

Influence of different levels of inorganic phosphorus in combination with biofertilizers on P-fractions in soil at different growth stages of sorghum

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ABSTRACT

Inorganic soil phosphorus fractions on sorghum at different growth stages were evaluated with the application of different levels of phosphorus fertilizer along with biofertilizers during *Rabi* 2022. The experiment was laid out in randomized block design and replicated thrice, conducted at Agricultural College Farm, Bapatla. The treatments comprised of T₁-0% RDP, T₂- 50% RDP, T₃- 75% RDP, T₄- 100% RDP, T₅- 0% RDP + PSB, T₆- 0% RDP + PSB + VAM, T₇- 50% RDP + PSB, T₈-50% RDP + PSB + VAM, T₉-75% RDP + PSB, T₁₀- 75% RDP + PSB + VAM. The soil samples were analysed for P- fractions by standard procedures. The results of the experiment indicated all the P- fractions *i.e.*, saloid-P, Al-P, Fe-P, Ca-P, total-P were significantly influenced by the imposed treatments. Phosphorus fractions was recorded highest in the treatment 100% RDP (T₄) and it is on par with 75% RDP (T₃) at different growth stages. The amount of inorganic P varied significantly and the distribution of added P into different fractions was in the order of Ca-P > Fe-P > Al-P > saloid-P.

Key words: Biofertilizers, P-fractions, Inorganic fertilizer Soil samples and Sorghum

Sorghum (*Sorghum bicolor* (L.) Moench) is the fifth world's most important nutritional cereal crop after wheat, maize, rice and barley and also the major staple food crop for millions of people in arid and semi-arid tropics and can be grown in different climates around the world in approximately 48 million hectares area annually. In India, it is grown in an area of 4.24 million hectares with a total production of 4.78 million tonnes and productivity of 1128 kg ha⁻¹ and in Andhra Pradesh, sorghum is cultivated in an area of 0.12 million hectares with an annual production of 0.37 million tonnes and productivity of 3070 kg ha⁻¹ (Ministry of Agriculture, Govt of India, 2020-21). Sorghum grain contains 10-12 per cent protein, 70 per cent carbohydrates, 3 per cent fats, vitamins, minerals and salts.

Phosphorus (P) is the second most important primary nutrient. Phosphorus is thought to be one of the most immobile, inaccessible and unavailable nutrient present in the soil. It is a constituent of ADP and ATP, two of the most important substances in life processes. An adequate supply of P is essential from the earliest stage of plant growth (Bertrand *et al.*, 2003).

Biofertilizers like phosphorus solubilizing bacteria (PSB) and Mycorrhiza (VAM) which solubilize and mobilize soil phosphates and make them available

to plants, can improve phosphorus use efficiency. PSB inoculum is used to increase phosphorus availability to crops. During phosphorus solubilization, some organic acids are produced which decrease the pH and acid phosphatases convert the organic phosphorus into inorganic form (Khan *et al.*, 2009).

In soils, the applied phosphate fertilizer enters into complex reactions with various constituents of soils such as Fe, Al, Ca and Mg which get quickly converted to less soluble or insoluble forms, as a result approximately only 20-25 per cent of the applied phosphatic fertilizer is utilized by the crop (Johnston and Syers, 2009). Thus, understanding the relationship between various forms of P, their interactions in soil and various factors influencing P availability to plants are essential for efficient P management in soil. In the above context, an experiment was conducted to assess the effect of biofertilizers in combination with inorganic phosphorus on yield and yield attributes of sorghum.

MATERIAL AND METHODS

A field experiment was conducted at Agricultural College Farm, Agricultural College, Bapatla during *Rabi*, 2022. MLSH- 296 was

seeded on claysoil with as pacing of 45 cm×15 cm in a randomized block design with three replications. The experimental soil was clay in texture, non-calcareous, slightly alkaline in nature (7.69), medium in organic carbon (0.41 dS m⁻¹), low in available nitrogen (210 kg ha⁻¹), high in available phosphorus (61.6 kg ha⁻¹) and available potassium (426 kg ha⁻¹) and sufficient in micro nutrients viz. Zn, Fe, Mn and Cu (1.66, 6.81, 3.86, 1.92, respectively).

The experiment consisted of ten treatment viz., T1-0% RDP, T2- 50% RDP, T3- 75% RDP, T4- 100% RDP, T5- 0% RDP + PSB, T6- 0% RDP + PSB + VAM, T7- 50% RDP + PSB, T8- 50% RDP + PSB + VAM, T9- 75% RDP + PSB, T10- 75% RDP + PSB + VAM. Well decomposed farmyard manure @ 10 t ha⁻¹ was applied 10 days before sowing. A recommended dose of nitrogen @ 100 kg ha⁻¹ was applied in the form of urea in two equal splits i.e., half as basal and half at 60 days after sowing. The recommended dose of P₂O₅ @ 60 kg ha⁻¹ was applied as per the treatments as basal just before sowing through single super phosphate. A recommended dose of K₂O 40 kg ha⁻¹ was applied as muriate of potash as basal application. Biofertilizers viz., PSB @ 1.25 L ha⁻¹ and VAM @ 12.5 kg ha⁻¹ were mixed with farm yard manure and applied as per the treatments. Recommended cultural practice and plant protection measures were taken through out the cropping season.

Phosphorus fractions

Saloid phosphorus (Sal-P)

One gram of soil sample in a 50 mL polyethylene centrifuge tube was treated with 25 mL of 1M NH₄Cl solution and shaken for 30 minutes. Supernatant solution after centrifugation was taken for saloid-P determination using the phosphomolybdate method by measuring the blue colour intensity in a spectrophotometer at 660 nm (Murphy and Riley, 1962).

Aluminium phosphorus (Al-P)

The residual soil left in the centrifuge tube after extraction of saloid-P was shaken for one hour with 50 mL 0.5 M NH₄F (pH 8.2) and centrifuged. The supernatant solution was collected and the residue was washed twice with a 25 mL portion of saturated NaCl and centrifuged. The washings were combined with NH₄F extract and the Al-P was estimated by the phosphomolybdate method.

Iron phosphorus (Fe-P)

The residual soil after Al-P extraction was treated with 50 mL of 0.1 M NaOH, shaken for 17 hours and the supernatant solution was collected after centrifugation. The residue was washed twice with a 25 mL portion of saturated NaCl and again centrifuged and the washings were combined with NaOH extract and Fe-P in the extract was estimated by phosphomolybdate method.

Calcium phosphorus (Ca-P)

After the extraction of Fe-P, 50 mL of 0.25 M H₂SO₄ was added to the residual soil and shaken for one hour, centrifuged and the extract was collected. The left over soil sample was washed twice with a 25 mL portion of saturated NaCl and again centrifuged. Finally, the washings were combined with H₂SO₄ extract. Then Ca-P was estimated by phosphomolybdate method.

Total phosphorus (Total P)

One gram of 0.5 mm sieved soil was weighed and transferred to a 250 mL Erlenmeyer flask to which 10 mL of di-acid mixture (HNO₃ and HClO₄ in 9:4 ratio) was added and digested on sand bath at 130° C till the dense fumes of HClO₄ were evolved. To the digested sample, 50 mL of distilled water was added, filtered by repeated washings and made up to 250 mL. An aliquot from this was used for estimation of total P (Jackson, 1967).

RESULTS AND DISCUSSION

The data recorded on different nutrient contents in sorghum plants at harvest stage of crop indicated that the treatmental effects were significant only on phosphorus and zinc content while other nutrient contents were not significantly influenced.

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Saloid - P (Sal-P)

Data regarding to the saloid phosphorus were significantly differed with the application of biofertilizers and different levels of inorganic phosphorus fertilizer at different growth stages of crop (Table 1 and fig 1).

The results shown that the saloid phosphorus was increased significantly with the application of biofertilizers along with inorganic P levels rather than the application of inorganic fertilizers alone. At flowering stage, the highest saloid phosphorus (20.4 mg kg^{-1}) was recorded in the treatment receiving 100% RDP (T_4) and it was on par with T_{10} (75% RDP + PSB+ VAM) (18.1 mg kg^{-1}) and it was significantly superior over all other treatments. Treatment T_9 (75% RDP+ PSB) (15.1 mg kg^{-1}) was on par with T_3 (14.4 mg kg^{-1}) (75% RDP) and T_8 (50% RDP+ PSB+VAM) (13.1 mg kg^{-1}) and significantly lowest (9.2 mg kg^{-1}) saloid phosphorus was recorded in 0% RDP (T_1). At harvest stage the highest saloid phosphorus (22.1 mg kg^{-1}) was recorded in the treatment receiving 100% RDP (T_4) was on par with T_{10} (75% RDP + PSB + VAM) (19.8 mg kg^{-1}) and it was significantly superior over all the other treatments. Treatment T_9 (75% RDP + PSB) (17.2 mg kg^{-1}) was on par with T_3 (75% RDP) (15.8 mg kg^{-1}) and significantly lowest saloid phosphorus was observed in 0% RDP (T_1) (10.1 mg kg^{-1}).

Saloid phosphorus was increased significantly with the application of increasing levels of phosphorus. These results are in agreement with the findings of Singaram and Kothandaraman (1993), reported greater concentration of saloid - P in soil with a water-soluble P source when PSB was combined with DAP and rock phosphate (RP), the saloid - P value increased marginally when compared to RP and DAP alone. They attributed it to P-fixing metallic cations complexing with organic acids released by P-solubilizing microorganisms, preventing P adsorption on soil particles. As a result, it aided in the extraction loosely bonded NH_4Cl extractable fraction of P.

Aluminium - P (Al-P)

Data regarding that effect of different levels of inorganic phosphorus and biofertilizers application on Al-P at different growth stages of crop are given in table 2 and figure 2.

Data clearly showed that effect of levels of inorganic phosphorus and biofertilizers application on Al-P at flowering and harvest stage was significant. The significant increase in Al-P was recorded with the application of inorganic fertilizers over biofertilizers alone. At flowering stage, the highest Al - P (82.0 mg kg^{-1}) was recorded with the application of 100% RDP (T_4) and it was superior over all other treatments. The significantly lowest (60.3 mg kg^{-1}) fixation of Al-P was

recorded in the treatment receiving without any application of inorganic phosphorus fertilizer *i.e.*, 0% RDP+ PSB + VAM (T_6) followed by (T_5) 0% RDP+ PSB (61.6 mg kg^{-1}) and (T_1) 0% RDP (62.3 mg kg^{-1}). The highest Al - P (83.2 mg kg^{-1}) was recorded with the application of 100% RDP (T_4) at harvest stage of the sorghum and it was significantly superior over all the other treatments. The significantly lowest (59.3 mg kg^{-1}) fixation of Al-P was recorded in the treatment (0% RDP+ PSB + VAM) (T_6).

The results on changes in Al-P fractions in soil at different growth stages of sorghum revealed that the application of P levels with or without biofertilizers significantly increased the Al-P fractions in soil at different growth stages of crop growth. Higher values of Al-P fractions were recorded in treatments involving only P levels without biofertilizers compared to application of P levels in combination with biofertilizers which might be due to the solubilisation of Al-P by P-solubilisers. The application of P fertilizers alone increased the Al-P content over control. This suggests that portion of added P was transformed into Al-P. Rao (1972) and Sheela (2006) reported that treatments comprising application of water-soluble P sources like SSP, favoured greater formation and accumulation of Al - P content of soil. This may be attributed to dissolution of aluminium of the clay in the acid produced as a result of hydrolysis of SSP in soil.

The lowest Al-P recorded in T_6 might be due to P-solubilizing microorganisms which are responsible mainly for organic acid production in soil leading to release of phosphorus from different pools either by lowering the pH, or by enhancing chelation of the cations bound to P or by competing with P for adsorption sites on the soil or by forming soluble complexes with metal ions associated with insoluble P (Ca, Al, Fe) and thus P is released. The lowering in pH of the soil suggested the release of organic acids by the P-solubilizing microorganisms via direct oxidation pathway that occurs on the outer face of the cytoplasmic membrane (Whitelaw 2000; Maliha *et al.*, 2004).

Iron - P (Fe-P)

Data pertaining to the Fe-P presented in table 3 and illustrated in figure 3 differed significantly due to the application of biofertilizers and different levels of inorganic phosphorus.

At flowering stage, the highest Fe-P was observed with the application of 100% RDP (T_4) (92.5 mg kg^{-1}) and the lowest (73.3 mg kg^{-1}) Fe-P was observed in the treatment receiving biofertilizers (0% RDP + PSB + VAM) (T_6). Fe-P was highest in the treatment receiving T_4 (94.8 mg kg^{-1}) followed by T_3 (92.4 mg kg^{-1}), T_2 (90.3 mg kg^{-1}) and T_9 (89.2 mg kg^{-1}) whereas significant lowest (74.1 mg kg^{-1}) Fe-P was observed in the treatment receiving (0% RDP+ PSB + VAM) (T_6) at harvest stage.

The results on changes in Fe-P fractions in soil at various growth stages of the sorghum crop revealed that applying P levels with or without biofertilizers dramatically improved the Fe-P fractions in soil at various growth stages of the crop. Higher values of Fe-P fractions were recorded in treatments that received only P levels without biofertilizers compared to treatments that received different P levels in combination with biofertilizers, which might be contributed to the solubilisation of Fe-P content over inorganics alone. According to Singh and Singh (1976), When compared to inorganic phosphorous fertilizers alone, the application of SSP resulted in the formation of more Fe-P during different periods of incubation in the soil as the soluble inorganic fertilizer would have been the reduction of Fe-P with the application of phosphate biofertilizers and farm yard manure. It is possible that this is due to the dissolution of iron oxide coatings with organic acids produced by P solubilizers, resulting in a decrease in Fe-P (Sheela, 2006).

Calcium-P (Ca-P)

The results furnished in the table 4 and figure 4 showed that application of different levels of inorganic phosphorus and biofertilizers was found to be significant at different growth stage of crop.

The highest Ca-P (254 mg kg^{-1}) was recorded with the application of 100% RDP (T_4) and it was superior over all the other treatments and the lowest fixation of Ca-P (218 mg kg^{-1}) was recorded in the treatment receiving without any application of inorganic phosphorus fertilizer (0% RDP+ PSB + VAM) (T_6) followed by T_5 (223 mg kg^{-1}) and T_1 (228 mg kg^{-1}) at flowering stage. At harvest stage, the Ca-P was highest in the treatment receiving 100% RDP (T_4) (249 mg kg^{-1}) and it was significantly superior over all the other treatments and significant lowest Ca-P (211 mg kg^{-1}) was recorded in the treatment receiving without any application of inorganic phosphorus fertilizer (0% RDP

+ PSB + VAM) (T_6) followed by T_5 (222 mg kg^{-1}) and T_1 (227 mg kg^{-1}).

Same as saloid, iron and aluminium phosphorus, calcium phosphorus increased with increased levels of phosphorus. The lowest Ca-P recorded in T_6 is due to P-solubilizing microorganisms are responsible mainly for organic acid production in soil which leads to release of phosphorus from different pools either by lowering the pH, or by enhancing chelation of the cations bound to P or by competing with P for adsorption sites on the soil or by forming soluble complexes with metal ions associated with insoluble P (Ca, Al, Fe) and thus P is released. The lowering in pH of the soil suggested the release of organic acids by the P-solubilizing microorganisms via direct oxidation pathway that occurs on the outer face of the cytoplasmic membrane (Whitelaw 2000; Maliha *et al.*, 2004).

Among the P fractions, the Al-P and Fe-P fractions contributed relatively less to the total P compared to the Ca-P. This could be mainly due to the fact that the solubility of Al-P and Fe-P decreases with increase in pH forming insoluble iron and aluminium phosphates, resulting in their reduced activity. Al-P was more than Fe-P. Singh *et al.* (2010) also reported higher concentration of Al-P fraction than that of Fe-P. They opined that, according to solubility product principle, soon after application of P fertilizer, Ca-P and Al-P are the immediate intermediate products and are comparatively more than Fe-P, due to relatively higher activities of Ca and Al ions than that of Fe ions.

Total - P

Data presented in table 5 and depicted in figure 5 indicating that the total P in the soil at different growth stages of crop significantly influenced by application of biofertilizers and different levels of inorganic phosphorus

The highest (667 and 662 mg kg^{-1}) total-P recorded with the application of 100% RDP (T_4) at flowering and harvest stages of the sorghum crop, respectively. The lowest total-P was recorded with the application of 0% RDP+ PSB + VAM (T_6) (559 and 519 mg kg^{-1} at flowering and harvest stages, respectively) without application of any inorganic phosphorus fertilizer. The total-P increased in all the treatments with crop growth stages except in T_1 , T_5 and T_6 where there was no inorganic P applied along

with organics and biofertilizers. There was a significant increase in total-P in the soil with the application of P

fertilizers. These results in accordance with the findings of Sheela (2006), Ranjit (2005).

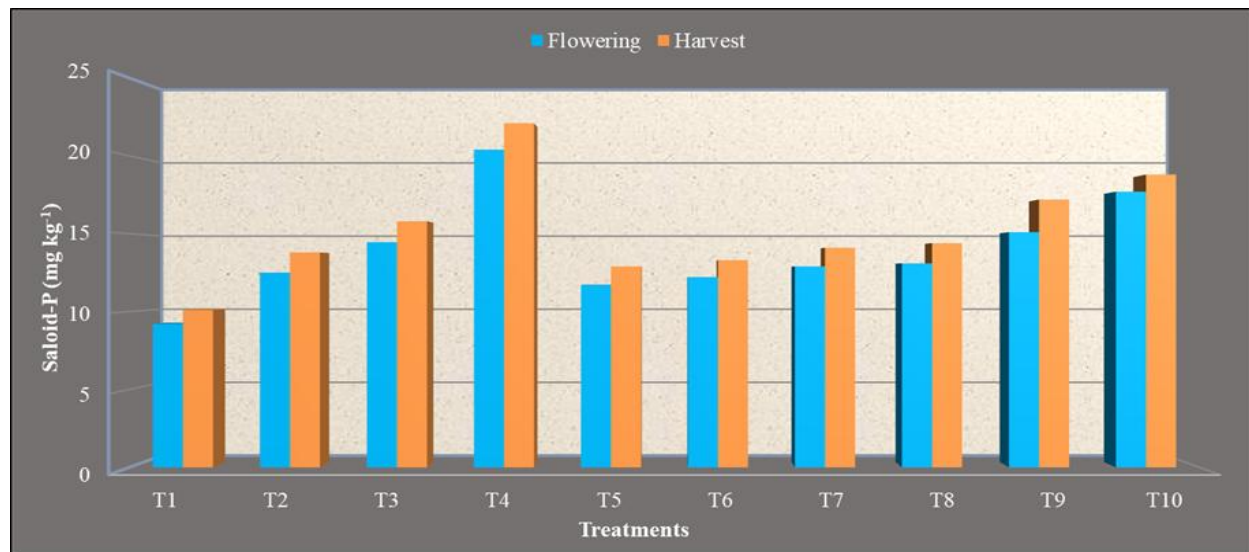


Fig1: Effect of biofertilizers in combination with inorganic phosphorus on saloid-P status of soil under sorghum

T₁-0% RDP T₄-100% RDP T₇- 50% RDP + PSB T₁₀-75% RDP +PSB +VAM
 T₂-50% RDP T₅- 0% RDP + PSB T₈- 50% RDP + PSB +VAM
 T₃-75% RDP T₆- 0% RDP + PSB + VAM T₉-75% RDP +PSB

CONCLUSION

Phosphorus fractions *i.e.*, saloid-P, Al-P, Fe-P and Ca-P, Total-P increased significantly with the application of 100% RDP (T₄) and it was significantly superior over all the other treatments. Significant lowest saloid phosphorus was observed with 0% RDP (T₁) without any application of inorganic phosphorus fertilizer but in Al-P, Fe-P and Ca-P, Total-P was observed in the treatment receiving only biofertilizers *i.e.*, (0% RDP + PSB + VAM) (T₆) followed by T₅ (0% RDP + PSB) at different growth stages. Among the P-fractions saloid-P, Al-P and Fe-P fractions contributed relatively less to the total P compared to the Ca-P. Hence, The potential of maximum P was bounded in Ca-P followed by Fe-P>Al-P> Sa-P fractions

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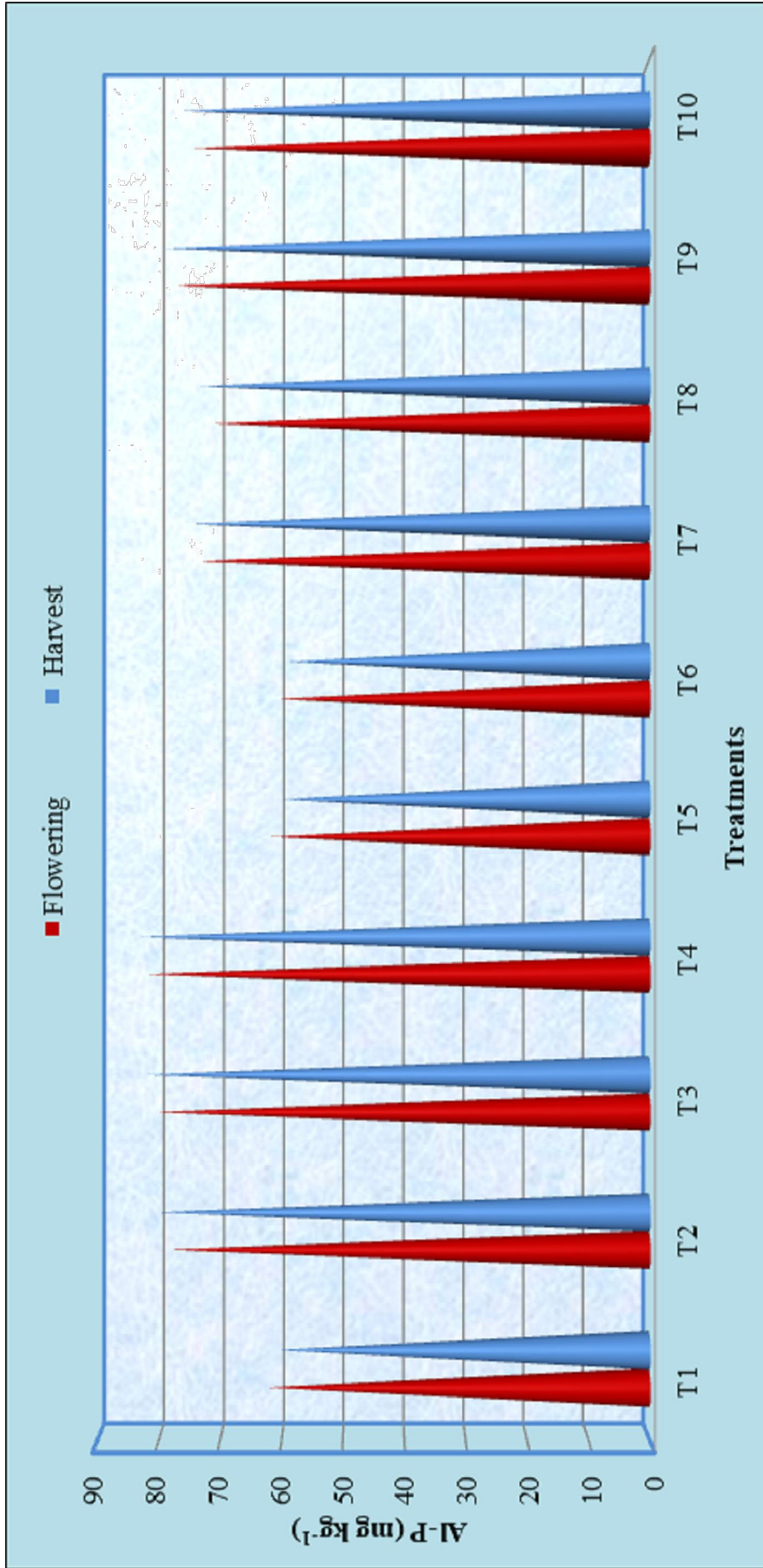


Fig2:Effect of biofertilizers in combination with inorganic phosphorus on Al- P status of soil under sorghum

T ₁ -0% RDP	T ₄ -100% RDP	T ₇ - 50% RDP + PSB	T ₁₀ -75% RDP +PSB + VAM
T ₂ -50% RDP	T ₅ - 0% RDP + PSB	T ₈ - 50% RDP + PSB + VAM	
T ₃ -75% RDP	T ₆ - 0% RDP + PSB + VAM	T ₉ -75% RDP +PSB	

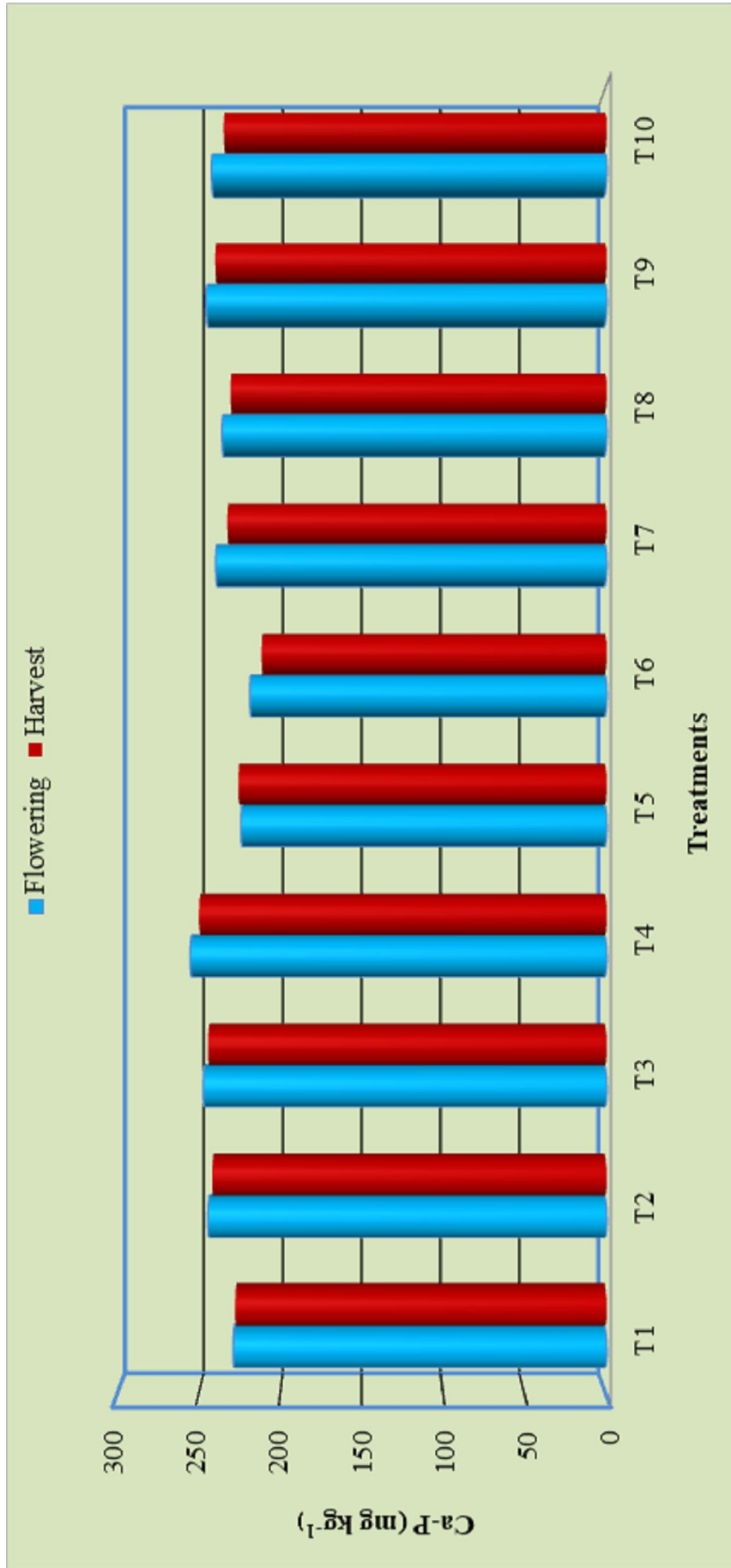


Fig4:Effect of biofertilizers in combination with inorganic phosphorus on Ca- P status of soil under sorghum

- T₁-0% RDP
- T₂-50% RDP
- T₃-75% RDP
- T₄-100% RDP
- T₅- 0% RDP + PSB
- T₆- 0% RDP + PSB + VAM
- T₇- 50% RDP + PSB
- T₈- 50% RDP + PSB +VAM
- T₉-75% RDP +PSB
- T₁₀-75% RDP +PSB +VAM

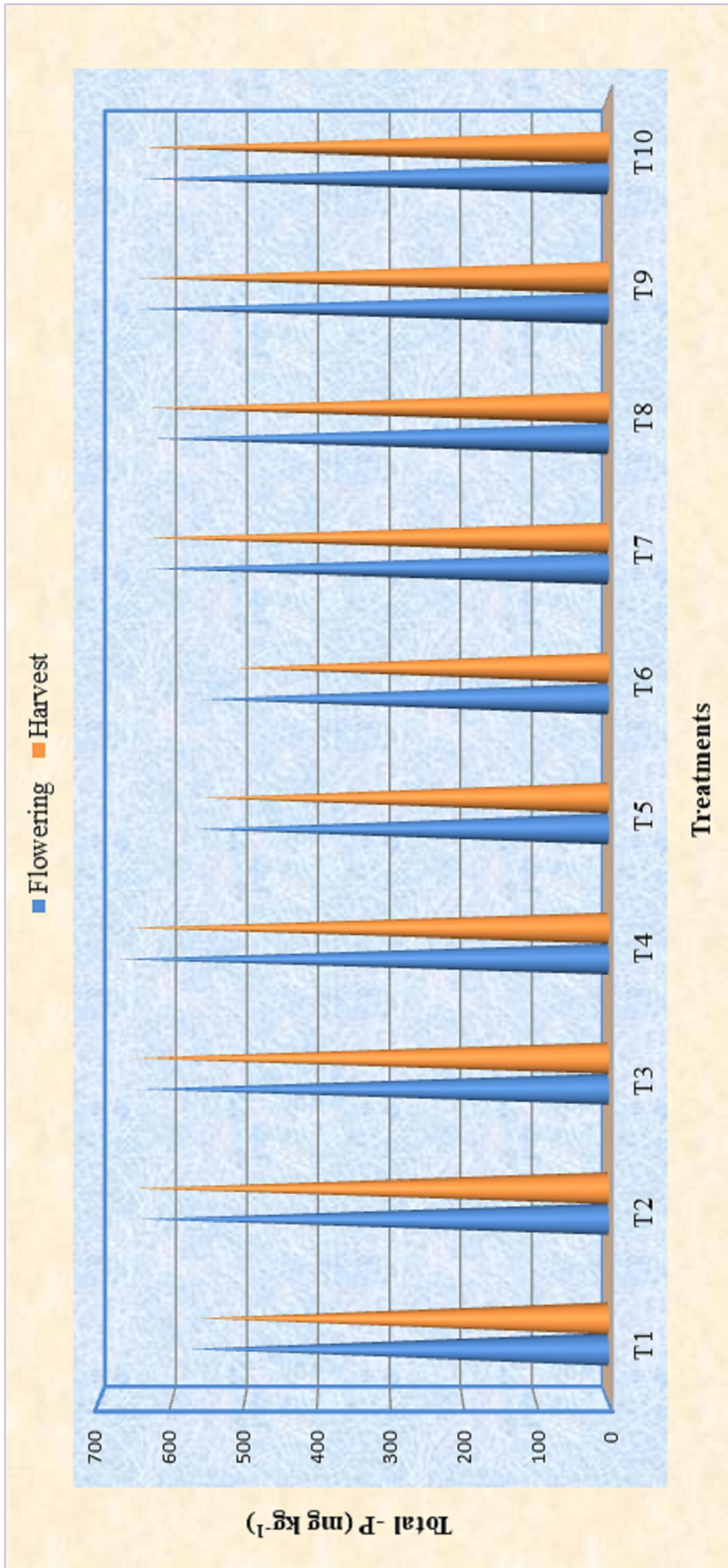


Fig5:Effect of biofertilizers in combination with inorganic phosphorus on total-P status of soil under sorghum

- T₁-0% RDP T₄-100% RDP T₇- 50% RDP + PSB T₁₀-75% RDP +PSB +VAM
- T₂-50% RDP T₅- 0% RDP + PSB T₈- 50% RDP + PSB +VAM
- T₃-75% RDP T₆- 0% RDP + PSB + VAM T₉-75% RDP +PSB

Table 1. Effect of biofertilizers in combination with inorganic phosphorus dynamics of P- fractions in soil under sorghum

Treatments	Saloid-P (mg kg ⁻¹)		Aluminium - P (mg kg ⁻¹)		Iron - P (mg kg ⁻¹)		Calcium - P (mg kg ⁻¹)	
	Flowering	Harvest	Flowering	Harvest	Flowering	Harvest	Flowering	Harvest
T ₁ : 0% RDP	9.2	10.1	62.3	60.7	78.2	76.4	228	227
T ₂ : 50% RDP	12.5	13.8	78.3	80.4	88.2	90.3	244	241
T ₃ : 75% RDP	14.4	15.8	80.6	82.3	89.5	92.4	246	244
T ₄ : 100% RDP	20.4	22.1	82.0	83.2	92.5	94.8	254	249
T ₅ : 0% RDP + PSB	11.7	12.9	61.6	59.8	76.7	74.8	223	222
T ₆ : 0% RDP + PSB + VAM	12.2	13.3	60.3	59.3	73.3	74.1	218	211
T ₇ : 50% RDP + PSB	12.7	14.1	73.4	74.8	83.4	85.2	239	232
T ₈ : 50% RDP + PSB + VAM	13.1	14.4	71.1	72.3	81.2	82.2	235	230
T ₉ : 75% RDP + PSB	15.1	17.2	77.3	79.3	87.3	89.2	245	239
T ₁₀ : 75% RDP + PSB + VAM	18.1	19.8	74.0	76.2	86.1	88.3	241	234
SEm (±)	0.72	0.84	3.35	3.37	3.62	4.01	7.01	6.94
CD (P = 0.05%)	2.31	2.68	10.7	10.7	10.9	12.2	22.4	22.2
CV (%)	8.99	9.54	8.04	8.00	7.47	8.15	5.11	5.15

Table 2. Effect of biofertilizers in combination with inorganic phosphorus on total-P status of soil under sorghum

Treatments	Total- P (mg kg ⁻¹)	
	Flowering	Harvest
T ₁ : 0% RDP	578	566
T ₂ : 50% RDP	646	654
T ₃ : 75% RDP	649	655
T ₄ : 100% RDP	677	662
T ₅ : 0% RDP + PSB	568	559
T ₆ : 0% RDP + PSB + VAM	559	519
T ₇ : 50% RDP + PSB	632	638
T ₈ : 50% RDP + PSB + VAM	628	634
T ₉ : 75% RDP + PSB	648	648
T ₁₀ : 75% RDP + PSB + VAM	642	646
SEm (±)	25.2	28.7
CD (P = 0.05%)	75.7	86.8
CV (%)	7.02	8.03

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