

 ESRF	Title Structural bases for allosteric regulation of acetylcholinesterase catalysis. Project 2: Search for the route for substrate entry into the fasciculin-acetylcholinesterase complex.	Experiment number: LS-895	
	Beamline: ID14-EH3	Date of experiment: from: 16/02/98 to: 17/02/98	Date of report: 28 Feb 98
	Shifts: 6	Local contact(s): Wim Burmeister	<i>Received at ESRF:</i>

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Report:

Two data sets were collected on crystals of the AChE-Fas2 complex, a « native » form of the complex, and an « inhibited » form, in which a ternary inhibitor acting at the catalytic site of AChE was soaked. The crystals belong to the hexagonal space group P6522 with cell dimensions: $a=b=75\text{Å}$ and $c=550\text{Å}$. The diffraction quality of these frozen crystals were first tested with the Mar CCD with a crystal-to-detector distance of 147.8Å giving a maximal resolution of 2.2Å ($\lambda=0.947\text{Å}$). The Large Imaging Plate detector (8000 x 4000 pixels) was then rapidly installed to allow data collection with a crystal-to-detector distance of 590 mm. Typically, 18 films were recorded for each crystal that correspond to a total oscillation of 36° along the c-axis with an oscillation step of 3° . Exposure time were setup to 15-20 sec per pass with a total of 12 passes (2/3 fill mode, $I=150\text{ mA}$). Films were then automatically scanned and the resulting files were then transformed to a format suitable for DENZO. On-line partial indexing of the initial images was successful; processing of these data is underway. The expected resolution is 2.6Å and 2.8Å for the native and inhibited form of these crystals, respectively.

A large number of crystals of the homologous AChE-Fas1 complex were also tested; however, no data could be collected on these crystals, which all appeared to be twinned or of a poor diffraction quality (maximal resolution achieved was about 5Å).

In addition, crystals of Civ1, a protein kinase for which beamtime has been allocated on the non-operational beamline EH4 (LS-897), were tested using the Mar CCD. These crystals diffracted weakly up to 3.5Å ; benefits of these preliminary data will be used to prepare new crystals for project LS-897.

Finally, we tested a few crystals of a new project for which beamtime will be requested starting March 1.

Full-time assistance from Wim Burmeister is much appreciated.