

Common sequence motifs in DNA relaxases and nick regions from a variety of DNA transfer systems

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Site and strand specific nicking at a transfer origin (*oriT*) is the central event in initiation of transfer DNA replication during bacterial conjugation. Recently, sequence identity in a DNA region adjacent to the nick sites of IncP plasmids RP4 and R751 and part of the border sequences of agrobacterial Ti and Ri plasmids has been demonstrated (Figure 1 and ref. 1). A search for this 'nick region' in a variety of transfer origins of other plasmids demonstrates its conservation in R64 (IncI1) and pTF-FC2, a mobilizable plasmid from *Thiobacillus ferrooxidans* (Figure 1, ref. 2). This finding is paralleled by occurrence of a common sequence motif in proteins (DNA-relaxases) encoded by the respective plasmids that interact with their cognate nick regions (Figure 2). Using the corresponding consensus sequence, a search in protein data bases (NBRF-Protein Release 27.0; Swiss-Prot Release 16.0) revealed that the 'relaxase motif' is also occurring in potential DNA-relaxing proteins (Rlx) of mobilizable plasmids from *S.aureus* (Figure 2).

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RP4/RP4		T G G G C C T A C T T C	A C C T A T C C T G C C C	(3)
R751		T T A G C T A A C T T C	A C A C A T C C T G C C C	(3)
R64		G G T A T T A C A A T T	G C A C A T C C T G T C C	(4)
pTF-FC2		G T G T G T T A C A A C	G G T C A T C C T G T A T	(5)
pTIT37	Nop (LB)	G T T T A C A C C A C A	A T A T A T C C T G C C A	(6)
pTIT37	Nop (RB)	G T T T A C C C G C C A	A T A T A T C C T G T C A	(6)
pTIC58	Nop (LB)	G T T T A C A C C A C A	A T A T A T C C T G C C A	(7)
pTIC58	Nop (RB)	G T T T A C C C G C C A	A T A T A T C C T G T C A	(7)
pTIA6	Oct (LB a)	A T T T A C A A T T G A	A T A T A T C C T G C C G	(8)
pTIA6	Oct (RB b)	A A T T A C A A C G G T	A T A T A T C C T G C C A	(8)
pTIA6	Oct (LB a)	A T T T A C A A T T G A	A T A T A T C C T G C C G	(9)
pTIA6	Oct (RB b)	A A T T A C A A C G G T	A T A T A T C C T G C C A	(9)
pT115955	Oct (LB a)	T T T T A C A A T T G A	A T A T A T C C T G C C G	(6)
pT115955	Oct (RB b)	A A T T A C A A C G G T	A T A T A T C C T G C C A	(6)
pT115955	Oct (LB c)	T T T T A C A C C T C G	A T A T A T C C T G C C A	(6)
pT115955	Oct (RB d)	A A T T A C A A C C G C	A T A T A T C C T G C C A	(6)
pRIA4	(LB a)	G T T T A C A T C A C A	A T A T A T C C T G C C A	(6)
pRIA4	(RB b)	C A T G A C A G G A A C	A T A T A T C C T G T C A	(6)
pRIA4	(LB c)	T T T T A C G T T G G C	A T A T A T C C T G C C A	(6)
pRIA4	(RB d)	C C T G A C C A C A A G	A T A T A T C C T G T C A	(6)
Consensus:			A T A T C T A T C C T G C C G G C C T A	

Figure 1. Alignment of defined and potential nick regions of various plasmids. Conserved nucleotide positions are boxed. Locations of nick-sites are indicated by wedges. References are given in brackets.

Tral (RP4)	104	R V S A V	H H D T D N L H I H I A I N	K I H P T	(10)
Tral (R751)	104	R I S A V	H N D T D N L H I H I A I N	K I H P T	(10)
NikB (R64)	153	Y V S A V	H T D T D N L H V H V A V N	R V H P E	(4)
MobA (pTF-FC2)	51	A I Y G L	H A D T D N L H L H L A I N	R V H P E	(5)
VirD2 (pTIA6)	126	Y L T A Y	H V D R D H P H L H V V V N	R R E L L	(11)
VirD2 (pRIA4)	126	Y L T A F	H I D R D H P H L H V V V N	R R E L L	(12)
Rlx (pC221)	94	V A V Y T	H T D K D H Y H N H I V I N	S V D L E	(13)
Rlx (pS194)	94	V A V Y T	H T D K D H Y H N H I I I N	S V N L E	(14)
Rlx (pC223)	94	V A V Y T	H N D T D H V H N H I V I N	S I D L E	(15)
Consensus		a	H D D ^N _H H H a a a N		

Figure 2. Alignment of DNA-relaxases. Numbers indicate the sequence positions of the first amino acid residues shown. A common motif is boxed. References are given in brackets.

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