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Biochemical responses of lentil (*Lens culinaris* Medik) to zinc and iron nutrition in zinc deficient soil

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Abstract
The experiment was carried out in pot culture during *rabi* 2016-17 at Institute Agricultural Farm and Department of Plant Physiology of Institute of Agricultural Sciences, Banaras Hindu University, Varanasi, with the soil and foliar application of zinc sulphate hepta-hydrate ($ZnSO_4.7H_2O$) as zinc nutrition and Fe-EDTA as iron nutrition on lentil crops and was arranged in completely randomized design (CRD) with 11 treatments and three replications. Measured biochemical parameters were soluble leaf protein content, nitrate reductase activity, total soluble sugar, chlorophyll 'a', chlorophyll 'b' and carotenoid pigments. The zinc foliar spray showed better performance with protein content, total soluble sugar, nitrate reductase activity, chlorophyll and carotenoid content in zinc deficient soil. When iron was applied as soil and foliar separately i.e. T_4 and T_5, did not show significant increase but T_7 (both soil and foliar treatment of iron) showed a significant increase in case of nitrate reductase activity. Results showed the significant increase in case of foliar treatment in comparison to soil treatment at 1% level of significance. In general, mean comparison showed that the effect of soil and foliar application of both zinc and iron (T_{10}) was overall significant and considered to be prime treatment in case of zinc deficient soil.

Keywords: lentil, biochemical parameters, zinc, iron, soil application, foliar spray

Introduction
Lentil is one of the early domesticated plant species, as old as those of emmer, barley and pea (Harlan, 1992) [11]. It is a deploid with chromosome no. 14 (2n = 14) and self-pollinating annual species with a haploid genome size of an estimated 4063 Mbp. The origin of lentil is considered to be the Near East and Egypt at the Central and Southern Europe, the Mediterranean basin, Ethiopia, Afghanistan, India and Pakistan, China and later it was spread to Latin America (Cubero 1981) [5]. Its production in India has always been important as it is the one of the most important rabi crops in the country. In fact, India was the largest producer of the Lentil crop in the world until recently Canada took over the lead leaving India at the second place. According to a report, the total cultivated area in the world is around 4.6 million hectares producing 4.2 million tons of seeds with an average production of 1095 kg/ha (FAO, 2010) [7]. Migliozzi *et al.* (2015) [18] reported that lentil (*Lens culinaris* Medik.) is a nutritious and a staple food which is consumed by millions of people as it is being the good source of energy and also contains a range of micronutrients and prebiotic carbohydrates.
Migliozzi *et al.* (2015) [18] reported that lentil (*Lens culinaris* Medik.) is a nutritious and a staple food which is consumed by millions of people as it is being the good source of energy and also contains a range of micronutrients and prebiotic carbohydrates. Thavarajah (2011) [29] estimated its richness in micronutrient content and potential to provide adequate dietary amounts, especially for iron (Fe), zinc (Zn), and selenium (Se); a 50 g serving provides 3.7–4.5 mg Fe, 2.2–2.7 mg Zn, and 22–34 µg Se. It also a good source of vitamin A, thiamin, folate, and β-carotene and also contained with considerable amounts of riboflavin, niacin, pantothenic acid, pyridoxine, vitamin K, and vitamin E (Bhatty, 1988) [1].
Singh *et al.* (2015) [26] reported that zinc (a bluish-white metallic element) is one of the essential micronutrients required for optimum crop growth and accounted for about 0.02% of the earth's crust. Plants take up zinc in its divalent (Zn^{+2}) form. It is also associated with more than 50 distinct metallo-enzymes possessing different functions like synthesis of nucleic acids, and specific proteins such as hormones and their receptors. Zinc response is quite common in plant and animals and is an essential part in dietary supplements. They also reported that the deficiency of zinc causes some symptoms in pulses and so what in lentil crop too. It takes 5-6 weeks after sowing of the lentil crop to show deficiency symptoms which include the colour change of matured leaves from green to yellowish white starting, the severity of the deficiency

results into the turning of leaflet brown and ultimately fall down, stunted growth of plants with poor pod formation whereas Chaney (1993) [4] reported that zinc toxicity can strongly inhibit photosynthesis, cause phyto-toxicity and reduction in yield can also be observed when plant leaf Zn concentrations reaches about 300 - 1000 µg Zn g^{-1}.

Iron is used by almost all living organisms as an important micronutrient because it plays an essential role in metabolic processes such as DNA synthesis, respiration, and photosynthesis. Instead of these, many other metabolic pathways are activated by iron, and it also serves as prosthetic group constituent for many enzymes (Rout and Sahoo, 2015) [22]. It is the third most limiting nutrient for plant growth and metabolism, primarily due to the low solubility of the oxidised ferric form in aerobic environments (Zuo and zhang, 2011; Samaranayke *et al*., 2012) [31, 23].

Iron deficiency is common nutritional disorder which results in reduced nutritional quality and poor yield of many crop plants. It is generally involved in chlorophyll synthesis and maintenance of chloroplast structure. The soil usually consists of a large amount of iron but it may or may not be in soluble form needed by the plant. Predominantly, iron exists as Fe^{+3}chelate form in soil which cannot be absorbed by the plant at higher pH or in alkaline soil. Thus, at high pH plants are not able to synthesize and stabilize chlorophyll that results in yellowing of leaves, poor growth and reduced yield. To deal with this problem, plants develop a mechanism to absorb the small amount of iron from the soil e.g. non- graminaceous crops release protons, secrete phenolics, reduce Fe^{+3} and take up iron (Jeong and Guerinot, 2009; Cesco *et al*., 2010) [13, 3]. Tognetti *et al*. (2007) [30] reported that Iron deficiency is a topic of major concern with the agriculture point of view because it affects one-third of the cultivable land on the earth and also causes the decline of many photosynthetic components such as Fe-s protein ferredoxin, involved in oxido-reductive pathway of chloroplast. The visual symptoms of iron deficiency in plants, is the interveinal chlorosis of young leaves and stunted root growth (Schimdt, 1993) [24].

Generally, micronutrients can be applied directly into the soil (in the solid form or dissolved in water) or sprayed on the leaves but Soil feeding is the most ancient normal fertilization practice, however it cannot be generalized as it depends on many factors from soil type to plant characteristics and its physiological state (Bratasevec *et al*., 2013) [2]. Foliar application was developed around 60 years ago, does not completely replace the soil application of nutrients but can perform better with uptake of nutrients by the plants and availability to the plants in comparison to soil application (Kannan, 2010) [14]. Darwesh (2011) [6] reported the effectiveness of foliar application of micronutrient that it supplies nutrients to higher plants better than any other means. In soil application of nutrients, there may be possibility of leaching of nutrients that might not be available to crops.

Thus, in the context of above facts and the importance of lentil crop, the present work was carried out to study the changes in certain biochemical parameters as influenced by soil and foliar application of zinc and Iron in lentil.

Materials and Methods

The present investigation on was carried out in pot culture during *rabi* 2016-17 at Institute Agricultural Farm and Department of Plant Physiology, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi. The experimental site is located in the south eastern part of Varanasi city at $25^0 18^{'}$N latitude, $83^0 3^{'}$E longitude and at an altitude of 75.5 meter above mean sea level. Varanasi falls in a sub-tropical climate and is subjected to extremes of weather condition i.e., extremely hot summer and cold winter. The temperature begins to rise from middle of February and reaches its maximum by May-June (mean maximum temperature 43.6^0C) but has a tendency to decrease from July onwards touching minimum in December-January (mean minimum temperature 8.2^0C). The normal rainfall is about 1100 mm of which 88 percent in June to September as a monsoon season rains, 5 to 7 percent in October to December as a post monsoon season rains. The mean relative humidity of the area is about 66 percent, which rises up 92 percent during July to September and falls down to 39 percent during the end April to early July. Disease free and healthy seeds of Lentil (*Lens culinaris* Medik.), genotype HUL-57 (microsperma), which is a mutant of variety HUL-11, was procured from the Department of Genetics and Plant Breeding, Institute of Agricultural Sciences, BHU, Varanasi. It was released by BHU, Varanasi in the 2005, having 121 days of maturity and average grain yield of 14 q/ha. This variety is mostly grown in the North Eastern Plain Zone (NEPZ).

Soil was collected from the dry land agriculture part of the experimental farm, Institute of Agricultural Sciences, Banaras Hindu University. It was cleaned by removing the stones, weeds etc. and the soil to be used in the pots were dried, powdered and mixed thoroughly. Available Zn and Fe content in the collected soil were 0.33 ppm and 19.8 ppm, respectively which deficient in zinc content. Thirty three plastic pots (2.5 kg of soil capacity) were taken and cleaned. Zinc was applied at the concentration of 3.6 mg zinc (16.5 mg of Zinc sulphate Hepta-hydrate) & Iron was applied at a concentration of 3.6 mg iron (30 mg of Fe-EDTA) and according to the treatment mixed with the soil (2.5 kg) along with control that contained no added zinc and Iron. After pot filling, 10 lentil seeds were sown in all the pots. The foliar treatments of Zinc and Iron included two sprays, the first foliar spray at 30 DAS and second at 45 DAS according to treatments with 0.5% zinc sulphate hepta-hydrate and 3% Fe-EDTA.

Observations pertaining to Biochemical parameters i.e. chlorophyll ‘a’ and chlorophyll ‘b’ content, carotenoids content, total soluble sugar content, protein content and nitrate reductase activity were recorded at 15 days after first foliar spray and 15 days after second foliar spray of nutrients corresponding to 45 DAS and 60 DAS. Plant samples were collected in the morning, between 07:00 am to 09:00 am. Fully expanded leaves were plucked and brought to the laboratory in the ice bucket. Observations were further analysed using Duncun’s Multiple Range Test (DMRT).

Result and Discussions

Chlorophyll ‘a’ content (mg g^{-1}F.W. of leaves)

The maximum chlorophyll ‘a’content was obtained in treatment T_{10} (0.31 mg g^{-1}F.W.) consisting of soil and foliar application of both the nutrients and minimum was obtained in T_0 i.e. control (0.12 mg g^{-1}F.W.) at 45 DAS and same pattern was also observed in case of analysis done at 60 DAS where T_{10} took the value 0.35 mgg^{-1} fresh weight and T_0 was 0.13 mgg^{-1}fresh weight. Treatments T_1 (soil application of Zinc), T_4 (soil application of Iron), T_5 (foliar application of iron), T_6 (soil and foliar application of Iron), T_8 (soil application of Iron and Zinc and foliar Application of Zinc), T_9 (soil and foliar application of Iron and Foliar application of

Zinc) and T_{10} (soil and foliar application of both Zinc and Iron) were significantly different when compared with control at both 45 DAS and 60 DAS.

Chlorophyll 'b' content (mg g^{-1}F.W. of leaves)

Mean value comparison showed that the maximum chlorophyll 'b' was found in treatment T_{10} (soil and foliar application of both Zinc and Iron) and minimum was found in treatment T_7 (soil application of both Zinc and Iron) at both stages. Treatment T_6, T_7 and T_8 are significantly inferior to control as compared according to critical difference at both stages whereas treatment T_3 and T_{10} are showing significant increase in comparison to control.

In the present study, the symptom of chlorosis was not observed and positive growth was observed, supporting the views of Goos and Johnson (2000) [9] that foliar sprays of FeEDTA generally corrected Fe deficiency chlorosis in Fe-inefficient soybean varieties, resulting in yield increases at some, but not all locations tested. Sultana *et al*. (2001) [27] reported that iron played essential roles in the metabolism of chlorophyll pigments and External application of Fe increased photosynthesis, net assimilation and relative growth in seawater-stressed rice. Pandaey and Gautam (2009) [21] observed that Chlorophyll 'a'content in leaves of lentil plant was significantly increased with the increase of Zn concentration upto 25.0 ppm, while it decreased at excess (100.0 ppm) Zn supply whereas chlorophyll 'b' and total chlorophyll contents were not affected at Zn above 0.25 ppm. Ghasemian *et al*. (2010) [8] declared the essentiality of zinc nutrition in chlorophyll production and pollen function. Laleluo *et al*. (2013) [15] carried out a study on naked pumpkin and discussed that the zinc concentration increased to 30 mM, increased the amount of chlorophyll 'a' and 'b', but higher concentrations of zinc i.e. above 30 mM decreased their amount.

Carotenoids content (mg g^{-1}F.W. of leaves)

Carotenoids consist of xanthophyll and carotene pigments. Comparing mean value, it was observed that the maximum carotene content was in treatment T_{10} (soil and foliar application of both Zinc and Iron) at both stages and the minimum was found in T_5 (foliar application of Iron) at 60 DAS and the difference between the control and the treatment T_5 was not significant whereas at 45 DAS, the minimum carotenoids content was obtained in T_6 (0.215 mg g^{-1}) consisting soil and foliar application of Iron. There was no significant difference between the T_6 and the control at 45 DAS too, whereas T_{10} showed a significant increase in comparison to control at both the stages. Nenova (2006) [19] while working with pea found that different levels of iron i.e. from deficient to toxic resulted in higher plant growth and carotenoids content.

Total soluble sugar content (mg g^{-1}F.W. of leaves)

Mean comparison showed that the maximum soluble sugar content was observed in the treatment consisting of soil and foliar application of both Zinc and Iron (T_{10}) at both the stages which were 36.527 mg g^{-1} at 45 DAS and 46.447 mg g^{-1} at 60 DAS whereas minimum i.e. 13.053 mgg^{-1} of F.W. and 17.453 mg g^{-1}F.W. were observed in controls (T_0) of both the stages respectively. There was significant increase in the treatments T_5, T_8, T_9 and T_{10} in comparison to control at both the stages and soil applied Zinc and Iron (T_7) did not show significant effect on concerned parameter in comparison to control at both stages.

Lalelou *et al.* (2013) [15] discussed that with the right amount of zinc concentration, the concentration of soluble sugars increased in roots and shoots of wheat plant. Accumulation of soluble sugars helps to regulate osmotic stress in plant cells and leads to preservation of biological molecules and membranes. Pandey *et al*. (2013) [20] reported that Providing Zn as a foliar spray at pre-flowering stage minimized the severity of Zn deficiency on reproductive structure development and enhanced the seed nutritional status by enhancing seed Zn density, seed carbohydrate (sugar and starch content) and storage proteins (albumins, globulins, glutenins, and prolamines).

Protein content (mg g^{-1}F.W. of leaves)

The data on total protein content is presented in table 4.2.1 and figure 4.2.1. At 45 DAS, the maximum protein content was obtained in treatment consisting soil and foliar application of both Zinc and Iron i.e. T_{10} (49.1 mg g^{-1} of F.W.) and minimum was obtained in treatment control i.e. T_0 (8.3 mg g^{-1} of leaf fresh weight), while comparing the mean values. The same pattern was also observed at 60 DAS with T_{10} (54.5 mg g^{-1} of F.W.) and T_0 (11.0 mg g^{-1} of leaf fresh weight). Ultimately i.e. at 60 DAS, it was observed that protein content was higher in the treatment containing foliar application of zinc (T_2) than the treatments having both soil and foliar application of zinc (T_3) and only soil application of zinc (T_1). In case of iron too, only foliar application performed better than others in zinc deficient soil. All the treatments showed significant increase in comparison to control at both the stages. There was also observed a significant increase between the treatments T_1, consisting of soil applied Zinc and T_2, consisting of foliar applied zinc at both the stages.

Ibrahim and Shabbir (2014) [12] reported that the low concentration of Zn addition support soluble protein accumulation in lentil leaves, although protease activity greatly arrested the protein content and appeared as a negative factor with increasing Zn levels. Singh and Bhatt (2015) [25] reported that zinc plays a vital role in the synthesis of proteins and nucleic acid and helps in the utilization of nitrogen and phosphorus in the plant. Hafeez *et al*. (2013) [10] reported the essentiality of zinc for the growth in animals, human beings, and plants and its vital participation in crop nutrition as required in various enzymatic reactions, metabolic processes, and oxidation-reduction reactions. In addition to these, Zn is also essential for many enzymes which are needed for nitrogen metabolism, energy transfer and protein synthesis. Thalooth *et al.* (2006) [29] showed that using of zinc sulfate increases grain protein content of mungbean. In addition, iron is involved in the metabolism of nitrogen and increases leaf area and has a direct impact on the process of protein production. So it can be expected that iron foliar application, increased the plant protein production.

Nitrate reductase activity (µ moles NO_2^- formed g^{-1}F.W. h^{-1})

Treatments T_2, T_6 and T_{10} showed the significant increase in comparison to T_0 i.e. control at both the stages. Mean value comparison showed that the maximum enzyme activity was observed in treatment T_{10} and minimum was observed in treatment T_0. In the present study, nitrate reductase performed better either in case of only foliar applied nutrients or where the nutrients were applied as both soil and foliar together. At 45 DAS, T_6 performed better than T_2 but the reverse result was obtained at 60 DAS. There was significant decrease in

NR activity in the treatments foliar application of only Zinc in zinc deficient soil.

Luna *et al.* (2000) [16] reported an inhibitory effect of Zinc on nitrate reductase activity in wheat leaves which increases with increasing concentration of Zinc. The increased NR activity in treatment T_{10} might be the result of positive effect of iron and antagonistic effect among themselves. Mann *et al.* (2017) [17] reported that nitrate reductase activity was increased with the supply of Iron source. A significant increase in NR was observed with the Foliar Application in comparison to basal application. Iron directly influences the activity of nitrate reductase enzyme as it constituted the prosthetic group (4Fe-4S) of this enzyme.

Table 1: Mean values of biochemical parameters at 45 days after sowing of lentil crop.

		Mean Values of Parameters (45 DAS)#					
S. No.	**Treatments**	**Chlorophyll 'a' content (mg g^{-1}F.W. of leaves)**	**Chlorophyll 'b' content (mg g^{-1}F.W. of leaves)**	**Carotenoids Content (mg g^{-1}F.W. of leaves)**	**Total Soluble Sugar content (mg g^{-1}F.W. of leaves)**	**Protein content (mg g^{-1}F.W. of leaves)**	**Nitrate Reductase Activity (µ moles NO_2^- formed g^{-1}F.W. h^{-1})**
1.	T_0 [Control$_{(no\ added\ zinc\ and\ iron)}$]	0.122^{f}	0.091^{cd}	0.243^{cd}	13.053^{e}	8.338^{h}	0.610^{d}
2.	T_1 Zinc$_{(Soil)}$	0.169^{cd}	0.125^{bc}	0.247^{cd}	19.407^{cde}	22.904^{e}	1.736^{cd}
3.	T_2 Zinc$_{(Foliar)}$	0.150^{de}	0.075^{de}	0.298^{b}	22.220^{bcde}	42.363^{b}	2.295^{bc}
4.	T_3[Zinc$_{(Soil)}$+Zinc$_{(Foliar)}$]	0.128^{ef}	0.160^{b}	0.302^{b}	21.753^{bcde}	18.313^{g}	1.276^{cd}
5.	T_4 [Iron $_{(Soil)}$]	0.226^{b}	0.099^{cd}	0.302^{b}	17.547^{de}	14.896^{h}	0.630^{d}
6.	T_5 [Iron$_{(Foliar)}$]	0.304^{a}	0.088^{cd}	0.220^{d}	29.987^{abc}	36.413^{d}	0.864^{d}
7.	T_6 [Iron$_{(Soil)}$+ Iron$_{(Foliar)}$]	0.175^{cd}	0.040^{ef}	0.215^{d}	22.793^{bcde}	21.725^{f}	3.405^{b}
8.	T_7 [Zinc$_{(Soil)}$ + Iron $_{(Soil)}$]	0.122^{f}	0.021^{f}	0.275^{bc}	15.793^{e}	44.300^{b}	0.786^{d}
9.	T_8 [Zinc$_{(Soil)}$ + Iron $_{(Soil)}$ + Zinc$_{(Foliar)}$]	0.184^{c}	0.036^{ef}	0.288^{b}	28.893^{abcd}	39.071^{c}	1.121^{cd}
10.	T_9 [Iron$_{(Soil)}$ + Iron $_{(Foliar)}$ + Zinc$_{(Foliar)}$]	0.187^{c}	0.096^{cd}	0.215^{d}	32.840^{ab}	18.250^{g}	1.840^{cd}
11.	T_{10} [Zinc$_{(Soil)}$ + Iron $_{(Soil)}$ + Zinc$_{(Foliar)}$+ Iron $_{(Foliar)}$]	0.314^{a}	0.200^{a}	0.356^{a}	36.527^{a}	49.129^{a}	4.705^{a}
	SE (m)±	0.009	0.014	0.013	4.051	0.785	0.436
	C.D.*	0.027	0.04	0.038	11.957	2.316	1.287

* Denotes significance at P= 0.01. Means followed by same letters in a column are not significantly different but different letters are significantly different, using Duncun's Multiple Range Test (DMRT).

As per treatments, the first foliar spray was given at 30 DAS and the observations were recorded after 15 days of spray application i.e. 45 DAS.

Table 2: Mean values of biochemical parameters at 60 days after sowing of lentil crop.

		Mean Values of Parameters (60 DAS)#					
S No.	**Treatments**	**Chlorophyll 'a' content (mg g^{-1}F.W. of leaves)**	**Chlorophyll 'b' content (mg g^{-1}F.W. of leaves)**	**Carotenoids Content (mg g^{-1}F.W. of leaves)**	**Total Soluble Sugar content (mg g^{-1}F.W. of leaves)**	**Protein content (mg g^{-1}F.W. of leaves)**	**Nitrate Reductase Activity (µ moles NO_2^- formed g^{-1}F.W. h^{-1})**
1.	T_0 [Control$_{(no\ added\ zinc\ and\ iron)}$]	0.130^{f}	0.096^{cd}	0.250^{e}	17.453^{d}	11.071^{i}	0.801^{c}
2.	T_1 Zinc$_{(Soil)}$	0.189^{cd}	0.146^{bc}	0.355^{c}	23.293^{cd}	29.521^{e}	2.021^{c}
3.	T_2 Zinc$_{(Foliar)}$	0.172^{de}	0.101^{c}	0.362^{bc}	24.687^{cd}	50.963^{b}	4.343^{ab}
4.	T_3[Zinc$_{(Soil)}$+Zinc$_{(Foliar)}$]	0.152^{ef}	0.182^{b}	0.386^{ab}	26.757^{bcd}	20.908^{g}	1.602^{c}
5.	T_4 [Iron $_{(Soil)}$]	0.253^{b}	0.122^{c}	0.327^{d}	23.460^{cd}	18.246^{h}	0.895^{c}
6.	T_5 [Iron$_{(Foliar)}$]	0.337^{a}	0.118^{c}	0.243e	35.527^{abc}	40.783^{d}	1.069^{c}
7.	T_6 [Iron$_{(Soil)}$+ Iron$_{(Foliar)}$]	0.205^{c}	0.048d^{e}	0.252^{e}	27.967^{bcd}	27.158^{f}	4.143^{b}
8.	T_7 [Zinc$_{(Soil)}$ + Iron $_{(Soil)}$]	0.146^{ef}	0.037^{e}	0.343^{cd}	21.187^{d}	51.104^{b}	0.938^{c}
9.	T_8 [Zinc$_{(Soil)}$ + Iron $_{(Soil)}$ + Zinc$_{(Foliar)}$]	0.208^{c}	0.039^{e}	0.325^{d}	38.607^{ab}	46.513^{c}	1.638^{c}
10.	T_9 [Iron$_{(Soil)}$ + Iron $_{(Foliar)}$ + Zinc$_{(Foliar)}$]	0.213^{c}	0.120^{e}	0.256^{e}	38.833^{ab}	22.050^{g}	2.052^{c}
11.	T_{10} [Zinc$_{(Soil)}$ + Iron $_{(Soil)}$ + Zinc$_{(Foliar)}$+ Iron $_{(Foliar)}$]	0.352^{a}	0.254^{a}	0.407^{a}	46.447^{a}	54.521^{a}	6.207^{a}
	SE (m)±	0.01	0.017	0.009	4.446	0.533	0.660
	C.D.*	0.029	0.052	0.027	13.123	1.573	1.948

* Denotes significance at P= 0.01. Means followed by same letters in a column are not significantly different but different letters are significantly different, using Duncun's Multiple Range Test (DMRT).

As per treatments, the second foliar spray was given at 45 DAS and the observations were recorded after 15 days of spray application i.e. 60 DAS.

Conclusion

On the basis of present investigation, it was found that all the biochemical parameters were found significant at 0.01 level of significance. Zinc foliar spray showed better performance with protein content, total soluble sugar, nitrate reductase activity, chlorophyll and carotenoid contents. When iron was applied as soil and foliar separately i.e. T_4 and T_5, did not show significant increase but T_7 (both soil and foliar treatment of iron) showed a significant increase in case of nitrate reductase activity. Treatment T_{10} consisting both soil and foliar application of zinc and iron performed extensively with both morphological as well as biochemical parameters in zinc deficient soil and considered as prime treatment that can be applied in zinc deficient soil.

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