IS1151, an IS-like element of <u>Clostridium perfringens</u>

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Mobile genetic elements have been found in most microorganisms where they have been sought (for a review see 1). However, insertion sequences (IS) have not yet been described in <u>Clostridium perfringens</u>. From a <u>C. perfringens</u> type D (strain NCTC2062) plasmid library constructed in pUC19 (2), an IS-like element, IS1151, was identified and located 96 bp upstream from the epsilon-toxin structural gene (3). This sequence (1696 bp) possesses two 23 bp stretches, one at each extremity, which constitute a perfect inverted repeat. A search in the EMBL data bank revealed homologies with the IS231 family of Bacillus thurigiensis insertion elements (4, 5), and with IS4, IS186, and IS421 insertion sequences of E. coli (6, 7, 8). The inverted repeats of IS1151 are homologous to the corresponding sequences of the IS231 and IS4 insertion elements but diverge from those of IS186 and of IS421. The left inverted repeat sequence of IS231A diverges also from those of IS4 and IS1151 (by 3 internal base-pairs) but the left inverted repeat sequences of IS4 and IS1151 differ even more (by 6 internal basepairs). The internal sequence of IS1151 possesses potential boxes at positions -35 and -10 (TCAACC-17-TATAAT), a Shine-Dalgarno sequence, a 1422 bp open reading frame, and a potential transcriptional stop signal. The amino-acid sequence deduced from the open reading frame (474 aa) resembles the potential transposase sequences of the IS231 family and of IS4, IS186, IS421, and Tn5 (9). The amino-acid sequence between positions 339 to 355 (YSLRWQIEIIFKVWKSL) is particularly conserved. After multiple alignment of the various amino-acid sequences with the ClustalV package (10), IS1151 appears to be more closely related to IS231A (32 % homology) than to IS4 (16 %), IS186 (17 %), IS421 (17 %), and Tn5 (17 %). A phylogenetic tree

constructed from the inverted repeated sequences and from the deduced amino acid sequences suggests that IS1151 and IS4 are derived from IS231.

Like IS231 in <u>B. thurigiensis</u>, IS1151 may be involved in virulence gene transfer in <u>C. perfringens</u>. Indeed, an IS1151-like element was located near the epsilon-toxin structural gene in all 19 type B and type D isolates studied by Southern blotting (3). A DNA-DNA colony hybridization study (11) performed with an IS1151 probe internal to the ORF sequence (nucleotides 314 to 1303) also revealed homologous sequences in some non-epsilon-toxin-producing strains: in all 7 type C strains (beta-toxin-producing), in 3 out of 8 enterotoxin-producing type A strains, but only in a very few non-enterotoxigenic type A strains (5 of 108). Moreover, some type C strains having lost the capacity to produce beta-toxin failed to hybridize with the IS1151 probe. Further studies are under way to explain the role of IS1151.

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