

The toxic guardians — Multiple toxin-antitoxin systems provide stability, avoid deletions and maintain virulence genes of *Pseudomonas syringae* virulence plasmids

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Table S1 List and application of primers used in this work.

Primer name ^a	Sequence ^b	5'pos ^c	Remarks
Cloning of putative stability determinants			
pPsv48A (accession no. FR820585)			
SD1_F	<u>GGATCC</u> GATGGTAACCGCCAATGTTC	21649*	Cloning of SD1 into pKMAG-C
SD1_R	<u>GGATCC</u> TTCTGGGATTCCGGCTTAC	20277	
TA1_F	<u>GGATCC</u> ACATAGGCTTGCGATTCAGC	24281*	Cloning of TA1 into pKMAG-C
TA1_R	<u>GGATCC</u> TTGATTGCTAATTCCGACACTG	23449	
TA2_F	<u>GGATCC</u> TTCCATCTCGTTTGCTGATG	37991*	Cloning of TA2 into pKMAG-C
TA2_R	<u>GGATCC</u> CGTTCCTCTATCGGTCTTCG	37061	
SD2_F	<u>GGATCC</u> ATGCACTTCATCGGGTAGATTTC	53332*	Cloning of SD2 into pKMAG-C
SD2_R	<u>GGATCC</u> CATGAGCGAAGTGTAGTGCAG	52067	
TA3_F	<u>GGATCC</u> CCACGTAGTAGCCCGATGC	53072	Cloning of TA3 into pKMAG-C

TA3_R	<u>GGATCCTCAGGGAGCACGAAACATC</u>	54708*	
TA4_F	<u>GGATCCGATCACCGCTGACATCGAG</u>	64737*	Cloning of TA4 into pKMAG-C
TA4_R	<u>GGATCCCGGCCAAGAACAAGAAAGAC</u>	63811	
SD3_F	<u>GGATCCACCCGTGTAGCGGTTAATTC</u>	79421	Cloning of SD3 into pKMAG-C
SD3_R	<u>GGATCCTTCCAAAAAGCCCGGTTAC</u>	79884*	
pPsv48B (accession no. FR820586)			
TA5_F	<u>GGATCCCTTGGATACGCACCGAAAAC</u>	11832*	Cloning of TA5 into pKMAG-C
TA5_R	<u>GGATCCAGGTGTCAATTGGCGATACC</u>	11108	
SD4_F	<u>GGATCCCCCAAATAGACCCAGCATATC</u>	11884	Cloning of SD4 into pKMAG-C
SD4_R	<u>GGATCCGGATGGTTACCGCCATACC</u>	13274*	
SD5_F	<u>GGATCCACCGTCGAAGCCAAGGTG</u>	31991	Cloning of SD5 into pKMAG-C
SD5_R	<u>GGATCCGCTGCCGCACTCGAAATAG</u>	32672*	
SD6_F	<u>GGATCCAGCGCACACTTGACGCTAC</u>	35775	Cloning of SD6 into pKMAG-C
SD6_R	<u>GGATCCCTATGACACCCGCTGACTTTC</u>	37405*	
pPsv48C (accession no. FR820587)			
TA6_F	<u>GGATCCCTGCCAAGAAAGTCGTCAAG</u>	1760	Cloning of TA6 into pKMAG-C
TA6_R	<u>GGATCCCGAAGCATGAATACCAGCAC</u>	2442*	

TA7_F	<u>GGATCCTTTTCGACCGGAATTGATTG</u>	4752*	Cloning of TA7 into pKMAG-C
TA7_R	<u>GGATCCGTACCCCATCATGCGATTG</u>	3866	
SD7_F	<u>GGATCCCTTCATGGAAAATGCTACCG</u>	11121	Cloning of SD7 into pKMAG-C
SD7_R	<u>GGATCCCTTAATAGGTAGGTGTGAGCTATCC</u>	9861*	
TA8_F	<u>GGATCCGCAATACAGTGCAAACCAATG</u>	39466	Cloning of TA8 into pKMAG-C
TA8_R	<u>GGATCCTGATCAACATCCCCCAACAG</u>	40245*	

Regulated expression of toxin genes in *E. coli*

pPsv48A

TA1_tox_F	<u>TATGGTACCAGCAAAGAAGCCCGCAAGAC</u>	23850*	Cloning of toxin from TA1 into pBAD24
TA1_tox_R	<u>TATCTGCAGGCCATTCCACAGGCATAAAG</u>	23496	
TA2_tox_F	<u>TATGGTACCAGAATTGAAGTGGACAAGCAAG</u>	37340*	Cloning of toxin from TA2 into pBAD24
TA2_tox_R	<u>TATCTGCAGCGTTCCTCTATCGGTCTTCG</u>	37061	
TA3_tox_F	<u>TATGGTACCAATCCTGCTCGATACGAACG</u>	53548	Cloning of toxin from TA3 into pBAD24
TA3_tox_R	<u>TATCTGCAGCCTGATTCAACACGACGTTTAC</u>	53982*	
TA4_tox_F	<u>TATGGTACCCGTGCTAACAATTCGCTG</u>	64278*	Cloning of toxin from TA4 into pBAD24
TA4_tox_R	<u>TAACTGCAGACGGTTCAAGGGTATTGCTG</u>	63995	

pPsv48B

TA5_tox_F	<u>TATGGTACCGATGCGAGGTGACCTAGTCA</u>	11434*	Cloning of toxin from TA5 into pBAD24
TA5_tox_R	<u>TATCTGCAGGCGACACAAGGAAAAGGTGT</u>	11094	
pPsv48C			
TA6_tox_F	<u>TATGGTACCACTACCCATTTTCTGGCTAG</u>	2108	Cloning of toxin from TA6 into pBAD24
TA6_tox_R	<u>TATCTGCAGCGAAGCATGAATACCAGCAC</u>	2442*	
TA7_tox_R	<u>TATGGTACCACAAGTAATCGTCACAGAAG</u>	4269*	Cloning of toxin from TA7 into pBAD24
TA7_tox_F	<u>TATCTGCAGGTCGATCTTCTCGGCCAGTC</u>	3938	
TA8_tox_F	<u>TATGGTACCACAAATCATCTGGCGGCAAC</u>	39831	Cloning of toxin from TA8 into pBAD24
TA8_tox_R	<u>TATCTGCAGATCAACATCCCCCAACAGC</u>	40243*	
Construction of pRK3A and pRK3C			
pPsv48A			
A1_R	<u>GGATCCATCCCTGGCTTCGGTCATAG</u>	23716	with TA1_F
A2_R	<u>GGATCCGGCCAGAACTCGTATAGCC</u>	37281	with TA2_F
A3_R	<u>GGATCCGCTGCGGTTCTGAAATTACG</u>	53584*	with TA3_F
pPsv48C			
A6_R	<u>GGATCCGTTCTGAAAGGGGCAGAACG</u>	2240*	with TA6_F

A7_R	<u>GGATCC</u> AGCTTCTGGGTCCTTTGCAG	4189*	with TA7_F
A8_R	<u>GGATCC</u> GCTTGCGGGTCTTCGTTG	39907*	with TA8_F
PCR multiplex			
pPsv48C			
RepL_F	TACGCTACGCGAGCTCAATC	33336	Detection of PSPSV_C0043
RepL_R	<u>TCTAGAGG</u> CCTTCCTTATTCATGATGC	34355*	
7C_F	AAACAGCCCCCGACATTG	4142*	Detection of PSPSV_C0007
7C_R	GCCGAAATTTCCGTCAGG	3411	
RepJ_F	ATAGGCTCCTAGCGTCGAG	30484	Amplifies a region within RepJ replicon
RepJ_R	AGAGCGATTGACCATGCAC	31028*	
RepA_F	CAATAGCAGCTTGTCGCTCA	882	<i>repA</i> detection
RepA_R	GTTACCGTTGGCCTCTCGTC	1148*	
ipt_F	GCGTCTCCAGGAAGAATTGG	18715	To detect gene <i>idi</i> and estimate copy number
ipt_R	ACCTCATCCGGATTCAAGC	18870*	
Plasmid copy number estimation by qPCR			
Chromosome (accession n° CP008742)			
gyrA_F	GACGAGCTGAAGCAGTCCTACC	3774872*	Internal to <i>gyrA</i>

gyrA_R	TTCCAGTCGTTACCCAGCTCG	3774736	
pPsv48A			
ptz_F	AAGACCCATACTGGTGCGATAG	28623*	Internal to <i>ptz</i>
ptz_R	CCGCAACCAACTCGTCTAAC	28471	
pPsv48B			
AO1_F	AGGGAAACCGAAGACCAATG	10026	Internal to <i>hopAO1</i>
AO1_R	AACCAGCCAATTTTCAGATCG	10176*	
pPsv48C			
ipt_F	GCGTCTCCAGGAAGAATTGG	18715	To detect gene <i>idi</i> and estimate copy number
ipt_R	ACCTCATCCGGATTCAAGC	18870*	
Other purposes			
pk18mob-km-sac_F	<u>GATATC</u> TTATGGACAGCAAGCGAACC		Used to amplify Km ^R - <i>sacB</i> cassette from pK18 <i>mobsacB</i>
pk18mob-km-sac_R	<u>GATATC</u> TCGTGGACTATGAGCACGTC		
IS801_R	ACGCATCAGCGTCTTCCTAC		Anneals 74 nt upstream of the 3' end of CRR1; outwards
IS50_F	ACACAGATTTAGCCCAGTCG		Anneals in Tn5GDYN1

^a The name of the primer is indicated, as well as the molecule used as target DNA and the purpose of designing each pair.

^b Adaptors introduced in primers to facilitate cloning are underlined.

^c The coordinates of the annealing point of the first nucleotide of each primer is indicated. Asterisks indicate that the primer anneals in the reverse strand of the DNA molecule.