Strain	Genotype/Phenotype	Source
DH5aMCR	F_mcrA (mrr-hsdRMS- mcrBC) 80dlacZM15 (lacZYAargF) U169 endA1 recA1 supE44 I-thi-1 gyrAa96 relA1	Gibco BRL
BL21 (DE3)	F ompT hsdS _B (r _B m _B) gal dcm (DE3)	Promega
BL21(DE3) <i>ihfA</i>	Deletion of <i>ihfA</i>	Laboratory strain

Table S1: Bacterial strains, plasmids, and oligonucleotides used in this study.

Plasmid	Description	Reference
pET28_intN1	<i>intN1</i> under T7 control in pET28	(5)
pJWS200	pGEM-T containing <i>attN1</i> ; template for site-directed mutagenesis of arm-type sites	(10)
pLR20	Cointegrate plasmid formed following recombination between pJWS200 (<i>attN1</i>) and pJWS14 (<i>attBT1-11</i>); contains <i>attL</i> and <i>attR</i>	L. Rajeev, unpublished results
pLR21	<i>orf2x</i> under T7 control in pET27b	L. Rajeev, unpublished results

Oligonucleotide	Sequence (5'-3')	Purpose	Reference
NdeI-orf2x	CCC GAA GCA TAT	Cloning of Orf2x into	L. Rajeev,
	GAC AGA CAT ATT	overexpression vector	unpublished
	GGC AAT TAT CC		results
orf2x-EcoRI	TCG AAT TCT TAG	Cloning of Orf2x into	L. Rajeev,
	ATT AAA GGA TTG	overexpression vector	unpublished
	TGT TCA CC		results
$LR207^{1}$	CCT TCT GGT AGT	attL amplification	L. Rajeev,
	GCA CAT TAG AAA		unpublished
	GAA ATA CCC TAT		results
	AAC		
LR200 ¹	ATA TTT TCC CCA	attL and attN1	L. Rajeev,
	CAT TTT CCC CAC	amplification	unpublished

	ATC TGC T		results
LR193 ¹	GAC TTA CTG CTA	attR an attN1	L. Rajeev,
	TAT TTT TTG CAC	amplification	unpublished
	GTG TGG GG	-	results
LR212 ¹	CGT ATC TTT GCA	attR amplification	L. Rajeev,
	CCG CAA TTG AGA		unpublished
	AAT CAA GC		results
MM208 ¹	CAT AGA CTT TCA	attL amplification and	This study
	GGT TGA ATT TTA	sequencing	
	CTC TGC TGC		
attBT1-1 T4	TCT TAG CTT TTC	IntN1 cleavage assays	L. Rajeev,
	GTG GTA CCC AGA		unpublished
	С		results
attBT1-1 bottom	CAT CCC GGT TCG	IntN1 cleavage assays	L. Rajeev,
	ACC CCG GGT CTG		unpublished
	GGT ACC ACG		results
	AAA AG		
DR1a mut corr ²	GCT ATA TTT TTT	Site-directed	This study
	GCG ACG TCG	mutagenesis of DR1a	
	GGG AAA ATG		
	TGG GGA AAA TTC		
	AAG C		
DR1b mut ²	GCA CGT GTG GGG	Site-directed	This study
	AAA GAC GTC	mutagenesis of DR1b	
	GGA AAA TTC		
	AAG CAA AAG		
	AAA AAG C		
$DR2a mut^2$	GAA ATA ATT AGA	Site-directed	This study
	CGT CGG AAA ATG	mutagenesis of DR2a	
	TGG GTA AAA		
	AGA AAA ATG		
	CGG		
DR2b mut^2	GAA ATA ATT AAA	Site-directed	This study
	GTG GGG AAA	mutagenesis of DR2b	
	GAC GTC GTA AAA		
	AGA AAA ATG		
	CGG		
DR3a mut ²	GCA AAA TAT TTA	Sire-directed	This study
	GCA GGA CGT	mutagenesis of DR3a	
	CGG AAA ATG		
	TGG GGA AAA TAT		
2	TTA TAT TTG C		
DR3b mut ²	GCA GAT GTG	Site-directed	This study
	GGG AAA GAC	mutagenesis of DR3b	
	GTC GGA AAA TAT		
	TTA TAT TTG CAG		

	С		
¹ Primers were ordered with 5' 6-carboxyfluorescein phosphoramidate (FAM) labels and paired			

with an unlabeled reverse primer for amplification of footprinting substrates. ² Only the top strand of each pair of mutagenesis primers is shown.



Figure S1: SDS-PAGE analysis of IntN1 protein samples following purification. IntN1 was overexpressed and purified as described in Materials and Methods. The predicted molecular weight of IntN1 is 53 kDa. Lane 1, *E. coli* crude extract containing IntN1; lane 2, IntN1 following heparin-agarose chromatography; lane 3, IntN1 following heparin agarose chromatography and gel filtration chromatography before dialysis into storage buffer; lane 4, IntN1 as described in lane 3 but following dialysis into storage buffer; lane 5, Benchmark ladder (Invitrogen).



Figure S2A: SDS-PAGE analysis of Orf2x following partial purification. Orf2x was overexpressed in an *ihfA E. coli* background and partially purified as described in Materials and Methods. Lanes 1 and 5, Precision Plus Kaleidoscope Protein Standard (Bio-Rad); Lane 2, Orf2x pellet fraction; Lane 3, Orf2x following heparin-agarose chromatography, Lane 4, Orf2x following heparin and SP cation exchange chromatography.



Figure S2B: Gel shift assays with a DNA substrate containing *attL* were used to detect Orf2x activity following purification steps. pET27b empty vector was partially purified using the same protocol and served as a negative control. Lane 1, free *attL* DNA; lane 2, Orf2x (pLR21) extract following heparin-agarose chromatography; lane 3, pET27b extract following heparin and SP chromatography; lane 4, Orf2x (pLR21) extract following heparin and SP chromatography.