



ESRF

Experiment title: Investigation of photoinduced structural changes with ns time resolution in protein crystals.

Experiment number:
LS 666

Beamline:

ID9

Date of experiment:

from: 14 June 1997

to: 19 June 1997

Date of report:

28 August 1997

Shifts:

6 sb+32/3fill

+6 16bunch

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Report:

In the June 1997 experiment LS666 we continued to collect high quality ns time-resolved Laue crystallographic data on the photocycle of photoactive yellow protein. We used 480 nm light from a dye laser pumped by the third harmonic of a ns Nd:YAG laser. According to experiments conducted prior to the run, this will improve the fraction of photoactivated molecules in the crystal over earlier studies (LS540, November 1996). For these studies we used wiggler W70 and undulator U46 in series in single bunch mode and wiggler W70 alone in hybrid and 16 bunch mode. The photocycle of PYP is a reversible process and therefore, in principle, allows signal averaging by repeated crystal photolysis where the repeated X-ray exposures are accumulated on the CCD detector prior to read-out. This allows a relatively rapid data collection (about 1h per data set of 50 images). In Nov 96 we recorded 10 images at each angular setting with a 4° step between them. This strategy acquired highly redundant data sets in about 1 hour so we were inclined to employ the same strategy in June 97. We collected data at laser / X-ray delays identical to those studied in November: 5ns, 10ns, 60ns, 420ns, **1.5µs, 8µs, 50µs** and 1ms and added several additional time points: 25ns, 250ns, 500ns, 750ns, **1.8µs, 200µs, 300µs**, 10ms, 50ms, 100ms, 500ms. Data processing for the June 97 data and structural refinement of results from the November 96 experiment are in progress, and clearly reveal structural changes at the chromophore and in the surrounding protein associated with various steps of the complex photocycle of PYP.