

PRIMARY DIAGNOSTICS OF DROUGHT RESISTANCE OF WHEAT GENOTYPES UNDER OSMOTIC STRESS

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Summary: The primary drought resistance of wheat plants of Moldavian and Ukrainian selection to induced drought caused by different sucrose solutions (3, 5, 10, and 16%) was studied. Testing of six wheat genotypes from Moldova and Ukraine revealed a distinct distribution of seed germination rates at sucrose concentrations above 16%. Genotypes showed a sharp response in root development to 3% and 5% sucrose compared to aboveground parts. These reactions of the root system can allow the classification of genotypes according to drought tolerance. This method provides an effective tool for the early selection of drought-resistant genotypes, supporting sustainable wheat cultivation under water stress conditions.

Keywords: *Triticum aestivum* L., drought resistance, osmotic stress.

Introduction

In light of global climate change, characterized by an increase in the average annual temperature, the typical weather patterns are transforming, resulting in more frequent and prolonged dry spells [4]. Approximately 40% of the global land surface is significantly affected by this phenomenon [19]. Consequently, the absence of moisture during the plant's developmental phase compels it to adapt to its surroundings to survive. Depending on the severity of the stress, growth processes slow down, respiration and photosynthesis rates fluctuate, and enzymatic activity diminishes [16]. Dehydration of cells can lead to a reduction in the production of certain substances, increase plasma membrane permeability, alter hormonal levels, and impact nitrogen and phosphorus metabolism. The selection of genotypes with a high level of viability under conditions of soil moisture deficit and the capacity for water absorption under high osmotic stress is critical for stable grain production and breeding in general. Scientific literature indicates [12] that extreme factors, such as hyperthermia, significantly inhibit the juvenile period and growth processes of winter wheat plants. Water scarcity arises when the rate of transpiration exceeds the rate of water uptake [17], which is often observed during periods of drought, salinity [4], and low temperatures [6], causing dehydration in plants even when there is sufficient moisture in the soil. Under the influence of drought, high osmotic pressure in the soil's aqueous solution occurs, making it difficult for the plant's root system to absorb water from the soil as a result, there is a water deficit, and the concentration of ions in the plant's cytoplasm increases [5]. This phenomenon can result in disruptions to the structure and functionality of biomolecules, as well as the integrity of the lipid bilayer in cell membranes. Moreover, plasmolysis may occur in cells due to a shortage of water. At the plant level, there is a disruption in water exchange between the soil, roots, and shoots, leading to inhibited growth indicators for the plant organism [15]. The survival of plants in such conditions depends on protective mechanisms evolved through time and encoded in DNA [1].

Wheat plants are exposed to adverse temperatures throughout their life cycle, from seedling to harvest maturity. Exposure to adverse environmental factors, such as high temperatures and long periods without precipitation, often accompanies the germination of winter wheat seeds [9]. For successful germination, the initial moisture content of the seeds should be at least 10% by weight,

and the water supply should be 70-100% of the seed weight [14, 10]. In this context, it is crucial to investigate the initial drought resistance of winter wheat genotypes at the stages of seed germination and seedling development. The early germination of seeds with varying osmotic potentials allows for the selection and isolation of genotypes that exhibit varying levels of resistance, which can be utilized for both breeding purposes and agricultural production. This approach helps to mitigate the risk of low crop yields. When conducting laboratory tests to evaluate the resistance of winter wheat plants to environmental stressors, it is essential to follow strict protocols to ensure accurate and reliable results. Firstly, the seeds used should be healthy, from the same year of harvest, from the same geographic region, with high germination rates, and with high germination energy. The degree of negative impact on plants is largely contingent upon the environmental conditions, such as temperature, which accompany it. Therefore, it is essential to maintain specific environmental conditions during the testing process to achieve accurate results [11, 2]. The objective of our research was to devise a novel method for laboratory diagnosis of drought tolerance in winter wheat samples and to identify genotypes with high and low indices of primary resistance to osmotic stress.

Materials and methods

The study was conducted on six genotypes of winter wheat (*Triticum aestivum* L.), selected from experimental fields in Moldova and Ukraine, under the auspices of the Institute of Genetics, Physiology, and Plant Protection at the State University of Moldova. To assess the drought tolerance of these wheat plants under controlled laboratory conditions, the seeds were subjected to a process of sieving using a Fadeev sieve with a mesh size №2, which allowed for the selection of pure and uniform seed material. The selected seed samples were then cultivated on a nutrient medium comprising water (control group) and sucrose solutions at concentrations of 3%, 5%, 10%, and 16%, corresponding to osmotic potential values of 2.4, 4.0, 8.2, and 13.5 atmospheres, respectively (experimental groups).

The seeds of the studied genotypes were germinated in a thermostat at a temperature of 20°C in Petri dishes (5 cups in total containing 20 seeds for each variant) at the bottom of which was filter paper moistened with water (control) and various concentrations of sucrose solution (experiment). The percentage of seed germination was determined over time on days 3, 5 and 10 of germination.

The growth parameters of wheat seedlings were determined on the 3rd 5th and 10th days of germination in a sucrose solution with different osmotic potential and a day/night lighting regime for 16/8 hours. Each experiment was repeated five times on at least 100 plants. The results were statistically processed in Microsoft EXCEL 2016 and Statistica 10 [13].

Results and discussions

It has been well documented [8, 17, 7] in the literature that the assessment of preliminary drought tolerance in wheat plants relies on their capacity to germinate in environments with high osmotic potential. However, it is important to note that seed germination in such conditions may not always accurately reflect the true picture, it is heavily influenced by the climatic circumstances under which the plant grows, as well as the quality of the seed. In the context of rapidly changing climate trends characterized by increasing temperatures and shorter periods of precipitation accumulation in soil, the urgent need for timely identification of different wheat genotypes based on their drought tolerance becomes paramount. The analysis of seed germination under varying osmotic potentials revealed that the wheat varieties under investigation exhibited three distinct groups of drought resistance when exposed to high sucrose concentrations. The first group consisted of drought-tolerant genotypes with low germination rates, such as Moldova16 and Bijuteria. The group of cultivars showing moderate resistance to drought included Meleag and Pisanca, while the group exhibiting high drought tolerance included Lăutar and Moldova 614 (Fig. 1).

Analysis of the data shown in Figure 1 shows that at lower concentrations of sucrose solutions (3 and 5%) seed germination was almost the same as the control variant (Water). Consequently, these concentrations have virtually no negative effects on seeds. It should be noted that the osmotic potential of dry seeds is one level higher than the osmotic potentials of these solutions, which may lead to an incorrect assessment of the drought resistance of varieties since the seedling only initially uses the nutritional components of the seed and then switches to self-maintenance. Another important component in assessing the drought resistance using this method is the climatic conditions that determine the fullness of the seed. Poor filling of seeds with reserve substances reduces the osmotic potential of seeds, thereby reducing their germination rate. When comparing different genotypes from different cultivation zones, can lead to a misleading conclusion.

In this connection, there is a question about the ability of wheat germ to develop evenly under the influence of osmotic stress for a longer time. For a more accurate assessment of the drought resistance of various wheat genotypes after reaching the third day of germination the seedlings were transferred to cultivation chambers consisting of a solution with an osmotic potential of 2.4; 4.0 atm with constant aeration and 400-600 FAR lighting with a day/night mode of 16/8 hours. Under these conditions, it was possible to obtain normally developed plants that recorded the growth of both the aboveground part of the seedling (the primary leaf) and the underground part of the seedling (the main root).

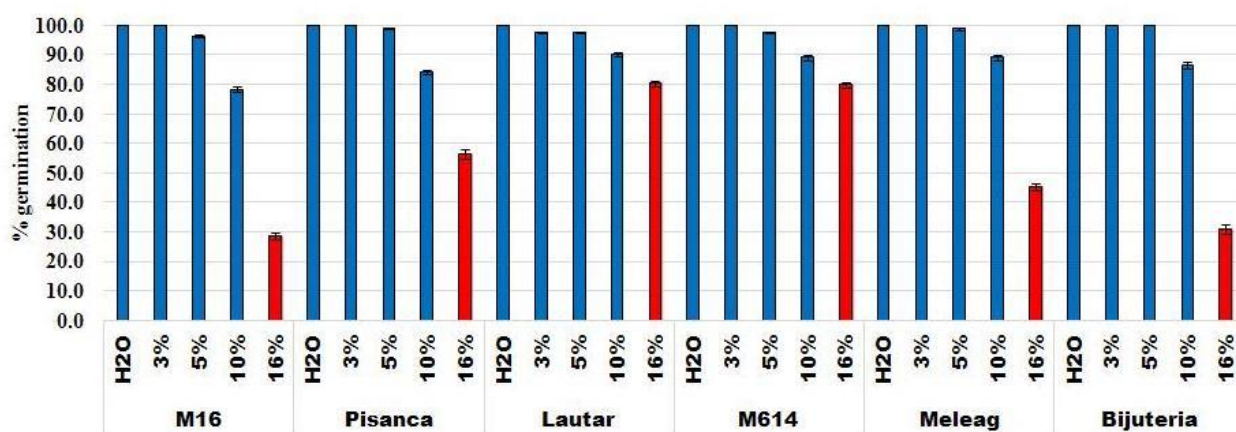


Figure 1. The percentage of germination of seeds of different wheat genotypes in sucrose solutions with different osmotic potential on the 10th day of vegetation

An analysis of the assessment of the length of the central root showed that under the influence of osmotic stress the central root is more inhibited than the primary leaf of seedlings. The data presented in Figure 2 demonstrate that different wheat genotypes respond differently to induced drought caused by sucrose solution and are more dependent on the specifics of the variety.

As the level of induced drought increases the distribution between wheat genotypes becomes more pronounced which can later be used to describe different varieties and identify their cultivation zones. As shown in Figure 2 the studied wheat genotypes can be roughly divided into three groups of resistance to induced drought. The first group includes varieties with a pronounced response to induced drought caused by osmotic stress (Moldova16 and Pisanca varieties); the second group includes genotypes with tolerant properties to the effects of induced drought (Meleag and Bijuteria) and the third group includes wheat genotypes with a selective attitude to the effects of induced drought which can change their growth rates as stress intensifies (Lăutar and Moldova 614 varieties). Due to the clear separation of different genotypes to the effect of induced drought caused by the osmotic properties of sucrose solutions, it is possible not only to initially distribute wheat

genotypes according to resistance but also to use these parameters (central root length) as a breeding feature when developing new genotypes.

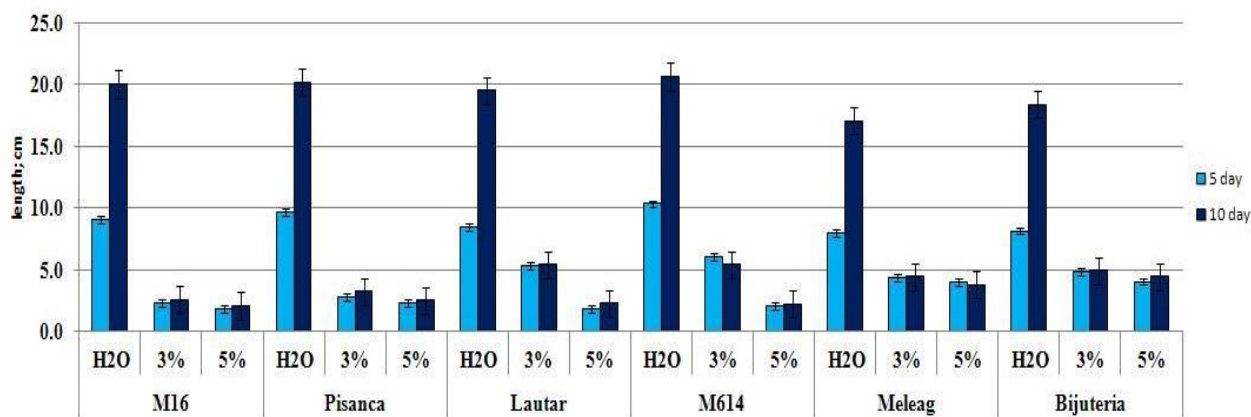


Figure 2. Average length of the central root of plants of different wheat genotypes grown on water (control) and sucrose solutions with different osmotic potential (experiment) on 5 and 10 days of germination

It was found that under the influence of osmotic stress from 2.4 to 4.0 atm. the increase in the length of the central root decreases on average from control (growth on water) by 75 – 85% while significant differences in the length of roots grown on water are not observed in different varieties. It is especially worth noting that the increase in the root system in the dynamics of vegetation in variants grown on sucrose solutions with different osmotic pressure does not change and is almost the same while variants grown on water have an average increase between 5 and 10 days of vegetation of about 50 – 60% in all studied genotypes.

Stopping the development of the root system under the influence of induced drought caused by sucrose solutions with different osmotic pressures indicates that wheat seedlings may experience significant difficulties in absorbing moisture into the plant body which in turn can lead to inhibition of the growth of the aerial part of the plant (primary leaf) as well as to a decrease in photosynthetic activity due to impaired activity of photosystem 2 and the fixation of CO₂ by a protein complex RuBisCo. The data presented in Figure 3 demonstrate that the growth parameters of the aerial part of the plant (primary leaf) are more of a varietal trait and do not have such a clear separation of genotypes as the assessment of central root growth.

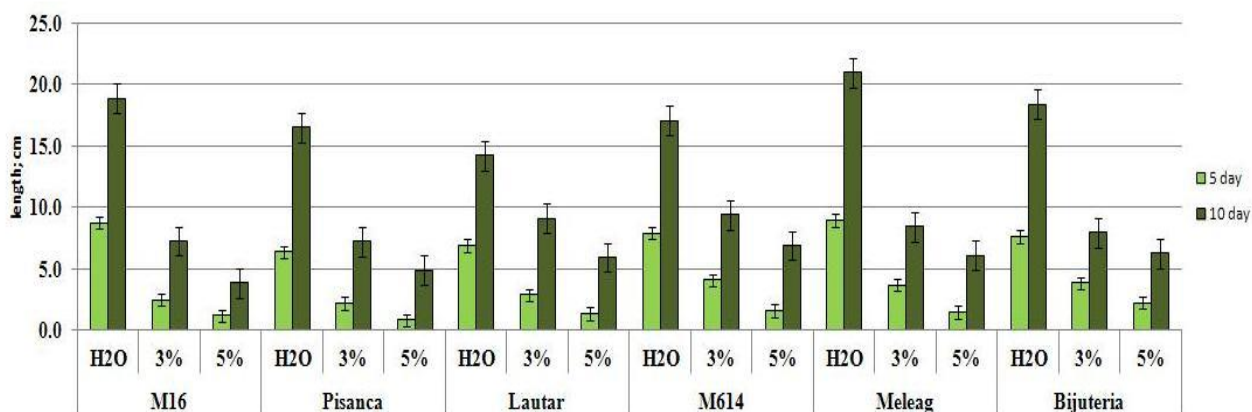


Figure 3. Average length of the above-ground part of plants of different wheat genotypes grown on water (control) and sucrose solutions with different osmotic potential

It is also worth noting that the increase in the above-ground part of the seedling in the dynamics of time between days 5 and 10 of vegetation in the control variants (grown on water) averaged from 60 to 75% which characterizes the ratio between the growth of the above-ground and underground part of the plant as 1:1 under the action of drought-induced osmotic sucrose solution 2.4 atm the ratio of the above-ground part to the underground part is 4:0.3 and under the action of osmotic stress 4.0 atm the ratio is 3:0.3 respectively. Thus it can be concluded that with an increase in osmotic stress from 2.4 atm to 4.0 atm a decrease in the increase in the aboveground part of wheat plants between the experimental variants has a ratio of 2:1 while there are no significant changes in the increase in root length (Fig. 2). Consequently, growing wheat seedlings in solutions with an osmotic pressure of 2.4 to 4.0 atm significantly inhibits the growth of the root system and to a lesser extent the growth of the aboveground part of plants which can be fatal under natural conditions due to the large area of transpiration (loss of moisture by the plant) and the extremely low collection area of available moisture (the development of the root system).

Conclusions

The assessment of wheat seed germination percentages under varying osmotic potentials offers limited insights into drought tolerance, as it is heavily influenced by extrinsic factors such as environmental conditions and intrinsic factors like seed quality. The method we propose, which centers on analyzing the primary root growth under controlled osmotic stress conditions, provides a more precise categorization of genotypes in terms of drought tolerance. This method holds significant potential for application in breeding programs aimed at enhancing drought resistance.

Our findings indicate that osmotic stress exerts a more profound influence on the development of the root system compared to the aerial parts of the plant. This differential impact underscores the importance of root system vigor for plant survival under natural drought conditions. Therefore, evaluating the dynamics of primary root growth across varying osmotic stress levels emerges as a critical criterion for the early selection of drought-resistant genotypes.

Incorporating selection strategies based on these observations is anticipated to contribute significantly to the development of wheat varieties with enhanced productivity and stability in environments subject to water stress.

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