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PHYTOMORPHOLOGY OF WHEAT AND ITS GRAIN QUALITY RESPONSE TO DIVERSE APPROACHES OF IRON APPLICATION

Muhammad Yousif Shaikh^{1*}, Aijaz Ahmed Soomro¹, Muhammad Nawaz Kandhro¹ and Inayatullah Rajpar²

¹Department of Agronomy, ²Department of Soil Science; Sindh Agriculture University, Tandojam

*Corresponding author: yousifghanishaikh@gmail.com

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Abstract

Agriculture soils of Pakistan are already deficient of macronutrients, and now inadequacy for micronutrients has also been reported causing adverse effects on crop productivity. The study was carried out to investigate the effect of soil and foliar applied iron (Fe) on wheat. The treatments included: T₁=Control (without Fe), T₂=Soil applied Fe 3 kg ha⁻¹, T₃=Soil applied Fe 6 kg ha⁻¹, T₄=Foliar applied Fe 0.2%, T₅=Foliar applied Fe 0.4%, T₆=Soil applied Fe 1.5 kg ha⁻¹+foliar applied Fe 0.1%, T₇=Soil applied Fe 1.5 kg ha⁻¹+foliar applied Fe 0.2%, T₈=Soil applied Fe 3 kg ha⁻¹+foliar applied Fe 0.1% and T₉=Soil applied Fe 3 kg ha⁻¹+foliar applied Fe 0.2%. All the treatments as alone or in combination, affected growth, yield and quality of wheat significantly (P<0.05). The results showed that T₉ and T₃ were most promising for almost all studied traits. Under T₉ and T₃, variety "TD-1" produced plants of 59.44 and 59.09 cm, while "Sindhu" showed 92.9 and 75.98 cm height, respectively. Higher grain yield (4799.7 and 4760.5 kg ha⁻¹), grain protein (17.22 and 17.13%) were also recorded in T₉ and T₃, respectively; while T₁ remained the least. In varieties, "TD-1" produced higher grain yield (4788.9 kg) than "Sindhu" (4341.8 kg ha⁻¹). Inversely, grain protein was higher in variety "Sindhu" (17.10%) than "TD-1" (16.72%). It was concluded that soil applied Fe 3 kg ha⁻¹+foliar Fe 0.2% concentration or in absence of foliar application, soil applied Fe at 6 kg ha⁻¹ remained most appropriate dose for wheat growers in addition to recommended NPK.

Keywords: Wheat, phytomorphology, iron, soil applied, foliar, grain yield, quality

INTRODUCTION

Wheat (*Triticum aestivum* L.) is the major source of plant based human nutrition and is an essential part of daily dietary needs (Bouis, 2003; Dewal and Pareek, 2004; Ghasemi-Fasaei and Ronaghi, 2008; Nawab *et al.*, 2006). This crop ensures food security of the country and cultivated over 22 million acres; accounts for 7.8 percent of the value added in agriculture and 1.8 percent of GDP (GoP, 2023). Self-sufficiency in wheat has always been a national objective and thus challenges for the agriculture experts and policy makers. As strategic crop, any shortfall in wheat production can create a crucial situation leading to political uncertainty, significant amount of foreign reserves is spent on imports, rise in prices of wheat flour and pocket shortages in vulnerable areas. During 2021-22, area sown decreased to 8,976 thousand ha⁻¹ (2.1%) against last year's area (9,168 thousand ha⁻¹). The production of wheat declined to 26.394 million tons (3.9 %) over 27.464 million tons of last year (2020-21). The declined production was associated with decline in area sown, acute shortage of irrigation water and drought conditions at sowing, less fertilizers uptake and heat wave at grain formation

stage (GoP, 2023). Wheat is the source of food for more than 60 percent of population in Pakistan and thus, the health of millions of people is directly influenced by the grain quality and nutrients (Iftikhar *et al.*, 2019; Khan *et al.*, 2010). More than 25% of world population is affected by micronutrient deficiencies in food products, a problem which is known as hidden hunger (Muthayya *et al.*, 2013). The application of micronutrients mixed with macronutrients increases plant growth, yield and grain quality (Murphy *et al.*, 2008; Sanchez & Swaminathan, 2005; Saqee *et al.*, 2023). Among micronutrients, the effect of iron (Fe) in wheat based food is crucial for human health (Abbas *et al.*, 2009; Afshar *et al.*, 2020; Chen *et al.*, 2018; Inayat *et al.*, 2014; Nadim *et al.*, 2013; Ziaieian & Malakouti, 2006). Iron is among essentially required 17 distinct nutrients for plants to achieve their full potential in terms of growth and development (Iqbal *et al.*, 2011; Wasaya *et al.*, 2021). Fe is essential for chlorophyll production in wheat, which keeps the plant leaves looking healthy and green; in result, it is existed in most of enzymes to help the plant produce energy by checking soil nitrate and soil sulfate (Rawashdeh *et al.*, 2023; Saqee *et al.*, 2023). Fe concentrations in plant tissues can range anywhere from 200 to 400

ppm. However, for healthy plants, this rate has to be over 100 ppm in order to avoid showing any signs of chlorosis, which most frequently manifests itself in the younger leaves (Shahrokhi *et al.*, 2012; Soetan *et al.*, 2010). Iron chlorosis, which is caused by Bicarbonate (HCO_3) prevents body to absorb Fe (Ali *et al.*, 2013; Ali *et al.*, 2020; Aziz *et al.*, 2019). Fe deficiencies are not always visible in plants grown in most calcareous soils, frequently suffer from Fe shortages (Assunco *et al.*, 2022). In combination with macronutrients, the application methods that are employed to supply crops with micronutrients have an effect on the growth and output of the crops. Chaudry *et al.* (2007) found that the most effective strategy to increase wheat grain yield was to apply Fe and other micronutrients to the leaves of plant in foliar form but irrespective of application methods, the combined use of micronutrients results in improvement of the plant quality and output (Malakouti, 2008). According to Arif *et al.* (2006) foliar applied micronutrients at tillering, jointing, and booting stages of wheat caused an increase in yield and grain quality. They also concluded that the use of foliar fertilizer is potentially productive method for increasing the concentration of micronutrients in wheat grain and the bioavailability of applied elements (Ali *et al.*, 2020). Foliar Fe application had a substantial impact on the grain Fe content of wheat; however, there was no significant interaction between genotype and Fe treatment ($P>0.05$). After foliar Fe treatment, there was no noticeable difference between cultivars and landraces in terms of the amount of Fe present or the yield (Hao *et al.*, 2021). Among various fortification techniques, soil applied Fe is considered as most cost-effective, rapid, and sustainable strategy to improve the contents of micronutrients in wheat grains to alleviate the widespread Fe deficiencies in humans. FeSO_4 is widely applied inorganic fertilizer as Fe source, due to its high solubility and low cost. It is evident that Fe fertilizer effectively improves wheat grain concentration and economic yield in Fe-deficient regions (Rawashdeh *et al.*, 2023). The FeSO_4 application through soil enhances grain quality of wheat. According to Niyigaba *et al.* (2019), soil applied Fe was more effective method for increasing the grain Fe as compared to its foliar application. Wheat output and quality are both increased when the micronutrient is used through soil (Moreno-Lora & Delgado, 2020). Similarly, Hao *et al.* (2021) concluded that grain Fe was 41.0 mg kg^{-1} when wheat fields were supplied with FeSO_4 at 20 kg ha^{-1} . The Fe content of grains, as well as the bioavailability of Fe, were unaffected by foliar Fe treatment, but influenced significantly ($P<0.05$) when Fe was applied through soil regardless of varieties (Hao *et al.*, 2021). In regions of less precipitation, micronutrients can be effectively used both through soil and by foliar method. Although, the Fe required by wheat is very small but in order to completion of

crop life cycle, Fe would be essentially required. Adequate Fe concentration in plant tissue may be in the range of 50-250 ppm, while in grain it may be in the range of 25-35 ppm (Cakmak *et al.*, 2010). In conditions, when plenty of irrigation water is available, Fe application through soil serves the purpose effectively (Naz *et al.*, 2015); while in drought conditions of water shortage circumstances, in combined to soil application, foliar application on leaves works more effectively to improve crop productivity and grain quality (Fernandez *et al.*, 2009). It was hypothesized that proper fertilization of Fe may play crucial role in improving growth, yield and nutritional quality of wheat and can help in resolving the malnutrition issue of populations. Therefore, this research was conducted to investigate the impact of Fe application through soil at various rates and through foliar application at different concentrations on the plant morphology, yield and grain quality of two wheat varieties.

MATERIALS AND METHODS

In order to examine the effect of soil and foliar applied Fe (separately and in integration) on growth, yield and grain quality of two wheat varieties (TD-1 and Sindhu) the trial was conducted during Rabi, 2020-21 and 2021-22 at the Students' Experimental Farm, Sindh Agriculture University Tandojam ($25^{\circ}26'0\text{N}$ latitude and $68^{\circ}32'E$ longitude). The land was prepared by two dry plowings followed by heavy soaking dose. When soil came in workable condition, precision leveling of land was carried out, followed by two cross-wise plowings with cultivator and planking with patio to achieve fine seedbed, bunds and feeding water channels were prepared. Sowing of wheat was done by single coulter hand drill in each experimental unit. The experiments were laid out in Randomized Complete Block Design (Factorial) with 3-replicates in net plot size of $6 \text{ m} \times 3 \text{ m} = 18 \text{ m}^2$, keeping 75 cm row spacing. The research comprised of nine treatments i.e T_1 = Control (without Fe), T_2 = Soil applied Fe 3 kg ha^{-1} , T_3 = Soil applied Fe 6 kg ha^{-1} , T_4 = Foliar applied Fe 0.2%, T_5 = Foliar applied Fe 0.4%, T_6 = Soil applied Fe 1.5 kg ha^{-1} + foliar applied Fe 0.1%, T_7 = Soil applied Fe 1.5 kg ha^{-1} + foliar applied Fe 0.2%, T_8 = Soil applied Fe 3 kg ha^{-1} + foliar applied Fe 0.1% and T_9 = Soil applied Fe 3 kg ha^{-1} + foliar applied Fe 0.2%. Ferrous Sulphate Crystal (FeSO_4), density 2.84 g/cm^3 , melting point 70°C , molar mass 151.908 g/mol , was purchased from Al-Beruni Scientific Store, Hyderabad. When applied through soil, the per hectare quantity as per the treatment plan was calculated on the basis of plot size, and applied as basal dose. In case of foliar application, FeSO_4 was mixed in water at certain percentage (as per treatment plan) in a 100 litre water tanks and sprayed with knapsack hand sprayer on crop leaves at tillering, flowering and grain filling stages. Six irrigations were applied throughout the growing season of wheat. The first irrigation was

applied at 30 to 35 days after sowing (DAS); whereas, subsequent irrigations were applied as per the crop requirement. The NPK fertilizers were applied as recommended (120-90-60 kg ha⁻¹) locally for Sindh province. The N was applied in the form of urea (46% N) and DAP (18% N); P in the form of Di-Ammonium Phosphate, 46% P₂O₅(DAP) and K in the form of Sulphate of Potash, 50% K₂O (SOP). All P, K and Fe in combined to half of N were applied as basal application; while remaining N was applied in two splits. The weeds in the experimental wheat crop were removed manually. The harvesting of experimental crop in all the treatments was manually done in the last week of March in both the years.

For recording the data, the plant was measured for height (cm) from ground to apex before harvest using a measuring tape; while for counting tillers number, one meter square area was selected in each treatment and the total number of tillers were counted manually. For spike length measurement, the measuring tape was used; while for recording the number of grains spike⁻¹, entire grain of the labelled plants was gathered, manually counted and averaged in grams. The seed index value was worked out on the basis of 1000 grains weight from the grain lot of each treatment and averaged in grams. The biological yield is the total crop biomass, and it was collected from all treatments separately, the bundles were tied with ropes and shifted to the threshing yard. Before threshing to separate the grain and straw/husk, the total biological yield was weighed using a field balance and averaged in kilograms per plot. Later, the per plot yield was calculated to achieve biological yield ha⁻¹ using the following formula:

$$\text{Biological yield (kg ha}^{-1}\text{)} = \frac{\text{Biological yield plot}^{-1} \text{ (kg)} \times 10000}{\text{Net plot size (m}^2\text{)}}$$

Similarly, for achieving the grain yield, each of the tied bundles were threshed separately for each treatment and grain yield per plot (kg) was converted into grain yield ha⁻¹ by using the following formula:

$$\text{Grain yield (kg ha}^{-1}\text{)} = \frac{\text{Grain yield plot}^{-1} \text{ (kg)} \times 10000}{\text{Net plot size (m}^2\text{)}}$$

The harvest index was calculated on the basis of biological yield and grain yield; considering grain yield as percentage of the biological yield per hectare using the following formula:

$$\text{Harvest index (\%)} = \frac{\text{Grain yield (kg ha}^{-1}\text{)} \times 100}{\text{Biological yield (kg ha}^{-1}\text{)}}$$

The leaf area (cm²), leaf area index (%) and crop growth rate (CGR) of wheat varieties were determined as per the respective formula given hereunder:

$$\text{Leaf Area (cm}^2\text{)} = \text{Leaf length} \times \text{Leaf breadth} \times \text{Factor (0.75)}$$

$$\text{Leaf area index (\%)} = \frac{\text{Leaf area (cm}^2\text{)} \times 100}{\text{Net plot size (m}^2\text{)}}$$

$$\text{CGR g m}^{-2} \text{ day}^{-1} = \frac{\text{Ground area (cm}^2\text{)} \times (\text{W}_2 - \text{W}_1)}{\text{T}_2 - \text{T}_1 \times \text{GA}}$$

Where;

W₁ = Weight at T₁ of the period

W₂ = Weight at T₂ of the Period

T₁ = Time in date at the start of the period

T₂ = Time in the date at the end of the period

GA = Ground area

The total dry matter was observed by oven drying samples at 70°C for 48 hours (2 days) and then weighed and averaged. In case of grain quality analysis, Similarly, the grain Fe was determined by DTPA Method The diethylenetriaminepentaacetic Acid (DTPA) test of Lindsay and Norvell (1978). DTPA method has a capacity to complex each of the micronutrient cations as 10 times of its atomic weight. The capacity ranges from 550 to 650 mg kg⁻¹ depending upon the micronutrient cations as suggested by ICARDA (2013). However, grain protein content was determined on the basis of grain N content by multiplying a factor i.e 6.25. Grain protein content in seed = N % in seed multiplied by factor 6.25 (AOAC, 1984).

The collected data were subject to statistical analysis using Statistix 8.1 computer software. The two-way ANOVA was developed to examine significance of the effects on studied parameters due to treatments and their interactions. The LSD test was applied to compare treatments' mean considering the significance of the treatments' effect (Steel et al., 1997).

RESULTS

Plant height: Plant height showed similarity (P>0.05) in higher side (76.16 and 75.98 cm) when crop was given soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉) or higher rate of soil applied Fe 6 kg ha⁻¹ (T₃), respectively (Table 1). The plant height followed a decreasing trend with values 75.12, 73.79, 73.15 and 73.14 cm in treatments comprised of soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇); only soil applied Fe 3 kg ha⁻¹ (T₂) and soil applied Fe 1.5 kg ha⁻¹ and foliar Fe 0.1% (T₆), respectively. Whereas, plant height declined considerably (73.0 and 72.41 cm) when only foliar Fe was applied 0.4% (T₅) and 0.2% (T₄), respectively; while least plant height (71.78 cm) was noticed in control (T₁). In varieties, Sindhu (V₂) produced plants of significantly greater height as compared to companion variety TD-1 (V₁) which is a dwarf variety produced shorter plants. The treatment interaction (T×V) maximized plant height (92.9 cm) under T₉×V₂ interaction, followed by 91.97 and 90.61 cm plant height in T₈×V₂ and T₇×V₂ interactions, respectively. Statistically the difference in plant height between T₉ and T₃, and amongst T₆, T₅ and T₂ were non-significant (P>0.05).

Tillers m⁻²: Similarity in tillers m⁻² (303.17 and 302.24) was seen in crop given soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉); and soil applied Fe 6 kg

ha⁻¹ (T₃), respectively (Table 1). The tillers number decreased to 298.67, 293.08 and 292.81 m⁻² in crop fertilized with soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇); and only soil applied Fe 3 kg ha⁻¹ (T₂), respectively (Table 1). The treatments comprised of soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and foliar Fe 0.4% (T₅) resulted in decreased tillers number (290.61 and 290.356 m⁻²), respectively. However, lower number of tillers (288.18 and 285.70m⁻²) was found in crop provided with only foliar Fe 0.2% (T₄) and control (T₁), respectively. The varietal effect indicated that TD-1 (V₁) produced more tillers m⁻² (P<0.05) than Sindhu (V₂). The treatment interaction study showed that maximum tillers (309.1 m⁻²) were found in interaction of T₉×V₁, followed by the 303.04 and 297.24 tillers m⁻² in T₈×V₁ and T₉×V₂ interactions, respectively.

Tillers plant⁻¹: The maximized tillers plant⁻¹ (7.75) were counted in crop supplied with soil applied Fe 3

kg ha⁻¹ + foliar Fe 0.2% (T₉); followed by 7.56, 7.47, 7.33 and 7.33 tillers plant⁻¹ noted under soil applied Fe 6 kg ha⁻¹ (T₃) and soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇); and only soil applied Fe 3 kg ha⁻¹ (T₂), respectively (Table 1). The soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆), foliar Fe 0.4% (T₅) and 0.2% (T₄) or control (T₁) showed similarity (P>0.05) for this trait with 7.27, 7.26, 7.21 and 7.15 tillers plant⁻¹ respectively. TD-1 variety produced more tillers plant⁻¹ than Sindhu; while highest tiller number (8.34 plant⁻¹) was counted T₉×V₁ interaction and minimum (6.44 plant⁻¹) in T₁×V₁₂ interaction.

Number of plants m⁻²: The number of plants (41.14, 41.03 and 40.55m⁻²) were greater in treatments based on soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉), soil applied Fe 6 kg ha⁻¹ (T₃) and soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), respectively (Table 2). The crop fertilized with soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇); soil applied Fe 1.5 kg

Table 1. Plant height, number of tillers m⁻² plant⁻¹ of wheat varieties (V) as affected by levels and methods of Fe

Soil and Foliar application of Fe	Plant height (cm)			No. of Tillers m ⁻²			No. of tillers plant ⁻¹		
	V ₁	V ₂	Mean	V ₁	V ₂	Mean	V ₁	V ₂	Mean
T ₁ = Control (Without Fe)	56.01 ^l	87.54 ^g	71.78 ^f	291.27	280.13	285.70 ^f	7.86	6.44	7.15 ^d
T ₂ = Soil Fe 3 kg ha ⁻¹	56.13 ^{kl}	90.17 ^{cd}	73.15 ^d	297.09	288.53	292.81 ^{cd}	8.02	6.63	7.33 ^{cd}
T ₃ = Soil Fe 6 kg ha ⁻¹	59.09 ^h	92.87 ^a	75.98 ^a	307.29	297.19	302.24 ^a	8.29	6.83	7.56 ^b
T ₄ = Foliar Fe 0.2%	56.46 ^{kl}	88.36 ^f	72.41 ^e	293.60	282.76	288.18 ^{ef}	7.92	6.50	7.21 ^d
T ₅ = Foliar Fe 0.4%	56.74 ^k	89.26 ^e	73.00 ^d	295.06	285.65	290.35 ^{de}	7.96	6.56	7.26 ^d
T ₆ = Soil Fe 1.5 kg ha ⁻¹ +foliar Fe 0.1%	56.56 ^{kl}	89.72 ^{de}	73.14 ^d	294.12	287.09	290.61 ^{de}	7.94	6.60	7.27 ^d
T ₇ = Soil Fe 1.5 kg ha ⁻¹ +foliar Fe 0.2%	56.96 ^l	90.61 ^c	73.79 ^c	296.20	289.96	293.08 ^c	7.99	6.67	7.33 ^{cd}
T ₈ = Soil Fe 3 kg ha ⁻¹ +foliar Fe 0.1%	58.28 ⁱ	91.97 ^b	75.12 ^b	303.04	294.30	298.67 ^b	8.18	6.77	7.47 ^{bc}
T ₉ = Soil Fe 3 kg ha ⁻¹ +foliar Fe 0.2%	59.44 ^h	92.90 ^a	76.16 ^a	309.10	297.24	303.17 ^a	8.34	7.17	7.75 ^a
Mean	57.30 b	90.378^a	--	298.53^a	289.20^b	--	8.06^a	6.68^b	--

ha⁻¹ + foliar Fe 0.1% (T₆), soil applied Fe 3 kg ha⁻¹ (T₂), foliar Fe 0.4% (T₅) and foliar Fe 0.2% (T₄) produced averagely 39.8, 39.75, 39.46, 39.41 and 39.11 plants m⁻², respectively; while the lowest number of plants (38.77 m⁻²) was recorded in control (T₁). Variety Sindhu had greater number of plants than TD-1; while plants m⁻² maximized (43.07 m⁻²) in T₉×V₂ interaction and least (36.96 m⁻²) in T₁×V₁ interaction.

Spike length: Length of spikes showed similarity in the higher side (13.85, 13.68, 13.67 cm) in case of the crop obtaining soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉); soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), and only soil applied Fe 6 kg ha⁻¹ (T₃), respectively (table 2). The spike length showed relative decrease (12.75, 12.67 and 12.59 cm) under soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇); soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and only foliar Fe 0.4% (T₅), respectively. Further decrease in spike length (12.57 cm) was seen under soil applied Fe 3 kg ha⁻¹ (T₂). However, almost equal and

minimized spike length (12.34 and 12.32 cm) was noticed in crop obtaining only foliar Fe 0.2% (T₄) and control (T₁), respectively. In varieties, TD-1 produced longer spikes as compared to Sindhu. Similarly, spike length was maximum (14.21 cm) in treatments interaction of T₉×V₁.

No. of grains spike⁻¹: The grains spike⁻¹ were markedly higher (48.76) in crop obtaining soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉); followed by 48.47, 47.80 and 46.73 grains spike⁻¹ recorded in crop given soil applied Fe 6 kg ha⁻¹ (T₃), soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈) and soil applied Fe 1.5 kg ha⁻¹+foliar Fe 0.2% (T₇), respectively. A decline in grains spike⁻¹ upto 46.54, 46.40 and 46.31 grains spike⁻¹ in treatments comprised of foliar Fe 0.4% (T₅), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and foliar Fe 0.2% (T₄), respectively. The least grains (46.04 and 45.94) spike⁻¹ were recorded in crop obtaining only soil applied Fe 3 kg ha⁻¹ (T₂) and control (T₁), respectively (Table 2). TD-1 variety resulted in more grains spike⁻¹ than Sindhu; while interaction for more grains spike⁻¹ (50.523) were

counted in $T_9 \times V_1$ interaction, followed by 49.533 and 48.42 grains spike⁻¹ found in $T_8 \times V_1$ and $T_7 \times V_1$ interactions, respectively.

Table 2 Plants m⁻², spike length and grains spike⁻¹ of wheat varieties (V) as affected by levels and methods of Fe

Treatments	No. of plants m ⁻²			Spike length (cm)			No. of grains spike ⁻¹		
	V ₁	V ₂	Mean	V ₁	V ₂	Mean	V ₁	V ₂	Mean
T ₁ =Control (No Fe)	36.96	40.58	38.77 ^d	12.64 ^{efg}	12.00 ^{hi}	12.32 ^c	47.61 ^{bcd}	44.28 ^f	45.94 ^d
T ₂ =Soil Fe 3 kg ha ⁻¹	37.70	41.81	39.75 ^{bc}	12.89 ^{de}	12.24 ^{ghi}	12.57 ^{bc}	47.71 ^{bcd}	44.37 ^f	46.04 ^{cd}
T ₃ =Soil Fe 6 kg ha ⁻¹	38.99	43.06	41.03 ^a	14.02 ^a	13.32 ^{bc}	13.67 ^a	50.23 ^a	46.71 ^{de}	48.47 ^{ab}
T ₄ =Foliar Fe 0.2%	37.25	40.97	39.11 ^{cd}	12.77 ^{def}	11.92 ⁱ	12.34 ^c	47.99 ^{bc}	44.63 ^f	46.31 ^{cd}
T ₅ =Foliar Fe 0.4%	37.44	41.39	39.41 ^{bcd}	12.91 ^{cde}	12.26 ^{ghi}	12.59 ^{bc}	48.23 ^b	44.86 ^f	46.54 ^{cd}
T ₆ =Soil Fe 1.5 kg ha ⁻¹ +foliar Fe 0.1%	37.32	41.60	39.46 ^{bc}	13.00 ^{cde}	12.35 ^{fgh}	12.67 ^b	48.08 ^b	44.71 ^f	46.40 ^{cd}
T ₇ =Soil Fe 1.5 kg ha ⁻¹ +foliar Fe 0.2%	37.59	42.01	39.80 ^b	13.08 ^{bcd}	12.43 ^{fg}	12.75 ^b	48.42 ^b	45.03 ^f	46.73 ^c
T ₈ =Soil Fe 3 kg ha ⁻¹ +foliar Fe 0.1%	38.45	42.64	40.55 ^a	14.03 ^a	13.33 ^{bc}	13.68 ^a	49.533 ^a	46.07 ^e	47.80 ^b
T ₉ =Soil Fe 3 kg ha ⁻¹ +foliar Fe 0.2%	39.22	43.07	41.14 ^a	14.21 ^a	13.50 ^b	13.85 ^a	50.523 ^a	46.99 ^{cde}	48.76 ^a
Mean	37.88 b	41.90 a	---	13.28 ^a	12.59 ^b	---	48.70 ^a	45.30 ^b	---

Seed index: The higher seed index (48.06 and 47.91 g) was recorded in crop given soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉) and only soil applied Fe 6 kg ha⁻¹ (T₃), respectively (Fig 1); followed by 47.35 and 46.46 g seed index in crop treated with soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈) and soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇), respectively. A declined seed index (46.07, 46.03 and 46.01 g) was noticed in crop given soil applied Fe 1.5

kg ha⁻¹ + foliar Fe 0.1% (T₆), foliar Fe 0.4% (T₅) and soil applied Fe 3 kg ha⁻¹ (T₂), respectively. Almost equal and least seed index (45.68 and 45.29 g) was found in crop obtaining only foliar Fe 0.2% (T₄) and control (T₁), respectively. Varietal effect on this trait showed that TD-1 showed greater seed index than Sindhu; while seed index was highest (48.74 g) in T₉ × V₁ interaction and least (44.65 g) in T₁ × V₂ interactio

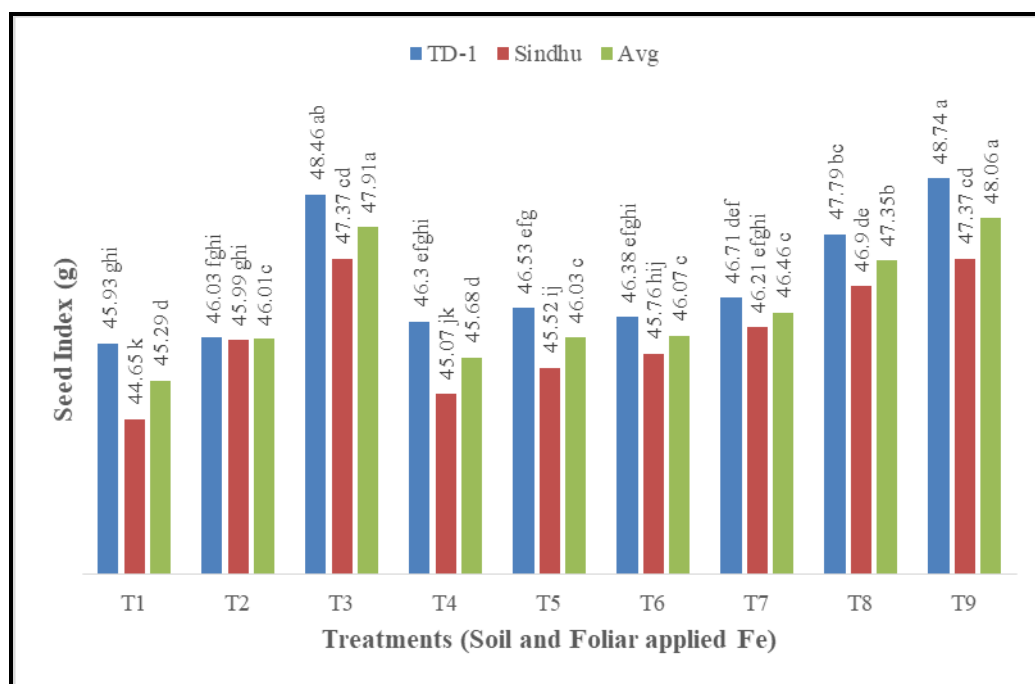


Fig 1: Effect of soil and foliar applied Fe (T) on Seed index (g) of wheat varieties TD-1 and Sindhu

Biological yield (kg ha⁻¹): It is evident that the biological yield was higher (9163 and 9136.8 kg ha⁻¹) in crop supplied with soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉) and only soil applied Fe 6 kg ha⁻¹ (T₃), respectively (Fig 2); followed by 9030.4 and 8864.3 kg ha⁻¹ biological yield obtained from the crop treated with soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈) and soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇), respectively. The biological yield

diminished to 8788.4, 8780.9 and 8777.8 kg ha⁻¹ in fields supplied with soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆), soil applied Fe 3 kg ha⁻¹ (T₃) and foliar Fe 0.4% (T₅), respectively. The lowest biological yield (8634.9 kg ha⁻¹) was realized in control where the crop was left untreated of any form of Fe (T₁). In case of varieties, Sindhu produced higher biological yield than variety TD-1; while interaction of T₉ × V₂ resulted in highest

biologicallyiedl (9706.9 kg ha⁻¹) and least (8121.9 kg ha⁻¹) in T₁×V₁ interaction

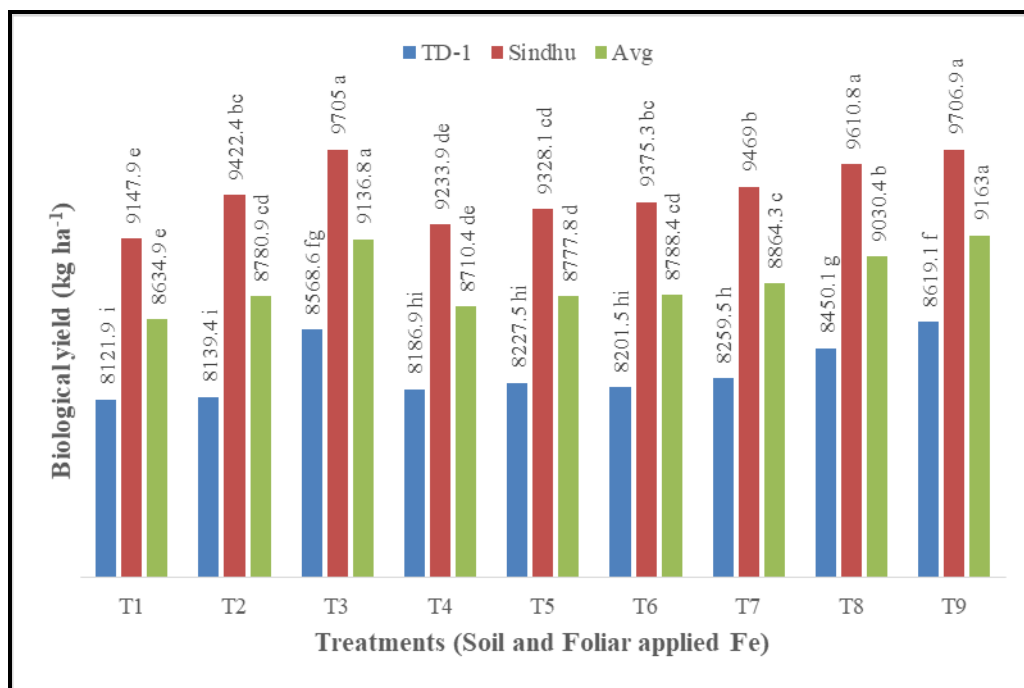


Fig 2: Effect of soil and foliar applied Fe (Treatments) on biological yield (kg ha⁻¹) of wheat varieties

Grain yield (kg ha⁻¹): The grain yield was highest (4799.7 kg ha⁻¹) in crop supplied with soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉), followed by the grain yield of 4760.5 and 4707.3 kg ha⁻¹ grain yield in treatments comprised of soil applied Fe 6 kg ha⁻¹ (T₃) and soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), respectively (Fig 3). The grain yield considerably decreased (4595.8, 4562.7 and 4504 kg ha⁻¹) in treatments comprised of soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and soil applied Fe 3 kg ha⁻¹ (T₂), respectively. The least grain yield (4348.2 kg ha⁻¹) was obtained in control (T₁). In varieties, TD-1 produced higher grain yield than its companion variety Sindhu. The T × V interaction indicated that grain yield was maximum (5084.7 kg ha⁻¹) in T₉×V₁ interaction; while minimum grain yield (4190.3 kg ha⁻¹) was achieved in T₁×V₂ interaction.

Harvest index: It is obvious from the data that maximized harvest index (52.77%) was determined in crop supplied with soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉), followed by the harvest index of 52.52, 52.49, 52.38 and 52.32 percent recorded in wheat crop treated with soil applied Fe 3 kg ha⁻¹ +

foliar Fe 0.1% (T₈), soil applied Fe 6 kg ha⁻¹ (T₃); soil applied soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇), respectively (table 4). A decline in the harvest index of wheat was noted in case of treatments based on foliar Fe 0.4% (T₅), control (T₁) and foliar Fe 0.2% (T₄), with average harvest index of 50.80, 5.65 and 5.60 percent, respectively. The varietal effect on this parameters showed that TD-1 (V₁) resulted in markedly higher harvest index as compared to other tested variety Sindhu. The treatment interaction T₉×V₁ maximized harvest index (59.02%); while minimum harvest index (45.62%) in T₄ × V₂ interaction (table 3).

Dry matter (g plant⁻¹): The dry matter was highest (59.63 g plant⁻¹) in treatment comprised of soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉), followed by the average dry matter of 59.35, 59.31, 59.19 and 59.12 g plant⁻¹ recorded in treatments comprised of soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), soil applied Fe 6 kg ha⁻¹ (T₃), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇), respectively (Table 3).

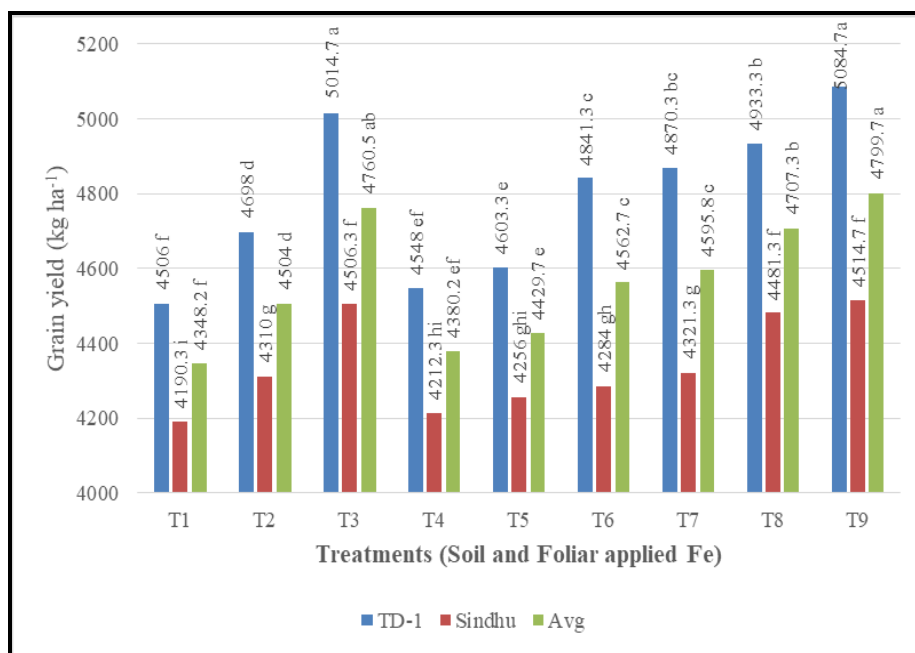


Fig 3: Effect of soil and foliar applied Fe on grain yield (kg ha⁻¹) of wheat varieties

Table 3. Harvest index, spike length and grains spike⁻¹ of wheat as affected by levels and methods of Fe

Treatments	Harvest index (%)			DM Yield (g plant ⁻¹)			Leaf area (cm ²)		
	V ₁	V ₂	Mean	V ₁	V ₂	Mean	V ₁	V ₂	Mean
T ₁ =Control (No Fe)	55.49 ^c	45.81 ^d	50.65 ^c	51.76 ^c	62.71 ^b	57.24 ^d	60.57 ^d	65.84 ^c	63.20 ^e
T ₂ =Soil Fe 3 kg ha ⁻¹	57.72 ^b	45.75 ^d	51.74 ^b	51.69 ^c	65.23 ^a	58.46 ^{bc}	60.48 ^d	68.49 ^b	64.49 ^b
T ₃ =Soil Fe 6 kg ha ⁻¹	58.54 ^{ab}	46.44 ^d	52.49 ^{ab}	52.47 ^c	66.15 ^a	59.31 ^{ab}	61.40 ^d	69.46 ^{ab}	65.43 ^{ab}
T ₄ =Foliar Fe 0.2%	55.56 ^c	45.62 ^d	50.60 ^c	51.56 ^c	62.79 ^b	57.17 ^d	60.32 ^d	65.93 ^c	63.12 ^c
T ₅ =Foliar Fe 0.4%	55.96 ^c	45.63 ^d	50.80 ^c	51.56 ^c	63.24 ^b	57.40 ^{cd}	60.33 ^d	66.40 ^c	63.36 ^c
T ₆ =Soil Fe 1.5 kg ha ⁻¹ +foliar Fe 0.1%	59.06 ^a	45.70 ^d	52.38 ^{ab}	51.64 ^c	66.74 ^a	59.19 ^{ab}	60.42 ^d	70.07 ^a	65.25 ^{ab}
T ₇ =Soil Fe 1.5 kg ha ⁻¹ +foliar Fe 0.2%	58.99 ^a	45.64 ^d	52.32 ^{ab}	51.58 ^c	66.66 ^a	59.12 ^{ab}	60.34 ^d	70.00 ^a	65.17 ^{ab}
T ₈ =Soil Fe 3 kg ha ⁻¹ +foliar Fe 0.1%	58.41 ^{ab}	46.63 ^d	52.52 ^{ab}	52.69 ^c	66.00 ^a	59.35 ^{ab}	61.65 ^d	69.30 ^{ab}	65.48 ^a
T ₉ =Soil Fe 3 kg ha ⁻¹ +foliar Fe 0.2%	59.02 ^a	46.51 ^d	52.77 ^a	52.56 ^c	66.69 ^a	59.63 ^a	61.49 ^d	70.03 ^a	65.76 ^a
Mean	57.64 ^a	45.97 ^b	---	51.95 ^b	65.13 ^a	---	60.78 ^b	68.39 ^a	---

The dry matter declined to 58.46, 57.40 and 57.17 g plant⁻¹ in treatments comprised of soil applied Fe 3 kg ha⁻¹ (T₂), foliar Fe 0.4 % (T₅) and foliar Fe 0.2 % (T₄), respectively. However, the least dry matter (57.24 g plant⁻¹) was obtained in control (T₁). In varieties, Sindhu had greater dry matter than TD-1. The treatment interaction T₉×V₂ resulted in maximum DM yield (66.69 g plant⁻¹), while minimum (51.69 g plant⁻¹) in T₂×V₁ interaction.

Leaf area (cm²): The leaf area was higher (65.76 and 65.48 cm²) in crop given soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉) and soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), respectively; followed by average leaf area of 65.43, 65.25 and 65.17 cm² measured in crop treated with soil applied Fe 6 kg ha⁻¹ (T₃), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇), respectively. A considerable reduction in leaf area (64.49, 63.36 and 63.12 cm²) was recorded in crop given soil applied Fe 3 kg ha⁻¹ (T₃), foliar Fe 0.4% (T₅) and foliar Fe 0.2% (T₄), respectively. The minimum leaf area (63.20 cm²) was measured in crop kept untreated of Fe (control). There was linear association of leaf area with the dry matter and

variety Sindhu was measured with greater leaf area as compared to variety TD-1. The treatment interaction T₉×V₂ resulted in highest leaf area (70.03 cm²); while minimum (60.32 cm²) in T₄×V₁ interaction.

Grain Fe (µg g⁻¹): The grain received from the crop supplied with soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉) had highest Fe content (66.69 µg g⁻¹), followed by the grain Fe contents of 66.41 and 66.35 µg g⁻¹ determined in grain received from the plots given soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈) and soil applied Fe 6 kg ha⁻¹ (T₃), respectively (Fig 4). A considerable decrease in grain Fe was noticed (66.07, 65.48 and 65.40 µg g⁻¹) in plots given soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and soil applied Fe 3 kg ha⁻¹ (T₂), respectively. However, the wheat grain with lowest Fe content (64.17 µg g⁻¹) was measured from the control plots (T₁). In varieties, the grain of variety Sindhu contained relatively higher Fe content as compared to TD-1; while treatments interaction T₉×V₂ showed maximum grain Fe (68.92 µg g⁻¹); and minimum (63.22 µg g⁻¹) in T₁×V₁ interaction.

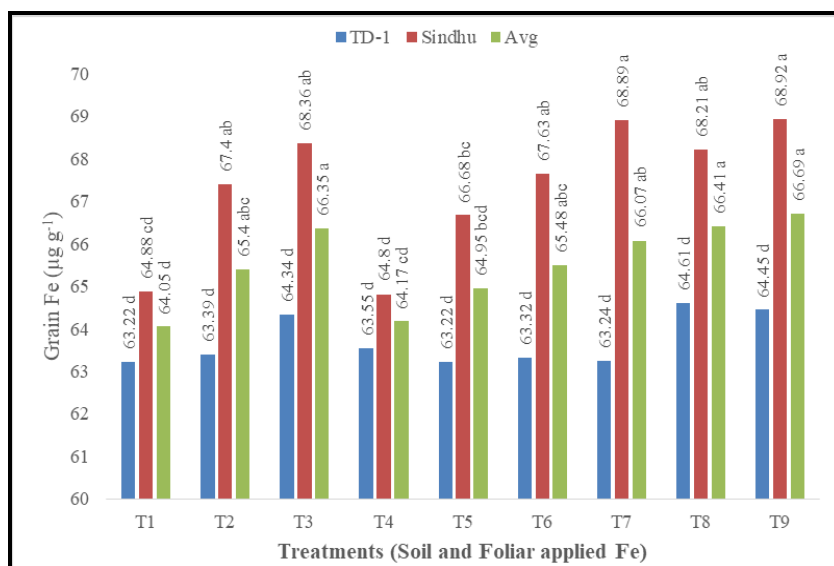


Fig 4: Effect of soil and foliar applied Fe (Treatments) on grain Fe (µg/g) of wheat varieties

Grain protein (%): The grain protein varied significantly in wheat varieties ($P < 0.05$); while soil or foliar applied Fe (T) and treatment interaction did not influence the grain protein significantly ($P > 0.05$). The grain protein was relatively higher (17.22%) in crop given soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.2% (T₉), followed by 17.15, 17.13, 17.07 and 17.05 % grain protein determined in crop given soil applied Fe 3 kg ha⁻¹ + foliar Fe 0.1% (T₈), soil applied Fe 6 kg ha⁻¹ (T₃), soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.1% (T₆) and soil applied Fe 1.5 kg ha⁻¹ + foliar Fe 0.2% (T₇), respectively. Slight further decrease in grain protein (16.89, 16.60 and 16.57%) determined in crop given soil applied Fe 3 kg ha⁻¹ (T₂), foliar Fe

0.4% (T₅), foliar Fe 0.2% (T₄), respectively. However, the lowest grain protein (16.54%) was determined control (T₁). The varietal impact showed that the grain of wheat variety Sindhu contained markedly higher protein than the grain of variety TD-1. The interactive effect (T×V) for T₂, T₃, T₆, T₇, T₈ and T₉ × V₂ interaction showed almost similar results for grain protein and least grain protein was determined in treatment interaction of T₁ × V₂. The results indicated that the grain of variety Sindhu were genetically superior to its companion variety TD-1 in protein content. However, apart from the slight variation in the grain protein, there was similarity ($P > 0.05$) in grain protein among the treatments evaluated (Fig 6).

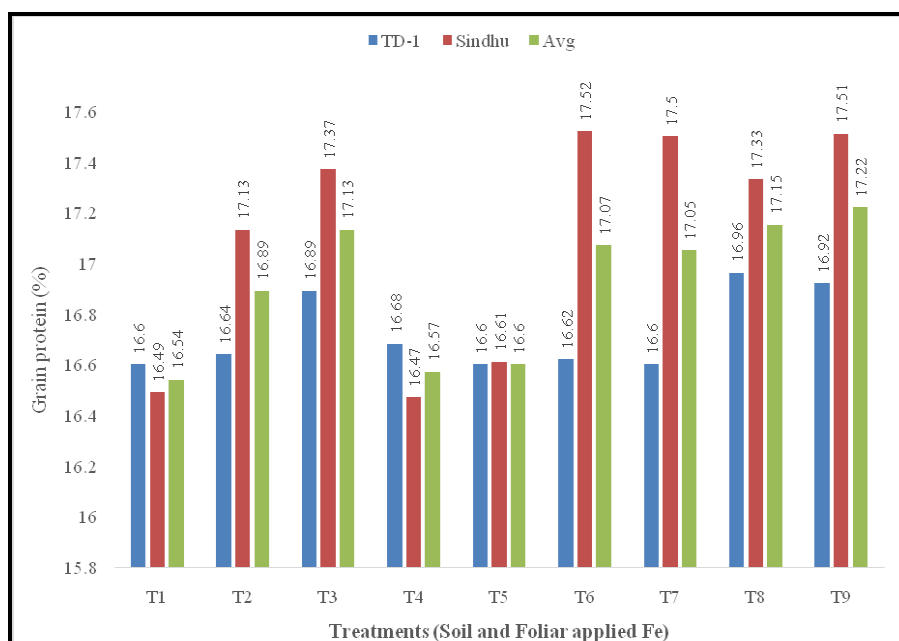


Fig 6: Effect of soil and foliar applied Fe (Treatments) on grain protein (%) of wheat varieties

DISCUSSION

Frequent use of NPK by the farmers to achieve higher crop yields with least or no awareness about the significance of micronutrients, the agriculture soils have become severely inadequate of organic matter and essential needed macro- and micronutrients (Bloemberg & Lugtenberg, 2001). The success of wheat production is reliant on soil available N, P and K. Thus, application of micronutrients at increased concentrations can potentially increase wheat yields and improve grain quality as well. The wheat crop needs distinct nutritional requirements at various stages of its lifecycle, and these requirements may change over time. There are three major critical phases throughout the growth cycle of wheat crop in which nutrients play an especially vital role. At budding stage, leaf feeding stimulates the development of the main shoot as well as the emergence of side shoot buds in the axils of the germinating leaves and the expansion of the germ system; while at tillering stage, the emergence of tillers activates morphophysiological processes, guaranteeing the development of a secondary root system. At the flag leaf stage, which marks the beginning of the emergence of the ear. Thus, using certain methods at the most suitable application stage can maximise the availability of micronutrients (Cakmak *et al.*, 2010; Goel *et al.*, 2021; Iftikhar and Ali, 2017; Rawashdeh *et al.*, 2023).

Among micronutrients, Fe (Fe) has crucial role in crop growth dynamics, yield and produce quality. In this study the effect of Fe (soil, foliar and integrated use) on the growth, yield and grain quality of wheat varieties. The study showed that soil applied Fe (Fe) 3 kg ha⁻¹+foliar Fe 0.2% concentration and soil applied Fe 6 kg ha⁻¹ only showed most promising results for almost all the agronomic parameters including grain yield and grain quality traits recording greater values for representative traits such as plant height (76.16 and 75.98 cm), grain yield (4799.7 and 4760.5 kg ha⁻¹), grain protein (17.22 and 17.13%), respectively.

The findings of the present research are in agreement with many past workers. According to a recent study, the Fe is essential to the grain and chlorophyll production in wheat, which is necessary for the plant to keep leaves healthy and green. Moreover, due to majority of enzymes after Fe application in plants, the plant produce energy by reducing the amounts of nitrate and sulfate in the soil (Impa *et al.*, 2019; Jalal *et al.*, 2020; Kihara *et al.*, 2020; Rawashdeh *et al.*, 2023). Similarly, the interveinal chlorosis also known as Fe deficiency in plants caused yellow-green bands on young leaves caused by insufficient amount of chlorophyll production in result Fe deficiency in the plant occurs (Saquee *et al.*, 2023). In case the Fe is not added to the soil under inadequate conditions, the leaves

become yellow, and the chlorosis spreads to older leaves (Shahrokhi *et al.*, 2012). The Fe cause the production of chlorophyll due to cytochromes and electron transport systems both need Fe as constituent (Soetan *et al.*, 2010). Fe chlorosis caused by HCO₃ that prevents plants from Fe absorption (Makawita *et al.*, 2021). According to Ziaieian & Malakouti (2006), the increased concentrations of Fe boost productivity and quality of wheat grain. He also indicated that in combined to NPK, the application of Fe or mixture of micronutrients had positive influence on grain production and quality in wheat. Abbas *et al.* (2009) showed that Fe applied 12 kg ha⁻¹ not only improved grain yield, but the grain quality (grain protein) was also improved. However, higher Fe rates than the above levels had no positive and significant effect on grain yield and quality. Hafeez *et al.* (2021) acquired highest starch content in grain when Fe was added in combined to usual NPK fertilizers in wheat. Similarly, Niyigaba *et al.* (2019) revealed that utilization of a 100 percent Fe supplement was the most effective method for increasing the amount of Fe found in grain (13 kg ha⁻¹). It is essential to apply Fe to wheat crop at the foliar stage in order to increase the grain Fe, grain protein contents and improve the overall quality of the crop produce. When Fe was used in conjunction other micronutrients, it was shown to be an effective strategy. In combined, the grain crude fat content was unchanged by any of the Fe-containing treatments, and the grain crude fiber content was enhanced by as much as three times the average when using 60% Zn + 40% Fe 5.5 (5.5 kg ha⁻¹ of 60% Zn + 40% Fe) (Li *et al.*, 2016; Maryami *et al.*, 2020; Niyigaba *et al.*, 2019).

The wheat yield and quality both increased when the micronutrient Fe was used as fertilizer. Moreover, the incidence of anemia caused by a lack of Fe in food also lowers (Ali *et al.*, 2022; Pallavi and Sudha, 2017; Ramzan *et al.*, 2020).

Since, Fe has pivotal role in chlorophyll synthesis, because it is a component of cytochromes and electron transport. Its inadequacy adversely affects the activity of various enzymes including catalase and peroxidase having porphyrin as a prosthetic group and iron chlorosis impairs the Fe uptake mechanism. To alleviate nutrient imbalance, plants use different mechanisms to decrease water loss when increasing water uptake, that may reduce leaf area and osmotic adjustment when organic compounds and minerals elements are applied (Hussain *et al.*, 2021; Soetan *et al.*, 2010). Makawita *et al.* (2021), and Rehman *et al.* (2020) clearly showed the evidence about the effectiveness of Fe fertilizer to improve wheat yield and grain nutrient concentration. The application of FeSO₄ enhanced quality of wheat grains and increased nutrient contents in wheat grain.

CONCLUSIONS

The results concluded that soil applied Fe when applied in wheat (3 kg ha⁻¹) in integration to foliar applied Fe (0.2%) or soil applied Fe at higher level (6 kg ha⁻¹) resulted in most promising results for growth, yield and grain quality traits; with greater values for grain yield (4799.7 and 4760.5 kg ha⁻¹) and grain protein (17.22 and 17.13%), respectively.

RECOMMENDATION

It is recommended for the wheat growers that they should choose either soil applied Fe (3 kg ha⁻¹) in combination with foliar applied Fe (0.2% concentration) or double the soil applied Fe (6 kg ha⁻¹) in absence of foliar application of Fe, for achieving the desired wheat yield and grain quality.

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Appendixes/Supplementary Materials

A

Appendix 1. Picture Gallery

