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Full Length Article

Terminal Drought-priming Improves the Drought Tolerance in *Desi* and *Kabuli* Chickpea

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Abstract

Chickpea productivity, throughout the world, is being threatened by water deficit. In this study, the influence of terminal drought-priming on the performance of *desi* and *kabuli* chickpea types was evaluated under drought and well-watered conditions. During first season, chickpea plants were grown under well-watered conditions (75% water holding capacity). At flower initiation, drought was imposed in half of the pots by maintaining soil moisture at 50% water holding capacity while remaining half continued to grow under well-watered conditions. Terminal drought stress significantly affected the seed composition of both chickpea types as indicated by increase in total proteins (10%), zinc (9.5%), potassium (3.2–0.9%), calcium (2.5–1.3%), and total soluble phenolics (4–57%) than the plants raised under well-watered conditions. During second growing season, chickpea seeds collected from well-watered and droughted source were grown in soil filled pots under well-watered and drought conditions maintaining soil moisture at 75% and 50% water holding capacity, respectively. Drought suppressed the stand establishment, seedling growth, total chlorophyll contents, rate of photosynthesis, PSII efficiency, α-amylase activity, sugar metabolism, and trehalose contents of both chickpea types. Chickpea types also differed in their response to drought; *kabuli* chickpea type was more affected by drought than the *desi* type. The *desi* chickpea type had better stand establishment and growth than the *kabuli* chickpea type. However, under drought, terminal drought-priming improved the performance of both types of chickpea; nonetheless the improvement was more pronounced in *desi* chickpea types. Terminal drought-priming stimulated the build-up of trehalose, proline and total phenolics, and improved the germination metabolism, which assisted improve drought tolerance in *desi* chickpea. In conclusion, changes in seed composition induced by drought-priming improved drought tolerance in chickpea owing to better germination and carbon assimilation, and more accumulation of trehalose, free proline and total soluble phenolics.

Keywords: Drought priming; Proline; Phenolics; PSII; Carbon assimilation

Introduction

Drought is one of the principal factors, which hinder the growth and development of chickpea. Drought stress disrupts the assimilation of carbon and other minerals, which cause decrease in plant growth and productivity (Farooq *et al.*, 2009). Drought suppresses the photosynthesis owing to decrease in the size of photosynthetic machinery (Wahid and Rasul, 2005; Chaves *et al.*, 2011; Zlatev and Lidon, 2012), CO_2 influx and activities of carboxylation enzymes, (Awasthi *et al.*, 2014), chlorophyll degradation and oxidative damages to the thylakoid membrane (Tas and Tas, 2007; Farooq *et al.*, 2009). Under drought, the abscission and senescence increases, and formation of new leaves is decreased (Karamanos, 1980) causing decrease in the size of photosynthetic machinery (Farooq *et al*., 2017a).

Reduction in influx of CO_2 into the leaves under drought stress is the early response of plants due to closing of stomata to decrease the transpiration water loss (Awasthi *et al.*, 2014). Upon sensing the water deficit in the rhizosphere, plants tend to increase the apoplastic abscissic acid concentration through chemical signaling by roots, which causes stomatal closure (Liu *et al.*, 2005). Drought-induced decease in CO_2 influx (Farooq *et al.*, 2009) and the degradation of chlorophyll may cause disturbance to the photosynthetic machinery (Tas and Tas, 2007) resulting in dilation of thylakoid membrane and destabilization of protein complexes (Farooq *et al.*, 2009). This leads to

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increase in generation of reactive oxygen species (ROS). The major types of ROS include superoxide (O_2^-), hydroxyl radicle (OH) and hydrogen peroxide (H_2O_2), etc., (McCord, 2000; Ruelland *et al.,* 2009), which may cause oxidative damages to biological membranes, and destablize the vital cellular molecules including nucleic acids, DNA, proteins, lipids and may even lead to cell lysis (Foyer, 2005).

The reproductive phase of chickpea growth is more sensitive to the drought than the vegetative phase. Any episode of drought during the reproductive phase (terminal drought) heavily tax the grain yield in chickpea due to flower abortion, impaired pollen viability and stigma/style functionality (Fang *et al.,* 2010; Pang *et al.,* 2017), decrease in seed set, pod abscission, and reduced seed size (Davies *et al.,* 1999; Pang *et al.,* 2017).

Priming (pre-exposure of plants to stress), permits plants to become more tolerant to stresses (abiotic or biotic) later (Bruce *et al.,* 2007). The pre-exposure to drought enhances the elasticity of plants to cope with re-occurring of same stress (Ding *et al.,* 2012). During the developmental cascades, plants retrieve preceding stress to a succeeding one (Molinier *et al.,* 2006; Wang *et al.,* 2014). The stress-primed plants facing an early drought stress event had better photo-protection and a higher biomass than non-primed in second drought episode (Walter *et al.,* 2011). Drought-primed plants exhibited improved photosynthesis rate, leaf water status, ascorbate peroxidase and lower membranes damage than un-primed plants at later stages of drought stress (Wang *et al.,* 2014). One type of stress faced by the plants improves its ability to tolerate the subsequent stresses during the next generation (Cuk *et al.,* 2010) by retaining trans-generational stress memory (Walter *et al.,* 2013). These stresses involve modifications in proteins, compatible solutes (Joyce *et al.,* 2003) and up-regulation of antioxidative enzymes in next generation (Cuk *et al.,* 2010).

Increase in the accumulation of sugars, specifically trehalose, provides protection against abiotic stresses including drought. Trehaloses help in stabilizing the biological membranes by scavenging the free radicles through detoxifying ROS with binding at polar region of membranes of phosphate and proteins hydroxyl group (Benaroudj *et al.,* 2001), shielding protein molecules from denaturation (Benaroudj *et al.,* 2001; Elbein *et al.,* 2003) and maintaining the carbon assimilation (Farooq *et al.,* 2017b, 2018).

In our recent work, we reported better salt tolerance in wheat, during next generation, by drought-priming owing to the osmolytes accumulation and improved water relations (Tabassum *et al.,* 2017). However, to the best of our knowledge, no information is available on the effect of terminal drought-priming on drought tolerance of chickpea. This study was, therefore, conducted to evaluate the influence of drought-priming on drought tolerance in *kabuli* and *desi* types of chickpea.

Material and Methods

Plant Material

Seeds of *kabuli and desi* chickpea cultivars Noor-2013 and Bitall-2016, respectively, used in this study, were received from the Pulses Research Institute, Ayub Agricultural Research Institute, Faisalabad, Pakistan.

Year 1

Drought-Priming

Seeds of both chickpea types were planted, (six in each pot), in soil filled earthen pots (10 kg) with 75% water holding capacity (well-watered conditions) and the pots were kept in a net-house under natural conditions. Upon the competition of seedling emergence, plants were thinned for maintaining three plants in each pot. At onset of flowering, half of pots continued to grow under well-watered conditions while drought (50% water holding capacity) was imposed in rest half. At harvest maturity, plants were harvested and threshed to separate the seeds.

Seed Composition Analysis

The chickpea seeds, from both drought stressed and well-watered sources, were milled to make powder. Total soluble proteins were estimated by Bradford method (Bradford, 1976). To determinate the total soluble phenolics, the flour were overnight soaked in 80% acetone then Folin-Ciocalteu reagent and Na_2CO_3 solution were added. Total soluble phenolics were estimated as gallic acid equivalent (GAE) (Singleton and Rossi, 1965). Seed phosphorus (P), potassium (K), calcium (Ca^{+2}) and zinc (Zn^{+2}) were analyzed by inductively coupled plasma (ICP) atomic absorption spectrometer (OES; Shelton, CT, USA).

Year 2

Experimental Details

Seeds of both chickpea types, from droughted and well-watered sources, were sown (ten seeds in each pot) in 5 kg soil-filled pots. The pots were maintained at soil moisture of 75% and 50% water holding capacity for well-watered and drought stress treatments, respectively. After completion of seedling emergence, plants were thinned to maintain three plants in each pot. These pots were placed in a climate chamber having temperature (day/night) (18/15°C) with a photosynthetically active photon flux with a photoperiod (light/dark) (of 16/8 h) having density (350 mM m^{-2} s^{-1}). Completely randomized design in factorial arrangement was used in the study with six replications. One replication was consisted of five pots each pot having three plants. The experiment was harvested, to record different observations, four weeks after the sowing.

Stand Establishment

The experiment was visited daily, and number of seedlings emerged were counted daily until a constant count was achieved. Emrgence index (EI) and the coefficient of uniformity of emergence (CUE) were estimated following Association of Official Seed Analysts (1983) and Bewley and Black (1985), respectively. The final emergence percentage (FEP) was recorded as ratio of emerged seedlings to the number of planted seeds and was expressed in percentage.

α-Amylase Activity and Suagrs

Two days after sowing five germinating seedlings were collected and crushed to record the activity of α-amylase. The extract was added in phosphate buffer of pH 0.7 and the mixture was incubated at 4°C for 24 h and was vortexd. The supernatant was used to determine activity of α-amylase using dinitrosalicylic acid (DNS) method (Bernfeld, 1955) with slight modification following Lee and Kim (2000). To determine the total soluble sugars, distilled water was added in ground seed samples and the mixture was incubated for 24 h at 25°C (Lee and Kim, 2000). The mixture was then filtered with Whatman No. 42 paper. Total soluble sugars were determined, from the filtrate, following the phenol-sulfuric acid method (DuBois *et al.*, 1956). The reducing sugars were estimated, from the same filtrate, using glucose as standard as described by Miller (1959) whereas sucrose contents were estimated as described by Stitt *et al.* (1989). Trehalose contents were estimated by measuring the glucose produced by hydrolysis of trehalose using a glucose oxidase-peroxidase kit (Spainreact) following Čižmárilk *et al.* (2004).

Total Chlorophyll Contents, Leaf Photosynthesis and PSII Efficiency

These observations were recorded one day before the final harvest. Total leaf chlorophyll contents were determined from the fully mature leaves following the method described by Arnon (1949). Rate of leaf photosynthesis was measured from the youngest fully mature leaf with a portable photosynthesis system (LI-6400; LiCor, Inc., NE, USA). The maximum efficiency of photosystem II (PSII), in chickpea leaves, was determined with Plant Efficiency Analyzer (Hansatech, Norfolk, UK) at excitation light energy of 3000 µmol m^{-1} s^{-1}.

Leaf Free Proline, Total Soluble Phenolics and Lipid Peroxidation

For the determination of leaf free proline, chickpea leaf samples were homogenized in sulphosalicylic acid, the filtrate was mixed with glacial acid and ninhydrin in equal proportion. The mixture was incubated in a water bath at 100°C for 60 min. The mixture was then vortexed and put into an ice bath; toluene was added into the mixture and the chromophore containing proline was aspirated. The leaf free proline was determined as described by Bates *et al.* (1973). Lipid peroxidation was estimated by measuring the malondialdehyde (MDA) contents. Leaf samples were homogenized in 0.1% trichloroacetic acid solution and the MDA contents were determined as described by Heath and Packer (1968). Leaf total soluble phenolics were estimated as described above for seed composition analysis.

Plant Growth

Leaf area was noted with a leaf area meter (Delta-T Devices, Cambridge, UK) using the leaves detached from the stem. The same leaves were then dried in an electric oven to record the leaf dry weight. The specific leaf area was recorded as the ratio of leaf area to leaf dry weight. Dry weight of all above ground plant material was recorded as seedling dry weight.

Leaf Mineral Analysis

For the determination of leaf minerals, plant leaves were dried and ground to powder. Total leaf nitrogen (N) was estimated with CHN-1000 analyzer by combustion analysis (LECO Corp., St. Joseph, MO). Leaf P, K and Ca^{+2} were estimated as described above for seed composition analysis.

Statistical Analysis

The experimental data were statistically analysed by analysis of variance technique (Steel *et al.*, 1997) using statistical software Co-Stat (CoHort, Berkeley, CA, USA). Treatment means were separated with least significant difference (LSD) test at probability level of 5%.

Results

Seed Composition

Terminal drought-priming stress significantly affected the seed composition of both chickpea types. Terminal drought stress increased total proteins (10%), Zn (9.5%), K (3.2 and 0.9%), Ca^{+2} (2.5 and 1.3%), and total soluble phenolics (41 and 57%) in *desi* and *kabuli* chickpea types, respectively, compared with well-watered control (Table 1).

Stand Establishment and Growth

Drought stress significantly suppressed the stand establishment and seedling growth of chickpea. However,

Table 1: Effect of terminal drought on seed composition of *desi* and *kabuli* chickpea types

Characteristic	*Desi* chickpea		*Kabuli* chickpea	
	Well-watered	Terminal drought	Well-watered	Terminal drought
Total proteins (g 100 g^{-1})	22.25c	24.47a	21.19d	23.39b
Potassium (mg 100 g^{-1})	625d	645c	725b	732a
Phosphorus (mg 100 g^{-1})	244b	245b	255a	254a
Calcium (mg 100 g^{-1})	238b	244a	232d	235c
Zinc (mg 100 g^{-1})	4.91b	5.38a	3.65d	4.00c
Total soluble phenolics (µg g^{-1})	0.43d	1.47a	0.47c	1.21b

Any two means, for a parameter, not sharing a letter in common differ significantly at $p \leq 0.05$

Table 2: Effect of drought-priming on coefficient of uniformity of emergence (CUE), emergence index, seedling dry weight and specific leaf area (SLA) of *desi* and *kabuli* chickpea types under well-watered (WW) and drought stress (DS) conditions

Chickpea types	Seed source	CUE		Emergence index		Final emergence (%)		Seedling dry weight (g)		SLA (cm^2 g^{-1} leaf DM)	
		WW	DS	WW	DS	WW	DS	WW	DS	WW	DS
Desi	Well-watered	0.67b	0.37d	68.75a	43.21c	97a	75c	7.45c	5.32e	20.61a	10.16f
	Drought-priming	0.68b	0.47c	68.95a	49.55b	98a	83b	7.42c	6.19d	18.65b	13.15e
Kabuli	Well-watered	0.71a	0.23f	68.78a	38.44d	99a	68e	8.74a	4.71f	16.85c	9.32g
	Drought-priming	0.72a	0.34e	69.61a	42.39c	98a	72d	8.15b	5.15e	15.44d	10.51f

Any two means, for a parameter, not sharing a letter in common differ significantly at $p \leq 0.05$

both tested chickpea types significantly differed for response to drought stress from both seed sources (drought-priming and well-watered) (Table 2).

There was no difference between drought-priming and well-watered seed sources for CUE, emergence index, and final emergence percentage in both chickpea types under well-watered conditions. Under well-watered conditions, although there was no difference in seedling dry weight of *desi* chickpea type from either seed source, however, drought-priming caused reduction in seedling dry weight of *kabuli* chickpea type and SLA in both *desi* and *kabuli* chickpea types (Table 2). However, better seedling emergence and growth were recorded from drought-primed seeds than the well-watered seed source in both chickpea types under drought stress (Table 2).

α-Amylase Activity and Sugars Metabolism

Both tested chickpea types significantly differed for activity of α-amylase, reducing sugars, total soluble sugars, sucrose, and trehalose contents under well-watered and drought conditions irrespective of the seed source (Table 3). However, more α-amylase activity, reducing sugars, total soluble sugars, sucrose and trehalose contents were recorded in *desi* chickpea than the *kabuli* chickpea irrespective of seed source under well-watered conditions (Table 3).

In both tested chickpea types, drought stress significantly reduced the α-amylase activity, total soluble sugars, sucrose, reducing sugars and trehalose contents from both seed sources. However, drought-priming had more activity of α-amylase, reducing sugars, total soluble sugars, sucrose and trehalose contents than the well-watered seed source under drought stress (Table 3).

Total Chlorophyll Contents, Leaf Photosynthesis and PSII Efficiency

The both tested chickpea types significantly differed for total chlorophyll contents, leaf rate of photosynthesis and maximal PSII efficiency from both seed sources under both drought stress and well-watered conditions (Table 4). Under well-watered conditions, drought-priming and well-watered seed sources did not differ for total chlorophyll contents, leaf rate of photosynthesis and maximal PSII efficiency. Drought stress significantly reduced the total chlorophyll contents, leaf rate of photosynthesis and maximal PSII efficiency in both chickpea types from both seed sources (Table 4).

Under drought stress, *kabuli* chickpea had lower total chlorophyll contents, leaf rate of photosynthesis and maximal PSII efficiency, irrespective of seed source, than the *desi* chickpea (Table 4). However, under drought, drought-priming increased the total chlorophyll contents, leaf rate of photosynthesis and maximal PSII efficiency in both chickpea types than the well-watered seed source (Table 4).

Leaf Free Proline, Total Soluble Phenolics and Lipid Peroxidation

Under well-watered conditions, except for total soluble phenolics, the both tested chickpea types significantly differed for leaf free proline and leaf malondialdehyde contents irrespective of seed source (Table 5). However, drought stress caused significant increase in these parameters. Under drought stress, highest leaf malondialdehyde contents were noted in *kabuli* chickpea from well-watered seed source (Table 5). However,

Table 3: Effect of drought-priming on α-amylase activity, total soluble sugars, reducing sugars, sucrose and trehalose of *desi* and *kabuli* chickpea types under well-watered (WW) and drought stress (DS) conditions

Chickpea types	Seed source	α-amylase activity (IU mg^{-1} protein)		Total soluble sugars (mg g^{-1})		Reducing sugars (mg g^{-1})		Sucrose (mg g^{-1})		Trehalose (µg g^{-1})	
		WW	DS	WW	DS	WW	DS	WW	DS	WW	DS
Desi	Well-watered	8.77a	5.54d	10.15a	6.45d	6.53a	4.73d	2.71a	1.89d	44.31a	31.17d
	Drought-priming	8.55a	6.75c	9.94a	7.15c	6.47a	5.21c	2.64a	2.12c	43.42a	35.29c
Kabuli	Well-watered	7.67b	4.25e	8.75b	4.75f	5.84b	3.87e	2.28b	1.61e	39.25b	26.34f
	Drought-priming	7.53b	5.15f	8.51b	5.28e	5.81b	3.35f	2.22b	1.92d	38.75b	28.32e

Any two means, for a parameter, not sharing a letter in common differ significantly at $p \leq 0.05$

Table 4: Effect of drought-priming on leaf CO_2 net assimilation rate, PSII efficiency and total chlorophyll of *desi* and *kabuli* chickpea types under well-watered (WW) and drought stress (DS) conditions

Chickpea types	Seed source	Leaf CO_2 net assimilation rate (µmol s^{-1} m^{-2})		PSII efficiency (F_v/F_m)		Total chlorophyll (mg g^{-1} FW)	
		WW	DS	WW	DS	WW	DS
Desi	Well-watered	10.68a	7.26d	0.71a	0.41d	16.91a	11.16d
	Drought-priming	10.72a	8.44c	0.69a	0.51c	16.82a	12.14c
Kabuli	Well-watered	9.42b	6.96e	0.59b	0.34e	13.56b	9.12f
	Drought-priming	9.48b	7.34d	0.56b	0.39d	13.21b	10.23e

Any two means, for a parameter, not sharing a letter in common differ significantly at $p \leq 0.05$

Table 5: Effect of drought-priming on leaf malondialdehyde (MDA) contents, Total soluble phenolics and free leaf proline of *desi* and *kabuli* chickpea types under well-watered (WW) and drought stress (DS) conditions

Chickpea types	Seed source	Leaf MDA contents (µmol g^{-1} FW)		Total soluble phenolics (µg g^{-1} FW)		Free leaf proline (µmol g^{-1} FW)	
		WW	DS	WW	DS	WW	DS
Desi	Well-watered	11.25f	16.43c	15.51d	22.86b	6.27f	14.14b
	Drought-priming	11.34f	14.44d	15.68d	26.17a	6.29f	17.33a
Kabuli	Well-watered	12.33e	19.76a	15.29d	20.69c	6.93e	12.17c
	Drought-priming	12.49e	17.32b	15.48d	22.24b	6.90e	11.95d

Any two means, for a parameter, not sharing a letter in common differ significantly at $p \leq 0.05$

Table 6: Effect of drought-priming on leaf nitrogen, leaf phosphorus, leaf potassium and leaf calcium of *desi* and *kabuli* chickpea types under well-watered (WW) and drought stress (DS) conditions

Chickpea types	Seed source	Leaf N (mg g^{-1} DM)		Leaf P (mg g^{-1} DM)		Leaf K (mg g^{-1} DM)		Leaf Ca^{+2} (mg g^{-1} DM)	
		WW	DS	WW	DS	WW	DS	WW	DS
Desi	Well-watered	3.45a	2.88d	1.88a	1.73b	1.94a	1.58d	1.00a	0.71c
	Drought-priming	3.43a	2.96c	1.91a	1.75b	1.92a	1.70c	1.01a	0.82b
Kabuli	Well-watered	3.33b	2.56f	1.86a	1.76b	1.82b	1.50e	0.99a	0.61d
	Drought-priming	3.31b	2.72e	1.87a	1.74b	1.81b	1.56d	1.02a	0.63d

Any two means, for a parameter, not sharing a letter in common differ significantly at $p \leq 0.05$

highest free leaf proline contents and total soluble phenolics were recorded in *desi* chickpea from drought-primed seeds, but, the lowest leaf malondialdehyde contents were noted in *desi* chickpea from drought-primed seeds (Table 5).

Leaf Mineral Analysis

Except for Ca^{+2} under well-watered conditions and leaf P under both well-watered and drought conditions, the tested chickpea types significantly differed for leaf mineral contents irrespective of seed source (Table 6). Mineral contents of *kabuli* chickpea were more strongly affected than the *desi* chickpea from well-watered seed source (Table 6). Drought caused significantly decrease in leaf mineral contents in both chickpea types irrespective of seed source, however, the drought-induced decrease was less from drought priming seed.

Discussion

The progeny of drought stressed chickpea types (both *desi* & *kabuli*) performed better than that of well-watered under drought stress, due to decrease in total lipids and increase in total proteins, Zn, K, Ca^{+2} and total soluble phenolics (Table 1). These phenolic compounds help in scavenging the ROS while acting as antioxidants (Weidner *et al.*, 2009). Any stress during grain development phase may change the quality and composition of grains with the accumulation of certain secondary metabolites (Tabassum *et al.*, 2017). This change in seed composition helps the plants to tolerate the

reoccurrence of the same or any other stress (Cuk *et al.,* 2010; Tabassum *et al.,* 2017) during later ontogeny of the plants through increased expression of proteins and compatible solutes (Joyce *et al.,* 2003) as was observed in this study (Tables 3 and 5). Plants maintain trans-generational stress memory in physiological, morphological and metabolic forms (Walter *et al.,* 2013; Tabassum *et al.,* 2017).

Stress-priming improves the accumulation of osmolytes through altered metabolic processes and these metabolites help in stress tolerance (abiotic stresses) during next growing season through revealing the preceding stress memory (Tabassum *et al*., 2017) acting as antioxidants, defense compounds and osmoregulators (Rivas-Ubach *et al.,* 2012). Progeny of the stressed plants store more proline and glycine betaine than non-stressed plants (Tabassum *et al.,* 2017). Plants tolerate abiotic stresses through alterations in gene expression, soluble sugars, proline contents, higher antioxidant, and through by biosynthesis of stress proteins (Sung *et al.,* 2003; Yamada *et al.,* 2007).

During stress-priming, plants attain cross tolerance to successive stresses through improved gene expression for osmolytes and heat shock proteins by buildup of transcription factors (Kibinza *et al.,* 2011; Chen *et al.,* 2012) and during the transcriptional drought memory, RNA polymerase II is involved (Ding *et al.,* 2012). Priming enhances photosynthesis, up-regulate Rubisco activase, and Rubisco while decreases malondialdehyde contents in primed plants under moisture stress (Wang *et al.,* 2014) as was observed in this study (Tables 4–5).

Drought stress caused delayed and erratic germination and seedling emergence, which resulted in poor growth of chickpea plants (Table 2). This may be attributed due to drought-induced decrease in the activity of α-amylase, sugar metabolism, trehalose accumulation (Table 3), total chlorophyll contents, rate of photosynthesis and maximum efficiency of PSII (Table 4), increase in oxidative damages (Table 5), and decrease in uptake of mineral elements (Table 6). Water deficit causes cessation of mitosis, cell elongation and expansion process (Farooq *et al*., 2009). Drought stress suppresses the plant growth through deregulation of elongating cells as water influx from xylem to the elongating cells is disrupted (Nonami, 1998; Farooq *et al.,* 2009).

Sugars are responsible for the regulation of α-amylase gene (Yu *et al.,* 1996) and under drought stress decrease in sugars (Table 3) might impact α-amylase activity as well as trehalose contents (Table 3) as trehalose protects the plants against abiotic stresses via acting as scavenger, stabilizing the cell membranes and ceasing the protein denaturation (Benaroudj *et al.,* 2001). Drought-induced decrease in α-amylase activity strongly impedes the carbohydrate metabolism resulting in reduction in food supply to the developing seedlings, which causes erratic stand establishment and restricts the seedling growth (Farooq *et al*., 2017b). During germination, α-amylase modulates the hydrolysis and mobilization of starch (Fincher, 1989; Farooq *et al*., 2006). These starch metabolites not only act as carbon source for the germinating seedling (Farooq *et al.,* 2017b) but also regulates the water potential during germination and seedling development (Murtaza and Asghar, 2012).

Nutrient uptake (specifically N) is reduced under drought stress, which led to the increased apoplastic abscisic acid concentration with result in stomatal closing due to xylem sap alkalization (Liu *et al.,* 2005). Under the moisture stress the accumulation of malondialdehyde contents (an index of oxidative stress) (Table 5) increases which damages the membranes.

The increase in total sugars, reducing sugars, trehalose contents, total soluble phenolics, free proline and reduction in malondialdehyde contents induced by drought-priming under drought (Tables 3 and 5) helped in improving tolerance to the chickpea plants against drought through maintenance of specific leaf area (Table 2) increasing the total chlorophyll contents, rate of photosynthesis and efficiency of PSII (Table 4), as the accumulation of same in the plants lowers the osmotic potentials of cells, which results in drawing the H_2O into the tissues and cells; hence maintains the turgor, and carbon influx (Subbarao *et al.,* 2000; Farooq *et al.,* 2018), and helps improving the plant growth.

However, enhanced accumulation of trehalose, total soluble phenolics and free leaf proline (Tables 3 and 5), under drought stress protects plants from ROS damage (Farooq *et al.,* 2009; Tabassum *et al.,* 2017), as phenolics contains aromatic ring, which shields from the oxidative damages by scavenging the ROS (Takahama and Oniki, 1997) and thus helps in stabilizing the biological membranes (Taiz *et al.,* 2015). Likewise, increase in the proline accumulation under drought (Table 5) also assist to protect the macromolecules from oxidative damages (Zhu, 2002; Wahid and Close, 2007) and acts as a sink for excessive reductant and a store house of nitrogen and carbon (Zhu, 2002), hence imparts tolerance against numerous stresses (Farooq *et al.,* 2009).

Conclusion

Drought-priming-induced alteration in seed composition, buildup of trehalose contents, total soluble phenolics and free leaf proline improved the chickpea performance under drought stress by modulating the oxidative stress, germination metabolism, carbon assimilation, PSII efficiency and uptake of minerals.

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