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Synergistic effects of bio-stimulant and nutrient application on growth, yield, and grain quality of chickpea (*Cicer arietinum* L.) grown under rain-fed environment

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Improving chickpea productivity under rain-fed environment requires nutrient management strategies that enhance growth, yield stability, and grain quality. A two-year field study (2019–20 and 2020–21) evaluated the effectiveness of integrated application of bio-stimulant (Actibion), macro (NPK) and micro (Zn and B) nutrients on chickpea under rain-fed environment. Foliar spray of Actibion @ 1250 mL ha⁻¹, NPK (16 : 18 : 18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution were applied independently or in combination at flower initiation stage in comparison to control (no spray) and water spray. In both studied years, foliar application of Actibion, macro and micro-nutrients either applied individually or in combination significantly enhanced growth, yield and quality of chickpea. However, the integrated application of Actibion @ 1250 mL ha⁻¹, NPK (16 : 18 : 18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution had a more pronounced impact than all tested treatments, enhanced the average plant height (55.75%) nodulation (49.87%), crop growth rate (49.17%), seed yield (54.52%) as compared to control during both years of study. Nutrient content analysis showed that highest nitrogen, phosphorus, potassium, zinc, boron and protein contents were observed under integrated application. This treatment also yielded the highest net income, demonstrating both agronomic and financial benefits than rest of the treatments. In light of these results, we recommend the integrated application of Actibion @ 1250 mL ha⁻¹, NPK (16 : 18 : 18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution, as an effective strategy for improving productivity and nutritional quality of chickpea under rain-fed environment.

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1 Introduction

Chickpea (*Cicer arietinum* L.) is one of the most important grain legumes cultivated in semi-arid and arid regions, where it serves as a major source of protein (23.3–28.9%), carbohydrates (61.5%), fats (4.5%), vitamin B, and mineral sources for human nutrition.^{1,2} In addition to its nutritional importance, chickpea contributes to soil fertility through biological nitrogen fixation, making it a key component of sustainable cropping systems.³ However, chickpea productivity under rain-fed conditions remains low and unstable due to erratic rainfall, poor soil fertility, and limited nutrient availability, particularly in arid

and semi-arid regions.⁴ A substantial gap exists between potential and actual yield of this crop which may be due to many constraints such as inadequate nutrient management, drought stress, heavy insect pests and disease infestation. To address these challenges, new strategies for sustainable crop cultivation have been explored.

A range of physio-chemical, biological, and integrated approaches are available to minimize the yield losses.⁵ Among these approaches, balanced nutrient management has a critical role in improving chickpea growth and yield under rain-fed environment.⁶ The use of organic bio-stimulants and inorganic nutrient inputs are observed as effective methods to mitigate the yield losses.⁷ Bio-stimulants enhance nutrient uptake efficiency, improve root architecture, stimulate physiological processes, and increase plant tolerance to abiotic stresses which is particularly relevant under rain-fed conditions.⁸ One of the most promising bio-based compounds is Actibion which has bio-chemical qualities; it contains nearly all of the key nutrients (aspartic acid 1.21, alanine 0.52, cystine 0.78, arginine 1.73, glycine 0.78, isoleucine 0.59, glutamic acid

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3.14, leucine 0.78, lysine 0.72, histidine 0.22, phenylalanine 0.54, proline 1.15, methionine 0.08, serine 1.51, tyrosine 0.26, threonine 1.14 and valine 0.85). Actibion has a positive effect on the yield of important crops.⁹ While bio-stimulants alone can positively influence crop performance, their integrated application with macro and micro-nutrients may produce synergistic effects by improving nutrient availability, uptake, and utilization efficiency. Macro-nutrients, including nitrogen (N), phosphorus (P) and potassium (K), are necessary to support vegetative growth, energy transfer, root development and assimilate partitioning.¹⁰ Although chickpea has nitrogen-fixing capabilities, supplemental N is normally needed during the initial stages of growth, as well as in high-yielding varieties, whereas P and K deficiency usually limit nodulation, flowering, and grain development at rain-fed conditions.¹¹ Besides the macronutrients, the role of the micronutrients like boron (B) and zinc (Zn) are also being considered as yield-limiting factors in legume production. B plays an essential role in cell wall development, pollen integrity as well as the seed setting, and Zn controls the enzyme activity, synthesis of auxins, as well as photosynthetic efficiency.^{12,13} Arid or semi-arid soils are usually alkaline and calcareous with a large content of calcium carbonate, that significantly reduces the solubility and plant accessibility of Zn and B.^{14,15} In contrast, the availability of other micro-nutrients are generally less affected under these conditions. Chickpea yield is particularly vulnerable to Zn and B deficiencies, especially during the flowering and pod-development stages, and the crop is more sensitive to shortage of these nutrients.¹⁶

Most of the previous studies have evaluated the effects of individual application of bio-stimulant, macro and micro-nutrients in legumes.^{17–19} Synergistic analyses of multiple nutrient sources under true rain-fed conditions are lacking. More importantly, no published work has investigated the integrated use of amino-acid-based bio-stimulant (Actibion), macro and micro-nutrients applied at flowering stage of chickpea. By integrating two years of field data with physiological assessments, nutrient profiling, and economic analysis, this study provides the first field-based evidence to evaluate whether the combined application of these inputs confers synergistic advantages over their individual use in improving chickpea growth, yield, quality, and economic returns under rain-fed agro-ecosystems.

2 Material and methods

2.1 Experimental location and site description

In the growing seasons of 2019–20 and 2020–21, two independent field experiments were carried out at “Thal” region of district Mianwali, Punjab, Pakistan, between 32.36 °N latitude and 71.69 °E longitude at an elevation of 200 meters (Fig. 1A). The area is located in agro-ecological zone III, and is characterized by sandy soils and an arid to semi-arid climate, with a minimum average temperature of 11 °C and 11.2 °C and a maximum average temperature of 22 °C and 21.7 °C respectively, in crop growing seasons of 2019–20 and 2020–21. However, the maximum and lowest temperature (°C) and

rainfall (mm) data throughout the crop growing periods are presented in Fig. 1B. The physio-chemical study of soil showed that soil at the experimental site was dominantly sandy in texture. The detailed soil properties of both growing seasons are described in Table 1.

2.2 Experimental design and treatments

Field experiments were conducted with a randomized complete block design (RCBD) with four replications. The current study consists of nine treatments: T_1 = control (no-spray), T_2 = water spray, T_3 = Actibion @ 1250 mL ha⁻¹, T_4 = NPK (16 : 18 : 18) @ 3% solution, T_5 = B @ 0.2% + Zn @ 0.3% solution, T_6 = Actibion @ 1250 mL ha⁻¹ + NPK (16 : 18 : 18) @ 3% solution, T_7 = Actibion @ 1250 mL ha⁻¹ and B @ 0.2% + Zn @ 0.3% solution, T_8 = NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution, T_9 = Actibion @ 1250 mL ha⁻¹ and NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution. The bio-stimulant used was “Actibion”, which is a commercial product and containing a blend of amino acids including aspartic acid (1.21), alanine (0.52), cystine (0.78), arginine (1.73), glycine (0.78), isoleucine (0.59), glutamic acid (3.14), leucine (0.78), lysine (0.72), histidine (0.22), phenylalanine (0.54), proline (1.15), methionine (0.08), serine (1.51), tyrosine (0.26), threonine (1.14) and valine (0.85) @ 1250 mL ha⁻¹. The applied dose of 1250 mL ha⁻¹ was selected according to the manufacturer's recommended foliar rate for field crops and has been previously validated in agronomic trials as an effective and farmer-relevant concentration. While, the doses of NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution were selected because these concentrations ensure efficient absorption without causing phytotoxic effects in chickpea. The chickpea cultivar ‘Bittal-2016’ was used as the test crop. Certified seeds of this cultivar were obtained from the Pulses Research Institute, Ayub Agricultural Research Institute (AARI), Faisalabad, Punjab, Pakistan. “Bittal-2016” was used as it is recommended by the Pakistan Agriculture Research Council (PARC) for cultivation in rain-fed environment.

2.3 Experimental procedure

Soil was cultivated two times during the preparation of the seed bed with a tractor-mounted cultivator, after which planking was done. Single row hand drill at 60 kg ha⁻¹ seed rate was used in 30 cm apart rows to spread the seed. Before sowing, the seeds were treated with fungicide. Immediately after crop emergence, overcrowded seedlings were thinned twice to minimize a 10 cm plant-to-plant spacing. Weeds were hoed at different intervals. The chickpea crop was entirely under rain-fed throughout the season, and no flood irrigation was applied. The foliar application of Actibion, macro (NPK) and micro (B and Zn) nutrients were applied according to the respective treatment plan at flower initiation stage. Foliar sprays were applied using a knapsack sprayer calibrated to 175 deliver 300 L ha⁻¹, corresponding to 4.5 L per plot. Sprays were applied uniformly until foliage was fully wetted but without runoff. In arid conditions, foliar fertilizer application is the most effective method to meet plant nutrient demands throughout the growing season and the flowering stage incenses the grain production of pulses. The



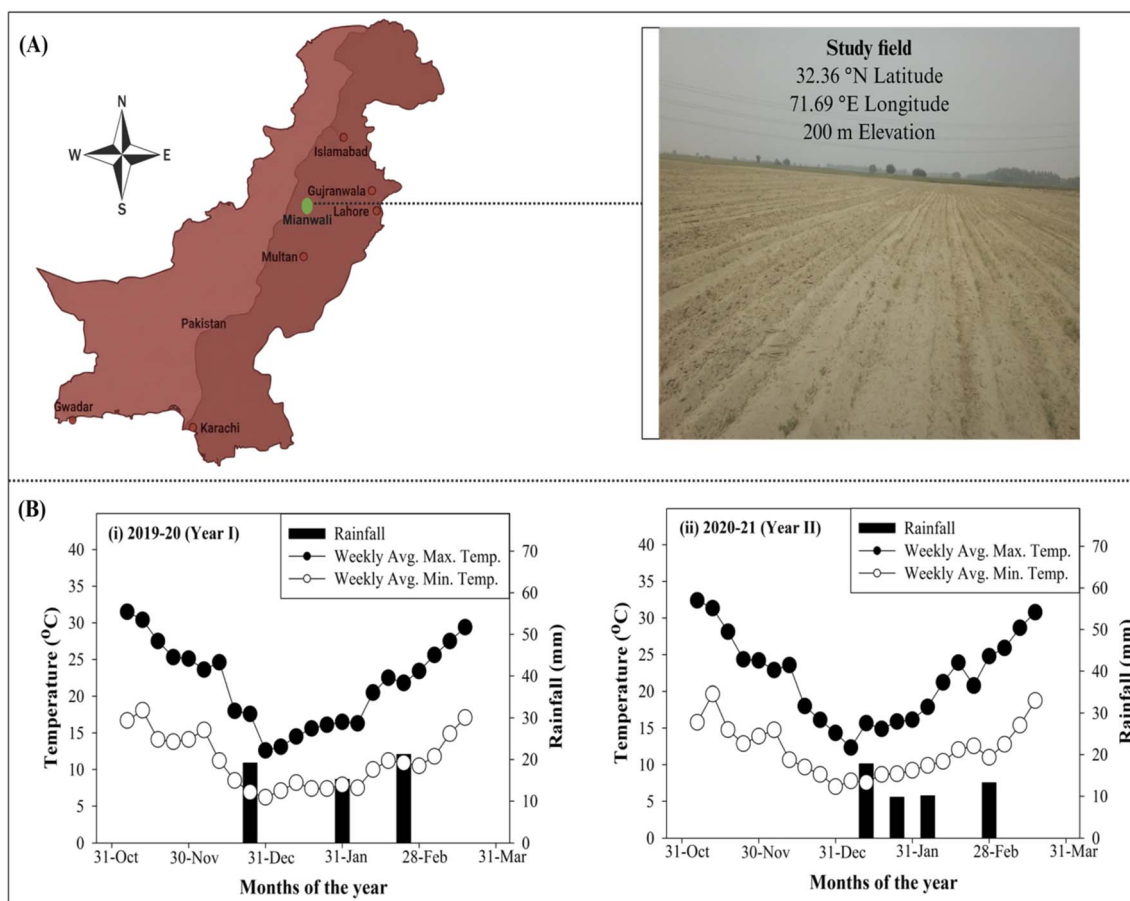


Fig. 1 (A) Geographical location of experimental site in Mianwali, Punjab, Pakistan. (B) Weekly average maximum or minimum temperature and rainfall of experimental site throughout the chickpea growing seasons (i) 2019–20 (Year I) and (ii) 2020–21 (Year II).

first experiment was initiated on October 10th, 2019, and plants were harvested on March 15th, 2020. The second experiment was a repeat of the 1st-year experiment, done on November 3rd, 2020, and harvested on March 28th, 2021. After washing the roots with distilled water and drying, the plant samples were stored in paper bags for growth, yield, and quality analysis.

Table 1 The physico-chemical properties of the experimental field during growing seasons of 2019–20 (Year I) and 2020–21 (Year II)

Characteristics	Values	
	Year I	Year II
Sand (g kg^{-1})	839	850
Silt (g kg^{-1})	91	84
Clay (g kg^{-1})	70	66
Soil texture class	Sandy soil	Sandy soil
Soil pH	8.51 ± 0.56	8.42 ± 0.48
EC (dS m^{-1})	3.19 ± 0.25	3.10 ± 0.31
Saturation percentage	20.8 ± 1.18	18.2 ± 1.05
Organic matter (%)	0.23 ± 0.12	0.20 ± 0.09
Organic C content (%)	0.134 ± 0.08	0.128 ± 0.06
Total soil N content (%)	0.012 ± 0.04	0.013 ± 0.03
Available P (mg kg^{-1})	3.86 ± 0.55	3.75 ± 0.43
Extractable K (mg kg^{-1})	207 ± 2.56	195 ± 3.12

Harvesting of chickpea was done when approximately 90% of the pods had reached maturity, and grains were separated manually.

2.4 Measurement of growth and yield attributes

During both years, the growth and yield traits such as plant height (cm), fresh and dry weight (g), number of seeds plant^{-1} , number of nodules plant^{-1} , 100-seed weight (g), biological yield (kg ha^{-1}), and harvest index (%) were recorded according to standard procedures. For seed yield (kg ha^{-1}), entire treatment plots were manually harvested by sickle, weighed on digital field balance, and converted in kg ha^{-1} . For the calculation of the leaf area index, the leaf area (one surface only) is divided by the land area occupied by plants.²⁰

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

Leaf area duration (days) was determined at 30, 60, and 90 days after sowing (DAS) in accordance with the methodology established by ref. 21.

$$\text{LAD} = \frac{\text{LAI1} - \text{LAI2}}{2} \times (t_2 - t_1) \text{ days}$$



where, LAI = leaf area index, t = time.

Crop growth rate ($\text{g m}^{-2} \text{d}^{-1}$) was calculated at 45, 90 DAS, and at the harvesting stage. It is expressed in $\text{g m}^{-2} \text{d}^{-1}$, as suggested by²² formula.

$$\text{CGR} = \frac{(W_2 - W_1)}{(T_2 - T_1)}$$

where, W_1 = initial dry weight, W_2 = final dry weight, T_1 = date of previous sampling, T_2 = date of current sampling.

The total chlorophyll content (SPAD) was measured between 9:00 and 10:00 am *via* a chlorophyll meter (SPAD-502 Konika Minolta Sensing, Inc., Japan).

2.5 Estimation of mineral elements in seed and plant

Grain samples were oven-dried and ground into a fine powder using a Willey mill (Model 4, Thomas Scientific, U.S.A.) before nutrient analysis. Grain nitrogen content (%) was determined using the Kjeldahl method as described by ref. 23. The percentage of nitrogen was calculated using the formula:

$$\text{Nitrogen}(\%) = \frac{(V_1 - V_2) \times N}{W} \times 1.4$$

where, V_1 is the volume of acid used for the sample (mL), V_2 is the volume of acid for the blank (mL), N is the normality of standard acid, W is the sample weight (g).

Phosphorus content (%) was determined following,²³ using the formula.

$$\text{Phosphorus}(\%) = (\text{ppm } P \times V_1/W_t) \times (100/V_2) \times (1/10\,000)$$

where, V_1 is the total digest volume (mL), V_2 is the digest volume used for measurement (mL), W_t is the dry sample weight (g).

Potassium content (ppm) was measured using flame photometry (PFP7, Jenway, United Kingdom) according to²³ using the formula.

$$\text{Potassium}(\text{ppm}) = \frac{C \times V}{W} \times 100$$

where, C is the potassium concentration from the calibration curve, V is the final volume of digested sample (mL), W is the sample weight (g).

Protein content (%) in grain was estimated using the Lowry method for protein determination.²⁴ Zinc content was measured using an atomic absorption spectrophotometer (AAnalyst 200, PerkinElmer, U.S.A.), while boron content was estimated through flame photometry at 420 nm.²³

2.6 Economic analysis

To examine the economic analysis, all the agronomic operations during both years were considered, and then the total cost of production was estimated. The benefit-ratio was determined by dividing the gross income by the total cost of cultivation under each treatment.

2.7 Statistical analysis

For statistical analysis, the Statistix 8.1 computer software (Statistix 8.1, Analytical Software, Tallahassee, FL, USA) was

used. To compare the individual treatment means, Tukey's Honest Significant Difference (HSD) test was done at the 5% probability level.²⁵ The results were presented as mean values \pm standard error ($n = 3$). The data were graphically demonstrated using the Sigma plot 11.0 (Systat Software, Inc., San Jose, CA, USA) and Biorender Softwares.

3 Results

3.1 Growth and yield parameters

Foliar application of bio-stimulant (Actibion), macro (NPK), and micro (B and Zn) nutrients applied individually or in combinations significantly ($p \leq 0.05$) affected the growth and physiological traits of chickpea in both growing seasons (Year I and II) as described in Table 2. During both years of study, the integrated application of Actibion @ 1250 mL ha⁻¹, NPK (16 : 18 : 18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution produced the tallest plants, recorded 75.50 and 76.50 cm in Year I and Year II, respectively, followed by Actibion @ 1250 mL ha⁻¹ + NPK (16 : 18 : 18) @ 3% solution. Whereas, the no-spray (control) resulted in the shortest plants (42.25 and 42.50 cm in Year I and Year II, respectively). Maximum fresh weight (127.50 and 128.00 g) and dry weight (48.75 and 46.50 g) were obtained under the combined treatment, while the lowest values of fresh weight (74.25 and 75.25 g) and dry weight (25.03 and 26.00 g) were observed under control (no-spray) in Year I and Year II, respectively. Similarly, nodulation was maximized under the combined treatment, followed by the foliar spray of Actibion @ 1250 mL ha⁻¹ + NPK (16 : 18 : 18) @ 3% solution whereas the control produced the least number of nodules.

Significant differences were observed in leaf area index (LAI), leaf area duration (LAD), crop growth rate (CGR), and chlorophyll content due to foliar treatments across both growing seasons (Table 3). The control (no-spray) and water spray treatments consistently exhibited the lowest values for all growth parameters in both years. The negligible differences between these two treatments indicated that improvements in plant growth were primarily due to nutrient application rather than the spraying effect. Among the individual treatments, Actibion @ 1250 mL ha⁻¹ and NPK (16 : 18 : 18) @ 3% solution significantly enhanced the LAI, LAD, CGR, and chlorophyll content compared to the control (no-spray). Actibion treated plants recorded slightly higher values than those receiving NPK alone, suggesting a stronger influence of the bio-stimulant on physiological activity and canopy development. In contrast, the micro-nutrient treatment (B @ 0.2% + Zn @ 0.3% solution) showed moderate improvements, particularly in chlorophyll content, but its effect on structural growth parameters remained comparatively lower.

Integrated applications of Actibion @ 1250 mL ha⁻¹, NPK (16 : 18 : 18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution produced the highest values for all growth parameters in both years. LAI reached 6.02 and 6.00, LAD 68.61 and 68.41 days, CGR 3.92 and 3.95 $\text{g m}^{-2} \text{d}^{-1}$, and chlorophyll content 55.97 and 55.68 in Year I and Year II, respectively. These values were significantly higher than all other treatments. The consistency of treatment effects across both growing seasons demonstrates





Table 2 Impact of foliar applied bio-stimulant (Actibion), macro (NPK), and micro (B and Zn) nutrients on plant height, fresh weight, dry weight, and number of nodules plant⁻¹ of chickpea cultivated under rain-fed conditions during the growing seasons of 2019–20 (Year I) and 2020–21 (Year II)^a

Treatments	Plant height (cm)		Fresh weight (g)		Dry weight (g)		Number of nodules plant ⁻¹	
	Year I	Year II	Year I	Year II	Year I	Year II	Year I	Year II
Control (no-spray)	42.25 ± 3.21i	42.5 ± 4.96i	75.25 ± 4.29h	74.25 ± 3.04h	26.0 ± 1.85h	25.03 ± 1.55i	14.75 ± 1.41g	15.00 ± 1.57g
Water spray	46.75 ± 3.56h	45.0 ± 3.25h	75.75 ± 2.63h	75.25 ± 3.35h	26.50 ± 1.56h	26.13 ± 1.45h	15.75 ± 1.25g	16.00 ± 1.35g
Actibion @ 1250 mL ha ⁻¹	68.50 ± 4.14e	69.5 ± 3.96e	116.0 ± 3.56e	116.5 ± 2.18e	41.50 ± 1.65e	40.00 ± 1.64e	21.0 ± 1.14de	22.00 ± 1.56e
NPK (16 : 18 : 18) @ 3% solution	66.50 ± 3.86f	67.75 ± 2.05f	112.0 ± 3.43f	111.0 ± 3.11f	39.00 ± 1.96f	38.25 ± 1.67f	20.0 ± 2.48e	21.00 ± 1.43e
B @ 0.2% + Zn @ 0.3% solution	62.00 ± 2.65g	65.50 ± 3.11g	89.75 ± 4.72g	90.50 ± 3.56g	30.25 ± 1.74g	30.75 ± 1.55g	18.5 ± 2.29f	18.50 ± 1.45f
Actibion @ 1250 mL ha ⁻¹ + NPK (16 : 18 : 18) @ 3% solution	74.00 ± 4.95b	74.75 ± 3.24b	123.7 ± 2.13b	124.7 ± 3.49b	46.75 ± 1.56b	44.75 ± 1.81b	26.50 ± 2.61b	29.75 ± 2.49b
Actibion @ 1250 mL ha ⁻¹ and B @ 0.2% + Zn @ 0.3% solution	71.00 ± 3.65d	71.25 ± 3.35d	118.5 ± 4.56d	119.2 ± 3.45d	43.25 ± 2.58d	41.50 ± 2.62d	21.75 ± 1.22d	24.75 ± 1.35d
NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution	72.50 ± 2.42c	73.00 ± 2.95c	120.75 ± 4.15c	122.0 ± 3.68 c	45.00 ± 2.61c	43.00 ± 1.96c	23.75 ± 1.71c	27.75 ± 2.67c
Actibion @ 1250 mL ha ⁻¹ and NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution	75.50 ± 3.68a	76.50 ± 3.16a	127.50 ± 3.25a	128.0 ± 4.31a	48.75 ± 2.75a	46.50 ± 2.32a	28.50 ± 1.75a	31.25 ± 1.86a
HSD (0.05)	1.42	1.71	2.12	1.87	1.72	0.95	1.33	1.49

^a Means sharing the same letter did not differ significantly at $p = 0.05$.

Table 3 Impact of foliar applied bio-stimulant (Actibion), macro (NPK), and micro (B and Zn) nutrients on leaf area index, leaf area duration, crop growth rate and chlorophyll content of chickpea cultivated under rain-fed conditions during the growing seasons of 2019–20 (Year I) and 2020–21 (Year II)^a

Treatments	Leaf area index (LAI)		Leaf area duration (days)		Crop growth rate (g m ⁻² d ⁻¹)		Chlorophyll content (SPAD)	
	Year I	Year II	Year I	Year II	Year I	Year II	Year I	Year II
Control (no-spray)	3.08 ± 0.56h	3.07 ± 0.25i	39.83 ± 2.52h	39.67 ± 1.96i	1.93 ± 0.62i	1.94 ± 0.58i	41.55 ± 2.18f	43.31 ± 2.24g
Water spray	3.32 ± 0.63g	3.33 ± 0.43h	40.47 ± 1.95h	40.51 ± 1.46g	2.28 ± 0.46h	2.3 ± 0.85h	43.23 ± 2.48e	43.32 ± 2.18g
Actibion @ 1250 mL ha ⁻¹	5.61 ± 0.52d	5.55 ± 0.42e	62.56 ± 2.05e	60.67 ± 2.35e	3.52 ± 0.58e	3.55 ± 0.46e	53.71 ± 2.05c	52.18 ± 1.83d
NPK (16 : 18 : 18) @ 3% solution	5.32 ± 0.46e	5.35 ± 0.35f	61.49 ± 3.21f	58.77 ± 3.12f	3.46 ± 0.81f	3.48 ± 0.43f	53.06 ± 1.58d	51.19 ± 2.01e
B @ 0.2% + Zn @ 0.3% solution	4.57 ± 0.36f	4.53 ± 0.29d	51.00 ± 2.59g	51.00 ± 2.85g	3.05 ± 0.69g	3.07 ± 0.76g	52.84 ± 1.69d	50.17 ± 2.33f
Actibion @ 1250 mL ha ⁻¹ + NPK (16 : 18 : 18) @ 3% solution	5.88 ± 0.29b	5.91 ± 0.31b	67.52 ± 2.56b	66.76 ± 3.12b	3.82 ± 0.49b	3.85 ± 0.55b	55.08 ± 2.19b	54.07 ± 1.86b
Actibion @ 1250 mL ha ⁻¹ and B @ 0.2% + Zn @ 0.3% solution	5.67 ± 0.49 cd	5.74 ± 0.55d	64.53 ± 3.54d	62.56 ± 3.19d	3.63 ± 0.71d	3.66 ± 0.39d	54.02 ± 1.69c	53.26 ± 1.58c
NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution	5.74 ± 0.35c	5.83 ± 0.85c	66.74 ± 3.41c	64.69 ± 2.58c	3.74 ± 0.39c	3.76 ± 0.48c	54.88 ± 1.81b	53.76 ± 1.19b
Actibion @ 1250 mL ha ⁻¹ and NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution	6.02 ± 0.41a	6.00 ± 0.96a	68.61 ± 2.96a	68.41 ± 3.10a	3.92 ± 0.72a	3.95 ± 0.68a	55.97 ± 1.65a	55.68 ± 1.92a
HSD (0.05)	0.12	0.07	0.76	0.32	0.05	0.06	0.56	0.43

^a Means sharing the same letter did not differ significantly at $p = 0.05$.

the stability and reliability of these responses under rain-fed conditions.

All yield components, including number of seeds plant⁻¹, 100-seed weight, seed yield, biological yield, and harvest index, were significantly ($p \leq 0.05$) affected by the foliar treatments (Table 4). The control and water spray treatments recorded the lowest values for all yield parameters in both years, confirming the limited productivity under untreated conditions. The highest number of seed (43.00 and 45.00 seeds plant⁻¹ in Year I and Year II, respectively) was recorded with combined application, compared with 26.75 in Year I and 27.25 seeds per plant in Year II under control. The maximum 100-seed weight was recorded under combined treatment, producing 20.50 g in Year I and 21.25 g in Year II, while the control recorded the minimum 100-seed weight during both years. In case of seed yield, the highest values of 2658.5 kg ha⁻¹ and 2624.8 kg ha⁻¹ in Year I and II, respectively was recorded with combined treatment, followed by the application of Actibion @ 1250 mL ha⁻¹ + NPK (16 : 18 : 18) @ 3% solution. In contrast, the lowest seed yield (1455.0 kg ha⁻¹ in Year I and 1426.3 kg ha⁻¹ in Year II) was observed under the control (no-spray) treatment, followed by the water spray treatment. A similar response was observed for biological yield, with maximum values under the combined treatment during both years. Harvest index varied significantly among treatments and years. The highest harvest index (37.98 and 37.87% in Year I and II, respectively) was recorded with combined treatment, whereas the control exhibited the lowest harvest index. This indicates that combined application not only increased biomass production but also more efficient partitioning of assimilates toward grain yield.

3.2 Quality attributes

Foliar application of Actibion, macro (NPK), and micro (B + Zn) nutrients, applied individually or in combination, significantly influenced grain nutrient and protein concentration of chickpea under rain-fed conditions during both cropping seasons (Fig. 2 and 3). The N content in grain varied significantly among treatments in both years (Fig. 2A and B). The highest N content was recorded with the combined treatment (Actibion @ 1250 mL ha⁻¹, NPK (16 : 18 : 18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution) recorded 4.80% and 4.75% N during Year I and II, respectively. The lowest N content was observed in control (2.55% in Year I and 2.57% in Year II), followed by water spray treatment. A similar trend was observed for P and K contents in grain of chickpea as highest values of these nutrients were noted under combined treatment followed by Actibion @ 1250 mL ha⁻¹ + NPK (16 : 18 : 18) @ 3% solution during both years (Fig. 2C–F).

In case of Zn and B content in grain, the combined treatment resulted in the highest Zn and B concentration in chickpea grains followed by the application of Actibion @ 1250 mL ha⁻¹ + B @ 0.2% + Zn @ 0.3% solution during both studied years (Fig. 3A–D). Whereas, the lowest Zn and B contents were observed in the control and water spray treatments. Fig. 3E and F demonstrated that combined treatment produced best performance, resulting in the highest protein content (30.0% in

Table 4 Impact of foliar applied bio-stimulant (Actibion), macro (NPK), and micro (B and Zn) nutrients on number of seed plant⁻¹, 100-seed weight, seed yield, biological yield and harvest index of chickpea cultivated under rain-fed conditions during the growing seasons of 2019–20 (Year I) and 2020–21 (Year II)^a

Treatments	Number of seed plant ⁻¹		100-Seed weight (g)		Seed yield (kg ha ⁻¹)		Biological yield (kg ha ⁻¹)		Harvest index (%)	
	Year I	Year II	Year I	Year II	Year I	Year II	Year I	Year II	Year I	Year II
Control (no-spray)	27.25 ± 1.85g	26.75 ± 1.68h	13.65 ± 1.05i	15.30 ± 0.86h	1455 ± 8.2i	1426 ± 7.5i	5100 ± 6.6i	5117 ± 7.5h	28.19 ± 1.5g	28.11 ± 1.2h
Water spray	27.25 ± 1.25g	27.00 ± 1.46h	14.37 ± 0.86h	15.62 ± 1.14h	1505 ± 7.2h	1537 ± 6.8h	5200 ± 8.4h	5201 ± 6.8h	29.50 ± 1.1f	29.55 ± 1.4g
Actibion @ 1250 mL ha ⁻¹	33.25 ± 1.86f	33.00 ± 1.71e	17.8 ± 0.91e	18.45 ± 1.33e	2297 ± 9.1e	2215 ± 7.4e	6350 ± 8.2e	6450 ± 4.8e	35.98 ± 1.5d	34.48 ± 2.5e
NPK (16 : 18 : 18) @ 3% solution	35.00 ± 1.94e	31.00 ± 2.12f	17.12 ± 0.76f	17.95 ± 1.09f	2210 ± 6.2e	2159 ± 7.1f	6200 ± 9.5f	6300 ± 7.9f	35.65 ± 1.4d	33.85 ± 1.6e
B @ 0.2% + Zn @ 0.3% solution	32.00 ± 2.01f	29.00 ± 1.88g	16.45 ± 1.12g	17.42 ± 1.15g	1998 ± 8.4g	1996 ± 4.5g	6050 ± 8.6g	6132 ± 6.4g	33.02 ± 2.1e	32.97 ± 1.3f
Actibion @ 1250 mL ha ⁻¹ + NPK (16 : 18 : 18) @ 3% solution	41.00 ± 2.25b	42.00 ± 2.15b	19.82 ± 1.06b	20.55 ± 0.95b	2557 ± 5.6b	2555 ± 8.2b	6875 ± 7.6b	6859 ± 5.8b	37.35 ± 1.7b	37.06 ± 1.7b
Actibion @ 1250 mL ha ⁻¹ and B @ 0.2% + Zn @ 0.3% solution	37.00 ± 2.45d	36.00 ± 1.86d	18.47 ± 1.10d	19.15 ± 0.91d	2343 ± 7.8d	2326 ± 6.8d	6500 ± 7.8d	6586 ± 6.5d	36.08 ± 1.2d	35.32 ± 1.4d
NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution	39.00 ± 2.16c	39.00 ± 1.68c	19.15 ± 1.21c	19.85 ± 0.85c	2443 ± 4.6c	2429 ± 5.8 c	6650 ± 8.4c	6722 ± 6.3c	36.70 ± 1.8c	36.20 ± 1.5c
Actibion @ 1250 mL ha ⁻¹ and NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution	43.00 ± 2.45a	45.00 ± 2.25a	20.5 ± 1.34a	21.25 ± 1.24a	2658 ± 6.4a	2624 ± 4.9a	6966 ± 6.5a	6995 ± 7.1a	37.98 ± 1.4a	37.87 ± 1.6a
HSD (0.05)	1.66	0.76	0.52	0.48	30.3	31.47	87.34	111.94	0.59	0.73

^a Means sharing the same letter did not differ significantly at $p = 0.05$.



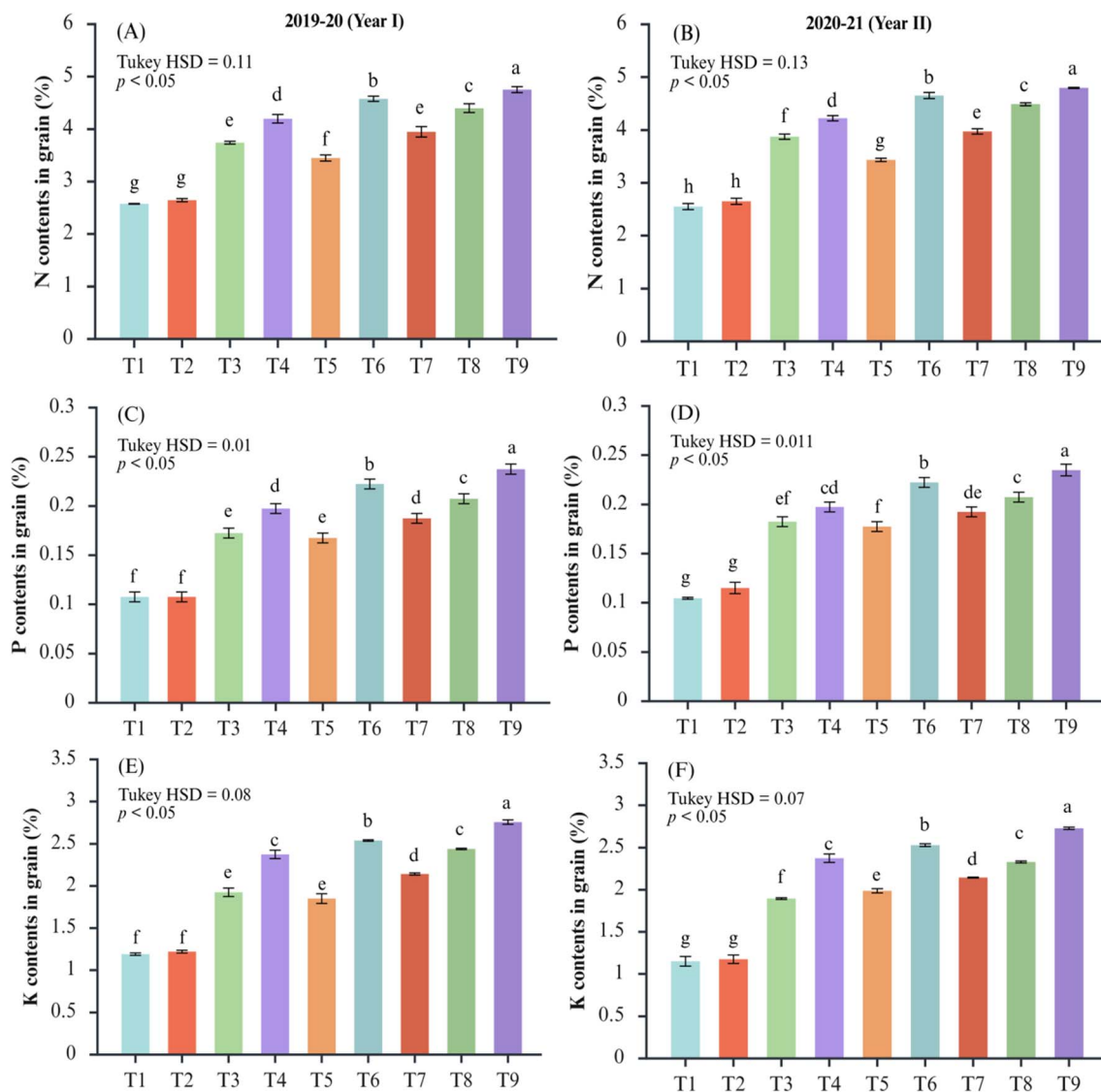


Fig. 2 Impact of foliar applied bio-stimulant (Actibion), macro (NPK), and micro (B and Zn) nutrients on N (A & B), P (C & D) and K (E & F) contents in chickpea during the growing seasons of 2019–20 (Year I) and 2020–21 (Year II), respectively. T_1 = control (no-spray), T_2 = water spray, T_3 = Actibion @ 1250 mL ha⁻¹, T_4 = NPK (16 : 18 : 18) @ 3% solution, T_5 = B @ 0.2% + Zn @ 0.3% solution, T_6 = Actibion @ 1250 mL ha⁻¹ + NPK (16 : 18 : 18) @ 3% solution, T_7 = Actibion @ 1250 mL ha⁻¹ and B @ 0.2% + Zn @ 0.3% solution, T_8 = NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution, T_9 = Actibion @ 1250 mL ha⁻¹ and NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution. Means sharing the same letter did not differ significantly at $p = 0.05$.

Year I and 29.68% in Year II), followed by the application of Actibion @ 1250 mL ha⁻¹ + NPK (16 : 18 : 18) @ 3% solution. In contrast, the control (no spray) treatment yielded the lowest protein content and showed non-significant difference to water spray treatment during both years.

3.3 Correlation analysis

The correlation analysis of Year I and Year II demonstrated a distinct tendency, the majority of the growth, yield, and nutritional characteristics clustered together, and the associations were very strong (Fig. 4). The coefficients were found above 0.77 in both seasons, and several trait pairs crossed the range of 0.80–0.99 indicating the gain in one parameter were frequently

accompanied by improvement in others. Indicators of vegetative growth (PH, LAI, LAD, CGR, and CC) exhibited positive and significant correlations with yield measures (NSPP, and HGW) which eventually led to an increase in seed and biological yield. Growth and yield attributes were also closely associated with nutrient contents (N, P, K, Zn, B) as well as protein indicating improved nutrient uptake and deposition in the seed. The correlation structure is also similar between the two years, which points to the fact that such relationships were constant and support the idea that the use of bio-stimulant (Actibion), macro (NPK) and micro (Zn and B) nutrients is likely to improve the growth, productivity, and nutritional quality, in a coordinated way.



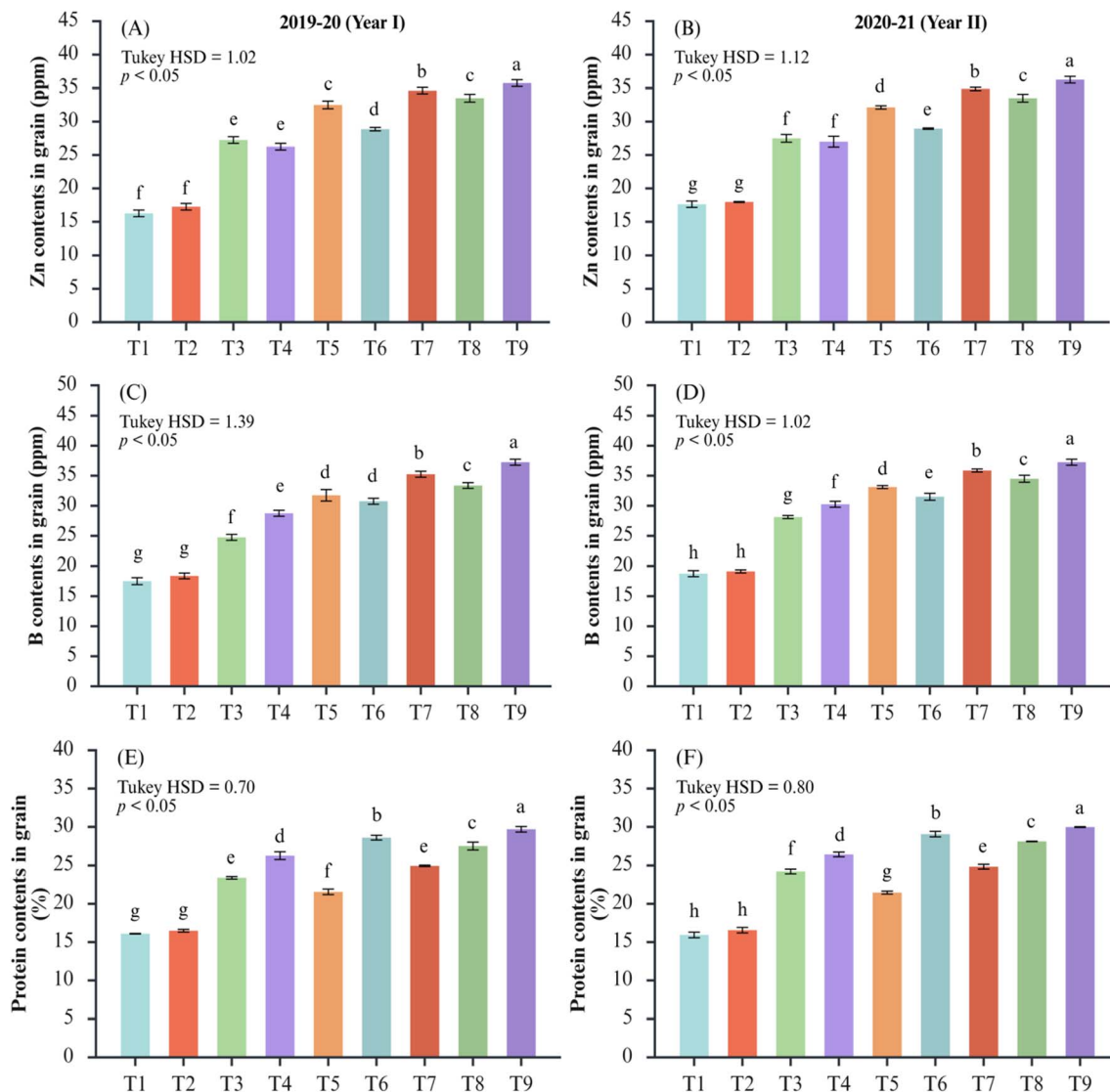


Fig. 3 Impact of foliar applied bio-stimulant (Actibion), macro (NPK), and micro (B and Zn) nutrients on Zn (A & B), B (C & D) and protein (E & F) contents of chickpea during the growing seasons of 2019–20 (Year I) and 2020–21 (Year II), respectively. T_1 = control (no-spray), T_2 = water spray, T_3 = Actibion @ 1250 mL ha⁻¹, T_4 = NPK (16 : 18 : 18) @ 3% solution, T_5 = B @ 0.2% + Zn @ 0.3% solution, T_6 = Actibion @ 1250 mL ha⁻¹ + NPK (16 : 18 : 18) @ 3% solution, T_7 = Actibion @ 1250 mL ha⁻¹ and B @ 0.2% + Zn @ 0.3% solution, T_8 = NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution, T_9 = Actibion @ 1250 mL ha⁻¹ and NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution. Means sharing the same letter did not differ significantly at $p = 0.05$.

3.4 Economic analysis

The economic analysis revealed significant variation among treatments in terms of gross income, total variable cost, net benefits, benefit over control, and benefit-cost ratio (BCR) (Table 5). Overall, the profitability of chickpea production improved progressively with the integration of bio-stimulant, macro, and micro-nutrients. The control (no spray) treatment recorded the lowest economic returns, with a gross income of 826.92 USD ha⁻¹ and net benefits of 715.02 USD ha⁻¹. Consequently, it served as the baseline with zero additional benefit and BCR. Application of water spray resulted only a marginal economic improvement over the control (no-spray), increasing net benefits to 759.88 USD ha⁻¹ and producing an additional

return of 44.86 USD ha⁻¹, with a low BCR of 0.40. The highest average gross income (USD 1516.08 ha⁻¹), net benefit (USD 1365.88 ha⁻¹), total variable cost (USD 150.20 ha⁻¹) and BCR (4.33) were achieved under combined application of Actibion @ 1250 mL ha⁻¹, NPK (16 : 18 : 18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution as compared to other treatments. This indicates that spraying alone, without nutrient supplementation, has limited economic value.

4 Discussion

Integrated nutrient management is an approach to preserving the environment for posterity while simultaneously improving the quality of the production.²⁶ It has been demonstrated that



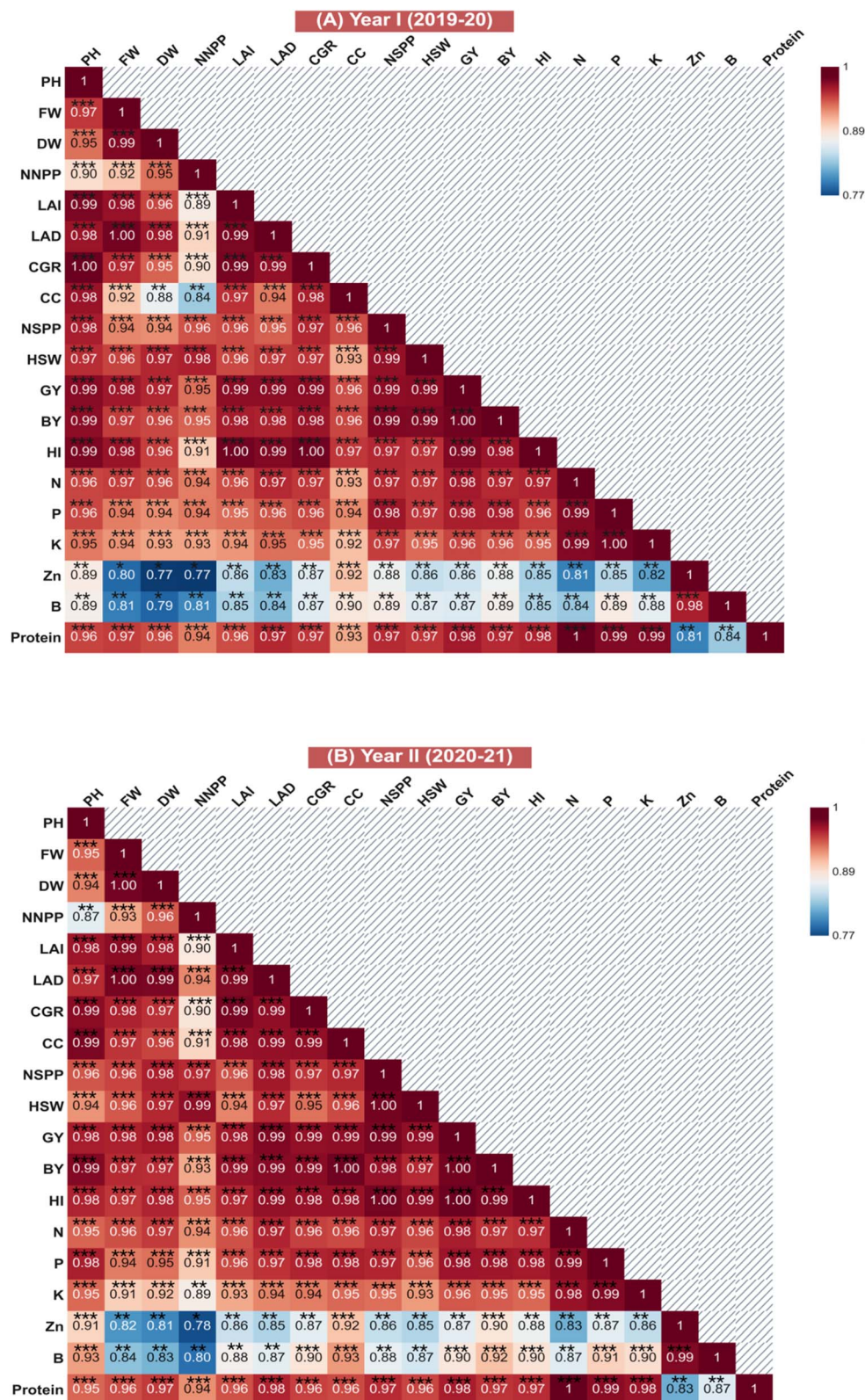


Fig. 4 Pearson's correlation matrix among growth, yield, and nutritional traits of chickpea following bio-stimulant (Actibion), macro (NPK), and micro (B and Zn) nutrients during the growing seasons of (A) Year I (2019–20) and (B) Year II (2020–21). PH: plant height, FW: fresh weight, DW: dry weight, NNPP: number of nodules plant⁻¹, LAI: leaf area index, LAD: leaf area duration, CGR: crop growth rate, CC: chlorophyll contents, NSPP: number of seed plant⁻¹, HSW: 100-seed weight, GY: grain yield, BY: biological yield, HI: harvest index, N: nitrogen, P: phosphorus, K: potassium, Zn: zinc, B: boron. Significance levels are indicated as * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$.



Table 5 Average economic analysis of chickpea production in both growing seasons of 2019–20 (Year I) and 2020–21 (Year II)^a

Treatment	Gross income (USD ha ⁻¹)	Total variable cost (USD ha ⁻¹)	Net benefits (USD ha ⁻¹)	Benefit over control (USD ha ⁻¹)	Benefit-cost ratio
Control (no-spray)	826.92	111.90	715.02	0.00	0.00
Water spray	872.39	112.50	759.88	44.86	0.40
Actibion @ 1250 mL ha ⁻¹	1295.46	131.05	1164.41	449.39	3.43
NPK (16 : 18 : 18) @ 3% solution	1254.28	125.84	1128.44	413.42	3.28
B @ 0.2% + Zn @ 0.3% solution	1146.03	126.02	1020.01	304.99	2.42
Actibion @ 1250 mL ha ⁻¹ + NPK (16 : 18 : 18) @ 3% solution	1466.26	147.20	1319.06	604.04	4.10
Actibion @ 1250 mL ha ⁻¹ and B @ 0.2% + Zn @ 0.3% solution	1339.95	144.57	1195.39	480.37	3.32
NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution	1398.10	141.19	1256.91	541.89	3.84
Actibion @ 1250 mL ha ⁻¹ and NPK (16 : 18 : 18) @ 3% and B @ 0.2% + Zn @ 0.3% solution	1516.08	150.20	1365.88	650.86	4.33

^a Cost was calculated in PKR and converted into USD.

integrated nutrient management significantly increases crop yields by controlling the nutrient supply and minimizing nutrient losses to the environment. This leads to a high resource utilization efficiency, lower costs, and better tolerance of biotic and abiotic challenges²⁷ and maintaining the agricultural productivity while simultaneously increasing farmers' profitability through the rational and effective use of its components such as organic and inorganic sources under stressful environments.

Similarly in current study, foliar application of bio-stimulant (Actibion), macro (NPK), and micro (B and Zn) nutrients either individually or combined significantly improved the chickpea growth attributes under rain-fed conditions (Table 2). The increase in plant height, fresh and dry weight might be due to physiological and biochemical mechanisms operating at the leaf, cellular, and whole-plant levels. Foliar feeding bypasses soil moisture constraints, enabling rapid nutrient absorption.²⁸ Actibion improves membrane permeability, enzyme activity, and hormonal balance, promoting cell division and elongation. The NPK supports chlorophyll synthesis, energy transfer, and osmotic regulation, sustaining photosynthesis and turgor under water stress.²⁹ Zn enhances enzyme function and auxin synthesis, stimulating vegetative growth, while B stabilizes cell walls and facilitates carbohydrate transport.³⁰ Together, these inputs improve photosynthetic efficiency, antioxidant defense, and assimilate translocation, resulting in increased plant height, fresh biomass, and dry matter accumulation in chickpea under moisture-limited environment. Similar improvements in chickpea plant height under foliar nutrition of Panchagavya have been reported by ref. 31 as it contains the favourable macro and micro-nutrients, growth hormones and biofertilizers in liquid formulation. The similar responses were also observed in green gram and chickpea through foliar application of B, Zn, and urea applied at critical growth stages.^{2,32} Outcomes of³³ were also confirmed that foliar nutrition with DAP in chickpea led to the highest fresh weight compared to the control. Moreover, the use of foliar sprays containing micro-nutrients (B and Zn), along

with bio-stimulants, has been demonstrated to significantly increased the plant height and dry matter accumulation in chickpea grown in sandy loam soil.³⁴ Such integrated approaches not only improve the fresh and dry weight of chickpeas but also enhance their nutritional quality, making them a sustainable and economically viable option for rain-fed agriculture.³⁵ In our study, the nodulation in chickpea boosted significantly with combined application of nutrients maybe due to enhanced supply of NPK and boron through foliar application, all of which are critical for nodulation and nitrogen fixation.³⁶ Increased nodule number with foliar application of urea, humic acid-based bio-stimulants, and micro-nutrients has also been reported in black gram and chickpea.^{17–19}

In this study, the combined application of nutrients enhanced plant performance, as reflected by increased chickpea growth-related attributes like LAI, LAD, and CGR (Table 3). It might be due to foliar application of integrated bio-stimulant and nutrients that enhances nutrient availability, which improves amino acids, maintains higher photosynthetic activity for longer periods,³⁷ promoting cell division, membrane integrity, and carbohydrate transport, cell elongation thereby stimulating leaf expansion and increasing photosynthetic surface area. Collectively, these synergistic effects increased the duration and efficiency of photosynthetically active leaf area (higher LAI and LAD), leading to greater biomass accumulation and an elevated CGR through improved carbon assimilation and partitioning towards growing tissues. The results of³⁸ also mentioned that foliar spray of macro-nutrient fertilizers (urea @ 2%, DAP @ 2%, and NPK @ 2%) significantly improved the LAI of soybean as compared to the un-sprayed. The outcomes of³⁹ reported that the foliar spray of NPK (19 : 19 : 19 mixture @ 2%) also contributed to higher LAD in black gram is likely due to the retention of leaves for a longer period. Exogenous application of bio-stimulants and micro-nutrients have been shown to stimulate physiological processes and growth in chickpea and other legumes.^{18,40} Current study reported a significant enhancement in chlorophyll content of chickpea due to synergistic effect of



applied bio-stimulant and nutrients as foliar N directly enhances chlorophyll biosynthesis, while K regulates stomatal function, optimizing photosynthesis.⁴¹ Furthermore, bio-stimulant induced antioxidant activity and Zn-mediated membrane stabilization reduce oxidative damage and chlorophyll degradation.⁴² Together, these mechanisms enhance chlorophyll synthesis, protect photosynthetic structures, and prolong chlorophyll retention under moisture stress conditions. Similar increases in chlorophyll concentration due to foliar application of growth stimulants (urea, and B) have been reported in black gram, chickpea, and cowpea.^{30,43–45}

A significantly higher yield attributes, including seed number, 100-seed weight and seed yield of chickpea was observed with integrated application of bio-stimulant and nutrients than rest of the treatments (Table 4) due to synergistic role of Actibion and nutrients application that supplies essential nutrients required for chlorophyll formation, ATP production, protein synthesis, facilitate sugar transport across membranes, ensuring efficient movement of carbohydrates to developing seeds that strengthening assimilate partitioning toward developing pods thereby increasing seed development.^{9,46,47} Similar results regarding yield improvements in chickpea with the use of foliar bio-stimulants, urea, KNO₃, and the micro-nutrients have been reported by ref. 47 and 48 Foliar application at reproductive phase contributed to a notable rise in seed yield with fast nutrient uptake and resulted in higher flower retention, pod setting, and assimilate partitioning that eventually increased the crop yield.⁴⁹

The greater biological yield in our research may be ascribed to the foliar combined application of bio-stimulant and nutrients through an integrated network of biochemical and physiological processes that collectively strengthen photosynthetic efficiency by improving chlorophyll biosynthesis, ATP formation, and key enzymatic reactions, that facilitate biomass accumulation. Simultaneously, this integrated application also supports structural integrity of cell walls *via* borate pectin cross-linking and facilitates efficient carbohydrate translocation by enhancing sugar-borate complex formation, ensuring effective phloem loading and assimilate movement toward actively growing tissues.^{50–52} The synergistic interaction of these components minimizes oxidative damage caused by ROS, maintains stomatal functionality and photosynthetic stability, and sustains protein and carbohydrate metabolism.⁵³ Consequently, this integrated biochemical and physiological enhancement leads to improved source-sink coordination, greater dry matter accumulation, and ultimately a substantial increase in biological yield of chickpea. These results are agreed with the reports in chickpea where foliar nutrition has been shown to increase biological yield at flowering and pre-flowering stages.^{54–56} The increase in harvest index indicates greater biomass partitioning towards economic yield in the presence of both integrated nutrient and bio-stimulant nutrient application. The probable causes of this response were enhanced flower retention, seed setting, and translocation of assimilates, which agrees with the previously reported results in chickpea and other legumes.^{57–59}

Our study further revealed that foliar application of Actibion, NPK, and micro-nutrients (B and Zn) increased the NPK content in the grains of chickpea (Fig. 2A–F) is rooted in coordinated chemical and physiological mechanisms. Actibion, containing plant growth regulators and signaling molecules, stimulates root proliferation, membrane permeability, and enzyme activation, facilitating more efficient nutrient uptake. Foliar applied NPK directly supplies essential macronutrients, contributing to amino acid and protein synthesis.⁶⁰ Zn acts as a cofactor for enzymes involved in N metabolism and auxin synthesis,⁶¹ while B stabilizes cell walls and enhance sugar translocation to developing seeds.⁵¹ Together, these interactions optimize nutrient absorption, assimilation, and partitioning, leading to significantly higher NPK accumulation in chickpea grains. The findings of⁶² mentioned that the application of 2% solution of the water-soluble N, P and K fertilizer at flowering and pod formation stages led to the highest concentration of nitrogen (3.41%) in the grains of chickpea due to better nutrient uptake and availability. Similarly the gain in P content have been recorded in cowpea, lentil, and lettuce with foliar application of the nutrient and bio-stimulant.^{62,63} Similarly,⁶⁴ reported that the highest K content in chickpea grains (0.68%) was achieved with foliar application of RDF + Speedfol pulses-50% (5 kg ha⁻¹) at 30 and 45 days after sowing. The results of⁶⁵ also confirmed that application of Zn, B, and Mo resulted in maximum K contents in lentil grains. Our outcomes were also supported by ref. 66 who observed that foliar spraying of moringa leaf extract (Bio-stimulant) increased the inorganic nutrients (N, P, and K) in snap beans compared to the control with no bio-stimulant application.

The synergistic application of Actibion, alongside macro (NPK) and micronutrients (B and Zn), applied either individually or in combination increased the Zn and B concentrations in chickpea grains (Fig. 3A–F) reflect the high availability and mobility of these micronutrients under foliar application, as well as synergistic interactions that enhance uptake and utilization.⁶⁷ Similar findings were reported by ref. 64 in chickpea, where a prescribed dosage of NPK fertilizer combined with 1% multi micro-nutrients solution led to maximum Zn content. Similarly,⁶⁸ reported the considerable increases in Zn, B, and Mo levels in chickpea. A significant improvement in grain protein content of chickpea could be due to synergistic role of applied bio-stimulant and nutrients that increased N accumulation, protein synthesis and enzyme activation.⁶⁹ Outcomes of⁶⁶ found that chickpea grain crude protein content was significantly affected by the application of Zn and B at 0.5% each, as Zn is a key structural element of the enzymes responsible for synthesizing proteins and amino acids in plants. Similarly,⁷⁰ noted that higher nitrogen concentrations in the grain, facilitated by organic treatments, boosted the protein synthesis in chickpea, contributing to increases in protein content and yield.

5 Conclusion

The current study demonstrates that integrated foliar application of a bio-stimulant (Actibion) with macro (NPK) and micro-nutrients (B and Zn) provides consistent and substantial



improvements in chickpea growth, physiological efficiency, yield, grain quality, and profitability under rain-fed conditions. The combined treatment outperformed individual nutrient applications by enhancing biomass accumulation, nodulation, photosynthetic capacity, and assimilate partitioning, resulting in marked enhancement in seed yield, grain protein and mineral content during both growing seasons. The strong economic advantage further highlights the practical relevance of this approach for semi-arid regions where soil nutrient availability and moisture limit the crop performance. Overall, the integrated application of Actibion @ 1250 mL ha⁻¹, NPK (16:18:18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution represents a viable, resource-efficient strategy for reducing chickpea yield gaps and improving the nutritional quality, and should be considered in sustainable intensification programs for rain-fed legume systems.

Further studies are required across diverse agro-ecological zones to assess regional adaptability. Moreover, in future, the synergistic impact of different bio-stimulants on growth, yield and quality attributes of chickpea will be investigated. There is need to explore nano-fertilizers and slow-release formulations for better nutrient absorption and efficiency. Need to evaluate site-specific nutrient application strategies using remote sensing and AI-based decision support system. Moreover, the analysis of bioactive compounds such as phenolics, flavonoids, and other metabolites should be considered as an important direction for future research to further elucidate the effects of biostimulant (Actibion) combined with macro (NPK) and micro-nutrients (B and Zn) on chickpea grain quality and nutritional value.

6 Implications

Nutrients depletion and moisture stress are the key constraints for the farmers growing crops under rain-fed environments. However, the findings of current study have important practical implications for farmers, growing crops under rain-fed environment. The combined application of bio-stimulant (Actibion) with macro (NPK) and micro-nutrients (B and Zn) can be effectively integrated into existing nutrient management practices to enhance yield and grain nutritional quality of chickpea. Actibion can be applied as a foliar spray at critical growth stages (e.g., vegetative and early reproductive stages), in combination with recommended basal doses of NPK fertilizers. Micro-nutrients such as B and Zn may also be applied either as soil amendments or foliar spray, depending on soil deficiency status and resource availability. This integrated approach ensures improved nutrient uptake efficiency, better physiological performance, and enhanced resilience of chickpea plants to intermittent drought stress commonly experienced in rain-fed systems.

In field-based agriculture systems, the use of foliar applied bio-stimulants like Actibion is particularly advantageous under limited soil moisture conditions, as nutrient absorption through leaves bypasses soil related constraints such as low nutrient mobility and reduced root activity. This can lead to improved flowering, pod formation, and seed filling, ultimately

translating into higher yield stability. Regarding economic feasibility, although the initial cost of integrated application of Actibion @ 1250 mL ha⁻¹, NPK (16:18:18) @ 3%, and B @ 0.2% + Zn @ 0.3% solution may increase the total cost of cultivation, the improved nutrient use efficiency and potential yield gains can offset these expenses. Under rain-fed conditions, where yield variability is high, such inputs may reduce the risk of crop failure and enhance profitability through better grain quality and market value. Furthermore, the relatively low quantities required for foliar application makes nutrients and bio-stimulant cost-effective compared to bulk fertilizers. Overall, the integration of Actibion with balanced macro and micro-nutrient management represents a promising and practical strategy for improving chickpea productivity and nutritional quality under rain-fed agricultural systems.

Author contributions

Conceptualization: M. A. B. K., M. A.; writing – original draft: M. A. B. K., A. G., M. A. M. M. I.; supervision & methodology: M. A.; formal analysis: A. G., M. A. B. K.; writing – review & editing: M. M., B. M. A. A. B., H. W.; visualization: A. G., M. M. H. W.

Conflicts of interest

The authors declare no competing interests.

Data availability

The data supporting the findings of this study are included in the article. Additional raw data are available from the corresponding author upon reasonable request.

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References

- 1 R. S. Bochalya, K. Choudhary, A. Katoch and A. Sood, *Int. J. Environ. Clim. Change*, 2023, **13**, 661–668.
- 2 R. Praveena, G. Ghosh and V. Singh, *Int. J. Curr. Microbiol. Appl. Sci.*, 2018, **7**, 1422–1428.
- 3 R. K. Mahto, B. Chandana, R. K. Singh, A. Talukdar, K. Swarnalakshmi, A. Suman, Vaishali, D. Dey and R. Kumar, *BMC Plant Biol.*, 2025, **25**, 693.
- 4 B. Bhargavi, R. Singh and S. Babu, *Agricultural Diversification for Sustainable Food Production*, 2025, p. 319.
- 5 S. Khan, S. Basra, M. Nawaz, I. Hussain and N. Foidl, *S. Afr. J. Bot.*, 2020, **129**, 74–81.
- 6 M. M. Javaid, H. Waheed, N. Nazami, M. Ashraf, F.-M. Li and A. Tanveer, *Semina: Cienc. Agrar.*, 2020, **41**, 3053–3066.



- 7 S. Batool, S. Khan, S. M. Basra, M. Hussain, M. S. Saddiq, S. Iqbal, S. Irshad and M. Hafeez, *Int. Lett. Nat. Sci.*, 2019, **76**, 51–59.
- 8 K. S. Reddy, G. Wakchaure, P. Khapte and S. Changan, *Indian J. Fert.*, 2023, **19**, 788–800.
- 9 M. Asif, M. Adnan, M. E. Safdar, N. Akhtar, A. Khalofah and F. M. Alzuaibr, *J. King Saud Univ. Sci.*, 2022, **34**, 102236.
- 10 W. Cao, H. Sun, C. Shao, Y. Wang, J. Zhu, H. Long, X. Geng and Y. Zhang, *Horticulturae*, 2025, **11**, 930.
- 11 A. Sadeque, S. Ahmed, M. A. Islam, M. A. Mukhtadir, M. Z. Rahman, M. S. Islam, U. B. Antu, M. M. Islam, Z. Ismail and A. M. Idris, *J. Plant Nutr.*, 2026, **49**, 203–245.
- 12 M. A. Hassan and J. H. Hamza, Iron, Zinc and Boron in Sunflower, *IOP Conference Series: Earth and Environmental Science*, 2025, **1487**, 12109.
- 13 S. K. Kohli, H. Kaur, K. Khanna, N. Handa, R. Bhardwaj, J. Rinklebe and P. Ahmad, *Plant Growth Regul.*, 2023, **100**, 267–282.
- 14 G. P. Nortjé, R. Kronenbergh, S. Bardhanh, S. Satyanarayana, N. Khank, H. Wang and J. Rinklebe, *Adv. Agron.*, 2023, **182**, 81.
- 15 R. Vahedi, M. H. Rasouli-Sadaghiani, M. Barin and R. R. Vetukuri, *Processes*, 2022, **10**, 343.
- 16 M. Hussain, M. Banoo, B. Sinha and G. Chand, *J. Pharmacogn. Phytochem.*, 2022, **11**, 270–275.
- 17 P. S. Pavan Shinde, S. Doddagoudar and S. Vasudevan, *Legume Res.*, 2017, **40**, 704–709.
- 18 D. Beulah Esther and G. Gautam, *J. Pharmacogn. Phytochem.*, 2020, **9**, 1754–1756.
- 19 B. Sharanya, A. M. Gowda and K. Srinivasappa, *Pharma Innovation*, 2022, **11**, 399–404.
- 20 Z. Sesták, J. Catský and P. G. Jarvis, In *Plant photosynthetic production*, Dr. W Junk N.V publishers, The Hague, 1971.
- 21 V. Powar, *Indian J. Biochem.*, 1967, **4**, 135.
- 22 D. Watson, *Proceedings-Easter School in Agricultural Science*, University of Nottingham, 1956, vol. 3.
- 23 G. Estefan, R. Sommer and J. Ryan, *Methods of Soil, Plant, and Water Analysis: A Manual for the West Asia and North Africa Region*, International Center for Agricultural Research in the Dry Areas, 2013.
- 24 O. H. Lowry, N. J. Rosebrough, A. L. Farr and R. J. Randall, *J. Biol. Chem.*, 1951, **193**, 265–275.
- 25 R. Steel, J. Torrie and D. Dicky, Principles and Procedures of Statistics, *Multiple Comparisons*, McGraw Hill Book Co., New York, NY, USA, 1997.
- 26 N. Kushwah, V. Billore, O. P. Sharma, D. Singh and A. P. S. Chauhan, *Plant Sci. Arch.*, 2024, **8**, 10–12.
- 27 P. S. Hindoriya, R. Kumar, R. K. Meena, H. Ram, A. Kumar, S. Kashyap, B. Biswal, K. Bhakuni, P. S. Pyati and K. Garg, *Agronomy*, 2024, **14**, 339.
- 28 A. Pooja and M. Ameena, *Agric. Rev.*, 2021, 42.
- 29 R. N. Shabbir, M. Y. Ashraf, E. Waraich, R. Ahmad and M. Shahbaz, *Pak. J. Bot.*, 2015, **47**, 1207–1216.
- 30 S. Surendar, B. Renjan and B. Bindu, *J. Sci. Res. Rep.*, 2024, **30**, 1072–1085.
- 31 P. Panchal, P. Patel, A. Patel and A. Desai, *Int. J. Chem. Stud.*, 2017, **5**, 265–267.
- 32 V. Pal, G. Singh and S. S. Dhaliwal, *J. Soil Sci. Plant Nutr.*, 2020, **20**, 738–750.
- 33 I. U. Rahman, F. Ijaz, A. Afzal and Z. Iqbal, *Bangladesh J. Bot.*, 2017, **46**, 111–118.
- 34 M. Dudwal, S. L. Bijarnia and D. Choudhary, *Asian Res. J. Agric.*, 2024, **17**, 288–298.
- 35 P. Subhasmita, L. Zurika and S. Arya, *Int. J. Adv. Biochem. Res.*, 2023, **7**, 118–123.
- 36 S. Sulieman and L.-S. P. Tran, *Plant Sci.*, 2015, **239**, 36–43.
- 37 T. A. Abd El-Mageed, W. M. Semida and M. M. Rady, *Agric. Water Manag.*, 2017, **193**, 46–54.
- 38 A. Dass, G. A. Rajanna, S. Babu, S. K. Lal, A. K. Choudhary, R. Singh, S. S. Rathore, R. Kaur, S. Dhar and T. Singh, *Sustainability*, 2022, **14**, 5825.
- 39 S. Samanta and J. K. Thakur, *Front. Plant Sci.*, 2015, **6**, 757.
- 40 K. Nouri, M. Janmohammadi, A. A. Aliloo, M. Nouraein and A. Abbasi, *Acta Univ. Agric. Silv. Mendelianae Brun.*, 2022, **69**, 665–675.
- 41 W. Hou, M. Tränkner, J. Lu, J. Yan, S. Huang, T. Ren, R. Cong and X. Li, *BMC Plant Biol.*, 2019, **19**, 302.
- 42 M. Shahid and S. Ali, *Physiol. Plant.*, 2025, **177**, e70680.
- 43 K. Sujatha and V. Vijayalakshmi, *Int. J. Adv. Res. Technol.*, 2013, **2**, 216–230.
- 44 N. Bellaloui, *Am. J. Plant Sci.*, 2011, **2**, 692–701.
- 45 B. Nandan, B. Sharma, G. Chand, K. Bazgalia, R. Kumar and M. Banotra, *Act. Sci. Nutr. Health*, 2018, **2**, 12–19.
- 46 R. Hänsch and R. R. Mendel, *Curr. Opin. Plant Biol.*, 2009, **12**, 259–266.
- 47 A. Sahu, D. Baghel, A. K. Singh, T. Chowdhury and R. Soni, *Curr. J. Appl. Sci. Technol.*, 2023, **42**, 128–135.
- 48 H. Singh, R. Lodhi, D. Dwivedi, V. Bharati, A. Kumar, S. Kumar and A. Singh, *Pharma Innov. J.*, 2023, **12**, 2200–2202.
- 49 A. A. H. Abdel Latef, M. F. Abu Alhmad and K. E. Abdelfattah, *J. Plant Growth Regul.*, 2017, **36**, 60–70.
- 50 T. Zhou, Y. Hua, B. Zhang, X. Zhang, Y. Zhou, L. Shi and F. Xu, *Plant Cell Physiol.*, 2017, **58**, 1991–2005.
- 51 F. Shireen, M. A. Nawaz, C. Chen, Q. Zhang, Z. Zheng, H. Sohail, J. Sun, H. Cao, Y. Huang and Z. Bie, *Int. J. Mol. Sci.*, 2018, **19**, 1856.
- 52 M. T. Qureshi, N. Iqbal, I. R. Noorka and H. Waheed, *J. Plant Nutr.*, 2020, **44**, 120–129.
- 53 R. Pasala, R. Kulasekaran, B. B. Pandey, C. Manikanta, K. Gopika, P. J. Daniel, S. Elthury and P. Yadav, *Plant Nutrition and Food Security in the Era of Climate Change*, 2022, pp. 377–398.
- 54 R. Sadoyan, H. Martirosyan, A. Avetisyan and L. Suvaryan, *E3S Web Conf.*, 2023, **460**, 01007.
- 55 V. Kirnapure, A. Choudhary, A. Gawate and S. Potkile, *J. Pharmacogn. Phytochem.*, 2020, **9**, 202–204.
- 56 R. Kumar, B. Kumar-Saren and S. Kumar-Patel, *Int. J. Plant Soil Sci.*, 2023, **35**, 958–965.
- 57 J. Mandloi, Doctoral diss., RVSKVV, Gwalior, 2022.
- 58 J. B. V. Montenegro, J. A. B. Fidalgo and V. M. Gabella, *Spanish J. Agric. Res.*, 2010, 797–807.
- 59 M. Shafiq, M. Arif, N. Akhtar, M. Yousaf, A. G. Saggoo and M. Zafar, *Pak. J. Agri. Sci.*, 2021, **58(1)**, 35–42.



- 60 M. Rajasekar, D. U. Nandhini, V. Swaminathan and K. Balakrishnan, *Int. J. Chem. Stud.*, 2017, **5**, 304–309.
- 61 A. Suganya, A. Saravanan and N. Manivannan, *Commun. Soil Sci. Plant Anal.*, 2020, **51**, 2001–2021.
- 62 B. Chaudhary, P. Chaudhari, R. Parikh, K. Patel and N. Chaudhari, *Int. J. Environ. Clim. Change*, 2023, **13**, 4046–4050.
- 63 M. M. Islam, M. R. Karim, M. M. H. Oliver, T. A. Urmi, M. A. Hossain and M. M. Haque, *Agronomy*, 2018, **8**, 100.
- 64 T. Kachave, H. Kausadikar and M. Deshmukh, *Int. J. Conserv. Sci.*, 2018, **6**, 1660–1662.
- 65 M. S. N. Chowdhury, M. N. H. Sani, A. B. Siddique, M. S. Hossain and J. W. H. Yong, *Plant Stress*, 2024, **12**, 100452.
- 66 A. F. El Sheikha, A. Y. Allam, M. Taha and T. Varzakas, *Appl. Sci.*, 2022, **12**, 776.
- 67 Y. Long and J. Peng, *Genes*, 2023, **14**, 130.
- 68 J. Nandaniya, D. Hirpara, N. Makwana and N. Sarvaiya, *Trends Biosci.*, 2016, **9**, 548–551.
- 69 I. Cakmak, *Plant Soil*, 2008, **302**, 1–17.
- 70 D. Jagtap, *Chem. Sci. Rev. Lett.*, 2021, **10**, 269–273.

