

Original Research Article

Effect of Zinc and Bio-fertilizers on growth and yield of Wheat (*Triticum aestivum* L.).

ABSTRACT

A field experiment was conducted during *Rabi* season of 2021-22 at the experimental field of the Crop Research Farm, Department of Agronomy, Naini Agricultural Institute, Sam Higginbottom University of Agriculture, Technology And Sciences, Prayagraj, Uttar Pradesh, India. The Experiment was laid out in Randomized Block Design with Nine treatments replicated thrice. ~~To and to~~ study the Response of Zinc and Bio-fertilizers on growth and yield of Wheat. The treatments consist of zinc 5, 7.5, 10kg/ha and bio-fertilizers *Azotobacter* (20 g/kg seeds), *Azospirillum* (20 g/kg seeds), *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds). The soil of experimental plot was sandy loamy in texture, nearly neutral in soil reaction (pH 7.8), low in organic carbon (0.35%). The results revealed that treatment ~~9 with the 9th~~ application of [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)] was recorded significantly higher plant height ~~(of 96.10 cm)~~, higher plant dry weight ~~(of 20.31 g/plant)~~, higher number of spikes/m² ~~(of 371.84)~~, higher number of grains/spike ~~(of 55.17)~~, higher grain yield ~~(of 5.697 t/ha)~~, higher straw yield ~~(of 12.59 t/ha)~~ and highest harvest index ~~(of 31.14 %)~~. ~~Higher-%. Including higher gross INR returns (INR of 152544.4/ha)~~, higher net returns ~~(INR of 108712.4/ha)~~ and higher B:C ratio ~~(of 2.48)~~ were also recorded in ~~the 9th~~ treatment ~~9 of~~ [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)].

Keywords: *Wheat, zinc, bio-fertilizers, azotobacter, azospirillum, growth parameters, yield attributes and economics.*

Style Definition: Normal: Font: (Default) Calibri, English (India), Space After: 8 pt, Line spacing: Multiple 1.08 ii, Widow/Orphan control, Adjust space between Latin and Asian text, Adjust space between Asian text and numbers

Style Definition: Heading 2

Style Definition: Header: Font: (Default) Calibri, English (India), Widow/Orphan control, Adjust space between Latin and Asian text, Adjust space between Asian text and numbers, Tab stops: 3.25", Centered + 6.5", Right + Not at 3.13" + 6.27"

Style Definition: Footer: Font: (Default) Calibri, English (India), Widow/Orphan control, Adjust space between Latin and Asian text, Adjust space between Asian text and numbers, Tab stops: 3.25", Centered + 6.5", Right + Not at 3.13" + 6.27"

INTRODUCTION

Wheat is cultivated in most of parts of the world. It is staple diet for majority of the population in both developed and developing countries. Wheat compares well with other important cereals in the nutritive value. It contains more protein than other cereals. The nutritive values of wheat are starch (60-68%), protein (8-15%), fat, sugar, cellulose, minerals, vitamins, etc. (Singh S.S., 2000).

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

In India wheat crop is cultivated in *Rabi* season. It is normally sown during November and harvested between March and April. The cultivated area under wheat at national level has shown increasing trend, from 29.04 million hectare to 30.54 million hectare with a magnitude of 1.5-million-hectare (5%) net gain in terms of area. Uttar Pradesh has largest share in area with 9.75 million hectare (32%), followed by Madhya Pradesh (18.75%), Punjab (11.48%), Rajasthan (9.74%), Haryana (8.36%) and Bihar (6.82%). The national productivity trend for wheat showed a marginal improvement, which has increased from 3009 kg/ha to 3100 kg/ha from 2012-2013 to 2017-2018. The rise in productivity is due to adoption of high-yielding varieties coupled with other inputs (Sendhil et al., 2019).

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

According to the fourth advance estimates of production of major crops for 2021-22 released, record food grains production of 315.72 million tonnes estimated in the country, which is higher by 4.98 million tonnes than the production of food grains during 2020-21. Production of wheat during 2021-22 is estimated at 106.84 million tonnes. It is higher by 2.96 million tonnes than the last five years average wheat production of 103.88 million tonnes (GOI-2021-22).

Wheat is known to respond to the application of several macro and micronutrients during its growing stages and results in enhanced output in terms of yield. Micronutrients comprising zinc, copper, iron, manganese, boron, molybdenum and chlorine are though required by plants in much smaller amounts, yet are as essential as the major nutrients such as nitrogen, phosphorus, potassium etc (Singh et al., 2015).

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

Zinc has diverse physiological functions in biological systems. ~~It~~, it interacts with a large number of enzymes and other proteins in the body and performs critical structural, functional and regulatory roles. It is estimated that about 10% of all the proteins in the human body, corresponding to nearly 3000 proteins, are Zn-dependent. About a third of the world's population is estimated to be at risk of Zn deficiency. Deficiencies of Zn and other micronutrients in developing countries are also reported to cause great economic losses and have a considerable effect on the gross national product by decreasing productivity and increasing the health care costs. (Cakmak and Kutman *et al.*, 2018).

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Italic

According to recent report, deficiency of zinc is widespread and covers about 48% area in the country. Zinc deficiency is a very important nutrient problem in the Indian soils. Total Zn concentration is sufficient in many agricultural areas, but available Zn concentration is deficient because of different soil and climatic conditions. Zinc was one of the first micronutrients, essentiality of which for plant growth has been confirmed. Zinc also plays a role in nucleic acid and protein synthesis and helps in the utilization of phosphorus and nitrogen, as well as in seed formation. Zinc is an important element for terrestrial life since it is required as either a structural component or reaction site in numerous proteins. Zinc deficiency in wheat resulted in severe reduction in growth, grain yield and seedling vigour, and enhances sensitivity of plants to pathogens. Decrease in grain quality is another typical consequence of zinc deficiency in wheat. (Singh *et al.*, 2015).

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

To correct Zn deficiency and prevent yield losses in crop plants, Zn is applied to deficient soil, typically in the form of ZnSO₄, at rates that range typically from 5 to 25 kg Zn/ha. The rates of soil Zn application vary depending on the crop species, soil characteristics and method of application; higher rates are associated with crops sensitive to Zn deficiency, alkaline or calcareous soil and broadcasting rather than banding. Development of Zn-rich cultivars or the use of Zn-fertilizers is significant practices to enrich the Zn-contents of grains. (Cakmak and Kutman *et al.*, 2018).

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Azotobacter is a free living N² fixing bacterium. It can successfully grow in the rhizospheric zone of wheat, maize, rice, sorghum, sugarcane, cotton, potato and many others and fix 10-20kg N/ha cropping season. Besides N² fixation, *Azotobacter* synthesizes and secretes considerable amounts of biologically active substances like vitamin B, nicotinic acid, pantothenic acid, biotin, heteroauxins, gibberellins, etc. which enhance root growth of plants. Another important characteristic of *Azotobacter* association with crop improvement is excretion of ammonia in the rhizosphere in the presence of root exudates; which helps in modification of nutrient uptake by the plants. *Azotobacter* has the ability to produce

Formatted: Indent: First line: 0"

antifungal antibiotics and fungistatic compounds against pathogens like Fusarium, Alternaria, Trichoderma (Reddy and Singh, 2021; Wani et al., 1988).

Formatted: Font: Not Bold, Italic

Azospirillum is a diazotrophic bacteria that play a valuable role in the rhizosphere of many plants, fixes atmospheric dinitrogen and converts it into plant-available form (Rehman et al., 2017). Inoculation of wheat seeds with *Azospirillum* spp. has been widely applied to promote crop yields on different soils (Ahmed and Kibret et al., 2014). Inoculation of seed with *Azospirillum* give host plant drought tolerance and disease resistance benefits apart from nitrogen fixation (Singh et al., 2007). The bacterium obtains the nutrients necessary to replicate and survive from the plant and the plant benefits from the association with the bacterium because the bacteria produce hormones that increase the volume of the roots bulk thereby leading to improved plant nutrition. The beneficial effects of *Azospirillum* on plant growth have been applied in agricultural practices and inoculants containing *A. brasilense* have been used as biofertilizer for cereals all over the world (Reddy and Singh et al., 2021).

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Increasing and extending the role of biofertilizers can reduce the need for chemical fertilizers and decrease adverse environmental effects. They can play a significant role in fixing atmospheric N and production of plant growth promoting substances. Therefore, in the development and implementation of sustainable agricultural techniques, biofertilization has great importance in alleviating environmental pollution and deterioration of nature. *Azotobacter* sp. and *Azospirillum* sp. are used as biofertilizers in the cultivation of many agricultural crops. The estimated contribution of these free-living N fixing prokaryotes to the N input of soil ranges from 0–60 kg/ha per year.

Existence of microbial communities like *Azotobacter* sp. and *Azospirillum* sp. in the rhizosphere promotes the growth of the plant through the cycling and availability of nutrients, increasing the health of roots during the growth stage by competing with root pathogens and increasing the absorption of nutrients and water (Namvar and Khandan et al., 2013; Namvar et al., 2012; Vessey, 2003). (Kandil et al., 2011) studied the effects of inoculation with *Azotobacter* sp. and *Azospirillum* sp. on wheat and observed that inoculated wheat plants gave higher plant height, spike per unit of area, grains per spike, grain weight, biological yield, grain yield and straw yield compared to non-inoculated cultivars. (Zorita and Ganigja et al., 2009) found that seed inoculation can increase the number of harvested grains by 6.1% and grain yield by 260 kg ha⁻¹ (8.0%) in wheat. (Piccinin et al., 2013) suggested that the combination between chemical fertilizers and microbial strains *Azotobacter* sp. and *Azospirillum* sp. caused an enhancing in plant growth parameters and yield component in wheat plants.

Formatted: Font: Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Therefore, the objective of the present work was to assess the effects of different levels of zinc in combination with bio-fertilizer (*Azotobacter* sp., *Azospirillum* sp.) on growth parameters, yield attributes and economic analysis of wheat.

Formatted: Indent: First line: 0"

MATERIALS AND METHODS

The experiment was conducted during *Rabi* season of 2021-22 at the Crop Research Farm, Department of Agronomy, Naini Agricultural Institute, Sam Higginbottom University of Agriculture, Technology And Sciences, Prayagraj (U.P.) India, which is located at 25° 30' 42" N latitude, 81° 60' 56" E longitude, and a height of 98 metres above sea level. The soil of experimental plot was sandy loamy in texture, nearly neutral in soil reaction (pH 7.8), low in organic carbon (0.35%). The crop was sown on 11 December 2021 using variety HD-2967. The experiment was set up in a Randomized Block Design with three replications and nine treatments in total *Viz.*, T₁: Zinc (5 kg/ha) + *Azotobacter* (20 g/kg seeds), T₂: Zinc (5 kg/ha) + *Azospirillum* (20 g/kg seeds), T₃: Zinc (5 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds), T₄: Zinc (7.5 kg/ha) + *Azotobacter* (20 g/kg seeds), T₅: Zinc (7.5 kg/ha) + *Azospirillum* (20 g/kg seeds), T₆: Zinc (7.5 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds), T₇: Zinc (10 kg/ha) + *Azotobacter* (20 g/kg seeds), T₈: Zinc (10 kg/ha) + *Azospirillum* (20 g/kg seeds), T₉: Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds). The growth parameters and yield attributes were recorded at harvest from randomly selected plants in each plot. The data were subjected to statistical analysis by analysis of variance method (Gomez and Gomez *et al.*, 1984).

Formatted: Indent: First line: 0", Right: 0"

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Bold

RESULT AND DISCUSSION

Effect of Zinc and Bio-fertilizers on growth parameters of wheat.

Plant height (cm)

At 100 DAS, the significantly and higher plant height (of 96.10 cm) as seen in Table-1 was observed in the 9th treatment-9 [of Zinc (at 10 kg/ha) + *Azotobacter* (of 10 g/kg seeds) + *Azospirillum* (at 10 g/kg seeds)]. However, treatment-7 [Zinc (10 kg/ha) + *Azotobacter* (20 g/kg seeds)] and treatment-8 [Zinc (10 kg/ha) + *Azospirillum* (20 g/kg seeds)] were statistically at par with treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. Significant and higher plant height was recorded with the application of Zinc (10 kg/ha) may be due to the Zn major role in the shoots and roots elongation due to auxin hormones activation in the wheat crop plant. Similar findings were obtained by (Arshad *et al.* (2016) and (Khan *et al.* (2007). Further, inoculation with

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

biofertilizer *Azotobacter* (10g/kg seeds) and *Azospirillum* (10g/kg seeds) increased plant height about 6.04% compared to non-inoculated plants. These results are in line with the findings of (Namvar and Khandan (et al, 2013), (Zorita and Canigia (et al, 2009), (Daneshmand et al. (2012) and (Namvar et al. (2012). Further, *Azospirillum* inoculation have suggested that Nitrogen fixation was major mechanism of plant growth. This increase in the soil nutrient level was responsible for plant growth and development. Similar findings were obtained by (Reddy and Singh (et al, 2021).

Plant dry weight (g)

At 100 DAS, the significantly higher dry weight (20.31 g) [Table-1] was observed in treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. However, treatment-8 [Zinc (10 kg/ha) + *Azospirillum* (20 g/kg seeds)] was statistically at par with treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. Significant and higher plant dry weight was recorded with the application of *Azospirillum* (10 g/kg seeds) may be due to *Azospirillum* inoculation on wheat, sorghum and panicum, which significantly increased the total shoot and root weight, plant and leaf length which and were ultimately increased dry matter accumulation and in winter, wheat inoculated with *Azospirillum brasilense* showed significantly increase in the number of fertile tillers, shoot and root dry weight and root to shoot ratio. Similar results were reported by (Reddy and Singh (et al, 2021), (Kapulnik et al. (1981) and (Warembourg et al. (1987). Further, application of *Azotobacter* (10 g/kg seeds) and *Azospirillum* (10 g/kg seeds) significantly increased dry matter accumulation at harvest over no inoculation and may be due to partly owing to their additive effect of nitrogen fixed atmosphere and partly owing to synthesis of biologically active substances like vitamins, auxin, and gibberellin etc, which in turn might have stimulated the plant growth parameters. These results are akin to the findings of (Singh et al. (2013), and (Kachroo and Razdan (et al, 2006). Further, application of zinc (10 kg/ha) resulted in significantly more dry matter production which may be due to the involvement of zinc in auxin metabolism which results in improvement in overall biomass. Similar findings were obtained by (Sharma et al. (2016).

Effect of Zinc and Bio-fertilizers on yield and yield attributes of wheat.

Number of Spikes/m²

The significant and higher number of spikes/m² (371.84) [Table-2] was observed in treatment-9 with [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. which was significantly higher over the rest of the treatments and treatment-8 [Zinc

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Indent: First line: 0"

(10 kg/ha) + *Azospirillum* (20 g/kg seeds)], was statistically at par with treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. Significant and higher number of spikes/m² was recorded with application of Zinc (10kg/ha), the increase in the number of spikes per unit area was due to the higher number of fertile tillers per plant at the end of the cycle. The lowest Zn supply to wheat plants promotes reduction in the number of tillers mainly because stems become thinner and lose their turgidity, a phenomenon called soft stems. The emergence, development and survival of tillers are of extreme importance for the crop. Similar results were reported by, (Zoz et al. (2012) and (Seadh et al. (2009). Further, with the inoculation of *Azotobacter* (10 g/kg seeds) and *Azospirillum* (10 g/kg seeds) showed no significant effects on number of spike per unit of area but the plants that were treated with these biofertilizer showed more values of this trait than non-inoculated plants. Similar results were reported by (Namvar and Khandan (et al, 2013).

Formatted: Font: Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Number of Grains/Spike

The significant and higher number of grains/spike (55.17) [Table-2] was observed in treatment-9 with [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)], which was significantly higher over rest of the treatments and treatment-8 [Zinc (10 kg/ha) + *Azospirillum* (20 g/kg seeds)], was statistically at par with treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. Significant and higher number of grains/spike was observed with the application of Zinc (10kg/ha), might be due to zinc play a significant role in enzyme activation, chlorophyll biosynthesis, pollen tube formation and pollen viability, starch utilization ensuing in greater seed set. Similar results were reported by (Arif et al. (2017). Further, *Azotobacter* (10 g/kg seeds) and *Azospirillum* (10 g/kg seeds) inoculated wheat plants had statistically more number of grains per spike than non-inoculated plants. Inoculation with biofertilizer increased this trait by 5.54% compared to plants that were not treated with inoculum. *Azotobacter* sp. and *Azospirillum* sp. increased the available nitrogen in the soil which could enhance the grain number. Similar results were reported by (Namvar and Khandan (et al, 2013) and (Vessey (2003).

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Grain yield (t/ha)

The significant and higher grain yield (5.697 t/ha) [Table-2] was observed in treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. However, treatment-8 [Zinc (10 kg/ha) + *Azospirillum* (20 g/kg seeds)], was statistically at par with

Formatted: Indent: First line: 0"

treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. Significant and higher grain yield was observed with the application of Zinc (10kg/ha), might be due to the greater photosynthesis efficiency or more nutrients availability due to increasing decomposition rate of organic matter or improved individual plant performance might the possible reasons for higher grain yield in zinc applied plots compared to other plots. These results are in conformity with the findings of (Arshad et al. (2016) and (Jan et al. (2013). Further, *Azotobacter* (10 g/kg seeds) and *Azospirillum* (10 g/kg seeds) inoculated plants indicated higher grain yield than non-inoculated plants. Biofertilizer inoculation increased grain yield about 8.68% compared to control. The increase in the yield components, grain yield and protein content in the inoculated plants could be attributable to the exudation of plant growth regulators (PGRs) such as auxins and gibberellin and cytokinin by *Azotobacter* sp. and *Azospirillum* sp. beside the increase of the nutrient availability. Similar results were obtained by (Namvar and Khandan (et al., 2013), (Kizilkaya (2008) and (Vessey (2003).

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Straw yield (t/ha)

The significant and higher straw yield (12.59 t/ha) [Table-2] was observed in treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. However, treatment-8 [Zinc (10 kg/ha) + *Azospirillum* (20 g/kg seeds)], was statistically at par with treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. Significant and higher straw yield was observed with the application of Zinc (10kg/ha), could be associated to more zinc content in the soil, which might have increased the wheat straw yield. Increasing zinc application dose increased the straw yield. Similar results were reported by (Arshad et al. (2016) and (Curtin et al. (2008). Further, the increase in straw yield with increasing dose of zinc and boron application (soil+foliar) might be due to increased photosynthetic efficiency and carbohydrate metabolism resulting in superior vegetative growth and yield attributes. Similar results were reported by (Singh et al. (2015).

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Harvest Index (%)

From the observations, the significantly highest harvest index (31.14 %) [Table-2] was recorded in treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)]. However, treatment-4 [Zinc (7.5 kg/ha) + *Azotobacter* (20 g/kg seeds)], treatment-5 [Zinc (7.5 kg/ha) + *Azospirillum* (20 g/kg seeds)], treatment-6 [Zinc (7.5 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)], treatment-7 [Zinc (10 kg/ha) +

Formatted: Indent: First line: 0"

Azotobacter (20 g/kg seeds)], treatment-8 [Zinc (10 kg/ha) + *Azospirillum* (20 g/kg seeds)], were statistically at par with treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)].

Economic Analysis

Gross Returns

Highest gross return (152544.4 INR/ha) [Table-3] was obtained in treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)] as compared to other treatments.

Formatted: Indent: First line: 0"

Net Returns

Net returns (108712.4 INR /ha) [Table-3] was found to be highest in treatment-9 [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)] as compared to other treatments.

Formatted: Indent: First line: 0"

Benefit Cost Ratio

Benefit Cost ratio (2.48) [Table-3] was found to be highest in treatment-9 with [Zinc (10 kg/ha) + *Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)] as compared to other treatments.

Formatted: Indent: First line: 0"

The statistically Maximum Gross returns (Rs. 1,00,500 INR/ha), Net returns (Rs. 72,002.50 INR/ha) and B:C ratio (2.52) was obtained with application of *Azospirillum* + 70 kg/ha Phosphorus. These results are supported by the findings of (Reddy and Singh *et al*, 2021) in wheat. Further, Applyingapplying Zn up to 12 kg Zn/ha increased the net return, however value cost ratio (VCR) was decreased by increasing Zn doses and this may be due to the fact that all applied Zn was not taken up by wheat crop, so residual effect of Zn can affect the yield next year. Soil application of Zn was economical and had long-term effects on enhanced wheat grain grown on Zn deficient soils. These results are supported by the findings of (Abbas *et al.* (2009) and (Yilmaz *et al.* (1997) in wheat.

Formatted: Indent: First line: 0"

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold, Italic

Formatted: Font: Not Bold

UNDER PEER REVIEW

Table-1: Influence of zinc and bio fertilizers on growth parameters of wheat at 100 days after sowing.

S.No.	Treatments	Plant height (cm)	Plant Dry weight (g/plant)
1.	Zinc 5 kg/ha + Azotobacter at 20 g/kg seeds	93.17	17.69
2.	Zinc 5 kg/ha + Azospirillum at 20 g/kg seeds	93.37	17.72
3.	Zinc 5 kg/ha + Azotobacter at 10 g/kg seeds + Azospirillum at 10 g/kg seeds	94.07	18.24
4.	Zinc 7.5 kg/ha + Azotobacter at 20 g/kg seeds	94.62	18.42
5.	Zinc 7.5 kg/ha + Azospirillum at 20 g/kg seeds	94.96	18.91
6.	Zinc 7.5 kg/ha + Azotobacter at 10 g/kg seeds + Azospirillum at 10 g/kg seeds	95.13	19.07
7.	Zinc 10 kg/ha + Azotobacter at 20 g/kg seeds	95.76	19.44
8.	Zinc 10 kg/ha + Azospirillum at 20 g/kg seeds	95.92	19.68
9.	Zinc 10 kg/ha + Azotobacter at 10 g/kg seeds + Azospirillum at 10 g/kg seeds	96.10	20.31
	F-test	S	S

Formatted: Space After: 0 pt, Line spacing: single

SEd (±)	0.14	0.23
CD (P=0.05)	0.43	0.68

S/n. (cm)	Treatments Weight (g/plant)	Plant height	Plant Dry
1.	Zinc 5 kg/ha + Azotobacter at 20 g/kg seeds	93.17	17.69
2.	Zinc 5 kg/ha + Azospirillum at 20 g/kg seeds	93.37	17.72
3.	Zinc 5 kg/ha + Azotobacter at 10 g/kg seeds + Azospirillum at 10 g/kg seeds	94.07	18.24
4.	Zinc 7.5 kg/ha + Azotobacter at 20 g/kg seeds	94.62	18.42
5.	Zinc 7.5 kg/ha + Azospirillum at 20 g/kg seeds	94.96	18.91
6.	Zinc 7.5 kg/ha + Azotobacter at 10 g/kg seeds + Azospirillum at 10 g/kg seeds	95.13	19.07
7.	Zinc 10 kg/ha + Azotobacter at 20 g/kg seeds	95.76	19.44
8.	Zinc 10 kg/ha + Azospirillum at 20 g/kg seeds	95.92	19.68
9.	Zinc 10 kg/ha + Azotobacter at 10 g/kg seeds + Azospirillum at 10 g/kg seeds	96.10	20.31
	F test	S	S

		SEd (±)				
		0.14	0.23			
S. No.	Treatments	Number of spikes/m ²	Number of Grains/Spike	Grain yield (t/ha)	Straw yield (t/ha)	Harvest index (%)

		CD (P=0.05)				
		0.43	0.68			

Table-2: Influence of zinc and bio fertilizers on yield attributes of wheat

1.	Zinc 5 kg/ha + <i>Azotobacter</i> at 20 g/kg seeds	324.70	43.03	4.257	10.36	29.12
2.	Zinc 5 kg/ha + <i>Azospirillum</i> at 20 g/kg seeds	332.81	43.48	4.433	10.58	29.52
3.	Zinc 5 kg/ha + <i>Azotobacter</i> at 10 g/kg seeds + <i>Azospirillum</i> at 10 g/kg seeds	343.40	45.06	4.640	10.83	30.00
4.	Zinc 7.5 kg/ha + <i>Azotobacter</i> at 20 g/kg seeds	340.50	47.17	4.957	11.28	30.53
5.	Zinc 7.5 kg/ha + <i>Azospirillum</i> at 20 g/kg seeds	346.15	49.48	5.143	11.44	31.02
6.	Zinc 7.5 kg/ha + <i>Azotobacter</i> at 10 g/kg seeds + <i>Azospirillum</i> at 10 g/kg seeds	348.25	50.58	5.280	11.72	31.06
7.	Zinc 10 kg/ha + <i>Azotobacter</i> at 20 g/kg seeds	355.58	52.25	5.417	12.23	30.69
8.	Zinc 10 kg/ha + <i>Azospirillum</i> at 20 g/kg seeds	364.73	54.73	5.570	12.44	30.93
9.	Zinc 10 kg/ha + <i>Azotobacter</i> at 10 g/kg seeds + <i>Azospirillum</i> at 10 g/kg seeds	371.84	55.17	5.697	12.59	31.14
	F-Test	§	§	§	§	§
	SEm±	5.35	0.37	0.06	0.07	0.24
	CD (P=0.05)	16.03	1.12	0.18	0.22	0.71

S/n.	Treatments	No. of spikes/ m ²	No. of Grains/Spike	Grain yield (t/ha)	Straw yield (t/ha)	Harvest index (%)
1.	<u>Zinc 5 kg/ha + Azotobacter</u>					
	at 20 g/kg seeds	324.70	43.03	4.257	10.36	29.12
2.	<u>Zinc 5 kg/ha + Azospirillum</u>					
	at 20 g/kg seeds	332.81	43.48	4.433	10.58	29.52
3.	<u>Zinc 5 kg/ha + Azotobacter at</u>					
	<u>10 g/kg seeds+Azospirillum</u>					
	at 10 g/kg seeds	343.40	45.06	4.640	10.83	30.00
4.	<u>Zinc 7.5 kg/ha + Azotobacter</u>					
	at 20 g/kg seeds	340.50	47.17	4.957	11.28	30.53
5.	<u>Zinc 7.5 kg/ha+Azospirillum</u>					
	at 20 g/kg seeds	346.15	49.48	5.143	11.44	31.02
6.	<u>Zinc 7.5 kg/ha+Azotobacter at</u>					
	<u>10 g/kg seeds +Azospirillum</u>					
	at 10 g/kg seeds	348.25	50.58	5.280	11.72	31.06
7.	<u>Zinc 10 kg/ha+Azotobacter</u>					
	at 20 g/kg seeds	355.58	52.25	5.417	12.23	30.69

S.No.	Treatments	Cost of cultivation (INR/ha)	Gross return (INR/ha)	Net return (INR/ha)	B:C Ratio
8.	Zinc 10 kg/ha + Azospirillum				
	at 20 g/kg seeds	364.73	54.73	5.570	12.44 30.93
9.	Zinc 10 kg/ha + Azotobacter				
	at 10 g/kg seeds+Azospirillum				
	at 10 g/kg seeds	371.84	55.17	5.697	12.59 31.14
	F-Test	S	S	S	S
	SEm+	5.35	0.37	0.06	0.07 0.24
	CD (P=0.05)	16.03	1.12	0.18	0.22 0.71

Table-3: Influence of zinc and bio fertilizers on economics of wheat.

Formatted: Font: 10.5 pt, Not Bold

Formatted: Left, Space After: 0 pt, Line spacing: single

1.	Zinc 5 kg/ha + <i>Azotobacter</i> at 20 g/kg seeds	42968	116847.4	73879.4	1.719405
2.	Zinc 5 kg/ha + <i>Azospirillum</i> at 20 g/kg seeds	42488	121074.0	78586.0	1.849603
3.	Zinc 5 kg/ha + <i>Azotobacter</i> at 10 g/kg seeds + <i>Azospirillum</i> at 10 g/kg seeds	42728	125974.0	83246.0	1.948277
4.	Zinc 7.5 kg/ha + <i>Azotobacter</i> at 20 g/kg seeds	43528	133703.4	90175.4	2.071664
5.	Zinc 7.5 kg/ha + <i>Azospirillum</i> at 20 g/kg seeds	43048	137939.5	94891.5	2.204317
6.	Zinc 7.5 kg/ha + <i>Azotobacter</i> at 10 g/kg seeds + <i>Azospirillum</i> at 10 g/kg seeds	43288	141552.0	98264.0	2.270006
7.	Zinc 10 kg/ha + <i>Azotobacter</i> at 20 g/kg seeds	44072	145822.4	101750.4	2.308731
8.	Zinc 10 kg/ha + <i>Azospirillum</i> at 20 g/kg seeds	43592	149543.5	105951.5	2.430526
9.	Zinc 10 kg/ha + <i>Azotobacter</i> at 10 g/kg seeds + <i>Azospirillum</i> at 10 g/kg seeds	43832	152544.4	108712.4	2.480206

S/n.	Treatments	Cost of cultivation (INR/ha)	Gross return (INR/ha)	Net Return (INR/ha)	Grain B:C Ratio
1.	Zinc 5 kg/ha + Azotobacter				
	at 20 g/kg seeds	42968	116847.4	73879.4	1.719405
2.	Zinc 5 kg/ha + Azospirillum				
	at 20 g/kg seeds	42488	121074.0	78586.0	1.849603
3.	Zinc 5 kg/ha + Azotobacter at 10 g/kg seeds+Azospirillum				
	at 10 g/kg seeds	42728	125974.0	83246.0	1.948277
4.	Zinc 7.5 kg/ha + Azotobacter				
	at 20 g/kg seeds	43528	133703.4	90175.4	2.071664
5.	Zinc 7.5 kg/ha+Azospirillum				
	at 20 g/kg seeds	43048	137939.5	94891.5	2.204317
6.	Zinc 7.5 kg/ha+Azotobacter at 10 g/kg seeds +Azospirillum				
	at 10 g/kg seeds	43288	141552.0	98264.0	2.270006
7.	Zinc 10 kg/ha+Azotobacter				
	at 20 g/kg seeds	44072	145822.4	101750.4	2.308731
8.	Zinc 10 kg/ha + Azospirillum				
	at 20 g/kg seeds	43592	149543.5	105951.5	2.430526
9.	Zinc 10 kg/ha + Azotobacter at 10 g/kg seeds+Azospirillum				
	at 10 g/kg seeds	43832	152544.4	108712.4	2.48020

CONCLUSION

From the experimental findings, it is concluded that with the application of Zinc (10 kg/ha) along with Bio-fertilizers [*Azotobacter* (10 g/kg seeds) + *Azospirillum* (10 g/kg seeds)] performs positively and improves the growth parameters, yield attributes and economics of wheat. These findings are based on one season therefore, further trails may be required for further confirmation.

Formatted: Indent: First line: 0"

Formatted: Font: 14 pt

Formatted: Left

UNDER PEER REVIEW

REFERENCES

Abbas, G., Muhammad, O. K., Jamil, M., Tahir, M. and Hussain, F. (2009). Nutrient uptake, growth and yield of wheat (*Triticum aestivum*) as affected by zinc application rates. *International Journal of Agriculture & Biology*, 11: 389-396.

Formatted: Width: 8.5", Height: 11", Header distance from edge: 0.5", Footer distance from edge: 0.5"

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Ahemad, M., Kibret, M. (2014). Mechanisms and applications of plant growth promoting rhizobacteria: current perspective. *Journal of King Saud Univ Sci University Science*, 26:1-20.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Alloway, B. J. (2008). Zinc in Soils and Crop Nutrition, 2nd edition. IZA Brussels, Belgium and IFA Paris, France.

Formatted: Font: Not Bold

Arif, M., Tasneem, M., Bashir, F., Yaseen, G., Anwar, A. (2017). Evaluation of Different Levels of Potassium and Zinc Fertilizer on the Growth and Yield of Wheat. *International Journal of Biosen Bioelectron* 3(2): 00057.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Arshad, Muhammad. & Adnan, Muhammad. & Ahmed, Sher. & Khan, Abdul. & Ali, Irshad. & Ali, Muhammad. & Ali, Azaz. & Khan, Azam. & Kamal, Muhammad. & Gul, Farhana. & Khan, Muhammad. (2016). Integrated Effect of Phosphorus and Zinc on Wheat Crop. *American-Eurasian J. Agric. & Environ. Sci.*, 16. 455-459.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Cakmak, I. and Kutman, U.B. (2018). Agronomic biofortification of cereals with zinc: a review. *Eur J Soil Sci*, 69: 172-180.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Curtin, D., Martin, R.J., and Scott, C.L. (2008). Wheat (*Triticum aestivum*) response to micronutrients (Mn, Cu, Zn, B) in Canterbury, New Zealand. *New Zealand Journal of Crop and Horticulture Sciences*, 36: 169-181.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

GOI, 2021-22. Fourth Advance Estimates for Production of Major Crops 2021-22, Ministry of Agriculture & Farmers Welfare, pib.gov.in.

Formatted: Font: Not Bold

Gomez, K.A. and Gomez, A.A. (1984). Statistical procedures for agricultural research (2 ed.). John Wiley and Sons, New York, 680p.

Formatted: Font: Not Bold

Jan, A., M. Wasim and Jr. Amanullah. (2013). Interactive effects of zinc and nitrogen application on wheat growth and grain yield. *Journal of Plant Nutrition*, 36: 1506-1520.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Kachroo, D. and Razdan, R. (2006). Growth, nutrient uptake and yield of wheat (*Triticum aestivum*) as influenced by biofertilizers nitrogen. *Indian Journal of Agronomy* 51(1): 37-9.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Kandil, A. A., M. H. El-Hindi, M. A. H. Badawi, S. M. A. El-Morarsy, S. A. and Kalboush, F. A. H. M. Kalboush, (2011). Response of wheat to rates of nitrogen, biofertilizers and land leveling. *Crop & Environment*, 2(1): 46-51.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Khan, M. & Fuller, Michael M. & Baluch, F. (2008). Effect of Soil Applied Zinc Sulphate on Wheat (*Triticum aestivum* L.) Grown on a Calcareous Soil in Pakistan. *Cereal Research Communications* 36. 571-582. 10.1556/CRC.36.2008.4.6.

Kizilkaya, R. (2008). Yield response and nitrogen concentrations of spring wheat (*Triticum aestivum*) inoculated with *Azotobacter chroococcum* strains. *Ecological Engineering*. Vol. 33(2): 150- 156.

Lal, B S., Rajkumar, Y. & Thomas, A. (2015). Studies on the effect of Zinc levels, and methods of boron application on growth, yield and protein content of Wheat (*Triticum aestivum* L.). *Bull. Env. Pharmacol. Life Sci.*, Vol 4 [2] January 2015: 108- 113.

Formatted: Font: Not Bold

Namvar, **Ali A.** & Khandan, **Teymur T.** (2013). Response of wheat to mineral nitrogen fertilizer and biofertilizer (*Azotobacter* sp. and *Azospirillum* sp.) inoculation under different levels of weed interference. *Ekologija*. 59. 10.6001/ekologija.v59i2.2711.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Piccinin, G. G., Braccini, A.L., Dan, L.G.M., Scapim, C.A., Ricci, T.T. and Bazo, G.L. (2013). Efficiency of seed inoculation with *Azospirillum brasilense* on agronomic characteristics and yield of wheat. *Industrial Crops and Products*, 43: 393–397.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Reddy **Vinay Kumar Reddy, V. K. R. and Shikha Singh, S.** (2021). Response of biofertilizers and phosphorus levels on growth and yield of wheat (*Triticum aestivum* L.). *Pharma Innovation*. 10(11):293-296.

Formatted: Font: 11 pt, Not Bold

Formatted: Font: 11 pt, Not Bold

Formatted: Font: 11 pt, Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Rehman, A., Khan, **Aza Z.**, Muhammad, **A. T.** and Jalal, A. (2017). Performance of nitrogen fixing bacteria with increasing nitrogen ratios on growth, maturity and biomass of winter wheat varieties. *Biosci Res*. 14:114–121.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Seadh, S.E. et al., Seadh, S.E., EL-Abady, M.I., El -Ghamry, A.M. and Farouk, S. (2009). Influence of micronutrients foliar application and nitrogen fertilization on wheat yield and quality of grain and seed. *Journal of Biological Sciences*, v.9, n.8, p.851-858.

Formatted: Font: 11 pt, Not Bold

Formatted: Font: Not Bold

Formatted: Font: 11 pt

Sendhil, **R. &** Kiran, T.M. & Singh, G. (2019). Wheat Production in India: Trends and Prospects. 10.5772/intechopen.86341.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Sharma, S. K., Kapoor, S., Rana S.S. and Sankhyan, N. (2016). Effect of nitrogen, zinc and boron on growth, yield attributes and yield of wheat under mid hill conditions of Himachal Pradesh. *Himachal Journal of Agricultural Research* 42(1): 99-103.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Singh, **RKR.K.**, Agarwal, **RL.R.L.** and Singh, **SKS.K.** (2007). Integrated nutrient management in wheat (*Triticum aestivum*). *Annals of Agricultural Research*. 28(1):20-24.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Singh, S.S. (2000). Crop management under irrigated and rainfed condition. Kalyani publishers PP 51.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Singh, **Vinay & V.**, Singh, S.P. & Singh, **Sandeep S.** & Shivay, **Yashbir Y.** (2013). Growth, yield and nutrient uptake by wheat (*Triticum aestivum*) as affected by biofertilizers, FYM and nitrogen. *Indian Journal of Agricultural Sciences*. 83. 331-334.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Vessey, J. K. (2003). Plant growth promoting rhizo-bacteria as biofertilizers. *Journal of Plant and Soil*. Vol. 255: 571-586.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Italic

Wani, **SPS.P.**, Chandrapalaiha, S., Zambre, **MA.M.A.** and Lee, **KKK.K.** (1988). Association between nitrogen fixation bacteria and pearl millet plants, responses mechanism and resistance. *Journal of Plant and Soil*, 110:284-302.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Italic

Warembourg, **FRF.R.**, Dressen, R., Vlassak, **K.** and Lafont, F. (1987). Peculiar effect of *Azospirillum* inoculation on growth and nitrogen balance of winter wheat (*Triticum aestivum*). *Biol. Fertil. Journal of Biology and Fertility of Soils*, 4:55-59.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Yilmaz, A., Ekiz, H., Torun, B., Gultekin, I., Karanlik, S., Bagci, S.A. & Cakmak, I. (1997). Effect of different zinc application methods on grain yield and zinc concentration in wheat cultivars grown on zinc-deficient calcareous soils, *Journal of Plant Nutrition*, 20:4-5, 461-471.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Zorita, M. D. and Canigia, V.F. (2009). Field performance of a liquid formulation of *Azospirillum brasilense* on dryland wheat productivity. *European Journal of Soil Biology*, 45(1): 3-11.

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Font: Not Bold

Formatted: Indent: Left: 0", Hanging: 0.59"

Zoz, **Tiago & T.**, Steiner, **Fábio & F.**, Fey, **Rubens & R.**, Castagnara, **Deise & D.** and Seidel, **Edlousa E.** (2012). Response of wheat to foliar application of zinc. *Ciência Rural*. 42. 784-787. 10.1590/S0103-84782012005000015.