Rhizobacteria-induced systemic resilience in *Sorghum bicolor* (L.) Moench against *Fusarium pseudograminearum* crown rot under drought stress conditions

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Supplementary material

Considering the enormous scale of the raw data in addition to the extent of the treatment interactions [Rhizobacteria treatment (4) × Stress regime (4) × ESI-charge (2)], not all of the chemometric models were included here, but can be made available upon request. The ESI-negative data are not given here but can be made available upon request.

Table S1 Screening of rhizobacterial isolates for induction of drought stress tolerance in *S. bicolor* seedlings.

	Growth parameters[1]			Disease parameters[1]			Percentage stress alleviation (%)[2]						SAF-ISR[3]
Isolate code	Plant height (cm)	Dry roots mass (g)	Dry shoot mass (g)	Severity	Incidence	Isolations	Height	Dry roots	Dry leaves	Disease severity	Disease incidence	Isola-tions	(%)
A02	0.28 a-i	0.17 b-i	0.17 a-e	1.10 a	53.43 a-j	41.76 b-o	10.60	14.36	13.29	43.73	21.22	38.49	23.61
A03	0.28 a-i	0.15 bi	0.16 a-e	1.76 a	58.50 a-j	53.65 c-y	10.08	5.57	7.99	10.03	13.74	20.98	11.40
A05	0.28 a-i	0.13 b-i	0.16 a-e	1.61 a	57.64 a-j	38.09 a-j	10.08	-8.32	5.17	17.85	15.01	43.89	13.95
A07	0.28 a-g	0.17 b-i	0.19 a-e	1.87 a	35.95 a-j	47.01 c-y	12.20	14.36	27.58	4.60	46.99	30.76	22.75
A08	0.29 a-g	0.13 c-i	0.14 b-e	1.86 a	58.20 a-j	43.26 a-p	14.11	-8.04	-8.32	5.01	14.18	36.28	8.87
A10	0.25 c-j	0.12 f-i	0.14 b-e	1.04 a	43.16 a-j	65.48 c-y	0.52	-18.35	-9.26	46.60	36.36	3.55	9.90
A11W	0.28 a-g	0.15 b-i	0.16 b-e	0.59 a	27.80 a-g	47.05 c-y	10.80	6.19	4.63	70.03	59.01	30.70	30.22
A11Y	0.29 a-e	0.15 b-i	0.16 a-e	1.24 a	44.97 a-j	44.4 c-t	16.79	4.67	5.57	36.68	33.69	34.60	22.00
A12	0.25 c-j	0.13 c-i	0.14 b-e	1.37 a	30.87 a-g	91.06 n-y	1.80	-9.48	-8.99	30.03	54.48	-34.13	5.62
A13	0.26 c-j	0.12 f-i	0.15 b-e	1.66 a	55.33 a-j	46.94 c-x	2.64	-16.70	-1.81	15.04	18.42	30.86	8.07
A14	0.28 a-g	0.15 b-i	0.15 b-e	1.86 a	79.60 h-j	81.03 h-y	13.91	0.27	3.49	5.01	-17.37	-19.35	-2.34
A15	0.28 a-g	0.13 c-i	0.16 a-e	1.27 a	53.76 a-j	51 c-y	13.79	-8.25	9.93	35.04	20.73	24.88	16.02
A17	0.28 a-g	0.15 b-i	0.13 b-e	0.59 a	20.71 a-c	34.4 a-h	10.92	0.21	-13.76	70.08	69.46	49.33	31.04
A18B	0.25 c-j	0.11 g-i	0.10 de	1.45 a	47.10 a-j	41.74 a-n	0.00	-25.57	-30.40	25.63	30.55	38.52	6.45
A18W	0.28 a-g	0.15 b-i	0.12 c-e	1.01 a	51.74 a-j	63.98 c-y	12.00	5.91	-19.13	48.13	23.71	5.76	12.73
A18Y	0.27 a-i	0.14 b-i	0.17 a-e	0.98 a	24.50 a-e	51.04 c-y	9.76	-2.54	12.42	50.03	63.87	24.82	26.39
A19	0.26 b-j	0.16 b-i	0.14 b-e	1.12 a	47.77 a-j	43.85 a-s	2.68	13.06	-3.96	42.76	29.56	35.41	19.92
A20	0.28 a-h	0.13 c-i	0.12 c-e	2.05 a	59.77 a-j	49.11 a-y	10.60	-9.62	-18.59	-4.96	11.87	27.66	2.83
A21	0.29 a-g	0.19 bc	0.18 a-e	1.37 a	61.52 a-j	51.24 c-y	13.99	33.33	21.88	30.03	9.29	24.52	22.17
A22	0.30 a-d	0.18 b-f	0.22 ab	0.85 a	26.71 a-g	71.61 c-y	19.39	21.86	47.99	56.68	60.62	-5.48	33.51
A23	0.29 a-f	0.14 b-i	0.15 b-e	0.85 a	21.86 a-d	43.62 c-r	16.27	-2.61	0.27	56.68	67.77	35.75	29.02
A25W	0.28 a-g	0.14 b-i	0.18 a-e	1.50 a	55.83 a-j	75.32 g-y	11.48	-2.20	18.93	23.38	17.68	-10.94	9.72
A26	0.28 a-g	0.15 b-i	0.16 a-e	1.01 a	38.84 a-j	60.46 c-y	11.72	5.70	5.91	48.34	42.73	10.94	20.89
A27	0.28 a-g	0.17 b-i	0.16 a-e	1.66 a	55.52 a-j	70.19 c-y	13.39	14.50	6.44	15.04	18.14	-3.39	10.69
A28	0.29 a-e	0.15 b-i	0.19 a-e	2.09 a	59.62 a-j	55.49 c-y	16.51	3.71	27.32	-6.80	12.09	18.26	11.85
A31	0.28 a-g	0.16 b-i	0.17 a-e	1.47 a	59.57 a-j	45.38 a-y	11.32	8.04	13.49	25.01	12.16	33.16	17.20
A35	0.28 a-g	0.15 b-i	0.15 b-e	2.15 a	68.96 g-j	86.66 n-y	11.28	1.31	2.42	-9.97	-1.68	-27.65	-4.05
A39	0.22 hj	0.10 hi	0.11 c-e	2.46 a	70.77 g-j	68.58 c-y	-10.40	-31.55	-27.25	-25.88	-4.35	-1.02	-16.74
A44	0.25 c-j	0.12 d-i	0.14 b-e	2.31 a	81.45 j	84.64 k-y	1.80	-15.12	-7.79	-18.31	-20.10	-24.67	-14.03
A47A	0.29 a-f	0.16 b-i	0.16 b-e	1.27 a	43.85 a-j	31.6 a-g	16.31	8.25	4.97	35.04	35.34	53.45	25.56
N09	0.26 b-j	0.14 b-i	0.16 a-e	0.91 a	34.41 a-j	21.3 a-d	5.44	-5.91	9.13	53.35	49.26	68.63	29.98
N1	0.28 a-g	0.14 b-i	0.17 a-e	1.17 a	55.33 a-j	37.99 a-j	11.32	-3.23	12.95	40.00	18.42	44.04	20.58
N10W	0.27 a-j	0.13 c-i	0.14 b-e	1.79 a	69.00 g-j	64.14 c-y	8.52	-10.52	-6.38	8.34	-1.74	5.52	0.62
N11	0.25 c-j	0.10 i	0.11 c-e	2.25 a	78.73 h-j	55.24 c-y	1.72	-29.35	-23.76	-14.99	-16.09	18.63	-10.64
N16W	0.24 g-j	0.13 c-i	0.13 b-e	1.79 a	61.08 a-j	43.85 a-s	-4.84	-13.47	-15.70	8.44	9.94	35.41	3.30

Table S1 *Cont.*

Isolate code	Growth parameters: Plant height (cm)		Dry roots mass (g)		Dry shoot mass (g)		Disease parameters[1]: Severity		Incidence		Isolations		Percentage stress alleviation (%)[2]: Height	Dry roots	Dry leaves	Disease severity	Disease incidence	Isola-tions	SAF-ISR[3] (%)
N17	0.26	c-j	0.12	f-i	0.12	c-e	1.37	a	43.59	a-j	26.27	a-f	2.04	-19.31	-16.51	30.03	35.73	61.31	15.55
N18	0.28	a-g	0.13	c-i	0.17	a-e	1.50	a	47.70	a-j	57.84	c-y	11.24	-8.32	15.77	23.38	29.67	14.80	14.42
N19	0.27	a-j	0.13	b-i	0.11	b-e	1.22	a	48.25	a-j	60.38	c-y	7.40	-7.70	-23.22	37.49	28.86	11.06	8.98
N2	0.26	c-j	0.17	b-h	0.15	b-e	2.18	a	76.43	h-j	47.83	c-y	2.20	15.26	-2.55	-11.66	-12.70	29.55	3.35
N20	0.28	a-i	0.13	c-i	0.16	b-e	1.69	a	26.07	a-f	44.26	c-t	10.56	-7.84	4.97	13.35	61.56	34.81	19.57
N22A	0.29	a-g	0.13	c-i	0.15	b-e	1.66	a	43.72	a-j	44.66	a-u	13.95	-9.42	-1.88	15.04	35.54	34.22	14.58
N23	0.27	a-j	0.14	b-i	0.13	b-e	0.62	a	28.63	a-g	60.28	c-y	8.52	-2.61	-12.82	68.34	57.79	11.21	21.74
N26	0.30	a-c	0.17	b-i	0.16	a-e	1.65	a	47.22	a-j	45.53	a-w	20.11	13.75	6.31	15.70	30.37	32.94	19.86
N28	0.30	a-e	0.18	b-g	0.17	a-e	1.34	a	60.50	a-j	45.14	c-v	18.71	21.10	16.31	31.66	10.79	33.51	22.01
N29	0.27	b-j	0.12	f-i	0.14	b-e	1.30	a	66.57	c-j	48.55	c-y	6.20	-15.88	-8.52	33.35	1.84	28.49	7.58
N30	0.29	a-g	0.16	b-i	0.15	a-e	1.27	a	40.33	a-j	39.75	a-m	15.19	12.30	2.48	35.04	40.53	41.45	24.50
N31	0.29	a-f	0.19	b-e	0.19	a-e	1.56	a	49.37	a-j	49.31	c-y	16.19	29.35	27.05	20.00	27.20	27.37	24.53
N36	0.27	a-j	0.16	b-i	0.21	a-d	1.97	a	75.11	a-j	16.57	a-c	6.36	8.59	39.93	-0.82	-10.75	75.59	19.82
N4	0.29	a-e	0.15	b-i	0.14	b-e	0.91	a	49.81	a-j	39.88	a-m	17.51	6.39	-3.49	53.35	26.56	41.26	23.60
N40	0.28	a-g	0.14	b-i	0.16	b-e	1.17	a	41.82	a-j	71.72	c-y	13.07	-2.20	4.36	40.00	38.34	-5.64	14.66
N43	0.31	ab	0.14	b-i	0.13	b-e	1.35	a	53.44	a-j	54.22	c-y	22.91	-5.15	-12.68	30.84	21.20	20.14	12.88
N45	0.27	a-j	0.14	b-i	0.12	a-e	1.96	a	63.89	a-j	72.83	c-y	6.96	-3.85	-17.38	0.00	5.79	-7.28	-2.63
N47	0.28	a-g	0.13	c-i	0.14	b-e	1.63	a	50.15	a-j	76.98	g-y	11.72	-9.35	-4.23	16.68	26.05	-13.39	4.58
N49	0.27	b-j	0.14	b-i	0.16	a-e	1.76	a	59.93	a-j	50.15	c-y	6.20	-4.95	7.11	10.03	11.63	26.13	9.36
N52	0.27	a-j	0.17	b-h	0.15	b-e	1.76	a	18.97	ab	37.23	a-j	7.92	15.26	0.27	10.03	72.03	45.16	25.11
N53	0.29	a-g	0.16	b-i	0.17	a-e	2.12	a	71.89	g-j	44.2	a-t	15.27	10.10	14.63	-8.29	-6.00	34.89	10.10
N54[4]	**0.30**	**a-e**	**0.20**	**ab**	**0.19**	**a-e**	**1.04**	**a**	**47.34**	**a-j**	**50.43**	**a-y**	**18.99**	**38.76**	**29.93**	**46.70**	**30.20**	**25.72**	**31.72**
N55B	0.27	a-j	0.13	c-i	0.19	a-e	1.57	a	44.53	a-j	43.32	a-q	9.04	-9.69	28.32	19.90	34.34	36.19	19.68
N59	0.28	a-g	0.16	b-i	0.17	a-e	1.73	a	47.42	a-j	45.23	a-w	11.40	6.67	14.23	11.66	30.08	33.38	17.90
N60	0.29	a-g	0.19	b-d	0.26	a	1.23	a	37.69	a-j	38.27	a-k	15.31	31.89	72.35	37.03	44.43	43.63	40.77
N61	0.24	f-j	0.12	f-i	0.13	c-e	1.37	a	63.18	a-j	21.65	a-e	-2.60	-16.63	-15.44	30.03	6.84	68.11	11.72
N62	0.26	b-j	0.16	b-i	0.15	b-e	1.90	a	63.08	a-j	35.52	a-j	3.16	9.00	-2.55	2.71	6.99	47.68	11.17
N65	0.29	a-g	0.13	b-i	0.17	a-e	1.37	a	41.00	a-j	74.52	c-y	14.07	-7.56	11.74	30.18	39.55	-9.77	13.04
N66	0.28	a-i	0.15	b-i	0.14	b-e	1.96	a	55.83	a-j	34.62	a-i	10.48	0.55	-5.64	0.00	17.68	49.01	12.01
N67	0.26	a-j	0.15	b-i	0.17	a-e	2.23	a	72.16	c-j	38.79	a-l	5.88	5.02	14.97	-13.81	-6.40	42.86	8.09
N69	0.26	b-j	0.13	c-i	0.13	b-e	1.08	a	44.11	a-j	45.88	c-w	5.80	-7.49	-10.34	45.01	34.96	32.42	16.73
N71	0.28	a-i	0.15	b-i	0.16	a-e	2.15	a	74.46	h-j	65.45	c-y	10.40	1.79	9.73	-9.97	-9.79	3.59	0.96
N74	0.26	b-j	0.12	f-i	0.11	c-e	1.89	a	68.15	f-j	68.81	c-y	5.60	-19.86	-23.96	3.38	-0.49	-1.36	-6.12
N76	0.25	d-j	0.13	c-i	0.12	c-e	1.69	a	58.28	a-j	67.99	c-y	1.08	-8.04	-19.40	13.35	14.07	-0.15	0.15

Table S1 *Cont.*

Isolate code	Growth parameters						Disease parameters[1]						Percentage stress alleviation (%)[2]						SAF-ISR[3]
	Plant height (cm)		Dry roots mass (g)		Dry shoot mass (g)		Severity		Incidence		Isolations		Height	Dry roots	Dry leaves	Disease severity	Disease incidence	Isola-tions	(%)
N77	0.28	a-g	0.16	b-i	0.16	a-e	2.15	a	67.62	f-j	37.2	a-j	11.68	6.87	10.67	-9.97	0.29	45.21	10.79
N77Y	0.28	a-g	0.15	b-i	0.18	a-e	1.37	a	50.99	a-j	51.24	c-y	13.27	2.13	18.99	30.03	24.82	24.52	18.96
N79	0.30	a-e	0.15	b-i	0.17	a-e	1.66	a	61.52	a-j	39.75	a-m	18.71	5.09	17.05	15.04	9.29	41.45	17.77
N80	0.26	b-j	0.15	b-i	0.17	a-e	1.53	a	59.86	a-j	58.88	c-y	3.96	0.48	15.17	21.69	11.74	13.27	11.05
NAS6G6	0.29	a-f	0.16	b-i	0.15	b-e	1.86	a	47.42	a-j	45.23	c-w	16.07	9.21	-2.08	5.01	30.08	33.38	15.28
RhizoVital®	0.26	b-j	0.15	b-i	0.15	a-e	1.57	a	37.42	a-j	99.93	b-y	4.68	2.06	3.09	19.90	44.82	-47.19	4.56
S5	0.27	b-j	0.14	c-i	0.15	b-e	2.22	a	64.70	a-j	82.32	h-y	6.08	-6.87	0.20	-13.76	4.60	-21.25	-5.17
T29	0.27	a-j	0.15	b-i	0.15	b-e	1.34	a	49.31	a-j	89.71	l-y	8.08	4.26	-0.20	31.36	27.29	-32.14	6.44
UCNS[5]	0.31	a	0.27	a	0.21	a-c	0.03	a	0.03	a	6.49	ab	25.51	83.78	42.42	98.47	99.96	90.44	73.43
UCNSNF[6]	0.25	d-j	0.14	b-t	0.10	e	0.03	a	0.03	a	0.01	a	1.00	-1.24	-34.50	98.47	99.96	99.99	43.95
UCS[7]	0.25	e-j	0.15	b-i	0.15	b-e	1.96	a	67.82	f-j	67.89	c-y	0.00	0.00	0.00	0.00	0.00	0.00	0.00

[1] Figures followed by the same letter within a column does not differ significantly according to Tukey's LSD test at a significance level of $p < 0.05$.
[2] The percentage stress alleviation was calculated as the percentage increase in the growth parameters (plant height, root- and shoot mass) and the percentage reduction in disease parameters (severity, incidence and isolations) by treating *S. bicolor* seedlings with each of the 77 rhizobacterial isolates compared to that of the untreated stress control inoculated with *F. pseudograminerum* and subjected to drought stress (UCS).
[3] The stress alleviation factor (SAF) was calculated with the formula as outlined in section 2.1.6 and incorporated the percentage increase in the growth parameters and the percentage reduction in disease parameters.
[4] The rhizobacterial isolate N54, selected as best-performing isolate for eliciting ISResilience in *S. bicolor* seedlings against combined biotic and abiotic stress, is indicated in **bold**.
[5] Untreated control receiving no stress (UCNS).
[6] Untreated control receiving no stress and no fertiliser (UCNSNF).
[7] Untreated control inoculated with *F. pseudograminerum* and subjected to drought stress (UCS)**.**

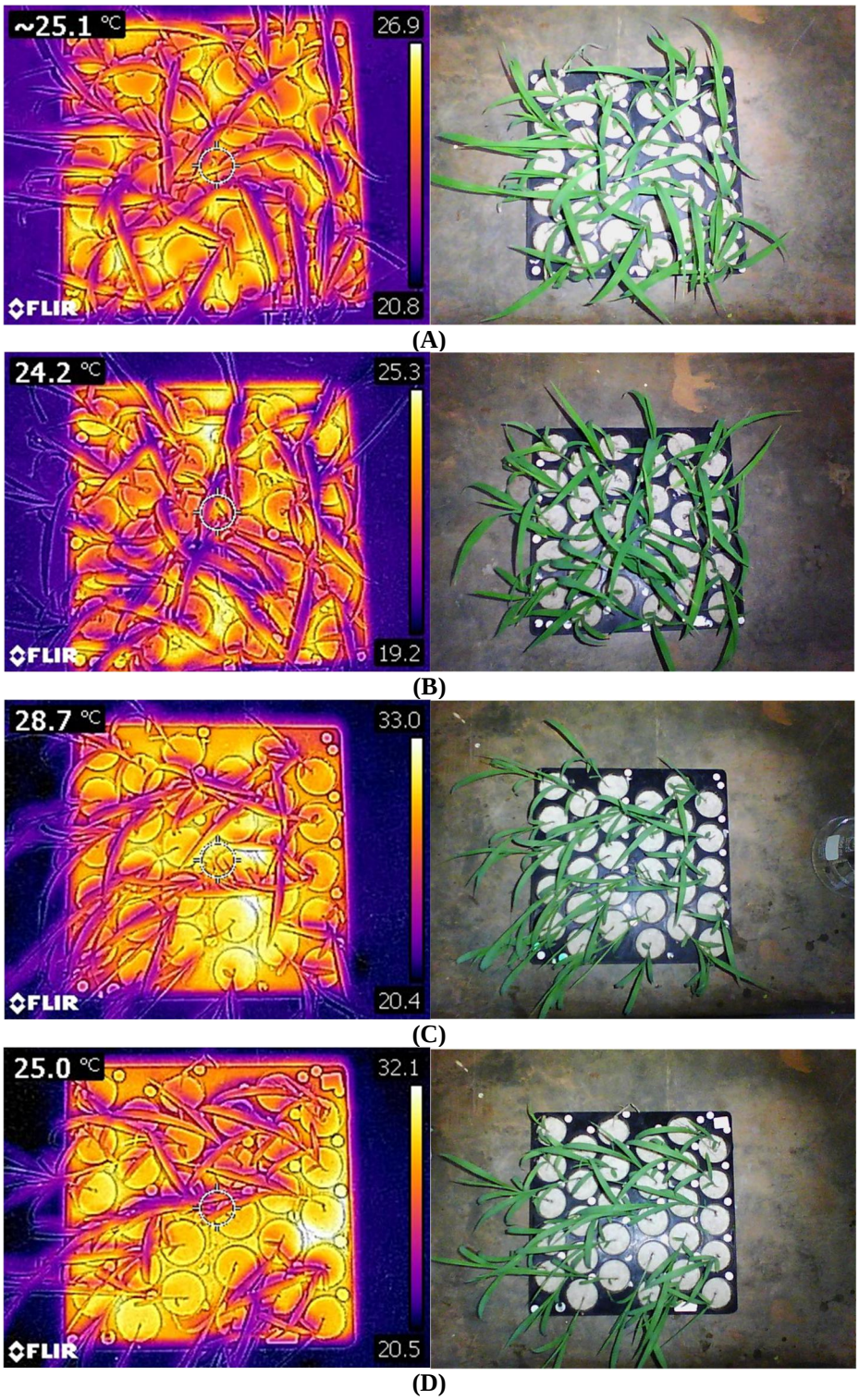


Figure S1 IR thermography images of *S. bicolor* seedlings demonstrating the effects of inoculation with *F. pseudograminearum* under drought stress conditions, visualized by means of a FLIR thermal imager: (A) untreated control, (B) untreated control inoculated with *F. pseudograminearum*, (C) untreated control with drought stress, (D) untreated control with combined stresses. The IR thermography images (the images on the left) provide a visual range of the temperature gradient, ranging from purple (indicating low leaf temperatures corresponding with low stress levels) to bright yellow/white (indicating high leaf temperatures corresponding with high stress levels) according to the temperature scale shown on the right of each thermal images.

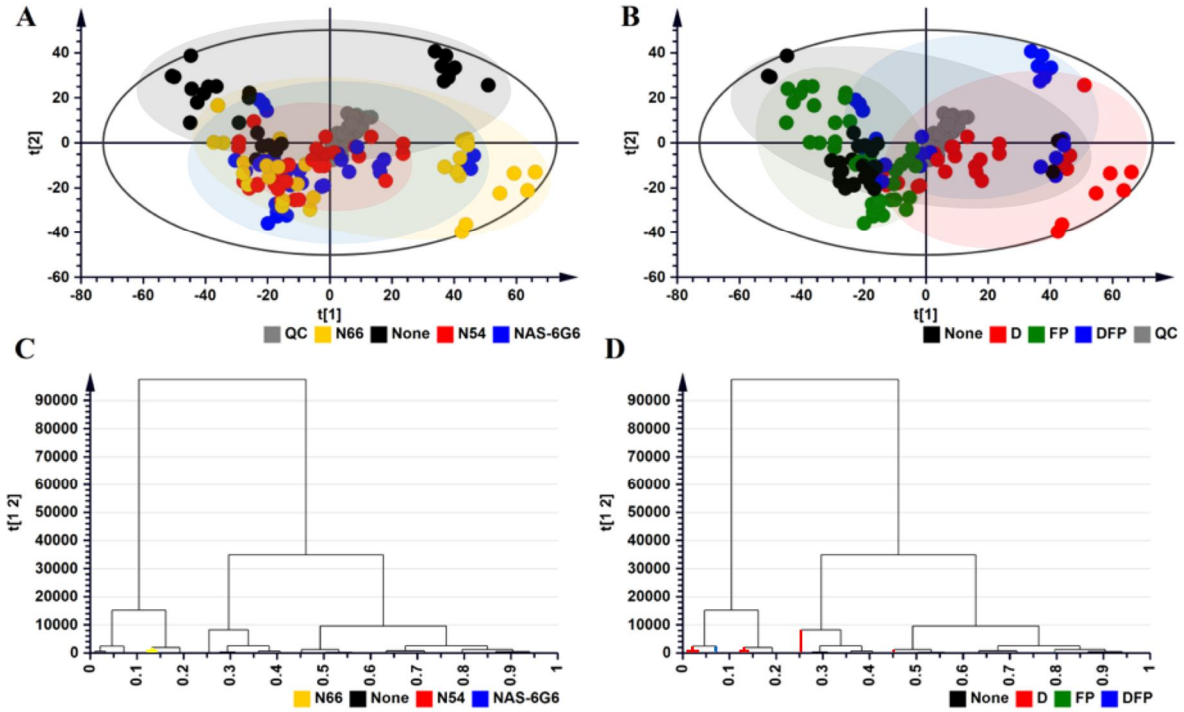


Figure S2 Unsupervised chemometric overview of ESI-negative data obtained from *S. bicolor* shoots. PCA score scatter plot computed from the first two PCs of an 12-component PCA model. The model explains 66.9 % variation in the Pareto-scaled data ($R^2X = 0.669$) and 43.6% predicted variation according to cross-validation ($Q^2 = 0.436$). (A and B) represents the same PCA scores plot with (A) showing the rhizobacteria treatment-related clustering and (B) showing the stress regime-related clustering. (C and D) HCA dendrogram corresponding to (A and B). Legend: (A and C) QC: Quality control samples (grey); N66: primed with isolate N66 (yellow); None: untreated control (black); N54: primed with isolate N54 (red); NAS-6G6: primed with isolate NAS-6G6 and (B and D) None: no-stress control (black); D: drought stress (red); FP: inoculated with *F. pseudograminearum* (green); DFP: drought stress and inoculated with *F. pseudograminearum* (blue); QC: Quality control samples (grey).

Table S2 Summary of metabolites significantly upregulated ($p < 0.05$) during the priming phase under conditions of no-stress in rhizobacteria-treated *S. bicolor* seedlings *versus* those left naïve. Discriminating metabolites were identified based on OPLS-DA S-plots.

							Metabolite fold change (fc) and *p*-value of primed versus naïve					
							NAS-6G6		N66		N54	
	Metabolite	***m/z***	**RT (min)**	**ESI mode**	**MF**	**Metabolic pathway**	**fc**	***p***	**fc**	***p***	**fc**	***p***
1	L-Leucine	335	8.00	Pos	$C_6H_{13}NO_2$	Amino acid	4.28	1.32E-04	4.49	1.40E-07	4.71	3.00E-09
2	N,N-Dihydroxy-L-Tyrosine	234	8.11	Neg	$C_9H_{11}NO_5$	Amino acid	5.09	4.71E-08	5.09	4.71E-08	4.32	2.53E-12
3	L-Histidine	309	6.00	Pos	$C_6H_9N_3O_2$	Amino acid	2.67	1.09E-03	3.99	3.46E-06	-	-
4	Riboflavin	437	9.07	Pos	$C_{17}H_{19}N_4O_6$	Antioxidant	-	-	-	-	-	-
5	Hydroxyproline	343	4.05	Neg	$C_5H_9NO_3$	Antioxidant	4.81	2.10E-04	-	-	4.22	7.39E-07
6	Myo-inositol	249	3.57	Pos	$C_6H_{12}O_6$	Carbohydrate	5.10	3.31E-05	5.21	2.61E-09	-	-
7	N-Caffeoylputrescine	309	5.99	Pos	$C_{13}H_{18}N_2O_3$	Diamine	2.67	1.09E-03	3.99	3.46E-06	-	-
8	Apigenin 7-O-neohesperidoside	623	7.34	Pos	$C_{27}H_{30}O_{14}$	Flavone / Flavonol	2.08	9.15E-04	2.01	1.07E-05	2.43	5.27E-07
9	Hesperetin 7-O-glucoside	532	6.11	Neg	$C_{22}H_{24}O_{11}$	Flavonoid	2.02	1.91E-05	2.02	1.91E-05	2.45	3.43E-08
10	Pentahydroxyflavanone	305	4.61	Pos	$C_{15}H_{12}O_7$	Flavonoid	11.73	6.79E-08	6.65	7.72E-06	9.47	9.65E-07
11	3,4-Epoxybutyl-Alpha-D-Glucopyranoside	335	4.96	Pos	$C_{10}H_{18}O_7$	Glycogen	-	-	3.17	6.01E-04	2.18	2.79E-03
12	Heptaethylene Glycol	349	1.69	Pos	$C_{14}H_{30}O_8$	Glycogen	-	-	-	-	-	-
13	Isopropylammelide	207	4.48	Neg	$C_6H_{10}N_4O_2$	Histidine	20.10	1.29E-06	20.10	1.29E-06	23.41	6.87E-09
14	(9Z,12Z,15Z)-Octadecatrienoic acid	317	3.66	Pos	$C_{18}H_{30}O_2$	Lipid	4.20	1.16E-06	5.91	4.08E-06	3.94	3.40E-10
15	L-Serine-phosphoethanolamine	301	3.46	Neg	$C_5H_{13}N_2O_6P$	Lipid	21.96	1.36E-08	21.96	1.36E-08	109.83	2.50E-05
16	(3Z,6Z)-Nonadienal	183	4.93	Pos	$C_9H_{14}O$	Lipid	-	-	-	-	-	-
17	3,5-Dimethoxyphenol	177	9.00	Pos	$C_8H_{10}O_3$	Phenylpropanoid	-	-	3.26	1.58E-03	2.18	1.26E-03
18	Trans-beta-D-Glucosyl-2-hydroxycinnamate	325	6.90	Neg	$C_{15}H_{18}O_8$	Phenylpropanoid	2.75	1.04E-03	2.75	1.04E-03	4.08	1.39E-06
19	Shikimate	195	5.59	Neg	$C_7H_{10}O_5$	Phenylpropanoid	2.84	1.16E-07	2.84	1.16E-07	3.53	1.95E-10
20	Trans-5-O-caffeoyl-D-quinate	561	6.09	Neg	$C_{16}H_{17}O_9$	Phenylpropanoid	-	-	-	-	3.29	2.09E-05
21	Gibberellin A36	415	9.07	Pos	$C_{20}H_{26}O_6$	Phytohormone	2.12	4.59E-02	2.79	1.50E-04	-	-
22	(-)-Phenylalanine Jasmonate conjugate	450	4.30	Pos	$C_{21}H_{29}NO_4$	Phytohormone	3.69	6.04E-04	-	-	3.33	4.90E-03
23	Dihydrozeatin-9-N-glucoside-O-glucoside	584	1.74	Pos	$C_{22}H_{35}N_5O_{11}$	Phytohormone	-	-	-	-	-	-
24	Thermozeaxanthin	948	14.12	Pos	$C_{51}H_{84}O_{12}$	Phytohormone	2.51	1.20E-03	-	-	-	-
25	Gibberellin A36	407	8.94	Neg	$C_{20}H_{26}O_6$	Phytohormone	11.46	1.23E-06	11.46	1.23E-06	12.30	8.94E-11
26	(-)-11-Hydroxy-9,10-dihydrojasmonic acid 11-beta-D-glucoside	475	8.00	Pos	$C_{18}H_{30}O_9$	Phytohormone	10.76	4.08E-05	9.41	2.83E-08	10.11	1.77E-08
27	3-Oxopropionyl-CoA	853	1.96	Neg	$C_{24}H_{38}N_7O_{18}P_3S$	Propanoate	-	-	-	-	-	-
28	Dimethylbenzimidazole	205	5.59	Pos	$C_9H_{10}N_2$	Riboflavin	2.02	2.90E-04	2.27	6.12E-05	-	-
29	Demethylphylloquinol	557	11.07	Pos	$C_{30}H_{46}O_2$	Ubiquinone	-	-	-	-	-	-
30	Ubiquinone	357	4.58	Pos	$C_{14}H_{18}O_4$	Ubiquinone	73.44	1.31E-11	42.54	1.46E-04	52.14	2.36E-11

Table S3 Summary of metabolites significantly upregulated ($p < 0.05$) during ISResistance under conditions of biotic stress (inoculated with *F. pseudograminearum*) in rhizobacteria-treated *S. bicolor* seedlings *versus* those left naïve. Discriminating metabolites were identified based on OPLS-DA S-plots.

							Metabolite fold change (fc) and *p*-value of primed versus naïve					
							NAS-6G6		N66		N54	
	Metabolite	***m/z***	**RT (min)**	**ESI mode**	**MF**	**Metabolic pathway**	**fc**	***p***	**fc**	***p***	**fc**	***p***
1	L-Leucine	335	8.00	Pos	$C_6H_{13}NO_2$	Amino acid	2.28	3.26E-05	2.53	1.72E-03	2.92	2.91E-06
2	N,N-Dihydroxy-L-Tyrosine	234	8.11	Neg	$C_9H_{11}NO_5$	Amino acid	4.68	4.69E-08	2.98	3.69E-04	4.19	2.40E-07
3	L-Histidine	309	6.00	Pos	$C_6H_9N_3O_2$	Amino acid	4.86	3.09E-05	-	-	5.99	5.84E-05
4	Riboflavin	437	9.07	Pos	$C_{17}H_{19}N_4O_6$	Antioxidant	2.48	7.53E-08	-	-	-	-
5	Hydroxyproline	343	4.05	Neg	$C_5H_9NO_3$	Antioxidant	3.96	1.84E-07	-	-	3.50	3.96E-05
6	Myo-inositol	249	3.57	Pos	$C_6H_{12}O_6$	Carbohydrate	6.72	4.89E-07	6.12	5.72E-08	7.20	3.09E-06
7	N-Caffeoylputrescine	309	5.99	Pos	$C_{13}H_{18}N_2O_3$	Diamine	4.86	3.09E-05	-	-	5.99	5.84E-05
8	Apigenin 7-O-neohesperidoside	623	7.34	Pos	$C_{27}H_{30}O_{14}$	Flavone / Flavonol	2.65	2.97E-04	-	-	3.00	1.30E-05
9	Hesperetin 7-O-glucoside	532	6.11	Neg	$C_{22}H_{24}O_{11}$	Flavonoid	2.73	1.29E-06	3.01	5.28E-06	3.29	1.00E-05
10	Pentahydroxyflavanone	305	4.61	Pos	$C_{15}H_{12}O_7$	Flavonoid	18.62	5.58E-05	-	-	17.67	2.00E-07
11	3,4-Epoxybutyl-Alpha-D-Glucopyranoside	335	4.96	Pos	$C_{10}H_{18}O_7$	Glycogen	3.17	1.69E-05	-	-	-	-
12	Heptaethylene Glycol	349	1.69	Pos	$C_{14}H_{30}O_8$	Glycogen	59.66	3.04E-11	21.76	2.26E-07	16.57	2.32E-07
13	Isopropylammelide	207	4.48	Neg	$C_6H_{10}N_4O_2$	Histidine	41.65	2.03E-07	30.56	1.99E-05	49.86	2.25E-09
14	(9Z,12Z,15Z)-Octadecatrienoic acid	317	3.66	Pos	$C_{18}H_{30}O_2$	Lipid	4.31	3.95E-08	5.13	6.41E-06	7.25	4.60E-05
15	L-Serine-phosphoethanolamine	301	3.46	Neg	$C_5H_{13}N_2O_6P$	Lipid	16.83	1.29E-02	23.67	3.11E-04	24.17	3.86E-03
16	(3Z,6Z)-Nonadienal	183	4.93	Pos	$C_9H_{14}O$	Lipid	84.30	3.89E-03	-	-	-	-
17	3,5-Dimethoxyphenol	177	9.00	Pos	$C_8H_{10}O_3$	Phenylpropanoid	2.15	9.00E-05	-	-	-	-
18	Trans-beta-D-Glucosyl-2-hydroxycinnamate	325	6.90	Neg	$C_{15}H_{18}O_8$	Phenylpropanoid	5.51	6.18E-06	3.22	2.02E-03	5.16	5.25E-06
19	Shikimate	195	5.59	Neg	$C_7H_{10}O_5$	Phenylpropanoid	2.36	3.84E-08	-	-	3.46	6.45E-08
20	Trans-5-O-caffeoyl-D-quinate	561	6.09	Neg	$C_{16}H_{17}O_9$	Phenylpropanoid	12.21	4.04E-03	13.05	4.32E-04	8.35	2.32E-02
21	Gibberellin A36	415	9.07	Pos	$C_{20}H_{26}O_6$	Phytohormone	3.22	2.07E-05	2.83	1.98E-03	2.87	3.24E-03
22	(-)-Phenylalanine Jasmonate conjugate	450	4.30	Pos	$C_{21}H_{29}NO_4$	Phytohormone	3.00	1.76E-03	-	-	7.11	1.38E-04
23	Dihydrozeatin-9-N-glucoside-O-glucoside	584	1.74	Pos	$C_{22}H_{35}N_5O_{11}$	Phytohormone	6.28	5.21E-04	-	-	4.82	2.68E-05
24	Thermozeaxanthin	948	14.12	Pos	$C_{51}H_{84}O_{12}$	Phytohormone	4.19	1.36E-02	-	-	-	-
25	Gibberellin A36	407	8.94	Neg	$C_{20}H_{26}O_6$	Phytohormone	6.61	3.32E-05	5.06	6.96E-06	6.24	3.24E-08
26	(-)-11-Hydroxy-9,10-dihydrojasmonic acid 11-beta-D-glucoside	475	8.00	Pos	$C_{18}H_{30}O_9$	Phytohormone	4.35	4.61E-05	-	-	3.90	2.00E-05
27	3-Oxopropionyl-CoA	853	1.96	Neg	$C_{24}H_{38}N_7O_{18}P_3S$	Propanoate	2.42	1.81E-04	2.72	1.31E-06	2.40	4.13E-07
28	Dimethylbenzimidazole	205	5.59	Pos	$C_9H_{10}N_2$	Riboflavin	2.61	8.98E-05	2.09	1.10E-03	3.53	6.27E-07
29	Demethylphylloquinol	557	11.07	Pos	$C_{30}H_{46}O_2$	Ubiquinone	2.14	4.56E-03	2.35	2.18E-03	2.43	7.43E-06
30	Ubiquinone	357	4.58	Pos	$C_{14}H_{18}O_4$	Ubiquinone	9.92	8.41E-03	-	-	24.73	3.14E-06

Table S4 Summary of metabolites significantly upregulated ($p < 0.05$) during ISTolerance under conditions of abiotic stress (drought stress) in rhizobacteria-treated *S. bicolor* seedlings *versus* those left naïve. Discriminating metabolites were identified based on OPLS-DA S-plots.

							Metabolite fold change (fc) and *p*-value of primed versus naïve					
							NAS-6G6		N66		N54	
	Metabolite	***m/z***	**RT (min)**	**ESI mode**	**MF**	**Metabolic pathway**	**fc**	***p***	**fc**	***p***	**fc**	***p***
1	L-Leucine	335	8.00	Pos	$C_6H_{13}NO_2$	Amino acid	3.68	6.66E-08	6.30	7.24E-10	3.21	1.49E-04
2	N,N-Dihydroxy-L-Tyrosine	234	8.11	Neg	$C_9H_{11}NO_5$	Amino acid	3.81	1.09E-12	6.06	1.51E-10	3.61	2.90E-07
3	L-Histidine	309	6.00	Pos	$C_6H_9N_3O_2$	Amino acid	-	-	2.67	4.07E-04	-	-
4	Riboflavin	437	9.07	Pos	$C_{17}H_{19}N_4O_6$	Antioxidant	3.36	1.01E-05	5.67	6.77E-07	-	-
5	Hydroxyproline	343	4.05	Neg	$C_5H_9NO_3$	Antioxidant	2.28	1.65E-04	-	-	-	-
6	Myo-inositol	249	3.57	Pos	$C_6H_{12}O_6$	Carbohydrate	4.01	9.74E-06	4.11	2.55E-07	5.30	1.33E-07
7	N-Caffeoylputrescine	309	5.99	Pos	$C_{13}H_{18}N_2O_3$	Diamine	-	-	2.67	4.07E-04	-	-
8	Apigenin 7-O-neohesperidoside	623	7.34	Pos	$C_{27}H_{30}O_{14}$	Flavone / Flavonol	3.17	3.96E-08	3.11	4.46E-05	-	-
9	Hesperetin 7-O-glucoside	532	6.11	Neg	$C_{22}H_{24}O_{11}$	Flavonoid	2.10	5.42E-04	2.10	5.42E-04	-	-
10	Pentahydroxyflavanone	305	4.61	Pos	$C_{15}H_{12}O_7$	Flavonoid	4.94	1.96E-05	7.90	1.92E-04	4.58	9.40E-05
11	3,4-Epoxybutyl-Alpha-D-Glucopyranoside	335	4.96	Pos	$C_{10}H_{18}O_7$	Glycogen	-	-	-	-	-	-
12	Heptaethylene Glycol	349	1.69	Pos	$C_{14}H_{30}O_8$	Glycogen	-	-	-	-	-	-
13	Isopropylammelide	207	4.48	Neg	$C_6H_{10}N_4O_2$	Histidine	6.94	2.65E-04	3.62	4.43E-03	5.67	7.90E-08
14	(9Z,12Z,15Z)-Octadecatrienoic acid	317	3.66	Pos	$C_{18}H_{30}O_2$	Lipid	2.96	2.91E-10	4.31	3.92E-06	4.09	2.45E-05
15	L-Serine-phosphoethanolamine	301	3.46	Neg	$C_5H_{13}N_2O_6P$	Lipid	17.35	1.87E-05	18.07	3.29E-06	2.82	5.75E-03
16	(3Z,6Z)-Nonadienal	183	4.93	Pos	$C_9H_{14}O$	Lipid	55.70	2.60E-09	54.20	1.55E-11	76.42	7.31E-11
17	3,5-Dimethoxyphenol	177	9.00	Pos	$C_8H_{10}O_3$	Phenylpropanoid	2.91	1.07E-04	3.63	1.88E-05	-	-
18	Trans-beta-D-Glucosyl-2-hydroxycinnamate	325	6.90	Neg	$C_{15}H_{18}O_8$	Phenylpropanoid	2.47	2.74E-05	2.77	2.64E-04	-	-
19	Shikimate	195	5.59	Neg	$C_7H_{10}O_5$	Phenylpropanoid	2.28	2.43E-05	2.29	3.62E-04	-	-
20	Trans-5-O-caffeoyl-D-quinate	561	6.09	Neg	$C_{16}H_{17}O_9$	Phenylpropanoid	6.62	1.15E-02	11.96	2.79E-04	7.43	7.65E-04
21	Gibberellin A36	415	9.07	Pos	$C_{20}H_{26}O_6$	Phytohormone	2.35	1.92E-05	-	-	-	-
22	(-)-Phenylalanine Jasmonate conjugate	450	4.30	Pos	$C_{21}H_{29}NO_4$	Phytohormone	2.63	1.03E-03	-	-	2.48	1.59E-04
23	Dihydrozeatin-9-N-glucoside-O-glucoside	584	1.74	Pos	$C_{22}H_{35}N_5O_{11}$	Phytohormone	2.35	1.18E-04	3.59	6.48E-05	3.38	8.44E-06
24	Thermozeaxanthin	948	14.12	Pos	$C_{51}H_{84}O_{12}$	Phytohormone	2.31	3.37E-04	-	-	-	-
25	Gibberellin A36	407	8.94	Neg	$C_{20}H_{26}O_6$	Phytohormone	2.88	1.11E-07	5.39	4.65E-10	2.50	3.33E-04
26	(-)-11-Hydroxy-9,10-dihydrojasmonic acid 11-beta-D-glucoside	475	8.00	Pos	$C_{18}H_{30}O_9$	Phytohormone	-	-	16.43	1.35E-08	8.56	3.57E-06
27	3-Oxopropionyl-CoA	853	1.96	Neg	$C_{24}H_{38}N_7O_{18}P_3S$	Propanoate	2.10	6.35E-05	2.38	2.00E-03	2.46	1.39E-03
28	Dimethylbenzimidazole	205	5.59	Pos	$C_9H_{10}N_2$	Riboflavin	2.34	4.68E-05	2.20	3.15E-03	2.39	2.17E-03
29	Demethylphylloquinol	557	11.07	Pos	$C_{30}H_{46}O_2$	Ubiquinone	3.81	9.89E-05	2.74	1.22E-02	4.40	7.06E-05
30	Ubiquinone	357	4.58	Pos	$C_{14}H_{18}O_4$	Ubiquinone	9.83	7.57E-04	17.94	1.17E-04	6.72	1.07E-02

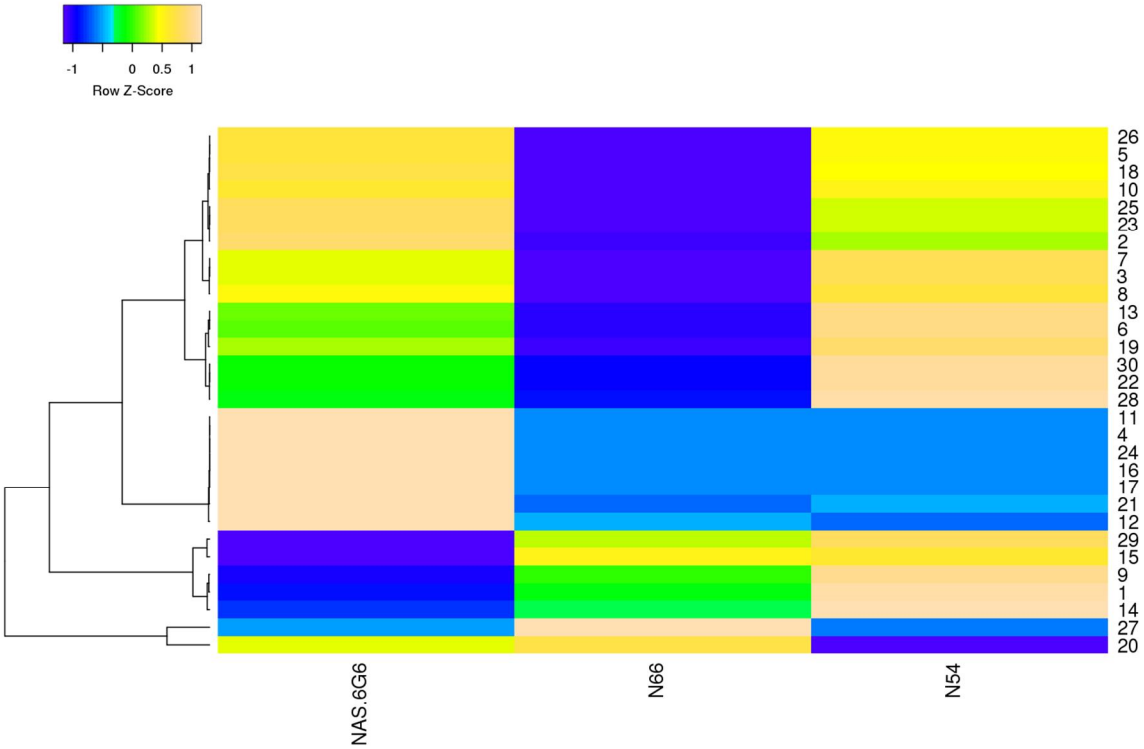


Figure S3 Heatmap comparing the fold change (primed *versus* naïve) of the top 30 metabolites significantly upregulated in *S. bicolor* seedlings primed with rhizobacterial isolates NAS-6G6, N66 and N54 in response to biotic stress (inoculation with *F. pseudograminearum*). The number to the right corresponds to the metabolite number in Table S3.

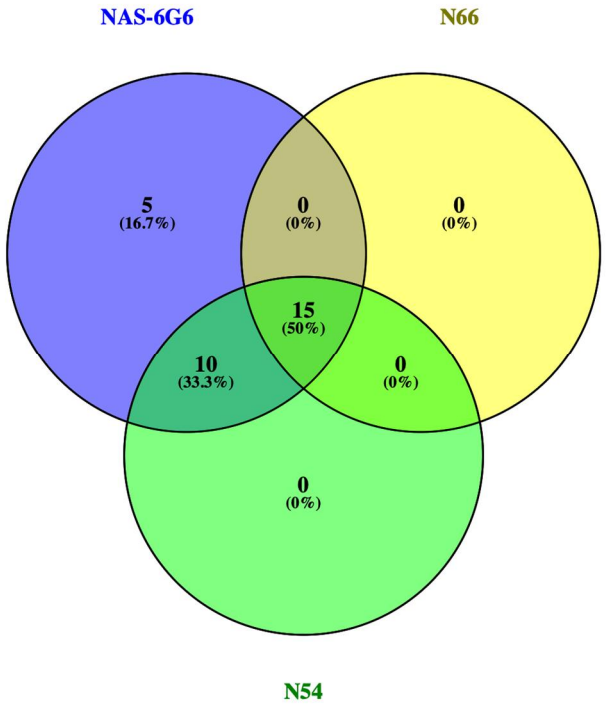


Figure S4 Venn diagram comparing the number of metabolites, of the top 30 metabolites significantly upregulated in *S. bicolor* seedlings primed with rhizobacterial isolates NAS-6G6, N66 and N54 in response to biotic stress (inoculation with *F. pseudograminearum*).

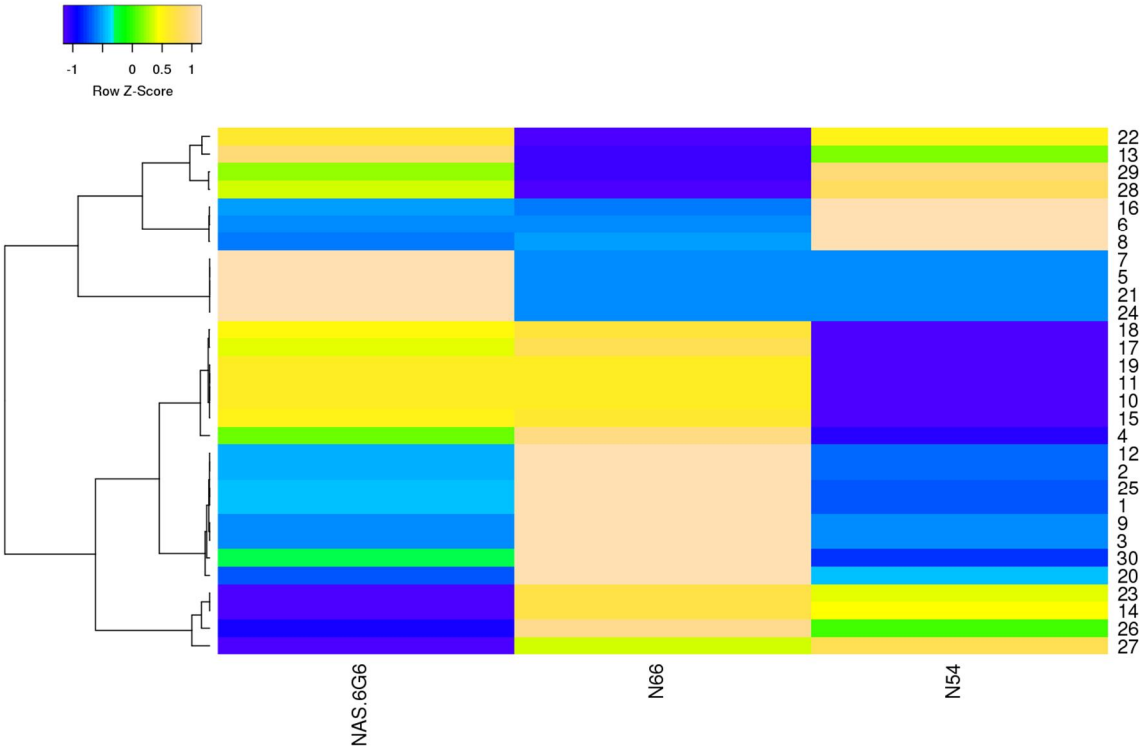


Figure S5 Heatmap comparing the fold change (primed *versus* naïve) the top 30 metabolites significantly upregulated in *S. bicolor* seedlings primed with rhizobacterial isolates NAS-6G6, N66 and N54 in response to abiotic stress (drought). The number to the right corresponds to the metabolite number in Table S4.

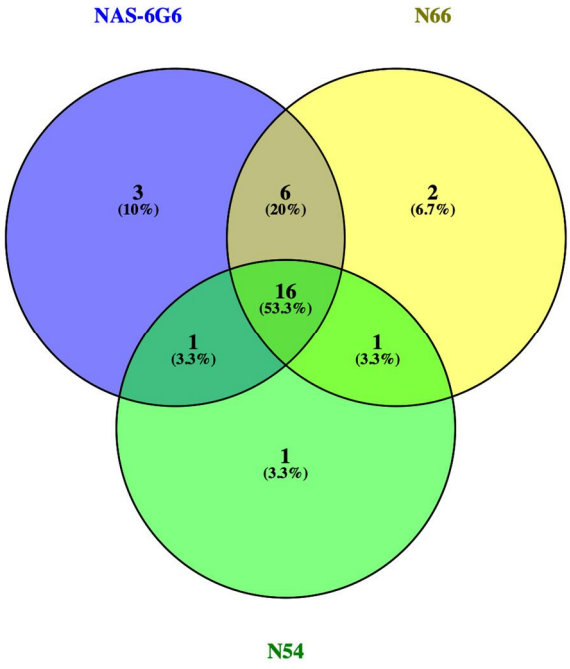


Figure S6 Venn diagram comparing the number of metabolites, of the top 30 metabolites significantly upregulated in *S. bicolor* seedlings primed with rhizobacterial isolates NAS-6G6, N66 and N54 in response to abiotic stress (inoculation with *F. pseudograminearum*).

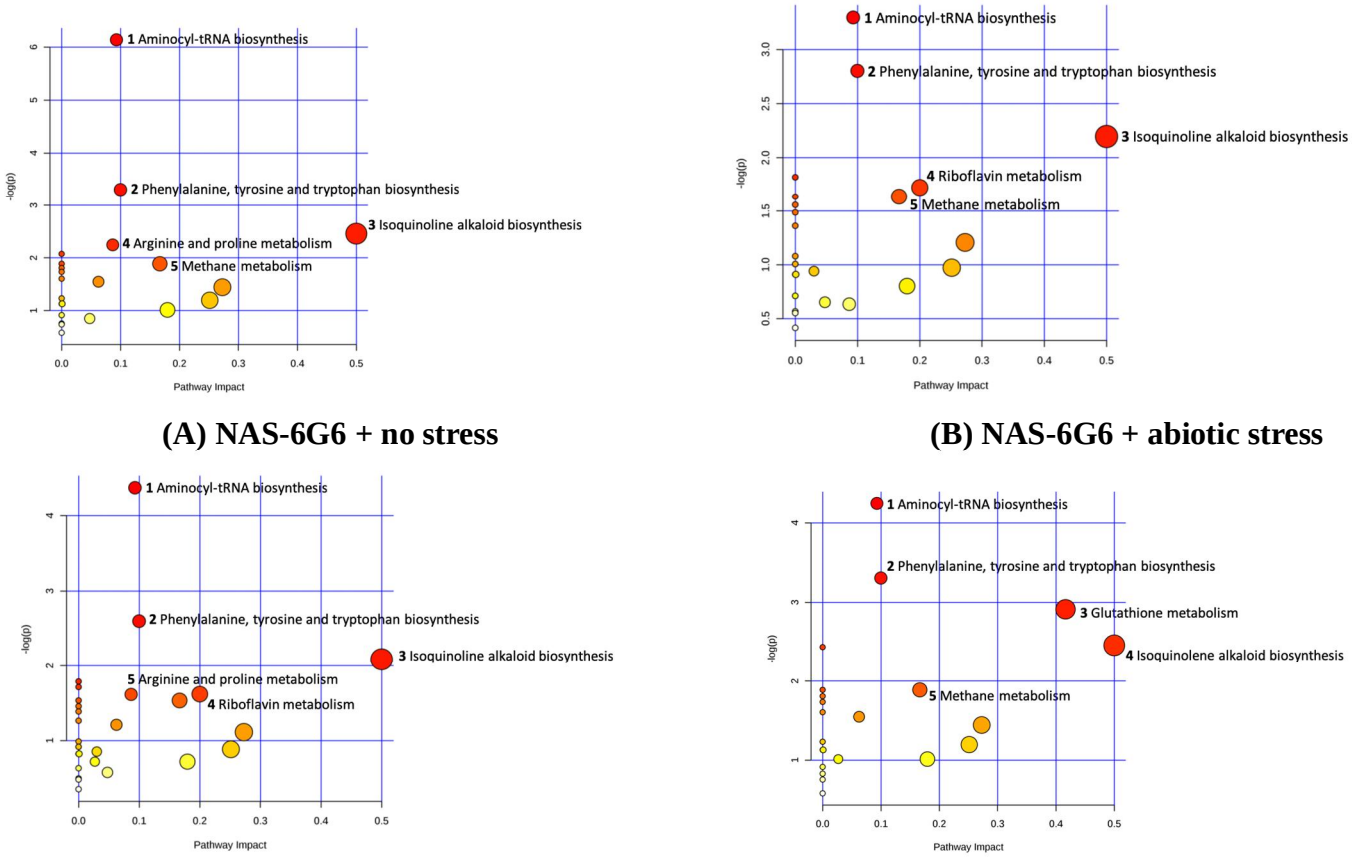
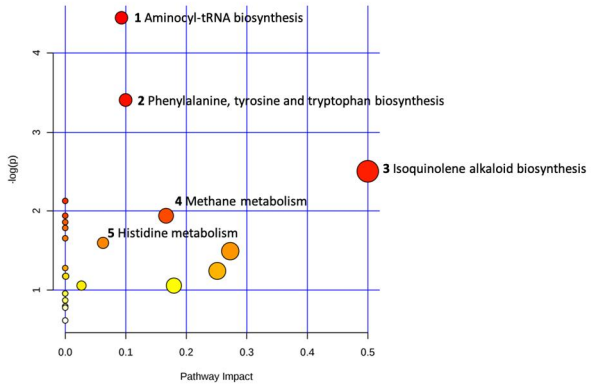
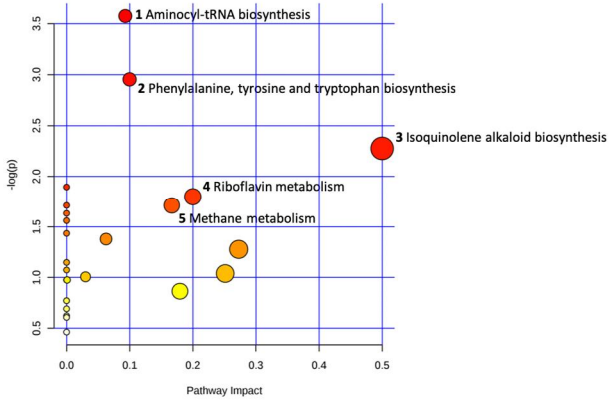


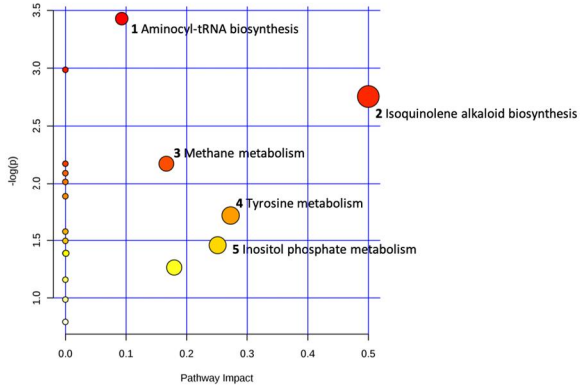
Figure S7 Summary of pathway analysis with MetPA of the effect of priming *S. bicolor* with *Pa. alvei* NAS-6G6 under conditions of (A) no stress, (B) drought, (C) inoculation with *F. pseudograminearum* and (D) combined stresses: Representation of all MetPA-computed metabolic pathways displayed per their significance or pathway impact. The graph, "metabolome view" contains all the matched pathways (the metabolome) arranged by *p*-values (pathway enrichment analysis) on *y*-axis, and the pathway impact values (pathway topology analysis) on *x*-axis. The node colour is based on the *p*-value and the node radius is defined by the pathway impact values. The latter is the cumulative percentage from the matched metabolite nodes. Thus, the graph indicates pathways with high impact. Only pathways of high impact (red) are labelled.



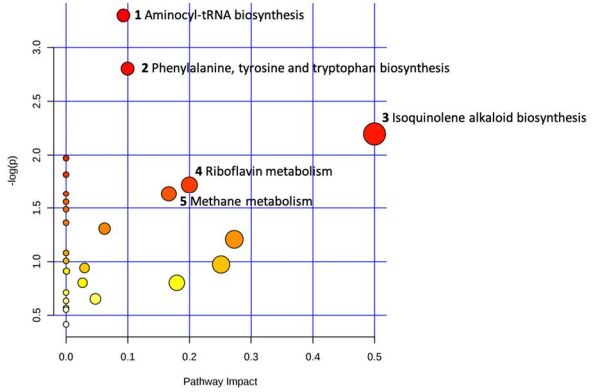
(A) N66 + no stress



(B) N66 + abiotic stress



(C) N66 + biotic stress



(D) N66 + combined stresses

Figure S8 Summary of pathway analysis with MetPA of the effect of priming *S. bicolor* with *Ps. taiwanensis* N66 under conditions of (A) no stress, (B) drought, (C) inoculation with *F. pseudograminearum* and (D) combined stresses: Representation of all MetPA-computed metabolic pathways displayed per their significance or pathway impact. The graph, "metabolome view" contains all the matched pathways (the metabolome) arranged by *p*-values (pathway enrichment analysis) on *y*-axis, and the pathway impact values (pathway topology analysis) on *x*-axis. The node colour is based on the *p*-value and the node radius is defined by the pathway impact values. The latter is the cumulative percentage from the matched metabolite nodes. Thus, the graph indicates pathways with high impact. Only pathways of high impact (red) are labelled.