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Research Article

Physiologic and Seed Yield Responses of Different Alkali Grass (*Puccinellia ciliata)* Populations to Salinity Stress

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Abstract

It was investigated the effects of salinity on some physiological characters such as maximal photochemical efficiency of PSII (Fv/Fm), SPAD value and stomatal conductance and seed yield and yield components in *Puccinellia ciliata* B. For this purpose, 7 populations were subjected to salinity stress. Salinity applications to plants started when plants were at the ~4 week old, and continued until the end of the experiment. Seedlings were watered with 30 g/l NaCl solution. Physiological observations in plants were carried out at stem elongation stage. The response of populations to salinity shown significant differences for Fv/Fm and stomatal conductance, whereas SPAD values not affected. Population III exhibited longer plant height, higher number of grains per panicle, spikelet number per panicle, grain yield per plant in salinity conditions. Therefore, it was suggested to be used as a female parent in puccinellia breeding.

Keywords: Chlorophyll content, PSII (Fv/Fm), *Puccinellia ciliata*, salinity stress, stomatal conductance.

Farklı *Puccinellia ciliata* Popülasyonlarının Tuzluluk Stresine Fizyolojik ve Tohum Verimi Tepkileri

Öz

Puccinellia ciliata B.'nın PSII (Fv/Fm)'nin maksimum fotokimyasal etkinliği, SPAD değeri ve stomatal iletkenlik gibi bazı fizyolojik özellikleri ve tohum verimi ve verim komponentleri üzerinde tuzluluğun etkisi araştırılmıştır. Bu amaçla 7 populasyon tuzluluk stresine maruz bırakılmıştır. Tuz uygulamaları, bitkiler ~ 4 haftalık iken başlamış ve denemenin sonuna kadar devam etmiştir. Fideler 30 g/l NaCl çözeltisi ile sulanmıştır. Bitkilerdeki fizyolojik gözlemler, gövde uzama döneminde gerçekleştirilmiştir. Popülasyonların tuzluluğa tepkisi Fv/Fm ve stoma iletkenliği için önemli farklılıklar gösterirken SPAD değerleri etkilenmemiştir. III numaralı populasyon tuzlu koşullarda daha uzun bitki boyu, salkım başına daha fazla tane sayısı ve daha fazla başakçık sayısı ve tane verimi vermiştir. Bu nedenle, puccinellia yetiştiriciliğinde ana ebeveyn olarak kullanılması önerilmektedir.

Anahtar Kelimeler: Klorofil içeriği, PSII (Fv / Fm), *Puccinellia ciliata*, tuzluluk stresi, stoma iletkenliği.

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1. Introduction

There are 1.125 million hectares of land affected by salinity in the world, of which 76 million hectares originated from anthropogenic factors. One-fifth of irrigated areas are affected by salt, and 1.5 million hectares of land become unusable for agricultural production due to high salinity levels. If the salinization of soils continues in this way, 50% of the arable land will be unusable by 2050. The countries affected by salinity are predominantly located in Australia, Bangladesh, China, Egypt, India, Iran, Iraq, Mexico, Pakistan, the former USSR, Syria, Turkey and the United States (Hossain, 2019). Salinity has an adverse effect on plant growth and metabolism and is one of the important environmental stresses affecting agricultural productivity. This situation is becoming more widespread in the world due to intensive agricultural practices and global climate change (Kumar et al., 2018). The negative effects of salt on plant growth are first associated with osmotic stress caused by the limitation of water intake by the roots as a result of a decrease in the amount of water in the soil (De Oliveira et al., 2013).

Puccinellia (*Puccinellia ciliata* B.), a perennial plant, is generally used as a feed for livestock on salinity agricultural land (Jenkins et al., 2010). Presumably the proliferation of aerenchymatous adventitious roots prevents O_2 from being limiting, and *P. ciliata* roots continue active H + pumping under hypoxia (Teakle et al., 2013).

In general stomatal conductance and water content reduced with the increase of salinity concentration the duration of salinity treatment. Salt-induced osmotic influences outside the roots immediately affect stomatal conductivity and reducing stomatal aperture (Wang, 2013). Approximately 20% of growth reduction in plants is probably explained by a decrease in stomatal conductivity. Leaf stomatal conductivity is essential to prevent significant desiccation and CO_2 acquisition in plants. Decrease in stomatal conductivity and consequently transpiration is very important in halophytes, coping with extreme salinity condition (Rangani et al., 2016).

Stomatal conductivity and net photosynthetic ratio in faba bean plants are significantly reduced with water deficiency. Furthermore, this reduction in photosynthesis may result from a decrease in stomatal conductivity, which corresponds to a water loss protective mechanism to improve water use efficiency (de Oliveira et al., 2013). Fv/Fm decreases with increasing salt concentration, especially at high salt concentrations. Decrease in photosynthesis with increased salinity may result from low stomatal conductivity, depression of specific metabolic processes in carbon uptake, inhibition in photochemical capacity, or a combination of these. Also salinity can reduce the efficiency of photosynthesis (Jamil et al., 2007). The maximum quantum efficiency of PSII photochemistry (Fv/Fm) was not significantly altered and stomatal conductance exhibited little changes under 38 mM Na_2CO_3, but was reduced remarkably under 95 mM Na2CO3 (Yu et al., 2013). Chlorophyll contents (SPAD value), in rice leaves are damaged by the increasing salt stress levels (Hussain et al., 2018).

Adequate seed production of adapted *Puccinellia* populations have allowed their use in vegetation of marginal areas. In our previous studies, it was emphazised the dry matter differences of populations in locations where populations grow as habitat (Yavas et al., 2017), the response of photosynthetic pigments to salinity at seedling stage in improved *Puccinellia* population via recurrent selection (Yavas et al., 2020). As such, to evaluate salt tolerance and seed yield together is an area of needed investigation in *Puccinellia*. Therefore, the changes in the levels of various physiological parameters and seed yield have been studied to elucidate the salinity tolerance mechanisms of Puccinellia ciliata to protect the plant from salinity damage.

2. Material and Method

2.1. Plant Materials and Growth Conditions

The study area is mainly characterized as Mediterranean climate (dry summers and mild and rainy winters) with mean annual temperature and total annual rainfall for growing season (from December to March) are around 9.6 ºC and 382.3 mm, respectively.

Plant seeds of *Puccinellia ciliata* were collected from 7 different locations mentioned in the Table 1 below. The soil charactersitics of these locations and morphological charactersitics of base plant populations were published by Yavas et al. (2017). Then, in our breeding study, superior genotypes were allowed to pollination according to recurrent selection methods. In this study, superior genotypes obtained by selection from these populations were used.

2.2. Method

There was filled a potting substrate consisting of 1/3 peat, 1/3 sand and 1/3 field soil in the plastic pots with diameter 12 cm and the high 11 cm. There were 20 seeds sown to every pot, after emergence they were thin out to 10 plant. They were exposed to the natural outdoor conditions. Two treatments were given: control (no NaCl; well drained), salinity (referred as "NaCl"; well drained pots watered with 30 g l-1 NaCl solution). Salinity applications to plants started when plants were at the ~4 week old, and continued until the end of the experiment. The control pot was established without any addition of NaCl. The experiment was planned in completelly randomized design in split plots with three replications. Plants of both species were irrigated with water [control, with electrical conductivity (EC) = 1 $dS·m^{-1}$], 37.5 mg l-1 NaCl solution (EC = 20 $dS·m^{-1}$), to induce salinity stress for 30 d. Leaf chlorophyll content (SPAD units) was measured with a SPAD meter (SPAD-502, Konica Minolta Sensing, Inc., Tokyo, Japan) every week on the middle part of the oldest fully expanded leaves. The stomatal conductance of the leaves was recorded every week with a portable phytosynthesis system DECAGON SC-1. Maximal photochemical efficiency of PSII (Fv/Fm) could be acquired weekly directly from the Plant Efficiency Analyser (Hansatech Instruments Ltd.). Soil salinity levels controlled weekly with a ProCheck (Decagon). All observations were made at stem elongation stage.

Table 1. Locations where Puccinellia ciliata is collected

Locations	Explanation	Coordinate
I	Aydın Söke I	37°,49',53.1984" K, 27°,30',4.14" D
II	Aydın Merkez I	37°,50',39.8328" K, 27°,50',44.88" D
III	Aydın Merkez II	37°,52',19.452" K, 27°,36',6.516" D
IV	Aydın Merkez III	37°,52',19.452" K, 27°,36',6.516" D
V	Aydın Merkez IV	37°,49',1.506" K, 27°,50',7.0476" D
VI	İzmir Kaklıç	38°50′,6.142" K, 26°99′,1.563" D
VII	Aydın Söke II	37°,38',22.1172" K, 27°,14',44.034" D

3. Results and Discussion

3.1. Physiological Parameters

Analysis of variance showed significant P x S interaction for Fv/Fm and stomatal conductance and significant differences between salinity and control for SPAD value (Table 2). The significant interactions indicated different responses populations to salinity for Fv/Fm and stomatal conductance. Therefore, it was compared the population mean values under salinity and control parcels (Table 3). In terms of SPAD, the means of salinity and control were evaluated.

Table 2. Analysis of variance for studied traits in seven Puccinellia populations under salinity stress at stem elongation stage

Source of Variance	Mean of Square			
	df	Fv/Fm	SPAD	SC
Populations (P)	6	0.01*	184.27	99.25
Salinity (S)	1	0.02*	3078.01 **	12785.62**
P x S	6	0.01**	33.30	640.68**
Error	28	0.00	90.68	434.50
CV		6.43	8.92	13.22

*, **: Significant at 5% and 1%, respectively; df: degree of freedom, SC: Stomatal conductance

Fv/Fm value increased in salt stress conditions in population II whereas it decreased in population I and VII. Hovewer, there was no statistical change in genotypes III, IV, V and VI (Table 3). It was shown that the reponse of populations to salinity were significantly different. Fv/Fm significantly reduced at 50 and 100 mM salinity level for pea plants (Sidana, 2017), at 300 mM and 500 mM NaCl concentrations for *Eremochloa ophiuroides* and *Paspalum vaginatum* (Liu et al., 2011) and at 95 mM Na_2CO3 for *Puccinellia tenuiflora* (Yu et al., 2013). It could be speculated that *Puccinellia ciliata*, a facultative halophyte, reacts to salinity unlike other plants and species, and genotypic variation and local differentiation shown among puccinellia populations in terms of Fv/Fm.

As a result of the study, the differences between saline and control for SPAD was non-significant. These results coincide with the earlier findings which show that there was no significant effect on chlorophyll concentration with 20 dSm^{-1} salinity (Akhzari and Aghbash, 2013). On the other hand, SPAD reductions with the increasing salinity have been observed for different crops (Zan et al., 2011; Daba et al., 2019).

The highest stomatal conductance value among the genotypes was obtained from the 4th population with 117.0 in both control and salinity parcels, while the lowest value was observed in the first population for both conditions. Other researchers have observed that stomatal conductance decreased with the salt stress in turfgrass (Liu et al., 2011), in *Phaseolus vulgaris* (Bhuiyan et al., 2015) in *Elytrigia intermedia* (Host.) (Zan et al., 2011).

3.2. Yield and Yield Components

Analysis of variance showed significant differences among populations and salinity for plant height; salinity for panicle length; populations for number of grains per panicle (Table 4). Also, salinity x populations interactions for number of spikelets per panicle and grain yield per plant were significant.

All populations showed better performance for PH at control parcels, whereas it decreased significantly at 20 ds/m salinity levels. However, plant height of population VII was the highest at EC: 20 ds/m (Table 5). Abdel Gadir (1993) stated that soil salinity caused to significantly decrease in barley plant height. The minimum plant height was obtained from control and 8 mmohs/cm salinity concentration while the maximum plant height was obtained from 2, 4 and 6 mmohs/cm. Increasing salinity concentration were caused that *Agropyron elongatum*'s plant height was increased by 5.6 % while plant heiht of *Agropyron cristatum* was decreased by 4.6% (Koc and Acar, 2017). Girma et al. (2017) stated that at 12 dSm^{-1} salinity decreased the plant height by 21-64% even 100% for some susceptible genotypes. Similar results were also determined in rice plants (Hakim et al., 2014).

Salt stress significantly affected panicle length in puccinellia (Table 5) and it recorded at 4.52 of control and 12.64 of salinity parcel. This agrees with Girma et al. (2017) that observed rice genotypes were inversely affected by salinity. This results in line with the results of Mahmoud et al. (2009), Rad et al (2012) and Hakim et al. (2014) where the reduction in panicle length were reported at salinity conditions.

Table 3. Means comparison of salinity and populations on Fv/Fm, SPAD and stomatal conductance (mmol m^{-2} s^{-1}) at stem elongation stage

	Salinity (ds/m)					
	0	**20**	**0**	**20**	**0**	**20**
Populations	**Fv/Fm**		**SPAD**		**SC**	
I	0.70 b	0.78 a	34.0	33.5	67.7 d	75.2 d
II	0.80 a	0.75 ab	31.2	36.6	106.3 a	80.8 bcd
III	0.75 ab	0.76 ab	34.2	35.7	90.6 b	87.7 bc
IV	0.73 ab	0.70 ab	30.8	31.9	117.0 a	108.1 a
V	0.76 ab	0.75 ab	31.3	34.7	87.0 bc	84.7 bc
VI	0.70 b	0.70 b	30.6	30.1	106.7 a	86.3 bc
VII	0.75 ab	0.78 a	30.6	33.2	87.5 bc	82.3 bc
LSD	0.10		ns		13.17	

Table 4. Analysis of variance for plant height, panicle height, number of grains per panicle, number of spikelets per spike, grain yield per plant in seven Puccinellia populations under salinity stress

Source of Variance	**df**	**Mean Square**				
		PH	**PL**	**GN**	**SN**	**GYP**
Populations (P)	6	102.79 **	10.15	2172.33 **	44.83 **	29.74 *
Salinity (S)	1	148.60 *	37.15 *	337.17	6.88	1.35
P x S	6	9.23	7.79	2343.44	47.83 **	46.98 **
Error	28	23.40	7.64	479.50	9.79	9.30
CV		9.98	20.35	18.58	18.58	18.93

*, **: Significant at 5% and 1%, respectively; df: degree of freedome, PH: plant height, PL: panicle length, GN: number of grains per panicle, SN: number of spikelets per panicle, GYP: grain yield per plant

While the population III performed better number of grains per panicle (189.00) under salinity conditions, the population VII (133.00) had better values under control conditions. Salinity highly significantly affected number of seeds per spica (Abdel Gadir, 1993). The salinity concentrations would reduce *Agropyron cristatum*, *Agropyron desertorum* and *Agropyron elongatum* seeds per spica by 62.5%, 61.7% and 42.8% respectively (Koc and Acar, 2017). Similarly, salinity decreased the total grain number per plant (Khanam et al., 2018). At 20 dSm^{-1} NaCl, the highest number of spikelet per panicle was obtained in population III. The highest value (19.0) was obtained from control conditions. It was stated by Abdel Gadir (1993) that salinity had no significant effect on the number of spikes per barley plants.

The interaction between salinity and population was found to be significant. Seed yield per panicle of *Puccinellia* populations significantly affected by salinity and, the means varied from 11.34 mg (I) to 25.53 mg (III) in salinity parcels. The populations VI and V had the second highest yield. Liu and Coulman (2015) also found signficant diferences of *Puccinellia* populations in terms of seed yield per plot. Moreover, seed yield of some *Puccinellia* populations such as III, VI and V were positively affected by salinity. Grime, (1979) and Tarasoff et al. (2007) revaled that less competitive *Puccinellia* plants had shorter plant height, fewer tillers, lower biomass and seed yield in normal condition than that of saline soil. Contrary to *Puccinellia*, rice, a member of same family, was negatively affected by salinity (Girma et al., 2017; Senanayake et al., 2017).

Table 5. Means comparison of salinity and populations on plant height (PH), panicle length (PL), and number of grains per panicle (GN)

	Salinity						
	0	**20**		**0**	**20**	**0**	**20**
Populations	**PH (cm)**		**Mean**	**PL (cm)**		**GN**	
I	48.67	43.83	46.25 b	14.83	9.33	91.00 b	84.00 d
II	48.50	45.00	46.75 b	13.67	12.00	116.67 ab	102.67 cd
III	50.0	49.00	49.50 b	13.33	13.50	105.00 ab	189.00 a
IV	46.0	44.00	45.00 b	13.00	13.17	123.67 ab	102.67 cd
V	49.83	46.33	48.08 b	15.50	12.67	112.00 ab	121.33 bc
VI	50.67	42.00	46.33 b	13.17	13.50	123.67 ab	147.00 b
VII	58.67	55.83	57.25 a	18.17	14.33	133.00 a	98.00 cd
Mean	50.33 a	46.57 b		14.52 a	12.64 b		
	LSD (P): 5.72			LSD (S): 1.75		LSD (P XS): 36.62	

Table 6. Means comparison of salinity and populations on spikelet number per panicle (SN) and grain yield per plant (GYP)

	Salinity			
	0	**20**	**0**	**20**
Populations	**SN**		**GYP (mg)**	
I	13.00 b	12.0 d	15.12 a	11.34 c
II	16.67 ab	14.67 cd	15.75 a	13.86 c
III	15.0 ab	27.00 a	14.18 a	25.53 a
IV	17.67 ab	14.67 cd	16.70 a	13.83 c
V	16.00 ab	17.33 bc	15.12 a	16.40 bc
VI	17.67 ab	21.00 b	16.70 a	19.83 b
VII	19.0 a	14.00 cd	17.96 a	13.23 c
	LSD (P X S): 5.23		LSD (P X S): 5.1	

4. Conclusions and Recommendations

Fv/Fm and SC values changing response depending on populations were demonstrated physiological tolerance of *Puccinellia ciliata*. Populations III shown persistent tolerance to salinity in terms of all physiological parameters. Also, seed yield per panicle of same population was stimulated by salinity. It could be concluded that population III can be used as female parent in future breeding programme.

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