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ORIGINAL ARTICLE

Effect of quantitative and qualitative performance of four canola cultivars (*Brassica napus* L) to salinity conditions

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ABSTRACT

In order to studying the salinity stress effects resulted from sodium chloride on germination, vegetative growth, elements concentration and proline accumulation of four canola cultivars two experiments were conducted in germinator and greenhouse attached in the Center of Agriculture and Natural Resources Research in East Azarbaijan. The experimental designs were Completely Randomized Design and Randomized Complete Block arrangement in factorial with four replications. The first factor was four canola cultivars included; Licord, Okapi, Elite, SLM_{046} and second factor was consist of different salinity stress levels included 0, 50, 100, 150 and 200 mM NaCl in germination experiment and 0, 75, 150, 200, 250 and 300mM NaCl in pot experiment. The results demonstrated that, different salinity stress levels had significant effect on germination percentage, germination speed and shoot and root length. Also, in pot experiment there was significant effect on plant height, leaf area, dry matter percentage, and seed yield due to salinity stress. In addition, it observed that, there was significant different among canola cultivars traits except leaf area trait. Comparison of means showed that, response of canola cultivars were different at germination and vegetative growth stages. In germination experiment, the most sensitive and most tolerate cultivars were Elite and Licord, respectively. But in pot experiment, Licord cultivar had least growth and yield than SLM_{046} and Okapi. also it observed that, there was significant different among these cultivars. Thereupon, it seems that, evaluation of germination properties is not helpful for assessment of salinity tolerance in canola cultivars.

Key words: Canola, Germination, Salinity stress, Vegetative growth

Introduction

Salinity is one of the major abiotic stresses in arid and semi-arid regions that substantially reduce the average yield of major crops by more than 50%[1]. Salinity affects 7% of the world's land area for around 930 million ha [22]. Salinity is one of the most serious factors limiting crops production, especially the sensitive one's [3]. Currently, high soil salinity affects the agricultural production in a large proportion in the world's territorial areas [4]. Although salt stress affects all growth stages of a plant, seed germination and seedling growth stages are known to be more sensitive in most plant species[5]. Furthermore, germination and seedling stage is predictive of plant growth responses to salinity[5,6]. Therefore, seeds with more rapid germination under salt stress and/or normal conditions may be expected to achieve a rapid seedling establishment and more salt tolerance, resulting in good stand establishment and hence higher yields [2]. Salt tolerance in plants is a

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complex phenomenon, which depends on a number of inter-related factors based on morphological, biochemical and physiological processes [7]. Salinity reduces the ability of plants to take up water, leading to growth reduction as well as metabolic change similar to those caused by the water stress [2]. High salt concentration in root affects the growth and yield of many important crops. The salinity may reduce the crop yield by upsetting water and nutritional balance of plant[9]. Water availability and nutrient uptake by plant roots is limited because of high osmotic potential and toxicity of Na and Cl ions [10]. Saline soils and saline irrigation waters present potential hazards to canola production. Germination failures on saline soils are often the results of high salt concentrations in the seed planting zone because of upward movement of soil solution and subsequent evaporation at the soil surface. The most common adverse effect of salinity on the crop of Brassica is the reduction in plant height, size and yield as well as deterioration of the product quality [11]. Seed germination has been reported to decline with increasing salinity levels [12]. There are differences in sensitivity to salinity among canola cultivars. The genetic role in seed germination resistance to salinity is probably one of the most important advantages that can be used in breeding programs. Significant variation in seed germination among canola cultivars grown under salinity condition is widely reported by Zheng *et al*[14] and Puppala *et al*[13].

The present study was undertaken to assess the effect of salt stress on some characteristics of four canola cultivars such as seed germination, seedling growth as well as some morphological traits and yield and yield components. The tolerance of canola to salinity during germination however has not been widely reported. Therefore, the objective of this study was to evaluate the germination response of canola cultivars to different levels of salinity, as well as their interactions.

Material and methods

Germination experiment

Four canola cultivars (*Brassica napus* L. c.v's Licord, Okapi, Elite, SLM_{046}) were collected from Seed and Plant Improvement Institute (SPII) Karaj, Iran. The same size seeds were surface-sterilized for 5 min in sodium hypo chloride solution (10%) and then were rinsed with distilled water for 3 to 5 times. After sterilization, 25 seeds were transferred into 9-cm sterile Petri dishes on filter paper and then were wetted with 7 ml distilled water (control) or saline water solution at 0, 50, 100, 150 or 200 mM NaCl. To prevent infection and evaporation of solution, all of the plates were closed with parafilm. All operations were performed under laminar flow. The Petri dishes were labeled and incubated in a germinator at 25°C and 18/6 h day/night illumination.

Computation of germinated seed was done daily until end of seventh day. After that five germinated seed was removed and morphological traits of them were assayed. Germination percentage and germination speed were calculated according to equation 1 and 2, respectively.

Equation1:Germination percentage $= \frac{n}{N} \times 100$

Where, n: Number of germinated seed at seventh day; N: number of seeds

Equation 2: Germination speed $= \frac{\sum Dn}{\sum n}$

Where, n: Number of germinated seed at lightly day; D: days after beginning

Pot experiment

The seeds of above mentioned cultivars were surface-sterilized for 5 min in sodium hypo chloride solution (10%) and then were rinsed with distilled water for 3 to 5 time. Then five seeds of canola were sowed in each plastic pot (35 cm height and 30 cm diameter) contain perlite and vermiculite (1:1) at depth of 1 cm.

The bottom of each pot was delved for drainage of extra water. Pots were transferred to glasshouse under conditions of 25/18°C day/night temperature and natural light. The pots were irrigated at field capacity level by nutrient solution (Hoagland's solution). After full germination; the number of plants was reduced to two seedlings per pot.

Salinity stress induction was done when fourth leaf was completely expanded. Different concentration of NaCl solution was added to each pot gradually.

In order to vernalization of plants, at early of January the pots were transferred to outdoor during 60 days at 5°C.

Morphological traits, yield and yield components

At physiological maturity stage, plant height, leaf area, number of silique in plant, number of seed in silique, 1000 seed weight and final yield were measured. In order to evaluate dry matter, harvested plants were oven-dried at 70°C for 48 h to constant weight. Dry matter percentage was calculated according to equation 3.

Equation 3: Dry matter percentage $= \frac{d}{w} \times 100$

Where, d: dry weight and w: fresh weight

Experimental design and analysis of data

The experimental designs were Completely Randomized Design for germination experience and Randomized Complete Block for pot experiment. The treatments were arranged in factorial with four replications. All data were analyzed by SPSS software and Duncan's Multiple Range Tests was used to measure statistical differences between treatment methods and controls ($p<0.05$).

Results and discussion

Germination experiment

Germination percentage and speed

The results demonstrated that effects of cultivars and salinity were significant on shoot length, root length, germination percentage and germination speed (Table 1). Interaction effect between cultivar and salinity was significant on germination percentage and germination speed. It shows that, different levels of salinity had different effect on these traits, and there were differences among cultivars. According to this result Elite cultivar had the lowest germination percentage and germination speed than other cultivars (Table 2). Thus this cultivar was known as sensitive cultivar to salinity stress. There was not significant different between SLM_{046} and Licord in aspect of germination parentage and germination speed but Licord cultivar was introduced as a tolerant cultivar (Table 3). Comparison of means showed that, reduction of germination percentage was significant at 150 and 200 mM and there was not any difference among other salinity levels such as 50 and 100 mM. Also, germination speed was decreased when salinity level was raised. The highest germination speed was related to control treatment and the lowest was related to 200 mM treatment (Table 3). On base of these results, germination percentage is more resistant and germination speed is more sensitive factor to salinity. The results are similar to findings of Moss and Hoffman[15]; they have reported that, high salinity level leads to ion imbalance, osmotic regulation disorders and finally decrease in water absorption by seeds.

Root and shoot length

It observed that, effect of salinity and cultivar were significant on root and shoot length (Table 1). The Licord had the highest root and shoot length. While there was no difference among other cultivars on root and shoot length. Comparison of means showed that enhancement of salinity decreases shoot and root length in all of cultivars (Table 3). The lowest shoot length and root length were related to 100, 150 and 200 mM and 150 and 200 mM, respectively (Table 3). In this study, it seems that, root length is more affected by salinity than shoot length. Findings of EL-Melegi and El-sayed *et al*., [16] on tomato under salinity stress showed that, dry weight of plants had positive correlation with root length and these plants were more tolerate to salinity stress. Also similar results were reported by François and Bahizire[17] regarding canola under salinity stress.

Yield and yield components

Salt stress had significant effect on yield and yield components. Interaction between cultivar and salinity stress was significant on yield and yield component too (Table 4). There was a decrease in yield components due to increment of salinity stress (Table 6). The results showed that, there was significant difference among cultivars in aspect of number of silique in plant, number of seed in silique, 1000 seed weight and seed yield (Table 5).

The highest and the lowest seed yield were achieved from SLM046, Okapi, Licord and Elite, respectively (Table 5). The SLM046 had high phenotypic value as this cultivar was better than other cultivars in aspect of number of silique in plant, number of seed in silique and 1000 seed weight (Table 5).Comparison of means demonstrated that, the highest silique per plant was related to control treatment and the lowest silique was observed in 300 mM salinity level (Table 6). The most lose of silique number was obtained from 150 mM salinity to up levels (Table 6). Already, it has been reported by Lin [18] that, decrease in slique number is associated to increase of ABA and pollen death. In canola plants, time of flowering is a critical stage, in other hand, salinity stress decreases growth period, consequently, plants decreases silique number to attain its survival. According to results of Sinaki *et al* [19] salinity stress at flowering stress, decreases silique number. With consider on these results, it seems that, the most important reason for silique number reduction is low tolerance of canola plants to low salinity level. Moss and Hoffman [15] have been reported that, salinity stress at 10 ds.m-1 is threshold of salinity tolerance in canola.

Comparison of means of seed number in silique showed that, salinity stress decreased significantly seed number (Table 6). The lowest salinity level had not difference with control treatment also 150 mM and 200 mM salinity level had not significant different with each other. Decrease in seed number was started from 200 mM salinity level. According to results, it seems that, one of the reasons of seed number decrease is silique size reduction. Sakr *et al* [20] have been reported that, major of growth parameters such as seed number are decreased by

Table 1: Analysis of variance on germination traits under salinity stress

S.O.V	df	Mean square			
		Shoot length	Root length	Germination percentage	Germination speed
Cultivar	3	20.673**	36.265**	3767.700**	1.230**
Salinity	4	42.492**	116.468**	3283.731**	0.121**
Cultivars × Salinity	12	6.531ns	6.023ns	480.523**	0.008**
Error	57	6.479	7.541	38.758	0.0001
C.V		44.2	33	8.18	1.02

*, ** significant at the 0.05 and 0.01 probability levels, respectively. ns, not significant

Table 2: Comparison of main effect of cultivars on germination traits

Cultivar	Shoot length (cm)	Root length (cm)	Germination percentage	Germination speed
SLM_{046}	3.6050b	6.1425b	89.0000a	0.6595b
Okapi	3.7430b	5.8530b	72.0000b	0.3755c
Licord	5.3990a	8.3690a	89.8000a	0.8795a
Elite	3.0338b	5.3053b	53.4000c	0.3630d

For a given means within each column followed by the same letter are not significantly differences ($p < 0.05$).

Table 3: Comparison of main effect of salinity levels on germination traits

Salinity levels	Shoot length (cm)	Root length (cm)	Germination percentage	Germination speed
0 mM NaCl	6.2263a	9.8600a	91.0a	0.6819a
50 mM NaCl	5.1312a	8.0431ab	90.68a	0.6175b
100 mM NaCl	2.8506b	6.5656b	88.77ab	0.5694c
150 mM NaCl	2.7466b	4.5147c	58.25b	0.5231d
200 mM NaCl	2.7712b	3.1037c	53.0c	0.4550e

For a given means within each column followed by the same letter are not significantly differences ($p < 0.05$).

Table 4: Analysis of variance on yield and yield component under salinity stress

S.OV	df	Numberof silique in plant	Number of seed in silique	1000 seed weight	Seed yield	Plant height	Leaf area	Dry matter
Cultivar	3	298.6**	70.9**	7.9**	19.5**	204.7**	1123.7ns	68.2**
Salinity	5	11704.8**	688.7**	29.6**	223.3**	1606.9**	61411.1**	77.1**
Replication	3	6.7ns	3.6*	0.02ns	0.336ns	115.0*	1038.3ns	36.8**
Cultivars× Salinity	15	16.7**	2.7**	0.365**	2.0**	13.6ns	545.1ns	1.7ns
Error	69	2.90	0.935	0.008	.175	40.1	1346.9	2.4
C.V		6.9	8.6	4.7	9.7	16.8	25.9	14.6

*, ** significant at the 0.05 and 0.01 probability levels, respectively. ns, not significant

Table 5: Comparison of main effect of cultivars on yield and yield component

Cultivar	Number of silique in plant	Number of seed in silique	1000 seed weight	Seed yield	Plant height	Leaf area	Dry matter
SLM_{046}	29.75a	13.16a	2.58a	5.50a	40.25a	150.87a	17.79a
Okapi	23.77b	12.14b	2.15b	4.37b	39.04a	134.70a	16.08b
Licord	24.22b	9.97c	1.60c	3.82c	37.79a	141.00a	15.25b
Elite	21.41c	9.58c	1.29d	3.43d	33.54b	139.00a	13.75c

For a given means within each column followed by the same letter are not significantly differences ($p < 0.05$).

Table 6: Comparison of main effect of salinity levels on yield and yield componen

Salinity levels	Number of silique in plant	Number of seed in silique	1000 seed weight	Seed yield	Plant height	Leaf area	Dry matter
0 mM NaCl	66.3 a	19.1a	3.7a	10.3a	56.2a	256.2a	19.2a
75mMNaCl	50.3 b	18.5 a	2.9 b	7.3 b	38.8b	156.7b	16.9b
150 mMNaCl	18.1 c	10.9 b	2.4c	3.3c	37.9b	125.2c	16.0b
200 mMNaCl	7.37d	10.2 b	1.4 d	1.8 d	34.4bc	125.3c	14.5c
250 mMNaCl	4.03e	4.81 c	0.69 e	1.5 d	30.2cd	77.5	14.6c
300 mMNaCl	2.46f	3.6 d	0.204	1.2e	28.1d	107.3c	13.0d

For a given means within each column followed by the same letter are not significantly differences ($p < 0.05$).

salinity stress in addition findings of Mendham and Salisbury[21] shows that, salinity stress decreases seed setting in canola plants. There are not significant differences between first level and 150 and 200 mM salinity level, because number of seed in silique is less sensitive to salinity stress at low salinity level.

It observed that, 1000 seed weight was decrease due to salinity stress (Table 6). Comparison between control treatment and each salinity stress showed that, there is significant difference between them (Table 6). Among salinity stress levels, the highest 1000 seed weight was related to 75mM salinity level (Table 6). There was gradual reduction in seed weight at low salinity level but at high levels it was increased (Table 6). Decrease in seed weigh can be due to prevention of assimilate transport to the seeds and decrease in growth during seed filling stage.

Munns *et al*., [22] have been showed that, when barley plants were exposure to salinity stress there was many disorders in reproductive stages. Gradual decrease in seed weight at low salinity level than high salinity level is because of low sensitivity of canola to salinity at vegetative growth stage. Canola is sensitive to salinity at seedling and early of vegetative stage and this sensitivity will be decrease at end of growth stage such as seed filling stage[23]. Comparison of means among salinity treatment showed that, the lowest and the highest seed yield were achieved from 300 mM salinity stress level and control treatment, respectively (Table 6). Decrease in yield components due to salinity stress lead to lose of final yield. It seems that, ions accumulation in plant tissues at different growth stage is main reason of lose yield. According to results of 1000 seed weight and number of silique in plants, it conclude that, these parts of yield components are more sensitive to salinity and decreased in final yield is related to decrease of them. Sana *et al*., [24] have been reported that, number of silique in plan is critical component to determine of yield. Also, Engqvvist beker [25] showed that, seed weigh and numbers of silique are more important component fro select of high yield genotypes.

Conclusion

1. The results of this study showed that although increasing salinity in both germination and vegetative growth phase on the negative effects of rapeseed, but salt sensitivity and tolerance for each of these two figures was not the same. So that the figure Licord and Okapi, respectively Although the highest record in the germination stage salinity tolerance than the other two varieties have accounted for, but the vegetative and reproductive growth stage, although superior varieties Elite but were unable at this stage in terms of concentration and transfer of mineral elements and the accumulation of proline and practice, to compete with SLM046 figure. Therefore, the results will be concluded that the amount of salt tolerance at germination stage communication with plant tolerance level is no later.
2. SLM046 suitable varieties cultivated areas at the beginning of the growth period of canola quality of irrigation water salinity is considered appropriate. Because this figure was sensitive to salinity at germination stage, but the end stage of the study attributes the growth of the charge has shown a high yield.
3. and figures Licord priority and Okapi, respectively, suitable cultivation areas in all that period of growth with low quality water resources is enjoyed. Because sensitivity and salt tolerance of these two cultivars for both stages was somewhat similar.

4. Elite figure and low yield due to both the current stage without further investigation, to be planted in area salinty are not economical and it is better not recommended.

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