

EVALUATION OF SALT TOLERANCE IN MAIZE HYBRIDS AT SEEDLING STAGE

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Abstract

Twenty four maize hybrids named as STM1 to STM24 were studied at five different salt concentrations of 0; 50; 100; 150; 200 mM NaCl at the National Maize Research Institute. After 7 days germination in room temperature, seedlings were transplanted to nutrient solutions salinized with salt. 17 day old seedlings were harvested for calculating of growth parameters such as root and shoot length, seedling dry matter production, ion Na⁺ and K⁺ content. As the results of the study, salt stress reduced plant growth of all hybrids but the STM10 and STM21 showed better salt tolerance while STM17 and STM18 presented salt-sensitive. The results of analysis for Na⁺ and K⁺ showed that among the hybrids, STM21 presented the lowest ion Na⁺ (2.057) and the highest ion K⁺ (2.763) in the seedlings.

Keywords: Maize hybrid, salt tolerance, seedling

INTRODUCTION

Salinization is the accumulation of salts in the soil, that is strong impact on agricultural production, the environment and economy of the countries (Rengasamy, 2006). According to Wild (2003) about 15% of the land in the world has been eroded, the physical and chemical characteristics of the soil has been modified, including soil salinization.

In Vietnam, saline soils are primarily formed by salt or saltwater from the sea or underground movement to the soil surface. Another reason is the using of saline water from the canals to the fields due to lack of fresh water. Evaporation during crop farming also causes of soil surface salinity. There is about three million hectares of land salinization and acidification, distributed mainly in the provinces of the Mekong Delta as Bac Lieu, Ca Mau, Kien Giang, Tra Vinh, Ben Tre, and the provinces in the Red River Delta as: Quang Ninh, Hai Phong, Thai Binh, Nam Dinh, Ninh Binh, Thanh Hoa (Soils and Fertilizers Institute, 2010).

Most crops tolerate salinity to a threshold level and above which will decrease in productivity (Khan *et al.*, 2006). Maize is pollinated crop which is able to tolerate to salinity (Paterniani, 1990). Research in maize genotypes is necessary to determine the materials for maize breeding programs with high yield and salt tolerance. The studies on salt tolerance in maize have been published by some national institutions and international organizations, which have given some scientific basis for salinity tolerance in maize. In Vietnam, salt tolerance in maize has not been studied yet. Facing of adverse climate change and the increasing salinization of agricultural land, the research on breeding salt-tolerant crops in general

and maize in particular is urgent for development of agriculture. Studies on salinity tolerance of maize on the field is very difficult, because the heterogeneity of soil physical and chemical properties and fluctuations in seasonal rainfall. Therefore, we conducted "Evaluation of salt tolerance in maize hybrid at seedling stage" with the objective assessment of maize hybrids for salt tolerance.

MATERIALS AND METHODS

Materials

The research was conducted at Maize Research Institute, Dan Phuong, Hanoi with 24 test crosses named as STM1, STM2, STM3, ..., STM24.

Methods

The seeds were soaked in distilled water and germinated at individual line for 5 to 7 days after transplant to Yoshida nutrition solution (Yoshida, 1976), which salinized with NaCl at 4 different salt (S) concentrations: S1 (50 mM); S2 (100 mM); S3 (150 mM); S4 (200 mM); and control 0 mM (S0). Salt was added in small amounts, 50 mM at 5 days after transplanting, the remain salt was provided after 7 days. The nutrition solution was changed twice a week. The experiment was arranged in randomized complete block design (RCBD) with 3 replications. After 17 days the plants were harvested separately in line and treatment. Harvested plants were washed in tap water and twice in distilled water and the following observation parameters were recorded:

- Survival date and salt tolerance ability: Number of dead plants after transplanting to solution culture.

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- Shoot length: 5 plants were taken for plants measurement and the length between the collar and tip of the longest leaf were measured in cm and the mean value was reported as shoot length.

- Root length: The plants taken for measuring of shoot length were used for root length measurement. The length between collar and tip of longest root was measured in cm and mean value was reported as root length.

- Plant dry weight: Five plants used for shoot and root length measurement that were first dried under shade for a while and then dried in a hot oven maintained at $70\text{ }^{\circ}\text{C} \pm 2$ for 48 hours, then cooled and weighed. The plant dry weight was expressed in gram/plant.

- Salt Tolerant Index (STI):

$$\text{STI (\%)} = \frac{\text{Total plant dry weight at treatment } S_x}{\text{Total plant dry weight at treatment } S_0} \times 100$$

(S_x : Treatment at salt concentration x; S_0 : Treatment at salt concentration 0).

- Ion Na^+ , K^+ accumulation: Ion Na^+ , K^+ accumulation were measured using a flame photometer.

- Statistical analysis: Data were analyzed with analysis of variance for factorial design with 3 replications using IRRISTAT statistical software.

RESULTS AND DISCUSSION

Survival date and salt tolerance ability

The observation showed that all crosses reduced growth and wilt after transplanting into saline solution, which presented differently between the

hybrids. In S4 treatment the plants presented the most in degradation but without dead plants after 17 days cultured in nutrition solution with salt. Among of them, STM21 showed the best tolerance in comparison to others.

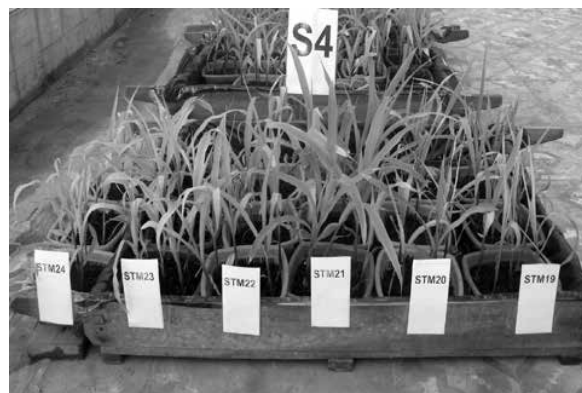


Figure 1. Effect of salinity on different maize crosses after 17 days of transplanting to 200 mM NaCl (S4)

Shoot length

Data in Table 1 shows that the shoot length of all crosses reduced when grown in saline solution. STM17 presented highest reduced shoot length percentage (45.78%) followed by STM18 (43.98%), whereas that decreased lowest in STM21 with only 19.72%. The average of shoot length of all hybrids were decreased 36.05% when grown in nutrition solution with 200 mM NaCl. The above result demonstrated that salinity makes a great impact on growth of plants. This was also found by André et al, 2004; Carlos et al, 2007 when researching on salt tolerance in maize.

Table 1. Effect of salinity on shoot length of different maize crosses at seedling stage

No.	Crosses	Shoot length (cm)					Mean	Reduced (%)
		Treatments						
		S0 (0mM NaCl)	S1 (50mM NaCl)	S2 (100mM NaCl)	S3 (150mM NaCl)	S4 (200mM NaCl)		
1	STM1	31.57	31.03	30.03	25.73	20.13	27.70	36.22
2	STM2	35.67	33.73	32.18	27.48	24.47	30.71	31.40
3	STM3	33.17	32.87	31.87	27.57	21.63	29.42	34.77
4	STM4	34.02	32.62	32.72	27.30	20.88	29.51	38.61
5	STM5	31.97	31.67	30.67	25.97	19.37	27.93	39.42
6	STM6	30.55	29.47	28.47	23.77	17.17	25.88	43.81
7	STM7	32.60	32.30	31.30	26.70	20.10	28.60	38.34
8	STM8	33.80	33.50	31.33	27.97	21.37	29.59	36.78
9	STM9	33.80	32.67	31.33	26.87	21.70	29.27	35.80
10	STM10	33.48	33.18	34.37	28.47	25.10	30.92	25.04

Table 1. Effect of salinity on shoot length of different maize crosses at seedling stage (*continued*)

No.	Crosses	Shoot length (cm)					Mean	Reduced (%)
		Treatments						
		S0 (0mM NaCl)	S1 (50mM NaCl)	S2 (100mM NaCl)	S3 (150mM NaCl)	S4 (200mM NaCl)		
11	STM11	34.40	32.53	31.53	27.67	22.80	29.79	33.72
12	STM12	32.93	32.63	31.63	27.33	21.33	29.17	35.22
13	STM13	33.73	33.43	33.53	28.00	25.02	30.74	25.84
14	STM14	29.87	29.57	28.57	23.87	17.27	25.83	42.19
15	STM15	31.53	31.23	30.23	25.53	18.93	27.49	39.96
16	STM16	34.50	32.67	33.20	25.93	21.90	29.64	36.52
17	STM17	33.20	31.40	28.90	22.33	18.00	26.77	45.78
18	STM18	31.13	29.90	28.90	23.00	17.60	26.11	43.98
19	STM19	32.07	31.77	30.77	26.07	20.63	28.26	35.66
20	STM20	34.20	31.23	30.23	25.40	20.93	28.40	38.79
21	STM21	33.13	33.90	32.90	28.80	26.60	31.07	19.72
22	STM22	32.27	31.97	30.97	25.13	22.30	28.53	30.89
23	STM23	32.68	32.38	31.38	23.63	21.80	28.38	33.30
24	STM24	27.13	26.83	25.83	21.13	15.20	23.23	43.47
	<i>Mean</i>	32.64	31.85	30.95	25.90	20.93	28.46	36.05

LSD_{.05}: 3.49

CV% : 17.6%

Root length

Root length was also affected by salinity, increasing salt concentration that means reduced root growth (Table 2) and which decreased positively in increasing salt concentration. However, the response of maize hybrids to salinity was different. The results showed that the average root length of STM18 was the shortest (9.53 cm) while STM10 was the longest (16.93 cm). However, in reduction of root length between the S0 and S4 treatments, the STM21 reduced lowest with 18.15%. When studying on maize in salt stress, Khan and McNeilly (2005) also showed that maize could be grown in saline conditions but root length decreased rapidly with increasing salt concentration.

Plant dry weight

As the shoot length and root length, dry weight of the seedling decreased with increasing salt concentration, the average plant dry weight of all crosses at S0 treatment was 0.535 gram/plant, whereas it decreased to 0.448 gram/plant at S4 concentration. Dry weight of the seedling in saline conditions obtained were very different between crosses, STM21 showed highest with 0.521 gram/plant while the lowest obtained in STM17 (0.404 gram/plant). The crosses which present high plant dry weight in salt concentration mean that have high salt tolerant index. The results

showed that STM21, STM10 and STM13 were better salt tolerant ability than other crosses. When researching on salt tolerance in maize, Muhammad et al (2010) found that salt tolerance in maize varieties was very different, the higher dry matter production could be, the better grew and developed in salt stress.

Ion Na⁺, K⁺ accumulation

Through the results of experiment, the crosses of tolerance and sensitive to salinity were selected for measurement of ion Na⁺, K⁺ accumulation. The results in table 4 showed levels of Na⁺ accumulation in plants increased with increasing salt concentration, the average Na⁺ content in S0 was 0.375%, increased to 4.951% in S4 treatment. Among the crosses, STM18 accumulated highest Na⁺ with 5.552% while STM21 showed lowest (4.392%) in the same salt concentration of 200 mM NaCl.

For potassium, the accumulation of K⁺ in all crosses was decreased with increasing salt concentration (Table 5). The average ion K⁺ accumulated of the crosses in S0 (0 mM NaCl) was 3.016% and which reduced to 1.931% in S4 concentration (200 mM NaCl). The STM21 maintaining K⁺ absorption was highest in all treatments, followed by STM10. The STM18 recognized as the cross which showed lowest of K⁺ uptake.

Table 2. Effect of salinity on root length of different maize crosses at seedling stage

No.	Crosses	Root length (cm)					Mean	Reduced (%)
		Treatments						
		S0 (0mM NaCl)	S1 (50mM NaCl)	S2 (100mM NaCl)	S3 (150mM NaCl)	S4 (200mM NaCl)		
1	STM1	17.87	17.67	16.67	14.87	12.87	15.99	27.99
2	STM2	16.07	15.87	14.87	13.07	11.07	14.19	31.12
3	STM3	16.80	16.60	15.60	13.80	11.73	14.91	30.16
4	STM4	17.82	17.62	16.62	15.15	14.22	16.28	20.21
5	STM5	19.10	18.90	16.83	14.63	13.90	16.67	27.23
6	STM6	16.20	16.00	15.00	13.20	11.00	14.28	32.10
7	STM7	18.67	18.47	16.23	15.93	13.73	16.61	26.43
8	STM8	15.93	19.60	14.73	13.47	11.33	15.01	28.87
9	STM9	18.60	18.40	16.80	14.60	13.67	16.41	26.52
10	STM10	19.80	15.73	18.33	16.27	14.53	16.93	26.60
11	STM11	16.20	16.00	15.00	13.40	11.53	14.43	28.81
12	STM12	17.93	17.73	16.73	15.07	13.07	16.11	27.14
13	STM13	18.25	18.05	17.05	16.00	14.38	16.75	21.19
14	STM14	17.23	17.03	16.03	14.23	12.03	15.31	30.17
15	STM15	17.05	16.85	15.85	14.05	11.85	15.13	30.50
16	STM16	16.73	16.53	15.53	13.73	11.53	14.81	31.08
17	STM17	16.75	16.55	14.82	12.62	10.48	14.24	37.41
18	STM18	17.20	17.00	14.47	12.20	9.53	14.08	44.57
19	STM19	16.07	15.87	14.87	13.07	11.80	14.33	26.56
20	STM20	18.00	17.80	15.87	15.20	13.33	16.04	25.93
21	STM21	18.00	17.80	18.20	16.33	14.73	17.01	18.15
22	STM22	17.87	17.67	16.33	15.27	13.87	16.20	22.39
23	STM23	16.23	16.03	15.03	13.23	12.07	14.52	25.67
24	STM24	16.58	16.38	15.38	13.58	11.38	14.66	31.36
	<i>Mean</i>	<i>17.37</i>	<i>17.17</i>	<i>15.95</i>	<i>14.29</i>	<i>12.49</i>	<i>15.45</i>	<i>28.26</i>

LSD_{.05}: 3.42

CV% : 13.80%

CONCLUSION

Shoot length, root length and plant dry weight of all crosses were reduced when increasing salt concentration, the STM17, STM18 reduced highest while the STM21 presented lowest in reduction of both shoot length and root length. The STM21, produced salt tolerant index followed by STM13 and STM10.

The accumulation of ion K⁺ decreased and increased in absorption of Na⁺ in salt stress. The STM21 showed the best accumulation of K⁺ and elimination of Na⁺.

Effect of salinity on the crosses were very different, among them STM21 and STM10 were identified to have high salt tolerant index.

Table 3. Effect of salinity on plant dry weight of different maize crosses at seedling stage

No.	Crosses	Plant dry weight (gram/plant)						Salt tolerant index (%)
		Treatments					Mean	
		S0 (0mM NaCl)	S1 (50mM NaCl)	S2 (100mM NaCl)	S3 (150mM NaCl)	S4 (200mM NaCl)		
1	STM1	0.502	0.491	0.451	0.425	0.371	0.448	73.79
2	STM2	0.547	0.508	0.469	0.441	0.387	0.470	70.81
3	STM3	0.515	0.476	0.427	0.396	0.341	0.431	66.19
4	STM4	0.503	0.465	0.419	0.424	0.317	0.425	63.00
5	STM5	0.490	0.499	0.440	0.392	0.362	0.437	73.93
6	STM6	0.569	0.529	0.468	0.422	0.349	0.467	61.34
7	STM7	0.528	0.490	0.441	0.405	0.339	0.441	64.20
8	STM8	0.501	0.460	0.414	0.380	0.312	0.413	62.28
9	STM9	0.487	0.513	0.468	0.373	0.300	0.428	61.58
10	STM10	0.577	0.536	0.509	0.491	0.450	0.513	77.94
11	STM11	0.528	0.490	0.439	0.405	0.344	0.441	65.15
12	STM12	0.548	0.510	0.460	0.419	0.358	0.459	65.27
13	STM13	0.560	0.525	0.498	0.487	0.442	0.503	78.99
14	STM14	0.515	0.469	0.408	0.362	0.317	0.414	61.53
15	STM15	0.523	0.482	0.424	0.374	0.301	0.421	57.62
16	STM16	0.516	0.476	0.419	0.369	0.293	0.415	56.78
17	STM17	0.541	0.448	0.395	0.350	0.289	0.404	53.51
18	STM18	0.554	0.449	0.403	0.357	0.276	0.408	49.73
19	STM19	0.547	0.508	0.460	0.433	0.367	0.463	67.05
20	STM20	0.569	0.511	0.460	0.416	0.354	0.462	62.21
21	STM21	0.575	0.541	0.522	0.502	0.465	0.521	80.81
22	STM22	0.538	0.495	0.460	0.436	0.405	0.467	75.34
23	STM23	0.549	0.510	0.454	0.417	0.357	0.457	65.07
24	STM24	0.550	0.511	0.451	0.404	0.327	0.449	59.36
	<i>Mean</i>	<i>0.535</i>	<i>0.496</i>	<i>0.448</i>	<i>0.412</i>	<i>0.351</i>	<i>0.448</i>	<i>65.63</i>

LSD_{.05}: 0.067

CV% : 9.30%

Table 4. Effect of salinity on ion Na⁺ accumulation of different maize crosses

No.	Crosses	Na ⁺ content (% dry weight)					Mean
		Treatments					
		S0 (0mM NaCl)	S1 (50mM NaCl)	S2 (100mM NaCl)	S3 (150mM NaCl)	S4 (200mM NaCl)	
1	STM10	0.363	0.795	1.733	3.355	4.509	2.151
2	STM13	0.373	0.848	1.786	3.408	4.988	2.281
3	STM17	0.390	1.022	2.579	4.352	5.472	2.763
4	STM18	0.363	1.102	2.659	4.432	5.552	2.822
5	STM21	0.380	0.702	1.639	3.262	4.392	2.075
6	STM22	0.380	0.943	2.043	3.665	4.795	2.365
	<i>Mean</i>	<i>0.375</i>	<i>0.902</i>	<i>2.073</i>	<i>3.746</i>	<i>4.951</i>	<i>2.409</i>

LSD_{.05}: 0.146

CV% : 13.40%

Table 5. Effect of salinity on ion K⁺ accumulation of different maize crosses

No.	Crosses	K ⁺ content (% of dry weight)					Mean
		Treatments					
		S0 (0mM NaCl)	S1 (50mM NaCl)	S2 (100mM NaCl)	S3 (150mM NaCl)	S4 (200mM NaCl)	
1	STM10	3.037	2.927	2.787	2.473	2.183	2.681
2	STM13	3.027	2.933	2.807	2.517	2.227	2.702
3	STM17	3.003	2.730	2.220	1.780	1.480	2.243
4	STM18	2.977	2.733	2.223	1.693	1.393	2.204
5	STM21	3.023	3.080	2.860	2.570	2.280	2.763
6	STM22	3.030	2.937	2.587	2.312	2.022	2.577
Mean		3.016	2.890	2.581	2.224	1.931	2.528

LSD_{.05}: 0.113 CV% : 12.10%

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RESEARCH ON PRODUCTION OF BIOLOGICAL PRODUCTS OF *CHAETOMIUM* FOR CONTROLLING FUNGAL DISEASES ON TEA, COFFEE AND RUBBER

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Abstract

Two strains of *Chaetomium* which were popular and with high antagonistic activity from tea, coffee and rubber soil in Northern Vietnam were identified, including *Chaetomium bostrychoides* and *Chaetomium globosum*. These strains had strongly antagonistic activity to *Fusarium* sp. causing root rot on tea, coffee and rubber and *Colletotrichum* sp. causing tea anthracnose. New identified *Chaetomium* strains were successfully used to produce biological products for controlling disease called CP2-VMNPB. The product composition was capable to exclude fungal diseases well such as *Fusarium* sp. (85.62%); *Colletotrichum* sp. (81.80%) after one month in laboratory conditions; in greenhouse conditions, the effect on root rot tea and coffee reached 83.30%, with powdery mildew disease on rubber *Oidium Hevea*, the effect reached 81.17% after 3 months.

Keywords: Tea, coffee, rubber, bio-fungicide, *Chaetomium*, *Fusarium*, *Colletotrichum theae-sinensis*, *Oidium heveae*

INTRODUCTION

Tea, coffee and rubber are perennial plants which survive and grow throughout the year and they are continuously damaged by many types of pestilent insect and diseases.

The control of disease on tea, coffee and rubber has mainly based on chemical methods which seem less effective and leave residues in the environment and are harmful to the health of consumer as well as producer. There have been studies and applications of beneficial microorganisms to control plant diseases, one of the microorganisms were *Chaetomium* (K. Soyong 1989). *Chaetomium* falls under the list of saprophytic fungi which have strong competitiveness against disease fungi, especially *Chaetomium globosum* and *Chaetomium bostrychoides* with strong resistance to pathogenic fungi of the *Fusarium* and *Helminthosporium* (Tveitand Moore, 1954), *Alternaria*, *Collectotrichum* (Vannacci *et al.*, 1978; C. Talubnuc *et al.*, 2010), and so on. The antagonistic activity of *Chaetomium* is due to the synthesis of antibiotic *Chaetoglobosin*, which breaks down cell membranes, making the cytopla broken and lose toxicity of fungal diseases (Di Petro, 1992; K. Soyong, 2007). In addition, *Chaetomium* helps stimulate growth and development, increase the resistance of plants (Le Thi Anh Hong, 2005; Doke *et al.*, 1991).

Based on the useful characteristics of *Chaetomium*, the Northern Mountainous Agriculture and Forestry Science Institute (NOMAFSI) has carried out the research and production of *Chaetomium* compositions to eliminate diseases on tea, coffee, rubber and other crops.

MATERIALS AND METHODS

Materials

Soil samples from the tea, coffee and rubber gardens for isolating *Chaetomium*; fungal pathogens were *Fusarium* sp., *Colletotrichum theae-sinensis* and some other pathogenic fungi from sources stored at the Department of Biotechnology & Plant Protection, NOMAFSI.

Isolate antagonistic *Chaetomium*; Review of active resistant characteristics of *Chaetomium* against root rot on tea, coffee and rubber caused by *Fusarium* sp. Tea leave rot caused by *Colletotrichum camelliae* nursery (C. sinensis - theae).

Methods

- Isolation and purification of *Chaetomium*: The soil samples were cut into small pieces and put on the surface of a petri dish with filter paper of 1 cm², humidifying and moisturizing regularly at room temperature to trap fungus by the method of K. Soyong 1989. Each fungus was separated by sterilized implants and implanted onto PDA to track the generation and development of spores.

- To assess the activity of the *Chaetomium* against fungus disease: Perform the experiment by conducting transplants symmetry 2 antagonist fungus and pathogens on the surface of potato dextrose agar in petri dishes 9cm, transplanting in place 1cm from the edge of the petri dish:

Disc 1: Mushrooms antagonists (*Chaetomium*);

Disc 2: Mushrooms antagonists (Changing domain name) and fungal pathogens (NGB);

Disc 3: Pathogens (control);

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