

## Appendix A. Strains and plasmids

Strain or plasmid	Relevant genotype and characteristics <sup>a</sup>	Reference or source
<b>Strains</b>		
HD12	<i>B. thuringiensis</i> subsp. <i>Morrisoni</i> strain containing multiple insecticidal genes and toxic to <i>Lepidoptera</i> insects.	Laboratory collection
HD73	<i>B. thuringiensis</i> subsp. <i>Kurstaki</i> carrying the <i>cryIAc</i> gene	Laboratory collection
HD73 <sup>-</sup>	HD73 mutant without the large plasmid pHT73	Laboratory collection
B-Pr-88	<i>B. thuringiensis</i> wild strain carrying the <i>cry2Ab</i> gene	Laboratory collection
HD( $\Delta$ <i>sigK</i> )	<i>B. thuringiensis</i> HD73 <i>sigK</i> gene mutant; Kan <sup>R</sup>	Yang <i>et al.</i> 2013
HD( $\Delta$ <i>sigE</i> )	<i>B. thuringiensis</i> HD73 <i>sigE</i> gene mutant	Du <i>et al.</i> 2012
HD(P <i>cry1Da</i> )	<i>B. thuringiensis</i> HD73 carrying pHT-P <i>cry1Da</i> plasmid; Em <sup>R</sup>	This study
HD(P <i>cry1Ae</i> )	<i>B. thuringiensis</i> HD73 carrying pHT-P <i>cry1Ae</i> plasmid; Em <sup>R</sup>	This study
HD(P <i>cry1Bb</i> )	<i>B. thuringiensis</i> HD73 carrying pHT-P <i>cry1Bb</i> plasmid; Em <sup>R</sup>	This study

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HD( <i>Pcry1Fb</i> )	<i>B. thuringiensis</i> HD73 carrying pHT- <i>Pcry1Fb</i> plasmid; Em <sup>R</sup>	This study
HD( <i>Pcry1Ja</i> )	<i>B. thuringiensis</i> HD73 carrying pHT- <i>Pcry1Ja</i> plasmid; Em <sup>R</sup>	This study
$\Delta sigK$ ( <i>Pcry1Da</i> )	HD( $\Delta sigK$ ) carrying pHT- <i>Pcry1Da</i> plasmid; Em <sup>R</sup> , Kan <sup>R</sup>	This study
$\Delta sigK$ ( <i>Pcry1Ae</i> )	HD( $\Delta sigK$ ) carrying pHT- <i>Pcry1Ae</i> plasmid; Em <sup>R</sup> , Kan <sup>R</sup>	This study
$\Delta sigK$ ( <i>Pcry1Bb</i> )	HD( $\Delta sigK$ ) carrying pHT- <i>Pcry1Bb</i> plasmid; Em <sup>R</sup> , Kan <sup>R</sup>	This study
$\Delta sigK$ ( <i>Pcry1Fb</i> )	HD( $\Delta sigK$ ) carrying pHT- <i>Pcry1Fb</i> plasmid; Em <sup>R</sup> , Kan <sup>R</sup>	This study
$\Delta sigK$ ( <i>Pcry1Ja</i> )	HD( $\Delta sigK$ ) carrying pHT- <i>Pcry1Ja</i> plasmid; Em <sup>R</sup> , Kan <sup>R</sup>	This study
$\Delta sigE$ ( <i>Pcry1Da</i> )	HD( $\Delta sigE$ ) carrying pHT- <i>Pcry1Da</i> plasmid; Em <sup>R</sup>	This study
$\Delta sigE$ ( <i>Pcry1Ae</i> )	HD( $\Delta sigE$ ) carrying pHT- <i>Pcry1Ae</i> plasmid; Em <sup>R</sup>	This study
$\Delta sigE$ ( <i>Pcry1Bb</i> )	HD( $\Delta sigE$ ) carrying pHT- <i>Pcry1Bb</i> plasmid; Em <sup>R</sup>	This study
$\Delta sigE$ ( <i>Pcry1Fb</i> )	HD( $\Delta sigE$ ) carrying pHT- <i>Pcry1Fb</i> plasmid; Em <sup>R</sup>	This study
$\Delta sigE$ ( <i>Pcry1Ja</i> )	HD( $\Delta sigE$ ) carrying pHT- <i>Pcry1Ja</i> plasmid; Em <sup>R</sup>	This study
HD(J-1A)	HD73 <sup>-</sup> carrying 315-P1Ja-1Ac plasmid; Em <sup>R</sup>	This study
HD(J-2A)	HD73 <sup>-</sup> carrying 1618-P1Ja-2Ab plasmid; Tet <sup>R</sup>	This study
HD(F-1A)	HD73 <sup>-</sup> carrying 315-P1Fb-1Ac plasmid; Em <sup>R</sup>	This study
HD(F-2A)	HD73 <sup>-</sup> carrying 1618-P1Fb-2Ab plasmid; Tet <sup>R</sup>	This study
HD(J1A- J2A)	HD73 <sup>-</sup> carrying 315-P1Ja-1Ac and 1618-P1Ja-2Ab plasmids; Em <sup>R</sup> , Tet <sup>R</sup>	This study
HD(J2A-F1A)	HD73 <sup>-</sup> carrying 1618-P1Ja-2Ab and 315-P1Fb-1Ac plasmids; Em <sup>R</sup> , Tet <sup>R</sup>	This study
HD(J1A-F2A)	HD73 <sup>-</sup> carrying 315-P1Ja-1Ac and 1618-P1Fb-2Ab plasmids; Em <sup>R</sup> , Tet <sup>R</sup>	This study

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HD(F2A-F1A)	HD73 <sup>-</sup> carrying 1618-P1Fb-2Ab and 315-P1Fb-1Ac plasmids; Em <sup>R</sup> , Tet <sup>R</sup>	This study
<i>E. coli</i> TG1	$\Delta(lac-proAB)$ <i>supE thi hsd-5</i> ( <i>F'</i> <i>traD36 proA</i> <sup>+</sup> <i>proB</i> <sup>+</sup> <i>lacI</i> <sup>q</sup> <i>lacZ</i> $\Delta$ M15), general purpose cloning host	Laboratory collection
<i>E. coli</i> ET12567	<i>F'</i> <i>dam-13::Tn9 dcm-6 hsdM hsdR recF143 zjj-202::Tn10 galK2 galT22 ara14 pacY1 xyl-5 leuB6 thi-1</i> , for generation of unmethylated DNA	Laboratory collection

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### Plasmids

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pHT304-18Z	Promoterless <i>lacZ</i> vector, Em <sup>R</sup> , Ap <sup>R</sup>	Laboratory collection
pHT315	<i>E. coli-B. thuringiensis</i> shuttle, Ap <sup>R</sup> , Em <sup>R</sup>	Arantes, <i>et al.</i> 1991
pHT1618	<i>E. coli-B. thuringiensis</i> shuttle, Ap <sup>R</sup> , Tet <sup>R</sup>	Lereclus, <i>et al.</i> 1992
pHT-Pcry1Da	pHT304-18Z carrying promoter upstream from <i>cry1Da</i>	This study
pHT-Pcry1Ae	pHT304-18Z carrying promoter upstream from <i>cry1Ae</i>	This study
pHT-Pcry1Bb	pHT304-18Z carrying promoter upstream from <i>cry1Bb</i>	This study
pHT-Pcry1Fb	pHT304-18Z carrying promoter upstream from <i>cry1Fb</i>	This study
pHT-Pcry1Ja	pHT304-18Z carrying promoter upstream from <i>cry1Ja</i>	This study
315-P1Ja-1Ac	pHT315 carrying <i>cry1Ja</i> promoter and <i>cry1Ac</i> gene	This study
315-P1Fb-1Ac	pHT315 carrying <i>cry1Fb</i> promoter and <i>cry1Ac</i> gene	This study
1618-P1Ja-2Ab	pHT1618 carrying <i>cry1Ja</i> promoter and <i>cry2Ab</i> gene	This study

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**Appendix B. Primers sequences**

oligonucleotides	sequence (5' - 3') <sup>a</sup>
Pcry1Da-F	CCCAAGCTTGATGGCTTCCTGCCA
Pcry1Da-R	CTAGTCTAGACCATAAACTATCCCC
Pcry1Ae-F	CCCAAGCTTGGGTCTATGGTTCTG
Pcry1Ae-R	CTAGTCTAGAAAGTTACCTCCATCT
Pcry1Bb-F	CGCGGATCCGGGTCTATGGTTCTG
Pcry1Bb-R	TGCACTGCAGGTATAGTTTTATAAT
Pcry1Fb-F	CGCGGATCCGGGTCTATGGTTCTG
Pcry1Fb-R	TGCACTGCAGATGTTCCCTCCACTT
Pcry1Ja-F	CGCGGATCCCGTGAGGGATGGTTA
Pcry1Ja-R	TGCACTGCAGAAGTTACCCCTTTCT
1DaRACE	TCTGCTACAGTATTCCTGTTTCTA
1AeRACE	TTCTCCACCTAATACTTCTACTCA
1BbRACE	GAATTATATAAGCAAATTGTTATAC
1FbRACE	TAAGCGACAAGGATATATCTAACGG
1JaRACE	GAAACTTCGAGTGGATCGATATCAG
UPM	AAGCAGTGGTATCAACGCAGAGTACATGGG
J1A-F1	CTTGCATGCCTGCAGCGTGAGGGATGGTTA

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J1A-R1	CGGATTGTTATCCATAAGTTACCCCTTTCTC
F1A-F1	CTTGCATGCCTGCAGGTCTATGGTTCTGCTG
F1A-R1	CGGATTGTTATCCATATGTTCCCTCCACTT
J1A-F2	GAGAAAGGGGTAACCTTATGGATAACAATCCG
1A-R2	GACGGCCAGTGAATTCCTATTCCTCCATAAGG
F1A-F2	AAGTGGAGGGAACATATGGATAACAATCCG
J2A-F1	CTTGCATGCCTGCAGCGTGAGGGATGGTTA
J2A-R1	CAATACACTATTCATAAGTTACCCCTTTCTC
F2A-F1	CTTGCATGCCTGCAGGTCTATGGTTCTGCTG
F2A-R1	CAATACACTATTCATATGTTCCCTCCACTT
J2A-F2	GAGAAAGGGGTAACCTTATGAATAGTGTATTG
2A-R2	CATGATTACGAATTCATAAAGTGGTGAAAGATI
F2A-F2	AAAGTGGAGGGAACATATGAATAGTGTATTG

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<sup>a</sup>Restriction enzyme sites are underscored.

## REFERENCES

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- Lereclus D, Arantes O. 1992. *spbA* locus ensures the segregational stability of pTH1030, a novel type of gram-positive replicon. *Mol Microbiol*, **6**, 35-46.

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