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Structure and Function of Adenylate Forming Enzymes: A ~140° C-terminus Domain Rotation. Albert S. Reger, Jill M. Carney, Andrew M. Gulick, SUNY, Dept. of Structural Biology, Hauptman-Woodward Medical Inst., Buffalo, NY 14203-1102.

The adenylate-forming enzymes, including Acyl-CoA Synthetases and the adenylation domains of Non-Ribosomal Peptide Synthetases, perform two half-reactions in a ping-pong mechanism. We have proposed that upon completion of the initial adenylation reaction, these enzymes use a 140° C-terminus domain rotation to perform the second thioester forming half reaction. Structural and kinetic data of mutant enzymes support this hypothesis. Mutations were made to Acetyl-CoA Synthetase that specifically affect the individual half-reactions. Substitution of K609A results in an enzyme unable to catalyze the adenylate reaction, while G524L is unable to catalyze the thioester-forming half reaction. Kinetic data including hinge mutants D517P and D517G and structures of the mutants R194A, R584A, R584E, and K609A will be presented in support of the stated hypothesis.

