

Enzymes I of Parallel Phosphotransferase Systems in *E. coli*

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The sugar transport system discovered by Roseman et al. [1] is commonly referred to as the phosphoenolpyruvate(PEP):sugar phosphotransferase system (PTS). In addition, a parallel pathway in *E. coli* having to do with the regulation of nitrogen metabolism has been identified [2]. The first enzyme of the latter pathway is enzyme I^{Ntr} (EI^{Ntr}).

Our previous studies on the conformational stability and dimerization of phosphotransferase enzyme I from *E. coli* [3-6] can be briefly summarized as follows: The activity of enzyme I (EI), the first protein in the bacterial PEP:sugar phosphotransferase system, is regulated by a monomer-dimer equilibrium where a Mg²⁺-dependent autophosphorylation by PEP requires the homodimer [7]. Using inactive EI(H189A), in which alanine is substituted for the active-site His189, substrate binding effects can be separated from those of phosphorylation [4,5]. Whereas 1 mM PEP (with 2 mM Mg²⁺) strongly promotes dimerization of EI(H189A) at pH 7.5 and 20 °C, 5 mM pyruvate (with 2 mM Mg²⁺) has the opposite effect. A correlation between the coupling of N- and C-terminal domain unfolding, measured by DSC, and the dimerization constant for EI, determined by sedimentation equilibrium, is observed [5,6]. Similar studies are underway to characterize a truncated EI^{Ntr} (with a deletion of the N-terminal 127 amino acids comprising the GTP-binding GAF domain), which is homologous to other enzymes I.

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