# Crystal Structure of a *Thermotoga maritima* S-Adenosylmethionine-dependent Methyltransferase

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## Introduction

The National Institutes of Health NIH Protein Structure Initiative is exploring the range of biologically important protein structures. As a part of the Midwest Center for Structural Genomics, we are determining representative structures of members of evolutionarily conserved sequence families. These structures also furnish insight into the biological functions of these proteins. The *Thermotoga maritima* Tm0872 protein is representative of a large sequence family of putative S-adenosylmethionine-dependent methyltransferases. The determination of the crystal structure of the Tm0872 protein reveals a typical S-adenosylmethionine binding domain and a unique substrate recognition domain.

## Methods and Materials

The *Thermotoga maritima* Tm0872 protein was prepared and crystallized as described previously [1]. X-ray diffraction data were collected at 100K at DND-CAT beamline station 5-ID-B at the APS with a Mar165 charge-coupled device (CCD) detector. Data sets were collected for the seleno-methionine-substituted protein and its complex with added S-adenosyl homocysteine. Complexes of the protein with either endogenous S-adenosyl methionine (SAM) or S-adenosylhomocysteine (SAH) were crystallized in the cubic space group $P2_13$ with a = b = c = 133.4 Å and had two polypeptides each having 299 amino acid residues per asymmetric unit. The structure of the seleno-methionine-substituted protein was determined by using anomalous diffraction data that were collected at three wavelengths near the selenium K edge. The data were integrated and merged with DENZO/SCALEPACK. The structure was refined by using data to 1.9 Å resolution and had R = 19.7 and $R_{free}$ = 21.2 for the SAH complex and R = 20.5 and $R_{free}$ = 22.6 for the SAM complex.

## Results and Discussion

The structure determination revealed that one domain of the protein had a typical S-adenosylmethionine (SAM) binding domain, while the second, primarily helical domain represents the presumed substrate recognition domain. Amino acid residues that are conserved in the sequence family are located primarily in the interface between the two domains, suggesting that this is the location of the active site. This is also indicated by the location of the SAM and SAH molecules. Surprisingly, except for the amino group of the adenine ring, the SAM and SAH are buried within the protein structure. It is clear that binding of the second substrate, allowing transfer of the methyl group, must be accompanied by a conformational change that provides access to the methyl group. A more detailed description of the structures has been presented elsewhere [1].

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## References

[1] D.J. Miller, N. Ouellete, E. Evdokimova, A. Savchenko, A. Edwards, and W.F. Anderson, "Crystal complexes of a predicted SAM dependent methyltransferase reveal a typical SAM domain and a substrate recognition domain," Protein Science **12,** 1432-1442 (2003).