

## Influence of salinity and drought stresses on seed germination and seedling growth characteristics in sesame (*Sesamum indicum* L.)

Tuz ve kuraklık stresinin susamda (*Sesamum indicum* L.) çimlenme ve fide gelişim karakterleri üzerine etkisi

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### ABSTRACT

In the present study, the potential for salt and drought tolerance of sesame genotypes were assessed. Seeds of three registered cultivars (Golmarmara, Muganlı-57 and Uzun) were treated to seven levels (0, 25, 50, 75, 100, 150 and 200 mM) of NaCl to determine salinity influence on seeds. The drought condition was provided by Polyethylene glycol (PEG) induced water deficit. Seeds were germinated under stress of aqueous PEG solutions mixed to create water potentials of -2, -4, -6, -8 and -10 MPa. Effects of these abiotic stresses were assessed with the use of germination rate and early seedling growth traits which were root and shoot length, root fresh and dry weight, shoot fresh and dry weight. There is no germination at -8 and -10 MPa PEG applications for all the cultivars. Increasing concentrations of PEG from -2 to -6 MPa drastically reduced root length and the lowest value was recorded at -6 MPa. The cultivar, The treatment of different NaCl levels showed that increasing salt stress significantly decreased germination rate and seedling growth traits. The traits of the germination rate, root fresh weight and shoot dry weight were less affected at lower levels of salinity. However the concentration of >150 mM NaCl drastically reduced all the studied traits. The overall results suggest that germination and seedling growth traits were strongly inhibited by > -6 MPa PEG and 150 mM NaCl levels therefore lower concentrations should take into consideration for sustainable sesame production.

### MAKALE BİLGİSİ

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### ÖZ

Yürütülen bu çalışmada susam genotiplerinin tuzluluk ve kuraklığa karşı potansiyel toleransları değerlendirilmiştir. Üç tescilli çeşidin (Gölmarmara, Muganlı-57 ve Uzun) genetik materyal olarak kullanıldığı çalışmada çimlenme ve fide gelişim karakterleri üzerindeki tuzluluk etkisini belirleyebilmek amacıyla tohumlar yedi farklı NaCl (0, 25, 50, 75, 100, 150 and 200 mM) konsantrasyonuna maruz bırakılmıştır. Kuraklık stresi yaratmak amacıyla ise PEG solüsyonu kullanılmış olup beş farklı (-2, -4, -6, -8 and -10 MPa) kuraklık seviyesi altında tohumlar çimlendirilmiştir. Stres faktörlerinin etkileri ise çimlenme oranı, kök ve kökçük uzunluğu, taze ve kuru kök ve kökçük ağırlığı karakterleri kullanılarak belirlenmeye çalışılmıştır. -8 and -10 MPa PEG solüsyonunda bulunan tohumların ise çimlenmediği ortaya konmuştur. Özellikle PEG solüsyon değerinin artması kök uzunluğu üzerinde önemli derece negatif etki yaparken en düşük değer -6 MPa'da gözlenmiştir. NaCl stresi altında da hem çimlenme oranı hem de fide gelişim karakterlerinin önemli derecede negatif etkilendiği çalışmada ortaya konmuştur. Çimlenme oranı, taze kök ağırlığı ve kuru kökçük ağırlıklarının düşük seviyedeki tuzluluktan diğer karakterlere göre daha az etkilenmiştir. Ancak 150 mM üzeri NaCl konsantrasyonunda tüm karakterlerde yüksek düzeyde negatif etkilene gözlenmiştir. Tüm sonuçlar dikkate alındığında -6 MPa ve 150 mM üzeri kuraklık ve tuz stresinin çimlenme ve fide gelişim karakterlerini önemli ölçüde negatif etkilediği ortaya konmuş olup bu değerlere sürdürülebilir susam tarımı yapabilmek adına dikkat edilmelidir.

## 1. Introduction

Sesame is an important oilseed crop grown in well tropical and subtropical regions from 40°N to 40°S (Ashri 2007). The crop is called “the queen of oil seeds” because of high and quality of nutritive ingredients of seeds which contain about 40-62.7% oil and 19-30% protein (Ashri 1998; Arslan et al. 2007). This oil is rich in unique antioxidants lignans, sesamin and sesaminol, providing oxidative stability (Erbaş et al. 2009). Sesame oil mainly consists of oleic and linoleic acids (unsaturated fatty acids) accounted for about 80% of total fatty acids (Yol et al. 2015). Sesame makes also positive contribution of human health such as reducing the rate of incidence of certain cancers (Miyahara et al. 2001) and preventing hypertension (Noguchi et al. 2004). Despite these agro-food and health advantages, the productivity of sesame is very low compared to important oilseed crops because of wildish traits include capsule shattering, indeterminate growth habit, non-synchronized maturity and biotic and abiotic stress factors (Ashri 2007).

Soil salinity is one of the important abiotic constraints affecting more than 45 million hectares of cultivated areas (Munns and Tester 2008). Plant growing and productivity are negatively affected by extreme salinity cause to cell damages affecting any stage of crop growth (Jewell et al. 2010). This stress factor reduces the yield of commercial crops by more than 50% (Ashraf 2009). Higher salt concentration leads to osmotic inhibition and this limit to nutrient transferring in roots resulting physiological drought stress and ion toxicity in plant development (Sairam et al. 2002). Drought is another important constraint for crops because of water limitation especially in arid and semi-arid areas having a high temperature, solar radiation and evaporation (Hassanzadeh et al. 2009). Affected plants showed dehydration and restricted gas exchanges followed by inhibition of biochemical mechanisms and photosynthesis resulting considerable yield losses (Jaleel et al. 2011). Like the other major crops, sesame is sensitive the salinity mostly to NaCl ions in soil and drought (Yousif et al. 1972). Sesame is mostly grown in arid and semi-arid lands where its productivity is limited by salinity and drought therefore tolerances of these abiotic stress factors are important for sustainable sesame production (Islam et al. 2016).

Early stages of plant growth, seed germination and seedling emergence, are more sensitive for many plant species although abiotic stress factors affect all growing periods (Cuartero et al. 2006). These stages start by water uptake therefore soil salinity (Tabatabaei and Naghibalghora 2014) and drought (Hassanzadeh et al. 2009) are highly crucial for initial growing and acquiring optimal seedling numbers that result in higher seed yield. Several studies showed that salt stress level (Purohit et al. 2005; Ramirez et al. 2005; El Harfi et al. 2016), drought (Bahrami et al. 2012) and cultivar differences affected the performance of seed germination and seedling traits of sesame seeds. In these studies different NaCl levels were selected understanding salinity effect on seeds for the reason that NaCl is considered as one of the dominant salts in soils. On the other hand, osmotic solutions are one of the important chemicals in the assessment of drought tolerance in germination stage (Muscolo et al. 2014). Polyethylene glycol (PEG), is a non-ionic water, has ability to create water stress artificially (Larher et al. 1993) and generally used to regulate water potential in germination (Kulkarni and Deshpande 2007). Sesame is mostly exposed to salinity and drought in its grown areas and from this perspective we aimed to examine the effects of salinity and

drought stress provoked by different concentrations by NaCl and PEG on germination characteristic and seedling growth traits of sesame seeds.

## 2. Materials and Methods

Sesame seeds of three released cultivars, Golmarmara, Muganli-57 and Uzun, were used as a genetic material in this study. The 2% sodium hypochlorite was used to sterilize seeds for 10-15 min and later washed with double-distilled water. Four replications of 30 seeds of each cultivar were spread on Whatman No. 1 filter paper in 10-cm-diameter petri dishes during the incubation. Moisturizing was provided with 10 ml of distilled water (control) and different NaCl solutions; 25, 50, 75, 100, 150 and 200 mM for salinity experiment. The seeds were exposed to different stress level of polyethylene glycol solution (PEG 6000) as 0 (control), -2, -4, -6, -8 and -10 MPa. The petri dishes were placed in a growth chamber for 10 days at 20 °C under a 14 h light 10 h<sup>-1</sup> dark photoperiodic condition for both treatments. Relative humidity was settled as 70% at day/night. The seeds with at least 1 mm shoot length were considered as germinated and the number of germinated seeds was counted each 3 days. Root and shoot length and fresh weight were measured at after 10 days of germination and calculated as mg plant<sup>-1</sup>. Selected 10 germinated seeds were oven dried at 65 °C to determine dry root and shoot weight. ANOVA and Duncan's multiple range test for comparisons were determined by SAS 9.3 (SAS Institute 2011).

## 3. Results

Drought stress significantly influenced germination rate and all seedling growth traits in this study (Table 1). There are also significant interactions among sesame cultivars and drought stress levels except for germination rate. The use of different PEG levels adversely affected to germination rate (Table 2). The highest ratio was observed in absence of drought stress (control) (Table 2). There is no germination at -8 and -10 MPa PEG applications for all cultivars. A reduction of up to 23 and 53% germination rate observed when seeds were exposed to -4 and -6 MPa compared to the control treatment (97.5%), respectively. There is no significant difference among cultivars in consideration of 0 to -6 MPa PEG (Table 3) for germination rate. The mean germination is 88.4, 83.5 and 82.04% for Golmarmara, Uzun and Muganli-57, respectively (Table 3). Increasing concentrations of PEG from -2 to -6 MPa drastically reduced root length and the lowest value was recorded at -6 MPa. The root length was observed as 25.4, 18.9 and 17.7 mm for Golmarmara Muganli-57 and Uzun cultivars, respectively. There is no data for -8 and -10 MPa PEG concentrations because of non-germinated seeds. Shoot length decreased with increase in drought level, significantly. The highest value for shoot length was observed as 20.0 mm in control condition. At the osmotic potential of -4 and -6MPa, corresponding to the moderate drought stress in this study, the shoot length was 9.1 and 5.4 mm, respectively. It was the highest for Muganli-57 followed by Golmarmara and Uzun. There was a significant reduction in fresh weight of root and shoot with increasing level of PEG concentration. The highest value for fresh weight of root and shoot were 310.1 and 718.2 mg were observed in control condition, respectively. Root fresh weight among sesame cultivars ranged between 107.5 and 237.33 mg (Table 3) and Golmarmara was found significantly higher from the others. Drastic decrease in shoot fresh weight was identified at -4 and -6 MPa PEG. The cultivars Golmarmara and Muganli-57 had

significantly differed from Uzun for this trait. The shoot dry weight also declined in higher drought effect however there is no significant difference among cultivars.

The ANOVA analysis revealed that salt stress had a significant effect on all studied traits (Table 4). However there is no difference among cultivars except for root fresh weight and shoot dry weight traits, significantly. The salt treatments reduced germination rate compared to control in all cultivars (Table 5). It was highly affected with the NaCl concentration of 150 mM. It was generally high, and close to the control level with the treatment of 25, 50 and 75 mM. The highest amount of NaCl concentration (200 mM) in this study completely inhibited seed germination. Although there is no statistical difference, Uzun had the highest average germination rate compared to Golmarmara and Muganli-57 (Table 6). Significant differences were observed among NaCl concentrations with respect to root and shoot length traits. Genotype by treatment interaction effect

was also significant for these traits. Root length was remarkably suppressed in concentrations of 75, 100 and 150 mM. The highest root length was recorded in control (26 mm) and the lowest was measured in concentration of 150 mM (2.9 mm). The longest shoot length was observed at 25 and 50 mM concentrations with 9.3 and 8.9 mm comparing to the control, respectively. Uzun and Golmarmara had higher average shoot length with the values of 9.3 and 9.0 mm, respectively. The root fresh and dry weights were significantly affected by salinity. These traits were decreased with the increase in salinity level, significantly. Lower values were observed at 100 and 150 mM of NaCl. The fresh weight of root was declined significantly by 14% and 47% compared to control in concentrations of 50 and 75 mM of NaCl, respectively. Increasing salinity levels reduced shoot dry weight drastically in the seeds exposed to 150 mM NaCl (Table 2). The highest shoot dry weight was observed in Golmarmara followed by Muganli-57 and Uzun.

**Table 1.** ANOVA on mean of squares of measured traits in sesame cultivars under control and different levels of PEG.

Source of variation	df	Germination rate	Root length (mm)	Shoot length (mm)	Root fresh weight (mg)	Root dry weight (mg)	Shoot fresh weight (mg)	Shoot dry weight (mg)
Cultivar (C)	2	22.1 <sup>ns</sup>	183.5**	23.0*	45707.0**	374.3**	108839.5**	853.3*
Drought level (DL)	5	23959.4**	2096.2**	753.4**	248391.1**	2024.6**	903116.5**	16117.5**
C x DL	10	13.6 <sup>ns</sup>	36.9**	29.2**	1886.1**	86.0**	65693.7**	796.0**

ns, \* and \*\* represent non significant, significant at 5% and 1% probability levels, respectively; df represents degree of freedom.

**Table 2.** Mean comparison of main effects of drought stress levels.

Drought stress level (bar) <sup>†</sup>	Germination rate	Root length (mm)	Shoot length (mm)	Root fresh weight (mg)	Root dry weight (mg)	Shoot fresh weight (mg)	Shoot dry weight (mg)
0	97.5 a*	32.2 a	20.0 a	310.1 a	32.9 b	718.2 a	74.2 a
-2	92.9 b	25.4 b	13.8 b	281.1 a	25.5 a	396.7 b	73.2 a
-4	79.7 c	15.0 c	9.1 c	40.1 b	16.0 c	226.8 c	68.4 ab
-6	63.7 d	9.9 d	5.4 d	38.6 b	9.6 d	149.6 d	57.9 b

\*Means with different letter(s) in each trait is significantly different at 5% probability level according to Duncan's multiple range test.

<sup>†</sup>There is no germination in -8 and -10 bars of PEG.

**Table 3.** Mean comparison of main effects of sesame cultivars for drought.

Cultivars <sup>†</sup>	Germination rate	Root length (mm)	Shoot length (mm)	Root fresh weight (mg)	Root dry weight (mg)	Shoot fresh weight (mg)	Shoot dry weight (mg)
Golmarmara	88.4 a*	25.4 a	12.5 ab	237.3 a	26.8 a	448.6 a	62.3 a
Uzun	83.5 a	17.7 b	10.4 b	107.5 c	15.3 b	258.1 b	73.0 a
Muganli-57	82.0 a	18.9 b	13.2 a	157.7 b	18.6 b	411.8 a	69.9 a

\* Means with different letter(s) in each trait is significantly different at 5% probability level according to Duncan's multiple range test.

<sup>†</sup>There is no germination in -8 and -10 bars which data was not evaluated.

**Table 4.** ANOVA on mean of squares of measured traits in sesame cultivars under control and different levels of NaCl.

Source of variation	df	Germination rate	Root length (mm)	Shoot length (mm)	Root fresh weight (mg)	Root dry weight (mg)	Shoot fresh weight (mg)	Shoot dry weight (mg)
Cultivar (C)	2	49.9 <sup>ns</sup>	12.68 <sup>ns</sup>	9.9**	9245.1 <sup>ns</sup>	0.67 <sup>ns</sup>	1191.5 <sup>ns</sup>	489.4**
Salinity level (SL)	6	17122.0**	1116.7**	210.7**	192625.9**	663.9**	454682.6**	8026.2**
C x SL	12	105.0 <sup>ns</sup>	26.1**	12.1**	26155.1**	4.6 <sup>ns</sup>	20939.6**	153.7 <sup>ns</sup>

ns, \* and \*\* represent non significant, significant at 5% and 1% probability levels, respectively; df represents degree of freedom.

**Table 5.** Mean comparison of main effects of salinity stress levels.

Salinity (NaCl) level (Mm) <sup>†</sup>	Germination rate	Root length (mm)	Shoot length (mm)	Root fresh weight (mg)	Root dry weight (mg)	Shoot fresh weight (mg)	Shoot dry weight (mg)
0	98.8 a*	26.0 a	13.9 a	332.1 a	22.6 a	525.1 a	70.3 a
25	95.3 ab	21.1 b	9.3 b	273.7 ab	14.2 b	420.9 bc	66.7 ab
50	95.5 ab	15.9 c	8.9 b	292.8 ab	11.1 c	460.0 ab	67.1 ab
75	95.8 ab	10.0 d	7.5 c	227.3 bc	9.0 cd	360.0 c	60.0 b
100	89.2 b	6.3 e	6.7 c	164.3 c	6.5 d	292.1 d	62.0 ab
150	44.8 c	2.9 f	6.3 c	47.2 d	3.4 e	93.2 e	21.8 c

\* Means with different letter(s) in each trait is significantly different at 5% probability level according to Duncan's multiple range test.

<sup>†</sup>There is no germination in 200 Mm of NaCl.

**Table 6.** Mean comparison of main effects of sesame cultivars for salinity.

Cultivars	Germination rate	Root length (mm)	Shoot length (mm)	Root fresh weight (mg)	Root dry weight (mg)	Shoot fresh weight (mg)	Shoot dry weight (mg)
Gölmarmara	86.1 a	14.6 a	9.0 a	246.3 a	11.0 a	365.6 a	61.2 a
Uzun	88.3 a	13.1 a	9.3 a	217.5 a	11.0 a	353.2 a	53.8 b
Muganli-57	85.3 a	13.5 a	8.0 b	204.8 a	11.3 a	351.8 a	58.8 ab

\*: Means with different similar letter(s) in each trait is significantly different at 5% probability level according to Duncan's multiple range test, respectively. †There is no germination in 200 mM of NaCl which data was not evaluated.

#### 4. Discussion

Drought stress inhibited to germination rate and seedling growth characteristics in this study. Especially, increasing drought stress level drastically decreased seed germination rate (Table 2). The results were in agreement with the reports of Bahrami et al. (2012), Keshavarzi (2012) and El Harfi et al. (2016) who showed drought inhibitory effect on germination of sesame seeds. Our results revealing PEG levels of -4 and -6 MPa caused to lower germination rate. Reduction in germination might be sourced of lower infusibility of water through the seed coat (Bahrami et al. 2012). Metabolic changes of seeds might also cause to lower seed germination ratio (Ayaz et al. 2000). There is no germination at -8 MPa and over PEG stress levels and similar result was obtained by Keshavarzi (2012) in sesame. Although cultivars have different response to drought tolerance (Bahrami et al. 2012), there is no statistical difference among cultivars for germination rate in our study showing similar drought response of genotypes for this trait. The trait of root length is an important selection criterion because advanced root systems provide tolerance for drought (Turner 1997). The trait of root length drastically decreased with increasing PEG levels compared to control condition. The decrease in root length might be sourced of reduced cellular reproduction and expansion in germination stage (Frazer et al. 1990). The root traits are generally affected firstly under drought stress conditions therefore the genotypes which showed better performance might have a kind of tolerance (Saxena et al. 1993). Hence, the cultivar Golmarmara could be evaluated more tolerant to drought stress than Uzun and Muganli-57 in this particular stage however its tolerance should be confirmed at advanced growth periods. Reduced root lengths under drought conditions have been reported in sesame (Bor et al. 2009) and also tomato (George et al. 2013) and safflower (Jajarmi 2009). Remarkable decrease in shoot length was monitored in higher PEG level which was supported by Bor et al. (2009). Muganli-57 had significantly longer shoot length than other cultivars showing higher productivity under drought conditions. Restricted water conditions inhibited plant development resulting in decline in biomass (Mujtaba et al. 2016). Correlatively, increasing drought stress level caused to reducing of root and shoot fresh weight. Although -2 MPa drought stress level had a similar effect with control, the trait of root fresh weight drastically reduced in -2 to -4 MPa therefore the range of these stress level should be critical values for sesame production.

The results indicated that increasing salinity reduced to germination rate. This was in accordance with the findings of El Harfi et al. (2016) and Bahrami and Razmjoo (2012) who identified salinity adverse effect on sesame seeds. The Higher salt rate in germination phase might cause an osmotic effect which leads to lower rate of water absorption and/or ion toxicity resulted in reduced germination rate (Huang and Redmann 1995). There were also many studies which were in

concordance with the obtained results showing that NaCl stress reduces germination rate in other species such as safflower (Khodadad 2011), wheat (Hampson and Simpson 1990) and chickpea (Murillo-Amador et al. 2002). Although a maximum reduction (44.8%) was recorded in highest dose of applied salts (150 mM) in our study, cultivars can tolerate up to 100 mM salinity during seed germination, only 10% reduction rate was observed in compared to control. This result was endorsed by Gehlot et al. (2005) who obtained limited reduction in germination rate of sesame seeds exposed to 100 mM NaCl. There is no germination in 200 mM NaCl level and this dose also significantly affected seed production in European sea rocket plant (Debez et al. 2004). The root and shoot lengths are one of the significant traits of salt stress tolerance because roots have direct association with soil and shoot supply water from soil for plant development (Gogile et al. 2013). Increasing salinity level strongly inhibited root and shoot length of sesame (Table 5). At low salt concentration (25 mM) root and shoot length traits of sesame seeds were reduced slightly. Increasingly stronger inhibitory effects were monitored with NaCl levels over >75 mM. Higher salt concentration in intercellular spaces (Zhang et al. 2006), specific ion toxicity (Saboor and Kiarostami 2006) and reduced water use efficiency might cause an adverse effect on root and shoot length of seeds (Grewal 2010). Similar to our result, various reports on sesame indicated that increasing level of salinity inhibits growth by reducing root and shoot length (Koca et al. 2007; Bekele et al. 2017). Although there was a varietal difference in response to salinity was reported by Bor et al. (2009), our study showed that cultivars had a similar response to salinity with respect to root length. Shoot and root fresh weights decreased with the high level of salinity stress compare the control level. Because NaCl significantly hinder normal metabolic pathway, it causes osmotic changes and toxic ion effects (Tafouo et al. 2010). Similar results were obtained in sesame (Koca et al. 2007; Bekele et al. 2017) and other species such as safflower (Ghazizade et al. 2012) and rice (Abbas et al. 2013). Increasing salt concentration up to 100 mM limited effect on shoot dry weight compared other traits and Golmarmara was less affected by higher salinity than other two cultivars with respect to shooting dry weight.

#### 5. Conclusion

Germination is the most critical step for plant development and drought and salinity are one of the hazardous stress factors for germination rate and latter growth stages. Sesame is largely grown arid and semi-arid areas therefore limitations of these stress factors are highly important for sesame seed germination. Our study clearly showed that up to -4 bar for drought and 100 mM NaCl level for salinity should be tolerated for sustainable sesame production. The cultivar, Golmarmara showed better performance especially for drought tolerance and it should be evaluated in field conditions with agro-morphological traits.

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