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Locations of the *speA*, *speB*, *speC*, and *metK* Genes on the Physical Map of *Escherichia coli*

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The *speA*, *speB*, *speC*, and *metK* genes have been located on the physical map of *Escherichia coli* (4) (Table 1). The ability of bacteriophages from the Kohara phage library to complement temperature-sensitive mutations in *metK* (8) and to hybridize to the cloned copies of the respective genes (2) was determined (4).

The genetic map locations of these genes at 63.0 to 65.0 min on the *E. coli* chromosome (1) and their relative positions on the physical map (4) were used to identify a group of phages from the Kohara collection which might be expected to carry the appropriate DNA. Appropriate lambda phages were obtained from Y. Kohara, lysates were prepared by growth on strain W3110 (5), and DNA was prepared (5). The DNA was digested with several restriction enzymes and used for Southern blotting (5). The blots were consecutively probed by using DNA from the transcribed sequences of the *metK* (6), *speA* (7), *speB* (9), and *speC* (unpublished results; GenBank accession no. 33766) genes. These lambda phages were also tested for the ability to complement the temperature-sensitive (cold-sensitive) *metK* mutation in strain DM101 (*metK501*) (8).

The DNA sequences (6, 7, 9) and hence restriction maps were available for all four genes. These data were used to define the physical gene locations. The restriction maps were mostly in agreement with the physical map except that the *Eco*RI site 3′ to *metK* was absent at the expected 3101.5-kilobase-pair site on the physical map and the *Hin*dIII and *Pvu*II sites in *speC* at the 3122-kilobase-pair site of the physical map were inverted with respect to each other. Extensive restriction data outside of these gene locations were available (unpublished data) for the Clarke and Carbon collection plasmids pLC2-5 and pLC20-5 (3), from which the cloned copies of the genes (2) were obtained. These have allowed precise mapping of the genes as shown in Table 1.

TABLE 1. Physical locations of the *speA*, *speB*, *speC*, and *metK* genes

Gene	Genetic map location (min)[a]	Physical map location (kb)[b]	Phages positive on test[c]
metK	63.7	3100–3101	473, 474[d,e]
speA	63.5	3099–3097	473, 474[e]
speB	63.5	3097–3096	473, 474[e]
speC	64.0	3123–3121	476, 477[e]

[a] From reference 1.

[b] From Southern blot results; locations in kilobase pairs (kb) as assigned in reference 3. The direction of transcription is clockwise for *metK* and counterclockwise for the *spe* genes with respect to the *E. coli* chromosome. The locations indicate the transcriptional "sense."

[c] Phages 470 to 480, inclusive, of the "Miniset" available from Y. Kohara were tested. The reference numbers of the tested phages as used by Kohara et al. (4) in order are: 10B4, 1A2, 6C5, 1H10, 23G4S, 12C6, 3A9, 3D11, 1G7, 21H2, and 3B2. The phages were tested by their ability to complement the *metK501* mutation in strain DM101 and by Southern blot analyses of restricted phage DNA probed with the DNA that encodes the *speA*, *speB*, *speC*, and *metK* genes.

[d] Phages 473 and 474 were able to complement the *metK501* mutation in strain DM101 (8) harboring a tetracycline-resistant derivative of plasmid pGp1-2 (10), which carries the gene (*c*I857) for a temperature-sensitive lambda repressor.

[e] These phages were identified as being positive by their ability to hybridize to the DNA probes derived from protein-coding regions of the respective genes and from the sizes of the hybridizable restriction fragment of the DNA of these phages for various enzymes, used by Kohara et al. (4).

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