Original Research Article

Effect of Zinc Fertilization on Nodulation, Chlorophyll, Yield and Quality of Soybean Grown in a Vertisol of Central India

Abstract

Under intensive cultivation, Zinc (Zn) fertilizer is very commonly used in deficient soils of central, India. Therefore, it is required to find out the optimum Zn dose for sustainable productivity and maintenance of soil fertility. The study comprised of three frequency levels, *i.e.*, Zn application in the 1st year only (F1), alternate year (F2), and every year (F3), with five different rates of Zn, *i.e.*, 2.5, 5.0, 7.5, and 10.0 kg Zn ha^{-1} imposed in the *Kharif* season. Result revealed the highest symbiotic traits *viz.*, nodule number of 36.78, fresh and dry weight (1.09 and 0.31 g plant^{-1}) were found with the application of 5 kg Zn ha^{-1} at 45 DAS at alternate year. The highest SPAD value of 44.98 and relative water content of 62.41 % were recorded with 5 kg Zn ha^{-1} at 60DAS and found significant at alternate year. The Zn addition @ 5.0 kg of Zn ha^{-1} through $ZnSO_4.7H_2O$ to soybean in alternate year which increase by 1.28 to 1.53 (19.53 %) over only single year Zn addition. This was the best practice in a typic haplusterts as it resulted in an average production of 1.54 t ha^{-1} of seed yield ha^{-1} over no zinc application (1.24 t ha^{-1}) which increase by 24.19 %. The zinc application improved quality of seed in terms of oil and protein over control.

Key word: Zinc fertilization, Soybean, Vertisol, SPAD, Nodulation, Relative Water Content

1.0 Introduction

Soybean (*Glycine max* L.) is one of the most valuable food crops in the global oilseed cultivation scenario due to its high productivity and vital contribution to maintaining soil fertility (Mathu et al., 2012). It fixes up to 200 kg N ha^{-1} year^{-1} under optimal field conditions (Cheminingwa et al., 2007). It is composed of 40 per cent protein and 20 per cent oil contributing about two-thirds of the global protein concentrates and 25% of the world's vegetable oil production (Mauyo et al., 2010). It contains essential amino acids, carbohydrates, unsaturated fatty acids, vitamins and minerals. The nutritional value of the crop is highly dependent on the presence of essential nutrients which plays a vital role in the quality and quantity of soybean crops and deficiencies of these nutrients will drastically reduce their growth and yield. In India, soybean is grown an area of 12.07 M ha with a production 13.98 m tonnes and productivity of 1158 kg ha^{-1}. Madhya Pradesh is the leading state with an area of 6.50 m ha with a production 4.61 m tonnes and productivity of 710 kg ha^{-1} (IISR, 2022). The yield of soybean is low due to nutrient deficiencies (Mahasi et al., 2011).

Zinc (Zn) is an important micronutrient for plants since it is involved in many key cellular functions such as metabolic and physiological processes, enzyme activation, and ion homeostasis (Yang et al., 2020; Alsafran et al., 2022). Additionally, Zn is a crucial nutrient for plants and is involved in a number of their bio-physicochemical responses including gene regulation and activation, protein synthesis, involvement in carbohydrate metabolisms, and morphological and anatomical participation in bio-membranes (Zaheer et al., 2022). Zn has an important role as a constituent of over 300 enzymes from all six enzyme classes. This is the only element found across all six enzyme classes (lyases, transferases, hydrolases, isomerases, oxidoreductases, and ligases). Zn influences the activity, structural integrity, and folding of numerous proteins as a fundamental or catalytic enzyme. Therefore, it is crucial for plants to properly absorb, transport, and distribute Zn in their tissues, cells, and intracellular locations to ensure the proper functioning of the plant (Zlobin, 2021). Despite its importance, Zn deficiency disrupts the basic operations of plant metabolism, causing growth retardation and leaf chlorosis, which can impair nutrient intake and eventually contribute to Zn deficiency in the human diet (Li et al., 2013).

Zn deficiency is a serious problem in agricultural soils across the world, due to limited amount of Zn present in soils is available for plant uptake, because the entire Zn is available in structural minerals and absorbed by other soil components (Ali et al., 2022). Zn deficiency is becoming a severe agricultural concern due to its poor availability in arable soils globally, resulting in decreased crop output and nutritional quality (Zeng et al., 2021). Numerous factors negatively affect the available Zn in soils for plants, including low/total Zn contents, high pH >7, low redox potential, prolonged flooding, microbial communities in the rhizosphere, high organic matter, high calcium carbonate and bicarbonate contents, high iron/manganese oxide contents, high exchangeable magnesium/calcium ratio, high sodium and phosphorus availability (Zhang et al., 2012; Moreno-Lora and Delgado, 2020). In India, soil zinc deficiency is estimated to be 49% (Sharma, 2008). The deficiencies were found to be even more severe (57 %) in the soils of Madhya Pradesh India (Shukla and Tiwari 2016). Nowadays, zinc fertilizer used for quality food production globally. Knowledge about proper application time and rates of Zn fertilizers is necessary to obtain higher crop production.

2.0 Material and Methods

2.1 Location of experimental site:

A field experiment was conducted at Research Farm of JNKVV, Jabalpur (M.P.) during *Kharif* season of 2016. There were three main treatments (Frequencies of Zn application) as single year application (Zn applied during *Kharif* 2012), alternate year application (Zn applied during *Kharif* 2012, 2014 and 2016) and each year application (Zn applied during *Kharif* 2012, 2013, 2014, 2015 and 2016) and five levels of Zn (0, 2.5, 5, 7.5 and 10 kg ha^{-1} through zinc sulphate) as sub treatments were applied to soybean. These fifteen treatments randomly allocated in split plot design with three replications. The basal dose of 20 kg N, 80 kg P_2O_5 and 20 kg K_2O ha^{-1} was applied to soybean crop at the time of sowing. Soybean variety (JS-9752) was sown at 40 cm row to row distance 18/7/2016 at the rate of 100 kg seed ha^{-1} and was harvested in first week of Nov 2016 (01/11/2016). The soil and plant samples were collected, processed and analyzed in laboratory using standard method. The sample of seed and stover of soybean was taken for chemical analysis. The grain yield was subtracted from bundle weight for obtaining the stover yield.

2.2 SPAD reading

The SPAD-502 a hand-held chlorophyll meter (Minolta Corporation) was used for rapid and non-destructive estimation of chlorophylls in leaves. The mean of three readings from the portable chlorophyll meter (SPAD-502, Minolta, Japan) was recorded for different treatment of soybean leaf. This instrument uses a silicon photo-iodide to detect transmittance of light emitted by two light emitting diodes through a leaf sample, one with peak emittance 650 nm where absorbance by chlorophylls is high and relatively unaffected by carotene and one with peak emittance at 940 nm where absorbance by chlorophyll content index was recorded in upper 4^{th} and 5^{th} leaf at 30, 45 and 60 DAS.

2.3 Relative water content % (RWC)

At the mid of flowering, heading and grain filling stages, the flag leaves of plants were selected for measuring the relative water content, chlorophyll content, stomatal conductance, quantum yield and electrical conductivity. Weight of fresh tissue of flag leaves was measured just after detached from the plants then taken turgid weight after leaf was incubated in distilled water for 24 h to obtain a full turgidity. Dry weight of leaf was measured after it was dried at 60°C for 24 h in an oven.

RWC (%) = [(FW − DW)/ (TW − DW)] ×100, where, RWC, FW, DW and TW are relative water content, fresh weight, dry weight and turgid weight, respectively.

2.4 Symbiotic traits

Nodulation study was done by uprooting three plants per plot by placing notch at 30, 45 and 60 DAS. The rhizosphere soil was washed in the running water and no. of nodules per plant was counted. Fresh weight of nodules was recorded and kept in small paper bags then then kept in hot air oven at 70 ± 2°C for 24 hours (till constant weight) to record their dry weight.

2.5 Statistical analysis

Analysis of variance and mean comparisons were performed computer software packages. The main effects and interactions were tested using the least significant difference (LSD) test at the 0.05 probability level.

3.0 Results and Discussion

3.1 Plant height, SPAD and RWC

3.1.1 Plant height

The data presented in Table 1 revealed that the application of Zn at different frequencies did not bring about the significant increase in plant height at 30 DAS. Only, each year application of Zn increased plant height significantly but it was at par with alternate year Zn application at 45 and 60 DAS. At 60 DAS, plant height was observed to be 44.21 cm with single year application of Zn which significantly increased to 46.26 cm and 46.18 cm with alternate and each year Zn application, respectively.

At 30 DAS, the height of plant was increased significantly up to 7.5 kg Zn ha^{-1} application which varied from 25.22 cm at control to 28.28 cm at 7.5 kg Zn ha^{-1}. The levels of Zn @ 2.5, 5.0, 7.5 and 10 kg Zn ha^{-1} significantly superior over control but among themselves they were statistically at par. At 45 DAS, height of plant increased significantly with the application of Zn up to 7.5 kg Zn ha^{-1}. It varied from 39.19 cm at control to 44.28 cm at 7.5 kg Zn ha^{-1}. However, levels of Zn were statistically at par among themselves at 45 and 60 DAS. The plant height was also increased significantly with the increasing levels of Zn over control at 60 DAS. The lowest height 42.11 cm was observed at control and maximum height 47.19 cm was observed at 7.5 kg Zn ha^{-1}. The application of 7.5 kg Zn ha^{-1} also showed its superiority over control and 2.5 kg Zn ha^{-1} rest of treatment. During advanced growth stages, the control plot was unable to fulfill the need of nutrients by the crop; which resulted in production of the shortest plants. The deficiency of Zn was limited growth (Fageria 2000). But other treated plots might be able to fulfill the minimum need of nutrients by the crop, hence they produced plants of identical or more height. Zinc is required as structural and catalytic components of protein and enzymes for normal growth and development (Broadley et al., 2007). Application of Zn has been reported significant positive effects, on growth by 50% in soybean (Gadallah, 2008).

3.1.2 SPAD reading (Chlorophyll content index)

The maximum SPAD value was recorded with alternate year Zn application which was significantly higher than single and each year of Zn application at 30, 45 and 60 DAS. Result showed increased with the advancement of crop age and reached its peak at general flowering stage (60 DAS). Significant elevation due to treatment was observed over control at 45 and 60 DAS. Significantly maximum content of chlorophyll (37.00, 36.98 and 37.73, respectively) was exhibited by the application of 5.0, 7.5 and 10.0 kg Zn ha^{-1} at 30 DAS. However, the highest SPAD value (44.98) was noted under the application of 5.0 kg Zn ha^{-1} which was statistically at par with the levels consisting 2.5, 7.5 and 10 kg Zn ha^{-1} at 60 DAS.

Zn maintains chlorophyll synthesis through sulphydryl group protection, a function primarily associated with Zn. The lowest SPAD of 35.68, 36.51 and 41.17 was recorded in control at 30, 45 and 60 DAS, respectively. The increased chlorophyll contents are due to zinc which acts as a structural and catalytic component of proteins, enzymes and as co-factor for normal development of pigment biosynthesis (Balashouri, 1995). The reduction of SPAD value was probably related to the enhanced activity of the enzyme chlorophyllase and inducing the destruction of chloroplast structure and the instability of pigment protein complex. Zinc deficiency can cause a drastic decrease in chlorophyll content as well as a severe damage to the fine structure of chloroplasts (Hisamitsu et al. 2001;Pandey et al., 2006; Kobraee et al., 2011) Zn inhibited the chlorophyll production by interfering with Fe metabolism, but not by lowering the Fe content of the leaves (Rosen et al., 1977). Therefore, Zn disrupts photosynthetic activity, stomatal conductance, and numerous other vital metabolic functions. Zn toxicity reduces plant development and structural stability (Chakraborty and Mishra, 2020), and promotes chlorosis of leaves (Bankaji et al., 2019).

3.1.3 Relative water content (RWC %)

The highest RWC % was recorded with an alternate-year Zn application but could not significantly increase. Treatment 5.0 kg Zn ha^{-1} possessed the highest RWC% (62.41) but it was at par with 2.5 and 10 kg Zn ha^{-1}. The interaction between frequencies and levels of Zn was non-significant. Sharma et al., 1995 observed a decrease in the K^+ content of guard cells in non-zinc application plants. Zinc may participate in stomatal regulation due to its role in maintaining membrane integrity. Silva et al. (2010) stated that these parameters decrease in most plants under water deficit. A decrease in RWC in plants under drought stress may depend on plant vigor reduction and have been observed in many plants (Liu et al. 2002). The decrease in leaf relative water content could be related to low water availability under stress conditions or to root systems, which are not able to compensate for water lost by transpiration through a reduction of the absorbing surface. The amelioration role of Zn in maintenance of relative water content might be attributed to improvement of vascular tissue (Gadallah 2000).

3.2 Symbiotic traits

3.2.1 Nodule number plant^{-1}

The number of nodules was significantly affected due to treatment but the interaction was found to be statistically non-significant. At 30 DAS, alternate year Zn application was found statistically non-significant while, at par with each year and it produced significantly higher nodule at 45 and 60 DAS over single year application. The lowest number of nodules was observed to be 10.67, 32.02 and 29.98 with single year Zn application at 30, 45 and 60 DAS, respectively. The no of nodules significantly increased to 34.84 and 34.53 with alternate and each year application of Zn at 45 DAS. Similarly, no of nodules increased to 31.16 and 30.84 with alternate and each year application at 60 DAS but both the frequencies were found to be on par at 45 and 60 DAS.

The maximum number of nodules plant^{-1} was found at 45 DAS thereafter showed decline at 60 DAS. At 30 DAS, the application of 2.5 kg Zn ha^{-1} bring significant enhancement in nodule number over control but it was also at par with 7.5 kg Zn ha^{-1}. The number of nodules, increased significantly due to application of 5.0, 7.5 and 10 kg Zn ha^{-1} over control and 2.5 kg Zn ha^{-1} at 45 and 60 DAS. Furthermore, the application of 5 kg Zn ha^{-1} produced the highest mean nodule number (36.78) which was closely followed by 7.5 kg Zn ha-1 (36.41) and 10 kg Zn ha^{-1} (35.70) at 45 DAS and these were at par among themselves.

3.2.2 Nodule fresh weight (g plant^{-1})

The fresh weight of nodules was observed to be 0.32 g with single year application of Zn which was non-significant at 30 DAS but at 45 and 60DAS it was significantly increased to 1.048 g and 0.897 g with alternate year application of Zn. Alternate and each year were found to be statistically at par at 60 DAS but at 45 DAS each year Zn application was non-significant.

At 30, 45 and 60 DAS, the highest nodule fresh weight was observed to be 0.34, 1.09 and 0.92 g plan^{t-1} respectively with of 5 kg Zn ha^{-1} application. Fresh weight of nodules increased significantly with the level of 5.0, 7.5 and 10 kg Zn ha^{-1} at 30 DAS over control. However, At 45 and 60 DAS all the Zn levels were recorded significantly higher nodule fresh weight over control but 5.0, 7.5 and 10 kg Zn ha^{-1} were statistically at par at 45 DAS. The levels of 5.0 and 7.5 kg Zn ha^{-1} produced identical at 60 DAS.

3.3.3 Nodule dry weight (g plant^{-1})

Each year, Zn applications could bring about only numerical rise in nodule dry weight over the control but it was statistically non-significant at 30 DAS. The alternate-year Zn application produced significantly higher nodule dry weight at 45 and 60 DAS than single - year Zn application. However, alternate and each-year of Zn application showed similar fresh weight. The nodule dry weight plant^{-1} at 30, 45 and 60 DAS was significantly affected by the increasing levels of Zn. At 30 DAS, not much difference was recorded in dry weight of nodules but with the advancement of crop ages it was increased and the highest with 7.5 kg of Zn ha^{-1} (0.310 g plant^{-1}) and 5.0 kg Zn ha^{-1} (0.283 g plant^{-1}) application at 45 and 60 DAS, respectively. Yet these levels were at par with just higher and lower levels of Zn.

The data on symbiotic traits of soybean indicated that nodule number, fresh and dry weight of nodules increased progressively at 30 and 45 DAS, but the decline was noted at 60 DAS. This variation observed in soybean seems to be due to decay of nodular tissues at pod formation stage. The decline in quantity of nodule leghaemoglobin is logical as the nodules formed on the plant root system start decaying with the initiation of podding in the leguminous crops.

Zn is needed for chlorophyll formation, nodulation, growth hormone stimulation, lipid and protein metabolism, carbohydrate synthesis, enzymatic activity and reproductive processes (Thenua et al., 2014). It plays a vital role in photosynthesis, synthesizing RNA and DNA, synthesis of auxins, nitrogen fixation and production of biomass. The findings are in agreement with those reported by Awlad et al. (2003) Surywanshi et al. (2015); Kulhare et al. (2014); Lamptey et al. (2014) and Vollmann et al., 2011) which showed that increasing level of zinc increased the number of nodules. symbiosis and nodulation.

3.3 Effect of Zn application on yield attributes, yield and quality of soybean

The data on various parameters, *viz.*, number of pods plant^{-1}, test weight (g), seed and stover yield (t ha^{-1}) and quality of soybean as influenced by the application of zinc, are presented in Table 3. The interaction effect between Zn levels and frequencies was found to be statistically non-significant.

3.3.1 Number of pods plant^{-1}

Data revealed that, the number of pods plant^{-1} was observed to be 65.73 with single - year application of Zn which significantly increased to 68.55 and 68.81 with alternate and each-year application of Zn. However, alternate and each-year application of Zn were statistically on par. The number of pods plant^{-1} was minimum (61.57) at control and maximum (71) at 5.0 kg Zn ha^{-1}. The treatments of 7.5 and 10 kg ha^{-1} levels were statistically at par but significant over control and 2.5 kg Zn ha^{-1}. Moreover, the application of 5 kg Zn ha^{-1} produced significantly higher pods $plant^{-1}$ than all treatments.

3.3.2 Seed test weight (g)

Data indicated that the seed test weight was observed of 7.62 g with single year application of Zn which significantly increased to 7.87 g and 7.79 g with alternate and each year application of Zn, respectively. However alternate and each year application of Zn were statistically on par. The interaction effect between Zn levels and frequencies was found to be statistically non-significant. Seed test weight increased significantly with 5.0, 7.5 and 10 kg Zn ha^{-1} application over control but these levels of Zn were statistically at par among themselves. Application of 2.5 kg Zn ha^{-1} application did not bring a significant increase in seed test weight but it was equal with higher levels. The minimum seed test weight was obtained with the control (7.50 g ha-^{1}) and the highest was found at 5 kg Zn ha^{-1} (8.01 g).

3.3.3 Seed and stover yield (t ha^{-1})

The data pertaining to the seed and stover yields of soybean have been presented in Table 3. The seed yield was observed to be 1.28 t ha^{-1} with single-year application of Zn, which significantly increased to 1.48 and 1.53 t ha^{-1} with each and alternate-year application of Zn, respectively. Alternate and each-year application of Zn were statistically equal. The interaction effect of Zn levels and their frequencies produced non-significant result. Seed yield due to different Zn levels varied from 1.24 t ha^{-1} (control) to 1.54 t ha^{-1} (5 kg Zn ha^{-1}). Application of 5.0 kg Zn ha^{-1} recorded significant and the highest seed and stover yield (1.54 t ha^{-1} and 3.28 t ha^{-1}) over control which, was on par with that obtained with 7.5 and 10 kg Zn ha^{-1}. Alternate year Zn application increased the stover yield significantly. However, single and each year application of Zn were statistically non-significant. Minimum stover yield 2.61 t ha^{-1} was obtained in control and maximum 3.28 t ha^{-1} with the application of 5 kg Zn ha^{-1}. Application of increasing levels of Zn significantly increased the stover yield of soybean over control but the increasing levels of Zn levels were found statistically at par among themselves for stover yield of soybean. The increase in yield may be attributed to beneficial effect of zinc on growth parameter yield attributing parameters, which finally reflected in increased both grain and straw yield. The similar result reported by Nandanwar et al., (2007) reported that grain and straw yield of soybean increased significantly with Zn 5.0 kg Zn application as compared to control. Pable et al. (2010) reported that zinc application increased the grain and straw yield of soybean over control. Similar results have been also reported by Kanase et al. (2008) Kulhare et al. (2014). Application of Zn has been reported significant positive effects, on growth dry matter production by 50% in soybean (Gadallah, 2008; Cakmak, 2008).

3.3.4 Quality of seed of soybean

The data given in Table 3 on protein and oil content in seed were resulted 39.34 % and 39.26 % and 19.30 % and 19.71 % with alternate and each year application of Zn, respectively, and these both were statistically at par. The oil content was observed to be 18.25% with single year application of Zn which significantly increased with alternate and each year application of Zn. However, it significantly increased with increasing Zn levels over control. The highest protein content in seed, 39.76% was analyzed with application of 10 kg Zn ha^{-1}. Yet it was on par with 7.5 and 5 kg Zn ha^{-1}. The increasing levels of Zn significantly increased the oil content in seed over control and 2.5 kg Zn ha^{-1}. The minimum oil content of 18.03 % was found at control and maximum of 19.94 % was found at 5 kg Zn ha^{-1}. The interaction effect was found to be statistically non-significant. According to Hafeez et al., (2013) low Zn levels actually inhibit the synthesis of RNA and proteins since Zn plays a significant role in several enzymes that are essential for protein synthesis, including glutamate dehydrogenase, ATPase, and ribonuclease. Zn deficiency causes the ribosomes to break down, which affects protein metabolism (Castillo-González et al ., 2018). These findings were in accordance with Krishna (1995) and Hisamitsu et al., 2001;Zeng et al., 2021) who also reported a significant positive effect of zinc treatment on crude protein content.

4.0 Conclusions

Zn application at alternate year and 5 kg Zn ha^{-1} was observed positively significant on SPAD value and RWC per cent. The alternate year and 5 kg Zn ha^{-1} application showed significantly higher response with respect to plant height, symbiotic traits and yield attributes over the control. Zn application at alternate year increased seed yield by 19.53% over single year and 5.0 kg Zn ha^{-1} Zn application significantly increased seed yield by 24.19 % over control (1.24 t ha^{-1}).

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Table 1 Effect of zinc application on plant height, SPAD and relative water content in leaves of soybean

Treatment		**Plant height (cm)**			**SPAD value**			**RWC (%)**
		30 DAS	**45 DAS**	**60 DAS**	**30 DAS**	**45 DAS**	**60 DAS**	
Frequencies								
F1 (Single)		27.19	41.37	44.21	35.29	36.19	42.17	59.39
F2 (Alternate)		27.59	42.48	46.26	37.46	39.44	44.92	61.01
F3 (Each)		27.01	44.28	46.18	37.38	38.24	44.74	60.16
SEm±		**1.048**	**0.473**	**0.408**	**0.513**	**0.6**	**0.577**	**0.279**
CD(P=0.05)		**NS**	**1.858**	**1.603**	**2.015**	**2.358**	**2.269**	**1.097**
Zn Levels (kg ha^{-1})								
0		25.22	39.19	42.11	35.68	36.51	41.17	54.25
2.5		27.26	42.87	45.61	36.17	37.92	43.94	61.98
5		28.15	43.66	46.48	37	38.29	44.98	62.41
7.5		28.28	44.28	47.19	36.98	38.11	44.87	60.43
10		27.39	43.56	46.35	37.73	38.96	44.75	61.86
SEm±		**0.48**	**0.586**	**0.523**	**0.452**	**0.394**	**0.591**	**0.558**
CD(P=0.05)		**1.371**	**1.675**	**1.493**	**1.29**	**1.125**	**1.689**	**1.595**
FXZn*	SEm±	**0.831**	**1.016**	**0.905**	**0.782**	**0.682**	**1.024**	**0.967**
	CD(P=0.05)	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**
FXZn**	SEm±	**2.224**	**1.311**	**1.149**	**1.241**	**1.346**	**1.474**	**1.029**
	CD(P=0.05)	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**

**Comparison of Two Zn levels at the same frequency.* RWC – Relative Water Content *NS – Non-Significant*
***Comparison of Two frequencies at the same or different levels of Zn.*

Table 2 Effect of zinc application on symbiotic traits of soybean

Treatment		No. of nodule plant^{-1} 30 DAS	45 DAS	60 DAS	NFW plant^{-1} (g) 30 DAS	45 DAS	60 DAS	NDW plant^{-1} (g) 30 DAS	45 DAS	60 DAS
Frequencies										
F1 (Single)		10.67	32.02	29.98	0.324	0.972	0.835	0.122	0.3	0.276
F2 (Alternate)		11.37	34.84	31.16	0.352	1.048	0.897	0.125	0.308	0.281
F3 (Each)		11.31	34.53	30.84	0.324	1.001	0.879	0.125	0.308	0.278
SEm±		**0.248**	**0.304**	**0.203**	**0.0086**	**0.0079**	**0.0114**	**0.0008**	**0.0016**	**0.0014**
CD(P=0.05)		**NS**	**1.195**	**0.797**	**NS**	**0.0312**	**0.0446**	**NS**	**0.0064**	**0.0057**
Zn Levels (kg ha^{-1})										
0		9.28	28.56	28.63	0.315	0.886	0.803	0.121	0.296	0.271
2.5		10.85	31.56	29.63	0.319	0.973	0.853	0.123	0.304	0.277
5		12.07	36.78	32.26	0.34	1.099	0.92	0.126	0.31	0.283
7.5		11.48	36.41	31.26	0.349	1.029	0.9	0.125	0.309	0.281
10		11.89	35.7	31.52	0.344	1.048	0.877	0.123	0.307	0.28
SEm±		**0.256**	**0.545**	**0.332**	**0.0073**	**0.0135**	**0.0167**	**0.0009**	**0.0017**	**0.0016**
CD(P=0.05)		**0.73**	**1.557**	**0.949**	**0.0209**	**0.0385**	**0.0476**	**0.0025**	**0.0049**	**0.0046**
FXZn*	SEm±	0.443	0.944	0.575	0.0126	0.0234	0.0289	0.0015	0.003	0.0028
	CD(P=0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS
FXZn**	SEm±	0.635	1.041	0.655	0.0207	0.0262	0.0344	0.0021	0.0042	0.0038
	CD(P=0.05)	NS	NS	NS	NS	NS	NS	NS	NS	NS

*Comparison of Two Zn levels at the same frequency. **Comparison of Two frequencies at the same or different levels of Zn.

NFW – Nodule Fresh Weight, NDW – Nodule Dry Weight, NS – Non Significant T

Table 3 Effect of zinc application on yield attributes, yield and quality

Treatment		No. of pods plant^{-1}	Test weight (g)	Yield (t ha^{-1})		Seed quality (%)	
				Seed	Stover	Oil	Protein
Frequencies							
F1 (Single)		65.73	7.62	1.28	2.84	18.25	37.95
F2 (Alternate)		68.55	7.87	1.53	3.43	19.3	39.34
F3 (Each)		68.81	7.79	1.48	2.96	19.71	39.26
SEm±		**0.41**	**0.036**	**0.042**	**0.099**	**0.257**	**0.32**
CD(P=0.05)		**1.613**	**0.143**	**0.167**	**0.388**	**1.012**	**1.25**
Zn Levels (kg ha^{-1})							
0		61.57	7.5	1.24	2.61	18.03	37.43
2.5		67.08	7.74	1.41	3.19	19.02	38.89
5		71.57	8.01	1.54	3.28	19.94	39.39
7.5		69.29	7.82	1.47	3	19.88	39.53
10		68.98	7.78	1.46	3.19	19.73	39.76
SEm±		**0.502**	**0.097**	**0.042**	**0.13**	**0.169**	**0.257**
CD(P=0.05)		**1.435**	**0.278**	**0.12**	**0.372**	**0.482**	**0.735**
	SEm±	**1.468**	**0.168**	**0.073**	**0.87**	**0.292**	**0.446**
FXZn*	CD(P=0.05)	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**
	SEm±	**1.702**	**0.167**	**0.107**	**1.131**	**0.577**	**0.755**
FXZn**	CD(P=0.05)	**NS**	**NS**	**NS**	**NS**	**NS**	**NS**

**Comparison of Two Zn levels at the same frequency. NS – Non-Significant*

***Comparison of Two frequencies at the same or different levels of Zn.*