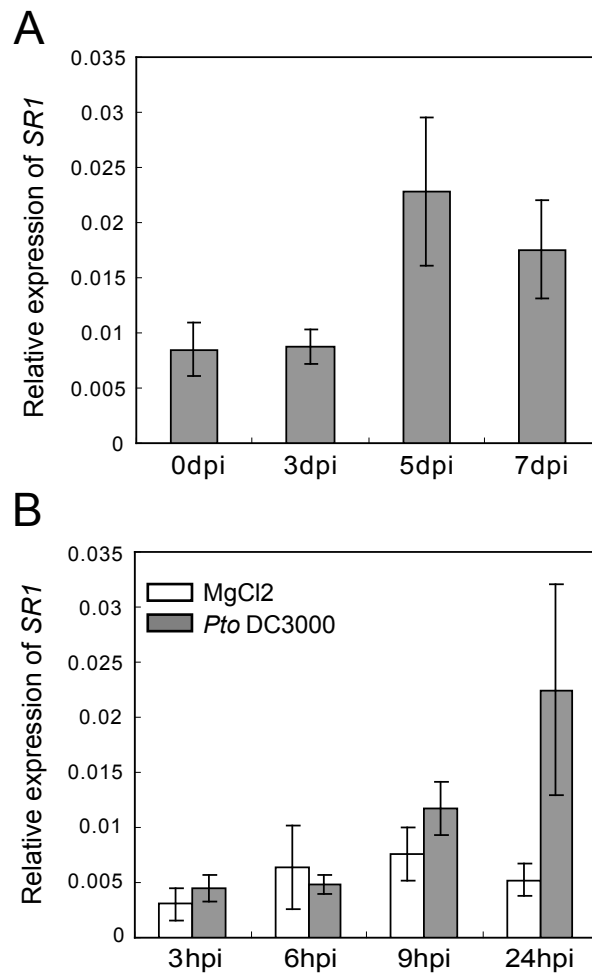


Figure S1 Nie *et al*



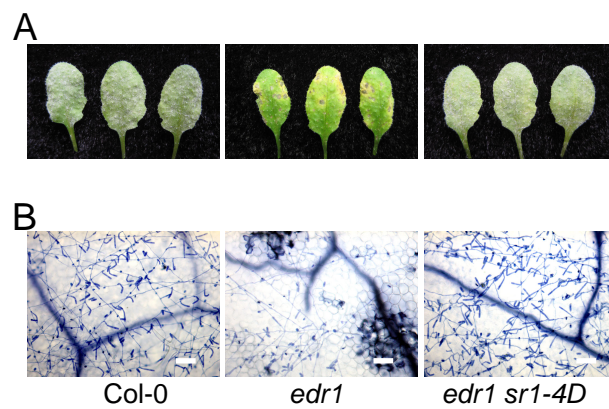
Supplemental Figure 1. *SR1* was induced by powdery mildew and *Pto* DC3000

Four-week-old plants were inoculated with *G. cichoracearum* or *Pto* DC3000. The relative expression of *SR1* was examined at various time points.

A. *G. cichoracearum*

B. *Pto* DC3000

The bars represent the mean and standard deviation of values from three independent experiments.

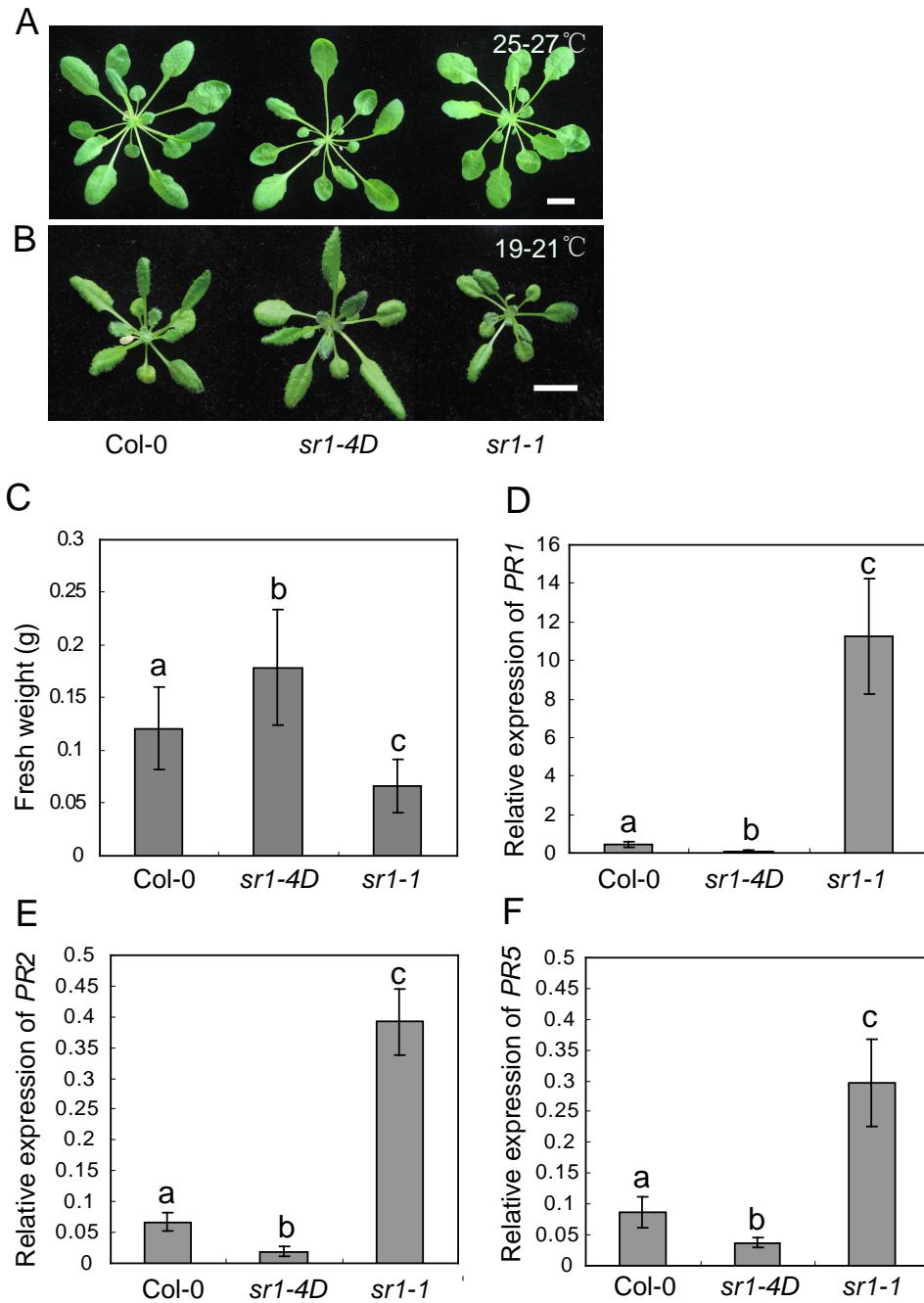


Supplemental Figure 2. *sr1-4D* suppressed *edr1*-mediated powdery mildew resistance
Four-week-old plants were infected with *G. cichoracearum*.

A. The representative leaves were removed and photographed at 8 dpi.

B. Infected leaves were stained with trypan blue to visualize fungal growth and plant cell death. Bar = 100 μ m.

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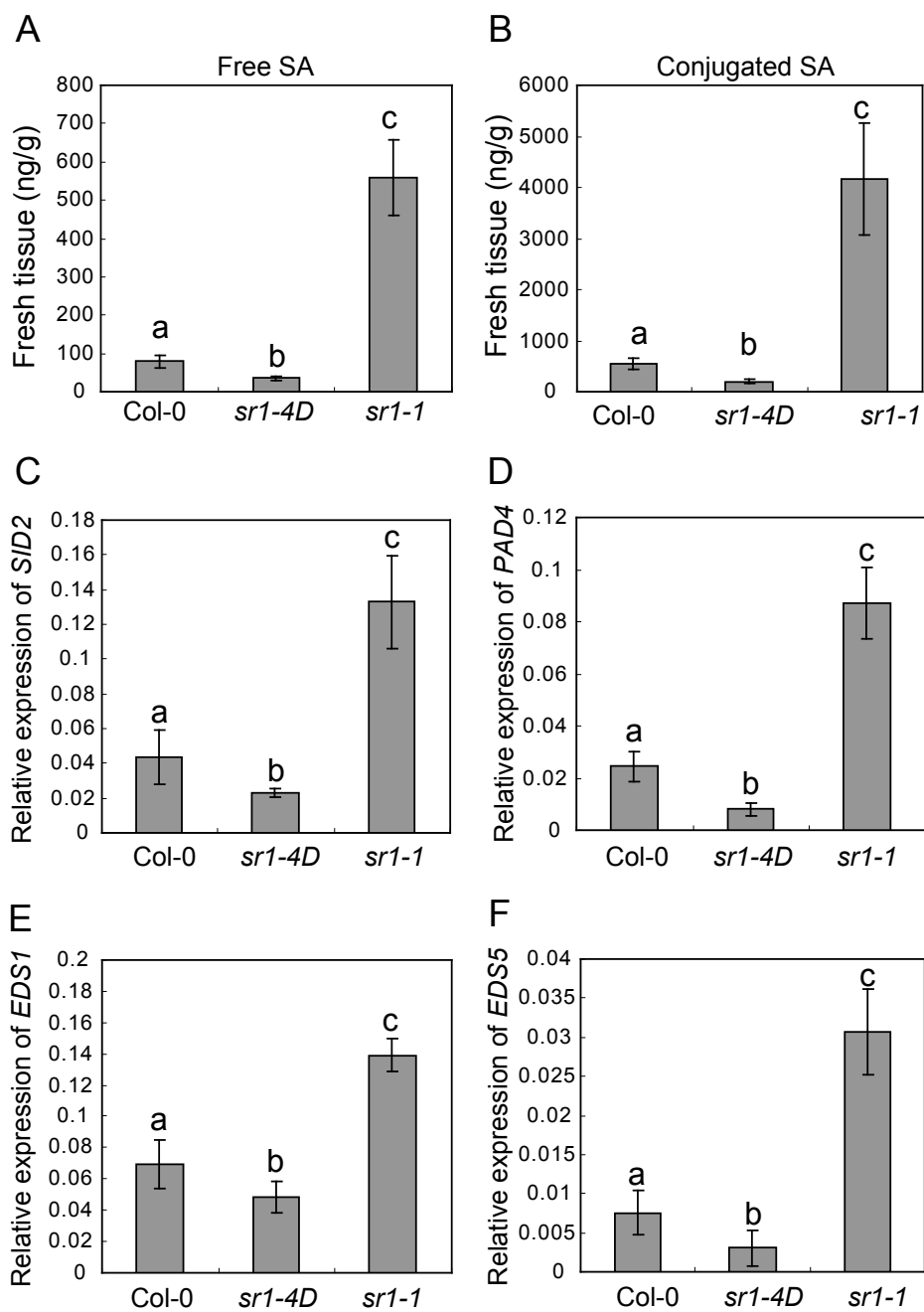
Supplemental Figure 3. Temperature dependent growth phenotype of *sr1-4D*

A-B. Wild type, *sr1-4D* and *sr1-1* plants were grown at 25-27°C (A) or 19-21°C (B) for 4-5 weeks. Bar=1cm

C. Fresh weight of five-week-old plants grown at 19-21°C. The bars represent mean and standard deviation of samples (n=12).

D-F. Relative expression of defense related genes *PR1* (D), *PR2* (E) and *PR5* (F) in 4-5 week old plants grown at 19-21°C. The bars represent the mean and standard deviation of values from three independent experiments. In C-F, low-case letters indicate statistical significance ($p < 0.01$, t-test).

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Supplemental Figure 4. SA accumulation of *sr1-4D*.

A-B. Accumulation of free (A) and Conjugated (B) SA in five-week-old plants grown at 19-21°C.

C-F. Relative expression of *SID2* (C), *PAD4* (D), *EDS1* (E) and *EDS5* (F) in 4-5 week old plants grown at 19-21°C.

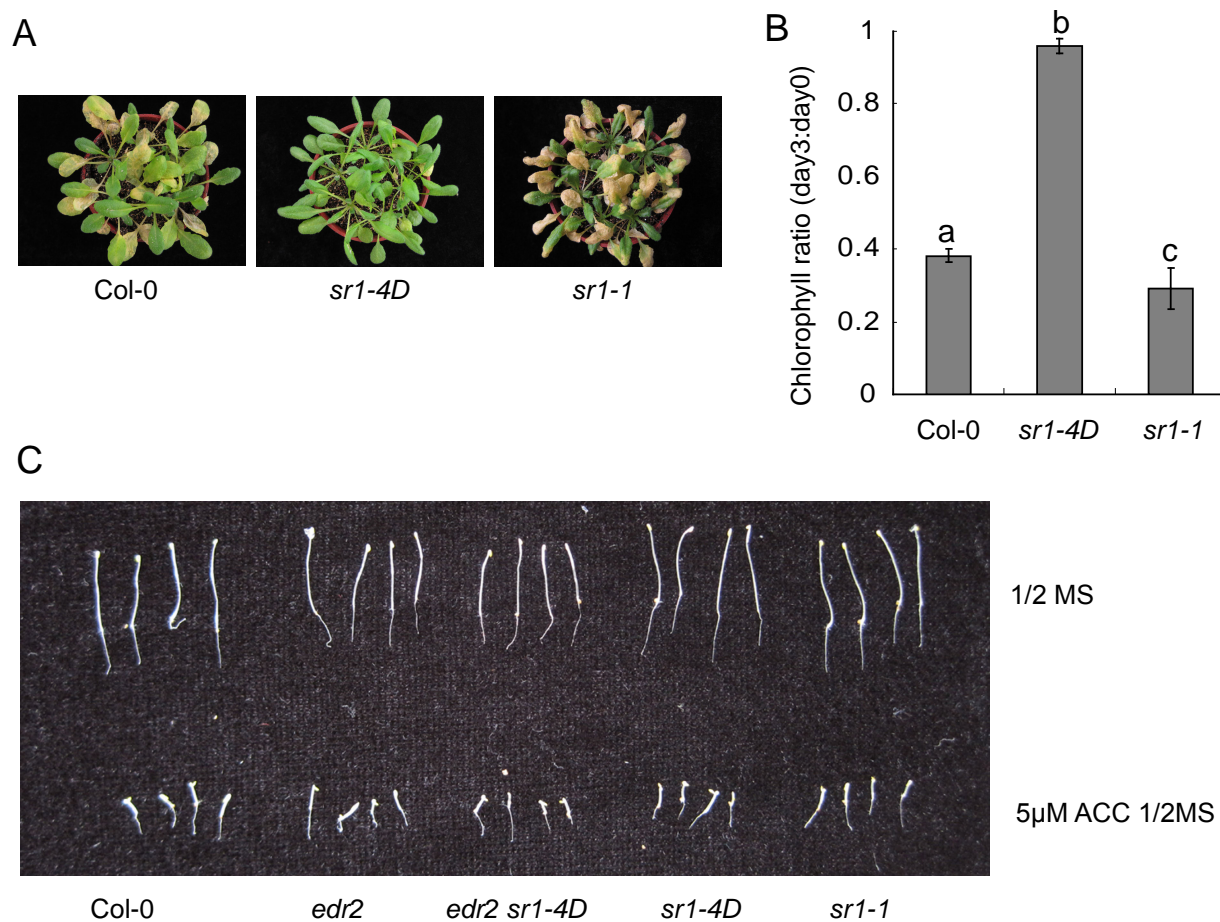
The bars represent mean and standard deviation of three independent biological samples. Low-case letters indicate statistical significance ($p < 0.01$, t-test).

Figure S5

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GTTAATTCTTGTTTTGGTTCTTTTTGATAACCCAAAGTTTATATAGGTTTTT
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-141 -137
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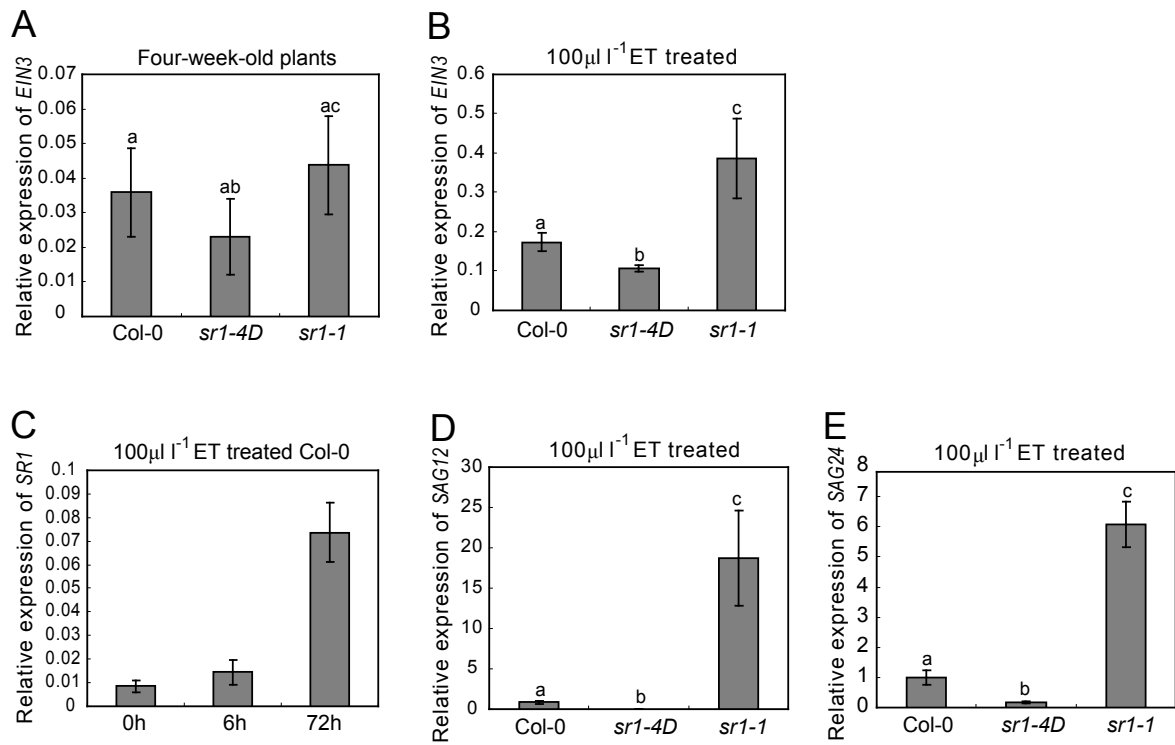
Supplemental Figure 5. The *NDR1* promoter sequence contains a CGCG box



Supplemental Figure 6. SR1 is involved in ethylene induced senescence, but is not involved in ACC induced triple response.

- A. Four-week-old plants were treated with $100 \mu\text{l l}^{-1}$ ethylene for 3 days.
- B. Chlorophyll ratio of day 3 over day 0, for the fourth to the sixth leaves treated with $100 \mu\text{l l}^{-1}$ ethylene. The bars represent mean and standard deviation of values from samples. Statistical differences are indicated with lower-case letters ($n=4$, $P<0.01$, one-way ANOVA). The experiment was repeated 3 times with similar results.
- C. The wild type, *edr2*, *edr2 sr1-4D*, *sr1-4D* and *sr1-1* seeds were plated on 1/2 MS medium with $5\mu\text{M ACC}$ for 4 days in dark.

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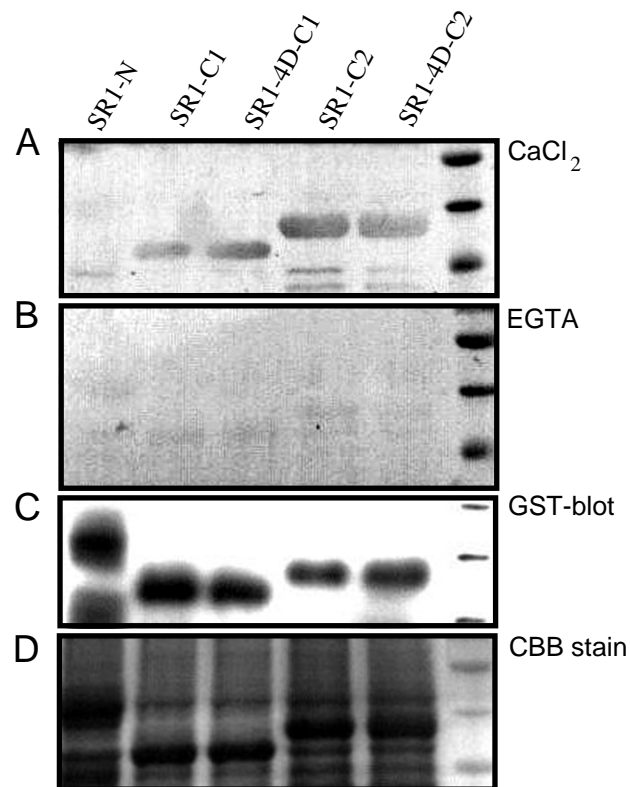


Supplemental Figure 7. Relative expression of several defense and senescence related genes.

A-B. Relative expression of *EIN3* in four-week-old plants. (A) Untreated plants (B) Plants treated with 100 $\mu\text{l l}^{-1}$ ethylene for 3 days. The bars represent mean and standard deviation from three biological replicates. Low-case letters indicate statistical significance ($p < 0.01$, t-test).

C. Relative expression of *SR1* in wild type before and after ethylene treatment (at 0h, 6h and 72h).

D-E. Relative expression of *SAG12* (D) and *SAG24* (E) in four-week-old plants treated with 100 $\mu\text{l l}^{-1}$ ethylene for 3 days. The bars represent mean and standard deviation from three biological replicates. Low-case letters indicate statistical significance ($p < 0.01$, t-test).

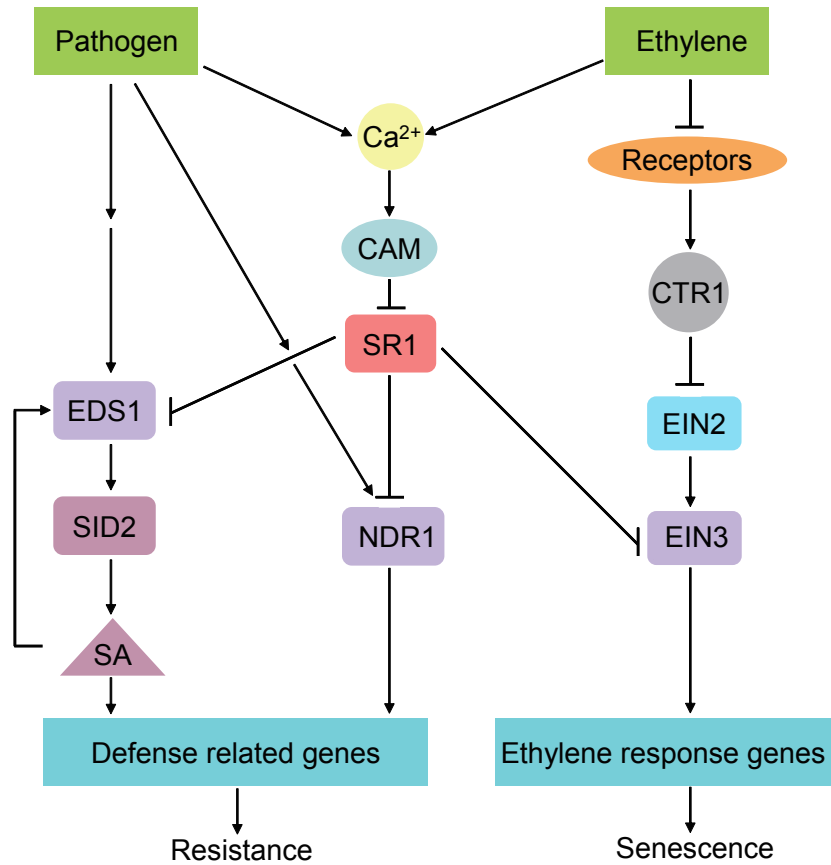


Supplemental Figure 8. Calcium is needed for SR1-4D binding to the calmodulin *in vitro*. (An animal version of calmodulin was used in the experiments)

- A. Calmodulin binding assay performed in the presence of 1mM CaCl_2 .
 - B. Calmodulin binding assay performed in the presence of 5mM EGTA
 - C. Immunoblot with GST antibody showing the correct size of the expressed protein.
 - D. Coomassie brilliant blue (CBB) staining showed the equal loading of each sample.
- SR1-C1-GST: Truncated SR1 protein (801aa-900aa)
 SR1-C2-GST: Truncated SR1 protein (801aa-930aa)
 SR1-4D-C1-GST : Truncated SR1-4D protein (801aa-900aa)
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Figure S9

Nie *et al*



Supplemental Figure 9. A model illustrating the role of SR1 in defense responses and senescence. SR1 negatively regulates defense responses and senescence by repressing the expression of *EDS1*, *NDR1* and *EIN3*. The SR1 protein functions at the critical point where plant immunity and ethylene signaling are fine-tuned to reach a well-balanced defense against pathogens.

Table 1 Primers used in this study.

Primer name	Sequence(5' to 3')	Used for
Salk_01152 LP	TGAAAACCTGATGAATCCGAG	<i>sr1-1</i>
Salk_01152 RP	GGTTGTGAAGTGGTGGTAAGC	<i>sr1-1</i>
Caps-F	AGTGATGATTCCGTGCAAGGCG	<i>sr1-4D</i>
Caps-R	GCACCATTGCCAGTAAAAACCGTT	<i>sr1-4D</i>
SR1-C-F	GGATCCTCCAAATAGATAAATAGGGTTC	Complementation
SR1-C-R	GTCGACCAGTAAGCATCAATAACATTTTCAGG	Complementation
NDR1-Gelshift F	ATTTGGCTAAACGCGTGTGTGCGTGTGTGT	EMSA
NDR1-Gelshift R	ACACACACGCACACACGCGTTTAGCCAAAT	EMSA
NDR1-Gelshift mF	ATTTGGCTAAACGATTGTGTGCGTGTGTGT	EMSA
NDR1-Gelshift mR	ACACACACGCACACAATCGTTTAGCCAAAT	EMSA
SR1-Gex F	CGCGGATCCATGGCGGAAGCAAGACGATT	EMSA
SR1n-Gex R	CCGCTCGAGCCGGTAAAAGAAGTAGAAACTC	EMSA
SR1-RT-F	ATGGCGGAAGCAAGACGATTCAG	Real time PCR
SR1-RT-R	ATCAAACATAAAAACAGACCCACTTG	Real time PCR
NDR1-RT F	CTTTTCTTATGGCTTAGTCTCCGTG	Real time PCR
NDR1-RT R	ATCTTGGTCGTGTTGATGGTGG	Real time PCR
Actin8-RT-F	CGAGGCTCCTCTTAACCCAAA	Real time PCR
Actin8-RT-R	GGCACAGTGTGAGACACACCA	Real time PCR
PR1-RT-F	AAAGCTCAAGATAGCCCACA	Real time PCR
PR1-RT-R	AGCCTTCTCGCTAACCCACA	Real time PCR
PR2-RT-F	GAATCAAGGAGCTTAGCCTCAC	Real time PCR
PR2-RT-R	GTAGAGCCGCATTCGCTGGAT	Real time PCR
PR5-RT-F	GCACAGAGACACACACAAAA	Real time PCR
PR5-RT-R	TGTTCTTAGAGTGAAGTCTG	Real time PCR
SID2-RT-F	CGCAAGAAGTATGAGTCATGTTCCG	Real time PCR
SID2-RT-R	AACCTGTAACCGAACGACGC	Real time PCR
PAD4-RT-F	CGGCTTATCCTCCGATGAACCT	Real time PCR
PAD4-RT-R	CCAAAGGTGATACAAAAGACGC	Real time PCR
EDS1-RT-F	GTCTACGCTCAATGACCTTGGAGTG	Real time PCR
EDS1-RT-R	CATTTTTATGGGCTTGACACTTTGG	Real time PCR
EDS5-RT-F	GGAACAGATGAAAGAGATAGTGAAG	Real time PCR
EDS5-RT-R	GATGTAGCCACGGAGAGGAA	Real time PCR
EIN3-RT-F	GACAGAACCGTTTTACCTGCGAGA	Real time PCR
EIN3-RT-R	CTGAGGAAATCCAACCTACAGGCTTA	Real time PCR
SAG12-RT-F	CTGGTTTCAAAGGTGTCTCGG	Real time PCR
SAG12-RT-R	CTGAAAACGCCCAACAACATC	Real time PCR
SAG24-RT-F	AATCATGGAGTTCATGCTCAGG	Real time PCR
SAG24-RT-R	CAGGCACAATCCTCTTCATCG	Real time PCR
NDR1 ChIP-F	TTGGTTCTTTTTGATAACCCAAAAGT	ChIP
NDR1 ChIP-R	TTTGGTTTGCTGATTGGTTGATATT	ChIP
EIN3-ChIP-F	TAGCACACAATAAATAAACTCCG	ChIP
EIN3-ChIP-R	TTACACATAAAAATTTAAGACATCAT	ChIP

EDS1-ChIP-F	TGGTTATGCAATTTGGTTTAGCCAA	ChIP
EDS1-ChIP-R	ACCGAATTA ACTAACTACACCTTCTT	ChIP
ACTIN2-ChIP-F	GATCCTAGTCTTTTAGTGTGCATTC	ChIP
ACTIN2-ChIP-R	ATTAAATGATTGATCGGTTTTTCGTG	ChIP
SR1-AttB1	ACAAGTTTGTACAAAAAAGCAGGCTTCGAAGGAGATA GAACCATGGCGGAAGCAAGACGATTCAGCC	ChIP
SR1-AttB2	ACCACTTTGTACAAGAAAGCTGGGTC ACTGGTC CACAAAGATGAGGACATAGGCAACATCAA	ChIP
SRcam-2401 F	CGCGGATCCGCTCGGATTCATCAGGTTTTTCAGGGC	CAM binding
SRcam-2700 R	ACGCGTCGACCCAAATTATCTTTCTGTAGTTTTTCCTA	CAM binding
SRcam-2880 R	ACGCGTCGACTGCAAGAGCTTTTTGTAGCCTATCCTCT	CAM binding

Supplemental Figures

Supplemental Figure 1. *SRI* was induced by powdery mildew and *Pto* DC3000

Four-week-old plants were inoculated with *G. cichoracearum* or *Pto* DC3000. The relative expression of *SRI* was examined at various time points.

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B. *Pto* DC3000

The bars represent the mean and standard deviation of values from three independent experiments.

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- D. Coomassie brilliant blue (CBB) staining showed the equal loading of each sample.

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SR1-C1-GST: Truncated SR1 protein (801-900aa)

SR1-C2-GST: Truncated SR1 protein (801-930aa)

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SR1-4D-C2-GST: Truncated SR1-4D protein (801-930aa)

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