

**Experiment title:**

Structural Investigation of imidazole glycerol phosphate dehydratase

Experiment**number:**

LS 1875

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| Beamline: ID 14.4 | Date of experiment: from: 25 April 2001 to: 26 April 2001 | Date of report: 30-8-01 <i>Received at ESRF:</i> |
| Shifts: 3 | Local contact(s): Dr. Hassan BELRHALI | |

Names and affiliations of applicants (* indicates experimentalists):

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Report:

Preliminary report on LS1875, ID 14.4.

Three shifts were allocated on station ID14.4, primarily to collect selenomethionine multiple wavelength anomalous dispersion data on crystals of imidazole glycerol phosphate dehydratase (IGPD). We had previously shown that our crystals of this enzyme were perfectly twinned (see report LS1745) and, despite checking scores of crystals on our home detector, had been unable in the intervening time to grow any crystals that were not twinned. Crystals of six other proteins were thus taken to the ESRF to use this beam time. Each of these six proteins had an unknown structure, and had been overexpressed in the presence of selenomethionine. Crystals of each were grown which showed good diffraction at Sheffield and were thus transported to the ESRF at 100K. Full MAD data sets were collected at three wavelengths for each crystal,

corresponding to the peak, inflection and high energy remote Selenium positions, as determined from a fluorescence scan of each crystal.

Of these six different proteins, the structures of four have been determined using either SOLVE, SHAKE'n'BAKE or SHELXC and MLPHARE and all four are now fully refined. Manuscripts are in preparation for each.

Of the two remaining proteins, selenium substructures have not yet been determined. For these two proteins, analysis of the data in XPREP suggests that the anomalous scattering signal is poor. As we have successfully solved the structures of four other proteins on this trip, it is unlikely that there were beam problems and we have thus concluded that the levels of selenium incorporation for these two proteins were low. Experiments are in progress to redo the selenium overexpression and to check incorporation levels using mass spectrometry.

A full report on this beam time, detailing the structures of these proteins will be added later.