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The effect of salt stress on the germination of maize (*Zea mays* L.) seeds and photosynthetic pigments

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ABSTRACT

The objective of this study was to investigate the effect of salinity stress on seed germination and chlorophyll content in maize. In the study, two maize hybrids were included (Bc 678 and Bc 408) originating from the Bc Institute at Rugvica near Zagreb (Croatia) and two maize populations (LMP-1 and LMP-2) originating from Kosovo. The experiment was conducted in four replicates of 100 seeds, which were germinated on top of double-layered papers, each with 10 ml of salt solution of NaCl and CaCl₂ in Petri dishes. Germinated seeds were counted every 24 h for 15 days. The photosynthetic pigments, chlorophylls 'a' and 'b' as well as carotenoids were extracted with 80 % acetone. Chlorophyll and carotenoid contents were calculated using absorbance values at 662, 644 and 440 nm. The effects of the NaCl and CaCl₂ concentrations accounted for a high proportion of the variance in all analyses. The results showed that both germination percentage and germination index decreased significantly in all cultivars at the highest salt concentrations. The significant differences between different concentrations of salinity were also found in all cultivars for the content of chlorophyll 'a' and 'b' and for the content of carotenoids.

Key words: maize, salinity stress, germination, NaCl, CaCl₂, chlorophyll, carotenoids

IZVLEČEK

VPLIV SLANOSTNEGA STRESA NA KALIVOST IN FOTOSINTEZNE PIGMENTE KORUZE (*Zea mays* L.)

Namen raziskave je bil proučiti vpliv slanosti tal na kalivost zrnja ter vsebnost klorofila in karotinoidov pri koruzi. V proučevanje sta bila vključena dva hibrida 'Bc 678' and 'Bc 408', vzgojena na Zavodu za koruzo Inštituta za žlahtnjenje rastlin v Zagrebu ter dve domači populaciji (LMP-1 and LMP-2) s Kosova. Poskus je bil izveden v 4 ponovitvah in sicer za kalivost v petrijevkah po 100 zrn, za vsebnost klorofila in karotinoidov pa v lončkih po 2 rastlini z 1 kg substrata. Vsak genotip je bil, poleg kontrole, tretiran s 4 različnimi koncentracijami (50, 100, 200 in 400 mMol NaCl in CaCl₂). Kalivost smo ugotavljali prvih 15 dni vsakih 24 ur. Klorofil 'a' in 'b' in karotinoide smo ekstrahirali z 80 % acetonom. Vsebnost klorofila in karotinoidov smo računali s pomočjo absorpcijske vrednosti pri 662, 644 and 440 nm. Pri največji slanosti (400 mMol NaCl in CaCl₂) je pri vseh kultivarjih ugotovljen statistično značilno manjši odstotek in indeks kalivosti. Prav tako so pri vseh kultivarjih ugotovljene statistično značilne razlike med različnimi koncentracijami slanosti tudi za vsebnost klorofila 'a' in 'b' ter vsebnost karotinoidov.

Ključne besede: koruza, slanost tal, kalivost, NaCl, CaCl₂, klorofil, karotinoide

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1 INTRODUCTION

Salinity stress negatively impacts agricultural yields throughout the world, affecting production, whether for subsistence or economic gain. At present, about 20 % of the world's cultivated land and approximately half of all irrigated land and 2.1 % of the dry agriculture land is affected by salinity (FAO, 2000). Salinization is spreading more rapidly in irrigated lands because of inappropriate management of irrigation and drainage. Moreover, rain, cyclones and wind add NaCl to coastal agricultural lands (FAO, 2008). Maize (*Zea mays* L.) is the important cereal crop, providing basic food and oil for human consumption, as well as feed for livestock throughout the world, but this crop is normally submissive to salt stress. Maize, a plant with a C4 metabolism, is also classified as moderately sensitive to salinity (Katerji et al., 1994). The rapid increase in the world's population requires an expansion of crop areas to raise food production. In this context, a significant part of agricultural crops is cultivated on low quality soils, which are sometimes affected by salinity (Allen et al., 1983). Different strategies for diminishing the adverse effects of salinity stress on plants are currently in practice. Salinity due to the over-accumulation of NaCl is usually of great concern and is the most damaging factor in arid and semi-arid regions. Saline soils are widespread throughout the world, and their genesis may be natural or accelerated by irrigated agriculture, the intensive use of water resources combined with high evaporation rates and human activity (Lambers, 2003; Arzani, 2008). The osmotic adjustment, i.e. the reduction of cellular osmotic potential by net solute accumulation, has been considered to be an important mechanism of salt and drought tolerance in plants. This reduction in osmotic potential in salt-stressed plants can be a result of inorganic ion (Na⁺, Cl⁻, and K⁺) and compatible organic solute (soluble carbohydrates, amino acids, proline,

betaines, etc.) accumulations (Hasegawa et al., 2000). Salinity-induced crop yield reduction takes place due to a number of physiological and biochemical disfunctions in plants grown under salinity stress, which have been listed in a number of comprehensive reviews (Kaya et al., 2013). Salinity is considered to be a major abiotic stress affecting germination, seedling growth, and crop production in arid and semi-arid regions (Yohannes and Abraha, 2013). Moreover, salinity has an adverse effect on seed germination of many crops, by creating an osmotic potential outside the seed, thereby inhibiting the absorption of water, or by the toxic effect of Na⁺ and Cl⁻ (Khajeh-Hosseini et al., 2003). Therefore, salinity is one of the most significant abiotic factors limiting crop productivity (Munns, 1993; Gama et al., 2007). The ability of seeds to germinate at high salt concentrations in the soil is of crucial importance for the survival of many plant species. Although salinity stress mostly reduces the germination percentage and delays the onset of germination, its effects are modified by interactions with other environmental factors, such as temperature and light (Bojović et al., 2010). In saline habitats, satisfactory seed germination takes place after high precipitation, when the soil salinity is reduced (Khan and Rizvi, 1994). Seed priming stimulates many of the metabolic processes involved in the early phases of germination, and it has been observed that seedlings from primed seeds grow more vigorously, and perform better in adverse conditions (Cramer, 2002). It has been shown that soil salinity increases P, Mn, and Zn and decreases K and Fe concentrations in plant tissues (Turan et al., 2010).

The present study was to investigate the response of maize seed germination, the content of chlorophyll 'a', 'b' and carotenoid content to different salinity concentration of NaCl and CaCl₂.

2 MATERIAL AND METHODS

2.1 Plant material

The plant material that was included in our study was two maize hybrids ('Bc 678' and 'Bc 408') originating from the Maize Dept. of Bc Institute at

Rugvica near Zagreb (Croatia) and two maize populations (LMP-1 and LMP-2) originating from Kosovo. The experiment was done in the Department of Crop Science, Laboratory of Plant Breeding, University of Prishtina. The seeds were

disinfected in NaOCl 1% for 60 minutes and then rinsed three times with distilled and sterilized water. Maize seeds were germinated on moistened filter paper. The prepared seeds were placed on the germinator for germination (after addition of 10 ml H₂O) for ten days in temperature 25 °C. Pots were filled with compost (minimum 1 kg/pot) for each cultivar and for each treatment. In total, 32 pots were prepared for salt treatment including NaCl and CaCl₂, and a control. During the experiment, solutions with different concentrations for each salt treatment was prepared in the growth period. For two salts (NaCl and CaCl₂), the concentrations were 50, 100, 200 and 400 mMol. After 20 days of exposure, the following parameters were determined in different parts of the plants: chlorophyll pigments and concentration, and seed germination. After disinfection, the seeds were divided into nine treatment groups for each salt solution: H₂O (Control), 50, 100, 200, 400 mMol NaCl and 50, 100, 200, 400 mMol CaCl₂.

2.2 Soil material

The compost consisted of pH (CaCl₂)=5.8; salt concentration (g L⁻¹ KCl=0.9; Nitrogen (NH₄+NO₃)=155 mg L⁻¹, CaCl₂=120; Phosphorus (P₂O₅) mg L⁻¹ CAL=150 and potassium (K₂O) mg L⁻¹ CAL=200. The maize seedlings were transferred to compost in 1 kg weight pots in controlled environment cabinets with 12-hour photoperiods and temperatures of 25/19 °C day/night and 75 % relative humidity.

2.3 Seed germination assays

Germination and early seedling growth were compared at 25 °C (optimum temperature) in the dark. The filter papers were moistened with 20 ml distilled water. For all the seeds groups, the experiment was conducted with four replicates of 100 seeds. Seeds were germinated on top of double-layered papers (ISTA, 1996) with 10 ml of each of the salt solutions of NaCl and CaCl₂ in 10 cm Petri dishes (4 Petri dishes × 25 seeds = 100 seeds × 4 replications = 400 seeds per treatment). These Petri dishes were placed in sealed plastic bags to avoid moisture loss. Seeds were allowed to germinate at 24±1 °C and for 16 hours on the light (day) and 8 hours in the dark (night). The germination percentage is an estimate of the viability of seeds. Germinated seeds were counted every 24 hours for 15 days. According to Sharma

(2010), seeds were considered to have germinated upon the emergence of radicles (≥ 2 mm).

Full Germination Percent (FGP) was calculating according to equation:

$$FGP = \frac{n}{N} \times 100$$

where N – is total seed number;
n – number of germinated seeds.

The Mean Germination Time (MGT) was calculated for each lot, using the daily counts, according to the equation (Moradi et al., 2008):

$$MGT = \frac{\sum nD}{\sum n}$$

where n= number of seeds newly germinated at day D,
D – days from the beginning of the germination test,
Σ n –number of all germinated seeds (final germination).

Germination Index (GI) was calculated as described in the Association of Official Seed Analysis (AOSA, 1983) according to the following formula:

$$GI = \left(\frac{G_1}{1}\right) + \left(\frac{G_2}{2}\right) + \left(\frac{G_x}{x}\right);$$

where GI – is Germination Index,
G₁, G₂, ..., G_x – is germination at 1, 2, ..., x day,
1, 2, ..., x – day of counting of germinated seeds.

2.4 Pigments analysis

Pigments were extracted by grinding 60–80 mg freshly sampled leaves. At the time of sampling, the plants reached the stage of five leaves; we took the third leaf and put in an 80% acetone/water solution containing MgCO₃ (0.5% w/v), at room temperature for 24 hours in the dark. Photosynthetic pigments of all samples were extracted in triplicate to minimize experimental errors. Concentrations of chlorophyll and carotenoid contents were measured by using absorbance recorded at 662, 644 and 440 nm for maximum absorption of chlorophyll 'a' (*Chl 'a'*), chlorophyll 'b' (*Chl 'b'*) and carotenoids, respectively. The extinction coefficients were

determined by a UV-Vis spectrophotometer (SECOMAM, Anthelie Advanced 5). Pigment contents were calculated in mg g⁻¹ fresh leaf

weight (FW) by applying the absorption coefficient equations, described by Lichtenthaler (1986); Aliu et al. (2013 and 2014); Gashi et al. (2013):

$$\begin{aligned} \text{Chl 'a'} \text{ (mg g}^{-1} \text{ FW)} &= [9.784 \text{ (OD662)} - 0.99 \text{ (OD644)}] \times \text{V/FW}, \\ \text{Chl 'b'} \text{ (mg g}^{-1} \text{ FW)} &= [21.426 \text{ (OD644)} - 4.65 \text{ (OD662)}] \times \text{V/FW}, \\ \text{Carotenoids (mg g}^{-1} \text{ FW)} &= [4.695 \text{ (OD440)} - 0.268 \text{ (Chl a + Chl b)}] \times \text{V/FW}. \end{aligned}$$

Where is:

FW – fresh leaf weight,

OD – optical density,

V – volume of sample.

2.5 Statistical analysis

SPSS version 19 was used for analysis of variance for all parameters and to compare of treatment

means with Duncan's Multiple Range Test. Relationships among the traits were estimated with Pearson correlation analysis.

3 RESULTS AND DISCUSSION

Analyses of variance showed a wide range and highly significant effects of NaCl and CaCl₂ concentrations on the parameters of seed germination. The effects of the NaCl and CaCl₂ concentrations accounted for a high proportion of the variance in all analyses include Full Germination Percent (FGP), Mean Germination Time (MGT) and Germination Index (GI) (Table 1).

The FGP at all maize genotypes ranged from 14 to 100%, depending on treatments. FGP for the hybrid 'Bc 678' was low (16 and 44 %) after treatments with 400 mMol CaCl₂ and 400 mMol NaCl, respectively. When comparing these values (16 and 44%) to the any other values (Control, 50, 100 and 200 mMol CaCl₂ and mMol NaCl), they were significantly lower. In the case of hybrid 'Bc 408', between different treatments the same differences for FGP are also present, while lower values (20 and 74%) were recorded in treatment with 400 mMol NaCl and CaCl₂. In comparison with 'Bc 678', the hybrid 'Bc 408' had the highest (87.8%) average values of FGP. The applied high concentrations with NaCl and CaCl₂ for FGP on treatments, as well as 400 mMol in seed Local Maize Populations (LMPs) had negative effects or inhibited the physiological processes. LMP-1 and LMP-2 had significantly lower values of FGP after the treatments of 200 and 400 mMol CaCl₂ and NaCl than the control.

In both hybrids, the differences in FGP, in treatments with 50, 100, 200 mMol, both for NaCl and CaCl₂ concentrations, resulted in no significant differences, except in the case of the hybrid 'Bc 678' after the treatment with 200 mMol CaCl₂. In both populations (LMP-1 and LMP-2) significant differences exist almost among all treatments. Therefore, the populations could be more susceptible to salinity stress than hybrids. However, the seed germination percentage of populations decreased at the highest level of salinity, but the hybrid 'BC 678' expresses significantly higher values of FGP at 50 and 100 mMol CaCl₂ and at 50, 100 and 200 mMol NaCl than the control does.

Many authors (Amzallag et al., 1990; Djanaguiraman et al., 2006) found that plants' exposure to low level salinity activates an array of processes leading to an improvement of plant stress tolerance. High salt concentrations negatively affect maize growth. Rahman et al. (2000) reported that maize cultivars were significantly more tolerant to salt stress at germination than at later stages of growth. In order to determine the usefulness of *Tripsacum* in improving salt tolerance in maize, and the effects of NaCl, *in vitro* and *in vivo*, Pesqueira et al., (2006) evaluated an intergeneric hybrid obtained

from crossing *Zea mays* L. with *Tripsacum dactyloides* L.

The different levels of NaCl and CaCl₂ concentrations also significantly affected the mean germination time (MGT) and germination index (GI) (Table 1). The significantly greater number of days for MGT at all genotypes were obtained from treatments in 400 mMol NaCl and CaCl₂ concentrations; furthermore, the concentration of 400 mMol CaCl₂ also resulted in a significantly higher number of days when compared to the same concentration of NaCl.

Similar findings were also obtained for GI. For both parameters, the greatest differences between control and treatments in 400 mMol NaCl and CaCl₂ concentrations for maize populations than for hybrids were obtained.

For maize hybrids, the MGT at 400 mMol CaCl₂ and NaCl concentration the 6.87 and 6.50 days were obtained, respectively; while for maize populations (LMP) the values 7.94 and 7.88 were obtained. On the basis of these results, we can conclude that the maize populations are more responsive to soil salinity than hybrids. Taiz and Zeiger (2002) concluded that the high concentration of NaCl in the salt solution increases its osmotic potential.

In addition, the high absorption of Na and Cl ions during seed germination can be due to the cell toxicity that finally inhibits or slows the rate of germination and thus decreases the germination percentage. Moreover, the germination indices of all the cultivars decreased with increasing salt stress (Carpici et al., 2009).

The leaf is a very important photosynthetic organ, in which light energy is transformed through the green pigment chlorophyll into the potential energy of assimilates. Our results show that the chlorophyll content concentration was significantly changed under different salinity concentrations (Table 2).

In many cases, the significantly higher content of chlorophyll 'a' was found at lower salinity concentrations of only NaCl. The significantly higher content of chlorophyll 'a' than in control was obtained at concentrations of 100 mMol NaCl ('Bc 408' and LMP-1), at concentrations of 50 and

200 mMol NaCl ('Bc 678'), and at concentrations of 50, 100 and 200 mMol NaCl (LPM-2). Significantly lower contents of chlorophyll 'a' were found at higher concentrations of 100, 200 and 400 mMol CaCl₂ ('Bc 678'), 200 and 400 mMol CaCl₂ ('Bc 408' and LPM-2) and at 400 mMol CaCl₂ (LMP-1).

NaCl was affected on lower chlorophyll 'a' at all cultivars only at 400 mMol concentrations. Similar results for chlorophyll 'a' in different treatments of maize were obtained by Daughtry et al. (1999), ranging from 10.4 to 34.6 mg g⁻¹. The content of chlorophyll 'b' is less variable than chlorophyll 'a' under different salinity concentrations.

Significantly higher contents of chlorophyll 'b' were found only at 'Bc 678' (50 mMol CaCl₂ and 200 mMol NaCl), at LMP-2 (50, 100 and 200 mMol NaCl); while the lowest chlorophyll 'b' contents at 200 and 400 mMol CaCl₂ ('Bc 678', 'Bc 408' and LMP-2), at 400 mMol CaCl₂ (LMP-1) and only at 400 mMol NaCl ('Bc 408', LMP-1 and LMP-2) were found.

In general, the highest salinity concentrations reduced content of both chlorophyll 'a' and 'b' compared to the control.

The significantly lower content of carotenoides was determined at higher concentrations of treatments, 400 mMol CaCl₂, at all cultivars and at 400 mMol NaCl at both LMP (Table 2). The most responsive to salinity stress relating to carotenoides content was LMP-2; at both highest concentrations, it showed the lowest carotenoid content, while at 50, 100 and 200 mMol NaCl concentrations it showed significantly higher carotenoides content than the control.

Table 1: The effect of salinity on seed germination in maize cultivars

Treatment	'Bc 678'			'Bc 408'			LMP-1			LMP-2		
	FGP * (%)	MGT (days)	GI %	FGP (%)	MGT (days)	GI %	FGP (%)	MGT (days)	GI %	FGP (%)	MGT (days)	GI %
Control	96 ^b	4.0 ^c	59.3 ^{ab}	100 ^a	4.0 ^c	61.7 ^a	100 ^a	4.0 ^{cd}	61.7 ^a	88 ^b	4.1 ^{de}	53.4 ^b
50 mM CaCl ₂	100 ^a	4.0 ^c	61.7 ^a	100 ^a	4.0 ^c	61.7 ^a	100 ^a	4.0 ^{cd}	61.4 ^a	82 ^b	4.0 ^e	50.5 ^b
100 mM CaCl ₂	100 ^a	4.0 ^c	61.7 ^a	100 ^a	4.0 ^c	61.5 ^a	98 ^a	4.1 ^{bc}	59.9 ^a	96 ^a	4.1 ^{de}	57.9 ^a
200 mM CaCl ₂	96 ^b	4.1 ^c	57.9 ^b	96 ^a	4.0 ^c	55.8 ^b	68 ^b	4.7 ^{ab}	35.3 ^c	70 ^c	4.2 ^d	39.5 ^d
400 mM CaCl ₂	16 ^d	6.9 ^a	3.6 ^d	20 ^c	6.5 ^a	5.2 ^d	28 ^c	7.9 ^a	3.7 ^e	14 ^d	7.9 ^a	2.0 ^f
50 mM NaCl	100 ^a	4.0 ^c	61.7 ^a	100 ^a	4.0 ^c	61.7 ^a	100 ^a	4.0 ^{cd}	60.5 ^a	86 ^b	4.0 ^e	53.1 ^b
100 mM NaCl	100 ^a	4.0 ^c	61.7 ^a	100 ^a	4.0 ^c	61.7 ^a	94 ^a	4.0 ^{cd}	57.3 ^a	94 ^a	4.0 ^e	58.0 ^a
200 mM NaCl	100 ^a	4.0 ^c	61.7 ^a	100 ^a	4.0 ^c	61.0 ^a	72 ^b	3.4 ^d	46.1 ^b	88 ^b	4.6 ^c	46.8 ^c
400 mM NaCl	44 ^c	4.6 ^b	23.1 ^c	74 ^b	5.3 ^b	33.6 ^c	20 ^c	5.0 ^b	9.7 ^d	16 ^d	6.2 ^b	5.9 ^e
Average (μ)	83.6	4.4	50.3	87.8	4.4	51.6	75.6	4.6	44.0	70.2	4.8	40.8

*FGP – final germination percentage; MGT – mean germination time; GI – germination index;

* – values within individual columns indicated by at least one equal letter are not significantly different at 0.05 probability level

Table 2: Effect of salinity on photosynthetic pigments content (mg g⁻¹ FW) of maize cultivars

Treatment	'Bc 678'					'Bc 408'					LMP-1					LMP-2				
	Chl <i>a</i>	Chl <i>b</i>	Carot	Total Chl	Ratio <i>a/b</i>	Chl <i>a</i>	Chl <i>b</i>	Carot	Total Chl	Ratio <i>a/b</i>	Chl <i>a</i>	Chl <i>b</i>	Carot	Total Chl	Ratio <i>a/b</i>	Chl <i>a</i>	Chl <i>b</i>	Carot	Total Chl	Ratio <i>a/b</i>
Control	38.8 ^{c*}	8.8 ^{bc}	5.6 ^{bc}	47.5 ^c	4.4 ^{bc}	37.3 ^b	10.1 ^a	5.8 ^{ab}	47.3 ^c	3.8 ^a	39.5 ^{bc}	10.1 ^a	6.2 ^{ab}	49.6 ^{bc}	4.0 ^a	37.9 ^b	8.9 ^c	5.6 ^c	46.7 ^b	4.2 ^a
50 mM CaCl ₂	44.8 ^{abc}	10.9 ^a	6.2 ^{ab}	55.8 ^{ab}	4.1 ^{ab}	38.9 ^b	9.5 ^{abc}	5.7 ^{ab}	48.4 ^c	4.2 ^a	38.8 ^{bc}	10.8 ^a	5.2 ^b	49.6 ^{bc}	3.7 ^{ab}	41.1 ^c	9.6 ^c	5.9 ^{bc}	50.7 ^b	4.3 ^a
100 mM CaCl ₂	31.2 ^d	7.8 ^{cd}	4.9 ^{cd}	39.1 ^d	4.0 ^{cd}	37.3 ^b	9.4 ^{abc}	5.4 ^{ab}	46.7 ^c	4.0 ^a	43.8 ^b	10.1 ^a	6.0 ^{ab}	53.9 ^{bc}	4.3 ^a	23.4 ^d	5.7 ^e	3.2 ^d	29.1 ^d	4.1 ^a
200 mM CaCl ₂	27.3 ^d	6.8 ^{de}	3.9 ^d	34.1 ^d	4.0 ^d	30.6 ^c	7.4 ^c	5.6 ^b	38.0 ^d	4.2 ^a	36.7 ^c	8.6 ^a	5.5 ^{ab}	45.2 ^c	4.3 ^a	33.0 ^c	7.5 ^d	4.7 ^c	40.5 ^c	4.4 ^a
400 mM CaCl ₂	20.1 ^e	5.3 ^e	3.8 ^d	25.4 ^e	3.8 ^d	9.7 ^d	3.5 ^d	2.1 ^c	13.2 ^e	2.8 ^b	6.1 ^c	2.2 ^b	1.3 ^d	8.2 ^e	2.8 ^b	10.0 ^c	2.8 ^f	2.2 ^d	12.9 ^c	3.6 ^b
50 mM NaCl	46.2 ^{ab}	11.6 ^a	7.1 ^a	57.8 ^{ab}	4.0 ^a	40.9 ^{ab}	9.8 ^{ab}	5.9 ^{ab}	50.7 ^{bc}	4.2 ^a	38.5 ^c	9.2 ^a	5.8 ^{ab}	47.7 ^{bc}	4.2 ^a	48.0 ^a	11.3 ^b	7.3 ^a	59.3 ^a	4.3 ^a
100 mM NaCl	41.3 ^{bc}	9.9 ^{ab}	6.2 ^{ab}	51.3 ^{bc}	4.2 ^{ab}	44.3 ^a	10.6 ^a	6.4 ^a	55.2 ^a	4.2 ^a	47.0 ^a	11.2 ^a	6.9 ^a	58.2 ^a	4.2 ^a	52.3 ^a	12.6 ^a	8.3 ^a	64.9 ^a	4.2 ^a
200 mM NaCl	48.5 ^a	11.1 ^a	6.6 ^{ab}	59.6 ^a	4.3 ^{ab}	42.3 ^{ab}	10.9 ^a	6.3 ^a	53.1 ^{ab}	3.9 ^a	39.2 ^{bc}	9.7 ^a	5.4 ^b	49.0 ^{bc}	4.1 ^a	49.7 ^a	11.9 ^{ab}	7.1 ^{ab}	61.7 ^a	4.2 ^a
400 mM NaCl	21.5 ^c	6.2 ^{ab}	6.1 ^{ab}	27.7 ^d	3.5 ^d	29.8 ^c	7.7 ^{bc}	4.9 ^{ab}	37.5 ^d	3.9 ^a	15.1 ^d	4.2 ^b	2.7 ^c	19.2 ^d	3.6 ^{ab}	20.0 ^d	5.9 ^e	3.4 ^d	25.1 ^d	3.9 ^b
Average	35.5	8.7	5.6	47.0	4.1	34.5	8.8	5.3	43.3	3.9	33.9	8.4	5.0	42.3	3.9	35.1	8.5	5.3	43.4	4.1

* – values within individual columns indicated by at least one equal letter are not significantly different at 0.05 probability level

In general, the correlation coefficients between all the studied properties for most cultivars were positive and statistically significant, except for the MGT, for which there was a negative and significant correlation (Table 3). Only for hybrid 'Bc 678' were a lower value of correlation

coefficients between the ratio of chlorophyll 'a' and 'b' and other properties obtained; statistically nonsignificant correlation coefficients were obtained only for MGT and chlorophyll 'a'. Wue et al. (2008) investigated similar issues and have obtained similar results.

Table 3: The correlation coefficients between investigated traits in maize cultivars

Treatment	FGP	MGT	GI	Chl 'a'	Chl 'b'	Carotenoids	Chl (a+b)	
Bc 678	FGP	1						
	MGT	-0.90**	1					
	GI	1**	-0.90**	1				
	Chl 'a'	0.50**	-0.62**	0.51**	1			
	Chl 'b'	0.48*	-0.58**	0.48*	0.95**	1		
	Carot.	0.50**	-0.62**	0.51**	0.99**	0.96**	1	
	Total Chl	0.39*	-0.48**	0.40*	0.92**	0.92**	0.93**	1
	Ratio a/b	0.30	-0.38*	0.30	0.42*	0.13	0.37	0.24
Bc 408	FGP	1						
	MGT	-0.97**	1					
	GI	0.98**	-0.99**	1				
	Chl 'a'	0.92**	-0.93**	0.92**	1			
	Chl 'b'	0.86**	-0.89**	0.87**	0.93**	1		
	Carot.	0.91**	-0.93**	0.92**	0.99**	0.95**	1	
	Total Chl	0.85**	-0.87**	0.85**	0.93**	0.98**	0.95**	1
	Ratio a/b	0.76**	-0.72**	0.73**	0.75**	0.50**	0.71**	0.54**
LMP-1	FGP	1						
	MGT	-0.69**	1					
	GI	0.98**	-0.80**	1				
	Chl 'a'	0.88**	-0.84**	0.92**	1			
	Chl 'b'	0.86**	-0.82**	0.90**	0.93**	1		
	Carot.	0.89**	-0.85**	0.92**	0.99**	0.95**	1	
	Total Chl	0.85**	-0.82**	0.88**	0.96**	0.91**	0.96**	1
	Ratio a/b	0.55**	-0.65**	0.58**	0.72**	0.45*	0.68**	0.69**
LMP-2	FGP	1						
	MGT	-0.91**	1					
	GI	0.99**	-0.93**	1				
	Chl 'a'	0.75**	-0.76**	0.75**	1			
	Chl 'b'	0.74**	-0.73**	0.74**	0.99**	1		
	Carot.	0.75**	-0.75**	0.75**	1**	0.99**	1	
	Total Chl	0.67**	-0.66**	0.67**	0.96**	0.97**	0.97**	1
	Ratio a/b	0.61**	-0.75**	0.62**	0.60**	0.51**	0.58**	0.49**

* - Correlation is significant at the 0.05 level,

** - Correlation is significant at the 0.01 level.

4 CONCLUSIONS

The study involving NaCl and CaCl₂ with different concentrations indicated that maize seed germination and chlorophyll content are sensitive to salt stress. In general, no significant decrease at 50 and 100 mMol salt concentrations for all maize cultivars and all investigated traits were found, while the highest concentrations of 400 mMol NaCl and CaCl₂ negatively affected all cultivars and for all properties. In some cases, mainly

moderate concentrations (50 or 100 mMol) of NaCl had a positive impact on investigated traits. Hybrids are less sensitive to salinity than populations because they were not genetically improved. Therefore, due to the genetic variability of populations and their responsiveness to salinity, they can serve as a good starting material for breeding of genotypes resistant to salinity stress.

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6 REFERENCES

- Aliu S., Fetahu Sh., Rozman L., Salillari A. 2008. General and specific combining ability studies for leaf area in some maize inbreds in agroecological conditions of Kosovo. *Acta agriculturae Slovenica*, 91, 1: 67-73; DOI: 10.2478/v10014-008-0007-4
- Aliu S., Gashi B., Rusinovci I., Fetahu Sh. Vataj R. 2013. Effects of some heavy metals in some morphological and physiological parameters in maize seedlings. *American Journal of Biochemistry and Biotechnology*, 9, 1: 27-33; DOI: 10.3844/ajbbsp.2013.27.33
- Aliu S., Rusinovci I., Gashi B., Kaul H.P., Rozman L., Fetahu Sh. 2014. Genetic diversity for mineral content and photosynthetic pigments in local bean (*Phaseolus vulgaris* L.) populations. *Journal of Food, Agriculture & Environment*, 12, 2: 635-639
- Allen S.G., Dobrenz A.K., Schonhorst M.H. Stoner J.E. 1983. Heritability of NaCl tolerance in germinating alfalfa seeds. *Agronomy Journal*, 77: 99-101; DOI: 10.2134/agronj1985.00021962007700010023x
- Amzallag N., Lerner H.R., Poljakoff Mayber A. 1990. Induction of increased salt tolerance in Sorghum bicolor by NaCl pretreatment. *Journal of Experimental Botany*, 41: 29-34; DOI: 10.1093/jxb/41.1.29
- Arzani A. 2008. Improving salinity tolerance in crop plants: a biotechnological view. *Vitro Cell. Dev. Biol. Plant* 44: 373-383; DOI: 10.1007/s11627-008-9157-7
- Association of Official Seed Analysis (AOSA). 1983. Seed Vigour Testing Handbook. Contribution No. 32 to the handbook on Seed Testing, 18-19
- Bojović B., Đelić G., Topuzović M., Stanković M. 2010. Effects of NaCl on seed germination in some species from families *Brassicaceae* and *Solanaceae*, *Kragujevac Journal of Science*, 32: 83-87
- Carpıcı E.B., Celik N., Bayram G. 2009. Effects of salt stress on germination of some maize (*Zea mays* L.) cultivars. *African Journal of Biotechnology*, 8, 19: 4918-4922.
- Cramer, G.R. 2002. Sodium-calcium interaction under salinity stress, University of Nevada, Reno, NV 89557 USA. Chapter 10: 205-228.
- Daughtry C., Walthall L., Kim K., Brown E., McMurtrey E. 1999. Estimating Corn Leaf Chlorophyll Concentration from Leaf and Canopy Reflectance. *Remote sens. Environment*, 74: 229-239 ; DOI : 0.1016/S0034-4257(00)00113-9
- Djanaguiraman M., Sheeba J.A., Shanker A.K., Devi D.D., Bangarusamy U. 2006. Rice can acclimate to lethal level of salinity by pretreatment with sublethal level of salinity through osmotic adjustment. *Plant and Soil* 284: 363-373; DOI: 10.1007/s11104-006-0043-y
- FAO 2000. Global network on integrated soil management for sustainable use of salt-affected soils. Available in:

- <http://www.fao.org/ag/AGL/agll/spush/intro.htm>
(28.Jan.2015)
- FAO. 2008. Land and Plant Nutrition Management Service. <http://www.fao.org/ag/agl/agll/>. Accessed on November/ 15/2012.
- Gama P.B.S., Inagana S., Tanaka K., Nakazawa R. 2007. Physiological response of common bean (*Phaseolus Vulgaris*. L.) seedlings to salinity stress. African Journal of Biotechnology, 2: 79-88
- Gashi B., Babani F., Kongjika E. 2013. Chlorophyll fluorescence imaging of photosynthetic activity and pigment contents of the resurrection plants *Ramonda serbica* and *Ramonda nathaliae* during dehydration and rehydration. Physiology Molecular Biology of Plants, 19, 3: 333-341; DOI: 10.1007/s12298-013-0175-5
- Hasegawa P.M, Bressan R.A., Zhu J.K., Bohnert H.J. 2000. Plantcellular and molecular responses to high salinity. Annu. Rev. Plant Physiology, 51: 463-499; DOI: 10.1146/annurev.arplant.51.1.463
- Katerji N., Van Hoorn J.W., Hamdy. A., Karam F., Mastroruilli M. 1994. Effect of Salinity on Emergence and on Water Stress and Early Seedling Growth of Sunflower and Maize. Agric. Wat. Mang., 26: 81-91; DOI: 10.1016/0378-3774(94)90026-4
- Kaya C., Ashraf M., Murat Dikilitas M., Atilla L. 2013. Alleviation of salt stress-induced adverse effects on maize plants by exogenous application of indoleacetic acid (IAA) and inorganic nutrients – A field trial. Australian Journal of Crop Science, 7, 2: 249-254
- Khajeh-Hosseini M., Powell A.A., Bingham I.J. 2003. The interaction between salinity stress and seed vigor during germination of soybean seeds. Seed Science Technology, 31: 715-725; DOI: 10.15258/sst.2003.31.3.20
- Khan A. M., Rizvi Y. 1994. Effect of salinity, temperature and growth regulators on the germination and early seedling growth of *Atriplex griffithii*. Canadian Journal of Botany, 72, 475-479; DOI: 10.1139/b94-063
- Lambers H. 2003. Introduction, dry land salinity: a key environmental issue in Southern Australia. Plant Soil, 257: 5-7; DOI: 10.1023/B:PLSO.0000003909.80658.d8
- Lichtenthaler H. 1986. Laser-Induced Chlorophyll Fluorescence of Living Plants, Proceedings of the Remote Sensing Symposium, Band III, ESA Publication Division, Noordwijk, 1571-1579
- Moradi D.P., Sharif-Zadeh F., Janmohammadi M. 2008. Influence of priming techniques on seed germination behavior of maize inbred lines (*Zea mays* L.). Journal of Agricultural and Biological Sciences, 3, 3: 22-25
- Munns R. 1993. Physiological processes limiting plant growth in saline soils: some dogmas and hypotheses. Plant Cell Environment, 16: 15-24; DOI: 10.1111/j.1365-3040.1993.tb00840.x
- Pesqueira J., Garcia M.D., Staltari S., Molina M.D.C. 2006. NaCl effects in *Zea mays* L. x *Tripsacum dactyloides* L. hybrid calli and plants. Electronic Journal of Biotechnology, 9, 3: 285-290; DOI: 10.2225/vol9-issue3-fulltext-29
- Rahman M., Kayani S.A., Gul S. 2000. Combined Effects of Temperature and Salinity Stress on Corn Sunahry Cv., Pakistan Journal of Biological Sciences, 3, 9: 1459-1463; DOI: 10.3923/pjbs.2000.1459.1463
- Sharma R.K., Sharma S. 2010. Effect of storage and cold-stratification on seed physiological aspects of *Bunium persicum*: A threatened medicinal herb of Trans-Himalaya. Int. J. Bot., 6, 2: 151-156; DOI: 10.3923/ijb.2010.151.156
- Taiz L., Zeiger E. 2002. Plant Physiology. 3rd Edn., Sunderland, Sinauer Associates, Inc.: 85-87
- Turan M., Elkarim H., Taban N., Suleyman Taban S. 2010. Effect of salt stress on growth and ion distribution and accumulation in shoot and root of maize plant. African Journal of Agricultural Research, 5, 7: 584-588
- Wue Ch., Niu Zh., Tang Q., Huang W. 2008. Estimating chlorophyll content from hyperspectral vegetation indices: Modeling and validation. Agricultural and forest meteorology, 148: 1230–1241; DOI: 10.1016/j.agrformet.2008.03.005
- Yohannes G., Abraha B. 2013. The role of seed priming in improving seed germination and seedling growth of maize (*Zea mays* L.) under salt stress at laboratory conditions. African Journal of Biotechnology, 12, 46: 6484-6490