Interactive effects of CO_2 and nitrogen supply on growth and physiological traits of millet cultivars under drought stress

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Supplemental material

Table S1. Analysis of variance for different measured traits of two millet cultivars (C_1: Bastan and C_2: Pishahang) under effect of two environment (Ambient "390±50 µmol mol^{-1}" and Elevated CO_2 "700±50 µmol mol^{-1}"), two irrigation regimes (Control "40% MAD", Stress "75% MAD") and two nitrogen levels (0 and 85 mgN kg^{-1} soil).

Sources of variance	DF	APX	CAT	POX	Chla	Chlb	Car	LPC	MSI	H_2O_2	MDA	LA	ShN	RDW	ShDW
E	1	**	**	**	**	**	**	**	**	**	**	**	**	**	**
I	1	**	**	**	**	**	**	**	**	**	**	**	**	**	**
N	1	**	**	**	**	**	**	**	**	**	**	**	**	**	**
C	1	**	**	**	**	**	**	**	**	**	**	*	**	**	**
E×I	1	**	*	ns	**	**	ns	ns	ns	**	ns	**	**	*	ns
E×N	1	ns	ns	ns	**	**	ns	ns	ns	**	ns	**	ns	ns	ns
E×C	1	**	**	ns	ns	ns	**	ns	ns	ns	**	ns	ns	ns	ns
E×I×N	1	ns	ns	ns	**	**	ns	ns	ns	ns	ns	ns	ns	**	ns
E×I×C	1	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	*	ns	ns
E×N×C	1	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	**	ns	ns
E×I×N×C	1	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
I×N	1	ns	ns	ns	**	**	ns	ns	ns	ns	**	**	ns	ns	**
I×C	1	ns	**	*	ns	ns	ns	ns	ns	ns	**	ns	ns	ns	ns
N×C	1	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns
I×N×C	1	ns	ns	*	*	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

APX ascorbate peroxidase specific activity, CAT catalase specific activity, POX peroxidase specific activity, Chla chlorophyll a content, Chlb chlorophyll b content, Car carotenoids content, LPC leaf prolin content, MSI membrane stability index, H_2O_2 Hydrogen peroxide, MDA Malondialdehyde, LA leaf area, ShN Shoot nitrogen concentration, RDW root dry weight, ShDW Shoot dry weight .

^{ns} Non significant.* Significant at the 0.05 probability level.** Significant at the 0.01 probability level.

Table S2. Interaction effects of CO_2 concentration, irrigation regimes and nitrogen levels on some of the traits studied in millet cultivars.

Traits	Irrigation	Environments				LSD 5%, P value
		Ambient CO_2		Elevated CO_2		
		0	85	0	85	
Chla	Control	1.17c	1.44b	1.49b	2.12a	0.106, 0.01
	Stress	0.877d	1.15c	1.11c	1.45b	
Chlb	Control	0.566d	0.642c	0.708b	1.01a	0.065, 0.01
	Stress	0.431e	0.515d	0.541d	0.627c	
RDW	Control	0.922cd	1.29b	1.59a	1.66a	0.175, 0.01
	Stress	0.847d	0.932cd	1.05c	1.35b	

Chla chlorophyll a content and Chlb chlorophyll b (mg g^{-1}FW), RDW root dry weight (g $plant^{-1}$).
Environments: Ambient (390±50 µmol mol^{-1}) and Elevated CO_2 (700±50 µmol mol^{-1}). Irrigation regimes: Control (40% MAD), Stress (75% MAD). Nitrogen levels: 0 and 85 (mgN kg^{-1} soil).

Table S3. Interaction effects of CO_2 concentration× irrigation regimes× cultivars and, CO_2 concentration× nitrogen levels× cultivars on shoot nitrogen.

Treatments		Environments				LSD 5%, P value
		Ambient CO_2		Elevated CO_2		
		Bastan	Pishahang	Bastan	Pishahang	
Irrigation	Control	37.3d	35.1e	32.3f	27.4g	1.48, 0.05
	Stress	49.4a	45.1b	40.5c	36.7d	
Nitrogen levels	0	41.7b	37.8c	33.4e	30.8f	0.16, 0.01
	85	44.9a	42.4b	34.9d	33.4e	

Environments: Ambient (390±50 µmol mol^{-1}) and Elevated CO_2 (700±50 µmol mol^{-1}). Irrigation regimes: Control (40% MAD), Stress (75% MAD).

Table S4. Interaction effects of irrigation regimes, nitrogen levels and cultivars on some of the traits studied in millet cultivars.

Traits	Cultivars	Irrigation				LSD 5%, P value
		Control		Stress		
		0	85	0	85	
POX	Bastan	5.81cd	4.46e	6.53ab	5.89cd	0.521, 0.05
	Pishahang	6.33bc	5.68d	7.04a	6.04bcd	
Chla	Bastan	1.51c	1.87a	1.12d	1.45c	0.106, 0.05
	Pishahang	1.15d	1.68b	0.870e	1.15d	

POX peroxidase specific activity (units mg^{-1} protein), Chla chlorophyll a content (mg g^{-1}FW), Irrigation regimes: Control (40% MAD), Stress (75% MAD). Nitrogen levels: 0 and 85 (mgN kg^{-1} soil).

Table S5. Correlation coefficients among the traits measured in millet cultivars under ambient CO_2 (lower triangle) and elevated CO_2 (upper triangle).

	APX	CAT	POX	Chla	Chlb	Car	LPC	MSI	H_2O_2	MDA	LA	ShN	RDW	ShDW
APX	1	0.97**	0.12	-0.16	-0.12	0.27	0.00	0.01	0.89**	0.19	-0.58	0.58	-0.79	-0.84*
CAT	0.94**	1	0.16	-0.19	-0.19	0.22	0.17	-0.09	0.91**	0.22	-0.61	0.68	-0.81*	-0.86*
POX	-0.01	-0.18	1	-0.92**	-0.93**	-0.84*	0.38	-0.89	0.19	0.66	-0.66	0.10	-0.58	-0.50
Chla	0.12	0.26	-0.93**	1	0.98**	0.85*	-0.40	0.86*	-0.32	-0.76*	0.81*	-0.15	0.57	0.62
Chlb	0.07	0.23	-0.93**	0.99**	1	0.87*	-0.52	0.89**	-0.28	-0.70*	0.81*	-0.23	0.55	0.60
Car	-0.27	-0.09	-0.83*	0.90**	0.91**	1	-0.53	0.92**	0.14	-0.64	0.46	-0.07	0.29	0.19
LPC	0.12	0.05	0.28	-0.44	-0.55	-0.51	1	-0.68	0.21	0.48	-0.32	0.75*	-0.40	-0.27
MSI	-0.23	-0.02	-0.81*	0.84*	0.91**	0.92**	-0.69	1	-0.15	-0.78*	0.54	-0.33	0.58	0.39
H_2O_2	0.79*	0.68	0.56	-0.48	-0.53	-0.74*	0.41	-0.71*	1	0.47	-0.65	0.63	-0.80*	-0.87*
MDA	-0.28	-0.33	0.79*	-0.90**	-0.87*	-0.70*	0.43	-0.70*	0.30	1	-0.48	0.33	-0.68	-0.46
LA	-0.48	-0.36	-0.77*	0.77*	0.79*	0.91**	-0.49	0.86*	-0.88**	-0.61	1	-0.41	0.67	0.92**
ShN	0.53	0.44	0.10	-0.20	-0.32	-0.44	0.88**	-0.60	0.61	0.10	-0.49	1	-0.68	-0.58
RDW	-0.80	-0.67	-0.43	0.30	0.34	0.59	-0.18	0.50	-0.88**	0.03	0.72*	-0.47	1	0.83*
ShDW	-0.66	-0.55	-0.60	0.58	0.64	0.79*	-0.62	0.79*	-0.96**	-0.39	0.90**	-0.74*	0.84*	1

APX ascorbate peroxidase specific activity, CAT catalase specific activity, POX peroxidase specific activity, Chla chlorophyll a content, Chlb chlorophyll b content, Car carotenoids content, LPC leaf prolin content, MSI membrane stability index, H_2O_2 Hydrogen peroxide, MDA Malondialdehyde, LA leaf area, ShN Shoot nitrogen concentration, RDW root dry weight, ShDW Shoot dry weight.

* Significant at the 0.05 probability level.** Significant at the 0.01 probability level.

Table S6. Correlation coefficients among the traits measured in millet cultivars under non-application of nitrogen (lower triangle) and nitrogen application (upper triangle).

	APX	CAT	POX	Chla	Chlb	Car	LPC	MSI	H_2O_2	MDA	LA	ShN	RDW	ShDW
APX	1	0.96**	0.08	-0.17	-0.21	-0.18	0.33	-0.22	0.75*	-0.39	-0.53	0.73*	-0.77*	-0.68
CAT	0.94**	1	0.06	-0.20	-0.25	-0.21	0.31	-0.15	0.68	-0.35	-0.58	0.68	-0.70*	-0.64
POX	0.26	0.15	1	-0.90**	-0.92**	-0.84*	0.79*	-0.93**	0.19	0.58	-0.66	0.60	-0.67	-0.59
Chla	-0.02	0.09	-0.92**	1	0.99**	0.89**	-0.86*	0.89**	-0.32	-0.65	0.83*	-0.70*	0.68	0.71*
Chlb	-0.07	0.05	-0.93**	0.98**	1	0.90**	-0.86*	0.89**	-0.31	-0.56	0.86*	-0.69	0.69	0.70*
Car	-0.08	0.12	-0.88**	0.93**	0.94**	1	-0.92**	0.93**	-0.40	-0.47	0.66	-0.64	0.59	0.65
LPC	0.33	0.24	0.82*	-0.87*	-0.88**	-0.77*	1	-0.92**	0.67	0.53	-0.74*	0.84*	-0.73*	-0.86*
MSI	-0.34	-0.18	-0.92**	0.93**	0.92**	0.90**	-0.93**	1	-0.45	-0.50	0.66	-0.73*	0.72*	0.72*
H_2O_2	0.89**	0.84*	0.52	-0.33	-0.37	-0.29	0.67	-0.61	1	0.01	-0.53	0.86*	-0.70*	-0.87*
MDA	0.02	0.00	0.80*	-0.88**	-0.83*	-0.69	0.88**	-0.84*	0.44	1	-0.31	0.28	-0.16	-0.31
LA	-0.54	-0.54	-0.77*	0.70*	0.67	0.52	-0.86*	0.82*	-0.83*	-0.83*	1	-0.81*	0.79*	0.84*
ShN	0.77*	0.67	0.73*	-0.62	-0.64	-0.57	0.82*	-0.83*	0.91**	0.59	-0.90**	1	-0.95**	-0.98**
RDW	-0.73	-0.70*	-0.69	0.51	0.53	0.46	-0.56	0.64	-0.73*	-0.37	0.74*	-0.86*	1	0.90**
ShDW	-0.85*	-0.81*	-0.59	0.41	0.42	0.34	-0.70*	0.67	-0.98**	-0.53	0.89**	-0.93**	0.76*	1

APX ascorbate peroxidase specific activity, CAT catalase specific activity, POX peroxidase specific activity, Chla chlorophyll a content, Chlb chlorophyll b content, Car carotenoids content, LPC leaf prolin content, MSI membrane stability index, H_2O_2 Hydrogen peroxide, MDA Malondialdehyde, LA leaf area, ShN Shoot nitrogen concentration, RDW root dry weight, ShDW Shoot dry weight.

* Significant at the 0.05 probability level.** Significant at the 0.01 probability level.

Table S7. Correlation coefficients among the traits measured in Bastan (lower triangle) and Pishahang (upper triangle).

	APX	CAT	POX	Chla	Chlb	Car	LPC	MSI	H_2O_2	MDA	LA	ShN	RDW	ShDW
APX	1	0.91**	0.86*	-0.91**	-0.88**	-0.90**	0.61	-0.95**	0.95**	0.94**	-0.79*	0.64	-0.81*	-0.94**
CAT	0.87**	1	0.63	-0.74*	-0.75*	-0.75*	0.82*	-0.86*	0.97**	0.87*	-0.67	0.75*	-0.65	-0.89**
POX	0.87*	0.94**	1	-0.89**	-0.85*	-0.75*	0.40	-0.92**	0.71*	0.89**	-0.74*	0.57	-0.92**	-0.83*
Chla	-0.84*	-0.94**	-0.96**	1	0.99**	0.89**	-0.48	0.87*	-0.76*	-0.80*	0.95**	-0.59	0.86*	0.93**
Chlb	-0.83*	-0.96**	-0.96**	0.97**	1	0.83*	-0.53	0.84*	-0.74*	-0.77*	0.97**	-0.64	0.86*	0.92**
Car	-0.94**	-0.94**	-0.91**	0.91**	0.93**	1	-0.38	0.79*	-0.83*	-0.76*	0.79*	-0.42	0.68	0.86*
LPC	0.64	0.54	0.47	-0.47	-0.58	-0.72*	1	-0.69	0.73*	0.66	-0.45	0.95**	-0.53	-0.74*
MSI	-0.94**	-0.85*	-0.87*	0.87*	0.87**	0.97**	-0.78*	1	-0.90**	-0.98**	0.73*	-0.77*	0.89**	0.93**
H_2O_2	0.88**	0.62	0.77*	-0.69	-0.63	-0.75*	0.43	-0.83*	1	0.93**	-0.63	0.69	-0.68	-0.89**
MDA	0.49	0.41	0.50	-0.58	-0.43	-0.53	0.13	-0.55	0.59	1	-0.65	0.70*	-0.81*	-0.89**
LA	-0.66	-0.83	-0.90**	0.92**	0.95**	0.77*	-0.46	0.74*	-0.53	-0.34	1	-0.55	0.73*	0.86*
ShN	0.73*	0.60	0.52	-0.51	-0.59	-0.78*	0.98**	-0.83*	0.53	0.23	-0.42	1	-0.71*	-0.79*
RDW	-0.88**	-0.92**	-0.92**	0.89**	0.88**	0.95**	-0.60	0.90**	-0.74*	-0.49	0.74*	-0.68	1	0.82*
ShDW	-0.87**	-0.77*	-0.90**	0.86*	0.85*	0.86*	-0.59	0.92**	-0.90**	-0.49	0.81*	-0.63	0.85*	1

APX ascorbate peroxidase specific activity, CAT catalase specific activity, POX peroxidase specific activity, Chla chlorophyll a content, Chlb chlorophyll b content, Car carotenoids content, LPC leaf prolin content, MSI membrane stability index, H_2O_2 Hydrogen peroxide, MDA Malondialdehyde, LA leaf area, ShN Shoot nitrogen concentration, RDW root dry weight, ShDW Shoot dry weight.

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