

Effect of NaCl on Physiological Performance and Yield of Wheat Hybrids

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Abstract

The effect of sodium chloride on the physiological parameters of wheat plants was studied in laboratory and field conditions. The effect of NaCl at concentrations of 0, 100 and 200 μM on seed germination and the development of seedlings of third generation hybrids was studied. Hybrids of the third generation were cultivated on the territory of the field experiment (0.98% NaCl salt in the soil); the physiological processes occurring in the leaves, changes in the amount of photosynthetic pigments, in the relative content of water, and in the yield were studied. Salt reduced the germination energy of hybrid seeds, as well as the amount of photosynthetic pigments in the leaves, photosystem II activity, and the relative amount of water, with different effects. The percentage of germination at a NaCl concentration of 200 μM was higher in hybrids of Tale-38 X Kyrmyzy Gul-1, Gobustan X Sheki-1, Vugar X Barakatly-95, Mirbashir-50 X Shiraslan-23, Gobustan X Barakatly-95, 24-Karabakh X Shark. It was found that the sum of pigments chl(a+b) and the amount of carotenoids, the activity of photosystem II and the relative amount of water were higher in the hybrids Lyagatly-80 x Mirbashir-128, Vugar x Barakatly-95, Tale-38 x Kyrmyzy Gul-1, Gobustan x Sheki-1, Mirbashir-50 x Shiraslan-23 in compared to other hybrids, and these hybrids were more tolerant to salt.

Keywords: wheat, NaCl, hybrid, pigment, activity, tolerance

Introduction

Salinity is one of the main abiotic environmental stresses affecting crop productivity [Grewal et al., 2010]. Approximately 7 percent of the total area of the

globe is already saline [Musyimi et al., 2007]. It is known that most of the arable land in Azerbaijan is subject to varying degrees of salinity. Salinization is caused by both natural factors and anthropogenic factors. Natural factors include rising groundwater levels, while anthropogenic factors include unbalanced supply of mineral fertilizers and improper irrigation. At the same time, the rise in the level of the World Ocean and, as a result, the outflow of groundwater to the surface is one of the main natural factors of soil salinity. Soil salinity causes water deficiency in the plant, which, as a result, disrupts the normal course of physiological processes in cells [Greenway and Munns, 1980]. Due to the influence of salts, the osmotic pressure of the soil solution increases, which leads to a stronger binding of water to soil particles. Absorption of water by the root becomes difficult, osmotic stress occurs. On the other hand, an excess of Na^+ ions entering the plant negatively affects metabolic processes. Plants have physiological responses to the effects of salt, which are manifested to varying degrees in different plant species.

In the studies carried out, it was found that plants are more sensitive to salt in the early stages of development, during seedling growth and biomass accumulation [Greenway and Munns, 1980; Chartzoulakis, 1994]. It has been established that chloride salinity reduces the amount of water in the leaves, disrupts the mechanism of stomata movement and limits the absorption of carbon dioxide. In plants, this is done through the synthesis of proline and the accumulation of NADP at the level of the whole organism under saline conditions. These compounds regulate the formation of reactive oxygen species and lipid peroxidation. This situation leads to a slowdown in plant growth and development, as well as a decrease in productivity and quality [Siddiqui et al., 2008; Basalah, 2010; Fahid et al., 2005].

Wheat (*Triticum aestivum* L.) is considered the main food source for more than 35 percent of the world's population. Salt stress, like other plants, negatively affects the growth and productivity of wheat plants [Sabura and Kiarostami, 2006; Mehmet et al., 2006]. The detrimental effects of salinity on germination and early germination of crops are mainly due to osmotic stress, which inhibits water uptake, or specific ionic toxicity [Wakeel et al., 2011]. However, different plant species may exhibit different responses to saline conditions [Mehmet et al., 2006; Shahid et al., 2011]. The main goal of the research work is to study the effect of various molar concentrations of NaCl on the germination energy of seeds, content of photosynthesis pigments, relative water content and productivity of F3 generation wheat hybrids under laboratory and field conditions.

Materials and methods

The experiment was carried out in laboratory conditions by studying the germination of seeds of F3 generation hybrids and the development of 7-day-old seedlings. Seeds of 24 F3 hybrids were soaked for 24 hours and after disinfection in 1% hydrogen peroxide solution were germinated in Petri dishes on filter paper at a temperature of 20 degrees in concentrations of 0, 100 and 200 μM NaCl salt solution. The percentage of seed germination was determined. Hybrid seeds were

sown in parallel in saline soil conditions. Generation F3 as an object of study: Layagatly-80 X Mirbashir-128, Kyzyl bugda X Guneshli, Terter X Karabakh, Tale-38 X Kyrmyzy Gul-1, Gobustan X Sheki-1, Murov X ♂ Dagdash, Bezostaya -1 X Kyrmyzy Gul-1, Dagdash X Murov, Nurlu -99 X Layagatly-80, Karabakh X Karakylchyk, Sheki-1 X Gobustan, Vugar X Barakatly - 95, Kyrmyzy Gul- 1 X Tale-38, Barakatly -95 X Vugar, Karabakh X Terter, Aran x Kyrmyzy Gul-1, Mirbashir-50 X Shiraslan-23, Karabakh X Gobustan, Barakatly-95 X Gobustan, Gobustan X Kyrmyzy Gul-1, Gobustan X Barakatly-95, Gobustan X Karabakh, Karabakh X Mirbashir-128, Karabakh X Shark hybrids were taken. The seeds were sterilized in 1.5% sodium hypochlorite solution for 10 minutes and washed thoroughly with sterile distilled water. Although field screening for salt tolerance has the advantage of screening germplasm *in vivo*, it is less effective at the germination and early growth stages and more expensive than screening under controlled conditions [Shannon and Noble, 1990]. To do this, ten seeds were placed in sterile Petri dishes 10 cm in diameter on filter paper with the addition of salt solutions. The seeds were subjected to salt stress by exposing them to 30 ml of control (with the addition of NaCl) 0, 100, 200 mM NaCl solutions. The dishes were sealed with paraffin and placed under controlled conditions ($25 \pm 2^\circ$ C. during the day; 16/8 h light/dark; illumination 4500 lux). Seeds were considered germinated when the root size exceeded 2 mm.

The average percentage of germination was calculated by the number of seedlings with a length of at least 5 mm in each Petri dish on the 7th day of growth [Maguire, 1962].

Hybrids were planted under normal conditions in the field experiment area and were also cultivated on the territory of the field experiment (0.98% NaCl salt in the soil). The amount of chlorophyll and carotenoids in the leaves was determined at the stage of flowering. 0.1 g sample of leaves was homogenized by grinding in 96% alcohol with the addition of CaCO₃ in a mortar, centrifuged at 200 g, and precipitated. The amount of chlorophyll and carotenoids was determined by measuring the optical density of a solution of chlorophyll in alcohol at a wavelength of 665, 649, and 440 nm on a sp-2000 spectrophotometer. [Wintermans, De Mots, 1965]. The activity of the second photosystem was determined by Fv/Fm using a MINI-PAM photosynthesis analyzer (photosynthesis yield analyzer, Germany). Here, $F_v = F_m - F_0$; F_0 is the fluorescence of leaves illuminated with weak light after exposure in the dark, F_m is the fluorescence of leaves saturated with light. Leaf water loss (LWL) was determined by the method of Tambussy et al. [Tambussi E.A., Noges S, 2005].

Observations of plants and structural elements of the product were carried out according to the existing method [Musaev A.S., Guseynov Kh.S., 2008]. To determine the resistance of varieties to salt stress, we used the stress resistance index given by Rosielle and Hambelen in 1981 [Rosielle and Hambelen., 1981].

Statistical analyses

The experiment employed three biological replicates and each replicate was reproduced independently three times. Statistical processing of the results was done

using the licensed IBM SPSS Statistics software package. The assessment of the reliability of variations in arithmetic means was carried out based on the Student's coefficient. Differences between groups were considered significant at a two tailed level of significance $p \leq 0.05$. The diagram was constructed using the Graph Pad Prism-8 software.

Results and discussion

It has been established that the percentage of seed germination of hybrids of the F3 generation at various concentrations (100 and 200 mmol) of NaCl salt in laboratory conditions decreases compared to the control variant. In general, NaCl salt had a negative effect on the germination of hybrids. Significant ($p \leq 0.05$) differences in various germination rates were observed between wheat hybrids, as well as salinity levels. In addition, wheat genotypes responded differently to different levels of salinity due to a significant interaction ($p \leq 0.05$) between these two factors. An increase in salinity not only reduced the percentage of germination, but also delayed the onset of germination in all wheat hybrids. Although a smaller decrease was observed in the 100% NaCl medium than in the control, a greater decrease was observed in the 200% NaCl medium compared to other hybrids. When comparing hybrid forms of salt stress, the greatest resistance at a salinity level of 200 mmol NaCl is shown by Layagatli-80 X Mirbashir -128, Taleh-38 X Kyrmyzy Gul-1, Gobustan X Sheki-1, Murov X ♂ Dagdash. , Vugar X Barakatly-95, Mirbashir-50 X Shiraslan-23, Gobustan X Barakatly-95, Karabakh X Mirbashir-128, Karabakh X Shark hybrids. (Figure 1).

Salinity is one of the main factors limiting the germination of plant seeds (Khan and Gulzar, 2003).). The inhibitory effect of salinity on the germination of various plants has been studied previously (Farooq et al., 2011; Elouaer and Hannachi, 2012; Afzal et al., 2012). The decrease in germination with salinity is attributed to the combined effect of osmotic pressure [(Moud and Maghsoudi, 2008)] and salt toxicity [Saboora and Kiarostami, 2006] or the effect of additional chlorine. [Almodares et al., 2007] Rahman and colleagues [Rahman et al., 2008] found that salinity significantly retards germination, mainly due to changes in the water regime caused by high salt accumulation in intercellular spaces [Khan and Gulzar, 2003; Zhang et al., 2007].

The inability of seeds to germinate under saline conditions may be associated with damage to the embryo. Na^+/Cl^- ions [Khajeh-Hosseini et al., 2003] inhibit seed water uptake [Mehmat et al., 2006; Saboora and Kiarostami, 2006]. In this case, salt-tolerant *Triticum* spp. had lower Na^+ accumulation than salt-sensitive ones [Ali et al., 2004]. In our studies, the greatest decrease in the effect of 200 mmol salt stress NaCl on the germination energy was recorded in the hybrids Bezostaya-1 X Qyrmyzy Gul-1, Dagdash X Murov, Gobustan X Karabakh, Terter X Karabakh, Karabakh X Gobustan (Figure 2).

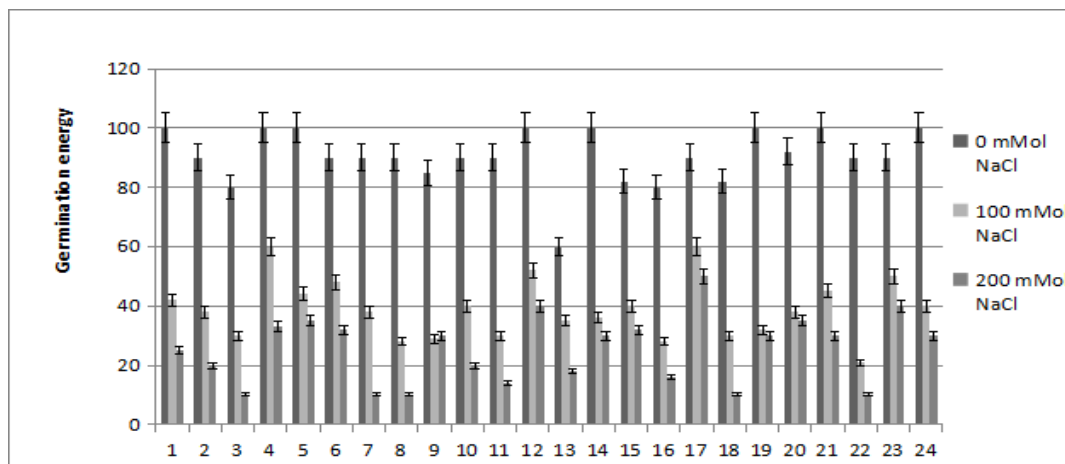


Figure 1. Effect of various NaCl salt concentrations (0.100, 200 mmol) on the germination energy (%) of hybrid seeds.

1- Layagatly-80 X Mirbashir -128, 2- Kyzyl bugda X Gunashli, 3- Terter X Karabakh, 4- Tale-38 X Kyrmyzy Gul-1, 5- Gobustan X Sheki-1, 6- Murov X ♂ Dagdash, 7 -Bezostaya -1 X Kyrmyzy Gul-1, 8- Dagdash X Murov, 9-Nurlu -99 X Layagatly-80, 10-Karabakh X Karakylchyk-2, 11-Sheki-1 X Gobustan, 12- Vugar X Barakatly-95, 13-Kyrmyzy Gul-1 X Tale-38, 14-Barakatli-95 X Vugar, 15-Karabakh X Terter, 16-Aran X Qyrmyzy Gul-1, 17-Mirbashir-50 X Shiraslan-23, 18-Karabakh X Gobustan, 19 - Barakatly-95 X Gobustan, 20-Gobustan X Kyrmyzy Gul-1, 21-Gobustan X Barakatly-95, 22-Gobustan X Karabakh, 23-Karabakh X Mirbashir-128, 24-Karabakh X Shark.

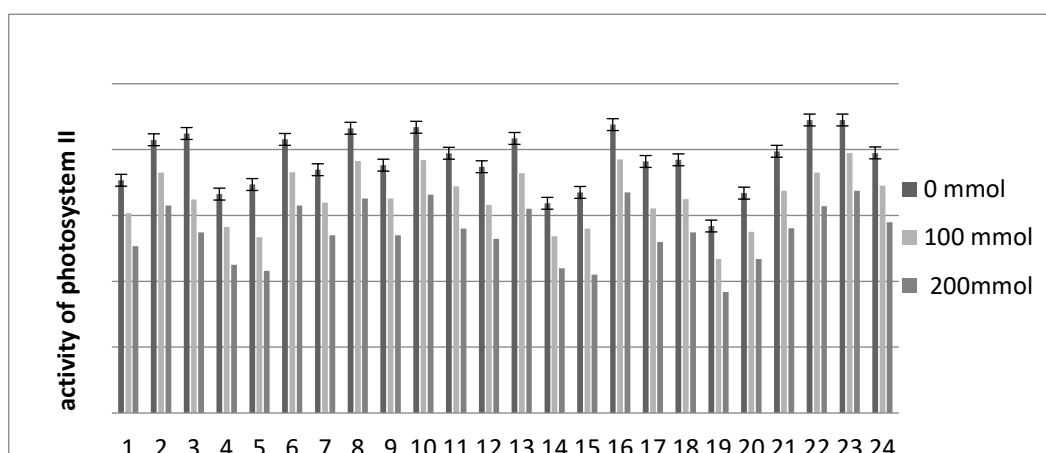
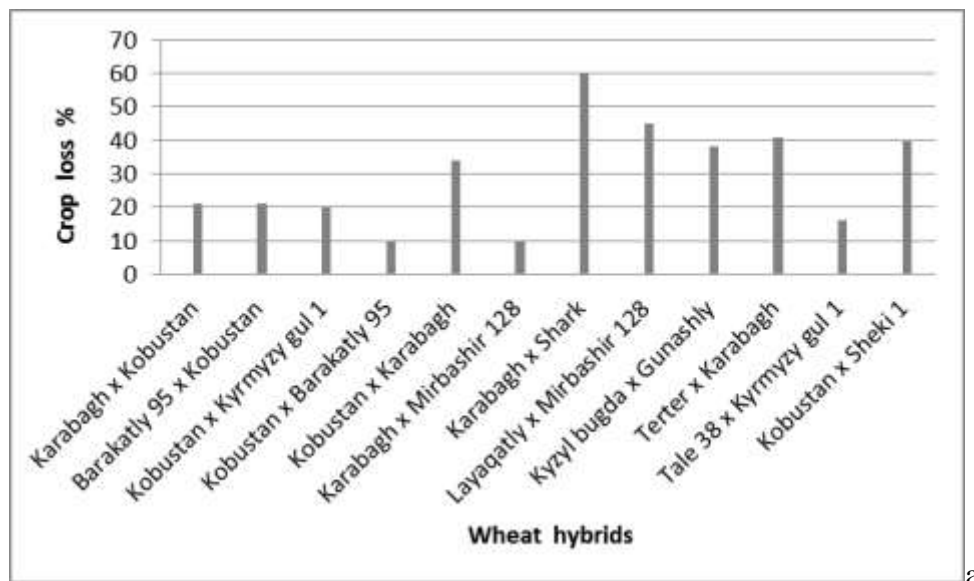


Figure 2. Effect of different concentrations of NaCl salt (0, 100, 200 μ M) on the activity of photosystem II (F_v/F_m) in wheat hybrids. (.Names of hybrids as in figure 1.)

One of the most useful indicators of wheat is grain yield. A high concentration of salt leads to a decrease in grain yield. The study of crop losses as the main indicator of yield is one of the main conditions. The smallest crop losses in Karbagh x Gobustan-21%, Karabakh x Terter-5%, Mirbashir-50 x Shiraslan-23-5%, Barakatly-95 x Vugar-15%, Barakatly-95 x Gobustan-19%, Karabakh x Mirbashir 128-10%, Karabakh x Terter-5%, Murov x Dagdash-8% hybrids are determined (Figure 3. (a,b)). Comparing hybrid forms, it can be seen that some of the hybrids suffered relatively less yield loss due to salt exposure. This suggests that while maintaining these long-lived hybrids, they can be planted in the future on moderately saline soils. The studied indicators are reflected in the following histograms (Fig. 1-3). The authors also studied the effect of salinity on the reduction rate of NADP and noted inter-sort differences in this indicator. Based on the obtained results, it was concluded that the main reason for the decrease in the rate of reduction of NADP under salinity is the decrease in the rate of the Calvin cycle [Villora, Pulgar et al., 1997]. During a comparative analysis of the effect of salt stress on the chlorophyll pigments of the studied hybrid forms in saline soil (0.98% NaCl), differences in the booting phase appeared between the hybrids. The effect of stress caused an increase in the amount of photosynthesis pigments in some hybrids and a relatively smaller amount in others (table 1). As can be seen from the table, the influence of salt in the soil had a specific effect on individual hybrids. Thus, under the influence of salt, the content of chl(a+b) and carotenoids in the leaves of the hybrids Layagatli X Mirbashir-128, Terter × Karabakh, Gobustan x Sheki-1, Mirbashir-50 X Shiraslan -23 was higher compared to other hybrids. Hybrids Kyrmyzy Gul -1 x Tale-38, Nurlu-99 x Layagatly 80 and Gobustan x Barakatly-95 were more sensitive to salt stress according to these indicators.



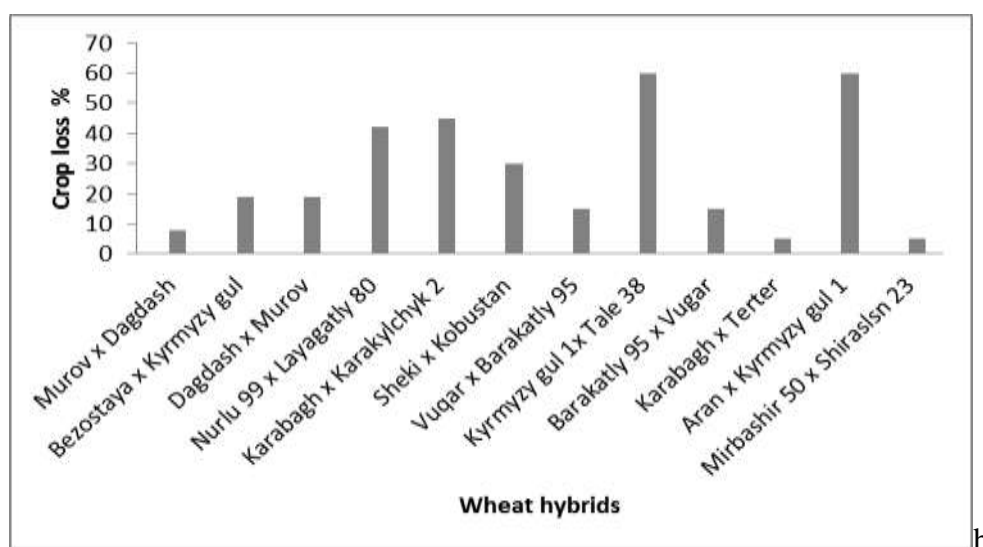


Figure 3. (a, b) Productivity indicators in F3 generation hybrid forms (according to yield loss (%)).

The environmental stresses affect photosynthesis and development of plants [Villora, et al.1997.], which is also associated with its toxic effects. Under the action of Na⁺ ions, the activity of ATPase pumps that regulate the balance of Na⁺ and K⁺ ions in the cell is disrupted, Na⁺ ions enter the cell, and the cell organelles exit the turgor state to the state of plasmolysis.

In the process of evolution, all organisms, including plants, develop protective mechanisms against environmental stress factors. Therefore, when assessing resistance to stress factors, it is necessary to take into account the individual characteristics of each plant genotype [Azizov I.V., Khanyshova M.A., 2019]. Under saline stress, a number of destructive changes occur in plant chloroplasts that prevent the normal course of photosynthesis. At the same time, it is noted that in plants resistant to salinity, there is a greater breakdown of chloroplasts and a decrease in the intensity of photosynthesis [Khosravinejad., et al 2008].

As can be seen from the table, Mirbashir-50 x Shiraslan-3, 39% Barakatly-95 x Vuqar and Karabakh x Mirbashir-128 are characterized by a relatively high water content and are more tolerant than other hybrids. Among the hybrids, a greater decrease in the relative amount of water was observed in the forms Kyrmyzy Gul-1 x Taleh-38, Nurlu-99 x Layagatly-80, Gobustan x Barakatly-95. A decrease in the relative amount of water due to stress indicates the absence of turgor, which is necessary for the process of tension in the cell [Katerji, N., Van Hoorn, J.W., 1997]. The effect of prolonged NaCl salinity on tolerant (Karchia 65) and sensitive (HD-2687) wheat genotypes was studied. Salinity decreased the relative amount of water, chlorophyll and ascorbic acid and increased H₂O₂. In sensitive varieties, this

change was observed more sharply than in resistant varieties [Khan. M.A. 2008]. , Thus, on the basis of the study, it was found that salt stress has a negative effect on the relative water content in the leaves of wheat hybrids.

Table 1. The effect of salt stress on the amount of chlorophyll a, chlorophyll b and carotenoids in hybrid (mg/g wet weight)

F ₃ hybrids	Chl (a+b)		Carotenoids		RWC, %	
	Control	Salt	Control	Salt	Control	Salt
1. Layaqatly -80 × Mirbashir-128	2.6±0.11	4.42±0.37	1,09±0.21	3,09±0.21	86	38
2. Qyzyl bugda × Gunashli	3.4±0.17	2.61±0.41	1,04±0.33	2,04±0.33	87	31
3. Terter × Karabagh	2.2±0.19	3.77±0.39	3,09±0.17	4,09±0.17	86	35
4. Tale-38 × Kyrmyzy Gul-1	2.7±0.13	1.22±0.41	0,78±0.26	0,78±0.26	83	36
5. Sheki-1 × Gobustan	2.4±0.17	2.83±0.35	1,05±0.14	1,15±0.14	88	38
6. Murov × Dagdash	3.1±0.21	1.59±0.43	1,61±0.33	1,06±0.33	82	37
7. Bezostaya-1 × Kyrmyzy Gul-1	2.1±0.33	1.17±0.31	1,11±0.21	1,01±0.21	81	35
8. Dagdash × Murov	3.3±0.32	1.35±0.28	1,71±0.32	1,61±0.32	81	32
9. Nurlu-99 × Layaqatli-80	3.2±0.33	1.44±0.19	1,32±0.19	1,12±0.19	84	35
10. Karabagh × Garagilciig-2	2.30.28	1.53±0.14	1,32±0.27	1,22±0.27	83	27
11. Gobustan × Sheki-1	2.60. ±24	3.38±0.17	1,17±0.16	2,17±0.16	83	37
12. Vugar × Barakatli-95	2.8±0.16	1.26±0.29	1,18±0.13	1,08±0.13	87	38
13. Kyrmyzy Gul-1 × Tale-38	2.7±0.13	1.32±0.22	1,17±0.41	1,11±0.41	86	28
14. Barakatli-95 × Vugar	2.9±0.23	1.51±0.18	1,35±0.15	1,35±0.15	89	39
15. Karabagh × Tartar	2.5±0.12	1.04±0.31	0,93±0.37	0,88±0.37	88	37
16. Aran × Kyrmyzy Gul-1	3.1±0.31	1.29±0.35	1,44±0.44	1,14±0.44	91	32
17. Mirbashir-50 × Shiraslan-23	2.4±0.22	3.22±0.16	1,08±0.34	2,08±0.34	91	38
18. Karabagh x Gobustan	2.9±0.27	1.59±0.32	2,12±0.16	1,12±0.16	85	33
19. Barakatli-95 × Gobustan	2.2±0.26	1.23±0.12	2,58±0.27	1,58±0.27	80	36
20. Gobustan × Kyrmyzy Gul-1	3.3±0.36	1.52±0.13	2,01±0.13	2,01±0.13	88	34
21. Gobustan × Barakatli-95	2.9±0.36	2.01±0.44	2,22±0.39	1,22±0.39	84	44
22. Gobustan × Karabagh	2.5±0.12	1.75±0.41	1,92±0.36	1,32±0.36	82	38
23. Karabagh × Mirbashir-128	2.2±0.22	2.31±0.39	1,17±0.22	1,07±0.22	84	39
24. Karabagh × Shark	2.3±0.21	1.93±0.19	1,92±0.33	1,22±0.33	76	32

Each value represents the mean ± SD (standard deviation) for the mean n = 3 independent experiments p = 0.05.

Changes in the water potential of leaves of different species and varieties of plants and other physiological parameters under salinity conditions have also been established in the works of a number of researchers. Thus, under the influence of

salt, the content of chl(a+b) and carotenoids in the leaves of the hybrids Layagatli X Mirbashir-128, Terter × Karabakh, Gobustan x Sheki-1, Mirbashir-50 X Shiraslan -23 was higher compared to other hybrids. Hybrids Kyrmyzy Gul -1 x Tale-38, Nurlu-99 x Layagatly 80 and Gobustan x Barakatly-95 were more sensitive to salt stress according to these indicators. According to the activity of Photosystem II, we can say that Terter X Karabakh, Tale-38 X Kyrmyzy Gul-1, Nurlu-99 X Layagatly-80, Sheki-1 X Gobustan, Barakatly-95 X Vugar, Mirbashir-50 X Shiraslan-23, Karabakh X Mirbashir-128 hybrids light fluorescence was observed, the photosystem showed high activity. By inhibiting the entry of potassium and calcium into cells, sodium and chlorine ions also prevent their transportation to intensively growing organs [Downton et al., 1985; Essa, 2002]. purchase of plants that can have a special place [Belovalova., 2011]. However, it has been determined that the effect of salt may be less at one stage of development and more at another. According to most authors, the stage when plants are most sensitive to salinity is the beginning of ontogenesis. Plants are more sensitive to the effect of salt during the period when the beginnings of spikers and bushing nodes are formed in grains [Maas et al.,1997].

Conclusions

When comparing the percentage of germination, the amount of pigments, the activity of photosystem II and the relative amount of water in F3 hybrids according to the influence of salt stress (NaCl 100, 200 mmol), Layagatly-80 X Mirbashir -128, Tale-38 X Kyrmyzy Gul-1, Gobustan X Sheki -1, Murov X ♂ Dagdash, Vugar X Barakatly-95, Mirbashir-50 X Shiraslan-23, Gobustan X Barakatly- 95, Karabakh X Mirbashir-128, Karabakh X Shark was higher. In saline soil (0.98 NaCl), the effect of salt on productivity indicators, including the weight of grain in one spike, caused yield loss. Reduction of grain weight per spike in Gobustan X Barakatly-95, Karabakh X Mirbashir-128, Tale-38 X Kyrmyzy gul-1, Murov X ♂ Dagdash, Vugar X Barakatli-95, Karabakh X Terter, Mirbashir-50 X Shiraslan-23 hybrids and yield loss was less than other hybrids. In the future, these hybrids can be used for growing in relatively saline soils.

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