

Experiment title: Structural studies of neural Hb-like hemoprotein from Cerebratulus lacteus Experiment number: LS1803

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A hemoprotein very loosely related to Hb occurs in circulating red blood cells, neural tissue and body wall muscle tissue of the nemertean worm *Cerebratulus lacteus*. The proteins isolated from neural and body wall muscle are composed of 109 residues only, which makes them the smallest naturally occurring Hb-related proteins known. This finding should make the *C.lacteus* Hb particularly valuable for studies of folding and stability, besides shedding light on the recently discovered role of "neural hemoglobins".

Cerebratulus Hb has a relatively high oxygen affinity and is present at high concentration within the neural tissue. This suggests that the Hb also may function as oxygen store when the animal encounters anoxic conditions during burrowing. However, other functions *in vivo* have also been considered.

Multiple sequence alignment with 'truncated Hbs' from the unicellular green alga *Chlamydomonas eugamentos* and the protist *Paramecium caudatum* (121 and 116 amino acid residues per chain, respectively), does not show sufficient similarities to suggest neatly comparable 3D structures.

Crystals of neural *C.lacteus* Hb belong to the tetragonal space group P4₂ and diffract up to 2.0 Å resolution in house (cell constants: a = b = 42.3 Å, c = 60.1 Å, one molecule per asymmetric unit).

In this experiment were collected a high resolution native data set of *C.lacteus* Hb (1.23 Å resolution) and a possible heavy atom derivative (see Table for the data collection statistics). Inspection of Patterson maps didn't reveal the presence of heavy atoms bound to the protein. One problem was that this derivative was not isomorphous to the native data set. In fact, the space group changed from tetragonal to orthorhombic.

The heavy atom derivative search is still in progress, as well as a MAD experiment on the heme Fe-atom absorption edge is planned.

Table: Summary of *C.lacteus* Hb data collection statistics:

	Native	Mercuryl derivative
Space Group	P4 ₂	P222 ₁
Unit cell (Å)	a=b=43.2, c=60.4	a=42.0, b=43.3, c=61.2
Resolution (Å)	35-1.23	30-2.5
Completeness (%)	97	78
Rsym (%)	11.2	5.7
Mosaicity (°)	0.6	0.6

