

Supplementary Table 1: Arabidopsis lines used in this study.

	Mutant line	Gene	Type of mutation(s)/transgene	Source
mdl mutant set 1	<i>mdl1-1</i> (SAIL_772_G01)	<i>MDL1</i> (AT5G57170)	T-DNA insertion, exon 2	NASC: N834538
	<i>mdl2-2</i> (SALK_078883)	<i>MDL2</i> (AT5G01650)	T-DNA insertion, intron 2	NASC: N656281
	<i>mdl3-3</i> (GK_750H06)	<i>MDL3</i> (AT3G51660)	T-DNA insertion, intron 1	NASC: N471994
	<i>mdl1-1 mdl2-2</i>	<i>MDL1, MDL2</i>	T-DNA insertions	Crossing, this study
	<i>mdl1-1 mdl3-3</i>	<i>MDL1, MDL3</i>	T-DNA insertions	Crossing, this study
	<i>mdl2-2 mdl3-3</i>	<i>MDL2, MDL3</i>	T-DNA insertions	Crossing, this study
	<i>mdl1-1 mdl2-2 mdl3-3</i>	<i>MDL1, MDL2, MDL3</i>	T-DNA insertions	Crossing, this study
mdl mutant set 2	<i>mdl1-2</i> (GK_786B08)	<i>MDL1</i> (AT5G57170)	T-DNA insertion, intron 2	NASC: N475380
	<i>mdl2-1</i> (SALK_024488C)	<i>MDL2</i> (AT5G01650)	T-DNA insertion, intron 1	NASC: N578883
	<i>mdl3-2</i> (SM_3_31346)	<i>MDL3</i> (AT3G51660)	T-DNA insertion, 3' UTR	NASC: N118057
	<i>mdl1-2 mdl2-1</i>	<i>MDL1, MDL2</i>	T-DNA insertions	Crossing, this study
	<i>mdl1-2 mdl3-2</i>	<i>MDL1, MDL3</i>	T-DNA insertions	Crossing, this study
	<i>mdl2-1 mdl3-2</i>	<i>MDL2, MDL3</i>	T-DNA insertions	Crossing, this study
	<i>mdl1-2 mdl2-1 mdl3-2</i>	<i>MDL1, MDL2, MDL3</i>	T-DNA insertions	Crossing, this study
Other mutants	<i>eds1-2</i> [Col-0]	<i>EDS1</i> (AT3G48090)	Point mutation	(84)
	<i>sid2-1</i>	<i>ICS1</i> (AT1G74710)	Point mutation	(46)
	<i>co-10</i>	<i>CO</i> (AT5G15840)	T-DNA insertion	(33)
	<i>svp-41</i>	<i>SVP</i> (AT2G22540)	Transposon insertion	(32)
	<i>pen2-1</i>	<i>PEN2</i> (AT2G44490)	Point mutation	(37)
	<i>mlo2-5 mlo6-2 mlo12-1</i>	<i>MLO2</i> (AT1G11310), <i>MLO6</i> (AT1G61560), <i>MLO12</i> (AT2G39200)	T-DNA insertion	(35)
	<i>mlo2-6 mlo6-4 mlo12-8</i>	<i>MLO2</i> (AT1G11310), <i>MLO6</i> (AT1G61560), <i>MLO12</i> (AT2G39200)	T-DNA insertions	(36)
	<i>mdl1-1 mdl2-2 mdl3-3 sid2-1</i>	<i>MDL1, MDL2, MDL3, ICS1</i>	T-DNA insertions, point mutation	Crossing, this study

Transgenic lines	<i>mCherry-MDL1.1</i>	<i>MDL1 (AT5G57170)</i>	<i>35S::mCherry-MDL1.1</i>	Transformation, this study
	<i>mCherry-MDL1.2</i>	<i>MDL1 (AT5G57170)</i>	<i>35S::mCherry-MDL1.2</i>	Transformation, this study
	<i>mCherry-MDL2.1</i>	<i>MDL2 (AT5G01650)</i>	<i>35S::mCherry-MDL2.1</i>	Transformation, this study
	<i>mCherry-MDL2.2</i>	<i>MDL2 (AT5G01650)</i>	<i>35S::mCherry-MDL2.2</i>	Transformation, this study
	<i>mCherry-MDL3</i>	<i>MDL3 (AT3G51660)</i>	<i>35S::mCherry-MDL3</i>	Transformation, this study

Supplementary Table 2: Vectors used in this study.

Vector	Description	Source
pJawohl2b 3xFLAG-GWY	Plant expression vector (N-terminal FLAG tag)	Parker lab, MPIPZ Cologne
p35S-mCherry-GWY	Plant expression vector (N-terminal mCherry tag)	This study
p35S-GWY-mCherry	Plant expression vector (C-terminal mCherry tag)	This study
pAMPAT nLUC-GWY	Split luciferase (N-terminal Luciferase ^{N-terminus})	This study
pAMPAT cLUC-GWY	Split luciferase (N-terminal Luciferase ^{C-terminus})	This study

Supplementary Table 3: Primers used in this study.

	Target	Primer name	Primer sequence (5' to 3')
RT-PCR	<i>AT5G57170</i>	RT-MDL1_Fwd2	TCATCGGCAAACCTGAATCCT
	<i>AT5G57170.1</i>	RT-MDL1.1_Rev	GTAACCGAAGAAAGGTCGCG
	<i>AT5G57170.2</i>	RT-MDL1.2_Rev	GAATTTGAAGCAAAAGGGGACTTAC
	<i>AT5G01650</i>	RT-MDL2_Fwd2	CATCGGCAAGCCTGAGAACT
	<i>AT5G01650.1</i>	RT-MDL2.1_Rev2	CCAACCAAAGAAGGATCCCTTG
	<i>AT5G01650.2/3</i>	RT-MDL2.2_Rev3	GCATATTCTTGACTTTGATGGTCCTT
	<i>AT5G01650.4</i>	RT-MIF2.4-Rev	TCAGAGAGAGGGAATTGCTTGC
	<i>AT3G51660</i>	RT-MDL3_Fwd	GGACGACCTCAAACTTAGTGA
	<i>AT3G51660</i>	RT-MDL3_Rev	AGTTTAGAAGGAAGAGGCAAAGA
	<i>AT4G26410</i>	RT-EXPR_Fwd	CTATTGGGATTGGTGTGCTG
	<i>AT4G26410</i>	RT-EXPR_Rev	AAGTCATGGAAGCCACTTCAG
Mutant screen	<i>AT5G57170</i>	At_md11-1_LP	TACATTTGGGCTGAAATTTGC
	<i>AT5G57170</i>	At_md11-1_RP	AGGTTGAATTCGCCTCTTTTC
	<i>AT5G57170</i>	At_md11-2_LP	ACTCTAACCTCTGTTTCGGC
	<i>AT5G57170</i>	At_md11-2_RP	GAAGTCGCGTTTGAAAAGTTG
	<i>AT5G01650</i>	At_md12-1_LP	CTAACCAATTAGGTGCGTTTCG
	<i>AT5G01650</i>	At_md12-1_RP	GAGGAAGAATCGAGACTTGGG
	<i>AT5G01650</i>	At_md12-2_LP	TCTCCACCAACGTTAACCTTG
	<i>AT5G01650</i>	At_md12-2_RP	AAGAAGGATCCCTGCAAGAAG
	<i>AT3G51660</i>	At_md13-2_LP	TCCATTTCTGAGTTTTGATTG
	<i>AT3G51660</i>	At_md13-2_RP	TACTTGACCCCTCAAACCAAG
	<i>AT3G51660</i>	At_md13-3_LP2	GCTCTATATATAATTGGCCATGTG
	<i>AT3G51660</i>	At_md13-3_RP2	AGATTATAGGGTCAAGATCTACATG
	<i>T-DNA</i>	GABI_LB	ATATTGACCATCATACTCATTGC
	<i>T-DNA</i>	SAIL_LB3	TAGCATCTGAATTTTCATAACCAATCTCGATACAC
	<i>T-DNA</i>	SALK_LBb1.3	ATTTTGCCGATTTTCGGAAC
	<i>T-DNA</i>	SM_Spm32_LB	TACGAATAAGAGCGTCCATTTTAGAGTGA
	<i>AT1G74710</i>	At_sid2-1_Fwd	GTGTCTGCAGTGAAGCTTTGG
	<i>AT1G74710</i>	At_sid2-1_Rev	GCTGGAAGCCCACAAACAGC

Supplementary Table 4: Mass transitions and optimized parameters for detection of phytohormones and defense metabolites by mass spectrometry ^a.

Phytohormone	RT [min]	Ionization mode	Q1 [<i>m/z</i>]	Q3 [<i>m/z</i>]	DP [V]	EP [V]	CE [V]	CXP [V]
SA	2.0	Negative	137	93	-25	-6	-20	-10
SAG	1.0	Negative	299	137	-30	-4	-18	-2
JA-Ile	5.0	Negative	322	130	-45	-5	-28	-2
Pip	0.7	Positive	130	84	90	8	22	4
NHP	0.7	Negative	144	82	-60	-8	-15	-13
ABA	3.5	Negative	263	153	-35	-4	-14	-2
Camalexin	5.0	Negative	199	141	-60	-4	-32	-2
ICA	3.0	Negative	160	116	-40	-6.5	-22	-2
D ₄ -SA	2.0	Negative	141	97	-25	-6	-22	-6
¹³ C ₆ -SAG	1.0	Negative	305	137	-30	-4	-18	-2
D ₃ -JA-Leu	5.0	Negative	325	133	-65	-4	-30	-2
D ₉ -Pip	0.7	Positive	139	93	90	8	22	4
D ₉ -NHP	0.7	Negative	153	90	-60	-8	-15	-13
D ₆ -ABA	3.5	Negative	269	159	-30	-5	-16	-2
D ₅ -IAA	3.1	Negative	179	135	-35	-9	-14	-2

^a RT is the retention time. The ionization mode depicts the polarity of the nanoESI source. Q1 and Q3 are the parent and product ion, respectively. CP, EP, CE, and CXP indicate the declustering potential, entrance potential, collision energy, and cell exit potential for each phytohormone/metabolite.