X	**Experiment title:** Squid neuronal regulatory protein Sec1	**Experiment number:** LS1535
Beamline: various	**Date of experiment:** from: various to:	**Date of report:** 1/3/00
Shifts:	**Local contact(s):** Gordon Leonard, Marc van Raij, Anastassis Perrakis	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

Winfried Weissenhorn*, Andreas Bracher*, EMBL

Report:

Background:

Sec1 protein family members are involved in the regulation of all intracellular SNARE-mediated (soluble N-ethylmaleimide sensitive fusion protein attachment protein receptor) vesicle fusion processes in a step preceding membrane fusion and have been shown to interact with t-SNAREs. To better understand the structural basis and the role of Sec1 in the regulation of the SNARE complex formation neuronal Sec1 from squid *Loligo pealei* has been expressed and crystallized; this invertebrate protein shows a high sequence homology to the human neuronal Sec1, Munc18a. Here the production of diffraction quality native crystals, which belong to space group $P3_121$ and diffract to 3.3 Å resolution is described. In addition, selenomethionyl n-Sec1 crystals in space groups $P3_121$ and $P2_1$ have been generated. The crystals belonging to space group $P2_1$ space group diffracted to 2.3 Å.

(i) A sec1 native data set was collected at the microfocus beamline ID13. Crystals belonging to space group $P3_121$ (or$P3_221$) diffracted to 3.3 Å resolution.
(ii) A MAD experiment on selenomethionine derivatized crystals of Sec1 was performed on BM14; three datasets around the selenium absorption edge were collected with diffraction to 3.5 Å. Although a clear signal for Se was observed in the fluorescence scan, the difference pattersons revealed no major peaks.
(ii) New selenomethionine derivatized protein reulted in a new crystal form belonging to space group $P2_1$. A MAD experiment was performed at BM14, two data sets were collected

at the peak wavelength and at a remote wavelenght. Diffraction data to 3.2 Å was collected. These datasets were used to solve the structure.

(iv) A high resolution "native"dataset was collected at EH4. Crystals diffracted to 2.3 Å.