

The Effects of Salt Stress in Zinnia (*Zinnia* sp.) Cultivars During Seed Germination and at the Early Stages of Seedling Growth

Sara YASEMİN1*, Ayşin GÜZEL DEĞER², Nezihe KÖKSAL³

¹Siirt University, Faculty of Agriculture, Department of Horticulture, Siirt, TURKEY ²Mersin University, Vocational School of Technical Sciences, Department of Food Processing, Mersin, TURKEY ³Cukurova University, Faculty of Agriculture, Department of Horticulture, Adana, TURKEY

 Received: 13.03.2020
 Accepted: 20.10.2020

 ORCID ID (By author order)
 Image: Control org/0000-0003-2193-6791

 Image: Control org/0000-0003-2193-6791
 Image: Control org/0000-0002-5401-9730

 'Corresponding Author: sara.yasemin@siirt.edu.tr

Abstract: The Zinnia genus which belongs to the Asteraceae family is an annual, multipurpose ornamental plant. Zinnia plants are cultivated not only in landscape but also as the potted plant and cut flower. One of the most important problems in the world is salinity in soil and water. The aim of this study was to determine the effects of salinity on twenty Zinnia cultivars during seed germination and early seedling growth. The salt was applied by irrigating seeds with 0 and 100 mM salt solutions. Radicle emergence, seed germination, root, hypocotyl and cotyledon lengths, relative growth index of the root, and seedling fresh weight were evaluated. At the end of the study, the radicle emergence reduction was the highest value in Zinnita Rose (52%). The highest reduction rates on seed germination were at Swizzle Cherry-Ivory, Double Zahara Raspberry Ripple (48%), and Double Zahara Yellow (48%). Root lengths of all Zinnia cultivars were dramatically decreased by salt stress. According to weighted ranked evaluation, Dreamland Ivory and Dreamland Coral were more tolerant to salt stress than other Zinnia cultivars. However, in general Zinnia cultivars were relatively sensitive to salt stress at the germination stage.

Keywords: Asteraceae, cotyledon, germination, hypocotyl, salinity, ornamental plants

1. Introduction

Soil salinity and water scarcity are one of the most forceful abiotic stresses that limit agricultural yield and productivity. Water scarcity is the result of global warming and excessive consumption. Soil salinity increases with many factors such as overfertilization and irrigation with saltwater. especially in arid and semi-arid regions (Koksal et al., 2014; Yasemin et al. 2017; Hannachi and Van Labeke, 2018). In parallel, the reduction of potable water resources leads to using alternative wastewater containing salt for agricultural growing processes. As a result of these inferences, the necessity to select salinity resistant plants have emerged (Koksal et al., 2016; Rámila et al., 2016; Feng et al., 2018; Liu et al., 2018).

Salt stress affects plants at different rates in different stages of development. Determining the

threshold of salt damage at all developmental stages is an essential step for the studies of salt tolerance during different growth phases (Zapata et al., 2004; Parida and Das, 2005; Hannachi and Van Labeke, 2018). Seed germination, which is the beginning of the life cycle of the plant, is of great importance for the future vital activities of the plant. Many plants need favorable conditions, mostly non-saline, for germination (Cassaniti et al., 2009, 2012, 2013). With the water imbibition, germination stage starts, and different pathways occur (molecular, biochemical, and cellular) in the seed. As a result of these pathways, radicle and hypocotyl protrusion occurs (Nonogaki et al., 2010; Mohamed et al., 2018). Depending on seed characteristics (morphological, genetic, physiological, etc) and environmental effects, seed germination occurs specifically for each plant. Since salinity affects all pathways during

germination, it not only causes germination inhibition but also reduces and delays it (Kaur et al., 1998; Sebei et al., 2007; Mohamed et al., 2018). Seed germination is affected by salinity with osmotic stress and ion toxicity. Water uptake and enzyme activation are inhibited due to osmotic effects. Seed reserves are also negatively affected by stress conditions. Moreover, the essential metabolic stages for dividing and expanding cells were inhibited with ion toxicity (Khan et al., 2000; Ashraf and Foolad, 2007; Munns and Tester, 2008; Mbarki et al., 2018).

Zinnia, Asteraceae family, is cultivated worldwide as annual bedding plants and some of Zinnia sp. could be used as cut flowers. Zinnia plants have wide variation considering their phenotypic characteristics, such as flower and leaf morphology, and ray floret colors. For these reasons, these plants are widely used in gardens or landscape arrangements (Stimart and Boyle, 2007).

The main aim of this study was to evaluate the relative salt stress response in the early stages of seedling growth of twenty *Zinnia* cultivars which have very important value in the bedding plant groups. To interpret the cultivar reactions during germination and early seedling growth for salinity tolerance, we analyzed the rates of radicle emergence and seed germination, radicle, hypocotyl and cotyledon length, relative growth index of the root, and fresh weight of seedlings.

2. Materials and Methods

This study was carried out in the plant physiology laboratory at the Department of Biology Mersin University/Turkey. Seeds of cultivars belong to Zinnia elegans (1 to 14) and Z. marvlandica (15 to 20) were supplied from a local seed distributor (Tasaco Farm, Antalya, Turkey) (Table 1). Zinnia cultivars seeds were placed in 20 x 30 mm dishes lined with double-layer Whatman No 1 filter paper. Seeds were irrigated with two different solutions containing 0 (control) and 100 mM NaCl. The filter papers were replaced every 2 days to prevent microbial contamination. Seeds were allowed to germinate at 25 ± 1 °C in the growth chamber and under dark conditions. The study was terminated when seeds reached the maximum germination number (5 days).

To determine the effects of salinity, seed germination and seedling growth parameters were evaluated in the study. Radicle emergence was defined when the radicles were <2 mm long and germination was considered to have occurred when the radicles were ≥ 2 mm long (plumule and radicle were fully visible) (Kaya et al., 2006; Gao et al., 2018). The number of seeds that emerged and

germinated was recorded every 24 h for 4 days. Radicle emergence and seed germination percentages were calculated using the following Equations 1 and 2 (Koksal et al., 2015; Gao et al., 2018).

$$RE(\%) = NES / TNS X 100$$
(1)

RE: Radicle Emergence, NES: Number of emerged seeds, TNS: Total number of seeds

$$SG(\%) = NGS / TNS X 100$$
(2)

SG: Seed Germination, NGS: Number of germinated seeds, TNS: Total number of seeds

Root length was measured daily with a digital caliper. Based on daily measurement, the relative growth index (RGI) of the root was calculated with the following Equation 3:

$$RGI (mm \, day^{-1}) = (RL_2 - RL_1) / (t_2 - t_1)$$
(3)

The RGI formula was modified from Ren et al. (2016) and Acosta-Motos et al. (2017).

Where, RL_2 - RL_1 , is the root length at the end the beginning of the experiment; t_2 - t_1 was the time duration for the treatment.

Table 1. Zinnia cultivars used in the study

Code	Cultivars
1	Dreamland Coral F1
2	Dreamland Ivory F1
3	Dreamland Pink F1
4	Dreamland Red F1
5	Dreamland Rose F1
6	Dreamland Scarlet F1
7	Dreamland Yellow F1
8	Zinnita Orange F1
9	Zinnita Rose F1
10	Zinnita Scarlet F1
11	Zinnita White F1
12	Zinnita Yellow F1
13	Swizzle Cherry And Ivory F1
14	Swizzle Scarlet Yellow F1
15	Double Zahara Raspberry Ripple F1
16	Double Zahara Yellow F1
17	Double Zahara Cherry F1
18	Double Zahara Fire Improved F1
19	Double Zahara Salmon F1
20	Double Zahara Strawberry F1

Fresh weights were weighed using a digital top loading weighing balance after the 5th day. Plantlets formed hypocotyl was registered after 5th day. Hypocotyl and cotyledon lengths were also measured by a digital caliper.

Reduction rates of radicle emergence, seed germination, root length, hypocotyl formation, hypocotyl and cotyledon lengths, and fresh weights of plantlets were calculated depending on values of control and saline conditions. Radicles and root tips of Zinnia cultivars were photographed with a Zeiss Discovery-V8 Stereo microscope attached to a Zeiss AxioCam ICc 3 digital camera. Images were obtained using a 1.0X lens and 1.5X ocular micrometer. Radicles were photographed after 24 h and root tips were monitored at the end of the germination period.

The experiment was carried out using a completely randomized design with a single factor and conducted with three replicates and each replication included 25 seeds. All quantitative data

expressed as percentages were subjected to arcsine transformation. Data were subjected to ANOVA and the means were separated using the Least Significant Difference (LSD) multiple range test at $p \leq 0.05$. All statistical analyses were performed using the JMP software package (JMP®, Version 8. SAS Institute Inc., Cary, NC, 1989-2019). The weighted ranked method was used to determine the tolerance level of cultivars (Sarıdaş, 2018). The weighted ranked method, relative values, and value range applied to the data which explained the reduction rate was presented in Table 2.

Table 2. The weighted ranked method, relative values, and value ranges

Dougon at ang	Relative		Ι	/alue ranges		
Parameters	values	1	2	3	4	5
Radicle emergence	20	39-53	24-38	9-23	(-6)-8	(-21)-(-7)
Seed germination	20	30-49	10-29	(-10)-9	(-30)-(-11)	(-50)-(-31)
Forming hypocotyl	15	85-98	71-84	57-70	43-56	29-42
Cotyledon length	15	84-100	67-83	50-66	33-49	16-32
Hypocotyl length	15	92-100	83-91	74-82	65-73	56-64
Root length	15	57-67	46-56	35-45	24-34	13-23

3. Results and Discussion

At the end of the study, it was determined that salt stress was effective on seed germination in Zinnia. In addition, there was a wide variation among cultivars in terms of germination parameters examined at both control and saline conditions. The differences in germination parameters were demonstrated both in daily observations and at the end of the germination process.

The cultivar differences in terms of radicle emergence on daily observations were seen in Figures 1a and 1b. As seen in Figures, the seed emergence number was fixed after the third day. In all Zinnia cultivars, the negative effects of salt stress on radicle emergence occurred from the first day. However, in general, Dreamland was the least affected cultivar from salinity. Different effects of salinity on radicle emergence were also shown in Figure 2.

At the end of the germination, we determined statistically significant differences among the cultivars in terms of radicle emergence percentages at both control and salt-containing conditions (Table 3). In the control group, genotypic differences were effective on radical emergence, however, when all cultivars were considered in terms of radicle emergence, the success rate was almost above 50%. In the salt-treated group, differences caused by the genotype were more pronounced. In general, cultivars of Dreamland and Double Zahara series generally had higher radicle emergence ratios, compared to other cultivars, under saline condition. The lowest

radicle emergence percentages under salt stress were in Zi. Scarlet and Zi. Rose with the ratios of 32% and 37%, respectively. To explain the effects of salt stress on the radical emergence, it is more accurate to consider the reduction rate results instead of raw data at the control and 100 mM. The highest radicle emergence reduction was seen in Zi. Rose (52%). However, in most Dreamland and Zahara cultivars, in terms of radical emergence, the reduction rate remained unchanged or minimum level. Even, the percentage of radicle emergence at D. Ivory (-19%), D. Scarlet (-7%), and D. Coral (-7%) slightly increased with salinity.

From daily observations, it was also found that the germination numbers were stable after the third day. Daily changes in germination percentage of cultivars were presented in Figures 1c and 1d. Seed germinations of some Double Zahara cultivars and most Zinnita cultivars were negatively affected by salt treatment.

Genotypic differences made a statistical difference in seed germination (Table 3) in both control and salt-containing conditions. The cultivar which had the highest seed germination number in control treatments was D. Za. R. Ripple followed by other Double Zahara cultivars. However, D. Red, D. Scarlet, and Zi. Orange had higher germination rates than others under stress conditions. D. Coral, D. Ivory, D. Pink, D. Za. Fire Improved and D. Za. Salmon followed them. According to the reduction rate results which compared seed germination percentage under control and salt-containing conditions, we noted significant differences among cultivars. The seed



Figure 1. Daily changes of radicle emergence percentage (RE-%) (a-control, b-saline) seed germination percentage (%) (c-control, d-saline), root length (mm) (e-control, f-saline) of Zinnia cultivars on control (0 mM NaCl) condition and saline (100 mM NaCl) condition

Table 3. The effects of salinit	y on radicle en	nergence, seed {	germination, and n	oot length					
	Ra	dicle emergence	(%)	Se	ed germination (⁹	(0)		Root length (mm)	
Cultivars	0 mM	100 mM	Reduction (%)	0 mM	100 mM	Reduction (%)	0 mM	100 mM	Reduction (%)
D. Coral	64±7 b-e	66±6 ab	4-	64±7 bcd	64±7 a-d	0	10.7±1.8 c-f	6.9±0.5 ab	36
D. Ivory	56±3 de	66±6 ab	-19	46±3 ef	69±9 ab	-49	$13.4\pm1.9 \text{ bc}$	7.7±0.7 a	43
D. Pink	69±5 a-d	66±6 ab	4	56±3 def	66±6 abc	-19	$11.9\pm1.5 \text{ cd}$	7.0±0.5 ab	41
D. Red	74±7 abc	74±7 a	0	58±3 def	74±7 a	-27	12.4±1.2 cd	6.9±0.5 ab	44
D. Rose	58±3 cde	54±5 bcd	8	61±8 cde	51±4 c-g	17	13.3±1.7 bc	6.9±0.5 ab	48
D. Scarlet	74±7 abc	79±7 a	L-	72±8 a-d	74±7 a	4-	17.1±1.4 ab	7.8±0.7 a	54
D. Yellow	74±7 abc	49±5 cde	34	69±5 a-d	46±3 e-h	33	16.5±2.1 ab	7.0±0.4 ab	58
Zi. Orange	74±7 abc	74±7 a	0	72±8 a-d	74±7 a	4-	10.4±1.7 c-f	5.8±0.3 b-e	44
Zi. Rose	77±8 ab	37±3 ef	52	44±3 f	32±3 h	28	7.5±1.3 efg	6.5±0.6 abc	13
Zi. Scarlet	49±5 e	32±3 f	35	44±3 f	32±3 h	28	7.1±0.7 fg	5.4±0.5 cde	24
Zi. White	79±7 ab	44±3 def	45	69±5 a-d	39±4 gh	43	12.2±1.3 cd	4.6±0.3 ef	62
Zi. Yellow	66±6 b-e	48±2 cde	27	46±3 ef	49±5 d-g	-5	6.7±0.8 g	5.1±0.5 def	24
S. Cherry-Ivory	79±7 ab	46±5 def	42	79±7 ab	41±5 gh	48	18.0±1.5 a	6.6±0.6 abc	63
S. Scarlet Yellow	74±7 abc	56±3 bcd	25	77±8 abc	46±6 e-h	40	11.9 ± 1.1 cd	6.1±0.4 abc	49
D. Za. R. Ripple	85±5 a	66±6 ab	22	85±5 a	44±5 fgh	48	10.9±0.9 cde	3.8±0.4 f	65
D. Za. Yellow	66±6 b-e	46±3 def	30	61±3 cde	32±3 h	48	10.9±0.9 cde	5.5±0.3 cde	50
D. Za. Cherry	79±7 ab	51±5 b-e	35	79±7 ab	44±5 fgh	44	7.7±0.7 efg	4.7±0.5 ef	39
D. Za. F. Improved	77±8 ab	64±7 abc	17	72±8 a-d	61±8 a-e	15	10.8±1.3 c-f	4.4±0.4 ef	59
D. Za. Salmon	74±7 abc	66±6 ab	11	72±8 a-d	59±9 a-f	17	9.5±0.9 d-g	$3.7{\pm}0.3~{ m f}$	61
D. Za. Strawberry	72±8 a-d	64±11 abc	10	72±8 a-d	53±5 b-g	25	9.8±0.9 c-g	4.5±0.4 ef	54
LSD	17.933^{*}	15.947^{***}		16.852^{***}	16.157^{***}		3.768***	1.356^{***}	
*: p≤0.05, ***: p≤0.001									



Figure 2. Radicle emergence images of Zinnia cultivars at the end of the 24th hour

germination percentage of the cultivars in the same series was generally consistent with each other. The highest reduction rates in terms of seed germination percentage were calculated at S. Cherry-Ivory (48%), D. Za. R. Ripple (48%), and D. Za. Yellow (48%). In contrast, the germination percentage of Dreamland cultivars increased with salinity compared to others.

When root elongation was followed daily during the experiment, some differences were observed both between treatments and among cultivars. The changes in the root lengths of Zinnia cultivars under control and saline conditions were presented in Figure 1e and 1f, respectively. As seen in Figures, root lengths of all Zinnia cultivars were dramatically decreased by salinity. The adverse effect of salinity on root lengths were revealed from the second day.

There was a quite significant effect of salt stress on root elongation at the end of the germination (Table 3). The root lengths of the cultivars were different in the control group. However, root length was suppressed in 100 mM NaCl. The highest values of root lengths were generally obtained at Swizzle and Dreamland cultivars and the lowest ones were measured at Zinnita cultivars, under control conditions. With respect to salinity, the highest root lengths were observed at Dreamland cultivars. The most reduced rates of root length were in D. Za. R. Ripple, S. Cherry-Ivory, Zi. White and D. Za. Salmon with 65%, 63%, 62% and 61%, respectively. The lowest reduction rates of root length were in Zi. Rose, Zi. Yellow and Zi. Scarlet with 13%, 24% and 24%.

The relative root growth index (RGI) results demonstrated the effects of salt stress on the radicle development stage of the seeds (Figure 3). Generally, Dreamland cultivars and S. Cherry Ivory had higher values than the others in terms of root-RGI when the seeds were germinated under control conditions. The highest root-RGI was found in S.Cherry-Ivory under control condition. On the other hand, the highest root-RGI under saline condition was found in Zi.Scarlet. The results on the control were similar, root-RGI of Dreamland cultivars were higher than the others under saline conditions. When the reduction rate was evaluated in terms of root-RGI, the most reduction was found in D. Za. R. Ripple cultivar and D. Za. Salmon and Zi. White followed (data not shown).



Figure 3. Root relative growth index (RGI) of Zinnia cultivars

The limiting effects of salt stress were also evident during the developmental stage of germinated seeds (Table 4). There were statistically significant differences among the cultivars in terms of hypocotyl formation under the control condition. The hypocotyl formation percentages of cultivars were between 22% and 66% under control condition. However, hypocotyl and cotyledon formations were reduced by salt treatment and even completely inhibited in most cultivars. All Zinnita series cultivars, S. Cherry-Ivory, D. Za. R. Ripple, D. Za. Yellow and D. Za. Cherry could not form hypocotyl under saline conditions. So, the values related to hypocotyl formation, length of hypocotyl, and cotyledon were not obtained from these cultivars. Besides, the lowest reduction rates of hypocotyl formation were in D. Coral and D. Ivory, 31%, and 32%, respectively. The cotyledon length of cultivars was between 3.71 and 7.09 mm under the control condition. The hypocotyl length of cultivars was between 7.43 and 19.48 mm under the control condition. The length of the cotyledon and hypocotyl were reduced under salt condition. In terms of the cotyledon length, the lowest values were in D. Coral (16%) and D. Ivory (28%). And the lowest reduction rates on hypocotyl length were also in D. Coral (56%) and D. Ivory (64%). The negative effects of salt stress on the growth of germinated seeds can also be seen in the images in Figure 4.

The negative effects of salt stress during the germination were also reflected in the fresh weight results. As it was shown in Figure 5, the fresh weights of cultivars were affected by salinity. The highest reduction rates of fresh weight were obtained from D. Yellow and D. Za. Strawberry. The lowest reduction rate was found in D. Pink and Zi. Yellow.

The score values of the cultivars calculated by evaluating all parameters examined in the study according to the weighted grading method are given in Table 5. In the study, there was a wide variation of the affected level of cultivars by salt stress. Some of the cultivars were relatively tolerant, while some of them were quite sensitive. According to the values (Table 5), the total score average of twenty cultivars was found as 216. When cultivars series were considered separately, it was seen that Dreamland cultivars were 301, Zinnita cultivars were 168, Swizzle cultivars were 163 and Double Zahara cultivars were 173. Consequently, the average of Dreamland cultivars was more than the average of all cultivars. So, Dreamland cultivars generally were relatively more tolerant than others in terms of total weighted ranked values. D. Coral and D. Ivory cultivars had the highest points among all cultivars, with 410 and 500, respectively, followed by Double Zahara cultivars. Swizzle and Zinnita cultivars were more sensitive with the lowest value in Swizzle Cherry Ivory with 130 points.

In this study, we interpreted the salt stress tolerance of twenty Zinnia cultivars at the seed germination and early stage of seedling growth. Although most cultivars were adversely affected by salt treatment in general, there was a wide variation among cultivars in terms of tolerance to salt stress on seed germination and early seedling development. Besides, when seed germination was examined in different groups such as radicle emergence, germination and, plantlet growth after germination, it was determined that the cultivars were affected at different levels at each growth stage.

Seed germination starts via water uptake and ends via radicle and plumule emergence from the seed (Parvin et al., 2019). Radical emergence is the

Table 4. The effects of salin	ity on hypocoty	l and cotyledor	1 development of Z	Zinnia cultivars					
	Hy	pocotyl formati	ion (%)	Coty	ledon length (m	un)	Hyp	ocotyl length (mn	U)
Culuvars	0 mM	100 mM	Reduction (%)	0 mM	100 mM	Reduction (%)	0 mM	100 mM	Reduction (%)
D. Coral	39±5 de	27±6 a	31	6.07±0.4 bc	5.07±1.3 a	16	14.49±2.7 a-d	6.39±1.7 a	56
D. Ivory	39±4 de	27±6 a	32	7.09±0.4 a	5.09±1.3 a	28	17.42±1.8 abc	6.19±1.8 a	64
D. Pink	39±4 de	$0{\mp}0$	100	6.72±0.7 abc	0 ± 0	100	$13.31 \pm 1.6 bcd$	0 ± 0	100
D. Red	46±3 cd	17 ± 6 b	64	5.79±0.3 cd	3.33±1.4 ab	43	15.50±3.0 a-d	3.89±1.6 ab	75
D. Rose	32±3 ef	$2\pm0~{ m c}$	94	6.48±0.2 abc	0 ± 0	100	14.80±3.9 a-d	0 ± 0	100
D. Scarlet	66±6 a	$17\pm 6 b$	75	6.88±0.1 ab	3.30±1.4 ab	52	18.64±1.7 ab	3.94±1.7 ab	62
D. Yellow	51±4 bc	7±5 c	87	$6.08{\pm}0.4~{ m bc}$	1.13±1.1 c	81	19.48±3.1 a	1.40±1.4 bcd	93
Zi. Orange	39±4 de	$0{\mp}0$	100	4.75±0.4 e	0 ± 0	100	11.00±2.6 de	0 ± 0	100
Zi. Rose	37±3 de	$0{\mp}0$	100	4.72±0.4 e	0 ± 0	100	7.86±0.7 e	0 ± 0	100
Zi. Scarlet	29±3 ef	$0{\mp}0$	100	5.00±0.3 de	0 ± 0	100	7.43±1.1 e	0 ± 0	100
Zi. White	51±4 bc	$0{\mp}0$	100	5.08±0.5 de	0 ± 0	100	10.80±0.6 de	0 ± 0	100
Zi. Yellow	22±5 f	$0{\mp}0$	100	6.28±0.4 abc	0 ± 0	100	10.70±2.1 de	0 ± 0	100
S. Cherry-Ivory	61±3 ab	$0{\mp}0$	100	5.80±0.3 cd	0 ± 0	100	12.70±1.7 cde	0 ± 0	100
S. Scarlet Yellow	44±3 cd	7±5 c	85	5.91±0.2 bcd	0.98±1.0 c	83	11.28±1.1 de	1.41±1.4 bcd	100
D. Za.R.Ripple	58±3 ab	$0{\mp}0$	100	3.71±0.2 f	0 ± 0	100	12.42±1.0 cde	0 ± 0	100
D. Za. Yellow	58±3 ab	0 ± 0	100	4.45±0.3 ef	0 ± 0	100	13.48±1.5 bcd	0 ± 0	100
D. Za. Cherry	54±5 bc	$0{\mp}0$	100	4.30±0.2 ef	0 ± 0	100	13.35±1.2 bcd	0 ± 0	100
D. Za. F. Improved	54±5 bc	7±5 c	87	4.93±0.4 de	0.66±0.7 c	87	16.73±1.1 abc	0.78±0.8 cd	95
D. Za. Salmon	66±6 a	$17\pm 6 b$	75	4.23±0.2 ef	$1.67{\pm}0.7~{ m bc}$	61	10.20±0.8 de	2.89±1.2 bc	72
D. Za. Strawberry	46±3 cd	7±5 c	85	4.75±0.3 e	1.37±0.9 bc	71	14.72±0.7 a-d	2.01±1.2 bcd	86
LSD	11.326^{***}	9.088^{***}		0.986^{***}	2.087^{***}		5.424^{***}	2.768^{***}	
***: p≤0.001									

	ਬੁ	
•	2	
-	Ħ	
	5	
	la	
	Е	
	Ξ	
ç	2	
	0	
1	Б	
	g	
	E	
	5	
	ē	
	5	
	ð	
	Ĕ	
-	8	
	õ	
	⊵	
	g	
	2	
	Ĕ	
_	ھ	
1	Σ	
	<u>5</u>	
	ဗ	
	ē	
	2	
	q	
	0	
	⊵	
•	E	
;	E	
	ŝ	
د	H	
	s	
	ರ	
ç	Ę	
9	G	
	e O	
Ē	9	
10		

260



Figure 4. General view of Zinnia seedlings



Figure 5. Reduction of fresh weight (RFW - %) on Zinnia cultivars

Table 5.	The score of	cultivars re	lated to	weighted	ranked evaluation
----------	--------------	--------------	----------	----------	-------------------

Cultivora	Root	Seed	Hypocotyl	Cotyledon	Hypocotyl	Root	Total
Cultivals	emergence	germination	formation	length	length	length	Total
D. Coral	80	60	75	75	75	45	410
D. Ivory	100	100	75	75	75	75	500
D. Pink	80	60	15	15	15	30	215
D. Red	80	80	45	60	45	45	355
D. Rose	80	40	15	15	15	30	195
D. Scarlet	100	60	30	45	45	15	295
D. Yellow	40	20	15	30	15	15	135
Zi. Orange	80	60	15	15	15	15	200
Zi. Rose	20	40	15	15	15	45	150
Zi. Scarlet	40	40	15	15	15	30	155
Zi. White	20	20	15	15	15	60	145
Zi. Yellow	40	60	15	15	15	45	190
S. Cherry-Ivory	20	20	15	15	15	45	130
S. Scarlet Yellow	40	20	15	30	30	60	195
D. Za.R.Ripple	60	20	15	15	15	15	140
D. Za. Yellow	40	40	15	15	15	30	135
D. Za. Cherry	40	40	15	15	15	45	150
D. Za. F. Improved	60	40	15	15	15	15	160
D. Za. Salmon	60	20	30	45	60	15	250
D. Za. Strawberry	60	20	15	30	30	30	205

first stage of seed development. In our study, we determined that salt stress and genotypic differences affected radicle emergence. Radicle emergence was generally not affected at Dreamland and Double Zahara cultivars from salinity while Zinnita cultivars were negatively affected by salt stress. Similar to our results, negative effects of salt stress on radicle emergence, seed germination, and plant growth was expressed in previous studies (Sun et al., 2013; Anderson et al., 2015; Borsai et al., 2017; Hannachi and Van Labeke, 2018; Pinheiro et al., 2018).

Seed germination is inhibited by a high salinity level. Salinity affects the germination process via altering imbibition water by seeds because of osmotic stress, metabolic enzyme toxicity, changing protein metabolism and hormonal balance, and decreasing seed reserves (Promila and Kumar, 2000; Othman et al., 2006; Dantas et al., 2007; Gomes-Filho et al., 2008; Hasanuzzaman et al., 2013). In this study, the most reduction in terms of germination percentage was observed at S.Cherry-Ivory, D. Za.R.Ripple, and D Za.Yellow. The germination percentage of Dreamland cultivars were slightly increased with salinity compared to control. In parallel, Zivdar et al. (2011) found that increasing salinity decreased seed germination at Z. elegans. Hannachi and Van Labeke (2018) reported that seed germination parameters (germination percentage, mean germination time, and mean daily germination) were adversely affected by increasing saline stress in Solanum melongena L. Seed germination of eggplant strongly reduced at 80 mM NaCl. Furthermore, it was reported that the germination percentage in Brassica napus importantly decreased at 150 and 200 mM NaCl (Bybordi, 2010). When Vigna radiata irrigated with 250 mM NaCl, germination percentage decreased to 55% (Nahar and Hasanuzzaman, 2009). Carter and Grieve (2008) showed that snapdragon can be produced from seed when exposed to salinity up to 14 dS m⁻¹ because germination remained at 92%. Three Portuguese wild beet ecotypes and one sugar beet were evaluated in terms of salinity tolerance germination during and early seedling development by Pinheiro et al. (2018). They showed that the germination of Vaiamonte (VMT) was the least affected by salinity (98 \pm 2%) germination in the presence of 200 mM NaCl). Comporta (CMP) and Oeiras (OEI) populations had an intermediate sensitivity (53 \pm 7 and 57 \pm 7% germination, respectively). CMP was only able to initiate and maintain radicle emergence, despite a very small extension (< 3%) in 500 mM NaCl. Gao et al. (2018) studied five alfalfa cultivars with salt stress during germination. Salinity caused a

delay of germination on the five cultivars. They emphasized that the ZhongmuNo.3 cultivar was tolerant to 200 mM NaCl, but Daxiyang was sensitive to salinity at the germination stage. Mohamed et al. (2018) revealed that the maximum seed germination percentage was observed in distilled water (100%) followed by 50 mM NaCl (86%)at Pancratium maritimum. Thev demonstrated that the germination percentage was drastically decreased (50%) in 100 mM NaCl and was completely inhibited at both 200 mM and 400 mM NaCl. Terrones et al. (2016) analyzed the effects of salinity on seed germination of three western Mediterranean autochthonous Tamarix species (Tamarix africana, T. boveana, and T. and the eastern Mediterranean gallica) allochthonous Т. parviflora. In general, germination was higher at nonsaline conditions, while germination was delayed under higher salinities. It was shown that seed germination of T. africana dramatically dropped at 1% salinity (from 23.7% to 4.1%), whereas for T. boveana and T. parviflora germination decreased at 6% salinity (from 95% to 42% and from 89% to 14%. respectively), though T. boveana still retained a high germination percentage. Seeds of T. gallica showed intermediate behavior and no germination was recorded for 6% salinity. Similarly, we observed decreased germination similar to reports on literature.

Formation and development of cotyledon and hypocotyl are negatively affected by salinity due to limiting most of the metabolic pathways. Seedling parameters (forming hypocotyl, root, cotyledon and hypocotyl lengths, fresh weight) were analyzed as well as the emergence and germination parameters in this study. These seedling parameters were drastically decreased by salinity. Similarly, the highest root length was recorded in the control by Zivdar et al. (2011) and the lowest was at 12 dS m⁻¹ in Z. elegans L. Zanetti et al. (2019) revealed that plumular and radicular length decreased at switchgrasses [Alamo (lowland) and Shawnee (upland)] as salinity increased and Shawnee had a considerably lower germination rate than Alamo, in particular under critical salinity level (14 dS m⁻¹). Bybordi and Tabatabaei (2009) evaluated germination and seedling responses of five rapeseed cultivars to salinity stress levels [0 (control), 5, 10, 15, and 20 dS m⁻¹]. As salinity increased, rate and final germination, radicle and plumule length and fresh weight significantly decreased. Fresh biomass, total seedling length, hypocotyl, root, and cotyledon lengths of the beet populations under control and 200 mM NaCl treatment were analyzed by Pinheiro et al. (2018). It was revealed that seedling fresh biomass and

hypocotyl, root, and cotyledon lengths in all the beets decreased under salt stress. Esechie et al. (2002) emphasized that salinity had an adverse effect on seedling emergence. It was found that the highest salinity treatment (12.2 dS m^{-1}) caused the lowest seedling emergence percentages. Hypocotyl injury was implicated as a possible cause of poor seedling emergence in chickpea under saline water irrigation and was less severe when pre-germinated seeds were used. Borsai et al. (2017) revealed that seed germination and early seedling growth of all tested Portulaca taxa are inhibited in the presence of increasing concentrations of both, PEG (osmotic stress) and NaCl. Germination and all determined seedling parameters (germination percentages and velocity, seedling fresh weight, seedling vigor index, and radicle, hypocotyl, and cotyledon lengths) decreased under stress conditions. In the study of Memon et al. (2010), it was found that some of Pak choi seeds' ("Ak-1", "Ha-1" and "Sl-1") radicle weight increased in low salt level, whereas others ("Ba-1", "Qd-1" and "Li-1") radicles weight decreased. Hypocotyls weight increased in low salt concentration at "Ba-1", "Ak-1", "Ha-1" and "Sl-1" cultivars. Furthermore, 'Qd-1" and "Li-1" hypocotyls weight significantly reduced. Cotyledons weight of all cultivars increased under salt stress except"Li-1". According to the results of most studies, salt stress could cause reduced biomass production. Because plants respond by reducing leaf area to salinity. Thus, plant water potential could negatively be affected by salt stress. Consequently, plants' biomass could be reduced by the salinity.

4. Conclusions

As a summary, the germination and growth parameters of Zinnia cultivars were negatively affected by salinity (100 mM NaCl). Radicle emergence of D. Ivory, D. Scarlet, and D. Coral slightly increased with salinity. The most radicle emergence reduction was seen in Zi. Rose. The most reduction in terms of germination percentage was observed at S. Cherry-Ivory, D. Za. R. Ripple, and D. Za. Yellow. In contrast, the germination percentage of Dreamland cultivars increased with salinity comparing to control. The most reduction percentages of root length were in S. Cherry-Ivory, D. Za. R. Ripple, Zi. White and D. Za. Salmon. The lowest reductions of root elongation were in Zi. Rose, Zi. Yellow and Zi. Scarlet. The lowest reduction value of forming hypocotyl was in D. Coral and D. Ivory. It was found that cotyledon and hypocotyl length reduction was the lowest in D. Coral and D. Ivory. When all germination and growth parameters under saline conditions were evaluated, Dreamland cultivars, especially D. Ivory and D. Coral, were less affected from the salinity than other cultivars. Consequently, it could be pointed out that germination capability and the following developmental stages of Zinnia cultivars are sensitive to salt stress.

Acknowledgment

This study was supported by Çukurova University, Scientific Research Projects Coordinating Office (Project No: FBA-2019-11481). We thank Prof. Dr. Serpil ÜNYAYAR, Department of Biology, University of Mersin/Turkey, for laboratory support.

References

- Acosta-Motos, J.R., Ortuño, M.F., Bernal-Vicente, A., Diaz-Vivancos, P., Sánchez-Blanco, M.J., Hernández, J.A., 2017. Plant responses to salt stress: adaptive mechanisms. *Agronomy*, 7(18): 1-38.
- Anderson, E.K., Voigt, T.B., Sumin, K., Lee, D.K., 2015. Determining effects of sodicity and salinity on switchgrass and prairie cordgrass germination and plant growth. *Industrial Crops and Products*, 64: 79-87.
- Ashraf, M., Foolad, M.R., 2007. Roles of glycine betaine and proline in improving plant abiotic stress resistance. *Environmental and Experimental Botany*, 59(2): 206-216.
- Borsai, O., Al Hassan, M., Boscaiu, M., Sestras, R.E., Vicente, O., 2017. Effects of salt and drought stress on seed germination and seedling growth in Portulaca. *Romanian Biotechnological Letters*, 23(1): 13340-13349.
- Bybordi, A., 2010. The influence of salt stress on seed germination, growth and yield of canola cultivars. *Notulae Botanicae Horti Agrobotanici Cluj-Napoka*, 38(1): 128-133.
- Bybordi, A., Tabatabaei, J., 2009. Effect of salinity stress on germination and seedling properties in canola cultivars (*Brassica napus* L.). Notulae Botanicae Horti Agrobotanici Cluj-Napoka, 37(1): 71-76.
- Carter, C.T., Grieve, C.M., 2008. Mineral nutrition, growth, and germination of *Antirrhinum majus* L. (snapdragon) when produced under increasingly saline conditions. *American Society for Horticultural Science*, 43(3): 710-718.
- Cassaniti, C., Leonardi, C., Flowers, T.J., 2009. The effect of sodium chloride on ornamental shrubs. *Scientia Horticulturae*, 122(4): 586-593.
- Cassaniti, C., Romano, D., Flowers, T.J., 2012. The response of ornamental plants to saline irrigation water. In: I.G. Garizabal and R. Abrahao (Eds.), *Irrigation-Water management, pollution and alternative strategies*. InTech, Rijeka, Croatia, pp. 131-158.
- Cassaniti, C., Romano, D., Hop, M.E.C.M., Flowers, T.J., 2013. Growing floricultural crops with brackish

water. *Environmental and Experimental Botany*, 92: 165-175.

- Dantas, B.F., De Sá Ribeiro, L., Aragão, C.A., 2007. Germination, initial growth and cotyledon protein content of bean cultivars under salinity stress. *Revista Brasileira de Sementes*, 29(2): 106-110.
- Esechie, H.A., Al-Saidi, A., Al-Khanjari, S., 2002. Effect of sodium chloride salinity on seedling emergence in chickpea. *Journal Agronomy and Crop Science*, 188(3): 155-160.
- Feng, J., Lin, Y., Yang, Y., Shen, Q., Huang, J., Wang, S., Zhu, X., Li, Z., 2018. Tolerance and bioaccumulation of Cd and Cu in Sesuvium portulacastrum. Ecotoxicology and Environmental Safety, 147: 306-312.
- Gao, Y., Cui, Y., Long, R., Sun, Y., Zhang, T., Yang, Q., Kang, J., 2018. Salt-stress induced proteomic changes of two contrasting alfalfa cultivars during germination stage. *Journal of Science of Food and Agriculture*, 99(3): 1384-1396.
- Gomes-Filho, E., Machado Lima, C.R.F., Costa, J.H., Da Silva, A.C., Da Guia Silva Lima, M., De Lacerda, C.F., Prisco, J.T., 2008. Cowpea ribonuclease: properties and effect of NaCl-salinity on its activation during seed germination and seedling establishment. *Plant Cell Reports*, 27(1): 147-157.
- Hannachi, S., Van Labeke, M.C., 2018. Salt stress affects germination, seedling growth and physiological responses differentially in eggplant cultivars (Solanum melongena L.). Scientia Horticulturae, 228: 56-65.
- Hasanuzzaman, M., Nahar, K., Fujita, M., 2013. Plant response to salt stress and role of exogenous protectants to mitigate salt-induced damages. In: P. Ahmad, M.M. Azooz and M.N.V. Prasad (Eds.), *Ecophysiology and* Responses of Plants under Salt Stress, Springer, New York, pp. 25-87.
- Kaur, S., Gupta, A.K., Kaur, N., 1998. Gibberellin A3 reverses the effect of salt stress in chickpea (*Cicer* arietinum L.) seedlings by enhancing the amylase activity and mobilization of starch in cotyledons. Journal of Plant Growth Regulation, 26(2): 85-90.
- Kaya, M.D., Okçu, G., Atak, M., Çıkılı, Y., Kolsarıcı, Ö., 2006. Seed treatments to overcome salt and drought stress during germination in sunflower (*Helianthus annuus* L.). European Journal of Agronomy, 24(4): 291-295.
- Khan, M.A., Ungar, I.A., Showalter, A.M., 2000. The effect of salinity on the growth, water status, and ion content of a leaf succulent perennial halophyte *Suadea fruticosa* (L.) Forssk. *Journal of Arid Environments*, 45(1): 73-84.
- Koksal, N., Agar, A., Yasemin, S., 2015. The effects of top coat substrates on seedling growth of marigold. *Journal of Applied Biological Sciences*, 9(3): 66-72.
- Koksal, N., Alkan-Torun, A., Kulahlioglu, I., Ertargin, E., Karalar, E., 2016. Ion uptake of marigold under saline growth conditions. *SpringerPlus*, 5: 139.
- Koksal, N., Kulahlioglu, I., Ertargin, E., Alkan-Torun, A., 2014. Relationship between salinity stress and

ion uptake of hyacinth (*Hyacinthus orientalis*). *Turkish Journal of Agricultural and Natural Sciences*, Special Issue-1: 578-583.

- Liu, Q., Tang, J., Wang, W., Zhang, Y., Yuan, H., Huang, S., 2018. Transcriptome analysis reveals complex response of the medicinal/ornamental halophyte *Iris halophila* Pall. to high environmental salinity. *Ecotoxicology and Environmental Safety*, 165: 250-260.
- Mbarki, S., Sytar, O., Cerda, A., Zivcak, M., Rastogi, A., He, X., Zoghlami, A., Abdelly, C., Brestic, M., 2018. Strategies to mitigate the salt stress effects on photosynthetic apparatus and productivity of crop plants. In: V. Kumar, S. Wani, P. Suprasanna and L.S. Tran (Eds.), *Salinity Responses and Tolerance in Plants*, Springer, Cham, pp. 85-136.
- Memon, S., Hou, A., Wang, L.J., 2010. Morphological analysis of salt stress response of Pak Choi. *Electronic journal of environmental, agricultural* and food chemistry, 9(1): 248-254.
- Mohamed, E., Kasem, A.M.M., Farghali, K.A., 2018. Seed germination of Egyptian *Pancratium maritimum* under salinity with regard to cytology, antioxidant and reserve mobilization enzymes, and seed anatomy. *Flora*, 242: 120-127.
- Munns, R., Tester, M., 2008. Mechanisms of salinity tolerance. *Annual Review of Plant Biology*, 59: 651-681.
- Nahar, K., Hasanuzzaman, M., 2009. Germination, growth, nodulation and yield performance of three mungbean varieties under different levels of salinity stress. *Green Farming*, 2(12): 825-829.
- Nonogaki, H., Bassel, G., Bewly, H., 2010. Germination still a mystery. *Plant Science*, 179(6): 574-581.
- Othman, Y., Al-Karaki, G., Al-Tawaha, A.R., Al-Horani, A., 2006. Variation in germination and ion uptake in barley genotypes under salinity conditions. *World Journal of Agricultural Sciences*, 2(1): 11-15.
- Parida, A.K., Das, A.B., 2005. Salt tolerance and salinity effects on plants: a review. *Ecotoxicology and Environmental Safety*, 60(3): 324-349.
- Parvin, K., Nahar, K., Hasanuzzaman, M., Borhanuddin Bhuyan, M.H.M., Fujita, M., 2019. Calciummediated growth regulation and abiotic stress tolerance in plants. In: M. Hasanuzzaman, K. Hakeem, K. Nahar and H. Alharby (Eds.), *Plant Abiotic Stress Tolerance*, pp. 291-331.
- Pinheiro, C., Ribeiro, I. C., Reisinger, V., Planchon, S., Veloso, M. M., Renaut, J., Eichacker, L., Ricardo, C.P., 2018. Salinity effect on germination, seedling growth and cotyledon membrane complexes of a Portuguese salt marsh wild beet ecotype. *Theoretical* and Experimental Plant Physiology, 30(2): 113-127.
- Promila, K., Kumar, S., 2000. Vigna radiate seed germination under salinity. Biologia Plantarum, 43(3): 423-426.
- Rámila, C.D.P., Contreras, S.A., Domenico, C.D., Molina-Montenegro, M.A., Vega, A., Handford, M., Bonilla, C.A., Pizarro, G.E., 2016. Boron stress response and accumulation potential of the

extremely tolerant species *Puccinellia frigida*. *Journal of Hazardous Materials*, 317: 476-484.

- Ren, J., Sun, L.N., Zhang, Q.Y., Song, X.S., 2016. Drought tolerance is correlated with the activity of antioxidant enzymes in *Cerasus humilis* seedlings. *BioMed Research International*, 7(Special issue): 1-9.
- Sarıdaş, M., 2018. Determination of yield, quality properties of selected strawberry genotypes obtained by cross breeding and molecular characterization. PhD thesis, Çukurova University Agriculture Faculty Department of Horticulture, Adana. (In Turkish).
- Sebei, K., Debez, A., Herchi, W., Boukhchina, S., Kallel, H., 2007. Germination kinetics and seed reserve mobilization in twoflax (*Linum* usitatissimum L.) cultivars under moderate salt stress. Journal of Plant Biology, 50(4): 447-454.
- Stimart, D., Boyle, T., 2007. Zinnia. In: N.O. Anderson (Ed.), *Flower breeding and genetics*, Springer Dordrecht, pp. 337-357.
- Sun, Y., Niu, G., Osuna, P., Ganjegunte, G., Auld, D., Zhao, L., Peralta-Videa, J.R., Gardea-Torresdey, J.L., 2013. Seedling emergence, growth, and leaf mineral nutrition of *Ricinus communis* L. cultivars

irrigated with saline solution. *Industrial Crops and Products*, 49: 75-80.

- Terrones, A., Moreno, J., Agullo, J.C., Villar, J.L., Vicente, A., Alonso, M.A., Juan, A., 2016. Influence of salinity and storage on germination of *Tamarix taxa* with contrasted ecological requirements. *Journal of Arid Environments*, 135: 17-21.
- Yasemin, S., Koksal, N., Ozkaya, A., Yener, M., 2017. Growth and physiological responses of 'Chrysanthemum paludosum' under salinity stress. Journal of Biological and Environmental Sciences, 11(32): 59-66.
- Zanetti, F., Zegada-Lizarazu, W., Lambertini, C., Monti, A., 2019. Salinity effects on germination, seedlings and full-grown plants of upland and lowland switchgrass cultivars. *Biomass and Bioenergy*, 120: 273-280.
- Zapata, P.J., Serrano, M., Pretel, M.T., Amoros, A., Botella, M.A., 2004. Polyamines and ethylene changes during germination of different plant species under salinity. *Plant Science*, 167(4): 781-788.
- Zivdar, S., Khaleghi, E., Sedighi Dehkordi, F., 2011. Effects of salinity and temperature on seed germination indices of *Zinnia elegans* L. *Journal of Applied Horticulturae*, 13(1): 48-51.