

Protocol S1. Extraction of leaf samples for liquid chromatography/mass spectrophotometry (LC/MS) in 2016.

For each sample, 100 mg frozen ground tissue (FW) was weighed into a 2-mL Eppendorf tube on dry ice and extracted with 1 mL of chilled (4°C) 80:20 acetonitrile:water to which internal standards had been added (2.5 ng [$^2\text{H}_4$] SA, 12.5 ng [$^2\text{H}_5$] JA, and 6.25 ng [$^2\text{H}_6$] ABA). The samples were shaken for 30 min at 4°C on a flat-bed orbital shaker, extracted overnight at -20°C and then centrifuged at 13,000 g for 10 min at 4°C. An 800- μL aliquot of each sample was transferred to a 96-well deep plate and evaporated to dryness in a refrigerated CentriVap vacuum concentrator at -4°C (Labconco, Kansas City, MO, USA). Samples were reconstituted in 200 μL of chilled (4°C) 80:20 acetonitrile:water and then made basic with the addition of 1.8 mL of 0.67% ammonium hydroxide (aq) prior to SPE clean-up on a SOLA SAX 96-well plate (10 mg/2 mL; Thermo Scientific, CA, USA). Plates were activated using 2x 1.5 mL acetonitrile and equilibrated using 2x 1.5 mL 0.67% ammonium hydroxide (aq). After equilibration, samples were loaded and washed with 1.5 mL of 0.67% ammonium hydroxide (aq) followed by 1.3 mL of methanol. Acidic plant hormones were eluted with 600 μL 3.77% formic acid in acetonitrile and evaporated to dryness in a refrigerated centrivap vacuum concentrator at -4°C. Samples were reconstituted in 200 μL of 10:90 acetonitrile:water for analysis by LC/MS.

Protocol S2. Extraction of leaf samples for LC/MS in 2018.

The samples, 100 mg frozen ground tissue (FW), were weighed into 1.5-mL Eppendorf tubes on dry ice, and to each was added 1 mL chilled (4°C) extraction solvent (acetonitrile + 0.01% TFA), labelled internal standard mix (2.5 ng [$^2\text{H}_4$] SA, 25 ng [$^2\text{H}_5$] JA, 6.4 ng [$^2\text{H}_6$] ABA, 0.6 ng [$^2\text{H}_{10}$] JA-ile (2018 season only)) and 0.8 g stainless steel beads 0.9–2 mm (Next Advance Inc., NY, USA). Samples were worked up as described in Bulley et al. [1] and reconstituted in 200 μL of 10:90 acetonitrile:water for analysis by LC/MS.

Table S1. Compounds used in liquid chromatography/mass spectrophotometry (LC/MS)

Compound Name	Abbreviation	Source
Radio-labelled salicylic acid O- β -glucoside	[² H ₄] SAG	synthesised utilising [² H ₄] ¹ MeSA in place of MeSA [2]
Salicylic acid O- β -glucoside	SAG	synthesised using published methodology [2] ²
12-hydroxy jasmonic acid	12-OH-JA	Ol-chemim Ltd (Olomouc, Czech Republic)
Radio-labelled salicylic acid	[² H ₄] SA	CDN Isotopes (Pointe-Claire, Quebec, Canada)
Salicylic acid	SA	Sigma Aldrich (Auckland, New Zealand)
Radio-labelled abscisic acid	[² H ₆] ABA	Toronto Research Chemicals (Toronto, Ontario, Canada)
Absciscic acid	ABA	Sigma Aldrich (Auckland, New Zealand)
Radio-labelled jasmonic acid	[² H ₅] JA	CDN Isotopes (Pointe-Claire, Quebec, Canada)
Jasmonic acid	JA	Sigma Aldrich (Auckland, New Zealand)
9,10-dihydrojasmonic acid	DH-JA	Ol-chemim Ltd (Olomouc, Czech Republic)
Radio-labelled jasmonoyl-isoleucine	[² H ₁₀] JA-Ile	synthesised utilising [² H ₁₀] L-isoleucine in place of isoleucine [3] ¹
Jasmonoyl-isoleucine	JA-Ile	Ol-chemim Ltd (Olomouc, Czech Republic)
(+/-)-4-(3-oxo-2-(pent-2-enyl)cyclopentyl) butanoic acid	OPC-4	Ol-chemim Ltd (Olomouc, Czech Republic)
cis-(+)-12-oxo-phytodienoic acid	cis-OPDA	Ol-chemim Ltd (Olomouc, Czech Republic)

¹>99% pure by LC/MS.²>99% pure by ¹H- and ¹³C-NMR.

Table S2. Multiple reaction monitoring (MRM) transitions used for plant hormone analysis. Optimised Q1 (precursor ion) and Q3 (product ion) transitions, retention time (RT), declustering potential (DP), entrance potential (EP), collision energy (CE) and collision cell exit potential (CXP) for each of the phytohormones analyzed. The internal standard (IS) used for quantitation for each phytohormone is also given. SAG = salicylic acid O- β -glucoside; 12-OH-JA = 12-hydroxy jasmonic acid; SA = salicylic acid; ABA = abscisic acid; JA = jasmonic acid; DH-JA = 9,10-dihydrojasmonic acid; JA-Ile = jasmonoyl-isoleucine; OPC-4 = (+/-)-4-(3-oxo-2-(pent-2-enyl)cyclopentyl) butanoic acid; and cis-OPDA = cis-(+)-12-oxo-phytodienoic acid.

Compound	Q1	Q3	RT (min)	DP	EP	CE	CXP	IS
[² H ₄] SAG	303.00	96.90	4.53	-50	-10.0	-56.2	-8.6	-
SAG	299.30	136.80	4.54	-60	-10.0	-20.0	-12.0	[² H ₄] SAG
12-OH-JA	225.10	58.90	4.78	-60	-10.0	-20.0	-7.9	[² H ₅] JA
[² H ₄] SA	141.10	97.00	4.98	-40	-7.8	-40.0	-9.6	-
SA	136.90	93.00	4.99	-40	-11.0	-25.0	-7.0	[² H ₄] SA
[² H ₆] ABA	269.00	159.00	5.28	-25	-10.0	-17.0	-15.0	-
ABA	263.10	153.00	5.28	-60	-5.0	-15.0	-8.0	[² H ₆] ABA
[² H ₅] JA	214.00	62.10	5.51	-60	-6.0	-24.0	-8.6	[² H ₅] JA
JA	209.10	59.10	5.52	-60	-6.0	-24.0	-8.6	[² H ₅] JA
DH-JA	211.00	59.00	5.71	-100	-2.9	-20.0	-5.0	[² H ₅] JA
[² H ₁₀] JA-Ile	332.20	139.90	5.83	-60	-10.0	-26.9	-12.0	-
JA-Ile	321.90	129.80	5.84	-25	-10.0	-32.0	-15.0	[² H ₁₀] JA-Ile
OPC-4	237.20	58.80	5.84	-25	-10.0	-23.0	-15.0	[² H ₅] JA
cis-OPDA	291.00	165.00	6.40	-48	-10.0	-26.0	-3.0	[² H ₅] JA

Table S3. Identification of reference genes (RGs) and genes of interest (GoI) and their primer sequences used for 'Hort16A' and 'Hayward' kiwifruit leaf defence gene expression analysis by quantitative polymerase chain reaction in 2016 and 2018.

Gene name (Gene ID used in Figures).	RG or GoI	Identification (NCBI entry ¹ /Acc # ² /Achn number ³)	Forward primer (5'-3')	Reverse primer (5'-3')
<i>40s ribosomal protein</i>	RG	FG498176/Acc23370.1	GCAAAGGGATGTGAGGTGA T	CCCCCTGTCTCAGAAGAACA
<i>Glyceraldehyde 3-phosphate dehydrogenase (GAPDH)</i>	RG	FG499278/Acc18050.1	GCAGTCAACGATCCCCTTCAT	GACGCCGAAAACGTGTGACTT
<i>Protein phosphatase 2A (PP2A)</i>	RG	FG522516/Acc33246.1	GCAGCACATAATTCCACAG G	TTTCTGAGCCCATAACAGGAG
<i>Actin</i>	RG	EF063572/Acc05529.1	CACCCTGTGCTGCTTACAGA	GAGAGAGAACGGCCTGAATG
<u>SA associated genes</u>				
<i>Pathogenesis-related protein family 1 (PR1)</i>	GoI	FG499230/Acc06864.1	GCCCCCGGTAAGGTTTGT	CGAACCAAGACCCACTATTGC
<i>Glucan endo-1,3-β-glucosidase (β-1,3-glucosidase)</i>	GoI	FG455092/Acc03929.1	TTGGTTCAACATGTCAAAGG AG	TAGGCTGCTTGTGGGAAAG
<i>Pathogenesis-related protein family 5 (PR5)</i>	GoI	AJ871175/Acc28854.1	CGATGGAATTTAGCCCTACG	ATTTCGGAGTTGCAACAGT
<i>NPR1</i>	GoI	Acc00932.1	AGTTCCCATTTGTCCGTCAT C	AAGAAACGCTTCCCCAATTC
<u>JA associated genes</u>				
<i>Lipoxygenase (LOX2)</i>	GoI	Acc32487.1/Achn1236.21	CAGAGCTTGATCCCAAGAC A	CAGGATGGCTCCAATTTTAC
<i>APETALA2 Ethylene responsive factor 2 (AP2 ERF2)</i>	GoI	Acc20680.1/Achn033.321	ATGAGTCCAATGGCCAAAA C	GATTTCTGCCGCATATTTCC
<i>Jasmonoyl-L-amino acid synthetase (JAR1)</i>	GoI	Acc09200.1	TTCTACATTTGCCCACAGCA	TTTGGCTTGAGCAGTTTGA
<u>ABA associated genes</u>				
<i>Response to desiccation 22 (RD22)</i>	GoI	Acc29141.1	GACCTCAAAAAGGGCACAA A	AGTCGGGCTTCACAGAAAAG

Table S4. Identification of reference genes (RGs) and genes of interest (GoI) and their primer sequences used defence gene expression analysis by nCounter™ PlexSet™ NanoString on ‘Hort16A’ and ‘Hayward’ kiwifruit (*Actinidia chinensis*) leaf tissue in 2018.

Gene name (Gene ID used in Figures)	RG or GoI	Identification (NCBI entry ¹ /Acc # ² /Achn number ³)	Primer A	Primer B
<i>40s ribosomal protein</i>	RG	FG498176/ Acc23370.1	AAACGCCATAGCAGGCCCTGCG AACAGCGAGGCCACCAAGGAGCCTCACTAACAATCACCTCACATC TCATCCTCTTCTTTTCTTGGTGT	CGAAAGCCATGACCTCCGATCA AATCTCA
<i>Glyceraldehyde 3- phosphate dehydrogenase (GAPDH)</i>	RG	FG499278/ Acc18050.1	AATCCGAGCCTTGGCATCAAAG ATGCTCGATCTGCTGTACCAAC AAAGTCTGTTGAGATTATTGAGCCACCAGTTTCACGAACAAGTCGT TTCATCATGACCAGAAG	CGAAAGCCATGACCTCCGATCA CTCCACTCGTTGTCATACCAAGA TCAAAGC
<i>Protein phosphatase 2A (PP2A)</i>	RG	FG522516/ Acc33246.1	TAATCATGTCCAATACCTGTGGA ATTATGTGCTGCATTGCCATTTC TGGACAAAGACGCCTATCTTCCA GTTTGATCGGGAAACT	CGAAAGCCATGACCTCCGATCA CTCAAGAGAGATCGAGTGTAGT ATGGTCATACGGTACAGATAAT GTGGGTTGC
<i>Actin</i>	RG	EF063572/ Acc05529.1	GTTGAACCACCACTGAGCACA TGTTGCCATATAGATCTTTCTG ATATCCGAACCTAACTCCTCGCT ACATTCTATTGTTTTT	CGAAAGCCATGACCTCCGATCA CTCCAAGAGCAGTAATTTCTTA CTCATGCGGTCTGCGATACCAGG GAACATA
<i>Elongation Factor (EF)</i>	RG	FG526520/ Acc31629.1	TTCCCCAACTTTAATCGTCCCTTGT TCAACACGACCAGTGGCAACAGTT CCCTCAAGACCTAAGCGACAGCGT GACCTTGTTTCA	CGAAAGCCATGACCTCCGATCACT CACCAGTAGATTTTCATGGGAGCACC CTGCATCAGCCCTAAATCTCAAC ATC
<u>SA associated genes</u>				
<i>Pathogenesis- related protein family 1 (PR1)</i>	GoI	FG499230/ Acc06864.1	CTTCTCTGCAACCCATAGGTTCA CTGCAGCGGTGCCAGTAAAAGA TCCACCCAATTTGGTTTTACTCCCACAAGAGTTAGAGTTGATATCAT CTCGATTATGCGGAGT	CGAAAGCCATGACCTCCGATCA CTCTGCAAGCATTCTCCCCCGAC AGTTCGG
<i>Glucan endo-1,3-β- glucosidase (β-1,3- glucosidase)</i>	GoI	FG455092/ Acc03929.1	ACATACTCTATTTTTTGCTAGTGC CCTCTTTATGAGGGAAATCACAA GCACAACAGCCACTTTTTTCCA AATTTTGCAAGAGCC	CGAAAGCCATGACCTCCGATCA CTCTATTCTTTTATTAGATATTTT GTCTGTCTTCATAGGAGCAATCT CTCGGT
<i>Pathogenesis- related protein family 5 (PR5)</i>	GoI	AJ871175/ Acc28854.1	CCTGGAACGGCAGCGGCCCAAA CGGTGAAAGGGCAGTTGTTTATG ATATTCTTGAGTTTATGTATTG CCAACGAGTTTGTCTTT	CGAAAGCCATGACCTCCGATCA CTCCAGGATTGATGATCCAATTC TGGCCACGGTCAAGGCGTTTGCC ACCA
WRKY33	GoI	FG468929	CGAAAGCCATGACCTCCGATCA CTCAGATTTCTGTGGAGAAGCTT TGAATGGGTGCAAATTCTGATTT CCCTCCA	GGGTAAACGACGAGAATTGAG TTTCTCTGATGGTTTGTTCATGT TCCACACCGTGTGGACGGCAAC TCAGAGATAACGCATAT
WRKY75	GoI	FG474406	TCCAATTGAGAAGTCCACAGAC AACTTTGAGCACATCTTGACTCA GATGCCCTGGAGTTTATGTATTG CCAACGAGTTTGTCTTT	CGAAAGCCATGACCTCCGATCA CTCCCTAAAGATGAAGGAGTGG TGGTGACAACTTATGAAGGCAT GCACTCCCA

<i>Glutathione S-transferase (Glut-S-transferase)</i>	GoI	FG460267	ATCGATCTTGTTTCAGCTCTTCAA CGAAAGCCATGACCTCCGATCA TCCATGCTGCTAGTTTAGGCCGC CTCCTGTATTGTTTGACCAATGC AGATAAGGTTGTTATTGTGGAGG TTCAGGGTCGCATTTTGTCTGCG ATGTTACTACA TGTAAGC
<i>Hypersensitive response/ salicylic acid signalling (HS SA sig)</i>	GoI	FG431814	ACATATACTGCATAGTTTCTCTA CGAAAGCCATGACCTCCGATCA GCGGTTTCATCTCCAGCTTCTGCC CTCTACGGGTAGGTGAGAAGGT TGAGGCTGTAAAGCTGTAGCA TTACCAATGACAAGTTCACTGCC ACTCTTCCACGA TCCCTTTC
<i>Pathogenesis-related protein family 2 (PR2 Primer3)</i>	GoI	FG480859	TCAAACATGGCAAACACATAAG CGAAAGCCATGACCTCCGATCA TTTCTATCGGTCCACCGGACCTT CTCGGAGAAAGAGACCCCAATG CTAGGCTAGGACGCAAATCACT CTTCTCAAATTCTGGGTCTTTTTG TGAAGAAGTGAAAGCGAG ATTCTCA
<i>Pathogenesis-related protein family 5 (PR5 PrimerF)</i>	GoI	FG417283/ Acc28852.1	ATTGCCCGTGATATTGACAGAGT CGAAAGCCATGACCTCCGATCA ACGATATTGGCTGACACTTGCCT CTCAAAACCGTGACAGGTTATT GACACCACGCGATGACGTTCTG GCATTGCCTGGGTTTGTAACGGG CAAGAGTCGCATAATCT TTTGGGC
<i>Endochitinase (Endochit PrimerG)</i>	GoI	FG512537/ Acc24397.1	AAAAGCATCATAGGTATAAAAT CGAAAGCCATGACCTCCGATCA CCCTTCCCCTCGCAAGCGGCATC CTCGCAGCATCACCGGTGTTTCC GTTCCCATTTGGAATGATGTGTA AAAAGCCCCAAAAGACTTAGCA CTGGAATAAGACGACG GCAGAAAC
<u>IA associated genes</u>			
<i>Cytochrome oxidase subunit 1 (COI1)</i>	GoI	FG460354	CGTTGTCTTGCTCACTTTTCTGTC CGAAAGCCATGACCTCCGATCA TTCTTCATAAGATCTGAACCGC CTCTAGTCAATCACGCGCTGGAA AAACTTCCTTCTGTGTTCCAGC CACCAAATCCGGCATTCCGTTTC TACAAACTTAGAAAC CATCACT
<i>Lipoxygenase (LOX2)</i>	GoI	Acc32487.1/ Achn123621	TCTTCTCAGAATCATCAGACTTG CGAAAGCCATGACCTCCGATCA GAAGCAACCCAAGAGTTGCAAT CTCCTCTCTTAGCCTCTTTAACCC TCACACATAAAATTGGTTTTGCC ATTTGGTGTCTTGTTGGTGAAGA TTTCAGCAATTCAACTT AAACTC
<i>Plant defensin gene 1.2 (PDF1.2)</i>	GoI	Acc11160.1	TCTCGTCCGGCGCTGGCATAAC CGAAAGCCATGACCTCCGATCA TTTGCTGTTGCGAATTCAGAGTT CTCACTGTCTGTGCGAGTTATCA AGCTCTTTCGTTGGGACGCTTGA GTCGAGCCACAGGGCCCGGACC AGCGCAAGTAGAAAAC ATG
<u>Phenyl propanoid pathway genes</u>			
<i>Phenylalanine ammonia lyase (PAL)</i>	GoI	FG484623/ Acc32206.1 /Achn060261	CATCTTAGTCGATGAGCGGATGA CGAAAGCCATGACCTCCGATCA CTTCGATTAGCGGGCCCAACCACCTCGAGACATCGATCAATGGATT TGGGCTGGTCAAGACTTGCATGA ATCGTTACCGAGTTGATCTCCC GGACCCGCAAATTCT GTTCGAT
<i>Chalcone synthase (CHS)</i>	GoI	FG528287	AAGAGAGAGTTCCAGTCTGAGA CGAAAGCCATGACCTCCGATCA TGCCCAAGGGCTTGAAAGCCTCT CTCTCAGTTCGACTTGGTCTAAA ACTAGCCAGCAGACCTGCAATA ATAACAGGCCACCAGGGTGAG TCAAAGTTATAAGCGCGT CGATCCAA
<i>4-Coumarate-coenzyme A ligase (4CL)</i>	GoI	FG515208	AACTTCACCAGCAACTTCATCTT CGAAAGCCATGACCTCCGATCA TTTGCGGGACAACAGCAGCGTC CTCGCTTCTTCTGTAAGTTCAAA GGCAACCTGCCAATGCACTCGA CCCATTGACCGAACCACAAAT TCTTGTCATTTTTTTGCG GCTACAGG

Cinnamoyl- coenzyme A reductase (CCR)	GoI	FG397115	TCCAGCAGATCTACCTTGAAGAG CGAAAGCCATGACCTCCGATCAC TTTCAGATTCTCGGATGCTTTATC TCCATGGAACACTCCGTCGCATC AAGCAAACCTGGAGAGAGAAGTG CTGATATGGGTGCAGAAAGAGA AAGACGATTTAACCCA ATGGTAA
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Defence hub

gene

TCP14 hub protein (TCP14 hub)	GoI	FG496474	GACCGTCGCTTCCGTCCATGATC CGAAAGCCATGACCTCCGATCAC ACATGAGAAATACTCTCTCTCTCT TCCTTCCTCTTTCTTCTCGAGCAG CTCCGATTGCTGCATTCCGCTCAA TTGGAATGGGAAATTCGGGCGGT CGCTTGAGGAAGTA GCA
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¹NCBI = National Centre for Biotechnology Information

²Acc. numbers come from the manual annotation of the Red5 kiwifruit whole Genome Shotgun project

[4] that has been deposited at DDBJ/ENA/GenBank

³Achn numbers identify *A. chinensis* var. *chinensis* 'Hongyang' genome sequences from the project described by Huang et al. [5]. The sequences are found at <http://bioinfo.bti.cornell.edu/cgi-bin/kiwi/home.cgi>.

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