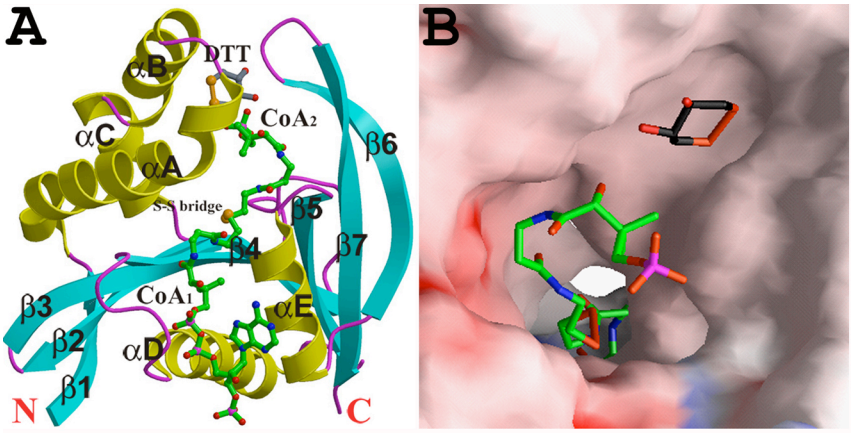
Northeast Structural Genomics Consortium

Structural and Functional Studies of the *Bacillus subtilis* PaiA Protein: NESG Target SR64

PaiA has been implicated in the negative control of sporulation in *B. subtilis*, as well as production of degradative enzymes (Honjo *et al*., 1990 J Bacteriol *172*, 1783). The NESG Consortium has determined the crystal structure of PaiA in complex with CoA at 1.9 Å resolution. The structure reveals that PaiA is a member of the N-acetyltransferase superfamily of enzymes. Unexpectedly, we observed the binding of an oxidized CoA dimer in the active site of PaiA (Fig. 1A). This unanticipated structural information suggests the substrates of the enzyme could be linear, positively charged compounds (Fig. 1B). In a collaboration with Dr. Porter group at the Roswell Park Cancer Institute, Buffalo, we subsequently demonstrated that PaiA possesses N1-acetyltransferase activity toward polyamine substrates including spermidine and spermine. The Porter group also found that conditional overexpression of PaiA in bacteria results in increased acetylation of endogenous spermidine pools.

Thus, our structural and biochemical analyses indicate that PaiA is a novel N-acetyltransferase capable of acetylating both spermidine and spermine. In this way, the pai operon may function in regulating intracellular polyamine concentrations and/or binding capabilities. In addition to preventing toxicity due to polyamine excess, this function may also serve to regulate expression of certain bacterial gene products such as those involved in sporulation.



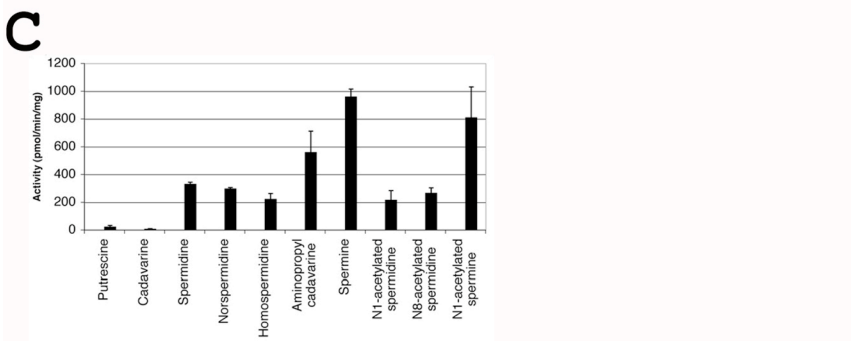


Fig. 1. **(A).** Structure of PaiA in complex with oxidized CoA dimer (in green for carbon atoms) and DTT (in gray). **(B).** Molecular surface of the PaiA molecule, showing that the active site of the enzyme is located at the center of the tunnel and viewed from the CoA_2 molecule. This view of PaiA also suggests that negatively-charged groups of CoA_2 and DTT are incompatible with the predominantly neutral and negatively charged surface of PaiA **(C).** The catalytic activity of PaiA towards a collection of polyamine and diamine substrates.

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Forouhar F, Kuzin A, Seetharaman J, Lee I, Zhou W, Abashidze M, Chen Y, Yong W, Janjua H, Fang Y, Wang D, Cunningham K, Xiao R, Acton TB, Pichersky E, Klessig DF, Porter CW, Montelione GT, Tong L J. Struct. Function. Genomics 2007 8: 37-44. Functional insights from structural genomics.