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Original Article

Exogenously applied nutrients can improve the chickpea productivity under water stress conditions by modulating the antioxidant enzyme system

Nutrientes aplicados exogenamente melhoram a produtividade do grão-de-bico em condições de estresse hídrico, modulando o sistema de enzimas antioxidantes

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Abstract

Water stress is one of the major factor restricting the growth and development of chickpea plants by inducing various morphological and physiological changes. Therefore, the present research activity was designed to improve the chickpea productivity under water stress conditions by modulating antioxidant enzyme system. Experimental treatments comprised of two chickpea genotypes i.e. Bhakhar 2011 (drought tolerant) and DUSHT (drought sensitive), two water stress levels i.e. water stress at flowering stage and water stress at flowering + pod formation + grain filling stage including well watered (control) and three exogenous application of nutrients i.e. KCl 200 ppm, $MgCl_2$ 50 ppm and $CaCl_2$ 10 mM including distilled water (control). Results indicated that water stress at various growth stages adversely affects the growth, yield and quality attributes of both chickpea cultivars. Exogenous application of nutrients improved the growth, yield and antioxidant enzyme activities of both chickpea genotypes even under water stress conditions. However, superior results were obtained with foliar spray of potassium chloride on Bhakhar 2011 under well-watered conditions. Similarly, foliar spray of potassium chloride on chickpea cultivar Bhakhar 2011 cultivated under stress at flowering + pod formation + grain filling stage produced significantly higher contents of superoxide dismutase, peroxidase and catalase. These results suggests that the application of potassium chloride mitigates the adverse effects of water stress and enhanced tolerance in chickpea mainly due to higher antioxidant enzymes activity, demonstrating the protective measures of plant cells in stress conditions.

Keywords: antioxidant, leaf K^+ contents, catalase, potassium chloride, foliar spray.

Resumo

O estresse hídrico é um dos principais fatores que restringem o crescimento e o desenvolvimento das plantas de grão-de-bico, induzindo várias alterações morfológicas e fisiológicas. Portanto, a presente atividade de pesquisa foi projetada para melhorar a produtividade do grão-de-bico em condições de estresse hídrico, por meio da modulação do sistema de enzimas antioxidantes. Tratamentos experimentais compostos de dois genótipos de grão-de-bico, ou seja, Bhakhar 2011 (tolerante à seca) e DUSHT (sensível à seca), dois níveis de estresse hídrico, ou seja, estresse hídrico na fase de floração e estresse hídrico na floração + formação de vagens + estágio de enchimento de grãos incluindo bem irrigado (controle) e três aplicações exógenas de nutrientes, ou seja, KCl 200 ppm, MgCl2 50 ppm e CaCl2 10 mM, incluindo água destilada (controle). Os resultados indicaram que o estresse hídrico em vários estágios de crescimento afeta negativamente os atributos de crescimento, rendimento e qualidade de ambas as cultivares de grão-de-bico. A aplicação exógena de nutrientes melhorou o crescimento, o rendimento e as atividades das enzimas antioxidantes de ambos os genótipos de grão- de-bico, mesmo em condições de estresse hídrico. No entanto, resultados superiores foram obtidos com pulverização foliar de cloreto de potássio em Bhakhar 2011, em condições bem irrigadas. Da mesma forma, a pulverização foliar de cloreto de potássio na cultivar de grão-de-bico Bhakhar 2011 cultivada sob estresse na fase de floração + formação de vagens + enchimento de grãos produziu teores significativamente maiores de superóxido dismutase, peroxidase e catalase. Esses resultados sugerem que a aplicação de cloreto de potássio atenua os efeitos adversos do estresse hídrico e aumenta a tolerância no grão-de-bico, principalmente em razão de mais atividade de enzimas antioxidantes, demonstrando as medidas protetoras das células vegetais em condições de estresse.

Palavras-chave: antioxidante, conteúdo foliar de K^+, catalase, cloreto de potássio, *spray* foliar.

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1. Introduction

Chickpea (*Cicer arietinum* L.) is a nutritionally important leguminous crop mainly cultivated on marginal lands in arid to semi-arid tropics (Jaleel et al., 2009). It plays an important role in human nutrition sectors in the developing world. It is ranked as a healthy food in many developed countries (Merga and Haji, 2019). Moreover, chickpea is also a crucial source of fodder and also fixes atmospheric nitrogen through symbiotic nitrogen fixation, hence plays a key role in enhancing and sustaining the overall productivity of the cropping systems (Khaitov et al., 2016). However, chickpea crop is facing severe water stress due to the unavailability of water, breaks-in precipitation associated with fewer rains at various growth stages causes the problem of erratic and terminal water stress and experiences up to 50% yield loss (Sinha et al., 2019). Plants experience water stress either when the water supply to roots is interrupted or when the transpiration rate becomes very high. When plants are subjected to water stress conditions, show a lot of morpho-physiological and biochemical changes (Reddy and Raghavendra, 2006).

Plants under water stress conditions are unable to absorb sufficient nutrients from the soil causing nutrient deficiencies and as consequently reduced productivity. Exogenous application of essential nutrients mitigates these deficiencies and improves water stress tolerance by maintaining vital physiological processes. Among various nutrients, exogenous application of potassium stimulates a wide range of enzymatic systems, regulating water use efficiency, protein building, nitrogen uptake, photosynthesis and translocation of photosynthates into sink organs. Similarly, the application of calcium play role in controlling mechanisms, which stimulate the plant to become adopted to adversarial environmental conditions (Upadhyaya et al., 2011). Calcium performs an essential role in the activation of various defense mechanisms that are induced by water stress and Ca^{2+} signaling is mandatory for the acquisition of drought tolerance or resistance (Cousson, 2009). In addition to these nutrients, magnesium is also involved in numerous biochemical and physiological processes. It is the central core of the chlorophyll molecule in plant tissue and plays a role in the light absorbing the complex of chloroplasts and its contribution to photosynthetic fixation of carbon dioxide (Gerendás and Führs, 2013). It also participates in activation of many enzymes, for instance, phosphoenol pyruvate (PEP), glutathionsynthetase, carboxylase, kinases and phosphatases etc., (Bose et al., 2011). Magnesium is a crucial element for plant growth and development and plays an important role in plant defense mechanisms under water stress conditions (Cakmak and Yazici, 2010). A few studies were conducted to explore the role of calcium chloride, potassium chloride and magnesium chloride on the chickpea productivity under water stress conditions. It is hypothesized that foliar spray of calcium chloride, potassium chloride and magnesium chloride may positively affect the morpho-physiological processes of chickpea and enhanced the growth and productivity under water stress conditions. Keeping these facts in view, the present investigation was therefore undertaken to study the role of nutrients to mitigate the adverse effect of water stress on chickpea genotypes by modulating the antioxidant enzymes system.

2. Materials and Methods

2.1. Experimental detail

The present research activity was designed to improve the chickpea productivity under water stress conditions by modulating antioxidant enzyme system under agro-ecological conditions of Arid Zone Research Institute, Bahawalpur (29.3871 °N, 71.653 °E) and Cholistan farm near Derawer (28.19°N, 71.80°E) of Southern Punjab, Pakistan during Rabi 2014-15. Experimental treatments comprised of two chickpea genotypes i.e. Bhakhar 2011 (drought tolerant) and DUSHT (drought sensitive), two water stress levels i.e. water stress at flowering stage and water stress at flowering + pod formation + grain filling stage including well watered (control) and three exogenous application of nutrients i.e. KCl 200 ppm (Bardhan et al., 2007), $MgCl_2$ 50 ppm (Thalooth et al., 2006) and $CaCl_2$ 10 mM (Xu et al., 2013) including distilled water (control). Foliar treatments were applied on 9th January and 12th February 2015 by using a hand sprayer. Field experiment was laid out in a Randomized Complete Block Design (RCBD) with factorial arrangement having three repeats.

2.2. Soil analysis

Prior to sowing, composite soil samples were collected from experimental areas at the depth of 0-30 cm and examined for physico-chemical features. The soil texture of experimental areas were loam and clay loam, having pH 8.2 and 7.9, organic matter 0.77 and 0.93%, EC 2.5 and 2.1 dS m^{-1}, total nitrogen 0.034 and 0.049%, available phosphorus 6.3 and 4.5 ppm and exchangeable potassium 192 and 94 ppm in Bahawalpur and Cholistan, respectively.

Pre-soaking irrigation was applied before seedbed preparation. When the soil reached a workable moisture level, seedbed was prepared by cultivating the field twice with tractor-mounted cultivator each followed by planking during Rabi 2014-15. Seeds of both chickpea cultivars were sown by hand drill using 60 kg ha^{-1} seed rate by keeping row to row distance of 25 cm and plant to plant distance 10 cm during last week of October 2014. Seeds were placed at 3 to 5 cm depth in each row. The seedlings were thinned out by maintaining one plant per hill at 15 days after sowing. The recommended dose of Urea @ 20 kg N ha^{-1} and Di-ammonium phosphate (DAP) @ 22 kg P ha^{-1} was applied at the time of sowing. The crop was maintained free from weeds, diseases and pests by adopting appropriate plant protection measures. All other agronomic practices were kept normal for all the treatments. Data of different weather elements were collected from Meteorological Observatory, Arid Zone Agricultural Research Institute, Bahawalpur, Pakistan during the growing season. Data on average maximum and minimum temperature, average relative humidity and total rainfall were recorded on daily basis and averaged each month (Figure 1).

2.3. Data collection

At 30 days after sowing, ten randomly selected plants were tagged to record the secondary branches per plant, number of pods per plant and 100 grain weight. Growth parameters were examined three times during growing seasons at both locations. Data regarding CGR was recorded by adopting the following Formula 1 of Watson (1952).

$$CGR\left(g\ m^{-2}day^{-1}\right)=\left(W_2-W_1\right)/\left(T_2-T_1\right) \quad (1)$$

whereas, W_1 and W_2 is the oven-dried weight at first and second sampling, T_1 and T_2 is the time of first and second sampling.

Relative growth rate (RGR) was calculated by using Equation 2 proposed by Radford (1967).

$$RGR\left(g\ g^{-1}day^{-1}\right)=\left(LnW_2-\ LnW1\right)/\left(T_2-T_1\right) \quad (2)$$

whereas, Ln= Natural Log, W_1 and W_2 is the oven-dried weight at first and second sampling, T_1 and T_2 is the time of first and second sampling.

The K^+ content in leaves were determined by flame photometer (Richards, 1954). Superoxide dismutase (IU $min^{-1}mg^{-1}$ protein) was calculated by adopting the methodology followed by (Giannopolitis and Ries, 1977). Moreover, peroxidase (IU $min^{-1}mg^{-1}$protein) and catalase (IU $min^{-1}mg^{-1}$protein) was calculated by adopting the procedure followed by Chance and Maehly (1955). $1m^2$ from each experimental unit was harvested and threshed manually to record the grain yield per unit area. The crop was harvested on 4th April 2015.

2.4. Statistical analysis

All the collected data was analyzed by using the MSTAT software. The Duncan's Multiple Range test (DMRt) was used to compare the differences among treatment means at 5% probability levels (Steel et al., 1997).

3. Results

3.1. Crop growth rate (g m^{-2} day^{-1})

Dry matter accumulation rate per unit land area is referred as crop growth rate (CGR). Data regarding CGR recorded at different growth stages were significantly affected by the interactive affects of chickpea genotypes, water stress at various growth stages and exogenous application of nutrients at both locations (Figure 1a and 1b). The CGR followed an increasing trend in the initial stages of crop growth and decreased thereafter. Exogenous application of potassium chloride on chickpea cultivar Bhakhar 2011 cultivated under well-watered condition produced significantly higher crop growth rate at all growth stages. Chickpea cultivar DUSHT cultivated under water stress at flowering + pod formation + grain filling stage produced minimum crop growth rate throughout the growing season.

3.2. Relative growth rate (g g^{-1} day^{-1})

Relative growth rate (RGR) is an obvious indicator of plant strategy with respect to productivity as related to environmental stress and disturbance regimes. RGR is the increase in size relative to the size of the plant present at the start of a given time interval. Data regarding RGR recorded at different growth stages were significantly affected by the interactive affects of chickpea genotypes, water stress at various growth stages and exogenous application of nutrients at both locations (Figure 2a and 2b). Exogenous application of potassium chloride on chickpea cultivar Bhakhar 2011 cultivated under well-watered condition produced significantly higher RGR at all growth stages. Chickpea cultivar DUSHT cultivated under water stress at flowering+ pod formation + grain filling stage produced minimum RGR throughout the growing season at both locations.

3.3. Secondary branches per plant

Secondary branches are mainly a genetic character and play an imperative role in improving the crop productivity.

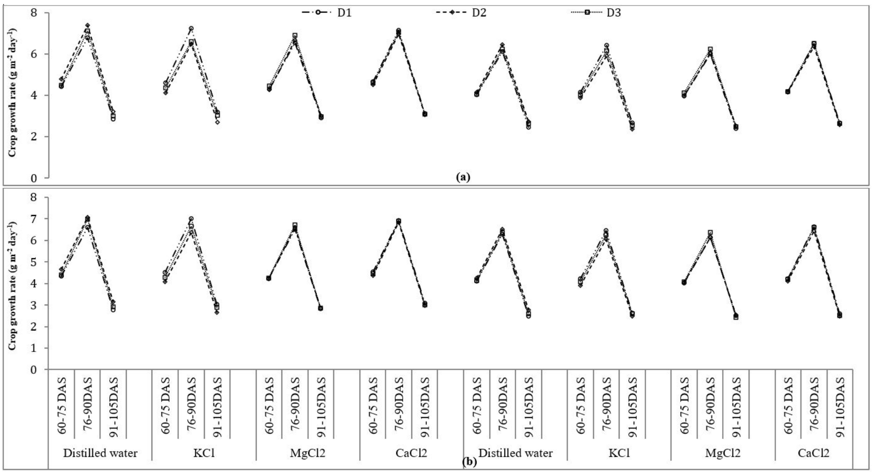

Figure 1. Effect of foliar application of nutrients on crop growth rate (g m^{-2} day^{-1}) of chickpea genotypes in Bahawalpur (a) and Cholistan (b). Whereas D1= well watered; D2=Drought at flowering+ pod formation + grain filling stage; D3= Drought at flowering stage; DAS= days after sowing.

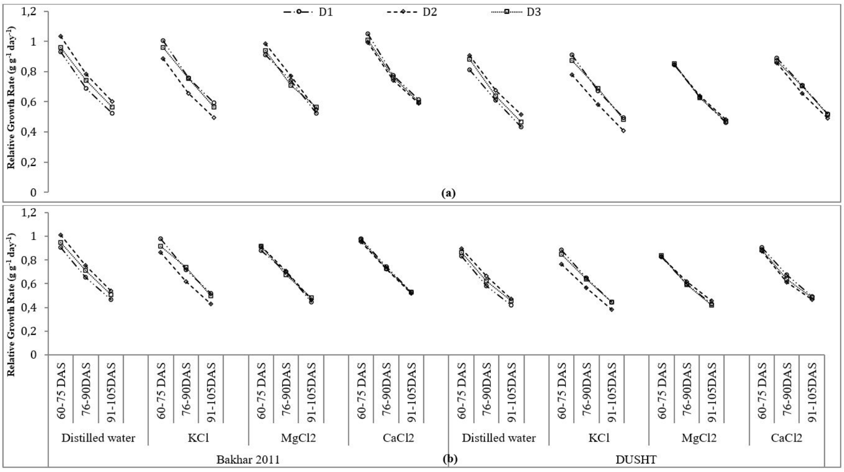

Figure 2. Effect of foliar application of nutrients on relative growth rate (g g^{-1} day^{-1}) of chickpea genotypes in Bahawalpur (a) and Cholistan (b). Whereas D1= well watered; D2= Drought at flowering+ pod formation + grain filling stage; D3= Drought at flowering stage; DAS= days after sowing.

Comparison of the treatment means showed that interaction among exogenous applications of nutrients, chickpea genotypes and water stress conditions at different growth stages significantly affected the number of secondary branches per plant. Foliar spray of nutrients significantly improved the secondary branches of both chickpea genotypes even under water stressed conditions. Exogenous application of potassium chloride on Bhakhar 2011 cultivated under well watered conditions produced maximum number of secondary branches per plant against the minimum was observed from same cultivar cultivated under water stress at flowering + pod formation + grain filling stage at both locations (Table 1).

3.4. Number of pods per plant

The results of analysis of variance values showed that interaction among exogenous applications of nutrients, chickpea cultivars and water stress conditions at different growth stages significantly affected the number of pods per plant (Table 1). Foliar spray of nutrients significantly improved the number of pods per plant of both chickpea genotypes even under water stressed conditions. Exogenous application of potassium chloride on Bhakhar 2011 cultivated under well watered conditions produced maximum number of pods per plant against the minimum was observed from DUSHT grown under water stress at flowering + pod formation + grain filling stage at both locations.

3.5. 100 grains weight (g)

Among the various parameters contributing towards final yield of a crop, hundred-seed weight is of prime importance. Interaction among exogenous applications of nutrients, chickpea cultivars and water stress conditions at different growth stages significantly affected the hundred grains weight (Table 1). Exogenous application of calcium chloride and potassium chloride on Bhakhar 2011 cultivated under well watered conditions produced maximum hundred grains weight in Bahawalpur and Cholistan, respectively. Whereas the minimum hundred grains weight was observed from DUSHT cultivated under water stress at flowering + pod formation + grain filling stage.

3.6. Biological yield (kg ha^{-1})

The biological yield refers to the total dry matter accumulation of a plant system. Interaction among exogenous applications of nutrients, chickpea genotypes and water stress at different growth stages significantly affected the biological yield (Table 2). Exogenous application of potassium chloride on Bhakhar 2011 cultivated under well watered conditions produced maximum biological yield against the minimum was observed from DUSHT grown under water stress at flowering + pod formation + grain filling stage at both locations.

3.7. Grain yield (kg ha^{-1})

Crop yield is a measurement of the amount of agricultural production harvested per unit of land area. It can also refer to the actual grains generation from the chickpea plant. Interaction among exogenous applications of nutrients, chickpea cultivars and water stress conditions at different growth stages significantly affected the grain yield (Table 2). Foliar spray of nutrients significantly improved the grain yield of both chickpea genotypes even under water stressed conditions at both locations. Exogenous application of potassium chloride on Bhakhar 2011 cultivated under well watered conditions produced maximum grain yield against the minimum was observed from DUSHT grown under water stress at flowering + pod formation + grain filling stage at both locations.

3.8. Leaf K^+ contents

The results of analysis of variance values showed that interaction among exogenous applications of nutrients, chickpea cultivars and water stress conditions at different

Table 1. Effect of foliar application of nutrients on secondary branches, pods per plant and 100 grains weight (g).

Chickpea genotypes	Water stress level	Foliar spray	Secondary branches per plant		Number of pods per plant		100 grains weight (g)	
			Bahawalpur	Cholistan	Bahawalpur	Cholistan	Bahawalpur	Cholistan
Bhakkar 2011	Well watered	Distilled water	9.93bc	10.15bc	28.51af	29.84 ad	203.48ad	186.48ad
		KCl	11.21a	11.35a	32.37a	33.71 a	210.18ab	193.83 a
		$MgCl_2$	10.47b	10.61b	29.56ac	32.39 ab	205.50ac	188.98 ad
		$CaCl_2$	10.51b	10.44b	30.08ab	31.42 ac	212.12a	191.21 ac
	Water stress at flowering+ pod formation + grain filling stage	Distilled water	8.17g	8.34ij	24.46fg	22.62gi	195.30be	176.20cd
		KCl	8.693dg	9.16dh	27.17bf	25.84 ci	204.43ac	187.34 ad
		$MgCl_2$	8.43fg	8.80fi	25.79cg	24.45 di	197.38ae	187.75 ad
		$CaCl_2$	8.42fg	8.76gi	25.17eg	24.07 di	194.89be	184.48 ad
	Water stress at flowering stage	Distilled water	8.77dg	9.17dg	27.29bf	26.62 bi	201.54ae	184.21ad
		KCl	9.27cd	9.75cd	30.15ab	29.38 ae	208.01ac	192.68 ab
		$MgCl_2$	9.19de	9.52ce	28.93ae	28.66af	203.78ad	187.67 ad
		$CaCl_2$	9.04df	9.45de	29.48ad	28.48ag	206.18ac	189.00 ad
DUSHT	Well watered	Distilled water	8.93df	9.23dg	25.17eg	27.47 bh	197.38ae	184.84ad
		KCl	9.20d	9.58cd	28.52af	30.77 ac	207.28ac	190.85 ac
		$MgCl_2$	8.98df	9.49de	25.76cg	28.75af	202.38ae	185.54 ad
		$CaCl_2$	9.02df	9.43df	26.18bg	28.09ah	201.17ae	185.72ad
	Water stress at flowering+ pod formation + grain filling stage	Distilled water	8.23g	8.06j	22.41g	21.07i	185.68e	174.27d
		KCl	8.83dg	8.66gj	25.29dg	23.41ei	191.34ce	181.33ad
		$MgCl_2$	8.69dg	8.52hj	24.91eg	22.41hi	187.43de	177.43bd
		$CaCl_2$	8.51eg	8.44ij	24.37 fg	23.02fi	193.69be	176.99cd
	Water stress at flowering stage	Distilled water	8.44fg	8.37ij	24.93eg	24.22 di	195.41ae	183.84ad
		KCl	9.06df	8.93ei	27.45bf	28.45ag	204.83ac	186.86 ad
		$MgCl_2$	8.76dg	8.76gi	26.13bg	27.07bh	203.06ad	184.81 ad
		$CaCl_2$	8.86dg	8.71gi	26.83bf	27.17 bh	199.28ae	184.32 ad
	LSD 0.05*p*=		0.6906	0.6402	4.2079	5.9757	16.707	15.679

LSD= Least significant difference.

growth stages significantly affected the leaf K^+ contents (Table 2). Exogenous application of potassium chloride on Bhakhar 2011 cultivated under well-watered conditions and water stress at flowering stage produced maximum leaf K^+ contents in Bahawalpur and Cholistan, respectively. Whereas, minimum leaf K^+ contents was observed from DUSHT grown under water stress at flowering + pod formation + grain filling stage at both locations.

3.9. Superoxide dismutase (IU $min^{-1}mg^{-1}$ protein)

Superoxide dismutase (SOD) is an enzyme that alternately catalyzes the dismutation of the superoxide (O_2^-) radical into either ordinary molecular oxygen (O_2) or hydrogen peroxide (H_2O_2). Interaction among exogenous applications of nutrients, chickpea cultivars and water stress conditions at different growth stages significantly affected the superoxide dismutase (Table 3). Exogenous application of potassium chloride on Bhakhar 2011 cultivated under water stress at flowering + pod formation + grain filling stage produced maximum superoxide dismutase against the minimum was observed from DUSHT grown under well watered conditions at both locations.

3.10. Peroxidase (IU $min^{-1}mg^{-1}$protein)

Interaction among exogenous applications of nutrients, chickpea cultivars and water stress conditions at different

Table 2. Effect of foliar application of nutrients on biological yield (kg ha^{-1}), grain yield (kg ha^{-1}) and leaf K^+ contents.

Chickpea genotypes	Water stress level	Foliar spray	Biological yield (kg ha^{-1})		Grain yield (kg ha^{-1})		Leaf K^+ contents	
			Bahawalpur	Cholistan	Bahawalpur	Cholistan	Bahawalpur	Cholistan
Bhakkar 2011	Well watered	Distilled water	5693.1ad	5793.1af	1783.1af	1792.1ad	50.49ab	47.15 dh
		KCl	6431.7a	6698.4a	2128.1a	2108.8 a	56.57a	62.41 ab
		$MgCl_2$	5984.5ac	6317.8 ac	1868.4ac	1976.7 ab	51.45ab	50.70ch
		$CaCl_2$	6197.7ab	6497.7ab	1923.3ab	1970.3abc	52.91ab	48.25 ch
	Water stress at flowering+ pod formation + grain filling stage	Distilled water	4765.6ch	4065.6gi	1405.3gi	1214.5fi	47.14b	42.08 gh
		KCl	5266.0af	4799.3ci	1629.5bh	1467.3ch	50.76ab	57.063 ad
		$MgCl_2$	5141.7bg	4375.0ei	1533.0ch	1382.0di	48.12ab	44.54 eh
		$CaCl_2$	4963.8bg	4463.8di	1503.1ch	1368.3di	51.64ab	47.48 dh
	Water stress at flowering stage	Distilled water	5494.0ae	4827.4ci	1635.1bh	1526.3 bh	51.28ab	50.54 ch
		KCl	6027.5ac	5827.5ae	1976.2ab	1802.4 ad	53.63ab	63.14 a
		$MgCl_2$	5968.7ac	5835.4ae	1811.3ae	1793.1 ad	51.06ab	52.46bg
		$CaCl_2$	5954.7ac	5954.7ad	1865.5ad	1781.5 ae	52.90ab	51.27 ch
DUSHT	Well watered	Distilled water	4547.6dh	5214.2ah	1420.5fi	1608.3 ag	46.73b	43.98fh
		KCl	5399.6af	5732.9af	1716.2bg	1815.0 ad	49.14ab	55.11 ae
		$MgCl_2$	4769.9ch	5336.6 ag	1489.7dh	1671.6 af	47.85ab	45.54 eh
		$CaCl_2$	5241.5ag	5274.8ah	1543.2ch	1662.5 af	50.19ab	47.51 dh
	Water stress at flowering+ pod formation + grain filling stage	Distilled water	3644.4h	3344.4i	1049.4i	955.5i	45.11b	41.05 h
		KCl	4566.7dh	3933.4gi	1375.1gi	1188.9fi	50.44ab	53.42 af
		$MgCl_2$	3991.2gh	3791.2gi	1268.9hi	1097.3hi	47.15b	45.30 eh
		$CaCl_2$	4311.5eh	3744.8hi	1269.4hi	1127.8gi	49.55ab	48.10 ch
	Water stress at flowering stage	Distilled water	4185.5fh	4252.1fi	1319.6hi	1281.1 ei	47.10b	43.38 fh
		KCl	5057.1bg	5323.8ag	1616.6bh	1674.0af	50.73ab	58.54ac
		$MgCl_2$	4690.8dh	4990.8 bh	1444.7eh	1503.7 bh	46.50b	47.81 dh
		$CaCl_2$	4653.0cdh	5253.0ah	1445.7eh	1594.0 bh	51.17ab	49.73 ch
	LSD 0.05*p*=		1262.7	1550.6	376.90	504.36	9.1396	10.586

LSD= Least significant difference.

growth stages significantly affected the peroxidase (POD) (Table 3). Exogenous application of potassium chloride on Bhakhar 2011 cultivated under water stress at flowering + pod formation + grain filling stage produced maximum peroxidase against the minimum was observed from DUSHT grown under well watered conditions.

3.11. Catalase (IU $min^{-1}mg^{-1}$ protein)

Interaction among exogenous applications of nutrients, chickpea cultivars and water stress conditions at different growth stages significantly affected the catalase (CAT) (Table 3). Exogenous application of potassium chloride on Bhakhar 2011 cultivated under water stress at flowering + pod formation + grain filling stage produced maximum catalase against the minimum was observed from DUSHT grown under well watered conditions at both locations.

4. Discussion

Water stress adversely affected chickpea growth and productivity by affecting the physiological and biochemical attributes. Exogenous application of mineral nutrients at 50% flowering and pod formation minimized the adverse affects of water stress by affecting the physiological

Table 3. Effect of foliar application of nutrients on SOD (IU min-1mg-1 protein), POD (IU min-1mg-1 protein) and CAT (IU min-1mg-1 protein).

Chickpea genotypes	Water stress level	Foliar spray	Superoxide dismutase (IU min-1mg-1 protein)		Peroxidase (IU min-1mg-1protein)		Catalase (IU min-1mg-1protein)	
			Bahawalpur	**Cholistan**	**Bahawalpur**	**Cholistan**	**Bahawalpur**	**Cholistan**
Bhakkar 2011	Well watered	Distilled water	70.67d	64.01 e	6.84e	6.46 d	38.98de	30.45 ef
		KCl	81.45d	75.27 e	7.10e	7.30 d	51.51c	40.05 e
		$MgCl_2$	77.85d	70.64 e	6.97e	7.07 d	46.59ce	37.77 ef
		$CaCl_2$	80.92d	73.90 e	7.03e	7.15 d	48.64cd	38.90 ef
	Water stress at flowering+ pod formation + grain filling stage	Distilled water	190.56bc	184.91bc	13.23d	15.04 c	88.65b	95.31 bc
		KCl	216.62a	214.38 a	19.11a	18.26 a	102.74a	110.96 a
		$MgCl_2$	197.89ab	201.16 ab	17.14bc	16.31 bc	93.80ab	102.88 ac
		$CaCl_2$	204.90ab	206.60 a	18.61ab	17.97ab	95.28ab	107.01 ab
	Water stress at flowering stage	Distilled water	79.69d	69.13 e	6.81e	6.65 d	42.55ce	29.57ef
		KCl	85.82d	74.15 e	7.23e	7.05d	52.29c	37.92 ef
		$MgCl_2$	77.92d	71.84 e	6.83e	6.81 d	45.70ce	36.71ef
		$CaCl_2$	79.13d	73.02 e	6.99e	6.91 d	47.48ce	39.32 ef
DUSHT	Well watered	Distilled water	66.64d	59.63 e	5.47e	6.04 d	36.86e	27.64 f
		KCl	78.83d	72.81 e	6.80e	6.57 d	43.56ce	34.35ef
		$MgCl_2$	76.63d	68.23 e	6.29e	6.38 d	41.69ce	30.75ef
		$CaCl_2$	78.20d	70.59 e	6.81e	6.54 d	43.03ce	33.67ef
	Water stress at flowering+ pod formation + grain filling stage	Distilled water	174.21c	163.52 d	16.49c	14.52 c	84.97b	81.36 d
		KCl	192.83bc	186.53 bc	17.85ac	16.39 bc	93.48ab	98.91ac
		$MgCl_2$	189.31bc	175.75 cd	16.52c	15.05 c	87.52b	90.88cd
		$CaCl_2$	194.45b	182.43 c	17.48ac	15.96 c	92.10ab	93.57 c
	Water stress at flowering stage	Distilled water	68.97d	63.33 e	6.30e	6.12 d	38.52de	28.36 ef
		KCl	77.16d	70.88 e	6.84e	6.68 d	44.85ce	34.70ef
		$MgCl_2$	74.70d	69.33 e	6.43e	6.33 d	41.72ce	32.97ef
		$CaCl_2$	78.16d	69.07 e	6.67e	6.53 d	43.94ce	33.93ef
	LSD 0.05*p*=		20.008	17.190	1.7849	1.8719	10.846	12.159

LSD= Least significant difference.

parameters of chickpea genotypes. Foliar spray of potassium chloride on chickpea cultivar Bhakhar 2011 resulted in significantly higher growth attributes even under water stress conditions. It might be due to the fact that the exogenous application of potassium improves the photosynthetic efficiency and the succeeding carbohydrate synthesis and translocation that ultimately improve the crop growth attributes (Pettigrew, 2008). Potassium chloride plays a key role in the growth of the plants that utilize it as a source of macromolecules, which are required for normal growth and metabolic pathways of plants (Tariq et al., 2019).

Chickpea cultivars were cultivated under water stress conditions at different growth stages significantly reduced the number of secondary branches per plant at both locations. However, exogenous application of potassium chloride produced significantly higher number of branches per plants as compared to the other mineral nutrients by enhancing the photosynthetic efficiency and translocated the photo-assimilates to lateral meristems (El-Sabagh et al., 2017). It was observed that water stress at any specific growth stage or throughout the growing period significantly reduced the yield contributing attributes of chickpea cultivars (Nasri, 2005). Our results showed that water stress at flowering + pod formation + grain filling stage significantly reduced the number of pods per plant and 100 grains weight that ultimately reduced the biological and grain yield in the drought-sensitive cultivar (DUSHT) than in the resistant one (Bhakhar 2011). Reduction in yield components might be due to disturbing the plant water relations, fewer and uneconomical nutrients uptake, reduced leaf photosynthesis, dry matter partitioning, less accumulation of photo-assimilates and its remobilization from source to pods resulted in accelerate maturity with shriveled grains (Zhang et al., 2014). Exogenous application of mineral nutrients accelerates nutrient uptake and reduces the detrimental influences of water stress (Nasri, 2005). Among these mineral nutrients, potassium plays a crucial role in enhancing the plant tolerance under water stress situations. Potassium plays a vital role in number of physiological processes such as photosynthesis, translocation of photosynthates, activation of enzymes and balance of turgidity (Mengel and Kirkby, 2001). It was also observed that potassium played a key role in the osmotic adjustment of plants under water stress and improves the adaptability of the plant to stress conditions, which ultimately enhanced the productivity (Yadov, 2006).

Water stress has a strong detrimental influence on the chickpea plants and it reduced the uptake of mineral nutrients including potassium. Inadequate supply of potassium negatively influences the stomatal roles in plant body, ensuing in late stomatal closure and even the incomplete closure of the pores. In present investigation it was observed that exogenous application of potassium chloride significantly improved the potassium uptake even under water stress conditions. Sufficient supply of potassium can apparently improve the organic osmolyte production particularly under water stress situations (Zain et al., 2014). As plants are exposed to water stress, Pro accumulation plays an extremely defensive role and it is involved in osmotic adjustments in plants (Teixeira and Pereira, 2007).

Water stress reduced the photosynthetic efficiency of chickpea plants due to modifications in chlorophyll contents and destruction of the photosynthetic apparatus (Salekjalali et al., 2012). One of the imperative reasons that water stress weakened the photosynthetic abilities of plants is the interruption of the balance between the production of reactive oxygen species (ROS) and the antioxidant defense triggering accumulation of ROS that stimulates the oxidative stress to membrane lipids, proteins and other cellular components (Shen et al., 2010). Plants have evolved several molecular and physiological means to handle with negative influence of water stress (Chen and Murata, 2011). Therefore, plants have developed antioxidant defense system to protect themselves from ROS. This system draws on a series of antioxidant enzymes i.e. SOD, POD and CAT etc. (Lee et al., 2007). In present study, the results showed that exogenous application of potassium chloride on Bhakhar 2011 cultivated under water stress at flowering + pod formation + grain filling stage significantly improved the antioxidant enzymes against the minimum antioxidants were observed from DUSHT cultivated under well watered conditions. A higher activity of antioxidant enzymes under water stress conditions is the basic cause for scavenging O^{2-} radical to produce H_2O_2 (Bano et al., 2012). There are various stress reactions concerning H_2O_2, the homeostasis of H_2O_2 is mainly due to catalase scavenging and regulating functions. Thus, H_2O_2 accumulation was possibly the trigger of SOD activation. Zheng et al. (2008) observed that foliar spray of potassium detoxified the ROS through increased SOD, CAT, and POD enzyme activities under water stress conditions. Similar results were reported by Jan et al. (2017) they concluded that the antioxidant enzymes activities were increased with the exogenous application of potassium in stress conditions thus detoxified the ROS. Hence, the equilibrium between ROS and the anti-oxidative system is vital for endurance and adaptation to stress (Zhang et al., 2014).

5. Conclusion

The present results showed that exogenous application of potassium chloride on Bhakhar 2011 produced significantly higher growth, yield and antioxidant enzymes activities even under water stress conditions as compared to other treatments. These results suggests that the application of potassium chloride mitigates the adverse effects of water stress and enhanced tolerance in chickpea mainly due to higher antioxidant enzymes activity, demonstrating the protective measures of plant cells in stress conditions.

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