

Supplementary Information for

Organic pollutant-induced long-distance ROS signaling drives plant systemic acquired acclimation via rhizomicrobiota

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Number of pages: 16

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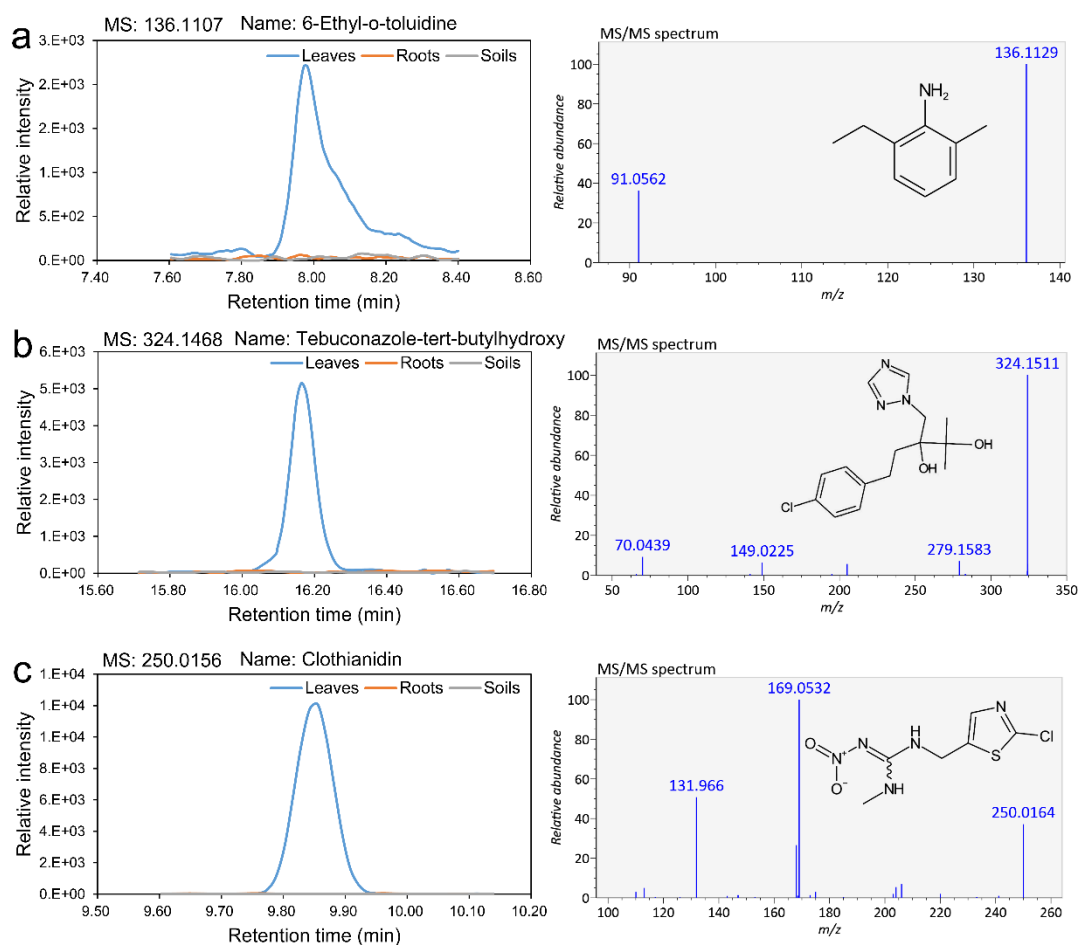


Figure 1 MS, MS/MS, retention time, and relative intensity of key metabolic intermediates derived from pollutants, including 6-ethyl-o-toluidine from acetochlor (a), tebuconazole-tert-butylhydroxy from tebuconazole (b), and clothianidin from thiamethoxam (c), in plant leaves, roots, and rhizosphere soil.

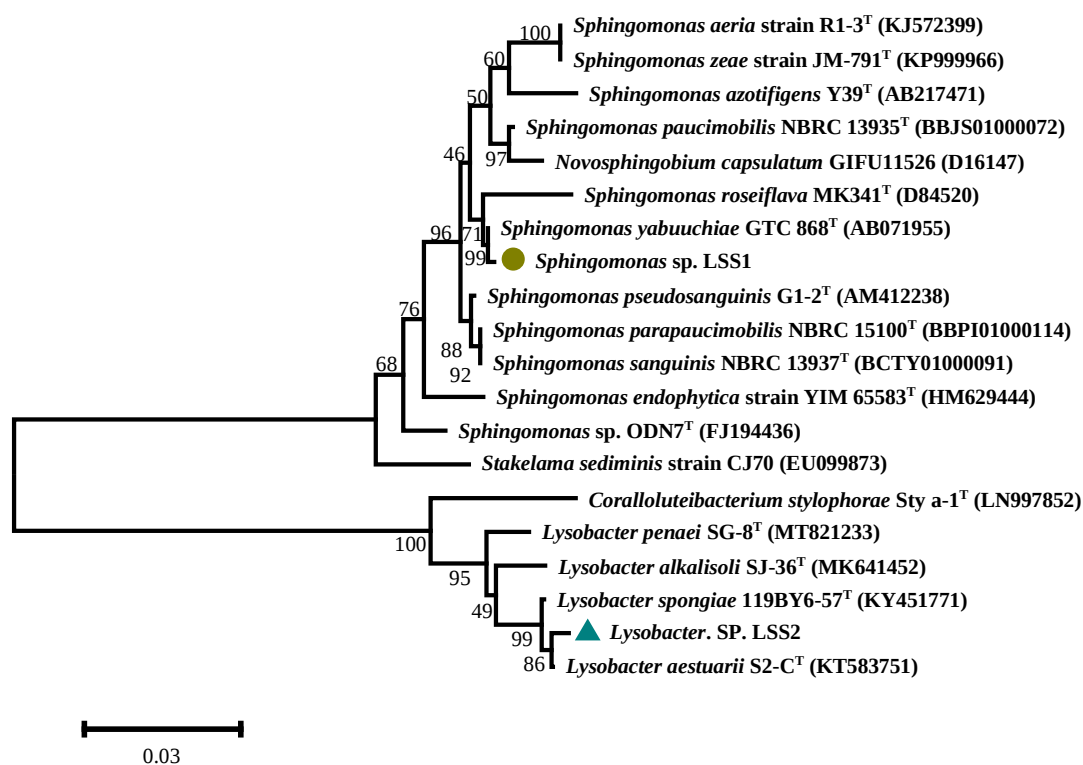


Figure 2 Phylogenetic tree based on the 16S rDNA sequences of *Sphingomonas* sp. LSS1 and *Lysobacter* sp. LSS2 and related species. The scale bar represents an evolutionary distance of 0.03. GeneBank accession numbers are given in brackets.

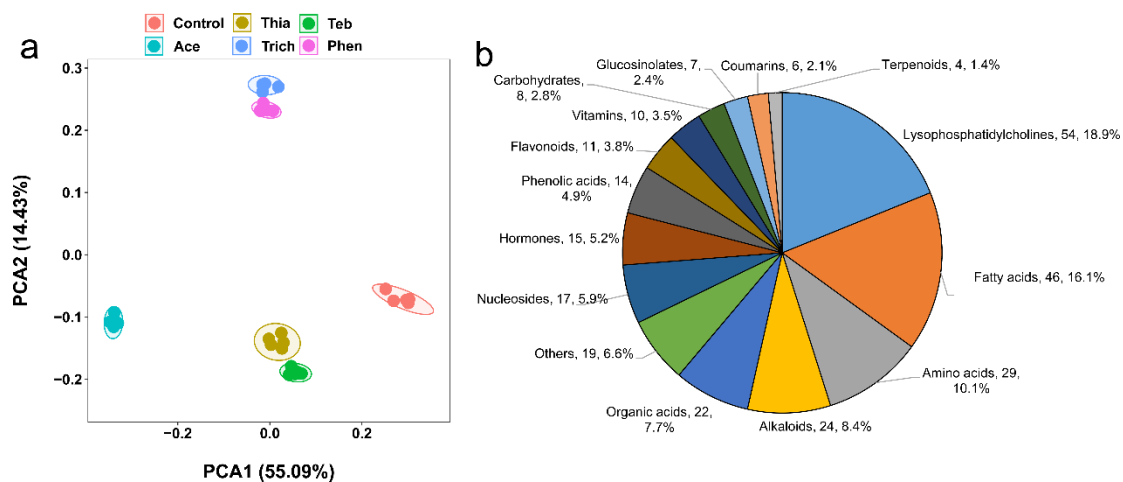


Figure 3 (a) PCA analysis showing the difference in the metabolic profile of vegetable root exudates. Each treatment includes 5 replicates. (b) Category percentage of identified metabolites with significant change in plant root exudates. Source data are provided as a Source Data file.

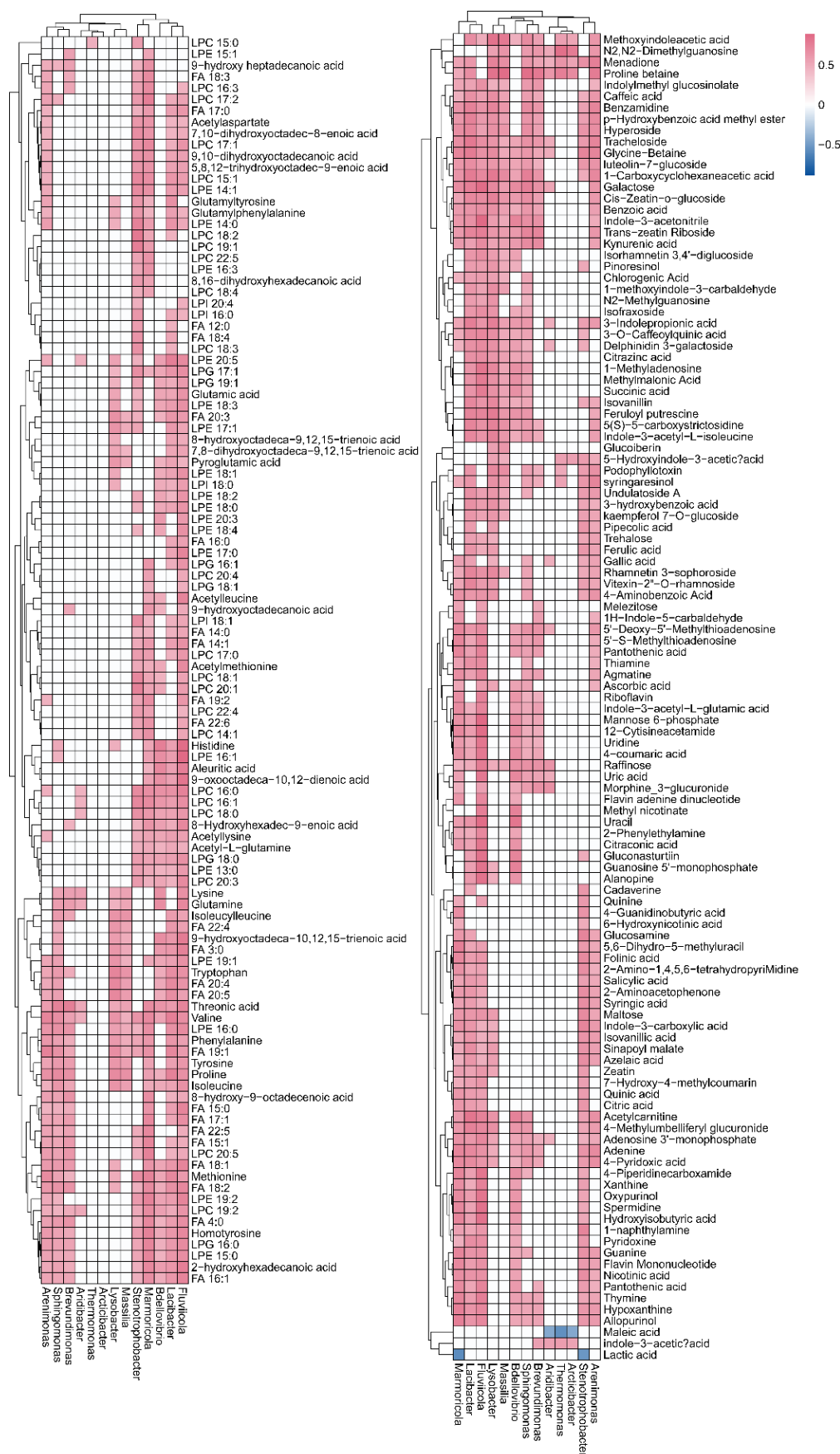


Figure 5 Spearman's rank correlation analysis of the altered root exudates and

rhizosphere genera commonly recruited by plants in response to various pollutant exposures, where red and blue represent significantly positive and negative correlation ($P < 0.01$), respectively and white represent no significant correlation ($P > 0.01$). Source data are provided as a Source Data file.

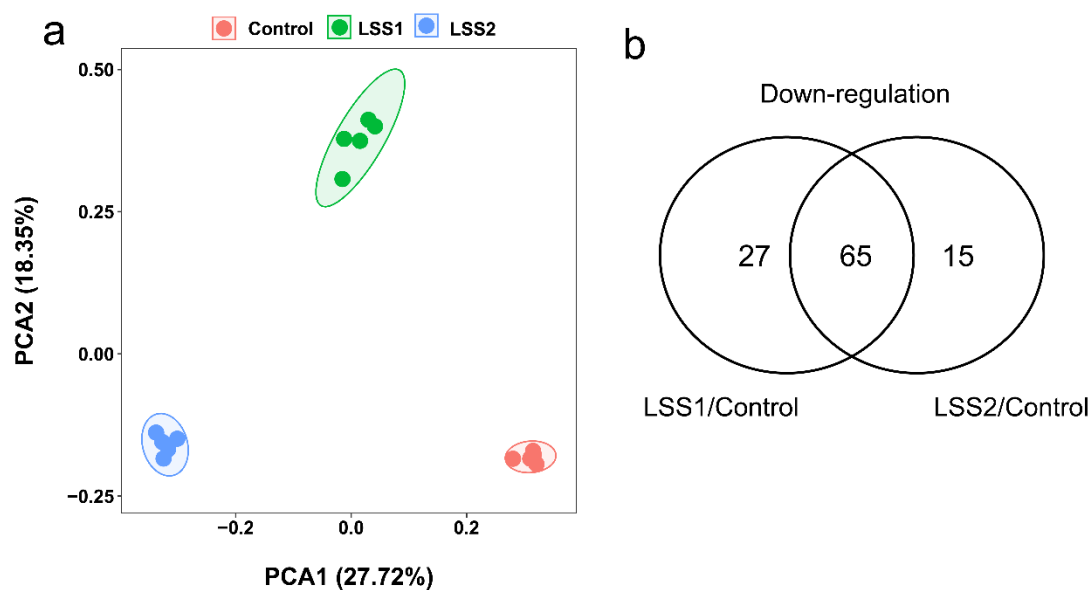


Figure 6 Preference utilizability of strains LSS1 and LSS2 on root exudate metabolites.

(a) PCA analysis of metabolic profiling of root exudates solution cultivating Strain LSS1 or LSS2 and without any stains (Control). Each data point represents the mean of 5 replicates. Source data are provided as a Source Data file. (b) Venn plot analysis the metabolites with significant down-regulation in the root exudates solution cultivating strains compared to the control. Source data are provided in Supplementary Data 3.

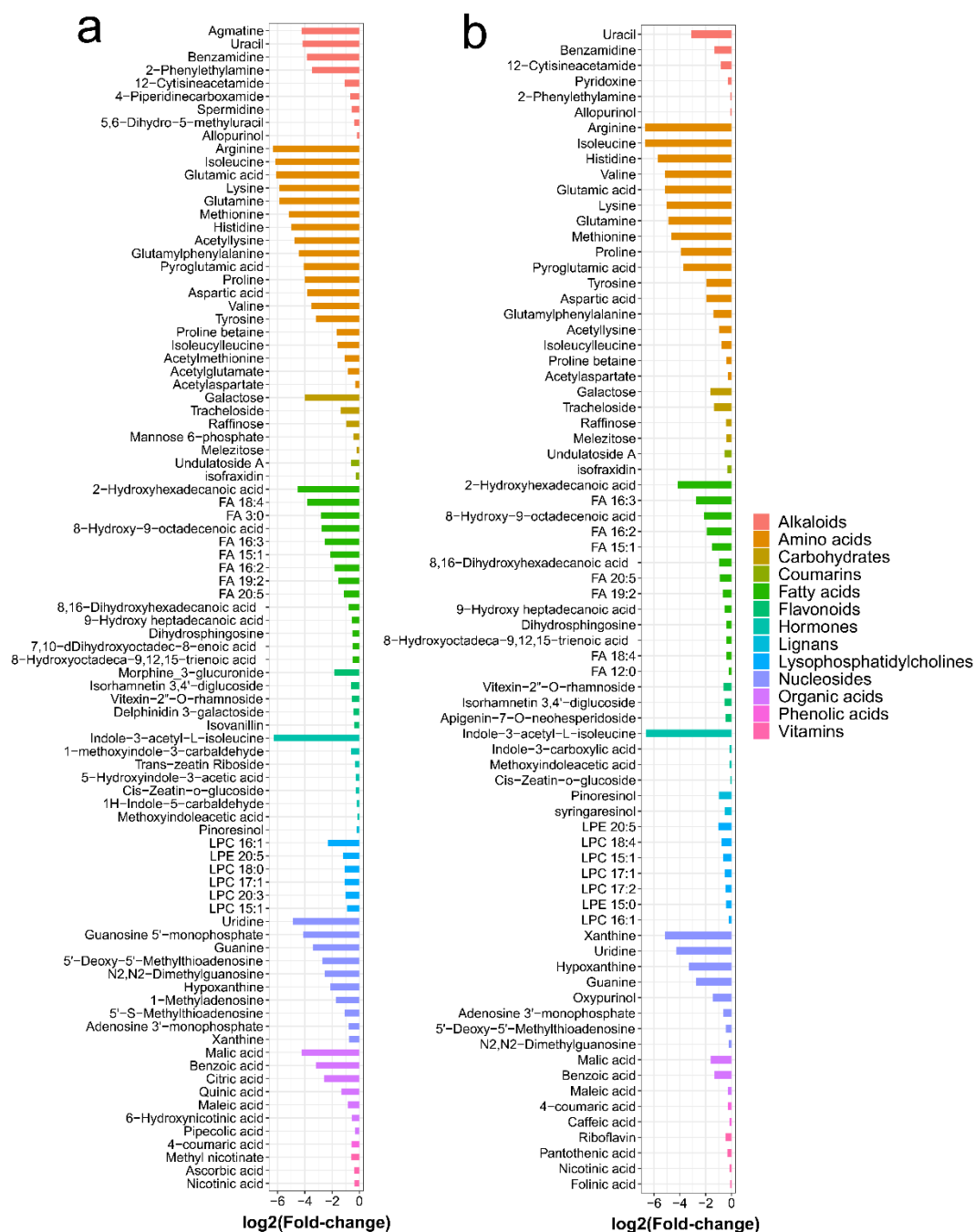


Figure 7 Barplot analysis of the preference utilizability of strains LSS1 (a) and LSS2 (b) on root exudate metabolites (two-sided Student's *t*-test with $P < 0.05$, PLS-DA with $VIP > 1$). Source data are provided in Supplementary Data 3.

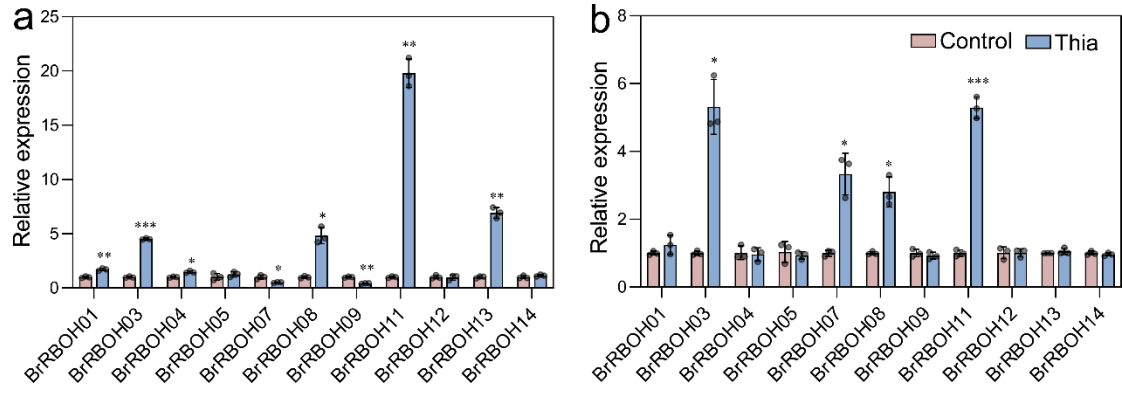


Figure 8 Relative expression of *BrRBOH* genes in plant leaves (a) and stems (b) after foliar application of Thia at 6 hours. Asterisks indicate statistically significant differences compared with the control group (two-sided Student's *t*-test, * for $P < 0.05$, ** for $P < 0.01$, *** for $P < 0.001$, **** for $P < 0.0001$; P values are shown in the Source Data file). Each data point represents the mean of 3 replicates and error bars represent standard deviations. Source data are provided as a Source Data file.

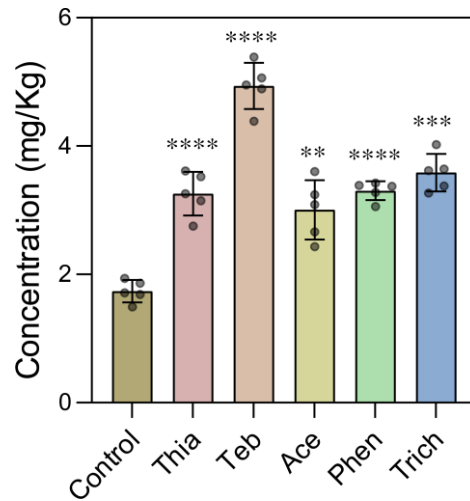


Figure 9 Linoleic acid concentration in root tissues. Each data point represents the mean of 5 replicates. Asterisks indicate statistically significant differences compared with the control group (two-sided Student's *t*-test, * for $P < 0.05$, ** for $P < 0.01$, *** for $P < 0.001$, **** for $P < 0.0001$; P values are shown in the Source Data file). Source data are provided as a Source Data file.

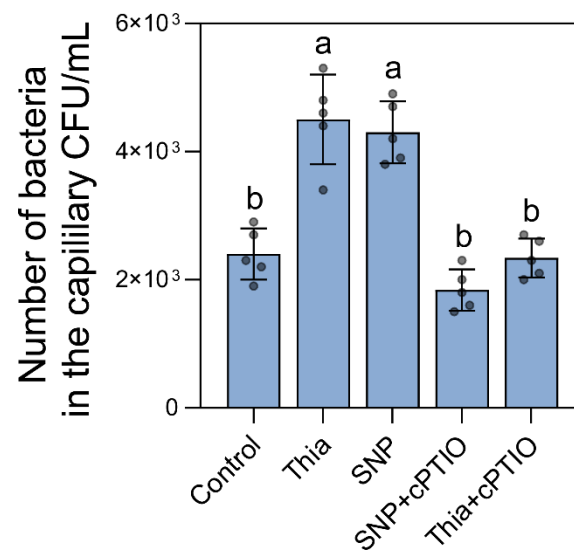


Figure 10 Colonization of Strain LSS2 in the root of plants treated by Thia, SNP and cPTIO. Different letters on the bars indicate statistical significance (one-way ANOVA, two-sided Tukey's test, $P < 0.05$; P values are shown in the Source Data file). Each data point represents the mean of 5 replicates and error bars represent standard deviations. Source data are provided as a Source Data file.

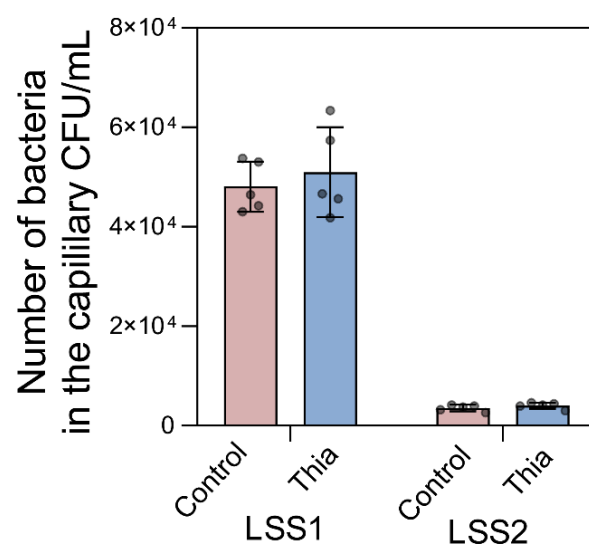


Figure 11 Capillarity response of Strain LSS1 and LSS2 toward H_2O_2 and water (Control). Each data point represents the mean of 5 replicates and error bars represent standard deviations. Source data are provided as a Source Data file.

Table 1 Primer sequence for respiratory burst oxidase homolog genes (*BrRBOH*) in vegetable roots.

Primer name	Primer sequence (5' to 3')	
	-F	-R
<i>BrRBOH01</i>	GTTCATCAGCATGACCGA	CACATCCAGCAGTAATTC
<i>BrRBOH03</i>	ATGAGCGAAATTGCTGACTCTG	GCCTCCCAAAGTGTGTTTCGT
<i>BrRBOH04</i>	ACGGAGTTGATGTCGTCTCG	GGTCCTTTGTCAGTCCTGGT
<i>BrRBOH05</i>	AGGATCCTGAGAAGTCAC	GATATTTGAAGGCATAAGA
<i>BrRBOH06</i>	GAGCGGTCCGATGAGCGGT	GACGCTTCACCACAGACG
<i>BrRBOH07</i>	TCAGGAATGGATCTCGTAG	TCTCTCTGCCAGAGCCGAT
<i>BrRBOH08</i>	CGGAGATGGTTACGACGAC	AGGCGTCGTGGATAGACTC
<i>BrRBOH09</i>	TACGAAGCAGGAGATGCAAGA	TTCGTATTCGGCTCTCAGAAACA
<i>BrRBOH10</i>	CTACGAGGAGCCCACTCAG	GACCTTTGAGAGCCGTGGAC
<i>BrRBOH11</i>	CGGAAGTGCATCCACCTC	ACGAGACCACGAGAACCGGT
<i>BrRBOH12</i>	ACGGAAGGAAGAGGTGAAGC	TTCATCGCAATCGCAGTCC
<i>BrRBOH13</i>	CATTTGCAAGACCCGACTG	GCCAGTCCCTTCAACTCTCC
<i>BrRBOH14</i>	ACAGAGAAGCCCTACAGCG	GCATGACGTACGCGTTTCA
<i>BrActin</i>	CTATCCTCCGTCTCGATCTCGC	CTTAGCCGTCTCCAGCTCTTGC

Table 2 Parameters of the LC-QQQ/MS system for detecting the content of Thia, Teb, Ace and linoleic acid.

Compound	Precursor ion (m/z)	Product ion (m/z)	Collision Energy (CE)	Declustering Potential (DP)
Thia	292.0	211.0*	16	30
	292.0	181.0	30	30
Teb	308.0	125*	47	95
	308.0	70.0	49	95
ACE	270.1	224.1*	15	20
	270.1	148.1	22	20
Linoleic acid	279.0	279.0*	-10	80
	279.0	261.0	-45	80

*, for quantitative analysis

Table 3 Parameters of the GC-MS system for detecting the content of Phen and Trich.

Item	Parameter
Carrier	$\geq 99.999\%$ pure helium gas with a 0.5 mL/min flow rate
Oven temperature program	Initial temperature of 140 °C was held for 0.5 min, increased at a rate of 2 °C min to 160 °C, increased at a rate of 50 °C min to 290 °C and held for 2 min
Inlet temperature	250 °C
Injection volume	1 μ L using a splitless mode
Mass spectrometry conditions	250 °C for EI source temperature, 150 °C for quadrupole temperature, 70 eV for ionization energy and 280 °C for transfer line temperature.
Detection mode	Selective ion monitoring mode (SIM)
m/z	Quantification: 178 and 256 for Phen and Trich, respectively; Qualitation: 221 and 89 for Phen and Trich, respectively