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## Effect of zinc mobilizing microbial cultures and zinc levels enhancing yield and enzymatic activity in soybean grown on Vertisol

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**Abstract**

Field experiment was conducted during *Kharif* season of 2017 at Research Farm, Department of Soil Science and Agricultural Chemistry at Vasandrao Naik Marathwada Krishi Vidyapeeth, Parbhani on zinc deficient Vertisol to study the zinc solubilization potential of different microorganisms in soybean crop. The experiment consist of sixteen treatment in which four laboratory pre-evaluated zinc solubilizing microbial cultures (Control, *Pseudomona strita*, *Bacillus megaterium*, and *Trichoderma viride*) and four graded doses of ZnSo<sub>4</sub> (0,10,20,30, kg ha<sup>-1</sup>) were used in the factorial randomized block design. The results emerged out indicated significant increase in yield and enzymatic activity of soybean grown soil was improved in the plots treated with *pseudomon strita* along with 30 kg ZnSo<sub>4</sub> ha<sup>-1</sup> and was found as the best combination in overall improvement of yield and enzymatic activity of soybean grown soil.

**Keywords:** Mobilizing, microbial, cultures, enzymatic activity, Vertisol

**Introduction**

Major soybean growing states in India are Madhya Pradesh, Maharashtra, Gujarat, Rajasthan, Karnataka and Andhra Pradesh. Maharashtra ranks 2<sup>nd</sup> in terms of production of soybean after Madhya Pradesh. In India area, production and productivity of soybean during 2017 is 101.5 lakh ha, 91.4 lakh million tonnes and 900 kg ha<sup>-1</sup>, respectively, in Maharashtra area, production and productivity of soybean during 2017 is 34.4 lakh ha, 31.8 lakh million tonnes and 925kg ha<sup>-1</sup>, respectively. (Anonymous, 2017) <sup>[1]</sup>. The area under soybean cultivation is increasing due to some reason such as soybean is short duration crop (90-110 days), good market price with its higher productivity compared to other pulses. It can be processed easily for different products *viz.*, soy cheese, soy milk, soy protein, soy yogurt, soybean oil, soy nut etc. Soybean is also used for making the soy ink, soy paint and soy molasses. It can give a boost to the food-processing industry in rural areas of India. Soybean is miracle golden bean of 21<sup>st</sup> century which possesses potential to revolutionized Indian economy by correcting the health of human being and soil.

The biochemical properties of soil have often been proposed as early and sensitive indicators of soil ecosystem health. Activities of soil enzymes indicate the direction and strength of all kinds of biochemical processes in soil and act as key biological indicator of soil. Soil enzymes play an essential role in energy transfer environmental quality, organic matter decomposition, nutrient cycling and crop productivity. (Usha Mina *et al.*, 2011) <sup>[2]</sup>. A bacterial based approach was devised to solve the micro nutrient deficiency problem. The basic principle behind this approach is decreasing the pH and making zinc soluble and as a consequence the available zinc will get increased in the soil system. The term called zinc solubilizing bacteria was coined for those bacteria that are capable of solubilizing the insoluble zinc compounds or minerals in agar plate as well as in soil. (Sarathamble *et al.*, 2010) <sup>[3]</sup>.

**Material and Methods**

The present investigation was carried at Research Farm, Department of Soil Science and Agricultural Chemistry at Vasandrao Naik Marathwada Krishi Vidyapeeth, Parbhani on Vertisol during 2017-18.

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The initial soil pH was 8.20, EC-0.22 dSm<sup>-1</sup>, Organic Carbon-4.43 g kg<sup>-1</sup>, Calcium carbonate -4.05%, available nitrogen-145 kg ha<sup>-1</sup>, Phosphorus -15.23, Potassium- 556 kg ha<sup>-1</sup> and Sulphur-9.38 mg kg<sup>-1</sup>. The initial micronutrient status was DTPA Copper-2.38, Manganese-7.34, Zinc-0.58 and Ferrous – 3.15 mg kg<sup>-1</sup>. The soil was clayey in texture, moderately alkaline in reaction, medium in available nitrogen, phosphorus and sufficient in available potassium and low in sulphur and iron.

The field experiment was carried out on soybean crop (Variety MAU-162) in kharif season during year 2017-18. After completion of preparatory tillage operations, the experiment was laid out in factorial randomized block design comprising (16) treatments and replicated (3) times.

### The experiment Consist of two factors

Factor-1- Zinc levels Factor-2- Zinc mobilizing cultures

1. Zn0- 0 kg ZnSO<sub>4</sub> ha<sup>-1</sup> 1. S0- Control
2. Zn1- 10 kg ZnSO<sub>4</sub> ha<sup>-1</sup> 2. S1- *Pseudomona striata*
3. Zn2- 20 kg ZnSO<sub>4</sub> ha<sup>-1</sup> 3. S2- *Bacillus megaterium*
4. Zn3- 30 kg ZnSO<sub>4</sub> ha<sup>-1</sup> 4. S3- *Trichoderma Viride*

Seed treatment was done immediate before sowing with liquid zinc mobilizing cultures @ 100 ml 10 kg<sup>-1</sup> seed. The crop was raised following recommended agronomic practices. The recommended dose of chemical fertilizer applied @ 30:60:30 NPK kg ha<sup>-1</sup>.

The soil sample was collected after harvest of soybean for

analysis of chemical properties and available nutrients status as per standard procedures.

## Results and Discussion

### Effect of zinc mobilizing cultures and zinc levels on seed and straw yield of Soybean

Data given in Table 1 reveal significant increase of seed and straw yield due to addition of zinc solubilizing microorganisms. Zinc solubilizers influenced the seed yield which ranged between 1941 to 2345 kg ha<sup>-1</sup> and straw yield 2731 to 2866 kg ha<sup>-1</sup> showing significantly higher seed and straw yield in *Pseudomona striata* treated plots follows by *Bacillus megaterium* and *Trichoderma viride*. Whereas, significantly lower seed and straw yield per plot were noted in uninoculated control. Similarly graded levels of zinc in the form of zinc sulphate also increased the seed and straw yield with each incremental dose up to 30 kg ZnSO<sub>4</sub> kg ha<sup>-1</sup>. The seed yield as influenced by Zn application ranged from 1741 to 2426 and straw yield 2591 to 2942 kg ha<sup>-1</sup>.

Interaction effect of zinc mobilizing cultures and zinc levels on seed and straw yield of soybean is given in Table 1a. Synergistic effect of both factor was recorded on each other showing significantly highest seed yield in *Pseudomona striata* X ZnSO<sub>4</sub> 30 kg ha<sup>-1</sup> (2426 kg ha<sup>-1</sup>) and straw yield (2786 kg ha<sup>-1</sup>) it was found a t par with *Bacillus megaterium* X ZnSO<sub>4</sub> 30 kg ha<sup>-1</sup>. However the lower values of seed yield (7141) and straw yield (2591) was recorded in without Zn application.

**Table 1:** Seed and Straw yield of soybean as influenced by zinc mobilizing cultures and zinc levels

Treatment	Seed yield (kg ha <sup>-1</sup> )	Straw yield (kg ha <sup>-1</sup> )
<b>Zinc solubilizers (S)</b>		
S0: Control	1941	2731
S1: <i>Pseudomona striata</i>	2345	2866
S2: <i>Bacillus megaterium</i>	2001	2817
S3: <i>Trichoderma viride</i>	2041	2786
S.Em.±	49.19	21.72
C.D. at 5%	142.0	62.73
<b>Levels of ZnSO<sub>4</sub> (Zn)</b>		
Zn0: ZnSO <sub>4</sub> 0 kg ha <sup>-1</sup>	1741	2591
Zn1: ZnSO <sub>4</sub> 10 kg ha <sup>-1</sup>	1967	2805
Zn2: ZnSO <sub>4</sub> 20 kg ha <sup>-1</sup>	2195	2862
Zn3: ZnSO <sub>4</sub> 30 kg ha <sup>-1</sup>	2426	2942
S.Em.±	49.19	21.72
C.D. at 5%	142.0	62.73
<b>Interaction (SxZn)</b>		
S.Em.±	98.38	43.45
C.D. at 5%	284.12	125.47
CV %	8.18	2.69

Zaidi and Singh (2001) [4] observed that inoculation with *Bradyrhizobium Japonicum* strains SB-12 and difference isolates of *Pseudomona* as well as their possible combination significantly increased yield of soybean over control. This might be due to reduction in infection by soil pathogen and greater seedling emergence. Similarly, Goudar *et al.* (2008) [5] found that grain yield of soybean was significantly influenced

by the seed inoculation of *Bradyrhizobium japonicum* strains along with micronutrients over uninoculated control. Further, Gupta (2006) [6] noted that the seed inoculation with *Rhizobium* + *PSB* + *Azotobacter* increased grain yield in chickpea. Thereafter, Prasad *et al.* (2017) indicate significant impact of Zn solubilizing microorganisms on dry pod and dry haulm yield of groundnut.

**Table 1a:** Interaction effect of zinc mobilizing cultures and graded level of zinc on seed yield and straw yield of soybean

Treatment	Zn0: ZnSO <sub>4</sub> 0 kg ha <sup>-1</sup>	Zn1: ZnSO <sub>4</sub> 10 kg ha <sup>-1</sup>	Zn2: ZnSO <sub>4</sub> 20 kg ha <sup>-1</sup>	Zn3: ZnSO <sub>4</sub> 30 kg ha <sup>-1</sup>
<b>Seed yield (kg ha<sup>-1</sup>)</b>				
S0: Control	1802	1896	1996	2072
S1: <i>Pseudomona striata</i>	1730	2053	2658	2940
S2: <i>Bacillus megaterium</i>	1723	1955	2046	2281
S3: <i>Trichoderma viride</i>	1709	1965	2079	2410

Interaction (SxZn)	S	Zn	SXZn	
SEm+	49.19	49.19	98.38	
CD at 5%	142.0	142.0	284.12	
Straw yield (kg ha <sup>-1</sup> )				
S0: Control	2622	2702.70	2744	2857
S1: <i>Pseudomona striata</i>	2507	2929.00	2972	3055
S2: <i>Bacillus megaterium</i>	2647	2805.07	2890	2927
S3: <i>Trichoderma viride</i>	2588	2786	2840	2930
Interaction (SxZn)	S	Zn	SXZn	
SEm+	21.72	21.72	43.45	
CD at 5%	62.73	62.73	125.47	

Significantly highest dry pod yield and dry haulm yield of groundnut was noted in RDF + *Rhizobium* + *Pseudomona striata* but treatment RDF + *Rhizobium* + *Pseudomona fluorescens*, RDF + *Rhizobium* + *Trichoderma viride*, RDF + *Rhizobium* + *Trichoderma harzianum* and RDF + *Rhizobium* + *Burkholderia cenocepacia* were found statistically at par. Lowest biological yields were obtained in treatment RDF + *Rhizobium*.

Further, Ekundayo (2003) [7] also observed greater seedlings emergence with inoculated seeds compared with uninoculated, maximum straw yield was recorded with *Pseudomona*. However, Patel *et al.* (2010) [10] also noted that application of 20 kg N + 40 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> and seed inoculation with *Rhizobium* + *PSB* increasing seed yield and straw yield in cowpea. Thereafter Pable *et al.* (2010) [9] reported the significant effect of the sulphur and zinc on yield of soybean in Vertisol and found the highest increase in grain and straw yield of 1755 kg ha<sup>-1</sup> and 3114 kg ha<sup>-1</sup> respectively with the application of 30 kg S ha<sup>-1</sup> + 2.5 kg Zn ha<sup>-1</sup> + RDF treatment. Similarly, (Bairagi *et al.*, 2007) [10] conducted a field experiment consisted of three levels of zinc (0, 10, 20, 30 kg ha<sup>-1</sup>) and four levels of P (0, 50, 75, 100 30 kg ha<sup>-1</sup>) in Maharashtra during kharif 2004 to study the effect of zinc and phosphorus on grain yield and straw yield due to application of zinc. The grain yield was highest with 10 kg Zn ha<sup>-1</sup> (15.15 and 26.75 q ha<sup>-1</sup>).

#### Effect of zinc mobilizing cultures and zinc levels on enzymatic activities in soil after harvest of soybean

Soil enzymatic activities can display the overall level of soil biological activity as well as transforming course of soil nutrients and are important indices to measure the fertility level of soil

#### Dehydrogenase activity in soil

Dehydrogenase is considered as an indicator of overall microbial activity because it occurs intracellularly in all living

microbial cells and it is linked with microbial respiratory processes. The dehydrogenase enzyme is known to oxidize soil organic matter by transferring protons and electrons from substrates to acceptors. These processes are part of respiration, pathways of soil microorganisms and closely related to the type of soil. related to dehydrogenase activity in soybean indicate significant effect of zinc solubilizers and graded level of zinc.

The results indicated that dehydrogenase activity in the rhizosphere soil gradually increased ranged between (49.50 to 60.33 µg TPF g<sup>-1</sup> soil 24 hr<sup>-1</sup>) showing significantly higher dehydrogenase activity in *Pseudomona striata* treated plots follows by *Bacillus megaterium* and *Trichoderma viride*. Whereas, significantly lower dehydrogenase activity per plot were noted in uninoculated control. Similarly, graded levels of zinc in the form of zinc sulphate also increase the dehydrogenase activity with each incremental dose up to 30 kg ZnSO<sub>4</sub> kg ha<sup>-1</sup>. The dehydrogenase activity as influenced by Zn application ranged from 48.67 to 59.97 TPF g<sup>-1</sup> soil 24 hr<sup>-1</sup>.

Interaction effect of zinc solubilizers and zinc levels on dehydrogenase activity in Table 2a. Showing significantly the highest dehydrogenase activity was noticed under in *Pseudomona striata* X ZnSO<sub>4</sub> 30 kg ha<sup>-1</sup> (67.33 TPF g<sup>-1</sup> soil 24 hr<sup>-1</sup>) it was found at par with *Bacillus megaterium* X ZnSO<sub>4</sub> 30 kg ha<sup>-1</sup>. However the lower values of dehydrogenase activity (44 TPF g<sup>-1</sup> soil 24 hr<sup>-1</sup>) was recorded in uninoculated control.

**Table 2:** Effect of zinc mobilizing cultures and zinc levels on enzymatic activities in soil after harvest of soybean

Treatments	Soil Dehydrogenase (µg g <sup>-1</sup> soil)	Acid phosphatase (µg g <sup>-1</sup> soil)	Alkaline phosphatase (µg g <sup>-1</sup> soil)	Soil Urease (µg g <sup>-1</sup> soil)
Zinc solubilizers (S)				
S0: Control	49.50	46.67	68.75	74.50
S1: <i>Pseudomona striata</i>	60.33	56.58	77.67	84.50
S2: <i>Bacillus megaterium</i>	58.38	52.66	73.83	81.08
S3: <i>Trichoderma viride</i>	50.37	49.89	70.75	76.00
S.Em.±	1.22	0.39	0.40	0.47
C.D. at 5%	3.54	1.13	1.15	1.37
Levels of ZnSO <sub>4</sub> (Zn)				
Zn0: ZnSO <sub>4</sub> 0 kg ha <sup>-1</sup>	48.67	47.00	67.58	73.83
Zn1: ZnSO <sub>4</sub> 10 kg ha <sup>-1</sup>	53.46	50.33	72.00	77.17
Zn2: ZnSO <sub>4</sub> 20 kg ha <sup>-1</sup>	56.48	52.77	74.75	81.17
Zn3: ZnSO <sub>4</sub> 30 kg ha <sup>-1</sup>	59.97	55.70	76.67	83.92
S.Em.±	1.22	0.39	0.40	0.47
sC.D. at 5%	3.54	1.13	1.15	1.37
Interaction (SxZn)				
S.Em.±	2.45	0.78	0.80	0.95
C.D. at 5%	7.08	2.27	2.31	2.75
CV %	7.77	2.65	1.91	2.09

Dehydrogenase activity reflects the oxidative activity or intensity of metabolism of soil microflora and can be used as an indicator of microbial activity or populations in soil (Beura and Rakshit, 2011) [11]. Our results are confirmed with the findings of Kohler *et al.*, (2007) [12]; Mader *et al.*, (2010) [13] and Rana *et al.*, (2012) [14] who reported that the higher dehydrogenase activity in the rhizosphere soils with inoculation can be ascribed to the availability of a high quantity of biodegradable substrates and hence an improvement in their microbial activity. Similarly, Ramesh *et al.*, (2014) [15] reported that there is significant increase in dehydrogenase activity with the inoculation of *Bacillus aryabhatai* strains. Further, Nihorimbere *et al.* (2011) [16] who reported more microbial activities increased the dehydrogenase activity in rhizosphere due to more availability of food material for its growth.

Dotaniya *et al.* (2014) [5] reported the maximum DHA (Dehydrogenase activity) was at 75 DAS. It might be due to vigorous growth of the crop at 75 DAS and root exudation also more in this period. Nihorimbere *et al.* (2011) [16] reported more microbial activities and increase in the DHA in rhizosphere due to more availability of food material for its growth. Thereafter, The dehydrogenase activity at flowering (72.89  $\mu\text{g g}^{-1} 24 \text{ h}^{-1}$ ) and harvest (49.78  $\mu\text{g g}^{-1} 24 \text{ h}^{-1}$ ) were highest with the treatment receiving combined application of 150 kg N ha<sup>-1</sup> + FYM + Bio fertilizers in rice (Goutami *et al.*, 2015) [18]. Further, Apoorva (2016) [19] reported that the highest dehydrogenase activity was observed in the treatment receiving RDF+ Soil application of biozinc @ 30 kg ha<sup>-1</sup> (86.5  $\mu\text{g TPF produced g soil d}^{-1}$ ) which was on par with RDF+ Soil application of ZnSO<sub>4</sub> @ 25 kg ha<sup>-1</sup> (81.9  $\mu\text{g TPF produced g soil d}^{-1}$ ). These treatments were followed by RDF+ Soil application of bio zinc @ 15 kg ha<sup>-1</sup> (76.8  $\mu\text{g TPF produced g soil d}^{-1}$ ). The lowest dehydrogenase activity was observed in control (30.2  $\mu\text{g TPF produced g soil d}^{-1}$ ) which was on par with RDF+ foliar application of 0.2% ZnSO<sub>4</sub> spray (39.2  $\mu\text{g TPF produced g soil d}^{-1}$ ).

#### Acid phosphatase activity in soils

The activities of acid phosphatase is predominate in acid soil and predominantly due to plants. Phosphatase activity has been closely correlates with pH. However, acid phosphatase is mostly of plant and associated fungal origin. The acid phosphatase activity would be more dependent upon the nutritional status of the plant. The acid phosphatase activity was significantly influenced by different zinc solubilizing microbial strains and data is presented in Table 2. Zinc solubilizers influenced the acid phosphatase activity which ranged from (46.67 to 56.58  $\mu\text{g g}^{-1}$  of soil.) showing significantly higher acid phosphatase activity in *Pseudomona striata* treated plots follows by *Bacillus megaterium* and *Trichoderma viride*. Whereas, significantly lower acid phosphatase activity per plot were noted in uninoculated control. Similarly, applied levels of zinc in the form of zinc sulphate also increase the acid phosphatase activity with each incremental dose up to 30 kg ZnSO<sub>4</sub> kg ha<sup>-1</sup>. The acid phosphatase activity as influenced by Zn application ranged from 47.00 to 55.70  $\mu\text{g g}^{-1}$  of soil.

Interaction effect of zinc mobilizing cultures and zinc levels on dehydrogenase activity in table 2a. Showing significantly the highest acid phosphatase activity was noticed under in *Pseudomona striata* X ZnSO<sub>4</sub> 30 kg ha<sup>-1</sup> (59.67  $\mu\text{g g}^{-1}$  of soil) it was found at par with *Bacillus megaterium* X ZnSO<sub>4</sub> 30 kg ha<sup>-1</sup>. However the lower values of acid phosphatase (42  $\mu\text{g g}^{-1}$  of soil) was recorded in without Zn application.

Our results are also concurrent with the findings of Rama Lakshmi *et al.*, (2014). However, the majority of the acid phosphatase is mostly of plant and associated fungal origin. Whereas, the alkaline phosphatase is more likely to be of microbial origin. The acid phosphatase activity would be more dependent upon the nutritional status of the plant (Naseby *et al.*, 2000) [20]. Further, the results emerged out indicated that the soil alkaline phosphatase and acid phosphatase enzyme activity was improved with RDF + *Bacillus megaterium* after harvest of soybean Kumar and Ismail (2016) [21]. Different crop growth stages showed different amount of acid phosphatase activity, it might be due to secretion of root exudates and biochemical changes in plant system (Dotaniya *et al.*, 2014) [17]. Similarly, Our results are also concurrent with the findings on the acid phosphatase activity revealed that there was no significant difference among different zinc solubilizing bacterial isolates, however, it was recorded in the range of 43.73 to 52.39  $\mu\text{g of PNP released g soil hr}^{-1}$  Vidyashree (2015) [22]. Further, Srilatha *et al.* (2013) [23] observed that soil enzyme activities increased with increasing rate of NPK application. Acid phosphatase ranged from 64.3 to 90.3, 77.3 to 127.9, 67.6 to 121.3 and 48.8 to 78.1 during *kharif* and 72.7 to 120.6, 169.8 to 206.1, 86.1 to 138.7 and 65.6 to 100.5  $\mu\text{g of p-nitrophenol released g}^{-1} \text{ soil h}^{-1}$  at 30, 60, 90 DAT and harvest respectively during *rabi*. Similarly, Apoorva (2016) [19] reported that the highest acid phosphatase activity was observed in the treatment receiving RDF+ Soil application of biozinc@30 kg ha<sup>-1</sup> (135.2  $\mu\text{g of p-nitrophenol g}^{-1} \text{ soil hr}^{-1}$ ) which was on par with RDF+ Soil application of ZnSO<sub>4</sub> @ 25 kg ha<sup>-1</sup> (129.8  $\mu\text{g of p-nitrophenol g}^{-1} \text{ soil hr}^{-1}$ ). The lowest acid phosphatase activity was observed in control (72.3  $\mu\text{g of p-nitrophenol g}^{-1} \text{ soil hr}^{-1}$ ) which was on par with RDF+ foliar application of 0.2% ZnSO<sub>4</sub> spray (72.3  $\mu\text{g of p-nitrophenol g}^{-1} \text{ soil hr}^{-1}$ ).

#### Alkaline phosphatase activity in soils

The data pertaining to alkaline phosphatase enzyme activity is narrated in Table 2. Which was significantly affected due to the inoculation of different ZnS microbial cultures. Zinc solubilizers influenced the alkaline phosphatase activity which ranged from (68.75 to 77.67  $\mu\text{g g}^{-1}$  of soil.) showing significantly higher acid phosphatase activity in *Pseudomona striata* treated plots follows by *Bacillus megaterium* and *Trichoderma viride*. Whereas, significantly lower acid phosphatase activity per plot were noted in control plots. Similarly, graded levels of zinc in the form of zinc sulphate also increase the alkaline phosphatase activity with each incremental dose up to 30 kg ZnSO<sub>4</sub> kg ha<sup>-1</sup>. The acid phosphatase activity as influenced by Zn application ranged from 67.58 (Zn0) to 76.67  $\mu\text{g g}^{-1}$  of soil (ZnSO<sub>4</sub>) 30 kg ha<sup>-1</sup>.

Interaction effect of zinc solubilizers and zinc levels on alkaline phosphatase activity in Table 2a. Increase the alkaline phosphatase activity significantly synergistic effect of each factor was recorded on each other showing significantly the highest alkaline phosphatase activity was noticed under in *Pseudomona striata* X ZnSO<sub>4</sub> 30 kg ha<sup>-1</sup> (82.33  $\mu\text{g g}^{-1}$  of soil) it was found at par with *Bacillus megaterium* and *Trichoderma viride* X ZnSO<sub>4</sub> 30 kg ha<sup>-1</sup>. However the lower values of alkaline phosphatase (66.33  $\mu\text{g g}^{-1}$  of soil) was recorded in without Zn.

The activities of both acid and alkaline phosphatases were significantly improved over control levels in the rhizosphere upon inoculation. This might be due to increased microbial and root activities as previously reported by Kaleeswari, (2007) Further Kumar and Ismail (2016) observed that alkaline

phosphatase activity was noted significantly highest in treatment RDF + *Rhizobium* + *Bacillus megaterium* over rest of treatments. It was closely at par with RDF + *Rhizobium* + *Pseudomonas fluorescens*. Whereas, acid phosphatase activity was also noted maximum in RDF + *Bacillus megaterium*. But it was found at par with the treatment RDF + *Pseudomonas fluorescens*, RDF + *Pseudomonas striata* and RDF + *Trichoderma viride*. Similarly, Badda *et al.*, (2013) who observed the highest increment in alkaline phosphatase activity in the plants inoculated with *A. laevis* + *T. viride* + *P. fluorescens*. Further, Vidyashree (2015) [22] observed that Maximum alkaline phosphatase activity was observed in the treatment inoculated with *B. aryabhatai* (68.9  $\mu\text{g}$  of PNP released/g soil/hour) which is significantly different from other treatments. Least activity of alkaline phosphatase was found in uninoculated control (47.6  $\mu\text{g}$  of PNP released/g soil/hour). Similarly Srilatha *et al.* (2013) [23] observed that soil enzyme activities increased with increasing rate of NPK application. Alkaline phosphatase activity ranged from 73.5 to 94.8, 81.8 to 135.2, 70.2 to 125.9, 52.8 to 92.6 in *kharif* and

81.7 to 126.1, 127.9 to 177.4, 85.6 to 151.4 and 69.1 to 109.4  $\mu\text{g}$  of *p*-nitrophenol released  $\text{g}^{-1}$  soil  $\text{h}^{-1}$  at 30, 60, 90 DAT and harvest respectively in *rabi*. Thereafter, Apoorva (2016) [19] reported that the highest alkaline phosphatase activity was observed in the treatment receiving RDF+ Soil application of biozinc @ 30  $\text{kg ha}^{-1}$  (206.1  $\mu\text{g}$  of *p*-nitrophenol  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$ ) which was on par with RDF+ Soil application of  $\text{ZnSO}_4$  @ 25  $\text{kg ha}^{-1}$  (190.0  $\mu\text{g}$  of *p*-nitrophenol  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$ ), RDF+ Soil application of bio zinc @ 15  $\text{kg ha}^{-1}$  (187.4  $\mu\text{g}$  of *p*-nitrophenol  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$ ), RDF+ Soil application of nano zinc @ 15  $\text{kg ha}^{-1}$  (176.0  $\mu\text{g}$  of *p*-nitrophenol  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$ ). The lowest acid phosphatase activity was observed in control (87.0  $\mu\text{g}$  of *p*-nitrophenol  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$ ) which was on par with RDF+ foliar application of 0.2%  $\text{ZnSO}_4$  spray (90.0  $\mu\text{g}$  of *p*-nitrophenol  $\text{g}^{-1}$  soil  $\text{hr}^{-1}$ ). Thereafter, Prasad and Ismail (2016) [24] showed that the enzymatic activity in soil after harvest of groundnut crop was influenced by different ZnS microbial inoculants. Activity of alkaline phosphatase was noted significantly highest in treatment RDF + *Rhizobium* + *Bacillus megaterium* (76.38  $\mu\text{g g}^{-1}$ ).

**Table 2a:** Interaction effect of zinc solubilizers and graded level of zinc on enzymatic activity in soil

Treatments	Zn0: $\text{ZnSO}_4$ 0 $\text{kg ha}^{-1}$	Zn1: $\text{ZnSO}_4$ 10 $\text{kg ha}^{-1}$	Zn2: $\text{ZnSO}_4$ 20 $\text{kg ha}^{-1}$	Zn3: $\text{ZnSO}_4$ 30 $\text{kg ha}^{-1}$
<b>Soil dehydrogenase (<math>\mu\text{g g}^{-1}</math> soil)</b>				
S0: Control	44.00	47.67	51.33	55.00
S1: <i>Pseudomonas striata</i>	50.67	58.00	65.33	67.33
S2: <i>Bacillus megaterium</i>	50.67	54.84	62.00	66.00
S3: <i>Trichoderma viride</i>	49.33	53.33	47.27	51.56
Interaction	S	Zn	SXZn	
SEm+	1.22	1.22	2.45	
CD at 5%	3.54	3.54	7.08	
<b>Acid phosphatase (<math>\mu\text{g g}^{-1}</math> soil)</b>				
S0: Control	42.00	44.00	48.00	52.67
S1: <i>Pseudomonas striata</i>	51.00	57.00	58.67	59.67
S2: <i>Bacillus megaterium</i>	48.67	51.33	53.33	57.30
S3: <i>Trichoderma viride</i>	46.33	49.00	51.08	53.16
Interaction	S	Zn	SXZn	
SEm+	0.39	0.39	0.78	
CD at 5%	1.13	1.13	2.27	
<b>Alkaline phosphatase (<math>\mu\text{g g}^{-1}</math> soil)</b>				
S0: Control	66.33	68.00	69.33	71.33
S1: <i>Pseudomonas striata</i>	70.00	77.67	80.67	82.33
S2: <i>Bacillus megaterium</i>	68.00	72.00	76.67	78.67
S3: <i>Trichoderma viride</i>	66.00	70.33	72.33	74.33
Interaction	S	Zn	SXZn	
SEm+	0.40	0.40	0.80	
CD at 5%	1.15	1.15	2.31	
<b>Urease (<math>\mu\text{g g}^{-1}</math> soil)</b>				
S0: Control	69.33	72.67	76.33	79.67
S1: <i>Pseudomonas striata</i>	79.00	82.67	86.33	90.00
S2: <i>Bacillus megaterium</i>	74.33	78.33	84.00	87.67
S3: <i>Trichoderma viride</i>	72.67	75.00	78.00	78.33
Interaction	S	Zn	SXZn	
SEm+	0.47	0.47	0.95	
CD at 5%	1.37	1.37	2.75	

### Urease activity in soils

The data clearly demonstrates a remarkable urease activity increased by the application of zinc solubilizing microbial inoculants compared to without application of inoculants. The data presented in Table 2. Shows zinc solubilizers influenced the urease activity which ranged from (74.50 to 84.50  $\mu\text{g g}^{-1}$  of soil.) showing significantly higher urease activity in *Pseudomonas striata* treated plots followed by *Bacillus megaterium* and *Trichoderma viride*. Whereas, significantly lower urease activity per plot noted in uninoculated control.

Similarly, graded levels of zinc in the form of zinc sulphate also increase the urease activity with each incremental dose of zinc up to 30  $\text{kg ZnSO}_4 \text{ kg ha}^{-1}$ . The urease activity as influenced by Zn application ranged between 73.83 (Zn0) to 83.92  $\mu\text{g g}^{-1}$  of soil ( $\text{ZnSO}_4$ ) 30  $\text{kg ha}^{-1}$ .

Interaction effect of zinc solubilizers in to zinc levels in Table 2a. Increase the urease activity significantly synergistic effect of each factor was recorded. Showing significantly the highest urease activity was noticed under in *Pseudomonas striata* X  $\text{ZnSO}_4$  30  $\text{kg ha}^{-1}$  (90.00  $\mu\text{g g}^{-1}$  of soil) it was found at par

with *Bacillus megaterium* in to ( $\text{ZnSO}_4$  30 kg ha<sup>-1</sup>). However the lower values of urease (69.33  $\mu\text{g g}^{-1}$  of soil) was recorded in Zn0 X S0 uninoculated control.

Rai and Yadav (2011)<sup>[25]</sup> reported that Application of RDF + FYM @ 10 tonnes ha<sup>-1</sup> recorded significantly highest urease activity (45.15 and 47.90 mg NH<sub>4</sub>N kg/24 hr) than any other treatments. Similarly, slight increase in the urease activity was observed at 70 DAS of sorghum as compared to 40 DAS, may be due to solubilization of complex organic compounds at steady rate. The increase in urease activity under RDF and RDF + FYM as compared to other treatments may be due to higher root biomass as a result of vigorous growth, thereby increasing urease activity in these treatments. The increased microbial population under organics and INM was responsible for increase in urease activity of the soil; this can be ascribed to the organic manure which acts as a source of carbon and energy for heterotrophs and provides sufficient nutrition for the proliferation of microbes and their activities in terms of soil enzymes. Thereafter, Vidyashree (2016)<sup>[22]</sup> reported that significantly higher urease activity (24.80  $\mu\text{g NH}_4\text{-N/g soil/hour}$ ) was recorded in *B. aryabhatai* inoculated treatment which was followed by *Bacillus* sp. (PAN-TM1) (21.40  $\mu\text{g NH}_4\text{-N/g soil/hour}$ ).

### Conclusion

From the study, it can be concluded that, the incremental levels of zinc solublizers and ZnSO<sub>4</sub> caused significant results show that, inoculation of *Pseudomona striata*, *Bacillus megaterium* and *Trichoderma viride* also increases yield and enzymatic activity. Similarly, graded levels of zinc in the form of zinc sulphate also increased with each incremental dose up to 30 kg ZnSO<sub>4</sub> kg ha<sup>-1</sup>.

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