

Supplementary Material

Soil origin impacts *Acacia longifolia* above and belowground development: water and nutrition as players

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Supplementary Tables

Table S1: Summary table of the average (mean \pm SD) shoot increment, root length, root/shoot length ratio, total number and weight of nodules, number of phyllodes, hydric content of phyllodes, total chlorophylls (C_{a+b}) and carotenoids content in the tip and the base of the phyllodes in *A. longifolia* grown for 20 weeks with twelve different treatment combinations. * C_{a+b} and carotenoids content at phyllode tip in DW+N- treatment had no standard deviation associated since it was only possible to analyze one individual due to the size of the phyllodes.

Treatments	Shoot increment (cm)	Root length (cm)	Root/shoot length ratio	Number of nodules	Total nodule weight (g)	Number of phyllodes	Phyllode water content	C_{a+b} content		Carotenoids content	
								Phyllode tip (mg/cm ²)	Phyllode base (mg/cm ²)	Phyllode tip (mg/cm ²)	Phyllode base (mg/cm ²)
FW+N+	57.21 \pm 14.5	42.1 \pm 13.3	0.73 \pm 0.06	58.5 \pm 30.9	0.21 \pm 0.12	6.6 \pm 1.8	80.25 \pm 1.8	36.4 \pm 4.4	54.32 \pm 10.1	6.73 \pm 1.2	8.57 \pm 1.7
FW+N-	35.50 \pm 12.9	25.06 \pm 10.3	0.70 \pm 0.07	31.75 \pm 17.1	0.14 \pm 0.07	2.9 \pm 1.0	83.49 \pm 5.0	40.71 \pm 2.9	57.76 \pm 5.9	8.20 \pm 0.9	9.80 \pm 0.7
FW-N+	47.66 \pm 13.3	34.29 \pm 12.3	0.72 \pm 0.16	41.5 \pm 38.5	0.16 \pm 0.13	3.38 \pm 1.1	76.70 \pm 5.9	43.19 \pm 17.0	55.28 \pm 13.4	8.07 \pm 2.3	9.41 \pm 1.9
FW-N-	50.01 \pm 10.7	36.65 \pm 9.8	0.73 \pm 0.09	31.38 \pm 22.1	0.12 \pm 0.09	4.25 \pm 1.6	78.21 \pm 5.5	38.07 \pm 3.4	56.56 \pm 14.4	7.31 \pm 0.3	8.57 \pm 2.4
AW+N+	55.78 \pm 13.9	37.55 \pm 12.1	0.67 \pm 0.09	11.63 \pm 10.1	0.1 \pm 0.08	4.87 \pm 1.7	88.24 \pm 4.6	30.19 \pm 5.7	51.03 \pm 9.0	5.42 \pm 0.5	7.57 \pm 0.6
AW+N-	47.85 \pm 10.6	33.39 \pm 9.1	0.69 \pm 0.08	19.0 \pm 13.5	0.29 \pm 0.16	3.25 \pm 1.0	84.40 \pm 4.7	35.06 \pm 3.7	52.07 \pm 9.2	6.46 \pm 0.5	7.88 \pm 1.1
AW-N+	47.90 \pm 8.3	32.73 \pm 7.6	0.68 \pm 0.08	14.13 \pm 17.5	0.09 \pm 0.11	4.25 \pm 1.6	82.89 \pm 5.9	37.55 \pm 5.7	51.84 \pm 11.2	6.94 \pm 0.82	7.78 \pm 1.0
AW-N-	56.34 \pm 12.8	40.66 \pm 12.8	0.70 \pm 0.13	15.13 \pm 9.6	0.2 \pm 0.17	4.13 \pm 1.1	78.55 \pm 4.6	31.78 \pm 3.9	41.57 \pm 7.0	5.58 \pm 0.7	5.68 \pm 0.9
DW+N+	48.09 \pm 11.9	38.6 \pm 10.8	0.79 \pm 0.07	52.38 \pm 30.5	0.17 \pm 0.08	3.25 \pm 1.0	82.06 \pm 3.9	35.93 \pm 10.5	45.95 \pm 14.2	6.67 \pm 1.1	7.81 \pm 2.0
DW+N-	32.34 \pm 11.4	23.58 \pm 9.1	0.72 \pm 0.05	18.25 \pm 11.4	0.09 \pm 0.08	1.88 \pm 1.1	82.79 \pm 3.3	27.87*	53.82 \pm 19.0	5.62*	8.63 \pm 3.3
DW-N+	45.08 \pm 16.8	30.23 \pm 17.2	0.63 \pm 0.17	33.29 \pm 21.3	0.22 \pm 0.29	5.13 \pm 1.7	82.28 \pm 4.2	43.69 \pm 10.0	62.03 \pm 4.5	8.19 \pm 1.6	10.04 \pm 0.67
DW-N-	37.7 \pm 10.06	27.96 \pm 9.6	0.73 \pm 0.09	19.62 \pm 16.5	0.08 \pm 0.05	2.75 \pm 1.0	83.12 \pm 5.0	40.22 \pm 2.8	54.84 \pm 10.8	8.02 \pm 0.8	8.97 \pm 1.6

Table S2: Summary table of the average (mean \pm SD) phyllode and nodule $\delta^{15}\text{N}$, $\delta^{13}\text{C}$, nitrogen (N) and carbon (C) content and their ratio (C/N) in *A.longifolia* grown for 20 weeks with 12 different treatment combinations.

Treatments	Phyllode					Nodule				
	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	N%	C%	C/N	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	N%	C%	C/N
FW+N+	4.03 \pm 4.3	-31.9 \pm 0.17	3.54 \pm 0.6	43.9 \pm 0.7	12.7 \pm 2.4	7.80 \pm 1.4	-30.9 \pm 0.4	5.68 \pm 0.9	40.0 \pm 1.8	7.13 \pm 0.8
FW+N-	1.03 \pm 2.4	-32.1 \pm 0.5	3.68 \pm 0.8	43.8 \pm 0.3	12.4 \pm 3.1	7.20 \pm 2.9	-30.5 \pm 1.0	5.86 \pm 0.5	40.9 \pm 1.2	7.01 \pm 0.6
FW-N+	7.70 \pm 4.7	-31.6 \pm 0.7	3.02 \pm 0.1	43.6 \pm 1.0	14.5 \pm 0.8	7.97 \pm 0.6	-30.3 \pm 0.9	4.97 \pm 0.4	35.0 \pm 4.4	7.03 \pm 0.3
FW-N-	2.90 \pm 4.8	-31.9 \pm 0.4	3.29 \pm 0.2	44.1 \pm 0.7	13.4 \pm 0.5	5.43 \pm 5.4	-33.3 \pm 5.0	4.71 \pm 0.9	28.3 \pm 19.2	5.59 \pm 3.5
AW+N+	8.13 \pm 0.5	-32.5 \pm 0.9	3.61 \pm 0.3	44.0 \pm 0.1	12.3 \pm 1.1	9.67 \pm 0.6	-30.9 \pm 0.5	4.21 \pm 1.1	33.0 \pm 5.7	7.99 \pm 0.9
AW+N-	2.90 \pm 4.6	-32.3 \pm 0.3	3.41 \pm 1.1	44.1 \pm 0.03	13.9 \pm 4.7	7.73 \pm 1.8	-30.6 \pm 0.3	4.84 \pm 0.8	38.7 \pm 1.0	8.11 \pm 1.1
AW-N+	8.77 \pm 2.6	-31.7 \pm 1.4	3.38 \pm 0.4	44.0 \pm 1.2	13.1 \pm 1.8	10.3 \pm 1.5	-29.7 \pm 0.7	3.79 \pm 0.7	38.4 \pm 0.5	10.3 \pm 1.6
AW-N-	4.30 \pm 3.2	-31.3 \pm 1.2	3.27 \pm 0.5	44.6 \pm 0.1	13.8 \pm 2.2	6.60 \pm 1.6	-29.6 \pm 1.2	5.21 \pm 0.3	38.1 \pm 3.0	7.34 \pm 0.9
DW+N+	5.17 \pm 2.5	-31.7 \pm 1.0	3.33 \pm 0.4	43.5 \pm 0.4	13.2 \pm 2.0	6.87 \pm 2.7	-30.7 \pm 1.2	3.64 \pm 1.0	30.9 \pm 7.9	8.51 \pm 0.6
DW+N-	-0.13 \pm 0.6	-32.6 \pm 0.5	3.34 \pm 0.4	44.6 \pm 0.8	13.5 \pm 1.7	6.43 \pm 1.0	-31.8 \pm 0.4	4.85 \pm 1.0	37.4 \pm 5.4	7.80 \pm 0.7
DW-N+	1.23 \pm 0.8	-31.8 \pm 0.6	3.76 \pm 0.1	43.6 \pm 0.4	11.6 \pm 0.4	7.37 \pm 0.5	-30.9 \pm 1.2	5.0 \pm 0.9	38.6 \pm 2.9	7.82 \pm 0.8
DW-N-	-1.5 \pm 0.7	-31.9 \pm 1.0	3.66 \pm 0.7	43.8 \pm 0.3	12.3 \pm 2.4	5.67 \pm 2.7	-31.0 \pm 0.7	4.78 \pm 1.8	38.2 \pm 5.9	8.14 \pm 0.9

Table S3: Summary of the post-hoc analysis following 3-way-ANOVA for shoot vigour and root development - results from the Tukey multiple comparisons of means differences considering 95% family-wise confidence level. Only statistically significant results are presented.

Factors	Shoot vigour				Root development			
	Difference	95% family-wise CI		Adjusted p-value	Difference	95% family-wise CI		Adjusted p-value
		Lower	Upper			Lower	Upper	
<i>Soil</i>								
A - D	17.56	6.66	28.47	0.001	-	-	-	-
F - D	14.29	3.38	25.19	0.007	-	-	-	-
F - A	-	-	-	-	12.58	3.58	21.58	0.004
<i>Nutrition</i>								
N+ - N-	17.80	10.38	25.22	<0.001	9.30	3.17	15.42	0.003
<i>Soil: Watering</i>								
AW- - DW+	24.93	6.08	43.78	0.003	-	-	-	-
AW+ - DW+	21.31	2.46	40.16	0.017	-	-	-	-
FW+ - DW+	20.92	2.07	39.77	0.021	-	-	-	-
<i>Soil: Nutrition</i>								
FN+ - DN-	35.79	16.94	54.64	<0.001	23.45	7.90	39.00	<0.001
FN+ - AN+	-	-	-	-	21.17	5.62	36.72	0.002
FN+ - AN-	-	-	-	-	17.89	2.33	33.44	0.015

AN+ - DN-	35.22	16.37	54.07	<0.001	-	-	-	-
DN+ - DN-	24.93	6.08	43.78	0.003	17.28	1.72	32.83	0.021
AN- - DN-	24.84	5.99	43.69	0.003	-	-	-	-
<i>Watering:Nutrition</i>								
W+N+ - W+N-	30.35	16.51	44.18	<0.001	16.28	4.86	27.69	0.002
W+N+ - W-N-	-	-	-	-	12.31	0.90	23.73	0.029
W-N+ - W+N-	22.00	8.17	35.83	<0.001	-	-	-	-
W-N- - W+N-	16.75	2.92	30.58	0.011	-	-	-	-
<i>Soil:Watering:Nutrition</i>								
FW+N+ - DW+N-	54.48	23.75	85.20	<0.001	31.77	6.42	57.12	0.003
AW+N+ - DW+N-	46.44	15.71	77.17	<0.001	-	-	-	-
FW+N+ - AW-N+	-	-	-	-	28.57	3.21	53.92	0.014
FW+N+ - DW-N-	41.94	11.21	72.67	0.001	28.50	3.15	53.86	0.014
FW+N+ - FW+N-	40.77	10.04	71.50	0.001	-	-	-	-
AW-N- - DW+N-	39.68	8.95	70.41	0.002	-	-	-	-

Table S3 (continued)

Shoot vigour	Root development
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Factors	95% family-wise CI				95% family-wise CI			
	Difference	Lower	Upper	Adjusted p-value	Difference	Lower	Upper	Adjusted p-value
<i>Soil: Watering: Nutrition</i>								
AW-N+ - DW+N-	36.53	5.81	67.26	0.007	-	-	-	-
DW-N+ - DW+N-	36.05	5.32	66.78	0.009	-	-	-	-
DW+N+ - DW+N-	-	-	-	-	26.51	1.15	51.86	0.032
FW-N- -DW+N-	34.26	3.53	64.99	0.016	-	-	-	-
AW+N+ - DW-N-	33.91	3.18	64.63	0.018	-	-	-	-
AW+N+ - FW+N-	32.74	2.01	63.47	0.027	-	-	-	-
FW+N+ - AW+N-	31.95	1.22	62.68	0.034	25.65	0.30	51.01	0.045
FW+N+ - AW+N+	-	-	-	-	27.14	1.79	52.50	0.025

Supplementary Figures

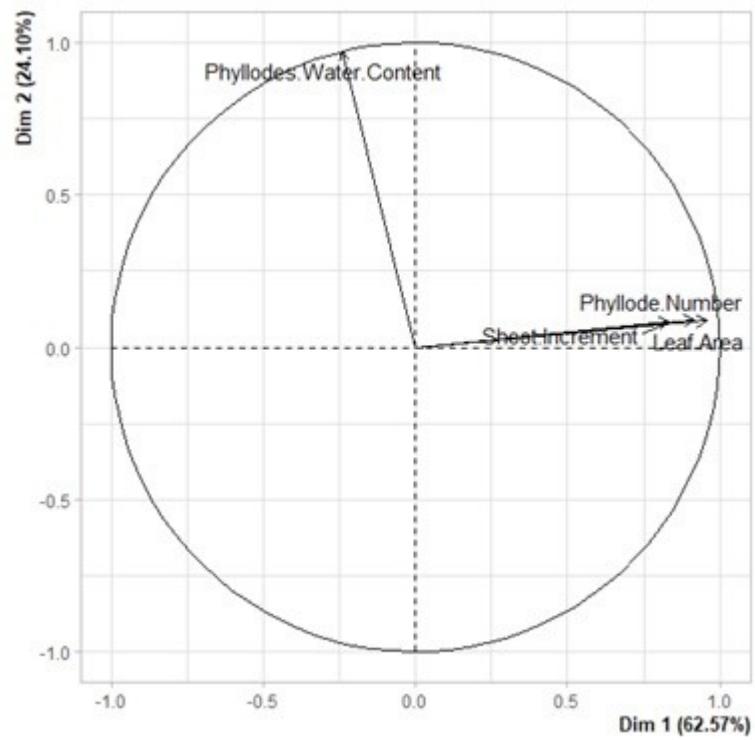


Fig. S1: Correlation circle obtained by principal component analysis (PCA) of the shoot vigour variables: shoot increment, number of phyllodes, leaf area and phyllodes water content.

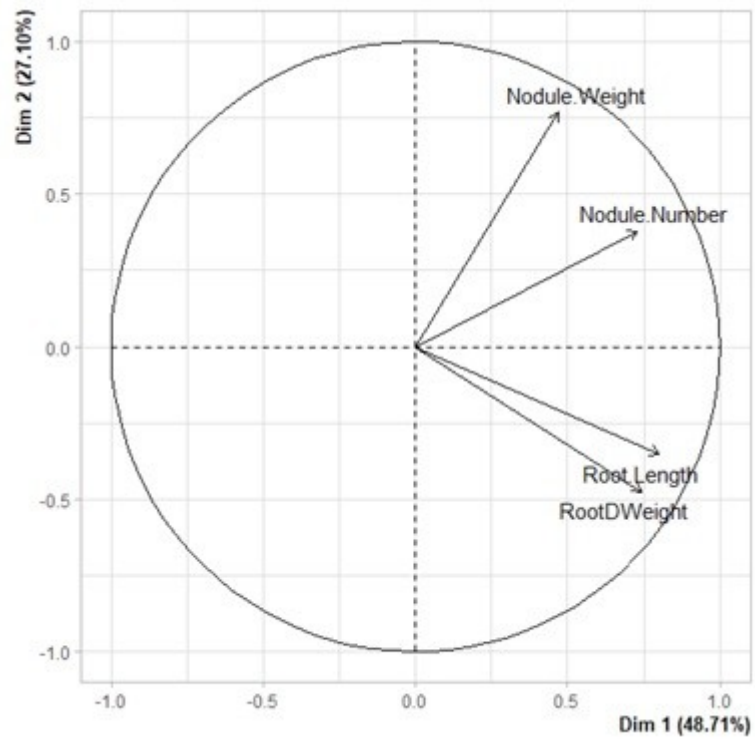


Fig. S2: Correlation circle obtained by principal component analysis (PCA) of the root development variables: root length, number of nodules, total nodule weight and roots dry weight.

Fig. S3: Dendrogram based on cluster analysis of fingerprinting PCR profiles of the isolates from young *A. longifolia* nodules, using the Pearson correlation coefficient and the unweighted pair-group method with arithmetic mean algorithm (UPGMA). Primers used for PCR amplification were PH and GTG5. On the right are represented the treatments from where the bacteria were isolated (soil type, watering, and nutrition regime).