

# Biochemical and structural investigation of a new prokaryotic specific GTP cyclohydrolase I enzyme family

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GTP cyclohydrolase I (GCYH-I) catalyzes the first step in folic acid biosynthesis in bacteria and plants, biopterin biosynthesis in mammals, and the biosynthesis of 7-deazaguanosine modified tRNA nucleosides in bacteria and archaea. The reaction is an especially complex transformation involving the hydrolysis and deformylation of GTP at C8, ring opening and isomerization of the ribosyl moiety, and cyclization to generate the pterin ring system. The mechanistic complexity of the reaction and the fact that enzymes from diverse organisms (e.g. humans and *E. coli*) are homologous suggested that this enzymatic activity was unique to a single homologous enzyme family. However, we have recently discovered a new prokaryotic specific GCYH-I enzyme (denoted GCYH-IB) that is distinct from the canonical GCYH-I. Analysis of the distribution of GCYH-IA (the new designation for the canonical GCYH-I enzyme) and GCYH-IB among all sequenced genomes revealed a great diversity in the distribution of these enzymes; while most contain either the IA or IB enzymes, some contain both, and a small number possess neither. Notably, the newly discovered enzyme is found in a number of human pathogens (e.g. *Staphylococcus* and *Neisseria*). While both enzyme families belong to the tunneling-fold (T-fold) superfamily, we report here studies that demonstrate significant biochemical and structural differences between the type IA and IB enzymes, including metal-dependence, active-site architecture, and catalytic strategies.

