

Original Research Article

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Physiological Parameters of Salt Tolerance in Different Genotypes of Oats (*Avena sativa* L.)

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ABSTRACT

Keywords

Oat, Salt stress, Germination percentage, Seed vigour, *Avena sativa*

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Eight genotypes of oats were screened under different salt stress levels (25, 50, 75, 100, 125, 150, 175 and 200 mM). The effect of salt stress on physiological characteristics of eight oat genotypes (Kent, OL-10, OL-1862, OL-1869, OL-125, OL-1966, OL-1876, OL-1895) cultivated under salt stress conditions was investigated in this study. The experiment was conducted in triplicates in the Punjab Agricultural University, Ludhiana. The results show that the percentage germination of seeds declined significantly beyond 100 and 125mM of NaCl in most of the genotypes. These genotypes showed a marked decline in shoot length, root length, seed vigour I, seed vigour II, dry weight. The varieties could be divided into two groups based on the decrease in shoot length, root length at 100mM which were comparatively sensitive and comparatively tolerant.

Introduction

Salt stress is a main abiotic stress or affects the crop production. Salinity mainly affects the more than 6% of the worlds total land area (Gao *et al.*, 2016). According to other report, 7% of the land region is affected by salt stress (Neto *et al.*, 2006). Salinity of soil may be attributed to the seepage from Rajasthan feeder canals and appears to be the cause for the development of water logging and salinity in these areas. Salinity affects crop development by affecting many physiological, biochemical and metabolic processes. Salt stress affects the different development

processes which include photosynthesis, ion regulation, water relations, etc (Ashraf 2004). Salinity, which not only affects crop physiological, morphological, and biochemical processes, but also germination of seedling, development with nutrient/water uptake in oat (Willenborg *et al.*, 2005). High concentration of soil salinity is difficult for roots to take up water and it can be toxic within plant cells (Zhang *et al.*, 2018).

Oat is known as moderately salt tolerant, however it is more sensitive to salt stress as compared to other cereals like barley and wheat. Salt stress has 3 possible effects on

plants which are as (i) reduction in water potential, (ii) toxicity of any Na⁺ and Cl⁻ absorbed and (iii) intrusion with the absorb of necessary nutrients. By changes in salt ion concentration, kind of salt present, or kind of plant species, the fresh and dry weights of the shoot system are affected either negatively or positively (Memon *et al.*, 2010). In Oat, Total leaf area, dry weight of shoot decreased by salt stress (Zhao *et al.*, 2017). Salinity amount can effect numerous process in plants, which includes the photosynthesis regulation, balance of ions (Prisco *et al.*, 2007). Propagation of salt-tolerant oats has been restricted by many factors, such as the lack of a standard, which is effective method for evaluate salt tolerance (Talei *et al.*, 2013). Germination is a necessary phase of vegetation and opposition against salt stress during the germination is very significant for stability. Germination proportion slowly decreased in all oat cultivated plants as the amount of salinity increased from 25 to 100 mM (Chauhan *et al.*, 2016).

This study dealt with the analysis of salt tolerant, salt moderant and salt sensitive oat varieties. NaCl as a neutral salt is applied as stress treatment to oat seedlings to talk over the salt tolerance procedure and adaptability under NaCl stress (Zhanwu 2015).

Materials and Methods

Oat seed was collected from fodder section of department of Plant Breeding and Genetics, Punjab Agricultural University, Ludhiana. For each treatment ten seeds were counted and disinfected in this experiment. At first, the entire Petri dishes were cleaned with ethanol. Oat seeds were surface sterilized by 0.1 % solution of mercuric chloride, followed by thorough washing by distilled water to avoid any fungal infection. This study work was performed at the laboratory of the Botany Department of Basic Sciences and Humanities College, Punjab Agricultural University,

Ludhiana. We used NaCl salt with nine concentrations of 0, 25, 50, 75, 100, 150, 175 and 200 mM. To stimulate salinity stress, the germination paper in petri dishes was moistened with solutions of different salinity levels (Control, 25mM, 50mM, 75mM and 100mM, 125mM, 150mM, 175mM and 200mM NaCl). The petri dishes were placed in an incubator at 25°C and 60 ± 15 % relative humidity for 14 days to record the effect of salinity on germination parameters.

During the experiment, the germinated seeds were counted each day at a certain time of the daytime. At the end of the experiment (after 12 days), the no and length of root and shoot, fresh and dry weight of root and shoot (oven dried for 72 hr at 70°C) and the no of normal and abnormal seedlings were measured. Seedlings having dead root tips as well as dead or severely distorted cotyledons were considered as abnormal plants.

Percent Germination

$$\text{Percent germination(\%)} = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds}} \times 100$$

Root length (cm)

On 14th day of germination five normal seedlings for each treatment were chosen from the germination test for measuring the root length. Root length was measured from the collar region to the tip of root. Average root length of five seedlings was computed and expressed in centimetres.

Shoot length (cm)

For measuring shoot length, previous five seedlings was used which was used for the root length measurement. The length of shoot was measured from the collar region to the point of attachment of cotyledons. The average of five seedlings was computed and expressed in centimeters.

Seedling Dry Weight (mg)

For the calculation of seedling dry weight, randomly five normal seedlings were taken and dried using oven at 110°C for 17 hours and their dry weight had been recorded by using weighing balance and average weight was computed and expressed in milligrams.

Vigour Index (VI)

Vigour index of seeds were calculated as suggested by (Abdul Baki's Anderson 1973).

Vigour Index I = Germination (%) x Seedling length (cm)

Vigour Index II = Germination (%) x Seedling dry weight (g)

Epicotyl length of seedlings were measured by using centimetre scale.

Results and Discussion

Overall, the salt stress reduced the growth in all varieties, while OL-10 variety showed sensitivity, Kent Variety showed tolerance and OL-1862 was moderate for almost all parameters. By increasing concentration levels the germination of seed and other traits decreased. The maximum inhibitory effect was seen in 200 mM.

Percentage Germination (PG)

Seed germination and seedling growth are critical determining for seedling growth during the initial stages of life. By increasing concentration the percentage germination was decreased to 0% at 200 mM concentration. At 75mM and 100mM, the genotypes showed a differential response to salinity stress. The decline in percentage germination varied from 96 to 49.3. On the basis the genotypes could be divided into comparatively salt sensitive, salt moderate and salt tolerant. It decreased

from 90 percent to 0 percent in OL-10 genotype thus making it salt sensitive and 100 percent to 26.7 percent in Kent genotype, as salt concentration increased from 25mM to 200mM thus making it salt tolerant (Table 1).

This reduction in the percentage of seeds germinating induced by an increase of salinity stress has been described by numerous authors (Akbarimoghaddam, 2011; Zhang, 2013; Kumar 2014; Chauhan, 2019; Berk and Ozkan, 2016). By the increase of salinity level, the reduction in germination was possibly due to the reduced soluble osmotic potential, high toxic ions and seed nutrient imbalance (Abbasian and Moemeni, 2013). It is also assumed that in addition to toxic effects of certain ions, higher concentration of salt reduces the water potential in the medium that hinders water absorption by germinating seeds and thus decreases germination (Maas and Nieman, 1978).

Length and weight of root and shoot (SL, RL, SRDW)

The root and shoot lengths are the most important parameters for salt stress because roots are in direct contact with soil and absorb water from soil and supply it to the rest of the plant. So, root and shoot length provide an important indication plant response to salt stress (Jamil *et al.*, 2006). The results showed that the Major decline of shoot length was observed in all the genotypes at 200 mM as compared to control. Differential response was seen at 75, 100 and 125mM NaCl concentration and the decline in shoot length range varied from 18.7 percent to 73.9 percent from 75 to 100mM salt concentration in different oat genotypes. In comparatively sensitive genotypes, the decrease was more than 50 percent at 100mM. Drastic changes were observed at 100 mM salt concentration. In OL-10 shoot length reduced by 100 percent at 200mM making it comparatively salt sensitive whereas with a decrease of only 80.6

percent at 200mM, Kent was considered as comparatively salt tolerant genotype.

The maximum root and shoot lengths were recorded in control treatment (Table 2 and Table 3). However, there occurs more progressive and linear decreases in roots as compared to shoot length with increase in soil salinity levels. Root length decreased gradually as oat seedlings were exposed to salt stress. In OL-10 root length decreased by 36.2 percent at 100mM salt concentration. In Kent decline was 28.4 percent at 100mM salt concentration and in OL-1876 a decrease of 26 percent was observed at 100mM salt concentrations. At higher salinity levels, there occurs decrease in root hairs as there is no sufficient energy for formation of new organs. Similar results were seen in barley seedlings by (Agami, 2014).

The decrease in shoot and root growth may be due to toxic effects of the salt used and unbalanced nutrient uptake by the seedlings. Root length and shoot length decreases with the increase in salinity level that may be due to slowing down the water absorption by the plants (Demir and Arif, 2003).

The increase in root to shoot ratio or decrease in shoot to root ratio is a general response to salt stress, related to factors linked with water stress (osmotic effect) rather than a salt-specific effect (Hsiao, 2000). Normally, the reduction in seedling length, due to the increase in the salt concentration, are caused by physicochemical effects or by osmotic-toxic salts that occurs in salty solutions. Salinity, generally, has inhibitory effects on germination of seeds (Zhang, 2010; Kaveh, 2011; El-Sabagh, 2018) due to hyperosmotic stress and hyper-ionic toxicity (Hasegawa 2000). Similar trend was observed in dry weight as salt concentration was increased. No much reduction in dry weight was seen at 25mM salt concentration i.e. 25 mM was at

par with control. In OL-10 dry weight decreased significantly from 21.8 mg in control to 0 mg at 200 mM salt concentration level. In Kent decline in dry weight accounted for 2 mg at 200 mM salt level as compared to its control counterparts.

Lamina Length (cm)

Lamina growth decreases with increased salinity level. Differential response was seen at 75, 100 and 125mM NaCl concentration. The decline in lamina length range varied from 12.6 percent to 82.4 percent from 75 to 100mM salt concentrations in different oat genotypes.

In comparatively sensitive genotypes, the decrease was more than 60 percent. In moderate genotypes the range lied between 30 to 60 percent whereas in salt tolerant genotypes, the decline was less than 30 percent. With a 97 percent decline at 200mM as compared to control, salt stress was much pronounced in lamina length. Much differential effects were observed at 75mM and 100mM NaCl where a sudden decrease in length of lamina was observed in many genotypes. At 125mM much of lamina growth was diminished in most of the genotypes. In OL-10 a tremendous decrease of 60.5 percent at 100 mM was seen thus making it most sensitive among all the 8 genotypes. Kent was considered as most salt tolerant with a mere decrease of only 29.3 percent at 100 mM NaCl concentration.

Sheath

A structure at the base of a leaf's petiole that partly surrounds or protect the stem or another organ that it subtends. Sheath, but not root or leaf, were useful as indicators to identify salt stress. The results showed that the Sheath length also reduced gradually with the increase in salt concentration but the decline

was not very much pronounced. Sheath length showed a decline in all the genotypes. In OL-10 genotype sheath length declined by 37.5 percent at 100mM and in Kent it declined by 27.5 percent at 100mM. (De Leon *et al.*, 2015) reported that salinity tolerance is most likely controlled in the shoot.

Dry weight (mg)

Similar trend was observed in dry weight as

salt concentration was increased. No much reduction in dry weight was seen at 25 mM NaCl concentration i.e. 25 mM was at par with control. In OL-10 dry weight decreased significantly from 21.8 mg in control to 0 mg at 200mM salt concentration level. In Kent decline in dry weight accounted for 2 mg at 200mM salt level as compared to its control counterparts.

Table.1 Salt stress effect on percent germination in oat (*Avena sativa* L.)

Treatment	OL-10	Kent	OL-1862	OL-1895	OL-1966	OL-125	OL-1869	OL-1876
Control	90	100	95	83	100	91.6	96.6	100
NaCl(25mM)	71	100	90	78.3	100	90	96.6	98.3
NaCl(50mM)	65	95	85	75	100	87.3	88	90
NaCl(75mM)	65	84.3	75	75	96	81.6	81.6	80
NaCl(100mM)	49.3	80	61	74	60	61.6	53.3	75
NaCl(125mM)	48.7	70	35	44	50	60	39.3	70.6
NaCl(150mM)	0	58.3	25	35	47.3	51.6	23.3	70
NaCl(175mM)	0	45.7	20.3	30	23.3	36.6	0	55
NaCl(200mM)	0	26.7	20	17.6	0	0	0	28.3
CD at 5%	X=2.22		Y=2.36		XY=6.67			

X=Genotype
Y=Salt Concentration

Table.2 Salt stress effect on shoot length in oat (*Avena sativa* L.)

Shoot length(cm)	OL-10	Kent	OL-1862	OL-1895	OL-1966	OL-125	OL-1869	OL-1876
Control	16.3	17.6	16.8	18.9	21.1	16.8	16.1	16.0
NaCl(25mM)	15.1	16.1	13.9	16.9	16.0	14.7	8.1	13.7
NaCl(50mM)	14.1	14.3	12.1	14.7	12.3	13.7	7.7	13.0
NaCl(75mM)	11.0	14.3	12.3	14.0	9.1	11.8	7.4	11.9
NaCl(100mM)	6.8	10.8	8.7	11.2	5.5	6.6	5.4	9.8
NaCl(125mM)	2	8.5	5.6	9.5	5.2	3.7	5.0	7.5
NaCl(150mM)	0	5.7	4.5	6.9	3.6	3.1	2.7	5.0
NaCl(175mM)	0	4.4	3.4	2.7	2.1	2.3	0.0	2.7
NaCl(200mM)	0	3.4	2.0	2.3	0.0	0.0	0.0	1.5
CD at 5%	A=0.57		B=0.29		AB=0.82			

Table.3 Salt stress effect on root length (cm) in oat (*Avena sativa* L.)

Root length(cm)								
Treatment	OL-10	Kent	OL-1862	OL-1895	OL-1966	OL-125	OL-1869	OL-1876
Control	10.2	12.3	11.7	11.4	13.3	14.5	9.1	101.5
NaCl(25mM)	9.4	11.2	9.4	8.8	10.2	11.6	8	11.3
NaCl(50mM)	9.4	9	9	7.7	9.6	11.6	7.9	11
NaCl(75mM)	7.4	9	8.5	7.2	7.4	13.1	7.7	10
NaCl(100mM)	6.5	8.8	6.5	5	6.9	6.1	7.5	8.5
NaCl(125mM)	4.3	7.5	6.3	4.4	5.9	6.1	6.5	8.5
NaCl(150mM)	0	6	4.7	3.7	5.2	6.0	5.8	6
NaCl(175mM)	0	4.6	2.6	3.1	3.2	4.4	0	5.5
NaCl(200mM)	0	4.0	2.4	1.4	0	0	0	3.3
CD at 5%	X=0.47		Y=0.50		XY=1.42			

X=Genotype

Y=Salt Concentration

Table.4 Effect of salt stress on percent decrease in Lamina length in oat (*Avena sativa* L.) variety

% decrease								
Lamina	OL-10	Kent	OL-1862	OL-1895	OL-1966	OL-125	OL-1869	OL-1876
Control	0	0	0	0	0	0	0	0
25mM	18.3	10.9	26.7	11.5	28.6	10.7	64.1	31.8
50mM	30.6	26.4	16.9	25	25.1	31.5	65.8	15
75mM	47.6	27.5	32	61.5	12.6	33.8	85.8	39.8
100mM	60.5	29.3	56.2	71.1	82.4	69.2	61.6	29.2
125mM	67.3	48.2	70.5	71.1	84.2	86.1	95.8	55.7
150mM	100	81.6	83	90.3	92.3	89.2	76.6	85.8
175mM	100	82.2	89.5	90.3	95.3	96.9	100	92
200mM	100	89	89.5	100	100	100	100	100

Table.5 Effect of salt stress on percent decrease in sheath length in oat (*Avena sativa* L.) variety

Sheath	OL-10	Kent	OL-1862	OL-1895	OL-1966	OL-125	OL-1869	OL-1876
Control	0	0	0	0	0	0	0	0
25mM	3.5	17.3	6.7	2	0	0	19.6	2
50mM	5.3	21.7	20.3	8	10.7	11.3	27.4	14.5
75mM	19.6	24.6	18.6	12	16	28.3	29.4	20.8
100mM	37.5	27.5	30.5	12	35.7	58.4	45	27
125mM	53.5	27.5	30.5	12	55.3	62.2	66.6	45.8
150mM	100	49.2	54.2	26	55.3	62.2	86.2	58.3
175mM	100	63.7	69.4	46	67.8	75.4	100	72.9
200mM	100	75.3	74.5	60	100	100	100	85.4

Table.6 Salt stress effect on dry weight (mg) in oat (*Avena sativa* L.)

Dry weight								
Treatment	OL-10	Kent	OL-1862	OL-1895	OL-1966	OL-125	OL-1869	OL-1876
Control	21.8	36	26	45.3	34.6	25.7	27.7	45.4
NaCl(25mM)	20	23.3	17	25.8	32	21.3	26	45
NaCl(50mM)	16	19.5	15.9	25	27.9	20	26	42
NaCl(75mM)	7.8	18.3	9.6	21.5	23.7	20	25.7	32.4
NaCl(100mM)	6.4	15	7.6	21	23	16.3	24.7	26.5
NaCl(125mM)	6	8.9	7	4.5	21	16.3	11.6	24.6
NaCl(150mM)	0	4.7	2	4	5.8	13	7.5	21.6
NaCl(175mM)	0	4.7	1.5	3.5	2	5.7	0	11.5
NaCl(200mM)	0	2	0	1.8	0	0	0	11.3
CD at 5%	X=0.53			Y=0.56		XY=1.60		

X=Genotype, Y=NaCl concentration

Table.7 Effect of salt stress on seed vigour I

Seed Vigour(I)								
Treatment	OL-10	Kent	OL-1862	OL-1895	OL-1966	OL-125	OL-1869	OL-1876
Control	2433	2930	2714	2267.7	3436.7	2864.3	2437.3	2743.3
NaCl(25mM)	1873.9	2726.7	2097	1930	2620	2365.8	1552.7	2461.7
NaCl(50mM)	1529.8	2215.8	1789.4	1862.3	2190	2208.1	1374.8	2163
NaCl(75mM)	1283	1960.0	1458.2	1659.5	1584.9	2023.8	1232.8	1819.2
NaCl(100mM)	750.2	1567.2	811.0	1193.6	743.5	788.5	685.7	1461.3
NaCl(125mM)	309.5	1117.7	412.8	610.7	556.8	606.2	452.4	1181.3
NaCl(150mM)	0	683.8	229.8	372.8	415.6	472.5	195.3	796
NaCl(175mM)	0	414	124.7	174	123.7	245	0	492.2
NaCl(200mM)	0	198.7	86.8	59.9	0	0	0	127
	A= 56.9			B=60.4		AB=170.9		

X=Genotype

Y=NaCl

Table.8 Effect of salt stress on seed vigour II

Seed Vigour(II)								
Treatment	OL-10	Kent	OL-1862	OL-1895	OL-1966	OL-125	OL-1869	OL-1876
Control	2	3.6	2.3	3.3	3.4	2.3	2.6	4.5
NaCl(25mM)	1.4	2.3	1.5	1.9	3.2	1.9	2.5	4.4
NaCl(50mM)	1	1.8	1.3	2.0	2.7	1.7	2.2	3.7
NaCl(75mM)	0.5	1.4	0.7	1.7	2.3	1.6	2.0	2.5
NaCl(100mM)	0.3	1.2	0.3	1.4	1.4	1	1.3	1.9
NaCl(125mM)	0.3	0.6	0.2	0.1	1	1.0	0.4	1.7
NaCl(150mM)	0	0.2	0	0.1	0.2	0.4	0.1	1.5
NaCl(175mM)	0	0.1	0	0.1	0.0	0.2	0	0.7
NaCl(200mM)	0	0	0	0.0	0	0	0	0.3
CD at 5%	A=0.69			B=0.73		AB=0.20		

Seed vigour index (SVI) I and II

Seed vigour is a physiological property which governs the capability of a seed to produce a seedling quickly in soil and the extent to which that seed tolerate a different environmental factors (Perry, 1980). Perry (1972b) concluded that seeds which have high vigour are more tolerant to environmental stress. The deleterious effects of salinity were further confirmed by analyzing vigour index. According to Table 6, in OL-10, 2433 was recorded vigour I in control, 750.2 at 100mM and 0 at 200mM salt concentration. Seed vigour is a physiological property which governs the capability of a seed to produce a seedling quickly in soil and the extent to which that seed tolerate a different environmental factors (Perry, 1980). Perry (1972b) concluded that seeds which have high vigour are more tolerant to environmental stress. The deleterious effects of salinity were further confirmed by analyzing vigour index.

The outcome of this study showed that the maximum percentage germination was recorded in control conditions and minimum at 150mM salinity level; however, no germination occurred at higher salinity levels (200mM) in some varieties. Increased salinity caused a significant reduction in germination percentage, germination rate, root and shoot length and fresh weight of root and shoot (El-Shaieny, 2015). Growth arrest in plants during the early stage of salt stress is mostly due to salt-induced osmotic stress (Sobhanian *et al.*, 2010, Munns, 2011, Carassay *et al.*, 2012). Salt stress affects not only oat plant morphological, physiological processes but also germination of seed, growth and water/nutrient uptake (Willenborg *et al.*, 2004). (Tajbakhsh *et al.*, 2006) reported that selection of crop plants for salinity stress at germination stage is essential for determining salt tolerance potential of crops, since this stage mostly determines crop establishment. (Balkan *et al.*, 2015) also observed that

germination is moderately delayed by salinity stress and this may be the result of seed having a lower water uptake at high salinity levels due to osmotic stress.

Similarly, with increasing salinity seed vigor index also declines (Table 6). These findings also correlated with the findings of (Kader and Jutzi 2004) and they observed that with increase in salinity levels seed characteristics were decreased. (Keshavarzi *et al.*, 2011) also found the similar results and reported the impact of ions on declining the germination parameter.

There was a significantly decreased in the result of the root, shoot length of the studied plant under salt stress (Table 3 and Table 4). Length of root and shoot decreases with the increase in salinity level that may be due to slowing down the water absorption by the plants (Demir and Arif, 2003). Moreover, (Jamil *et al.*, 2006) observed that salt stress inhibited largely plant growth and this may be because of reduced supply of metabolites to young tissues because the production of this metabolite is significantly disturbed under the salt stress, either due to toxic effect of NaCl or low uptake of water. During salinity stress, less affected roots provide sufficient water and nutrients.

In this experiment the seedling fresh and dry weight of oat plant were decreased linearly with increasing salinity levels (Table 5 and Table 6). Decrease in fresh weight and dry weight has been seen in all plant tissues subjected to salt stress, but it is particularly noticeable in the aerial part. Salinity decrease plant growth through osmotic and toxic effects, and high sodium uptake ratio values cause sodicity, which increases soil resistance, decreases root growth, and reduces water movement through the root with a decrease in hydraulic conductivity (Table 7 and 8).

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