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Functional Plant Biology

Supplementary Material

Investigating the combined effects of β-sitosterol and biochar on nutritional value and drought tolerance in *Phaseolus vulgaris* under drought stress

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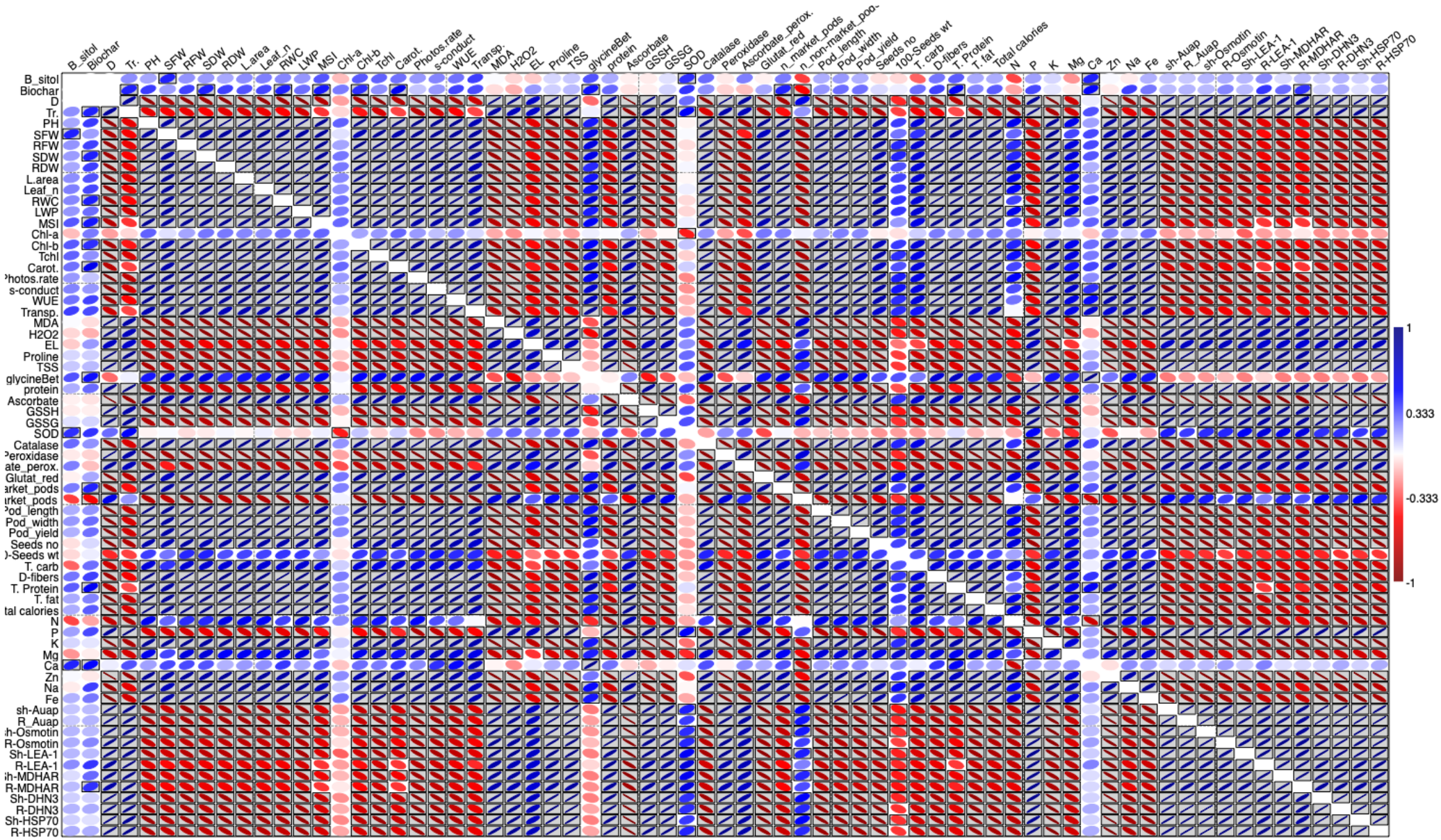
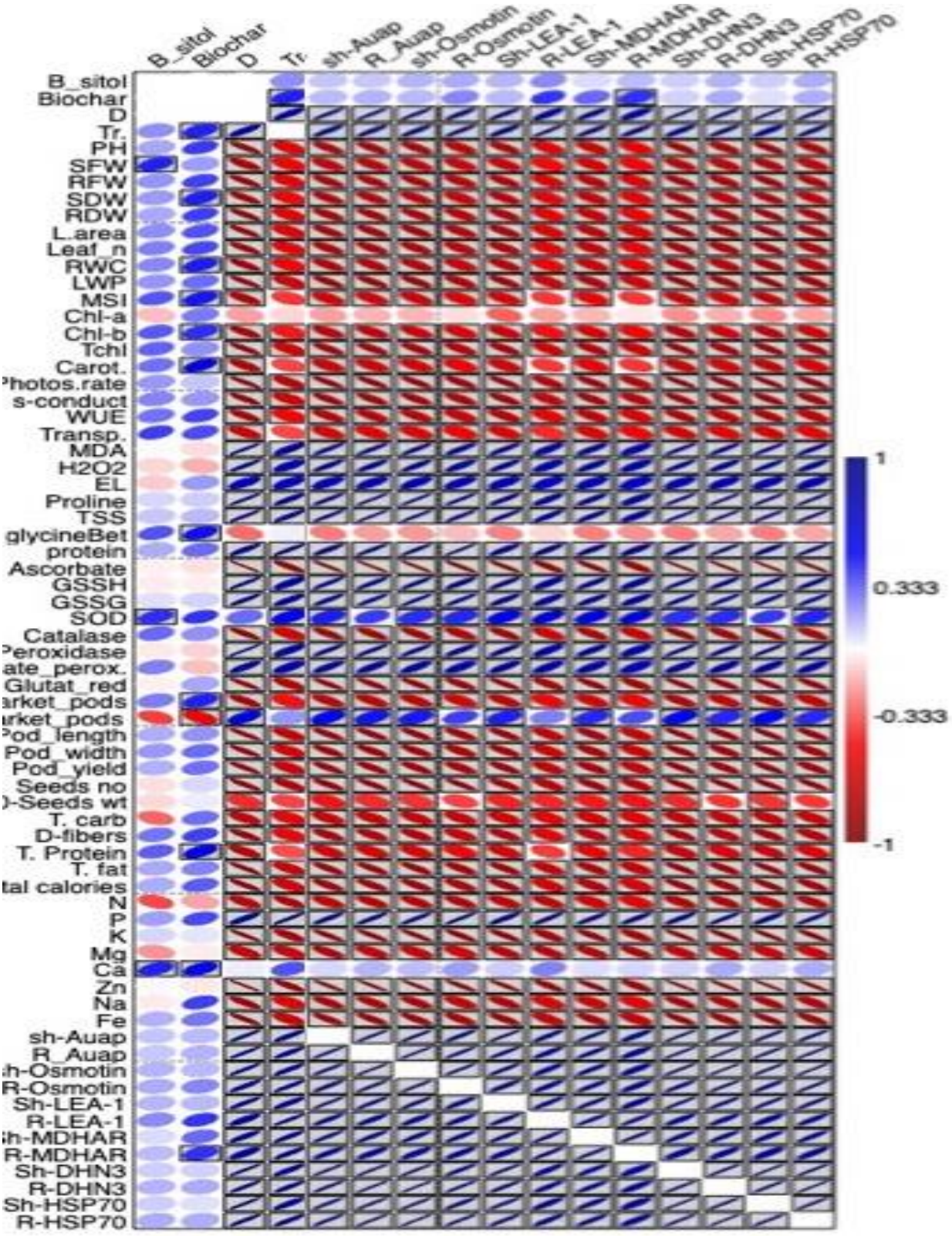
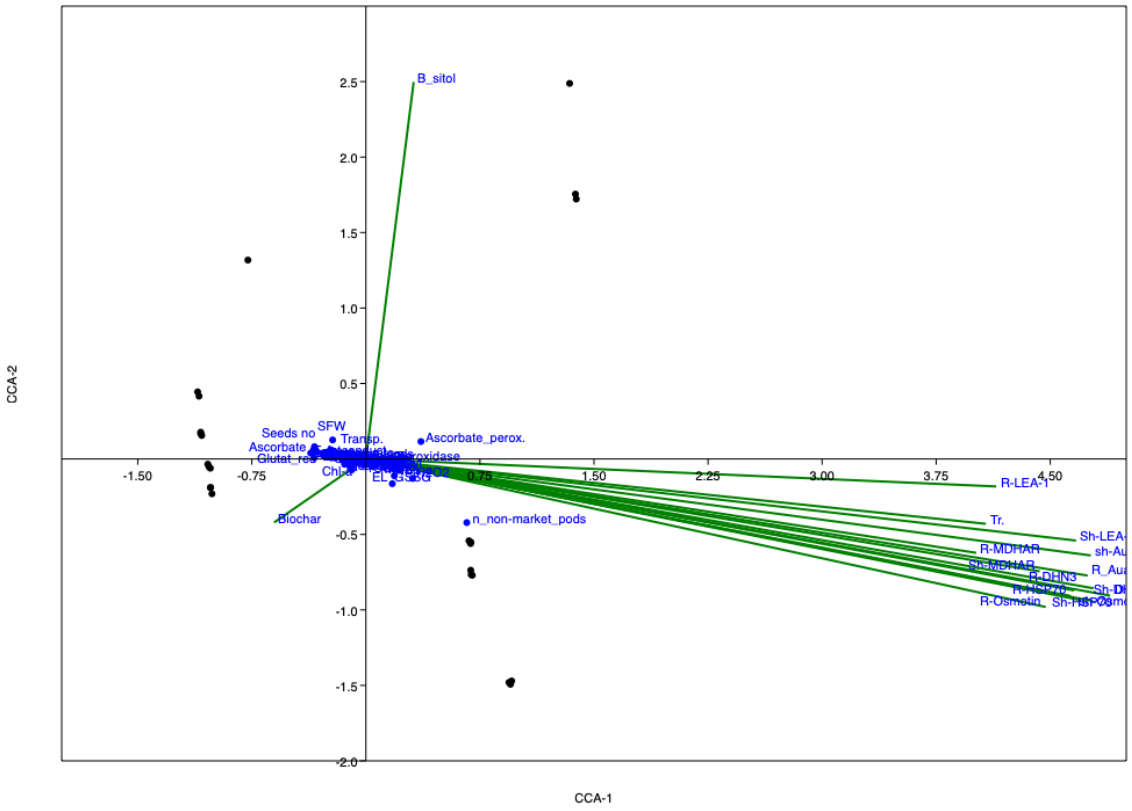


Supplementary Figure S1. Heatmap showing the relationship between study variables.



Supplementary Figure S2. Heatmap showing the relationship between study variables.



Supplementary Figure S3. Canonical correspondence analysis (CCA) shows the interaction between variables and the interaction between gene expression and various measure parameters.

Supplementary Table S1. Physico-chemical properties of *Eucalyptus* wood Biochar (EB) and sandy loam soil.

Chemical analysis	Sandy loam soil	EW Biochar
Organic matter (%)	31.05±0.41	75.00±0.062
Phosphorus (%)	2.19± 0.003	3.23±0.021
Nitrogen (%)	251±1.04	316.5±0.42
pH	7.26±0.24	7.6±0.176
ECe (dS m^{-1})	0.69±0.042	1.08±0.011
	Cations (meq L^{-1})	
Potassium	162±0.81	251.9±0.007
Sodium	0.003±0.014	0.009
Magnesium	0.025±0.004	0.334
Calcium	1.230.025	2.88
	Anions (meq L^{-1})	
Sulphate	2.03	4.5
Chloride	3.41	1.88
Bicarbonate	1.97	2.2
Carbonate	0.62	0.99
	Physical properties	
Soil texture		
Sand (%)	54.1	ND
Silt (%)	28.5	ND
Clay (%)	17.4	ND

Supplementary Table S2. List of Pprimers used

Gene Name		Sequence	Tm (C)
DHN3	F	F 5´- CATGGCGTCTACTGCTTGTA -3’	
	R	R 5´- CAGAGGACTTGAACCCAGATAC -3’	
HSP70	F	5′- CCATGAAGCTCTACAACGAG -3′	
	R	5′- GTAGAAGTAGGGCAGGTAGT -3′	
MDHAR	F	F5´- CCATGAAGCTCTACAACGAG -3’	
	R	R5´- GTAGAAGTAGGGCAGGTAGT -3’	

Aquaporin	F	5′- GTTCCTATCCTTGCCCCACT -3′	60
	R	5′- AGGCGTGATCCCTGTTGTAG -3′	
Osmotin-34	F	5‟- GAACGGAGGGTGTCACAAAATC -3'	
	R	5‟- CGTAGTGGGTCCACAAGTTCCT -3'	
LEA-1	F	5‟- CAGCGAAGTTTGGATGGAATG-3'	
	R	5‟- ACCTGTCGCCAATCAGAAGAT-3'	
β-Actin	F	5´-GTGCCCATTTACGAAGGATA- 3´	
	R	5´-GAAGACTCCATGCCGATCAT- 3´	

Supplementary Table S3. Yield parameters were recorded in the eight treatment groups.

	Yield parameters						
	Number of marketable pods	**Number of non-marketable pods**	**Pod length (mm)**	**Pod width (mm)**	**Pod yield (g)**	**Seeds plant^{-1}**	**100-Seeds weight (g)**
Control	11.7±0.58 cd	1.3±0.58 bc	106.0±1.00 c	5.9±0.06 b	3.1±0.01 b	72.0±1.00 d	37.5±0.02 ab
β-sitosterol	13.0±1.00 bc	0.0±0.00 d	109.3±0.58 b	6.0±0.01 c	3.2±0.01 a	75.0±1.00 c	39.2±0.01 a
Biochar	14.0±1.00 ab	0.0±0.00 d	113.0±1.00 a	5.9±0.01 b	3.2±0.01 a	77.0±1.00 b	39.3±0.01 a
Biochar +β-sitosterol	15.0±1.00 a	0.0±0.00 d	112.3±0.58 a	6.1±0.01 a	3.3±0.01 a	81.0±1.00 a	30.1±17.31 ab
Drought stress	6.7±0.58 f	3.7±0.58 a	87.0±1.00 g	5.3±0.01 g	2.6±0.02 f	37.0±1.00 g	27.2±0.01 b
Drought +β-sitosterol	8.3±0.58 e	1.7±0.58 b	93.0±1.00 e	5.4±0.01 f	2.7±0.01 e	46.0±1.00 f	30.1±0.02 ab
Drought +Biochar	9.3±0.58 e	1.0±0.00 bc	91.3±0.58 f	5.5±0.02 e	2.8±0.01 d	54.0±1.00 e	32.9±0.02 ab
Drought +Biochar +β-sitosterol	11.0±1.00 d	0.7±0.58 cd	95.0±1.00 d	5.6±0.02 d	2.9±0.01 c	29.0±1.00 h	33.6±0.01 ab
ANOVA (p-value)	**<0.001*** **	**<0.001*** **	**0.008** **	**<0.001*** **	**<0.001*** **	**<0.001*** **	**0.189 ns**

*, **, ***, significant at p<0.05, <0.01, <0.001; ns, non-significant at p>0.05

[a,b] Means followed by different letters vertically (in the same column are significantly different according to DMRT.