



	Experiment title: BAG Barcelona – Carbamoyl Phosphate from <i>P. furiosus</i>	Experiment number: LS-1377/78
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Report:

Two types of enzymes, carbamate kinase (CK) and carbamoyl-phosphate synthetase (CPS) are known to synthesize carbamoyl phosphate (CP) from mixtures of ATP, bicarbonate and ammonia. However, the reaction catalyzed by CPS differs from that catalyzed by CK not only in its irreversibility in the direction of CP synthesis, but also in the use of bicarbonate and ammonia as the true substrates and in the utilization of two ATP molecules per molecule made of CPS. In general, CK and CPS appear to be unrelated molecules: CK is a homodimer of a polypeptide of approximately 33 kDa, whereas CPS is a 120-kDa polypeptide that is either associated or fused to another polypeptide of approximately 40 kDa. Alignment of the amino acids sequences of CK and CPS failed to reveal the existence of a statistically significant sequence identity.

Given the important differences between between CPS and CK, the recent description of CK-like CPS in the hyperthermophilic archea *Pyrococcus furiosus* and *Pyrococcus abyssi* was most puzzling.

We obtained crystals from CPS from *Pyrococcus furiosus* isolated (78.6*78.6*278.0 Å, space group P6₁22) and cocrystalized with AMPPNP (60.0, 71.9 107.4 Å space group C222₁), that both diffracted till resolutions close to 1.0 Å at the ESRF.

Complete data sets, at 1.4 Å resolution, were collected and are now been used in the refinement and analysis of this CPS with a CK-like structure.