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IMPACT OF NITROGEN FERTILIZER MANAGEMENT ON DISTRIBUTION OF NITROGEN CONTENT IN DIFFERENT ORGANS OF WINTER WHEAT

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## **Article Info**





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Abstract However, increasing the rates and adjusting the time of nitrogen treatment might worsen nitrogen loss and lower grain yields, which will have a detrimental effect on the quality of the environment. The relationship between nitrogen nutrition and grain production has been extensively studied, but much has been discovered about how nitrogen concentration in winter wheat, higher up in components like leaves, stems, and grains, reacts to nitrogen nutrition. At Shanxi Agricultural University's Taigu Experimental Agricultural Centre in Shanxi Province, China, experiments in the field were carried out. Three repetitions of these split-plot tests were conducted. In the main plots, four nitrogen application levels—75, 150, 225, and 300 kg ha-1 were tested. Different nitrogen levels were used in the subplots, including 5:5 (50%, 50%) and 6:4 (60%, 40%). Nitrogen fertilizer was applied at pre-sowing, jointing, flowering, and grain filling stages, respectively. The 12  $m^2$  (3  $m \times 4$ m) experimental plots were used. As nitrogen fertilizer rates increased, there was a considerable increase in the total nitrogen content of the dry matter. In comparison to other growing phases and nitrogen rates, plants got nitrogen nutrition at 60% during sowing and 40% during the jointing stage, increasing the average total nitrogen content from 40.00 to 60.00% under 225 kg N ha-1. Less than 225 kg N ha-1 at the jointing stage, when nitrogen was given in 6:4 ratios, the maximum nitrogen content in leaves was recorded at 32-34 g kg-1. Similarly, at 225 kg N ha-1, the maximum nitrogen contents in the stem were measured at 13-15 g kg-1, meaning that plants absorbed 60% of their nitrogen nutrition during sowing and 40% during the jointing stage. 225 kg ha-1 of nitrogen, applied in the following proportions: 60%, 40%, 0%, and 0% (amount applied at the sowing, jointing, blooming, and grain filling stages) successfully increased the nitrogen content of winter wheat's leaves, stems, and grains above ground.

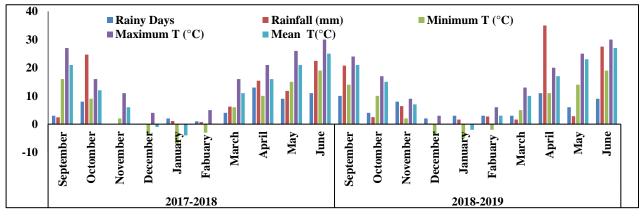
#### Introduction

Nitrogen is the essential component in achieving constantly high yields in cereals. It is complex in all of the plant's metabolic developments, its proportion of uptake and partition being mainly determined by amount and demand during the several stages of plant development. Soil nitrogen supply, for illustration, must be high at shoot elongation, jointing, flowering, and grain filling, requiring a better amount for the development and progress of its preparative organs and for an improved and high accretion of proteins in the spikes (Ranjan et al., 2019; Tolley et al., 2019; Moreira et al., 2017 Baloch et al., 2023 and Nageeb et al., 2024). Several different methods have been recommended to increase crop NUE, such as using the best amount, application period, and application technique for matching nitrogen supply with plant demand. Proportions of nitrogen fertilizer and application timing are crucial factors in the gaining of high yields (Wang et al., 2017; Hafiz et al., 2011). According to Yang et al., 2014, Munoz et al., 2013, and Kaleri et al., 2024. the nitrogen nutrition index is a plant-based diagnostic technique that may be used to assess a crop's nitrogen status during the growth season and, consequently, identify any areas that require more nitrogen fertilization. When the developing period comes to an end, the nitrogen nutrition concentration can also be utilized to evaluate and interpret the agronomic performance of crops. Overall, plant development rate increases around linearly with the absorptions of maximum nutrient components in the plant tissues up to an ideal nutrient absorption elsewhere, where a further increase in the element absorption is not accompanied by improved plant growth. It is frequently assumed that changes in the variation in tissue component absorptions are closely reflected by the changes in nitrogen concentration, provided nitrogen (and not other elements) is the most limiting factor for plant development. However, this common possibility in nutrient examination has been rarely tested, particularly for agricultural crops (Ort et al., 2010; Zhu et al., 2008 and Javed et al., 2024). Determining the assessment of the nitrogen status of the plant might be done with the help of leaf diagnostics, which is established on the association among nitrogen concentration and chlorophyll in the plant leaves (Yuan et al., 2016; Ali et al., 2017; Roll et al., 2018 and Ahmed et al., 2023). During grain filling, the nitrogen accumulated in the vegetative organs is redistributed to the ear. Various modifications, such as nitrogen fertilization, shading, or ear removal, can alter the nitrogen content in wheat grains.

#### 2. Materials and methods

#### 2.1. Experimental locations

At the Taigu Experimental Agricultural Station of Shanxi Agricultural University, which is situated in Shanxi Province, China (N 37°25', E 112°33'), field trials were carried out in the 2017–2018 and 2018–2019 seasons. With average annual temperatures of 13°C and 12°C, average annual rainfall of 442 mm and 600 mm, potential evapotranspiration of 1840.2 mm and 1872.2 mm, and sunshine hours of 2672 and 2697, respectively, the research region has moderate continental monsoon weather. The research region has a semiarid climate, with 60% to 70% of the annual rainfall falling between July and August. It is distinguished by a mountainous and dry environment typical of the Northeast Loess Plateau. The cultivated field's soil has a pH of 7.7, 51.12 mg kg<sup>-1</sup> of available nitrogen, 19.34 mg kg<sup>-1</sup> of available phosphorus, and 7.7 mg kg<sup>-1</sup> of surface organic matter. The minimum, maximum, and average temperatures are shown in the weather data (Figure 2.1), together with the mean monthly precipitation and the number of days with rainfall.



# Figure 2.1: In Shanxi Agricultural University, monthly rainy days, rainfall, minimum and maximum temperatures, and mean temperatures from 2017–2018 to 2018–2019.

#### 2.2. Treatment detail

Three repetitions of the trials were used to arrange them in a split-plot design. Total nitrogen (N) levels with 75, 150, 225, and 300 kg N ha-1 were the main plots. Two types of nitrogen ratio subplots were present: a 5:5 (50%, 50%) ratio and a 6:4 (60%, 40%) ratio. The nitrogen ratios were arranged in the following labels: 50%:50%:0% 0% (labelled as 5+5), 50%:0%:0%:0% (labelled as 5+5), and 50%:0%:0%:50% (labelled as 5+5). Two methods of applying nitrogen fertilizer were used to treat winter wheat at distinct growth stages: 50% sowing time + 50% at jointing, 50% sowing time + 50% at flowering, and 50% sowing time + 50% at grain filling. The nitrogen fertilizer was applied during the base period and top dressing. Additionally, there is a second method that involves sowing 60% of the seed at sowing time and 40% at the jointing stage, flowering 60% of the seed at sowing time and 40% at the grain filling stage, and so on. Each treatment comprised three replications, and the experimental plots measured  $12 \text{ m}^2$  (3 m × 4 m). In the 72 total plots, urea (46.4%) was applied as the nitrogen source for the experimental field prior to crop sowing, phosphorus (16%) was applied as triple super phosphate at a rate of 120 kg ha-1, and potassium (45%) was applied as potassium chloride at a rate of 60 kg ha-1 during the sowing period. In the experimental investigation, winter wheat (variety Jintai 182) was cultivated prior to seeding, with a sowing rate of 95 kg ha-1. Plants for winter wheat were harvested between June 15 and June 21, 2019, after being sown on September 31, 2017, and October 1, 2018. Data gathered during a 20–25 day field break in the months of March, April, and May. Using Shanxi province's traditional methods, all other agricultural operations, including weed management, irrigation, disease and pesticide application, were completed equally and on schedule depending on the crop's growth stage and need.

#### 2.2. Sampling, chemical analysis

Winter wheat plants at the jointing, blooming, and grain filling stages were sampled every 20 cm in order to calculate the dry matter from each treatment and the nitrogen content of the above-ground biomass. Utilizing sulfuric acid ( $H_2SO_4$ ) and hydrogen peroxide ( $H_2O_2$ ), a 0.25 g sample derived from the spike grain, stem, and leaf was processed and digested. Subsequently, nitrogen was determined using the Kjeldahl method (Derocher et al., 1993; Christians et al., 1991). The nitrogen content of the several organs of winter wheat plants was determined in grams per hectare by multiplying the nitrogen content of each organ by the total dry matter of the organ.

## 2.3. Statistical Analysis

The average of three replicates served as the data source for this investigation. ANOVA was used to analyze all of the data for a randomized block design. Each source's significance was ascertained using the F-test. Using SAS 9.3 (SAS Institute, Cary, North Carolina, USA), the significant difference from the Duncan's Multiple Range Test (DMRT) was employed as a post hoc mean separation test (P < 0.05). The least significant difference (LSD P < 0.05) was used to compare the treatments based on significant differences. Prior to evaluating the ANOVA, the normality of variance was evaluated using the Shapiro-Wilk test. For data calculation, Microsoft Excel 2013 was utilized. SAS version 9.3 and SPSS version 19.0 were used for all statistical analysis.

## 3. Results and analysis

## 3.1. Plant nitrogen contents as affected by N ratios and N timing

Higher nitrogen treatment rates resulted in a considerable increase in the total nitrogen content of the above-ground biomass (AGB) in dry matter (Fig. 3.1). The average nitrogen content in AGB was 84.75%, 110.85%, and 79.47% in the 2017–2018 season, and 91.10%, 90.97%, and 60.31% in the 2018–2019 season throughout different winter wheat growth stages. These results were obtained using a nitrogen ratio of 6:4 (225 kg N ha<sup>-1</sup>) administered during the jointing stage as opposed to 5:5 (nitrogen applied at the flowering and grain filling stages). In both years, the amount of nitrogen in above-ground biomass rose from the jointing to the grain filling stage; the maximum values were recorded at 225 kg N ha<sup>-1</sup> with a 6:4 ratio. When nitrogen was carried out at the jointing stage, it increased the nitrogen content to 33.29 in the 2017–2018 season and 31.25 in the 2018–2019 season, outperforming the 150 and 300 kg N ha<sup>-1</sup> treatments. When the 225 kg N ha<sup>-1</sup> treatment was applied, the nitrogen levels in the above-ground biomass were kept constant until the beginning of the flowering stage in both years. Furthermore, nitrogen content

levels surpassing 75 were observed in both years in the treatments with 150 and 300 kg N ha<sup>-1</sup> and nitrogen ratios of 5:5 and 6:4. Plant nitrogen content was found to be significantly impacted by both the rate and ratio of nitrogen application, as well as by the significant interaction between nitrogen treatments and ratios, according to variance analysis (Table 3.1). Based on dry matter, these results imply that nitrogen treatment efficiently raises the nitrogen content specifically in plant parts, such as the stem or leaf. These organs' pattern of nitrogen buildup is identical to the dry matter pattern of the whole plant. However, the ageing of the leaves, the increase in physical leaf tissue, and the shadowing impact of young leaves all contribute to the decrease in leaf nitrogen content.

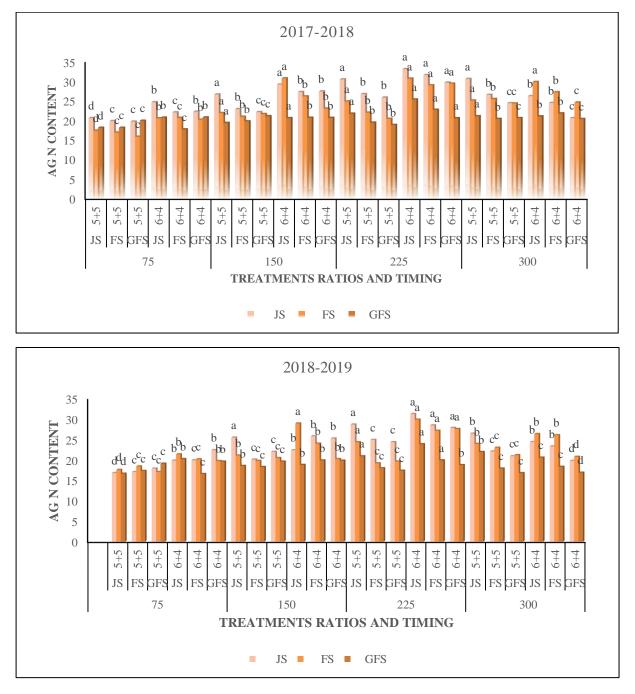
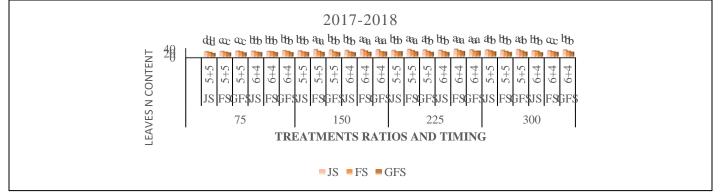


Figure 3.1 shows how nitrogen fertiliser management affects winter wheat's above-

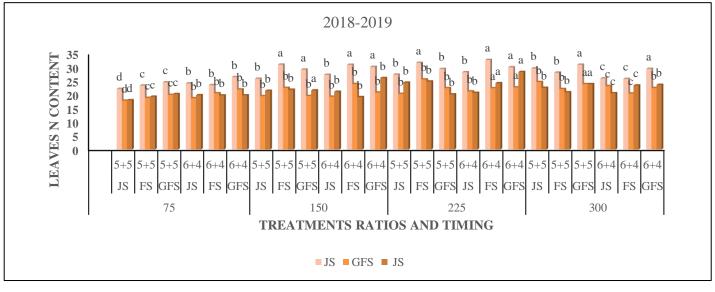
ground N content (g kg1). "5+5 and 6+4" stands for 50% + 50% and 60% + 40%, respectively. JS stands for jointin g stage, FS for flowering stage, and GFS for grain filling stage. The means in the separate columns that are separate d by similar letters do not differ significantly at p <0.05.

## 3.2. Leaf nitrogen contents as affected by N ratios and N timing



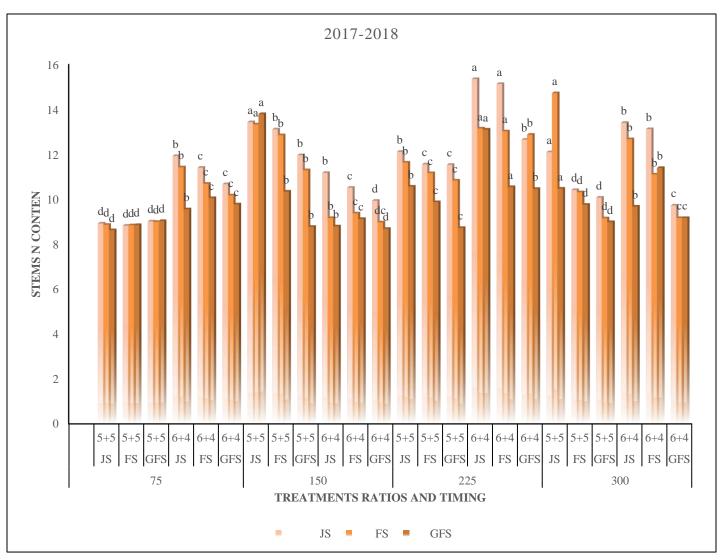
As nitrogen application increased, the amount of nitrogen in leaves increased significantly (Fig. 3.2). In both years, the total nitrogen concentration in leaves varied from 43.52% to 47.79% at the jointing stage, 63.52% to 57.34% at the grain filing stage, and 37.10% to 43.55% at the flowering stage under the ratio 5:5 when 225 kg N ha-1 was applied. The 6:4 ratio produced the best results, and the treatment of 225 kg Nha-1 varied from 34.82 to 32.75 g kg-1 when the nitrogen was introduced during the jointing stage. varied from 28.70 to 25.63 in the flowering stage when treated in the same way, but with a 5:5 ratio and jointing stage application period. Under the same conditions and a 6:4 ratio, the results at the grain filling stage varied between 26.34 and 28.34 when nitrogen was applied as amounts of 150 and 300 kg N ha-1 during the flowering stage in both years. (Figure 3.2). At the jointing and flowering stages, our results from 2017–2018 are higher than the decrees from 2018–2019; however, at the grain filling stage, the decrees from 2018–2019 are higher than the results from 2017–2018 while treating 225 kg N ha^1 at a ratio of 6:4 and applying the treatment at the flowering stage. Additionally, the minimum results for the treatment of 150 kg N ha-1 and 300 kg N ha-1 under the 5:5 ratio were 33.26 to 31.03 and 32.95 to 30.99. The nitrogen was applied at the jointing and flowering stage in both years as a dose of 75 kg ha-1. According to the variance analysis results, the nitrogen treatment had a substantial impact on the amount of nitrogen in the leaves, and there was also a significant interaction between the nitrogen treatments and ratio (Table 3.1). According to our findings, adding nitrogen fertilizer at the time of planting was beneficial for raising the study's leaf nitrogen content. It has long been known that the growth of roots and shoots during the nutrient-achievement phase plays a crucial role in determining winter wheat plants' capacity to absorb and enable the easy availability of energy-rich nutrients in the soil.

Figure 3.2 shows how winter wheat leaf nitrogen content (g kg-1) is affected by nitrogen fertilizer management. 50% + 50% and 60% + 40% are represented by "5+5 and 6+4".FS stands for flowering, JS for jointing, and GFS for grain filling. The identical letters are followed by the means values in separate columns, which do not differ substantially at p <0.05.

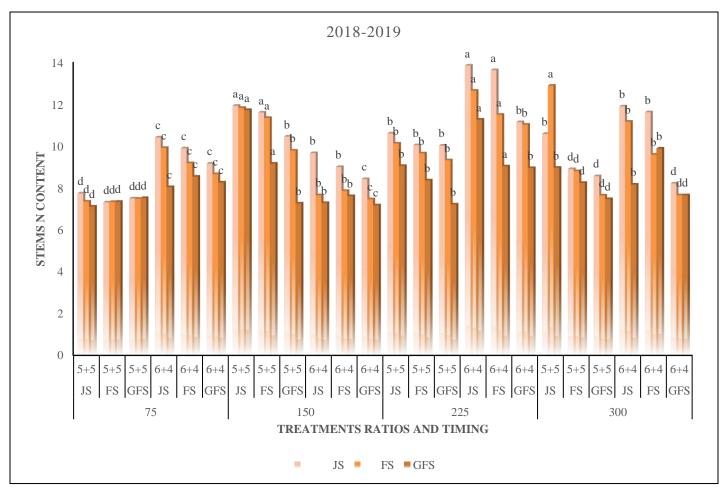


#### 3.3. Stem nitrogen contents as affected by N ratios and N timing

While stem nitrogen content in winter wheat decreased over the growth period in the second year, it increased in the first year from the jointing to grain filling stage (Fig. 3.3). The average total nitrogen content in stems increased from 87.14% to 106.65% at the jointing stage under the 225 kg N ha<sup>-1</sup> treatment with a 6:4 ratio when nitrogen was applied at the jointing stage in both years compared to the 75 kg N ha<sup>-1</sup> treatment. Under the 300 kg N ha<sup>-1</sup> treatment with a 5:5 ratio, it increased from 108.56% to 131.85% at the flowering stage. Similarly, with the 150 kg N ha<sup>-1</sup> treatment, it developed from 107.03% to 113.24% during the grain filling stage, with an overall 5:5 ratio. The highest results were recorded at 15.35 and 13.85 during the jointing stage with the 225 kg N ha<sup>-1</sup> treatment at a 6:4 ratio when nitrogen was applied at the jointing stage in both years, compared to the 150 and 300 kg N ha<sup>-1</sup> treatments. However, for both years, the results for the 225 kg N ha<sup>-1</sup> treatment decreased from the flowering to the grain filling stages. When compared to the 75 kg N ha<sup>-1</sup> treatment at the jointing stage in both years, the highest values were seen during the flowering and grain filling stages: 14.72–12.89 at the flowering stage and 13.79–11.73 under the 150 and 300 kg N ha<sup>-1</sup> treatments with a 5:5 ratio (Fig. 3.3). The 75 kg N ha<sup>-1</sup> treatment with the 5:5 and 6:4 ratios yielded the least beneficial outcomes overall. The probability levels for the differences between the treatments were 0.05. The results of the analysis of variance indicated that the nitrogen level and ratio had a substantial variable effect on the amount of nitrogen in stem cells, and that there was a significant interaction between the two (Table 3.1). It was noted that during the reproductive growth period, winter wheat is frequently affected by high temperatures and hot or dry air, which causes winter wheat to mature earlier than usual. Grown in conditions other than these, winter wheat experiences a reduced canopy of photosynthesis and developed nitrogen metabolism in the plant. Figure 3.3 shows

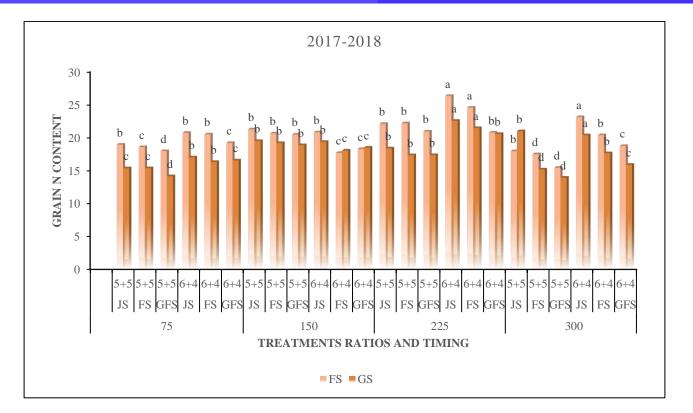


how winter wheat's spike nitrogen contents (g kg-1) are affected by nitrogen fertilizer treatment. 50% + 50% and 60% + 40% are represented by "5+5 and 6+4".FS stands for flowering, JS for jointing, and GFS for grain filling. The identical letters are followed by the means values in separate columns, which do not differ substantially at p <0.05.



## 3.4. Spike nitrogen contents as affected by N ratios and N timing

During the reproductive phase of the growing period, spike nitrogen contend dry matter was positively impacted by the nitrogen application rates and ratios (Fig. 3.4). spike nitrogen content varied from 75 kg N ha-1 to 13.33 and 225 kg N ha-1 to 26.33 in g kg based on treatments, ratios, application timing, sample date, and growth season. Spike nitrogen content increased gradually with the rising proportion of nitrogen rate. When nitrogen was applied at the jointing stage rather than the flowering and grain filling stages, the average nitrogen content in the spike was 83.90% and 52.09% at the flowering stage and 93.71% and 57.95% at the grain filling stage of winter wheat under the treatment of 225 kg N ha-1 under the ratio of 6:4 as camper of 5:5 ratio. Furthermore, when comparing 150 and 300 kg N ha-1, the highest results were found at the treatment of 225 kg ha-1 under ratio 6:4, which ranged from 26.33 to 24.83 (g kg-1) at the blooming stage and 22.56 to 21.06 g kg at the grain filling stage in both years. Additionally, in both years, the under treatment of 75 kg N ha-1 produced the lowest outcomes. The results of the analysis of variance indicated that the Spike N content was influenced by the nitrogen level at the 0.01 or 0.05 probability levels. The F values for the N treatments, ratio, and interaction of treatments and ratio were 294.67\*\*\*, 34.60\*\*\*, and 11.43\*\*\*, respectively (Table 3.1). These findings show that, in settings where nitrogen growth is modified, spike nitrogen content can be a powerful diagnostic tool for determining the nitrogen status of winter wheat.



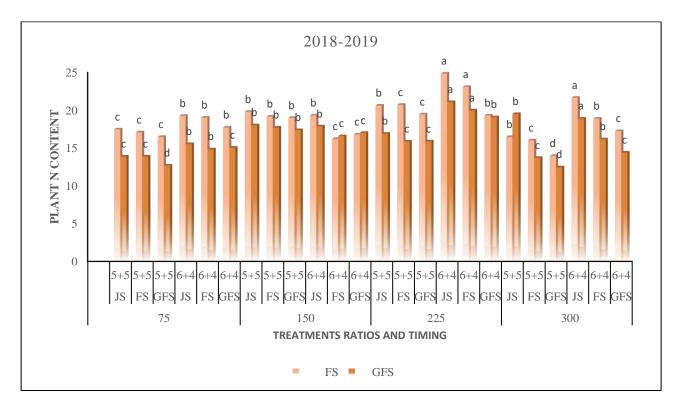


Figure 3.4 shows how nitrogen fertilizer management affects the amount of nitrogen in winter wheat spikes (g kg-1). 50% + 50% and 60% + 40% are represented by "5+5 and 6+4".FS stands for flowering, JS for jointing, and GFS for grain filling. The identical letters are followed by the means values in separate columns, which do not differ substantially at p <0.05.

Table 3.1 Significance of the F-value from the analysis of variance of different winter wheat parameters in relation to nitrogen management

Parameter	N-rates (N)	Ratios (R)	$\mathbf{N}  imes \mathbf{R}$
Plant N content	70.24***	5.83**	2.07 *
Leaf N content	161.38 ***	6.19***	5.20***
Stem N content	67.70***	10.29***	5.36***
Spike N content	294.67***	34.60***	11.43***

Note: The honestly significant difference (HSD) test yielded significance levels at alpha 0.05, 0.01, and 0.001 represented by the symbols \*, \*\*, and \*\*\*.

### 4. Discussion

#### 4.1. Effect of nitrogen fertilizer on plant nitrogen contents

Treatments and ratios had significantly increased N content in the whole plant of above-ground biomass at the jointing stage, and similar treatments and ratios impacts were found among the winter wheat growing periods of 2017-18 and 2018-19 (Fig. 3.1). The average nitrogen content in winter wheat organs (plant above-ground biomass, spike, stem, and leaf) was different among the jointing, flowering, and grain-filling stages. Our results match with Peltonen et al. (2007), who reported that the highest total nitrogen content in aboveground biomass greater than 28 g kg<sup>-1</sup> was observed at the flowering stage of the wheat crop in Finland. Instead, the nitrogen absorbed and stored through jointing stage and flowering stage was mostly complex in the nitrogen metabolism as mobile elements, which are remobilized to the growing grain through the reproductive stage (Ivanov et al., 2013). The average of total nitrogen content in the plant's aboveground biomass during the jointing stage was significantly greater than those during the flowering and grain filling stages. While an opposite trend was found in the total nitrogen content in the aboveground parts in second year.

The nitrogen content of above-ground increase during jointing and flowering stage was 84.75% and 110.85% and 91.10% and 90.97% higher than that grain-filling stage across the two growing seasons in the treatment of 225 kg N ha<sup>-1</sup> under the ratio of 6:4 plots, respectively. Sanchez et al. (2017) described that the root, stem, and leaves contribute 16%, 23%, and 40%, respectively, to the daily amount of grain nitrogen accumulation in wheat at mid-grain filling. The maximum results were obtained at 225 kg N ha<sup>-1</sup> under ratio 6:4 ratio as compared to 5:5 ratio from 33.29 and 31.25 at jointing stage in both years when nitrogen was applied at timing of jointing stage as compared to the flowering and grain-filling stage. Chen et al. (2019) informed us that maximum total nitrogen content in different organs of winter wheat ranged from 24 to 32 in above-ground biomass, from 16 to 20 in leaves, from 10 to 15 in stems, and from 20 to 25 in grains in g/kg were recorded (Chen et al., 2019; Jin et al., 2015). Jones et al. reported that highest total nitrogen content in aboveground biomass ranged from 25-33 g kg<sup>-1</sup> at the jointing stage of wheat crop in USA (Umara et al., 2019). Scientist Nikolic et al. (2012) originate that nitrogen concentration in the aboveground part of the plant is expressed very strongly and has a direct positive impact on wheat yield, which frequently agrees with our results.

As nitrogen application increased, the amount of nitrogen in leaves increased dramatically (Fig. 3.2). The 6:4 ratio produced the best results, and the treatment of 225 kg N ha-1 varied from 34.82 to 32.75 (g kg-1) when the nitrogen fertilizer was added at the jointing stage. varied from 28.70 to 25.63 in the flowering stage when treated in the same way, but with a 5:5 ratio and jointing stage application period. When nitrogen was applied as a mixture of 150 and 300 kg N ha-1 during the flowering stage in both years, the results at the grain filling stage varied from 26.34 to 28.34

under the same treatment and 6:4 ratio, respectively. Our findings corroborate those of Yan et al., who found that the greatest total nitrogen content in leaves ranged from 30 to 38 (g kg-1) and were seen throughout the Finnish wheat crop's jointing to flowering stage (Yan et al., 2022). The high leaf nitrogen contents found while applying 150 to 200 kg N ha-1 nitrogen levels are highlighted by Yadav et al., 2023; Lu et al., 2015; these nitrogen contents are significantly higher than the typical range of 20 to 34 (g kg-1) dry matter. Furthermore, minimal results in our study were observed in the jointing and flowering stage in both years as camper of 75 kg-1 ha-1 and 33.26 to 31.03 and 32.95 to 30.99 at the treatment of 150 kg ha-1 and 300 kg N ha-1 under the ratio of 5:5. According to You et al. (2023), this happened as a result of applying nitrogen as a side dressing closer to the wheat crop's blooming time, when the flag leaf was collected to measure the nitrogen content of the leaf. According to our findings, fertilizing with nitrogen at the time of seeding promoted the growth of the nitrogen content in the leaves in our investigation. Our findings were in line with those of Bashir et al. (2017) and Kubar et al. (2022), who found that nitrogen fertilizer positively impacted the concentration of nitrogen in young plants during their primary growth stage. In our study, a high dose of nitrogen fertilizer impeded root development and activity, which in turn decreased the amount of nitrogen absorbed and absorbed during the growing stage. Our findings are consistent with the prior research carried out by Mariano et al. (2015), who found that, especially in soil with less accessible nitrogen, a specific amount of nitrogen fertilizer was beneficial to root growth and greater nitrogen uptake. The important data on stem nitrogen concentration are provided by our field study (Fig. 3.3). One of the main elements in crops' ability to produce biomass above ground is an increase in nitrogen content. The consistent concentrations for the stems were 106.65%, 127.49%, and 104.83% higher (P < 0.05) and 87.14%, 86.24%, and 96.68%. However, at levels of 150 and 300 kg N ha-1, treatments and ratios considerably improve N concentrations by an average of 86.24 to 127% and 96.68 to 104% in both years at the flowering and grain filling stage. This finding demonstrates how nitrogen deposited in the stems affects the grain's ability to store nitrogen, which is increased during the last stages of vegetative growth before flowering. Rahimi et al.'s 2021 study, which found that stem N content increased significantly at the flowering stage, ranging from 34.4% to 47.1%, and at grain filling, increased from 33.4% to 60.6%, supported our findings and suggested that stem N contributed more to wheat grain quality. According to a 2004 study by Liao et al., the greatest total nitrogen level in stems ranged from 6 to 10 (g kg-1) throughout the Australian wheat crop's tillering to jointing stage. At the jointing periods of winter wheat, as camper of 150 and 300 kg N ha-1 in 207-18 15.35, the maximum nitrogen content was reached in 225 kg-1 ha-1 under the 6:4 ratio as camper of 5:5 ratio in 2018-19 13.85. The nitrogen content of the stem was found to be significantly improved with increasing nitrogen application, according to studies by Fang et al. (2022) and Ma et al. (2016). At low nitrogen levels, 180 kg N ha-1, the basal and top rates of fertilizer were reduced when the amount of nitrogen application was increased to 225 and 270 kg N ha-1.

In our investigation, we found that winter wheat spikes had higher nitrogen contents ever since plants began absorbing more nitrogen from the soil. Previous research has shown that high temperatures during the blooming and grainfilling growth stage may increase the nitrogen concentration in wheat grains due to changes in weather regulations that affect the development of starch in the grain (Zhang et al., 206; Gorlach et al., 202; Zubaidi et al., 2018). According to our research, spike nitrogen concentration increases dramatically as nitrogen rates rise. Grain nitrogen concentrations of winter wheat below the treatment of 225 kg N ha-1 and the ratio of 6:4 as camper of 5:5 ratio when the nitrogen was applied at the jointing stage as camper of flowering and grain filling stage, respectively, averaged 83.90% and 52.09% at the flowering stage and 93.71% and 57.95% at the grain filling stage. The results of Wille et al. (2017) show that the nitrogen content of winter wheat increases in the following ways: spikes 58.36 to 66.12%, leaves 12.19 to 17.88%, and stems 2.21 to 7.05%. Additionally, our findings are consistent with those of Tian et al. (2018), who demonstrated that when winter wheat growth improved, the influence of nitrogen fertilization tended to be larger. At the end of the jointing and grain filling stages, the nitrogen treatment had substantially more stem, spikes, and above-ground dry biomass than the control. While the effects of treatments and ratios on plant nitrogen concentrations varied between the vegetative and reproductive sections, the two growing seasons' impacts on plant nitrogen contents during the flowering stage were similar (Fig. 3.4). The treatment of 225 kg N ha-1 under ratio 6:4 produced the best results, ranging from 26.33 to 24.83 (g kg-1) at the flowering stage and 22.56 to 21.06 at the grain filling stage in both years as compared to the camper of 150 and 300 kg N ha-1. Consistent with our findings, Jalal et al. 2020 revealed that the total nitrogen content of grains per spike (Grain N•spike-1) was significantly higher than the total nitrogen content of all the other organs investigated. Furthermore, under fertilized settings, the total organ nitrogen and the nitrogen content on a dry matter basis were higher than under unfertilized conditions.

#### 5. Conclusion

In both years, the nitrogen content increased in above-ground biomass, stems, or leaves until the jointing stage; nevertheless, the nitrogen content spiked until the conclusion of flowering, during the grain filling stage. The nitrogen concentration in the plant's above-ground biomass, stem, and leaves was significantly influenced (p < 0.05) by the winter wheat growth stage and nitrogen nutrition method. 92% of grain yield was influenced by N timing, treatments, and ratios; 86% was influenced by fertilizer application rates. When nitrogen fertilizer was applied at 225 kg N ha-1 in both years, grain yield rose. Although there was a tight positive association between grain output and nitrogen concentration in stems during the jointing stage, there was also a close positive relationship between grain yield and nitrogen concentration in leaves and ears at the end of the flowering period. This study suggests that when plants require nitrogen nutrition at the right growth stage, it should be applied in the right amounts and ratios to sustainably increase grain output and increase agricultural productivity.

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