



EFFECT OF POTASSIUM OPTIMIZATION ON WHEAT DROUGHT TOLERANCE IN CONTROLLED CONDITIONS

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Abstract: Wheat (*Triticum* spp.) is an important cereal crop consumed worldwide, but it is highly susceptible to drought. Potassium plays an essential role in osmotic regulation, photosynthesis, and nitrogen assimilation, all of which are critical for maintaining plant growth and productivity under stress conditions. The aim of this study is to investigate how different potassium levels, including sufficient potassium (SK, 1 mM) and low potassium (LK, 0.05 mM), affect the drought tolerance of wheat during the early stages of seedling development under PEG-induced drought stress. Plant physiological development, canopy temperature, photosynthetic efficiency, antioxidant defense enzymes, and nitrogen assimilation enzymes were assessed in the experiment. In non-drought conditions, LK increased canopy temperature and reduced dry matter yield and photosynthetic performance, with these effects becoming more pronounced under drought stress. SK-treated plants exhibited higher biomass, chlorophyll content, maximum quantum efficiency of photosystem II, and lower canopy temperatures, even under drought conditions. Furthermore, LK restricted the accumulation of key osmotic regulators, including proline, amino acids, and soluble sugars. Under drought stress, LK plants also showed increased hydrogen peroxide and superoxide anion levels, while SK plants had lower reactive oxygen species accumulation and higher antioxidant enzyme activities (catalase and superoxide dismutase). Additionally, LK resulted in reduced activity of nitrogen assimilation enzymes (nitrate reductase, NR, and nitrite reductase, NiR) under both normal and drought conditions. In contrast, SK-treated wheat seedlings maintained higher NR and NiR activities and higher soluble protein content during drought stress. These findings underscore the critical role of potassium management in enhancing wheat yield, particularly in water-scarce regions, as optimal potassium supply strengthens essential physiological and biochemical mechanisms that improve plant tolerance to drought stress.

Keywords: Drought stress, Potassium, Nitrate assimilation, Oxidative stress

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Received: October 25, 2024

Accepted: December 04, 2024

Published: January 15, 2025

Cite as: Uğurlar F. 2025. Effect of potassium optimization on wheat drought tolerance in controlled conditions. BSJ Agri, 8(1): xxx-xxx.

1. Introduction

Wheat (*Triticum* spp.) is a significant cereal crop extensively produced globally and is essential for human nutrition (Igrejas and Branlard, 2020). Wheat, which is prone to higher yields under irrigated conditions (Mansour et al., 2020), is a highly water-sensitive plant (Zhao et al., 2020). Experiencing drought during various growth stages leads to considerable reductions in wheat yield (Hu et al., 2021). Drought stress is a critical environmental challenge that endangers global food security, particularly in areas with limited water resources and irregular precipitation (Begna, 2020; Pequeno et al., 2021). Drought stress severely impacts crop productivity by disrupting basic physiological processes of plants, including water uptake, nutrient absorption, and photosynthesis (Pamungkas and Farid, 2022; Wahab et al., 2022; Qiao et al., 2024; Ramazanoglu et al., 2024).

Plants subjected to drought stress experience significant physiological disturbances, primarily due to water loss, which leads to osmotic stress and a reduction in turgor pressure (Yang et al., 2021; Hemati et al., 2022). In the

later stages of water scarcity, plants close their stomata to minimize water loss through transpiration, but this also restricts CO₂ uptake, reducing photosynthesis and inhibiting plant growth (Song et al., 2020; Shohat et al., 2021). Additionally, drought stress induces oxidative stress, which harms cellular structures such as lipids, proteins, and DNA by promoting the accumulation of reactive oxygen species like hydrogen peroxide and superoxide anion (Zheng et al., 2023). This oxidative stress amplifies the detrimental impact of drought, resulting in disrupted cellular function and decreased biomass (Seleiman et al., 2021). In response, plants activate antioxidant defense systems to mitigate oxidative stress (Akbari et al., 2022) and accumulate osmotic regulators like proline, amino acids, and soluble sugars to preserve cellular homeostasis (Ahmad et al., 2020; Choudhary et al., 2023; Mehta and Vyas, 2023).

Nitrate, an important nitrogen source for plants in agricultural soils (Shafreen et al., 2021), is converted into ammonium within plant cells through the actions of the enzymes nitrate reductase and nitrite reductase (Kumari et al., 2022). The ammonium produced from nitrate



reduction is utilized in the synthesis of amino acids and proteins with the assistance of various enzymes (Taria et al., 2022). Drought stress impairs nitrate uptake and assimilation by diminishing the activity of these enzymes, thereby reducing the nitrogen use efficiency in plants (Khatoon et al., 2024; Wei et al., 2024). This reduction in nitrate assimilation leads to a decline in photosynthetic capacity and carbon uptake, adversely impacting plant health and yield (Zayed et al., 2023). Consequently, regulating nitrogen metabolism under stress conditions is crucial for improving plant tolerance (Baslam et al., 2020; Ye et al., 2022).

Potassium is a vital macronutrient that helps plants adapt to stress conditions by regulating key physiological and biochemical processes (Ul-Allah et al., 2020). Research shows that potassium enhances drought tolerance by promoting root growth, controlling stomatal movements, supporting photosynthesis, facilitating protein synthesis, and maintaining cellular osmotic balance (Xu et al., 2021; Johnson et al., 2022; Mostofa et al., 2022). Additionally, a positive relationship has been found between potassium and nitrate uptake and transport in plants. Ye et al. (2022) found that potassium boosts nitrate uptake and assimilation, while nitrate, in turn, facilitates potassium uptake and transport from roots to shoots. Feng et al. (2020) also observed that potassium improves plant resistance by regulating nitrogen uptake and assimilation. Furthermore, potassium reduces oxidative stress in plants by activating antioxidant enzymes that neutralize reactive oxygen species (Taha et al., 2020; Tittal et al., 2021). However, potassium availability can be limited by factors such as low potassium reserves (Reimer et al., 2020), leaching (Dianjun et al., 2022), and potassium fixation (Celik et al., 2023), all of which can contribute to potassium deficiency in plants.

Due to potassium's ability to regulate key physiological and biochemical processes under drought stress, plant responses to drought conditions largely depend on the availability of this nutrient. However, information on the effects of K deficiency on nitrogen metabolism, oxidative stress, and osmotic adjustment in plants under drought stress is limited. Therefore, this study aims to investigate the impact of potassium nutritional status on the stress tolerance of wheat plants exposed to drought stress during the early stages of seedling development. The research focuses on assessing the role of potassium under both optimal growth conditions and drought stress simulated using polyethylene glycol (PEG).

2. Materials and Methods

This study was conducted in the plant growth chambers at the Faculty of Agriculture, Harran University in 2023, using the wheat variety 'Edessa' as the research plant. Seedlings germinated in perlite were transferred to 5 L PVC containers filled with aerated Hoagland solution, which was refreshed every two days throughout the experiment. Half of the containers were supplied with a

nutrient solution containing low potassium (0.05 mM), while the other half received a sufficient potassium concentration (1 mM K₂SO₄).

Polyethylene glycol 6000 (PEG 6000) was chosen in this study to simulate drought stress. PEG 6000, as an inert and non-ionic molecule, reduces water potential uniformly without entering plant cells, making it a reliable option for creating drought stress under controlled conditions (Mohi-Ud-Din et al., 2021; Batool et al., 2022). PEG 6000 has been extensively used in drought tolerance studies across various plant species (Bukhari et al., 2021; Guo et al., 2022; Mahpara et al., 2022; Qi et al., 2023). However, the water deficit created by PEG does not fully capture all aspects of natural drought conditions, such as fluctuations in temperature and humidity. Therefore, the findings reflect a drought simulation under controlled conditions and should be further validated under natural settings.

After a 10-day growth period, PEG 6000 was added to the pots at a concentration of 200 g L⁻¹ (20%) to induce drought stress, and the plants were harvested six days later. The experiment consisted of 12 pots, with 4 treatments and 3 replicates for each treatment.

2.1. Biomass Yield and Potassium Content Measurement

Shoots and roots were washed with distilled water and kept in a 70 °C oven until they reached constant weight, and then their dry weights were recorded. The samples were then ashed at 500 °C, digested with HCl, and analyzed using a flame photometer to determine their potassium content (Kacar and Inal, 2008).

2.2. Chlorophyll Content and Chlorophyll Fluorescence

Chlorophyll content was measured by homogenizing fresh plant leaves in 80% acetone and measuring absorbance at 663 and 645 nm (Arnon, 1949). Chlorophyll fluorescence was assessed using a photosynthetic efficiency analyzer (Walz, Germany) after dark-adapting leaves for 30 minutes.

2.3. Proline, Total Free Amino Acids, and Soluble Sugar Content

Proline levels were measured following the method outlined by Bates et al. (1973). Leaf samples were homogenized in 3% sulfosalicylic acid and filtered through filter paper. An aliquot of the supernatant was then mixed with acid ninhydrin solution and acetic acid. The mixture was heated at 100°C for 1 hour and subsequently cooled in an ice bath. To isolate free proline, 4 ml of toluene was added and vortexed. The absorbance of the mixture was measured at 520 nm using a microplate spectrophotometer (Epoch, SN: 1611187, ABD), with pure toluene as the blank. Proline concentration was determined using a standard curve prepared with L-proline.

The method described by Lee and Takahashi (1966) was used to determine total free amino acids. Fresh plant leaves were incubated in distilled water at 45 °C for 1 hour. After incubation, 100 µl of the centrifuged sample

was transferred to a new tube, and 1.9 mL of ninhydrin-glycerol-citrate buffer was added. The mixture was heated in boiling water for 12 minutes, then cooled to room temperature in a water bath. The absorbance was measured at 570 nm using a microplate spectrophotometer. Total free amino acids were quantified using a standard curve prepared with glycine. The soluble sugar content of leaf samples was determined following the method of Dubois et al. (1956). Leaf samples were treated with 5 ml of 2.5 N HCl and incubated in a water bath at 80 °C for 2 hours. After cooling, concentrated sulfuric acid and 5% phenol were added, and the mixture was shaken. The absorbance of the solution was measured at 490 nm using a microplate spectrophotometer.

2.4. Oxidative Stress Markers

The hydrogen peroxide (H₂O₂) concentration was determined following the method described by Loreto and Velikova (2001). Fresh plant leaves were homogenized in 0.1% TCA and centrifuged at 15,000 rpm at 4 °C for 15 minutes. The H₂O₂ content was measured by adding 10 mmol L⁻¹ K-phosphate buffer and 1 M KI to the homogenate, and the absorbance was recorded at 390 nm.

Superoxide anion levels were measured according to the method by Elstner and Heupel (1976). Fresh leaf samples were homogenized in 65 mM phosphate buffer (pH 7.8) and centrifuged at 5000 x g for 10 minutes. The supernatant was treated with 10 mM hydroxylamine and homogenization buffer, then incubated at 25 °C for 20 minutes. Following incubation, 1 mL of aminobenzene sulfonic acid and α -naphthylamine were added and incubated again at 25 °C for 20 minutes. The absorbance was measured at 530 nm using a spectrophotometer, and the results were calculated using a standard curve prepared with NaNO₂.

2.5. Activities of Antioxidant Enzymes and Total Soluble Protein Content

Fresh plant leaves were homogenized by adding 0.1 mol L⁻¹ K-phosphate buffer (containing 1% PVP and 0.1 mmol L⁻¹ EDTA). The extract was centrifuged at 15,000 rpm and +4 °C for 15 min.

Superoxide dismutase (SOD) activity was measured following the method of Beauchamp and Fridovich (1971). The activity was determined by measuring the absorbance at 560 nm using a microplate spectrophotometer after incubating the reaction mixture (0.05 M K-phosphate buffer (pH 7.8), 0.05 M sodium carbonate, 13 mM L-methionine, 100 μ M EDTA, 75 μ M NBT, 5 μ M riboflavin, and 25 μ L supernatant) under a fluorescent lamp for 15 minutes. A tube without the enzyme extract served as the control, while a complete mixture that was not irradiated and did not develop color was used as the blank.

Catalase (CAT) enzyme activity was determined following the procedure described by Aebi (1984). The absorbance change of the reaction mixture (50 mmol L⁻¹ K-phosphate, 10 mmol L⁻¹ H₂O₂, 4 mmol L⁻¹ Na₂EDTA and

10 μ L supernatant) at 240 nm was measured for 5 min (Karakas 2013).

Total soluble protein content was measured using the Bradford method (1976). Briefly, 200 μ L of Bradford reagent (prepared according to Bradford (1976) recipe) was added to 10 μ L of protein extract. Samples were kept at room temperature for 15 min and their absorbance at 595 nm was determined by microplate spectrophotometer. The results were calculated with a standard curve prepared with Bovine Serum Albumin (BSA).

2.6. Nitrate Reductase (NR) and Nitrite Reductase (NiR) Activity

The NR activity was determined using the method described by Jaworski (1971). Fresh leaf samples were placed in test tubes with 0.1 M phosphate buffer (comprising 20 mM KNO₃ and 5% propanol) and incubated for 2 hours at 25 °C in darkness. At the conclusion of incubation, 0.3 mL of a mixture containing 1% sulfanilamide and 0.02% naphthylenediamine dihydrochloride was introduced to a 0.4 mL aliquot. After 20 minutes, the absorbance of samples diluted with 0.4 mL of deionized water was measured at 540 nm using a microplate spectrophotometer. Nitrite production was quantified with a NaNO₂ calibration curve.

To measure NiR activity, the method of Ramirez et al. (1966) was used. Leaf samples were homogenized in potassium phosphate buffer prepared as described in the method. 1.4 ml potassium phosphate buffer, 5 mmol L⁻¹ KNO₂ and 100 μ l 2 mg/ml methyl viologen and 100 μ l enzyme extract were mixed and the volume was completed to 1.8 ml with distilled water. The reaction was initiated by adding 200 μ l sodium dithionite and incubated at 30 °C for 30 min. After incubation, 100 μ l of the mixture was added with 1.9 ml water and vortexed. Then, 2 ml of 1% sulfanilamide and 0.05% (w:v) N-(1-naphthyl)-ethylenediamine hydrochloride solution was added. The solution was incubated again, and its absorbance was measured at 540 nm.

2.7. Statistical Analysis

All data obtained were analyzed using SPSS (Version 22.0) program with variance analysis (ANOVA) and Duncan Multiple Comparison Test at a significance level of P<0.05 and presented in graphs (Genç and Soysal, 2018).

3. Results

3.1. Effect of Potassium Availability and Drought Stress on Plant Temperatures, Potassium Uptake and Biomass Accumulation

Thermal imaging revealed that potassium (K⁺) deficiency elevated plant canopy temperature (Figure 1A). When K⁺ deficiency was combined with drought stress (DS), plant temperatures rose even further. However, supplementing K⁺ to the growth medium seemed to mitigate temperature increases under DS. Overall, low potassium (LK) levels tended to result in higher plant temperatures.

Shoot K^+ content (Figure 1B) significantly increased in the sufficient potassium (SK) treatment compared to the LK treatment, under both control and DS conditions. In control conditions, the shoot K^+ content of SK-treated plants was approximately twice that of LK-treated plants, whereas under drought stress, this ratio decreased to 1.4-fold.

The treatments also significantly changed the shoot dry weight (DW) and root DW (Figure 1C, D). Without drought stress, SK-treated plants had 34% higher shoot DW and 15% higher root DW than LK-treated plants. DS reduced shoot DW by 46% in LK plants and by 42% in SK plants, with root DW decreasing by 44% in both treatments. The reduction in biomass was more severe in LK plants under DS.

3.2. Effects of Potassium Deficiency and Drought Stress on Chlorophyll Content and Chlorophyll Fluorescence

Total chlorophyll (Chl) content significantly decreased under DS in both LK and SK treatments (Figure 1E). Under control conditions, there was no significant difference in Chl content between LK and SK treatments. However, under DS, Chl content in SK-treated plants was approximately 35% higher than in LK-treated plants. Notably, LK-treated plants experienced a pronounced reduction in Chl content when exposed to drought. Compared to control conditions, Chl content decreased by 54% in LK-treated plants and by 44% in SK-treated plants under DS.

Photosystem II efficiency (F_v/F_m) followed a similar trend, with DS reducing F_v/F_m values in both treatments (Figure 1F). LK plants showed a 39% reduction under DS, while SK plants had a smaller decrease of 25%, indicating that potassium helps maintain photosynthetic efficiency under drought conditions.

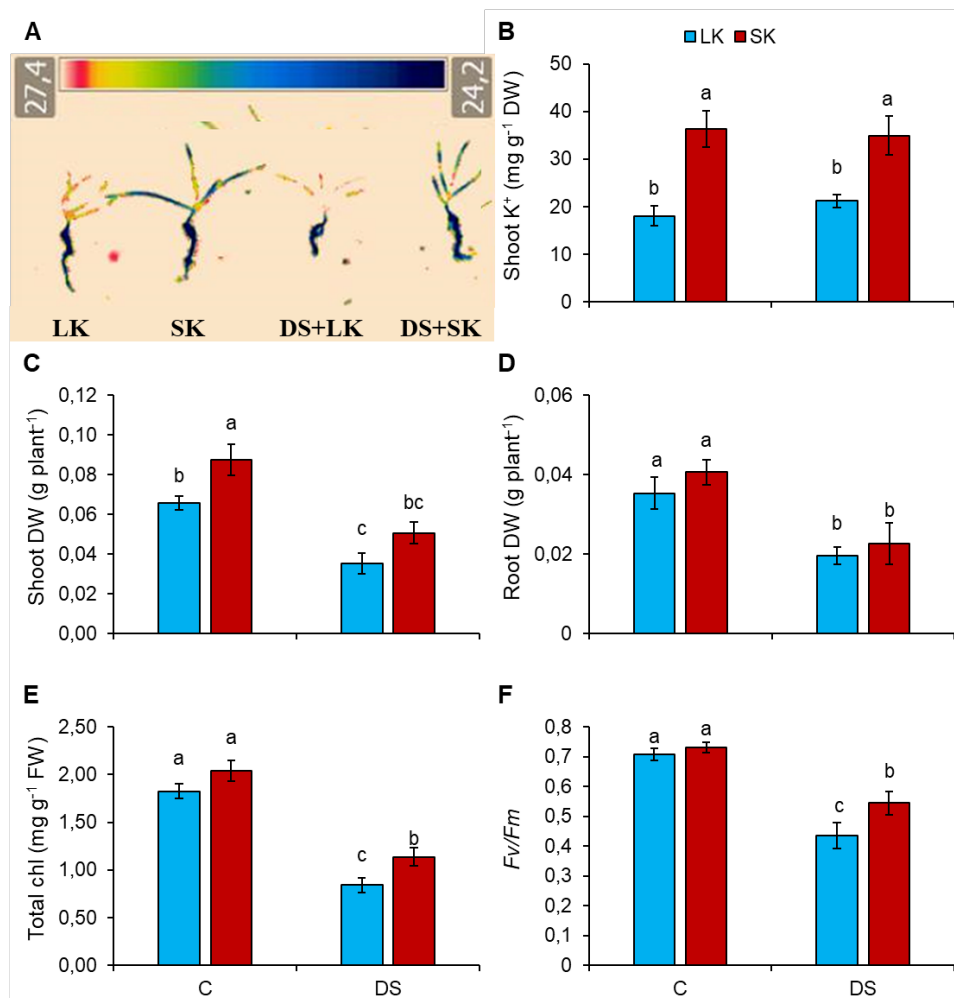


Figure 1. A) Thermal image, B) Shoot potassium (K^+) content, C) Shoot DW, D) Root DW, E) Total chlorophyll (chl) and F) Photosystem II quantum efficiency (F_v/F_m) in wheat seedlings grown under drought stress (DS) and low K (LK) or sufficient K (SK). Error bars represent the standard error of each treatment (n=3). Letters above the bars indicate statistical differences ($P < 0.05$) between treatments.

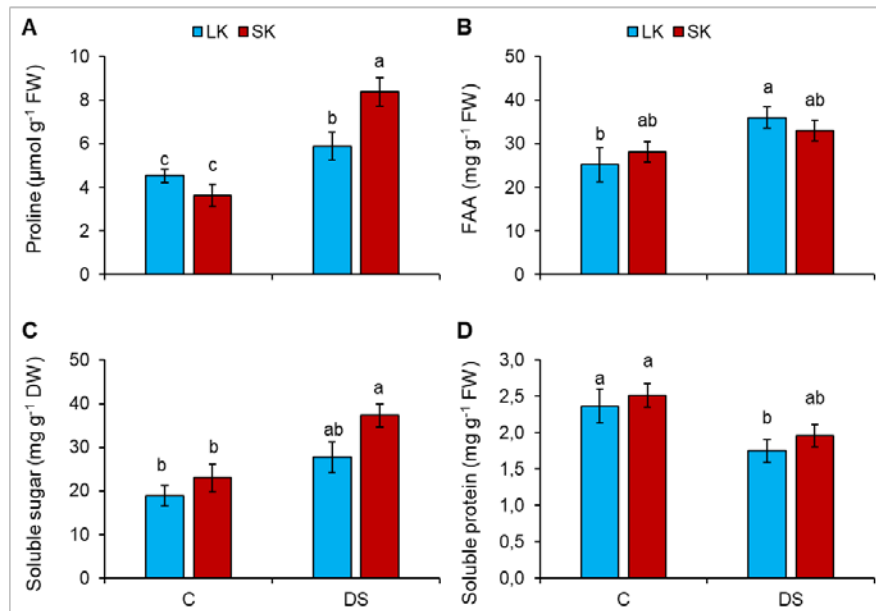


Figure 2. A) Proline, B) total free amino acids (FAA), C) soluble sugar and D) Soluble protein content in wheat seedlings grown under drought stress (DS) and low K (LK) or sufficient K (SK). Error bars represent the standard error of each treatment (n=3). Letters above the bars indicate statistical differences ($P < 0.05$) between treatments.

3.3. Effects of Potassium Deficiency and Drought Stress on Osmolyte Accumulation and Soluble Protein Levels

Under DS, proline content increased approximately 2.3-fold in SK plants, while it rose by about 1.3-fold in LK plants, indicating that potassium enhances proline accumulation for drought tolerance (Figure 2A). Total free amino acids (FAA) increased modestly under drought (Figure 2B), with SK and LK plants showing increases of 4% and 5.6%, respectively, compared to control conditions. Under DS, LK plants exhibited 8% higher FAA content than SK plants. Soluble sugars also increased significantly under DS, with SK showing a 62% rise compared to control, while LK increased by 46% (Figure 2C). Consequently, SK plants accumulated 35% more soluble sugars than LK plants under DS. No significant difference in soluble protein content was observed under control conditions (Figure 2D). However, under DS, protein content decreased by 22% in SK plants and 26% in LK plants, with SK plants having 12% higher protein content than LK plants.

3.4. Oxidative Stress and Antioxidant Enzyme Activity in Wheat Seedlings under Potassium Deficiency and Drought Stress

The results in Figure 3 demonstrate that DS and potassium availability significantly affect oxidative stress markers and antioxidant enzyme activities in wheat seedlings.

Hydrogen peroxide (H_2O_2) levels increased under DS in both LK and SK treatments (Figure 3A), with H_2O_2 content in LK-treated plants being approximately 21% higher than in SK-treated plants under DS. Superoxide anion (O_2^-) production rates also rose significantly under drought, with SK plants showing a 47% decrease in O_2^- levels compared to LK plants under stress (Figure 3B).

Catalase (CAT) activity increased under DS in both LK and SK treatments (Figure 3C), with LK-treated plants exhibiting approximately 37% higher CAT activity than SK plants. Superoxide dismutase (SOD) activity also increased slightly under drought conditions, with LK-treated plants showing about 14% higher SOD activity compared to SK under DS (Figure 3D).

3.5. Nitrogen Metabolism Enzymes in Wheat Seedlings under Potassium Deficiency and Drought Stress

The results for nitrate reductase (NR) and nitrite reductase (NiR) activities under potassium deficiency and DS are shown in Figures 3E and 3F. Under control conditions, NR activity in SK-treated plants was approximately 28% higher than in LK-treated plants. DS significantly reduced NR activity in both treatments, with activity decreasing by 47% in SK and 58% in LK compared to control. However, even under DS, SK plants had 59% higher NR activity than LK plants.

NiR activity followed a similar pattern to NR. Under control conditions, SK plants exhibited 41% higher NiR activity than LK plants. DS led to a 31% reduction in LK plants and a 38% reduction in SK plants. Despite the decline, SK plants maintained 25% higher NiR activity under DS compared to LK plants.

4. Discussion

Potassium (K^+) is essential for plants because it regulates stomatal openings (Ahammed et al., 2022), enhances gas exchange efficiency (Simões et al., 2020), maintains water balance (Lu et al., 2022), activates enzymes critical for photosynthesis and protein synthesis (Mostafa et al., 2022; Rawat et al., 2022), and boosts tolerance to environmental stresses such as drought (Hasanuzzaman et al., 2018).

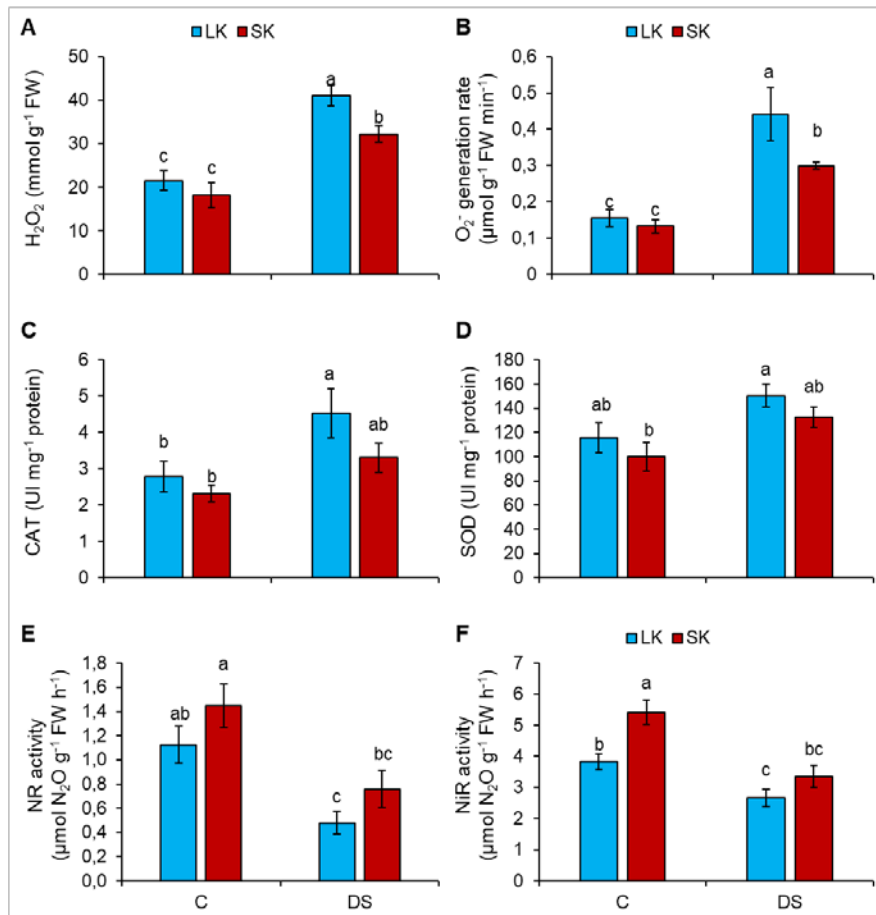


Figure 3. A) Hydrogen peroxide (H₂O₂) content, B) superoxide (O₂⁻) generation rate, C) Catalase (CAT) activity, D) Superoxide dismutase (SOD) activity, E) Nitrate reductase (NR) activity and E) Nitrite reductase (NiR) activity in wheat seedlings grown under drought stress (DS) and low K (LK) or sufficient K (SK). Error bars represent the standard error of each treatment (n=3). Letters above the bars indicate statistical differences (P<0.05) between treatments.

Numerous studies have observed that potassium-enriched plants exhibit greater resistance to environmental stressors (Wang et al., 2013; Sardans and Peñuelas, 2021; Johnson et al., 2022; Pantha et al., 2023). Potassium deficiency significantly reduced biomass accumulation, especially under drought stress. The reduction in biomass observed in potassium-deficient plants can be attributed to impaired water relations (Tavakol et al., 2018) and decreased photosynthetic capacity (Pan et al., 2017), both of which are exacerbated under drought stress. Potassium is necessary to regulate stomatal activity and supports cooling through transpiration (Hasanuzzaman et al., 2018). In LK-treated plants, reduced K⁺ availability likely led to impaired stomatal function, causing higher canopy temperatures and reduced water use efficiency. This resulted in lower shoot and root biomass, likely limiting carbon assimilation and reducing the energy available for growth (Tighe-Neira et al., 2018). Additionally, the decrease in biomass can be linked to potassium's role in maintaining chlorophyll content and photosynthetic efficiency (Tighe-Neira et al., 2018; Ju et al., 2021). Bo et al. (2020) demonstrated that potassium deficiency reduces chlorophyll content and the quantum efficiency of Photosystem II (*F_v/F_m*), which aligns with our findings

under drought stress. The decrease in photosynthetic parameters is probably due to increased oxidative stress or impaired chlorophyll synthesis (Khodabakhshi et al., 2023). In contrast, plants treated with adequate potassium with higher chlorophyll levels and photosynthetic efficiency were more resistant to drought stress, thus resulting in higher biomass compared to potassium deficient plants. Therefore, adequate nutrition of plants with potassium plays a critical role in maintaining plant growth in both normal and drought conditions by supporting photosynthesis and water relations (Wang et al., 2013; Rawat et al., 2022; Fang et al., 2023).

Plants accumulate organic substances such as proline, soluble sugars, and soluble proteins to reduce damage caused by drought stress, protect cellular structures, and maintain metabolic activities (Fang et al., 2022; Alagoz et al., 2023). These compounds play a key role in reducing the effects of reactive oxygen species (ROS), aiding osmotic adjustment, maintaining cell water balance, and enhancing stress tolerance (Afzal et al., 2021; Alagoz et al., 2023). In our study, drought stress induced the accumulation of proline, soluble sugars, and free amino acids. Many researchers have reported similar increases in these compounds under drought stress in various

plants (Holmstrup et al., 2015; Gurrieri et al., 2020; Nguyen et al., 2020; Semida et al., 2020; Živanović et al., 2020). Potassium enhances osmoregulation in plants by increasing cell turgor and maintaining water balance (Kumar et al., 2020; Pandey and Mahiwal, 2020). In our study, plants with adequate potassium under drought stress accumulated higher levels of proline and soluble sugars, emphasizing potassium's key role in regulating osmotic potential. Similar patterns have been reported in cotton (Zahoor et al., 2017), wheat (Ahanger et al., 2017), and sunflower (Shehzad et al., 2020), highlighting potassium's contribution to osmotic adjustment under stress. These results emphasize the importance of maintaining optimal potassium levels to enhance plant resilience to drought.

Drought stress induced oxidative stress in wheat seedlings, as indicated by increased levels of hydrogen peroxide (H_2O_2) and superoxide anion (O_2^-). These reactive molecules damage cellular structures such as lipids, proteins, and nucleic acids, leading to the disruption of cellular functions (Zheng et al., 2023). In our study, plants exposed to potassium deficiency exhibited higher O_2^- and H_2O_2 production rates, indicating that adequate potassium reduces the extent of oxidative damage under drought (Johnson et al., 2022). Similar patterns of increased ROS under potassium deficiency have been observed in barley (Tavakol et al., 2021), rapeseed (Zhu et al., 2020), tomato (Siddiqui et al., 2021), and sorghum (Tittal et al., 2021), reinforcing potassium's critical role in managing oxidative stress. Plants activate antioxidant defense mechanisms to mitigate ROS-induced damage (Nowroz et al., 2024). Superoxide dismutase (SOD), a widespread metalloenzyme, converts O_2^- into molecular oxygen and H_2O_2 , providing the primary defense against oxidative stress (Mishra and Sharma, 2019). Increased catalase (CAT) activity under stress conditions breaks down H_2O_2 into harmless compounds, playing a crucial role in safeguarding plants against oxidative stress (Dikilitas et al., 2016). Although both treatments showed increased CAT and SOD activity under drought, LK-treated plants had higher antioxidant enzyme activity than SK-treated plants, possibly as a compensatory response to elevated oxidative stress. Nevertheless, the results indicate that potassium sufficiency reduces oxidative stress, likely by maintaining better cellular homeostasis and membrane stability (Sardans and Peñuelas, 2021).

Nitrate (NO_3^-) is an important nitrogen source for plants in agricultural soils and must be reduced to ammonium (NH_4^+) form for the synthesis of proteins and other organic compounds in plants (Shafreen et al. 2021; Islam et al., 2022). Nitrate reductase (NR) reduces NO_3^- to nitrite (NO_2^-) in plant cells, and this NO_2^- is reduced to NH_4^+ by the nitrite reductase (NiR) enzyme (Kumari et al., 2022; Chen et al., 2023). NH_4^+ is assimilated into amino acids and proteins by glutamine synthetase-glutamate synthase enzymes or glutamate dehydrogenase in the plant (Taria et al., 2022). Stress

exposure leads to a reduction in the transcript levels of nitrogen-assimilating enzymes, including NR, NiR, and glutamate synthase (Sahay et al., 2021; Sathee et al., 2021). The suppressive effect of drought stress on NR and NiR enzyme activities has been previously reported in many plants such as soybean (Du et al., 2020; Qu et al., 2023), maize (Majeed et al., 2020), Brassica juncea (Sahay et al., 2021) and pepper (Kaya and Shabala, 2023). Our results showed that drought stress decreased NR and NiR enzyme activities in wheat plants, like other reports. However, potassium deficiency worsened the negative effects on nitrogen metabolism by further decreasing NR and NiR activities under both normal and drought stress conditions. This aligns with the findings of Hu et al. (2016), who reported that potassium deficiency significantly reduced NR activity and nitrogen assimilation in cotton, leading to impaired nitrogen metabolism and reduced growth. This reduction in nitrate assimilation not only disrupts nitrogen metabolism but also directly affects photosynthetic efficiency, as it limits CO_2 availability (Ivanov et al., 2023). Potassium supplementation, on the other hand, improved NR and NiR activities, leading to better nitrogen assimilation and photosynthetic efficiency under drought stress. This demonstrates potassium's vital role in regulating nitrogen metabolism, especially under stress conditions (Zhong et al., 2017; Liu et al., 2022). Consequently, maintaining sufficient potassium levels is crucial for enhancing stress tolerance by supporting nitrogen use efficiency and sustaining overall plant growth and productivity.

5. Conclusion

This study highlights that potassium plays an important role in regulating wheat plant responses to drought stress. Potassium supply alleviates the adverse effects of drought by regulating water relations in plants, increasing photosynthetic efficiency, promoting osmolyte accumulation, reducing oxidative damage, and maintaining nitrogen metabolism. The findings highlight the need to maintain potassium levels in agricultural soils to enhance wheat resilience in water-limited environments. However, the findings are based on controlled conditions using PEG to simulate drought, which does not fully capture natural environmental variables like temperature and humidity fluctuations. Thus, future studies should focus on evaluating the effects of potassium supplementation under field conditions where multiple environmental factors interact. Moreover, while the study focused on the early seedling stage of wheat, potassium's effects on drought tolerance at later developmental stages should be further investigated.

Author Contributions

The percentage of the author(s) contributions is presented below. The author reviewed and approved the final version of the manuscript.

	F.U.
C	100
D	100
S	100
DCP	100
DAI	100
L	100
W	100
CR	100
SR	100
PM	100
FA	100

C=Concept, D= design, S= supervision, DCP= data collection and/or processing, DAI= data analysis and/or interpretation, L= literature search, W= writing, CR= critical review, SR= submission and revision, PM= project management, FA= funding acquisition.

Conflict of Interest

The author declared that there is no conflict of interest.

Ethical Consideration

Ethics committee approval was not required for this study, as it did not involve any research on humans or animals.

Acknowledgments

The author expresses gratitude to Harran University for providing access to digital resources, laboratory facilities, and plant growth chambers.

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