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Construction of a Shuttle Vector Containing the Minimal Replication Origin of *Bacillus thuringiensis* to Allow Efficient Transformation

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To develop the improved *Bacillus thuringiensis* (Bt)-*Escherichia coli* (*E. coli*) shuttle vector system, we constructed two vectors to confirm the minimal replication region of Bt-*E. coli* shuttle vector,pHT3101. Both vectors have the fragment carrying multi-cloning site (MCS) of pUC18, the pUC replicon for the replication in *E. coli*, the erythromycin resistance (Em^r) and the ampicillin resistance (Amp^r) gene for selection of transformants. The first is a pHT261 carrying 261 base pairs as the Bt replication origin. The second is a pHT1K carrying the minimal replicon that resides within a 2.9 kb fragment of the plasmid pHT3101. In order to test their replication in Bt, electroporation and selection on NA medium plates containing erythromycin were performed. Transformants were obtained by pHT1K but not by pHT261. These results suggest that the Bt replication origin is in a 1.0 kb fragment. Therefore, pHT1K must be a very useful Bt-*E.coli* shuttle vector having minimal Bt replication region.