

中文題目：發現一種新型包含嵌入序列(IS26)與乙內醯氨酵素基因(SHV-12)之不完整第一型嵌入子於臺灣分離之成簇腸桿菌

英文題目：Discovery of a new IS26- and *bla*<sub>SHV-12</sub>-containing imperfect class 1 integron in *Enterobacter cloacae* isolated from Taiwan

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## 前言

Extended-spectrum  $\beta$ -lactamases (ESBLs), conferring resistance to broad-spectrum cephalosporins, have offer major challenges to antimicrobial therapy owing to their broadly expanded dissemination worldwide. SHV-type enzymes, encoded by *bla*<sub>SHV</sub> genes, have been one of the most commonly found ESBL families, implying their abilities to proliferate rapidly among clinically relevant species. However, the factors contributing to the highly spreading capability of the *bla*<sub>SHV</sub>-containing plasmids have seldom been addressed.

Integrans have been recognized as significant contributors to horizontal transfer of antibiotic resistance genes in gram-negative bacteria. Class 1 integrons, the most commonly found integrons in gram-negative bacteria, contain *intI*, encoding an integrase that can mobilize and insert gene cassettes. Although rarely confirmed, association between integron and *bla*<sub>SHV</sub> carriage, as located on a common plasmid, has been reported in *Klebsiella* species. **However, direct evidence of *bla*<sub>SHV</sub> genes as inserted gene cassettes contained within the integron structures has not been reported as yet.**

## 材料及方法

An SHV-12-producing strain of *Enterobacter cloacae* (E71) was investigated for the genetic environments of the SHV-12-encoding  $\beta$ -lactamase gene (*bla*<sub>SHV-12</sub>) by using molecular cloning experiment and subsequent DNA sequencing analysis. Plasmid DNA of E71 (designated pE71) was digested with restriction enzyme *Bam*HI and digested fragments were cloned into vector plasmid pOK12. The pE71 was also digested with *Sau*3A1 and digested fragments were cloned into vector pBK-CMV. Then *E. coli* DH5 $\alpha$  transformants were preselected to resistance to ampicillin followed by replicating onto Luria-Bertani agar plates containing 2 $\mu$ g of cefotaxime per ml.

## 結果

Three distinct plasmids including pE71-1 (containing a 3.6-kb *Bam*HI fragment), pE71-2 (containing a 4.5-kb *Bam*HI fragment) and pE71-3 (containing a 7.6-kb *Sau*3A1 fragment) were obtained. Nucleotides sequencing of these fragments revealed that the *bla*<sub>SHV-12</sub> linked to an insertion sequence of IS26 was contained within a distinct class 1 integron with a novel gene cassette configuration (Fig.). The 4.5-kb cassette array containing 5'-conserved segment-*intI*-IS26-*bla*<sub>SHV-12</sub> may be belonged to a novel imperfect class 1 integron, lacking 3'-conserved segment in the downstream region including additional 3.1-kb nucleotide sequence in pE71-3.

## 結論

**For the first time, the *bla*<sub>SHV</sub> gene as an inserted gene cassette contained within an integron structure was evidenced.** This study first describes the newly characterized imperfect class 1 integron that may contribute to transfer of the  $\beta$ -lactamase gene encoding SHV-12 in *Enterobacter cloacae*.

Fig.

