

Table S1 Details of primers used to amplify regions of the TotiEVE T1 (HITE) on chromosome 1 of BSFs.

Primer	Sequence	Region on BSF genome (bp)	Target on TotiEVE	Expected PCR product size (bp)	Annealing temperature (°C)
HITEHNE F & R	5'-GTT GTA ATC GGG AAT TTG GC-3' & 5'-GAA TGT GAA TAG AAA CTC CGC-3'	97,213,374 – 97,214,373	Host sequence outside EVE	~1000	52
HITEP F & R	5'-AAC AAA ATG TCG CAA CAA GG-3' & 5'-TAC AGA ATA TCC CAG AGT GC-3'	97,212,055 – 97,213,054	POL ORF end of TotiEVE	~1000	50
HITE F & R	5'-TTA TAA AGG TAC CGC TGG AC-3' & 5'-TGC TTA TCT ATT GGG CTA GC-3'	97,211,454 – 97,212,021	RdRP-like domain	~568	50
HITEG F & R	5'-ACT CAT GAT CGA AAA GGA CTC-3' & 5'-TTT CCA AAA CAT CGA TCT GC-3'	97,206,252 – 97,207,251	GAG ORF end of TotiEVE	~1000	50
HITEHNE F & HITEP R	5'-GTT GTA ATC GGG AAT TTG GC-3' & 5'-TAC AGA ATA TCC CAG AGT GC-3'	97,212,055 – 97,214,373	Host sequence outside EVE to POL ORF	~2300*	52
HITEHNE F & HITE R	5'-GTT GTA ATC GGG AAT TTG GC-3' & 5'-TGC TTA TCT ATT GGG CTA GC-3'	97,211,454 – 97,214,373	Host sequence outside EVE to RdRP-like domain	~2900*	52

*extension time was changed from 1 minute to 3 minutes.

Table S4 Abbreviations of virus names or accepted virus species names of those used in phylogenetic trees and paper.

NCBI Accession	Virus species/name	Paper abbreviation*
AB555544	Omono River virus	OmRV
GQ342961	Drosophila melanogaster totivirus SW-2009a	DmTV SW-2009a
EU715328	Armigeres subalbatus virus SaX06-AK20	AsTV SaX06 (AK20)
AY570982	Penaeid shrimp infectious myonecrosis virus	IMNV
BK061373	Hermetia illucens Toti-like virus 1	HiTV1
NC025218	Leptopilina boulardi Toti-like virus	LbTV
MH213243	Linepithema humile toti-like virus 1	LhTV1
MH727531	Solenopsis midden virus	SoMiV
NC032851	Shuangao toti-like virus	ShoTV
HQ158596	Tuber aestivum virus 1	TaV1
NC003745	Saccharomyces cerevisiae virus L-A L1	ScV L-A (L1)
KY207365	Puccinia striiformis totivirus 5	PsTV5
NC028480	Red clover powdery mildew-associated totivirus 1	RPaTV1
NC029096	Panax notoginseng virus A	PnV-A
KC610514	Scheffersomyces segobiensis virus L	SsV-L
NC028481	Red clover powdery mildew-associated totivirus 2	RPaTV2
U01060	Saccharomyces cerevisiae virus L-BC La	ScV L-BC (La)
JN997472	Xanthophyllomyces dendrorhous virus L1A	XdV-L1A
JN997473	Xanthophyllomyces dendrorhous virus L1B	XdV-L1B
LC075489	Red clover powdery mildew-associated totivirus 4	RPaTV4
NC028483	Red clover powdery mildew-associated totivirus 3	RPaTV3

KY207361	<i>Puccinia striiformis</i> totivirus 1	PsTV1
KY207363	<i>Puccinia striiformis</i> totivirus 3	PsTV3
KY207362	<i>Puccinia striiformis</i> totivirus 2	PsTV2
KY207364	<i>Puccinia striiformis</i> totivirus 4	PsTV4
NC028485	Red clover powdery mildew-associated totivirus 5	RPaTV5
NC028486	Red clover powdery mildew-associated totivirus 6	RPaTV6
NC028488	Red clover powdery mildew-associated totivirus 7	RPaTV7
LC075493	Red clover powdery mildew-associated totivirus 8	RPaTV8
KX148550	<i>Anopheles</i> totivirus	AtoV
NC027212	<i>Camponotus yamaokai</i> virus	CYV
NC029312	<i>Camponotus nipponicus</i> virus	CNV
NC029302	Piscine myocarditis-like virus	PMCLV
DQ238861	<i>Giardia canis</i> virus	GcV
NC003555	<i>Giardia lamblia</i> virus	GLV
AF356189	<i>Eimeria brunetti</i> RNA virus 1	EbRV1
KU597305	<i>Eimeria stiedai</i> RNA virus 1	EsRV1
NC026140	<i>Eimeria tenella</i> RNA virus 1	EtRV1
HE588147	<i>Aspergillus foetidus</i> slow virus 1	AfSV1
NC003607	<i>Helminthosporium victoriae</i> virus 190S	HvV190S
NC014823	<i>Tolypocladium cylindrosporum</i> virus 1	TcV1
NC021565	<i>Rosellinia necatrix</i> victorivirus 1	RnVV1
NC005074	<i>Helicobasidium mompa</i> totivirus 1-17	HmTV1-17
NC006367	<i>Magnaporthe oryzae</i> virus 1	MoV1
M92355	<i>Leishmania</i> RNA virus 1	LRV1
U32108	<i>Leishmania</i> RNA virus 2	LRV2
AF127178	<i>Trichomonas vaginalis</i> virus 2	TvV2
AF325840	<i>Trichomonas vaginalis</i> virus 3	TvV3
HQ607522	<i>Trichomonas vaginalis</i> virus 4	TvV4
NC027701	<i>Trichomonas vaginalis</i> virus 1	TvV1

*Abbreviations used were for convenience as some viruses do not have official abbreviations.

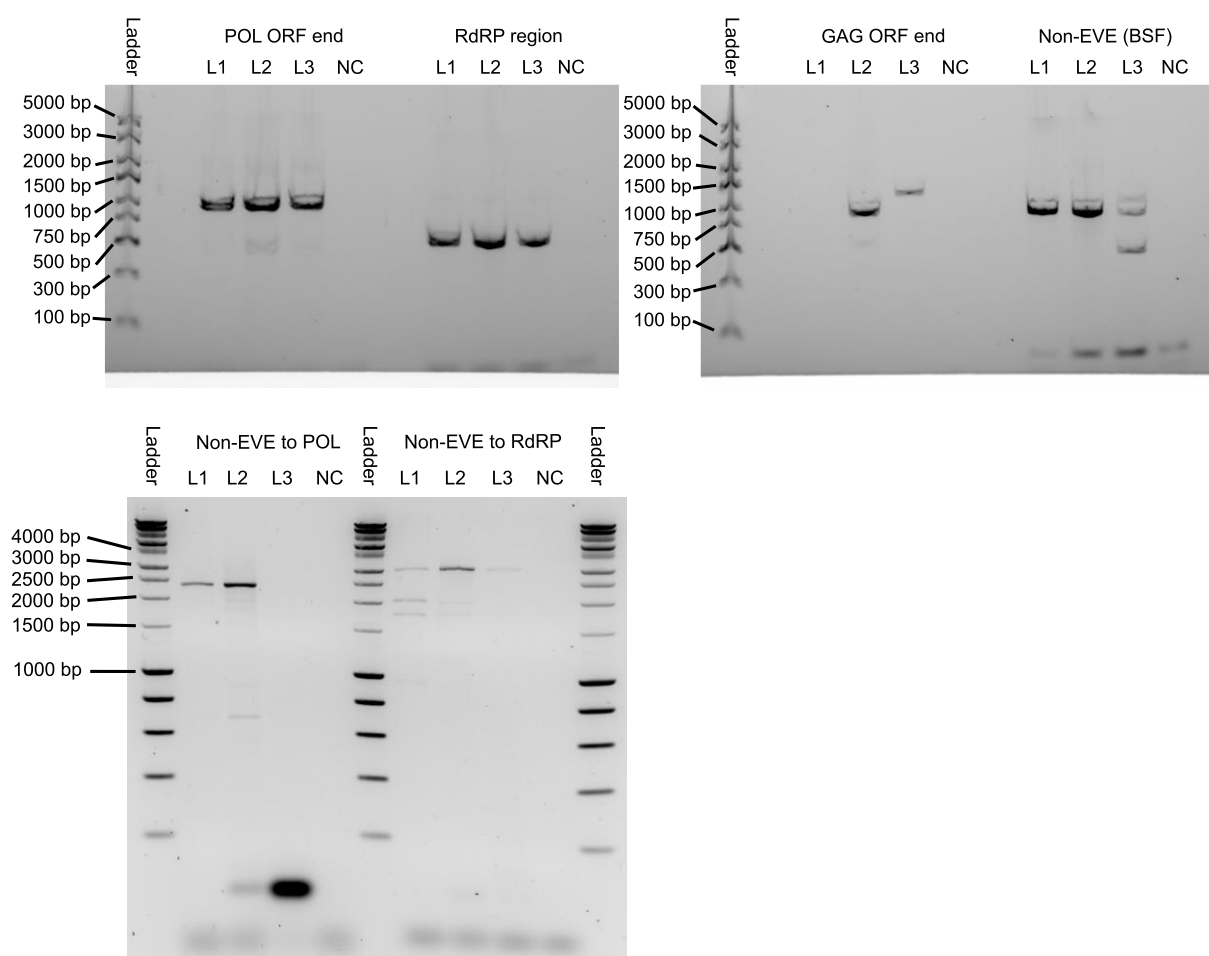


Figure S1 PCR validation of the integration of TotiEVE and expressed TotiEVE-ST within the genome of BSF larvae (L1, L2, L3) originating from three independant rearing facilities. (Top) Primers designed amplified short fragments (~1 kb), firstly at either extremity of the TotiEVE integration site on chromosome 1 according to the 5' to 3' orientation of the HiTV1 polymerase (POL) and capsid encoding (GAG) ORFs. Secondly a shorter fragment (~0.5 kb) to cover the region of the RdRP conserved domain-like sequence appearing in BSF transcriptomes and a region of BSF non-viral related sequence, which was outside of the alignment spanned by the HiTV1 contig, near the GAG ORF-like region of the TotiEVE integration. (Bottom) Primer HITEHNE F was used with HITEP R and HITE R to amplify fragments which overlapped the non-EVE BSF flanking sequence with the TotiEVE POL and RdRP ORFs. These fragments were migrated on a 1.2% agarose gel and were sized according to the SmartLadder (MW-1700-10, Eurogentec, Liège, Belgium). Positive bands on the gel demonstrate that the TotiEVE fragments in Larval L1 to L3 are indeed integrated in the BSF genome.

Table S3 List of sequences resulting from study and related transcriptome list. (a) contigs of H1TV1 which are longer than 5000 nt in length in cDNA and RNA forms. (b) short transcripts which contained/mapped to the RdRp 4-like conserved domain detected in BSF transcriptomes and closely related to TotiEVE site T1. (c) Sequences of short PCR amplified products which targeted different regions of TotiEVE site T1. (d) Sequences of EVE candidates obtained from BGA1, 2 and 3. (e) Regions of BSF genome which flanked the sites on the genome where the sequences of EVE candidates could map. (f) List of transcripts where contigs mapped to H1TV1 contig 1.

[illegible]

[illegible]

[illegible]

[illegible]

[illegible]

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List of transcriptomes where contigs mapped to HITV1 contig 1:

SRR14339796	SRR8242285
SRR14339795	SRR8242284
SRR14339794	SRR8242283
SRR14339793	SRR8242282
SRR14339791	SRR8242281
SRR14339790	SRR8242280
SRR14339789	SRR8242279
SRR14339788	SRR8242277
SRR14339787	SRR6656088
SRR14339786	SRR6656087
SRR14339785	ERR1801998
SRR14339784	ERR1801997
SRR14339783	ERR1801996
SRR14339782	ERR1801995
SRR10233312	ERR1801994
SRR10158821	ERR1801993
SRR8242297	ERR1801992
SRR8242293	ERR1801991
SRR8242292	ERR1801990
SRR8242291	ERR1801989
SRR8242289	ERR1801988
SRR8242288	ERR1801987
SRR8242287	ERR1801986
SRR8242286	ERR1801985