



## Drought tolerance of developed wheat genotypes based on early diagnostics

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Climate change is being observed worldwide, affecting all continents. Currently, it has the most devastating effects in the regions of the Southern Hemisphere, particularly in African countries, although its impact is felt across the entire planet. Global climate change necessitates the development of highly productive crop varieties and hybrids whose physiological mechanisms promote efficient metabolism and redistribution of assimilates from vegetative tissues to generative organs and seeds. A key challenge in this context remains the creation of source material resistant to abiotic stress factors, particularly drought. Mannitol can serve as a model agent for screening drought-tolerant genotypes. Its concentrations exert selective pressure on biological objects, enabling the identification of materials that exhibit neutral or reduced responses to a given stressor. As a result of intra-specific hybridization between geographically distant forms, spring durum and bread wheat accessions were developed and used as the subject of research aimed at selecting drought-tolerant, high-yielding genotypes. The experiments were conducted under moderately continental climatic conditions during 2021–2024 at Uman National University of Horticulture (Central Ukraine). Throughout the study years, insufficient precipitation and elevated air temperatures were recorded, resulting in specific phenotypic responses of the breeding material. The use of mannitol solutions during germination caused a decrease in seed germination rates, as well as reductions in root and shoot length and seedling biomass in wheat samples. At the same time, genotypes displayed differential responses to mannitol concentrations, allowing the identification of breeding materials that were less affected by the stress factor. These materials can be effectively used as sources of drought-tolerance genes in breeding programs aimed at developing high-yielding spring bread and durum wheat varieties. Early diagnostics of drought tolerance using mannitol-based germination tests represents a promising tool for accelerating the selection of high-yielding and drought-tolerant crop genotypes.

**Keywords:** temperature regime; precipitation amount; spring durum wheat; spring bread wheat; accession; concentration of the stress factor mannitol.

### Introduction

In recent decades, pronounced processes of climate change and a steady rise in global temperature regimes have been documented. These phenomena are increasingly associated with natural hazards such as prolonged heat waves, destructive downpours, droughts, floods, and hurricanes. In recent years, record-breaking extreme temperature maxima, coupled with insufficient precipitation – often concentrated in heavy rainfall events that limit efficient plant uptake – have resulted in substantial yield losses and productivity declines (Novak, 2025).

Ensuring sustainable agricultural production under the conditions of climate change requires the integrated application of agrotechnical, breeding, organizational, and land-reclamation strategies. Within this framework, the development of crop varieties tolerant to drought, heat, and salinity has become a priority (Vozhehova, 2019). Promising approaches include the use of early diagnostic methods that enable the identification and ranking of plant materials according to their tolerance to osmotically active substances (Almansouri et al., 2001; Pykalo et al., 2020).

Wheat remains the principal cereal crop in Ukraine. The breeding of drought-tolerant, high-yielding biotypes of both durum and bread wheat is expected to play a key role in securing stable grain production under the accelerating pace of global warming.

According to a number of authors, the primary driver of rising global temperatures is the anthropogenic factor (Lyzun et al., 2017). Over the past 30 years, the mean annual temperature in Ukraine has increased by 1 °C, particularly during the summer months. As a result, droughts have intensified, the hydrological regimes of rivers and lakes have shifted, and extreme weather phenomena atypical for Ukraine have emerged. Projections indicate that within the next two decades the global temperature will reach or exceed 1.5 °C. Such climatic anomalies affect the structure of the atmosphere, oceans, ice sheets,

and the Earth's surface. Nearly half of the world's countries experienced abnormal heat waves in 2024, lasting two to four months in some regions. Even in countries with temperate climates, such as the United Kingdom and the United States, three additional weeks of heat posed significant challenges for infrastructure and healthcare systems (Novak, 2025). Long-term data suggest that a 1 °C increase in mean air temperature reduces maize yields by 13.5%, soybean by 8.8%, rice by 2.6%, and wheat by 5.6%. However, the use of drought-tolerant biotypes mitigated these negative effects on yield by a factor of two to three (Jägermeyr et al., 2021).

Kumar & Rani (2025), analyzing 77 studies published between 2015 and 2024, concluded that climate change exerts a profound impact on the productivity of agricultural crops, with global food production expected to decline by 14% by 2050 unless adaptive measures are undertaken. They further estimated that climate-smart agriculture could increase agricultural productivity by 10.5% and profitability by 29.4% under global warming conditions. Nevertheless, implementation faces major obstacles, including financial constraints and underdeveloped infrastructure. Effective mitigation of climate change impacts requires a combination of policy instruments (carbon pricing, climate-resilient subsidies, and trade liberalization), technological innovations (precision farming, satellite-based climate monitoring, early warning systems for natural disasters, efficient irrigation, conservation tillage, and the deployment of drought-tolerant crop varieties), and improved farmer awareness.

Climate change has had a particularly severe impact on African countries. Uganda, Ethiopia, and Malawi face recurrent floods and droughts, underscoring the urgent need for early warning systems that integrate satellite weather data with community-level monitoring. In Mali and Senegal, efforts have been directed at combating desertification through reforestation and sustainable land use. Agriculture in Nigeria, Kenya, and Mali suffers from recurrent droughts, highlighting

the importance of drought-tolerant varieties and improved irrigation (Mohamed et al., 2025). In Angola, climate change is not a future threat but a present reality. Rainfall patterns are uncertain, with longer dry spells and severe flooding. Approximately 37% of the population (11.1 million people) lives in rural areas and depends on agriculture. In Southern Angola, the last decade has witnessed the most extreme droughts in 40 years. In 2021, more than 3.8 million people across six southern provinces faced food shortages, and over 1.2 million suffered from water scarcity due to drought. If global warming continues at the current rate, agricultural productivity in Angola alone may decline by 7% by 2050 (Andrade et al., 2024).

Globally, anthropogenic climate change has reduced agricultural production by 21% since 1961, rendering food systems increasingly vulnerable to ongoing climate variability (Ortiz-Bobea et al., 2021). Rising CO<sub>2</sub> emissions are the main driver of global warming, and climate projections typically rely on high- and low-emission scenarios. It is estimated that, under high- and low-emission scenarios respectively, an additional 20–36% and 11–33% of the global population may face hunger by 2050. In parts of South Asia, food requirements to offset these impacts are projected to triple current reserves, underscoring the urgency of crop adaptation and strategic food stockpiling (Hasegawa et al., 2021).

Recent evidence suggests that global warming reduces drought forecasting accuracy by up to 70%, particularly in North America, the Amazon Basin, Europe, South and East Asia, and Australia. Wu et al. (2025) stress the importance of crop adaptation to climate change, particularly through the development and dissemination of drought-tolerant cultivars. Indian researchers emphasize plant breeding as a key strategy to combat drought. The application of advanced technologies, including genetic engineering, has shown considerable potential to enhance drought tolerance in wheat. For example, transgenic approaches using barley genes have improved water-use efficiency, biomass accumulation, and root development under water stress (Sivamani et al., 2000). Similarly, the introduction of proline-inducing genes has been linked to improved drought tolerance in transgenic wheat lines, likely due to the antioxidant role of proline in mitigating oxidative damage (Vendruscolo et al., 2007). Establishing links between drought and other stressors, and mitigating their combined effects, is critical for sustaining crop productivity under limited water availability (Singh et al., 2025).

Wheat is among the most valuable cereal crops, serving as a primary source of protein and calories in the human diet. Drought is a major abiotic stress factor severely affecting wheat production worldwide (Diordiieva, et al., 2023). Shifts in precipitation patterns, elevated atmospheric CO<sub>2</sub> concentrations, rising air temperatures, and hot winds are key drivers of drought-induced stress. These factors alter morphological, physiological, and biochemical processes in plants, leading to yield reductions and associated traits. Negative effects include reduced germination and seedling vigor, premature leaf senescence, decreased chlorophyll content and photosynthetic activity, impaired starch accumulation, and accelerated grain maturation. Drought stress also induces reactive oxygen species (ROS) formation, causing oxidative damage and cell death.

Wheat plants have evolved a range of adaptive mechanisms to withstand drought, including increased trichome density and leaf wax deposition, altered root-to-shoot ratios, stay-green traits, proline accumulation, and the activation of antioxidant enzymes such as superoxide dismutase (SOD), ascorbate peroxidase (APX), peroxidase (POD), and catalase (CAT). Additional adaptations include osmotic adjustment, abscisic acid (ABA) accumulation, and dehydrin formation. Screening diverse genotypes for novel trait combinations, combined with genetic engineering, transgenesis, and targeted breeding for drought tolerance, are key strategies to enhance wheat productivity and ensure global food security under climate change (Nyaupane et al., 2024).

Swedish researchers emphasize the significance of early-maturing wheat varieties capable of generating substantial leaf and stem biomass prior to the onset of extreme heat in drought-prone regions. Crop stress responses can be monitored using satellite imagery at the landscape level to determine stress duration and intensity. Soil water availability has been identified as a critical factor in yield formation during drought years. Incorporating soil characteristics into remote sensing analyses significantly improves the accuracy of drought stress forecasting for specific crops (Velde et al., 2025).

The identification of drought-tolerant wheat lines through early diagnostic techniques offers a promising avenue for accelerating the selection of valuable breeding material and the development of drought-resilient varieties. This strategy will contribute to sustaining and enhancing wheat productivity under conditions of global warming and climate change.

## Materials and methods

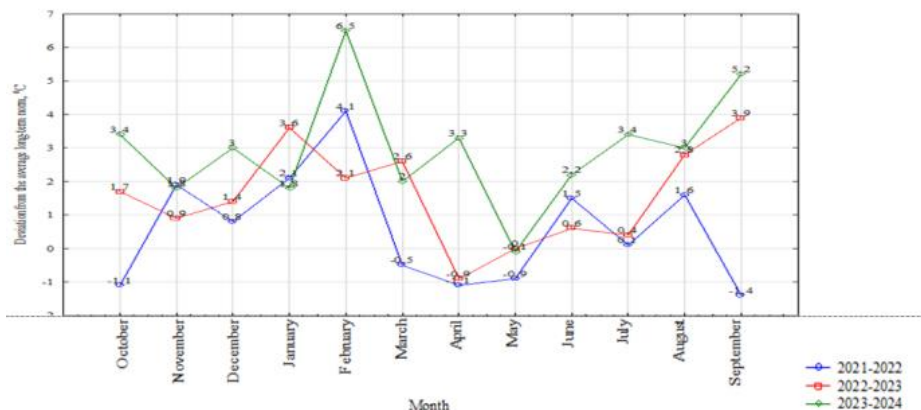
Environmental indicators for the period from October 2021 to September 2024 were obtained from observations conducted at the Uman meteorological station. Baseline (typical) values were established as 30-year means for the region (1991–2020).

Climatic Conditions: Temperature Regime and Precipitation (2021–2024). During the summer periods of 2022–2024, record-high temperatures were observed against the background of uneven precipitation distribution.

According to meteorological data, in the 2021–2022 agricultural year, deviations of air temperature from the norm were within  $\pm 0.5$  °C only in March and July (Fig. 1, Table 1). In October 2021, as well as in April, May, and September 2022, the mean monthly values were 0.9–1.4 °C lower than the long-term average.

For six months, the air temperature exceeded the climatic norm: in November (+1.9 °C), December (+0.8 °C), January (+2.1 °C), June (+1.5 °C), and August (+1.6 °C). The most significant deviation was recorded in February, when the temperature was 4.1 °C higher than the long-term average. The mean annual deviation amounted to +0.6 °C.

In 2022–2023, the excess of average air temperatures relative to the norm was even more pronounced. Only in April was a decrease observed (–0.9 °C), whereas May and July were characterized by values that were nearly identical to the long-term averages.



**Fig. 1.** Dynamics of air temperature deviations (°C) from long-term averages according to data from Uman meteorological station, 2021–2024

All other months were warmer. In November 2022 and June 2023, air temperatures exceeded the norm by 0.6–0.9 °C, while in October and December 2022, the deviations reached 1.4–1.7 °C. The largest anomalies were recorded in January (+3.6 °C), February (+2.1 °C), March (+2.6 °C), August (+2.8 °C), and September (+3.9 °C). On average, the 2022–2023 agricultural year was 1.6 °C warmer than the long-term mean.

In 2023–2024, no month was cooler than the long-term average, and only in May were temperatures nearly identical to the norm. All other months were characterized by elevated air temperatures. Deviations of +2.0 °C were recorded in November 2023, January, and March 2024; from +2.2 to +3.0 °C in December 2023, June, and August 2024; and +3.3–3.4 °C in October 2023, April, and July 2024. Extreme anomalies were observed in February and September, when the deviations reached +6.5 and +5.2 °C, respectively. On average, annual temperatures in 2023–2024 exceeded the norm by 3.0 °C.

Thus, throughout the 2021–2022, 2022–2023, and 2023–2024 agricultural years, elevated air temperatures were consistently observed, averaging 9.4, 10.4, and 11.8 °C, respectively, compared with the long-term average of 8.8 °C. Overall, all seasons were warmer than the climatic norm. Record summer temperatures were recorded in July and June 2024, reaching 24.3 and 23.1 °C, respectively.

Frost-free periods were recorded in winter, specifically in February 2022, January 2023, and December and February 2024. In contrast, May was within or even below the long-term temperature norm across all three years.

Regarding precipitation, the dynamics were inverse (Fig. 2, Table 1). In 2021–2022, precipitation deficits were observed during nine months of the agricultural year, with only December, April, and September exceeding the long-term average by 51.2, 16.7, and 59.0 mm, respectively. Severe deficits were noted in November 2021, as well as in March, May, June, and July 2022, when precipitation amounted to only 51–63% of the long-term norm, corresponding to decreases of 21.8, 22.6, 29.6, 44.7, and 39.9 mm. Even drier conditions were recorded in October 2021 and February 2022, with precipitation falling short by 84% (–36.0 mm) and 79% (–26.8 mm), respectively. In con-

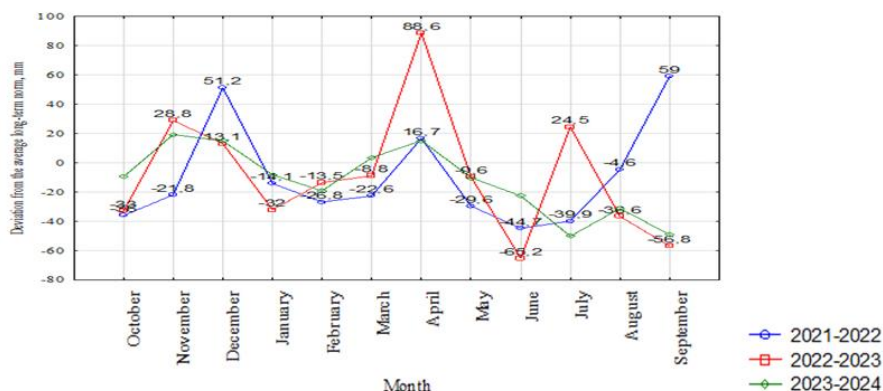
trast, December 2021 and September 2022 were marked by excessive rainfall, exceeding the norm by 128% (+51.2 mm) and 97% (+59.0 mm). Overall, total precipitation in the 2021–2022 agricultural year amounted to 473 mm, which was 113 mm (–19%) below the long-term mean.

**Table 1**

Air temperature and amount of precipitation according to the Uman weather station

Month	Air temperature, °C				Amount of precipitation, mm			
	long-term average	2021–2022	2022–2023	2023–2024	long-term average	2021–2022	2022–2023	2023–2024
October	8.3	7.2	10.0	11.7	43	7.0	10.0	33.5
November	2.8	4.7	3.7	4.6	43	21.2	71.8	62.3
December	–1.8	–1.0	–0.4	1.2	40	91.2	53.1	55.0
January	–3.4	–1.3	0.2	–1.6	38	23.9	6.0	29.8
February	–2.3	1.8	–0.2	4.2	34	7.2	20.5	14.9
March	2.5	2.0	5.1	4.5	36	13.4	27.2	39.5
April	9.7	8.6	8.8	13.0	41	57.7	129.6	56.2
May	15.4	14.5	15.4	15.3	52	22.4	42.4	41.8
June	19.0	20.5	19.6	21.2	81	36.3	15.8	58.5
July	20.9	21.0	21.3	24.3	68	28.1	92.5	17.9
August	20.1	21.7	22.9	23.1	49	44.4	12.4	17.7
September	14.5	12.6	18.4	19.7	61	120.0	4.2	12.1
average/total	8.8	9.4	10.4	11.8	586	473	486	439

In 2022–2023, alternating periods of drought and excess rainfall were recorded. November and December 2022, as well as April and July 2023, were characterized by precipitation exceeding the long-term average by 67%, 33%, 216%, and 36%, respectively. Notably, in April, precipitation reached 129.6 mm, more than triple the norm. The remaining months were predominantly dry. In October and January 2022, as well as in June, August, and September 2023, precipitation was 75–84% lower than the average, corresponding to decreases of 33.0, 32.0, 65.2, and 36.6 mm, respectively. In total, 486 mm of precipitation was recorded during the agricultural year, which was 100 mm (–17%) below the long-term mean.



**Fig. 2.** Dynamics of deviations in precipitation (mm) from long-term averages according to data from Uman meteorological station, 2021–2024

During eight months of the 2023–2024 agricultural year, a precipitation deficit was observed. In October 2023, January, May, and June 2024, precipitation reached only 72–80% of the long-term average, corresponding to deficits of 9.5, 8.2, 10.2, and 22.5 mm, respectively. Precipitation shortages in February and August amounted to 19.1 and 31.3 mm, or 56% and 64%, respectively. The driest months were July and September, with precipitation falling short of the norm by 74% and 80%, or 50.1 and 48.8 mm, respectively. Conversely, precipitation exceeded the norm in November and December 2023, as well as in March and April 2024, by 19.3, 15.0, 3.5, and 15.2 mm, corresponding to 45%, 38%, 10%, and 37%, respectively. Thus, among the reporting years, 2023–2024 was the driest, with total annual precipitation of 439 mm, which was 147 mm (–25%) below the long-term average.

The research material comprised spring durum wheat (*Triticum durum* Desf.) lines 326, 327, 328, 329, 330, 331, 332, 333, 334, and 335, as well as spring bread wheat (*Triticum aestivum* L.) lines 336

and 337, developed at Uman National University. Drought tolerance of the wheat samples was assessed by germinating seeds in mannitol solutions. Mannitol (Manit Pharm, Ukraine) was used because it induces osmotic stress.

The method is based on the ability of seeds of different genotypes to germinate under conditions of high osmotic pressure. Accessions whose seeds exhibited higher suction force than that of the osmotic solution were considered tolerant to the selective factor.

Selected seed batches (100 seeds per treatment) were soaked either in distilled water (control variant) or in mannitol solutions of different concentrations (4%, 6%, 8%, 10%, and 12%). After imbibition, the seeds were placed at equal distances on strips of filter paper (100 × 10 cm). The seeds were covered with another layer of filter paper, rolled into cylinders, and placed in vessels containing either distilled water or the respective mannitol solutions. Germination was carried out in thermostats at 20 °C with four replications. Seed germination percentage was determined on the eighth day of cultivation.

After germination assessment, root and shoot lengths were measured as indicators of seedling growth intensity under laboratory conditions. Root length was determined using a ruler to measure the longest root of each seedling, while shoot length was recorded for each seedling and averaged. Seedling mass was determined by weighing all seedlings to the nearest hundredth of a gram. The obtained results were compared with the control variant based on seed quality indicators.

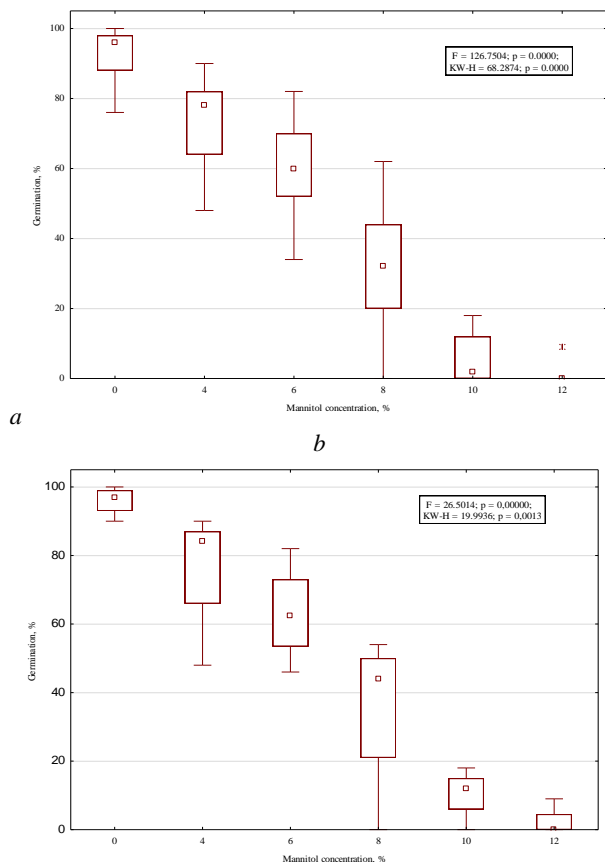
To assess the representativeness of mannitol's effect on each trait, we applied the effective concentration 50 (EC<sub>50</sub>). EC<sub>50</sub> indicates the mannitol concentration that results in a 50% decrease in seed germination or seedling growth compared to the control (Sebaugh, 2011). This value is used to assess the tolerance of wheat accessions to osmotic stress and to identify genotypes with higher drought resistance.

The effective concentration causing a 50% reduction (EC<sub>50</sub>) in seedling growth parameters was determined for multiple traits, including seed germination percentage, root length, shoot length, and seedling fresh weight. Seeds were exposed to increasing concentrations of the osmotic agent (mannitol) under controlled conditions. Maximum and minimum responses were considered as 100% and 0% for each parameter. EC<sub>50</sub> values were calculated using linear interpolation between the two concentrations flanking the 50% response. For each trait, mean values from three replicates per concentration were used to ensure accuracy and reproducibility (Alexander et al., 1999).

Statistical analysis of the research data was performed using Statistica 12 software (StatSoft, Inc., USA). The differences between samples were determined using Tukey's test, where the differences were considered significant at  $P < 0.05$ .

## Results

The detrimental combination of elevated temperatures and precipitation deficits negatively affects crop yields. However, genotypes



**Fig. 3.** Average germination of wheat accessions at different mannitol concentrations (%): *a* – spring durum wheat (*Triticum durum* Desf.); *b* – spring bread wheat (*T. aestivum* L.); 0% – control; abscissa shows the mannitol concentration; the y-axis is the germination of seeds, small square – median, upper and lower rectangle borders – 25% and

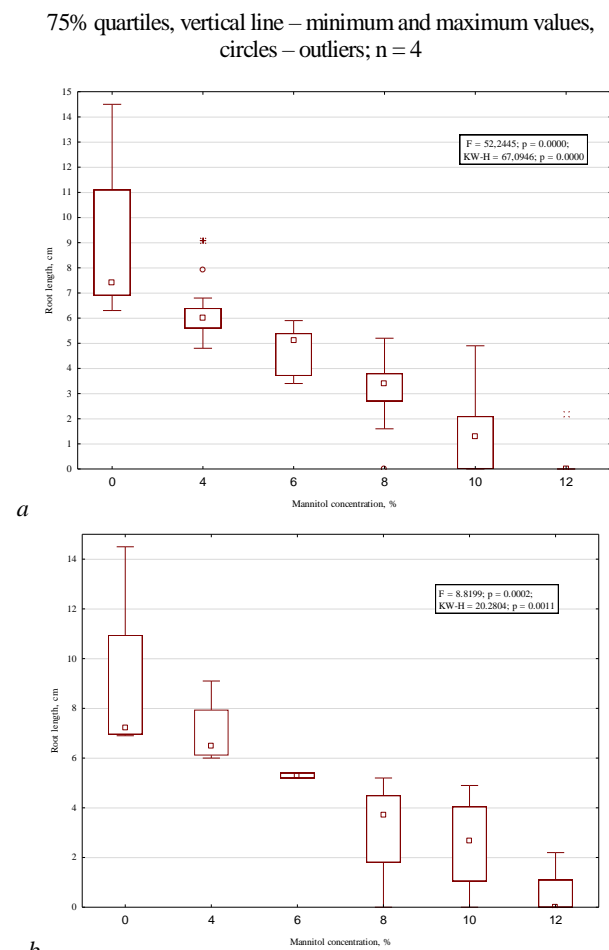
respond differently to these stress factors, highlighting the need to develop and implement drought-tolerant varieties capable of withstanding water scarcity and high temperatures.

Against the backdrop of altered and extreme abiotic conditions, drought tolerance of the Chado spring durum wheat variety, ten spring durum wheat lines, and two spring bread wheat lines was analyzed using early diagnostic methods.

At 0% mannitol concentration, the mean germination rate of spring durum wheat (*Triticum durum* Desf.) lines was 92.7%, while that of spring bread wheat (*T. aestivum* L.) was 93.0% (Figs. 3, 4). Exposure to a 4% mannitol solution decreased germination by 28.4% in durum wheat and by 16% in bread wheat. Further increases in mannitol concentration to 6%, 8%, and 10% resulted in a progressive reduction in germination to 59.1%, 27.0%, and 4.4% in durum wheat, and to 62.5%, 48.0%, and 15.0% in bread wheat, respectively. The 12% mannitol concentration was lethal to all durum wheat lines and reduced germination of bread wheat accessions to 4.5%.

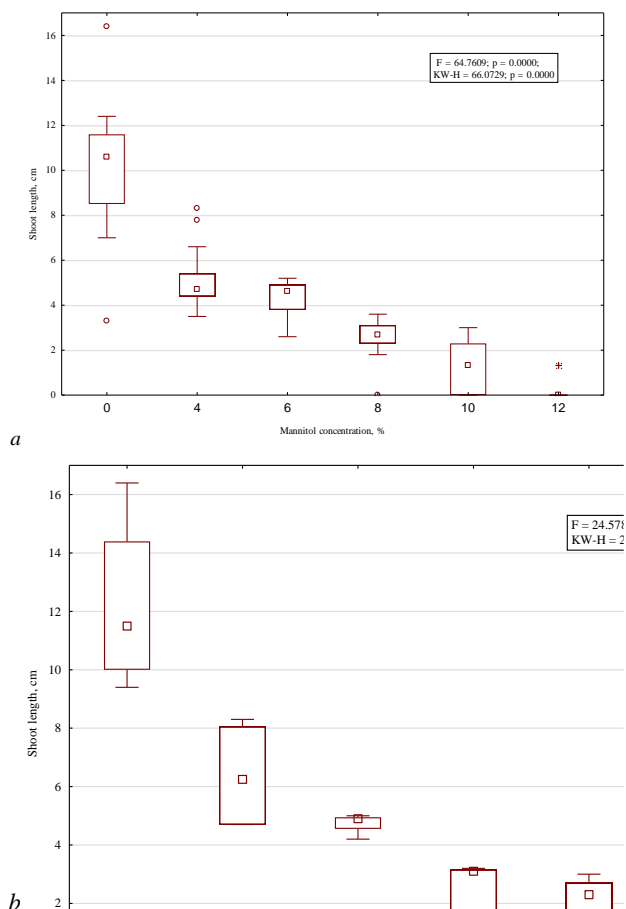
The analysis of root length dynamics under stress conditions revealed a decrease in this parameter in both durum and bread wheat samples (Fig. 4). At mannitol concentrations of 4% and 6%, the average root length of durum wheat seedlings decreased relative to the control by 29% and 46%, respectively, whereas that of bread wheat decreased by 10% and 25%. Increasing the stressor concentration to 8% and 10% mannitol caused a stronger inhibition of seedling growth, resulting in a reduction in root length by an average of 65% and 84% compared to the control (0% mannitol) in durum wheat biotypes, and by 37% and 52%, respectively, in the breeding material of bread wheat.

In the control variant without the stress factor, shoot length of the analyzed spring durum wheat selection material ranged from 5.3 to 11.6 and in spring bread wheat lines from 9.4–16.4 cm. (Fig. 5, 6).



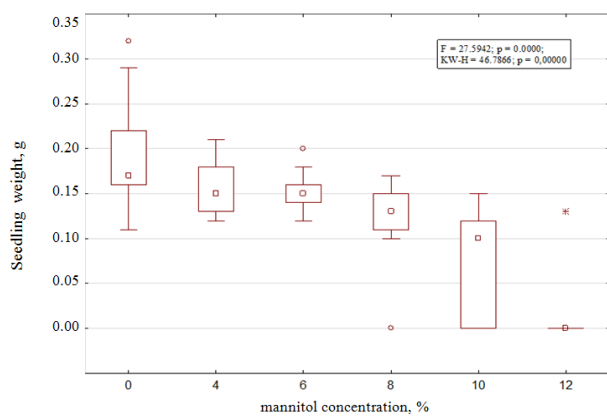
**Fig. 4.** Average root length of seedlings (cm) of wheat accessions at different mannitol concentrations: *a* – spring durum wheat; *b* – spring bread wheat; 0% – control; abscissa shows the mannitol concentra-

tion; the y-axis is the root length of seedling, small square – median, upper and lower rectangle borders – 25% and 75% quartiles, vertical line – minimum and maximum values, circles – outliers; n = 4



**Fig. 5.** Average shoot length of seedlings (cm) of wheat accessions at different mannitol concentrations: *a* – spring durum wheat; *b* – spring bread wheat; 0% – control; abscissa shows the mannitol concentration; the y-axis is the shoot length of seedlings, small square – median, upper and lower rectangle borders – 25% and 75% quartiles, vertical line – minimum and maximum values, circles – outliers; n = 4

The application of mannitol resulted in a reduction in shoot length of both durum and bread wheat seedlings within nearly the same range. At lower mannitol concentrations (4% and 6%), the values decreased relative to the control by 44–46% and 56–59%, respectively. Increasing the mannitol concentration to 8% and 10% led to



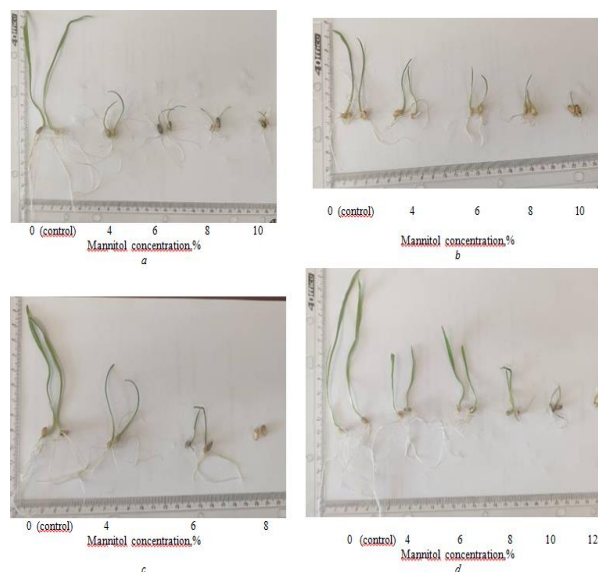
**Fig. 7.** Average seedling weight of seedlings, g of wheat accessions at different mannitol concentrations: *a* – spring durum wheat (*Triticum durum*); *b* – spring bread wheat; 0% – control; abscissa shows the mannitol concentration; the y-axis is the seedling weight, small square – median, upper and lower rectangle borders – 25% and 75% quartiles, vertical line – minimum and maximum values, circles – outliers; n = 4

According to the obtained data (Table 2), accession 328 exhibited the highest tolerance to the stressor in terms of germination, with 9.33%

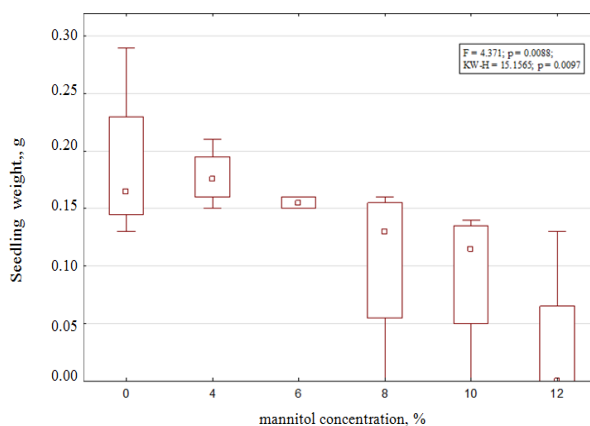
the formation of shoots with an average length of 2.48 and 1.10 cm in durum wheat samples, and 3.10 and 2.30 cm in bread wheat, respectively. Thus, the average reduction in shoot length of durum wheat lines reached 70% and 80% compared to the control, while in bread wheat lines it amounted to 74% and 81%, respectively.

The highest mannitol concentration in the experiment caused the formation of shoots in bread wheat seedlings with an average length of 0.65 cm, which was 92% shorter than that of the control.

Seedling mass in the control variant ranged from 0.13 g to 0.32 g (Fig. 7). In the control variant (0% mannitol), the average seedling mass of the durum wheat breeding samples was 0.20 g. The application of 4% and 6% mannitol reduced this parameter to 0.15 g, or by about 20%. Increasing the mannitol concentration to 8% and 10% caused a further decrease in seedling mass by 25% and 35%, respectively. In bread wheat lines, the use of lower concentrations of the stress factor (4% and 6% mannitol) led to an increase in seedling mass by 31% and 5%, respectively. However, a further increase in mannitol concentration resulted in a decrease in seedling mass by 14%, 23%, and 24%, respectively.



**Fig. 6.** Development of wheat seedlings at different mannitol concentrations: *a-c* – spring durum wheat (*Triticum durum*) accessions; *d* – spring bread wheat (*Triticum aestivum*) accession



mannitol causing a 50% reduction in this parameter. Lines 331 and 337 also demonstrated relatively high stress tolerance. Root length sho-

wed minimal variation in accessions 328, 330, 331, and 337, whereas shoot length was more stable in lines 327, 328, 330, and 332. Seedling weight was least affected in accessions 326, 331, and 333. Notably, the seedling weight of line 327 varied considerably across different mannitol concentrations, which prevented the construction of a reliable linear regression and the determination of an accurate EC<sub>50</sub> for this trait.

**Table 2**

Effective concentration (EC<sub>50</sub>)\* values of mannitol for durum wheat (*Triticum durum*) accessions (%)

Accessions	Germination	Root length	Shoot length	Seedling weight	Average
Chado	5.89	4.03	4.38	4.57	4.72
326	7.76	7.03	4.78	8.64	7.05
327	6.00	6.83	8.42	**	7.08
328	9.33	8.16	8.17	5.00	7.67
329	5.21	6.83	4.24	2.86	4.79
330	5.80	9.30	8.55	11.23	8.72
331	8.05	8.63	6.90	9.77	8.34
332	6.50	7.08	8.33	6.33	7.06
333	7.06	6.44	2.78	9.00	6.32
334	7.00	6.70	6.42	7.50	6.91
335	3.75	8.38	6.43	6.33	6.22

Note: \* – EC<sub>50</sub> (effective concentration 50) – the mannitol concentration that causes a 50% reduction in the studied parameter compared to the control; \*\* – due to the high variability of the response data of accession's 327 seedling weight, EC<sub>50</sub> could not be reliably determined for this parameter.

The application of EC<sub>50</sub> facilitates the identification of breeding material with higher tolerance to mannitol, allowing the selection of resilient genotypes for further breeding. In this study, durum wheat lines 330 and 331 were identified as such, and they are recommended as donors of drought tolerance in subsequent breeding programs.

**Table 3**

Effective concentration (EC<sub>50</sub>)\* values of mannitol for bread wheat (*Triticum aestivum*) accessions (%)

Accessions	Germination	Root length	Shoot length	Seedling weight	Average
336	7.12	6.66	6.33	5.40	6.40
337	8.20	8.04	6.60	5.43	7.07

Note: see Table 2.

## Discussion

Drought tolerance is a complex trait and one of the important components of wheat yield stability. In the study by Ahmed et al. (2022), 40 wheat genotypes were tested under typical regional conditions (control) and under drought conditions – 50% and 75% field capacity. They analyzed root and shoot length, fresh and dry mass, and chlorophyll content. These traits were positively correlated, and improvement in any of them enhanced the others. Genotypes G 47, G 48, G 65, G 68, and G 80 demonstrated the highest drought tolerance and can be used as source material for developing drought-resistant wheat varieties.

Research by Ukrainian scientists is also aimed at developing drought-tolerant wheat breeding material. Based on the analysis of winter wheat varieties for drought tolerance in the Steppe zone of Ukraine, seven main drought tolerance indices were identified: tolerance to drought (TOL), stress susceptibility index (SSI), yield stability index (YSI), relative drought index (RDI), drought index (DI), stress susceptibility percentage index (SSPI), and stress resistance (ISR), as well as three auxiliary indices: yield index (YI), second modified stress tolerance index (M2STI), and harmonic mean productivity (HMP). Using drought tolerance indices and biplot analysis, drought-resistant varieties such as Lyra Odeska, Kokhana, Zysk, and Koshova were identified, while Maria, Nyva Odeska, and Shchedrist Odeska were classified as drought-sensitive (Konovalova et al., 2023).

Recent decades have seen changes in weather and climate conditions, characterized by rising mean annual temperatures and increased

drought risk, prompting agricultural producers to grow intensive and high-yielding varieties for which drought tolerance is crucial. Under modern climate change conditions and severe hydrothermal coefficients, developing drought-tolerant wheat varieties for southern Ukraine is urgent. Scientists have shown that varieties Driada I, Khersonska 99, and Askaniiska have high leaf water-holding capacity, making them drought-tolerant. These varieties respond minimally to stress caused by significant temperature fluctuations and low natural moisture levels (Domaratskyi et al., 2023).

Early diagnosis is used to determine drought tolerance levels. Prokopik et al. (2019) studied seed germination in 17 soft winter wheat varieties developed in the Forest-Steppe and Steppe zones of Ukraine and Central Europe (Germany) using sucrose, mannitol, and sorbitol solutions of various molar concentrations, corresponding to 16 and 18 atm osmotic pressure. The criterion for drought tolerance was a high germination percentage (over 70–80%) under artificial water deficit in all osmotic solutions. On the third day of germination, the highest germination energy under 16 atm osmotic pressure was observed in varieties Valencia, Turunchuk, Knyazhna (Myronivka Wheat Institute), Misiya Odeska, and Blahodarka Odeska (Plant Breeding and Genetics Institute – National Center of Seed and Cultivar Investigation), and under 18 atm in Knyazhna, Valencia, Turunchuk, and Blahodarka Odeska. According to the adopted drought tolerance criterion, on the 7th and 10th day, varieties Albatros Odeskyi, Hratsia Myronivska, Vyshyvanka, Balada Myronivska, and the German variety Samurai also stood out.

Scientists have developed and patented a method for assessing drought tolerance in wheat plants in isolated culture, which involves analyzing callus survival on a selective medium with mannitol and the proportion of germinated seeds on a stressor medium. Correlations between these two indicators were established, and genotypes with increased water stress resistance were identified. Screening of soft winter wheat varieties developed in different ecological zones identified sources of genes for water deficit resistance. The response of varieties to osmotic stress was determined by the proportion of viable calli and the percentage of germinated seeds under the stress factor. Varieties Balada Myronivska and Horlytsia Myronivska demonstrated the highest resistance to water deficit, with the highest germination percentages and callus survival rates on osmotic media. Varieties Ovidiy and Wenzell were more sensitive to water deficit, showing the lowest stress resistance under selective conditions. A significant correlation ( $r = 0.86$ ) was found between drought tolerance indicators obtained by laboratory and biotechnological methods (Pykalo et al., 2019).

## Conclusions

Over the past three agricultural years, there has been an intensification of rising temperatures in the surrounding natural environment against the background of insufficient precipitation. During 2021–2022, the temperature increased by 0.6 °C relative to the long-term average, in 2022–2023 – by 1.6 °C, and in 2023–2024 – by 3.0 °C. These processes highlight the need to develop new drought-tolerant varieties and hybrids of agricultural crops, particularly spring bread wheat and spring durum wheat.

The process of creating drought-tolerant crop accessions can be accelerated using early diagnostic methods.

Through intra- and interspecific hybridization, initial material of spring durum and bread wheat was created, which, upon testing, showed individual characteristics of tolerance to the stress factor, particularly to different mannitol concentrations (4–12%).

Among the analyzed traits, shoot length responded most strongly to stress, while seedling mass was the most stable. Using the lowest 4% mannitol concentration, on average across the breeding accessions, germination decreased by 21%, root length by 26%, seedling mass by 14%, and shoot length by 46%. At 6% mannitol, germination and root mass decreased by 36% and 42% relative to control, shoot length by 56%, and seedling mass by 15%. Higher concentrations (8% and 10%) generally led to reductions in germination by 67% and 93%, root length by 61% and 79%, shoot length by 73% and 88%, and seedling mass by 29% and 58%, respectively.

The use of the EC<sub>50</sub> indicator for the analyzed parameters made it possible to identify lines characterized by lower sensitivity to the stressor.

It is advisable to use the created spring durum wheat accessions 330, 331, and 328 and spring bread wheat accession 337 as initial material in breeding for drought tolerance, as they demonstrated stability under high mannitol concentrations.

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