

Characterization of the methane monooxygenase (MMO) regulatory proteins, mmoS and mmoQ, from *Methylococcus capsulatus* (Bath)

Uchechi Ukaegbu, Shannon Henery, Amy Rosenzweig
Department of Biochemistry and Molecular Cell Biology and Chemistry,
Northwestern University, Evanston, IL 60208

Methanotrophs are gram-negative eubacteria that use methane as their only source of carbon and energy. The first and key enzyme in methane metabolism, methane monooxygenase (MMO), catalyzes the oxidation of methane to methanol. Several strains of methanotrophs, including *Methylococcus capsulatus* (Bath), express a membrane-bound or particulate MMO (pMMO) at high copper levels in the growth medium and a soluble form, sMMO, at low copper-to-biomass ratios.

The mechanism by which the cell senses copper and controls the expression of both MMOs is not known. Four genes located downstream of the sMMO gene cluster; *mmoS*, *mmoQ*, *mmoR* and *mmoG* have recently been identified. These four gene products are proposed to be involved in sMMO regulation. In particular, the copper sensing mechanism might involve mmoS and mmoQ. These proteins are ~ 50% identical to two-component sensor-regulator systems such as NifL and NifA that regulate nitrogenase expression and CheA and CheY of the chemotaxis system. We have cloned, expressed and purified both MmoQ (69.8kDa) and MmoS (128.6kDa) from *M. capsulatus* (Bath). We have analyzed their oligomerization states, and we have determined that MmoS is a flavoprotein that binds FAD. Additional biochemical and structural studies are focused on understanding the roles of these proteins in MMO regulation.