the enormous sensitivity of 'tracer' variants for the study of protein association and hetero-associations in dilute solution and highly crowded environments of concentrated solutions.

Subsequently, the final papers refer to applications to specific biological problems. T. Jovin and co-workers reveal the power of fluorescence technologies to the study of defined intermolecular interactions using lifetime imaging and resonance energy transfer techniques. Particularly novel was the incorporation of polarization techniques into these methodologies. This is followed by R. Cherry and co-workers, who describe, using single particle fluorescence imaging, how cell surface receptors associate. The theme of molecular recognition continues with T. Michaelsen's paper, which compares the different antigen binding properties of IgG and IgM classes of antibody possessing identical variable (V) regions. Antibodies are only relatively modestly glycosylated: the following paper by S. Harding describes interactions involving highly glycosylated systems, namely mucoadhesive interactions involving mucins with both a highly cationic protein (*mefp1*) and chitosans. Solution techniques of analytical ultracentrifugation and SEC-MALLs (size exclusion chromatography combined with multi-angle laser light scattering), combined with electron microscopy and atomic force microscopy, are providing useful molecular insights into the interactions involved and how they may be manipulated for therapeutic purposes. I. McEwan and co-workers then describe the interplay between protein conformation and protein–protein interactions. In the penultimate paper, A. Bain and co-workers suggest the possibility of applying excited state techniques involving polarized excitation then 'dumping' a component of the excited population to get round 'selection rules' constraints. This could conceivably offer the means of revealing a whole host of new dynamical modes of macromolecular behaviour. In the final paper, S. Allen and co-workers describe how atomic force microscopy (AFM) is being used to study interactions between single molecules, and they highlight its particular potential for the study of peptides and proteins with membranes. Although not a true selection technique, AFM does permit the high-resolution study of interaction in a lipid environment, and there are clearly exciting possibilities especially when used in conjunction with other methods. The virtue of combined approaches to the study of specific interactions was a recurrent theme throughout the Meeting.

We hope this published collection of papers will not only provide a permanent record of a stimulating meeting, but will also provide researchers investigating interactions in both membranes and aqueous media with a useful summary of the state of the art as in June 2003. These papers may also help readers share some of the excitement that we experienced in this meeting, and stimulate collaboration between researchers across the wide range of disciplines represented.

References

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