



RESEARCH ARTICLE

Enhancing zinc bio-fortification in wheat through agronomic interventions

Dipak Gite, Saumya Sharma, Sushant Patil, Bhupendra Mathpal & Mayur Darvhankar*

School of Agriculture, Lovely Professional University, Punjab 144 001, India

*Email: mayur.21878@lpu.co.in



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Abstract

Around half of the world's agricultural soils, including sandy loam soils of Punjab region, are deficient in zinc (Zn), leading to decreased crop production and nutritional value. Balanced application of macro nutrients along with Zn bio-fortification enhances crop productivity and reduces the deterioration of soil health. This study investigates the effect of zinc sulphate monohydrate ($\text{ZnSO}_4 \cdot \text{H}_2\text{O}$) on growth, yield and nutrient uptake of wheat (Var. PBW-343). The plants were subjected to the following treatments: T_1 (control), T_2 (100% recycling-derived fertilizers, RDF), T_3 (100% RDF + 5 kg $\text{ZnSO}_4 \cdot \text{H}_2\text{O ha}^{-1}$), T_4 (100% RDF + 10 kg $\text{ZnSO}_4 \cdot \text{H}_2\text{O ha}^{-1}$), T_5 (100% RDF + 15 kg $\text{ZnSO}_4 \cdot \text{H}_2\text{O ha}^{-1}$), T_6 (100% RDF + 20 kg $\text{ZnSO}_4 \cdot \text{H}_2\text{O ha}^{-1}$) and T_7 (100% RDF + two spray of $\text{ZnSO}_4 \cdot \text{H}_2\text{O}$ @ 0.5% at tillering and late jointing stages). The results revealed that the plant height, number of functional leaves per plant, number of tillers per plant, number of effective tillers per plant, spike length, number of grains per spike, test weight, grain yield and straw yield are significant and found maximum in treatment T_5 , which was 100% RDF and 15 kg $\text{ZnSO}_4 \cdot \text{H}_2\text{O ha}^{-1}$.

Keywords

bio-fortification; uptake; wheat; yield; zinc

Introduction

Wheat (*Triticum aestivum* L.) is the most widely cultivated annual plant from Poaceae family. It is the second most significant food crop after rice in terms of area and production (1). The estimated area used for wheat cultivation in world is 221.85 million hectares (Mha), with annual production of 776.10 million metric tonnes (MmT) and productivity of 3.50 MT ha^{-1} (2). India contributes a gross area of 31.36 Mha with annual production of 107.86 MmT and productivity of 3.44 MT ha^{-1} (2). Punjab contributes 3.53 Mha area and annual production is 17.14 MT with a productivity of 4862 kg ha^{-1} during the year 2020–21 (3).

Nutrient plays an important role in increasing production potential of wheat (4). Macronutrients and micronutrients have major role in growth and development of wheat crop. Each nutrient has its specific importance and regulates many of the plant's metabolic processes. (4). A balanced application of nitrogen (N), phosphorus (P) and potassium (K) is needed in wheat to enhance the growth. However, micronutrients are essential plant nutrients important for healthy growth and increased yield. Micronutrients play specific roles and are required to regulate essential physiological functions in plants (5).

Most of Indian soils are deficient in micronutrient, Zn (6). Zn is one of the seven essential plant micronutrients which promote stronger emergence, quicker stand establishment, better root growth, greater plant vigour, protein synthesis, metabolism of carbohydrate and increased yield. Plant-available Zn is low in most of the cereal-growing areas. Thus, Zn application is essential in deficient soils of India to enhance cereal yield and Zn concentration in grain (7). Wheat often has low grain Zn content in some regions. (8). Another important factor that can significantly impact a person's real Zn intake is wheat processing, as it often results in the removal of the wheat's bran and aleurone layers. A significant amount of Zn is lost throughout various wheat processing technologies (8).

The present study devised different combinations of RDF and Zn to understand the effects of Zn on wheat plants by observing various parameters like plant height, number of functional leaves per plant, number of tillers per plant, number of effective tillers per plant, spike length, number of grains per spike, test weight, grain yield and straw yield.

Materials and Methods

Location and climatic condition

The field experiment was conducted at Experimental farm, Lovely Professional University during *Rabi* 2020–2021. The experimental location is located in sub-tropical region with hot summer and cool winter's weather condition. At crop growth stage, the temperature ranged from 4–37°C and annual rainfall ranged from 500–750 mm, with most falling during monsoon season (July to September)(Fig. 1).

Initial soil chemical properties

Soil for the experiment was collected from the depth of 0–15 cm from different places of the experimental site. The experimental soil had the following characteristics: soil pH (7.81), electrical conductivity (0.21dSm⁻¹), organic carbon

(0.46 % i.e 4.6 gm kg⁻¹ soil), available soil nitrogen (145.5 kg ha⁻¹), available soil phosphorous (11.25 kg ha⁻¹), available soil potassium (180.56 kg ha⁻¹) and DTPA (Diethylene triamine penta acetic acid) extractable Zn (0.21 mg kg⁻¹).

Experimental materials and treatment details

The certified seed of wheat PBW-343 was used for the present investigation with the seed rate of 100 kg ha⁻¹. PBW-343 is resistant to brown rust and moderately resistant to yellow rust. Average plant height is 100 cm and has high yield potential in Punjab region. The experiment had seven treatments and three replicates and it was set up using a Randomized Block Design (RBD). The treatments used were T₁ (control), T₂ (100% RDF), T₃ (100% RDF + 5 kg ZnSO₄.H₂O ha⁻¹), T₄ (100%RDF + 10 kg ZnSO₄.H₂O ha⁻¹), T₅ (100%RDF + 15 kg ZnSO₄.H₂O ha⁻¹), T₆ (100%RDF + 20 kg ZnSO₄.H₂O ha⁻¹) and T₇ (100% RDF + two sprays of ZnSO₄.H₂O @ 0.5% at tillering and late jointing stages). Except the control (T₁), all treatments received the RDF (N:P:K) at the rates of 120:60:30 kg ha⁻¹. A 50% dose of nitrogen and a 100% dose of potassium and phosphorus were applied at the time of seeding in the form of urea, single super phosphate and murate of potash. At 30 days after seeding (DAS), the remaining 50% dose of N in the form of urea was delivered. ZnSO₄.H₂O was applied into the soil in treatments: T₃, T₄, T₅ and T₆ whereas its foliar spray was done in treatment, T₇, at tillering and late jointing stages. Zn application to soil will guarantee its spread throughout the root zone and longer availability of the micronutrient to the plant, probably throughout the crop life cycle, thus providing a more sustained nutrient supply and increasing the possibility to solve Zn deficits of wheat.

Growth and yield parameters

Five randomly chosen plants from each plot were tagged and used for measuring plant height, number of leaves per plant, number of tillers per plant and the results were averaged. The yield parameters, i.e. effective tillers per plant,

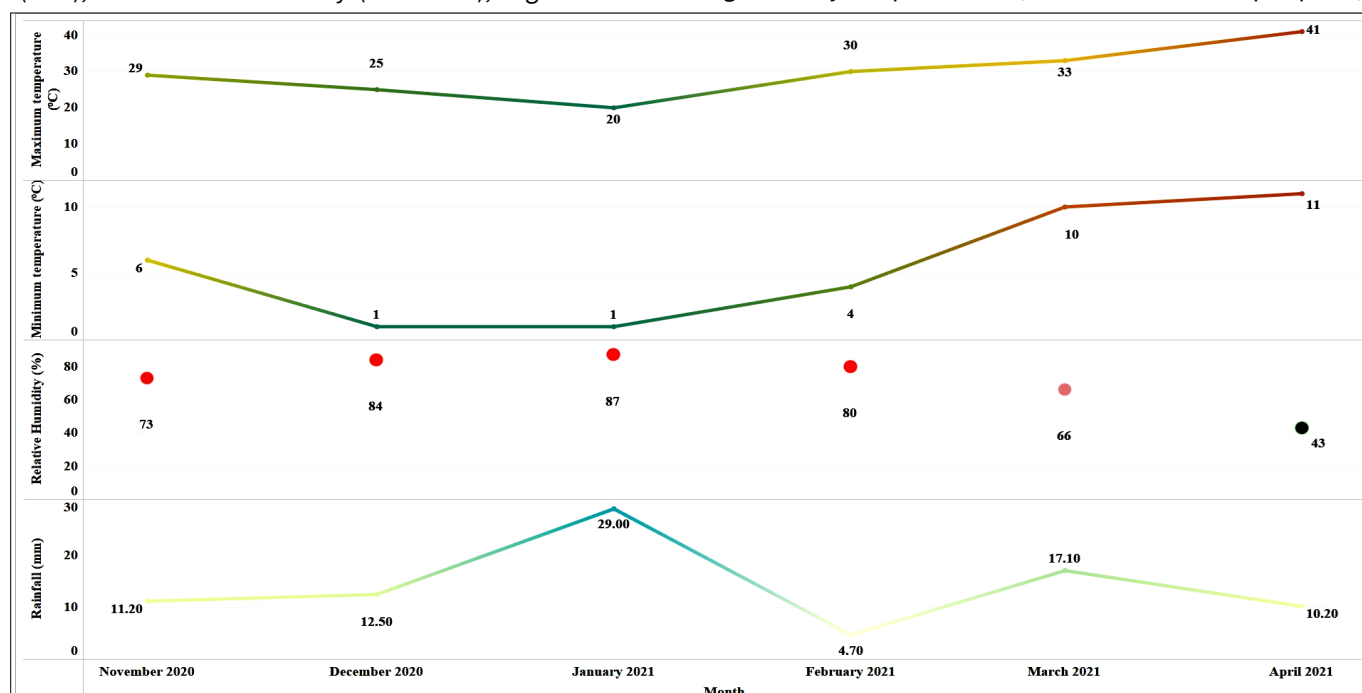


Fig. 1. Weather parameters during growing season of wheat crop.

spike length (cm) and number of grains per spike, was calculated from randomly selected 5 plants and then averaged. The wheat was harvested from net plot area and test weight (g), grain yield (q ha^{-1}) and straw yield (q ha^{-1}) were calculated after drying and threshing and converted into quintal ha^{-1} (9).

Nutrient uptake

The following formula was used to assess the uptake of specific nutrients (N, P, K & Zn) by comparing the concentration of each nutrient in each plot to the grain and dry matter yield at harvest in each plot (10):

$$\text{Nutrient intake (kg ha}^{-1}\text{)} = \text{Nutrient concentration (\%)} \times \text{Yield of grain or straw (kg ha}^{-1}\text{)} / 100$$

..... (Eqn. 1)

Statistical analysis

The data pertaining to the growth characteristics, yield characteristics and nutrient uptake of wheat crop was collected at regular intervals and analyzed statistically in RBD in web-based OPSTAT software.

Results

Growth parameters

Plant height (cm)

Plant height is one of the major growth parameters that is directly linked with yield contributing character of the plant. At harvest, plant height after treatment T_5 was recorded to be significantly highest (80.77 cm) than other treatments and treatment T_1 had the lowest plant height (46.86 cm). The plant height with treatment T_5 was found at par with treatments T_4 , T_6 and T_7 (Table 1, Fig. 2). It is observed that with the advancement of crop duration plant height also increased. The results indicated that minimum height of the plant was recorded under T_1 as com-

Table 1. Effect of zinc on growth parameters of wheat crop.

Treatments	Plant height (cm)	Number of leaves per plant	Number of tillers per plant
T_1	46.86 ^d	4.4 ^b	2.06 ^b
T_2	66.23 ^b	13.7 ^a	3.2 ^{ab}
T_3	55.97 ^c	13.8 ^a	3.4 ^a
T_4	75.96 ^a	14.1 ^a	3.46 ^a
T_5	80.77 ^a	14.3 ^a	3.93 ^a
T_6	74.02 ^a	14.2 ^a	3.33 ^a
T_7	76.98 ^a	11.9 ^a	3.26 ^a
SE (m)±	2.482	1.311	0.34
C.D 5%	7.732	4.085	NS

pared to other treatments, this might be low due to initial fertility status of that soil.

Number of leaves per plant

Number of leaves per plant was found significant at every periodical crop growth stage. At harvest stage, the number of leaves per plant was recorded to be the maximum in treatment T_5 (14.30) while minimum number of leaves per plant was recorded in the treatment T_1 (4.40). The number of leaves per plant recorded under treatment T_5 were found at par with rest of treatments except T_1 (Table 1, Fig. 2).

Number of tillers per plant

A crucial factor influencing spike production and grain yield is the tiller number. The data regarding number of tillers per plant was recorded at harvest stage. The effect of different levels of Zn along with RDF on number of tillers per plant was found to be statistically non-significant. Treatment T_5 had highest number of tillers per plant (3.93) and was found to be at par with rest of treatments except treatment T_1 which recorded the lowest number of tillers per plant (2.06) (Table 1, Fig. 2).

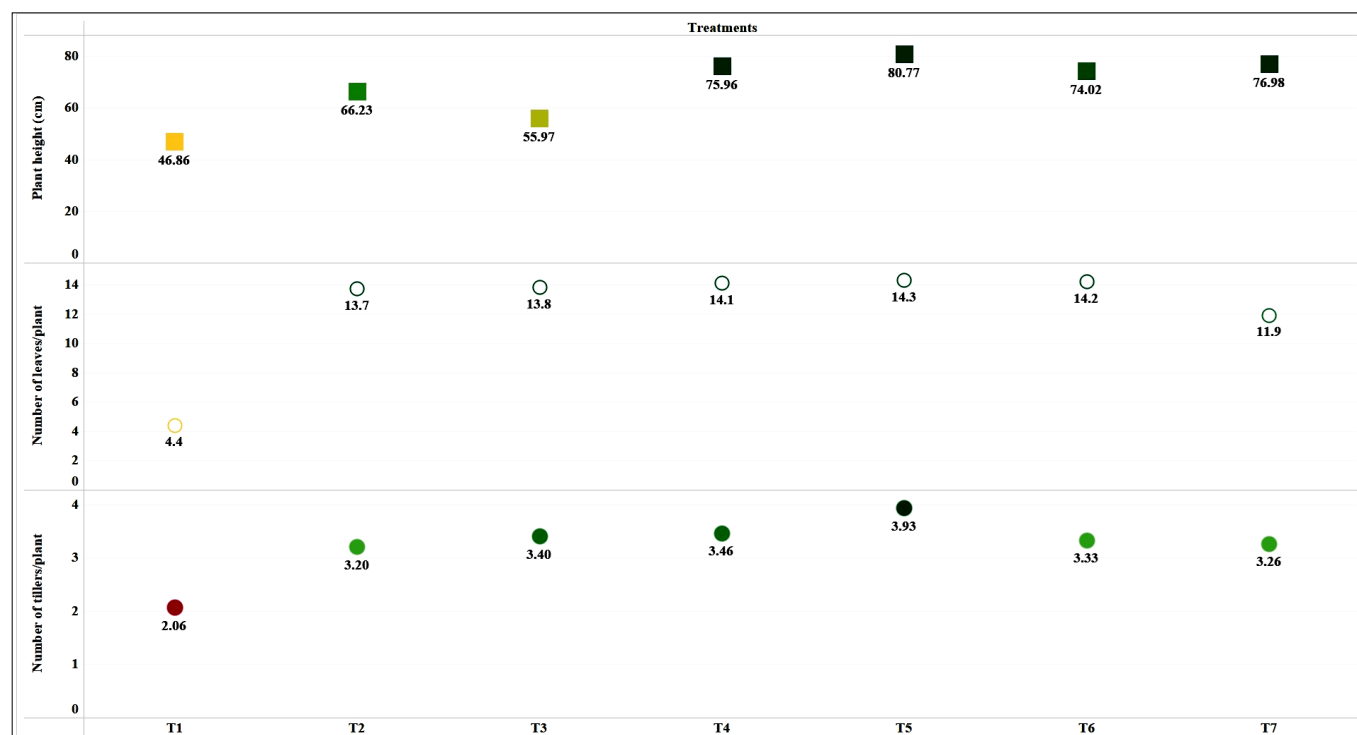


Fig. 2. Effect of zinc on growth parameters of wheat crop.

Yield parameters

Effective tillers per plant⁻¹

At harvest, effective tillers per plant was more under treatment T₅ (2.93) and that was found to be at par with rest of treatments except T₁. The T₁ treatment has least number of effective tillers per plant (1.03) (Table 2, Fig. 3). Various Zn levels showed significant differences in number of effective tillers at harvest.

Table 2. Effect of zinc on yield parameters of wheat crop.

Treatments	NET	LS	NGS	TW	GY	SY	HI
T ₁	1.03 ^b	6.16 ^b	22 ^c	35.6 ^e	7.62 ^d	9.91 ^c	43.25
T ₂	2.26 ^a	8.14 ^{ab}	33 ^b	37.8 ^{de}	36.87 ^c	50.88 ^b	41.95
T ₃	2.46 ^a	8.98 ^a	42.66 ^{ab}	38.5 ^{cd}	44.12 ^b	58.04 ^b	43.17
T ₄	2.66 ^a	9.99 ^a	43 ^{ab}	42.1 ^{ab}	51.61 ^a	69.67 ^a	42.51
T ₅	2.93 ^a	10.12 ^a	45.33 ^a	42.8 ^a	53.4 ^a	73.69 ^a	42.16
T ₆	2.4 ^a	9.55 ^a	42.33 ^{ab}	40.6 ^{abc}	41.05 ^{bc}	56.37 ^b	42.18
T ₇	2.26 ^a	8.63 ^a	36.33 ^{ab}	40.1 ^{bcd}	38.2 ^c	51.55 ^b	42.63
SE (m)±	0.327	0.657	2.997	0.766	1.643	3.009	-
C.D 5%	1.019	2.046	9.338	2.386	5.117	9.376	-

NET-Number of effective tillers per plant, **LS**-Length of spike, **NGS**-Number of grains per spike, **TW**-Test weight, **GY**-Grain yield, **SY**-Straw yield, **BY**-Biological yield, **HI**-Harvest index.



Fig. 3. Effect of zinc on yield parameters of wheat crop.

Spike length (cm)

Maximum spike length (10.12 cm) recorded under treatment T₅ which was at par with rest of treatments except T₁ and minimum length of spike (6.16 cm) was recorded under treatment T₁ at harvest (Table 2, Fig. 3).

Number of grains per spike

Grains per spike is crucial for determining the yield of wheat which contributes to final seed yield. At harvest, maximum number of grains per spike (45.33) was recorded under treatment T₅ which was at par with treatments T₃, T₄, T₆ and T₇ and minimum number of grains per spike (22.00) was recorded under treatment T₁ (Table 2, Fig. 3).

Test weight (g)

Highest test weight (42.80 g) was recorded under treatment T₅ which was found to be at par with treatments T₄ and T₆. The lowest value of test weight (35.60 g) was recorded in treatment T₁ (Table 2, Fig. 3). Zn is essential for growth hormone synthesis, formation of starch and maturation which increase the seed weight (8 and 11).

Grain yield (q ha⁻¹)

Zn application had a notable impact on grain yield; this is possible because wheat requires Zn as a necessary micro-nutrient. The data indicated that the highest grain yield was recorded under treatment T₅ (53.40 q ha⁻¹) which was at par with treatment T₄ and the lowest was recorded in treatment T₁ (7.62 q ha⁻¹) (Table 2, Fig. 3).

Straw yield (q ha⁻¹)

Different levels of Zn showed significant variation in straw yield. Highest straw yield was found under the treatment T₅ (73.69 q ha⁻¹) which was at par with treatment T₄ while

the lowest straw yield was found under treatment T₁ (9.91 q ha⁻¹) (Table 2, Fig. 3).

Uptake of nutrients

Nitrogen uptake (kg ha⁻¹)

Treatment T₅ shows the highest N uptake by wheat grains (90.78 kg ha⁻¹) and the straw (36.75 kg ha⁻¹) while the lowest value of N uptake by the wheat grains (11.81 kg ha⁻¹) and the straw (4.16 kg ha⁻¹) was found in T₁. Wheat showed maximum total N uptake (127.53 kg ha⁻¹) under T₅ and the least total N uptake (15.97 kg ha⁻¹) under T₁ (Table 3, Fig. 4).

Potassium uptake (kg ha⁻¹)

Treatment T₅ shows highest K uptake in wheat grains (18.69 kg ha⁻¹) and the straw (91.89 kg ha⁻¹) while the lowest K intake in wheat grains (2.13 kg ha⁻¹) and the straw (8.02 kg ha⁻¹) was found in T₁. Maximum total K intake in wheat (110.58 kg ha⁻¹) was found under T₅ and lowest K intake in wheat (10.15 kg ha⁻¹) was found in T₁ (Table 3, Fig. 4).

Zinc uptake (g ha⁻¹)

Treatment T₅ shows highest intake of Zn in wheat grains (146.95 g ha⁻¹) and the straw (72.65 g ha⁻¹) while the lowest value of Zn uptake by wheat grains (7.92 g ha⁻¹) and the

Table 3. Effect of zinc on nutrient uptake by wheat.

Treatments	Nitrogen uptake (kg ha ⁻¹)			Phosphorous uptake (kg ha ⁻¹)			Potassium uptake (kg ha ⁻¹)			Zinc uptake (g ha ⁻¹)		
	Grain	Straw	Total	Grain	Straw	Total	Grain	Straw	Total	Grain	Straw	Total
T ₁	11.81 ^e	4.16 ^c	15.97 ^e	1.75 ^d	1.78 ^c	3.53 ^d	2.13 ^c	8.02 ^e	10.15 ^d	7.92 ^e	4.75 ^d	12.67 ^e
T ₂	61.2 ^d	24.38 ^b	85.58 ^d	13.21 ^c	12.19 ^b	25.4 ^c	11.42 ^b	61.46 ^d	72.88 ^c	78.97 ^f	46.48 ^c	125.83 ^d
T ₃	74.12 ^b	27.85 ^b	101.97 ^b	15 ^b	13.92 ^b	28.92 ^b	14.55 ^b	71.38 ^b	85.93 ^b	104.25 ^c	55.19 ^b	159.44 ^c
T ₄	86.7 ^a	35.53 ^a	122.23 ^a	18.57 ^a	18.11 ^a	36.68 ^a	17.54 ^a	87.08 ^a	104.62 ^a	135.47 ^b	68.41 ^a	203.88 ^b
T ₅	90.78 ^a	36.75 ^a	127.53 ^a	19.75 ^a	18.37 ^a	38.12 ^a	18.69 ^a	91.89 ^a	110.58 ^a	146.95 ^a	72.65 ^a	219.6 ^a
T ₆	68.14 ^c	27.05 ^b	95.19 ^{bc}	13.95 ^{bc}	11.83 ^b	25.78 ^{bc}	13.13 ^b	69.89 ^{bc}	83.02 ^b	97.32 ^d	53.04 ^{bc}	150.36 ^c
T ₇	63.41 ^{cd}	27.74 ^b	91.15 ^{cd}	13.37 ^c	12.37 ^b	25.74 ^{bc}	11.84 ^b	62.89 ^{cd}	74.73 ^c	85.98 ^e	48.86 ^{bc}	134.84 ^d
SE (m)±	1.878	2.036	2.583	0.401	0.87	1.01	0.941	2.314	2.084	2.071	2.081	3.19
C.D 5%	5.851	6.344	8.046	1.251	2.71	3.145	2.932	7.209	6.493	6.453	6.484	9.938

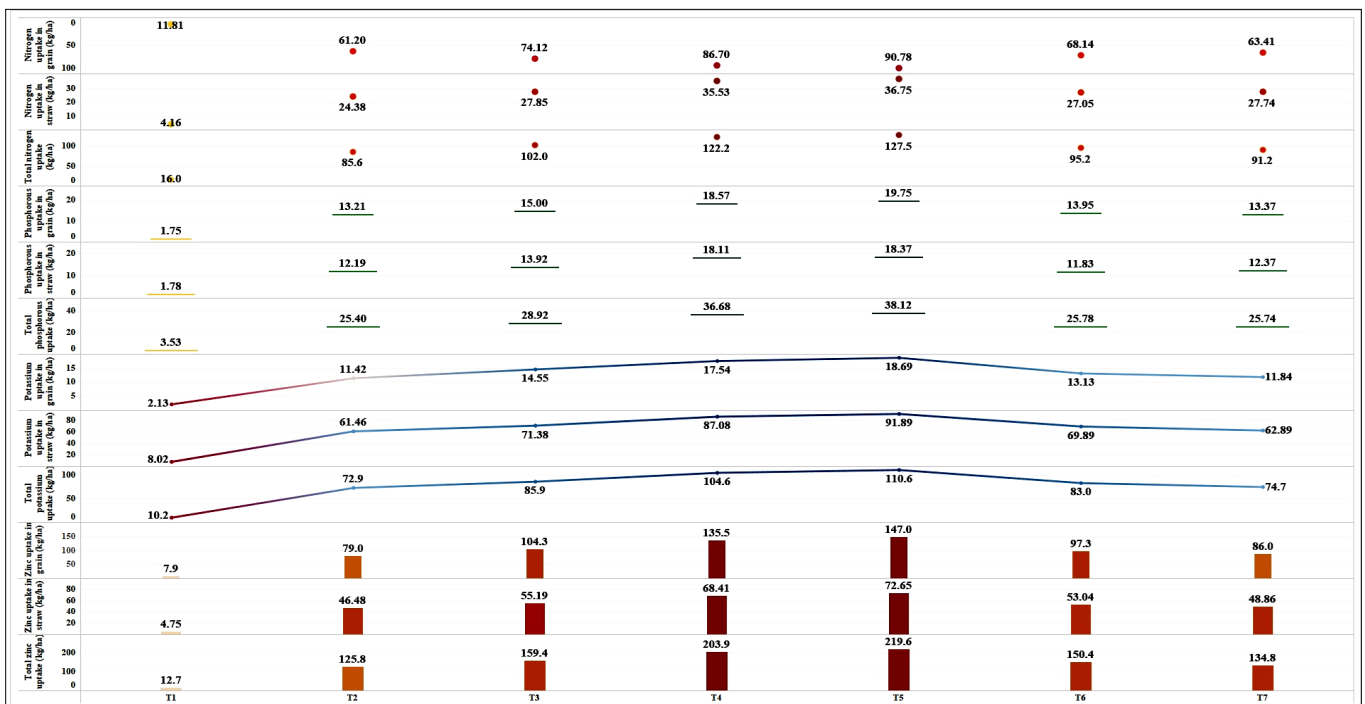


Fig. 4. Effect of zinc on nutrient uptake of wheat crop.

Phosphorous uptake (kg ha⁻¹)

Treatment T₅ shows highest intake of P in the wheat grains (19.75 kg ha⁻¹) and the straw (18.37 kg ha⁻¹) while the lowest uptake of P by wheat grains (1.75 kg ha⁻¹) and the straw (1.78 kg ha⁻¹) was found in T₁. Highest total P intake in wheat (38.12 kg ha⁻¹) was found under T₅ and lowest P intake in wheat (3.53 kg ha⁻¹) was found under T₁ (Table 3, Fig. 4).

straw (4.75 g ha⁻¹) was found under T₁. Maximum total Zn intake in wheat (219.60 g ha⁻¹) was found under T₅ and minimum total Zn intake in wheat (12.67 g ha⁻¹) was found under T₁ (Table 3, Fig. 4).

Discussion

The combined application of 100% RDF (120:60:30 kg NPK ha⁻¹) + 15 kg ZnSO₄.H₂O ha⁻¹ effectively enhanced the growth, yield and nutrient uptake in wheat. N and Zn help

in optimum plant growth, cell division and metabolism, which helps to increase plant height (11). Similarly, application of more N increases the plant height that may be attributed to vigorous vegetative growth which results in internodal extension and these observations corroborate with previous study and also the present study which reported maximum plant height due to ZnSO_4 (12–14). Increased number of leaves per plant is due to sufficient and continuous supply of major nutrients through the inorganic fertilizer and these fertilizers are responsible for prolific leaf production. The photosynthetic efficiency of crop is increased by nitrogen, which is one of the main constituents of chlorophyll, thus results in higher leaf counts. Similar trend of results was reported by other researchers (15).

Similarly, for yield attributing characteristics and yield, Zn treatment increases the number of effective tillers due to its function in numerous Zn-induced enzymatic activities and auxin metabolism, which influence plant growth and development (16). Increase in Zn levels caused considerable increase in number of effective tillers per plant (17). Observed number of effective tillers increased by the application of Zn and earlier findings reported similar results (18). Length of spike is important for the number of grains and their development in the spike of plant. Application of Zn increases the spike length (19). The result is in accordance with those of earlier studies (20, 21). Zn application results in an increase in grain setting because it participates in processes like pollination and fertilization, which have direct impact on formation of pollen tube (22). Similar results were described by other groups (14, 23). The test weight increased by Zn application because Zn increases the phloem movement from leaves to the roots, stem of the plant and developing grains (22). The present findings are similar to the previous reports (23). Increase in grain yield might be due to adequate amount of Zn, which has improved plant water relation, nutrients availability and enhanced cell physiology. Zn is a crucial element of enzymes, therefore an increase in yield may be caused by abundant efficiency of enzyme activities that affects the pigment of plants (18). Similar findings on grain yield were also observed by other research groups (23–25). Straw yield enhancement might be due to the balanced supply of Zn along with inorganic fertilizers which enhanced vegetative growth. Results are in accordance with the previous study (26).

Increased nitrogen uptake by wheat at various levels of Zn might be due to synergistic effect between nitrogen and Zn (27). Plants fertilized with Zn showed a much higher rate of N uptake (28). The nitrogen uptake by wheat grain and straw was increased due to the use of Zn in comparison to control (27). Similar results were obtained by the previous findings (14, 26). Antagonistic effects of high rates of Zn application on absorption and uptake of P have been reported in wheat (26). Observed phosphorous uptake was reduced at higher levels of Zn and the present study found similar results (29). For potassium uptake, findings of this study were similar to those obtained in the earlier outcomes (14). The Zn application increases its availability in soil and consequently in the plant system

that causes higher metabolism which resulted in increased uptake of nutrients (30). Earlier researchers reported similar results and found that the Zn fertilization increased Zn concentration and uptake in wheat (31).

Conclusion

This experimental study showed that the application of Zn along with N, P and K is important for various growth and yield contributing parameters of wheat. Furthermore, height of the plant, number of functional leaves per plant, tillers per plant, effective tillers per plant, grains per spike, length of spike, test weight (g), grain yield (q ha^{-1}) and the straw yield (q ha^{-1}) enhanced by the use of 100% RDF (120:60:30 kg NPK ha^{-1}) + 15 kg $\text{ZnSO}_4 \cdot \text{H}_2\text{O ha}^{-1}$ and this treatment considered as the best among all treatments. The grain yield and straw yield reported under treatment T_5 were at par with treatment T_4 [100% RDF (120:60:30 kg NPK ha^{-1}) + 10 kg $\text{ZnSO}_4 \cdot \text{H}_2\text{O ha}^{-1}$]. Since soil suffered from severe Zn deficiency, applying 15 kg ha^{-1} of Zn chemical fertilizer ($\text{ZnSO}_4 \cdot \text{H}_2\text{O}$) with 100% RDF (120:60:30 kg NPK ha^{-1}) significantly enhanced the N, P, K and Zn uptake by wheat.

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Authors' contributions

DG carried out research work and SS helped in recording the field and lab observations. SP and BM checked and modified the draft of manuscript while MD devised the research work plan and shaped the final manuscript.

Compliance with ethical standards

Conflict of interest: All authors do not have any conflict of interests to declare.

Ethical issues: None

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