

Location of the Protease II Gene (*ptrB*) on the Physical Map of the *Escherichia coli* Chromosome

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Protease II has been purified from *Escherichia coli* by Pacaud and Richaud (5). The enzyme has substrate specificity for basic amino acids and is inhibited by diisopropylphosphorofluoridate (DFP) and tosyllysylchloromethylketone, similarly to trypsin.

The protease II gene (*ptrB*) in *E. coli* HB101 was cloned and expressed in *E. coli* DH1 (1). The entire nucleotide sequence of an inserted *EcoRV-EcoRV* fragment in pPROII-12 was determined, and *ptrB* was found to start with an ATG codon and to comprise 2,121 bp, as judged by comparison with the protein sequence. The deduced amino acid sequence of the enzyme indicated that the molecular weight is 81,858. The reactive serine residue of protease II was identified as Ser-532 by reaction with tritium DFP. Interestingly, about 25% sequence homology was observed between protease II and prolyl endopeptidases from porcine brain (7) and *Flavobacterium meningosepticum* (8). Since dipeptidyl aminopeptidase IV (4) and acyl amino acid releasing enzyme (3) seem to belong to the same family as prolyl endopeptidase (6), protease II will also be included in this family.

The position of the *ptrB* gene on the *E. coli* chromosome

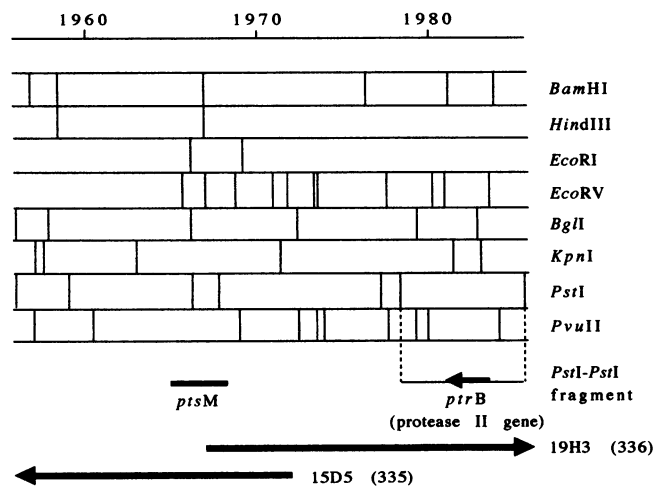


FIG. 1. Location of *ptrB* gene based on the revised map of Kohara et al. (2; personal communication). The arrows indicate the orientations of genes. Map units are indicated in kilobase pairs. *ptsM*, phosphotransferase system gene.

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was determined by hybridization to the Kohara library (2). With the *EcoRV-EcoRV* fragment of pPROII-12 as a probe, only one clone 19H3 (phage 336) hybridized with the probe. Comparison of the *ptrB* sequence with the restriction map of Kohara et al. (2) and revised data (Y. Kohara, personal communication) places the gene at 1981 to 1984 kb on the *E. coli* physical map (Fig. 1), with the direction of transcription as shown. The location of the *ptrB* gene on the *EcoRV-EcoRV* fragment of 19H3 was also confirmed by nucleotide sequencing.

Nucleotide sequence accession number. The nucleotide sequence of the *ptrB* gene has been assigned GenBank accession number D01148.

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