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INTER AND INTRA SPECIFIC VARIATION FOR SALT TOLERANCE IN TURNIP (*BRASSICA RAPA* L.) AND RADISH (*RAPHANUS SATIVUS* L.) AT THE INITIAL GROWTH STAGES

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Abstract

Exploration of inter- and intra-specific variation for salt tolerance is a pre-requisite for the improvement of this trait through selection and breeding. In order to assess, inter- and intra-specific variation for salt tolerance in radish (*Raphanus sativus* L.) and turnip(*Brassica rapa* L.), local cultivars of these two species were screened at varying levels of salinity stress (0, 60, 120, 180, and 240 m*M*) at the germination and seedling stages. Increasing level of salinity caused a marked reduction in germination percentage and fresh and dry biomass of the seedlings of all cultivars of both species. Both species displayed highly significant intra-specific variation in seed germination and seedling growth at higher levels of salt. However, the degrees of salt tolerance at the germination and seedling stage varied in both species. Of turnip cultivars, Shaljum desi surakh was highest in seed germination, while it was lowest in seedling shoot dry biomass production. However, Neela Shaljum having lower seed germination percentage produced maximum seedling shoot dry biomass. In the same way, cv. Desi of radish with minimum seed germination had highest shoot dry weight under saline conditions. Although a considerable magnitude of variation for salt tolerance was observed in a set of available local cultivars of both species at the germination and seedling stages, the performance of cultivars of both species was not consistent across different stages, thus this needs to be further investigated at later growth stages. On the whole, radish was found to be tolerant as compared to turnip in terms of tolerance index worked out using overall germination percentage and seedling shoot dry weight.

Introduction

Salt accumulation is a consistent process in the soils of arid and semi-arid regions of the world. However, besides improving water management practices to reduce the salt accumulation in plant root zone, there is a need to improve salinity tolerance of prospective crops. A way-forward to achieve this, is the exploration of inter- and intra-specific variation for salt tolerance.

Although a survey of salt tolerance of crops, vegetables and fruit trees is available on http://www.ussl.ars.usda.gov, the data generated by USDA Salinity Laboratory presented only few cultivars of a species with specific region; however, it shows a wide range of salt tolerance in crops, vegetables and fruit tree species. Furthermore, considerable genetic diversity at intra-specific level for salt tolerance has already been observed in different species such as wheat (Kingsbury & Epstein, 1984)**,** lentil (Ashraf & Waheed, 1993), barley (Belkhodja *et al*., 1994), cotton (Ashraf & Ahmad, 1999) pea (Noreen *et al*., 2007) and *Brassica napus* (Ulfat *et al*., 2007). In view of this information, it is suggested that although considerable magnitude of genetic variation occurs at inter- and intra-specific level, it has not been fully explored and exploited, particularly in local cultivars of most crops. With this fact in mind, it is imperative to explore intra-specific (inter-cultivar) and inter-specific variation for salt tolerance of turnip and radish by screening their available local germplasm.

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The principal objective of conducting this study was to determine variations in degree of salt tolerance at inter- and intra- specific levels particularly at the germination and seedling stages.

Materials and Methods

An experiment was conducted in the growth room of the Department of Botany, University of Agriculture, Faisalabad, Pakistan to assess intra- and inter- specific variation for salt tolerance in two potential vegetable crops viz., radish (*Raphanus sativus* L.) and turnip (*Brassica rapa* L.). For this purpose, 6 varieties of radish and 5 of turnip were used in the experiment. Their names and sources of origin are mentioned in Table 1. Three hundred seeds of each cultivar were surface sterilized in 5% Sodium hypochlorite solution for 5 min and then carefully rinsed with distilled water to remove the sterilizing agent. Five different levels of NaCl i.e., 0, 60, 120, 180 and 240 m*M* were used. Fifty seeds of each cultivar were allowed to germinate in each Petri plate double lined with a sterilized filter paper moistened with 10 mL of Hoagland’s nutrient solution with or without appropriate levels of NaCl. The treatment solution in each Petri plate was changed every day so as to ensure the desired salt level. Germination started after two days of sowing and a seed was considered germinated when the radicle emerged up to 5 mm in length. The data for germination was recorded daily up to day 15 of the start of the experiment after which time the experiment was terminated. After 15 days of the start of the experiment, plant seedlings were removed carefully from the Petri plates and separated into shoots and roots. After recording fresh weights, the plant samples were oven-dried at 65°C for five days and their dry weights measured.

Statistical analysis of data: A completely randomized design (CRD) with four replicates was used for data analysis using the COSTAT computer package (*CoHort software*, Berkeley, USA). For comparing the means, the Least Significance Difference (LSD) test (Snedecor & Cochran, 1980) was used.

Results

(I) Radish: Analysis of variance of the data for germination percentage of all radish cultivars shows that salt stress markedly suppressed the total germination percentage (Table 2). However, a significant inter-cultivar variation for this attribute was observed in the set of radish cultivars examined in the present study when grown under both normal and saline conditions (Fig. 1). Of all cultivars, Radish 40 days had the maximum germination percentage, while the minimum in Desi under saline conditions (Fig. 1).

Shoot fresh and dry weights of all radish cultivars were reduced significantly ($P \leq 0.001$) due to salt stress (Table 2). Cultivars differed significantly in both these growth attributes (Fig. 3). Cultivar Desi followed by cv. Mannu early was the least affected by salt stress in terms of shoot fresh and dry biomass as compared to the other cultivars. The worst hit cultivar in these attributes was Radish 40 days.

Although salt stress caused a significant reduction in root fresh and dry weights of all cultivars, the degrees of salt tolerance were not consistent in the two attributes (Fig. 3). However, cvs. Desi, Mannu early and Minu japani had higher root dry biomass than the other cultivars under saline conditions.

Table 1. Sources of origin of two root crops used in the experimentation.

Crop Name	Sr.#	Cultivar	Source
Radish	1	Radish red neck	AARI, Fsd
	2	Radish lal pari	AARI, Fsd
	3	Radish mino japani	AARI, Fsd
	4	Radish 40 days	AARI, Fsd
	5	Mannu early	Local market
	6	Desi	Local market
Turnip	1	Shaljum desi surakh	AARI, Fsd
	2	Shaljum purple top	AARI, Fsd
	3	Shaljum golden bal	AARI, Fsd
	4	Neela shaljum	Local market
	5	Peela shaljum	Local market

Table 2. Mean squares from analysis of variance (ANOVA) of data for germination percentage, shoot fresh wt., shoot dry wt., root fresh wt. and root dry wt. (mg/seedling) of radish seedlings grown under varying levels of NaCl (Mean ± S.E; *n* = 4).

Source of variation	df	Germination percentage	Shoot fresh wt.	Shoot dry wt.	Root fresh wt.	Root dry wt.
Main effects						
Varieties	5	1801.7 ***	4007.8 ***	68.8 ***	243.1 ***	1.22 ***
Salinity	4	1895.4 ***	18795.0 ***	27.1 ***	2440.3 ***	0.88 ***
Interaction						
Varieties x Salinity	20	102.5 ***	630.2 ***	0.58 *	64.6 ***	0.027 ^{ns}
Error	90	10.8	72.05	0.28	6.52	0.018

*, *** = Significant at 0.05 and 0.001levels, respectively.

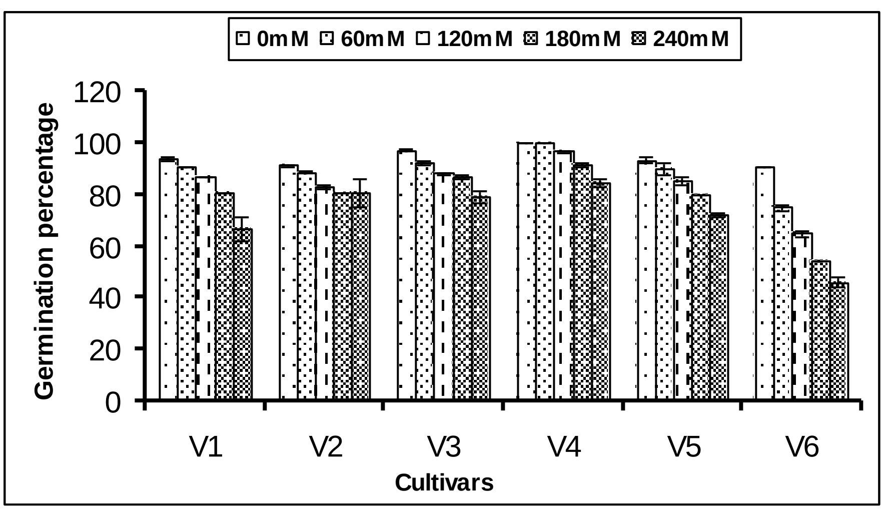

Fig. 1. Germination percentage of radish (*Raphanus sativus* L.) under varying levels of NaCl. (V1=Radish red neck, V2=Radish lal pari, V3=Radish mino jupani, V4=Radish 40 days, V5=Mannu early, V6=Desi)

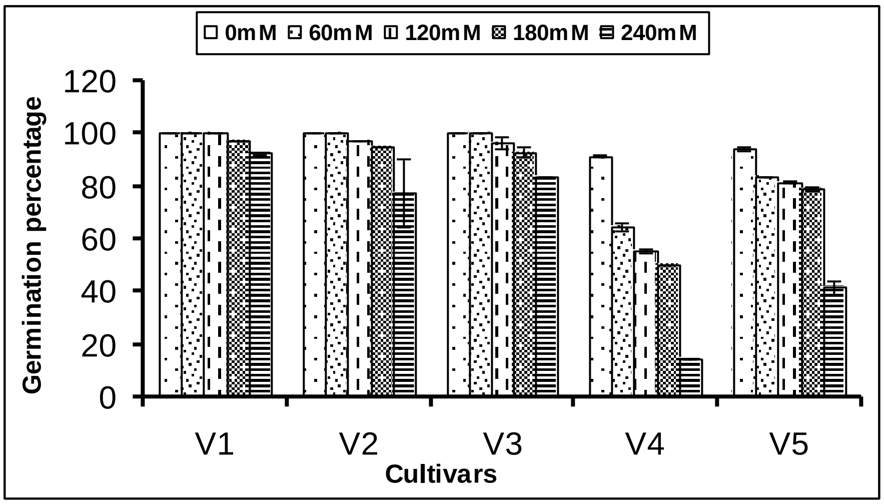

Fig. 2. Germination percentage of turnip (*Brassica rapa* L.) under varying levels of NaCl.
(V1=Shaljum desi surakh, V2=Shaljum purple top, V3=Shaljum golden bal, V4=Neela shaljum, V5=Peela shaljum)

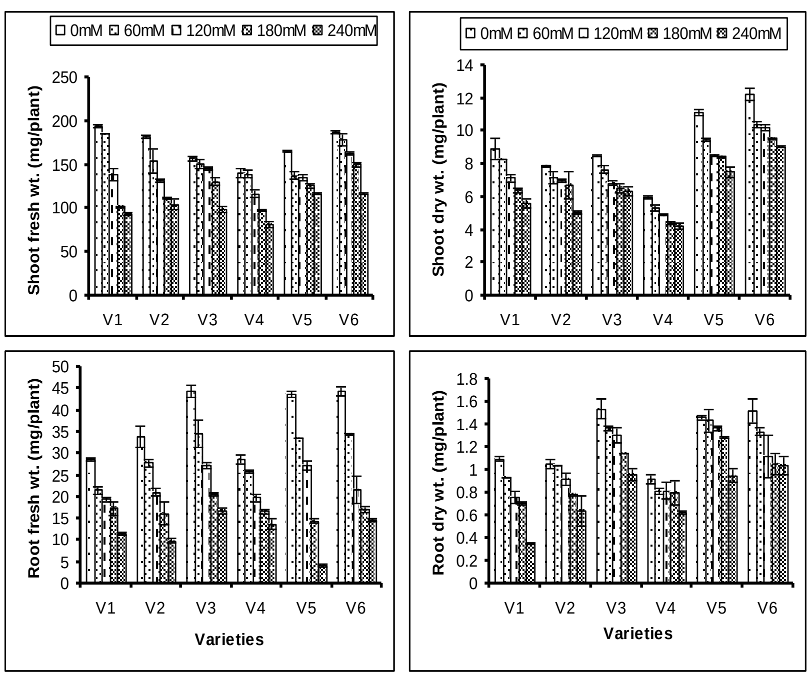

Fig. 3. Effect of different salinity levels on shoot fresh wt., shoot dry wt., root fresh wt. and root dry wt. of six varieties of rdish (*Raphanus sativus* L.).
(V1=Radish red neck, V2=Radish lal pari, V3=Radish mino japani, V4=Radish 40 days, V5=Mannu early, V6=Desi)

(II) Turnip: Germination percentage of all turnip cultivars was decreased significantly due to salt stress ($P \leq 0.001$). Maximum germination percentage was observed in Shaljum desi surakh followed by Shaljum golden bal under highest salinity level. In addition, most of the cultivars were less affected by salt stress at mild level except cv. Neela shaljum (Fig. 2).

Both shoot fresh and dry weights of the seedlings of all turnip cultivars were reduced significantly at varying levels of NaCl ($p \leq 0.001$). However, maximum reduction in shoot biomass was observed at the highest level. Of all cultivars, Neela shaljum produced highest shoot fresh and dry weights at 240 m*M* of salt, but the lowest shoot fresh and dry weights were recorded in cv. Shaljum desi surakh (Table 3; Fig. 4).

A marked reduction in root fresh and dry weights of the seedlings of all cultivars of turnip was observed due to salt stress. A considerable variation in the five turnip cultivars was also observed with respect to root fresh and dry weights (Table 3; Fig. 4). Peela shaljum and Neela shaljum were more sensitive to salt with respect to these two attributes.

Discussion

The main objective of the present study was to examine the extent of inter- and intra-specific variation in radish and turnip by screening available local germplasm, as a first step to identify specific salt tolerant traits at earlier growth stages and future efforts for breeding for salt tolerant cultivars. In the present study, salt stress adversely affected the germination percentage, and seedling growth of all cultivars of both radish and turnip, particularly at the highest level of salt stress. However, considerable variation in salt tolerance was observed among all cultivars of both radish and turnip with respect to seedling shoot biomass or germination percentage. From the results of the present study, it was obvious that Shaljum desi surakh (turnip) salt tolerant at the germination stage was salt sensitive at the seedling stage. Similarly, Neela Shaljum was salt tolerant at the seedling stage, while it was found sensitive at the germination stage. In the same way, 'Radish 40 days' salt tolerant at the germination stage was salt sensitive at the seedling stage. Thus, degree of salt tolerance in both species i.e., turnip and radish varied at these two growth stages. These findings are similar with those of Mano & Takeda (1997) who found a continuous and wide variation among 1300 wheat cultivars and Bayuelo-jimenez *et al*., (2002) who reported that inter-cultivar variation existed in *Phaseolus* species at the germination and seedling stage. Thus, the degrees of salt tolerance in both *Brassica* species were not consistent at the two early growth stages.

Although increasing levels of salinity reduced the germination and seedling growth of both species, the adverse effects of salt stress were less on germination of turnip at intermediate level of salinity. However, highest level of salt stress was very effective in discriminating both turnip and radish cultivars based on either germination or seedling biomass production. From the results of ranking of both turnip and radish cultivars based on germination and fresh and dry weight of shoots and roots, all cultivars of both turnip and radish showed inconsistent pattern in terms of salt tolerance ranking (Table 4). This finding also confirms that degree of salt tolerance at these two growth stages varies in both turnip and radish. These results are in accordance with findings of Van Hoorn (1991) who found that early seedling growth of safflower, sorghum, sunflower and wheat appear to be less tolerant than germination and later growth. But it is not in conformity with the findings of Ashraf & Waheed (1993), who found that the salt tolerant lines identified out of 133 accessions of lentil at the initial growth stages maintained their degree of salt tolerance at the later growth stages. In view of data for tolerance index worked out using overall germination percentage and seedling shoot dry weight of both species (Fig. 5), radish was found to be tolerant as compared with turnip.

Table 3. Mean squares from analysis of variance (ANOVA) of data for germination percentage, shoot fresh wt., shoot dry wt., root fresh wt. and root dry wt. (mg/seedling) of turnip seedlings grown under varying levels of NaCl (Mean ± S.E; *n* = 4).

Source of variation	df	Germination percentage	Shoot fresh wt.	Shoot dry wt.	Root fresh wt.	Root dry wt.
Main effects						
Varieties	4	6486.9 ***	121.6 ***	2.39 ***	8.63 ***	0.038 ***
Salinity	4	3479.9 ***	3485.8 ***	0.66 ***	90.98 ***	0.12 ***
Interaction						
Varieties x Salinity	16	450.7 ***	41.8 ***	0.031 ***	0.71 *	0.0019 **
Error	165	29.8	9.81	0.007	0.33	0.00086

*, **, *** = Significant at 0.05, 0.01 and 0.001 levels, respectively.

Table 4. Ranking of the two vegetable crops on the basis of their salt tolerance indices.

Crop name	Parameter	Sensitivity	Limits	Cultivars
	Germination %			
Turnip		Tolerant	≥ 0.90	Shaljum desi surakh, Shaljum golden bal, Shaljum purple top
		Moderate	0.7 to 0.9	Peela shaljum
		Sensitive	≤ 0.7	Neela shaljum
Radish		Tolerant	≥ 0.90	Radish 40 days, Radish lal pari
		Moderate	0.7 to 0.9	Radish mino japani , Radish red neck, Mannu early
		Sensitive	≤ 0.7	Desi
	Shoot dry weight			
Turnip		Tolerant	≥ 0.80	Neela shaljum, Shaljum purple top, Peela shaljum
		Moderate	0.6 to 0.80	Shaljum desi surakh
		Sensitive	≤ 0.6	Shaljum golden bal
Radish		Tolerant	≥ 0.80	Radish lal pari
		Moderate	0.76 to 0.8	Radish mino japani, Desi, Radish 40 days
		Sensitive	≤ 0.76	Radish red neck Mannu early

From the results of the present study, it is clear that although a considerable magnitude of variation for salt tolerance was observed in the available local cultivars of radish and turnip, degree of salt tolerance varied at two early growth stages. Thus, it is suggested that screening and selection for salt tolerance in turnip and radish should be carried out at all growth stages.

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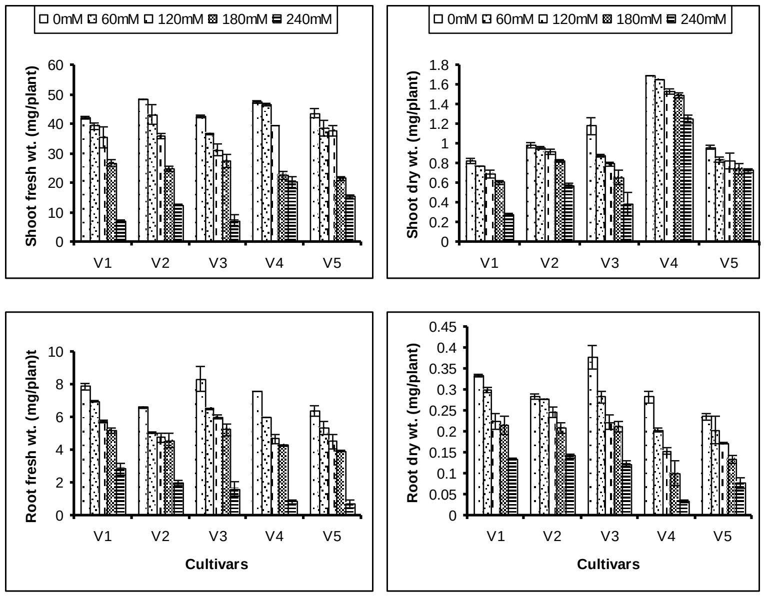

Fig. 4. Effect of different salinity levels on shoot fresh wt., shoot dry wt., root fresh wt. and root dry wt. of five turnip (*Brassica rapa* L.) varieties. (V1=Shaljum desi surakh, V2=Shaljum purple top, V3=Shaljum golden bal, V4=Neela shaljum, V5=Peela shaljum).

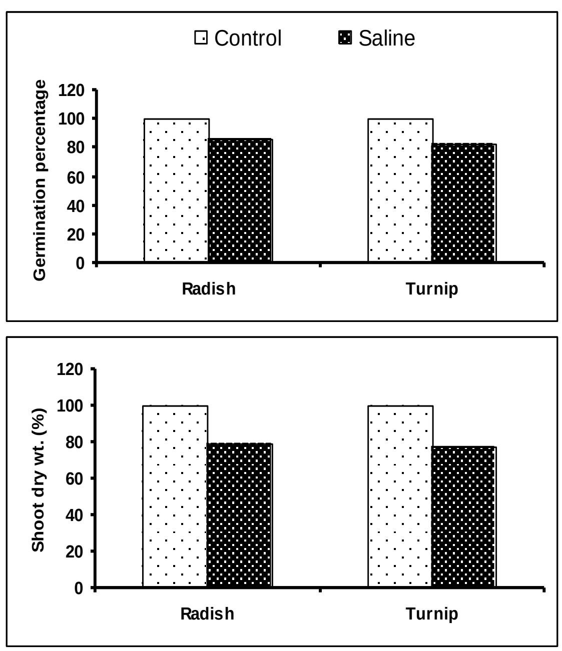

Fig. 5. Inter-specific variation for salt tolerance in turnip and radish with respect to overall germination percentage and shoot dry weight at the early seedling stage.

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