Laser and Optical Spectroscopy

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Light, chemistry and photophysics are the most natural, cooperative and synergistic of partners. The energy in a single particle (photon) of visible light is just that amount needed to perform chemical transformations. Nature takes supreme advantage of this synergy and teamwork in the process of photosynthesis, so essential to most life on the planet.

Spectroscopy delves into the subtle yet *absolutely distinctive* interactions of light, both visible and invisible, with the many forms of matter. Normally, light can be absorbed, emitted or just scattered. Using lasers, other far less familiar processes are possible. The wide range of spectroscopic techniques now available may be utilised as analytical or diagnostic tools, right down to the extreme limit – measurements on *single* molecules.

At a more fundamental level, spectroscopy provides potent methods with which to probe the innermost secrets of all chemical species. Spectroscopy also maps out the detailed electronic structure of all the different forms of matter. This information is critical to the understanding of both chemical bonding and chemical reactivity.

Laser technology continues to evolve and in the process revolutionise spectroscopy. Laser light is *very* different to ordinary light (sunlight or lamplight, etc.). Lasers can be made of one single colour, to a precision better than one part in a billion. Light pulses can be compressed into incredibly short pulses or amplified to a point where the very distinction between light and matter becomes blurred. Put simply, lasers induce processes to occur that are just not seen with "ordinary" light sources.

Our group performs spectroscopic measurements on a wide range of materials and systems: organic and inorganic, molecular, ionic, amorphous, crystalline, and increasingly, biological. Our great strength is the ability to design, develop and invent special experiments and apparatus to target particular questions. A molecule may behave very differently in solution to when it is 'trapped' in the special environment of a protein or crystal. These critical environmental influences may be identified and probed via the application of laser selective spectroscopy.

The study of Photosystem II continues to dominate our activities. Professor Elmars Krausz spoke on some of our recent Photosystem II results at the Conference on Physical Chemistry in Christchurch New Zealand in February. Dr Sindra Peterson also presented work at the Photosynthesis Gordon Conference in July in Rhode Island, US, as well as giving a well received seminar in Stanford. Dr Barry Prince and Dr Sindra Peterson presented work at the Australian Biophysics Society meeting in Melbourne.

A major success this year was granting of an ARC LEIF bid, to develop and construct *two new generation* metallo enzyme magneto-optical spectrometers. One system is to be located at the University of Queensland with Dr Mark Riley and the second at the ANU. Both systems are to be constructed at the RSC using experience and technologies we have gained over the last decade. Further funding success was the granting of a joint ARC Discovery grant with Professor Rob Elliman of the Research School of Physical Sciences to perform optical studies of silicon nanocrystals.

Electrochromism and EPR associated with Radical Formation in Photosystem II

We have discovered that PSII core complexes undergo surprisingly rapid and efficient photochemistry at low temperatures (1.7K), with high yields of the quinone radical anion. This anion gives rise to a large and characteristic electrochromic (Stark) shift on the close-by pheophytin, from details in optical spectra. In a 'physiological' PSII illumination process, the electron donor would be the Manganese cluster of the oxygen evolving centre, but this (so called S state) process is inhibited at low temperatures. Alternative electron donors usually considered are cytochrome b559, chlorphylls, □-carotenes and the two redox active tyrosines. None of the above candidates appear to be the majority donor in samples where the cytochrome is oxidised and we are attempting to identify the 'mystery radical' via optical and EPR studies. *(with S. Peterson, B. Prince, and R. Pace, P. Smith [Dept. Chemistry, ANU])*

Spectroscopy of Oriented Photosystem II

The crystal structure of Photosystem II has established many aspects of the overall displacement and orientation of the pigment molecules in PSII. This in turn has allowed an increasingly detailed analysis of high resolution optical spectra. It is possible to *orient* PSII enriched membrane fragments of PSII by evaporation of a solubilised sample onto a glass surface. The measurement of linear dichroism spectra, taken of a tilted plate which supports the stacked membranes, indicate that symmetry between the pigments in the D1 and D2 proteins in PSII is absent. Features attributed to the largest electronic coupling in P680 may arise from two pigments within D1 and not from the 'special pair' of chlorophylls. *(with S. Peterson, C. Dobson, and R. Pace, P. Smith [Dept. Chemistry, ANU]*

Single Crystal Spectroscopy of Photosystem II

Crystallisation of PSII core complexes can *initially* lead to the formation of very fine (green) needles, of only a few microns thickness. Although such crystals are too small for structural studies, they are the appropriate thickness to measure absorption features associated with the pigments in the PSII core complex. Our experience with single crystal spectroscopy has allowed us to perform low temperature polarised spectral measurements of these tiny, fragile crystals, although at present they appear to undergo photochemical damage during the sample mounting process. *(with S. Peterson, and R. Pace, P. Smith [Dept. Chemistry, ANU], M. Parker [St. Vincent's Inst. of Medical Research, Melbourne])*

Synechoccoccus Vulcanus, a Hot Opportunity

This thermophillic bacterium is one of two related species that have succumbed to crystallisation of its PSII core complex, leading to subsequent X-ray structural determination. We have been fortunate enough to be invited to collaborate with a group performing structural studies. We have already seen that there are significant differences between plant and cyanobacterial core complexes. Measurements on Vulcanus sample preparations having high activity, purity, and stability, and for which the structure is known, is certain to advance our understanding of P680 and PSII in general. We have shown (above) that it is feasible to perform spectroscopy on small single crystals of such materials. *(with S. Peterson, and R. Pace, P. Smith [Dept. Chemistry, ANU], J.-R. Shen [Riken Harima Inst., Kyoto, Japan])*

http://rsc.anu.edu.au/krausz.html