



	Experiment title: Structure determination of the glycosyltransferase amylosucrase using multiple anomalous dispersion techniques.	Experiment number: LS-1450
Beamline: BM-14	Date of experiment: From: 26 September 99 to: 27 September 99	Date of report: 29 February 2000
Shifts: 3	Local contact(s): Dr. Vivian STOJANOFF	<i>Received at ESRF:</i>
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Report:

Our MAD experiment on selenomethionine substituted amylosucrase was very successful. We managed to obtain 3 data set to 3.0 Å resolution at $\lambda = 0.9790$, $\lambda = 0.9791$ and $\lambda = 0.8850$ Å. The data collection was very time consuming and more data could have been obtained given more beam time (we had 3 shifts). The three data sets were processed and all 15 selenium positions were found with the program SOLVE. The resulting electron density map could subsequently be interpreted and about 80 % of the structure traced. A major improvement was phase extension to 1.6 Å and via a native data set. This electron density map was completely auto-traced using the WarpArp program and further fitting has given a model with 630 amino acid residues, one tris molecule and ca. 600 water molecule. The current R_{free} is 21.5 %.

A preliminary crystallization paper has been published where the data collection of the native data set is described.¹

1. *Acta Cryst.* (2000). D56, 203-205.

Crystallization and preliminary X-ray studies of recombinant amylosucrase from *Neisseria polysaccharea*.

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Abstract:

Recombinant amylosucrase from *Neisseria polysaccharea* was crystallized by the vapour-diffusion procedure in the presence of polyethylene glycol 6000. The crystals belong to the orthorhombic space group *P21212*, with unit-cell parameters $a = 95.7$, $b = 117.2$, $c = 62.1$ Å, and diffract to 1.6 Å resolution. A *p*-chloromercuribenzenesulfonate (pcmbs) derivative has been identified and a selenomethionine-substituted protein has been produced and crystallized.