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Physical Map Location of the Peptide Methionine Sulfoxide Reductase Gene on the *Escherichia coli* Chromosome

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The enzyme peptide methionine sulfoxide reductase (MsrA), the product of the *msrA* gene, catalyzes the reduction of methionine sulfoxide residues in proteins to methionine (1, 3, 4). During aerobic metabolism, methionine in proteins may be oxidized to methionine sulfoxide by biological reagents such as hydrogen peroxide, hydroxyl radicals, and hypochlorite and superoxide ions (reviewed in reference 2).

The *Escherichia coli* gene for MsrA has been cloned and sequenced (6). The position of this gene on the chromosome was determined by using a membrane which contains λ phages bearing the entire *E. coli* genome on overlapping fragments (5). The membrane was probed with a 765-bp, ^{32}P-5′-end-labeled probe which contained the coding region of the *msrA* gene. Figure 1 shows that two positive clones were found that correspond to overlapping clones numbered 656 (5B5) and 657 (E1H3) on the Kohara map (5). These same clones were also found to be positive when a 125-bp oligonucleotide covering the 5′ upstream region of the gene was used as a probe (data not shown). The overlap region is located approximately between 4520 and 4525 kb on the physical map and could easily accommodate the cloned 3-kb *Eco*RI fragment that contains the *msrA* gene. Restriction analysis using eight restriction endonucleases and sequence analysis defined the location of the gene even further, as shown in the physical map in Fig. 2. The restriction pattern corresponds to that reported by Kohara et al. (5) for this region of the *E. coli* chromosome. The *msrA* gene is transcribed in a counterclockwise direction at about 4523 kb on the physical map. This region lies between the *cysQ* gene and the gene coding for a pyrophosphatase (*ppa*), which are found at about 95.6 and 95.9 min, respectively, on the genetic linkage map (7). This would place the *msrA* gene at about 95.7 to 95.8 min on the genetic linkage map.

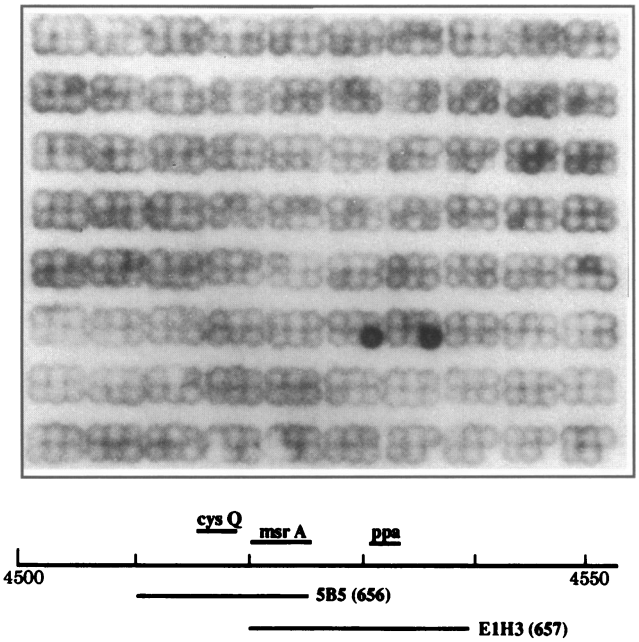

FIG. 1. Chromosomal mapping of the *msrA* gene. A ^{32}P-labeled DNA probe complementary to the coding region of the *msrA* gene was hybridized to a membrane containing a phage library of the *E. coli* genome. The membrane was washed and exposed to X-ray film. Top, autoradiogram of membrane; bottom, portion of physical map of the genome showing the area of hybridization and alignment of phages 5B5 and E1H3.

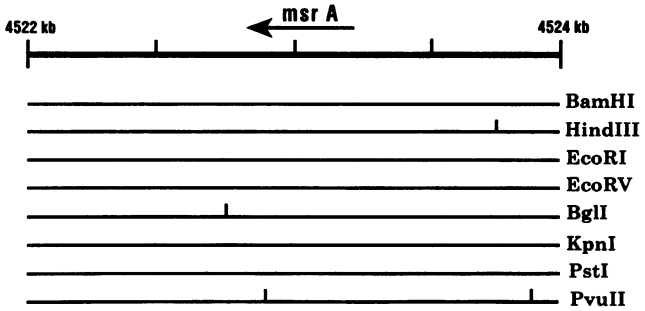

FIG. 2. Restriction map of the *msrA* region of the *E. coli* chromosome generated with eight restriction enzymes. The upper line gives the map units in chromosome kilobase pairs (5). The arrow indicates the position and transcriptional direction of the *msrA* gene.

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