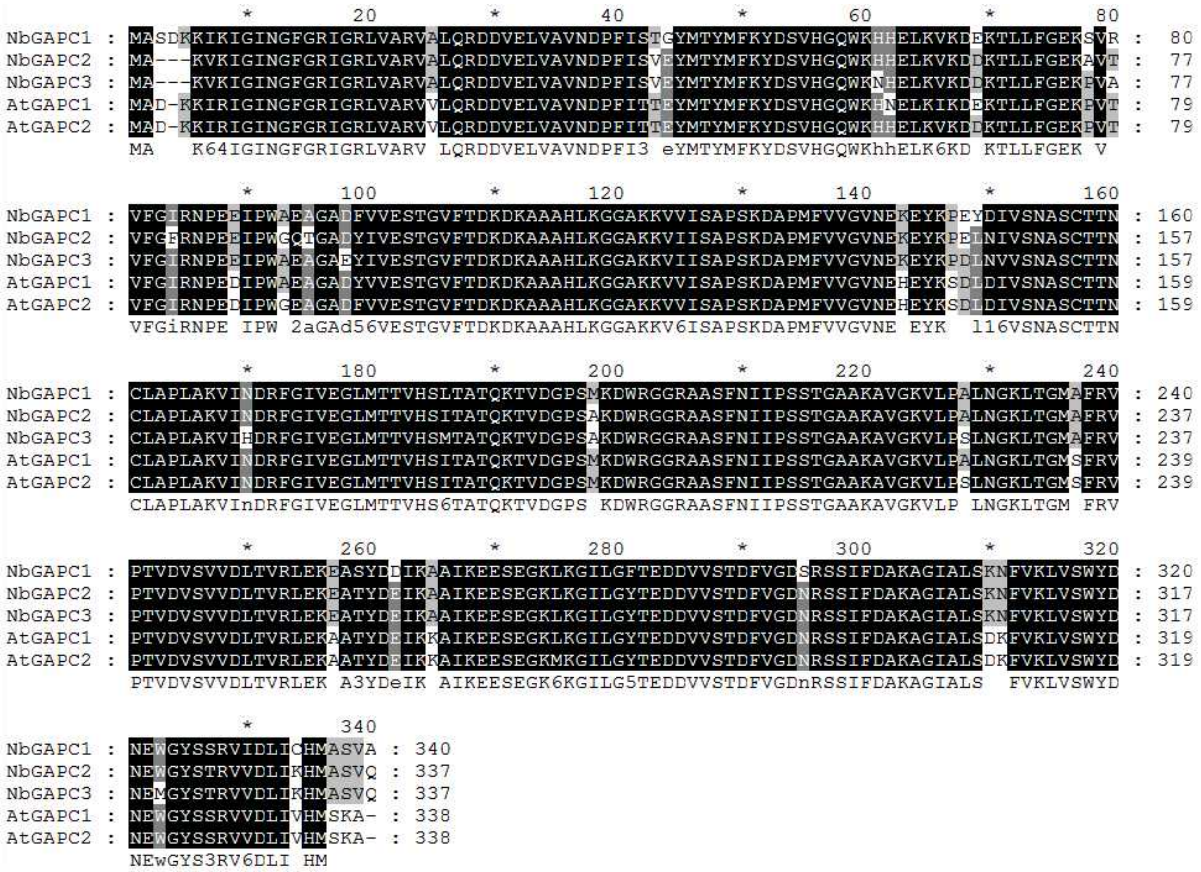
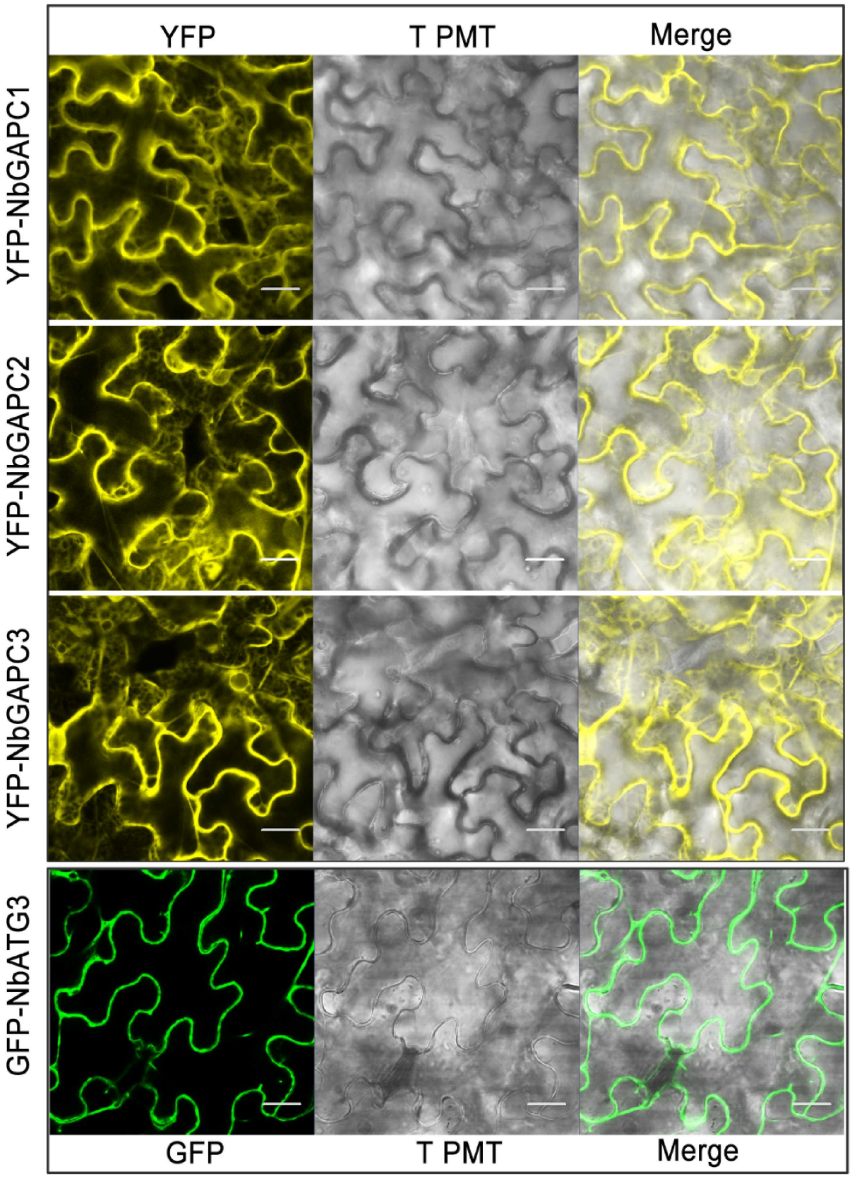


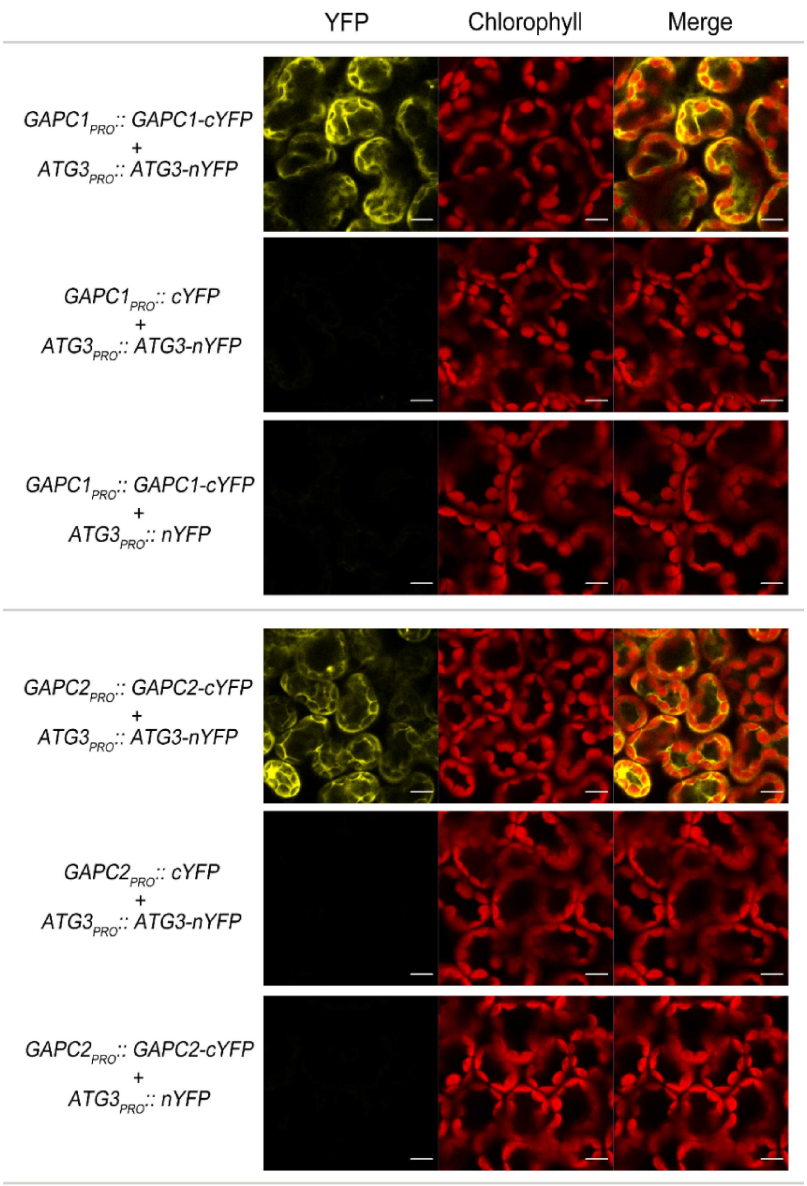

**Supplemental Figure 1. Alignment of the NbGAPC amino acid sequences with their *Arabidopsis* homologues.**

Homologs from *N. benthamiana* (NbGAPC1, NbGAPC2, NbGAPC3), *Arabidopsis* (*AtGAPC1, AT3G04120; AtGAPC2, AT1G13440)* were included. The alignment was generated using Clustal W2. Black, dark gray and white backgrounds represent residues that are conserved in 100%, above 80% or below 60% of the sequences at the corresponding positions. Capital letters under each block indicate residues that are consensus in all aligned sequences and the lowercase letters indicate mostly conserved residues other than consensus ones. Asterisk above indicates the marker line of ten amino acid residues. Numbers at the right indicate the positions of amino acid residues.



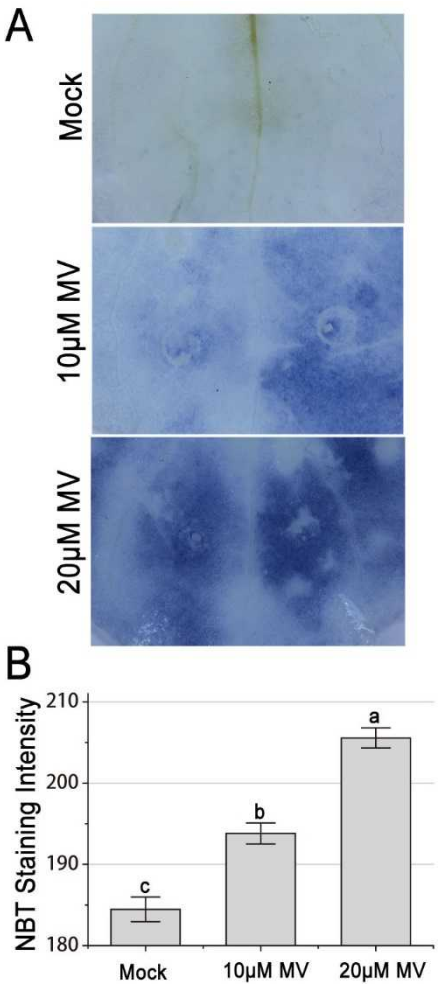

**Supplemental Figure 2. Subcellular localization of YFP-NbGAPCs and GFP-NbATG3 in *N. benthamiana* epidermal cells.**

YFP-NbGAPCs and GFP-NbATG3 was transiently expressed in *N. benthamiana* leaves via agroinfiltration and the images of epidermal cells (T PMT images showed) was taken at 60 hpi by LSM microscopes. Yellow color indicated YFP-NbGAPCs mainly localized in cytoplasm (upper three panels); Green color showed the cytoplasmic localization of GFP-NbATG3 (lower panel). Bars =20 μm.



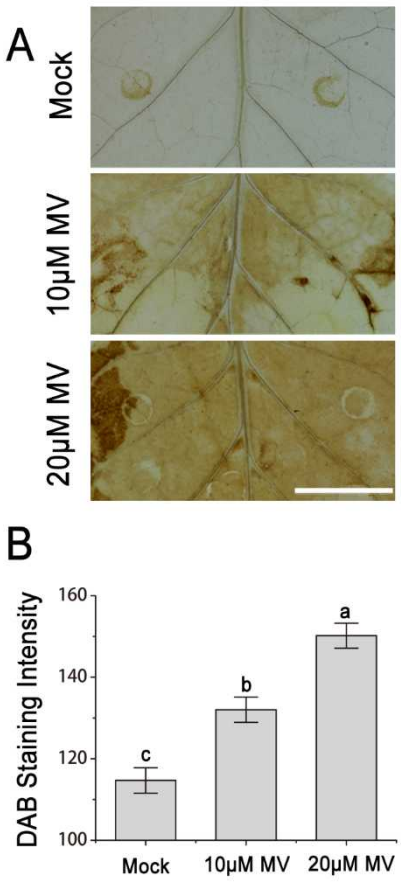

**Supplemental Figure 3. BiFC assays show the ATG3-GAPCs interactions when they are expressed under control of their native promoters.**

We used 2000bp of DNA fragment upstream of the start codon of *ATG3*, *GAPC1* or *GAPC2* as the native promoter of each corresponding gene, and named $ATG3_{PRO}$, $GAPC1_{PRO}$, $GAPC2_{PRO}$. $ATG3_{PRO}$*::ATG3-nYFP* or $ATG3_{PRO}$*::nYFP* was co-expressed transiently with $GAPC1_{PRO}$*::GAPC1-cYFP*, $GAPC1_{PRO}$*::cYFP* (upper panel), $GAPC2_{PRO}$*::GAPC2-cYFP* or $GAPC2_{PRO}$*::cYFP* (lower panel) in *N. benthamiana* leaves. Fluorescence was detected for mesophyll cells. Combinations of $ATG3_{PRO}$*::ATG3-nYFP* with either $GAPC1_{PRO}$*::GAPC1-cYFP* or $GAPC2_{PRO}$*::GAPC2-cYFP*, but not other combinations, gave YFP fluorescence signals (left). Yellow color indicates positive interaction signal while red color indicates the signals from chloroplasts (middle). The experiments were repeated three times with similar results. Bars =10 μm.



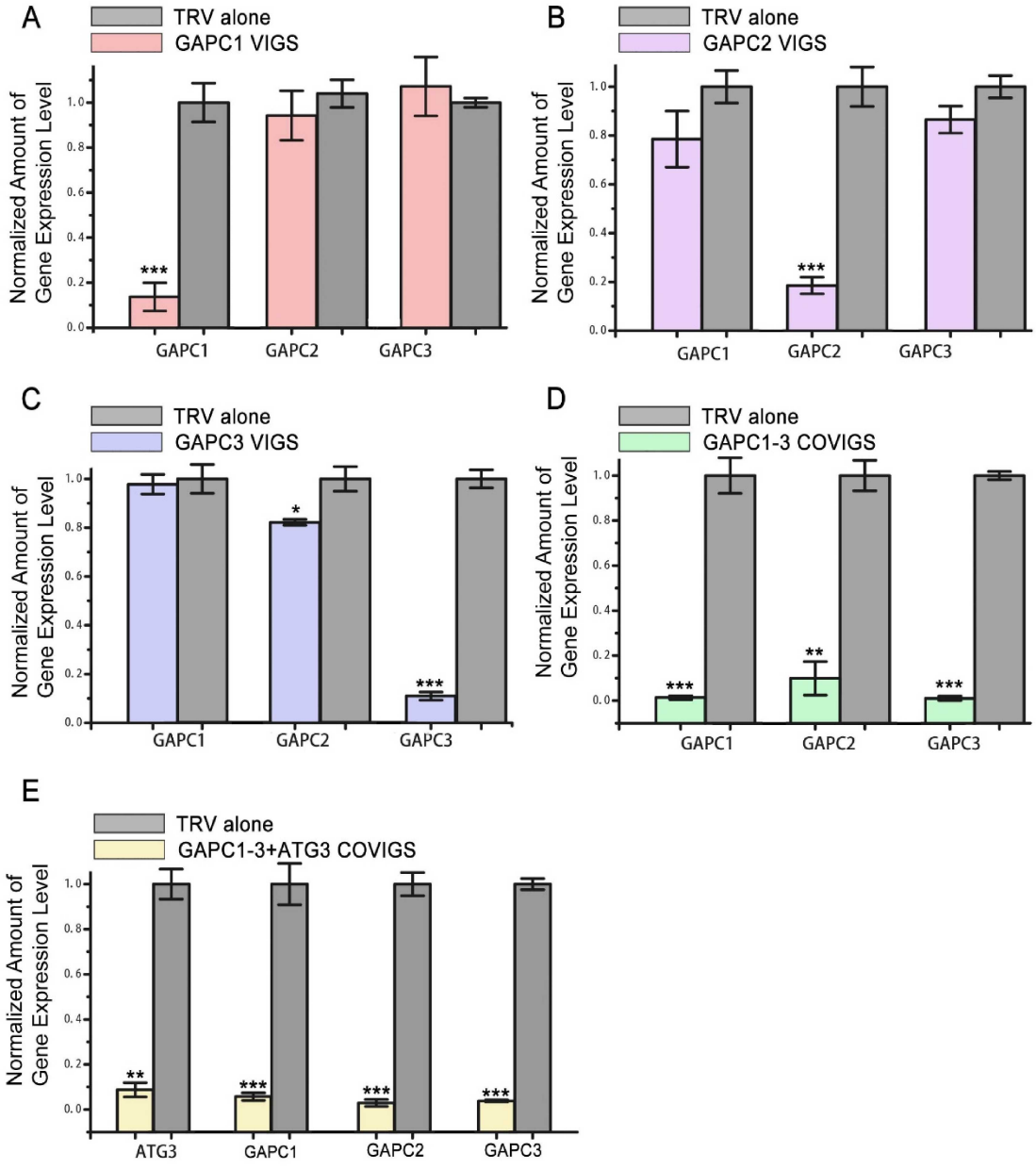

**Supplemental Figure 4. NBT staining indicates that MV concentration is positively correlated with superoxide anion accumulation.**

(A) Detection of superoxide anion accumulation in 10 μM MV treatment (middle panel) and 20 μM MV treatment leaves (low panel) other than in WT (up panel) leaves. Leaves detached from MV treatment or WT plants were vacuum-infiltrated with 0.1 mg/ mL nitroblue tetrazolium (NBT) in 25 mM Hepes buffer, pH 7.6. After 2 h incubation at room temperature in the dark, samples were transferred to 80% ethanol and destained at 90°C for 10 min. (B) For quantification of NBT staining intensity, images were converted to gray scale and inverted, and mean gray was calculated using ImageJ. Values represent means ± SE, n=8. Different letters indicate significant differences (ANOVA, $P < 0.05$).



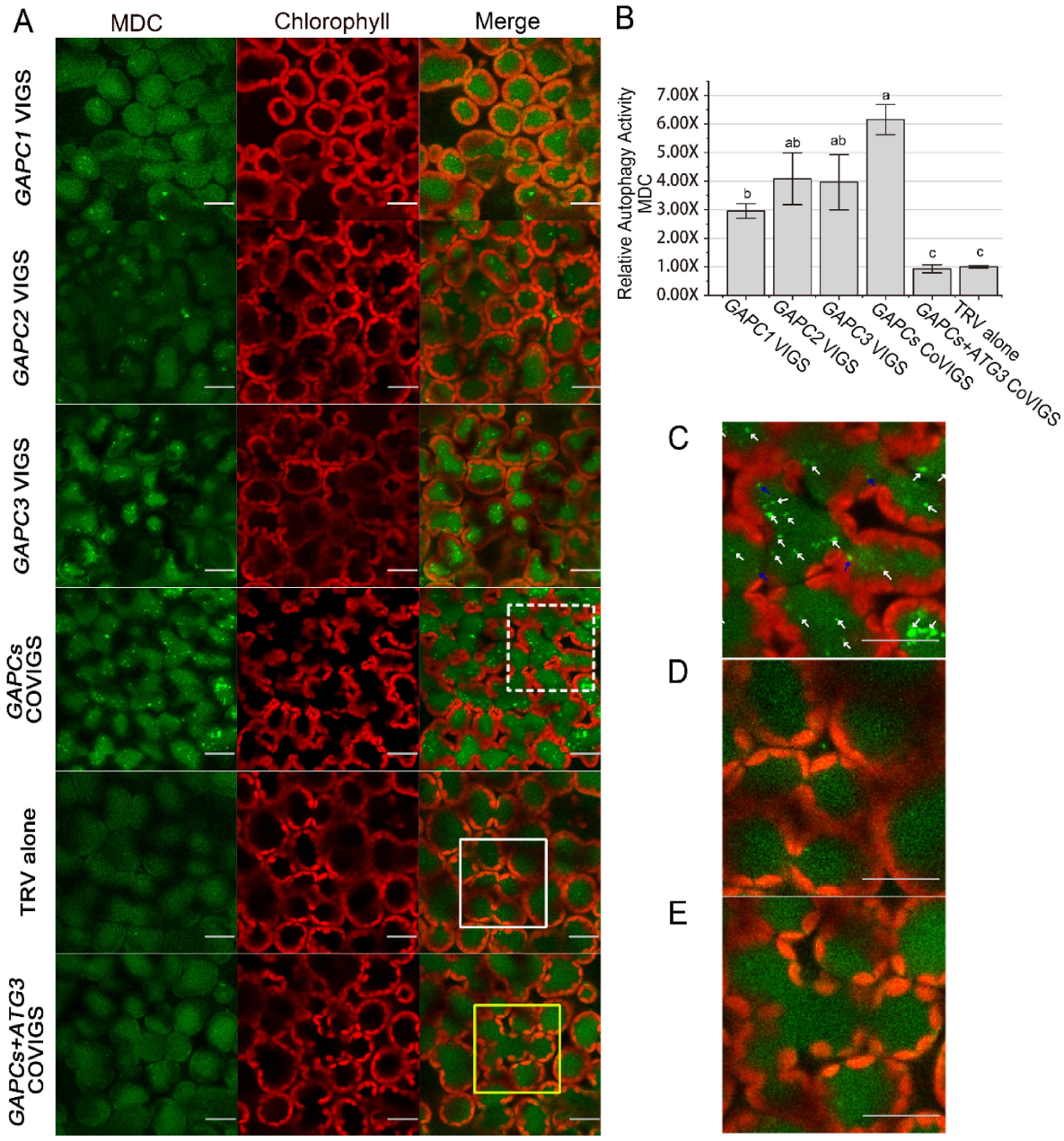

**Supplemental Figure 5. DAB staining indicates that MV concentration is positively correlated with $H_2O_2$ accumulation.**

(A) $H_2O_2$ accumulation was detected in 10 μM MV treatment (middle panel) and 20 μM MV treatment leaves (low panel), but not in $H_2O$ mock treatment leaves (up panel). Leaves detached from MV treatments or mock treatment plants were vacuum-infiltrated with 1 mg/mL DAB containing Tween 20 (0.05% v/v) and 10 mM sodium phosphate buffer (pH 7.0). After 4 hours incubation at room temperature in the dark, samples were transferred to 80% ethanol and destained at 90°C for 10 min. (B) For quantification of DAB staining intensity, images were converted to gray scale and inverted, and mean gray was calculated using ImageJ. Values represent means ± SE, n=8. Different letters indicate significant differences (ANOVA, $P < 0.05$).




**Supplemental Figure 6. Real-time RT-PCR to confirm VIGS efficiency.**

Real-time RT-PCR was performed using total RNA isolated from leaves of VIGS plants indicated on each chart. mRNA levels of *GAPC1*, *GAPC2*, *GAPC3* and *ATG3* were measured by real-time PCR normalized to *Nb eIF4A*. For each gene, the expression data of VIGS vector (TRV alone) was set as 1.0. Values represent means ± SE from three independent experiments. Student's t test was used to determine significant differences (***P < 0.001, 0.001<**P < 0.01, 0.01<*P<0.05).



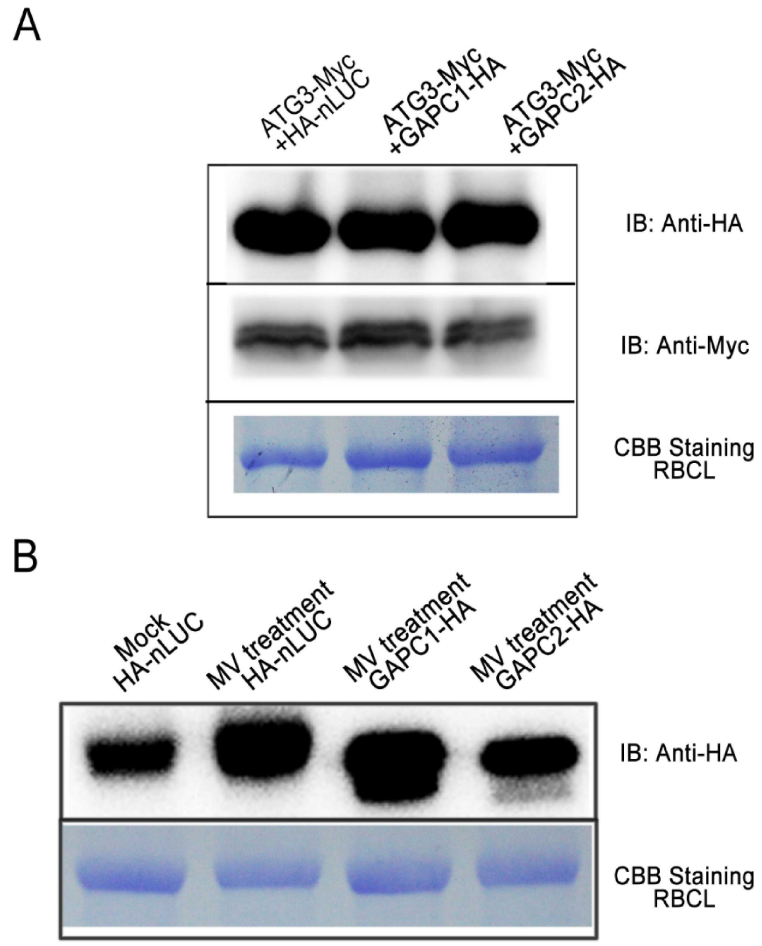

**Supplemental Figure 7. MDC staining indicates that silencing of *GAPCs* activates autophagy.**

(A) Representative images of MDC-stained mesophyll cells. MDC revealed that autophagy was activated in *GAPC* individually silenced plants (*GAPC1* VIGS, *GAPC2* VIGS and *GAPC3* VIGS), *GAPCs* co-silenced plants (*GAPCs* CoVIGS) but not in plants for co-silencing four genes *GAPC1-3* together with *ATG3* (*GAPCs+ATG3* CoVIGS). MDC-stained autophagic structures are in green and the chloroplasts are in red. Bars = 20 μm.

(B) Relative autophagic activity in *GAPC* silenced plants was normalized to that of TRV control plants, which was set to 1.0. Quantification of the MDC positive

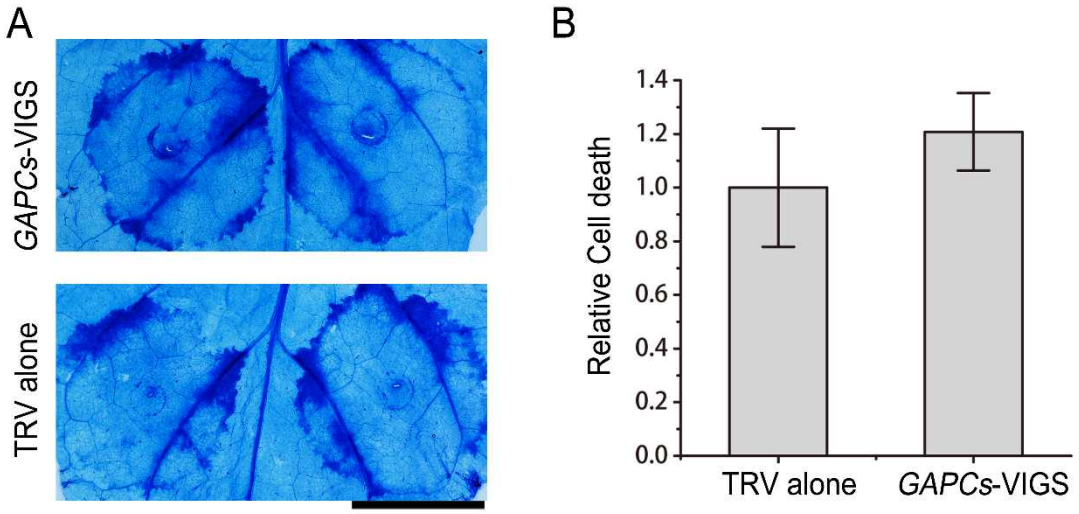
structures per cell was performed. More than 150 mesophyll cells for each treatment were used for the quantification. Values represent means ± SE from three independent experiments. Different letters indicate significant differences (ANOVA, $P < 0.05$).

(C)–(E) Magnification of the mesophyll cells in (A) surrounded by a dashed line (C), a solid line (D) and a yellow line (E). MDC-positive autophagic structures were indicated by arrows. The white arrows indicated the individual and aggregated autophagic structures in the vacuole. The blue arrows referred to the autolysosomal structures in the cytoplasm. Bars = 20 μm.



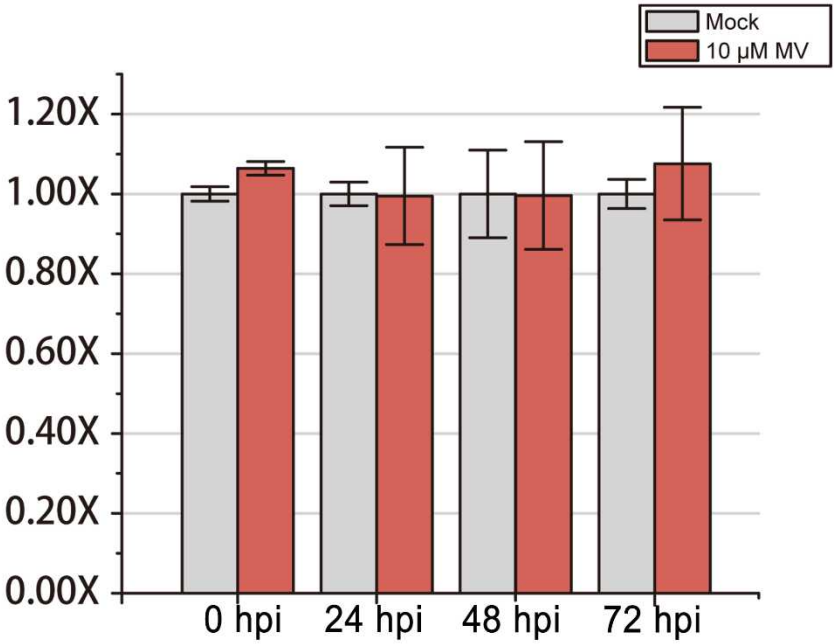

**Supplemental Figure 8. Protein expression tested by immunoblot.**

(A) *N. benthamiana* leaves co-expressing ATG3-Myc with HA-nLUC, GAPC1-HA and or GAPC2-HA were separately collected at 60 hpi. The leaf lysates were subjected to SDS-PAGE followed by immunoblotting with the indicated antibodies. Western blot assays confirmed similar expression level among various samples. (B) Leaves overexpressing HA-nLUC, GAPC1-HA and GAPC2-HA, followed by treatment with 10 μM MV or mock $H_2O$ at 60 hpi, were separately collected. The leaf lysates were subjected to SDS-PAGE followed by immunoblotting using anti-HA antibody. Western blot assays showed similar expression among HA tagged proteins under different treatments. The RuBisCO large subunit was used as a loading control and indicated by Coomassie Brilliant Blue staining.




**Supplemental Figure 9. Silencing of *GAPCs* does not have an obvious effect on HR cell death induced by nonhost pathogen *Pst* DC3000.**

(A) *NbGAPCs*-silenced (upper panel) and control plants (TRV alone, lower panel) were challenged with nonhost pathogens *P. syringae* pv. *tomato* strain DC3000 at $10^7$ cfu/mL. Representative photographs were taken at 15-hr after inoculation after trypan blue staining. Scale bars represent 1 cm.
(B) Quantitative representation of *Pst* DC3000-induced HR PCD. For quantification of death intensity, images were converted to gray scale, and mean gray value of inoculation area minus non-inoculation area was calculated using ImageJ. Values represent means ± SE (n=8). The experiments were repeated twice with similar results.




**Supplemental Figure 10. MV application does not affect mRNA level of *ATG3.***

Leaves infiltrated with 10 μM MV or $H_2O$ mock treatment was collected at indicated time points. hpi, hours post-infiltration. Real-time RT-PCR was performed to test mRNA level of *ATG3* by using *NbEif4a* as an internal control.

**Supplemental Table 1 Primers used in generating constructs.**

| Note | Primer Name | Sequence (5'-3') |
|---|---|---|
| For making NbATG3-GFP | LIC1-ATG3-F | CGACGACAAGACCGTAACCATGGTACTGTCACAGAAGATTCACGAAG |
| | ATG3-GFP fusion-R | TTTGCTAGCGGTGCTGCTGCTA |
| | ATG3-GFP fusion-F | AGCAGCACCGCTAGCAAAGGAGA |
| | LIC2-GFP-R | GAGGAGAAGAGCCGTCTATTTGTAGAGCTCAT |
| For making NbATG3-nLUC | LIC1-ATG3-F | See above |
| | ATG3-nLUC fusion-R | GGCGTCTTCGGTGCTGCTGCTAC |
| | ATG3-nLUC fusion-F | GCACCGAAGACGCCAAAAACATA |
| | LIC2-nLUC-R | GAGGAGAAGAGCCGTTCATCCATCCTTGTCAAT |
| For making NbATG3-nYFP | LIC1-ATG3-F | See above |
| | ATG3-nYFP fusion-R | CTCACCATGGTGCTGCTGCTACC |
| | ATG3-nYFP fusion-F | AGCACCATGGTGAGCAAGGGCGA |
| | LIC2-nYFP-R | GAGGAGAAGAGCCGTTCAGGCCATGATATAGACG |
| For making NbATG3-Myc | LIC1-ATG3-F | See above |
| | ATG3-Myc fusion-R | ATTAACCCGGTGCTGCTGCTACC |
| | ATG3-Myc fusion-F | CAGCACCGGGTTAATTAACGGTG |
| | LIC2-Myc-R | GAGGAGAAGAGCCGTCTAAGCGCTACCGTTCA |
| For making GFP-NbGAPC3 | LIC1-GFP-F | CGACGACAAGACCGTAACCATGGCTAGCAAAGGAGAAG |
| | GFP-ATG3 fusion-R | ACAGTACCATTTTGTAGAGCTCATC |
| | GFP-ATG3 fusion-F | GCTCTACAAAATGGTACTGTCACAG |
| | LIC2-ATG3-R | GAGGAGAAGAGCCGTTCAGGTGCTGCTGCTA |
| For making NbGAPC1-GFP | LIC1-GAPC1-F | CGACGACAAGACCGTAACCATGGCATCTGACAAGAA |
| | GAPC1-GFP fusion-R | TTGCTAGCAACAGAAGCCATATGGC |
| | GAPC1-GFP fusion-F | TGGCTTCTGTTGCTAGCAAAGGAGAA |
| | LIC2-GFP-R | See above |
| For making NbGAPC1-cYFP | LIC1-GAPC1-F | See above |
| | GAPC1-cYFP fusion-R | CTGCTTGTCAACAGAAGCCATATGGC |
| | GAPC1-cYFP fusion-F | GGCTTCTGTTGACAAGCAGAAGAACG |
| | LIC2-cYFP-R | GAGGAGAAGAGCCGTCATTACTTGTACAGCTCGTCC |
| For making NbGAPC1-HA | LIC1-GAPC1-F | See above |
| | GAPC1-HA fusion-R | TAACCCCATAACAGAAGCCATATGGC |
| | GAPC1-HA fusion-F | ATGGCTTCTGTTATGGGGTTAATTAAC |
| | LIC2-HA-R | GAGGAGAAGAGCCGTCAGCTGCACTGAGCA |

| | | |
|---|---|---|
| For making cLUC-NbGAPC1 | LIC1-cLUC-F | CGACGACAAGACCGTGACCATGTCCGGTTATGTAAA |
| | cLUC-GAPC1 fusion-R | GATGCCATCACGGCGATCTTTCCG |
| | cLUC-GAPC1 fusion-F | ATCGCCGTGATGGCATCTGACAAG |
| | LIC2-GAPC1-R | GAGGAGAAGAGCCGTCAAGCAACAGAAGCCAT |
| For making YFP-NbGAPC1 | LIC1-YFP-F | CGACGACAAGACCGTAACCATGGTGAGCAAGGGCGAG |
| | YFP-GAPC1 fusion-R | AGATGCCATCTTGTACAGCTCGTCCA |
| | YFP-GAPC1 fusion-F | TGTACAAGATGGCATCTGACAAG |
| | LIC2-GAPC1-R | See above |
| For making NbGAPC2-GFP | LIC1-GAPC2-F | CGACGACAAGACCGTAACCATGGCCAAGGTTAAGAT |
| | GAPC2-GFP fusion-R | TGCTAGCCTGAACCGATGCCAT |
| | GAPC2-GFP fusion-F | CGGTTCAGGCTAGCAAAGGAGAA |
| | LIC2-GFP-R | See above |
| For making NbGAPC2-cYFP | LIC1-GAPC2-F | See above |
| | GAPC2-cYFP fusion-R | TGCTTGTCCTGAACCGATGCCAT |
| | GAPC2-cYFP fusion-F | GGTTCAGGACAAGCAGAAGAACG |
| | LIC2-cYFP-R | GAGGAGAAGAGCCGTCATTACTTGTACAGCTCGTCC |
| For making NbGAPC2-HA | LIC1-GAPC2-F | See above |
| | GAPC2-HA fusion-R | ACCCCATCTGAACCGATGCCAT |
| | GAPC2-HA fusion-F | TCGGTTCAGATGGGGTTAATTAAC |
| | LIC2-HA-R | See above |
| For making cLUC-NbGAPC2 | LIC1-cLUC-F | See above |
| | cLUC-GAPC2 fusion-R | GGCCATCACGGCGATCTTTCCG |
| | cLUC-GAPC2 fusion-F | CGCCGTGATGGCCAAGGTTAAGA |
| | LIC2-GAPC2-R | GAGGAGAAGAGCCGTTACTGAACCGATGCC |
| For making YFP-NbGAPC2 | LIC1-YFP-F | See above |
| | YFP-GAPC2 fusion-R | TGGCCATCTTGTACAGCTCGTCCA |
| | YFP-GAPC2 fusion-F | CTGTACAAGATGGCCAAGGTTAAGA |
| | LIC2-GAPC2-R | See above |

| | | |
|---|---|---|
| For making cLUC-NbGAPC3 | LIC1-cLUC-F | See above |
| | cLUC-GAPC3 fusion-R | TGGCCATCACGGCGATCTTTCCG |
| | cLUC-GAPC3 fusion-F | TCGCCGTGATGGCCAAGGTTAAGA |
| | LIC2-GAPC3-R | GAGGAGAAGAGCCGTTACTGGACTGATGCC |
| For making cYFP-NbATG8f | LIC1-cYFP-F | CGACGACAAGACCGTGACCATGGACAAGCAGAAGA |
| | cYFP-ATG8f fusion-R | AGCCATCTTGTACAGCTCGTCC |
| | cYFP-ATG8f fusion-F | GTACAAGATGGCTAAGAGCTCAT |
| | LIC2-ATG8f-R | GAGGAGAAGAGCCGTCTACAGCTTGTTCAG |
| For making GFP-NbATG8f | LIC1-GFP-F | CGACGACAAGACCGTAACCATGGCTAGCAAAGGAGAAG |
| | GFP-ATG8f fusion-R | CTTAGCCATTTTGTAGAGCTCATC |
| | GFP-ATG8f fusion-F | TCTACAAAATGGCTAAGAGCTCAT |
| | LIC2-ATG8f-R | See above |
| For making ATG3 | LIC1-ATG3-F | CGACGACAAGACCGTAACCATGGTACTGTCACAGAAGATTCACGAAG |
| | LIC2-ATG3-R | GAGGAGAAGAGCCGTACGGTGCTGCTGCTACCAAGATCAAAG |
| For making NbGAPC1-3×FLAG-6×His | *Bam*HI-GAPC1-F | CGCGGATCCATGGCATCTGACAAGAAGAT |
| | *Xho*I-GAPC1-R | CCGCTCGAGTGCAACAGAAGCCATATGGC |
| For making NbGAPC2-3×FLAG-6×His | *Bam*HI-GAPC2-F | CGCGGATCCATGGCCAAGGTTAAGATTGG |
| | *Xho*I-GAPC2-R | CCGCTCGAGCTGAACCGATGCCATGTGCT |
| For making GST-ATG3 | *Nde*I-ATG3-F | CGCCATATGGTACTGTCACAGAAGATTCAC |
| | *Xho*I-ATG3-R | CGCCTCGAGTCAGGTGCTGCTGCTACCAAGATC |
| For making pTRV2-NbGAPC1 | LIC1-GAPC1 3'UTR-F | CGACGACAAGACCGTCAGCTCTCGTGTGATTG |
| | LIC2-GAPC1 3'UTR -R | GAGGAGAAGAGCCGTCAGGAACATAGG |
| For making pTRV2-NbGAPC2 | LIC1-GAPC2 3'UTR-F | CGACGACAAGACCGTGAGTGGTGGACTTGATTAA |
| | LIC2-GAPC2 3'UTR -R | GAGGAGAAGAGCCGTTCCCCATTGGATTTCCAGGGC |
| For making pTRV2-NbGAPC3 | LIC1-GAPC3 3'UTR-F | CGACGACAAGACCGTGCATCAGTCCAGTAAAGT |
| | LIC2-GAPC3 3'UTR-R | GAGGAGAAGAGCCGTTATGTGAGGGTCCAA |

| | | |
|---|---|---|
| For making pTRV2-GAPCs | LIC1-GAPC1 for fusion-F | CGACGACAAGACCGTCAGCTCTCGTGTGAT |
| | GAPC1-GAPC2 fusion-R | CACCACTCCAGGAACATAGGTAG |
| | GAPC1-GAPC2 fusion-F | GTTCCTG GAGTGGTGGACTTGAT |
| | GAPC2-GAPC3 fusion-R | GATGCTCCCCATTGGATTTCCA |
| | GAPC2-GAPC3 fusion-F | ATGGGGA GCATCAGTCCAGTA |
| | LIC2-GAPC3 for fusion--R | GAGGAGAAGAGCCGTATGTGAGGGTCCAACA |
| For making pTRV2-NbGAPCs+ NbATG3 | LIC1-GAPC1 for fusion-F | See above |
| | GAPC1-GAPC2 fusion-R | See above |
| | GAPC1-GAPC2 fusion-F | See above |
| | GAPC2-GAPC3 fusion-R | See above |
| | GAPC2-GAPC3 fusion-F | See above |
| | GAPC3-ATG3 fusion-R | AATATGACTTCCTCTTTATGTGAGGGTCCAACA |
| | GAPC3-ATG3 fusion-F | CCCTCACATAAAGAGGAAGTCATATTTACCTG |
| | LIC2-ATG3 for fusion-R | GAGGAGAAGAGCCGTCGCTCCCATGTCTGGTAT |
| ATG3 promoter | *Eco*RI-ATG3 promoter-F | CGCGAATTCCGAAAGAACTGACGAGAGGAAGATT |
| | *Sac*I-ATG3 promoter-R | CGCGAGCTCTTTCTCCGCCCTCCACTTTCACGGT |
| GAPC1 promoter | *Eco*RI-GAPC1 promoter-F | CGCGAATTCTTGAACGAACTCGGATTTGAACTAT |
| | *Sac*I-GAPC1 promoter-R | CGCGAGCTCGGCTTAGGAGAGAAGAATGGGGTTT |
| GAPC2 promoter | *Eco*RI-GAPC2 promoter-F | CGCGAATTCGCAGATCAAGAGGTTGAGCGTCCGT |
| | *Sac*I-GAPC2 promoter-R | CGCGAGCTCGATTTTCGAGAAATGAGCGGAGGAT |

**Supplemental Table 2. Primers used in RT-PCR.**

| NOTE | Primer Name | Sequence(5'-3') |
|---|---|---|
| For GAPC1 detection | GAPC1 5'UTR RT-F | CAGACTGCAACCCTACACTTTCGC |
| | GAPC1 5'UTR RT-R | CCACCAAACGACCAATCCTTCCAA |
| For GAPC2 detection | GAPC2 5'UTR RT-F | CCTCCGCTCATTTCTCGAAAATCA |
| | GAPC2 5'UTR RT-R | GCCACTAATCGGCCAATTCTTCC |
| For GAPC3 detection | GAPC3 5'UTR RT-F | TGCCGGTCTCCCAGTACCAACTA |
| | GAPC3 5'UTR RT-R | GCAACCAGTCGCCCAATTCTTCC |
| For ATG3 detection | ATG3 RT-F | GCCTCGTGTATGGCTCACTGGAT |
| | ATG3 RT-R | CCGTGCCGACAAGGATGTACTGAA |
| For TMV-GFP detection | TMV CP RT-F | GCCTGGAAACCTGTGCCTAGT |
| | TMV CP RT-R | GCATCGTCTACCCTCTGAGTCG |
| For internal control | eIF4A RT-F | GCTTTGGTCTTGGCACCTACTC |
| | eIF4A RT-R | TGCTCGCATGACCTTTTCAA |