

The roles of HicBA and a novel toxin-antitoxin-like system, TsxAB, in the stability of IncX4 resistance plasmids in *Escherichia coli*

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Objectives: To identify toxin-antitoxin (TA)-like plasmid stability loci on IncX4 plasmids. **Methods:** TA-like loci were identified bioinformatically and their contribution to stability of the IncX4 plasmid pJIE143 was tested in optimal growth conditions in vitro. The conservation of the TA-like loci identified was analysed within an updated IncX plasmid database. **Results:** A novel TA-like locus, tsxAB, was identified on the IncX4 plasmid pJIE143, carrying the important plasmid-borne antibiotic resistance gene blaCTX-M-15. pJIE143 (the WT plasmid) and its tsxA mutant are stable for 80 bacterial generations in the absence of selective pressure but a tsxB deletion mutant of pJIE143 is relatively quickly lost without positive selection (91.1%±1.5% loss after 50 generations). Nine IncX subclasses were identified among 272 fully sequenced IncX plasmids, dominated by those identified as IncX3, IncX1 and IncX4 subclasses in PlasmidFinder. The novel TA-like locus, tsxAB, appea