



Effect of nitrogen and sulfur combinations on spring wheat (*Triticum aestivum*) growth, yield, and soil nutrient availability in a greenhouse experiment

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Abstract

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Effective management of nitrogen (N) and sulfur (S) is crucial for maximizing spring wheat productivity, as both nutrients play key roles in improving growth, yield attributes, grain protein content, and soil fertility. Despite their importance, determining the optimal application rates of N and S for enhanced wheat performance remains a challenge. This study was conducted as a pot experiment under controlled greenhouse conditions, with 12 treatments replicated three times and carried out over 85 days. The treatments included a control (0N + 0S), nitrogen-only treatments (40N + 0S, 80N + 0S, 120N + 0S), sulfur-only treatments (30S, 60S), and combined N and S treatments (40N + 30S, 80N + 30S, 120N + 30S, 40N + 60S, 80N + 60S, 120N + 60S). The results revealed that the application of 120N + 60S significantly improved key growth parameters such as plant height, grains per spike, spike density (spike/m²), and 1000-grain weight. This treatment also resulted in higher grain nitrogen content, N uptake, and protein levels, confirming its superiority over other treatments. Additionally, post-harvest soil analysis indicated increased mineral N and available S levels, while showing a slight decrease in pH and an increase in electrical conductivity (EC). In conclusion, the 120N + 60S combination was identified as the most effective treatment for maximizing wheat yield, improving grain quality, and enhancing soil nutrient availability. However, it is recommended that future studies validate these findings under field conditions, across different soil types and climates, to ensure broader applicability of 120N + 60S as a best practice for wheat cultivation.

Keywords: Wheat productivity, Nitrogen and sulfur fertilization, Grain protein content, Nutrient use efficiency (NUE), Greenhouse pot experiment.

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Introduction

Wheat (*Triticum aestivum*) is one of the most important staple crops globally, contributing significantly to food security and human nutrition (Shewry, 2009). Its production is critical, accounting for approximately 20% of the daily caloric intake worldwide, and providing substantial amounts of carbohydrates (55%) and proteins (8-12%). The quality and yield of wheat are influenced by a variety of factors, including environmental conditions, fertilizer management, and the genotype of the cultivated variety (Souza et al., 2004). Most wheat varieties are selected for traits such as milling performance, protein content, and baking

properties (Branlard et al., 2001; Gupta et al., 2021). Therefore, improving wheat growth, yield, and quality is essential for both agricultural productivity and the health benefits wheat provides to the global population (Shiferaw et al., 2013).

A critical factor in maximizing wheat yield and quality is Nutrient Use Efficiency (NUE), which refers to the plant's ability to absorb, store, utilize, and redistribute nutrients efficiently (Ghafoor et al., 2021). NUE is largely influenced by root architecture and growth, particularly in soils containing a mixture of organic and inorganic matter (Panhwar et al., 2019). Additionally, NUE is affected by external factors such as irrigation and fertilizer management. Among essential nutrients, nitrogen (N) plays a pivotal role in enhancing crop growth, as it is a fundamental component of chlorophyll, directly impacting photosynthesis and energy production (Khalofah et al., 2021). Research has shown that effective nitrogen management can significantly improve grain weight and yield, especially in cereal crops like wheat. Furthermore, nitrogen application has been positively correlated with increased protein content in wheat grains, which is a key quality attribute associated with nutritional benefits (Zhang et al., 2021; Alimbekova et al., 2022).

In addition to nitrogen, sulfur (S) is another essential nutrient crucial for plant growth and development. Sulfur is involved in the synthesis of amino acids, such as methionine and cysteine, which are vital for protein formation (Hell, 1997). Studies have shown that balanced sulfur application plays a key role in reducing oxidative stress and improving the plant's internal nutrient reduction mechanisms, ultimately leading to better productivity (Ragab and Saad-Allah, 2021). Sulfur has also been found to enhance the efficiency of nitrogen uptake and utilization, especially in the biosynthesis of protein, as nitrogen cannot optimally perform this function without sufficient sulfur (Wang et al., 2021). This makes sulfur an indispensable component for maximizing wheat grain quality and yield.

Despite the established importance of nitrogen and sulfur for wheat production, there is limited research on the optimal application rates of these nutrients when applied together. Most studies have focused on nitrogen or sulfur individually, but their combined effects on wheat productivity and grain quality remain underexplored. Given that nitrogen and sulfur exhibit a synergistic relationship, particularly in protein synthesis, understanding the best application rates for both nutrients is critical to enhancing wheat's agronomic performance.

Thus, the primary aim of this study is to investigate the combined effects of nitrogen and sulfur on wheat growth, yield, and protein content, with the goal of identifying the optimal application rates for both nutrients. By addressing the gap in the literature, this research seeks to provide valuable insights for improving wheat productivity through better nutrient management strategies. To achieve this, a pot experiment was conducted under controlled greenhouse conditions, using the Omskaya 18 variety of spring wheat, allowing for precise monitoring of plant growth, nutrient uptake, and environmental factors. This experiment serves as an initial step in understanding how nitrogen and sulfur interact to influence wheat performance, and the results will inform future field trials to assess the applicability of these findings under real-world agricultural conditions.

Material and Methods

Soil characteristics

This study was conducted under controlled greenhouse conditions using a randomized complete block design (RCBD) with three replications. The soil properties were determined according to the methods described by Rowell (1996) and Jones (2001). The experimental soil had a silty clay texture, consisting of 45% clay, 15% sand, and 40% silt. The other soil characteristics were as follows: organic matter content of 2.3%, pH (1:1) of 7.65, electrical conductivity (EC) of 0.86 dS/m, CaCO₃ content of 13%, total nitrogen (N) of 0.105%, available phosphorus (P) of 4.5 mg/kg, available sulfur (S) of 2176 mg/kg, and available potassium (K) of 195 mg/kg.

Experimental design and applications

Wheat (*Triticum aestivum*) variety Omskaya 18 was used as the test plant material. Each pot was filled with 5 kg of air-dried soil passed through a 4 mm sieve, and 15 wheat seeds were sown in each pot. The moisture content of the soil was maintained at field capacity throughout the experiment by weighing the pots daily and replacing the water lost through evapotranspiration. The effects of different nitrogen (N) and sulfur (S) levels on wheat growth were investigated. Nitrogen was applied in the form of urea (46% N), and sulfur was applied in the form of potassium sulfate (18% S). The experiment included 12 treatment combinations, with a total of 36 pots. The treatments were as follows:

1. Control (0N + 0S)
2. 40 kg/ha N (40N + 0S)
3. 80 kg/ha N (80N + 0S)
4. 120 kg/ha N (120N + 0S)
5. 30 kg/ha S (30S)
6. 40 kg/ha N + 30 kg/ha S (40N + 30S)
7. 80 kg/ha N + 30 kg/ha S (80N + 30S)
8. 120 kg/ha N + 30 kg/ha S (120N + 30S)
9. 60 kg/ha S (60S)
10. 40 kg/ha N + 60 kg/ha S (40N + 60S)
11. 80 kg/ha N + 60 kg/ha S (80N + 60S)
12. 120 kg/ha N + 60 kg/ha S (120N + 60S)

Plants were harvested after 85 days, when they reached physiological maturity. Soil and plant samples were collected for subsequent analysis.

Measurements and Analysis

Plant analyses: Plant height, number of grains per spike, spikes per square meter, 1000-grain weight, biological yield, and grain yield were determined using gravimetric and counting methods. The results were expressed in kg/ha based on the soil weight in the pots. Nitrogen (N) and sulfur (S) contents in wheat grains were analyzed using the method described by Jones (2001). N and S uptake by plants was calculated using the biological yield and the nutrient content of the plant tissues.

Soil analyses: After the harvest, soil samples were analyzed for pH, electrical conductivity (EC), mineral nitrogen (N), and available sulfur (S) contents using the method outlined by Rowell (1996) and Jones (2001).

Statistical analysis: The data were subjected to analysis of variance (ANOVA), and the means were compared using the least significant difference (LSD) test. Statistical analyses were performed using the SPSS software package.

Results

The application of nitrogen (N) and sulfur (S) significantly affected various growth parameters of wheat, including plant height, grains per spike, spike density (spike/m²), and 1000-grain weight, as summarized in Table 1. Statistical analysis indicated that nitrogen and sulfur had a significant impact on these parameters, with differences between treatments determined to be statistically significant ($p \leq 0.05$). Below is a detailed breakdown of each parameter:

Table 1. Effect of Nitrogen and Sulfur on Plant Height, Grains per Spike, Spike/m², and 1000-Grain Weight

Treatment	Plant Height (cm)	Grains/Spike	Spike/m ²	1000-Grain Weight (g)
0N + 0S	75.3 ± 2.1c	25.4 ± 1.5b	270 ± 15b	40.5 ± 1.2b
40N + 0S	80.1 ± 2.2bc	26.5 ± 1.6b	285 ± 16b	41.2 ± 1.1b
80N + 0S	85.7 ± 2.4b	27.8 ± 1.3ab	310 ± 20ab	42.8 ± 1.3b
120N + 0S	90.5 ± 2.6a	29.1 ± 1.2a	350 ± 18a	44.3 ± 1.4a
40N + 30S	83.9 ± 2.0bc	27.2 ± 1.4b	300 ± 17b	42.3 ± 1.1b
80N + 30S	88.6 ± 2.5ab	28.5 ± 1.3a	335 ± 19a	43.5 ± 1.2a
120N + 30S	92.1 ± 2.7a	29.6 ± 1.1a	360 ± 20a	45.0 ± 1.3a
40N + 60S	84.7 ± 2.3b	27.5 ± 1.4ab	305 ± 18b	42.7 ± 1.2b
80N + 60S	89.4 ± 2.4ab	28.9 ± 1.3a	340 ± 19a	44.0 ± 1.3a
120N + 60S	93.2 ± 2.9a	30.2 ± 1.1a	370 ± 21a	46.1 ± 1.4a

Note: Values followed by different letters (a, b, c) indicate significant differences between treatments at $p \leq 0.05$, based on Fisher's LSD test.

Plant height

Nitrogen and sulfur applications led to a notable increase in plant height, with the highest values observed in treatments where both nitrogen and sulfur were applied at higher rates. The 120N + 60S treatment produced the tallest plants, reaching 93.2 ± 2.9 cm, which was significantly greater compared to the control (75.3 ± 2.1 cm). Plant height increased consistently with higher nitrogen levels, and the addition of sulfur further enhanced this growth. For example, treatments such as 120N + 30S (92.1 ± 2.7 cm) and 80N + 60S (89.4 ± 2.4 cm) also showed significantly higher plant heights compared to both the control and lower nitrogen levels. These results indicate that the combination of nitrogen and sulfur is essential for maximizing plant height.

Grains per spike

The number of grains per spike was also significantly influenced by nitrogen and sulfur application. The highest grain count per spike was recorded in the 120N + 60S treatment (30.2 ± 1.1 grains), which was significantly higher than the control (25.4 ± 1.5 grains). Other treatments, such as 80N + 30S (28.5 ± 1.3 grains) and 120N + 30S (29.6 ± 1.1 grains), also exhibited significantly higher grain counts compared to the control. These results suggest that increasing nitrogen levels, particularly when combined with sulfur, promotes greater spike fertility, leading to a higher number of grains per spike.

Spike/m²

Spike density, measured as the number of spikes per square meter, was significantly affected by nitrogen and sulfur applications. The highest spike density was observed in the 120N + 60S treatment (370 ± 21 spikes/m²), which was significantly greater than the control (270 ± 15 spikes/m²). Similarly, other treatments with higher nitrogen and sulfur levels, such as 120N + 30S and 80N + 60S, also resulted in significantly higher spike densities compared to both the control and lower nitrogen treatments. The results demonstrate that the combined application of nitrogen and sulfur improves spike production, which directly contributes to increased grain yield.

1000-grain weight

The 1000-grain weight, an indicator of grain size and quality, was significantly increased by nitrogen and sulfur applications. The 120N + 60S treatment produced the highest 1000-grain weight (46.1 ± 1.4 g), significantly higher than the control (40.5 ± 1.2 g). Treatments with nitrogen alone, such as 120N + 0S (44.3 ± 1.4 g) and 80N + 0S (42.8 ± 1.3 g), also showed significant improvements in grain weight, though the addition of sulfur further enhanced grain size. Other treatments, such as 120N + 30S (45.0 ± 1.3 g) and 80N + 60S (44.0 ± 1.3 g), also resulted in substantial increases in 1000-grain weight, confirming the importance of sulfur in enhancing grain quality when applied alongside nitrogen.

In summary, the application of nitrogen and sulfur significantly improved all measured growth parameters. Higher rates of nitrogen, particularly when combined with sulfur, consistently led to improved plant height, grain count per spike, spike density, and 1000-grain weight. These results indicate that both nitrogen and sulfur play crucial roles in maximizing wheat growth and yield.

The effects of nitrogen (N) and sulfur (S) on biological yield and grain yield of wheat were significant, as shown in Table 2. The results indicate that both nitrogen and sulfur had a considerable influence on these yield parameters, with statistically significant differences observed between treatments ($p \leq 0.05$).

Table 2. Effect of Nitrogen and Sulfur on Biological Yield and Grain Yield

Treatment	Biological Yield (kg/ha)	Grain Yield (kg/ha)
0N + 0S	5600 ± 150c	2400 ± 100c
40N + 0S	6100 ± 160bc	2650 ± 110b
80N + 0S	6800 ± 170b	2900 ± 120b
120N + 0S	7500 ± 180a	3150 ± 130a
40N + 30S	6400 ± 165bc	2750 ± 115b
80N + 30S	7100 ± 175ab	3000 ± 125ab
120N + 30S	7800 ± 185a	3250 ± 135a
40N + 60S	6600 ± 170b	2850 ± 120b
80N + 60S	7300 ± 180a	3050 ± 130ab
120N + 60S	8000 ± 190a	3350 ± 140a

Note: Letters indicate significant differences ($p \leq 0.05$).

Biological yield

Biological yield, representing the total above-ground biomass, increased significantly with higher nitrogen and sulfur applications. The 120N + 60S treatment produced the highest biological yield, reaching 8000 ± 190 kg/ha, which was significantly greater than the control (5600 ± 150 kg/ha). Treatments with higher nitrogen levels, such as 120N + 30S (7800 ± 185 kg/ha) and 80N + 60S (7300 ± 180 kg/ha), also showed significant improvements in biological yield compared to lower nitrogen or sulfur applications. These findings suggest that the combined application of nitrogen and sulfur maximizes biomass production, with nitrogen playing a primary role, further enhanced by sulfur supplementation.

Grain yield

Grain yield, a key indicator of crop productivity, was similarly influenced by nitrogen and sulfur application. The highest grain yield was recorded in the 120N + 60S treatment (3350 ± 140 kg/ha), significantly higher than the control treatment (2400 ± 100 kg/ha). Higher nitrogen levels alone, such as 120N + 0S (3150 ± 130 kg/ha), resulted in increased grain yield, but the addition of sulfur at both moderate (30S) and high (60S) levels further enhanced the yield. For example, the 120N + 30S treatment resulted in 3250 ± 135 kg/ha, while 80N + 60S produced 3050 ± 130 kg/ha, both significantly higher than lower nitrogen and sulfur treatments.

Overall, the results demonstrate that higher nitrogen levels, especially when supplemented with sulfur, significantly improve both biological and grain yield. These findings highlight the essential role of nitrogen in

enhancing wheat productivity, with sulfur acting as a synergistic element that further boosts the yield potential of the crop.

The effects of nitrogen (N) and sulfur (S) on nitrogen and sulfur uptake in wheat plants were significant, as shown in Table 3. The statistical analysis confirmed that both nitrogen and sulfur levels had a substantial impact on grain nitrogen content, sulfur content, and the total nitrogen and sulfur uptake by the plants ($p \leq 0.05$).

Nitrogen content and uptake

Nitrogen content in the wheat grains increased significantly with higher nitrogen applications, particularly when combined with sulfur. The highest nitrogen content in the grains was recorded in the 120N + 60S treatment, with a value of $2.1 \pm 0.1\%$, which was significantly higher than the control ($1.3 \pm 0.1\%$). Similarly, nitrogen uptake by the plants was highest in the 120N + 60S treatment, reaching 55.4 ± 2.7 kg/ha, compared to the control (29.5 ± 2.0 kg/ha). Treatments with moderate nitrogen levels, such as 80N + 60S (48.3 ± 2.5 kg/ha) and 120N + 30S (52.7 ± 2.6 kg/ha), also showed significantly increased nitrogen uptake. These results indicate that higher nitrogen rates lead to increased nitrogen accumulation in the plants, with sulfur further enhancing nitrogen uptake efficiency.

Table 3. Nitrogen and Sulfur Uptake in Wheat Plants

Treatment	Grain N Content (%)	N Uptake (kg/ha)	Grain S Content (%)	S Uptake (kg/ha)
0N + 0S	1.3 \pm 0.1c	29.5 \pm 2.0c	0.12 \pm 0.02c	1.8 \pm 0.1c
40N + 0S	1.5 \pm 0.1bc	35.7 \pm 2.1bc	0.15 \pm 0.02bc	2.2 \pm 0.1bc
80N + 0S	1.7 \pm 0.1b	43.2 \pm 2.3b	0.17 \pm 0.02b	2.6 \pm 0.1b
120N + 0S	1.9 \pm 0.1a	50.6 \pm 2.5a	0.19 \pm 0.02a	3.0 \pm 0.1a
40N + 30S	1.6 \pm 0.1b	38.5 \pm 2.2b	0.18 \pm 0.02b	2.8 \pm 0.1b
80N + 30S	1.8 \pm 0.1ab	45.8 \pm 2.3ab	0.20 \pm 0.02ab	3.1 \pm 0.1ab
120N + 30S	2.0 \pm 0.1a	52.7 \pm 2.6a	0.22 \pm 0.02a	3.4 \pm 0.1a
40N + 60S	1.7 \pm 0.1b	41.2 \pm 2.2b	0.19 \pm 0.02b	2.9 \pm 0.1b
80N + 60S	1.9 \pm 0.1a	48.3 \pm 2.5a	0.21 \pm 0.02a	3.2 \pm 0.1a
120N + 60S	2.1 \pm 0.1a	55.4 \pm 2.7a	0.23 \pm 0.02a	3.6 \pm 0.1a

Note: Values followed by different letters (a, b, c) indicate significant differences ($p \leq 0.05$) based on Fisher's LSD test.

Sulfur content and uptake

Sulfur content in the wheat grains followed a similar trend, with significant increases observed in treatments with higher sulfur applications. The highest sulfur content was recorded in the 120N + 60S treatment ($0.23 \pm 0.02\%$), which was significantly higher than the control ($0.12 \pm 0.02\%$). Sulfur uptake also peaked in the 120N + 60S treatment, reaching 3.6 ± 0.1 kg/ha, compared to the control (1.8 ± 0.1 kg/ha). Other treatments with combined nitrogen and sulfur, such as 80N + 60S (3.2 ± 0.1 kg/ha) and 120N + 30S (3.4 ± 0.1 kg/ha), also demonstrated significantly higher sulfur uptake than the control or nitrogen-only treatments. These results highlight the synergistic effect of nitrogen and sulfur on sulfur uptake, with higher sulfur availability leading to increased absorption by the wheat plants.

In summary, the combined application of nitrogen and sulfur significantly increased both nitrogen and sulfur content in the grains, as well as the total uptake of these nutrients by the wheat plants. Higher nitrogen levels, particularly when paired with sulfur, resulted in greater nutrient accumulation, suggesting that sulfur enhances nitrogen use efficiency and contributes to optimal plant growth.

The effects of nitrogen (N) and sulfur (S) on soil properties after harvest, including mineral nitrogen (N), available sulfur (S), pH, and electrical conductivity (EC), are presented in Table 4. The results demonstrate significant changes in soil nutrient levels and properties due to varying nitrogen and sulfur applications ($p \leq 0.05$).

Mineral nitrogen (N)

Post-harvest mineral nitrogen levels in the soil increased significantly with higher nitrogen applications. The highest mineral N content was recorded in the 120N + 60S treatment, reaching 27.4 ± 0.9 mg/kg, compared to the control (12.1 ± 0.5 mg/kg). Treatments such as 120N + 0S (23.5 ± 0.8 mg/kg) and 80N + 60S (23.1 ± 0.8 mg/kg) also exhibited significantly higher mineral nitrogen levels compared to the control and lower nitrogen treatments. These results suggest that increasing nitrogen application leads to a buildup of residual mineral nitrogen in the soil, which can be beneficial for subsequent crops.

Table 4. Soil Properties Post-Harvest (Mineral N, Available S, pH, EC)

Treatment	Mineral N (mg/kg)	Available S (mg/kg)	pH	EC (dS/m)
0N + 0S	12.1 ± 0.5c	210 ± 15c	7.8 ± 0.1a	0.85 ± 0.05b
40N + 0S	16.4 ± 0.6bc	220 ± 16bc	7.7 ± 0.1a	0.88 ± 0.05b
80N + 0S	19.8 ± 0.7b	230 ± 16b	7.6 ± 0.1ab	0.91 ± 0.05b
120N + 0S	23.5 ± 0.8a	240 ± 17b	7.5 ± 0.1b	0.93 ± 0.05ab
40N + 30S	17.2 ± 0.6bc	250 ± 17ab	7.6 ± 0.1ab	0.92 ± 0.05ab
80N + 30S	21.5 ± 0.7ab	260 ± 18ab	7.5 ± 0.1b	0.95 ± 0.06ab
120N + 30S	25.6 ± 0.8a	270 ± 19a	7.4 ± 0.1b	0.98 ± 0.06a
40N + 60S	18.6 ± 0.7b	260 ± 18ab	7.5 ± 0.1b	0.94 ± 0.05ab
80N + 60S	23.1 ± 0.8a	280 ± 19a	7.4 ± 0.1b	0.99 ± 0.06a
120N + 60S	27.4 ± 0.9a	290 ± 20a	7.3 ± 0.1b	1.02 ± 0.07a

Note: Letters indicate significant differences ($p \leq 0.05$).

Available sulfur (S)

Available sulfur in the soil increased significantly with sulfur applications. The highest available sulfur content was observed in the 120N + 60S treatment (290 ± 20 mg/kg), which was significantly higher than the control (210 ± 15 mg/kg). Other sulfur-supplemented treatments, such as 80N + 60S (280 ± 19 mg/kg) and 120N + 30S (270 ± 19 mg/kg), also showed substantial increases in available sulfur levels compared to nitrogen-only treatments and the control. These results confirm that sulfur applications not only improve plant sulfur uptake but also enhance sulfur availability in the soil after harvest.

Soil pH

Soil pH values showed slight but significant changes following nitrogen and sulfur applications. The control treatment had the highest pH value (7.8 ± 0.1), whereas the pH levels slightly decreased with increasing nitrogen and sulfur applications. The lowest pH value was recorded in the 120N + 60S treatment (7.3 ± 0.1), indicating that higher nitrogen and sulfur applications tend to slightly acidify the soil. However, these changes in pH remain within the neutral to slightly alkaline range, suggesting that nitrogen and sulfur applications did not drastically affect soil acidity.

Electrical conductivity (EC)

Electrical conductivity (EC), an indicator of soil salinity, increased with higher nitrogen and sulfur applications. The highest EC was observed in the 120N + 60S treatment (1.02 ± 0.07 dS/m), which was significantly higher than the control (0.85 ± 0.05 dS/m). Treatments such as 80N + 60S (0.99 ± 0.06 dS/m) and 120N + 30S (0.98 ± 0.06 dS/m) also showed increased EC values compared to lower nitrogen or sulfur applications. The increase in EC is likely due to the addition of nitrogen and sulfur, as these nutrients contribute to the ionic content of the soil.

Discussion

The results of this study clearly demonstrate that the combined application of nitrogen (N) and sulfur (S) significantly enhances wheat growth, yield attributes, and nutrient uptake. The increase in growth and yield characteristics is largely attributable to improved nitrogen and sulfur uptake by the plants. These findings are consistent with previous research, which highlights the critical role of N and S in promoting plant growth, optimizing nitrogen use efficiency, and enhancing protein synthesis in grains.

Nitrogen and sulfur effects on growth and yield

The significant improvement in wheat growth and yield observed in this study can be attributed to better uptake of nitrogen and sulfur, particularly in treatments with higher N and S application rates. The combined application of 120N + 60S led to the highest plant height, spike density, grain yield, and biological yield. This enhancement in growth is likely due to the role of nitrogen in improving leaf surface area and chlorophyll synthesis, which optimizes the photosynthetic process and provides the energy needed for plant growth. Zecevic et al. (2010) noted that nitrogen facilitates cell division, which plays a crucial role in shoot elongation and overall plant height, similar to the findings of this study.

Sulfur, when applied in conjunction with nitrogen, plays a synergistic role in enhancing wheat productivity by improving nitrogen use efficiency (Haneklaus et al., 1999). In the present study, treatments with combined N and S resulted in a significant improvement in grain number per spike and 1000-grain weight, likely due to the favorable environment created for tillering and grain filling. Assefa et al. (2021) also reported similar results, noting that sulfur application improved both grain and straw yields by optimizing nutrient availability. Moreover, Šiaudinis and Lazauskas (2005) found that higher nitrogen doses increased

biological yield and altered vegetative growth characteristics, which aligns with the increase in biological yield observed in this study.

Protein content and nutrient uptake

In addition to improving growth and yield, the application of nitrogen and sulfur significantly increased the protein content in wheat grains. This is in agreement with previous studies, such as those by [Wilson et al. \(2020\)](#) and [Liimatainen et al. \(2022\)](#), who reported that sulfur application enhances the synthesis of amino acids and proteins, leading to higher protein content in grains and leaves. The increase in protein content observed in the 120N + 60S and 120N + 30S treatments in this study can be attributed to sulfur's involvement in the formation of amino acids and its role in activating enzymes that stimulate the production of proteins and vitamins essential for plant metabolism ([Haneklaus et al., 1999](#)).

The synergistic interaction between nitrogen and sulfur is well documented in the literature, and this study further confirms the positive effect of S on N uptake. [Khandkar and Shinde \(1991\)](#) demonstrated that the combined effect of S and N fertilizers significantly improved nitrogen uptake in maize plants, a result that is consistent with the findings of this study, where sulfur supplementation significantly enhanced nitrogen assimilation in wheat. The improvement in nitrogen uptake is critical for promoting protein synthesis, as nitrogen is a key component of amino acids, the building blocks of proteins. As [Garrido-Lestache et al. \(2005\)](#) pointed out, the increase in nitrogen supply is strongly correlated with higher protein content, although the effect on protein quality also depends on the wheat variety used, due to differences in nitrogen utilization.

Soil properties post-harvest

The post-harvest soil analysis in this study revealed important changes in soil nutrient levels due to nitrogen and sulfur applications. The increase in residual mineral nitrogen in the soil after high nitrogen treatments, particularly when combined with sulfur, indicates that these nutrients can have a lasting impact on soil fertility. [Fuentes et al. \(2003\)](#), [İslamzade et al. \(2023; 2024\)](#) and [Kaliyeva et al. \(2024\)](#) reported similar findings, noting that residual nitrogen in the soil could benefit subsequent cropping cycles by maintaining higher nutrient availability.

Additionally, the slight decrease in soil pH with increasing nitrogen and sulfur rates aligns with the findings of [Barak et al. \(1997\)](#), who observed soil acidification due to nitrogen fertilization. The increase in electrical conductivity (EC) observed in this study is likely a result of the accumulation of salts from the fertilizers, as noted by [Choudhary et al. \(2011\)](#), and suggests that while the nutrient additions improved plant growth and nutrient uptake, there may be long-term implications for soil salinity that require monitoring in future seasons.

Practical implications for fertilizer management

The findings of this study underscore the importance of balanced nitrogen and sulfur fertilization to maximize wheat productivity. The combined application of 120 kg/ha nitrogen with 60 kg/ha sulfur was particularly effective in enhancing growth, yield, and protein content, indicating that sulfur plays a critical role in complementing nitrogen fertilization by improving nitrogen use efficiency and promoting protein synthesis. This is consistent with the broader body of literature that highlights sulfur's importance in nitrogen metabolism and amino acid formation, leading to improved grain yield and quality ([Carciochi et al., 2020](#); [Wilson et al., 2020](#)).

In terms of practical application, these results suggest that sulfur should be included in fertilizer programs, especially in soils where sulfur is deficient or where high rates of nitrogen are applied. The improvement in yield and protein content observed with combined N and S application suggests that wheat farmers can achieve higher productivity and better grain quality by incorporating both nutrients into their fertilization strategies. However, as the slight increase in soil salinity (EC) suggests, continued monitoring of soil conditions is essential to ensure long-term soil health and sustainability.

Conclusion

This pot experiment demonstrated the significant impact of nitrogen (N) and sulfur (S) on wheat growth, yield attributes, and grain protein content. The combined application of 120 kg/ha nitrogen and 60 kg/ha sulfur (120N + 60S) proved to be the most effective treatment, leading to substantial improvements in plant height, spike density, grain number per spike, biological yield, and nutrient uptake. The results indicate that the 120N + 60S combination is a balanced and optimal fertilization strategy for maximizing wheat productivity and enhancing grain quality. However, it is important to note that these findings were obtained under controlled pot experiment conditions. To fully validate the efficacy of the 120N + 60S treatment under real-world agricultural scenarios, further field trials are necessary. Conducting field experiments across

different wheat cultivars, soil types, and environmental conditions will provide more comprehensive insights into how nitrogen and sulfur interactions influence wheat growth in larger-scale farming systems. Wheat growers are encouraged to consider the 120N + 60S combination for improving yield and protein content, but it is also recommended that additional research be undertaken to bridge the gap between pot experiment results and field conditions. Such trials will help determine the most effective nutrient management strategies for achieving maximum crop productivity under variable and dynamic field conditions.

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