Comparative characterization of a third isoform of pantothenate kinase identified in pathogenic bacteria: Implications for Coenzyme A biosynthesis and drug design

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Pantothenate kinase catalyzes the first committed step of Coenzyme A (CoA) biosynthesis in all organisms: the phosphorylation of pantothenate (Vitamin B_5) to form 4'-phosphopantothenate. Two isoforms of the enzyme have been characterized. The first is predominantly found in eukaryotic organisms, while the other mainly occurs in prokaryotic systems. However, a search of bacterial genomes fails to locate a gene homologous to either isoform in some organisms, most of which are known pathogens. Since CoA is an essential cofactor, and no intermediate in the biosynthesis of CoA can enter cells from the medium, another protein capable of performing the pantothenate kinase reaction must be encoded by the genomes of these organisms. In this study we set out to clone, express and characterize a previously unidentified protein from *Bacillus subtilis* (and its homologue from *Helicobacter pylori*) that have been implicated in CoA biosynthesis by genetic complementation experiments. Our results show that the protein indeed has pantothenate kinase activity, but exhibits a variety of distinct characteristics which distinguishes it from the known isoforms of the enzyme. Most importantly we show that certain conserved aspartate residues in the enzyme are required for phosphate transfer to occur. This suggests that this protein is a third distinct isoform of pantothenate kinase which is more closely related to enzymes involved in transferring phosphate groups via phosphorelays, such as those found in bacterial two-component systems and certain members of the HAD superfamily of enzymes. This conclusion has important implications for the biosynthesis of CoA in pathogenic bacteria, and provides a new target for the design of pathogen-specific drugs.