Supplementary Figure 1. Sequence alignment of FHD1 protein from N. benthamiana (NbFDH1), tobacco (NtFDH1), tomato (SIFDH1), and Arabidopsis (AtFDH1). Sequence information was obtained from public database; TAIR, NCBI GenBank, and Sol Genomics Network. The software MEGA-X (Kumar et al., 2018) was used for sequence alignment. The colors amino acids were according to the default coloring schemes of ClustalX alignment, which depends on both residue type and the pattern of conservation within a column (http://www.clustal.org/clustal2/).
Supplementary Figure 2. The expression of NbFDH1 gene is reduced in NbFDH1-silenced N. benthamiana plants. Two weeks old N. benthamiana seedlings were inoculated with TRV1 + TRV::00 (control) or TRV1 + TRV::NbFDH1. Three weeks after TRV inoculation, leaf samples from three different biological replicates for each construct were collected and gene expression was measured by RT-qPCR. NbActin was used as internal control for normalization. Bars represent mean and error bars represent standard deviation for three biological replicates (four technical replicates for each biological sample). Asterisk represents statistical significance that was determined using Student’s t-test, (P < 0.01).
Supplementary Figure 3. *AtFDH1* is upregulated upon inoculation with host and nonhost pathogens in wild-type Col-0 and some defense related genes are differentially expressed in *Atfdh1* mutant. (A) Gene expression patterns of *AtFDH1* against *P. syringae* bacterial pathogen in Arabidopsis. This data was obtained from Arabidopsis eGFP Browser at bar.utoronto.ca (Winter et al., 2007). (B) *AtFDH1* is induced by host and nonhost pathogen inoculations. Four-weeks-old Arabidopsis wild-type (Col-0) were flood-inoculated with host (*P. syringae* pv. maculicola, Psm) or nonhost (*P. syringae* pv. tabaci, Pstab) pathogens. The 24 hours after inoculation, leaves were harvested, total RNA was extracted and subject to RT-qPCR using *AtFDH1* specific primers. *AtActin* was used as internal control for normalization. (C) Gene expression patterns of defense related genes in wild-type and the *Atfdh1* mutant without any biotic or abiotic stresses. Leaves of four weeks old Arabidopsis wild-type (Col-0) and *Atfdh1* mutant (*fdh1-1*) plants were collected, total RNA was isolated and subject to RT-qPCR to measure the transcripts of *PAD4*, *EDS1*, *NPR1*, and *PDF1.2*. Bars represent mean and error bars represent standard deviation for three biological replicates (four technical replications for each biological replicate). Asterisks represent statistical significance as determined using Student’s t-test, (P < 0.01).
Supplementary Figure 4. Subcellular localization of AtFDH1 in *N. benthamiana*. For *Agrobacterium*-mediated transient assay, GFP protein fused to the C-terminal of AtFDH1 was transformed into the *A. tumefaciens* strain GV3101. The *Agrobacterium* suspension was (5×10^7 CFU/ml) was infiltrated using a syringe into leaves, and the expression was observed 3 days after the agroinfiltration. Red channel (a 561 nm excitation, 570-620 nm emission filter), showing mitochondria stained with MitoTracker dye was used; green channel showing AtFDH1-GFP. Bars = 10 μm.
Supplementary Figure 5. Subcellular localization of AtFDH1 in Arabidopsis leaves. The expression and localization of AtFDH1-GFP was observed in detached (no stress) and peeled adaxial epidermal cells (pathogen stress) from leaves of transgenic Arabidopsis lines expressing AtFDH1-GFP in Col-0. The protein localization was also examined in detached leaf samples 1-hr after the treatment of *P. syringae* pv. tomato DC3000 (1×10^5 CFU/ml) and *P. syringae* pv. phaseolicola (1×10^5 CFU/ml). Red channel (a 561 nm excitation, 570-620 nm emission filter), showing chloroplast; green channel showing AtFDH1-GFP. Scale bar = 10 μm.