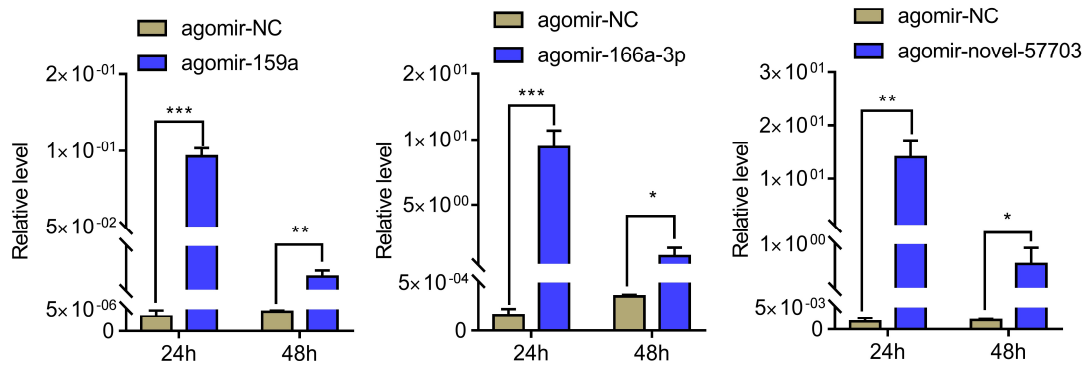


1				
2	A	aly-miR159a-3p	TTTGGATTGAAGGGAGCTCTA	21
		ath-miR159a	TTTGGATTGAAGGGAGCTCTA	21
3		bdi-miR159a-3p	CTTGGATTGAAGGGAGCTCT.	20
		bra-miR159a	TTTGGATTGAAGGGAGCTCTA	21
		cas-miR159a	TTTGGATTGAAGGGAGCTCT	21
		cme-miR159a	TTTGGATTGAAGGGAGCTCTA	21
4		cpa-miR159a	TTTGGATTGAAGGGAGCTCTA	21
		csi-miR159a-3p	TTTGGATTGAAGGGAGCTCTA	21
		fve-miR159a-3p	TTTGGATTGAAGGGAGCTCTA	21
5		gma-miR159a-3p	TTTGGATTGAAGGGAGCTCTA	21
		hbr-miR159a	TTTGGATTGAAGGGAGCTCTA	21
		htu-miR159a	TTTGGATTGAAGGGAGCTCTA	21
		hvu-miR159a	TTTGGATTGAAGGGAGCTCTG	21
6		mdm-miR159a	CTTGGATTGAAGGGAGCTCC.	20
		mes-miR159a-3p	TTTGGATTGAAGGGAGCTCTA	21
		mtr-miR159a	TTTGGATTGAAGGGAGCTCTA	21
7		osa-miR159a.1	TTTGGATTGAAGGGAGCTCTG	21
		pta-miR159a	.TTTGGATTGAAGGGAGCTCCA	20
		ptc-miR159a	TTTGGATTGAAGGGAGCTCTA	21
8		pvu-miR159a.1	TTTGGATTGAAGGGAGCTCTA	21
		sbi-miR159a	TTTGGATTGAAGGGAGCTCTG	21
		sof-miR159a	TTTGGATTGAAGGGAGCTCTG	21
		ssp-miR159a	TTTGGATTGAAGGGAGCTCTG	21
9		tae-miR159a	TTTGGATTGAAGGGAGCTCTG	21
		zma-miR159a-3p	TTTGGATTGAAGGGAGCTCTG	21
10	B	zma-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
		aly-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
		ata-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
		ath-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
11		bdi-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
		csi-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
		gma-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
		osa-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
		stu-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
		vca-miR166a-3p	TCGGACCAGGCTTCATTCCCC	21
12	C	Brassica napus	TCCAGCTGGGTTTGGATTGAAGGGAGCTCTACTCAACTGA	40
		Brassica oleracea	TCCAGCTGGGTTTGGATTGAAGGGAGCTCTACTCAACTGA	40
		Raphanus sativus	TCCAGCTGGGTTTGGATTGAAGGGAGCTCTACTCAACTGA	40
13		ath-miR-159a	TTTGGATTGAAGGGAGCTCTA.....	21
14	D	Brassica napus	TCCAGCTGGGTCGGACCAGGCTTCATTCCCCCTCAACTGA	40
		Brassica oleracea	TCCAGCTGGGTCGGACCAGGCTTCATTCCCCCTCAACTGA	40
		Raphanus sativus	TCCAGCTGGGTCGGACCAGGCTTCATTCCCCCTCAACTGA	40
		ath-miR-166a-3p	TCGGACCAGGCTTCATTCCCC.....	21
15	E	Brassica_napus	TCCAGCTGGGCGAGAATTCTGGAAGAGCTC.TCAACTGA	39
		Brassica_oleracea	TCCAGCTGGGCGAGAATTCTGGAAGAGCTC.TCAACTGA	39
		Raphanus_sativus	TCCAGCTGGGCGAGAATTCTGGAAGAGCTCTCAACTGA	40
16		ath-novel-7703-5p	CGAGAATTCTGGAAGAGCT.....	20

17 **Figure S1. Conservation analysis of plant-derived miRNAs. A and B.** Homologous
 18 sequences of miR159a (A) and miR166a-3p (B) in diverse plant lineages acquired from
 19 miRBase 22. **C-E.** Sequence alignments of miR159a (C) and miR166a-3p (D) and

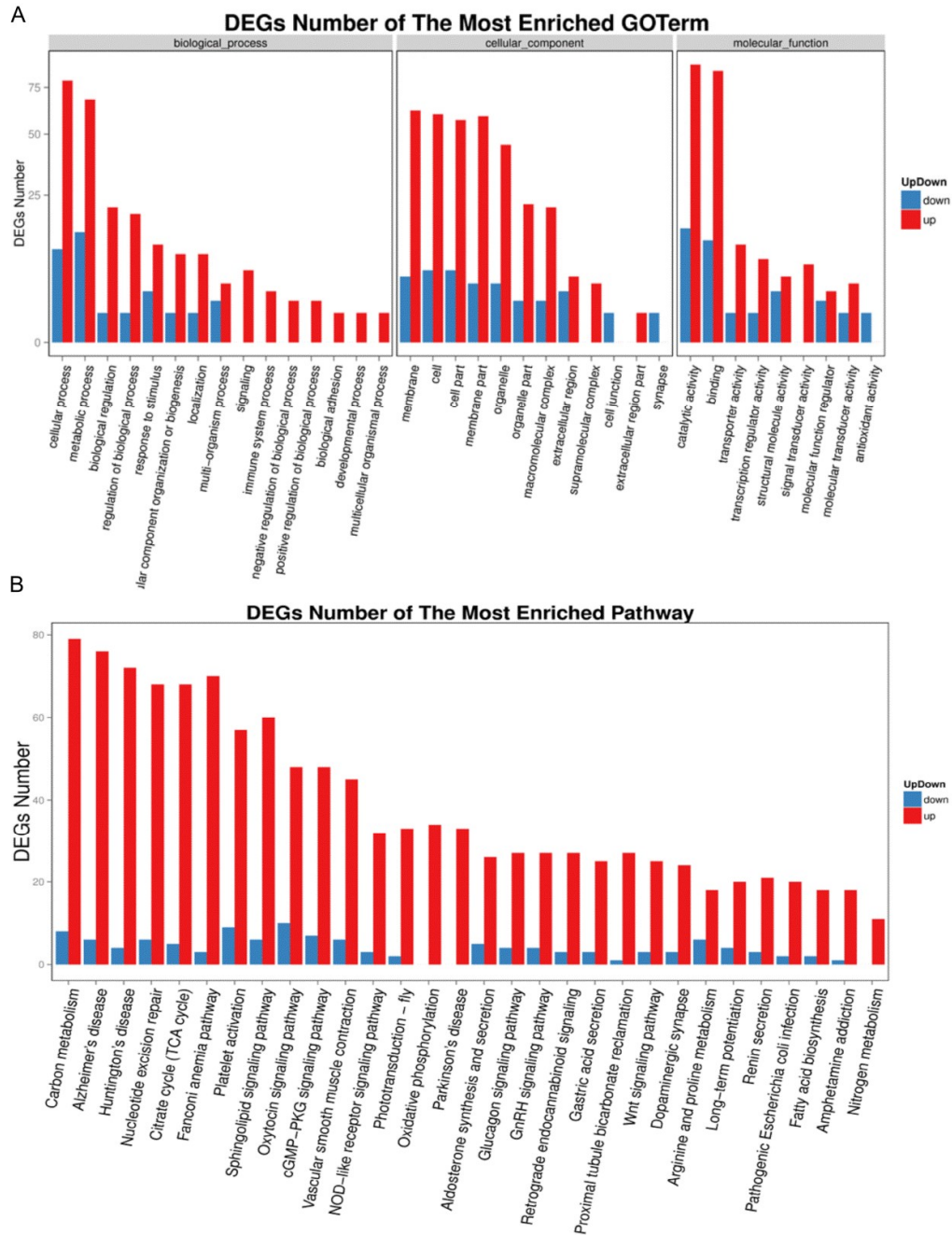
20 novel-7703-5p (E) cloned from four host plants of *P. xylostella* using stem-loop PCR.
21 The completely matched sequences were in dark blue background, while the vector
22 sequences were in pink background.

23

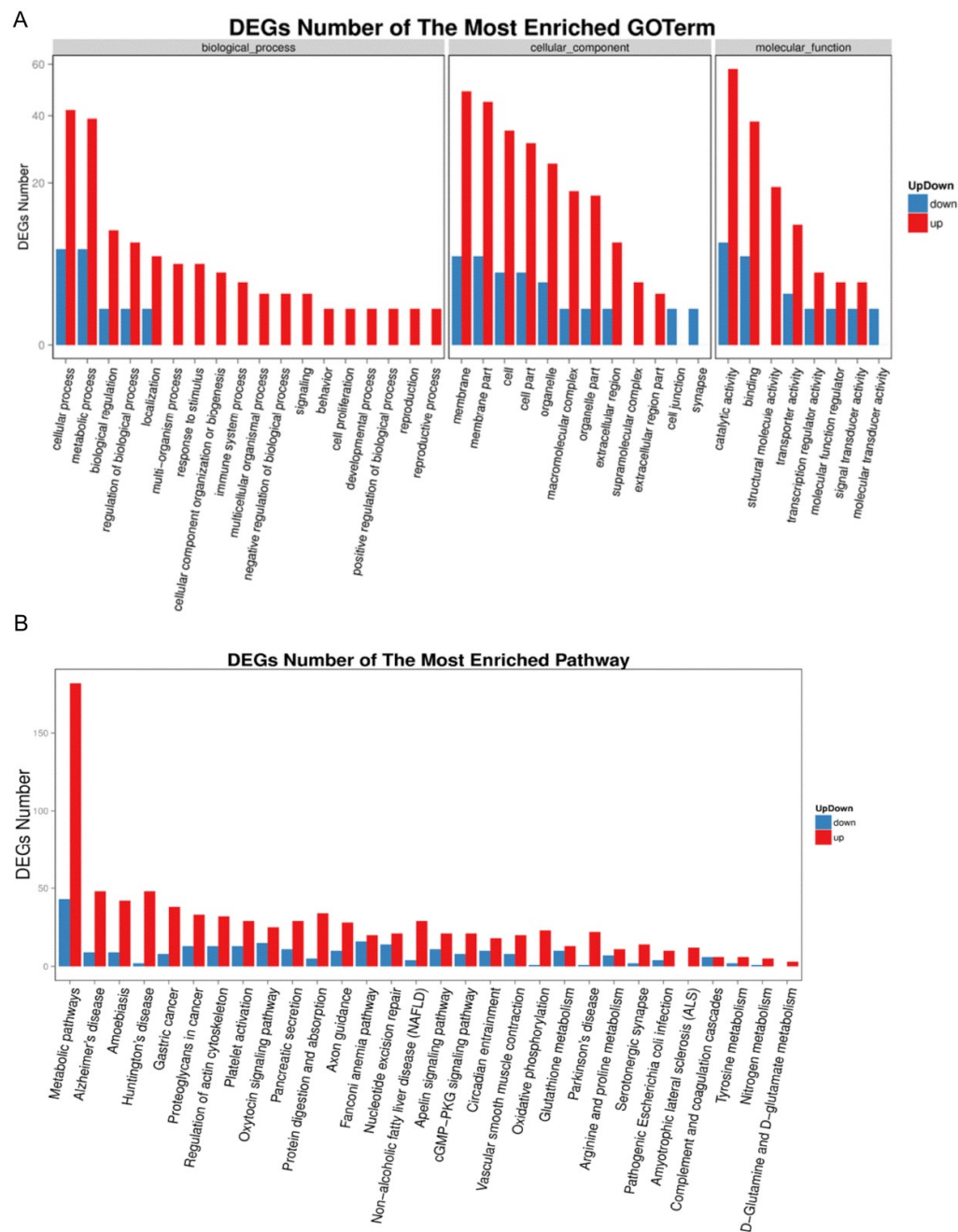


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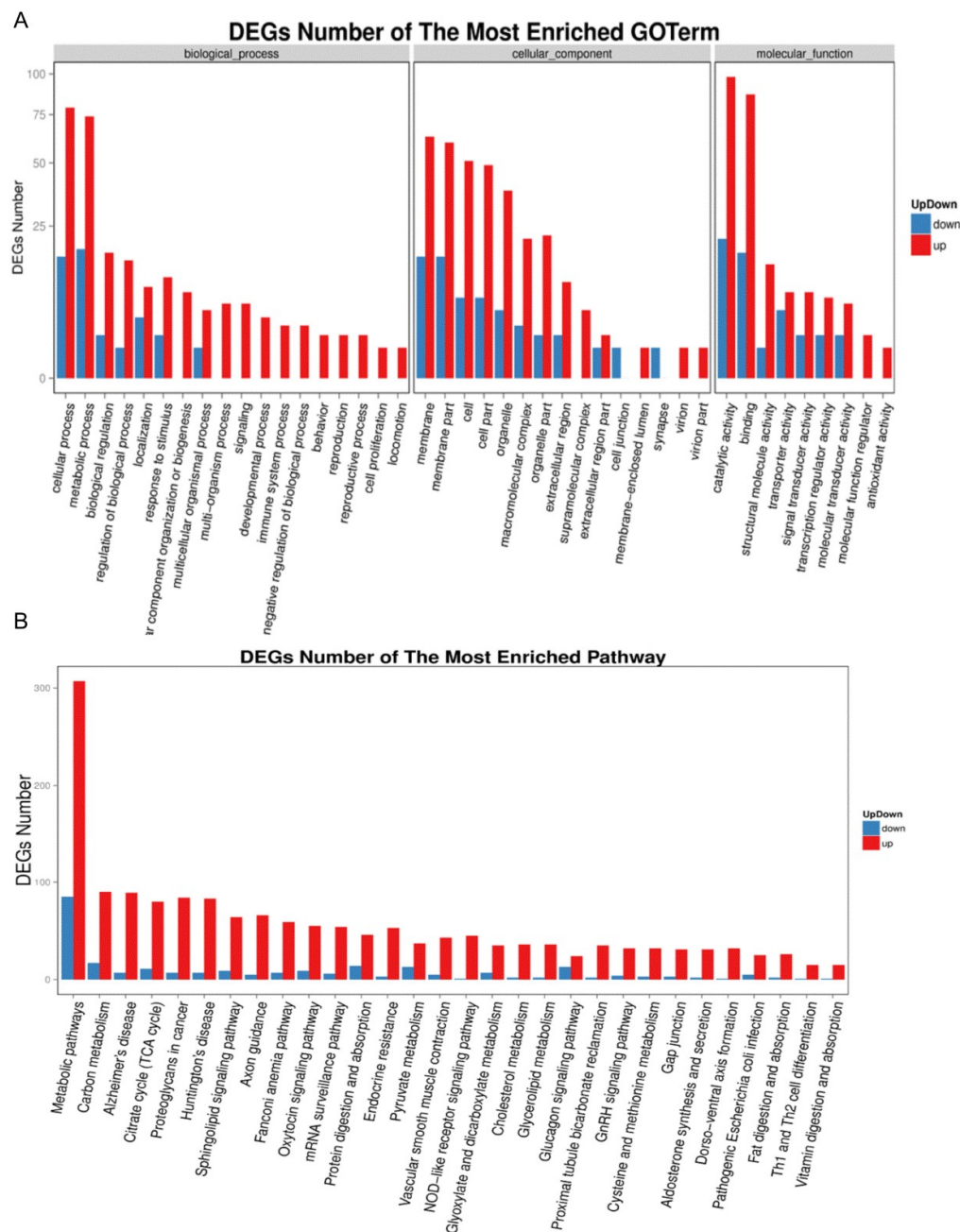
25 **Figure S2. Expression of three miRNAs at different time points after agomir**
 26 **treatments. A-C.** Relative abundance of miR159a (A), miR166a-3p (B) and novel-
 27 7703-5p (C) in hemolymph of *P. xylostella* at 24 h and 48 h after injection with
 28 corresponding miRNA agomir. Data were presented as mean $\pm$ SD (Student's *t* test, n
 29 = 3, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$).



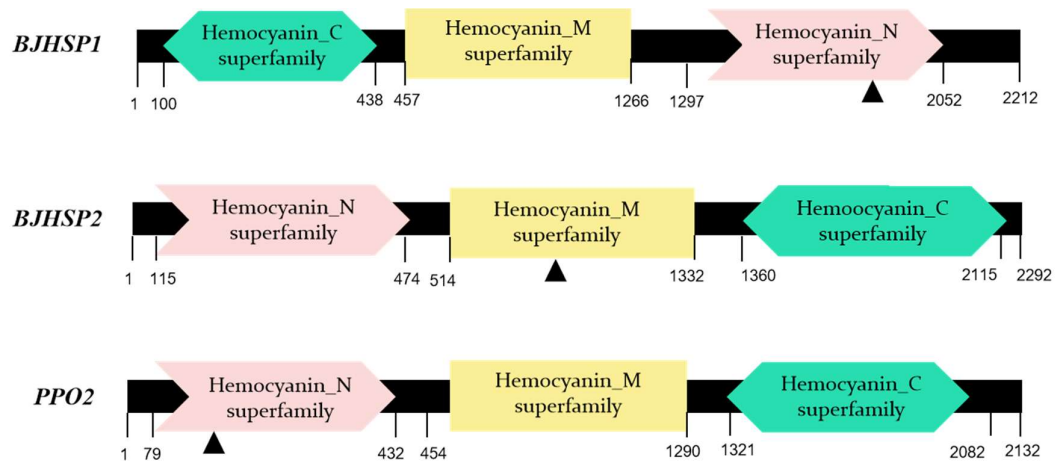
30 **Figure S3. Enrichment analysis of differentially expressed genes in agomir-159a**
 31 **treatment. A and B.** The most enriched GO terms (A) and KEGG pathways (B) for
 32 DEGs in agomir-159a treatment compared with the control.



33 **Figure S4. Enrichment analysis of differentially expressed genes in agomir-166a-**
 34 **3p treatment. A and B.** The most enriched GO terms (A) and KEGG pathways (B)
 35 for DEGs in agomir-166a-3p treatment compared with the control.

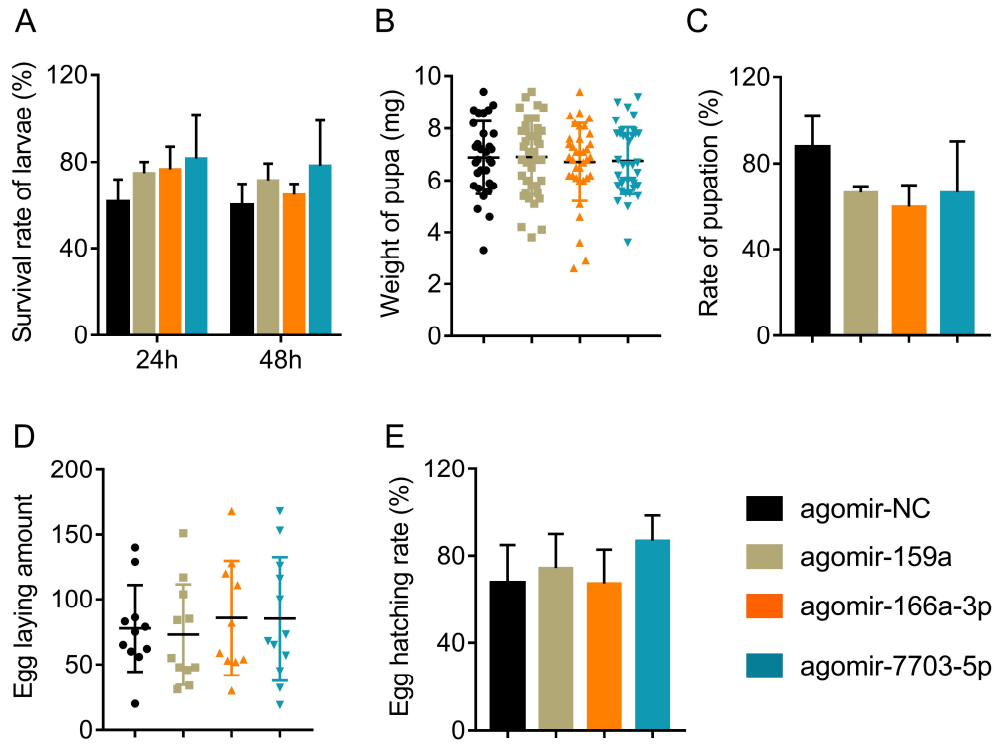


36 **Figure S5. Enrichment analysis of differentially expressed genes in agomir-7703-**
 37 **5p treatment. A and B.** The most enriched GO terms (A) and KEGG pathways (B)
 38 for DEGs in agomir-7703-5p treatment compared with the control.

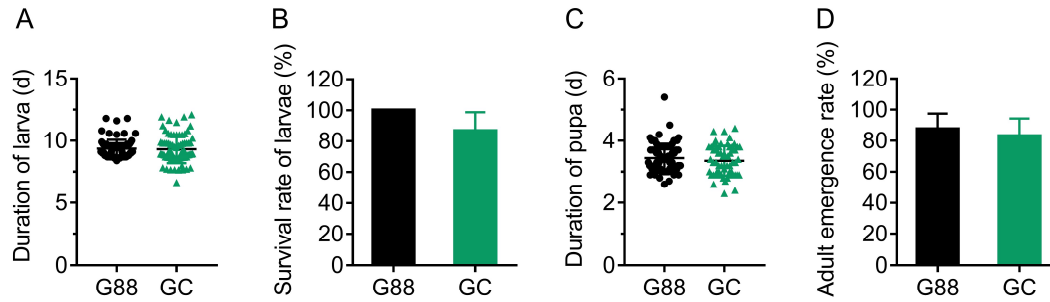


39 **Figure S6. Predicted conserved domains of *BZHSP1*, *BZHSP2* and *PPO2*.** The
40 numbers denote the locations of predicted conserved domains on target genes. The
41 binding sites of miR159a, miR166a-3p and novel-7703-5p were marked using black
42 triangles.

43



44 **Figure S7. The biological parameters of the G88 strain that were not affected by**
 45 **treatments with miRNA agomirs.** **A.** Survival rate of G88 larvae at 24 h and 48 h
 46 post-injection of miRNA agomir. Data were presented as mean $\pm$ SD ($n = 3$, Student's
 47 t test.) **B.** Weight of pupa after miRNA agomir treatment. Data were presented as mean
 48 $\pm$ SD ($n = 31 - 40$, Student's t test.) **C.** Rate of pupae after miRNA agomir treatment.
 49 Data were presented as mean $\pm$ SD ($n = 3$, Student's t test.) **D.** Total amount of eggs
 50 laid by single pair of male and female adults during the first three days. **E.** Hatching
 51 rate of eggs. Data were presented as mean $\pm$ SD ($n = 10 - 12$, Student's t test.)



52 **Figure S8. The biological parameters showing no difference between the G88 and**
53 **GC strains. A.** Duration of G88 and GC larva. Data were presented as mean $\pm$ SD
54 (Student's *t* test, $n = 78 - 90$). **B.** Survival rate of G88 and GC larva. Data were presented
55 as mean $\pm$ SD (Student's *t* test, $n = 3$). **C.** Duration of G88 and GC pupae. Data were
56 presented as mean $\pm$ SD (Student's *t* test, $n = 61 - 78$). **D.** Eclosion rate of G88 and GC
57 pupae. Data were presented as mean $\pm$ SD (Student's *t* test, $n = 3$).

58 **Table S1. Sequences of primer pairs, miRNA agomirs and target binding sites**

Description	Names	Sequences (5'-3')
	ath-miR159a-RT	CTCAACTGGTGTCGTGGAGTCGGCAATTC AGTTGAGTAGAGCTC
Reverse transcription ¹	ath-miR166a-3p-RT	CTCAACTGGTGTCGTGGAGTCGGCAATTC AGTTGAGGGGGAATG
	ath-novel-7703-5p-RT	CTCAACTGGTGTCGTGGAGTCGGCAATTC AGTTGAGAGCTCTTC
	ath-miR159a-F	ACACTCCAGCTGGGTTTGGATTGAAGG
	ath-miR166a-3p-F	ACACTCCAGCTGGGTCGGACCAGGCTT
miRNA	ath-novel-7703-5p-F	ACACTCCAGCTGGGCGAGAATTTCTG
qRT-PCR ²	universal Reverse	TGGTGTCGTGGAGTCG
	U6-F	CTCGCTTCGGCAGCACA
	U6-R	AACGCTTCACGAATTTGCGT
	Px006820-F	GACCTCAACACCTACTTC
mRNA qRT-PCR	Px006820-R	CTCCTTGTCCATCTTCTC
	Px007031-F	ATCAAGAAGACCGACAAG
	Px007031-R	CGTGGAAGTAGTAGTAGTAG

	Px002274-F	GCCTGATACCAAGAATGTA
	Px002274-R	CGATG TTCCTCTTCTAAGT
	EF1-F	GCCTCCCTACAGCGAATC
	EF1-R	CCTTGAACCAGGGCATCT
	ath-miR159a	UUUGGAUUGAAGGGAGCUCUA
miRNA	ath-miR166a-3p	UCGGACCAGGCUUCAU UCCCC
agomir	ath-novel-7703-5p	CGAGAAUUUCUGGAAGAGCU
	agomir-NC	UUCUCCGAACGUGUCACGUTT
	Px006820-WT	CGGCTTCCCCTTCGACCGC
	Px006821-Mut	CAATCTCTTTCCTACTTAT
Target	Px007031-WT	CATGACATTCTTGGTCCGC
binding sites	Px007031-Mut	CGCAGCATTTC CAACTTAC
	Px002274-WT	CCTTTCAGAGATATTCC
	Px002274-Mut	AGCGACTCTCGACGACT

¹ The miRNA-specific reverse primer was synthesized by adding the reverse complementary sequence of the last 8 bp (underlined) of mature miRNA to the 3' terminal of the common stem-loop structure.

² The remainder of the sequence of mature miRNA (underlined) was added with a universal adapter (*italic*) at the 5' end as the forward primer paired with a universal reverse primer.