Crystal Structure of an Archaeal Class I Aldolase

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Fructose 1,6-bisphosphate aldolase (FBPA) catalyses the reversible cleavage of fructose 1,6-bisphosphate to glyceraldehyde 3-phosphate and dihydroxyacetone phosphate in the glycolytic pathway. FBPAs from archaeal organisms have recently been identified and characterized as a divergent family of proteins (1). We have crystallised the first archaeal FBPA and collected X-ray diffraction data at the EMBL beam lines BW7B and X13 to a resolution of 1.9 Å and 2.4 Å. Self rotation functions revealed two pentamers in the asymmetric unit corresponding to 280 kDa of protein. Se-Met substituted protein was expressed, crystallised and SAD data collected at the EMBL beam line BW7A (2). Forty selenium atoms were located from the anomalous signal alone, providing initial phases that were further improved using ten fold non-crystallographic symmetry averaging in combination with solvent flattening yielding an interpretable electron density map. Additionally, a crystal structure of the archaeal FBPA covalently bound to the substrate dihydroxyacetone phosphate was determined at 2.1 Å resolution. This structure allows us to do a detailed comparison of the archaeal FBPA with the eukaryotic counterparts to establish their evolutionary relationship.
Table: Summary of the data collection statistics for the anomalous diffraction and the native data sets for Tt-FBPA

<table>
<thead>
<tr>
<th></th>
<th>Apo (P2₁)</th>
<th>Apo (P2₁2₁2₁)</th>
<th>Se-peak (P2₁)</th>
<th>Holo (P2₁)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wavelength (Å)</td>
<td>0.967/0.803 c</td>
<td>0.967</td>
<td>0.97642</td>
<td>0.8453</td>
</tr>
<tr>
<td>Resolution (Å)</td>
<td>40-1.90</td>
<td>20-2.39</td>
<td>20-2.27</td>
<td>40-2.10</td>
</tr>
<tr>
<td>Cell dimensions</td>
<td>a = 83.3 Å</td>
<td>b = 159.0 Å</td>
<td>c = 103.0 Å</td>
<td>a = 89.9 Å</td>
</tr>
<tr>
<td></td>
<td>β = 108.1 °</td>
<td>β = 108.1 °</td>
<td>β = 108.2 °</td>
<td></td>
</tr>
<tr>
<td>Unique reflections</td>
<td>188245</td>
<td>109747</td>
<td>114381</td>
<td>128643</td>
</tr>
<tr>
<td>Redundancy</td>
<td>4.0</td>
<td>5.4</td>
<td>4.8</td>
<td>3.6</td>
</tr>
<tr>
<td>I/σ(I) a</td>
<td>14.5 (2.4)</td>
<td>13.3 (2.7)</td>
<td>22.2 (4.1)</td>
<td>13.6 (2.3)</td>
</tr>
<tr>
<td>R_sym a, b</td>
<td>0.085 (0.365)</td>
<td>0.121 (0.372)</td>
<td>0.061 (0.301)</td>
<td>0.071 (0.272)</td>
</tr>
<tr>
<td>Completeness a</td>
<td>0.945 (0.870)</td>
<td>0.936 (0.690)</td>
<td>0.995 (0.988)</td>
<td>0.902 (0.593)</td>
</tr>
</tbody>
</table>

a The numbers in parentheses correspond to the last resolution shell.

b $R_{sym} = \frac{\sum |I - \langle I \rangle|}{\sum I}.$

c Native data were collected from two crystals on beamlines BW7B and X13.
