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Effect of Iron, Zinc and Microbial Inoculants on Soil Health and Nutrient Uptake of Urdbean (*Vigna mungo* L.) in Vertisols

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

An experiment was conducted on urdbean during *kharif* 2021 at Instructional Farm, College of Agriculture, Ummedganj, Kota (Rajasthan). The experiment comprised 10 treatments viz. (Control, 75 % RDF, 100 % RDF, 75 % RDF + *Rhizobium* @ 600 g ha^{-1} seed inoculation, 75 % RDF + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation, 75 % RDF + *Rhizobium* @ 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1}, 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage, 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Rhizobium* @ 600 g ha^{-1}, 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Bacillus megaterium* @ 600 g ha^{-1}, 75% RDF + 0.1 % $FeSO_4$ + 0.5 %

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$ZnSO_4$ at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1}) was carried out in randomized block design with three replications. Significantly higher nitrogen (3.87 and 1.84 %), phosphorus (0.34 and 0.154 %), potassium (0.56 and 1.82 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}) content in seed and straw of urdbean were recorded with application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation over rest of treatments. The significantly higher uptake of nitrogen (49.36, 27.84 and 77.20 kg ha^{-1}), phosphorus (4.32, 2.33 and 6.65 kg ha^{-1}), potassium (7.16, 27.44 and 34.60 kg ha^{-1}), Fe (573.25 ,1120.36 and 1693.60 g ha^{-1}) and Zn (413.78, 297.40 and 711.18 g ha^{-1}) by seed, straw and total by urdbean crop with application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation over rest of treatments. Application of Fe, Zn foliar spray and bio fertilizers seed inoculation did not significantly influence soil physico-chemicals properties during the experimentation. The maximum available nitrogen (285.21 kg ha^{-1}), phosphorus (25.41 kg ha^{-1}), potassium (252.66 kg ha^{-1}), Fe (5.75 mg kg^{-1}) and Zn (0.76 mg kg^{-1}) after urdbean crop harvest in soil were recorded with application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation over rest of treatments.

Keywords: Bio-fertilizer; nutrient content; nutrient uptake; Fe; urdbean and Zn.

1. INTRODUCTION

Pulses are the integral part of Indian dietary system due to richest source of protein and other nutrients. Indian population is predominantly vegetarian and protein requirement for the growth and development of the human being is mostly met with pulses. The availability of pulses per capita per day has proportionately declined from 71 g to 52 g against the minimum requirement of 70 g day^{-1}/capita [1]. Pulses are improving soil fertility by atmospheric nitrogen fixation through *Rhizobium culture*. In India pulses are grown an area of 28.34 million ha with total production of 23.15 million tonnes and productivity of 817 kg ha^{-1} [2]. Among the pulses, urdbean [*Vigna mungo* (L.) Hepper], is an important crop in India. Urdbean is the third most important pulse crop after chickpea and pigeonpea in India. It is highly nutritious containing 24-26% protein, 1.3% fat and 60% carbohydrates on dry weight basis and it is rich source of calcium, iron and vitamins [3].

The major constraints for low yield of urdbean are lack of micronutrient application and non-adoption of proper agronomic practices. Micronutrients are equally important in plant nutrition as major nutrients. [4] The incidence of micronutrient deficiencies in crops has increased markedly in recent years due to intensive cropping, loss of top soil by erosion, losses of micronutrients through leaching, liming of acid soils, decreased proportions of farmyard manure or other organic sources compared to chemical fertilizers and use of marginal lands for crop production [5]. The optimum supply of micronutrients under balanced condition is very important for achieving higher productivity of urdbean. Foliar application of micronutrients has been proved to be an important asset in fertilizer application with a specific aim of increasing nutrient availability at the time of need, especially at the later stage of plant growth. Foliar application of nutrients using water soluble fertilizers is one of the possible ways to enhance the productivity of pulses. The foliar fertilization of Zn and Fe at flowering stage improves growth and yield urdbean [6].

Bio-fertilizer plays a vital role in maintaining long term soil fertility and sustainability [7]. Incorporation of FYM alone or bio-fertilizers improves available nutrient status of the soil with enhanced soil biological activity which in turn provides a congenial physical condition and improved availability of nutrient in the rhizosphere, resulting in an improvement in the crop growth and providing a better source–sink relationship [8]. Among bio-fertilizers, *Rhizobium* inoculation is cheapest, easiest and safest method of supplying nitrogen to legumes through well-known symbiotic nitrogen fixation process. Inoculation of appropriate strain enhances nodule formation resulting better nitrogen fixation. *Rhizobium* species in association with plant roots in urdbean improved soil fertility by fixing atmospheric nitrogen and produces plant growth substances in the soil. *Rhizobium* inoculation can increase the grain yield of pulse crops [9].

2. MATERIALS AND METHODS

A field experiment was conducted on urdbean during *kharif* 2021 at Instructional Farm, College of Agriculture, Ummedganj, Kota (Rajasthan). Geographically, is situated at 25.11^0 North latitude and 75.50^0 East longitude at an altitude of 258 meters above mean sea level. In Rajasthan, this region falls under the Agro-Climatic Zone-V (Humid South Eastern Plain Zone). This zone possesses typical sub-tropical conditions with maximum temperature range in summer is 42.2- 43.0 ^{0}C and minimum 12 - 27^{0}C. In this zone annual average rainfall is received 1275 mm. The soil of experimental site was clay loam in texture, slightly saline in reaction, medium in available nitrogen (217.35 kg ha^{-1}) and phosphorus (19.82 kg ha^{-1}) while high in potassium (236.31kg ha^{-1}) and sufficient in DTPA extractable micronutrients with pH (7.60) and EC (0.49 dS m^{-1}). [5] Source of nutrients were applied urea for nitrogen, DAP for phosphorus and mutate of potash for potassium. The full dose of fertilizer 100 % RDF (20:40:30 NPK kg ha^{-1}) was applied as basal dose. Before sowing, seed of urdbean was inoculated with liquid *rhizobium* culture @ 10 ml kg^{-1} seed and liquid *Bacillus megaterium* culture @ 10 ml kg^{-1} seed as per treatment. Foliar fertilization of $ZnSO_4$ and $FeSO_4$ were applied at pre flowering and pod formation stage. Spraying was done with knapsack sprayer and the leaves were wetted thoroughly with fine mist. The absorption of solution by urdbean leaves, a sticker was added in the spray solution.

The experiment laid out in randomized block design with three replications. The experiment comprised 10 treatments viz. (control, 75 % RDF, 100 % RDF, 75 % RDF + *Rhizobium* @ 600 g ha^{-1} seed inoculation, 75 % RDF + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation, 75 % RDF + *Rhizobium* @ 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1}, 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage, 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Rhizobium* @ 600 g ha^{-1}, 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Bacillus megaterium*@ 600 g ha^{-1}, 75% RDF + 0.1 % $FeSO_4$ + 0.5 %

Table 1. Soil physico-chemical properties analysis methods

S. No.	Particulars	Values	Method employed
A.	**Mechanical composition**		
i.	Sand (%)	23.80	Hydrometer method [11]
ii.	Silt (%)	36.00	
iii.	Clay (%)	40.20	
iv.	Textural class	Clay loam	
B.	**Physical properties**		
i.	Bulk density (Mg m^{-3})	1.34	Core sampler method, [12]
ii.	Particle density (Mg m^{-3})	2.39	Pycnometer or RD bottle method
iii.	Porosity (%)	43.94	Calculated by BD and PD
C.	**Chemical composition**		
i.	Organic carbon (%)	0.41	Determination by rapid titration method, [13]
ii.	Available nitrogen (kg ha^{-1})	217.35	Estimation by alkali potassium permanganate method [14]
iii.	Available phosphorus (kg ha^{-1})	19.82	Olsen's P, 0.5 M $NaHCO_3$ method, pH 8.5 [15]
iv.	Available potassium (kg ha^{-1})	236.31	Extraction was done with 1 N neutral ammonium acetate at pH 7.0 and determined by flame photometer [16]
v.	Available Zn (mg kg^{-1})	0.57	Extraction by 0.005 M DTPA + 0.01 M $CaCl_2$ + 0.1 M tri ethanol amine at pH 7.3 [17]
vi.	Available Fe (mg kg^{-1})	3.93	Extraction by 0.005 M DTPA + 0.01 M $CaCl_2$ + 0.1 M tri ethanol amine at pH 7.3 [17]
vii.	EC at 25^{0}C (dSm $^{-1}$)	0.49	Measured in soil: water solution (1:2) [18]
viii.	pH (1 :2 soil water suspension)	7.60	Glass electrode pH meter in soil: water solution (1:2) [11]

$ZnSO_4$ at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1}) was carried out in randomized block design with three replications. For recording pre and post-harvest observations, five plants were randomly selected for each plot and tagged with labels for various observations on seed and straw. Soil samples were recorded plot wise as per standard procedures and statistically analysed by adopting appropriate method of standard analysis of variance [10].

2.1 Chemical Analysis in Plant

Plant samples were randomly collected from labelled plants in each treatment. The samples were then cleaned with water, air dried and then dried in hot-air oven at 60 °C for 18 hours. The samples were oven dried, powdered and stored in polythene bags. These powdered samples were analysed for nitrogen, phosphorus, potassium, zinc and ferrous content. Nitrogen was determined by Kjeldahl distillation method. Plant sample (0.5 g) was digested in digestion flasks using sulphuric acid and digestion mixture. After complete digestion, the digested material was distilled in alkaline medium and the liberated ammonia was trapped in four per cent boric acid solution containing mixed indicator. The trapped ammonia was titrated against standard sulphuric acid [11]. Phosphorus content in the di-acid digested extract was estimated by Vanadomolybdo-phosphoric yellow colour method in nitric acid medium and the colour intensity was measured at 420 nm wave length [11]. Potassium in the plant sample was estimated by atomizing the diluted di-acid extract to a flame photometer [11]. After making suitable dilution of di-acid extract, the samples were fed to the atomic absorption spectrophotometer using appropriate hallow cathode lamps to estimate the Zn content in plant sample. After making suitable dilution of di-acid extract, the samples were fed to the atomic absorption spectrophotometer using appropriate hallow cathode lamps to estimate the Fe content in plant sample. Nitrogen, phosphorus and potassium uptake by seed and stover at harvest was calculated by using following formula:

Nutrient uptake by seed (Kg ha^{-1}) = Nutrient content in seed (%) x Seed Yield (Kg ha^{-1}) / 100

Nutrient uptake by straw (Kg ha^{-1}) = Nutrient content in straw (%) x Straw Yield (Kg ha^{-1}) / 100

Zinc and Iron uptake by plants seed and stover at harvest was calculated by using following formula:

Nutrient uptake by seed (mg ha^{-1}) = Nutrient content in seed (%) x Seed Yield (Kg ha^{-1}) / 100

Nutrient uptake by straw (mg ha^{-1}) = Nutrient content in straw (%) x Straw Yield (Kg ha^{-1}) / 100

2.2 Soil Analysis after Crop Harvest

Soil samples (0-15 cm) were collected from each plot after harvest of urdbean crop. These were dried and passed through 2 mm sieve and prepared for subsequent analysis as per standard methods.

3. RESULTS AND DISCUSSION

3.1 Nutrient Content and Uptake

The data pertaining to nutrient content and uptake of urdbean crop significantly influenced under application of Fe, Zn and Bio-fertilizers were presented in Table 2. Significantly higher nitrogen (3.87 and 1.84 %), phosphorus (0.34 and 0.154 %), potassium (0.56 and 1.82 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}) content in seed and straw of urdbean were recorded with application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation over control nitrogen (1.66 and 1.37 %), phosphorus (0.22 and 0.177 %), potassium (0.40 and 1.33 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}), 75 % RDF nitrogen (1.94 and 1.48 %), phosphorus (0.24 and 0.123 %), potassium (0.44 and 1.48 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}) ,100 % RDF nitrogen (2.45 and 1.57 %), phosphorus (0.25 and 0.129 %), potassium (0.47 and 1.57 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}), 75 % RDF + *Rhizobium* @ 600 g ha^{-1} seed inoculation nitrogen (2.49 and 1.59 %), phosphorus (0.27 and 0.133 %), potassium (0.48 and 1.60 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}), 75 % RDF + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation nitrogen (2.46 and 1.58 %), phosphorus (0.26 and 0.131%), potassium (0.47 and 1.57 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}), 75 % RDF +

Rhizobium @ 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1} nitrogen (2.53 and 1.70 %), phosphorus (0.28 and 0.141 %), potassium (0.51 and 1.67 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}), 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage nitrogen (2.52 and 1.63 %), phosphorus (0.29 and 0.134 %), potassium (0.50 and 1.62 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}), 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Rhizobium* @ 600 g ha^{-1} nitrogen (3.11 and 1.77 %), phosphorus (0.31 and 0.149 %), potassium (0.53 and 1.80 %), Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}) , 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Bacillus megaterium* @ 600 g ha^{-1} nitrogen (2.99 and 1.75 %), phosphorus (0.30 and 0.145 %), potassium (0.53 and 1.72 %) Fe (45.15 and 73.89 mg kg^{-1}) and Zn (32.61 and 19.69 mg kg^{-1}) content in seed and straw of urdbean.

The nutrient uptake by seed, straw and by urdbean crop were significantly influenced under application of Fe, Zn and Bio-fertilizers were presented in Table 3. The significantly higher uptake of nitrogen (49.36, 27.84 and 77.20 kg ha^{-1}), phosphorus (4.32, 2.33 and 6.65 kg ha^{-1}), potassium (7.16, 27.44 and 34.60 kg ha^{-1}), Fe (573.25 ,1120.36 and 1693.60 g ha^{-1}) and Zn (413.78, 297.40 and 711.18 g ha^{-1}) by seed, straw and total by urdbean crop with application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation over control nitrogen (10.04,10.99 and 21.03 kg ha^{-1}), phosphorus (1.33, 0.94 and 2.27 kg ha^{-1}), potassium (2.41,10.68 and 13.09 kg ha^{-1}), Fe (167.06,344.14 and 511.20 g ha^{-1}) and Zn (120.59, 95.23 and 215.82 g ha^{-1}), 75 % RDF nitrogen (14.26,13.76 and 28.02 kg ha^{-1}), phosphorus (1.76,1.44 and 2.90 kg ha^{-1}), potassium (3.25,13.76 and 17.0 kg ha^{-1}), Fe (227.51, 461.25 and 688.76 g ha^{-1}) and Zn (159.97, 125.72 and 285.69 g ha^{-1}) ,100 % RDF nitrogen (20.0, 16.13 and 36.13 kg ha^{-1}), phosphorus (2.04,1.32 and 3.37 kg ha^{-1}), potassium (3.82,16.01and 19.83 kg ha^{-1}), Fe (278.19, 550.66 and 828.85 g ha^{-1}) and Zn (194.90,152.17 and 347.07 g ha^{-1}), 75 % RDF + *Rhizobium* @ 600 g ha^{-1} seed inoculation nitrogen (21.39,17.43 and 38.83 kg ha^{-1}), phosphorus (2.33,1.46 and 3.79 kg ha^{-1}), potassium (4.15,17.47 and 21.63 kg ha^{-1}), Fe (310.48, 625.75 and 936.23 g ha^{-1}) and Zn (218.81,171.05 and 389.87 g ha^{-1}), 75 % RDF + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation nitrogen (21.05, 17.22 and 38.27 kg ha^{-1}), phosphorus (2.22,1.43 and 3.64 kg ha^{-1}), potassium (4.02,17.11and 21.13 kg ha^{-1}), Fe (303.01, 607.60 and 910.61 g ha^{-1}) and Zn (212.47, 168.28 and 380.74 g ha^{-1}), 75 % RDF + *Rhizobium* @ 600 g ha^{-1} + *PSB* @ 600 g ha^{-1} nitrogen (22.93,19.57 and 42.51kg ha^{-1}), phosphorus (2.53, 1.62 and 4.15 kg ha^{-1}), potassium (4.62,19.21and 23.83 kg ha^{-1}), Fe (336.86,685.66 and 1022.51 g ha^{-1}) and Zn (236.50,187.88 and 424.38 g ha^{-1}), 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage nitrogen (22.20,18.20 and 40.40 kg ha^{-1}), phosphorus (2.56,1.50 and 4.05 kg ha^{-1}), potassium (4.35, 18.03 and 22.38 kg ha^{-1}), Fe (325.38,643.15 and 968.53 g ha^{-1}) and Zn (226.17, 178.83 and 404.99 g ha^{-1}), 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Rhizobium* @ 600 g ha^{-1} nitrogen (31.26, 21.99 and 53.25 kg ha^{-1}), phosphorus (3.11,1.85 and 4.96 kg ha^{-1}), potassium (5.32,22.44 and 27.76 kg ha^{-1}), Fe (419.13,809.61 and 1228.75 g ha^{-1}) and Zn (304.10, 226.82 and 530.92 g ha^{-1}), 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Bacillus megaterium* @ 600 g ha^{-1} nitrogen (29.72,21.63 and 51.35 kg ha^{-1}), phosphorus (2.98, 1.79 and 4.77 kg ha^{-1}), potassium (5.23, 21.22 and 26.46 kg ha^{-1}) Fe (410.23,804.38 and 1214.62 g ha^{-1}) and Zn (295.57,223.41and 518.98 g ha^{-1}) uptake by seed, straw and total by urdbean crop.

Application of Fe, Zn and Bio fertilizers significantly influenced the nutrient content and their uptake. The increase in nutrient content might be due to increased availability of native nutrients. These results supported by the findings of Keram et al. [19], who stated that the increase could be attributed to the synergistic effect between nutrients and bio-fertilizers which might be due to increase enzymatic activity by different nutrient sources. It is an established fact that nutrient accumulation depends upon dry matter accumulation and concentration of nutrient at cellular level. These results are in agreement with those of Choudhary et al. [20] and Ranpariya and Polara [21]. Significant increase in protein content has been observed in the present investigation because of increased nitrogen content in seed which might be the result of increased availability of nitrogen to plants. An improved metabolism to greater translocation of these nutrients to reproductive organs of the crop and ultimately increased the content in seed and stover. These results are in close conformity with those of Rajkhowa et al. [22]. Significant increase in nutrient uptake due to application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation recorded under investigation. Since the nutrient uptake is a function of its content in crop plant and seed and straw yield of the crop. The increase in these parameters due to nitrogen and phosphorus fertilization with bio inoculants led to an increased uptake of nutrients in the present study. These results are in confirmity with the findings of Sesode [23] and Rathore et al. [24].

3.2 Post Harvest Soil Physico-Chemical Properties

Soil status after harvesting of urdbean were measured in terms of pH, EC (dS m^{-1}), organic carbon (%), bulk density (Mg m^{-3}), particle density (Mg m^{-3}) and porosity (%) are presented in Table 4. Soil pH after harvesting of urdbean crop did not find to show any significant variation with application of Fe, Zn and Bio-fertilizers. Among the treatments, under absolute control was recorded maximum soil pH (7.32) and minimum pH (7.19) with application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation. It is evident from data the electrical conductivity did not significantly influence with application of Fe, Zn and Bio-fertilizers. Among the treatments, application of 100% RDF recorded maximum EC (0.68 dS m^{-1}) and least EC (0.49 dS m^{-1}) under application of absolute control during course of experimentation. Under application of Fe, Zn and Bio-fertilizers did not significantly influence organic carbon content in soil after crop harvest. It is evident from data the bulk density and particle density could not reach the level of significance due to application of Fe, Zn and Bio-fertilizers. The maximum bulk density (1.33 Mg m^{-3}) and particle density (2.45 Mg m^{-3}) were noted under absolute control. However, minimum value of bulk density (1.31 Mg m^{-3}) and particle density (2.41 Mg m^{-3}) was noted under application of 75% RDF + Rhizobium @ 600 g ha^{-1} seed inoculation. Soil porosity did not find to show any significant effect due to application of Fe, Zn and Bio-fertilizers. The maximum higher porosity (45.71%) was noted under control and application of 100% RDF over rest of treatments.

The results regarding residual effect of biofertilizers (*Rhizobium* and PSB) and spray of $FeSO_4$ and $ZnSO_4$ on soil pH, EC, porosity, bulk density, particle density and organic carbon was presented. The data showed that pH, EC, particle density, bulk density of soil did not significantly influence due to individual seed treatment with *Rhizobium* and *Bacillus megaterium* levels but the lower soil pH was recorded under the treatment 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering & pod formation stage + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation, similarly higher soil pH was observed with control. Singh et al. [4] revealed that the effect of various treatments on soil pH with addition of organic and inorganic fertilizer is not consistent. Govindan and Thirumurugan [25] did not found any change in EC with the treatment of bio-inoculants. Similar results were also observed in bulk density and particle density which ranged from 1.31 to 1.33, 2.41to 2.45 Mg m^{-3} and data indicated that the difference in soil bulk density and particle density values were not reach to the levels of significance due to application of Fe, Zn and biofertilizer. Similar results were also observed in per cent organic carbon and porosity.

3.3 Available Nutrients of Soil after Crop Harvest

The data pertaining to nutrient content and uptake of urdbean crop significantly influenced under application of Fe, Zn and Bio-fertilizers were presented in Table 4. The significantly higher available nitrogen (285.21 kg ha^{-1}), phosphorus (25.41 kg ha^{-1}), potassium (252.61 kg ha^{-1}), Fe (5.75 mg kg^{-1}) and Zn (0.76 mg kg^{-1}) content in soil after crop harvest were recorded with application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation over control nitrogen nitrogen (228.21kg ha^{-1}), phosphorus (17.46 kg ha^{-1}), potassium (230.10 kg ha^{-1}), Fe (4.30 mg kg^{-1}) and Zn (0.59 mg kg^{-1}), 75 % RDF nitrogen (238.88 kg ha^{-1}), phosphorus (20.72 kg ha^{-1}), potassium (241.87 kg ha^{-1}), Fe (4.64 mg kg^{-1}) and Zn (0.62 mg kg^{-1}),100 % RDF nitrogen (245.84 kg ha^{-1}), phosphorus (21.28 kg ha^{-1}), potassium (243.57 kg ha^{-1}), Fe (4.70 mg kg^{-1})

Table 2. Effect of Fe, Zn and Bio-fertilizers application on nutrient content in seed and straw of urdbean

Treatments	Nitrogen content (%)		Phosphorous content (%)		Potassium content (%)		Iron content (mg kg^{-1})		Zinc content (mg kg^{-1})	
	Seed	Straw	Seed	Straw	Seed	Straw	Seed	Straw	Seed	Straw
T_1: Control	1.66	1.37	0.22	0.117	0.40	1.33	27.22	42.94	20.05	11.87
T_2: 75 % RDF	1.94	1.48	0.24	0.123	0.44	1.48	31.13	39.87	21.79	13.58
T_3: 100 % RDF	2.45	1.57	0.25	0.129	0.47	1.57	34.05	53.46	23.84	14.85
T_4: T_2 + *Rhizobium* @ 600 g ha^{-1} seed inoculation	2.49	1.59	0.27	0.133	0.48	1.60	36.14	56.73	25.29	15.76
T_5: T_2 + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	2.46	1.58	0.26	0.131	0.47	1.57	35.57	55.84	24.90	15.51
T_6: T_2 + *Rhizobium* @ 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1}	2.53	1.70	0.28	0.141	0.51	1.67	37.58	58.99	26.30	16.39
T_7: T_2 + 0.1 % FeSO4 + 0.5 % ZnSO4 at pre flowering and pod formation stage	2.52	1.63	0.29	0.134	0.50	1.62	36.72	57.65	25.70	16.01
T_8: T_2 + T_7 + *Rhizobium* @ 600 g ha^{-1} seed inoculation	3.11	1.77	0.31	0.149	0.53	1.80	41.82	65.66	30.27	18.24
T_9: T_2 + T_7 + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	2.99	1.75	0.30	0.145	0.53	1.72	41.38	64.96	29.96	18.05
T_{10}: T_2 + T_7 0 + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	3.87	1.84	0.34	0.154	0.56	1.82	45.15	73.89	32.61	19.69
SEm±	0.12	0.03	0.01	0.003	0.01	0.05	1.95	4.87	0.99	0.61
CD (p=0.05)	0.37	0.10	0.03	0.008	0.03	0.13	5.80	14.47	2.93	1.83

Table 3. Effect of Fe, Zn and Bio-fertilizers application on nutrient uptake by seed, straw and total by urdbean crop

Treatments	Nitrogen uptake (kg ha^{-1})			Phosphorus uptake (kg ha^{-1})			Potassium uptake (kg ha^{-1})			Iron uptake (g ha^{-1})			Zinc uptake (g ha^{-1})		
	Seed	Straw	Total	Seed	Straw	Total	Seed	Straw	Total	Seed	Straw	Total	Seed	Straw	Total
T_1: Control	10.04	10.99	21.03	1.33	0.94	2.27	2.41	10.68	13.09	167.06	344.14	511.20	120.59	95.23	215.82
T_2: 75 % RDF	14.26	13.76	28.02	1.76	1.14	2.90	3.25	13.76	17.00	227.51	461.25	688.76	159.97	125.72	285.69
T_3: 100 % RDF	20.00	16.13	36.13	2.04	1.32	3.37	3.82	16.01	19.83	278.19	550.66	828.85	194.90	152.17	347.07
T_4: T_2 + *Rhizobium* @ 600 g ha^{-1} seed inoculation	21.39	17.43	38.83	2.33	1.46	3.79	4.15	17.47	21.63	310.48	625.75	936.23	218.81	171.05	389.87
T_5: T_2 + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	21.05	17.22	38.27	2.22	1.43	3.64	4.02	17.11	21.13	303.01	607.60	910.61	212.47	168.28	380.74
T_6: T_2 + *Rhizobium* @ 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1}	22.93	19.57	42.51	2.53	1.62	4.15	4.62	19.21	23.83	336.86	685.66	1022.51	236.50	187.88	424.38
T_7: T_2 + 0.1 % FeSO4 + 0.5 % ZnSO4 at pre flowering and pod formation stage	22.20	18.20	40.40	2.56	1.50	4.05	4.35	18.03	22.38	325.38	643.15	968.53	226.17	178.83	404.99
T_8: T_2 + T_7 + *Rhizobium* @ 600 g ha^{-1} seed inoculation	31.26	21.99	53.25	3.11	1.85	4.96	5.32	22.44	27.76	419.13	809.61	1228.75	304.10	226.82	530.92
T_9: T_2 + T_7 + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	29.72	21.63	51.35	2.98	1.79	4.77	5.23	21.22	26.46	410.23	804.38	1214.62	295.57	223.41	518.98
T_{10}: T_2 + T_7 0 + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	49.36	27.84	77.20	4.32	2.33	6.65	7.16	27.44	34.60	573.25	1120.36	1693.60	413.78	297.40	711.18
SEm±	2.01	1.12	2.32	0.13	0.09	0.15	0.25	1.09	1.12	21.34	72.82	64.02	11.66	9.11	11.87
CD (p=0.05)	5.98	3.33	6.91	0.39	0.28	0.46	0.74	3.25	3.32	63.43	216.35	190.21	34.65	27.08	35.28

Table 4. Effect of Fe, Zn and Bio-fertilizers application on soil physico-chemical properties and nutrient status after harvest of urdbean

Treatments	pH	EC (dS m^{-1})	Organic carbon (%)	Bulk density (Mg m^{-3})	Particle density (Mg m^{-3})	Porosity (%)	Available macro nutrients (kg ha^{-1})			Available micro nutrients (mg kg^{-1})	
							N	P	K	Fe	Zn
T_1: Control	7.32	0.49	0.41	1.33	2.45	45.71	228.21	17.46	230.10	4.30	0.59
T_2: 75 % RDF	7.28	0.61	0.42	1.32	2.43	45.68	238.88	20.72	241.87	4.64	0.62
T_3: 100 % RDF	7.27	0.68	0.43	1.33	2.45	45.71	245.84	21.28	243.57	4.70	0.65
T_4: T_2 + *Rhizobium* @ 600 g ha^{-1} seed inoculation	7.25	0.62	0.42	1.31	2.41	45.64	266.92	21.79	245.39	5.15	0.67
T_5: T_2 + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	7.22	0.61	0.44	1.31	2.41	45.64	260.33	23.77	247.51	5.07	0.66
T_6: T_2 + *Rhizobium* @ 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1}	7.20	0.63	0.46	1.31	2.41	45.64	272.88	24.00	250.82	5.18	0.69
T_7: T_2 + 0.1 % FeSO4 + 0.5 % ZnSO4 at pre flowering and pod formation stage	7.21	0.64	0.44	1.32	2.43	45.68	258.54	22.74	249.71	5.25	0.71
T_8: T_2 + T_7 + *Rhizobium* @ 600 g ha^{-1} seed inoculation	7.20	0.63	0.46	1.32	2.43	45.68	280.17	23.61	254.75	5.48	0.74
T_9: T_2 + T_7 + *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	7.23	0.65	0.45	1.32	2.43	45.68	275.88	24.47	250.15	5.44	0.72
T_{10}: T_2 + T_7 + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation	7.19	0.64	0.48	1.32	2.43	45.68	285.21	25.41	252.66	5.75	0.76
SEm±	0.07	0.10	0.02	0.01	0.02	0.13	8.02	1.06	13.30	0.10	0.02
CD (p=0.05)	NS	NS	NS	NS	NS	NS	23.84	3.15	NS	0.30	0.06

and Zn (0.65 mg kg^{-1}), 75 % RDF + *Rhizobium* @ 600 g ha^{-1} seed inoculation nitrogen (266.92 kg ha^{-1}), phosphorus (21.79 kg ha^{-1}), potassium (245.39 kg ha^{-1}), Fe (5.15 mg kg^{-1}) and Zn (0.67 mg kg^{-1}), 75 % RDF + *PSB* @ 600 g ha^{-1} seed inoculation nitrogen (260.33 kg ha^{-1}), phosphorus (23.77 kg ha^{-1}), potassium (247.51 kg ha^{-1}), Fe (5.07 mg kg^{-1}) and Zn (0.66 mg kg^{-1}), 75 % RDF + *Rhizobium* @ 600 g ha^{-1} + *Bacillus megaterium* @ 600 g ha^{-1} nitrogen (272.88 kg ha^{-1}), phosphorus (24.0 kg ha^{-1}), potassium (250.82 kg ha^{-1}), Fe (5.18 mg kg^{-1}) and Zn (0.69 mg kg^{-1}), 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage nitrogen (258.54 kg ha^{-1}), phosphorus (22.74 kg ha^{-1}), potassium (249.71 kg ha^{-1}), Fe (5.25 mg kg^{-1}) and Zn (0.71 mg kg^{-1}), 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Rhizobium* @ 600 g ha^{-1} nitrogen (280.17 kg ha^{-1}), phosphorus (23.61 kg ha^{-1}), potassium (254.75 kg ha^{-1}), Fe (5.48 mg kg^{-1}) and Zn (0.74 mg kg^{-1}), 75 % RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Bacillus megaterium* @ 600 g ha^{-1} nitrogen (275.88 kg ha^{-1}), phosphorus (24.47 kg ha^{-1}), potassium (250.15 kg ha^{-1}), Fe (5.44 mg kg^{-1}) and Zn (0.72 mg kg^{-1}) content in soil after crop harvest.

Application of 75% RDF + 0.1 % $FeSO_4$ + 0.5 % $ZnSO_4$ foliar spray at pre flowering & pod formation stage + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1} seed inoculation showed the significant effect on the available nitrogen, phosphorus, Zn and Fe in soil. Significant buildup of the soil available nitrogen could be attributed to increased activity of nitrogen fixing rhizobia there by resulting in higher accumulation of nitrogen in the soil leading to better nodulation and mineralization of organic nitrogen with phosphorus application [26]. These results were in close conformity to Sarawagi et al. [27], who reported that the application of microbial culture along with phosphorus enhances the nitrogen and phosphorus content in chickpea. Kumar et al. [28] reported that the PSB are known to have ability to solubilize phosphorus from insoluble source. The PSB secretes the different organic acids which act on insoluble phosphate to convert them in to soluble phosphate near the root of the plant and hence availability of phosphorus is increased. Konthoujam et al. [29] who had indicated that the judicious integration of organic and inorganic sources of nutrition significantly improved the soil available nitrogen, phosphorus and micronutrient in soil. Further, the buildup of available phosphorus with phosphorus application can be attributed to an increase in the available soil phosphorus after satisfying phosphorus fixation capacity and other chemical reaction [30]. Duraisami and Mani [31] and Nimje [32] who observed increased available phosphorus content in soil after the harvest of the crop due to phosphorus application. The increase in available phosphorus content of soil might be due to greater mobilization of native soil phosphorus by vigorous root proliferation and contribution through biomass. These results find support from the results of Kumar and Elamathi [33], Raja and Takankhar [34] in soybean. The application of 75% RDF with iron and zinc application significantly increased the available nitrogen and phosphorus content in soil [35]. As expected, a linear increase in available zinc and iron content in soil was observed with application of Zn @ 0.5% and Fe @ 0.1% through $ZnSO_4$ and $FeSO_4$, after harvest of the crop. The experimental soil being low in available zinc and iron might have resulted in increased available zinc and iron application. There could be a priming effect which possibly caused solubilization of native zinc and iron with increase in the rate of zinc and iron application. The result was in conformity to those of reported by Badiyala and Chopra [36] and Kannan et al. [37].

4. CONCLUSION

It is concluded that the application of 75% RDF + Fe 0.1 % $FeSO_4$ + Zn 0.5 % $ZnSO_4$ at pre flowering and pod formation stage + *Rhizobium* 600 g ha^{-1}+ *Bacillus megaterium* @ 600 g ha^{-1} found most suitable dose for urdbean to increase nutrient content & uptake and also beneficial for soil health.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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