



	<b>Experiment title:</b> NS3 full-length protein	<b>Experiment number:</b> LS-1517
<b>Beamline:</b> ID14-EH4	<b>Date of experiment:</b> from:25.09.99 to: 26.09.99	<b>Date of report:</b> 29.02.00
<b>Shifts:</b> 3	<b>Local contact(s):</b> E.Mitchell	<i>Received at ESRF:</i>
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**Report:***Aims:*

1. Carry out a MAD experiment using selenomethionine labelled protein
2. Characterise new crystal form
3. Screen for mercury derivatives optimising anomalous signal from Hg
4. Screen for crystals with low or no twinning.

**Results:**

- 1.) Selenomethionine labelled NS3 full-length has been crystallized. A MAD experiment was planned on ID14 EH4. No fluorescence signal (for selenium) was detected from the selenomethionine labelled crystals, which can be attributed to the very small size of the crystals. Data were collected above the selenium absorption edge from 3-4 different crystals and the data has been analysed. No significant signal was detected and thus no reasonable solution for the determination of the selenium sub structure of the protein has been obtained.

- 2.) Crystals of a different morphology of NS3 full length have been grown. These crystals were characterised and found to be tetragonal with unit cell dimensions of  $a = b = 78 \text{ \AA}$   $c = 138 \text{ \AA}$ . The crystals diffract beyond  $2.5 \text{ \AA}$  resolution but were found to be sensitive to radiation damage and degraded rapidly. A  $3 \text{ \AA}$  data set was collected, although the data were of poor quality due to the bad quality of crystals. Molecular replacement trials using these data have been carried out but were unsuccessful. This was attributed to the poor quality and lack of good data at low resolution. Conditions for better quality crystals are being searched.
- 3.) Two data sets were collected from crystals soaked (using different soaking times) with mercury acetate . The data were collected at  $12.45 \text{ keV}$  to optimise the anomalous scattering from mercury. These data are currently being analysed; although these crystals show significant twinning fractions. Results of data analysis suggest no mercury was bound.
- 4.) Additional data from the cubic crystal form of the NS3 full-length were collected with the aim of finding a crystal with little or no twinning. Three redundant data sets were collected but large twinning fractions (up to  $0.37$ ) were observed.