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Inorganic P fractions in rhizosphere and yield of onion amended with organics and inorganic P in soybean (*Glycine max* L.): onion (*Allium cepa* L.) cropping system

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Abstract
A field experiment was conducted with soybean-onion cropping sequence in a high P Alfisol to study the response to P levels either alone or in combination with PSB, biochar, humic acid and citric acid to study the direct, residual and cumulative effects of the treatments imposed on inorganic P fractions and yield of onion. Among the inorganic P fractions estimated, saloid-P is the smallest pool and Ca-P is the largest pool. Cumulative application of 30 kg P_2O_5 ha^{-1} along with biochar resulted in mean inorganic P fractions viz., saloid-P, Al-P, Fe-P, occluded-P and Ca-P of 3.11, 11.1, 50.5, 16.1 and 397 against 2.68, 9.3, 39.0, 14 and 358 mg kg^{-1} without organics. Among the organics, biochar application to soybean was found to be benefited more due to the cumulative application of 30 kg P_2O_5 ha^{-1} resulting in a mean bulb yield of 24.1 t ha^{-1} against 20.0 t ha^{-1} due to the residual effect, the yield response being 20.5 per cent. There was significant positive correlation between Ca-P and onion yield (r = 0.889**) in the high P Alfisol.

Keywords: inorganic P fractions, inorganic P, biochar, residual effect, cumulative effect, onion

Introduction
Phosphorus is an important nutrient especially for pulses as a high phosphorus supply is needed for nodulation. It also influences N availability through N fixation. Phosphorus plays a pivotal role for the structure and regulatory functions in photosynthesis, root development, energy conservation and transformation, carbon metabolism, redox reactions, enzyme activation and inactivation, signaling and nucleic acid synthesis (Vance *et al*., 2003) [1]. Adsorption/desorption and precipitation/dissolution equilibria control the concentration of P in the soil solution and, thereby, both its chemical mobility and bioavailability. Biochar, a carbonised and highly porous carbon rich biomass obtained from sustainable sources and sequestered in soils to sustainably enhance their agricultural and environmental value under present and future management, has been reported to increase nutrient availability in soils through increased cation retention and decreased phosphate adsorption.
Organic matter additions were found to mobilize the fixed phosphates in the soil thus increasing the available P to crops (Venkateswarlu, 2000) [1]. The available information on the role of organics like PSB, biochar, humicacid and citricacid in combination with inorganic P sources is scanty. Hence, the present work was aimed at studying the effect of combined application of organic and inorganic sources of P on inorganic P fractions of rhizosphere soils of soybean.

Materials and Methods
During *kharif* (soybean) 2012, the experiment was laid out in split plot design consisting 3 main levels of inorganic P (0, 30 and 60 kg P_2O_5 ha^{-1}) and 5 sub levels of organics (no organics, PSB, biochar, humic acid and citric acid). In *rabi* (onion) 2012-13, the experiment was laid out in split-split plot design, with 2 sub – sub levels (no application, application of best combination from *kharif* to study the residual and cumulative effects respectively). For this all the plots were divided into two equal halves. For one half, neither inorganic P nor organics were applied to know the residual effect on onion grown during *rabi* after harvest of soybean crop. In another half, the best combination from *kharif* was applied to study the cumulative effects.
The experimental soil was sandy clay loam in texture, slightly alkaline (pH 7.64) in reaction, non-saline (0.195 dS m^{-1}) in nature and medium in organic carbon (0.57 %). The soil was low

in available nitrogen (177 kg N ha^{-1}), high in available phosphorus (29.9 kg P ha^{-1}) and potassium (449 kg K ha^{-1}) (Table 1).

The plants were uprooted from the plot without disturbance and then the plant roots were shaken softly to remove the root-zone soils. The soil adhering to the surroundings of a root was called ‘rhizosphere soil’ in general, and the soil shaken off is ‘root-zone soil’. Collected soils were sieved in a 1 mm mesh removing root hair as much as possible. The soil samples from individual treatmental plot were collected at flowering and at harvest and assayed for inorganic P-fractions (Saloid-P, Al-P, Fe-P, Ca-P and Occluded-P) in the soil (Chang and Jackson, 1967).

Yield of onion was computed and the data on various parameters were statistically analysed following the method of variance for split and double split designs and the significance was tested by ‘F’ test (Snedecor and Cochran, 1967). Critical difference for comparing the treatment means and their interactions were calculated at 5 per cent level of probability.

Table 1: Salient soil characteristics of experimental site

S.No.	Name of the property	Value
I. Physical properties		
a.	Textural fraction	
	Sand (%)	72.04
	Silt (%)	7.4
	Clay (%)	20.56
b.	Textural class	Sandy clay loam
II. Physico-chemical analysis		
a.	Soil reaction (pH)	7.64
b.	Electrical conductivity (dSm^{-1})	0.195
III. Chemical properties		
a.	Organic carbon (%)	0.57
b.	Available Nitrogen (kg ha^{-1})	177
c.	Available phosphorus (kg P ha^{-1})	29.9
d.	Available potassium (kg K ha^{-1})	449
e.	Saloid-P (mg kg^{-1})	2.21
f.	Al-P (mg kg^{-1})	7.23
g.	Fe-P (mg kg^{-1})	35.2
h.	Ca-P (mg kg^{-1})	339
i.	Occluded-P (mg kg^{-1})	54.0

Results and Discussion

Inorganic P Fractions in Rhizosphere Soil

Saloid-P: At 45 DAT of onion crop, when organics were applied at 30 kg P_2O_5 ha^{-1}, the residual effect was significantly higher due to biochar with saloid-P of 2.36 mg kg^{-1} which was higher by 20 per cent when organics were not applied. However, humic and citric acid were at a par with biochar. While at higher levels of inorganic P biochar recorded significantly higher saloid-P of 3.52 mg kg^{-1} against 3.04 mg kg^{-1}. The response being 15.8 per cent, other organics were at a par with each other. When cumulative effect was considered, at 0 kg P_2O_5 ha^{-1} biochar was significantly higher with a saloid-P of 3.38 mg kg^{-1}. At 30 and 60 kg P_2O_5 ha^{-1}, biochar recorded significantly higher saloid-P of 4.25 and 5.66 mg kg^{-1} respectively (Table 2). Adsorption of Ca via cation exchange (Bolan *et al*., 1990, Rajan *et al*., 1996) [1, 11] or the complexation of Ca by an organic ligand such as citrate may lead to dissolution of phosphates. Such enhanced dissolution of Ca – phosphate can also occur as a consequence of removal of P or Ca from the soil solution by plant uptake or by the supply of H^+ or organic ligands that can complex Ca (Hinsinger, 1998., Nye, 1986 and Jurinak *et al*., 1986) [6, 10, 9].

There was a reduction to the extent of 11.3 per cent in saloid-P at harvest of onion as against at 45 DAP. This could be due to the depletion in the soluble saloid P by the crop or due to the reason that it may enter into non labile pools like Ca-P.

Al-P: When organics were applied alone the residual effects was significantly higher due to citric acid with Al-P of 8.6 mg kg^{-1} which was higher by 19.4 per cent when organics were not applied at 45 DAT of onion crop. While at higher levels of inorganic P the biochar recorded significanly higher Al-P of 12.9 and 12.5 kg ha^{-1} respectively, at 30 and 60 kg P_2O_5 ha^1 against no organics. At 60 kg P_2O_5 ha^{-1}, all the organics were at a par with each other. When cumulative effects were considered, at 0 kg P_2O_5 ha^{-1} humic acid recorded significantly higher Al-P of 12.5 mg kg^{-1} against 7.7 mg kg^{-1} when no organics were applied. But it was on par with citric acid. At higher levels of 30 and 60 kg P_2O_5 ha^{-1}, biochar proved to be superior with Al-P of 15.6 and 17.1 mg kg^{-1} respectively. However, at 60 kg P_2O_5 ha^{-1}, except PSB all other organics and no organic treatment were at a par (Table 3). Citric acid and humic acid resulted in a higher Al-P which might be due to the rhizosphere acidification that would enhance Al-P pool on one side, or indirectly by increasing root exudation that in turn result in reduced pH on the other side.

Al-P at harvest stage was influenced significantly by inorganic P levels, organics, mode of effects and their interaction. There was a decrease in Al-P by 13.1 per cent at harvest when compared to 45 DAP. The decrease in Al-P could be due to the dissolution by root exudates in the rhizosphere.

Table 2: Residual and cumulative effects of organics, inorganic P and their interaction on saloid-P (mg kg^{-1}) in rhizosphere soil at 45 DAT of onion

Organics- Sub treatments	Inorganic P levels (kg P_2O_5 ha^{-1}) -Main treatments: 0			30			60			Means		Mean for Organics
	Residual	Cumulative	Mean	Residual	Cumulative	Mean	Residual	Cumulative	Mean	Residual	Cumulative	
No organics	2.12	2.01	2.06	2.88	3.47	3.18	3.04	4.44	3.74	2.68	3.31	2.66
PSB	2.15	2.89	2.52	2.96	3.45	3.21	3.23	3.93	3.58	2.78	3.42	3.10
Biochar	2.36	3.38	2.87	3.46	4.25	3.86	3.52	5.66	4.59	3.11	4.43	3.77
Humic acid	2.34	3.21	2.78	3.17	3.93	3.55	3.45	4.74	4.10	2.99	3.96	3.48
Citric acid	2.27	3.13	2.70	3.21	3.73	3.47	3.21	4.59	3.90	2.90	3.82	3.36
Mean	2.25	2.92	2.58	3.14	3.77	3.45	3.29	4.67	3.98	2.89	3.79	
	MT	ST	SST	MT at ST	MT at SST	ST at MT	ST at SST	SST at MT	SST at ST	SST at MT,ST	ST,SST at MT	MT at ST,SST
SEm±	0.07	0.07	0.04	0.13	0.08	0.12	0.09	0.06	0.08	0.14	0.09	0.02
CD (P=0.05)	0.27	0.20	0.11	NS	0.29	NS	0.26	0.19	0.24	0.42	0.33	0.42

Table 3: Residual and cumulative effects of organics, inorganic P and their interaction on Al-P (mg kg^{-1}) in rhizosphere soil at 45 DAT of onion

Organics-Sub treatments	Inorganic P levels (kg P_2O_5 ha^{-1}) -Main treatments									Means		Mean for Organics
	0			30			60					
	Residual	Cumulative	Mean	Residual	Cumulative	Mean	Residual	Cumulative	Mean	Residual	Cumulative	
No organics	7.2	7.7	7.4	10.1	12.5	11.3	10.5	16.3	13.4	9.3	12.2	10.7
PSB	6.8	9.9	8.3	10.3	11.8	11.1	11.4	14.4	12.9	9.5	12.0	10.8
Biochar	8.1	10.9	9.5	12.9	15.6	14.2	12.5	17.1	14.8	11.1	14.5	12.8
Humic acid	8.3	12.5	10.4	11.5	14.3	12.9	11.6	16.8	14.2	10.4	14.5	12.5
Citric acid	8.6	11.4	10.0	11.6	13.7	12.6	11.6	16.8	14.2	10.6	14.0	12.3
Mean	7.8	10.5	9.1	11.3	13.6	12.4	11.5	16.3	13.9	10.2	13.4	
	MT	ST	SST	MT at ST	MT at SST	ST at MT	ST at SST	SST at MT	SST at ST	SST at MT,ST	ST,SST at MT	MT at ST,SST
SEm±	0.2	0.2	0.1	0.4	0.2	0.4	0.3	0.2	0.2	0.4	0.3	0.1
CD (P=0.05)	0.7	0.6	0.3	NS	0.8	NS	0.8	0.5	0.7	1.2	1.0	1.3

Fe-P: Fe-P at 45 DAT was influenced significantly by the individual effects of inorganic P, organics, mode of effects; however, their interactions were not significant. The proportion of Fe-P in total inorganic P at 45 DAT was 9.54 per cent.

When organics were applied alone the residual effect was significantly higher due to biochar with Fe-P at harvest stage of onion crop was 39.4 mg kg^{-1}. It was higher by 21.1 per cent when organics were not applied. While at higher levels of inorganic P the same biochar showed significantly higher Fe-P of 42.9 and 48.2 mg kg^{-1} respectively at 30 and 60 kg P_2O_5 ha^{-1}. The buildup in Fe-P was 25.8 and 36.2 per cent respectively. When cumulative effect was considered, at 0 kg P_2O_5 ha^{-1}, biochar recorded significantly higher Fe-P of 45.3 mg kg^{-1}, which was on par with humic acid. At 30 and 60 kg P_2O_5 ha^{-1}, the same biochar resulted in significantly higher Fe-P of 48.4 and 52.2 mg kg^{-1} respectively. The per cent increase being 19.5 and 25.8 per cent (Fig 1).

Ca-P: The Ca-P occupies the major proportion of 83.4 per cent out of the total inorganic P at 45 DAT of onion. Individual application of inorganic P, organics, and mode of effects was significant on Ca-P. However, their interactions were not significant (Table 4).

Application of 30 kg P_2O_5 ha^{-1} to soybean across organics and mode of effects increased the mean Ca-P to 379 mg kg^{-1} *i.e* 3.5 per cent as against 366 mg kg^{-1} in the control. However, the mean Ca-P obtained due to 30 and 60 kg P_2O_5 ha^{-1} were at a par. Jain and Sarkar (1979) conformed that the significant increase in saloid-P, Al-P and Ca-P as a result of inorganic fertilizations and organic amendments was attributed to the transformation of applied P at a faster rate into saloid-P, Al-P and Ca-P in the first instance and then to Fe-P with time.

There was a reduction in the Ca-P content to the tune of 4.1 per cent at harvest when compared to 45 DAT. The Ca-P at harvest of onion crop was influenced significantly by individual application of inorganic P, organics, mode of effects, while among the interactions, the inorganic P and mode of effect were significant in influencing the Ca-P. Indiati and Sharpley (1998) found that added P enhanced saloid P by 1-26 per cent (mean 9%), Al-P by 7-55 per cent (mean 32%), Fe-P by 1-70 per cent (mean 31%) and Ca-P by 6-86 per cent.

Table 4: Residual and cumulative effects of organics, inorganic P and their interaction on Ca-P (mg kg^{-1}) in rhizosphere soil at 45 DAT of onion

Organics-Sub treatments	Inorganic P levels (kg P_2O_5 ha^{-1}) -Main treatments									Means		Mean for Organics
	0			30			60					
	Residual	Cumulative	Mean	Residual	Cumulative	Mean	Residual	Cumulative	Mean	Residual	Cumulative	
No organics	328	374	351	335	383	359	344	394	369	336	384	360
PSB	347	389	368	364	392	378	374	390	382	361	391	376
Biochar	375	379	377	384	436	410	395	414	404	385	410	397
Humic acid	354	391	373	363	394	379	374	429	402	364	405	384
Citric acid	337	385	361	344	393	369	354	404	379	345	394	370
Mean	348	384	366	358	400	379	368	406	387	358	397	
	MT	ST	SST	MT at ST	MT at SST	ST at MT	ST at SST	SST at MT	SST at ST	SST at MT,ST	ST,SST at MT	MT at ST,SST
SEm±	3	4	2	7	4	7	6	4	5	9	6	1
CD (P=0.05)	12	12	7	NS	NS	NS	NS	NS	NS	NS	NS	NS

Occluded-P: Occluded-P at 45 DAT of onion accounted for 3.6 per cent.

At harvest, In the residual effect, all the organics were found to be significant over no organics. Among the organics, biochar was found to be superior with a mean occluded-P of 16.1 mg kg^{-1} against 14.0 mg kg^{-1} in no organic treatment. However, biochar and humic acid were at a par. In the cumulative treatment also similar response to the organics was exhibited, the mean occluded-P fraction increasing from 15.9 mg kg^{-1} in the no organic treatment to 20.0 mg kg^{-1} with biochar. Cumulative effect showed significant increase in the mean occluded-P to 16.5 mg kg^{-1}, which was higher by 13.8 per cent over the residual effect that showed 14.5 mg kg^{-1} in onion crop at harvest stage (Fig 1).

Rokima and Prasad (1991) found that most of the added P was transformed into organic P and Ca-P (40 to 45% of total P) and very little was transformed to water soluble P (0.23 to 0.34%), saloid-P (0.64 to 1.69%), Al-P (0.94 to 1.40%) and Fe-P (0.49 to 1.34%). The increase in content of Ca-P was due to conversion of insoluble tricalcium phosphate to more soluble mono or dicalcium phosphate besides mineralization of organic P. Large amount of P applied as fertiliser enters into immobile pools through precipitation reactions with highly reactive Al and Fe in acid soil and Ca in calcareous or normal soil (Gyaneshwar *et al*., 2002 and Hao *et al*., 2002) [4, 5].

Bulb yield of onion

Among the organics, biochar application to soybean was found to be benefited more due to the cumulative application of resulting in a mean yield of 24.1 t ha^{-1} against 20.0 t ha^{-1} due to the residual effect, the yield response being 20.5 per cent. While, when organics were not applied the corresponding yields were 14.2 and 17.5 t ha^{-1} resulted by residual and cumulative effects (Table 5). Chandrika and Reddy (2011) [2] also reported similar yields of onion i.e., 31.18 and 23.60 t ha^{-1} respectively in 2004 and 2005 years (Agrifound light red). Biochar could hold the native and applied nutrients and supply slowly over a long period and increase the use efficiency of applied inorganic P. This might be the reason for the residual effect of biochar. While, when the short term nutritional requirements of the crop are met from the conjunctively applied 30 kg P_2O_5 ha^{-1}, the long term requirements are met from the application of biochar.

Table 5: Residual and cumulative effects of organics, inorganic P and their interaction on onion yield (t ha^{-1})

Organics-Sub treatments	Inorganic P levels (kg P_2O_5 ha^{-1}) -Main treatments: 0			30			60			Means		Mean for Organics
	Residual	Cumulative	Mean	Residual	Cumulative	Mean	Residual	Cumulative	Mean	Residual	Cumulative	
No organics	11.9	16.5	14.2	14.5	18.3	16.4	16.3	17.5	16.9	14.2	17.5	15.8
PSB	13.0	18.7	15.8	17.5	22.9	20.3	18.4	22.5	20.4	16.3	21.4	18.8
Biochar	16.3	21.5	18.9	21.2	24.9	23.1	22.6	25.8	24.2	20.0	24.1	22.1
Humic acid	16.1	21.1	18.6	21.3	23.3	22.3	21.0	23.3	22.2	19.5	22.6	21.0
Citric acid	13.6	17.8	15.7	16.5	20.6	18.5	18.0	20.6	19.3	16.0	19.6	17.8
Mean	14.2	19.1	16.6	18.2	22.0	20.1	19.3	21.9	20.6	17.2	21.0	19.1
	MT	ST	SST	MT at ST	MT at SST	ST at MT	ST at SST	SST at MT	SST at ST	SST at MT,ST	ST,SST at MT	MT at ST,SST
SEm±	0.3	0.2	0.1	0.4	0.3	0.4	0.3	0.2	0.3	0.5	0.3	0.1
CD (P=0.05)	1.1	0.6	0.4	1.2	1.2	1.1	0.8	0.6	0.8	NS	NS	NS

Correlation among P fractions and bulb yield of onion crop at harvest

The saloid-P had significant positive correlation with Al-P (r = 0.998**), Fe-P (r = 0.674*), Ca-P (r = 0.805**), and with onion bulb yield (r = 0.820**). Also, Al-P had significant positive relation with Fe-P (r = 0.665*), Ca-P (r = 0.796**), and with onion bulb yield (0.814**). While, Fe-P had positive relation with Ca-P (r = 0.887**), occluded-P (r = 0.715**), and with onion bulb yield (r = 0.872**). Ca-P recorded significantly positive relation with occluded-P (r = 0.685**), and with onion bulb yield (r = 0.889*). Occluded-P had significantly positive relation with onion bulb yield (r = 0.677*) (Table 6). Saha *et al*. (2014) also established that the saloid and Al-P fractions seemed to be important as these correlated significantly with Olsen's available P and various yield and P uptake parameters in calcareous sols. The available P, saloid-P and Al-P were positively and significantly correlated with grain yield, straw yield, P concentration in grain and total P uptake by wheat.

According to Jiang and Gu (1989), Pi fractions had different contributions to plant-available P which could be reflected by the extraction sequence. However, the correlation coefficients did not decrease in the extraction order, and high correlations were also found among these Pi fractions except for Ca_{10}-P, so the transformation among these fractions would be more important to understand their potential of P release and direct availability.

Table 6: Correlation among P fractions and bulb yield of onion crop at harvest

	Saloid-P	Al-P	Fe-P	Ca-P	Occluded-P	yield
Saloid-P	1.000	0.998**	0.674*	0.805**	0.528	0.820**
Al-P		1.000	0.665*	0.796**	0.530	0.814**
Fe-P			1.000	0.887**	0.715*	0.872**
Ca-P				1.000	0.685*	0.889**
Occluded-P					1.000	0.677*
yield						1.000

*Significant at P = 0.05 **Significant at P = 0.01

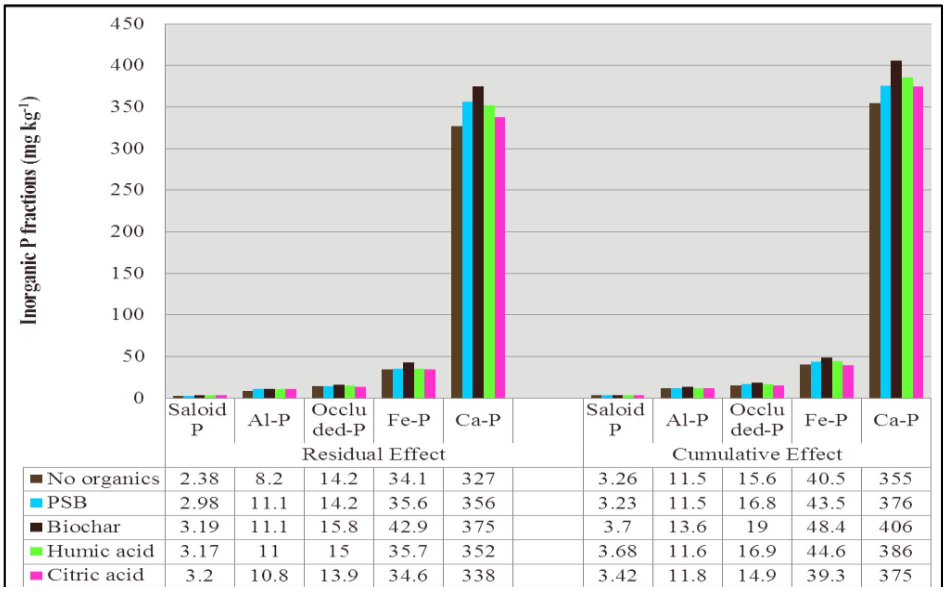

Fig 1: Inorganic P fractions (mg kg^{-1}) in the rhizosphere of onion at harvest due to integrated application of 30 kg P_2O_5 ha^{-1} with organics

Conclusion

In a high P Alfisol, among the inorganic P fractions estimated, saloid-P is the smallest pool and Ca-P is the largest pool accounting for 1 and 83 per cent respectively of the total inorganic P at harvesting stage of onion. In both the crops (soybean and onion), different inorganic P pools were larger with biochar followed by humic acid when they are integrated with inorganic P. Size of different P-fractions increased with increased levels of inorganic P application. In rhizosphere soils, the different inorganic P fractions followed the order: Ca-P > Occluded-P > Fe-P > Al-P > saloid-P. In onion crop, cumulative effect showed significantly higher inorganic P fractions than residual effect.

Biochar resulted in a significant increase in mean onion yield to 22.1 t ha^{-1} against 15.8 t ha^{-1} when organics were not supplemented, the yield response being 39.9 per cent across inorganic P. Cumulative effect was found to show significant influence resulting in a mean yield of 21t ha^{-1} which was higher by 22.1 per cent than 17.2 t ha^{-1} due to the residual effect. Simple correlations indicated that Ca-P recorded significant positive relation with onion yield (r = 0.889**) in the high P Alfisol.

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