Nucleotide sequence of the *phoP* gene encoding PhoP, the response regulator of the phosphate regulon of *Bacillus subtilis*

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We (1) and others (2) have shown that PhoP is necessary for alkaline phosphatase (APase) expression during phosphate starvation of *Bacillus subtilis* cultures. The *phoP* gene encodes a protein (3) that has sequence similar to PhoB of *E. coli* (4), a positive regulator of transcription that binds to the promoter region of PHO regulon genes. PhoP and PhoR of *B. subtilis* and PhoB and PhoR of *E. coli* are members of the family of two component signal transduction regulators which enable procaryotes to respond to environmental signals; *phoP* and *phoB* encode the respective response regulators while the *phoR* genes encode sensor kinases. In general, the sensor kinase is autophosphorylated upon sensing an environmental change, whereupon the kinase phosphorylates its cognate response regulator, often a transcription activator, thereby activating it. An important approach to understanding the interaction between the sensor-kinase and the response regulator involves the analysis of mutations in these genes that affect the response to the environmental signal.

During analysis of *B. subtilis* strains in which mutations linked closely to the *phoP/phoR* operon blocked APase production, we observed several differences between the *phoP* nucleotide sequence which was published (3) and the sequence we obtained. Because our clone had been generated by PCR, it might contain errors produced during the amplification process. Clones from two additional PCR reactions were sequenced and were identical to our original sequence. To determine if the differences in our sequence represented the mutations in the *phoP* gene that were responsible for the APase negative phenotype of strain JH703, we sequenced two clones derived from separate PCR reactions in which the template was chromosomal DNA from strain JH642, the parent strain of JH703. These two *phoP* clones were identical to each other and identical to the sequence from JH703, suggesting that the APase negative phenotype of JH703 was not due to mutations at the *phoP* locus. To determine if the sequence we obtained was peculiar to JH642 and strains constructed from JH642, we cloned and sequenced the *phoP* gene from *B. subtilis* 168, a commonly used laboratory strain and the strain chosen for the European *Bacillus* genome sequencing project. Again the sequence, was identical to the sequence we determined in JH642

and the JH642 derivative, JH703. In each case the entire *phoP* gene was sequenced using synthetic oligonucleotide primers and Sequenase (United States Biochemical Corporation). Table 1 summarizes the differences between the original and our new sequences. While it is possible that the inversions between basepairs 568 and 569 and between basepairs 658 and 659 are sequence reading errors, it seems likely that the sequence changes between basepairs 829 and 834 represent strain differences. Six nucleotides in the original sequence are replaced by three nucleotides in our sequence. As a result, Pro—Thr in the original deduced amino acid sequence was replaced with Asp in our sequence.

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Table 1. Changes in *phoP* sequence

Nucleotide Old position	Sequence New position	Change	Amino Acid Old position	Sequence New position	Change
568	568	C to G	124	124	R to A
569	569	G to C	124	124	R to A
658	658	A to T	154	154	I to Y
659	659	T to A	154	154	I to Y
829		-C	211 to 212	211	PT to D
830		-C			
834		-A			

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