



Influence of Enriched Zeolite on Chemical and Biological Properties of Black Soil (*Typic Calciusterts*)

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Authors' contributions

This work was carried out in collaboration among all authors. Author JB performed the laboratory and statistical analysis, wrote the protocol and first draft of the manuscript. Author GR designed the study. Authors SHKS and PCL managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

This study investigates the effects of zinc loaded zeolite (ZZ) on pH, electrical conductivity (EC), organic carbon content, and the activities of soil enzymes, namely dehydrogenase, urease, and phosphatase. A laboratory incubation study was conducted in in black soil (*Typic Calciusterts*)

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during 2023-24 with 6 graded levels of Zn loaded zeolite (2.5, 5, 7.5, 10, 12.5, 15 mg/kg) along with control at two different moisture regimes in a factorial completely randomized design (FCRD) at ICAR- Indian Institute of Rice Research. The results revealed that interaction of three factors (treatments, moisture regime and intervals) did not show any significant effects on soil physico-chemical and biological parameters, but on comparison with initial properties, slight variations were observed among all the parameters. Soil pH, EC and organic carbon content status, pH of (8.20) observed at 10 and 12.5 mg kg⁻¹ of ZZ, the highest soil EC (0.41 dS m⁻¹) recorded with the application of 15 and 12.5 mg kg⁻¹ of ZZ. The maximum organic carbon (0.57%) recorded at 60 and 80 DAI in the saturation capacity moisture. For enzymatic activities, the highest dehydrogenase activity the highest (203.4 µg of TPF h⁻¹) was recorded with the application of 10 mg kg⁻¹. Similarly, the peak acid phosphatase activity (25.6 µg p-nitrophenol g⁻¹ of soil h⁻¹) was observed at 80 days after incubation (DAI), while alkaline phosphatase activity was highest at 20 DAI and then declined over time. None of the three enzymatic activities showed significant differences among moisture levels, intervals and treatments. Hence, application of enriched zeolite has no significant effect on soil chemical and biological properties under saturation and field capacity with the three factors interaction.

Keywords: Zinc; zeolite; incubation; saturation capacity; field capacity.

1. INTRODUCTION

Zinc (Zn) is a crucial micronutrient necessary for various physiological and metabolic processes in living organisms. In plants, zinc is vital for chlorophyll synthesis, proper pollen function, effective fertilization, and successful germination [1]. It is uniquely found in all six enzyme classes: oxidoreductases, transferases, hydrolases, isomerases, lyases, and ligases [2], it also plays a significant role in protecting cells from reactive oxygen species (ROS) by regulating free radicals [3]. Additionally, it is involved in biomass production, photosynthesis, cell division, protein synthesis, maintaining membrane integrity, pathogen resistance, and enhancing sexual reproduction through pollen production and changes in stigma structure. Prolonged zinc deficiency leads to reduced vegetative growth and sexual development [4]. The effects of zinc deficiency are more pronounced in neutral to alkaline soils [5], where zinc solubility significantly decreases above a pH of 6.0-6.5 [6]. In plants, zinc deficiency is indicated by dusty brown patches on leaves, known as Khaira disease, which eventually leads to necrosis. This condition impairs photosynthesis rates and causes a 25-30% yield loss. Soil and plant sample analysis has shown that 49% of Indian soils are potentially deficient in zinc [7]. Zn deficiency ranks 11th in the world and 5th in developing countries (WHO, 2002). Deficiency of zinc is more prevalent, occurring in 51.2% of soils in India [8]. Applying zinc fertilizers is an effective and cost-efficient method to correct soil zinc deficiency and boost zinc concentration in grains [9,10]. However, it is important to note

that conventional soluble zinc fertilizers exhibit low use efficiency (2-5%) in neutral to alkaline soils due to zinc immobilization [11]. To prevent zinc deficiency, various methods can be employed, including soil application, biofortification, and conventional breeding [12]. Thus, it is essential to investigate alternative zinc nutrient management strategies that provide high performance and nutrient use efficiency as substitutes for traditional zinc fertilizers [13,14].

In this context, the application of zinc by porous clay minerals, known as zeolites, are strong materials to use as nutrient carriers. Natural zeolites are nanoporous, crystalline, hydrated alumino silicates of alkali and alkaline earth cations that are characterized by reversible dehydration, a large volume of free space, and high cation exchange capacity [15]. Zeolites enhance nutrient use efficiency and influence nutrient release dynamics by increasing availability and reducing losses [16]. The extensive honeycomb crystal structure offers ample storage space for cationic nutrient ions such as potassium (K⁺) and zinc (Zn²⁺), as well as water molecules within the zeolite crystal. This study aimed to investigate the effects of zinc loaded zeolite application on the physio-chemical and biological properties of soil in a different incubation period.

2. MATERIALS AND METHODS

A laboratory incubation study was conducted during 2023-24 at ICAR-Indian Institute of Rice Research. The soil was collected at a depth of 0-15 cm, was pounded and sieved with 2 mm sieve

and used for this study. The effect of Zn-loaded zeolite application on soil physio-chemical and Biological properties was investigated under laboratory incubation experiment in black soil. A sample of 1000g weight of air-dried soil (<2mm) was placed in an air-tight plastic container. Seven treatments were applied including six treatments of Zn-zeolite along with control (No zeolite). T1: Control (No Zeolite), T2: Zn-loaded Zeolite (2.5 mg/kg soil), T3: Zn-loaded Zeolite (5.0 mg/kg soil), T4: Zn-loaded Zeolite (7.5 mg/kg soil), T5: Zn-loaded Zeolite (10.0 mg/kg soil), T6: Zn-loaded Zeolite (12.5 mg/kg soil), T7: Zn-loaded Zeolite (15.0 mg/kg soil). Graded levels of Zn-loaded zeolite were applied to soil and incubated for 80 days. During this period, the soil moisture was maintained at field and saturation capacity at a temperature of $28 \pm 2^\circ\text{C}$. The incubation study was designed in a factorial completely randomized design with three replications. The effect of added Zn-zeolite in black soil at different day intervals was studied by estimating parameters like soil pH, EC, organic carbon and dehydrogenase, urease, acid and alkaline phosphatase at various intervals i.e. 20, 40, 60, and 80 days. The initial properties of the soil used are presented in Table (1). The analyses were conducted according to the standard procedures outlined in Table (2). The statistical analysis was carried out using WASP 1.0 software.

3. RESULTS AND DISCUSSION

3.1 Soil Physio-Chemical Properties

3.1.1 Soil pH

The data on soil pH presented in Table (3) revealed no significant difference between the two moisture levels. However, a significant difference was observed over time, with the highest pH (8.51) recorded at 20 DAI for both moisture levels. The application of graded levels

of Zn-loaded zeolite also showed a significant difference with a pH of (8.20) observed at 10 and 12.5 mg kg⁻¹ of Zn-loaded zeolite. The interactions between moisture and intervals were significant, but others were not significant. The application of zinc loaded zeolite did not significantly affect the soil pH because the small quantities were involved in this study, which did not make any noticeable change in the soil activity. The application of zinc sulphate at the rate of 0, 1.25, 2.50, 5, 10 and 20 kg ha⁻¹ in wheat crops did not show any significant effect on soil pH demonstrated with the results of Keram et al. [22]. No noticeable impact on soil pH was observed with zeolite application (0%, 2%, and 5%) in lettuce crops by Kavvadias et al. [23]. Similar results that zeolite Z₀:0, Z₃:30, Z₆:60, Z₉:90, and Z₁₂:120 t ha⁻¹ application in common bean crop did not show much effect on the soil pH Ozbahce et al. [24].

3.1.2 Soil EC

Soil EC showed a significant difference among the moisture regimes, with the highest mean EC (0.41 dS m⁻¹) observed at field capacity (Table 4). Similarly, a significant difference was noted among intervals, with the highest EC value (0.41 dS m⁻¹) found at 80 DAI for both moisture regimes, which was on par with the values at 20 and 40 DAI at the field capacity. Significant differences were also observed among treatments, with the highest EC (0.41 dS m⁻¹) recorded with the application of 15 and 12.5 mg kg⁻¹ of Zn-loaded zeolite. The interactions among moisture-intervals, moisture-treatments, and intervals-treatments were found to be significant. However, the results indicated that the interactions among moisture regimes, intervals and treatments were not significant. Ozbahce et al. [24] demonstrated that graded levels of zeolite application (0, 30, 60, 90 and 120 t ha⁻¹) had no effect on soil EC in red soil.

Table 1. Initial properties of soil

Sl.No.	Properties	Values
1.	pH	8.60
2.	EC (dSm ⁻¹)	0.44
3.	Organic carbon (%)	0.52
4.	Dehydrogenase (µg TPF g ⁻¹ soil h ⁻¹)	72.0
5.	Urease (µg NH ₄ ⁺ -N g ⁻¹ dry soil)	120.5
6.	Acid phosphatase (µg p-nitrophenol g ⁻¹ of soil h ⁻¹)	17.4
7.	Alkali phosphatase (µg p-nitrophenol g ⁻¹ of soil h ⁻¹)	42.3

Table 2. Soil Analysis methods

S.No.	Parameter	Method	Reference
Physico-chemical Properties			
1.	Soil pH (1:2.5 soil water suspension)	Digital pH meter	Jackson [17]
2.	Soil EC (dS m ⁻¹) (1:2.5 soil water suspension)	Conductivity meter	Jackson [17]
3.	Organic Carbon (%)	Wet Oxidation Method	Walkley and Black [18]
Biological Properties			
4.	Dehydrogenase (µg TPF g ⁻¹ soil h ⁻¹)	Triphenylformazan method	Casida et al. [19]
5.	Urease (µg NH ₄ ⁺ -N g ⁻¹ dry soil)	PMA- KCL method	Tabatabai and Bremmer [20]
6.	Acid and alkali phosphatase (µg p-nitro phenol g ⁻¹ of soil h ⁻¹)	p-nitrophenol method	Eivazi and Tabatabai [21]

3.1.3 Soil OC

There was no significant difference was observed between the moisture regimes and treatments of organic carbon content in Table 5. However, there was a significant difference among intervals, with the maximum organic carbon (0.57%) recorded at 60 and 80 DAI in the saturation capacity moisture regime. The interactions among moisture-intervals, moisture-treatments, and intervals-treatments on soil organic carbon were found to be non-significant. Additionally, the interaction effect of moisture, intervals, and treatments on soil organic carbon was also found to be non-significant. This was in accordance with Ravali et al. [25], that interaction of nitrogen (100, 150, 200 kg ha⁻¹) with zeolite (0, 2.5 5, 7.5 t ha⁻¹) had no significant effect on soil OC and also combined application of clinoptilolite zeolite and compost had no significant impact on SOC (Filcheva and Tsadilas et al. [26]).

3.2 Soil Biological Properties

3.2.1 Dehydrogenase activity

From the results presented in the Table.6, there was a significant difference observed among moisture regimes on dehydrogenase activity, the highest mean (177.1 µg of TPF g soil⁻¹ h⁻¹) was observed in saturation capacity. Similarly significant difference was noted among intervals, the peak dehydrogenase activity (196.7 µg of TPFg soil⁻¹ h⁻¹) was observed at 60 DAI. Significant difference was observed among the treatments, the maximum dehydrogenase activity (203.4 µg of TPF g soil⁻¹ h⁻¹) was found with the application of 10 mg kg⁻¹ Zn loaded zeolite (T5), this level of activity was on par with (T6) in field capacity Fig. 2 and (188.5 µg of TPF g soil⁻¹ h⁻¹) was recorded with application of 10 mg kg⁻¹ of Zn loaded zeolite (T5) of saturation capacity Fig. 1.

A significant effect on dehydrogenase was observed among the interactions of moisture-intervals and intervals-treatments, but there was no significant difference with the interaction of intervals-treatments on dehydrogenase activity. The results presented in the table showed that the interaction among moisture, intervals and treatments was found non-significant. Zinc acts as a cofactor for many enzymes, stabilizing their structure and ensuring proper function. Adequate zinc levels enhance soil microorganism growth and dehydrogenase enzyme production, boosting soil dehydrogenase activity. However, excessive zinc can be toxic and inhibit this activity, necessitating a balanced supply to avoid harmful concentrations. These results were in accordance with Borowik et al. [27], the application of ZnCl₂ was applied in liquid form (0, 300, 600, 1200, 2400 mg Zn²⁺ kg⁻¹) decreased the dehydrogenase activity in black soil.

3.2.2 Urease activity

The urease activity presented in the Table 7 found to be significant among intervals and it is recorded non-significant with moisture regimes and treatments of Zn loaded zeolite. The significant difference on urease activity was observed (214.31 NH₄⁺ -N g⁻¹ dry soil) among intervals was recorded at 20 DAI in field capacity in Fig. 2, this is on par (212.43 NH₄⁺ -N g⁻¹ dry soil) at 20 DAI in saturation capacity Fig. 1. There was no significant difference observed on soil urease activity among the interactions effect of moisture-intervals, moisture-treatments and intervals-treatments. The interaction of moisture, intervals, and treatments didn't show any significant effect on soil urease activity. Zinc application can change the soil chemical environment, potentially affecting the availability of substrates or cofactors necessary for urease

activity. The presence of other ions can influence enzyme stability and function. The decrease in urease concentration due to zinc application is mainly attributed to direct enzyme inhibition, microbial toxicity, changes in soil chemistry, and competition with other essential metal ions. The results were consistent with the findings of Wyszowska et al. [28] with 150 mg.kg⁻¹ of (ZnSO₄.7H₂O) application in sunflower crop where declined activity of urease was recorded and with application of ZnCl₂ in the form of aqueous solution (0, 300, 600, 1200, 2400 mg kg⁻¹) Borowik et al. [27]. but contradictory to decline, Moreno et al. [29], documented that urease activity was found significant with the application of leonardite-enriched zeolite at 38 and 90 t ha⁻¹ [30-32].

3.2.3 Acid phosphatase

The acid phosphatase activity was recorded at different days of incubation (DAI) in soil (Table 8). The results indicated that there was no significant difference among the interactions of

moisture regimes with the application of Zn-loaded zeolite. However, there was a significant difference observed within the moisture regimes, with the highest mean (21.1 µg p-nitrophenol g⁻¹ of soil h⁻¹) observed in saturation capacity Fig. 1. A significant difference was observed in acid phosphatase among intervals, the addition of Zn-loaded zeolite significantly and positively improved the acid phosphatase activity (25.6 µg p-nitrophenol g⁻¹ of soil h⁻¹) at 80 DAI, and no significant difference was observed in treatments and their interactions. The enzyme's activity naturally decreases because the pH is not optimal for its function. The presence of zinc loaded zeolite might not be enough to alter the soil pH to an acidic level where acid phosphatase can be more active. These results were in conformity with the findings of Wyszowska et al. [28] and Moreno et al. [29] that application of 150 mg.kg⁻¹ (ZnSO₄.7H₂O) and graded level (38 and 90 t ha⁻¹) leonardite-enriched zeolite were not improved the acid phosphate activities in sunflower and Barley grown soils, respectively [33,34].

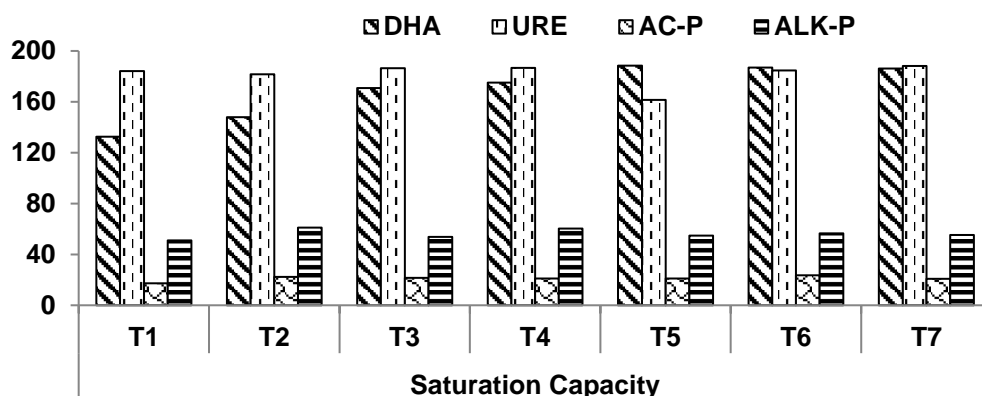


Fig. 1 Representation of the enzymatic activities (Dehydrogenase - µg TPF g⁻¹ soil h⁻¹, Urease- µg NH₄⁺ -N g⁻¹ dry soil; and phosphatase- µg p-nitrophenol g⁻¹ of soil h⁻¹) in saturation capacity

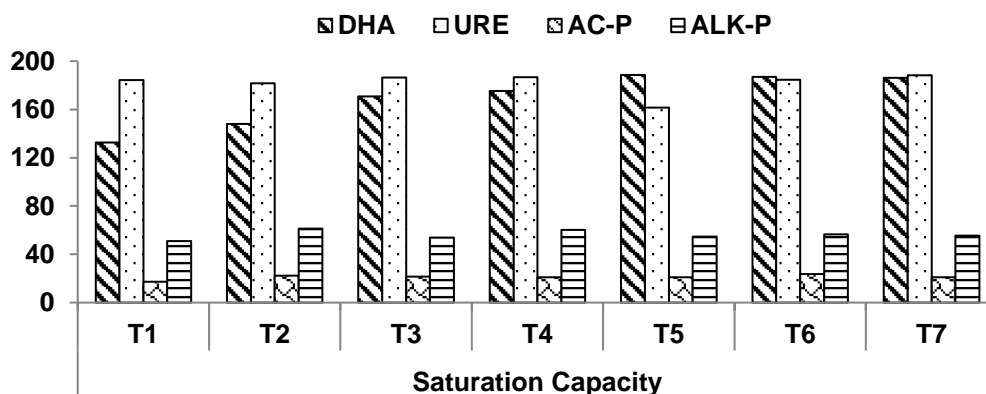


Fig. 2. Representation of enzymatic activities (Dehydrogenase - µg TPF g⁻¹ soil h⁻¹, Urease- µg NH₄⁺ -N g⁻¹ dry soil; and phosphatase- µg p-nitrophenol g⁻¹ of soil h⁻¹) in field capacity

Table 3. Effect of Zn-loaded zeolite application on soil pH under different moisture regimes

Treatments	Saturation Capacity					Field Capacity					Overall mean
	20 DAI	40 DAI	60 DAI	80 DAI	Mean	20 DAI	40 DAI	60 DAI	80 DAI	Mean	
Control	8.56±0.12	8.36±0.04	8.10±0.03	7.46±0.13	8.12	8.56±0.01	8.26±0.02	8.03±0.08	7.56±0.14	8.10	8.11
ZZ- 2.5 mg/kg	8.50±0.31	8.36±0.05	8.06±0.05	7.73±0.10	8.16	8.46±0.26	8.33±0.06	8.16±0.06	7.70±0.07	8.16	8.16
ZZ- 5.0 mg/kg	8.50±0.16	8.33±0.07	8.03±0.08	7.73±0.03	8.15	8.50±0.06	8.30±0.08	8.16±0.10	7.83±0.08	8.20	8.17
ZZ- 7.5 mg/kg	8.56±0.11	8.33±0.03	8.16±0.33	7.66±0.05	8.18	8.50±0.11	8.33±0.08	8.20±0.03	7.66±0.08	8.17	8.17
ZZ- 10.0 mg/kg	8.43±0.09	8.36±0.16	8.20±0.12	7.76±0.09	8.19	8.53±0.03	8.40±0.05	8.16±0.01	7.80±0.01	8.22	8.20
ZZ- 12.5 mg/kg	8.53±0.30	8.43±0.09	8.16±0.02	7.63±0.01	8.19	8.53±0.20	8.40±0.06	8.13±0.06	7.86±0.02	8.23	8.20
ZZ- 15.0 mg/kg	8.50±0.37	8.10±0.11	8.00±0.01	7.56±0.03	8.13	8.50±0.21	8.30±0.32	8.13±0.01	7.46±0.02	8.10	8.10
Mean	8.51	8.37	8.11	7.65	8.16	8.51	8.33	8.14	7.70	8.16	
CD(P=0.05)	Moisture regime		Intervals		Treatments		M x I	M x T	I x T	M x I x T	
	NS		0.028		0.037		0.039	NS	NS	NS	
C.V(%)	0.795										
SE(m) ±	0.021										

* ZZ – Zinc loaded zeolite, DAI = Days after Incubation

Table 4. Effect of Zn loaded zeolite application on soil EC (dS m⁻¹) under different moisture regimes

Treatments	Saturation Capacity					Field Capacity					Overall mean
	20 DAI	40 DAI	60 DAI	80 DAI	Mean	20 DAI	40 DAI	60 DAI	80 DAI	Mean	
Control	0.39±0.01	0.38±0.01	0.36±0.01	0.40±0.01	0.38	0.39±0.01	0.37±0.01	0.39±0.01	0.38±0.01	0.38	0.38
ZZ- 2.5 mg/kg	0.38±0.01	0.40±0.01	0.40±0.01	0.40±0.01	0.39	0.40±0.01	0.40±0.01	0.41±0.01	0.41±0.01	0.40	0.40
ZZ- 5 mg/kg	0.38±0.02	0.39±0.01	0.39±0.02	0.41±0.01	0.39	0.41±0.02	0.40±0.01	0.42±0.01	0.40±0.02	0.40	0.40
ZZ- 7.5 mg/kg	0.38±0.01	0.39±0.02	0.39±0.01	0.42±0.01	0.39	0.42±0.02	0.40±0.01	0.42±0.01	0.43±0.02	0.41	0.40
ZZ- 10 mg/kg	0.39±0.01	0.38±0.02	0.40±0.01	0.42±0.01	0.39	0.41±0.01	0.41±0.01	0.41±0.02	0.41±0.01	0.41	0.40
ZZ- 12.5 mg/kg	0.38±0.02	0.37±0.02	0.42±0.01	0.42±0.02	0.40	0.41±0.01	0.41±0.01	0.42±0.02	0.42±0.02	0.42	0.41
ZZ- 15 mg/kg	0.39±0.02	0.41±0.02	0.42±0.01	0.39±0.02	0.40	0.41±0.01	0.43±0.01	0.42±0.01	0.41±0.02	0.42	0.41
Mean	0.38	0.39	0.39	0.41	0.39	0.40	0.40	0.41	0.41	0.41	
CD(P=0.05)	Moisture regime		Intervals		Treatments		M x I	M x T	I x T	M x I x T	
	0.01		0.01		0.01		0.01	0.02	0.03	NS	
C.V(%)	4.13										
SE(m) ±	0.01										

* ZZ – Zinc loaded zeolite, DAI = Days after Incubation

Table 5. Effect of Zn-loaded zeolite application on organic carbon (%) under different moisture regimes

Treatments	Saturation Capacity					Field Capacity					Overall mean
	20 DAI	40 DAI	60 DAI	80 DAI	Mean	20 DAI	40 DAI	60 DAI	80 DAI	Mean	
Control	0.50±0.03	0.51±0.03	0.50±0.03	0.54±0.01	0.51	0.52±0.02	0.51±0.01	0.50±0.03	0.56±0.02	0.52	0.51
ZZ- 2.5 mg/kg	0.49±0.02	0.50±0.02	0.51±0.02	0.56±0.02	0.51	0.49±0.01	0.52±0.02	0.51±0.01	0.56±0.01	0.52	0.51
ZZ- 5 mg/kg	0.47±0.03	0.51±0.01	0.54±0.02	0.56±0.03	0.52	0.49±0.03	0.50±0.02	0.52±0.03	0.55±0.01	0.51	0.51
ZZ- 7.5 mg/kg	0.51±0.02	0.53±0.02	0.49±0.03	0.60±0.02	0.53	0.52±0.04	0.51±0.01	0.51±0.02	0.55±0.02	0.52	0.52
ZZ- 10 mg/kg	0.53±0.04	0.51±0.02	0.55±0.02	0.62±0.02	0.55	0.51±0.02	0.49±0.02	0.52±0.02	0.53±0.03	0.51	0.53
ZZ- 12.5 mg/kg	0.51±0.03	0.50±0.01	0.53±0.02	0.59±0.03	0.53	0.53±0.03	0.50±0.03	0.50±0.03	0.53±0.02	0.51	0.52
ZZ- 15 mg/kg	0.52±0.02	0.51±0.02	0.52±0.02	0.56±0.02	0.52	0.49±0.01	0.51±0.03	0.53±0.02	0.56±0.02	0.52	0.52
Mean	0.50	0.51	0.57	0.57	0.52	0.50	0.51	0.51	0.54	0.52	
CD(P=0.05)	Moisture Regime		Intervals		Treatments		M x D	M x T	D x T	M x D x T	
C.V(%)	NS		0.017		NS		NS	NS	NS	NS	
SE(m) ±	7.74										
	0.01										

* ZZ – Zinc loaded zeolite, DAI = Days after Incubation

Table 6. Effect of Zn loaded zeolite on soil dehydrogenase activity ($\mu\text{g TPF g}^{-1} \text{ soil h}^{-1}$)

Treatments	Saturation Capacity					Field Capacity					Overall mean
	20 DAI	40 DAI	60 DAI	80 DAI	Mean	20 DAI	40 DAI	60 DAI	80 DAI	Mean	
Control	135±7.9	132±10.5	132±6.5	130±8.5	132	112±5.2	149±14.8	152±5.2	134±12.6	136	134.70
ZZ- 2.5 mg/kg	149±12.9	134±11.4	153±12.4	154±16.4	147	121±8.6	173±12.8	175±5.7	156±9.8	156	152.21
ZZ- 5 mg/kg	161±18.1	177±3.03	182±13.1	161±11.5	170	130±11.6	194±15.9	195±10.6	170±13.0	172	171.70
ZZ- 7.5 mg/kg	163±15.0	187±10.2	197±12.4	152±13.6	175	148±4.9	202±6.7	212±7.6	186±12.7	187	181.30
ZZ- 10 mg/kg	148±17.6	219±4.51	214±8.04	172±13.0	188	173±10.6	221±5.5	212±1.1	206±5.7	203	196.00
ZZ- 12.5 mg/kg	136±16.2	221±1.63	199±7.8	191±7.11	187	159±18.1	220±14.3	213±9.6	195±3.2	197	192.10
ZZ- 15 mg/kg	139±18.3	206±1.90	218±4.7	180±8.54	186	135±7.2	205±3.8	217±3.2	186±5.7	185	186.06
Mean	147.0	182.58	185.50	163.29	169	140.05	195.07	196.76	176.56	177.1	
CD(P=0.05)	Moisture Regime		Intervals		Treatments		M x I	M x T	I x T	M x I x T	
C.V(%)	5.50		7.78		10.30		11.01	NS	20.6	NS	
SE(m) ±	10.49										
	6.06										

* ZZ – Zinc loaded zeolite, DAI = Days after Incubation

Table 7. Effect of Zn loaded application on soil urease activity ($\mu\text{g NH}_4^+ \text{-N g}^{-1}$ dry soil)

Treatments	Saturation Capacity					Field Capacity					Overall mean
	20 DAI	40 DAI	60 DAI	80 DAI	Mean	20 DAI	40 DAI	60 DAI	80 DAI	Mean	
Control	192±4.0	176±2.4	175±2.59	184±6.0	182.19	205±12.9	191±7.9	180±6.9	170±0.8	187.00	184.63
ZZ- 2.5 mg/kg	197±11.5	204±9.9	181±7.2	181±7.8	191.56	209±6.0	182±8.0	181±4.5	171±1.9	186.52	189.04
ZZ- 5 mg/kg	301±8.4	186±7.7	192±4.8	186±6.4	216.78	210±8.9	190±6.7	179±4.2	176±3.3	189.40	203.09
ZZ-7.5 mg/kg	228±26.0	193±5.1	183±7.4	186±10.4	198.22	212±12.9	181±2.8	185±7.7	169±0.6	187.31	192.77
ZZ-10 mg/kg	197±4.6	186±3.3	180±8.8	161±10.6	180.30	220±18.5	187±7.8	184±3.4	171±1.9	190.94	185.62
ZZ-12.5 mg/kg	186±9.4	183±1.4	180±1.87	184±5.9	183.80	213±16.7	184±8.1	195±1.3	175±7.5	191.96	187.88
ZZ- 15 mg/kg	181±3.1	172±10.9	190±3.8	188±5.5	183.21	227±25.4	195±4.0	189±15.1	171±2.7	196.12	189.67
Mean	212	185	183	181	190	214	187	185	172	189.90	
CD(p=0.05)	Moisture Regime		Days		Treatments		M x D	M x T	D x T	M x D x T	
	NS		11.34		NS		NS	NS	NS	NS	
C.V(%)	13.93										
SE(m) ±	8.83										

* ZZ – Zinc loaded zeolite, DAI = Days after Incubation

Table 8. Effect zinc loaded zeolite application on soil acid phosphatase activity ($\mu\text{g p-nitrophenol g}^{-1}$ of soil h^{-1})

Treatments	Saturation Capacity					Field Capacity					Overall mean
	20 DAI	40 DAI	60 DAI	80 DAI	Mean	20 DAI	40 DAI	60 DAI	80 DAI	Mean	
Control	13±0.9	16±0.7	18±2.4	21±0.7	17.35	12±0.9	15±2.3	18±1.3	22±3.3	17.36	17.36
ZZ- 2.5 mg/kg	14±0.6	20±3.3	25±2.3	27±2.9	22.20	13±0.6	15±0.3	20±3.6	23±3.4	18.24	20.22
ZZ- 5 mg/kg	14±0.5	23±3.1	21±1.3	27±1.0	21.53	13±0.5	15±0.3	20±3.7	21±2.4	17.85	19.69
ZZ- 7.5 mg/kg	15±1.5	20±0.6	23±1.4	25±2.3	21.07	14±1.5	16±3.0	19±4.2	20±2.7	17.83	19.45
ZZ- 10 mg/kg	15±1.0	20±0.9	22±2.1	25±1.4	21.03	14±1.0	15±3.0	18±1.8	25±1.5	18.17	19.60
ZZ- 12.5 mg/g	22±0.8	24±4.1	21±3.3	26±2.1	23.67	17±0.8	16±0.9	19±1.8	19±1.5	18.00	20.84
ZZ- 15 mg/kg	18±2.0	18±2.3	21±2.7	25±1.0	20.90	17±2.0	12±0.5	15±3.0	21±2.8	16.64	18.77
Mean	16.28	20.58	21.98	25.60	21.11	14.64	15.32	18.95	22.05	17.73	
CD(P=0.05)	Moisture Regime		Intervals		Treatments		M x I	M x T	D x T	M x I x T	
	1.21		1.71		NS		NS	NS	NS	NS	
C.V(%)	20.59										
SE(m) ±	1.33										

Table 9. Effect of Zn loaded zeolite application on soil Alkaline phosphatase activity ($\mu\text{g p-nitrophenol g}^{-1}$ of soil h^{-1})

Treatments	Saturation Capacity					Field Capacity					Overall mean
	20 DAI	40 DAI	60 DAI	80 DAI	Mean	20 DAI	40 DAI	60 DAI	80 DAI	Mean	
Control	62±2.3	57±1.8	54±0.6	51±0.7	56.10	50±3.9	86±5.4	51±2.1	52±2.4	53.50	54.80
ZZ- 2.5 mg/kg	69±5.0	57±2.2	52±2.7	61±4.1	60.30	61±1.4	59±3.4	57±2.2	51±3.5	57.30	58.80
ZZ- 5 mg/kg	69±4.9	56±3.9	52±5.1	53±2.7	58.01	59±2.6	61±6.8	53±2.8	53±1.6	56.90	57.40
ZZ- 7.5 mg/kg	75±0.7	60±0.6	59±2.3	60±0.5	63.90	58±2.1	63±5.9	59±2.9	54±5.4	58.90	61.40
ZZ- 10 mg/kg	75±4.5	67±6.0	55±1.8	54±3.7	63.40	56±2.7	65±5.1	61±4.7	55±1.5	59.50	61.40
ZZ- 12.5 mg/kg	79±6.2	62±2.4	52±5.2	56±3.8	62.60	57±0.8	67±6.0	59±4.4	52±1.6	59.20	60.90
ZZ- 15 mg/kg	68±5.7	54±3.5	54±5.4	55±1.0	58.40	55±4.3	68±9.3	58±3.8	51±2.8	58.70	58.50
Mean	71.39	59.54	54.50	56.14		56.95	63.61	57.38	52.94		
	Moisture regime		Intervals		Treatments	M x I	M x T	I x T	M x I x T		
CD(P=0.05)	2.05		2.9		3.84	4.11	NS	NS	NS		
C.V(%)	11.70										
SE(m) ±	2.26										

* ZZ – Zinc loaded zeolite, DAI = Days after Incubation

3.2.4 Alkaline phosphatase

The results indicated that there was no significant difference among interactions of moisture regimes with the application of graded levels of Zn-loaded zeolite (Table 9). However, significant influence was observed among the moisture regimes, the highest mean ($60.3 \mu\text{g p-nitrophenol g}^{-1}$ of soil h^{-1}) was observed at saturation capacity. Similarly significant difference was observed among intervals, with the highest mean (71.3 and $63.6 \mu\text{g p-nitrophenol g}^{-1}$ of soil h^{-1}) observed at 20 DAI in saturation capacity moisture regime Fig. 1 and 40 DAI in field capacity moisture regime Fig. 2, respectively. Significant difference was observed among treatments, the maximum alkaline phosphatase activity ($63.9 \mu\text{g p-nitrophenol g}^{-1}$ of soil h^{-1}) was observed with T4 at saturation capacity and ($59.5 \mu\text{g p-nitrophenol g}^{-1}$ of soil h^{-1}) was observed with T5 at field capacity. Alkaline phosphatase operates optimally in alkaline conditions, so the enzymes already functioning near their peak activity in alkaline soils. Adding zinc zeolite might not significantly enhance its activity further and it may not require zinc as a cofactor, but excess zinc can also inhibit enzyme activity due to toxicity. Wyszowska et al. [28] reported, similar findings in sunflower with $150 \text{ mg}\cdot\text{kg}^{-1}$ ($\text{ZnSO}_4\cdot 7\text{H}_2\text{O}$) application. Moreno et al. [29] reported non-significant results with leonardite-enriched zeolite application at 38 and 90t ha^{-1} .

4. CONCLUSION

The application of Zn-loaded zeolite application did not have any significant influence on pH, EC and organic carbon of black soil. However, over time, the release of Zn from Zn-Zeolite has enhanced the soil pH to the neutral level followed by slight increase in EC where a higher concentration of Zn-Zeolite is added, but a slight decrease in soil pH was observed as time progressed and the application of Zn-loaded zeolite slightly increased soil electrical conductivity. In case of soil enzymatic activities, graded level of Zn-Zeolite enhanced the activities *i.e.* dehydrogenase, urease and phosphatases over the period, which could be ascertained that Zn released from zeolite acted as a co-factor for enzymatic reactions in black soil but not significantly. Hence, it can be concluded that the addition of zinc rich zeolite may support the crop and nutrition by improving the enzymatic activities in black soil.

5. FUTURE RESEARCH WORK

Study the impact of zinc-loaded zeolite on a variety of crops beyond rice, including cereals, legumes, and vegetables.

Assess the long-term effects of zinc-loaded zeolite on soil health and crop productivity.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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