

Crystal Structure Analysis of Valyl-tRNA Synthetase in a complex with tRNA^{Val}

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T. thermophilus valyl-tRNA synthetase complexed with tRNA

Valyl-tRNA synthetase (ValRS) specifically attaches L-valine to tRNA^{Val}. ValRS misacylates minimally-distinct L-Thr, but hydrolytically edits its own misproducts upon binding with tRNA^{Val}. To elucidate the mechanism of editing activation by tRNA, we have crystallized ternary complex of *T. thermophilus* ValRS, tRNA^{Val}, and Val-AMP analogue. The crystals belong to spacegroup $P4_22_12$ with extremely-large cell constants of $a=b=410\text{\AA}$ and $c=82.8\text{\AA}$. In the previous beam time (Dec., 1997), we collected the diffraction data upto 2.8\AA resolution for 180° with oscillation angle of 4° ($\lambda=1.0\text{\AA}$, camera distance=500 mm, and Temp.=100K). However, the oscillation angle was so large that we could not process the data. Therefore, in this beam time, we collected the diffraction data with oscillation angle of 0.5° (camera distance=400 mm at Raxis-IV) (Fig. 1).

By the use of shorter wave length ($\lambda=0.7\text{\AA}$) and data collection at lower temperature (100 K), we could significantly reduce the X-ray damage. To successfully process diffraction data of crystals with such extremely-large unit cells, low mosaicity is critical. Since the mosaicity of our crystals largely depend on crystals, it takes much time to select the crystals. Within the beam time, we collected 120 frames (60°) with exposure time of 4 min. The highest resolution of the diffraction data was 2.5\AA . We could process the native data up to 2.8\AA resolution with completeness of 95.1% and R_{merge} of 10.2%.

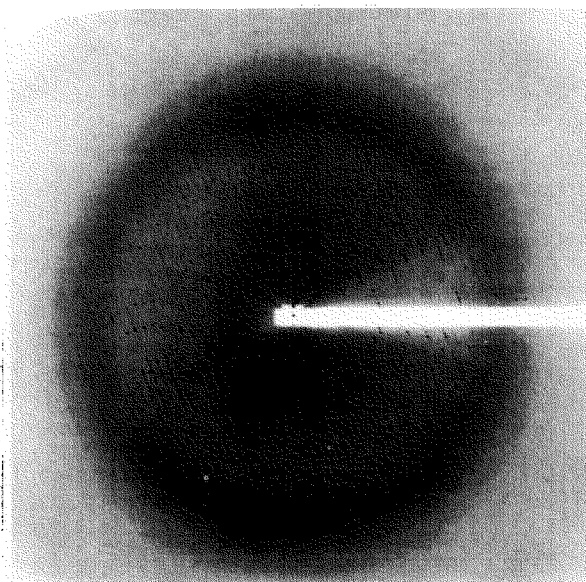


Fig. 1 Native data of *T. thermophilus* valyl-tRNA synthetase complexed with tRNA and Val-AMP analogue