Data Collection of Phenylethylamine Oxidase from *Arthrobacter globiformis* at Low Temperature

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Introduction

Phenylethylamine oxidase from *Arthrobacter globiformis* (AGAO) catalyzes the two electron oxidative deamination of primary amine. The holoenzyme of AGAO has a topaquinone (TPQ) cofactor formed from the apoenzyme by the post-translational modification of the tyrosine residue in the presence of Cu^{2+}. We have already determined the structures of both the holo and the apo forms at 2.2Å resolution, and proposed the mechanism of biogenesis of TPQ based on the differences between both the structures[1]. In order to detect the intermediate structures, we carried out cryocrystallography.

Experiment

Crystals of AGAO were obtained by the hanging-drop vapor diffusion method by using 1.1M potassium and sodium tartrate as a precipitant and 25mM HEPES buffer, pH6.8. The crystals are plate shape, and belong to the space group $C2$ with cell dimensions of a=158.7 Å, b=64.6 Å, c=93.3 Å and β=112.3°.

In order to collect the standard data sets at the low temperature for the analysis of the structure changes, we tried to collect diffraction data from the apo-form crystals at liquid nitrogen temperature. The crystals were transferred to the solution which contained 28% treharose as a cryoprotectant. The data collection at SPring8 was done with a screenless Weissenberg camera equipped with a cylindrical cassette with a 560 mm radius installed in the BL41XU station. An Oxford cryosystem was used and controlled at -150°C. The crystals were mounted by using cryoloops in the cryostream. The crystals showed anisotropic diffraction patterns at low temperature. We could record diffraction patterns of apo-form crystals only over angles of 120°. The wavelength of 0.71 Å was used. The oscillation range of each frame was 3.0°. The diffraction patterns were recorded on 40cm × 80cm Fuji Film imaging plates and were digitized on a Rigaku imaging plates photo reader system. The crystal diffracted at least up to 2.2 Å resolution. Data reduction is in progress.

Reference

1) Wilce MC, *et al. Biochemistry*, (1997), **36**, 16116-16133.