E313 A Extracytoplasmic Sigma Factor SigE50 from *Mycobacterium bovis* BCG

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The extracytoplasmic function (ECF) sigma factors constitute a diverse group of alternative sigma factors that have been demonstrated to regulate gene expression in response to environmental conditions in several bacterial species. The ECF sigma factor-regulated gene expression play a important role in bacterial pathogenesis. A gene encoding an ECF sigma factor, *sigE50*, was cloned from *Mycobacterium bovis* BCG. Sequence analysis showed that the SigE50 shared highly conserved domains with the RNA polymerase ECF sigma factors. Expression of the *sigE50* in *Mycobacterium smegmatis* up-regulated the expression of several genes containing *groEL*.

E314 Topography of Interaction of Enzyme IIA^{glc} of the *Escherichia coli* Phosphotransferase System with Lactose Permease

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The unphosphorylated form of enzyme IIA^{glc} of the *Escherichia coli* phophoenolpyruvate:sugar phosphotransferase system inhibits transport catalyzed by lactose permease. Using radioactive enzyme IIA^{glc}, we previously (Seok et al., Proc. Natl. Acad. Sci. USA, 94, 13515-13519, 1997) characterized the area on the cytoplasmic face of lactose permease that interacts with enzyme IIA^{glc} and consensus binding sequences on proteins that interact with enzyme IIA^{glc} were suggested (Sondej et al., Proc. Natl. Acad. Sci. USA, 96, 3525-3530, 1999). The present study characterizes the region of the surface of enzyme IIA^{glc} that interfaces with lactose permease. Sulfosuccinimidyl acetate treatment of enzyme IIA^{glc}, but not lactose permease, substantially reduced the degree of interaction of two proteins. To localize the lysine residue(s) on enzyme IIA^{glc} that, upon acetylation by sulfosuccinimidyl acetate, lead to inactivation of the regulatory interaction, selected lysine residues were mutagenized. Conversion of nine separate lysines to glutamic acid resulted in proteins that were still capable of phosphoryl acceptance from HPr. Except for Lys69, all the modified proteins were as effective as the wild-type enzyme IIA^{glc} in a test for binding to lactose permease. The Lys69 mutant was also defective in phosphoryl transfer to glucose permease. To derive further information concerning the contact surface, some additional selected residues in the vicinity of Lys69 were mutagenized. On the basis of these studies, a model for the region of enzyme IIA^{glc} that interacts with lactose permease is proposed.