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Genetic Platform Of *rmtD2*, A New Allele Of A 16s rRNA Methylase

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Abstract:

Background. *rmtD2*, a new allele of *rmtD*, was identified in a survey of aminoglycoside (AG) resistance mediated by 16S methylation among enterobacteria in Argentina (Chung, C et al, 109th ASM General Meeting, abst A-079, 2009). The aim of this study was to analyze the genetic platform of *rmtD2*.

Methods. *Xba*I DNA libraries from *E. cloacae* Q4010, *E. aerogenes* Q4079 and *C. freundii* Q1174 were constructed in *E. coli*. Cloned fragments containing *rmtD2* were sequenced. Conjugation was used to transfer *rmtD2* from these 3 strains to *E. coli*. *rmtD2* was amplified and cloned in *E. coli*. MICs to AMK, GEN, KAN, TOB and NET were determined by Etest.

Results. *E. coli* transconjugants harboring *rmtD2* (obtained in all cases) as well as *E. coli* harboring cloned *rmtD2* were highly resistant to the AGs tested (MICs ≥ 256 μ g/ml). Sequences of the cloned fragments from strains Q1174 and Q4010 (~10Kb) were 100% identical. They differed from the corresponding sequence in the cloned fragment from strain Q4079 (~23 kb) in the cassette array of a class 1 integron. In this longest fragment, *rmtD2* was immersed in a Tn2670-like element. The *rmtD2* flanking regions showed similar architecture as that from *rmtD* (GenBank DQ914960): a putative tRNA ribosyl-transferase gene and a Δ *groEL* were found upstream of *rmtD2*, all flanked by 2 ISCR14 elements. This ISCR14-bracketed structure was downstream to the 3'-CS of the class 1 integron. However, some intergenic regions showed key differences between the environments of *rmtD* and *rmtD2*. Of note, the ISCR14 located upstream to *rmtD2* is a chimeric element that might be the result of a recombination between ISCR14 and ISCR5B.

Conclusion. *rmtD2* was harbored in a conjugative plasmid. Its flanking regions showed a similar structure to the reported for *rmtD* but probably having an independent assembly. The finding of *rmtD2* in a Tn2670-like element constitutes a potential way for a broader dissemination.