

# Crystallographic analyses of type III polyketide synthase reaction intermediates

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Type III polyketide synthases are structurally simple yet biochemically complex family of homodimeric proteins involved in the biosynthesis of a wide variety of metabolites in plants, bacteria and fungi [1]. Each monomeric subunit iteratively condenses malonyl-Coenzyme A onto a Coenzyme A-tethered starting substrate. Despite progress in the structural characterization of several type III PKSs, direct visualizations of reaction intermediates in crystals has been elusive.

Here we present a study of the combined use of crystallography and gene mutations to trap natural and near-natural intermediates in biphenyl synthase, a benzoic acid-specific type III PKS. This research builds on our previous work in which we characterized the apo form of biphenyl synthase [2]. Here we present crystallographic structures that approximate several acyl-enzyme intermediates thought to occur during biphenyl synthase catalysis. These structures give us a window to understand the role of ordered solvent and keto-enol tautomerization in stabilizing polyketide reaction intermediates. Ultimately, we anticipate that these crystal structures will provide insight into fundamental mechanisms of carbon-carbon bond formation in these enzyme systems.

1. M. B. Austin and J. P. Noel, *Nat. Prod. Rep.*, 20, (2003), 79

2. C. Stewart Jr, K. Woods, G. Macias, A. C. Allan, R. P. Hellens, J. P. Noel, *Acta Crystallogr. D Struct. Biol.*, **73**, 1007