Supplemental Materials:

Figure S1. Survival curves generated from two independent virulence assays where *Drosophila suzukii* was used as the insect host.

Table S1. *SWN* gene homologues and swainsonine production in fungal cultures.

Table S2. Results of assays for effects of swainsonine on virulence of *Metarhizium robertsii* on *Drosophila suzukii*.

Table S3. Oligonucleotide primers used in this study.

Table S1. *SWN* gene homologues and swainsonine production in fungal cultures.

Taxon and strain	Swainsonine production [b]	GenBank accessions [a]						
		swnA	***swnH*1**	***swnH*2**	***swnK***	***swnN***	***swnR***	***swnT***
Alternaria oxytropis Raft River	+	–	KY365741	KY365741	KY365741	KY365741	KY365741	–
Arthroderma benhamiae MYA-4681 CBS 112371	+	XP_003014122	XP_003014119	XP_003014123	XP_003014124	XP_003014121	XP_003016302	XP_003014120
Arthroderma otae (= *Microsporum canis*) MYA-4605 CBS 113480	–	XP_002850893 EEQ28109	XP_002850896 EEQ28112	XP_002850892 EEQ28108	XP_002850891 EEQ28107	XP_002850894 EEQ28110	XP_002850890 EEQ28106	XP_002850895 EEQ28111
Chaetothyrialean endophyte of *Ipomoea carnea* (ICE)	+	KY365740	KY365740	KY365740	KY365740	KY365740	KY365740	KY365740 [c]
Fungal sp. No.14919	n.t.	–	GAW19807	GAW19802	GAW19803	GAW19805	GAW19804	GAW19806
Metarhizium acridum ARSEF 324	+	s.n.a.	s.n.a.	s.n.a.	s.n.a.	s.n.a.	s.n.a.	s.n.a.
Metarhizium acridum ARSEF 3341	+	s.n.a.	s.n.a.	s.n.a.	s.n.a.	s.n.a.	s.n.a.	s.n.a.
Metarhizium acridum CQMa 102	n.t.	XP_007815894	XP_007815893	XP_007815888	XP_007815889	XP_007815891	XP_007815890	XP_007815892

Metarhizium album ARSEF 1941	–	–	–	–	–	–	–	–
Metarhizium anisopliae BRIP 53284	n.t.	(KJK92562)	KJK85334	KJK85339	KJK85338	KJK85336	KJK85337	KJK85335
Metarhizium anisopliae BRIP 53293	n.t.	(KJK74447)	KJK74448	KJK74453	KJK74452	KJK74450	KJK74451	KJK74449
Metarhizium anisopliae ARSEF 549	+	KID61013	KID61012	KID61007	KID61008	KID61010	KID61009	KID61011
Metarhizium anisopliae E6	n.t.	–	KFG82168	KFG82173	KFG82172	KFG82170	KFG82171	KFG82169
Metarhizium brunneum ARSEF 3297	+	XP_014543161	XP_014543162	XP_014543167	XP_014543166	XP_014543164	XP_014543165	XP_014543163
Metarhizium guizhouense ARSEF 977	tr	KID83598	KID83599	KID83604	KID83603	KID83601	KID83602	KID83600
Metarhizium majus ARSEF 297	tr	XP_014575317	XP_014575316	XP_014575311	XP_014575312	XP_014575314	XP_014575313	XP_014575315
Metarhizium robertsii ARSEF 23	+	XP_007824806.2	XP_007824807	XP_007824812	XP_007824811	XP_007824809	XP_007824810	XP_007824808
Metarhizium robertsii ARSEF 2575	+	EXU97977	EXU97978	EXU97983	EXU97982	EXU97980	EXU97981	EXU97979

Nannizzia gypsea CBS 118893 (= *Microsporum gypseum*) MYA-4604	+	XP_003176905	XP_003176902	XP_003176906	XP_003176907	XP_003176904	XP_003176908	XP_003176903
Pseudogymnoascus sp. VKM F-4515 (FW-2607)	n.t.	–	KFY51104	KFY51100	KFY51099	KFY51103	KFY51102	–
Slafractonia leguminicola ATCC 26280	+	–	KY365745	KY365745	KY365746	KY365744	KY365743	KY365742
Trichophyton interdigitale MYA-3108 tinea pedis H6	+	EZF30504	EZF30351	EZF30503	EZF30502	EZF30348	EZF32734	EZF30349
Trichophyton equinum CBS 127.97 (= *Trichophyton tonsurans*) MYA-4606	+	EGE01984	EGE01987	EGE01983	EGE01982	EGE01985	EGE02341	EGE01986
Trichophyton rubrum MYA-4607 CBS 118892	tr	XP_003238868	XP_003238865	XP_003238869	XP_003238870	XP_003238867	XP_003238559	XP_003238866
Trichophyton interdigitale MR816	n.t.	KDB22224	KDB22220	KDB22225	KDB22226	KDB22223	KDB22462	KDB22222
Trichophyton rubrum CMCC(F)T1i	n.t.	OAL63128	OAL63131	OAL63127	OAL63126	OAL63129	LHPM01000018	OAL63130

Trichophyton rubrum CBS 289.86	n.t.	EZF58568	EZF58564	EZF58569	EZF58570	EZF58567	EZF48315	EZF58566
Trichophyton rubrum CBS 735.88	n.t.	EZG01519	EZG01515	EZG01520	EZG01521	EZG01518	EZG01771	EZG01517
Trichophyton rubrum CBS 100081	n.t.	EZF37321	EZF37322	EZF37317	EZF37323	EZF37320	AOKT01000327	EZF37319
Trichophyton soudanense CBS 452.61	n.t.	EZF69146	EZF69142	EZF69147	EZF69148	EZF69154	EZF69504	EZF69144
Trichophyton tonsurans CBS 112818	n.t.	EGD97141	EGD97144	EGD97140	EGD97139	EGD97142	EGD97826	EGD97143
Trichophyton verrucosum HKI 0517	n.t.	XP_003020761	XP_003020758	XP_003020762	XP_003020763	XP_003020760	XP_003022179	XP_003020759
Trichophyton violaceum CMCC(F)T3l	n.t.	OAL75149	OAL75146	OAL75150	OAL75151	OAL75148	OAL69278	OAL75147

[a] Genes are identified by GenBank accession numbers either for the predicted protein or the assembled genomic sequence bearing the genes. Symbols and abbreviations: – = not found, s.n.a. = sequence not available.

[b] Symbols and abbreviations: + = detected at > 10 µg/g, tr = trace detected at ≤ 10 µg/g, – = undetected, n.t. = not tested. Literature reports production of swainsonine by the respective strains of ICE (COOK *et al.* 2013) and *A. oxytropis* (GRUM *et al.* 2013), and by another isolate of *S. leguminicola* (HARRIS *et al.* 1988b).

[c] The *swnT* homologue in ICE is probably a pseudogene.

Table S2. Results of assays for effects of swainsonine on virulence of *Metarhizium robertsii* on *Drosophila suzukii*.

Assay ID	Strain [a]	Swainsonine (Strain/WT) [b]	Mean Survival Time (days)		Hypothesis test [c]	
			Reference (WT applied)	Strain applied [c]	Log-rank	Wilcoxon
1	Δ*swnK*	0	6.90	6.09	0.096	0.187
2	Δ*swnK*	0	6.74	**<u>5.75</u>**	**<u>0.007</u>**	**<u>0.010</u>**
3	Δ*swnK*	0	6.11	**7.21**	**0.001**	**0.001**
1	*swnK*-C18	0.25	6.90	**7.60**	0.054	**0.011**
2	*swnK*-C18	0.25	6.74	6.60	0.517	0.660
1	*swnK*-C5	2	6.90	**7.72**	**<.0001**	**<.0001**
2	*swnK*-C5	2	6.74	**8.30**	**<.0001**	**<.0001**
3	*swnK*-C5	2	6.11	**7.71**	**<.0001**	**<.0001**
3	*swnK*-C12	2	6.11	**8.71**	**<.0001**	**<.0001**
3	*swnK*-C19	4	6.11	**6.93**	0.061	**0.005**

[a] *Metarhizium robertsii* strain, where WT = wild type ARSEF 2575, Δ*swnK* = the *swnK* deletion mutant, and *swnK*-C18, *swnK*-C5, *swnK*-C12 and *swnK*-C19 are four independent transformants of Δ*swnK* with *swnK* from WT (complemented strains).

[b] Approximate ratio of swainsonine produced in cultures of the respective strain over swainsonine produced in cultures of wild-type *M. robertsii* ARSEF 23 (WT).

[c] Bold-face type indicates significant difference in mean survival times of insects treated with the respective mutant compared to those treated with WT. Underline indicates significant reduction in mean survival time compared to those treated with WT.

Table S3. Oligonucleotide primers used in this study.

Name	Sequence (5` to >3`) [a]	Remarks
SWKO-AF	GGGAAAGdUGAAGATGCCGGGCTCAGACT	*swnK* knockout construct, flank A
SWKO-AR	ATCATCCdUGTTGACCTTTTCGCTGGGAGAC	*swnK* knockout construct, flank A
SWKO-BF	ACTTGTGGdUAACGATCCGGCTATACACGAG	*swnK* knockout construct, flank B
SWKO-BR	GGAGACAdUTCCGCTTGTGCCCTGAGAG	*swnK* knockout construct, flank B
BarExprSF	AGGATGAdUAGAAGATGATATTGAAGGA	*bar* cassette
BarExprSR NOS	ACCACAAGdUCATGTTTGACAGCTTATCAT	*bar* cassette
SW-T2F	CGCGCTCCATCGACCTCATT	Anneals outside the region targeted for homologous integration
SW-T2R	CATCACGCAAGGGGCTGTC	Anneals outside the region targeted for homologous integration
ptrpc80R	CCGCCTGGACGACTAAACC	Anneals at the 5` end of the *trpC* promoter of the *bar* expression cassette
bar848F	ACTGGCATGACGTGGGTTTCTGG	Anneals at the 3` end of the *bar* ORF
SW-T3F	TGATCGTCTGGTGGCGTATTGTGT	Anneals inside the targeted region.
SW-T3R	ATCGGACAGGCGCTTGAGAACTAT	Anneals inside the targeted region.
SW-CF	GGGAAAGdUTTTGAAGATGGGCTGGTG	complementation
SW-CR	GGAGACAdUCAGGCTTGGGAACAGAATGAGACC	complementation

[a] dU = internal deoxyuracil, which is coded as /ideoxyU/ by Integrated DNA Technologies.

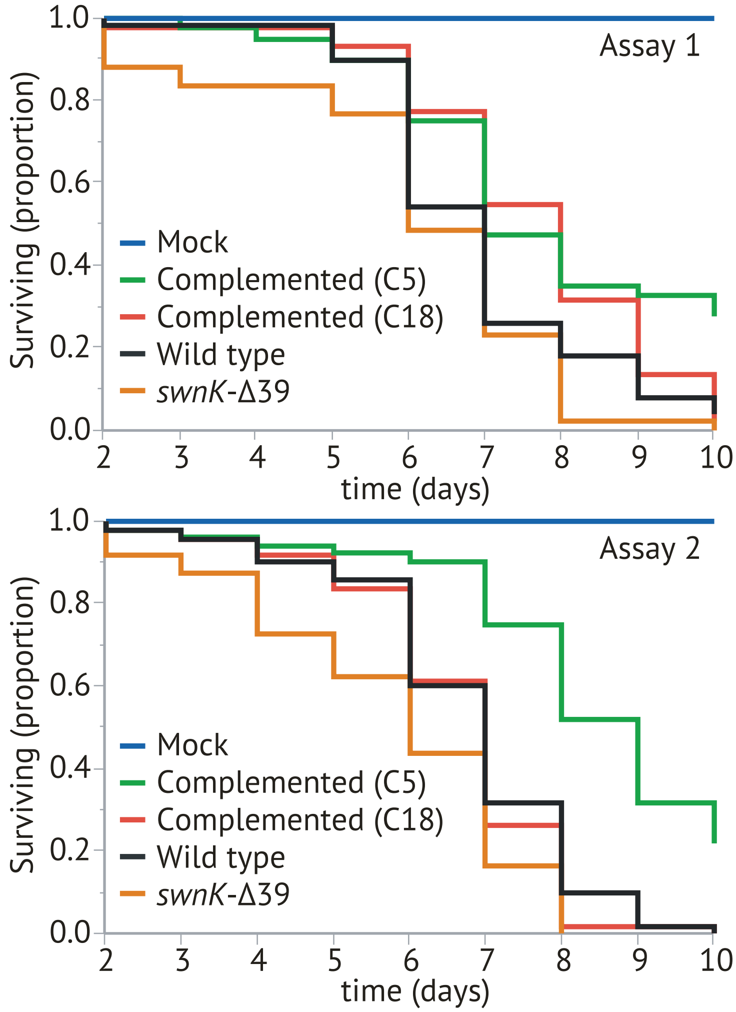

Figure S1 Survival curves generated from two independent virulence assays where *Drosophila suzukii* was used as the insect host. Cox's proportional hazards presented here were generated using the wild type as the control and represent the increase of risk associated to the inoculation of a mutant compared to the WT. Thus, a CPH of 1 indicates no risk change compared to the WT; a CPH of 2 indicates a mortality risk twice that inflicted by the WT in the same assay conditions.