Cloning, Expression, Purification, Crystallization and Preliminary X-ray Crystallographic Study of the Putative SAICAR Synthetase (*PH*0239) from *Pyrococcus horikoshii* OT3

Jeyakanthan Jeyaraman (桀肯特)

National Synchrotron Radiation Research Center, Hsinchu, Taiwan

The study of proteins involved in the *de novo* biosynthesis of purine nucleotides is central in the development of antibiotics and anticancer drugs. In view of this, a protein from hyperthermophile *Pyrococcus horikoshii* OT3 was isolated, purified and crystallized using microbatch method. The primary structure is found to be similar to that of SAICAR synthetase which catalyses the seventh step of *de novo* purine biosynthesis. Diffraction quality crystal is obtained with Hampton crystal screen II 34 consisting of 0.05 M cadmium sulfate

hydrate, 0.1 M HEPES buffer with pH 7.5 and 1.0 M sodium acetate trihydrate and 40% v/v 1,4-butanediol as an additive. Crystal belongs to the space group $P3_1$ with unit cell parameters a = b = 95.62, c = 149.13 Å. Assuming a hexamer in the asymmetric unit results in a Matthews coefficient (V_M) of 2.3 Å 3 Da $^{-1}$ corresponding to a solvent content of about 46%. A detailed study of this protein could yield meaningful insights regarding the structural stability at high temperatures.