

EFFECT OF BIOCHAR WITH AND WITHOUT PHOSPHORUS FERTILIZERS ON PHOSPHORUS FRACTIONS, WHEAT YIELD AND MICROBAL BIOMASS CARBON IN ALKALINE SOIL

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Received: Aug. 9, 2018

Accepted: Aug. 16, 2018

ABSTRACT: *The present work was carried out to investigate the effect of biochar and phosphorus fertilizer application, on phosphorus fractions, soil microbial biomass carbon in alkaline soil during two seasons for wheat (*Triticum aestivum* L. cv. Gemmiza) in field experiments. Phosphorus was added at 0%, 50%, 100% and 150% P of recommended P fertilizers, with or without biochar at a rate of 10 t h⁻¹ arranged in a randomized complete block design with three replicates. Results showed that the wheat grain yield increased by 56 to 69% in plots treated with the interaction of biochar and P mineral during the 2015 and 2016 growing seasons. Sequential extraction of the biochar-treated with P revealed that HCl-P decreased, whereas others fractions increased with increasing P rate. The results of this study reveal that the co- application of biochar with inorganic P can be a promising strategy to improve soil productivity and soil quality in alkaline soil.*

Key words: *Phosphorus fractions; Biochar; Soil quality; Vertisol; Wheat yield.*

INTRODUCTION

In arid and semi-arid regions, soils are low in organic matter (<1%) and plant available nutrients. Although the total amount of P in the soil may be high, it is not meeting the nutritional requirements of crops (Mohamed *et al.* 2000), because in the soil more than 80% of the P becomes immobile and unavailable for plant uptake because of adsorption, precipitation, or converted to the organic form (Holford 1997). Phosphorus (P) is one of the main growing plants, limiting nutrients although it is abundant in soils in both inorganic and organic forms. Use of mineral P fertilizers and biochar can be a promising strategy to improve soil fertility and increase the efficiency of the use of P fertilizers in alkaline soils (Gunes *et al.* 2014). Agricultural waste is a great importance to soil quality by

recycling of these as biochar (Widowati & Asnah 2014). Biochar is become as a simple technology that can provide multiple environmental benefits when added to soil, including long-term carbon sequestration (C) and increased P use efficiency in soil (Woolf *et al.* 2010). (Mahmoud *et al.* 2017) found that availability of phosphate in soils increased with biochar applied as soil amendments. Several studies have explained that the biochars applied in the soil could enhance P availability in soils and decrease P fixation (Novak *et al.* 2009; Silber *et al.* 2010; Chintala *et al.* 2014). Moreover, the biochar application could decrease P loss of applied P fertilizers during the leaching, as a result, reducing the pollution risk in the water and soils (Laird *et al.* 2010 & Kumari *et al.* 2014). Biochar has the potential to

increase P availability by nutrient inputs, increasing CEC, or altering soil pH (Enders *et al.* 2012; Jones *et al.* 2012 & Yuan *et al.* 2011).

P fraction is important to assess the P status of soils and to study the chemistry of soils that influence environmental and soil quality. In general, the P distribution in the soils examined that after Headley fraction; more than 40% of P remained in the residual fraction in all soils (Turner *et al.* 2005; Condrón & Newman 2011). Inorganic P in all fractions increased significantly by P fertilization (Solomon & Lehman 2000). Biochar with fertilizers significantly increased Olsen P and labile P concentrations (Chaturika *et al.* 2016).

Wheat is one of the main important crops for global food security. Cultivated cereal in the world is more than 220 million ha to produce about 670 million tons. In Egypt, wheat is one of the main and strategic crops that have caused a complex problem, especially the gap between high production and consumption. Wheat production in Egypt reached 9.46 million tons in 2012 (covers 40% of our needs), produced from an area of 1.419 million ha (CAPMAS 2014). The aim of this study was to investigate the effects of the co-application of P fertilizers and a biochar on soil P fractionation and wheat yield over two growth seasons in alkaline soil.

MATERIALS AND METHODS

Biochar production

Citrus trees pruning biochar used in this study is locally produced using a batch pyrolysis facility at a final temperature (500 °C) with a retention time of 2 h. Biochar samples were ground and sieved <0.5mm, prior to use and characterization. The main characteristics of the biochar are pH (8.09), OC (46.85%) and available

nutrients N (1.4 %), P (0.92 %) and K (1.42%), as described in (Table 1).

Field experiment

A field experiment was carried out during the two winter seasons of 2015/2016 and 2016/2017 at Experimental Farm of El-Gemmiza Agric.Res-Station, Agric. Res-center, El-Gharbia governorate, Egypt (30° 43 N - 31° 07') to evaluate the effect of biochar along with different P fertilizer rates on phosphorus fractionation, microbial biomass carbon and wheat yield (*Triticum aestivum*). Soil type was a *Vertic Torrifuvents* (Gad & Ali 2011). The soil temperature regime of the studied area could be defined as *Thermic* and soil moisture regimes as *Torrif*, main soil characteristics were shown in (Table 2). Treatments consisted of a factorial combination of four phosphate fertilizer levels (0% P, 50% P, 100% P and 150% P of recommended P fertilizers) and two biochar levels (0.0 and 10 t h⁻¹) arranged in a randomized complete block design with three replicates. The field was plowed and mixed of biochar at 10 t h⁻¹. Plot area was 14 m² (4 m x 3.5 m). Basic application of N and K were applied to all plots i.e. 178 kg N h⁻¹ and 57.12 kg K₂O h⁻¹ in the forms of urea (46%N) and potassium sulphate (48% K₂O), respectively. The other usual agronomic processes of wheat plants (variety of Gememiza 9) were practiced. Wheat seeds were sown at the second week of November by using 142.8 kg ha⁻¹ grain rate. Wheat plants were harvested after 190 days of sowing when the grains were reopened. After harvesting, straw and grain weight was recorded by using a weighting balance. Soil samples at a depth of 0-30 cm and 30-60 cm was taken after wheat harvesting from each plot and analyzed for the selected soil chemical properties. The soil samples collected were air-dried and crushed to pass

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through a 2 mm sieve for chemical properties. Grain yield was recorded by using weighing balance after harvesting;

plants were taken from all plots by randomly selected 1 m².

Table (1): The main characteristics of biochar.

Parameters	pH (1 : 10)	EC ds m-1 (1 : 10)	OC %	CEC Cmole kg-1	N %	P %	K %	Ca %	Mg %	C / N
Values	8.09	0.52	46.8	30.31	1.4	0.92	1.42	1.5	2.7	33.40

Table (2): The main properties of the soil.

Soil properties	Unit	Season 2015		Season 2016	
		Soil depth, cm			
		0 – 30	30 - 60	0 - 30	30 - 60
Particle size distribution					
Sand	%	16.42	16.71	16.31	16.48
Silt	%	25.77	27.37	30.12	27.75
Clay	%	57.81	55.39	53.57	55.57
Texture		clay	clay	clay	clay
bulk density	mg m ⁻³	1.41	1.5	1.46	1.51
pH (1:2.5)soil and water		7.97	8.09	7.98	8.01
EC soil paste extract	dS m ⁻¹	1.32	1.23	1.37	1.25
OM	%	1.38	1.23	1.39	1.25
Soluble cations					
Ca	meq L ⁻¹	6.4	6.01	6.13	6.0
Mg	meq L ⁻¹	3.2	2.6	3.23	2.51
Na	meq L ⁻¹	5.1	4.89	5.29	5.0
K	meq L ⁻¹	0.81	0.8	0.84	0.8
Soluble anions					
HCO ₃	meq L ⁻¹	4.31	5	4.32	5.14
CO ₃	meq L ⁻¹	0	0	0	0
Cl	meq L ⁻¹	7.11	6.89	7.22	6.84
SO ₄	meq L ⁻¹	4.09	2.41	3.95	2.33
CEC	Cmole kg ⁻¹	46.12	45.01	47.1	45.12
Available nutrients					
N	mg kg ⁻¹	40	38	43	39
P	mg kg ⁻¹	6	5.01	6.01	4.98
K	mg kg ⁻¹	300	289	313	284

Analysis of soil and biochar

The soils were characterized for their physical and chemical properties following standard laboratory procedures (Klute & Dirksen 1986 & Page *et al.* 1982). Soil pH and electrical conductivity (EC) were measured in a saturated soil paste extract and biochar in a water (1:10 biochar: water ratio) using a pH/conductivity meter. Organic carbon was determined by sulphuric acid and dichromate oxidation according to (Walkley & Black 1934). Soil organic carbon content of the biochar was determined by combustion method (Page *et al.* 1982). The cations and anions were determined in a saturated soil paste extract as described by (Rhoades 1954). The soil texture was determined using the international pipette method, as described by (Kim 1996). Cation exchange capacity (CEC) was determined by the (1 M NH₄OAc at pH 7.0) method (Jackson 1974). Exchangeable Na and K were measured by the flame photometer (Rich 1965). Total nitrogen (N) was determined by the macro-Kjeldahl method (Page *et al.* 1982).

Microbial biomass carbon

Microbial biomass carbon (MBC) was determined by the fumigation-extraction technique. Ten grams of soil was fumigated with chloroform (CHCl₃) for 24 h at 25°C, and samples were extracted with 50 ml 0.5 M K₂SO₄ for 30 min on a horizontal shaker at 200 rev min⁻¹ and filtered through paper (Whatman No. 42). Similarly, 10 g soil was extracted for non-fumigation at the same time (Brookes *et al.* 1985). Soil organic carbon (SOC) in the extracts was measured by the titration method. Then MBC was calculated as: Microbial biomass C = (C_{fumigated} - C_{unfumigated}) x 2.64.

Phosphorus fractionation

Soil samples after harvesting of wheat 2015-2016 season were collected from

each plot at a depth 0 -30 cm and 30-60 cm. the fractionation of P in the soil was carried out the following the sequential extraction, analysis according to (Hedley *et al.* 1982) procedure and modified by Chen *et al.* 2000. The extraction was: exchangeable phosphorus extracted by 0.5 g air dried soil with 30 ml NH₄Cl (1M) in centrifuge tubes, labile phosphorus by 30 ml of 0.5 M NaHCO₃ (pH 8.5), Al and Fe phosphates by 0.1 M NaOH, Ca-phosphate by 1M HCl, unlabeled P by 0.1 M NaOHii. All centrifuge tubes used in this process were shaken for 16 h at room temperature, and filtered with Whatman No. 42. The organic P (Po) in NaHCO₃, NaOHi and NaOHii extracts was obtained by digesting 5ml of each extractable with H₂SO₄ and H₂O₂ and calculation as the (Pt mince Pi) of respective extracts. Residual P in soil samples was measured after digestion with (HNO₃-HClO₄). The P concentration in all extracts analyzed calorimetrically with the molybdate-blue method at 880 nm (Murphy & Riley 1962).

$$P_T = \text{NH}_4\text{Cl} + \text{NaHCO}_3 + \text{NaOH}_i + \text{HCl} + \text{NaOH}_{ii} + \text{residual}$$

$$P_{o=} = \text{NaHCO}_3 - P_o + \text{NaOH}_i - P_o + \text{NaOH}_{ii} - P_o + \text{residual}$$

$$P_i = \text{NH}_4\text{Cl} + \text{NaHCO}_3 - P_i + \text{NaOH}_i - P_i + \text{HCl} + \text{NaOH}_i - P_i$$

Statistical analysis

Values of treatments were analyzed statistically using SAS software (1996). Compare the differences between all values of treatments used Duncan's multiple range tests statistical significance level of P<0.05 was used.

RESULTS AND DISCUSSION

Effect of biochar and different amounts of inorganic P on grain yield of wheat and microbial biomass carbon

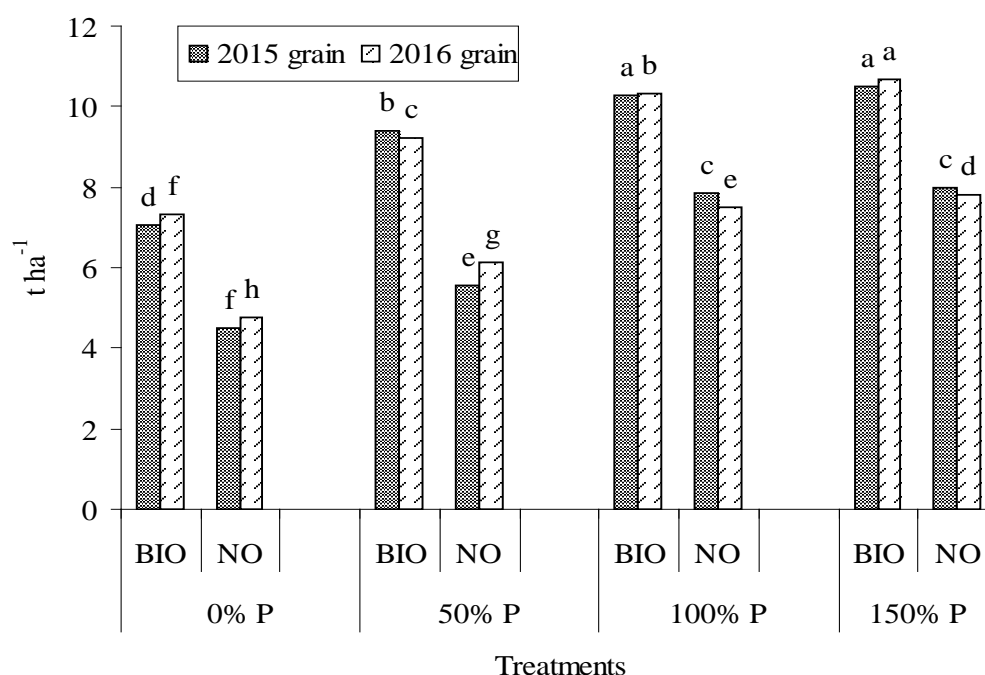
Grain yield of wheat

It has been observed that fertilization

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of P or combinations between P fertilizer levels and biochar significantly increased grain yield of wheat crops in the growing 2015 and 2016 seasons (Fig. 1). Biochar with 150 % P treatment gave the highest significant increase in the grain yield of wheat compared to other treatments. The interaction between biochar and phosphorus led to above 30 % increase in grain yield compared with the use of the P mineral fertilizer alone. This indicates the ability of the mixed addition of biochar and fertilizer P to maintain soil fertility through an increase in the labile P fraction. Similarly, increases in grain yield of wheat plants with P fertilization and biochar are probably due to improve in soil physical properties and organic matter (Mahmoud *et al.* 2017). And also, with the addition of biochar, yield increase of plants can be related to soil quality improvement Azeez *et al.*

2010, Demir *et al.* 2010), enhance nutrient supply to the plants (Gaskin *et al.* 2010), and increase microbial biomass and activity in soil (Gunes *et al.* 2014). Grain yield of wheat in the soil amended biochar and P did not increase significantly between the application at 100% and 150% P. The increase in available phosphorus has increased grain yield. Biochar has been found to alter P availability through its electrostatic repulsion and ion competition (Cui 2011). (Atkinson *et al.* 2010) reviewed several mechanisms which can enhance availability and plant uptake of P, which led to increase crop productivity after biochar addition to soil. It acts as a source of soluble P salts and exchangeable P forms; soil avoids P precipitation by modifying pH or enhances microbial activity leading to changes in P availability.



Bio: biochar at 10t/ha.
No: without biochar

Figure 1. Effect of biochar and different amounts of inorganic P on grain yield of wheat crops in the growing 2015 and 2016 seasons.

Soil microbial biomass carbon

Soil microbial biomass carbon (MBC) in the soil is a good indicator of biological activity of soil, soil quality, and it's generally positively correlated with the soil organic matter (SOM) content and soil pH. Thus, the changes in MBC may lead to changes in organic matter (OM) decomposition and nutrient cycling (Dick *et al.* 1996 and Caldwell 2005). In general, soil MBC was significantly increased with the application rate of the phosphorus and biochar (Fig 2). The highest MBC was recorded in soil amended with 150% P amended biochar (209.60 mg kg⁻¹). P application, microbial biomass in 0–30 cm soil depth increased by 52%, 49%, and 46% when fertilizer P rates were 50, 100, and 150% P, respectively, (Brendecke *et al.* 1993) found similar results, noting that the addition of organic carbon and nutrients through sewage sludge increased the population of microbes in the soil, which

in turn stimulated enzymatic activity in the soil. The obtained results point out The increase in the MBC is correlated with soil organic matter ($R^2=0.94$, $p< 0.05$, Fig. 3). Microbial biomass increased with biochar addition is due to the presence of labile C fractions, un-pyrolised feedstocks and biochar have been surfaces supplying nutrients for the microbes (Bruun *et al.* 2011; Zimmerman 2011,; Luo *et al.* 2013).The higher in P, C and other nutrients with the phosphorus and biochar amended plots could contribute to increase soil microbial biomass. The co-application of biochar with inorganic P produced up to 1.5 fold more microbial biomass C than either biochar or inorganic P applied alone. (Gichangi *et al.* 2009) observed the similar results and reported that microbial biomass increased with the co-application of inorganic P with goat manure in small-holder farms in South Africa.

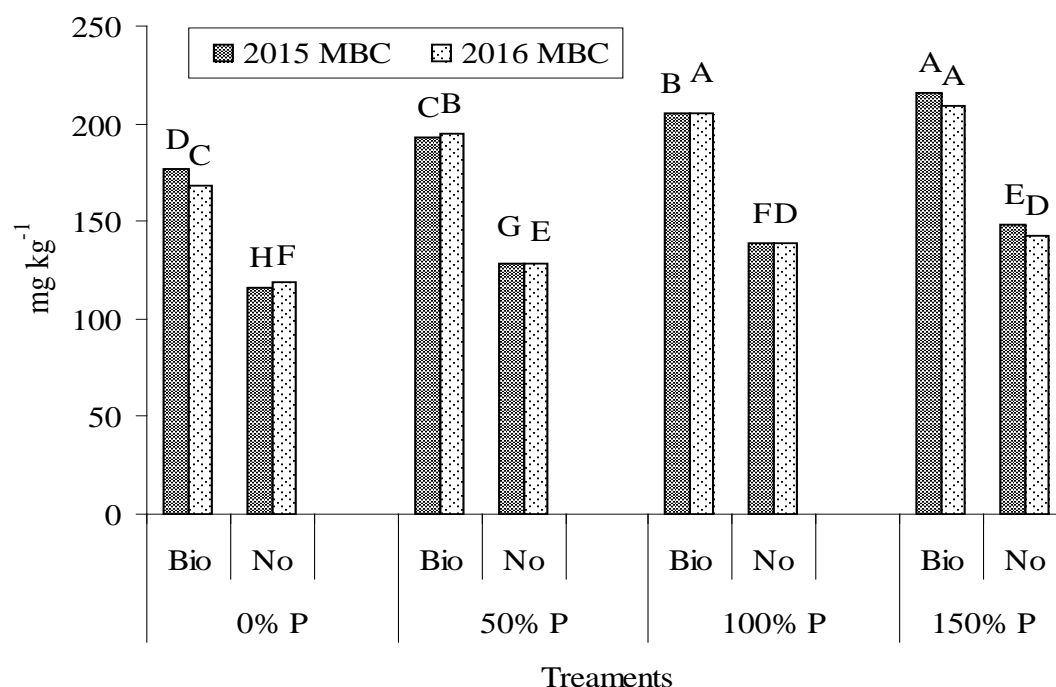


Figure 2. Effect of biochar and different amounts of inorganic P on soil microbial biomass carbon (MBC) in the growing 2015 and 2016 seasons.

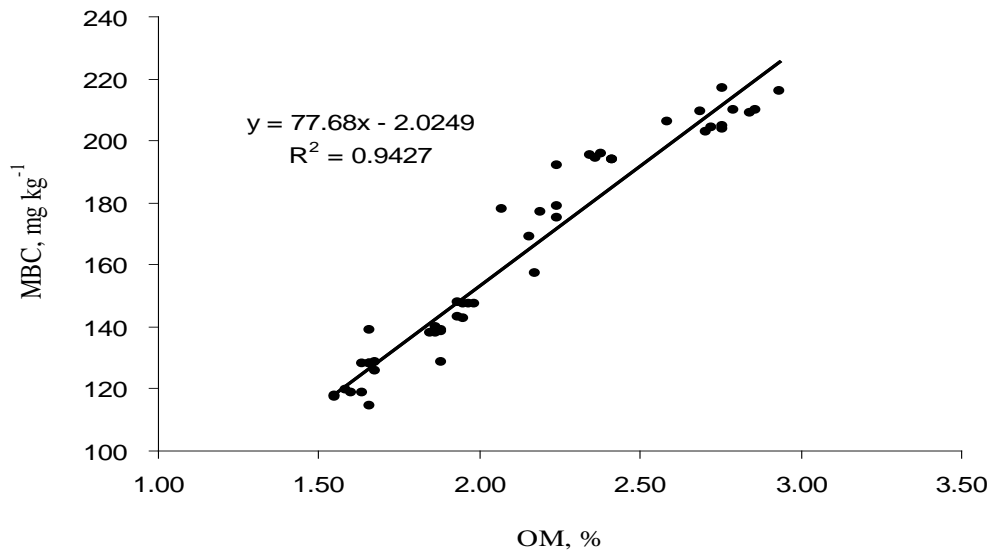


Figure 3. Relationship between soil organic matter (SOM) and microbial biomass carbon (MBC)

Phosphorus fractionation

NH₄Cl-P (exchangeable phosphorous) significantly increased in soil treated with P fertilizers or in combination with biochar in both two depths (Table 3). NH₄Cl fraction P increased with increasing P rates in the plots treated with biochar or without biochar, The NH₄Cl-P in the soil amended biochar did not increase significantly between the application at 50%P and 100%P in 30 – 60 depth. Fertilization of P increased significantly NH₄Cl-P in both depths. Similarly, (Amaizah *et al.* 2012) found that the content of water-soluble phosphorus increased with the soil treated with fertilization with mineral phosphorus as compared with the control. This increase is due to the accumulation of P and was influenced by the saturation of free spaces for adsorption of P in soil. The increase of NH₄Cl-P in the 30–60 cm depth was lower in the soil treated by biochars with the mineral phosphorus, which could be explained by movement of P along the depth was very limited and

slow due to its strong binding (Amaizah *et al.* 2012).

The NaHCO₃-P labile fraction ranged between 24.95 and 40.65 mg P kg⁻¹, which is equivalent to 11.14% and 12.26% of total P in the growing 2015 season (Table 3). Fertilization of P increased significantly NaHCO₃-P in both depths. Similar results were reported by (Sharpley & Smith 1983). NaHCO₃-P (labile phosphorus) did not increase significantly in the amended P and biochar plots except 150% P in the 0-30 cm depth and increased significantly in 30 – 60 depth. NaHCO₃-P labile fraction was higher in the amended biochar plots at different P rates than in the un-amended biochar plots, except of 150% P in the un-amended biochar plots at 30-60 cm depth.

Phosphate bound to (Al and Fe) were higher in the soil amended biochar (ranged from 7.9 to 18.44 mg P kg⁻¹) than without biochar (ranged from 2.49 to 10.45 mg P kg⁻¹) at two depths (Table 3). Phosphate bound to (Al and Fe)

Table 3

increased significantly with increasing P rates in the soil amended biochar or without biochar except of 50% P in 0-30 cm depth. In alkaline soils, as in the present case, P is fixed mainly by calcium, while in acidic mineral soils by aluminum and iron (Tunési 1999). Phosphate bound to (Al and Fe) represented a very small fraction (about 1.25 to 4.24 % without biochar but 3.6 to 5.56 % with biochar) of total P. The hydroxide P (Pi and Po) fraction is considered a moderately labile P (Pi and Po) pools that are bound with amorphous and some crystalline Al and Fe (Tiessen & Moir 2007), with low availability to plants (Cui 2011).

The HCl extractable fraction ranged between 60.74 and 87.48 mg P kg⁻¹, which are equivalent to 18.32 and 32.48% of total P in the amended biochar plots (Table 3). HCl -P decreased non-significantly in soil treated with the biochar and P in 0-30 cm depth. In the non-biochar treatments, the HCl extractable increased with increasing P rates in the two depths. Similarly, increases in HC1- P with inorganic P fertilization are most likely due to the effect of exchangeable Ca²⁺ or Mg²⁺ which can significantly affect the absorption of P in tropical soils (Smillie *et al.* 1987) including savanna soils (Agbenin 2003). The content of HCl-P is more than those observed by (Adhami *et al.* 2007) in calcareous soils of Iran. The difference is due to a type of soils in this study.

NaOHii extractable (unlabeled P) increased significantly with the soil treated by phosphorus and biochar or without biochar in the two depths (Table 3). Unlabeled P concentrations were increased by 19.36% and 32.52% under 50% P in the no-biochar plots at 0-30 cm and 30-60 cm depths, respectively. (Motavalli & Miles 2002) observed the similar results and reported that

phosphorus in the NaOH fractions increased significantly above the control with P additions and manure.

Residual P ranged from 66.33 to 165.2 mg kg⁻¹ in amended soil with biochar and fertilizer P (Table 3), and accounted for 24.63% to 40.51% of the total P. Residual P increased over the control when the biochar with fertilizer P at 150% were applied in both depths. P application, residual P at 0–30 cm soil depth increased by 40.9%, 44.2%, and 44.9% when fertilizer P rates were 50, 100, and 150% P, respectively.

Total P

In the amended biochar and P plots, total P ranged from 269.3 to 407.8 mg kg⁻¹ (Fig. 4). Biochar contributed about 35% and 20% P_t in the surface and sub-surface layers, respectively. Most of the total P in amended biochar was about 28.54% and 28.25% in organic forms and inorganic, respectively. (Hong & Lu 2018) observed the similar and found that that the total P content increased with biochar addition.

The contents of P_t fractions increased after fertilizer P applications as shown in Fig (4), there is significant differences were found between fertilizer P levels and treatment compared to control. (Blackwell *et al.* 2010) studied the effect of biochar and fertilizer in several dry land cropping soils of Australia; they found reduced P fertilizer requirements in biochar addition because biochar itself as source of P a similar manner to the present study.

Inorganic phosphorus

The soil inorganic P constituted between 39-59% of the P_t in all treatments in both depths. Fertilization of P increased significantly inorganic-P in both depths (Fig. 4). Inorganic-P increased with the increasing P rates. These results are in are agreed with

(Sharpley & Smith 1983), who found that inorganic P increased with P fertilization applications in cultivated soil. The inorganic P fractions were increased significantly when the biochar was combined with fertilizer P. This indicates the ability of the mixed addition of biochar and fertilizer P to maintain soil fertility through an increase in the labile P fraction (Zhang & MacKenzie 1997). According to (Tisdale & Nelson 1990), concentrated P solution may cause the release of reactive cations such as Ca^{2+} ,

Mg^{2+} , Fe^{3+} and Al^{3+} the P in the solution reacts with these cations forming active inorganic P fractions with varying solubility. The observed relatively lower soil inorganic P at 100% and 150% P application may have resulted from higher P concentration in soil solution leading to higher sorption and formation of more complexes with the dissolved cations (Abekoe & Sahrawat 2007). (Schmidt *et al.* 2011) found that all for fraction increased by phosphorus fertilization.

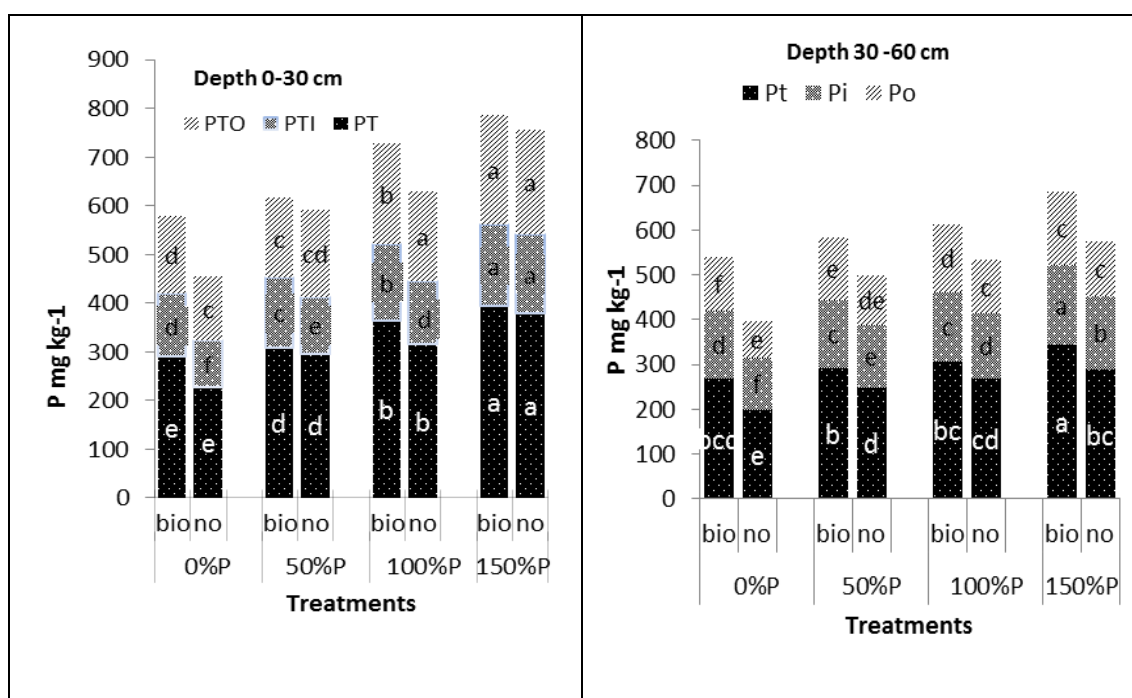


Figure 4. Effect of biochar and different amounts of inorganic P on organic and inorganic P fractions in 0-3- cm and 30-60 cm depths.

Organic phosphorus

Soil P_o contents ranged from 120.9 to 165.1 $mg\ kg^{-1}$ in the amended biochar and P plots at 0-30 cm depth (Fig. 4). Biochar, along with different P fertilizer applications increased the contents of P_o fraction, there is significant differences were found between fertilizer P levels and biochar compared to control. Biochar contributed about 48% and 21% P_o in the surface and sub-surface layers, respectively. Organic-P increased with

the increasing P rates. These results are in are agreeing with (Velásquez *et al.* 2016), who found that organic P increased with P fertilization applications in arable soils. The organic P fractions were increased significantly when the biochar was combined with fertilizer P. Farrell *et al.* 2013 observed the similar results and reported that organic P fractions increased significantly with biochar combined with fertilizer P.

Conclusions

The results demonstrated that the effects of the co-application of P fertilizers and a biochar contributed to improving wheat yield, labile P fraction and soil quality in alkaline soil. Co-application of P fertilizers with biochar significantly increased soil microbial biomass carbon, grain yield of wheat crops and organic and inorganic phosphorus compared to biochar or P fertilizers alone application. labile P fraction ranged between 24.95 and 40.65 mg P kg⁻¹, which is equivalent to 11.14% and 12.26% of total P. increases in the grain yield of wheat crops and microbial biomass carbon in the soil treated by combinations between P mineral and biochar are important for sustainable good soil properties and soil productivity. Therefore, combinations between P mineral and biochar can be used to improve soil productivity, sequester C and soil quality.

Acknowledgements

The author would like to thank the Laboratory of Soil and Water Sciences, Agriculture faculty, Tanta University, Egypt, for their assistance during this work.

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تأثير اضافة البيوشار مع التسميد الفوسفاتي على مفصلات الفسفور محصول القمح و الكتله الميكروبيه بالاراضي القلوية

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الملخص العربى

اجريت تجربه حقلية لدراسه تاثير البوشار و مستويات مختلفه من التسميد الفوسفاتي على مفضلات الفسفور والكتل الميكروبيه في التربه و محصول القمح في موسم ٢٠١٦/٢٠١٥ و ٢٠١٦/٢٠١٧ وذلك بالاضافه سوبر فوسفات الكالسيوم بمعدل ٠ في الميه ٥٠ في الميه ميه في الميه و ١٥٠ في الميه من المعدل الموصى بها من التسميد الفوسفاتي سواء باضافته منفردا او خليطا مع البوشار بمعدل (١٠ طن/الهكتار) في نظام القطع العشوائيه الكامله في ثلاث مكررات لكل معامله وقد اوضحت النتائج ان زياده محصول القمح بمعدل الى ٦٩ في الميه في المعاملات بخليط البوشار والفسفور وكذلك اعطى خليط البوشار والفسفور اعلى كتله ميكروبيه بمقدار واحد ونصف مره عن اضافه الفسفور او البوشار منفردا . وبتقدير مفضلات الفسفور في المعاملات بخليط البوشار والفسفور تبين انها تعطي انخفاضا في الفسفور المرتبط بالهيدروكلوريك (HCL-P) بينما باقى المفصولات زادت بزياده التسميد الفوسفاتي اضافته البوشار ساهمت بالزياده حوالي ٤٨ في الميه ٢١ في الميه في الفسفور العضوي في الطبقة السطحيه وتحت السطحيه على التوالي مفسرات الفسفور العضويه وغير العضويه زادت معنويا باضافه البوشار مع التسميد الفوسفاتي عما اذا تم اضافته التسميد الفوسفاتي منفردا هذه النتائج تبين ان اضافته البوشار مع التسميد الفوسفاتي تمثل استراتيجيه افضل لتحسين جوده التربه في الاراضي القلويه .

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