



ESRF

Experiment title:

Protein Crystallographic Studies of Flagellar HAP2
And F41 Fragment of Flagellin

Experiment

number:

LS-1327

Beamline:

ID14-3

Date of experiment:

from: April 16, 1999 to: April 18, 1999

Date of report:

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Shifts:

6

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Report:

We came with samples of 1) flagellar HAP2 crystals and 2) flagellin fragment F41 crystals containing heavy atom. The 6 shifts were to collect native data from the HAP2 crystals and derivative data from the F41 crystals.

1- HAP2 was crystallized in two crystal forms: C2 ($a=225 \text{ \AA}$, $b=157 \text{ \AA}$, $c=212 \text{ \AA}$, $\beta=102^\circ$) and hexagonal ($a=125 \text{ \AA}$, $c=271 \text{ \AA}$). The crystals were obtained by the hanging drop vapor diffusion method. Both crystals diffracted around 3.5 \AA by several hours of exposure on a conventional x-ray source and to 2.9 \AA by 45 sec of exposure on ID14-3. Data collected on ID14-3 showed that the crystals had some distortion due to freezing. We could overcome the distortion problem by annealing the crystals for a very short time. Unfortunately HAP2 crystals showed some stacking problems and it was not possible to collect useful data set from the HAP2 crystals.

2- The F41 fragment of flagellin was crystallized by the hanging drop vapor diffusion method. Native data sets were collected at ESRF on beam line ID14-3 in April 1998. The space group of the crystal is P21 ($a=52 \text{ \AA}$, $b=37 \text{ \AA}$, $c=120 \text{ \AA}$, $\beta=91.6^\circ$). This time we collected heavy atom derivative data. Derivative crystals were obtained by soaking native crystals in heavy atom containing solution. Two types of heavy atoms reagents were used: mercury (HgCl_2 , PHMBS) and lead (PbAc). In the case of the mercury compounds different soaking time were applied. During these 6 shifts we were able to collect a full data set from a derivative crystal containing PbAc and five data sets from derivative crystals containing HgCl_2 . Two of the five data sets from the mercury containing crystals were not processed because of high mosaicity. Other data sets were processed to 2.0 \AA resolution with a completeness of 99% and a Rmerge of 4%. The isomorphism between native and derivative crystals was very good. Further analysis of these data and comparison with the native data collected last year at ESRF are underway.