Effect of Drought-induced Stress on Seed Germination and Seedling Growth of *Zea mays* L.

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**ABSTRACT**

**Background:** Drought is one of the major abiotic factors leading to diminishing growth, development and productivity of plants worldwide. Considering that germination is the first phase of growth which in large measure determines plant quality and yield, knowing the effects of different factors on this process is of major importance. This paper studies the effect of drought-induced stress on seed germination and seedling growth of *Zea mays* L. (the Sweet corn and the hybrid Pioneer B23).

**Methods:** The effect of water stress was caused by different concentrations of mannitol: 5%, 10% and 20%. In the control, we used distilled water. The germination test was performed in three trials of 45 seeds each. The germination percentage, germination potential, drought resistance index were calculated at 3, 5 and 7 days. Growth of seedlings and biomass content were calculated at 14 days.

**Result:** The results show significant differences between the variety and hybrid examined. The pioneer B23 seed germinated in larger number and more quickly. The Sweet corn variety seedling growth was completely absent after treatment with mannitol. The observed difference is certainly not just a consequence of higher mannitol concentrations, but also a difference in the water-retention capability of the variety and hybrid studied.

**Key words:** Corn, Drought resistance, Germination, Mannitol, Stress.

**INTRODUCTION**

In the last few decades, climate changes have grown more pronounced, causing rising temperatures and an increasing number of countries have faced the adverse effects of drought (Godfray et al. 2010) and limited agricultural production (Liu et al. 2015). As drought is a factor that substantially disrupts physiological processes in plants (inhibiting germination, reducing growth, decreasing yields and disrupting major metabolic processes), researchers have been paying increased attention to this problem, making significant progress in mitigating adverse effects of drought and producing increasingly drought-tolerant varieties (Machado Neto et al. 2004; Farooq et al. 2009; Khodarahmpour, 2011; Jain et al. 2013; Liu et al. 2015; Koka et al. 2015; Duan et al. 2017; Mahantesh et al. 2018; Noori et al. 2018). Studies on the genetic variation of plants in relation to water stress have become increasingly important in recent times, both between plant species and between individual genotypes (varieties and hybrids) within a species. The difference in drought resistance of different maize genotypes have been examined by Grzesiak (2001), Grzesiak et al. (2012), Khodarahmpour (2011), Liu et al. (2015). This research is of great interest from a practical as well as from a theoretical point of view. In other words, finding resistance to water stress can help breeders create new varieties and hybrids that would be more tolerant and resistant to drought stress (Farooq et al. 2009; Khodarahmpour, 2011; Jain et al. 2013; Liu et al. 2015; Koka et al. 2015; Duan et al. 2017).

As a consequence, numerous tests have been carried out in recent times to determine, based on a number of physiological criteria, which varieties and hybrids are more resistant to insufficient plant water supply, i.e. to drought. This paper aims to contribute to this effort, testing the effect of water deficit (stress), caused by different concentrations of mannitol, on seed germination and seedling growth of certain variety or hybrid of maize, in order to determine the sensitivity - or tolerance - of the tested variety and hybrid to stress conditions.

**MATERIALS AND METHODS**

We monitored the impact of water deficit on seed germination and seedling growth of maize (the variety Sugar corn and the hybrid Pioneer B23). The maize seeds were first sterilized with alcohol, then rinsed and left overnight to cause imbibition. The effect of water stress was caused by different concentrations of mannitol: 5%, 10% and 20%. In the control, we used distilled water. The germination test was performed in three trials of 45 seeds each. After experiment set-up, seed germination took place at a...
temperature of 26°C in the thermostat. The germination percentage was calculated at 3, 5 and 7 days and after seven days the germination potential, drought resistance index, growth of seedlings, biomass and photosynthetic pigment content were calculated.

Germination percentage

The germination percentage was calculated as follows:
\[
\text{Germination percentage} = \left( \frac{\text{number of germinated seeds}}{\text{total number of seeds}} \right) \times 100\% \quad (Liu \ et \ al. \ 2015).
\]

Germination potential

The germination potential is an index that indicates the ratio of the germination percentage and the uniformity of germination and is calculated as follows: Germination potential = (Germinated seed number at germination peak/Test seed number) \times 100\% (Liu \ et \ al. \ 2015).

Germination index of drought resistant

Germination index of drought resistant were calculated as follows: Seed germination index of drought resistance = seed promptness index under water stress (PIS)/controlled seed promptness index (PIC); Promptness index PI = nd\(_x\) (1.00) + nd\(_x\) (0.75) + nd\(_x\) (0.50) + nd\(_x\) (0.25); nd\(_x\) = number of germinated seeds by the xth day of measurement (Grzesiak \ et \ al. \ 2013).

From each group treated with mannitol, five seeds were separated and given distilled water over the next seven days to determine the possibility of recovery from stressful conditions, while the remaining seeds continued to be treated with mannitol for the seven days. After the 14 day period, plant growth, fresh and dry biomass were measured. All results are presented as the average value of the three ± standard deviation (SD). The statistical significance of the analyzed parameters was tested by ANOVA test (p<0.05) two-way analysis of variance.

RESULTS AND DISCUSSION

The percentage of germination in variety and hybrid tested decreased with increasing mannitol concentration compared to the control (Fig 1). Seed germination of the hybrid Pioneer B23 treated with the 5% mannitol solution was the same as in the control, while treatment with 10% mannitol solution resulted in seed germination 6.67% lower than the control and the highest (20%) concentration of mannitol completely inhibited seed germination.

The Sugar corn variety had a lower germination percentage compared to the hybrid Pioneer B23 in all trials (Fig 1). Comparing the results, it was determined that the germination at 5% mannitol was reduced by 8.89%, at 10% mannitol by 17.78% and at 20% mannitol by 37.78% compared to the control. We observed a decrease in the germination percentage due to increased drought stress for all varieties, though at the influence of 5% and 10% mannitol the germination rate was higher in the hybrid Pioneer B23, while at 20% mannitol the germination rate was higher in the sugar corn variety. We also observed that the germination percentage rose from day 3 to day 5 in all trials, but was regardless significantly lower than the control. Based on the results of the ANOVA test, it was concluded that there are statistically significant differences between the same treatment depending on the day for investigated hybrid and variety. Also, a statistically significant difference was obtained between different groups depending on the concentration of mannitol, for the variety Sugar corn and hybrid Pioneer B23.

The germination potential of the examined maize variety and hybrid dropped significantly under the influence of different mannitol concentrations compared to control (Fig 2). The germination potential of the hybrid Pioneer B23 dropped by 6.67% at 5% mannitol, by 46.67% at 10% mannitol, while at 20% mannitol zero seeds germinated. The germination potential of the Sugar corn variety, on the other hand, dropped by 26.67% at 5% mannitol, by 33.33% at 10% mannitol and by 40% at 20% mannitol, compared to the control. The results of the ANOVA test for the germination potential parameter show that there are statistically significant differences between variety and hybrid. Statistically significant differences also exist between different concentrations of mannitol in the same variety and hybrid maize.

The germination index (PIS/PIC) of drought resistance of the variety and hybrid dropped as mannitol concentration increased. PIS/PIC of the Sugar corn variety at 10% and 20% mannitol was higher than that of the Pioneer B23 hybrid,
while at 5% mannitol there was no visible difference (Fig 3). As for the germination potential parameter, the test shows that for the germination index there are statistically significant differences between different variety and hybrid at a mannitol concentration of 10% and 20%.

To determine the ability of plants to recover from stressful conditions, five seeds were taken out from each trial group treated with 5%, 10% and 20% mannitol to be watered with distilled water for the next seven days. Comparing the results of the shoot and root length of the treated plants and the so-called “refresh” plants, we can see that, for the 5% mannitol group, the shoot of the “refresh” plants was 18.05% longer and the root 15.72% longer than the treated plants. With 10% mannitol the aboveground part of the “refresh” plants was 62.53% longer and the root 73.2% longer than in the treated plants. With 20% mannitol there was no plant growth in the treated plant group, while the “refreshed” plants recovered and formed both the root and the aboveground part. There is a statistically significant difference between the length of the roots and the stem of the treated and the so-called refresh plants (Fig 4).

The fresh mass of the aboveground part and root of the Pioneer B23 hybrid dropped with increasing mannitol concentrations. With 5% mannitol, the mass of fresh seedlings dropped by 70.84% and the mass of dry seedlings by 71.74%, while the mass of fresh roots dropped by 17.62% and the mass of dry roots by 25% compared to the control. With 10% mannitol, the mass of fresh seedlings dropped by 89.51% and the mass of dry seedlings by 84.78%, while the mass of fresh roots dropped by 86.04% and the mass of dry roots by 80% compared to the control. The results of the statistical analysis also show a significant difference between fresh and dry mass at different concentrations of mannitol (Fig 5).

The water deficit and high temperatures limit growth and reduce the yield of maize crops (Chaves et al. 2002; Iwuala et al. 2020). The effect of drought on the whole of the maize plant is substantial, as shown in its reduced ability to germinate and sprout seeds, poorer development of the roots and aboveground part of the plant, reduced ability to create and accumulate dry matter and negative impact on flower formation, pollen formation, pollination, grain formation and quality (Chaves et al. 2003; Khodarahmpour, 2011; Jain et al. 2013).

The germination percentage in variety and hybrid was significantly reduced compared to the control, with the Pioneer B23 hybrid having higher germination rates and thus showing a higher degree of drought resistance. Chaves et al. 2002; Khodarahmpour, 2011; Jain et al. 2013; Liu et al. 2015 have shown that different maize varieties are highly sensitive to drought, suggesting that different genotypes of maize might respond differently to drought conditions. Liu et al. (2015) examined the effect of different mannitol concentrations on two maize varieties (Zhengdan 958 and Liansheng 15), finding significant differences in the germination percentage, with the Zhengdan 958 variety proving the more resilient. Khodarahmpour (2011) reported a 71.2% decrease in the germination rate and percentage of different maize hybrids under the influence of polyethylene-glycol-induced drought. Barbosa et al. (2004) have done similar studies on certain soybean varieties, finding that, depending on the mannitol concentrations used, the germination percentage decreased significantly, which ties to our research results.
We prove in this paper that the germination potential of both tested maize varieties decreased under the influence of drought, but that it decreased more in the Sugar corn variety than in the Pioneer B23 hybrid (Fig 2). This suggests that the germination rate was higher in the case of the Pioneer B23 hybrid than in the Sugar corn variety, meaning it is more drought resistant. The effect of drought was greater on the germination potential than on the germination rate, showing a greater impact of drought in the early germination stages. This may be due to the increased absorption ability of the variety under the influence of mannitol in the early germination phase, which ties in with the results of other researchers (Koka et al. 2015; Liu et al. 2015).

In both maize, the drought resistance index dropped with increasing mannitol concentration, but in the Pioneer B23 hybrid it decreased less than in the sugar corn variety, which means that the Pioneer B23 showed higher drought resistance.

Koka et al. (2015) examined the effect of different mannitol concentrations on plant growth, discovering in the process the resultant root growth inhibition, which ties in with the results of our research. The negative impact of drought on seedling growth of four soybean species was examined by Machado Neto et al. (2004), showing that the dry mass of the aboveground part and root did not differ between the control plants and those exposed to a mannitol-induced water deficit. The impact of drought induced by polyethylene glycol 6000, mannitol, as well as by sodium and potassium salts on oats (Koka et al. 2015) showed a drastic decrease in dry and fresh plant root mass.

The mannitol concentrations applied had an inhibitory effect on both the fresh and dry aboveground part and root biomass. Under the influence of different concentrations of mannitol, the seeds of Pioneer 23 germinated faster and grew better than those of the sugar corn variety, as can be seen from the higher biomass and length of its seedlings (Fig 5). As well, the plants that were left to recover from drought-induced stress had higher growth compared to treated plants. Lekshmi et al. (2018) reached similar conclusions in their work, finding that increased mannitol concentrations lowered both fresh and dry mass.

CONCLUSION

The research presented in this paper show that drought-induced stress substantially disrupts the germination process as well as the growth and development of the maize tested. The results show significant differences between the variety and hybrid examined. The Pioneer B23 seed germinated in larger number and more quickly and had a better-developed aboveground part, entirely absent in the Sugar corn variety. As well, the Pioneer B23 hybrid had a stronger root and so a greater capability of osmotic adaptation to drought conditions, which is one parameter that demonstrates greater Pioneer B23 seed resilience to drought-induced stress. Research such as this shows that three germination indexes can be used to determine which maize varieties have superior drought tolerance at germination and at different stages of seedling growth. Taking these characteristics into account, this study determined that the Pioneer B23 hybrid was more drought-tolerant and possessing of greater genetic potential to maintain better growth in such conditions.

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