International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
IJCS 2019; 7(3): 24-27

Received: 14-03-2019
Accepted: 18-04-2019

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Effect of seed inoculation of Zinc and Iron solubilizers on soil properties of pH, EC, organic carbon and calcium carbonate at panicle initiation and harvest stage of wheat

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Abstract
A field experiment was conducted during the year 2015-16 at Post Graduate Institute Farm, Mahatma Phule Krishi Vidyapeeth, Rahuri, to study the "Effect of Seed Inoculation of Zinc and Iron Solubilizing Microorganisms on Yield and Nutrient Uptake by Wheat in Inceptisol''.
The experiment was laid out in randomized block design with three replications and seven treatments. There were various levels of zinc sulphate and iron sulphate and recommended dose of fertilizer (GRDF) with FYM, seed inoculation of zinc and iron solubilizers.
The soil pH, EC and organic carbon at panicle initiation stage of wheatwas not influenced by different treatments. However a decrease in $CaCO_3$ of soil was observed in the treatment with seed coating of iron and zinc solubilizers and doses of $FeSO_4$ and $ZnSO_4$. The lowest $CaCO_3$ content was observed in treatment T_7.
The decrease in $CaCO_3$ content may be attributed to the synthesis of organic acids by bacteria, fungi and formation of carbonic acid due to release of CO_2 by microbes and formation of mineral acid (H_2SO_4) due to dissociation of sulphate (SO_4^{2-}).
The same trend of pH, EC, organic carbon and calcium carbonatestatus was observed at harvest stage with a slight decrease in pH, EC, organic carbon and calcium carbonate.

Keywords: wheat, zinc and iron solubilizng microorganisms, pH, EC, $CaCO_3$ and organic carbon

Introduction

Wheat (*Triticum aestivum*) is the second most important cereal crop in India next to rice in respect of area and production. In 2016, global wheat production was 749 million tonnes. Wheat is the primary food staple in North Africa and the Middle East, and is growing in uses in Asia. Unlike rice, wheat production is more widespread globally, though 47% of the world total in 2014 was produced by just four countries – China, India, Russia and the United States. In India, area under wheat cropping in 2015-16 was 29.25 million hectares with the annual production of 85.93 million tones with average productivity of 2938 kg ha^{-1}. In Maharashtra, wheat occupied 1.08 million hectare and annual production was 1.74 million tones with average productivity of 1483 $kgha^{-1}$ (Anonymous, 2015) [1]. The average productivity of wheat in Maharashtra is quite low. Therefore, it is very essential to increase the production and productivity of wheat in the state. The deficiencies of micronutrients (Zn and Fe) have been increasing on many agricultural soils. It can be grown in tropics, sub tropics and temperate region. Wheat is cultivated in alluvial soil and black cotton soils. Wheat is an important source of carbohydrate, proteins and minerals like P, Mg, Fe, Cu and Zn and vitamins like thiamine, riboflavin, niacin and Vitamin E. The micronutrient deficiencies have been verified in many soils through soil testing and plant analysis. The application of micronutrient fertilizers have proved better in many agricultural crops *viz*., wheat, maize, rice etc.

Zinc is one of the most important micronutrients. It has vital role in transformation of carbohydrates, regulation of consumption of sugar and increase source of energy for the production of chlorophyll. Zinc is also required for maintenance of auxin in an active state. Zinc is essential for the synthesis of tryptophan, a precursor of auxin. The basic function of zinc in plants relates to metabolism of carbohydrate, protein and phosphate, auxin and ribosome formation. The intensive cropping, imbalanced fertilization, non-use of micronutrients and inadequate supply of organic manures have resulted in the depletion of soil fertility. Iron is involved in the production of chlorophyll and iron chlorosis is easily recognized on iron sensitive crops growing on calcareous soil. Iron also is a component of many enzymes associated with energy transfer, nitrogen reduction and fixation and lignin formation. Iron is associated with sulphur in plants to form compounds that catalyse other reactions. Iron deficiencies are mainly manifested by yellow leaves due to low levels of chlorophyll. Leaf yellowing first appears on the younger upper leaves in interveinal tissues. Severe iron deficiencies cause leaves to turn completely yellow or almost white and then brown as leaves die. Iron deficiencies are found mainly on high pH soil, although some acid, sandy soil low in organic matter also may be iron deficient. Cool, wet weather enhances iron deficiencies, especially on soil with marginal level of available iron. Poorly aerated or compacted soil also reduce iron uptake by plants, uptake of iron decreases with increase in soil pH and is adversely affected by high level of available phosphorus, manganese and zinc in soil. Wheat is the crop species which is most susceptible to zinc deficiency. About 96 to 99 percent of the applied zinc and iron is concerted to different insoluble forms depending upon the soil types, physico-chemical reactions of the soil. The solubility of zinc and iron is highly dependent on soil pH and moisture. Zinc occurs in soil as sphalerite, olivine, hornblende, augite and biotite. Adoption of recommended package of practices is a need of the day. Macro and micronutrients play a vital role in the physiology of plants. The application of micronutrient either foliar or through soil is very essential for higher production and quality improvement of wheat. Amongst the micronutrients, iron and zinc have recently assumed greater importance in crop production. The information on seed coating of iron and zinc solubilizing microorganisms to solubilize the soil mineral zinc and iron is very scanty and staggered.

Materials and Methods

Materials

1. Experimental site

The experimental plot belonging to Inceptisol order, deficient in Zn and Fe and low status of organic carbon content was selected for conduct of experiment.

2. Soil

Composite soil sample from the experimental site was collected and processed for analysis of soil properties and fertility. After collection soil, the soil was air dried under diffused sunlight and processed for initial chemical properties.

3. FYM

Well decomposed farmyard manure was procured from cattle project, M.P.K.V., Rahuri and applied as per recommendation @ 10 t ha^{-1}.

4. Seed inoculation

The Fe-Zn solubilizing culture required for seed coating for this experiment, was brought from the Vasantdada Sugar Institute, Manjari, Dist. Pune. The culture consisted of a consortium of zinc and iron solubilizing bacteria and fungi. The zinc solubilizers included a consortium of bacterial strains viz. *Bacillus Polymyxa, Bacillus megaterium, Pseudo Monasstriata, Psuedomonasfluroscense, Gluconacetobacter Diazotrophicus* and *Aspergillus Awamori Ea* fungal strain. The iron solubilizing microorganisms included bacterial strains viz. *Thiobacillus Thiooxidans*, *Thiobacillus Ferrooxidans* and *Aspergillus Niger* and *Trichoderma viridae*, which are the fungal strains. This consortium of iron and zinc solubilizing organisms were used for wheat seed inoculation. The iron and zinc solubilizing culture was inoculated @ 250 ml per 5 kg wheat seed in a bowl. This seed was partially dried in shade and was again treated with Azotobactor @ 250 g per 5 kg seed.

5. Experimental details

1. Location : Department of Soil Science and Agril. Chemistry, PGI Farm, M.P.K.V., Rahuri.
2. Crop : Wheat
3. Soil type : Inceptisol (deficient in Zn and Fe)
4. Season : *Rabi-2015-16*
5. Variety : Trimbak
6. Treatments : 7
7. Replications : 3
8. Design : RBD
9. Spacing : 22.5 cm
10. Plot size : Gross: 3.60X 4.50 m.
Net: 3.15 X 4.10 m.

6. Treatments details

T_1 : Absolute control
T_2 : Absolute control + seed treatment of Zn and Fe solubilizers
T_3 : GRDF only (120:60:40 kg ha^{-1} N, P_2O_5 and K_2O +10 t ha^{-1} FYM)
T_4 : GRDF + seed treatment of Zn and Fe solubilizers
T_5 : GRDF + 5 kg ha^{-1} $ZnSO_4$ + 10 kg ha^{-1} $FeSO_4$ + Zn and Fe solubilizers
T_6 : GRDF+ 10 kg ha^{-1} $ZnSO_4$+ 15 kg ha^{-1} $FeSO_4$ + Zn and Fe solubilizers
T_7 : GRDF + 20 kg ha^{-1} $ZnSO_4$ + 25 kg ha^{-1} $FeSO_4$ + Zn and Fe solubilizers

Note: Half of N, total P_2O_5 and K_2O was applied the time of sowing; remaining half of N was given at 30 DAS

Methods

1. Soil analysis

Representative surface soil samples of the experimental site were collected up to 22.5 cm depth at panicle initiation and harvesting stage of wheat from each plot to know the fertility status of soil. The collected soil samples were air dried under shade, grinded in wooden mortar and pestle, sieved through 2 mm sieve and were analyzed for pH, EC, $CaCO_3$,

Table 1: Standard methods for analysis of soil and plant samples

Sr. No.	Parameter	Method used	Reference
I.	Soil analysis		
1.	pH (1:2.5)	Potentiometric	Jackson (1973)
2.	EC (1:2.5)	Conductometric	Jackson (1973)
3.	$CaCO_3$ content (%)	Acid neutralization	Piper (1966)
4.	Organic carbon	Walkely and Black, Wet oxidation method	Nelson and Sommer (1982)

Results and Discussion

1. Effect of seed inoculation of Zn and Fe solubilizers on soil pH, EC, organic carbon and calcium carbonate at panicle initiation stage of wheat

1.1 pH

A slight decrease in soil pH (Table 2) was observed in all the treatments with seed inoculation of zinc and iron solubilizers and treatments of $ZnSO_4$ and $FeSO_4$.However, this decrease was statistically non-significant between the treatments. This decrease in soil pH at panicle initiation stage may be ascribed to synthesis of organic acids by the consortium of Fe and Zn solubilizing fungi and bacteria. These observations are in close confirmity with the findings of El-Azouni (2008) [2].

1.2 EC

The soil electrical conductivity (Table 2) at panicle initiation stage of wheat was found to increase in the treatments T_3 to T_7. The slight increase in EC may be due to addition of chemical fertilizer and dissolution of fixed phosphate and iron and zinc in the soil. However, the increase in EC was observed to be non significant. Hati *et al*. (2007) also observed an increase in electrical conductivity due to seed inoculation of PSM.

1.3 Organic carbon

The per cent organic carbon (Table 2) of soil at panicle initiation stage of wheat was not much influenced by all the treatments, except for a slight increase in the seed inoculation treatments. The results were non significant. Similar results were reported by Yadav and Singh (1991) [11].

1.4 Calcium carbonate

The $CaCO_3$ content of the soil at panicle initiation stage of wheat was found to decrease in all the treatments of seed inoculation of zinc and iron solubilizers (T_2, T_4, T_5, T_6 and T_7). The lowest $CaCO_3$ content (6.83%) of the soil was observed in treatmentT_7. This decrease of $CaCO_3$was significantly lowest than all other treatments. The decrease in $CaCO_3$ in T_4, T_5 and T_6 was significantly lower than T_3, T_2 and T_1.The decrease in $CaCO_3$content may be attributed to the synthesis of organic acids by bacteria and fungi, formation of carbonic acid due to CO_2 release by microbes and formation of mineral acids (H_2SO_4) due to dissociation of sulphate (SO_4^{--}). These observations are close in confirmity with the findings reported by Said (2011) [8].

2. Effect of seed inoculation of Zn and Fe solubilizers on soil pH, EC, organic carbon and calcium carbonate at harvest stage of wheat

2.1 pH

A slight decrease in soil pH was observed in all the treatments with seed inoculation of zinc and iron solubilizers and treatments of $ZnSO_4$ and $FeSO_4$. However, this decrease was statistically at par within the treatments. The decrease in soil pH in T_6 and T_7 was significant over the initial soil pH (8.28). This decrease in soil pH at harvest stage may be ascribed to synthesis of organic acids by the consortium of Fe and Zn solubilizing fungi and bacteria. Similar findings were reported by Katkar *et al*. (2002) [5].

2.2 EC

The soil electrical conductivity (Table 3) at harvest stage was found to increase in the treatments T_3 to T_7.The slight increase in EC may be due to addition of chemical fertilizer and dissolution of fixed phosphate and iron and zinc in the soil.

2.3 Organic carbon

The organic carbon (Table 3) of soil at harvest stage of wheat was not much influenced by all the treatments except for a slight increase in the seed inoculation treatments. However, the results were non-significant. Similar findings were reported by Vikram *et al*., (2007) [10].

2.4 Calcium carbonate

The $CaCO_3$ content of the soil at harvest stage was found to decrease in all the treatments of seed inoculation of zinc and iron solubilizers (T_4, T_5, T_6 and T_7).The lowest $CaCO_3$ content of the soil in treatment T_7 (6.17 %). This decrease of $CaCO_3$was significantly lowest than all other treatments. The decrease in $CaCO_3$in T_4, T_5 and T_6 was significantly lower than T_3 and T_1.The decrease in $CaCO_3$content may be attributed to the synthesis of organic acids by bacteria and fungi formation of carbonic acid due to CO_2 release by microbes and formation of mineral acids (H_2SO_4) due to dissociation of sulphate (SO_4^{2-}). These observations were in close confirmity with Shinde (2009) [9].

3. Summary

3.1 Effect of seed inoculation of Zn and Fe solubilizers on pH, EC and organic carbon at panicle initiation and harvest stage wheat.

The soil pH, EC and organic carbon at panicle initiation stage of wheat was not influenced by different treatments. However a decrease in $CaCO_3$ of soil was observed in the treatment with seed coating of iron and zinc solubilizers and doses of $FeSO_4$ and $ZnSO_4$. The lowest $CaCO_3$ content was observed in treatment T_7.

The decrease in $CaCO_3$ content may be attributed to the synthesis of organic acids by bacteria, fungi and formation of carbonic acid due to release of CO_2 by microbes and formation of mineral acid (H_2SO_4) due to dissociation of sulphate (SO_4^{2-}).

The same trend of pH, EC, organic carbon and calcium carbonate status was observed at harvest stage with a slight decrease in pH, EC, organic carbon and calcium carbonate.

Table 2: Effect of seed inoculation of Zn and Fe solubilizers on soil pH, EC, organic carbon and calcium carbonate at panicle initiation stage of wheat

Tr. No.	Treatment	pH (1:2.5)	EC (dSm^{-1})	Org. C (%)	$CaCO_3$ (%)
T_1	Absolute control	8.24	0.20	0.50	8.08
T_2	Absolute control + seed treatment of Zn and Fe solubilizers	8.26	0.21	0.49	7.83
T_3	GRDF only (120:60:40 kg ha^{-1} N, P_2O_5& K_2O +10 t ha^{-1} FYM)	8.25	0.24	0.51	8.75
T_4	GRDF + seed treatment of Zn and Fe solubilizers	8.23	0.25	0.52	8.33
T_5	GRDF + 5 kg ha^{-1} $ZnSO_4$ + 10 kg ha^{-1} $FeSO_4$ +Zn and Fe solubilizers	8.22	0.25	0.52	7.67
T_6	GRDF + 10 kg ha^{-1} $ZnSO_4$ + 15 kg ha^{-1} $FeSO_4$ + Zn and Fe solubilizers	8.22	0.27	0.53	7.17
T_7	GRDF + 20 kg ha^{-1} $ZnSO_4$ + 25 kg ha^{-1} $FeSO_4$ + Zn and Fe solubilizers	8.21	0.27	0.53	6.83
Initial		8.28	0.22	0.51	8.25
SE$\pm$		0.010	0.017	0.073	0.164
CD at 5%		NS	NS	NS	0.507

Table 3: Effect of seed inoculation of Zn and Fe solubilizers on soil pH, EC, organic carbon and calcium carbonate at harvest stage of wheat

Tr. No.	Treatment	pH (1:2.5)	EC(dSm^{-1})	Org. C (%)	$CaCO_3$ (%)
T_1	Absolute control	8.23	0.19	0.49	7.58
T_2	Absolute control + seed treatment of Zn and Fe solubilizers	8.25	0.20	0.48	6.83
T_3	GRDF only (120:60:40 kg ha^{-1} N, P_2O_5& K_2O +10 t ha^{-1} FYM)	8.24	0.23	0.51	8.17
T_4	GRDF + seed treatment of Zn and Fe solubilizers	8.23	0.24	0.50	7.83
T_5	GRDF + 5 kg ha^{-1} $ZnSO_4$ + 10 kg ha^{-1} $FeSO_4$ +Zn and Fe solubilizers	8.21	0.26	0.50	7.25
T_6	GRDF + 10 kg ha^{-1} $ZnSO_4$ + 15 kg ha^{-1} $FeSO_4$ + Zn and Fe solubilizers	8.19	0.25	0.51	6.83
T_7	GRDF + 20 kg ha^{-1} $ZnSO_4$ + 25 kg ha^{-1} $FeSO_4$ + Zn and Fe solubilizers	8.19	0.26	0.51	6.17
Initial		8.28	0.22	0.51	8.25
SE±		0.010	0.008	0.018	0.243
CD at 5%		0.031	0.027	NS	0.750

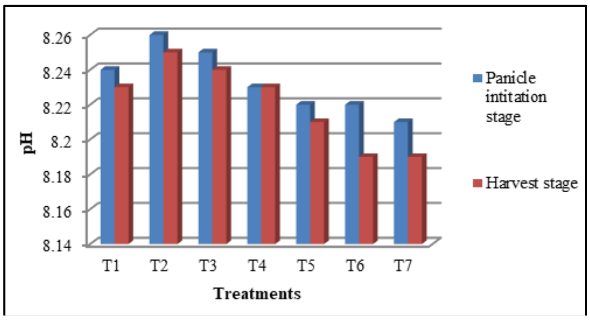

Fig 1: Effect of seed inoculation of Zn and Fe solubilizers on soil pH at panicle initiation and harvest stage of wheat

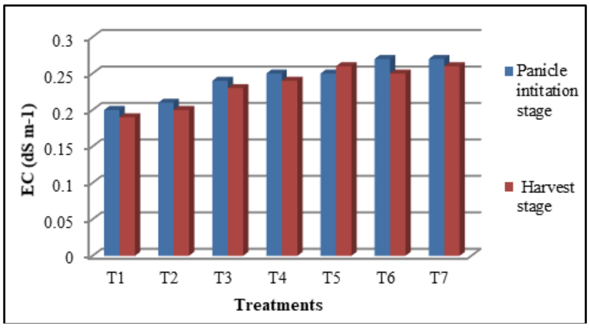

Fig 2: Effect of seed inoculation of Zn and Fe solubilizers on Soil EC at panicle initiation and harvest stage of wheat

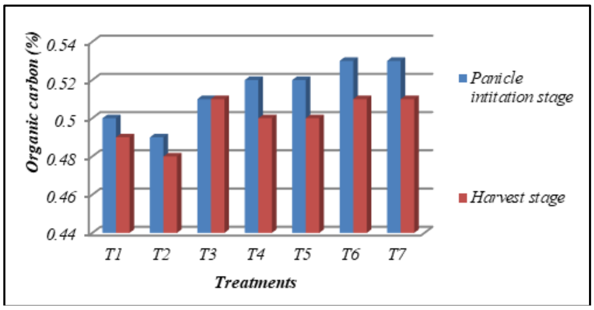

Fig 3: Effect of seed inoculation of Zn and Fe solubilizers on soil Organic carbon at panicle initiation and harvest stage of wheat

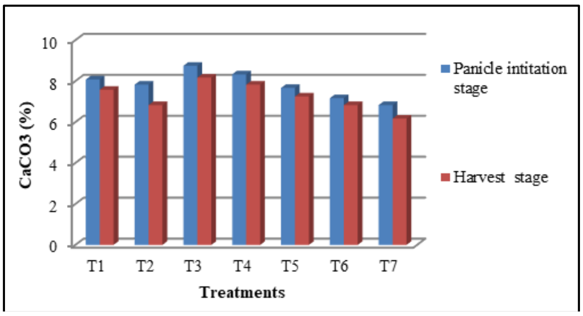

Fig 4: Effect of seed inoculation of Zn and Fe solubilizers on Soil calcium carbonate at panicle initiation and Harvest stage of wheat

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