BIOFERTILIZERS AND INORGANIC FERTILIZERS APPLICATION IMPACT ON AVAILABLE NUTRIENTS IN SOIL OF *KHARIF* MAIZE

ABSTRACT

A field experiment entitled **"Biofertilizers and inorganic fertilizers application impact on available nutrients in soil of *kharif* maize"** was conducted at Agriculture College Farm, Bapatla, during *kharif* of 2020-21. The experiment was laid out in randomized block design (RBD) with seven treatments and replicated thrice. The treatments consisted of T_1- Control; T_2- 100% RDF; T_3- 125% RDF; T_4 – 100% RDF +Vesicular Arbuscular Mycorrhizae (VAM); T_5- 100% RDF +VAM + *Azospirillum* + Phosphate Solubilizing Bacteria (PSB); T_6- 75 % RDF + VAM; T_7- 75 % RDF + VAM + *Azospirillum* + PSB. We recorded the available macro nutrients (nitrogen, phosphorus and potassium) and micronutrients (zinc, iron, manganese and copper) of maize during the study. The results revealed that available nitrogen during kharif of 2020-21 was significantly ($P < 0.05$) higher with 125 % RDF (T_3) that was on par with T_5 (100% RDF + VAM + *Azospirillum* + PSB) and T_4 (100% RDF + VAM). Higher available phosphorus and potassium were recorded in the treatment T_5 and it was on par with T_7, T_3 and T_4 at knee high, tasseling and harvest stage of maize. However, micronutrient status was higher with application of 100% RDF +VAM + *Azospirillum* + PSB but non-significant at all the growth stages of maize during both the years of study. The biofertilizers have to be applied side-by-side with inorganic fertilizers to improve nutrient availability and increased the fertility status of soil and productivity.

Key words: Biofertilizers, Fertilizers levels, Available nutrients and Soil health.

INTRODUCTION

"Maize (*Zea mays* L.) is one of the most important cereal crops next only to wheat and rice in the world. In India, it ranks fourth after rice, wheat and sorghum. Maize is the principal staple food in many countries, particularly in the tropics and subtropics and it is being consumed as food and fodder and required by the various industries. The crop has high genetic yield potential hence, it is called Miracle crop and "Queen of Cereals. It is a nutrient- exhaustive crop than other cereals and absorbs a large number of nutrients from the soil during its growth. Maize responds well to fertilizer but under field conditions due to over- reliance on nitrogenous fertilizers and no or negligible use of organic manure its yield potential is difficult to exploit. biofertilizers not only help to provide balanced nutrients but also support sustainable production due to their pivotal role in soil health enhancement". (Janardhan et. Al., 2022)

"Biofertilizers are products containing live or latent microorganisms that are capable of mobilizing nutrients from unavailable form to available form through biological processes. Proper use of biofertilizers and inorganics maintains the fertility of agricultural soils. *Azospirillum* is a free- living bacteria that colonize near the root zone and enhance the available nitrogen in the soil by fixation, whereas phosphate solubilizing bacteria (PSB) solublize the unavailable phosphorus in the soil and make it available for the plants" (Kachari and Korla, 2009). "*Vesicular Arbuscular Mycorrhizae* (VAM) help in the development of a stronger root system, increase root surface area, and improve growth" (Zandavalli *et al.*, 2004). The use of nitrogen- fixing microbes helps in reducing the dependence on urea, while phosphorus-solubilizing microbes will increase the availability of P from relatively unavailable pools, thus the use of the integrated source of nutrients will help in curtailing over- dependence on inorganic fertilizers alone for nutrient supply to maize. Application biofertilizers along with inorganic fertilizers improved available soil nutrient status.

Soil fertility is influenced by biofertilizers, which play an important role in fixing atmospheric nitrogen, solubilizing insoluble form of phosphorous, potash and mobilizing the immobile nutrients in the soil. These processes can enhance the nutrient status of the soil. In this study, a microbial consortium consisting of biofertilizers *viz.*, nitrogen- fixing, P solubilizer bacteria and VAM were used.

MATERIAL AND METHODS

Site Description

The field experiment was carried out during the *kharif* of 2020-21 at Agricultural College Farm, Bapatla. Geographically located at an altitude of 5.49 m above sea level, $15^{o}54'$ North latitude, $80^{0}30'$ East longitude and about 8 km away from the Bay of Bengal. It is located in the Krishna agro-climatic zone of Andhra Pradesh. The experimental soil was clay loam in texture, slightly alkaline in reaction (pH 7.56), non- saline (0.64 dS-m^{-1}), medium in organic carbon (5.4 g-kg^{-1}), medium in available nitrogen (283 kg-ha^{-1}), medium in available phosphorus (42.5 kg-ha^{-1}), high in potassium (426 kg-ha^{-1}) and soil micronutrients *viz;* iron (6.81 mg -kg^{-1}), manganese (5.43 mg-kg^{-1}), copper (1.37 mg -kg^{-1}) and zinc (0.58 mg -kg^{-1}) in initial soil characters of the experimental field.

Experimental design and treatments

The experiment was laid out in randomized block design (RBD) with seven treatments and replicated thrice. The experimental treatment details are as following T_1- Control; T_2- 100% RDF; T_3- 125% RDF; T_4 – 100% RDF + Vesicular Arbuscular Mycorrhizae (VAM); T_5- 100% RDF +VAM +

Azospirillum + Phosphate Solubilizing Bacteria (PSB); T_6- 75 % RDF + VAM; T_7- 75 % RDF + VAM + *Azospirillum* + PSB. RDF for maize 200:60:50 kg-ha^{-1} N, P_2O_5 and K_2O through applied Urea, Single super phosphate and Muriate of potash and biofertilizers like VAM -12.5 kg-ha^{-1}, *Azospirillum* -5 kg-ha^{-1} and PSB -5 kg -ha^{-1} through applied vermicompost.

Collection and Preparation of Soil Samples: Initial soil samples were collected from the entire field randomly and made into composite samples, from the composite sample representative sample was taken by a quartering method. Plot- wise surface (0-15) soil samples were collected at knee-high, tasseling and at harvest stages of maize. The soil samples were air dried in the shade, ground and screened through a 2_mm sieve and used for laboratory analysis. These soil samples were further estimation of available soil nutrient status. "Available nitrogen was estimated by the alkaline permanganate method by using a macro Kjeldahl distillation unit" (Subbiah and Asija, 1956). "Available phosphorus in the soil samples was extracted with 0.5 M $NaHCO_3$ buffered at pH 8.5 and the phosphorus in the extract was estimated by the ascorbic acid method using a spectrophotometer at 660 nm" (Watanabe and Olsen, 1965). "Available potassium in the samples was extracted with neutral normal ammonium acetate and estimated with the help of a flame photometer" (Jackson, 1973). "Available zinc, iron, manganese and copper in the soils were determined in DTPA extract, using atomic absorption spectrophotometer" (Lindsay and Norvell, 1978).

RESULTS AND DISCUSSION

Available macronutrients

Available nitrogen

The results (Table-1) indicated that various levels of fertilizers and biofertilizer treatments imposed on maize crops have shown a significant ($P < 0.05$) effect on available nitrogen at all the growth stages of maize. Significantly ($P < 0.05$) higher nitrogen was recorded in the treatment T_3 (372, 361, 349 kg -ha^{-1} and 383, 366, 351 kg -ha^{-1}) in kharif, 2020-21, respectively at knee high, tasseling and harvest stages of maize and it was on par with the treatments T_5 (349, 338, 319 kg -ha^{-1} and 357343, 317 kg -ha^{-1}) and T_4 (338, 327, 314 kg -ha^{-1} and 346, 332, 316 kg -ha^{-1}) and these were significantly($P < 0.05$) superior over other treatments. The lowest available nitrogen was recorded in control (T_1) (262, 234, 204 kg -ha^{-1} in 2020 and 269, 239, 206 kg -ha^{-1} in 2021) which received no fertilizers at all three stages of crop growth. The moist soil conditions might have helped the mineralization of soil nitrogen and greater multiplication of soil microbes, which could convert organically bound nitrogen into readily available form leading to building up of higher available nitrogen. The combined application of biofertilizers and inorganic fertilizers increased the available nitrogen in the soil. *Azospirillum* excrete ammonia in the rhizosphere in the presence of root exudates. This might be ascribed to the fixation of nitrogen by the *Azospirillum.* The increase in the availability of nitrogen in the plots where biofertilizers were applied could also be attributed to the positive relationship between the added and native microorganisms in the soil that mineralized the organic matter (Ponmurugan and Gopi, 2006). The role of *Azospirillum* and phosphobacteria in enhancing the availability of nitrogen in the soil was reported by Ram *et al*. (2011).

Available phosphorus

Treatment T_5 (62.08, 58.69, 53.53 kg ha^{-1} and 64.11, 60.07, 55.65 kg ha^{-1}) recorded significantly ($P < 0.05$) highest available phosphorus in soil and it was on par with the treatments T_7 , T_4 and T_3 at knee high, tasseling and harvest stages of maize crop. These were significantly superior over other treatments. The lowest available phosphorus was recorded with the treatment T_1 control (39.52, 35.33, 31.35 kg ha^{-1} in 2020 and 41.59, 37.54, 32.15 kg ha^{-1} in 2021) which received no fertilizers at all the three stages of crop growth. "The results revealed that available soil phosphorus significantly ($P< 0.05$) increased with the combined application of biofertilizers and inorganic fertilizers. The use of biofertilizer with chemical fertilizer can play an important role in improving P availability. The increase in soil P content might be due to the P-solubilizing potential of the isolates in biofertilizer. This may be attributed to the production of organic acids and solubilization of inorganic insoluble phosphates by microorganisms. The potential role of soil microorganisms for increasing the amount of available P by phytase activity" has also been reported by Richardson (2001). "Amending soil with biofertilizers and inorganic fertilizers helps in enhancing the P concentration in solution through the mineralization of organic P and solubilization of native soil P compounds by producing organic acids" (Roy *et al.*, 2017).

Available Potassium

Results (Table-3) indicated that combined application of inorganics and biofertilizers have shown significant effect on available potassium at all three growth stages of maize. The results revealed a significantly ($P < 0.05$) highest available potassium in the soil was recorded in the treatment T_5 (493, 475, 457 kg ha^{-1} and 500, 482, 460 kg ha^{-1}) and it was on par with the treatments T_7, T_3 and T_4 at knee high, tasseling and harvest stages of maize crop. These were significantly (P <

0.05) superior over other treatments. The lowest available potassium was recorded with the treatment T_1 control (389, 363, 332 kg ha^{-1} in 2020 and 391, 365, 335 kg ha^{-1} in 2021). “The present study indicated that application of biofertilizers along with inorganic fertilizers increased the available potassium content in the soil. This may be due to a variety of soil microbes that can release soluble potassium from potassium-bearing minerals. These microbes release organic acid, which quickly dissolves rock and chelate ions, releasing K ions into the soil” (Friedrich *et al*., 2004). The presence of indigenous potassium-solubilizing microbes might have increased the concentration of available soil potassium. The organic acids released during the decomposition of manures mobilize the native or non-exchangeable forms of potassium and charge the soil solution with potassium ions, so that it will be readily available. The results were in close conformity with Thakur *et al*. (2010) and Pande *et al*. (2013).

Available micronutrients

Available Iron

The results (Table-4) revealed that available iron at all the stages of maize was non significantly influenced by different levels of fertilizers and biofertilizers during both the years of study. Numerically, higher available iron in soil was recorded in T_5 *i.e*., 100% RDF+ VAM + *Azospirillum* and PSB (7.54, 7.27, 7.08 mg kg^{-1} and 7.69, 7.38, 7.14 mg kg^{-1}) and lower values (6.70,6.46,6.23 mg kg^{-1} and 6.85,6.55,6.27 mg kg^{-1}) were recorded in T_1 control at knee high, tasseling and harvest stages of maize respectively. Critical observation of the data revealed that there was no much influence of inorganics alone or their combination with biofertilizers on available micronutrient status of the soil. However, the treatments received combined application of inorganics with biofertilizers have shown a slight increase in micronutrients, which might be due to formation of chelated forms and enhance mineralization and solubilisation of the native nutrients (Kharache *et al.*, 2013). Increase in available Fe might be due to lowering of pH as a result of decomposition of organics which was known to increase the solubility of metallic elements (Prasad *et al*., 2010b).

Available Manganese

The available manganese content presented in the table-5 and revealed that available manganese at all the stages of maize was non significantly influenced by different levels of fertilizers and biofertilizers during both the years of study. Numerically, higher available manganese in soil was recorded in T_5 *i.e*., 100% RDF+ VAM + *Azospirillum* and PSB (6.44, 6.27, 6.17 mg kg^{-1} and 6.52, 6.39, 6.24 mg kg^{-1}) and lower values (5.30, 5.10, 5.00 mg kg^{-1} and 5.38, 5.17, 5.06 mg kg^{-1}) were recorded in T_1 control at knee high, tasseling and harvest stages of maize, respectively. The combined application of inorganics and biofertilizers non significantly influenced available manganese content but slightly increased when compared to control. “This might be due to the solubility of Mn under relatively acid and reducing conditions like Fe. Most of the total Mn in soils was found in the Mn - oxide and organic fractions the later are more soluble and therefore, easier to redistribute in plant available forms than the Fe - oxide and residual forms” (Das, 2000). However, the available manganese was decreased with advancement of crop stage during both the years. This decrease in Mn might be attributed to the depletion of micronutrients from soil due to crop uptake (Veeranagappa *et al*., 2011a).

Available Copper

The results pertaining to soil available copper (Table-6) revealed that irrespective of the year of study, the available copper status at knee high, tasseling and harvest stages of maize crop was non significantly influenced by the treatments that received inorganics along with the biofertilizers. The higher available copper content in soil recorded with the treatment T_5 *i.e.*, 100% RDF+ VAM + *Azospirillum* and PSB (1.69,1.59,1.55 mg kg^{-1} and 1.77,1.65,1.59 mg kg^{-1}) and lower values (1.32, 1.22, 1.18 mg kg^{-1} and 1.38,1.28,1.22 mg kg^{-1}) were recorded in T_1 control at knee high, tasseling and harvest stages of maize, respectively. “The enhancement in the available Cu due to the addition of organic substances might be ascribed to their ability to form stable water soluble complexes preventing the reaction with other soil constituents and also increasing the Cu content by releasing it from the native reserves” (Gupta *et al*., 1988). Copper has a strong affinity for the nitrogen atom of amino groups and it appeared quite likely that soluble nitrogen compounds like amino acids act as copper carriers in xylem and phloem.

Available Zinc

Higher available zinc in soil was recorded in T_5 *i.e.*, 100% RDF+ VAM + *Azospirillum* and PSB (0.67, 0.62, 0.60 mg kg^{-1} and 0.73, 0.68, 0.64 mg kg^{-1}) and lower values (0.54, 0.50, 0.47 mg kg^{-1} and 0.59, 0.54, 0.51 mg kg^{-1}) were recorded in T_1 control at knee high, tasseling and harvest stages of maize during both *kharif*, 2020 and 2021, respectively (Table-6). Application of 100% RDF+ VAM + *Azospirillum* and PSB (T_5) slightly increased zinc content compared to control (T_1), but non significantly influenced by the treatments. The available Zn decreased with advancement of crop stage during both the years of study. The decrease might be attributed to uptake of Zn by the growing plants. On the other hand, all the available micronutrients were gradually decreased with advancement of crop stage *i.e.,* from knee high to harvest stages of maize. The lowest content was obtained at harvest stage and the highest content was obtained at knee high stage. This might be due to the continuous depletion of nutrients by crop uptake. Similar type of results were noted by Subhalakshmi and Pratapkumarreddy (2017).

Conclusion

The combined application of different levels of fertilizers and biofertilizers significantly improved available soil nutrient status. The efficacy of biofertilizers as good organic amendments in the soil for enhanced improvement of soil chemical properties. The application of biofertilizers has beneficial effects on maintaining the soil nutrients as it increases the available macro nutrients (nitrogen, phosphorus and potassium) and micro nutrients (iron, manganese, copper and zinc) of the soil and improved soil health.

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Table 1. Effect of different levels of fertilizers in combination with biofertilizers on available nitrogen (kg ha^{-1}) at different stages of maize

Treatments	*Kharif* (2020)			*Kharif* (2021)		
	Kneehigh	**Tasseling**	**Harvest**	**Kneehigh**	**Tasseling**	**Harvest**
T_1: Control	262	234	204	269	239	206
T_2: 100% RDF	309	294	277	316	299	281
T_3: 125% RDF	372	361	349	383	366	351
T_4: 100% RDF + VAM	338	327	314	346	332	316
T_5: 100% RDF + VAM+ *Azospirillum* + PSB	349	338	319	357	343	317
T_6: 75% RDF + VAM	299	278	247	309	283	253
T_7: 75% RDF + VAM + *Azospirillum* + PSB	313	297	278	320	302	281
SEm (±)	11.3	11.4	11.8	12.5	11.4	11.8
CD (P=0.05)	35	35	36	38	35	36
CV (%)	6.2	6.5	7.2	6.6	6.4	7.2

Treatments	*Kharif* (2020)	*Kharif* (2021)

Table 2. Effect of different levels of fertilizers in combination with biofertilizers on available phosphorus (kg ha^{-1}) at different stages of maize

	Kneehigh	**Tasseling**	**Harvest**	**Kneehigh**	**Tasseling**	**Harvest**
T_1: Control	39.52	35.33	31.35	41.59	37.54	32.15
T_2: 100% RDF	51.35	47.26	43.62	53.38	49.36	44.34
T_3: 125% RDF	55.28	51.24	47.17	57.33	53.31	48.61
T_4: 100% RDF + VAM	57.24	52.56	48.19	59.30	55.27	50.23
T_5: 100% RDF + VAM+ *Azospirillum* + PSB	62.08	58.69	53.53	64.11	60.07	55.65
T_6: 75% RDF + VAM	49.38	45.02	42.62	51.49	47.46	43.44
T_7: 75% RDF + VAM + *Azospirillum* + PSB	59.95	55.67	50.26	62.01	57.97	52.93
SEm (±)	2.46	2.63	2.54	2.45	2.44	2.36
CD (P=0.05)	7.57	8.12	7.81	7.54	7.53	7.28
CV (%)	7.95	9.24	9.70	7.62	8.21	8.78

Table 3. Effect of different levels of fertilizers in combination with biofertilizers on available potassium (kg ha^{-1}) at different stages of maize

Treatments	*Kharif* (2020)			*Kharif* (2021)		
	Kneehigh	Tasseling	Harvest	Kneehigh	Tasseling	Harvest
T_1: Control	389	363	332	391	365	335
T_2: 100% RDF	445	423	403	449	425	405
T_3: 125% RDF	483	453	436	486	457	439
T_4: 100% RDF + VAM	476	446	426	479	450	429
T_5: 100% RDF + VAM+ *Azospirillum* + PSB	493	475	457	500	482	460
T_6: 75% RDF + VAM	439	417	387	443	420	386
T_7: 75% RDF + VAM + *Azospirillum* + PSB	484	463	445	488	467	448
SEm (±)	16.5	16.6	16.8	16.0	16.5	15.9
CD (P=0.05)	49	51	52	49	51	49
CV (%)	6.3	6.6	7.0	6.0	6.6	6.6

Treatments	*Kharif* (2020)	*Kharif* (2021)

Table 4. Effect of different levels of fertilizers in combination with biofertilizers on available iron (mg kg^{-1}) at different stages of maize

	Kneehigh	**Tasseling**	**Harvest**	**Kneehigh**	**Tasseling**	**Harvest**
T_1: Control	6.70	6.46	6.23	6.85	6.55	6.27
T_2: 100% RDF	7.27	7.01	6.82	7.42	7.11	6.87
T_3: 125% RDF	7.34	7.09	6.91	7.49	7.20	6.94
T_4: 100% RDF + VAM	7.31	7.06	6.83	7.46	7.16	6.88
T_5: 100% RDF + VAM+ *Azospirillum* + PSB	7.54	7.27	7.08	7.69	7.38	7.14
T_6: 75% RDF + VAM	7.18	6.94	6.71	7.33	7.05	6.76
T_7: 75% RDF + VAM + *Azospirillum* + PSB	7.46	7.22	7.04	7.61	7.33	7.10
SEm (±)	0.31	0.31	0.31	0.31	0.31	0.31
CD (P=0.05)	NS	NS	NS	NS	NS	NS
CV (%)	7.45	7.57	7.92	7.33	7.46	7.88

Table 5. Effect of different levels of fertilizers in combination with biofertilizers on available manganese (mg kg^{-1}) at different stages of maize

Treatments	*Kharif* (2020)			*Kharif* (2021)		
	Kneehigh	Tasseling	Harvest	Kneehigh	Tasseling	Harvest
T_1: Control	5.30	5.10	5.00	5.38	5.17	5.06
T_2: 100% RDF	5.72	5.59	5.50	5.81	5.67	5.57
T_3: 125% RDF	6.14	5.93	5.81	6.23	6.02	5.88
T_4: 100% RDF + VAM	5.93	5.74	5.65	6.03	5.85	5.72
T_5: 100% RDF + VAM+ *Azospirillum* + PSB	6.44	6.27	6.17	6.52	6.39	6.24
T_6: 75% RDF + VAM	5.61	5.43	5.37	5.69	5.49	5.44
T_7: 75% RDF + VAM + *Azospirillum* + PSB	6.33	6.11	6.02	6.41	6.23	6.09
SEm (±)	0.25	0.24	0.25	0.25	0.25	0.25
CD (P=0.05)	NS	NS	NS	NS	NS	NS
CV (%)	7.37	7.38	7.80	7.31	7.42	7.69

Table 6. Effect of different levels of fertilizers in combination with biofertilizers on available copper (mg kg^{-1}) at different stages of maize

Treatments	*Kharif* (2020)			*Kharif* (2021)		
	Kneehigh	**Tasseling**	**Harvest**	**Kneehigh**	**Tasseling**	**Harvest**
T_1: Control	1.32	1.22	1.18	1.38	1.28	1.22
T_2: 100% RDF	1.57	1.48	1.44	1.63	1.56	1.48
T_3: 125% RDF	1.64	1.55	1.51	1.71	1.61	1.55
T_4: 100% RDF + VAM	1.60	1.51	1.47	1.67	1.58	1.51
T_5: 100% RDF + VAM+ *Azospirillum* + PSB	1.69	1.59	1.55	1.77	1.65	1.59
T_6: 75% RDF + VAM	1.50	1.40	1.37	1.56	1.46	1.41
T_7: 75% RDF + VAM + *Azospirillum* + PSB	1.67	1.58	1.52	1.75	1.64	1.56
SEm (±)	0.08	0.08	0.08	0.08	0.08	0.08
CD (P=0.05)	NS	NS	NS	NS	NS	NS
CV (%)	8.43	9.09	9.27	8.22	8.92	9.01

Treatments	*Kharif* (2020)	*Kharif* (2021)

Table 7. Effect of different levels of fertilizers in combination with biofertilizers on available zinc (mg kg^{-1}) at different stages of maize

	Kneehigh	Tasseling	Harvest	Kneehigh	Tasseling	Harvest
T_1: Control	0.54	0.50	0.47	0.59	0.54	0.51
T_2: 100% RDF	0.59	0.55	0.53	0.64	0.60	0.57
T_3: 125% RDF	0.63	0.59	0.57	0.69	0.64	0.61
T_4: 100% RDF + VAM	0.61	0.58	0.56	0.66	0.62	0.60
T_5: 100% RDF + VAM+ *Azospirillum* + PSB	0.67	0.62	0.60	0.73	0.68	0.64
T_6: 75% RDF + VAM	0.57	0.53	0.51	0.62	0.57	0.55
T_7: 75% RDF + VAM + *Azospirillum* + PSB	0.65	0.62	0.58	0.71	0.67	0.62
SEm (±)	0.03	0.03	0.03	0.03	0.04	0.03
CD (P=0.05)	NS	NS	NS	NS	NS	NS
CV (%)	8.35	8.26	8.22	8.06	9.80	7.66