

APPENDIX B

Fungus *Metarhizium robertsii* and neurotoxic insecticide affect the gut immunity and microbiota in Colorado potato beetle

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Table S1. Putative identification of 16S rRNA gene sequences of isolates of bacteria of the genus *Serratia* from the gut of the Colorado potato beetle using the BLAST algorithm compared to similar sequences obtained from GenBank

Isolated numbers	GenBank accession numbers	Nearest match	GenBank accession numbers	Identity (%)
0218	MT256306	<i>Serratia nematodiphila</i>	NR_044385.1	100
3318	MT256278	<i>Serratia quinivorans</i>	NR_037112.1	99.77
10918	MT256307	<i>Serratia marcescens</i>	NR_114043.1	99.86
5618	MT256279	<i>Serratia quinivorans</i>	NR_037112.1	99.78

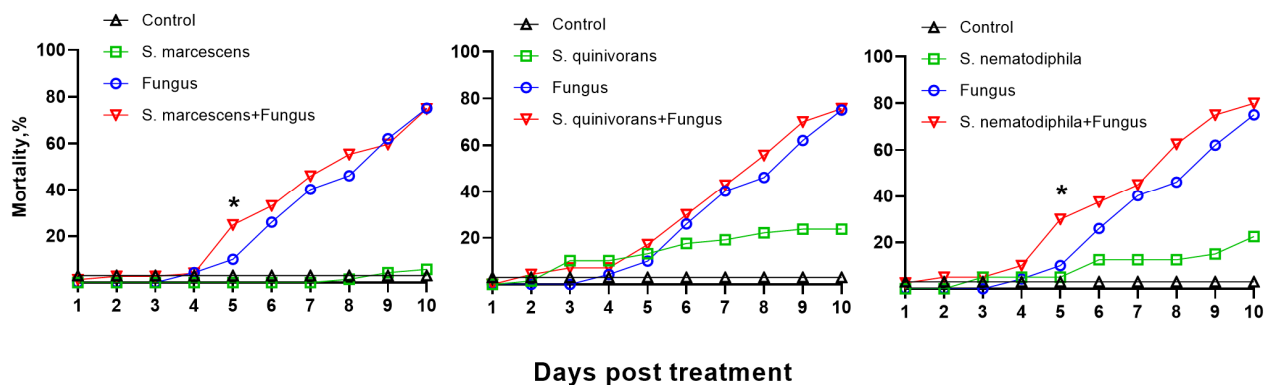


Fig. S1. Mortality dynamics of Colorado potato beetle larvae after topical treatment with *M. robertsii*, oral administration of *Serratia* species and the combined treatment; * - synergistic effect ($\chi^2 > 3.8$, $df = 1$, $P < 0.05$).

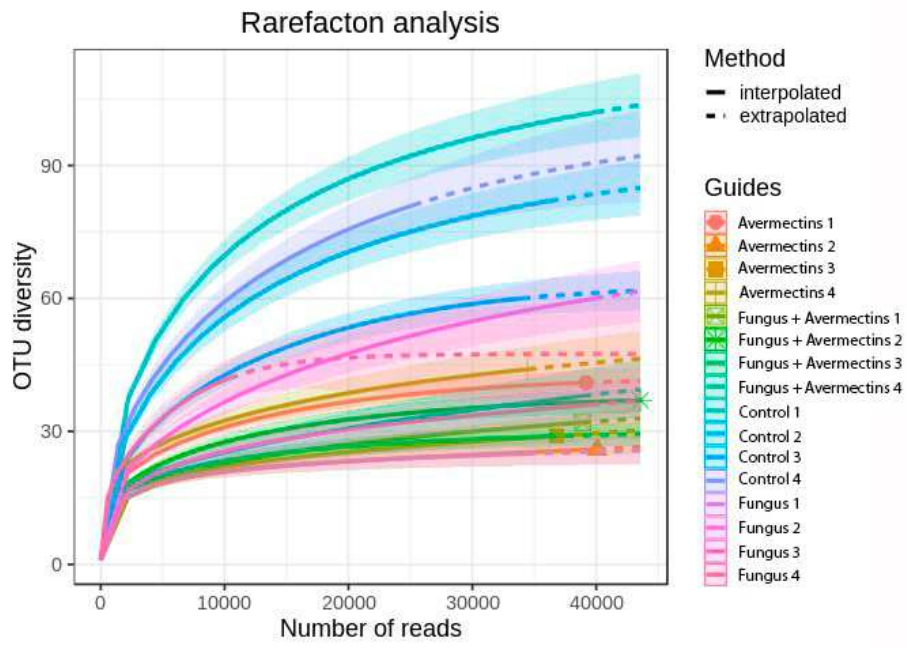


Fig. S2. Rarefaction curves of the OTU number for each sample.

Table S2. List and description of genes and primers sequences used in the qPCR.

Gene Name	NCBI GenBank Accession number	Gene symbol	Primer sequence (5' – 3')	Product size (bp)	PCR efficiency (\pm SD)	Tm in qPCR, °C	Primers source
60S ribosomal protein L4	XM_023165859.1	<i>Rp4</i>	For GAAACGAGCATTGCCCTTCC Rev TCGCTGACACTGTAGGGTTG	119	1,99 \pm 0,03	62	^a modified
ADF-ribosylation factor-like protein 1	XM_023169879.1	<i>ARF1</i>	For CGGTGCTGGTAAAACGACAATATT Rev TGACCTCCCAAATCCCAAACCTT	135	0,97 \pm 0,04	62	^a modified
DorsalDif-like protein	XM_023158121.1	<i>Dorsal-like</i>	For TGTGCGAAAAGGTGGCTAAAG Rev ACTTGGGAGGGTTGGAAGTC	94	1,93 \pm 0,07	62	^b ISaEA
NF-kappaB-like protein	XM_023174540.1	<i>NF-kB-like</i>	For AAGCAGCGGTTTGATTTCGTTT Rev AACTCGTCCAAGTTCTCCAGG	120	1,97 \pm 0,03	62	^b ISaEA
Signal transducer and activator of transcription protein	XM_023165198.1	<i>Stat</i>	For AGGAGCAGAACACAGGGTAC Rev TTTGCCTGGGAATTCTGTTGAC	140	1,93 \pm 0,08	62	^b ISaEA
Attacin_C domain contained protein	XM_023168834.1	<i>Att34</i>	For TGAGAACTCCACAAAATATTCCTTCG Rev CTAAGGGTATGGCAGCAACAAC	98	1,95 \pm 0,04	62	^b ISaEA
Heat shock protein 90	KC556802.1	<i>Hsp90</i>	For GGGTGTAGTCGACTCTGAAGAC Rev AGAGCTCCTCAAACAGTTCCAA	125	1,89 \pm 0,01	62	^b ISaEA
Gut membrane-associated protein Mesh	XM_023158552.1	<i>Mesh^a</i>	For CGCAATGGTTCAACGATATGGT Rev GCTTGGTACGCGACACAGT	124	1,98 \pm 0,01	62	^b ISaEA

^a Shi X.-Q. et al., Validation of reference genes for expression analysis by quantitative real-time PCR in *Leptinotarsa decemlineata*(Say) Shiet al. BMC Research Notes 2013, doi: 10.1186/1756-0500-6-93

^b Primers were designed by Rotskaya U. in programs Primer-BLAST <https://www.ncbi.nlm.nih.gov/tools/primer-blast/>, IDT OligoAnalyser Tool and IDT UNAFold Tool <https://eu.idtdna.com> .