

Physiological and Biochemical Characterisation of *Brassica juncea* (L. Czern and Coss) inbred lines for drought tolerance



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India has 18% of the world's population but has only 4% of total usable water resources (Ministry of Water Resources, Govt of India).

Agriculture sector is the biggest user of water in India followed by domestic sector and industrial sector.

Of 150 administrative blocks of Indian Punjab, 78 % are extreme dark zones (over exploited) in 2023



Introduction

- Oilseed Brassicas are cultivated on marginal lands of varying soil texture under rain fed and dry farming conditions and often prone to moisture stress during reproductive phase.
- The projected temperature increase and erratic rains due to global warming are likely to further limit water availability and adversely affect production of Brassica oilseeds in the region.
- Phenotyping newly developed germplasm for identified physiological and biochemical traits associated with water stress tolerance will aid to develop drought tolerant cultivars.

Objectives

- To study the effect of moisture stress and irrigated regime on identified physiological and biochemical traits of *Brassica juncea* genotypes.
- Characterization of genotypes for physiological responses under moisture stress and irrigated regime.
- Explore the selection possibility of stress indices on the physiological responses.

Material and Methods

Laboratory conditions : : A panel of 82 inbred lines of *Brassica juncea* (released varieties, natural germplasm, derived lines, exotic lines, introgression material and zero erucic acid lines) were tested for germination traits .

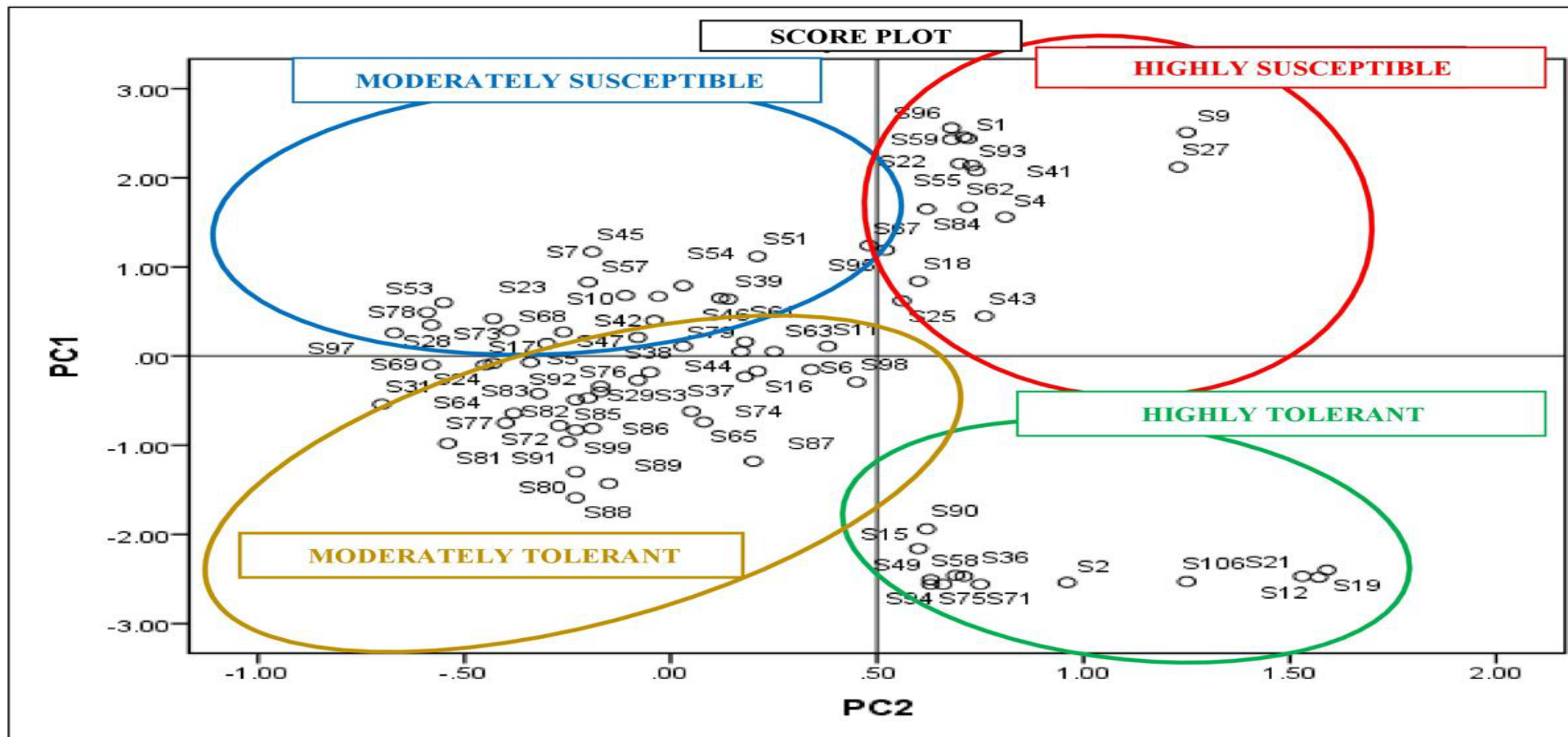
- Treatments: Control as distilled water
- Six osmotic potentials using PEG 6000 (-0.20, -0.40, -0.60, -0.80, -1.00, and -1.20 MPa).

Field trials : 30 lines from lab studies were selected and evaluated under moisture stress and irrigated module

Soil type: Loamy sand (*Typic ustochrept*)

Treatments	Water equivalence (mm)	
	2018-19	2019-20
No added moisture (water stress/Rainfed)	140.8	215.6
Normal moisture (Irrigated)	260.8	335.6

Observations	Instrument/Method
<p>➤ Germination traits Germination percentage Mean germination time (days), Speed of germination (days) Root length and Shoot length, Fresh and dry weight root and Shoot & Seedling vigour indices</p>	<p>Abdul Baki and Anderson (1973)</p>
<p>❖ Physiological traits SPAD</p>	<p>SPAD 502 Plus, Minolta</p>
<p>❖ Relative water content</p>	<p>Cornic 1994</p>
<p>❖ Membrane stability</p>	<p>Premchandra <i>et al</i> (1990)</p>
<p>❖ Total chlorophyll content</p>	<p>Hiscox and Israelstam (1979)</p>
<p>➤ Antioxidative Enzymes ❖ Superoxide dismutase ❖ Peroxidase ❖ Catalase</p>	<p>Marklund and Marklund 1974 (Shannon <i>et al</i> 1966) (Chance and Maehley 1955)</p>
<p>➤ Biochemical constituents</p> <ul style="list-style-type: none"> ▪ α-Tocopherols ▪ Ascorbic acid ▪ Total sugar ▪ Proline 	<p>Emmerie-Engel reaction (Rosenberg 1992) Roe and Keuther (1943) Nelson 1944) Bates 1973)</p>
<p>❖ Growth, yield and yield attributes at maturity</p>	
<p>❖ Drought susceptibility index (DSI): $(1-(Y_s/Y_p))/(1-Y_s / Y_p)$, ❖ Yield stability index (YSI) = Y_s/Y_p where Y_s and Y_p yield under stress and non-stress for each cultivar Y_s and Y_p yield mean in stress and conditions for all cultivars</p>	<p>Fischer and Maurer [1978] Bousslama and Schapaugh [1984]</p>



Categorization of germplasm through principal component analysis based on studied germination traits

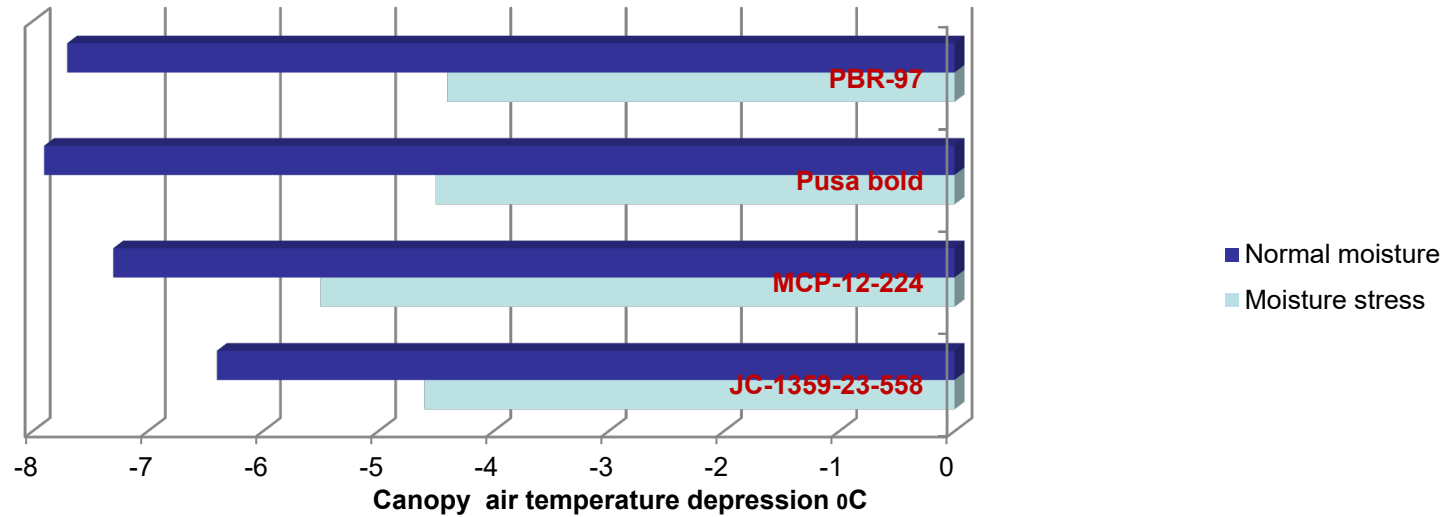
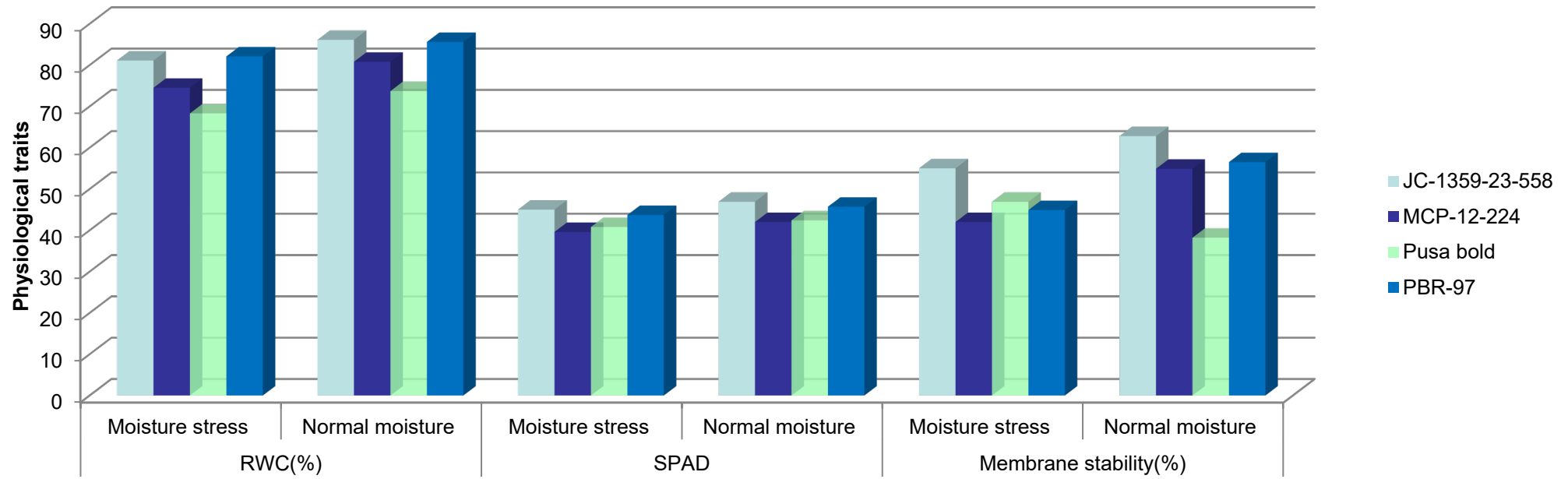
Physiological Traits	No added moisture	Normal moisture
Relative water content(%)	60.5±0.7(MCP-12-60) to 82.1±0.6 (PBR-97)	67.4±4.2 (MCP-12-60) to 86.1±1.8(JC-1359-23-558)
Mean	71.3±0.9	77.4±1.0
SPAD	34.9±0.2(JM-06-010) to 45.3±0.9 (EC564649)	38.1±0.3 (JM-06022) to 48.8±0.8 (EC-564649)
Mean	40.4±0.4	43.3± 0.3
Total Chlorophyll content (mg g⁻¹FW)	1.27±0.1 (Pusa bold) to 2.14±0.03 (JC-1359-23-55)	1.56±0.03 (Pusa bold) to 2.46±0.04(JC-1359-23-558)
Mean	1.55±0.03	2.00±0.04
Membrane stability (%)	21.9±0.7 (JM-06022) to 55.0±0.6(JC-1359-23-558)	36.6±1.4 (JT-152-2) to 62.8±1.0 (JC-1359-23-558)
Mean	27.8±0.8	49.8±1.1
Canopy temperature depression (°C)	-1.3±0.5(JM-06-010) to -5.5±0.2 (MCP-12-224)	-4.3±1.0 (JM-06022) to -7.9±0.5 (Pusa bold)
Mean	-6.5±0.5	-3.6±0.2

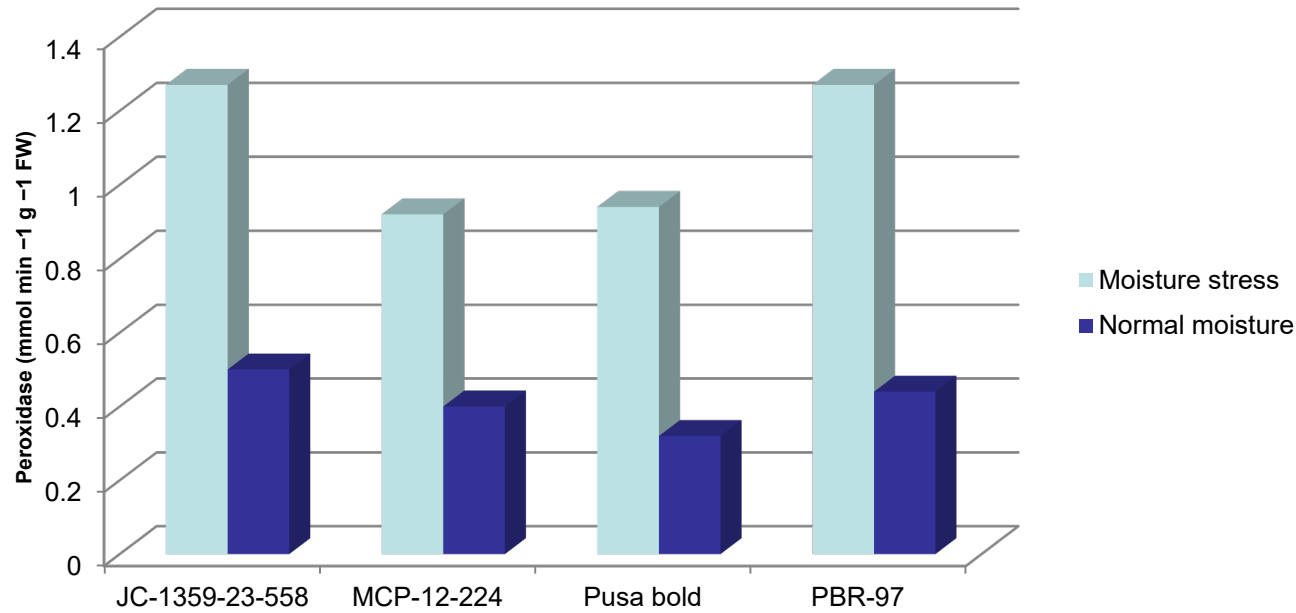
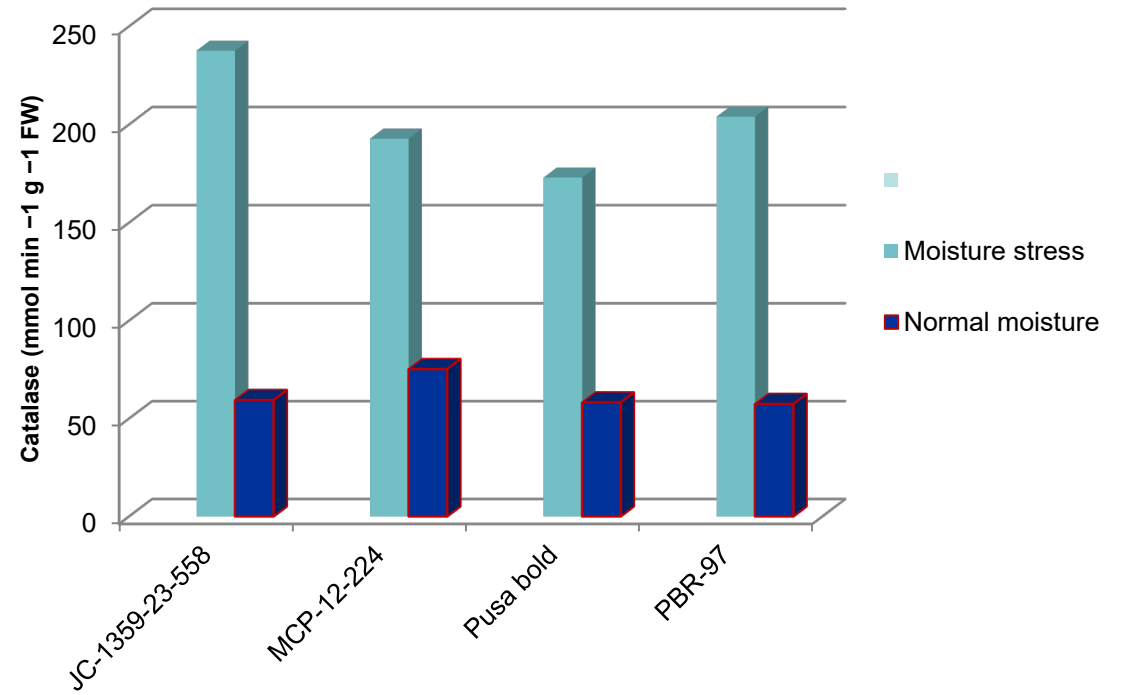
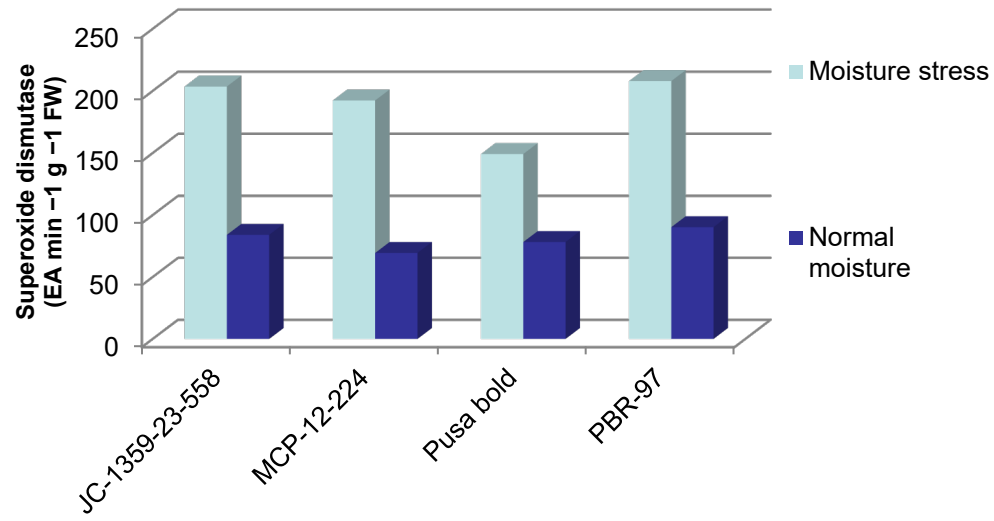
Biochemical Traits	No added moisture	Normal moisture
	Ranges	
Superoxide dismutase (EA min ⁻¹ g ⁻¹ FW)	80.7±0.6 (PRG-901) to 203.5±0.9 (JC-1359-23-558)	59.4±0.3 (PRG-901) to 102.0±0.8 (EC-564647)
Mean	141.1±0.8	80.6±0.7
Catalase (mmol min ⁻¹ g ⁻¹ FW)	124.7±0.9 (CSR-95-7) to 237.6±1.8 (JC-1359-23-558)	45.0±0.9 (CSR-95-7) to 89.5±0.6 (MC-633)
Mean	169.8±1.2	70.4±0.9
Peroxidase (mmol min ⁻¹ g ⁻¹ FW)	0.64±0.01 (MCN-12-45) to 1.31±0.04 (HUSM-1050)	0.32±0.06 (PBR-97) to 0.86±0.02 (JM-06022)
Mean	1.02±0.04	0.54±0.03
Ascorbic acid (mg g ⁻¹ DW)	1.37±0.07 (JM-06022) to 3.27±0.04 (MCP-12-224)	0.72±0.06 (JM-06022) to 1.83±0.04 (JT-152-2)
Mean	2.81±0.07	1.32±0.05
Total sugars (mg g ⁻¹ DW)	59.4±0.6 (PRG-901) to 92.1±0.5 (CSR-95-7)	54.2±0.5 (PRG-901) to 71.9±0.7 (ELM-123)
Mean	75.2±0.6	63.8±0.6
Proline (mg g ⁻¹ DW)	24.3±0.3 (JM-06-010) to 39.0±0.4 (JC-1359-23-558)	19.3±0.5 (JT-152-2) to 30.1±0.7 (RH0725)
Mean	31.0±0.5	23.8±0.6

Growth parameters	No added moisture		Normal moisture	
	Range	Means	Range	Means
Plant height (cm)	102.0 (JM-06-010) to 188.6 (EC-564647)	158.9	120.6 (JM-06-010) to 205.5 (EC-564647)	179.3
Length of main raceme (cm)	35.7 (JM-06-010) to 74.8 (PBR-97)	54.7	44.0 (JM-06-010) to 81.1 (PBR-97)	64.3
Primary branches	2.8 (JM-06022) to 8.3 (JC-1359-23-558)	4.8	3.6 (JM-06022) to 9.4 (JC-1359-23-558)	5.9
Secondary branches	5.2 (JM-06022) to 17.7 (JC-1359-23-558)	10.0	7.8 (JM-06022) to 20.1 (JC-1359-23-558)	13.0
Yield components				
Main shoot siliquae	23.6 (PRG-901) to 49.1 (JC-1359-23-558)	35.9	30.4 (JT-152-2) to 54.3 (JC-1359-23-558)	42.8
Total siliquae plant⁻¹	105.7 (PRG-901) to 375.1 (JC-1359-23-558)	206.3	177.0 (PRG-901) to 406.0 (JC-1359-23-558)	264.1
Siliqua length (cm)	2.5 (JM-06022) to 5.2 (Pusa bold)	4.2	3.5 (JM-06022) to 5.7 (MCP-12-224)	4.9
Seeds siliqua⁻¹	6.2 (JM-06022) to 12.3 (JC-1359-23-558)	10.0	7.7 (JM-06022) to 13.5 (EC-564647)	11.7
Seed yield (kg ha⁻¹)	450.0 (JM-06022) to 2387.5 (JC-1359-23-558)	1107.6	706.9 (JM-06022) to 2806.9 (EC-564647)	1646.1

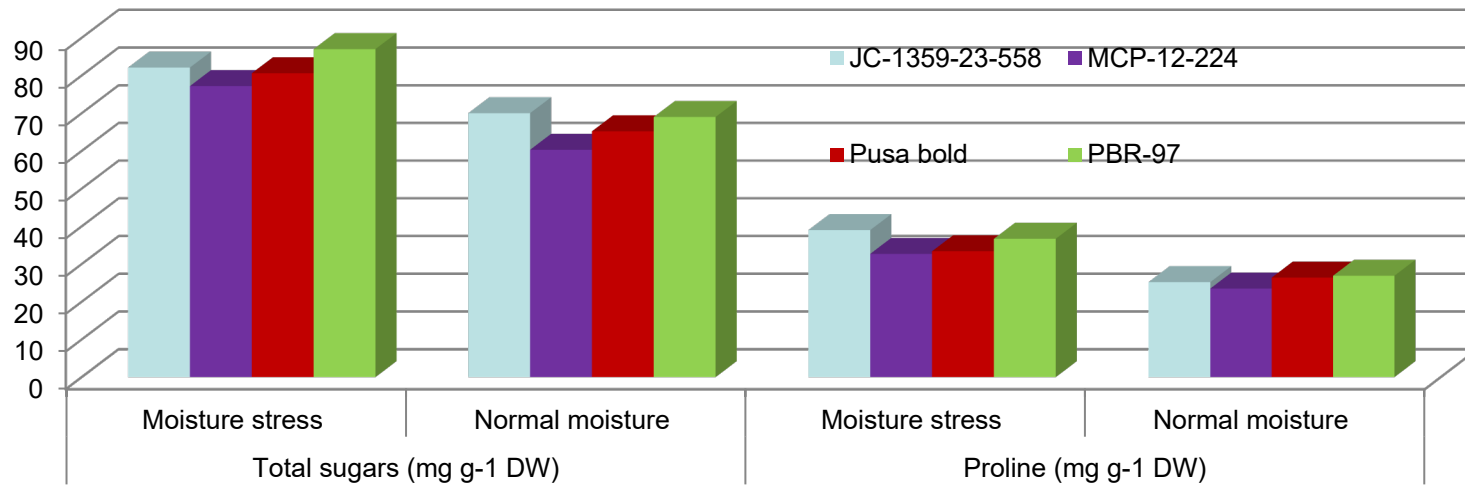
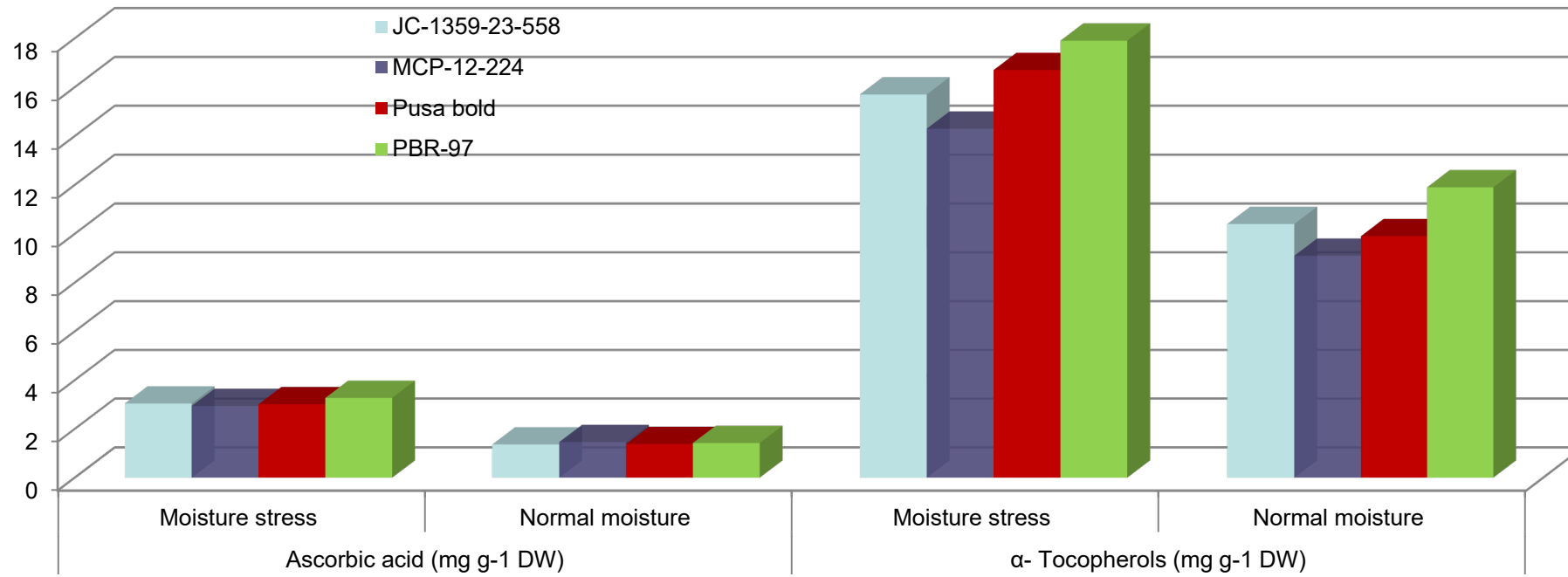
Over the years

JC-1359-23-558, MCP-12-224 and Pusa bold were higher yielders
as compared to PBR-97





Upregulation of antioxidative enzymes



Strains/varieties	Stress indices			Seed yield (Red %)	Rating
	Drought susceptibility index (DSI)	Drought tolerance index (DTI)	Yield stability index (YSI)		
JC-1359-23-558	0.16	2.46	0.85	≤20%	Tolerant
MCP-12-2-24	0.63	1.90	0.80		
Pusa Bold	0.63	1.75	0.80		
PBR97	0.50	2.13	0.84		
ELM151	0.88	0.40	0.72	≤30%	Moderately tolerant
RH725	0.72	0.31	0.80		
PLM4	0.84	0.56	0.73		
NRCHB101	0.69	1.28	0.78		
MCN-12-45	0.84	0.53	0.73		

Conclusions

- ❑ Moisture deficit reduced productivity but there existed genotypic variation for moisture tolerance .
- ❑ Upregulation of antioxidative enzymes and accumulation of proline in water stressed plants imparts drought tolerance.
- ❑ Drought susceptibility index (DSI), Drought tolerance index (DTI) as well as Yield stability index (YSI) may be used to categorize germplasm for tolerance to moisture stress.

Genotypes showing lower DSI, higher DTI and YSI indicate higher moisture stress tolerance.