



	Experiment title: Structural studies of poplar plastocyanin I21C E25C mutant	Experiment number: LS1803
Beamline: ID14-1	Date of experiment: from 23-11-2000 to 25-11-2000	Date of report: 14-06-01
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Plastocyanin (PC) is a key copper-protein involved in electron transfer from photosystem I to photosystem II. PC consists of 99 amino acids and includes one type I copper atom, coordinated by His87 N δ , Cys84 S γ and Met92 S δ atoms.

With the aim of studying PC electron transfer properties at the single molecule level using Scanning Tunneling Microscopy (STM), we engineered within PC a surface disulfide bridge, by site-directed mutagenesis of residues Ile21 and Glu25. The disulfide bond, located in the “southern pole” of PC opposed to the Cu-I site, located in the “northern pole”, will provide an anchoring group for chemisorption onto gold substrate, upon reduction.

The Ile21Cys, Glu25Cys PC mutant (MutPI) was characterised by different spectroscopic techniques: cyclic voltammetry, which suggested that no substantial change in the copper site geometry had occurred following engineering of the disulfide bridge. Moreover, preliminary STM investigations indicate that MutPI adsorbs onto Au substrates via the disulfide bridge, retaining the redox properties of wild type (w.t.) PC.

In this experiment I collected a high resolution data set of MutPI (1.6 Å resolution) (see Table for data collection statistics).

Table: Summary of MutPI data collection:

Space Group	P2 ₁ 2 ₁ 2 ₁
Unit cell (Å)	$a = 47.38, b = 49.31, c = 76.58$
Resolution (Å)	35-1.6
Completeness (%)	98.1
R _{sym} (%)	4.5
Mosaicity (°)	0.44

