

Supplementary Data

Light Intensity-Mediated Induction Of Trichome-Associated Allelochemicals Increases Resistance Against Thrips In Tomato

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This file contains information on supplementary Figures S1-S3, Table S1-S2 with legends, Method S1, and Notes S1.

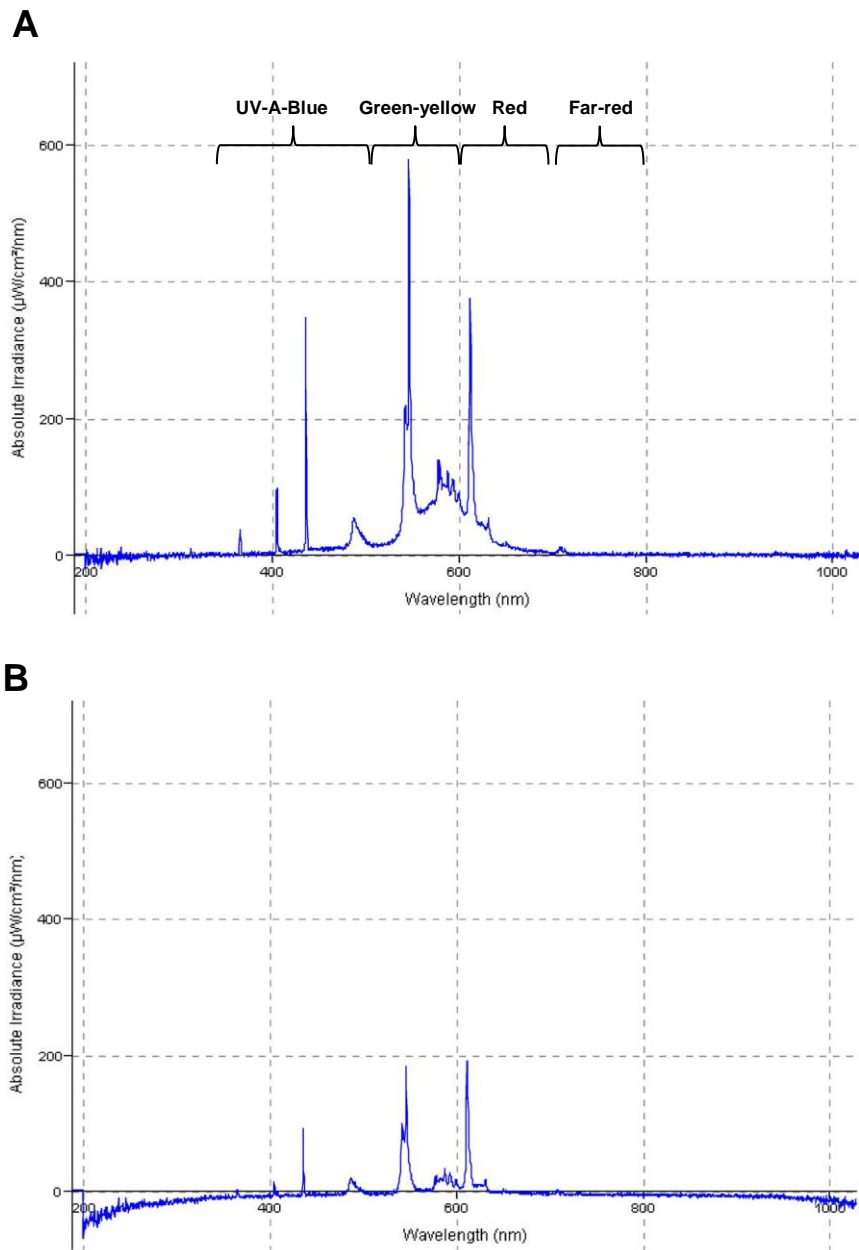


Fig. S1. Spectral quality in high (a) and low (b) PAR conditions were measured using a spectrometer UV-Vis equipped with a cosine corrector (Flame-S, Ocean Optics).

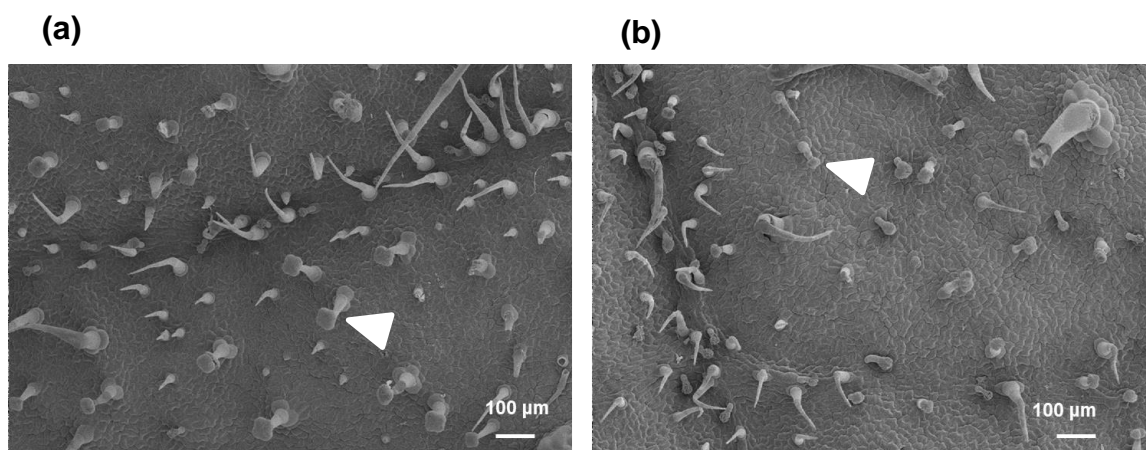


Fig. S2. Scanning electron micrographs of adaxial leaf surfaces of wild-type (a) and *od-2* (b) plants grown under high PAR conditions. Note that type-VI glandular trichomes (pointed by white triangles) in *od-2* leaves display smaller sizes and different shapes when compared to the wild-type.

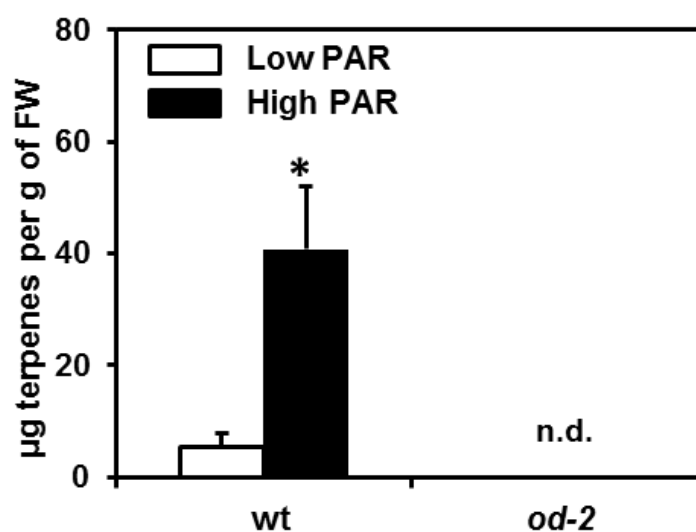


Figure S3. Effect of low and high photosynthetically active radiation (PAR) treatments on type-VI leaf trichome-associated defenses in wild-type (wt) and *odorless-2* (*od-2*) plants. Total terpene content (mean + SEM, $n = 4$) in leaf exudates of leaflets taken from the third/fourth youngest leaf were measured at 35 days after the initial light treatment. Asterisks denote significant differences between low and high PAR-treated wild-type plants analyzed by *t-test*. n.d. = not detected.

Table S1. Nucleotide sequence of primers used for qRT-PCR analysis.

Target gene	Gene identification	Forward Primer 5' → 3'	Reverse Primer 5' → 3'
<i>WIPI-II</i>	Solyc01g095200	GACAAGGTACTAGTAATCAATTATCC	GGGCATATCCCGAACCAAGA
<i>TD-2</i>	Solyc09g008670	TGCCGTAAAAATGTCACCA	ACTGGCGATGCCAAAATATC
<i>JIP-21</i>	Solyc03g098790	ACTCGTCCTGTGCTTTGTCC	CCCAAGAGGATTTTCGTTGA
<i>Actin</i>	Solyc03g078400	TTAGCACCTTCCAGCAGATGT	AACAGACAGGACACTCGCACT

Table S2. Transitions or specific pair of m/z values associated to the precursors and fragment ions of the analytes measured by LC/MS.

Analyte	Q1 [m/z] → Q3 [m/z]^a	CE [V]	Standard
ABA	(-)263.13 → 153.00	9	D6-ABA
JA	(-)209.12 → 59.00	12	D6-JA
JA-Ile	(-)322.20 → 130.00	19	D6-JA-Ile
SA	(-) 137.02 → 93.00	15	D6-SA
IAA	(+) 176.07 → 130.00	-14	D5-IAA
D6-ABA	(-) 269.17 → 159.00	10	
D6-JA	(-)215.15 → 59.00	10	
D6-JA-Ile	(-) 328.24 → 130.00	19	
D6-SA	(-)141.05 → 97.00	15	
D5-IAA ^b	(+) 181.10 → 135.00	-14	
	(+) 181.10 → 134.00	-14	
	(+) 181.10 → 133.00	-14	
OPDA	(-) 291,00 → 165.00	18	D6-JA-Ile

CE: collision energy

a Resolution: Q1: 0.7, Q3: 22

b Analyzed as the sum of all three transitions

Methods S1. Hormone extraction and analysis

Hormones extraction was performed in approximately 100 mg of frozen and homogenized leaf material aliquoted in 2 ml Eppendorf tubes. After adding 1 ml of ethyl acetate containing 40 ng of phytohormone standards D₆-ABA (Olchemin), D₆-JA (HPC), D₆-JA-Ile (HPC), D₆-SA (Olchemin) and D₅-IAA (Olchemin), samples were vortexed for 10 min and centrifuged at 14.000 rpm for 10 min at 4°C. Supernatants were transferred to a new Eppendorf tube and evaporated to dryness on a vacuum concentrator at room temperature. The residue was dissolved in 0.2 ml of 70% methanol (v/v) for 5 min using an ultrasonic bath, and centrifugated at 14.000 rpm for 5 min at room temperature. Supernatants were transferred to glass vials and then analyzed by means of LC-MS/MS

Measurements were conducted on a liquid chromatography-triple quadrupole mass spectrometry system (LC-MS/MS, EVOQ, Bruker). We injected 20 µL of each sample onto C18 Zorbax column (4.6 x 50 mm, 1,8 µm, 600 bar). The mobile phase comprised of solvent A (0.05 % (v/v) formic acid in LCMS-grade water) and solvent B (0.05% (v/v) formic acid in LCMS-grade methanol). The program with a constant flow rate of 0.4 ml/min was set as follows: 0 - 0.5 min 95% solvent A; 0.5 - 2.5 min 50% solvent A and 50% solvent B; 2.5 - 3.5 100% solvent B; 3.5 - 4.5 min 95% solvent A. The column temperature was set at 42°C. The cone, probe and nebulizer gas were set at the following flow conditions (arbitrary units/temperature): 35/350°C, 60/475°C and 60, respectively. Phytohormones were measured by monitoring the transition m/z described in Supporting Information Table S2. Phytohormones were quantified using the signal of their corresponding internal standard, and expressed as ng per gram fresh mass leaf material.

Note S1. For the second analysis of terpene content in leaf exudates of low and high-PAR treated plants we used benzyl acetate as an internal standard following the same procedure described in Materials and Methods.