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Role of biochar on phosphorus transformation in soil and/or tannery sludge mixtures

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Abstract

Biochar is used as a solid amendment to stabilize the soil nutrients in otherwise nutrient - deficient soils. In the present study, the mobility of soil phosphorous and phosphomoonoestrases were monitored with respect to biochar amendment. Biochar amendment was used at different application rates with soil and soi sludge mixtures. Soil samples collected at different time intervals were evaluated for phosphorus levels and (acidic and alkaline) phosphatases enzyme activities. The entire data set was subjected to kinetic modeling and results showed that the data was best described by parabolic diffusion in the available and total phosphorus. Acidic and alkaline phosphatase activities follow the rate equation of power function. Briefly, our study suggests that the biochar addition alter the initial adsorption rate, diffusion of available phosphorus and total phosphorus. The biochar acts as sustainable source of phosphorus and also sustain the available phosphorus in soil and sludge.

INTRODUCTION

It has been proposed that agricultural demand for P will outstrip mineable resources in 50–100 years [4].This estimation of phosphorus availability over a time scale is popularly known as ‘’Peak phosphorus” and it is connected with the concept of nine planetary boundaries. The concept of peak phosphorus generally describes the point in time at which the maximum global phosphorus production rate will be achieved. Phosphorus is a scare finite resource on earth and method of its production other than mining is unavailable because of its non-gaseous environmental cycle. Earth’s phosphorus reserves are expected to be completely depleted in 50-100 years and peak phosphorus stage is expected to be reached by approximately2030. Phosphorus belong to one of nine “Earth system processes” which are known to have boundaries. As long as the boundaries are not crossed, they are marked under the “safe zone” for the planet. Climate change, chemical pollution, loss of biosphere integrity, ozone depletion, ocean acidification, global hydrological cycle of water consumption, land usage patterns and aerosol loading are other major components of earth system processes. Phosphorus, therefore is a limiting natural resource and the only cost-effective method of its production is mining of phosphate rocks whose distribution is restricted to a few countries like Morocco, China, Algeria and Syria. At the same time phosphorus is an essential nutrient for all forms of life. It is a key element in our

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renewable resource, cannot be replaced and there is no synthetic substitute.

The availability of phosphorus in soil is influenced by soil organic matter, pH, soluble Al, Fe, and Ca [20]. The high base status and pH of calcareous soils make phosphorus sparingly soluble due to formation of metal complexes such as Ca–P and Mg–P [13] that causes the low availability of phosphorus. Organic matter additions have long been used in the tropics to enhance phosphorus cycling and bioavailability by reducing phosphorus sorption sites [13]. Biochar is a rich source of phosphorus acting as a liming agent and thus it can alter the phosphorus cycle leading to improvement in the soil phosphorus level. Biochar produced from the waste biomass can effectively address the problem of phosphorus deficiency in soil and improve its availability. Amendment of biochar in soil was found to influence the temporary phosphorus sorption/desorption pattern in soils. The addition of biochar has been reported to increase Mehlich-3 extractable phosphorus and bicarbonate extractable phosphorus (Olsen P) in soils [3].

Phosphorus release in soil is also governed by the activity of different enzymes. Phosphatase is one such enzyme that release inorganic phosphate from organic moiety or complex inorganic materials. Soil phosphatase hydrolyzes the ester bonds binding phosphorus to carbon (C-O-P) in organic matter and in this process the inorganic phosphorus is released from organically bound P. As per many previous reports the activities of phosphatases (like those of many hydrolases) depend on several factors such as soil properties, soil organism interactions, plant cover, leachate inputs and the presence of inhibitors and activators [11]. In soil ecosystems, these enzymes are believed to play critical roles in phosphorus cycles as evidence shows that they are correlated to phosphorus stress and plant growth. Phosphomonoesterases are ubiquitous in soil and have been studied extensively because they catalyze the hydrolysis of organic phosphomonoester to inorganic phosphorous, making it available for plant uptake [1]. Biochar amendment in soil and sludge may alter the enzymatic behavior of theses combinations leading to a shift in the phosphorus cycle. However, the fate of the nutrients and the enzymes associated is poorly understood when biochar is involved, particularly for soil phosphorus. Hence, the present study was undertaken to understand the mechanism of phosphorus mineralization in soil and soil + tannery sludge mixture after biochar addition. The hypothesis of this study was based on the presumption that biochar derived from waste biomass would have a positive effect on phosphorus mineralization. The pot experiments were designed for the soil-biochar and soil-sludge-biochar mixtures and their available and total phosphorus. Acidic and alkaline phosphorus were analyzed at different time points. The kinetic evaluation was also done to evaluate the release rate of available phosphorus and associated enzymatic activity.

MATERIALS AND METHODS

Characterization and treatments of soil, sludge and biochar

A sandy loam soil (Entisol), neutral to alkaline in nature, and maintained fallow for a year was collected from the CIMAP experimental farm in Lucknow, Uttar Pradesh, India. Tannery sludge was obtained from Central Leather Research Institute (CLRI), Kanpur. Soil and Sludge sample was dried, crushed and sieved before mixing with biochar. Biochar was prepared using *Cymbopogon flexuosus* biomass as reported earlier by our group [21]. Samples of biochar, tannery sludge and soil included in this study were characterized for pH, EC, total and available phosphorus, organic carbon and CEC the detailed properties of three constituents are shown in Table 1.

Biochar was added to soil at 2%, 4%, 6% and 8% application rates for obtaining four treatments: Viz,BC1, BC2, BC3 and BC4. Also, biochar at 4% was added to different combinations of soil and sludge (i.e. 25:75, 50:50, 75:25) obtaining the treatments BS1, BS2, BS3 and BS4 respectively. The non-amended soil and sludge were included as controls in there experiment.

Table 1. Physico-chemical properties of soil, biochar and tannery sludge used in the study.

Characteristics	Units	Garden soil	Biochar	Tannery sludge
pH		8.10	9.8	8.64
EC	dS m-1(25 °C)	162	985	272
Organic carbon	%	0.30	12.00	8.03
Available Phosphorus	mg/kg	15.03	20.39	19.10
Total phosphorus	mg/kg	785	3997.5	2712
Cation exchange capacity	meq/100gm	14.76	12.37	239.02

All treatments were arranged in polyvinyl plastic pots of 1 kg soil capacity with three replicates. Three different plants were sequentially planted at different times in above mentioned treatments during the study period between January to October. Soil sampling was done from the rhizosphere of these plants at the time of harvesting. (*Mentha pipereta, Withania sominifera* and *Bacopa moneri*)

Chemical and enzyme analysis

Total phosphorus was extracted by digestion with 60% $HClO_4$ after pretreatment with HNO_3 by following the methods reported by [9]. Olsen extractable P content of samples was determined based on protocol reported by Olson [14]. Acidic phosphorus and alkaline phosphatare activities were determined using the methods of Tabatabai and Bremner [19]. The activity of each of enzyme was expressed as microgram p-nitrophenol per gram per hour.

Kinetic models and statistical analysis

The concentration of total phosphorus, available phosphorus, acidic phosphatases and alkaline phosphatases (mean of triplicate) was plotted against time. The kinetics of these parameters were fitted using zero, first and second order, equations, the simple Elovich function, the power function and the parabolic diffusion law. The interpretation of the kinetic parameters was done as per the previous report [6]. Detailed descriptions of the equations are shown in the Table 2. Multivariate analysis was used to perform Hierarchical Cluster Analysis (HCA) using Euclidean distance Co-efficient generating the dendograms for visualization of resultant clustering of data.

Table 2. Kinetic models used in the study.

Model	Kinetic Equation	Parameters
Zero order	Po-Pt = a – ko t	a: is the initial desorption rate constant ko: is the rate constant
First order	In Pt = In Po – k_1 t	k_1: is the first rate constant
Second order	1/Pt = 1/Po + k_2	k_2: is second order rate constant
Simple Elovich	Pt = 1/β In (α β)+ 1/β In t	α: is the initial desorption rate β: is the desorption constant
Power function	In Pt = In a + b In t	a: Initial desorption rate constant b: desorption rate coefficient
Parabolic diffusion	Pt/Po = c + r $t^{½}$	r: diffusion rate constant

RESULTS AND DISCUSSION

Behavior of available and total phosphorus with time

The total and available phosphorus were higher in biochar as compared to sludge (Table 1). However total/available phosphorus ratios were significantly higher in sludge samples**.** Variations in different soil parameters of all treatments with time are depicted in Figure 1a-d. In the case of sludge, a steep decrease in total phosphorus was observed in S100. ANOVA was performed to assess the significant differences in the total phosphorus, available phosphorus, acidic phosphatase and alkaline phosphatase in different sampling of treatments. The changes in the all were significantly different in initial values as compared to their final values (Table 3). No significant difference was observed in values of acidic phosphatase of BS1, while content of total phosphorus was not varied significantly in S100 samples. Paired T test was applied to see the difference in the phosphorus mineralization activities among the treatments. Results of paired test were very interesting and indicated that control had significant variation in all parameters under study in all sludge treatments. A significant variation was observed in control with biochar treatments except alkaline phosphatase values of BC1 and BC2. No significant difference was observed between available phosphorus content in BC2 and BS1, BS2 and BS3.

The maximum increase in cumulative available phosphorus was found in BS3. S100 and BS2 had a similar rise in available phosphorus concentration. Cumulative available phosphorus increased with time with increase in sludge application rate. Release of available phosphorus in S100 and BC4 after 240 days (5740 hr) was 21.8 mg/kg and 28.1 mg/kg respectively. This shows that release in available phosphorus in biochar treated soil is more than the sludge treated soil. Both biochar and tannery sludge had different cation exchange capacity (Table 1) and availability of PO_4^- might alter through its anion exchange capacity or by influencing activity/ availability of the cations that interact with phosphorus [4]. Availability of PO_4^- can lead to formation of oxide with either heavy metal or with alkaline earth metal. Biochar addition in soil promotes this process [5]. It may also provide indirect effects on phosphorus availability and uptake through changes in the soil environment for micro-organisms this may also lead to mineralization of PO_4^- in the soil. Scot [17] reported that PO_4^- availability may be due to the retention of orthophosphates by surface functional groups

Activity of phosphatase in soil with time

The enzymatic activities of acidic and alkaline phosphorus were monitored with time and results are shown in the Figure 1. In sludge treatments, activity of acidic phosphorus (Fig. 1) showed no significant differences between control and BS1 (p = 0.059) and BS3 and S100 (p = 0.064). Similarly, in biochar treatments, Paired test showed no significant variation in enzymatic activities of BC1, BC2, BC3 and BC4 Table 3.

When alkaline phosphorus was estimated for sludge treated samples, BS3 and S100 were found to be similar (p =0.80). However, a significant variation was observed among the biochar treatments. The similarity in values of enzymatic activities between any two treatments may be due to dominance of their physico-chemical properties. Release of acidic phosphatase in both the treatments (Biochar and sludge) was found maximum at higher concentration at 90 days (2160 hr), while after 240 days (5760 hr) acidic phosphatase was maximum in 4% biochar treated soil. The result shows that activity of acidic phosphatase increase (from 0 to 2160 hr) in both biochar and sludge. Initially the increase in the activity of acidic phosphatase was rapid which might be due to the high pH at initial stage in soil. Effect on phosphates activity which is highly pH-dependent might be due to changes in soil pH which influence the phosphorus availability in soil. Atkinson [2] reported that increase in pH and the availability of minerals with application of biochar. In contrast organic amendment (biochar and/tannery sludge) acting as a source of soluble phosphorus-salts and exchangeable phosphorus, acts as a modifier of soil pH. It also acts as an enhancer of microbial activity and therefore indirectly subsequent mineralization of phosphorus [15] affecting the activity of phosphatase enzyme. Cumulative increase in the acidic phosphatase for sludge and biochar treated soil were 707.65 and 521.32 mg PNP $kg^{-1}h^{-1}$ respectively. There results suggested that activity of acidic phosphatase in sludge amended soil was more than the biochar amended soil.

Interaction between the minerals of sludge and or biochar with soil minerals may change the surface properties of soil minerals [10] and impact on phosphorus sorption and consequently phosphorus availability. This effect may develop more charge in soil which changes the soil pH.

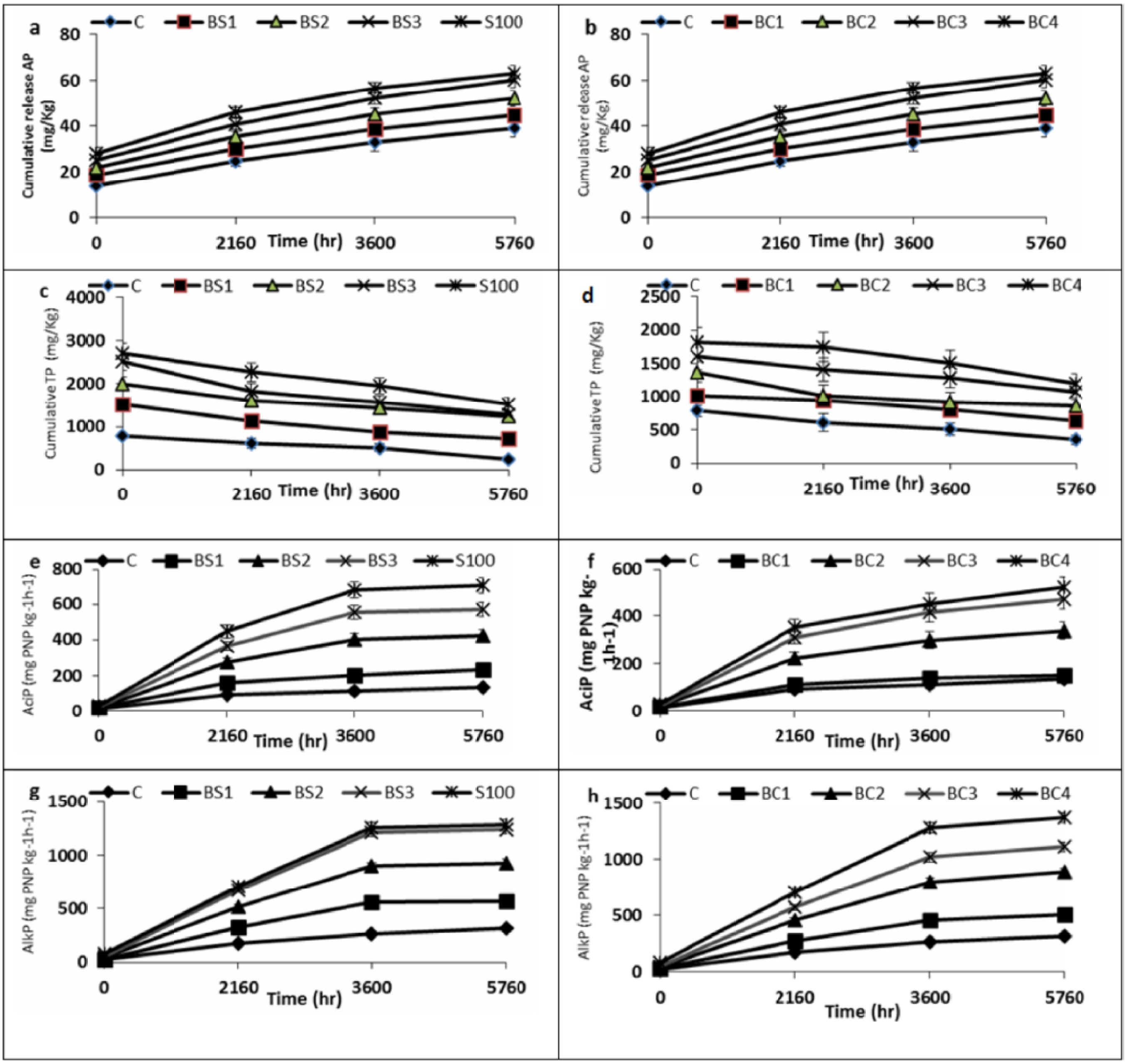

Fig. 1: Cumulative changes in total, available phosphorus, acidic and alkaline phosphatase with amendment of biochar and tannery sludge. (a) Cumulative changes in total phosphorus in tannery sludge treatments; (b) Cumulative changes in total phosphorus in biochar treatments; (c) Cumulative changes in available phosphorus in tannery sludge treatments; (d) Cumulative changes in available phosphorus in biochar treatments; (e) Cumulative changes in acidic phosphatase in tannery sludge treatments (f) Cumulative changes in acidic phosphatase in biochar treatments; (g) Cumulative changes in alkaline phosphatase in tannery sludge treatments; (h) Cumulative changes in alkaline phosphatase in biochar treatments.

[*AP-available phosphorus; TP-total phosphorus; AciP-Acidic phosphatase; AlkP-alkaline phosphatase BS1; BS2;BS3;S100 are different treatments and C is soil control].

The increase in enzyme activity observed in current study may also be due to the increase in the activity of the extracellular enzyme–organo complexes because they are added with the amendment or the added organic moieties stabilize extracellular enzymes produced by the stimulated microbial activity. In previous studies also reported that the addition of sewage sludge to soils increased the activity of alkaline phosphatase. Kýzýlkaya [10] reported that the activity of the acidic

Table 3. Paired sample test to compares the different treatments.

	TP		AvP		AlKP		AcidP	
	t	sig (2-tail)	t	sig (2-tail)	t	sig (2-tail)	t	sig (2-tail)
C - BS1	-4.436	.002	-8.281	.000	-13.635	.000	-2.252	.054
C - BS2	-4.258	.003	-9.389	.000	-8.311	.000	-2.856	.021
C - BS3	-4.217	.003	-7.772	.000	-7.906	.000	-2.847	.022
C - S100	-5.001	.001	-4.229	.003	-13.401	.000	-2.495	.037
C - BC1	-2.988	.017	-3.328	.010	-1.351	**.214**	-4.037	.004
C - BC2	-2.293	.040	-2.780	.024	-1.211	**.260**	-2.913	.019
C - BC3	-2.650	.029	-5.679	.000	-2.519	.036	-2.206	.058
C - BC4	-2.352	.005	-4.805	.001	-2.186	.027	-3.039	.016
S100 - BS1	5.144	.001	2.043	.045	13.033	.000	2.543	.035
S100 - BS2	5.211	.001	1.158	.002	3.322	.011	2.311	.050
S100 - BS3	4.383	.002	-3.274	.011	.257	**.803**	2.151	**.064**
BC2 - BS1	0.478	**.645**	2.082	**.071**	-8.027	**.000**	-.699	**.505**
BC2 - BS2	-4.431	.002	2.583	.015	-24.571	.000	-1.791	.111
BC2 - BS3	-5.427	.001	2.017	.039	-19.743	.000	-2.098	.069
BC1 - BC2	-2.100	**.069**	-2.072	**.072**	-.333	**.747**	-.516	**.620**
BC1 - BC3	-2.601	.032	-6.458	.000	-11.743	.000	-.854	**.418**
BC1 - BC4	-2.318	.049	-4.019	.004	-10.236	.008	-1.750	.118
BC2 - BC3	-3.475	.008	1.412	.196	-3.767	.005	-.334	**.747**
BC2 - BC4	-2.445	.040	1.198	**.265**	-.143	**.890**	-1.747	.119
BC3 - BC4	-1.930	**.090**	-2.205	**.059**	1.668	**.134**	-3.821	**.005**

phosphatase was partly inhibited at high dosages of applied sewage sludge [18]. However, the negative effect of inorganic phosphorus on phosphatase activities of soil can be partially detected because the measured enzyme activity also depends on the stabilized extracellular phosphatase activities, which are not related to microbial activity [16].

The relationship of enzymatic activity with sludge and biochar doses was also confirmed by HCA analysis. The HCA dendogram depicts two clusters I and II, Cluster I was again divided in to two sub clusters Cluster Ia and Cluster Ib (Figure 2). Cluster Ia comprised of all the biochar treatments (BC1 BC2 BC3 and BC4) while, Cluster Ib has BS1 and control. Second cluster contains all sludge treatments. It indicates that biochar and sludge treatments have different phosphorus dynamics whereas, control has a similar phosphorus cycle as in biochar treatments. The grouping of BS1 and control indicates that the addition of 4% biochar and 25% sludge enhances the release phosphorus, but may not alter the phosphorus release mechanism.

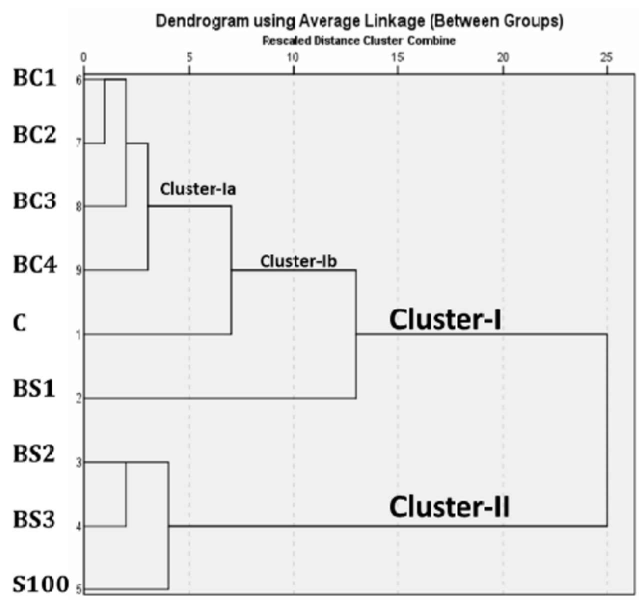

Fig. 2: Dendograms produced by Hirachieal cluster analysis (using Euclidean distance).

Kinetic models

Different kinetic models were applied to the available and total phosphorus and enzymatic data (Table 4). Here, r^2 values calculated of the different kinetics models were used for the most suitability for the reactions. For total phosphorus, power function model was most suitable at describing the kinetic data for both tannery sludge and biochar was better fitted to the power function (r^2 = 0.85 to 0.99 Table-2). The constants a and b in the power function can be regarded as the initial desorption rate constant and desorption rate coefficient respectively. The values of constant 'a' indicate that control, pure sludge and BC_4 has higher initial desorption.

Desorption rate coefficient were comparable for all treatments. Available phosphorous follow the desorption kinetic of parabolic diffusion in all 9 treatment (r^2 = 0.95 to 0.99 Table-2). Here, coefficient c denotes that diffusion rate constant. For available phosphate highest value of C were observed for BC4 followed by pure sludge. The diffusion of available phosphorus was increased with increasing amount of biochar/sludge in soil. Khater [7] reported that the parabolic diffusion equations were the best fitted equations used to describe desorption under varying soil pH [12].

Activity of phosphatase enzyme mainly promote by the presence of phosphate ions in the soil solution. Coefficient of determination (r^2) value of acidic phosphtase activity showed higher values for parabolic diffusion follows by simple Elovich and power function. Alkaline phosphatase activity also followed kinetic power function. All treatments had higher values of constant a and b as compared to control for both the enzymes. The greater a and/or lower b, might be considered indicative of greater rate of nutrient release. Heterogeneity of adsorption sites with different adsorption affinities of phosphorous in soil might be responsible for both alkaline and acidic phosphtase enzyme.

CONCLUSION

The amendments and kinetic study suggests that the biochar is capable of the mobilization of phosphorus in soil as well as sludge. It enhances

Table 4. Correlation coefficient and estimates of release parameter of different kinetics models.

			C	BS1	BS2	BS3	S100	BC1	BC2	BC3	BC4
AP	**PD**	r^2	0.95	1.00	1.00	0.99	0.97	0.99	1.00	0.98	0.96
		c	0.36	0.11	0.14	0.23	0.78	0.06	0.19	0.61	1.12
TP	**PF**	r^2	1.00	0.93	0.95	0.95	0.99	1.00	0.85	0.98	0.98
		a	-4.24	-2.85	-2.52	-2.38	-3.10	-4.45	-1.98	-3.41	-3.05
		b	0.69	0.56	0.66	0.71	0.72	0.59	0.57	0.62	0.64
AciP	**SE**	r^2	0.96	0.97	0.94	0.93	0.92	0.96	0.99	0.95	0.95
		1/β	0.07	0.04	0.02	0.02	0.01	0.07	0.03	0.02	0.02
		α	-2.11	-2.20	-1.99	-1.95	-1.91	-2.64	-2.02	-2.01	-2.06
	PF	r^2	0.998	0.998	0.998	0.997	0.997	0.997	0.998	0.998	0.998
		a	-4.47	-4.12	-4.12	-4.06	-4.02	-4.03	-4.15	-4.03	-3.93
		b	0.58	0.64	0.72	0.75	0.78	0.58	0.69	0.73	0.74
	PD	r^2	0.99	0.99	0.98	0.97	0.97	0.97	0.99	0.99	0.99
		c	2.55	5.51	6.84	8.37	8.58	5.38	5.46	7.25	9.21
AlkP	**PF**	r^2	0.99	0.99	0.99	0.99	0.99	0.97	0.99	0.99	0.99
		a	-3.33	-3.40	-3.59	-3.65	-4.18	-2.87	-3.27	-3.50	-3.38
		b	0.65	0.73	0.79	0.82	0.83	0.70	0.78	0.81	0.83

AP= Aviliable Phosphorous, TP = Total Phosphorous, AciP= Acidic Phsphtase, AlkP= Alkaline Phsphtase SE= Simple Elovich, PF= Power function, PD= Parabolic diffusion, Correlation coefficient (r2) the desorption and diffusion of phosphorus from soil particle and make them available for the plant.

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