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Radiotracer Studies on the Interaction of Phosphorus with Zinc in Okra Using Double Tagging Technique

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Abstract: *A pot experiment was conducted using P^{32} tracer technique at the Radioisotope Laboratory, Tamil Nadu Agricultural University, Coimbatore, during 2013-2014 to study the yield response of okra to P fertilization and its interaction with Zn. To study the fertilizer Zn use efficiency under varying levels of P, double tagging technique was employed using ^{65}Zn and ^{32}P as radiotracers. The treatments included four levels of P (0, 50, 100 and 150 kg P_2O_5 ha^{-1} applied as ^{32}P labeled single superphosphate) and two levels of FYM (0 and 25 t ha^{-1}), replicated thrice in a completely randomized design. Zinc sulphate, labeled with ^{65}Zn, was applied as common application to the all pots @ 25 kg $ZnSO_4$ ha^{-1}. Initial and postharvest soil analysis and plant analysis for P^{32} activity, ^{65}Zn activity and nutrients content were carried out. Application of P at different levels significantly and positively increased the dry matter yields of both fruit and shoot of okra, up to the P application level of 100 kg P_2O_5 ha^{-1}. 'A' value of P, was influenced significantly by P levels up to P_{100}. The 'A' values for Zn declined with increasing P levels. The %ZnUE for the applied $ZnSO_4$ increased significantly with the level of P application up to the 100 kg P_2O_5 ha^{-1} level.*

Keywords: *A value, Radiotracer technique, Specific activity, %Pdff, %Pdfs*

I. Introduction

Okra is an important vegetable crop grown in the tropics and sub-tropics of the world. Leading producers of okra are India, Nigeria, Sudan, Iraq and Pakistan. In the tropics, soil fertility is one of the major constraints to crop productivity. Phosphorus is often described as the second limiting nutrient in crop production after N (Thompson and Throch, 1975). Hence application of P fertilizers has become necessary under most condition for increasing crop yields. Phosphorus fertilization practices help to insure an adequate supply of P during peak growth demand periods of a crop. Interactions between nutrients in higher plants occur when the supply of one nutrient affects the absorption, distribution or function of another element. Phosphorus and Zn interact both in plants and soils and hence may affect the availability and utilization of each other. The effects of P on Zn or the effects of Zn on P are interactions significant to profitable crop production.Orabi*et al*. (1985) showed that the relationship between the two elements was positively correlative. Several field and greenhouse studies (Olsen 1972; Robson and Pitman, 1983) have suggested that high levels of available P in soil may induce Zn deficiency in various crops. Higher P availability in soil significantly decreased Mn, Zn and Cu concentrations in eight cultivars of barley (Zhu *et al*., 2002).

An estimate of interactions among plant nutrients could be made by determining whether any change in plant growth and/or concentration of nutrients in plants had taken place as a result of variation in the level of supply of one of the interacting elements. Tracer techniques enable tracing accurately and precisely nutrient elements taken up by the plants and also help study accurately the effect of one element upon the absorption of another. Double tagging studies with two different radioisotopes as ^{32}P (a pure beta emitter with a relatively short half-life) and ^{65}Zn (a beta and gamma emitter with a relatively long half-life) to elucidate information on interaction between P and Zn have not been attempted so far.

II. Materials and Methods

Surface soil was collected in bulk from a farmer's field in Annur Block of Coimbatore District, Tamil Nadu and used for potculture experiment. The bulk soil collected was air-dried in shade, gently powdered with a wooden mallet, and sieved to pass through a 2 mm sieve. Representative subsamples were analyzed for the mechanical composition and physical and chemical properties as per the standard procedures. Post-harvest soil samples were also subjected to chemical analysis. The processed soil samples were filled in glazed ceramic pots (@10 kg pot^{-1}) having an inner diameter of 28 cm and a height of 28 cm. There were 24 pots in all, comprising eight treatments and three replications. Radiotracer ^{32}P was applied to all the pots, except control as per the treatments. The treatments included four levels of P (0, 50, 100 and 150 kg P_2O_5 ha^{-1} applied as ^{32}P labeled single superphosphate) and two levels of FYM (0 and 25 t ha^{-1}). Zinc sulphate, labeled with ^{65}Zn, was applied as common application to the all pots @ 25 kg $ZnSO_4$ ha^{-1}.

Carrier free ^{32}P was obtained as orthophosphoric acid in dilute hydrochloric acid medium from the Board of Radiation and Isotope Technology, Mumbai and used for preparing labeled superphosphate. The tagging level was 11.1 MBq g^{-1} of P. The physical half-life ($T_{½}$) of ^{32}P is 14.3 days. It decays into ^{32}S by emitting negatrons (β^-) of E_{max} 1.71 MeV. Using the principle of isotope dilution technique, ^{65}Zn-tagged $ZnSO_4$ fertilizer was prepared, taking calculated quantities of analar grade $ZnSO_4$ and ^{65}Zn radioisotope. The radioisotope of ^{65}Zn was obtained from the Board of Radiation and Isotope Technology, Mumbai as $ZnCl_2$ in dilute HCl acid medium. The tagging level employed was 37 MBq g^{-1} of Zn. Zinc-65 emits β^+ rays (1.5%) of E_{max} 0.325 MeV and γ rays of energy 1.11 MeV (45%) by electron capture (98.5%). The $T_{½}$ of ^{65}Zn is 245 days.

Seeds of okra variety ArkaAnamika were sown in each pot @ three seeds per pot. Routine cultural practices and fertilization were adopted in raising the crop. The okra fruits were harvested at different times according to the maturity of fruits. At harvest, fresh weights and dry weights were recorded. The plant materials, after processing and proper subsampling, were analyzed for nutrients contentas per standard procedures. Plant P was determined by Vanadomolybdate yellow color method by Olsen and Sommers (1982) and Zn was analyzed by Atomic Absorption Spectrometry by Lindsay and Norvell (1978). The radioassay for ^{32}P was done following McKenzie and Dean (1948) procedure and ^{32}P activity was determined in proportional counter [Type PCS 13 B of Electronics Corporation of India Ltd., Hyderabad; counting efficiency 21.28%]. Radiochemical separation of Zn in the triple acid extract was done by precipitating Zn as Zn $(OH)_2$ by adding necessary quantity of NaOH. The precipitate was then filtered, the filter paper disc with the precipitate was transferred to a planchet and counted for ^{65}Zn activity in a NaI (Tl) crystal solid scintillation counter [Type GR 611 M of Nucleonix, Hyderabad; counting efficiency 33.2%]. Following equations were used to interpret the radio assay data.

$$\text{Bq} = \text{dps} = \frac{\text{Corrected count per second}}{\text{Counting efficiency (\%)}} \times 100$$

$$\text{Specific activity (dps mg}^{-1}) = \frac{\text{Disintegration rate in sample (dps)}}{\text{P content in sample (mg)}}$$

$$\%\text{Pdff} = \frac{\text{Specific activity of plant sample}}{\text{Specific activity of standard}} \times 100$$

$$\%\text{Pdfs} = 100 - \%\text{Pdff}$$

[Pdff = Phosphorus in the plant derived from applied fertilizer]
[Pdfs = Phosphorus in the plant derived from soil]

$$\text{Uptake from applied fertilizer (mg pot}^{-1}) = \frac{\%\text{Pdff}}{100} \times \text{Total P uptake (mg pot}^{-1})$$

$$\text{‘A’ value (mg 100g}^{-1}\text{ soil)} = \frac{\%\text{Pdfs}}{\%\text{Pdff}} \times \text{P applied (mg 100g}^{-1}\text{ of soil)}$$

$$\%\text{ P use efficiency} = \frac{\text{Uptake from applied source (mg P pot}^{-1})}{\text{P applied through fertilizer (mg P pot}^{-1})} \times 100$$

In a similar vein, the % Zn use efficiency was calculated.

III. Results and Discussion

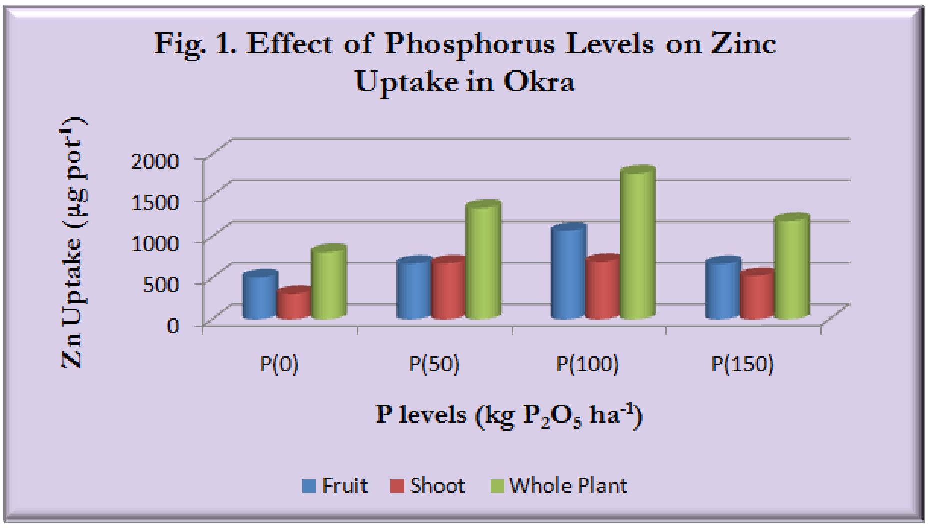
The initial analysis of the experimental soil revealed that the texture was clay loam with the pH of 8.08 and electrical conductivity of 0.26 dSm^{-1}. The soil was low in available N (217.70 kg ha^{-1}) and low in available P (8.7 kg ha^{-1})and high in available K (660 kg ha^{-1}). The DTPA extractant Zn was 1.22 $\mu g\ g^{-1}$. Application of P at different levels significantly and positively increased the dry matter yields of both fruit and shoot of okra, up to the P application level of 100 kg P_2O_5 ha^{-1}. An adequate supply of phosphorus has been associated historically with increased root growth which in turn resulted in better uptake of nutrients and water. This could be a possible explanation for the increase in yield with phosphorus application. Many studies have shown that application of phosphatic fertilizers generally has a great impact on crop yield because the P deficiency limits the response of plants to the other nutrients (Jones,1982; Nisar*et al.*,1992; Alaam*et al.*, 2002).The availability of P in soil consequent to fertilizer P application, determined as 'A' value, was influenced significantly by P levels up to $P_{100.}$ The 'A' value increased significantly from 1.52 mg $100g^{-1}$ (P_{50}) to 2.06 mg $100g^{-1}$ at P_{100}. The availability of Zn decreased significantly with the increase in the application of P. When No P was applied the highest availability of Zn was observed (9.46 µg g^{-1}). This trend observed in the present investigation reveals the antagonistic interaction between Zn and P. This was accentuated by the fact that the available Zn status determined by tracer technique employing the radiotracer ^{65}Zn also indicated a conspicuous decline 'A' values with increasing P levels (Table 1).

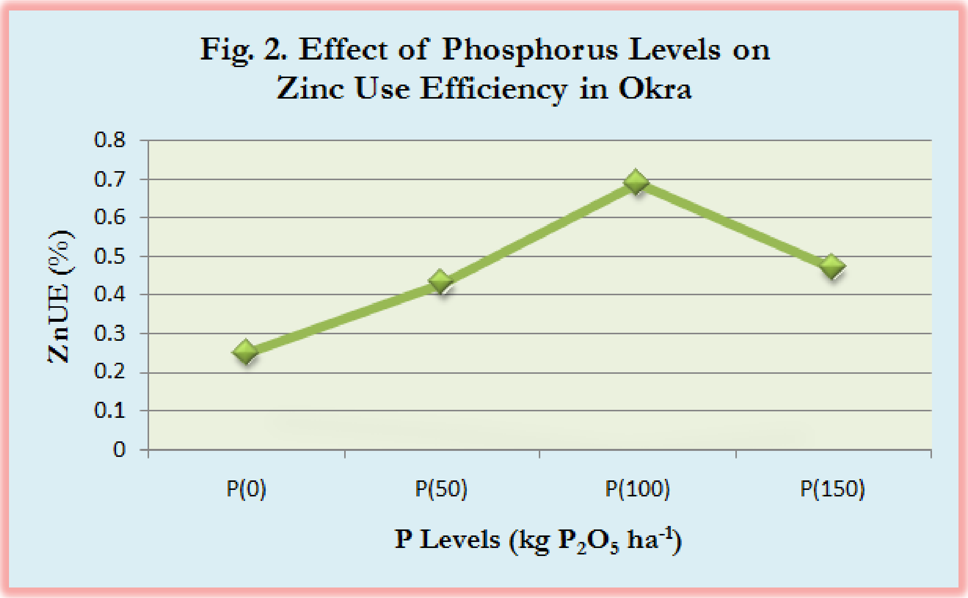
Table 1: Effect of Treatments on 'A' Value of Zinc (mg Zn $100g^{-1}$)

Levels of P ($Kgha^{-1}$)	Farm Yard Manure (t ha^{-1})		Mean
	M_0	M_{25}	
P_0	1.34	2.46	**1.90**
P_{50}	1.64	2.17	**1.91**
P_{100}	1.13	1.50	**1.32**
P_{150}	1.07	1.59	**1.33**
Mean	**1.30**	**1.93**	
Source		**SE_d**	**CD(p=0.05)**
P Levels(P)		0.13	0.28
FYM(M)		0.09	0.20
PxM Interaction		0.19	NS

The P uptake by fruit and shoot increased significantly with the P levels up to P_{100}. The %Pdff increased from 35.86 in P_{50} to 52.61 in P_{150}in fruit. In shoot it is increased significantly from 49.96 (P_{50}) to 76.72 (P_{150}). In fruit, a numerical decrease in %Pdfs was observed with the increase in P levels and in shoot, significantly decreased with the level of P application from 50.05 at P_{50} to the 23.28 at P_{150}. The %PUE decreased significantly from 19.09 (P_{50}) to 10.25 (P_{150}). The %PUE at P_{100} (18.85) was on par with that at P_{50}.

Application of P in increasing levels decreased the Zn content in fruit, obviously due to the negative interaction between Zn and P. This might be attributed to P-induced deficiency and the results are in agreement with that of Adams (1980) on brinjal.Total Zn uptake by whole plant was significantly influenced by P levels (Fig 1). The uptake increased from 840.83 µg pot^{-1}(P_0) to 1767.84 µg pot^{-1} (P_{100}), beyond which there was a sharp decline in total Zn uptake at P_{150} (1193.91 µg pot^{-1}).With regard to %Zndff, it was observed that in both fruit and shoot there was a general increasing trend with the increase in the level of P applied. Since the applied $ZnSO_4$ fertilizer was water soluble, it must have furnished to plants readily available Zn ions in soil solution as suggested by Raja Rajan (1991). The %Zndfs was not influenced by the application of different levels of P. Total fertilizer Zn uptake in okra plant increased significantly with the increase in the level of P applied up to the level of P_{100}. From 315.15 µg pot^{-1} at P_0 the uptake increased to864.88 µg pot^{-1} at P_{100}.The %ZnUE in the okra whole plant ranged from 0.25 to 0.69 in case of P treatments. Among the P levels the highest %ZnUE was recorded in the treatment P_{100}. Since the %Zndff and fertilizer Zn uptake in both fruit and shoot and the total fertilizer Zn uptake in the whole plant were all highest in the treatment P_{100}, this treatment resulted in the highest %ZnUE.





IV. Conclusion

Both positive and negative interactions are observed in the present investigation. The availability of Zn, as determined by radiotracer techniques, indicated conspicuous decline in 'A' values with increasing P levels. The %ZnUE for the applied $ZnSO_4$ increased significantly with the level of P application up to the 100 kg P_2O_5 ha^{-1} level.

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