

**Experiment title:**

Structural Analysis of the Dynamin GTPase

Experiment number:

LS-1615

Beamline:

ID13-3

Date of experiment:

from: 24.02.00 to: 25.02.00

Date of report:

29.02.00

Shifts:

3

Local contact(s):

David Flot

*Received at ESRF:***- 3 MAR. 2000****Names and affiliations of applicants (* indicates experimentalists):****F. Jon Kull*, MPI Heidleberg****Hartmut Niemann*, MPI Heidelberg****Report:**

The three shifts reported about here are the first half of 6 shifts allocated for investigating crystals of dynamin A from *Dictyostelium discoideum*. During test beam time (TC-59) we had diffraction to about 10Å from needle shaped crystals of dynamin A.

Despite considerable effort over the past 5 months, the needle shaped crystals could not be markedly improved. Their current size is up to over 1 mm long, 60 µm wide, but no more than 15 µm thick. The results obtained during the test time could be reproduced with spots going out to 10Å. However, there are now indications that the diffraction pattern is that of a helical fiber. This result complements the observation by EM that the protein forms filamentous spirals. A more thorough analysis of the data is on the way and will be summarized in the final report after the next three shifts in June.

Because improvement of the needle shaped crystals turned out to be almost impossible, we searched for alternative crystal forms under different crystallization conditions. Using different precipitants we obtained crystals that were nicely extended into all three dimensions (up to 100 x 100 x 200 µm). Crystals of two more crystal forms of dynamin A were screened during the beam time, but only very limited or no diffraction at all was observed. All in all 18 crystals of dynamin A were tested during these three shifts.

Meanwhile, we have obtained crystals of mammalian dynamin. These crystals are small hexagons (40 x 40 x 20 µm). They had been tested on ID 14-3 before, but no diffraction was observed. We tested 15 of these crystals during our beamtime on ID 13. All of the crystals diffracted. Three crystals diffracted so well, that we collected data until they died due to radiation damage. The diffraction limit is 3.8Å. The mosaicity is very low. We determined the unit cell (a=147 b=147 c=161 α=90 β=90 γ=120) and the space group to be P622. By combining the data collected on the three good crystals we hope to be able to obtain a complete medium-resolution data set. By comparing the diffraction of crystals frozen with different cryo-protectants, we could determine optimal cryo conditions.