

Soil Properties of Barley (*Hordeum vulgare* L.) Crop as Affected by Zinc-Based Fertilizers in Saline Conditions

ABSTRACT

The field experiment conducted during the rabi season of 2021-22 at the Agricultural Research Sub-station in Vallabh Nagar, Udaipur, Rajasthan, aimed to assess the influence of zinc-based fertilizers on soil properties including chemical and biological properties after harvest of barley (*Hordeum vulgare* L.) crop. The experimental design followed a split plot arrangement, with main plot treatments consisting of a control, 5 kg Zn per hectare as soil application, and seed treatment with zinc solubilizing bacteria (ZSB) at a rate of 5 ml per kg of seed. The sub plot treatments included a control and three foliar sprays of nano Zn at 5 ml per litre of water, applied at 15, 30, and 45 days after sowing. Each treatment was replicated three times. The soil application of zinc @ 5 kg Zn ha⁻¹ along with foliar spray of nano Zn @ 5 ml per litre of water at 45 days after sowing had significantly influenced availability of macronutrients (N and K), micronutrients (Zn, Fe, Mn and Cu) as well as microbial population (bacteria, fungi and actinomycetes) in post-harvest soil of barley crop over control. The combined application of conventional zinc fertilizer and foliar spray of nano zinc offers a promising strategy to improve nutrient availability and enhance soil microbial population in saline soil after the harvest of barley crop.

Keywords: Foliar application, Nano Zinc, Barley, Microbial population, Saline Soil

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1. Introduction

Soil is an immensely valuable and delicate resource that holds significant importance for any nation. It plays a pivotal role in providing vital ecological services necessary for nourishing and sustaining life. Thus, it is absolutely crucial to prioritize the maintenance of soil health to ensure the long-term sustainability of ecosystems (Lal, 2014). Among the factors that influence soil health, the physiochemical properties and microbial community of the soil hold particular significance. They serve as essential indicators and early warning signs of the overall state of soil health, owing to their remarkable responsiveness and

sensitivity to environmental changes. The physiochemical properties of the soil encompass various physical and chemical characteristics such as texture, structure, nutrient content, pH levels, and water-holding capacity. These properties directly influence the soil's ability to retain and supply essential nutrients, regulate water flow, and provide a supportive environment for plant growth. Any alterations in these properties can have far-reaching consequences on the overall health and productivity of the soil (Roldán *et al.*, 2015). Equally important are the soil microbes, which comprise an extensive array of microscopic organisms such as bacteria, fungi, archaea, and protozoa. These organisms inhabit the soil in vast numbers and perform crucial functions that contribute to soil fertility and ecosystem stability. Soil microbes are involved in processes like nutrient cycling, organic matter decomposition, soil structure formation, disease suppression, and symbiotic relationships with plants. As a result, their presence, diversity, and activity serve as reliable indicators of the overall soil health (Bardgett and van der Putten, 2014). Due to their rapid response to changes in environmental conditions, soil microbes act as valuable sentinels, providing early warnings of any disturbances or imbalances within the soil ecosystem. Their sensitivity enables them to reflect the impact of various factors such as land management practices, pollution, climate change, and the introduction of invasive species. By monitoring the abundance and diversity of soil microbes, scientists and land managers can gain valuable insights into the condition of the soil, allowing for timely interventions and sustainable soil management strategies (Philippot *et al.*, 2013).

Barley (*Hordeum vulgare* L.) holds a distinguished status as an ancient cereal grain that has undergone domestication, transforming from primarily a food grain into a valuable feed and malting grain. As an herb cultivated for centuries, barley predates wheat and is believed to be the oldest of all cultivated plants. Its cultivation has been documented in ancient civilizations across the world. In northern India, barley assumes importance as a **rabi** cereal crop. Globally, barley ranks among the most significant cereal grains, following rice, wheat, and maize. In India, it is cultivated during the summer in temperate regions and during the winter in tropical regions. Notably, barley exhibits a short growing season and demonstrates remarkable drought tolerance. While it was previously mainly used as livestock feed, barley has now emerged as a staple grain in human consumption. Additionally, barley plays a pivotal role as a rabi crop in various regions. It is also cultivated specifically for malting and brewing purposes in Haryana, Western Uttar Pradesh, Punjab, and Rajasthan, where careful management practices are employed to ensure high grain quality. Barley holds a relatively small share of agricultural statistics in India. It occupies approximately 0.46% of

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the total cropped area, 0.62% of the food grains, and 0.76% of the cereals in the country. Despite its modest acreage, barley contributes significantly to the total cereal production, accounting for 0.86% of the overall cereals produced and 0.81% of the food grains in India. Over the years, the area dedicated to coarse cereal crops has decreased from 37.67 million hectares in 1950-51 to 24.15 million hectares in 2019-20. However, barley production has witnessed a substantial increase from 15.38 million tonnes to 41.75 million tonnes during the same period (GoI, 2021). Furthermore, the yield of barley has seen improvement, rising from 1938 kg/ha in 2005-06 to 2881 kg/ha in 2019-20 (DAC & FW, 2020). In the state of Rajasthan, specifically during the 2020-21 period, barley was cultivated on approximately 2.69 lakh hectares, resulting in a production of 9.35 lakh tons and a productivity of 3469 kg/ha. Within the Udaipur region, barley was cultivated on 11.7 thousand hectares, yielding a production of 30.7 thousand tons and a productivity of 2584 kg/ha (Rajasthan Agriculture Statistics at a Glance, 2020-21).

Zinc is acknowledged as the fourth most critical nutrient that restricts crop yield, trailing behind nitrogen, phosphorus, and potassium, both on a global scale and in Indian soils (Singh *et al.*, 2020). Additionally, it is estimated that around 36.5% of Indian soils are deficient in zinc, emphasizing the widespread prevalence of this nutrient deficiency in the country (Dixit *et al.*, 2021). Zinc assumes critical importance as a nutrient for barley, particularly in the context of saline soils. Salinity poses significant challenges to nutrient uptake by plants, leading to imbalances and deficiencies. Zinc plays a pivotal role in various physiological processes of barley, making it essential for its growth and development in saline conditions. Firstly, zinc is necessary for the activation of numerous enzymes involved in key metabolic pathways, including carbohydrate metabolism, protein synthesis, and DNA synthesis (Gupta and Sharma, 2002). By facilitating these enzymatic reactions, zinc ensures the proper functioning of vital physiological processes in barley even under saline stress. Moreover, zinc is instrumental in regulating plant hormones, such as auxins and cytokinins, which are responsible for growth, development, and stress responses. Adequate zinc levels help maintain hormone balance in barley, enabling it to better adapt and tolerate the adverse effects of salinity (Zahedi *et al.*, 2019). Additionally, zinc plays a crucial role in the antioxidant defense system of barley. Saline soils induce oxidative stress in plants through the accumulation of reactive oxygen species (ROS). Zinc acts as a cofactor for several antioxidant enzymes, including superoxide dismutase (SOD), catalase (CAT), and peroxidase (POD), which protect barley plants by detoxifying ROS and minimizing oxidative damage caused by salinity stress (Balakhnina and Borkowska, 2013). Furthermore, zinc is involved in

maintaining ion homeostasis within barley plants. Saline soils disrupt the balance of ions, leading to nutritional imbalances. Adequate zinc levels help in the regulation of ion transport and uptake, ensuring proper nutrient absorption and utilization by barley in saline conditions (Ehsanzadehet *et al.*, 2013).

Saline soils pose significant challenges to agricultural productivity in various regions, including the arid and semi-arid areas of Vallabhagar, Udaipur. These soils are characterized by high concentrations of soluble salts, which negatively impact soil properties and constrain the growth and development of crops. To address these challenges, various strategies have been explored, including the use of zinc-based fertilizers. Zinc, an essential micronutrient for plant growth, has shown promise in mitigating the adverse effects of salinity on crops. Therefore, this study aims to assess the soil properties after harvest of barley crops in saline conditions at the Agriculture Research Sub Station in Vallabhagar, Udaipur.

2. MATERIALS AND METHODS

2.1 Description of the Study Site

The experiment took place at the Agriculture Research Sub Station Farm located in Vallabhagar, Udaipur (Rajasthan). The geographical coordinates of the farm are approximately 24° 38' North latitude and 73° 42' East longitude. Situated at an average altitude of 633 meters above sea level, it is in close proximity, only 45 km east of Udaipur. The experimental site falls within Rajasthan's agro-climatic zone IVa, characterized as Sub-Humid Southern Plain and Aravalli Hills.

To examine the physico-chemical properties of the soil, samples were collected randomly from a depth of up to 15 cm prior to the start of the experiment. A composite sample was created and subjected to analysis to determine the soil's physico-chemical characteristics. The results, including the analysis methods employed, are presented in Table 1. The data reveals that the soil in the experimental field exhibited a clay loam texture and had an alkaline pH of 8.79. The soil was found to have a moderate amount of available nitrogen ($272.24 \text{ kg ha}^{-1}$) and available phosphorus (26.19 kg ha^{-1}), while the available potassium content was high ($273.12 \text{ kg ha}^{-1}$). However, the available zinc content was low, measuring only 0.49 ppm.

2.2 Experimental design and treatments

To ensure comprehensive analysis of the study variables, a split-plot design was employed in the experiment. The design consisted of three main plot treatments that involved different zinc sources, and four sub-plot treatments that involved the application of nano zinc

at various time intervals. This design was replicated three times to enhance the reliability of the results and minimize potential variations. The split-plot design facilitated the simultaneous evaluation of the effects of both the main plot treatments (zinc sources) and the sub-plot treatments (nano zinc application timing) on the study variables, allowing for a thorough examination of their individual and combined impacts.

List 1 :Details oftreatments and their symbols

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Treatments	Symbols
Main Plot (Soil application and seed treatment)	
i. Control	Zn ₀
ii. 5 kg Zn ha ⁻¹ (soil application)	Zn _{SA}
iii. Zinc solubilizing bacteria @ 5 ml kg ⁻¹ of seed (Seed treatment)	Zn _{ST}
Sub Plot (Foliar application)	
i. Control	NP ₀
ii. Foliar spray of nano Zn @ 15 DAS	NP ₁₅
iii. Foliar spray of nano Zn @ 30 DAS	NP ₃₀
iv. Foliar spray of nano Zn @ 45 DAS	NP ₄₅

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2.3 Application protocol of fertilizers

In the experimental setup, the recommended doses of nitrogen (N) and phosphorus (P) were 60 kg ha⁻¹ and 20 kg ha⁻¹, respectively. To provide these nutrients, urea was used for nitrogen application, while diammonium phosphate (DAP) was utilized for phosphorus application. Zinc was applied in the form of zinc sulphate, with the quantity varying based on the specific treatment. During sowing, the total amount of phosphorus and zinc, along with half of the nitrogen, were applied by placing them in furrows. This allowed for direct placement of the nutrients in the root zone of the crops. The remaining half of the nitrogen was divided into two equal splits and applied during subsequent irrigations. This approach ensured that the nitrogen supply was distributed evenly over the crop's growth stages. Furthermore, a foliar application of nano zinc was carried out following the treatment requirements. Nano zinc was sprayed on the plants using a concentration of 5 ml per liter of water.

2.4 Soil Chemical Properties

To evaluate the fertility status of the soil, soil samples were collected from each plot at crop harvest, specifically from a depth of 0-15 cm. The collected soil samples were then passed through a 2 mm plastic sieve to eliminate any metallic contamination.

The soil samples were subjected to analysis to determine the availability of key nutrients such as nitrogen (N), phosphorus (P), potassium (K), as well as micronutrients like zinc (Zn), iron (Fe), manganese (Mn), and copper (Cu). The following methods were employed for the analysis are given below

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Chemical determinations

(a)	Available nitrogen	By alkaline permanganate method	Subbiah and Asija (1956)
(b)	Available Phosphorus	Extraction of soil with 0.5 M NaHCO ₃ at pH 8.5 and development of blue colour with SnCl ₂ and measurement through colorimetrically	Olsen <i>et al.</i> (1954)
(c)	Available potassium	Extraction was done with 1 N neutral ammonium acetate at pH 7.0 and determined by flame photometer	Richards (1954)
(d)	Available Zn, Fe, Mn and Cu	Analysis of suitable aliquot of DTPA extract with the help of atomic absorption spectrophotometer (Varian techtron AAS-120)	Lindsay and Norvell (1978)

2.5 Soil microbial properties

The enumeration of soil microbial populations including bacteria, fungi, and actinomycetes was conducted using the standard serial dilution and plate count method, as described by Vance *et al.* (1987). To determine the alkaline phosphatase activity, a spectrophotometric analysis was performed using B-nitrophenol phosphate in buffers with pH 5.4 and 9.4. This method was originally outlined by Tabatabai and Bremner (1969). Dehydrogenase activity was assessed by employing a colorimetric determination of TPF (triphenylformazon). The method used for this analysis was initially described by Casida *et al.* (1964).

2.6 Statistical Analysis

The experimental data were subjected to statistical analysis using the analysis of variance (ANOVA) procedure as outlined by Panse and Sukhatme (1985). The "F" test was utilized to interpret the results and determine the significance of the observed differences among the treatment groups. To compare the means of different treatment groups, the critical difference (CD) was calculated at a significance level of 5%.

3. Result and Discussion

3.1 Chemical Properties

3.1.1 Effect of soil application and seed treatment with zinc on chemical properties of soil after harvest of barley crop

The application of zinc in the soil and the seed treatment with zinc solubilizing bacteria had a significant effect on the availability of nitrogen (N), potassium (K), zinc (Zn), iron (Fe), manganese (Mn), and copper (Cu) in the soil. In comparison to the control group, the levels of these nutrients increased significantly. However, the available phosphorus (P) in the soil after the crop harvest was not significantly affected by the treatments. Among the different treatments, the highest levels of available N ($291.46 \text{ kg ha}^{-1}$), K ($294.99 \text{ kg ha}^{-1}$), Zn (0.64 mg kg^{-1}), Fe (6.25 mg kg^{-1}), Mn (5.77 mg kg^{-1}), and Cu (1.82 mg kg^{-1}) were observed when zinc was applied to the soil at a rate of 5 kg Zn ha^{-1} (Z_{NSA}). The decrease in available phosphorus (P) following the harvest of the barley crop, attributed to increased zinc application, can be explained by the antagonistic relationship between these two nutrients. It is known that high levels of zinc can interfere with phosphorus availability in the soil. This antagonistic interaction may have led to a reduction in the availability of phosphorus (Alloway, 2008). On the other hand, the increase in available nitrogen, potassium, and zinc after the harvest of the barley crop can be attributed to the synergistic relationship of zinc with these nutrients (Sharma *et al.*, 2012). Zinc has been shown to exhibit positive

interactions with nitrogen, potassium, and zinc, potentially enhancing their availability and uptake by plants. This synergistic relationship may have contributed to the observed increase in the availability of these nutrients in the soil (Kumar *et al.*, 2018). Furthermore, the higher content of DTPA-Zn in the soil after the barley crop harvest can be attributed to the increased solubility, diffusion, and mobility of the applied zinc. This enhanced solubility of zinc, combined with its improved movement in the soil, can lead to higher levels of DTPA-Zn. This finding aligns with previous research by Chatterjee *et al.* (1983). Saline soil typically contains high levels of salt, which can hinder nutrient availability and plant uptake. The application of zinc fertilizer can help alleviate the negative effects of salinity on nutrient availability. Soil application of zinc fertilizer **enhance** the soil's nutrient-holding capacity, allowing for better retention and availability of essential nutrients (Siddique *et al.*, 2017). This is particularly important in saline soil, where salt content can disrupt nutrient availability and increase leaching. Zinc fertilizer can improve soil structure and drainage, reducing the impact of salt accumulation and facilitating the movement of nutrients within the soil. This enhances the ability of plants to access and uptake the available N P K nutrients (Gupta and Gupta, 2016). Additionally, zinc fertilizer can promote root development and function, enabling plants to better extract nutrients from the soil, even in saline conditions. This enhanced root activity improves nutrient uptake efficiency and helps plants overcome the negative effects of salinity (Sharma *et al.*, 2017). Furthermore, zinc is an essential micronutrient for plant growth and plays a crucial role in various metabolic processes. By supplying plants with adequate zinc through fertilizer application, it can support their physiological functions, including nutrient uptake and utilization (Saikia *et al.*, 2018).

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3.1.2 Effect of foliar application of nano zinc on chemical Properties of soil after harvest of barley crop

The results of the **study** revealed a significant increase in the availability of nitrogen (N), potassium (K), zinc (Zn), iron (Fe), manganese (Mn), and copper (Cu) with the foliar application of nano-zinc compared to the control group. However, the availability of phosphorus (P) in the soil after the crop harvest remained unaffected by the treatment. Among the various foliar spray timings, the maximum levels of available N ($285.95 \text{ Kg Ha}^{-1}$), K ($289.11 \text{ Kg Ha}^{-1}$), Zn (0.63 mg kg^{-1}), Fe (6.12 mg kg^{-1}), Mn (5.67 mg kg^{-1}), and Cu (1.76 mg kg^{-1}) were observed when nano-zinc was applied as a foliar spray at 45 days after sowing (NP₄₅). The foliar spray of ZnO nanoparticles (NPs) had a positive impact on the available nitrogen (N) content in the soil, which is consistent with the findings of Sabaghet *et al.* (2020) who observed an increase in N-content in rice varieties due to foliar application of zinc. This

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improvement in N-content may be attributed to the residual fertilizer in the soil and the higher nutrient-holding capacity of nano fertilizers compared to conventional ones. However, the available phosphorus (P) content in the soil decreased, possibly due to the interaction between P and Zn in the soil, resulting in reduced translocation of P from roots to shoots and an imbalanced P:Zn ratio in the plant, as mentioned by El-Nagaret *et al.* (2020). Sabaghet *et al.* (2020) also reported a relationship between Zn application and the total potassium (K) percentage in the soil, demonstrating its impact on the K content. Similarly, the treatment with ZnO NPs affected the soil micronutrient contents, with an increase in soil zinc content observed through foliar application of zinc compared to the control, as supported by Ghoneim (2016), Rajoneet *et al.* (2016), and Jassimet *et al.* (2019).

3.2 Biological Properties

3.2.1 Effect of soil application and seed treatment with zinc on biological properties of soil after harvest of barley crop

The micro biological population (bacteria, fungi and actinomycetes) were significantly increased in the soil after the harvest of barley crop with application of zinc in the soil and the seed treatment with zinc solubilizing bacteria (Table 4). The significantly highest bacteria (39.84×10^7 cfu g⁻¹ of soil), fungi (23.97×10^5 cfu g⁻¹ of soil), actinomycetes (33.85×10^6 cfu g⁻¹ of soil) population in soil was recorded under soil application of zinc @ 5 kg Zn ha⁻¹ (Zn_{SA}). The application of zinc to saline soil after the harvest of barley crop has been found to have positive effects on the microbial population. Zinc application improves nutrient availability, as zinc is an essential micronutrient for microbial growth and metabolism. By supplying an adequate amount of zinc, it helps alleviate zinc deficiency in microorganisms, promoting their growth and activity (Islam *et al.*, 2016). Additionally, zinc acts as a cofactor for many enzymes involved in metabolic processes, enhancing enzymatic activity. This, in turn, supports microbial nutrient acquisition and utilization (Sabaghet *et al.*, 2020). Moreover, zinc application has been found to enhance microbial stress tolerance in saline soil. Saline conditions impose stress on microbial communities due to high salt concentrations, negatively impacting their growth and activity. However, zinc application can alleviate the negative effects of salinity stress, enhancing microbial survival and activity in saline conditions (Eftekhariet *et al.*, 2019). Furthermore, the application of zinc to the soil improves overall soil health. It enhances soil structure, promoting better soil aggregation, water-holding capacity, and nutrient cycling. These improvements create a more favorable environment for microbial populations to thrive and proliferate (Reddy *et al.*, 2018).

However, soil application of zinc and seed treatment with zinc solubilizing bacteria recorded non-significant effect on soil enzymatic activities (dehydrogenase and alkaline phosphatase). Saline conditions can affect the enzymatic activity directly by inhibiting enzyme production or indirectly by disrupting microbial metabolism and function. High salt concentrations can negatively impact microbial cell membranes, enzyme stability, and nutrient availability, ultimately affecting enzyme synthesis and activity (Singh *et al.*, 2018). Saline conditions can lead to shifts in microbial community composition. Salt-tolerant or halophilic microorganisms, which may dominate in saline soil, may have different enzymatic capabilities or lower activity compared to non-halophilic counterparts (Tripathi *et al.*, 2019).

3.2.2 Effect of foliar application of nano zinc on biological properties of soil after harvest of barley crop

The biological population (bacteria, fungi and actinomycetes) were significantly increased in the soil after the harvest of barley crop with foliar application of nano-zinc as compared to control (Table 4). The significantly maximum bacteria (39.88×10^7 cfu g⁻¹ of soil), fungi (23.85×10^5 cfu g⁻¹ of soil), actinomycetes (33.77×10^6 cfu g⁻¹ of soil) population in soil was observed under foliar spray of nano Zn at 45 DAS (NP₄₅) as compared to control. The foliar application of nano zinc has been shown to have a positive impact on the microbiological population of soil after the harvest of barley crop in saline soil. The application of nano zinc provides a readily available source of zinc, which serves as a crucial micronutrient for microbial growth and metabolism (Kumar *et al.*, 2020). By supplying an adequate amount of zinc, it helps alleviate zinc deficiency in microorganisms, promoting their growth and activity (Hussain *et al.*, 2020). Nano zinc application has been found to enhance microbial stress tolerance in saline soil. Saline conditions impose stress on microbial communities, negatively impacting their growth and activity (Singh *et al.*, 2018). However, the application of nano zinc can help alleviate the negative effects of salinity stress, enabling microorganisms to thrive and multiply in saline soil environments. The foliar application of nano zinc in saline soil after the harvest of barley crop has been found to increase the microbiological population. This increase is attributed to the improved availability of essential nutrients, enhanced enzymatic activity, microbial stress tolerance, improved soil health, and the indirect effects on organic matter decomposition. These combined effects contribute to a healthier and more abundant microbiological community in saline soil (Yadav *et al.*, 2019).

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Results revealed that with foliar application of nano zinc there is non-significant effect on soil enzymatic activities. According to Xu *et al.* (2015), their study revealed that the presence of TiO₂ and ZnO nanoparticles had a negative impact on soil microbial biomass and enzymatic activities in flooded paddy soil. Similar findings were reported by You *et al.* (2018) in their investigation of ZnO, TiO₂, CeO₂, and Fe₃O₄ nanoparticles' effects on soil enzymatic activities in saline-alkali and black soils. They observed changes in enzymatic activities and alterations in the soil bacterial community, which posed a potential threat to biological nitrogen fixation. Additionally, Chai *et al.* (2015) found that zinc oxide and CeO₂ nanoparticles affected the plate counts of beneficial bacteria such as Azotobacter, P-solubilizing, and K-solubilizing bacteria, and they inhibited enzymatic activities.

4. Conclusion

From the foregoing result, it was concluded that the combined application of the conventional and nano zinc fertilizers significantly affects the soil properties after harvest of barley crop. The treatment combination includes soil application of zinc @ 5 kg Zn ha⁻¹ (Zn_{SA}) along with foliar spray of nano zinc at 45 DAS (NP₄₅) increase soil available nutrient and microbiological population after harvest of barley crop maximum. The combined application of conventional zinc fertilizer and foliar spray of nano zinc offers a promising approach to increase nutrient availability and enhance soil microbial population in saline soil after the harvest of barley crop. By addressing nutrient deficiencies and promoting microbial activity, this integrated approach contributes to improving soil fertility, nutrient cycling, and overall productivity in challenging saline soil conditions.

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Table 1. Mechanical, physico-chemical and biological properties of soil of the experimental field

Comment [a10]: Correct the table number

Characteristics	Value	Method of analysis	Reference
A. Mechanical Composition			
Sand (%)	39.31	By International Pipette Method	Bouyoucos (1962)
Silt (%)	24.96		
Clay (%)	35.21		
Soil texture	Clay loam		Piper (1950)
B. Physical Properties			
Bulk density (Mg m^{-3})	1.29	Core sampler method	Piper (1950)
Particle density (Mg m^{-3})	2.54		Black (1965)
Porosity (%)	49.21		
C. Chemical Properties			
Available N (kg ha^{-1})	272.24	Alkaline KMnO_4 method	Subbiah and Asija (1956)
Available P_2O_5 (kg ha^{-1})	26.19	Olsen's method	Olsen <i>et al.</i> (1954)
Available K_2O (kg ha^{-1})	273.12	Flame photometer	Jackson (1973)
Available Zn (mg kg^{-1})	0.49	DTPA-extract with AAS	Lindsay and Norvell (1978)
Available Fe (mg kg^{-1})	5.84		
Available Mn (mg kg^{-1})	5.20		
Available Cu (mg kg^{-1})	1.51		
Organic carbon (%)	0.52	Walkley and Black's rapid titration method	Walkley and Black (1934)
Electric Conductivity (dS m^{-1} at 25°C)	5.59	Using soltbridge	Richards (1954)
pH (1:2 soil water suspension)	8.79	Glass electrode pH meter	Richards (1954)
D. Biological properties			
Bacterial population (cfu g^{-1} soil)	37.75	Standard serial dilution and plate count method	Schmidt and Colwell (1967)
Fungal population (cfu g^{-1} soil)	23.19		
Actinomycetes population (cfu g^{-1} soil)	32.91		
Microbial biomass carbon (mg kg^{-1})	340 mg kg^{-1}	An extraction method for measuring soil microbial biomass carbon	Vance <i>et al.</i> (1987)

Table 2. Effect of different sources of zinc on available N, P and K in soil after harvest of barley

Comment [a11]: Correct the table number

Treatments	Available Nitrogen (kg ha ⁻¹)	Available Phosphorus (kg ha ⁻¹)	Available Potassium (kg ha ⁻¹)
Main Plot (Soil application and seed treatment)			
Zn ₀ = Control	272.24	26.19	273.12
Zn _{SA} = 5kg Zn ha ⁻¹ (soil application)	291.46	27.33	294.99
Zn _{ST} = Z.S.B. @ 5 ml kg ⁻¹ of seed (Seed treatment)	278.58	26.73	283.56
SEm±	2.916	0.261	1.750
CD (P= 0.05)	11.448	NS	6.872
Sub Plot (Foliar application)			
NP ₀ = Control	272.73	26.27	273.96
NP ₁₅ = Foliar spray of nano Zn at 15 DAS	279.35	26.70	283.86
NP ₃₀ = Foliar spray of nano Zn at 30 DAS	285.01	26.89	288.64
NP ₄₅ = Foliar spray of nano Zn at 45 DAS	285.95	27.13	289.11
SEm±	3.304	0.246	2.050
CD (P= 0.05)	9.816	NS	6.091

Table 3. Effect of different sources of zinc on available micronutrients in soil after harvest of barley

Comment [a12]: Correct the table number

Treatments	Available Micronutrients (mg kg ⁻¹)			
	Zinc	Iron	Manganese	Copper
Main Plot (Soil application and seed treatment)				
Zn ₀ = Control	0.51	5.84	5.20	1.51
Zn _{SA} = 5kg Zn ha ⁻¹ (soil application)	0.64	6.25	5.77	1.82
Zn _{ST} = Z.S.B. @ 5 ml kg ⁻¹ of seed (Seed treatment)	0.61	5.93	5.54	1.68
SEm±	0.004	0.033	0.034	0.010
CD (P= 0.05)	0.017	0.129	0.134	0.041
Sub Plot (Foliar application)				
NP ₀ = Control	0.49	5.83	5.18	1.51
NP ₁₅ = Foliar spray of nano Zn at 15 DAS	0.60	5.97	5.51	1.66
NP ₃₀ = Foliar spray of nano Zn at 30 DAS	0.62	6.08	5.65	1.74
NP ₄₅ = Foliar spray of nano Zn at 45 DAS	0.63	6.12	5.67	1.76
SEm±	0.005	0.040	0.040	0.012
CD (P= 0.05)	0.014	0.118	0.118	0.036

Table 4. Effect of different sources of zinc on soil microbial population, dehydrogenase and alkaline phosphatase enzyme activity after harvest of barley

Comment [a13]: Correct the table number

Treatments	Microbial Population (cfu g ⁻¹ of soil)			Dehydrogenase (µg TPF g ⁻¹ 24 h ⁻¹ soil)	Alkaline Phosphatase (µg of PNP g ⁻¹ h ⁻¹ soil)
	Bacteria (10 ⁷)	Fungi (10 ⁵)	Actinomycete s (10 ⁶)		
Main Plot (Soil application and seed treatment)					
Zn ₀ = Control	38.02	23.24	32.91	5.62	9.93
Zn _{SA} = 5 kg Zn ha ⁻¹ (soil application)	39.84	23.97	33.85	5.69	9.97
Zn _{ST} = Z.S.B. @ 5 ml kg ⁻¹ of seed (Seed treatment)	39.77	23.73	33.56	5.66	9.95
S Em±	0.22	0.14	0.21	0.035	0.061
CD (P= 0.05)	0.86	0.54	0.81	NS	NS
Sub Plot (Foliar application)					
NP ₀ = Control	37.75	23.19	32.87	5.61	9.91
NP ₁₅ = Foliar spray of nano Zn at 15 DAS	39.35	23.70	33.44	5.66	9.96
NP ₃₀ = Foliar spray of nano Zn at 30 DAS	39.84	23.82	33.67	5.67	9.96
NP ₄₅ = Foliar spray of nano Zn at 45 DAS	39.88	23.85	33.77	5.68	9.96
S Em±	0.26	0.16	0.24	0.041	0.072
CD (P= 0.05)	0.77	0.48	0.72	NS	NS

UNDER PEER REVIEW