



RESEARCH ARTICLE

# Effect of different modes of zinc fertilization on growth, yield and economics of quality protein maize under rainfed conditions of Jammu

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## Abstract

A field experiment was conducted to evaluate the effect of different modes of zinc fertilization on quality protein maize, to assess the response of zinc on growth, yield attributes, yield and nutrient uptake in quality protein maize. The experiment was laid out in a randomized block design and was replicated thrice. The soil of the experimental field was sandy loam in texture, near to neutral in reaction, low in organic carbon, available nitrogen, potassium and zinc but medium in available phosphorus. The experimental results revealed that seeds treated with zinc-sulphate @ 4g zinc per kg of seed + 1.5% foliar spray with ZnSO<sub>4</sub> at knee high and silking stage recorded significantly higher growth, yield attributes and yield, which was statistically at par with seeds treated with ZnSO<sub>4</sub> @ 4g/kg of seed + 1% foliar spray of ZnSO<sub>4</sub> at knee high and silking stage and seed treatment with ZnSO<sub>4</sub> @ 2g/kg of seed + 1.5% foliar spray of zinc-sulphate at knee high and silking stage. In terms of economics, the similar treatment (zinc-sulphate @ 4g/ kg of seed + 1.5% foliar spray using zinc-sulphate at knee high and silking) recorded the highest gross returns, Net returns and B: C ratio, which was closely followed by zinc-sulphate @ 4g/ kg seed + 1% foliar spray of ZnSO<sub>4</sub> at knee high and silking stage. Hence, seed treatment with zinc sulphate @ 4g/ kg of seed + foliar application of zinc sulphate @ 1.5% emerged as an effective approach in enhancing growth and yield of quality protein maize under rainfed conditions.

## Keywords

fertilization; foliar application; quality protein maize; seed treatment; zinc

## Introduction

Maize is an important food crop after rice and wheat, contributing to nearly 9 percent of the national food basket. In India, maize crop is cultivated on an area of about 10.04 million hectares with production of 33.62 million tonnes and productivity of 3.34 tonnes/hectare (1). In the Union Territory of Jammu and Kashmir, maize is cultivated on an area of about 276.36 thousand hectares with a production of 5470.01 thousand quintals and productivity of 19.79 quintals/hectare. Out of 276.36 thousand hectares, maize is cultivated over an area of about 207.05 thousand hectares in Jammu region with the production of 4239.83 thousand quintals and productivity of about 20.48 quintals/hectare (2). Although maize is rich in protein, fat, carbohydrates and certain essential vitamins and minerals, it lacks two essential amino acids, lysine and tryptophan, which lowers its biological value. By introducing the

opaque-2 gene into quality protein maize, the quantity of these inadequate amino acids has been raised (3). Therefore, farmers can produce maize grains with greater nutritional value by cultivating quality protein maize. Among the many variables, crop nutrition has a significant impact on maize grain yield as well as other significant growth and yield characteristics. Zinc is the most crucial micronutrient for crops due to its essential function as a co-factor in the plant's enzyme system. Zinc deficiency is currently so common that, in intensive cropping systems, it ranks second next to nitrogen (4). However, insufficient zinc levels can cause physiological stress in plants, leading to malfunctions in many enzyme systems and other metabolic processes. Over the last three decades, intensive cropping has resulted in the continuous and indiscriminate use of high-analysis fertilizers without adding micronutrients. Additionally, the neglect of on-farm inputs such as crop residues, organic manures and other materials, particularly those that are nearly neutral to alkaline in reaction, has manifested widespread zinc deficiency and significantly reduced crop yield. Zinc is an essential micronutrient that supports many physiological processes in plants, including protein synthesis, gene expression, enzyme structure, energy production, Krebs cycle, carbohydrate metabolism, photosynthesis, chlorophyll formation, auxin metabolism, pollen formation, resistance to pathogen infection and crop yield. Hence, zinc has a significant impact on crop output, both qualitatively and quantitatively (5,6). As zinc is essential for many physiological processes in biological systems, including DNA synthesis and cell division, it is also known as the "Metal of Life" in humans. This is due to its role in immune system development and brain function (5).

Zinc deficiency, is a major problem in developing nations like India since it impacts soil, plant, animal and human resources, affecting the entire food chain. A zinc deficiency is estimated to affect over 50% of Indian agricultural soils, with maize being the most susceptible crop. Zinc concentrations in maize are typically below the critical level of 22 mg kg<sup>-1</sup>, which causes stunted growth, a shorter crop maturity period, low sterility and lower-quality grains (7,8). Even it has been reported that zinc deficiency reduced the quality and yield of maize by 25-35% thus possessing a significant impact on food security (7,5).

Agronomically, zinc can be applied via soil, seed treatment, foliar spray and seed priming or by dipping seedlings into a fertilizer solution. Micronutrient seed treatment enhances crop development and yield, promotes healthy crop stand and enriches grains with micronutrients. The benefit of seed treatment is its economic feasibility, as it reduces the micronutrient requirements for biofortification (9). Considering that seed treatment uses fewer nutrients than soil application, it's a better choice. Applying zinc to the soil can involve banding or distributing it beneath the seed. Zinc foliar application is promising to enrich zinc in the seed (10). But there is still a lack of information regarding the magnitude of this effect under the specific agro- ecological conditions of Jammu. The present study has been investigated to study the effect of different modes of zinc fertilization on quality protein maize under rainfed conditions of Jammu.

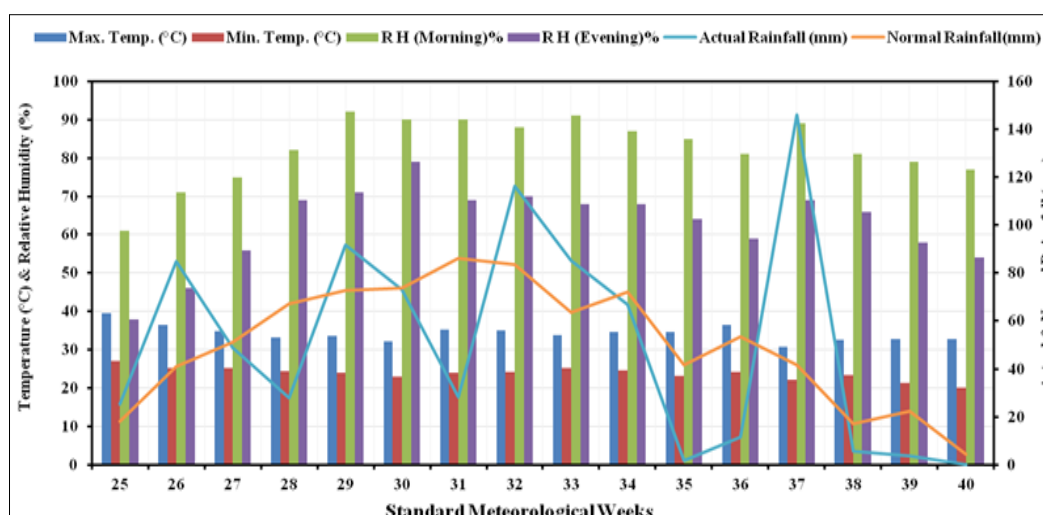
## Materials and Methods

### Location and soil of experimental field plot

The field study was conducted in the *Kharif* season 2023 at the Advanced Centre for Rainfed Agriculture (ACRA), Rakh Dhiansar, Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu. Located at 32°39' N latitude, 74° 53' E longitude and at an elevation of 332 meters above mean sea level, the site lies in the sub-tropical foothills of the Shiwaliks. The soil was sandy loam in texture, with a neutral pH (6.68), low organic carbon (2.65 g kg<sup>-1</sup>), limited available nitrogen (170.03 kg ha<sup>-1</sup>), potassium (102.43 kg ha<sup>-1</sup>) and zinc (0.50 mg kg<sup>-1</sup>), but moderate in available phosphorus (14.27 kg ha<sup>-1</sup>).

### Climatic condition

The climate of the experimental field comes under rainfed conditions. In general, the cropping season is characterized by hot and dry early summers followed by hot and humid monsoon seasons. The mean annual rainfall of the location varies from 1093 mm, out of which 70 -75 percent is received from June to September, whereas the remaining 25-30 percent of rains are received in a few showers during winter. The graphical representation of meteorological data of the experimental site is presented in Fig. 1.



**Fig. 1.** Graphical representation of meteorological data of quality protein maize grown during *Kharif* season 2023.

### Agronomic Practices and Fertilizer Application

The HQPM-1 cultivar of maize was sown by the dibbling method. As maize is the staple food of Jammu, its cultivation is suitable for rainfed conditions of Jammu. The spacing was maintained at a row-to-row of 75cm × plant-to-plant 20cm and a gross plot size of 6m × 4m. Seed treatment was done by mixing ZnSO<sub>4</sub> in viscous jaggery solution in a ratio of 1:1. The quantified amount of seed was then treated with a prepared solution and was dried in the shade before sowing (10). The crop was fertilized with 60:40:20 kg ha<sup>-1</sup> N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O, where full doses of phosphorus and potassium along with the half quantity of nitrogen was applied as basal dose at the time of sowing and the remaining half dose of nitrogen was applied at 30 DAS. However, zinc-sulphate was applied as per the technical programme of the experiment.

### Observation Recorded

The observations for various growth parameters, viz., plant height (cm) and dry matter accumulation (g plant<sup>-1</sup>), were recorded in periodic intervals of 30, 60 and 90 DAS at harvest. Five plants were tagged in each plot to measure the plant height, which was measured with the help of a meter scale from the ground base of the plant to the uppermost tip. For dry matter accumulation, plants were taken from the penultimate row and were cut closely to the soil surface from each plot. The samples were sundried and thereafter shifted in the oven to dry at a temperature of 65 ± 5°C till constant weight was achieved. Leaf area index (LAI) was recorded at an interval of 30, 60 and 90 DAS. The leaves are designated into small, medium and large categories and the number of leaves in each category was counted. Leaf area was determined with the help of the length and breadth method and it was calculated by using the formulae given by Watson, 1947 mentioned below:

Land area per plant = Row distance × plant distance

$$\text{Leaf area index (LAI)} = \frac{\text{Leaf area (cm}^2\text{)}}{\text{Land area (cm}^2\text{)}}$$

For yield attributes viz., number of cob plant<sup>-1</sup>, number of grains row<sup>-1</sup>, length of cob (cm), test weight (g) and number of grains cob<sup>-1</sup> were recorded at crop harvest. The total number of cobs plant<sup>-1</sup> was calculated from five tagged plants in each plot. The Number of grains in row<sup>-1</sup> was calculated by manually counting grains in each row of the cob. Similarly, the total number of grains cob<sup>-1</sup> was calculated by manually counting the grains. The cob length was measured from base to tip of the cob with the help of a measuring tape. For cob weight, cobs from five tagged plants were threshed, shade dried and their mean weight was calculated. In the case of cob weight, grains obtained from cobs of tagged plants were taken randomly and from it, 100 grains were counted and weighed.

To work out the yield. Grain yield was calculated by harvesting the cobs, sun-dried for 3-4 days and subsequently threshed (through a cob sheller) and cleaning. The grains thus obtained were weighed. The stover yield per net plot was recorded by subtracting the total grain weight from the total biomass. The Harvest index was computed by dividing the economic yield by the biological yield as per the formula

suggested by (11). The crude protein content in grain was calculated by multiplying nitrogen content (percent) in the grain by factor 6.25 and expressed in percentage (12).

The nitrogen, phosphorus, potassium and zinc uptake in grains and stover samples was calculated by multiplying the percent nutrient content by the respective dry matter accumulation.

The details of the methods employed for chemical analysis of plant samples are given below:

S. No.	Nutrient assessed	Method adopted
1	Nitrogen	Modified Kjeldhal's method
2	Phosphorous	Vanadomolybdo phosphoric acid yellow colour method
3	Potassium	Ammonium acetate method
4	Zinc	Atomic Absorption Spectrophotometer method

### Economic studies

For economic studies, various parameters viz., cost of cultivation, gross returns, net returns and B: C ratio, were recorded. The cost of cultivation was recorded by adding the expenditure involved in all the operations. Gross returns were calculated by multiplying the saleable products (maize grain and stover) by their respective sale prices. The net returns were computed by deducting the total cost of cultivation from the gross returns. To assess the economic feasibility of treatments, the benefit-cost ratio (B:C) was computed by dividing net returns by the entire cost of cultivation.

### Statistical Analysis

The experiment was conducted using a randomized block design and replicated thrice. The results were tested for the treatments' mean by applying an F-test of significance based on the null hypothesis (13).

## Results and Discussion

### Effect of different modes of zinc fertilization on growth of quality protein maize

The morphological parameters such as plant height, dry matter accumulation and leaf area index were noted to be influenced significantly by different modes of zinc application (Table 1). The results indicated that significantly higher plant height (221.50 cm) was recorded by seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed+ foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage recorded significantly higher plant height which was found to be statistically at par with seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed + foliar application of 1% ZnSO<sub>4</sub> at knee high and silking stage and seed treatment with ZnSO<sub>4</sub>@2g kg<sup>-1</sup> of seed + foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage. This might have happened because zinc is associated with tryptophan, which helps in the auxin synthesis, cell division and internode elongation, resulting in significantly higher plant height. Similar results were also reported by (14). Dry matter accumulation was increased significantly by various methods of zinc application, with significantly higher dry matter at harvest (167.70 g plant<sup>-1</sup>) by seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed+ foliar application of 1.5% ZnSO<sub>4</sub>

**Table 1.** Effect of different modes of zinc fertilization on growth of quality protein maize on plant height, dry matter accumulation and leaf area index

Treatments	Plant height (cm) (At harvest)	Dry matter accumulation (g plant <sup>-1</sup> ) (At harvest)	Leaf Area Index (90 DAS)
T <sub>1</sub>	184.99	140.47	2.01
T <sub>2</sub>	202.96	151.62	2.48
T <sub>3</sub>	185.84	141.97	2.03
T <sub>4</sub>	188.62	142.52	2.05
T <sub>5</sub>	185.75	140.90	2.02
T <sub>6</sub>	186.17	142.16	2.03
T <sub>7</sub>	189.51	142.34	2.04
T <sub>8</sub>	203.40	152.59	2.52
T <sub>9</sub>	203.51	152.68	2.55
T <sub>10</sub>	218.06	162.62	2.98
T <sub>11</sub>	203.58	154.84	2.56
T <sub>12</sub>	220.40	163.46	3.04
T <sub>13</sub>	221.50	167.70	3.06
SEm (±)	4.91	2.59	0.11
CD (5%)	14.31	7.56	0.32

at knee high and silking stage and was found to be statistically at par with seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed + foliar application of 1% ZnSO<sub>4</sub> at knee high and silking stage and seed treatment with ZnSO<sub>4</sub>@2g kg<sup>-1</sup> of seed + foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage. This might be attributed to the fact that zinc activates different physiological processes like stomatal regulation, chlorophyll formation, enzyme activation and biochemical processes, which resulted in significantly higher dry matter accumulation. These results are in close conformity with the findings of (15). Similarly, zinc contributes to the production of the substrates necessary for plant growth and development and functions as a coenzyme in photosynthesis (16).

Results of leaf area index revealed that a significantly higher leaf area index (3.06) was found through seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed+ foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage and was found to be statistically at par with seed treatment with ZnSO<sub>4</sub>@ 4g kg<sup>-1</sup> of seed + foliar application of 1% ZnSO<sub>4</sub> at knee high and silking stage and seed treatment with ZnSO<sub>4</sub>@ 2g kg<sup>-1</sup> of seed + foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage. This might be attributed to the foliar spray of zinc increasing leaf size by enhancing the length and width of the leaf, which led to a significant increase in LAI. Similar findings were reported by (10).

Different modes of zinc fertilization recorded significant yield attributing characters (Table: 2) viz. number of grains row<sup>-1</sup>, number of grains cob<sup>-1</sup>, cob length and cob weight. The results revealed that seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed+ foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage recorded significantly higher number of grains row<sup>-1</sup> (33.11), number of grains cob<sup>-1</sup> (319.04), cob length (22.08 cm) and cob weight (106.68 g) which was found to be statistically at par with seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed + foliar application of 1% ZnSO<sub>4</sub> at knee high and silking stage and seed treatment with ZnSO<sub>4</sub>@2g kg<sup>-1</sup> of seed + foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage. This might have happened due to the fact that zinc application enhanced photosynthetic activity that is involved in carbohydrate metabolism, protein synthesis and pollen formation, which led to a significant increase in yield attributes of quality protein maize. These results corroborate with the findings of (17).

Different methods of zinc application significantly improved both grain and stover yield (Table.3), seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed+ foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage recorded significantly higher grain yield (4298.75 kg ha<sup>-1</sup>) and stover yield ( 7159.77 kg ha<sup>-1</sup> ) which was found to be statistically at par with seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed + foliar application of 1% ZnSO<sub>4</sub> at knee high and silking stage and seed treatment with ZnSO<sub>4</sub>@2g kg<sup>-1</sup> of seed + foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage. This

**Table 3.** Effect of different modes of zinc fertilization on grain yield, stover yield and harvest index of quality protein maize

Treatments	Grain Yield (kg ha <sup>-1</sup> )	Stover Yield (kg ha <sup>-1</sup> )	Harvest Index
T <sub>1</sub>	3503.98	6237.20	35.96
T <sub>2</sub>	3849.89	6609.02	36.81
T <sub>3</sub>	3529.75	6278.36	35.98
T <sub>4</sub>	3548.26	6288.35	36.07
T <sub>5</sub>	3519.78	6263.54	35.97
T <sub>6</sub>	3531.94	6281.02	35.99
T <sub>7</sub>	3544.48	6284.35	36.06
T <sub>8</sub>	3865.09	6618.46	36.87
T <sub>9</sub>	3870.73	6623.80	36.88
T <sub>10</sub>	4179.25	6958.08	37.50
T <sub>11</sub>	3881.60	6634.12	36.91
T <sub>12</sub>	4180.54	6963.24	37.51
T <sub>13</sub>	4298.75	7159.77	37.52
SEm (±)	101.82	109.24	1.65
CD (5%)	297.20	318.84	NS

**Table 2.** Effect of different modes of zinc fertilization on yield attributing characters of quality protein maize

Treatments	No. of cobs plant <sup>-1</sup>	no. of grains row <sup>-1</sup>	No. of grains cob <sup>-1</sup>	Cob length (cm)	Cob weight (g)	100-grain weight (g)
T <sub>1</sub>	1.05	20.16	246.69	14.35	81.53	22.32
T <sub>2</sub>	1.06	24.72	278.85	16.97	94.46	22.43
T <sub>3</sub>	1.05	21.18	248.72	14.43	83.85	22.37
T <sub>4</sub>	1.05	21.34	255.49	14.74	86.93	22.42
T <sub>5</sub>	1.05	20.72	247.81	14.39	82.98	22.36
T <sub>6</sub>	1.05	21.25	249.55	14.49	85.40	22.40
T <sub>7</sub>	1.05	21.29	252.18	14.57	86.66	22.41
T <sub>8</sub>	1.06	25.31	280.54	16.99	95.83	22.44
T <sub>9</sub>	1.06	26.85	285.87	17.07	96.07	22.45
T <sub>10</sub>	1.07	30.93	307.42	20.50	103.72	22.47
T <sub>11</sub>	1.06	27.22	287.19	17.12	96.25	22.46
T <sub>12</sub>	1.07	31.96	310.49	21.57	104.47	22.48
T <sub>13</sub>	1.07	33.11	319.04	22.08	106.68	22.51
SEm (±)	0.06	1.07	6.71	0.75	2.35	0.49
CD (5%)	NS	3.13	20.14	2.20	6.87	NS

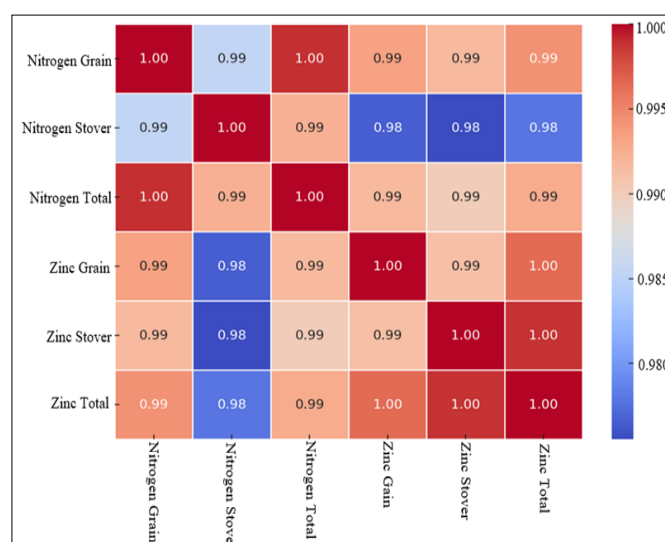


might have happened due to the higher physicochemical triggering of the biosynthesis of nucleic acids and proteins and the consequential enhancement of cell division besides the enhanced metabolic activity of the plants resulting in increased uptake of nutrients by the better root system. This could have possibly accounted for the improvement in crop performance; gaining more plant height and accumulation of dry matter helped in the the improvement in crop performance; gaining more plant height and accumulation of dry matter helped in the partitioning of photosynthates evenly towards the newly formed sink. Translocation of assimilates from source to sink helped obtain higher yield attributing characteristics, which was a direct function of yield and finally resulted in higher grain and straw yields. Similar results were also reported by (18, 19).

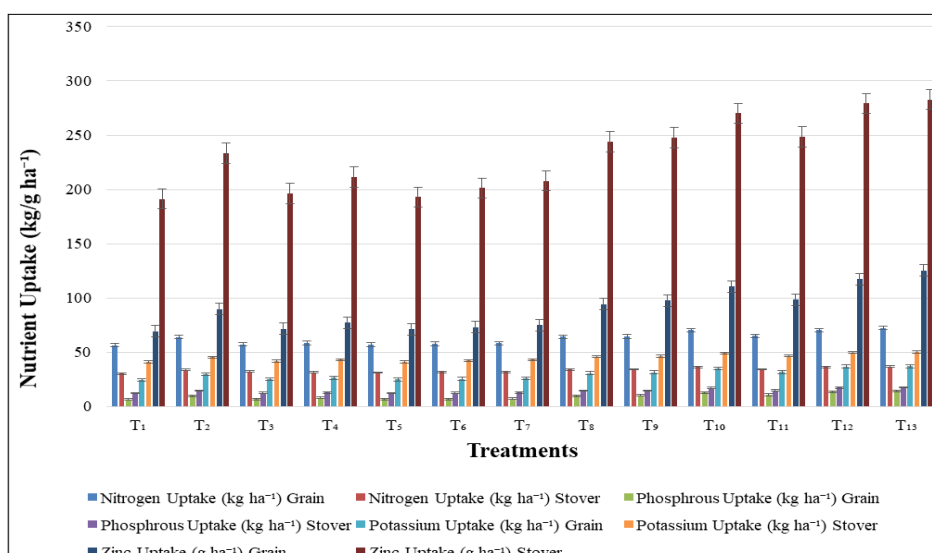
The results for nutrient uptake revealed that ), seed treatment with  $\text{ZnSO}_4$  @  $4\text{g kg}^{-1}$  of seed+ foliar application of 1.5%  $\text{ZnSO}_4$  at knee high and silking stage recorded significantly higher nutrient uptake (Fig. 3), which was found to be statistically at par with seed treatment with  $\text{ZnSO}_4$  @  $4\text{g kg}^{-1}$  of seed + foliar application of 1%  $\text{ZnSO}_4$  at knee high and silking stage and seed treatment with  $\text{ZnSO}_4$  @  $2\text{g kg}^{-1}$  of seed + foliar application of 1.5%  $\text{ZnSO}_4$  at knee high and silking stage. It might be due to the synergistic interaction between zinc and, nitrogen, potassium; many zinc-dependent enzymes are involved in carbohydrate metabolism in general and leaves in particular, impairment of potassium in stomata regulation, phloem export of assimilation from source to sink, maintained water balance in the soil-plant-atmosphere continuum. The nitrogen content and uptake in seed and stover could be due to zinc application, as zinc is essential for the synthesis of DNA and RNA and metabolism for the production of carbohydrates, lipids and proteins. The greater mobilization of phosphorus in the presence of nitrogen may be another reason for the higher phosphorus uptake. The increase in potassium content and uptake due to the interaction of potassium and zinc by the improvement of enzymatic activity and metabolic processes of plants, might have eventually made it easier to remove potassium. These results are in close agreement with the findings of (20, 21).

The correlation analysis reveals a strong positive relationship between nitrogen and zinc uptake across all components-grain, stover and total-with values ranging from 0.98 to 0.99 (Fig. 2). This indicates a synergistic effect: as nitrogen uptake increases, zinc uptake also rises proportionally. These findings suggest that practices enhancing nitrogen uptake may also improve zinc uptake, highlighting the importance of optimizing nutrients for better crop performance and more efficient nutrient management strategies.

The efficiency of various modes of zinc fertilization is finally decided in terms of economics. Results revealed that seed treatment with zinc sulphate @  $4\text{g kg}^{-1}$  seed+ foliar application of 1.5%  $\text{ZnSO}_4$  at knee high and silking stage recorded the highest cost of cultivation, gross returns, the net return and benefit-cost ratio (Table. 4). It is probably due to higher yields in this treatment that results in higher gross returns, net returns and benefit-cost ratio. These results closely conform with the findings of (10, 22).



**Fig. 2.** Correlation matrix between Nitrogen and Zinc Uptake in grain and stover of Quality Protein Maize.



**Fig. 3.** Effect of different modes of zinc fertilization on nitrogen ( $\text{kg ha}^{-1}$ ), phosphorus ( $\text{kg ha}^{-1}$ ), potassium ( $\text{kg ha}^{-1}$ ) and zinc uptake ( $\text{g ha}^{-1}$ ) by grain and stover of quality protein maize.

**Table 4.** Effect of different modes of zinc fertilization on the economics of quality protein maize

Treatments	Cost of cultivation (₹ ha <sup>-1</sup> )	Gross Returns (₹ ha <sup>-1</sup> )	Net Returns (₹ ha <sup>-1</sup> )	B: C Ratio
T <sub>1</sub>	28514	94047	65533	2.30
T <sub>2</sub>	29972	102600	72628	2.42
T <sub>3</sub>	29221	94725	65504	2.24
T <sub>4</sub>	29225	95153	65928	2.26
T <sub>5</sub>	30155	94466	64311	2.13
T <sub>6</sub>	30580	94780	64200	2.10
T <sub>7</sub>	31405	95059	63654	2.03
T <sub>8</sub>	30558	102955	72397	2.37
T <sub>9</sub>	30983	103092	72109	2.33
T <sub>10</sub>	31408	11078	79320	2.53
T <sub>11</sub>	30562	103357	72795	2.38
T <sub>12</sub>	30987	110771	79784	2.57
T <sub>13</sub>	31412	113903	82491	2.63

## Conclusion

In conclusion, seed treatment with ZnSO<sub>4</sub>@4g kg<sup>-1</sup> of seed + foliar application of 1.5% ZnSO<sub>4</sub> at knee high and silking stage significantly enhanced growth, yield attributes and yield along with improved economic results. Also, the synergistic interaction between zinc and other nutrients, particularly nitrogen and potassium, was observed with the same treatment, resulting in improved nutrient uptake and better crop performance. The results advocate for incorporating zinc fertilization as a key strategy for enhancing crop performance and profitability.

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## Authors' contributions

GK carried out the research part of the paper. NS carried out the corrections. APS participated in the modification. RS participated in the sequence alignment. PS participated in the table's arrangements. PSS helped in the rearrangement of the subheadings and finishing. JSM helped in arranging the sub topics. CL helped in searching relevant data. FF, HS and RB helped in the final assessment of the paper. All authors read and approved the final manuscript.

## Compliance with ethical standards

**Conflict of interest:** Authors do not have any conflict of interests to declare.

**Ethical issues:** None

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