

Small RNAs and Cross-Kingdom RNAi in Plant-Microbial Interaction

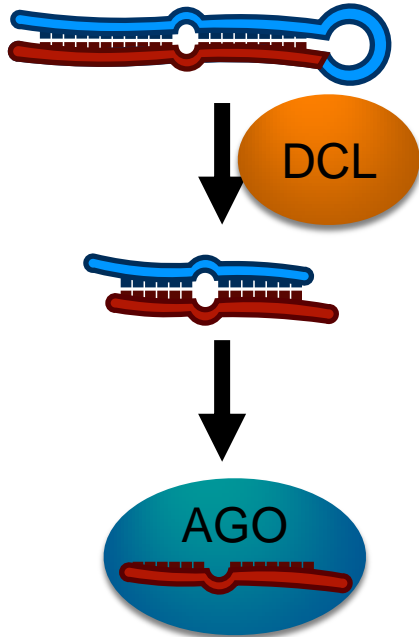
Hailing Jin

**Department of Plant Pathology & Microbiology
Institute for Integrative Genome Biology**

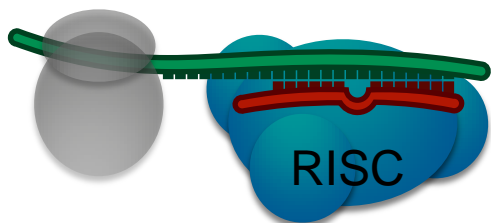
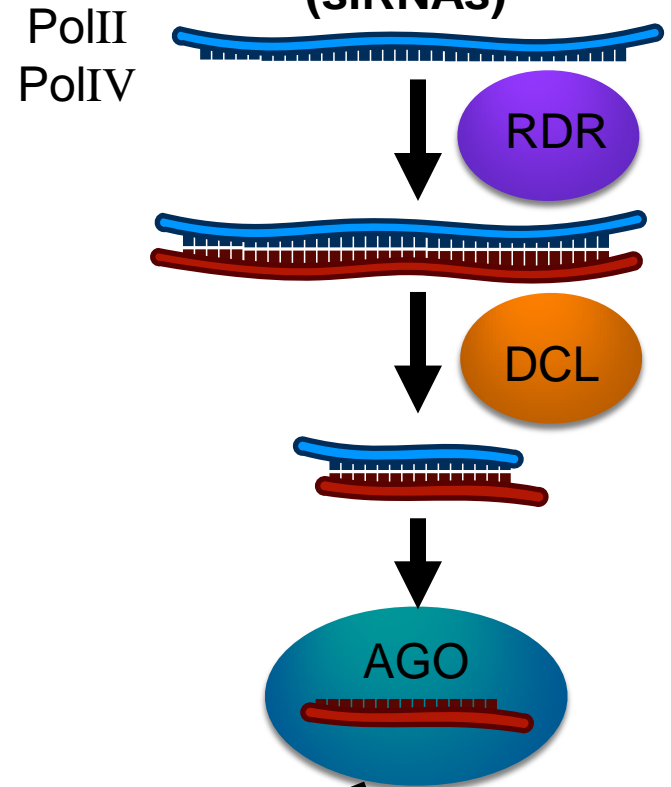


Small RNAs

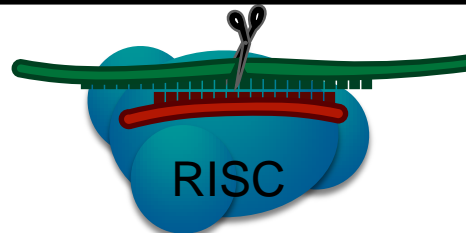
microRNAs
(miRNAs)



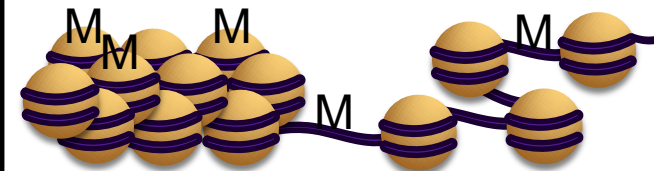
Small interfering RNAs
(siRNAs)



Translation Inhibition



mRNA Degradation



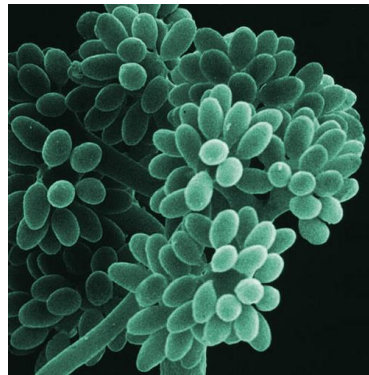
Chromatin Modification

Small RNAs and Cross-Kingdom RNAi in plant-pathogen interactions

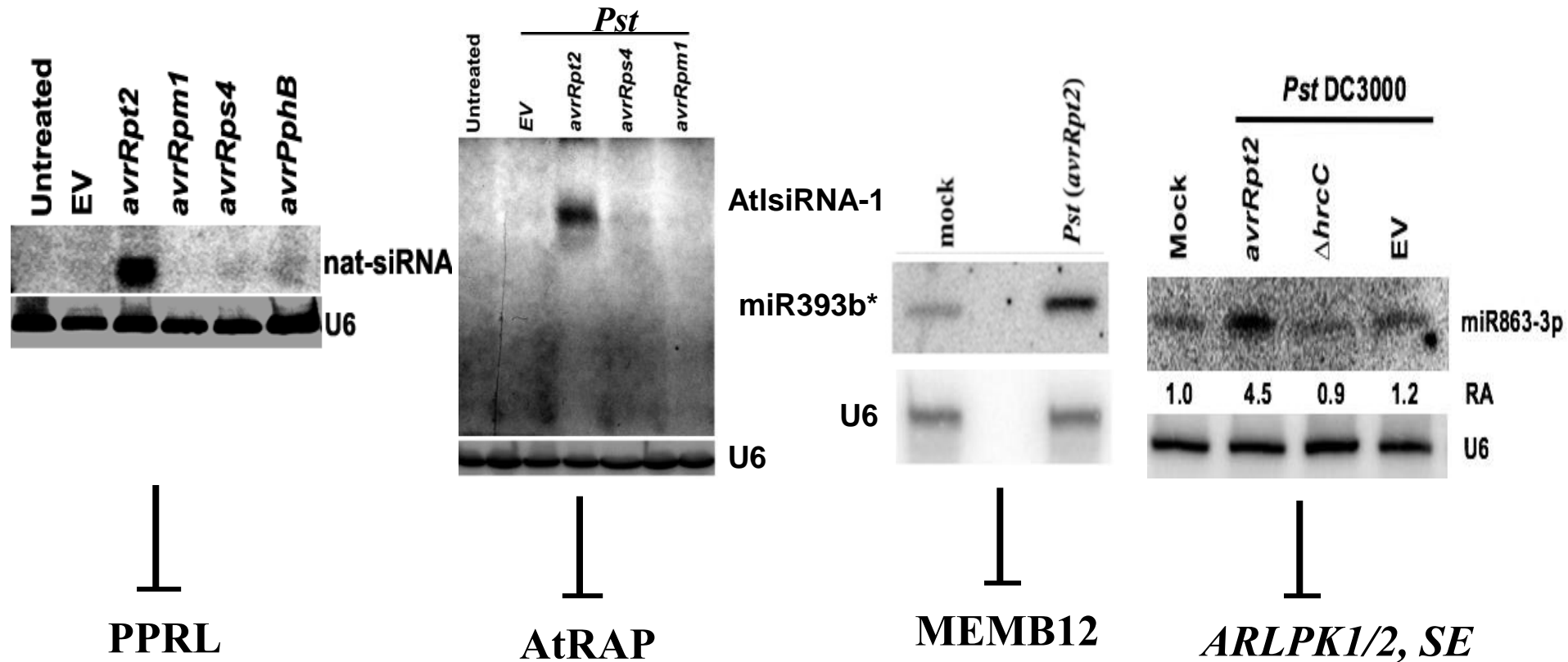
- **Host endogenous small RNAs and RNAi machinery**



- **Pathogen-derived small RNAs and Cross-Kingdom RNAi**



Pathogen-induced host small RNAs regulate immunity by suppressing negative regulators of plant defense



Negative Regulators of Plant Defense

Katiyar-Agawal et al., PNAS 2006

Katiyar-Agawal et al., Genes & Dev 2007

Zhang et al., Molecular Cell 2011

Niu et al., Nature Comm 2016

Fungal small RNAs in host pathogen interaction

Fungal Pathogen

Botrytis cinerea



Gray mold disease

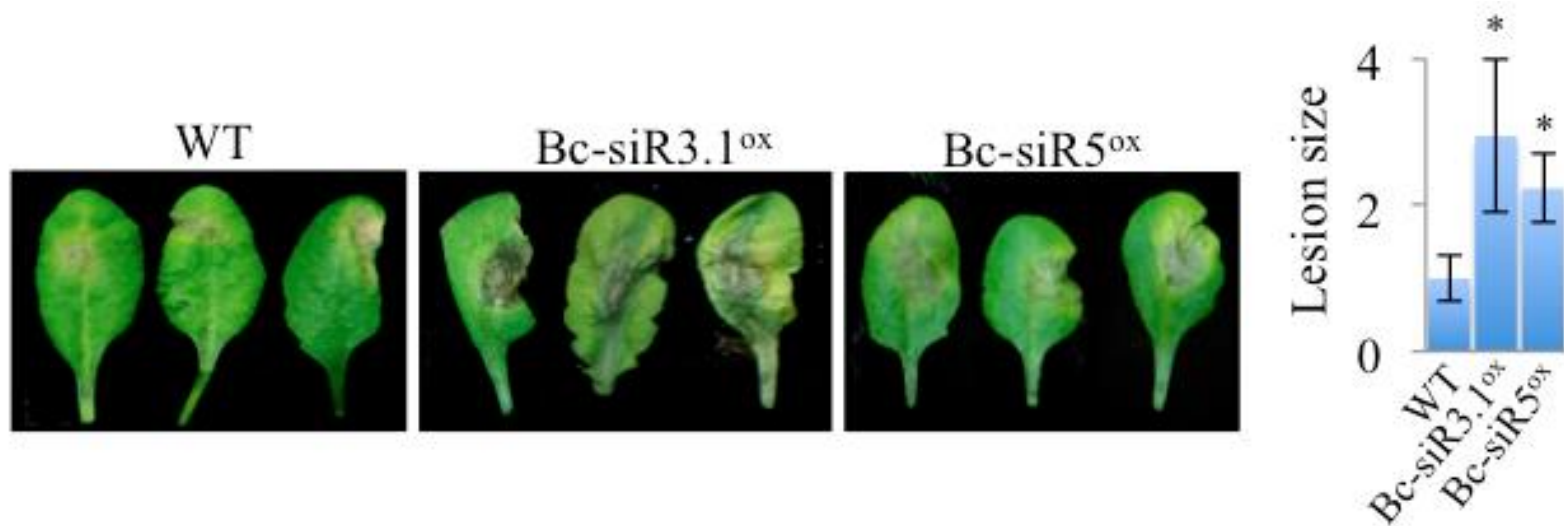
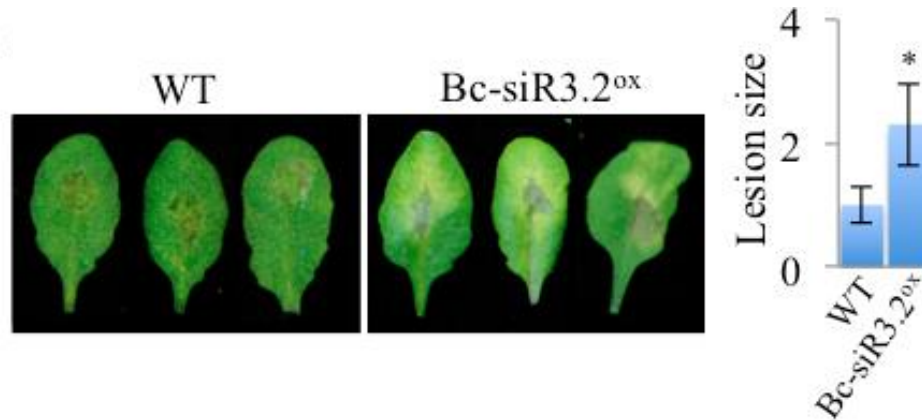


Plant Hosts



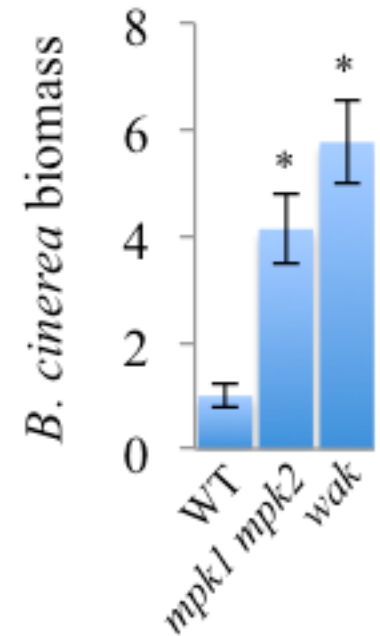
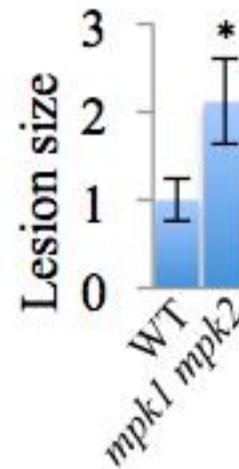
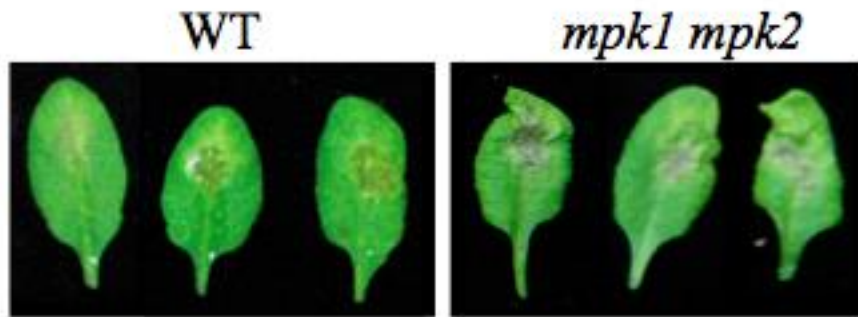
- Super aggressive
Infect more than 200 species including almost all the fruits and vegetables
- Has its own sRNA machinery

Transgenic plants expressing *Bc*-siRNAs display enhanced disease susceptibility to *B. cinerea*

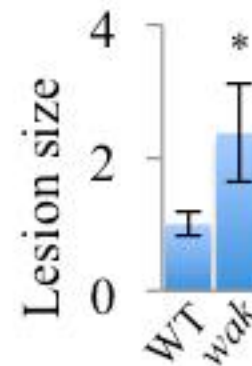
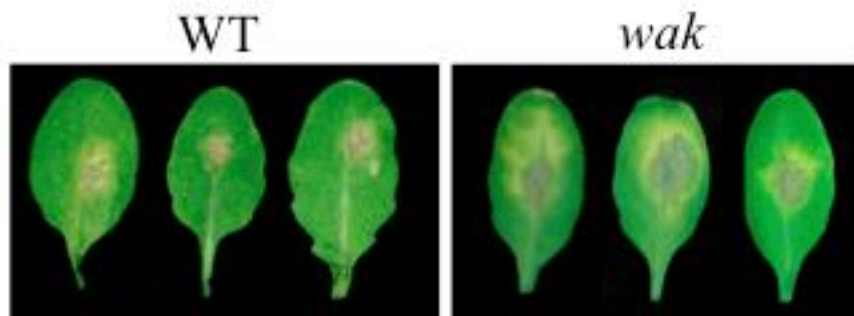


Arabidopsis target genes of *Bc*-siRNAs are important for host defense against *Botrytis*

Bc-siR3.2 Targets MPK1 and MPK2



Bc-siR5 Target WAK



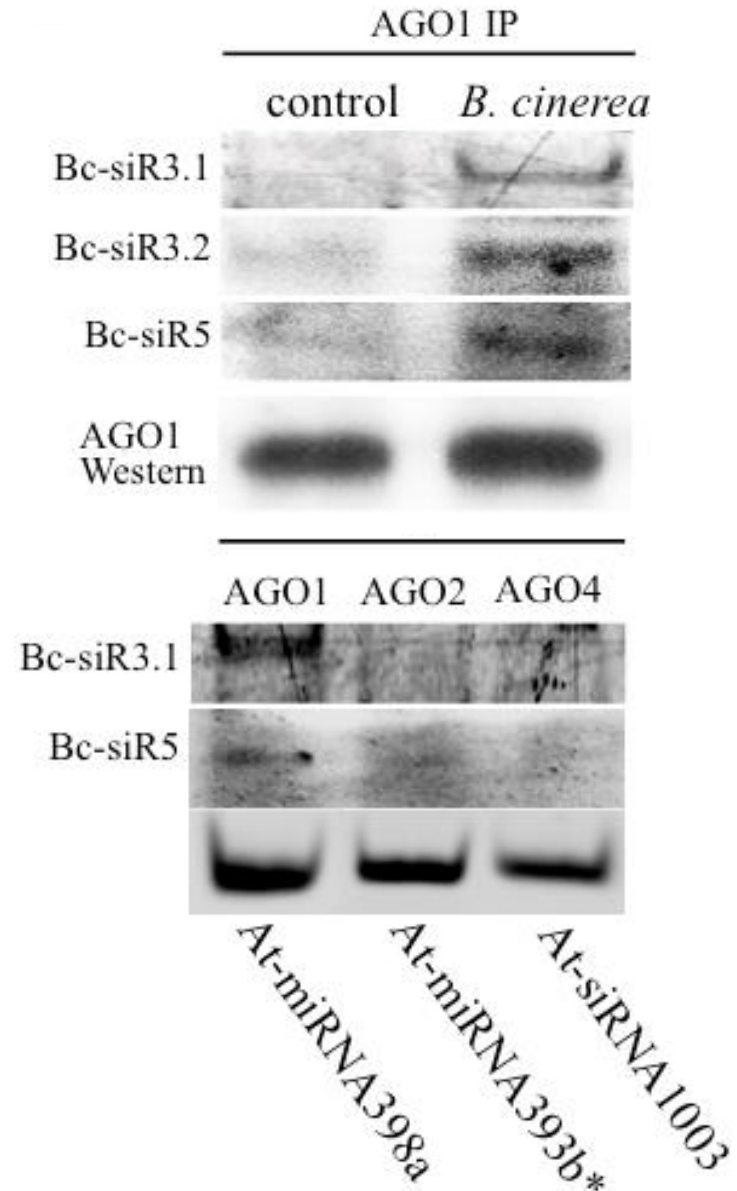
B. cinerea siRNAs hijack Arabidopsis AGO1 during infection to silence host genes

Bc-siR3.1 5`UUGUGGAUCUUGUAGGUGGGC
Bc-siR3.2 5`UACAUUGUGGAUCUUGUAGGU
Bc-siR5 5`UUUGACUCGAAUGUAUACUU

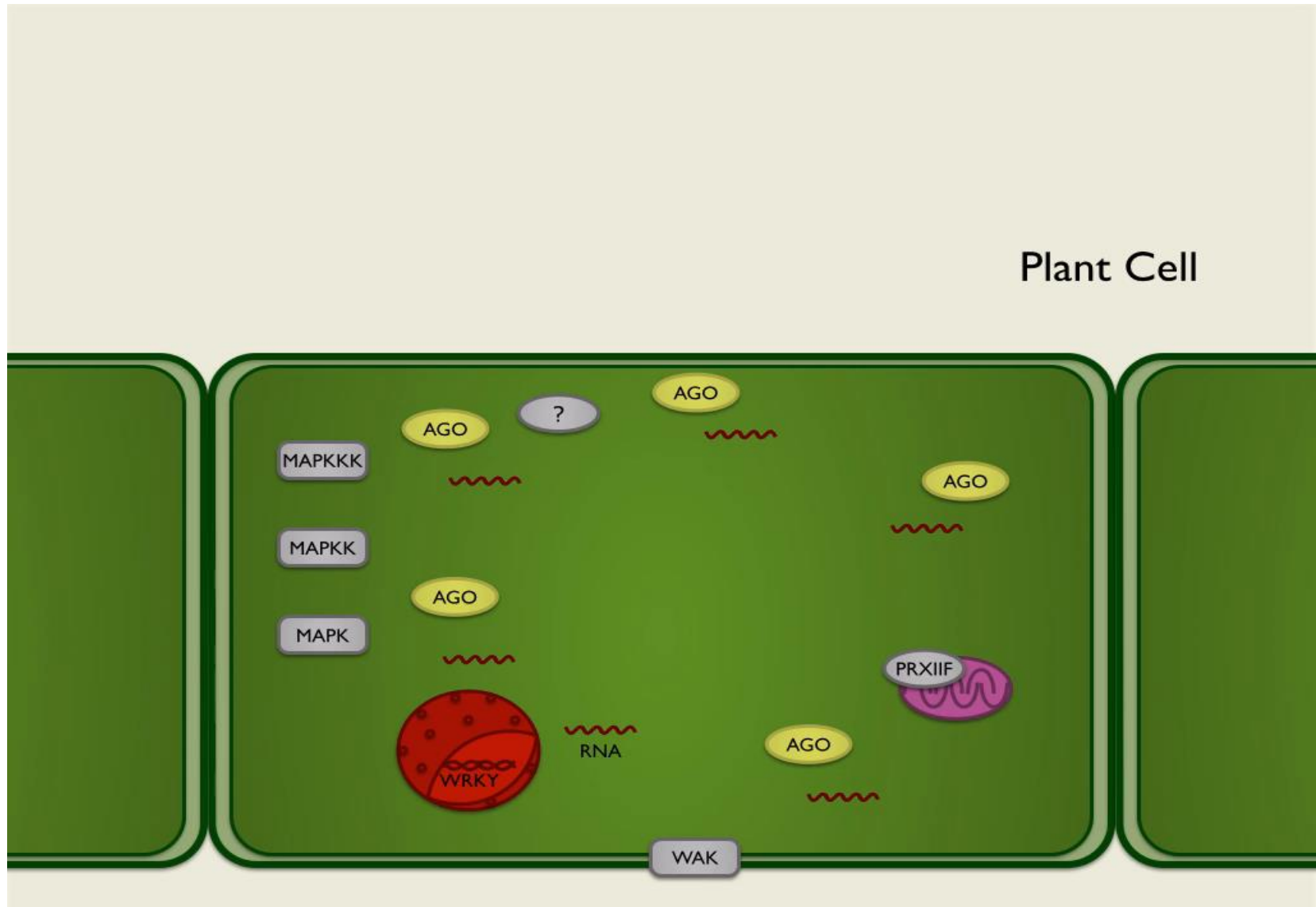
AGO-IP



sRNA Analysis



Naturally Occurring Cross-Kingdom RNAi (as an Advanced Virulence Mechanism)



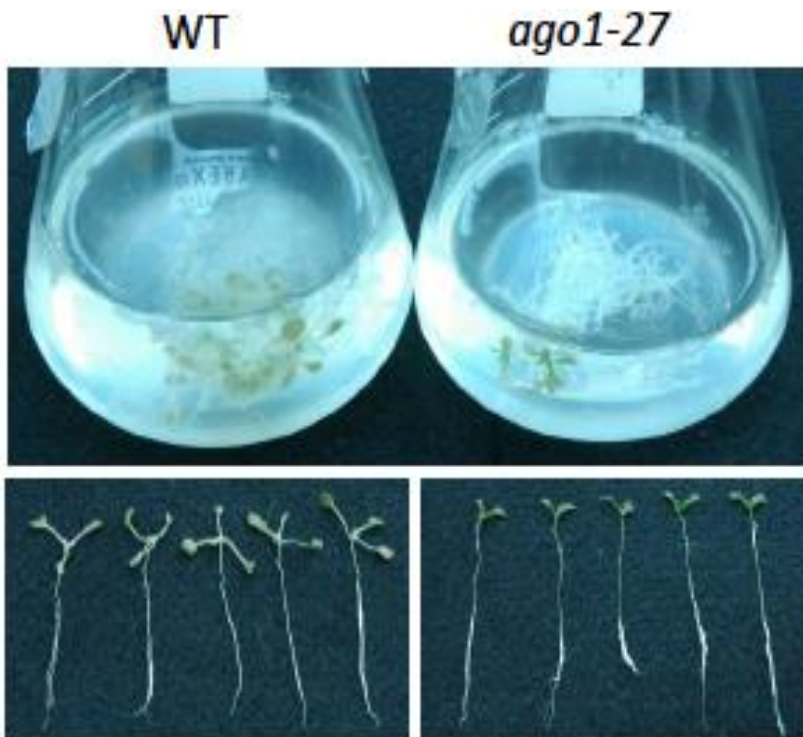
Is such cross-kingdom RNAi virulence mechanism present in other eukaryotic pathogens?

Another fungal plant pathogen, *Verticillium dahliae*, also depends on AGO1 function for its pathogenicity.

J Exp Bot. 2009;60(2):591-602. doi: 10.1093/jxb/ern306. Epub 2008 Dec 19.

RNA silencing is required for Arabidopsis defence against *Verticillium* wilt disease.

Ellendorff U¹, Fradin EF, de Jonge R, Thomma BP.



AGO1- and AGO2-IP



sRNA Analysis



We found more than 135 Vd-sRNAs have At Targets in AGO1-IP fraction, vs only 15 in AGO2-IP fraction
(using 100 rpm and 10 fold enrichment as cutoff)

Similar Findings in Animal Systems

Nat Commun. 2014 Nov 25;5:5488. doi: 10.1038/ncomms6488.

Exosomes secreted by nematode parasites transfer small RNAs to mammalian cells and modulate innate immunity.

Buck AH¹, Coakley G¹, Simbari F¹, McSorley HJ¹, Quintana JF¹, Le Bihan T², Kumar S³, Abreu-Goodger C⁴, Lear M¹, Marcus Y¹, Ceroni A⁵, Babayan SA⁶, Blaxter M⁷, Ivens A⁶, Maizels RM¹.

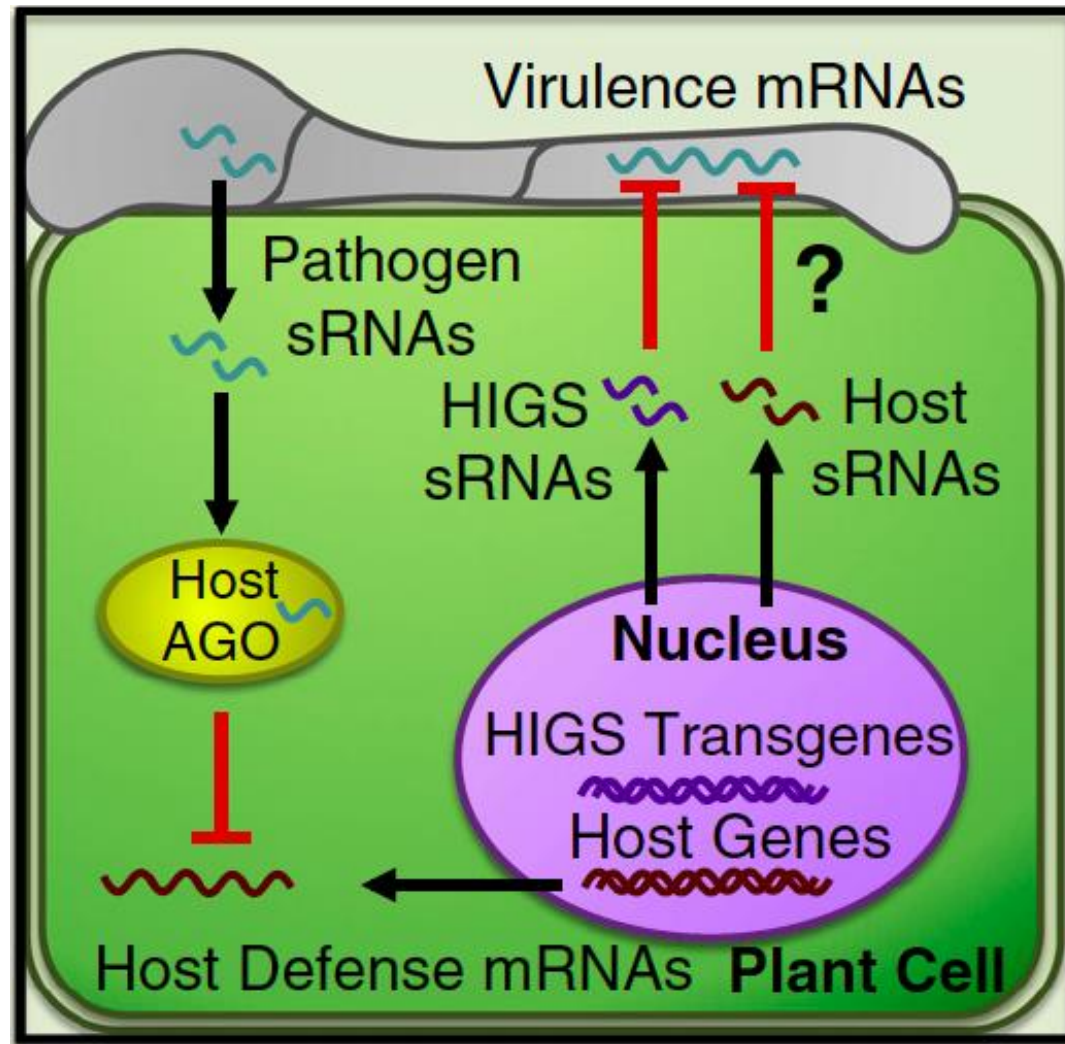
⊕ Author information

Table 2 | *Litomosoides sigmodontis*-derived miRNAs found in mouse serum.

Name	Mature sequence	Number of reads (infected)
miR-100a	UACCCGUAGCUCCGAAUAUGUGU	479
miR-86	UAAGUGAAUGCUUUGCCACAGUCU	57
Bantam-a	UGAGAUCAUUGUGAAAGCUAUU	45
Bantam-b	UGAGAUCACGUUACAUCGCCU	45
miR-100b	AACCCGUAGUUUCGAACAUGUGU	40
miR-71	UGAAAGACAUGGGUAGUGAGACG	32
miR-100c	AACCCGUAGAAUUGAAUUCGUGU	22
miR-50-5p	UGAUAUGUCUGAUAUUCUUGGGUU	10
miR-34-5p	UGGCAGUGUGGUUAGCUGGUUGU	8
miR-263/183	AAUGGCACUAGAUGAAUUCACGG	7
Bantam-c	UGAGAUCAUGCCACAUCGGUCU	4
miR-50-3p	CCAGCAUCUCAGACGUAUCGGC	3
miR-153	UUGCAUAGUCACAAAAGUGAUG	3
miR-87-5p	CGCCUGGGACUUCGACUCAACCU	2
miR-2	UAUCACAGCCAGCUUUGAUGU	2
miR-5866	UUACCAUGUUGAUCGAUCUCC	2

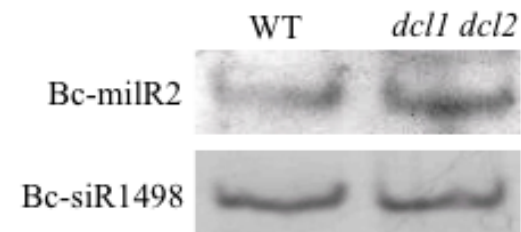
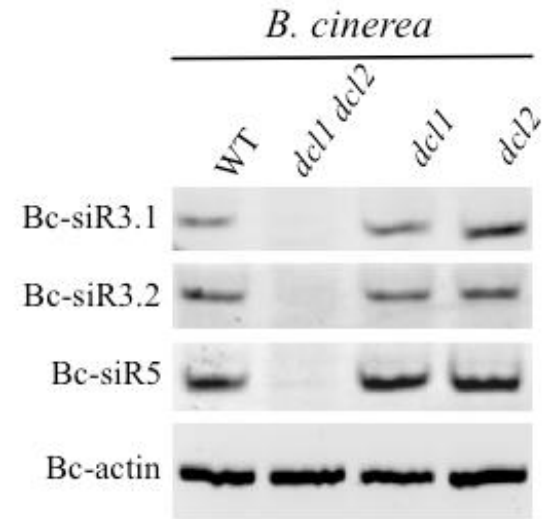
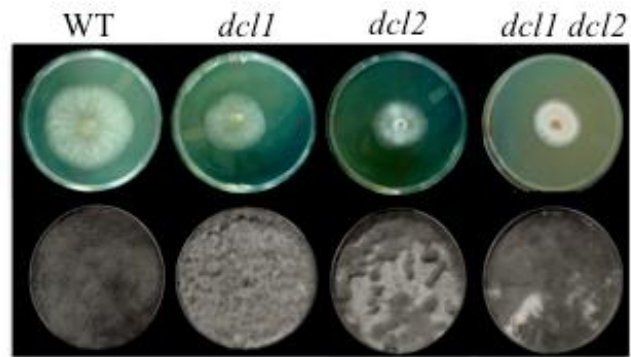
These parasite small RNAs suppress host genes involved in inflammation and immunity, including the receptor for the alarmin IL-33 and a key regulator of mitogen-activated protein kinase (MAPK) signaling

Can we develop an efficient way to control pathogens using small RNA effectors?



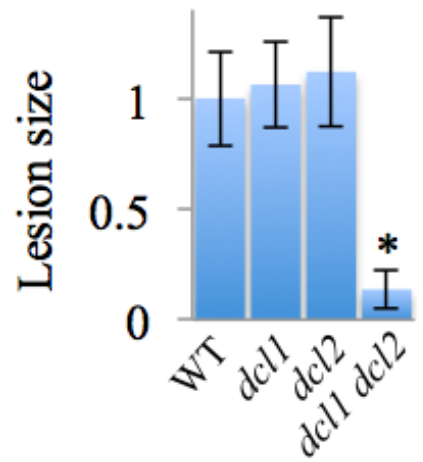
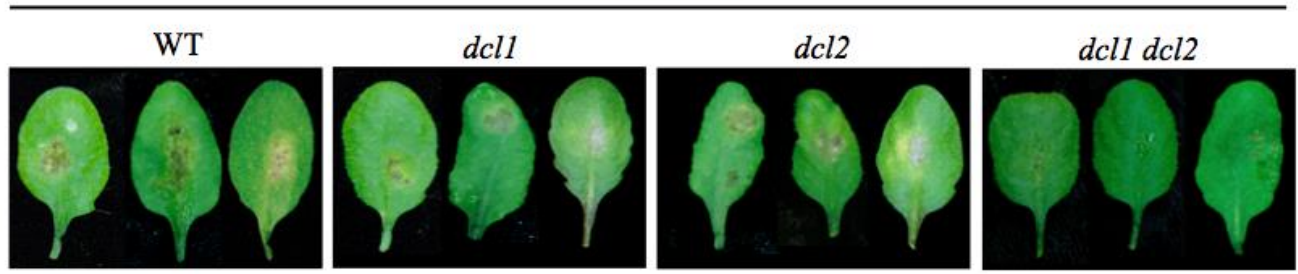
Is cross-kingdom RNAi bidirectional?

Bc-sRNA effectors are dependent on *B. cinerea* DCLs

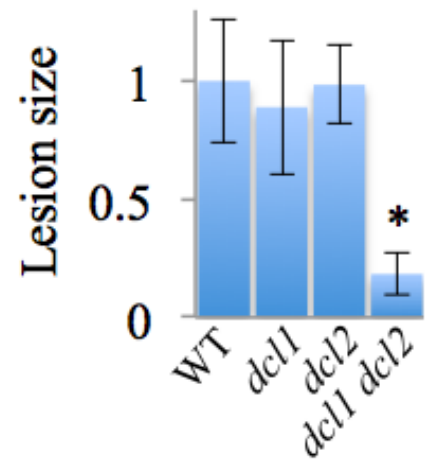
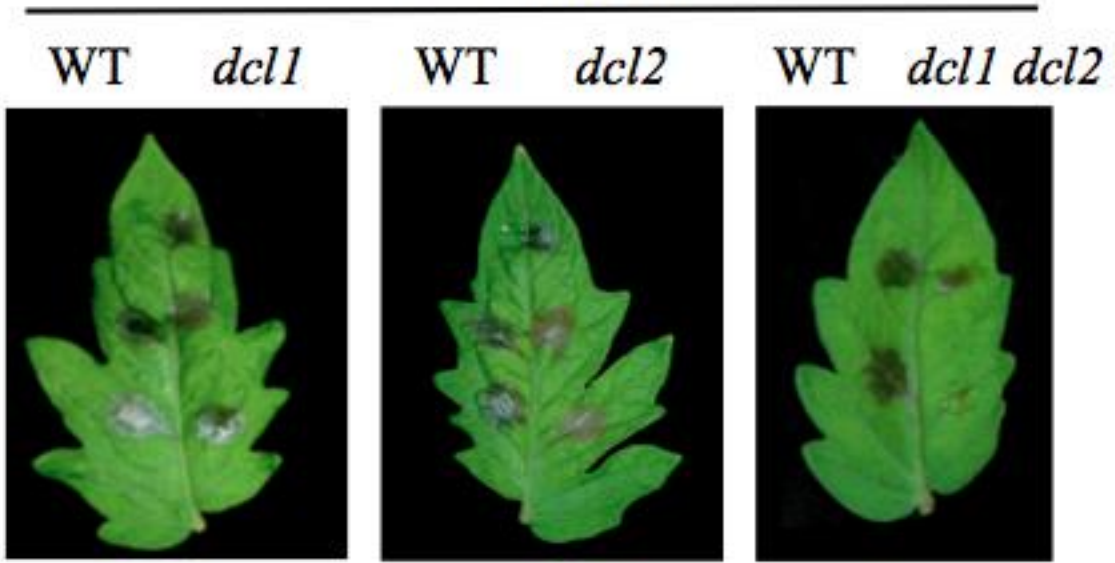


B. cinerea DCLs are essential for its pathogenicity

B. cinerea

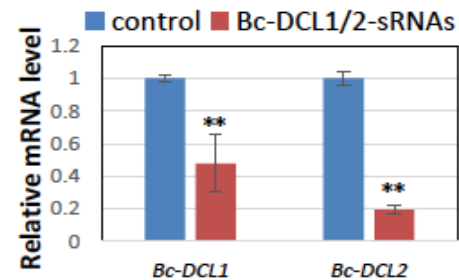
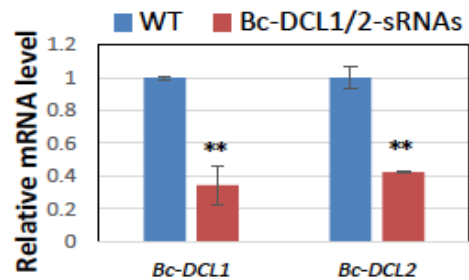
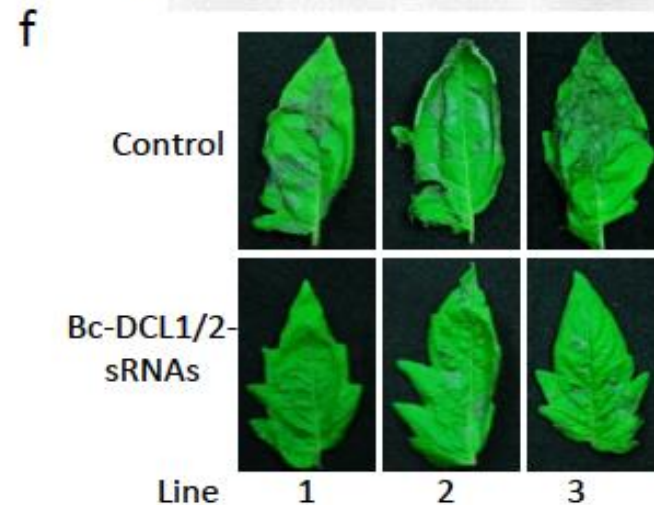
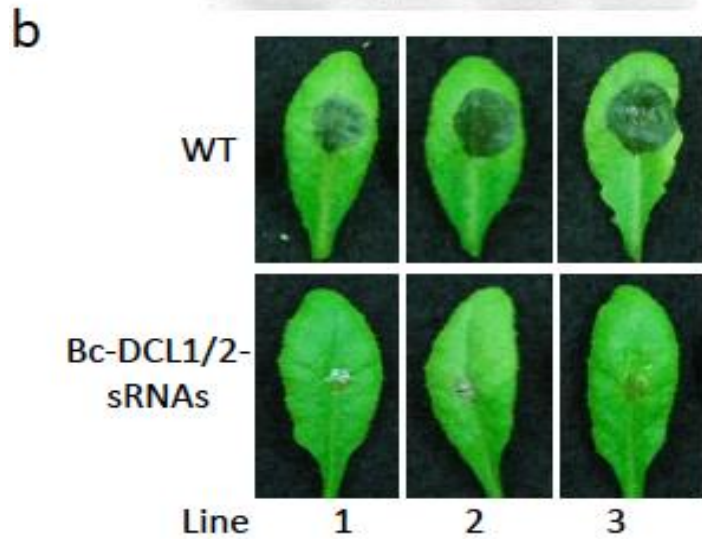
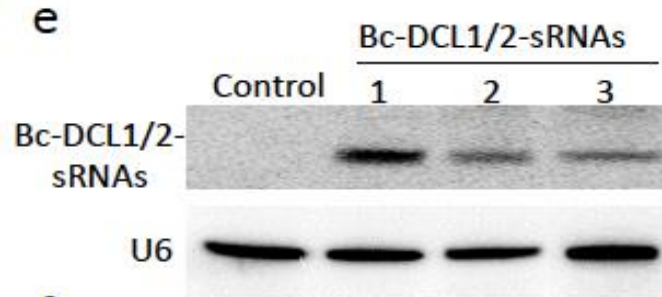
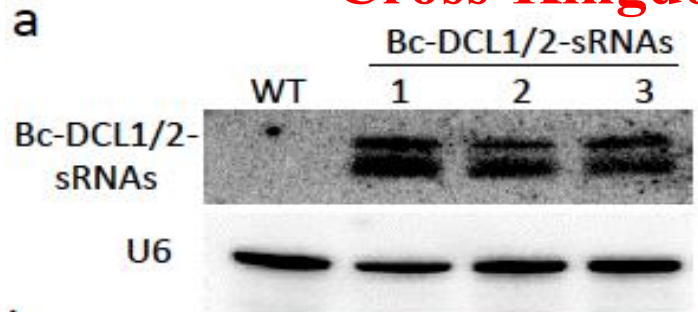


B. cinerea

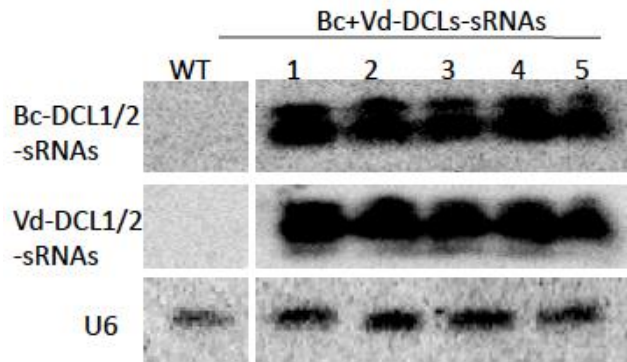


Plants expressing Bc-DCL1/2 targeting sRNAs are more resistance to *B. cinerea* infection

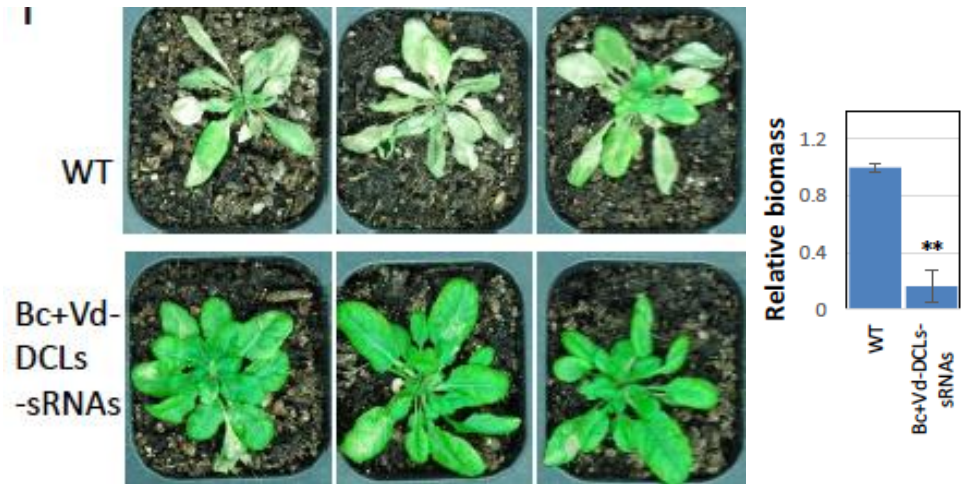
Cross-Kingdom RNAi is bidirectional



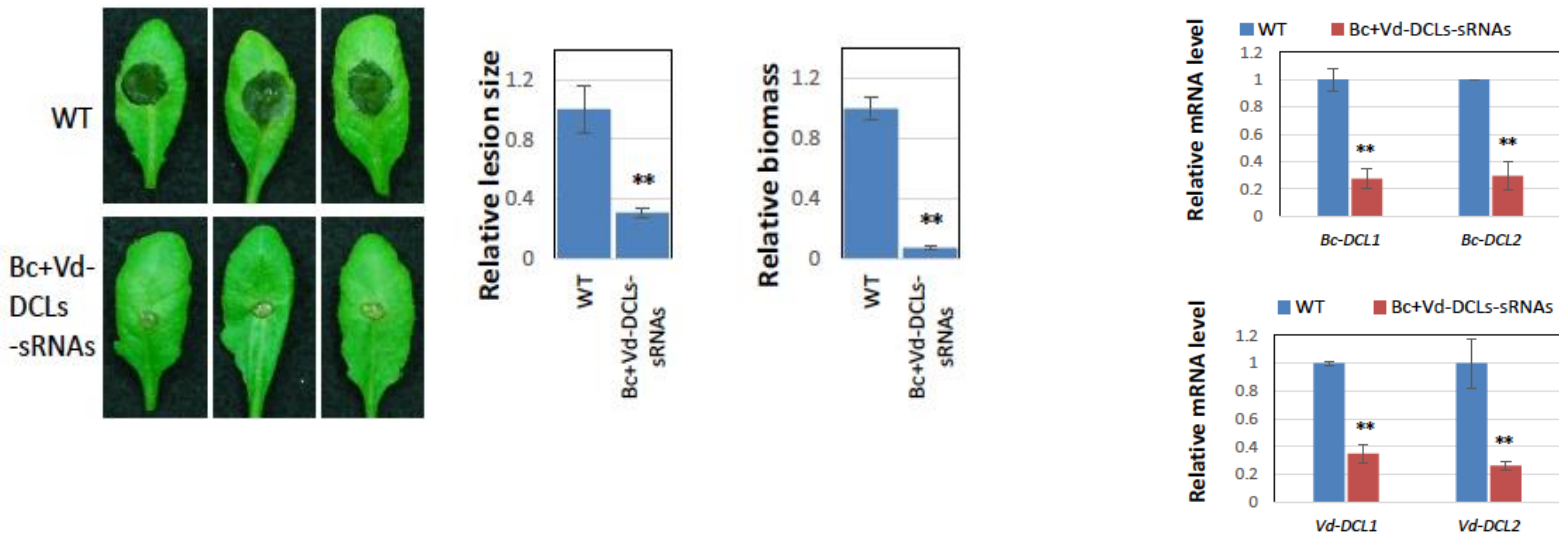
Cross-Kingdom RNAi-based disease control strategy allows targeting multiple pathogens



Verticillium resistance



Botrytis resistance

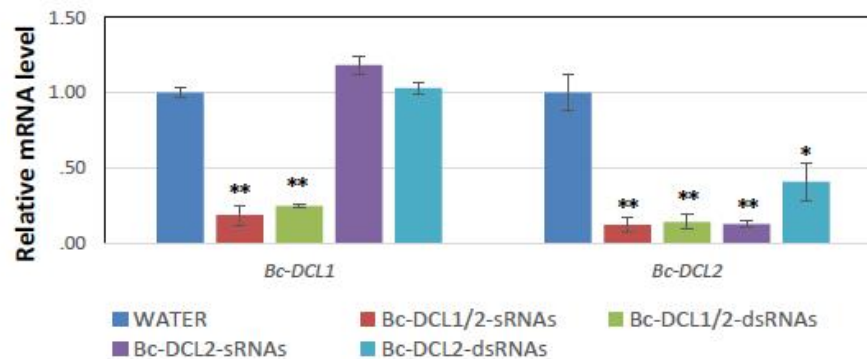
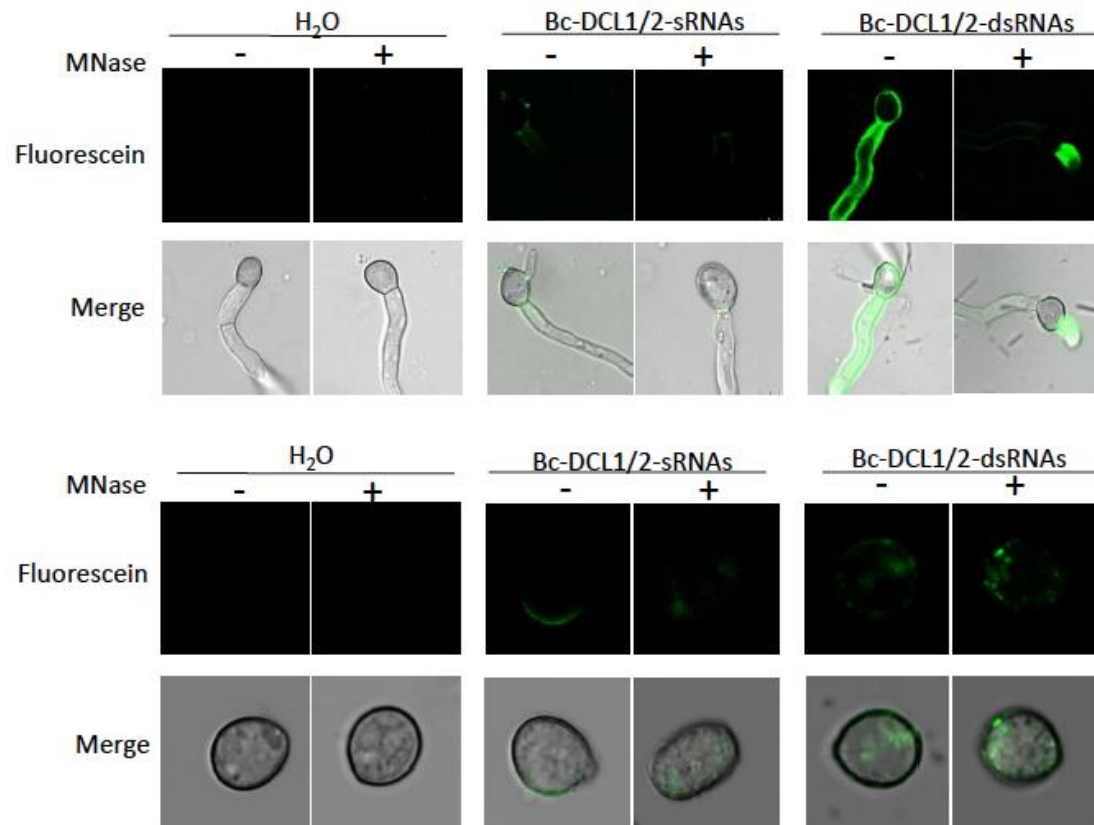


Translocation of Sickle Cell Erythrocyte MicroRNAs into *Plasmodium falciparum* Inhibits Parasite Translation and Contributes to Malaria Resistance

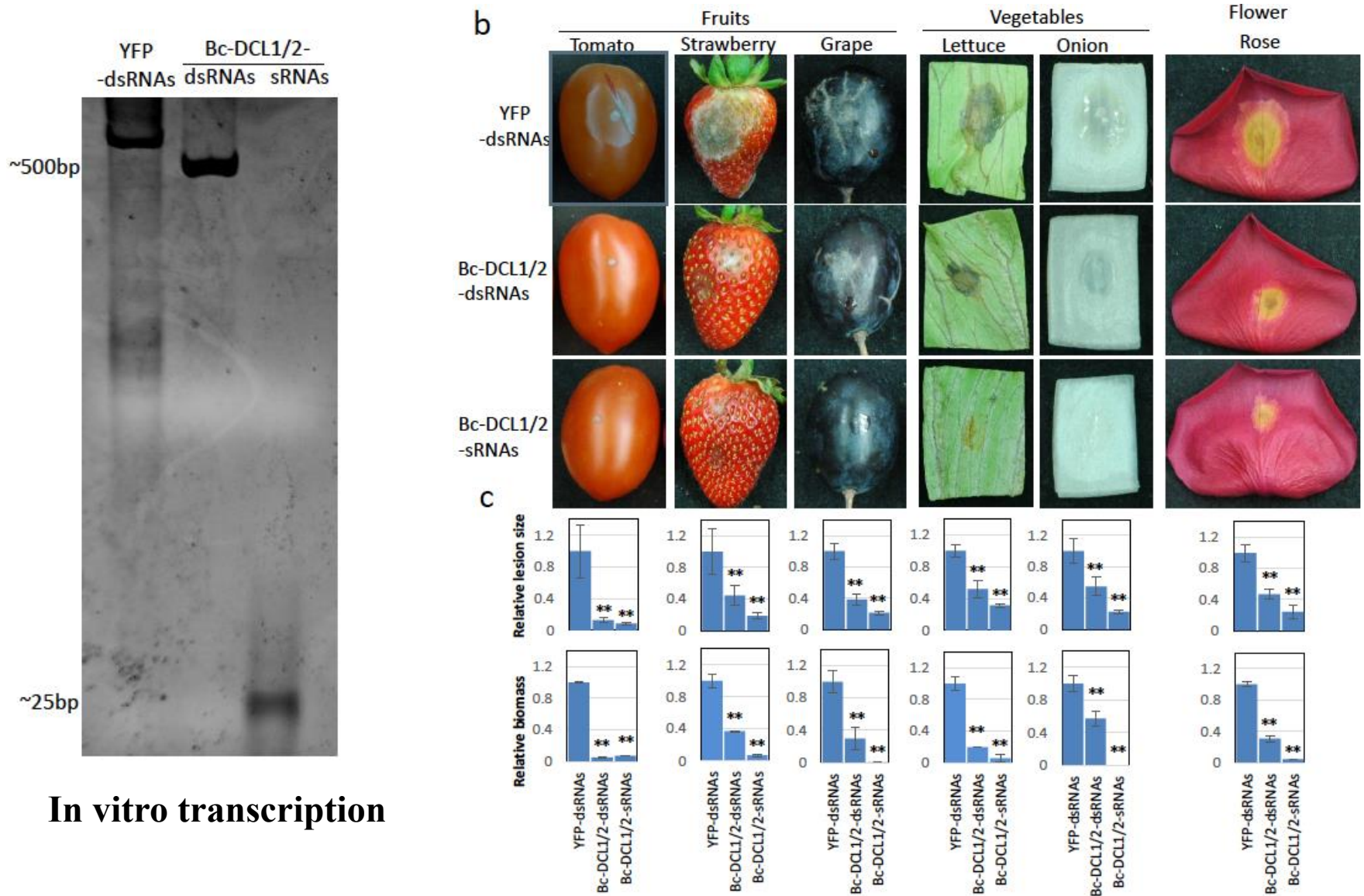
Gregory LaMonte,^{1,2,10,11} Nisha Philip,^{1,2,11,12} Joseph Reardon,² Joshua R. Lacsina,^{3,9} William Majoros,¹ Lesley Chapman,^{1,2} Courtney D. Thornburg,⁴ Marilyn J. Telen,⁵ Uwe Ohler,^{1,6,7} Christopher V. Nicchitta,^{3,9} Timothy Haystead,⁸ and Jen-Tsan Chi^{1,2,*}

miR451 and let-7i are highly enriched in hemoglobin allele (HbS) heterozygous and homozygous erythrocytes, which contributes to resistance against *P. falciparum*.

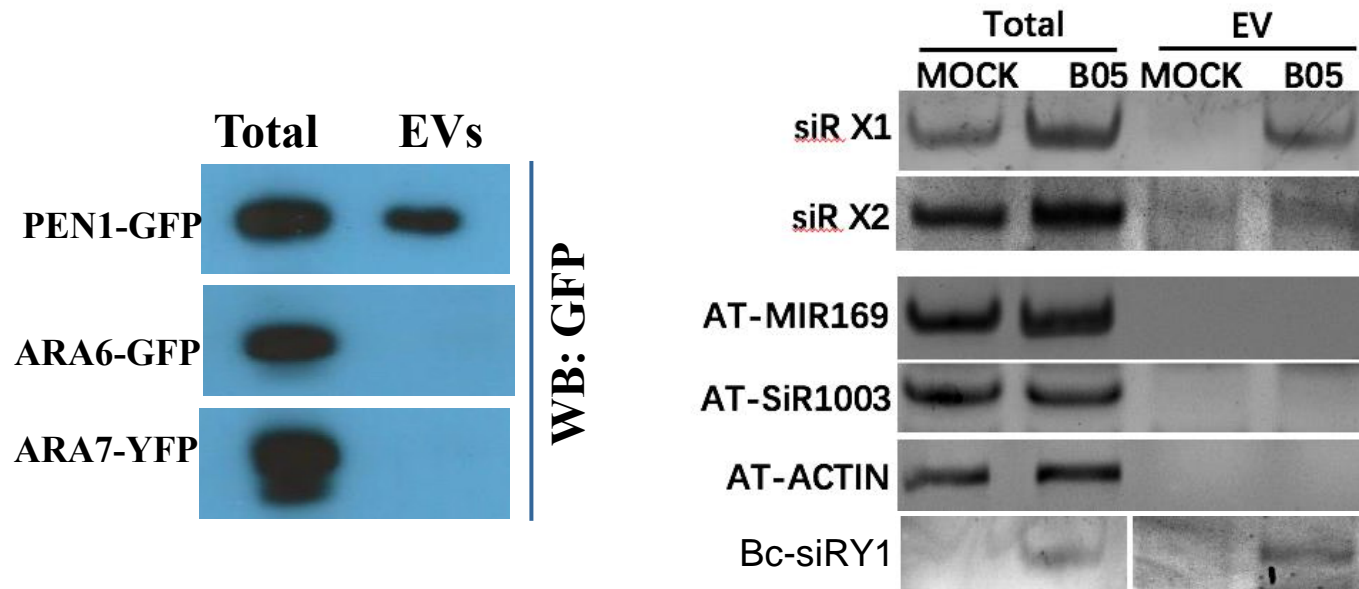
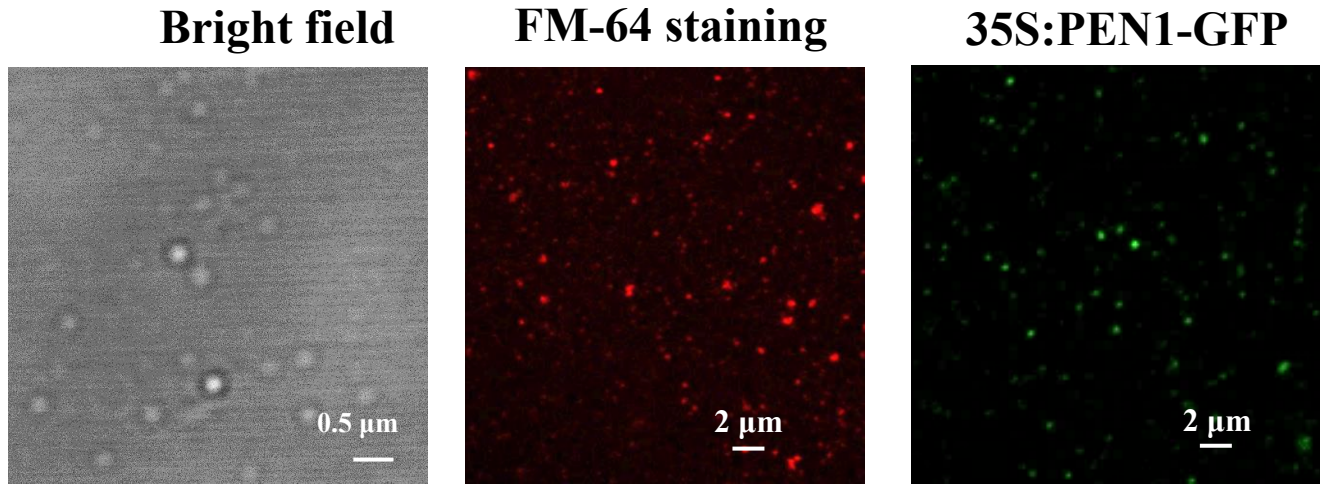
B. cinerea could uptake RNAs from environment



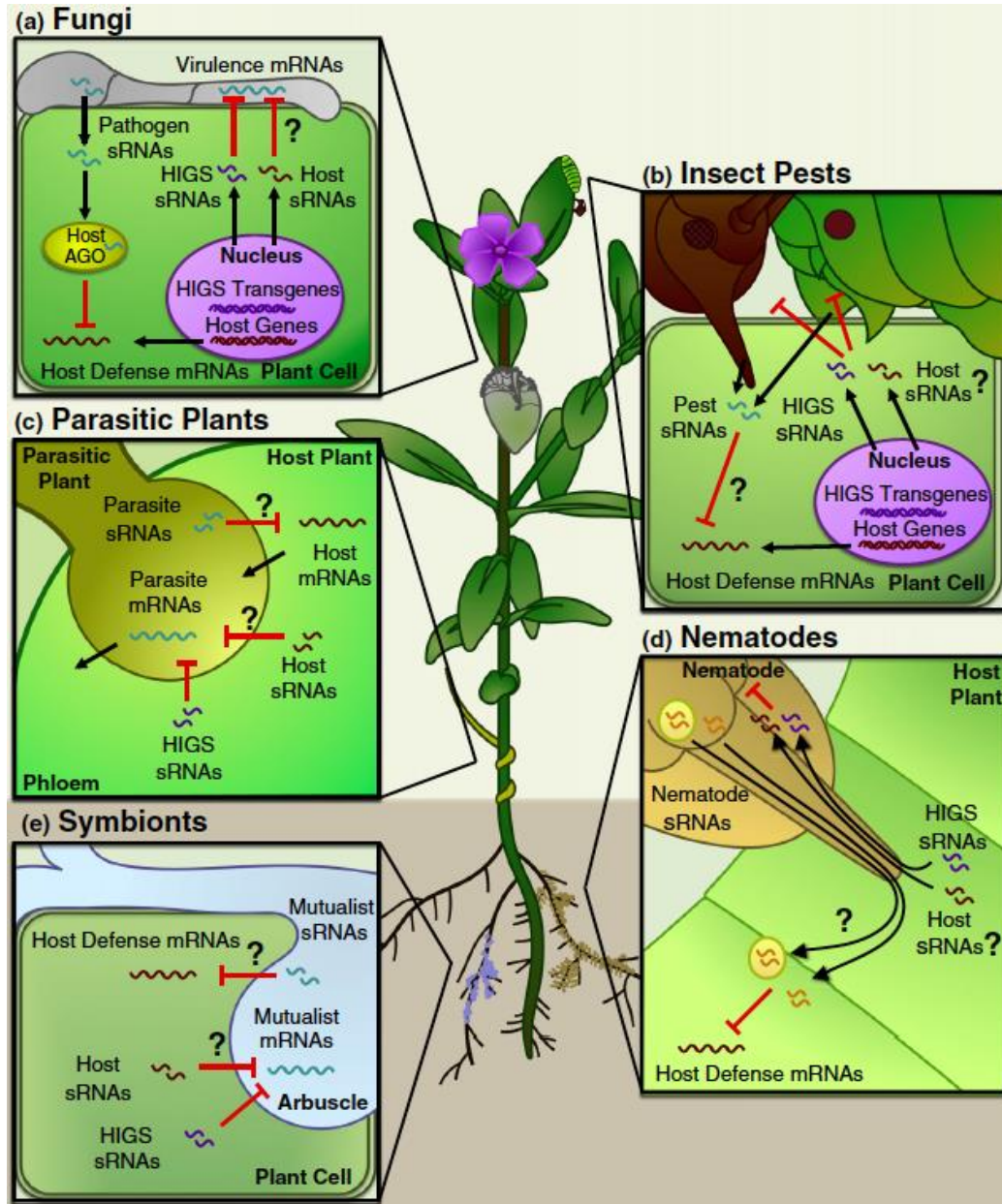
Spraying Bc-DCL1/2 dsRNAs or siRNAs inhibits infection



Plant and fungal sRNAs are present in extracellular vesicles



Cross-kingdom RNAi in host-pathogen/pest interaction



Acknowledgement



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Sascha Laubinger
Mike Prigge
David Baulcombe
Herve Vaucheret
María Marcote
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