wild type DLH crystals to study the time-dependent nature of the oxidation of the active site Cys123 residue. Data were collected both on the in house CCD detector system and image plates. Initial results will be presented from these two studies.

References:
Neimann A, Matzinger P & Hädener A (1994) *Helvetica Chimca Acta* ,77 1791 - 1809
Pathak D & Ollis D.L. (1990) *J. Mol. Biol.*, **214**, 497 - 525

MS02.02.09 LAUE STUDIES ON ACETYLCHOLINESTERASE. Mia L. Raves, (Weizmann Institute of Science), Raimond B.G. Ravelli, Jan Kroon, (Utrecht University), Michel Roth, Dominique Bourgeois, (IBS, Grenoble), Ling Peng, Maurice Goeldner, (Université Louis Pasteur, Strasbourg), Israel Silman and Joel L. Sussman, (Weizmann Institute)

In the last ten years it has become feasible to study dynamic processes in macromolecules at the atomic level. Most of these studies have used the Laue diffraction technique that permits data collection on a second to pico-second time scale by virtue of the polychromatic synchrotron radiation.

It is our aim to do time-resolved experiments on the enzyme acetyl-cholinesterase (AChE). AChE cleaves the neurotransmitter acetylcholine (ACh) in the synapse at a very high turnover rate. The enzyme has a strong dipole moment that draws the positively charged substrate towards the catalytic site, situated at the bottom of a narrow gorge lined with aromatic residues. One of the reaction products, choline, bears the same charge as the substrate, ACh, which raises the problem of its exit route from the active site. In order to attempt to observe the reaction process, photolabile precursors of choline and of carbamylcholine have been synthesized and characterized. These two probes generate choline in different ways, either by direct photocleavage (choline precursor) or by enzymatic hydrolysis of a substrate generated by photocleavage (These tools for the time-resolved crystallographic studies will hopefully permit monitoring of the exit of choline from the active site.

To test the feasibility of the use of the Laue method, trigonal crystals of Torpedo californica AChE soaked with the inhibitor edrophonium were used to collect data at the ESRF in Grenoble, beam line ID9 (BL3) on a CCD detector with an exposure time of 1 msec. The frames were indexed using a new program called Lauecell that is able to determine the relative cell parameters from the Laue pattern semi-automatically. Further processing with the CCP4 Laue software package yielded a data set to 3.1 Å resolution of 80% completeness with 15,000 unique reflections. Constrained refinement of the protein coordinates with X-PLOR gave a difference map in which the density for the inhibitor can be clearly seen.

1. Peng, L. et al. submitted to Biochemistry (1996).
2. Ravelli, R.B.G.et al. accepted in J. Appl. Cryst (199

PS02.02.10 THE DARESBURY LABORATORY LAUE SOFTWARE SUITE. S. Arzt, J.W. Campbell, Q. Hao, D. Nguti, CCLRC Daresbury Laboratory, Daresbury, Washington, WA4 4AD, Cheshire, U.K.; M.M. Harding, University of Liverpool, U.K.; J.R. Helliwell, G. Bradbrook, J. Habash, Y.P. Nieh & E.H. Snell, University of Manchester, U.K.

High quality Laue intensity data can be obtained using synchrotron radiation. The Daresbury Laboratory Laue Software Suite has been developed and 'calibrated' with a series of studies (pea lectin, carbonic anhydrase, concanavalin A, lysozyme, as well as smaller inorganic and organic structure). It is available for distribution from Daresbury (contact Dr. J.W. Campbell) and has been successfully worldwide (e.g. p21 catalysis, Heidelberg; trypsin, NSLS; isocitrate dehydrogenase, Seattle; ESRF Grenoble etc.; see also poster abstract of Carr et al.). The package consists of a series of programs written primarily in FORTRAN, but also using libraries written in 'C' and runs on UNIX and VMS based computer systems. The LAUEGEN program uses an Xwindows based toolkit (XDL_VIEW) also developed at the Daresbury Laboratory. The aim is to exploit the interactive and display facilities available using X-windows but at the same time to provide more automatic procedures for data processing where these are appropriate. A Laue Data Module defines a set of standard parameters describing the crystal, the X-ray detector and the scanned images. It also provides a set of program independent functions for handling these parameters. Other recent developments allow the automatic estimation of sizes and the soft limits λ_{min} and d_{min}. A particular recent emphasis has been to provide new algorithms for deconvolution of the small fraction of multiples; these are important to give fuller coverage at low resolution. A method based on the λ-curve has been implemented and tested with lysozyme (see Campbell et al. (1994), Bull. Mater.Sci. 17, 1-18) and explored in relation to angular sampling collection strategies (see Bradbrook et al. (1995) SPIE 2521, 160-177 for the Röntgen Centennial Celebration).

PS02.02.11 BAYESIAN APPROACH TO THE ANALYSIS OF TIME-RESOLVED PROTEIN LAUE DIFFRACTION DATA. G.P. Bourenkov, A.N. Popov and H.D. Bartunik, Max-Planck Research Unit for Structural Molecular Biology, Protein Dynamics Group, MPG-ASMB c/o DESY, Notkestraße 85, 22603 Hamburg, Germany

A new method of deconvoluting overlapping reflections in protein Laue diffraction patterns solves the problem of the "low resolution hole" without the need for redundancy. It is therefore of particular interest for single-shot time resolved studies. For the first time, non-cyclic reactions may be investigated by Laue diffraction on short time scales. In test application to orthorhombic bovine trypsin, the new method improved the resolution from 1.7 Å to 1.4 Å as compared to standard processing methods. It provided high completeness over the whole resolution range < 7Å. The contrast in electron density maps calculated with the Laue structure factors improved dramatically; the Laue maps are of similar quality as maps that are calculated with monochromatic high-resolution data. The method follows a Bayesian statistical approach. A-priori given information about structure factor amplitudes obeying Wilson's distributions is employed. The (single or redundant) measurement of the intensity of an energetic or spatial overlap yields the normal multivariate probability density function (PDF) of the intensities of the components. This information is associated with the prior PDF through the Bayes theorem. The moments of the resulting posterior PDF give expected values for the component intensities, the structure factor amplitudes and their uncertainties. These moments are always finite and positive, even if the initial normal matrix is degenerate. Due to the nature of the wavelength normalisation curve and the dependence of the scattering power on resolution, accurate estimates are obtained for the structure factors of the components, even in the case of a single observation of an energetic overlap. Furthermore, all data can be processed to the physically relevant wavelength-dependent diffraction limit. No "soft parameters" are involved. The power of the method may be further enhanced, if a (roughly) approximate structural model is available, e.g., of an initial state of a reaction. Then, conditional prior probability density functions may be employed.