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Linkage of *vat*(E) and *erm*(B) in Streptogramin-Resistant *Enterococcus faecium* Isolates from Europe

Quinupristin-dalfopristin is a streptogramin and has recently been approved for human treatment (7). Streptogramins have been considered valuable antimicrobial drugs for treatment of infections with methicillin-resistant *Staphylococcus aureus* (MRSA) and multiresistant enterococci (8). Low frequencies of streptogramin resistance have been detected among *Enterococcus faecium* strains of human origin in Europe and the United States (8, 11), whereas streptogramin resistance has been detected frequently among *E. faecium* strains of animal origin (2, 12, 14), especially among poultry isolates (2, 14). The streptogramin virginiamycin has been used as a growth promoter in both Europe and the United States. Streptogramins consist of two compounds: streptogramin A and streptogramin B (4). Several genes encoding resistance to streptogramins in *E. faecium* have been described (10). Two genes encoding resistance to streptogramin A in *E. faecium* have been identified. These are *vat*(D) (formerly called *satA* [9]) and *vat*(E) (formerly called *satG* [15]). For streptogramin B resistance, the *erm* genes are known to encode macrolide, lincosamide, and streptogramin B resistance (MLS_B) (13). Another gene encoding streptogramin B resistance in enterococci is *vgb* (1).

In an attempt to determine the genetic background for streptogramin resistance in *E. faecium* of poultry origin, a 7.5-kb *Eco*RI-digested DNA fragment was cloned from isolate F9731349-1 into pUC18Not (6), resulting in the plasmid pVIR1. The streptogramin-sensitive *E. coli* DB10 (9) (MIC of virginiamycin, 8 μg/ml; MIC of pristinamycin, 4 μg/ml) was used as a recipient. *E. coli* DB10 containing pVIR1 (MIC of virginiamycin, 64 μg/ml; MIC of pristinamycin, 128 μg/ml) was resistant to streptogramins. The entire DNA fragment was sequenced, and the sequence was deposited in GenBank under the accession number AF242872. The sequence revealed the presence of two insertion sequences (IS*1216V* and part of IS*1542*), *erm*(B), and *vat*(E) (Fig. 1). No other open reading frames were detected. This is the first evidence of a direct physical linking of the *vat*(E) and the *erm*(B) genes. A previous publication did not find a similar link between an *erm*(B) gene and the *vat*(D) gene in a streptogramin-resistant *E. faecium* strain (HM1032) of human origin (3).

The linkage of *erm*(B) and *vat*(E) was tested in 102 *vat*(E)-positive streptogramin-resistant *E. faecium* poultry isolates from Denmark isolated in 1997. The linkage was found in 74% of the isolates using the new primers satG1-out (5′-GCATTT GCGTCAGGTATAGT-3′) and ermB2-1 (5′-CGCCATACC ACAGATGTTCC-3′) on the basis of the finding that, in all PCR-positive isolates, an amplicon of 942 bp was obtained. Previously described *vat*(E)-positive strains from different origins and countries in Europe were also tested for the presence of the link between *vat*(E) and *erm*(B). The 942-bp PCR amplicon was obtained from UW1965K1 (a sewage isolate from Germany) (15), K14syn (a poultry isolate from The Netherlands) (5), and A33 (a chicken isolate from the United Kingdom) (12) (data not shown). This indicates the presence of a highly conserved genetic element, containing *erm*(B) and *vat*(E) and mediating resistance to both streptogramins and macrolides, in Europe.

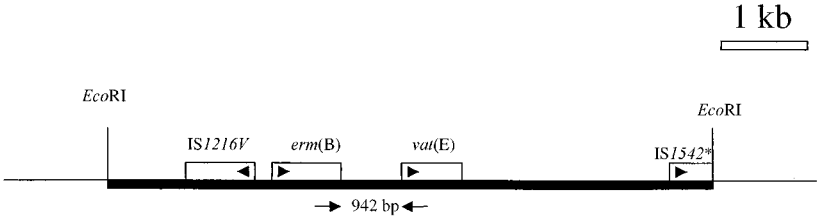

FIG. 1. pVIR1. The sizes and positions of the genes and insertion sequences are indicated. Filled bar, 7.5-kb insert cloned in the *Eco*RI site of pUC18Not. Restriction enzyme sites and open reading frames for IS*1216V*, *erm*(B), *vat*(E), and IS*1542* are indicated. Only the sequence to the internal *Eco*RI site in IS*1542* is present (asterisk). Arrowheads, orientations of the genes; arrows positions and orientations of the primers.

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