efficient drop inspection and analysis, these technological advancements have initiated the development of microplates with low birefringent background (LBR plates) to allow more effective use of polarized light in protein crystal detection. LBR plates are especially beneficial for identification of crystals out of focus, very small microcrystals, microcrystals hidden in or resembling precipitate or phase seperation and crystals located at the edge of droplets or crystallization wells.

As an alternative to classical microplates, plastic microstructured devices for liquid-liquid diffusion crystallography offer the benefits of low protein and reagent consumption, ease of handling and time conservation. Further advantages of plastic microstructures devices are a broad selection of available raw materials and surface treatments as well as reasonable costs of manufacture.

Keywords: high throughput crystallography, polarized light microscopy, microfluidics

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Lactate Dehydrogenases from Extremophile Organisms: Clues for Radio-resistance?

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Lactate dehydrogenases (LDH) catalyse the last step of glycolysis in which pyruvate is reduced by NADH to lactate. Our structural study of LDH from *D. radiodurans* finds its place in a broader project aimed at investigating the effect of ionising radiation on many enzymes of the malate/lactate dehydrogenase family from extremophile organisms (thermophiles, psychrophiles, halophiles and radio-resistants).

Ionising radiation, including synchrotron radiation, has specific effects on structure, activity and stability of biological macromolecules and represents at the same time the cause of the damage and the tool to study it [1].

In order to better understand how molecular adaptation to extreme environments is achieved, and if it confers radio-resistance, the structure of LDH from *D. radiodurans* has been solved, both in its native form and in complex with the allosteric cofactor fructose 1,6-biphosphate (FBP). A structural comparison with LDHs from other extremophile organisms is under way. Furthermore, the sensitivity to ionising radiation of the two allosteric forms and the possible protective effect of FBP and NADH against radiation damage is currently being investigated.

[1] Weik M., Ravelli R.B., Kryger G., McSweeney S., Raves M.L., Harel M., Gros P., Silman I., Kroon J., Sussman J.L., *Proc Natl Acad Sci USA*, 2000, **97**, 623.

Keywords: LDH, radiation damage, allostery

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Interactions of Phospholipids with Integral Membrane Proteins; Use of Brominated Lipids

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The lipid environment of integral membrane proteins is important to their structure and function, but little is known about specific protein-lipid interactions. Knowledge of these will help in devising better protocols for solubilisation and crystallisation of membrane proteins. Although some lipids may co-purify with the protein and show as residual electron density, the detergent used in solubilisation can confuse the interpretation. By co-crystallising the protein with brominated lipids their binding sites can be distinguished.

We have located two lipid binding sites in a complex of the reaction centre (RC) from *Rb. sphaeroides* with the 1-Palmitoyl-2-Stearoyl(6,7)-dibromo-sn-glycero-3-Phosphocholine (Br-PC) based on single wavelength data. We are now producing complexes of RC with variety of brominated lipids changing the lipid headgroup, the number and positions of bromine atoms, and concentration of the lipid in co-crystallisation with RC. We would also like to perform MAD experiments for some of these complexes at the Br K-edge (around 13.47 keV) to improve the anomalous signal and also to assess the possibility of using this method to phase structures of integral membrane proteins. Results of these experiments will be described.

The authors are members of the membrane protein structure initiative (MPSI), supported by the BBSRC.

Keywords: membrane protein crystallisation, brominated phospholipids, MAD and SAD experiments

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Structural Studies of Water-soluble Chlorophyll Protein from *Chenopodium album*

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Generally, chlorophyll (Chl) molecules functioning in photosynthesis are associated with hydrophobic integral membrane proteins. Water-soluble Chl protein (WSCP) was first found in *Chenopodium album* in 1963. WSCPs have then been detected in several species classified in the Polygonaceae, Chenopodiaceae, Amaranthaceae and Brassicaceae families. Although the physiological function of WSCPs has not yet been cleared, these WSCPs can be categorized into two classes according to their photoconvertibility: *Chenopodium*-type (Class I) and *Brassica*-type (Class II). The absorption spectrum of a Class I WSCP changes drastically on exposure to visible light, while a Class II WSCP does not. And there is no significant sequence homology between Classes I and II WSCPs. The X-ray structure analysis of Class II WSCPs containing *Lepidium*-, *Raphanus*- and Kale-WSCPs reveals that these WSCPs consist of 4 subunits and a Chl is contained in each subunit. In order to determine the crystal structure of a Class II WSCP and elucidate the photoconversion mechanism, *Chenopodium*-WSCP was extracted from leaves, purified, and crystallized in a dark room. Green rod crystals appeared in a week. A native data set was collected to 3.0 Å resolution at 100 K with synchrotron radiation at PF. The space group of the crystal was determined to be orthorhombic *I*222 with unit-cell parameters a = 47.08, b= 61.42, and c = 107.0 Å. Heavy atom derivative screening for structure determination is in progress. The photoconversion mechanism and the interaction between Chl and the protein are being studied.

Keywords: water-soluble chlorophyll protein, photoconvertibility, pigment protein

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Structure of the C-terminal Domain of DipZ from *Mycobacterium tuberculosis*

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DipZ (Rv2874) from *M. tuberculosis* is a member of the CcdA protein family. This family of proteins shares a conserved transmembrane electron transport domain with similarity to CcdA from *Rhodobacter capsulatus* and varies in size from 190 aa to over 750 aa. It has been proposed that the function of the larger proteins has been modified by the acquisition of extra-cytoplasmic protein domains. The transmembrane region functions by passing electrons from the cytoplasm of the cell across the membrane for use by these extra-cytoplasmic domains [1].

The C-terminal soluble domain from DipZ has been crystallised and the structure determined by SAD methods from crystals soaked in K_2PtCl_4. The model reveals a dimeric structure. Each subunit is comprised of two domains, an N-terminal thioredoxin-like fold predicted from earlier sequence alignments, and a C-terminal jelly-roll fold with similarity to the family 6 carbohydrate binding modules. A