Dubey S *et al.* (2020)
Notulae Botanicae Horti Agrobotanici Cluj-Napoca 48(2):954-966
DOI:10.15835/nbha48211861
Research Article

Effect of salinity stress on yield and quality parameters in flax (*Linum usitatissimum* L.)

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Abstract

Flax is one of the oldest utilitarian plants that have been grown for fibre as well as an oilseed crop. The species has been recognized as an excellent source of micronutrients, dietary fibre, protein, vitamin B1, lignan, and essential fatty acids (EFA), namely linoleic and α-linolenic acids. However, in spite of immense nutritional and industrial importance of the crop no work has been carried out on the effect of salinity induced by different salts on seed yield and quality traits in *Linum usitatissimum*. A high yielding germplasm line 'Mukta' was subjected to five concentrations (0 mM, 50 mM, 100 mM, 150 mM and 200 mM) of three salts viz. sodium chloride (NaCl), potassium chloride (KCl) and calcium chloride ($CaCl_2$) for two seasons. Data were collected for seed yield along with different quality traits and a range of antioxidant enzymes. Seed yield decreased with rise in salt concentration and was minimum at 200 mM for all the salts. Chlorophyll a, chlorophyll b and carotenoid content continuously decreased with increase in salt concentration for all the salt treatments. Catalase and GST content increased with increase in salt concentration and was maximum at 200 mM.

Keywords: antioxidant enzymes; carotenoid; flax; flavonoids; phenolics; salt stress

Introduction

Environmental conditions are known to exert a strong influence on plant growth and productivity (Feller and Vaseva, 2014; Pandolfi *et al*., 2017). A number of abiotic factors like salinity, drought and extreme temperatures play a major role in significantly reducing yield along with an adverse impact on several growth parameters (Tardieu, 2013; Bose *et al*., 2014). Salinity is one of the major abiotic stresses, which has significant impact on plant productivity and quality (Bhargava *et al*., 2003; Shahbaz and Ashraf, 2013). Soil salinity has significantly increased in recent years due to several factors like excessive irrigation, low precipitation, high surface evaporation, rock weathering, ion exchange and poor cultural practices (Munns and Tester, 2008; Bui,

Received: 27 Mar 2020. Received in revised form: 02 Jun 2020. Accepted: 09 Jun 2020. Published online: 30 Jun 2020.

2017). Recent reports indicate that about 20% of total cultivated and 33% of irrigated lands are afflicted by saline conditions at present, and that more than 50% of the arable land would be salinized by the year 2050 (Jamil *et al*., 2011; Shrivastava and Kumar, 2015). Salinity stress negatively affects all growth stages of plants in various ways like reduction of plant height, deterioration of the product quality and crop yields (Shahbaz and Ashraf, 2013; Shrivastava and Kumar, 2015; Rahneshan *et al*., 2018). Soil salinity is also known to affect different physiological and metabolic processes of plants such as reduction in water uptake, chlorophyll content, photosynthesis, transpiration rate, nutrient availability, stomata conductance and root hydraulic conductance (Khataar *et al*., 2018; Hernández, 2019).

Flax (*Linum usitatissimum* L.) is a member of the family Linaceae which comprises of 22 genera and approximately 180 species distributed over six continents (Hickey, 1988; Heywood, 1993; Vromans, 2006; McDill *et al*., 2009). The genus is divided in five subsections of which subsection *Linum* contains the cultivated species *L. usitatissimum* L. and the ornamentals *L. grandiflorum* and *L. perenne* (Tutin *et al*., 1968). It is considered that flax originated in southern Europe, the Near East, or Central Asia (Zeven, 1982; Zohary and Hopf, 2000) and was probably the first plant cultivated for fibre in the Old World (Zohary and Hopf, 2000). A wide karyological heterogeneity has been documented in *Linum* with chromosome numbers ranging from $2n$=12 to $2n$=84 (Kikuchi, 1929; Rogers and Harris, 1966; Harris, 1968; Bari and Godward, 1970; Robertson, 1971; Chennaveeraiah and Joshi, 1983). However, with respect to *L. usitatissimum* the diploid chromosome has been confirmed as $2n$=30 chromosomes (Muravenko *et al*., 2003) and the genome size (1C) as 686 mega base pairs (Mbp) (Bennett and Leitch, 2004). Flax is one of the oldest utilitarian plants that have been grown for fibre as well as an oilseed crop. Linseed oil has been extensively used in paint and flooring industries due to its rapid drying property in comparison with other traditional oils (Bayrak *et al*., 2010). The species has also been recognized as an excellent source of micronutrients, dietary fibre, protein, vitamin B1, lignan, and essential fatty acids (EFA), namely linoleic and α-linolenic acids (Singh *et al*., 2011; Kajla *et al*., 2015; Goyal *et al*., 2016).

However, inspite of immense nutritional and industrial importance of the crop no work has been carried out on the effect of salinity induced by different salts on seed yield, quality traits and antioxidant enzymes in *L. usitatissimum*. Therefore, the objective of the present study was to ascertain up to what extent can yield and quality be influenced by different salts and their concentrations? Principal component analyses (PCA) was carried out using 12 variables to ascertain the main factors that indicate stress situations effectively. This study reports for the first time the effects of different salts on seed yield and quality traits, as well as antioxidant enzyme systems in the species.

Materials and Methods

Experimental area

The experiments were conducted at Amity University Uttar Pradesh (Lucknow campus) which is located at 26.5 °N latitude and 80.5 °E longitude, and at 120 meter above sea level. This region is characterized by monsoonal climate and two major crop seasons, summer (Kharif- March to July) and winter (Rabi- October to February). *L. usitatissimum* is cultivated in the rabi season during which the minimum and maximum temperature ranges from 2.5-19 °C and 14-29 °C respectively.

Experimental material

The germplasm line Mukta having its origin at Chandra Shekhar Azad University of Agriculture and Technology, Kanpur, India was obtained from National Botanical Research Institute, Lucknow, India and used as the experimental material.

Experiment

The germplasm line Mukta was used in a split plot design for two experimental years (2017 and 2018) with sowing date as the main plot and salts and concentrations as subplots. Sowing was done at the beginning of January month in both 2017 and 2018. The weather parameters prevailing during both the experimental years have been provided in Table 1. The seeds were surface sterilized in 70% ethanol for 10s, and then washed with distilled water at least three times. Thereafter, ten seeds were sown in plastic pots containing vermiculite at depth of one cm. Five concentrations (0 mM, 50 mM, 100 mM, 150 mM and 200 mM) of three salts viz. sodium chloride (NaCl), potassium chloride (KCl) and calcium chloride ($CaCl_2$) were given to the germplasm line in two replications. Hoagland's solution was used as a nutrient medium with five additions during the entire growth season. A total of five salt treatments were provided to each pot during the growing season.

Table 1. Weather conditions during the experimental years

Experimental year	Temperature (°C)			Humidity (%)		
	Minimum	Maximum	Mean	Minimum	Maximum	Mean
2017						
January	2	28	15	29	100	79
February	9	33	19	21	100	63
March	9	42	25	14	100	48
April	17	43	33	9	89	38
2018						
January	3	27	12	29	100	82
February	8	34	19	15	100	66
March	13	40	26	16	88	53
April	19	42	31	13	94	45

Parameters recorded

Data were collected for seed yield along with different quality traits and antioxidant enzymes. The chlorophyll and carotenoid (mg/g) content of fresh leaves of each treatment were analysed following Jensen (1978). The total flavonoid and phenolic contents in leaf samples were spectrophotometrically determined by the method of Kevin *et al*. (2002) and Fatma *et al*. (2013), respectively. The flavonoid content was expressed in mg equivalent QE/gram of tissue and phenolic content was expressed as mg equivalent gallic acid/gram dry weight. Leaf protein (%) was estimated as per Bradford (1976). The scavenging ability of the natural antioxidants of the leaves towards the stable free radical DPPH (1,1-diphenyl-2-picrylhydrazyl) was determined by the method of Mensor *et al*. (2001) and expressed as µg/mL. Reduced glutathione was analysed by method of Ellman (1959) and expressed as µmol/g/min, while glutathione S-transferase (GST) activity was assessed following Habig *et al*. (1974) and expressed as mol/min/mg protein. Superoxide dismutase (SOD) activity was assayed according to Kakkar *et al.* (1984), while catalase activity was determined by the method of Aebi (1974) and both were expressed as unit/g fresh weight (U/g fw).

Data analysis

The results of each year (2017-18 and 2018-19) were statistically analysed separately by analysis of variance according to the split plot model using Windostat software program. Differences between means were separated using Tukey's test. Principal component analysis (PCA) (Hair *et al*., 1992) of a treatment/variable x year matrix, containing standardized trait data, was analysed through a biplot constructed by plotting the symmetrically scaled principal component 1 (PC1) scores against the principal component 2 (PC2) scores by using InfoStat® (Infostat, 2019) statistical software. All comparisons were made at the 5% level of significance ($p \leq 0.05$).

Results and Discussion

The analysis of variance showed significant differences among the salts for all the traits except flavonoids, phenolics and GST content for both the experimental years (data not shown). Also, significant differences were observed for salt concentrations for all the traits under study in both the years (data not shown).

In order to assess the patterns of variations, principal components analysis (PCA) was done considering all the 12 variables simultaneously. The first two principal components, PC1 and PC 2, that explained 63.9 and 22.0% of the total variance of measured traits were applied to make a score (Figure 1). All the variables except catalase, GST and DPPH had positive scores on PC1, while DPPH, SOD and chlorophyll a had highest value for PC2. It was interesting to note that in the PCA score plot, the treatments with lower salt concentrations (50 and 100 mM) were placed closely and were far apart from treatments with high salt concentrations (150 and 200 mM). This indicated that higher salt concentrations induced somewhat similar effects on quality traits and antioxidant parameters.

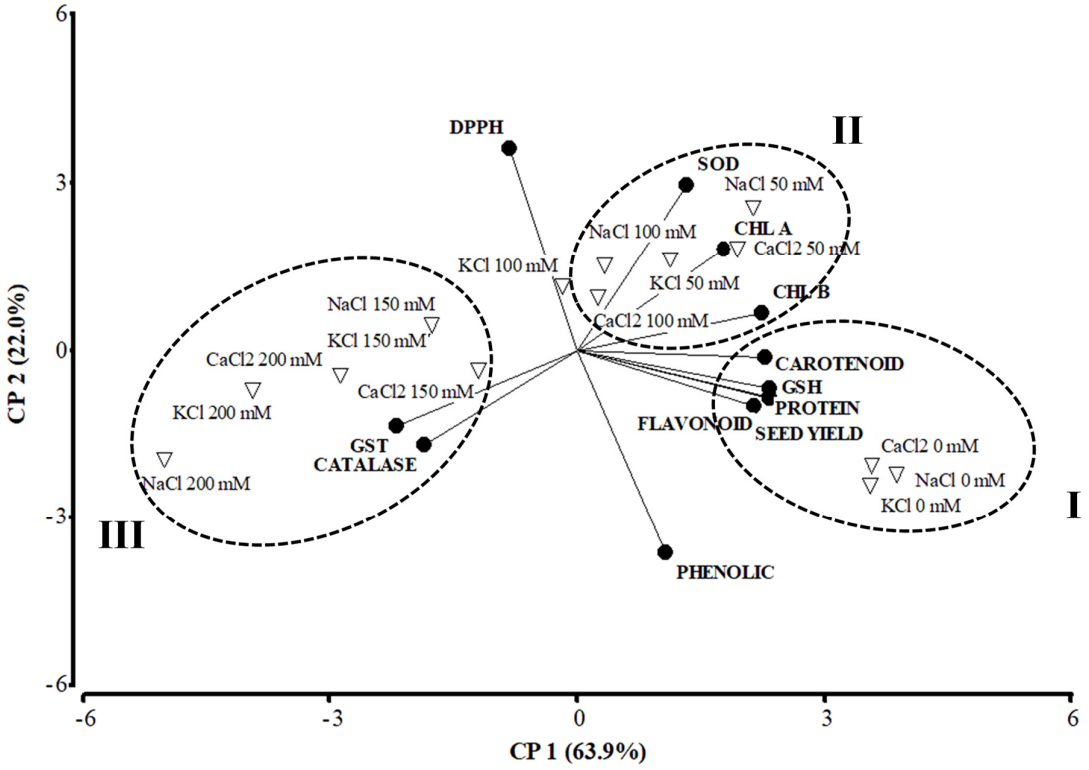

Figure 1. The score plot for the first two principal components explaining 85.9% of the variance of all salt treatments and measured variables included in the principal component's analysis

During K stress, both chlorophyll a and chlorophyll b initially increased on salt treatment at 50 mM, but thereafter continuously decreased for both the experimental years and were lowest for 200 mM KCl (Table 2). Chlorophyll a recorded a decrease of about 10% in 2017 at 200 mM KCl as compared to control plants. The fall was more drastic for chlorophyll b which reduced to about 50% at 200 mM in both the experimental years. As salt concentration increased, $CaCl_2$ and NaCl also had significant impact on chlorophyll content though not in a definite pattern. The decrease in chlorophyll during salt stress could be attributed to increased activity of chlorophyllase enzyme or salt induced weakening of protein-pigment lipid complex (Strogonov, 1970; Sivtsev *et al*., 1973). It was interesting to note that the carotenoid content continuously decreased with

increase in salt concentration for all the salt treatments (Table 2). However, the decrease was more pronounced during Ca and Na stress. Generally lowest reduction in the carotenoid content was witnessed in both the experimental years during K stress (Table 2). Similar decrease in leaf pigment concentrations in increasing levels of soil salinity has earlier been reported in other plants like *Phaseolus vulgaris* (Taïbi *et al*., 2016), *Acacia auriculiformis* (Rahman *et al*., 2017) and *Triticum aestivum* (Shah *et al*., 2017). The effect of salinity stress on plant pigments is highly specific and can be attributed to stomatal closure and mesophyll limitations for the diffusion of gases that leads to impaired photosynthetic process (Hu *et al*., 2005; Chaves *et al*., 2009; Tang *et al*., 2013).

Table 2. Mean values for flax leaf pigments during salt stress

Trait	2017						2018					
	CaCl2		KCl		NaCl		CaCl2		KCl		NaCl	
Chlorophyll a (mg/g)	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0	1.87 ±0.13 ab		1.77 ± 0.12 ab		1.89 ± 0.10 a		1.72 ± 0.11 a		1.67 ± 0.09 ab		1.77 ± 0.04 b	
50	1.91 ± 0.04 a		1.94 ± 0.01 a		1.96 ± 0.02 a		1.78 ± 0.01 a		1.78 ± 0.002 a		1.92 ± 0.01 a	
100	1.83 ± 0.0018 ab		1.76 ± 0.03 ab		1.78 ± 0.02 ab		1.76 ±0.0022 a		1.66 ± 0.01 ab		1.65 ± 0.01 c	
150	1.78 ± 0.01 ab		1.64 ± 0.01 b		1.77± 0.01 ab		1.75 ± 0.01 a		1.61 ± 0.02 ab		1.78 ± 0.02 b	
200	1.65 ± 0.0014 b		1.59 ± 0.01 b		1.64 ± 0.05 b		1.61 ± 0.0028 a		1.56 ± 0.03 b		1.62 ± 0.02 c	
Chlorophyll b (mg/g)												
0	1.56 ± 0.08 a		1.51 ± 0.07 a		1.42 ± 0.08 a		1.54 ± 0.08 a		1.64 ± 0.13 a		1.67 ± 0.04 a	
50	1.45 ± 0.00092 a		1.41 ± 0.02 ab		1.43 ± 0.00049 a		1.68 ± 0.01 a		1.61 ± 0.02 a		1.44 ± 0.07 b	
100	1.22 ± 0.00064 b		1.28 ± 0.07 bc		1.26 ± 0.04 ab		1.67 ± 0.0039 a		1.52 ± 0.01 a		1.63 ± 0.01 a	
150	1.23 ± 0.01 b		1.15 ± 0.01 c		1.07 ± 0.04 bc		1.58 ± 0.03 a		1.17 ± 0.02 b		1.17 ± 0.04 c	
200	0.98 ± 0.02 c		0.78 ± 0.01 d		0.90 ± 0.10 c		1.07 ± 0.04 b		0.83 ± 0.06 c		1.04 ± 0.01 c	
Carotenoids (mg/g)												
0	0.89 ± 0.09 a		0.96 ± 0.04 a		0.97 ± 0.11 a		0.78 ± 0.10 a		0.87 ± 0.08 a		0.9 ± 0.13 a	
50	0.85 ± 0.02 a		0.79 ± 0.01 b		0.80 ± 0.02 ab		0.78 ± 0.01 a		0.81 ± 0.02 ab		0.79 ± 0.01 ab	
100	0.63 ± 0.0011 b		0.70 ± 0.02 c		0.67 ± 0.0038 bc		0.64 ± 0.04 ab		0.73 ± 0.01 ab		0.68 ± 0.01 abc	
150	0.47 ± 0.0025 bc		0.61 ± 0.01 cd		0.49 ± 0.01 cd		0.52 ± 0.0049 b		0.68 ± 0.02 bc		0.57 ± 0.006 bc	
200	0.45 ± 0.01 c		0.56 ± 0.01 d		0.45 ± 0.01 d		0.49 ± 0.01 b		0.55 ± 0.01 c		0.46 ± 0.01 c	

Different letters within a column indicate significant differences according to Tukey's test ($p \leq 0.05$).

Phenolics are a large and diverse group of aromatic compounds found throughout the plant kingdom which possess at least one aromatic ring (C_6) and one or more hydroxyl groups (Boudet, 2007; Dai and Mumper, 2010). This ubiquitous group of most abundant secondary metabolites are ubiquitous in all plant organs with over 8000 structures reported so far (Manach *et al*., 2004; Bhattacharya *et al*., 2010; Dai and Mumper, 2010). Plant phenolics have recently drawn increasing attention due to their potent antioxidant properties, as protective agents, inhibitors, natural animal toxicants, pesticides and in the prevention of diseases associated with oxidative stress (Sowndhararajan and Kang, 2013; Arora *et al*., 2019; Durazzo *et al*., 2019). The concentration of phenolics reduced significantly at 50 mM salt concentration but thereafter continuously increased with increase in salt concentration and was maximum at 200 mM for all the three salts in both the experimental years (Table 3). This increase in phenolic content during salt stress has been attributed to disturbances in the secondary metabolic pathways (Ksouri *et al*., 2007). The phenolic content in flax leaves was higher than that reported in flax seeds (Oomah *et al*., 1995; Russo and Reggiani, 2015). Such high concentrations in phenolics in plant leaves as compared to other parts like flower, root, stem and fruit have also been earlier reported in *Lantana camara* (Family Verbenaceae) (Mahdi-Pour *et al*., 2012).

Flavonoids are low molecular weight secondary plant metabolites that exhibit wide distribution in most of the plant parts viz. leaves, seeds, bark and flowers (Tohge *et al*., 2013). Flavonoids, characterized by the flavan nucleus, have a broad range of functions in plants ranging from pollination and seed dispersal to protection against various abiotic and biotic stresses (Jiang *et al*., 2016; Yonekura-Sakakibara *et al*., 2019). Flavonoids have been associated with numerous biological properties and also confer various human health-related benefits (Kawai, 2018; Fraga *et al*., 2019). In the present study, the flavonoid content continuously decreased with increase in salt concentration for all the salt treatments. However, the decrease was maximum during K stress in 2017 and Na stress in 2018 (Table 3). The flavonoid content was highest during calcium stress for all salt concentrations in both the experimental years (Table 3). Also, moderate amount of protein content was reported in flax leaves which was highest during sodium stress (Table 3). Similar to flavonoids, the protein content was significantly lower at higher salt concentrations in both the experimental years. The protein content reduced to almost half at 50 mM K concentration and fell further at 200 mM K concentration in 2017. In 2018, the fall in protein was maximum for Na at 2.90% that indicated a fall of about 95% as compared to control in the quality parameter.

Seed yield presented interesting results during the course of the study. At higher salt concentrations (150 and 200 mM), seed yield reduced significantly in both the experimental years (Table 3). Seed yield continuously decreased with rise in salt concentration and was minimum at 200 mM for all the salts (Table 3). The reduction was most pronounced under Ca stress with seed yield reducing to 0.10 ± 0.01 g/plant and 0.08 ± 0.03 g/plant at 200 mM salt concentrations for 2017 and 2018, respectively. Less reduction in seed yield was observed during Na stress.

In nature, plants encounter a variety of stress conditions like supra-optimal light, water deficit, salinity, radiation, pathogens, chemicals and pollutants (Mhamdi *et al*., 2010). Saline conditions induce osmotic stress from low soil water potential, toxicity and oxidative stress from reactive oxygen species (ROS) (Nounjan and Theerakulpisut, 2012). The production of ROS such as superoxide (O_2^-), hydrogen peroxide (H_2O_2) and hydroxyl radical (OH^o) during salinity stress causes extensive oxidative damage to biomolecules like protein, DNA and lipids (Nounjan and Theerakulpisut, 2012; Weisany *et al*., 2012; Das and Roychoudhury, 2014). To counteract the ROS, plants generate non-enzymatic antioxidants as well as antioxidant enzymes i.e. peroxidase (POX), superoxide dismutase (SOD), ascorbate peroxidase (APX) and catalase (CAT) (Sairam and Tyagi, 2004). Several studies have pointed out that salt-tolerant species increased their antioxidant enzyme activities and antioxidant contents in response to salt treatment, whereas salt-sensitive species failed to do so (Shalata *et al*., 2001; Demiral and Türkan, 2005).

Catalase, a tetrameric heme-containing enzyme and the first antioxidant enzyme to be discovered, removes H_2O_2 by conversion into water and oxygen, and plays a key role in plant defense, aging, and senescence

(Scandalios, 2002; Yang and Poovaiah, 2002). The catalase content continuously increased with salt treatments as compared to control in both the experimental years during Ca stress (Table 4). However, maximum catalase content was observed during Na stress at 200 mM salt concentrations. There was about 9-fold increase in catalase content for Na in 2017 (7.15±0.44 U/g fw) and 2018 (6.77±2.39 U/g fw) as compared to control. Jakovljević *et al*. (2017) and Aghaei *et al*. (2009) have also reported increase in catalase content under high salt stress conditions in sweet basil (*Ocimum basilicum*) and potato (*Solanum tuberosum*).

Similar to catalase, the GST content increased with increase in salt concentration and was maximum at 200 mM (Table 4). This increase in GST content was most prominent for Na and exhibited a 9-fold enhancement in 2017 (8.10±1.05 mol/min/mg protein) and about 12-fold increase in 2018 (8.96±2.87 mol/min/mg protein).

DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) free radical method, developed by Marsden Blois (1958), is an easy, rapid and most frequently employed assay for the evaluation of the antioxidant potential of a biological source by spectrophotometry (Kedare and Singh, 2011; Lalhminghlui and Jagetia, 2018). DPPH, a stable free radical possessing deep purple colour is reduced in the presence of an antioxidant molecule, giving rise to pale yellow solution, thus allowing the spectrophotometric determination of the antioxidant activity (Akar *et al*., 2017). In this study, the DPPH content increased when low concentration of salt treatment was provided and was maximum at 50 mM salt concentration for K and Na (Table 4). After this initial rise in DPPH levels, the parameter gradually decreased with increase in salt concentrations and was lowest at 200 mM salt concentrations for both Na (20.21±3.07 µg/mL and 30.37±3.28 µg/mL for 2017 and 2018), and K (37.72±2.78 µg/mL and 32.23±1.41 µg/mL for 2017 and 2018 respectively) (Table 4).

Reduced glutathione is an NAD(P)H-dependent enzyme that catalyses the reduction of oxidized glutathione to reduced its reduced form thereby protecting the cell against oxidative damage (Foyer and Noctor, 2005). The GSH content showed a consistent pattern of decrease with rising salt concentrations for all the salts in both the experimental years (Table 4). Similar results have been obtained in tomato (*Solanum lycopersicum* L.) under salt stress by Parvin *et al*. (2019).

SOD, belonging to the family of metalloenzymes omnipresent in all aerobic organisms, plays a key role in catalysing the dismutation reaction of the superoxide radical (O^{2-}) to hydrogen peroxide (H_2O_2) and molecular oxygen and thus protects the cells from oxidative damage caused by ROS (Mohamed *et al*., 2016; Szymańska *et al*., 2017). In the present study low salt concentration of 50 mM led to a slight increase in the SOD levels (Table 4). Such increase in SOD content of salt treated plants has also been reported earlier in *Olea europaea* (Valderrama *et al*., 2006), *Robinia pseudoacacia* (Meng *et al*., 2016) and *Triticum aestivum* (Esfandiari and Gohari, 2017). But excess salt treatment resulted in a significant decline in SOD levels. The SOD content was lowest for Na at 200 mM in both the experimental years (2.71±0.34 U/g fw and 1.98±0.16 U/g fw, respectively) (Table 4).

Table 3. Changes in flavonoid, phenolics and protein in flax leaves and seed yield during salt stress.

	2017									
Trait	CaCl			KCl			NaCl			CaCl
Phenolics (mg equivalent gallic acid/g DW)	Mean	SD		Mean	SD		Mean	SD		Mean SI
0 mM	164.91	± 11.91	a	169.97	± 13.20	a	174.10	± 4.24	a	145.07 ± 7.5
50 mM	92.90	± 6.36	b	94.20	± 17.25	b	93.40	± 6.79	b	111.80 ± 1.1
100 mM	99.30	± 12.87	b	103.70	± 12.87	b	98.60	± 8.20	b	127.20 ± 12.4
150 mM	105.60	± 7.07	b	114.10	± 1.84	b	107.60	± 9.05	b	131.10 ± 9.4
200 mM	110.60	± 9.62	b	116.00	± 3.11	b	110.00	± 7.64	b	133.90 ± 8.3
Flavonoids (mg equivalent QE/g of tissue)										
0 mM	40.88	± 5.64	a	42.06	± 2.77	a	46.20	± 5.80	0	37.77 ± 3.5
50 mM	35.55	± 0.26	ab	30.45	± 1.59	ab	30.96	± 2.30	ab	35.57 ± 1.0
100 mM	33.57	± 0.04	ab	26.48	± 3.51	b	28.40	± 4.34	ab	34.19 ± 0.3
150 mM	32.19	± 0.19	ab	22.93	± 3.40	b	24.29	± 6.07	b	32.59 ± 0.3
200 mM	30.59	± 0.26	b	22.37	± 2.90	b	22.73	± 4.14	b	30.64 ± 0.3
Protein (%)										
0 mM	47.91	± 5.50	a	42.30	± 4.41	a	46.19	± 4.82	a	53.39 ± 2.8
50 mM	32.55	± 0.35	b	22.00	± 0.85	b	33.90	± 1.56	b	31.00 ± 1.4
100 mM	19.35	± 0.49	c	19.70	± 0.42	bc	25.70	± 0.71	b	15.10 ± 1.5
150 mM	9.40	± 0.28	d	12.20	± 0.28	c	7.10	± 0.57	c	8.00 ± 0.7
200 mM	4.15	± 0.21	d	4.00	± 0.00	d	4.05	± 0.07	c	5.10 ± 0.1
Seed yield (g/plant)										
0 mM	4.98	± 0.59	a	5.17	± 0.23	a	4.70	± 0.07	a	5.24 ± 0.2
50 mM	2.86	± 0.11	b	3.08	± 0.08	b	3.59	± 0.11	b	2.81 ± 0.1
100 mM	1.98	± 0.08	b	1.91	± 0.12	c	2.25	± 0.08	c	1.93 ± 0.1
150 mM	0.69	± 0.06	c	1.17	± 0.10	d	1.17	± 0.07	d	0.67 ± 0.0
200 mM	0.10	± 0.01	c	0.25	± 0.05	e	0.35	± 0.01	e	0.08 ± 0.0

Different letters within a column indicate significant differences according to Tukey's test ($p \leq 0.05$).

Table 4. Effect of salt stress on different antioxidant enzymes in flax leaves.

	2017							
Trait	CaCl		KCl		NaCl		CaCl	
Catalase (U/g fw)	Mean	SD	Mean	SD	Mean	SD	Mean	SD
0 mM	0.73 ± 0.07	a	0.82 ± 0.17	a	0.83 ± 0.04	a	0.67 ± 0.13	a
50 mM	0.83 ± 0.05	a	0.80 ± 0.08	a	0.31 ± 0.01	a	0.75 ± 0.05	a
100 mM	1.32 ± 0.04	a	0.60 ± 0.05	a	0.68 ± 0.06	a	1.39 ± 0.19	a
150 mM	2.85 ± 0.16	b	1.29 ± 0.04	b	1.72 ± 0.11	b	3.01 ± 0.34	b
200 mM	2.78 ± 0.30	b	2.53 ± 0.06	c	7.15 ± 0.44	c	2.51 ± 0.09	b
GST (mol/min/mg protein)								
0 mM	0.81 ± 0.11	a	0.96 ± 0.06	a	0.92 ± 0.09	a	0.77 ± 0.07	a
50 mM	0.94 ± 0.07	a	1.33 ± 0.05	ab	0.87 ± 0.04	a	0.93 ± 0.07	ab
100 mM	1.90 ± 0.05	b	1.53 ± 0.05	b	1.34 ± 0.002	ab	1.66 ± 0.24	b
150 mM	3.25 ± 0.26	c	2.85 ± 0.17	c	2.83 ± 0.05	b	2.93 ± 0.30	c
200 mM	5.41 ± 0.27	d	5.39 ± 0.11	d	8.10 ± 1.05	c	3.98 ± 0.26	d
DPPH (µg/mL)								
0 mM	9.18 ± 0.54	a	8.92 ± 0.93	a	8.68 ± 0.68	a	8.02 ± 0.38	a
50 mM	53.69 ± 0.53	c	42.22 ± 2.52	b	39.38 ± 3.46	c	54.65 ± 4.42	c
100 mM	48.37 ± 5.03	bc	41.90 ± 2.29	b	35.67 ± 1.21	c	55.04 ± 2.88	c
150 mM	28.77 ± 9.23	ab	39.33 ± 2.17	b	30.34 ± 2.98	bc	30.64 ± 3.15	b
200 mM	45.64 ± 3.72	bc	37.72 ± 2.78	b	20.21 ± 3.07	b	47.04 ± 1.39	c
GSH (µmol/g/min)								
0 mM	45.74 ± 10.55	a	41.32 ± 6.31	b	38.20 ± 4.81	b	53.02 ± 10.20	b
50 mM	45.05 ± 0.62	a	41.76 ± 0.26	b	38.87 ± 1.80	b	43.30 ± 1.14	ab
100 mM	41.05 ± 0.22	a	39.51 ± 0.49	b	36.14 ± 1.03	b	41.20 ± 1.02	ab
150 mM	37.81 ± 0.80	a	34.17 ± 1.01	ab	33.47 ± 1.00	ab	36.18 ± 1.14	ab
200 mM	32.91 ± 0.12	a	24.34 ± 1.02	ab	25.12 ± 1.57	a	34.13 ± 1.11	a
SOD (U/g fw)								
0 mM	8.17 ± 0.21	b	8.60 ± 0.25	b	8.33 ± 0.30	b	7.52 ± 0.23	bc
50 mM	14.33 ± 0.97	c	9.65 ± 0.68	b	18.87 ± 0.22	d	12.72 ± 0.41	d
100 mM	8.08 ± 1.07	b	8.99 ± 0.14	b	16.42 ± 0.71	c	8.92 ± 1.16	c
150 mM	6.49 ± 0.77	ab	6.19 ± 0.18	a	8.16 ± 0.32	b	5.72 ± 0.47	ab
200 mM	4.94 ± 0.11	a	5.17 ± 0.20	a	2.71 ± 0.34	a	4.80 ± 0.28	a

Different letters within a column indicate significant differences according to Tukey's test ($p \leq 0.05$)

Conclusions

The study showed that salt stress induced physiological changes in flax germplasm. Reductions in leaf pigment concentration were observed in plants exposed to high salt concentrations, indicating that salt stress reduced the capacity of the photosynthetic system for efficient energy conversion. The leaf protein content exhibited least reduction during Na stress, while seed yield was lowest during Ca stress. The catalase and GST content increased with increase in salt concentration and was maximum at 200 mM for all the salt treatments.

Acknowledgements

The authors are thankful to Director, National Botanical Research Institute to provide the germplasm for carrying out the present work.

Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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