Structure of Oxa-1, a Class D Beta-Lactamase

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Introduction: Bacterial beta-lactamases produce resistance to beta-lactam antibiotics (penicillins and cephalosporins) by hydrolyzing the drugs to inactive acids. The serine-reactive enzymes fall into 4 structural classes, the least studied of which is class D. The so-called Oxa-1 enzyme is a prototypical class D beta-lactamase that, in native and now 10-12 mutant forms, provides resistance to oxacillin in clinical strains of E. coli.

Results: The 28kDa enzyme crystallizes from PEG 8000 in P1 with 2 molecules and cell dimensions 36x51x74 Å and alpha = 110, beta = 84, gamma = 98. A UO2 derivative was found, and MAD data were collected to 2.3 Å resolution at X4A at 3 wavelengths near the uranium edge. Data were processed with DENZO. For the 3 data sets, the average I/signal is only 11-13, possibly because the beam intensity at X4A was weak at the monochromator setting needed to reach the LIII uranium edge near 17.2 keV.

Conclusions: A self-rotation Patterson search indicated the presence of 2-fold non-crystallographic symmetry. SOLVE was used to confirm the uranium sites found earlier in the Patterson map and to locate another minor site. The SOLVE solutions have low Z scores in the range 0.5 to 6.0, and the maps at 2.6 Å have so far been uninterpretable, even after density modification. Phasing with MLPHARE has produced similar results. It may be necessary to collect stronger data from larger crystals.