Experiment 01-02 289, february, 14.-15. 2001

Investigation of malat dehydrogenases (MDH) from thermophilic bacteria

A total of 3 full datasets were collected during the 3 shifts allocated for this experiment. These were of two different single-site mutants of the MDH-gene from the green gliding thermophilic bacteria *Chloroflexus Aurantiacus*. The mutants are E165Q (Where glutamic acid 165 is replaced by glutamine) and T187C (where tyrosine 187 is replaced by cysteine)

The dataset of E165Q has been processed and the final refinement of the structure will be performed in due course. The data is of average quality, with a maximum resolution of 2.5Å. The structure will be published in a paper describing the cloning, experssion, purifiaction and thermal analysis of several different MDH-mutants. The manuscript is in preparation. Analysis of the structure has given new insights into the thermostabilic properties of MDH from this thermophilic bacteria.



Figure: Overall arrangement of monomeric subunits in the bioactive tetramer.

As far as the other mutant is concerned, two datasets from two individual T187C-crystals were collected. Both datasets were of average quality with maximum resolution ca. 2.5 - 2.0 Å. Unfortunately, both datasets proved to be impossible to process with the current processing software. We have tried various aproaches with the intergration software – without any success. We now believe that the crystals are twinned with only partially overlapping Bragg lattices. All present protein X-ray integration software lack the possibility to handle such data. Attempts to crystallize this very interesting mutant in another space group is in progress. In fact, we believe to have managed this and new data will soon be collected.