DOI: https://doi.org/10.53040/cga12.30 UDC: 633.854.78:632.112

COMPARATIVE DROUGHT RESISTANCE IN SUNFLOWER UNDER CONTROLLED CONDITIONS

Maria DUCA[1], ORCID: 0000-0002-5855-5194
mduca2000@yahoo.com
Steliana CLAPCO[1], ORCID: 0000-0001-7147-2740
steliana.clapco@usm.md
Ana MUTU[1], ORCID: 0000-0001-8603-142X
ana.mutu@usm.md
Rodica MARTEA[1], ORCID: 0000-0002-1244-7147
rodica.martea@usm.md
Maria JOITA-PACUREANU[2,3], ORCID: 0009-0009-2797-6672

[1]Moldova State University, Chisinau, Republic of Moldova
[2]Center for Research for Agroforestry Biodiversity "Acad. David Davidescu", Romania
[3]National Agricultural Research and Development Institute, Fundulea, Romania

Abstract. *Under changing climatic conditions, sunflower (Helianthus annuus L.) cultivation is increasingly challenged by abiotic stresses such as drought. This study evaluates the response of three sunflower genotypes to osmotic stress induced by polyethylene glycol (PEG) at concentrations of 10%, 20%, and 30% during germination and early seedling development under controlled conditions. The germination rate was evaluated daily over a 10-day period. Seeds were exposed to PEG to assess germination rate, seedling growth, and stress tolerance indices. Genotype H3 demonstrated moderate tolerance, maintaining satisfactory germination at 10% PEG but showing a marked decline at higher concentrations. H1 was highly sensitive, with poor germination and severely restricted growth of 20% and 30% PEG. In contrast, H2 displayed strong resilience, maintaining consistent germination, root development even under severe stress. These highlight the genotypic variability in sunflowers drought tolerance and suggest that H2 is a promising candidate for cultivation in water-limited environments. H1 may be suitable under moderate stress, while H1 may require additional agronomic interventions. Overall, the selection of stress-resilient sunflower genotypes emerges as a key strategy for enhancing productivity in drought-prone regions.*

Keywords*: *sunflower, drought stress, PEG, germination, seedling

INTRODUCTION

Sunflower (*Helianthus annuus* L.) is a crop of high economic value due to its oil-rich seeds and its contributions to both global food and biofuel production. However, sunflower cultivation is often constrained by abiotic stresses, with

drought being the most critical. Water deficit particularly affects key developmental stages such as germination and early vegetative growth [1-3], ultimately reducing yield potential. These vulnerabilities underscore the importance of targeted breeding strategies to improve drought tolerance.

Enhancing osmotic stress resilience is a central goal of sunflower breeding programs, especially in actual conditions of climate changes. The identification and selection of genotypes with improved adaptive responses to drought are crucial for ensuring stable yields under water-limited conditions. Additionally, other abiotic factors, such as soil salinity and extreme temperatures, further compromise plant performance, reinforcing the demand for robust, multi-stress-tolerant cultivars [3-5].

Osmotic stress, especially that induced by drought, represents a major environmental challenge that significantly affects plant growth and development. The capacity of plants to tolerate such stress varies considerably across species and genotypes, highlighting the need for the understanding of the genetic and physiological mechanisms of resistance. This knowledge is essential for the development of crops with enhanced resilience to the adverse effects of climate change [5].

While field evaluations remain a valuable approach for identifying drought- and heat-tolerant genotypes, they are often constrained by seasonal dependency, high labor demands, and extended timelines. Alternatively, early-stage laboratory assessments under controlled conditions provide a more practical and time-efficient method for evaluating the stress response of genotypes. Polyethylene glycol (PEG), a hydrophilic polymer, is widely used to simulate drought conditions in laboratory settings. This method enables consistent and reproducible evaluation of plant responses to water stress, particularly during early growth stages, and provides critical insight into tolerance mechanisms. Systematic evaluations under PEG-induced osmotic stress are therefore essential for identifying genotypes with superior drought resilience [6-8].

The main objectives of this study are to evaluate the germination, growth, and physiological responses of three sunflower genotypes subjected to osmotic stress induced by different concentrations of PEG-6000.

MATERIALS AND METHODS

The experimental design involved three local sunflower genotypes (H1, H2, and H3), each characterized by a distinct response to drought stress under field conditions, with H2 identified as the most tolerant to hydric stress, followed by H1, and H3 being the most sensitive. The plants were subjected to controlled laboratory conditions (25±3°C) during the germination and early seedling stages. Before the experiment, the seeds of each sunflower genotype were surface-sterilized by

immersing them in 70% ethanol for 2 minutes, followed by treatment with a 10% sodium hypochlorite solution for 10 minutes, and finally rinsed thoroughly with distilled water. Each genotype was subjected to osmotic stress using three concentrations of polyethylene glycol (PEG-6000): 10%, 20%, and 30%, alongside a control group with distilled water. Twenty seeds per genotype were placed in Petri dishes lined with Whatman No. 1 filter paper and moistened with the respective PEG solutions or water. All treatments were performed in duplicate. After 10 days, germination percentage, plant height, root length, fresh weight, and dry biomass were recorded. The germination rate (GR) was calculated based on the formula by George (1967): *GR=(Germinated seeds/Total seeds) × 100* [9].

The stress tolerance indices for plant height (PHSI), root length (RLSI), and dry matter (DMSI) were calculated as follows:

- *PHSI=(Height of stressed plant / Height of control plant) × 100*
- *RLSI=(Root length of stressed plant / Root length of control plant) × 100*
- *DMSI=(Dry matter of stressed plant / Dry matter of control plant) × 100*

Statistical analyses were conducted using the methods outlined by Dospekhov (1985) to assess significant differences among treatments [10].

RESULTS AND DISCUSSIONS

Drought stress affects *Helianthus annuus* by disrupting essential physiological processes, particularly seed germination, a critical phase for successful crop establishment. In this context, the primary objective of the study was to evaluate the germination response and drought tolerance of three sunflower genotypes under osmotic stress conditions induced by polyethylene glycol (PEG).

Numerous studies have investigated the germination rate (GR) of sunflower under water deficit conditions, highlighting genotype-specific responses to osmotic stress. The germination behavior of sunflower genotypes under PEG-induced osmotic stress observed in the present study aligns with previous findings by Praveen et al. (2021), Razzaq et al. (2017), Toscano et al. (2017) [11-13], and others, who reported a gradual reduction in germination rate (GR) with increasing osmotic stress, accompanied by significant variability in drought tolerance and germination dynamics among different genotypes. Similar trends were also noted by Akhter et al. (2025) [14], who emphasized that higher PEG concentrations reduce seed moisture uptake, ultimately resulting in decreased seed germination percentages.

According to present study under control conditions, all genotypes exhibited high and stable GR, ranging from 93,3 to 96,7%. At a 10% PEG6000 concentration, the hybrids maintained successful germination, with GRs remaining stable around 90%. This suggests that a low concentration of PEG6000 imposes only a mild

osmotic stress that does not significantly affect the germination process in the tested genotypes (Figure 1).

At moderate stress levels (20% PEG6000), GR declined to approximately 80% across genotypes. Under severe osmotic stress (30% PEG6000), pronounced genotype-dependent differences were observed. Thus, GR decreased to 60% in genotype H2, while H3 declined further to 46.7%. In contrast, genotype H1 failed to germinate entirely. These findings underscore the varying degrees of drought tolerance among genotypes, with H2 demonstrating the highest resilience under severe osmotic stress during the germination stage. Similar results were reported by Meng et al. (2023) [15], who observed near-total germination failure at high PEG concentrations, including complete inhibition of germination in *Chamaecytisus palmensis* at 30% PEG.

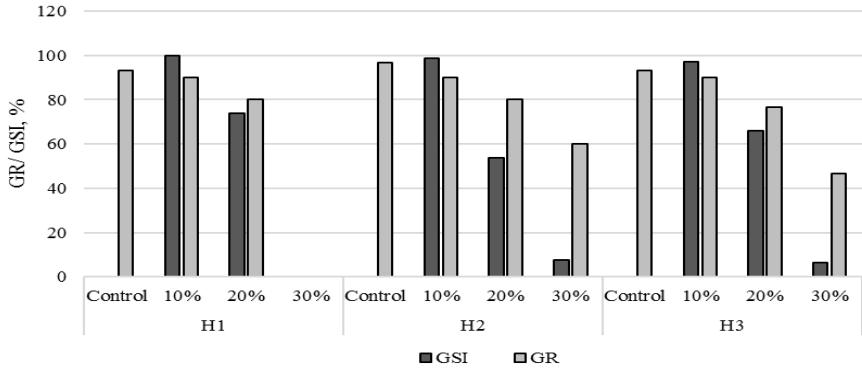


Figure 1. Effect of different PEG concentrations on final germination rate (GR) and germination stress tolerance index (GSI) in some sunflower hybrids

Compared to the control, the mean germination rate of the hybrids was notably delayed under 30% PEG-induced stress. Under control conditions and at low stress levels, nearly all seeds had germinated by the second day of incubation. In contrast, at the highest stress level, the first signs of germination were observed only on the seventh day. A delay in germination was also evident at the moderate stress level (20% PEG6000); by the second day, the germination rates were 40% for H1, 37% for H2, and 20% for H3, respectively. These results are in agreement with previous observations reported for sunflower by El Midaoui et al. (2001) and for *Brassica napus* L. by Willenborg et al. (2004) [16, 17].

The germination stress tolerance index (GSI) values for the tested sunflower hybrids varied notably across osmotic stress levels. Under mild stress conditions, the germination stress index (GSI) remained very high across all genotypes, averaging around 98%. At 20% PEG, GSI values decreased moderately, ranging from 53.9% for H2 to 73.9% for H1, indicating that H1 maintains relatively strong

germination performance even under moderate osmotic stress. In contrast, under severe stress (30% PEG6000), H1 exhibited a GSI of 0%, while the other hybrids showed a dramatic decline, with GSI values of 6.52% and 7.41%, respectively, highlighting a high degree of sensitivity to elevated osmotic stress.

Osmotic stress is known to affect sunflower shoot height, thereby influencing genotype performance during early developmental stages. This trend has been observed by several authors. For example, *Ahmad et al.* (2009) reported that plant height significantly decreased under osmotic stress conditions [8].

The physiological indices such as the Plant Height Stress Index (PHSI), Root Length Stress Index (RLSI), and Dry Matter Stress Index (DMSI) were used to evaluate the response of sunflower hybrids to PEG-induced water stress. A significant reduction in shoot length was observed in all tested genotypes. Shoot growth declined across all genotypes as PEG6000 concentration increased, and hypocotyl elongation was completely inhibited at 30% PEG in most of the analyzed hybrids, consistent with the findings of Toosi et al. (2014) [18].

At 10% PEG6000, hybrid H2 demonstrated the highest shoot height tolerance, with a Plant Height Stress Index (PHSI) of 96.6%, whereas H1 and H3 exhibited substantially lower values of 43.1% and 36.0%, respectively. At 20% PEG, shoot growth was further suppressed: H2 declined to 14.8%, and H3 to 10.9%, while no shoot elongation was recorded for H1. Under severe stress conditions (30% PEG6000), only H2 maintained measurable shoot growth (PHSI of 6.0%), suggesting greater relative tolerance compared to the other genotypes (Figure 2).

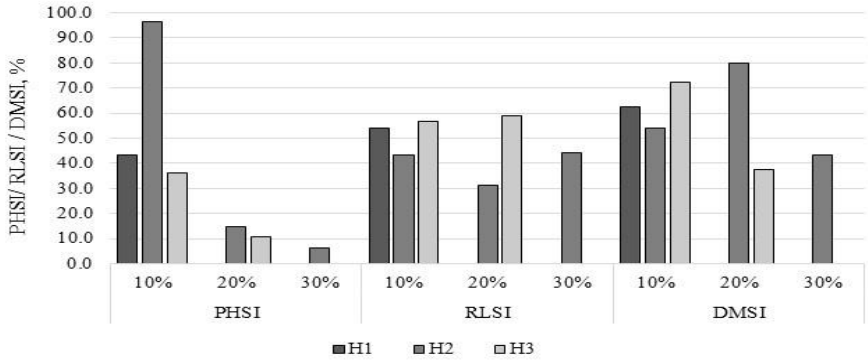


Figure 2. Effect of different PEG concentrations on:
plant height stress tolerance index (PHSI), root length stress tolerance index (RLSI) and dry matter stress tolerance index (DMSI) of some sunflower hybrids

Root growth responded differently, under 10% PEG, RLSI ranged from 43.4% in H2 to 56.7% in H3, with H1 showing 53.9%. At 20% PEG, H3 retained relatively stable root growth (58.9%), while H2 declined to 31.4%, and H1 data was not reported. Under severe stress (30% PEG), H2 recovered slightly (44.0%), while

data was unavailable for the others, indicating a relatively better root development capacity in H2 under high osmotic pressure.

DMSI values followed a decreasing trend with increased stress. At 10% PEG, dry matter accumulation was highest in H3 (72.4%), followed by H1 (62.3%) and H2 (53.8%). Under 20% PEG, H2 demonstrated a superior response with a DMSI of 79.7%, while H3 dropped to 37.6%. At 30% PEG, H2 still maintained a moderate DMSI (43.3%), suggesting greater biomass retention capacity under drought stress. The results are similar with those reported by Ahmad et al. (2009) and Saensee K. et al. (2012) [8, 19].

Overall, H2 demonstrated the highest level of tolerance to PEG-induced osmotic stress based on shoot height, root length, and dry matter accumulation indices. These results indicate that H2 could be a promising candidate for cultivation under moderate to severe drought conditions during the early phases of development.

CONCLUSIONS

The results demonstrate clear genotypic variation in drought tolerance among the three sunflower genotypes tested under PEG-induced osmotic stress. Genotype H2 emerged as the most drought-tolerant, maintaining high germination rates, vigorous seedling growth, enhanced root development, and relatively stable biomass accumulation across all stress levels. H3 exhibited moderate tolerance, with noticeable reductions under severe drought conditions, while H1 proved highly sensitive, showing poor germination and growth performance even under moderate stress.

These laboratory-based findings align closely with field observations, where H2 also demonstrated the highest drought resilience, confirming its adaptability to water-limited conditions. Similarly, H3 showed an intermediate response in both environments, while H1 consistently performed poorly under drought stress. This correlation between controlled laboratory screening and field performance highlights the reliability of PEG-based assays for early-stage genotype evaluation. It supports the use of H2 for cultivation in regions characterized by water deficiency, as well as its inclusion in breeding programs aimed at enhancing drought tolerance in sunflower.

Acknowledgement: This work was supported by the Subprogram "011101 Genetic and biotechnological approaches to agroecosystem management under climate change" funded by the Ministry of Education and Research of the Republic of Moldova.

REFERENCES

1. Puttha, R.; Venkatachalam, K.; Hanpakdeesakul, S.; Wongsa, J.; Parametthanuwat, T.; Srean, P.; Pakeechai, K.; Charoenphun, N. Exploring the Potential of Sunflowers: Agronomy, Applications, and Opportunities within Bio-Circular-Green Economy. In: *Horticulturae* 2023, 9, 1079. https://doi.org/10.3390/horticulturae9101079
2. Debaeke, P., Bedoussac, L., Bonnet, C., Bret-Mestries, E., Seassau, C., Gavaland, A., Raffaillac, D., Tribouillois, H., Véricel, G., & Justes, E. (2017). Sunflower crop: Environmental-friendly and agroecological. In: *OCL*, 24(3), D302. https://doi.org/10.1051/ocl/2017020
3. Kaya, M.D., et al. Seed treatments to overcome salt and drought stress during germination in sunflower (*H. annuus* L.). In: *Europ. J. of Agr.*, 2006, 24(4), 291–295.
4. Ashraf, M., Harris, P.J.C. Potential use of select plant species for amelioration of salt-affected soils. In: *Agriculture, Ecosystems & Environment*, 2004, 102(2–3), 85-94.
5. Rauf, S. Breeding sunflower (*Helianthus annuus* L.) for drought tolerance. In: *Communications in Biometry and Crop Science*, 2008, 3(1), 29-44.
6. Michel, B.E., Kaufmann, M.R. The osmotic potential of polyethylene glycol 6000. In: *Plant Physiology*, 1973, 51(5), 914-916.
7. Farooq, M., et al. Plant drought stress: effects, mechanisms and management. In: *Agronomy for Sustainable Developmen*t, 2009, 29, 185-212.
8. Ahmad, M., et al. Sunflower (*Helianthus annuus* l.) response to drought stress at germination and seedling growth stages. In: *Pak. J. Bot.*, 2009, 41(2): 647-654.
9. George, R.A.T. *Vegetable Seed Production.* London: Leonard Hill Books. 1967.
10. Dospekhov, B.A. *Metodika polevogo opyta* [Methodology of field experiment]. Moscow: Agropromizdat. 1985.
11. Praveen, H. G., Nagarathna, T. K., & Nanja Reddy, Y. A. (2021). Drought tolerance in popular sunflower hybrids at seedling stage. In: *Indian Journal of Agricultural Sciences*, 91(5), 689–694.
12. Razzaq, H., M.H.N. Tahir, H.A. Sadaqat and B. Sadia. 2017. Screening of sunflower (*Helianthus annus* L.) accessions under drought stress conditions, an experimental assay. *J. Soil Sci. Plant Nutr*. 17(3): 662-667.
13. Toscano S., et. al Effects of drought stress on seed germination of ornamental sunflowers. In: *Acta Physiologiae Plantarum*, 2017, 39(8):1-12.
14. Akhter, M. M., Hasan, M. A., Bahadur, M. M., Islam, M. R., Iqbal, M. A., Soufan, W., Aurib, K., Akhter, T., Soysal, S., Elsabagh, A., Heniesh, M., & Islam, M. S. (2025). Wheat genotypes respond differently under polyethylene glycol (PEG)-induced drought stress during germination and early seedling growth stages. *Pakistan Journal of Botany*, 57(1).
15. Meng, Z., & Gao, Y. (2020). Effects of polyethylene glycol (PEG)-simulated drought stress on Chamecytisus palmensis seed germination. IGC Proceedings (1989–2023), (33).
16. El Midaoui, M., et al. Effect of osmotic pressure on germination of sunflower seeds (*Helianthus annuus* L.). In: *Helia*, 2001, 24(35):129-134.
17. Willenborb CJ, Gulden RH, Jhonson EN, Shirtliffe SJ (2004) Germination characteristics of polymer-coat canola (*Brassica napus* L.) seeds subjected to moisture stress at different temperatures. Agron J 96(3):786–791.
18. Toosi, A. F., Bakar, B. B., & Azizi, M. (2014). Effect of drought stress by using PEG 6000 on germination and early seedling growth of *Brassica juncea* var. *Ensabi*. *Scientific Papers. Series A. Agronomy*, *57*, 360–363. ISSN 2285-5785.
19. Saensee, J., et al. (2017). Selection of drought-tolerant sunflower hybrids under water-limiting conditions. In: *Breeding Science*, 67(2), 158-167.