Available Phosphorous and Organic Carbon as an Indication for the Evaluation of Bone Char and Bone Ash Applied to Calcareous Soil

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ABSTRACT

Modern agriculture's biggest challenge is finding sustainable and environmentally peaceful fertilizers. This study aimed to compare the physicochemical properties of some non-traditional P-Sources such as bovine bone-ash (BA-600 and BA-700) and bone-char (BC-600 and BC-700), and commercial phosphate rock (PR). Various extraction techniques were applied to assess phosphorus solubility and availability of the P-Sources mentioned above compared to single-superphosphate (SSP). Additionally, an incubation experiment was carried out to investigate the phosphorus availability and organic carbon in calcareous soil, which received different P-Sources and N-K fertilizers and incubated for up to 120 days (~29 °C) arranged in a three-factor completely randomized design. The physicochemical properties of bone-char (BA-600 and BA-700) and bone-ash (BC-600 and BC-700) revealed that hydroxyapatite is present in both P-resources. Soluble and available phosphorus followed the order: BA-600>BC-600>PR for all extraction techniques. Relative to SSP fertilizer (100%), Soluble-P of water and diluted-H2SO4 (0.005 M) were nearly closed. In the incubation experiment, significant differences (P<0.05) were observed between the levels of N-K treatment, incubation time, and P-sources on soil pH, soluble calcium, and Olsen-P. Statistically, N-K fertilizers addition to P-sources-amended soil significantly (P < 0.05) increased the calcium concentration in all treatments. Olsen-P was in order: SSP > BC-600 > BA-600 > PR > control; the highest significant incubation time was 80 days. The highest percent organic carbon was recorded in the BC-600 amendment after 100 days of incubation, with no clear effect for N-K fertilizers addition. Both BA and BC are potential phosphorous resources of a sustainable nature, with a preference for

bone char as a source of non-microbially degradable organic carbon.

Keywords: Bone-ash; Bone-char; Calcareous soil; Olsen-P; Organic carbon; P-Sources

INTRODUCTION

Phosphorus (P) is a critical macronutrient for growing plants, which plays a vital role in many physiological and biochemical processes (Mengel et al., 2001). The primary phosphate fertilizer source is phosphate rock, a non-renewable resource (Warren et al., 2009), whose quantities are expected to run out during the next 50 or 100 years (Cordell et al., 2009). Phosphate badly affects the sustainability of phosphate fertilizer production. The most promising secondary Presource is the phosphorus recovery from nontraditional sources such as sewage waste (Cao et al., 2016; Worsfold et al., 2016; Yu et al., 2019 and Zhang et al., 2019), animal manure (Szogi & Vanotti, 2009 and EL-Saved, 2013), struvite mineral (Ghosh et al., 2019; Zhang et al., 2020 and Sena et al., 2021), solid wastewater ashes (Imai, 2019 and Sonoda, 2019), and P-rich 2CaO.SiO₂ phase steelmaking slag (Miki, 2019). Phosphorous fertilizer recovery from exhausted extinguishing powders (EEP) is an innovative technology called PHOSave. They comprise monoammonium phosphate (MAP, 40-50%), ammonium sulfate (in various concentrations), and other components to make the powder water-repellent. These raw materials are also used in agriculture as fertilizers due to their specific nature of releasing nitrogen and phosphorus (Dotelli and Viganò, 2020). Industrial meat and bone meal derivatives are characterized by high

DOI: 10.21608/asejaiqjsae.2023.305025

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Received, May 15 2023, Accepted, June 24, 2023.

calcium and phosphorus contents (up to 47%). Therefore, the value of these wastes can be realized through their application in the fertilizer industry. Soil enrichment with thermally treated bone meal subproducts has proved very efficient in the remediation of heavy metal-contaminated sites (Hodson et al., 2000 and Devdier et al., 2003). However, there is optimism that studies will soon focus on using industrial bone meal ash and bone char as novel and effective sources of P. Heat-treated bones have a low concentration of toxic metals, one of their most important benefits. Heavy metals (e.g., Cd, Pb, Cu, Se, etc.) are known to be present in phosphate rocks but are undetectable in bone char and bone ash (Deydier et al., 2005 and El-Refaey et al., 2015). These metals can be prevented in agricultural soils by employing non-traditional Psources.

It is reported by the Organization for Economic Cooperation and Development (OECD) that the meat industry will increase by 40 million metric tons within the next ten years, including 13% in poultry, 10% in pig meat, and >21% in sheep meat (OECD/FAO, 2018). Consequently, there will be a significant increase in the amount of meat and bone meal waste produced globally. Thermal treatment of animal bones is one of the methods used to safely dispose of bone waste through incineration to ensure the destruction of pathogens.

Appetites are minerals that are widely distributed in igneous rocks. At the same time, bio-apatite is the biological form of inorganic calcium phosphate salts (Liu *et al.*, 2013), with a general formula of Ca₅(PO₄, CO₃)₃(OH, F, Cl, CO₃) (Skinner and Jahren, 2004). Bone apatite is a carbonate apatite with a 6–9% carbonate composition in the apatite structure (Ishikawa *et al.*, 2018). It is reported that bone char samples were calcinated up to 600 °C, making carbonate apatite. While raising the calcination temperature to 900 and 1200 °C resulted in hydroxyapatite (HAP) formation (Figueiredo *et al.*, 2010). Thus, the amount and form of apatite content of a bone char are related to the charring temperature and thermal treatment duration time.

Additionally, calcination temperature significantly affects bone char's surface area and pore volume. For instance, raising the calcination temperature of cow bone char from 650 °C to 700 °C under a CO₂ atmosphere increased the specific surface area from 62 to 69 m² g⁻¹ and the total pore volume from 0.2 to 0.23 cm³ g⁻¹ (Rojas-Mayorga *et al.*, 2015a). However, increasing the temperature to 1000 °C reduced the surface area and the pore volume to 2 m² g⁻¹ and 0.02 m³ g⁻¹, respectively, and altered the color of the bone char to white.

Bone char has high pollutant removal efficiency due to its principal characteristics, represented by the textural properties of bone char and the hydroxyapatite content. It comprises 70-76% hydroxyapatite (HAP), 7-9% calcium carbonate, and 9-11% amorphous carbon (Rojas-Mayorga et al., 2015a; Mendoza-Castillo et al., 2015 and Reynel-Ávila et al., 2015). In a different scenario, bone char comprises 80-90% HAP and 10% amorphous carbon (Lambert and Graham, 1989). Leinweber (2017) reported that bone char improved porosity and water retention in sandy soil, which is particularly beneficial for crops grown in dry conditions. It is found that bone char applied to sandy soil with a ratio of 1, 5, and 10% increased water holding capacity at the permanent wilting point of plants $(-10^{4.2} \text{ hPa pressure})$ to 1.4, 3.2, and 5.5 w/w%, respectively (control without bone char was 1.3 w/w%) (Leinweber, 2017). The behavior of phosphorus in calcareous soils as a result of applying bone char and bone ash has not been studied, nor has a comparison been made between bone ash (calcined bone) and bone char (pyrolyzed bone) from the point of view of phosphorus availability and evolution of organic matter in the soil. Hence, this study aimed to compare the physicochemical characteristics of bovine bone char and bone ash produced at 600 and 700 °C relative to commercial phosphate rock and single superphosphate sources and to assess the solubility and availability of phosphorus from different P-Sources using various extraction techniques. Additionally, to investigate the availability of phosphorus and organic carbon in calcareous soil amended with the different P-Sources and treated with nitrogen and potassium fertilizers.

MATERIALS AND METHODS

Phosphorus Materials:

The bovine bone was collected from local butcher shops to prepare bone-ash (BA) and bone-char (BC). The cleaned, air-dried bone was crushed into 3.0 - 5.0 cm pieces. Incineration and pyrolysis (under a limited oxygen environment) processes were applied to produce BA and BC, respectively, using a muffle furnace at a temperature of 600 and 700 °C (heating rate of 25 °C min⁻¹). The cooled bone materials (BA and BC) were ground and passed through a 500-µm sieve (Fig. 1). The produced bone materials were symbolized BA-600 and BA-700 for bone-ash and BC-600 and BC-700 for bonechar (Fig. 1). Phosphate rock (PR) was obtained from Al-Nasr Mining Company - Cairo - Egypt. While, Single superphosphate fertilizer (SSP, $15.5\% P_2O_5$) was purchased from a local market. All different phosphorus sources (P-Sources) were passed through a 500-µm sieve and kept in plastic bottles for further analysis and testing of phosphorus availability.

The total elemental analyses of different P-Sources (PR, BA, and BC) were determined according to Brazilian Ministry of Agriculture, Livestock and Supply (MAPA) (Silva et al., 2014). 1.0 g of each sample was boiled in a concentrated HCl (10 mL) until complete evaporation of the acid (without burning the residue). Then, diluted HCl (20 mL, 2 mol L⁻¹) was added to the residue; the volume was completed to 50 mL with DW dissolution. Total after complete phosphorus concentration was measured in the clear solutions using the vanado-molybdate yellow method at a wavelength of 445 nm using a UV/VIS double beam JENWAY spectrophotometer model 6850 (Chapmann and Pratt, 1961). Calcium (Ca) and heavy metals concentrations were measured by atomic adsorption spectrophotometer (AAS Varian Spectra AA Model 220 using airacetylene flame). The Welkely-Black technique was followed to determine the total organic carbon (Jackson, 1973). The total content of carbonate was determined by calcimeter method (Pansu and Gautheyrou, 2006). Kjeldahel distillation technique was used to determine total nitrogen (TN). Digestion and determination was carried out according to Bremner and Mulvaney (1982). The surface area of different P-Sources was measured using N₂ sorption isotherms at 77 K on a gas sorption analyzer run on BEISORP-mini and the Brunauer-Emmett-Teller (BET). The total elemental composition and surface area parameters of different P-Sources are listed in Table 1. The X-ray Diffraction (XRD) analysis was performed to characterize the crystalline structure and identify the main components of PR, BA, and BC using a SHIMADZU XRD-7000 X-ray diffractometer at room temperature with Cu-Ka radiation operated at 30 kV and 30 mA; all samples were scanned from 5 to 90° 2Θ using a step size of 0.02° 2 Θ at a scanning rate of 4° min⁻¹ (Hughes et al., 1994). The Fourier Transform Infrared (FT/IR) technique was used to determine the surface functional groups of the PR, BA, and BC samples; the biomaterials were recorded in the range cm⁻¹ 400-4000 using SHIMATZU infra-red spectrophotometer, model FI/IR-5300, JASCO Corporation, Japan. The surface morphology of all bone materials was investigated using scanning electron microscopy (SEM, JEOL JSM-6360 LA analytical electron microscope). Before the investigation, the samples were coated with a thin layer of gold using a sputter coater (model: SI50B, Edwards High Vacuum Ltd., UK) to avoid the build-up of local electrical charges (Hughes et al., 1994).

To assess the soluble and available P-fraction in PR, BA-600, and BC-600, different extraction methods were applied, as summarized in Table (2). The sodium bicarbonate solution expresses the phosphorous available to the plant in basic and alkaline soils, while the formic or citric acid extractions represent the available form in acidic and near-neutral soils.

Soil Sample Collection:

A calcareous soil sample (0-30 cm) was collected from El-Hamam Experimental Station, Faculty of Agriculture (El-Shatby), Alexandria University, Marsa Matrouh Governorate (30° 51' 0.00" N and 29° 25' 9.70" E). The soil sample was air-dried, passed through a 2mm sieve, and kept in plastic bags for use. The main physio-chemical properties of the soil were determined according to Jackson (1973). The results of soil analyses are listed in Table 3. Soil pH was measured in the suspension of 1:2.5 (soil: DW) using a pH meter (WTW inoLab pH/Ion 735, Germany). Electrical conductivity (EC, dS m⁻¹) and soluble cations and anions were measured in the saturated soil paste extraction. Soil organic matter (OM) was determined by the wetoxidation of Welkelv-Black method (Jackson, 1973). Soil particle size distribution (soil texture) was determined by the hydrometer method (Gee and Bauder, 1986). The results showed that the soil texture was Sandy Clay Loam (SCL). The soil had a low organic carbon content (0.9%) and medium available phosphorus (avail. P, 5.6 mg kg⁻¹). According to pH (8.2) and EC (1.2 dS m^{-1}), this soil was classified as alkaline and non-saline soil.

Incubation Experiment

laboratory incubation experiment А was implemented to study the impact of wetting-drying cycles on the extractability and bioavailability of phosphorus released from BA-600 or BC-600 bone materials applied in calcareous soil compared to PR and SSP. Eighty grams of air-dried soil were mixed with 1% P₂O₅ of each P-Sources in a plastic can as following: 2.13 g BA-600, 2.21 g BC-600, 2.38 g PR, or 5.16 g SSP. The phosphorus treated soil was divided into two parts; one of them had no N-K fertilizers and the other one was thoroughly mixed with nitrogen $((NH_4)_2SO_2,$ 20.6% N) and potassium (K₂SO₄, 48% K₂O) fertilizers according to the recommended dose for maize plant growth. The fertilizers were mixed thoroughly with the treated soil just before incubation. The soil samples were wetted with distilled water up to 100% of the soil saturation ratio. The cans were incubated at laboratory conditions (29 \pm 2 °C) and wetted every five days for six cycles (20 days per cycle). A three-factor, completely randomized design was applied on the experiment (P-sources, incubation periods, and N-K fertilizer).

After each period, soil samples were collected, air-dried, ground, and passed through a 2-mm sieve. The amounts of Olsen-extractable P were determined in the incubated soil; the pH was measured in soil-water suspension (1:2.5, w:v); the soluble calcium of the clear extract of 1:5 soil suspension (W:V) was determined by titration using 0.01 N EDTA solution (Jackson, 1973)

Software and Statistical Analysis

XRD and FT-IR figures were generated from Origin Pro 8 SRI v8.0773 (B773) software, while incubation experiment figures were obtained from Excel programmer of Microsoft Office Professional Plus 2016. The data of treatments were statistically performed to the analysis of variance (ANOVA) and compare differences between the means at least significant differences (L.S.D., P < 0.05) using CoStat statistical analysis system (Version 6.303, CoHort, USA, 1998-2004).

RESULTS AND DISCUSSION

Characterization of Different P-Sources

Table (1) shows the elemental analysis and surface area parameters of the phosphate rock (PR), bone-ash (BA), and bone-char (BC). The three P-Sources are close in P and Ca contents and, consequently, in the Ca:P ratio (1.71–1.75). Additionally, increasing calcination and pyrolysis temperatures increased the mineral P content of both BA and BC by about 3.76% and 0.63%, respectively, compared to a lower temperature (600 °C). At the same time, there was an increase in calcium content in bone-ash (5.11%) and bone-char (2.58%), which reflected positively on the Ca:P molar ratios in the structural composition from 1.71 to 1.73 in BA and from 1.71 to 1.75 in BC, while this ratio was 1.72 in PR. Krzesińska and Majewska (2015) obtained a Ca:P for bone char of 1.67, which is equal to the value obtained for pure hydroxyapatite (Ca:P = 1.67) and confirms the predominance of hydroxyapatite (HAP) in the structural composition of bone-char. The total carbonate percentage was somewhat higher in the phosphate rock than in the other P-sources (BA and BC). At the same time, this percentage was relatively close in both BA and BC; this may be due to a convergence in the structural composition of both ash and char, as hydroxyapatite predominates over carbonate apatite. However, the opposite was found in the structural composition of phosphate rock. BC has the highest total carbon and nitrogen content compared to BA and PR (Table 1). This is because the pyrolysis process leads to the rearrangement of carbon atoms into the aromatic form. In contrast, calcination is accompanied by the total burning of the organic components in the structural composition, leading to the significant disappearance of C and N compounds from the structural composition of the BA. That illustrates the significantly higher percentage of carbon content in BC than in BA. On the other hand, PR has the greatest concentration of heavy

metals regardless of Fe and Mn compared to the concentrations of these minerals in BA and BC. The mineral analysis results of phosphate rock and bones were similar to those of several previous works (El-Refaey et al., 2015; Mahmoud et al., 2017 and Alotaibi et al., 2018). Bone sources have lower quantities of Cd and Cr than PR (Table 1). However, these metals pose a severe risk due to their accumulation in the soil and transfer to plants via uptake (Benavides et al., 2005 and Oliveira, 2012). Moreover, the accumulation of heavy metals in soil may inhibit beneficial biological activity, reducing the quantity and quality of agricultural production (Šmejkalová et al., 2003 and Zhao et al., 2020). Thus, the successive additions of commercial phosphate fertilizers made from PR may contain significant amounts of Cd and Cr that cause soil pollution. That confirms bone waste recycling is the potential and sustainable source of P with low content of heavy metals (Warren et al., 2009 and El-Refaev et al., 2015).

The temperature of BA calcination and BC pyrolysis has a significant effect on the surface area (SA), pore volume (PV), and mean pore diameter (MPA) of both bone productions. Raising calcination temperatures from 600 to 700 °C led to a reduction in SA of the produced bone-char about ten times, from 47.00 to 4.49 m² g⁻¹ for BA-600 and BA-700, respectively. In contrast, the same raising in the pyrolysis temperature led to a slight increase in the SA from 94.25 for BC-600 to 98.92 m² g⁻¹ for BC-700 (Table 1). Additionally, there is a direct relationship between the decrease in SA and pore volume (PV) and mean pore diameter (MPA) for both P-sources. According to IUPAC, the average pore diameter is compatible with a microporous and mesoporous structure (20 to 500 Å) (Dimovic et al., 2009). It is reported that the high temperature of pyrolysis (600 to 1000 °C) of animal bone increased the specific surface area of BC (Dimovic et al., 2009). Calcination or pyrolysis conditions of bone (temperature, residence time, oxygen limitation, and bone fraction of animals) significantly affect the surface properties, porosity, and solubility rate of P, reflecting their reactivity toward the soil components and interactions with soil biota. Moreover, there is no correlation between the SA of the tested samples (BA, BC, and PR) and the average pore diameter. Generally, the elevation of the calcination temperature (or pyrolysis) did not give a specific trend for the mean pore area for both BA and BC.

Elements	Unit	PR	BA-600	BA-700	BC-600	BC-700
Total Element						
Phosphorus	%	14.67	16.38	17.02	15.84	15.94
Calcium	%	25.3	28.01	29.44	27.12	27.82
Carbonate	%	11.03	8.92	9.41	6.52	6.74
Carbon	%	0.17	0.24	0.19	5.66	5.15
Nitrogen	%	0.21	0.34	0.28	1.65	1.16
Heavy metals						
Pb	mg kg ⁻¹	1.09	nd	nd	nd	nd
Cr	mg kg ⁻¹	89.33	5.88	6.01	7.52	7.87
Cd	mg kg ⁻¹	8.12	1.99	2.01	2.41	2.45
Fe	mg kg ⁻¹	13.09	15.20	15.66	17.22	16.98
Mn	mg kg ⁻¹	8.99	16.02	16.25	13.55	14.02
Cu	mg kg ⁻¹	8.01	nd	nd	nd	nd
Ni	mg kg⁻¹	nd	nd	nd	nd	nd
		Surface	Area Paramete	er		
SA (BTE)	$m^2 g^{-1}$	9.31	47.00	4.49	94.25	98.92
PV (P/P°)	cm ³ g	0.109	0.397	0.047	0.278	0.309
MPD	nm ²	46.67	33.77	42.08	11.81	12.57

Table 1. Total elements and surface area parameters in tested phosphate rock (PR), bone-ash calcinated at 600 °C (BA-600) and 700 °C (BA-700), and bone-char pyrolyzed at 600 °C (BC-600) and 700 °C (BC-700).

nd: not detected; SA: surface area; PV: pore volume; MPD: mean pore diameter.



Fig. 1. Images of calcinated bone-ash (BA) and pyrolyzed bone-char (BC) produced at 600 and 700 °C: (a) BA-600; (b) BA-700; (c) BC-600; and (d) BC-700, respectively.

X-Ray Diffraction

The X-ray diffraction patterns of bone-ash (BA-600 and BA-700), bone-char (BC-600 and BC-700), and phosphate rock (PR) are presented in Fig. (2). Regardless of the burning temperatures, there are no significant changes in the bands at 25.9, 31.7, 40.0, and 46.7 for all samples. Apatite crystallinity increased when the calcination temperature increased from 600 to 700 °C (Figs. 2a and b). The obtained results confirmed that the carbonate component increased in the bone-ash with temperature rises from 600 to 700 °C compared to the bone-char, as the carbonate component bands were not clearly shown in the X-Ray diffractograms (Figs. 2c and d). Wang *et al.* (2018) confirmed that the same bands were observed in the synthetic hydroxyapatite treated with different proportions of carbonates ranging

from 0.3 to 5.9% due to apatite carbonate formation (Wang et al., 2018). Additionally, it is confirmed that the crystallinity of synthetic hydroxyapatite (HAP) increased slowly from 400 to 600 °C, remarkably from 600 to 800 °C, and remained almost constant from 800 to 1000 °C (Smičiklas et al., 2005). The XRD patterns of the powders obtained at 800 and 1000 °C were similar to those of the well-crystallized synthetic HAP. The XRD patterns of BC show similar peaks; however, they are not as sharp as those of BA (Figs. 2a and b). The smooth XRD peaks of BC (Fig. 2c and d) may reflect the presence of organic carbon associated with the pyrolytic bone structure, which in turn may delay the crystallization process of the apatite formation. However, there is no conclusive evidence supporting the role of organic carbon in apatite crystallinity degree during pyrolysis of animal bone wastes, which encourages further study. The diffractograms of PR are related to HAP as the main crystalline phase present (Fig. 2e), while the broad and intense background is associated with carbonate and other inorganic compounds. The same diffractograms were observed for BA and BC (Fig. 2).

Fourier Transform Infrared (FT/IR)

Figure (3) presents the FT/IR spectra to identify the functional groups of the studied P-Sources (BA-600, BA-700, BC-600, BC-700, and PR). It was possible to verify the presence of absorbance peaks characteristic of hydroxyapatite centered at 3400, 1640, 1460, 1040, 870, and 580 cm⁻¹. The band around 3400 cm⁻¹ is characteristic of the hydroxyl stretching mode on the hydroxyapatite surface (Rojas-Mayorga *et al.*, 2015a and Reynel-Ávila *et al.*, 2016). The main indication of hydroxyapatite is the band between 950 and 1120 cm⁻¹, which is associated with the asymmetric stretching

vibration mode of the PO_4^{3-} group (Rojas-Mayorga *et al.*, 2015a&b and Reynel-Ávila *et al.*, 2016).

The absorbance band at about 1640 cm⁻¹ corresponds to stretching the carbon-oxygen double bonds in the carboxylic groups (Figueiredo et al., 2010 and Reynel-Ávila et al., 2016). The peak observed may be related to structural changes promoted by the thermal treatment of the adsorbent (Krzesińska & Majewska, 2015 and Patel et al., 2015). The band around 1440 cm⁻¹ may be attributed to the amine group (Reynel-Ávila et al., 2016). The band obtained around 2010 cm^{-1} is probably a result of overtones and combinations of peaks relating to the phosphate group (Markovic et al., 2004 and Ooi *et al.*, 2008). The carbonate group CO_3^{2-} peaks are evident at 877.64 and 1429.3 cm⁻¹ (Krzesińska & Majewska, 2015; Patel et al., 2015 and Reynel-Ávila et al., 2016). It is also possible to observe the presence of the phosphate group through the absorbance peak around 580 cm⁻¹ (Patel et al., 2015 and Reynel-Ávila et al., 2016).



Fig. 2. X-Ray Diffraction (XRD) patterns of investigated P-Sources: (a) bone-ash calcinated at 600 °C (BA-600); (b) bone-ash calcinated at 700 °C (BA-700); (c) bone-char pyrolyzed at 600 °C (BC-600); (d) bone-char pyrolyzed at 700 °C (BC-700); and (e) phosphate rock (PR).



Fig. 3. FT/IR spectra of investigated P-Sources: (a) bone-ash calcinated at 600 °C (BA-600); (b) bone-ash calcinated at 700 °C (BA-700); (c) bone-char pyrolyzed at 600 °C (BC-600); (d) bone-char pyrolyzed at 700 °C (BC-700); and (e) phosphate rock (PR).

Scanning Electron Microscopy

Scanning electron microscopy (SEM) images of investigated samples of bone-ash (BA-600 and BA-700) and bone-char (BC-600 and BC-700) produced at 600 and 700 °C (magnified at X5000) are shown in Fig. (4). In contrast to biochar (the most significant component of organic carbon), bone-ash and bone-char have an innate component of calcium phosphate (greater than 85%), so the porous surface is not clearly visible, as in the case with biochar. Increasing the temperature from 600 to 700 °C led to more pronounced calcium phosphate (or apatite), therefore, elevated levels of phosphorus and calcium, owing to the composition of the hydroxyapatite presence and dominance in both Presources (Wilson et al., 2003 and Nigri et al., 2019). Although no pores appeared on the surface of the studied samples, that does not mean the absence of pores. The surface analysis and the specific surface area the presence of results indicated nano-porous (microporous mesoporous) components and in constructing bone-ash and bone-char, as indicated in Table (1).

Solubility and Availability of P from Investigated P Sources:

The main objective of this study is to identify the solubility and availability of phosphorus from non-traditional sources of phosphorus (bone-ash "BA-600" and bone-char "BC-600") compared to non-sustainable traditional sources (phosphate rock "PR" and single superphosphate "SSP"). Table (4) shows several extraction solutions of dissolved phosphorous (distilled water (DW) and 0.005 mol L⁻¹ H₂SO₄) and available forms (NaHCO₃, CH₂O₂, and C₆H₈O₇) from the different P-Sources. The water-soluble phosphorous of bone-ash was the highest (299.25 mg P kg⁻¹), followed by bone-char (63.97 mg P kg⁻¹), and the lowest one was PR (21.85 mg P kg⁻¹). The relative water solubility to single superphosphate fertilizer (SSP), which represents 100%, was 0.64, 0.14, and 0.05%.



Fig. 4. SEM images of incinerated and pyrolyzed bovine bone: (a) bone-ash calcinated at 600 °C (BA-600); (b) bone-ash calcinated at 700 °C (BA-700); (c) bone-char pyrolyzed at 600 °C (BC-600); and (d) bone-char pyrolyzed at 700 °C (BC-700).

Extraction Method	Extracting solution	Ratio (W:V)	Shaking Time	Determination Method	Reference
Soluble Form					
Water	Distilled water	1:100	24 h	Ascorbic acid	Murphy and
				molybdate method at	Riley (1962);
Sulfuric acid	0.005 M H ₂ SO ₄	1:100	24 h	wavelength 882 nm	Watanab and
				(blue method)	Olsen (1965)
Available Form					
Olsen	$0.5 \text{ mol } L^{-1} \text{ NaHCO}_3$	1:20	30 min	Ascorbic acid	Watanab and
	(pH 8.5)			molybdate method	Olsen (1965)
Farmia anid	20 form is said (all 4)	1.10	1 1	Vanada malahdata	
Formic acid	2% formic acid (pH 4)	1:10	1 N	vanado-molybdate	Chapmann and
	at 25°C			445 nm vellow method	Pratt (1961);
Citric acid	2% citric acid (pH 4) at	1.10	1 h	The min years willow method	Hoffman and
	23 °C		1 11		Mager (1953)

Table 2. A summary of the investigated P-extraction techniques for different P-Sources.

Table 3. Physicochemical p	properties	of the	tested	soil	sampl	le
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Property		Unit	Value
Sand		%	56
Silt		%	19
Clay		%	25
Texture nomenclature			SCL
Olsen P		mg kg ⁻¹	5.6
OM		%	0.9
pH			8.2
EC,		dS m ⁻¹	1.5
Soluble ions			
	Na ⁺	meq L ⁻¹	7.6
	K ⁺	meq L ⁻¹	1.8
	Ca ²⁺	meq L ⁻¹	3.5
	Mg^{2+}	meq L ⁻¹	2.3
	Cl	meq L ⁻¹	4.5
	CO ₃ ²⁻	meq L ⁻¹	0.25
	HCO ₃ -	meq L ⁻¹	2.25

SCL Sandy Clay Loam; O.M. organic matter; EC electrical conductivity

Table 4. Soluble and available phosphorus (mg kg⁻¹) in various extractions from bone ash (BA-600), phosphate rock (PR), single superphosphate (SSP), and bone char (BC-600).

Fractions (mg kg ⁻¹)	SSP	PR	BA-600	BC-600
Water Soluble-P	46670.16 ± 1048.77	21.85 ± 0.80	299.25 ± 5.38	63.97 ± 1.89
H ₂ SO ₄ -Dissolved-P	59080.58 ± 6983.04	30.23 ± 2.18	315.49 ± 3.64	91.06 ± 5.38
Olsen-P	17596.51 ± 10.16	62.12 ± 1.32	667.40 ± 84.75	95.32 ± 2.12
Formic acid-P	33619.08 ± 1096.42	55786.27 ± 1566.27	85325.80 ± 1149.33	78268.17 ± 2186.46
Citric acid-P	28600.72 ± 1798.56	40873.30 ± 2105.45	82040.37 ± 1508.76	$67273.38 \pm \! 1903.42$

Moreover, the acid-soluble form was close to that of the concentrations of phosphorous dissolved in water (water-soluble form). The highest concentration of P was released from bone-ash (314.9 mg P kg⁻¹), followed by bone-char (91.06 mg P kg⁻¹) and then phosphate rock (30.23 mg P kg⁻¹). The results also showed a solubility rate in the diluted acid solution in order: BA-600 (0.53 %) > BC-600 (0.15 %) > PR (0.05%) relative to the acid-soluble form of the single superphosphate fertilizer (SSP).

The exposure of the bovine bone to high temperature (thermo-chemical transformation) is the main factor that plays a vital role in the quantities of phosphorous released from the bones. Nevertheless, Zwetsloot *et al.* (2015) reported that the phosphorus extraction from bone-char by water was low (3-20% of total phosphorus) but greater when extracted with formic acid (92-95% of total phosphorus), with a tendency to general for lower water solubility but greater solubility in formic acid with increasing pyrolysis temperature between 220 and 750 °C (Zwetsloot *et al.*, 2015). El-Refaey *et al.* (2015) reported that the concentration of

water-soluble P of bone-char (pyrolyzed at 650 °C for 2 hr) and phosphate rock was 54.15 and 3.06 mg P kg⁻¹, respectively, while the concentration of acidified water (0.01M H₂SO₄) was 20.96 and 134.5 mg P kg⁻¹ of bone-char and phosphate rock, respectively (El-Refaey *et al.*, 2015).

The phosphorus concentration of alkaline solution extraction (0.5 M NaHCO₃, pH 8.5) was much lower than the acid extraction (2% H_2CO_2 or $H_8C_6O_7$, pH 4) for all P-Sources (Table 4). Alkaline sodium bicarbonate solution is prescribed as one of the solutions for extracting phosphorous available to plants grown in limestone soils. Additionally, this solution represents the sources of phosphate tested (mainly calcium phosphate) that depends on the pH of the extraction environment. Accordingly, formic and citric acid solutions will dissolve significant amounts of phosphorous from different P sources corresponding to the bicarbonate solution (Table 4), which represented 898.04 and 657.97 folds for phosphate rock, 127.85, and 122.93 folds for bone-ash, and 821.11 and 705.76 folds, respectively.

The values of phosphorus availability (Olsen-P) of bone-ash and bone-char represent 10.74 and 1.53 times relative to the concentration of phosphate rock, respectively. The relatively low rate of Olsen P released from bone-char compared to that from bone-ash may be due to the organic carbon presence or, more precisely, to the organic component that contains a large amount of organic phosphorus. While the weaker phosphorus released from phosphate rock is because it is more alkaline than ash and char. In a comparative study of pyrolyzed bovine bone-char at 400 °C for 45 minutes and Gafsa phosphate rock (GPR), it was shown that the concentration of phosphorus extracted by Olsen method was 1498 mg P kg⁻¹ and water-soluble P was 244 mg kg⁻¹. These values represented 12.91 and 5.81 times the extracted from the GPR, respectively (Warren et al., 2009). In another study, it was found that the Olsen-P and soluble-P of bone-char pyrolyzed at 650 °C for 2 hours gave 1552.4 mg P kg⁻¹ and 4.9 mg P kg⁻¹, respectively (Amin, 2020). This may indicate that an increase in the pyrolysis temperature higher than 600 °C may lead to phosphorus retention in the structure of bone-char and reduce (or delay) the chances of its availability to plants in the short term. It is important to take into account the quantities of phosphorous extracted with the organic acids (formic and citric) mentioned in Table (4) and link that to the root secretions of plants grown in the soil and the role of these secretions in dissolving P. Consequently, it is expected that the phosphorous released from the phosphate sources that we are currently studying is greater than that extracted by the Olsen method.

Effect of Incubation Time on Phosphorus Availability

Table (5) shows the effect of incubation time (20, 40, 60, 80, 100, and 120 days) on the change values of soil pH, water-soluble calcium, and the available-P (Olsen method) of soil amended with different Psources (SSP, PR, BA-600, and BC-600) in untreated and treated soil with N-K fertilizers. Regardless of N-K fertilizers addition, soil pH of incubated calcareous soil amended with different P-Source significantly decreased (P < 0.05) compared to the control treatment except for the BA-600-amended soil (Fig. 5). BA-600amended soil was more alkaline (average pH: 8.63) than both PR (8.31) and BC-600 (8.40) over the course of the incubation periods. However, these changes were negligible for all treatments relative to soil treated with SSP, which had a negative effect on soil alkalinity for both treatments of N-K fertilizers addition as a result of the acidic features of SSP fertilizer. Statistically, Fig. (5) shows a significant decrease in soil pH was observed during the first 60 days of incubation, but there is no significant change in pH values in the last three periods (from 80 to 120 days). Additionally, treated soil with N- K fertilizers has a significant negative effect on soil pH during the incubation times. On average, compared to untreated soil with N-K fertilizers, the pH was decreased by 0.37, 0.08, 0.22, 0.47, and 0.33 units for and BC+N-K, SSP+N-K, PR+N-K, BA+N-K, respectively. Due to NPK nutrients may increase the biological that try to move towards neutralization or reduce the level of alkalinity in the soil, or reach the pH of the equilibrated soil (buffering capacity of soil). It is known that the level of dissolved phosphorus in the soil solution of calcareous soils depends entirely on the change in the pH of the soil solution and partly on both the dissolved calcium and the pressure of carbon dioxide (or the level of calcium carbonate in the soil). The higher the pH, the less the solubility of phosphorous that changes the phosphorous forms in soil solution from $H_2PO_4^-$ to HPO_4^{2-} and then to PO_4^{3-} (Lindsay, 1979). Based on the results in Table (5), the expected form of phosphorus in the soil solution is the most basic, such as HPO42- for the soil incubated with PR, BA-600, and BC-600, while those incubated with single superphosphate fertilizer are both H₂PO₄⁻ and HPO₄²⁻, according to the mole fraction curve proposed by Lindsay (1979). In the incubated soil amended with the examined P-sources, the lack change in the pH of soil solution over the incubation period may indicate the presence of an adequate level of phosphorous dissolved in the soil solution in the form of HPO_4^{2-} , which is the easier form absorbed by the plant (Table 5). In the study of the effect of applying three different nitrogen fertilizers (ammonium sulfate, ammonium nitrate, and urea) to calcareous sandy soil amended with bone-char (0.1%) and incubated for 48 days, the results confirmed that nitrogen fertilizers have an acidic effect on the soil (Amin, 2023). In the present study, the soil pH decreasing may be caused by ammonium sulfate which has acidified effect on soil pH due to the nitrification process during the transformation of ammonium to nitrate (Amin, 2023).

A slight significant (P < 0.05) increase in dissolved calcium was observed with an increase in the incubation period in all treatments except for soil treated with single superphosphate fertilizer. In general, the soluble calcium concentrations increased significantly with increasing incubation up to day 80 of incubation cycles. On the average soluble calcium concentration increased from 2.99 meq L⁻¹ of the control treatment to 26.56, 4.34, 1.76, and 1.85, respectively. The addition of nitrogen and potassium to the incubated soil amended with different P-Sources increased the soluble calcium concentration significantly for all treatments in order of SSP+N-K > PR+N-K > BA+N-K > BC+N-K > Control (Fig. 6). This may be due to the slight decrease in the pH of the applied soil by N and K fertilizers, and then to the occurrence of dissolution of calcium carbonate.

		,			0,,,	-					
Incubation Darra			Untreated	I N-K			Treated N-K				
Incubation Days	Cont.*	SSP	PR	BA-600	BC-600		Cont.	SSP	PR	BA-600	BC-600
						pН					
20	8.57	7.23	8.30	8.67	8.37		8.17	7.20	8.03	8.03	8.00
40	8.53	7.30	8.27	8.63	8.40		8.20	7.33	8.10	8.17	8.03
60	8.63	7.73	8.30	8.57	8.40		8.23	7.57	8.13	8.23	8.10
80	8.67	7.87	8.37	8.63	8.50		8.17	7.77	8.07	8.10	8.07
100	8.57	7.90	8.37	8.63	8.47		8.27	7.73	8.10	8.20	8.07
120	8.50	7.97	8.23	8.67	8.27		8.23	7.93	8.10	8.23	8.17
						Soluble-Ca					
20	3.13	17.77	3.43	1.87	1.93		8.87	22.67	11.57	7.50	6.30
40	2.50	21.43	4.33	1.30	1.30		8.10	24.27	11.97	6.10	5.67
60	2.53	26.00	2.50	1.30	1.30		7.50	17.77	29.30	11.67	6.63
80	3.37	37.87	5.13	1.70	2.17		8.07	40.37	13.70	6.83	7.47
100	3.33	29.17	5.70	2.37	2.37		9.90	32.77	9.20	7.50	7.50
120	3.10	27.10	4.93	2.00	2.00		8.70	34.50	12.67	7.57	5.53
						Olsen-P					
20	6.80	426.00	16.97	42.77	54.97		8.93	433.27	14.90	39.00	42.13
40	7.13	410.40	15.97	42.97	55.80		8.80	406.17	13.83	40.63	41.20
60	7.63	401.87	14.33	45.17	57.03		9.53	357.63	12.57	36.30	39.57
80	8.50	364.70	14.83	46.70	55.40		9.60	324.57	11.00	39.50	43.93
100	7.53	319.77	9.60	42.00	56.67		9.87	295.10	14.60	38.97	43.37
120	8.43	302.23	14.33	41.13	52.57		9.87	223.23	14.90	38.57	47.77

Table 5. Values of soil pH, Soluble-Ca, and Olsen-P in incubated calcareous soil treated with single superphosphate (SSP), phosphate rock (PR), boneash (BA-600), and bone-char (BC-600) in untreated and treated of nitrogen (N) and potassium (K).

*Cont. control treatment



Fig. 5. Soil pH of incubated soil for 120 days treated with single superphosphate (SSP), phosphate rock (PR), bone-ash (BA-600), and bone-char (BC-600) in untreated and treated N-K fertilizers. The different letters are significantly different at P < 0.05 according to L.S.D (least significant difference).



Fig. 6. Soluble calcium (meq L⁻¹) of incubated soil for 120 days treated with single superphosphate (SSP), phosphate rock (PR), bone-ash (BA-600), and bone-char (BC-600) in untreated and treated N-K fertilizers. The different letters are significantly different at P < 0.05 according to L.S.D (least significant difference).

The same behavior of soluble calcium, due to the incubation of sandy calcareous soil treated with bonchar and different nitrogen fertilizers was observed by Amin (2023), who found that the soluble calcium increased with the addition of different nitrogen fertilizers in order of $(NH_4)_2SO_4 > NH_4NO_3 >$ $CO(NH_2)_2 >$ control. Moreover, the expected increase in microbial activity in the soil may be accompanied by an increase in the pressure of carbon dioxide, which in turn helps in the dissolution of carbonate and the release of calcium. From another point of view, the transformation of the phosphate forms from the less basic to the more basic forms leads to the release of protons (H^+), and then provides an environment for the solubility of carbonates and the release of calcium to the soil solution (Lindsay, 1979).

Table (5) shows the behavior of available phosphorous (Olsen extraction method) in the incubated soil amended with various phosphorous sources in the presence of nitrogen and potassium fertilizers. Olsen-P decreased significantly with increasing incubation time and the addition of N-K fertilizers (Fig. 7). Generally, SSP was highly significant on the P-availability followed by BC-600 and BA-600, and the lowest one was PR compared to control (Fig. 7). Changes in available phosphorus concentrations with the incubation time were evident only for the soil amended with single superphosphate fertilizer (SSP), which concentration decreased from 426 mg P kg⁻¹ soil after 20 days of incubation and continued until it reached 302 mg P kg⁻¹ after 120 days of incubation in the untreated soil with N-K fertilizers and from 433 to 223 mg P kg⁻¹ for the same incubation periods in treated soil with N-K fertilizers. Olsen-P concentrations of BC-600 and BA-600 are nearly close compared to those of PR. Concerning phosphorus available in soils amended with the proposed P-Sources in untreated N-K fertilizers, the concentrations of P in bone-char amended soil (55 mg kg⁻¹ after 20 days to 52 mg kg⁻¹ after 120 days) were more significant than those in bone-ash amended soil (from 42 mg kg⁻¹ after 20 days of incubation to 41 mg kg⁻¹ after 120 days). In addition, both bone sources were significantly higher than that recorded for phosphate rock (from 16.97 mg kg⁻¹ after 20 days to 14.33 mg kg⁻¹ after 120 days). The average increases in available P relative to the control trial of untreated N-K fertilizers during incubation periods represented 7.28, 5.70, and

1.89 folds for BC-600, BA-600, and PR, respectively.

However, the average Olsen-P concentration in all treatments, compared to the original soil increased from 5.6 mg P kg⁻¹ soil (Table 3) to 7.67, 370.83, 14.34, and 43.46 mg P kg⁻¹ soil in untreated N-K fertilizers for control, SSP, PR, BA-600, and BC-600, respectively. While, in case of soil treated with N-K fertilizers, the average concentration of Olsen-P reached to 9.43, 340.00, 13.63, 38.83, and 43.00 for control, SSP, PR, BA-600, and BC-600, respectively. There is a significant decrease in the available phosphorus concentration in soil treated with N-K fertilizers relative to untreated N-K fertilizers (Fig. 7), which led to decreased Olsen-P on the average of 9.2%, 1.2%, 10.5%, and 22.3% for SSP, PR, BA, and BC, respectively. However, applying nitrogen and potassium to unamended soil with phosphorus in the control treatment led to an increase in Olsen-P in rates ranging from 31% to 17.1% (average, 23.5%) during incubation time (Table 5). The reduction in phosphorus availability in soil treated with N-K fertilizers may be due to the increased microbial activity and communities in soil that consume the available nutrients from the environment. Various studies reported that microbial communities have focused on fertilizer application responses. Compared to untreated soils, the P concentration reached 28% and 19% for the soil treated with char produced at 350 °C and 750 °C, respectively. The researchers concluded that bone-char is an effective P fertilizer, especially if root-arbuscular mycorrhizae (AM) interactions are simultaneously considered. Another incubation experiment for 78 days at laboratory temperature, which ranged between 24 and 28 to test bovine bone-char added to the calcareous sandy soil (pH 8.7) as a potential source of phosphorous (Amin, 2020) indicated an improvement in the level of phosphorus availability in the soil, especially when mixing the soil with elemental sulfur or the chelating compound Na-EDTA and attributed this improvement to a decrease in soil pH as a result of mixing with sulfur or calcium chelating due to mixing with Na-EDTA.

Effect of tested P sources application on soil organic carbon

According to the data in Table (1), bone-char contained a significant percentage of organic carbon (5.66% and 5.15% for BC-600 and BC-700 °C, respectively) in the structural composition compared to the rest of the sources under test that significantly impacted the soil organic carbon content (SOC). Table (6) shows the change in the percentage of soil organic carbon (SOC) due to adding different P-Sources after incubation of untreated and treated soil with N-K fertilizers for 20, 60, and 100 days. The incubation of soil with bone-char increased the content of SOC, reaching nearly double compared to the other P-Sources (SSP, PR, and BA). This increase in total carbon is expected to improve many of its physical, chemical, and biological properties; additionally, this will positively impact its productivity. Several studies have indicated the role of biochar (organic carbon is the dominant content) in improving soil properties due to the increasing soil content of organic matter (Saleh et al., 2020). Bone-char – a potential sustainable source of P – contains 5.5 – 11% organic carbon (El-Refaev et al., 2015 and Saleh et al., 2020), a potential source of stable organic carbon applied to soils. As known, pyrolysis of the various biomass through a thermo-chemical transformation produces organic matter with high stability against biodegradation (recalcitrant carbon). It stays in the soil for a long time, reaching hundreds of years. Additionally, our previous study (Saleh et al., 2020) indicated that bone-char is vital in reducing carbon dioxide (CO₂) emissions to the atmosphere and preserving carbon in the soil.

Figure (8) shows the values of increase or decrease in organic carbon (Δ SOC) in amended soil with different P-Sources and incubated for 20, 60, and 100 days in untreated and treated N-K fertilizers related to the control treatment. It was also shown that the obvious increase in carbon was in soils treated with bone-char. The addition of nitrogen and potassium did not have a clear effect, but the resulting increase in organic carbon in soil not treated with nitrogen and potassium decreased somewhat.



Fig. 7. Olsen-P (mg Kg⁻¹) of incubated soil for 120 days treated with single superphosphate (SSP), phosphate rock (PR), bone-ash (BA-600), and bone-char (BC-600) in the untreated and treated N-K fertilizers.

The different letters are significantly different at P < 0.05 according to L.S.D (least significant difference).





Fig. 8. Total organic carbon (Δ SOC %) of incubated soil treated with different P-Sources of untreated and treated nitrogen (N) and potassium (K) fertilizers compared to the total carbon percentage of control treatment.

G		Incubation days							
Source	20	60	100	20	60	100			
		Untreated	l N-K		Treated N-H	X			
Control	0.509	0.579	0.658	0.569	0.648	0.678			
SSP	0.529	0.608	0.648	0.579	0.658	0.678			
PR	0.569	0.618	0.638	0.599	0.648	0.668			
BA-600	0.599	0.638	0.668	0.628	0.668	0.698			
BC-600	0.778	0.998	1.117	0.818	0.978	1.137			

Table 6. Values of soil organic carbon (SOC) in incubated calcareous soil amended with different P-Sources in untreated and treated N-K fertilizers.

CONCLUSION

This study indicates that both bone-ash and bonechar are potential sources of phosphate that adhere to high phosphorous content, as is the case in phosphate rock on the one hand and sustainability on the other. Another advantage of bone-ash and bone-char usage is the absence of heavy metal contents compared to phosphate rock, reflecting environmental safety and preserving soil health. Continuing the availability of high levels of available phosphorous for up to 120 days (47.8 - 52.6 mg P kg⁻¹ soil in bone-char treated soil and 38.6 – 41.1 mg P kg⁻¹ soil in bone-ash treated soil) confirms the efficiency of these sources as a potential, sustainable, and safe phosphate fertilizer. Another advantage of bone-char is its organic carbon compounds, which improve the soil's organic matter content, and thus have a positive impact on its fertility. However, questions concerning temporal change related to phosphorus availability from bone-char and bone-ash in the calcareous soil and how the bone materials react to various fertilizer inputs, organic carbon, and microbial activity still need to be discovered and need further studies.

REFERENCES

- Alotaibi, K.D., J.J. Schoenau, G. Kar, D. Peak and T. Fonstad. 2018. Phosphorus speciation in a prairie soil amended with MBM and DDG ash: Sequential chemical extraction and synchrotron-based XANES spectroscopy investigations. Sci. Rep. 8, 3617.
- Amin, A.A. 2020. Sulfur, Na₂-EDTA and their mixture effects on phosphorus release from cow bone char in P-poor sandy soil. Environ. Technol. Innov. 17: 1-10.
- Amin, A.A. 2023. Effect of co-applying different nitrogen fertilizers with bone char on enhancing phosphorus release in calcium carbonate-rich soil: an incubation study. J. Soil Sci. Plant Nutr. 23: 1565–1575.
- Benavides, M.P., S.M. Gallego and M.L. Tomaro. 2005. Cadmium toxicity in plants. Braz. J. Plant Physiol. 17: 21-34.

- Bremner, J.M. and C.S. Mulvaney. 1982. Nitrogen-total. In: methods of soil analysis. Part 2. Chemical and microbiological properties, A.L. Page, R.H. Miller and D.R. Keeney (eds.). Amer. Soc. Agron. Inc. Madison, Wis. USA 595-624.
- Cao, X., Y. Wang, J. He, X. Luo and Z. Zheng. 2016. Phosphorus mobility among sediments, water and cyanobacteria enhanced by cyanobacteria blooms in eutrophic Lake Dianchi. Environ. Pollut. 219: 580–587.
- Chapmann, H.D. and P.F. Pratt. 1961. Methods of analysis for soils, plants and waters. Div. Calif., Div. Agric. Sci., Berkeley, CA 309.
- Cordell, D., J.O. Drangert and S. White. 2009. The story of phosphorus: Global food security and food for thought. Glob. Environ. Change 19: 292-305.
- Costat Version 6.303, copyright. 1998-2004. CoHort software 798 lighthouses Ave. PMB320. Monterey CA, 93940, USA.
- Deydier, E., R. Guilet and P. Scharrock. 2003. Beneficial use of meat and bone meal combustion residue: an efficient low cost material to remove lead from aqueous effluent. J. Hazard. Mater. 101: 55–64.
- Deydier, E., R. Guilet, S. Sarda and P. Sharrock. 2005. Physical and chemical characterization of crude meat and bone meal combustion residue: waste or raw material?. J. Hazard. Mater. 121:141–148.
- Dimovic, S., I. Smiciklas, I. Plećaš, D. Antonović and M. Mitrić. 2009. Comparative study of differently treated animal bones for Co²⁺ removal. J. Hazard. Mater. 164: 279–287.
- Dotelli, G. and E. Viganò. 2020. Phosphate recovery from exhausted extinguishing powders: a case study of circular economy in the chemical industry. In: S. Maranghi and C. Brondi (eds). Life Cycle Assessment in the Chemical Product Chain. Springer, Cham. 145-165.
- El-Refaey, A.A., A.H. Mahmoud and M.E. Saleh. 2015. Bone biochar as renewable and efficient P fertilizer: a comparative Study. Alex. J. Agric. Res. 60: 127-137.
- EL-Sayed, S.A.M. 2013. Relationship between the amount of available p and manure application by calcareous soils. Alex. Sci. Exch. J. 34: 354-359.

- Figueiredo, M., A. Fernando, G. Martins, J. Freitas, F. Judas and H. Figueiredo. 2010. Effect of the calcination temperature on the composition and microstructure of hydroxyapatite derived from human and animal bone. Ceram. Int. 36: 2383-2393.
- Gee, G.W. and J.W. Bauder. 1986. Particle-size analysis. In: A. Klute (ed.). Methods of soil analysis: Part 1. Physical and mineralogical methods. Agronomy monograph 9, 2nd edition. Am. Soc. Agron. Soil Sci. Soc. Am. Madison, Wisconsin 383-411.
- Ghosh, S., S. Lobanov and V.K. Lo. 2019. An overview of technologies to recover phosphorus as struvite from wastewater: advantages and shortcomings. Environ. Sci. Pollut. Res. 26: 19063-19077.
- Hodson, M.E., E. Valsami-Jones and J.D. Cotter-Howells. 2000. Bone meal additions as a remediation treatment for metal contaminated soil. Environ. Sci. Technol. 34: 3501– 3507.
- Hoffman, E. and D. Mager. 1953. Solubility of phosphoric acid and phosphate rocks. Z Pflanzenernaehr Dueng Bodenkd 62: 262-264.
- Hughes, R.E., D.M. Moore and H.D. Glass. 1994. Qualitative and quantitative analysis of clay minerals in soils. In: J.E. Amonette and L.W. Zelazny (eds.). Quantitative methods in soil mineralogy. SSSA miscellaneous publication. Mad. WI. USA.
- Imai, T. 2019. Calcination technology for manufacturing mineral fertilizer using cao-enriched sewage sludge ash. In: H. Ohtake and S. Tsuneda (eds.). Phosphorus Recovery and Recycling. Springer 179-187.
- Ishikawa K., Y. Miyamoto, A. Tsuchiya, K. Hayashi, K. Tsuru and G. Ohe. 2018. Physical and histological comparison of hydroxyapatite, carbonate apatite, and β-tricalcium phosphate bone substitutes. Mater. 11, 1993.
- Jackson, M.L. 1973. Soil chemical analysis. Advanced course Ed. 2. A manual of methods useful for instruction and research in soil chemistry, physical chemistry of soils, soil fertility and genesis.
- Krzesińska, M. and J. Majewska. 2015. Physical properties of continuous matrix of porous natural hydroxyapatite related to the pyrolysis temperature of animal bones precursors. J. Anal. Appl. Pyrol. 116: 202–214.
- Lambert, S. and N. Graham. 1989. Adsorption methods for treating organically coloured up land waters. Environ. Technol. Lett. 10: 785-798.
- Leinweber, P. 2017. Pyrolyse von schlachtknochen ein attraktiver ansatz im phosphorrecycling. In: U. Sauermann, M. Klätte (eds.). Thermopolyphos Steinbeis Edition Stuttgart 59–65.
- Lindsay, W.L. 1979. Chemical equilibria in soils. John willey and sons. New York.
- Liu, Q., S. Huang, J.P. Matinlinna, Z. Chen and H. Pan. 2013. Insight into biological apatite: physiochemical properties and preparation approaches, Review Article. Biomed. Res. Int. 2013: 1-13.

- Mahmoud, A.H., M.E. Saleh and A.A. El-Refaey. 2017. Evaluation of poultry manure and acidified water for improving phosphorus utilization from bone char: a comparative study. Egypt. J. Soil Sci. 57: 233 - 245.
- Markovic, M., B.O. Fowler and M.S. Tung. 2004. Preparation and comprehensive characterization of a calcium hydroxyapatite reference material. Res. Natl. Inst. Stand. Technol. 109: 553-568.
- Mendoza-Castillo, D.I., A. Bonilla-Petriciolet and J. Jáuregui-Rincón. 2015. On the importance of surface chemistry and composition of bone char for the sorption of heavy metals from aqueous solution. Desalin. Water Treat. 54: 1651-1662.
- Mengel, K., E.A. Kirkby, H. Kosegarten and T. Appel. 2001. Phosphorus. In principles of plant nutrition, 2nd edn. International Potash Inst. 453–477.
- Miki, T. 2019. Phosphorus separation and recovery from steelmaking slag. In H. Ohtake and S. Tsuneda (eds.). Phosphorus Recovery and Recycling. Springer Singapore 329-337.
- Murphy, J. and J.P. Riley. 1962. A modified single solution method for the determination of phosphate in natural waters. Anal. Chim. Acta. 27: 31-36.
- Nigri, E.M., A.L.A. Santos, A. Bhatnagar and S.D.F. Rocha. 2019. Chemical regeneration of bone char associated with a continuous system for defluoridation of water. Braz. J. Chem. Eng. 36: 1631-1643.
- OECD/FAO. 2018. OECD-FAO agricultural outlook 2018-2027, OECD Publishing, Paris/FAO, Rome. https://doi.org/10.1787/agr_outlook-2018-en.
- Oliveira, H. 2012. Chromium as an environmental pollutant: insights on induced plant toxicity. J. Bot. 2012: 1-9.
- Ooi, G.J., J. Fox, K. Siu, R. Lewis, K.R. Bambery, D. Mcnaughton and B.R. Wood. 2008. Fourier transform infrared imaging and small angle X-ray scattering as a combined biomolecular approach to diagnosis of breast cancer. Med. Phys. 35: 2151–2161.
- Pansu, M. and J. Gautheyrou. 2006. Carbonate. In handbook of soil analysis: Mineralogical, organic and inorganic methods. Springer-Verlag, Berlin Heidelberg New York. 593-604
- Patel, S., Han, J., W. Qiu and W. Gao. 2015. Synthesis and characterisation of mesoporous bone char obtained by pyrolysis of animal bones, for environmental application. J. Environ. Chem. Eng. 3: 2368-2377.
- Reynel-Ávila, H.E., D.I. Mendoza-Castillo, A. Bonilla-Petriciolet and J. Silvestre-Albero. 2015. Assessment of naproxen adsorption on bone char in aqueous solutions using batch and fixed-bed processes. J. Mol. Liq. 209: 187–195.
- Reynel-Ávila, H.E., D.I., Mendoza-Castillo, A. Bonilla-Petriciolet. 2016. Relevance of anionic dye properties on water decolorization performance using bone char: Adsorption kinetics, isotherms and breakthrough curves. J. Mol. Liq. 219: 425–434.

- Rojas-Mayorga, C.K., A. Bonilla-Petriciolet, F.J. Sánchez-Ruiz, J. Moreno-Pérez, H.E. Reynel-Ávila, I.A. Aguayo-Villarreal and D.I. Mendoza-Castillo. 2015b. Breakthrough curve modeling of liquid-phase adsorption of fluoride ions on aluminum-doped bone char using micro-columns: effectiveness of data fitting approaches J. Mol. Liq. 208: 114-121.
- Rojas-Mayorga, C.K., A. Bonilla-Petriciolet, J. Silvestre-Albero, I.A. Aguayo-Villarreal and D.I. Mendoza-Castillo. 2015a. Physico-chemical characterization of metal-doped bone chars and their adsorption behavior for water defluoridation. Appl. Surf. Sci. 355: 748–760.
- Saleh, M.E., A.A. El-Refaey and Y.A. Al-Damarawy. 2020. CO₂ emissions and soil organic carbon in calcareous soils as affected by bone char and phosphate rock. Egypt. J. Soil. Sci. 60: 365-375.
- Sena, M., M. Seib, D.R. Noguera and A. Hicks. 2021. Environmental impacts of phosphorus recovery through struvite precipitation in wastewater treatment. J. Clean. Prod. 280, 124222.
- Silva, Y.J.A.B.D., C.W.A.D. Nascimento, C.M. Biondi and W. Preston. 2014. Comparison of digestion methods to determine heavy metals in fertilizers. Rev. Bras. Cienc. Solo 38: 650–655.
- Skinner, H.C.W. and A.H. Jahren. 2004. Biomineralization. In: Biogeochemisty, Treatise on Geochemistry, W. Schlesinger (ed.). Elsevier 8: 118-184.
- Šmejkalová, M., O. Mikanová and L. Borůvka. 2003. Effects of heavy metal concentrations on biological activity of soil micro-organisms. Plant Soil Environ. 49: 321–326.
- Smičiklas, I., A. Onjia and S. Raicevic. 2005. Experimental design approach in the synthesis of hydroxyapatite by neutralization method. Sep. Purif. Technol. 44: 97–102.
- Sonoda, K.I. 2019. Alkaline leaching of phosphate from sewage sludge ash. In: H. Ohtake and S. Tsuneda (eds.). Phosphorus Recovery and Recycling. Springer Singapore 143–148.
- Szogi, A.A. and M.B. Vanotti. 2009. Prospects for phosphorus recovery from poultry litter. Bioresour. Technol. 100: 5461–5465.

- Wang, M., R. Qian, M. Bao, C. Gu and P. Zhu. 2018. Raman, FT-IR and XRD study of bovine bone mineral and carbonated apatites with different carbonate levels. Mater. Lett. 210: 203-206.
- Warren, G.P., J.S. Robinson and E. Someus. 2009. Dissolution of phosphorus from animal bone char in12 soils. Nutr. Cycl. Agroecosyst. 84: 167–178.
- Watanabe, F.S. and S.R. Olsen. 1965. Test of an ascorbic acid method for determining phosphorus in water and NaHCO₃ extracts from soil. Soil Sci. Soc. Am. Proc. 29: 677 – 678.
- Wilson, J.A., I.D. Pulford and S. Thomas. 2003. Sorption of Cu and Zn by bone charcoal. Environ. Geochem. Health. 25: 51–56.
- Worsfold, P.J., I. McKelvie and P. Monbet. 2016. Determination of phosphorus in natural waters: a historical review. Anal. Chim. Acta 918: 8–20.
- Yu, L., Y. Zhang, C. Liu, Y. Xue, H. Shimizu, C. Wang and C. Zou. 2019. Ecological responses of three emergent aquatic plants to eutrophic water in Shanghai, P. R. China. Ecol. Engin. 136: 134-140.
- Zhang, T., X. He, Y. Deng, D. Tsang, R. Jiang, G.C. Becker and A. Kruse. 2020. Phosphorus recovered from digestate by hydrothermal processes with struvite crystallization and its potential as a fertilizer. Sci. Total Environ. 698, 134240.
- Zhang, W., S.G. Pueppke., H. Li, G. Jianwei, Y. Diao and D.W. Hyndman. 2019. Modeling phosphorus sources and transport in a headwater catchment with rapid agricultural expansion. Environ. Pollut. 255, 113273.
- Zhao, X., Y. Sun, J. Huang, H. Wang and D. Tang. 2020. Effects of soil heavy metal pollution on microbial activities and community diversity in different land use types in mining areas. Environ. Sci. Pollut. Res. 27: 20215–20226.
- Zwetsloot, M.J., J. Lehmann and D. Solomon. 2015. Recycling slaughterhouse waste into fertilizer: how do pyrolysis temperature and biomass additions affect phosphorus availability and chemistry?. J. Sci. Food Agric. 95: 281–288.

الملخص العربى

الفوسفور والكربون العضوي المتاح كمؤشر لتقييم الفحم العظمي ورماد العظام المضاف للتربة الجيرية

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التحدى الأكبر للزراعة الحديثة هو توفر أسمدة مستدامة وصديقة للبيئة. هدفت هذه الدراسة إلى مقارنة الخصائص الفيزيائية والكيميائية لمصادرغير تقليدية للأسمدة الفوسفاتية المنتجة من عظام الأبقار في صورة رماد (BC-600 و-BA) 700 وفحم العظام (BC-600 و BC-600) ، بالإضافة إلى صخر الفوسفات (PR). اختبار مدى ذائبية تيسير الفوسفورمن المصادر المذكورة منسوبة لسماد السوبر فوسفات الأحادى (SSP) باستخدام مستخلصات مختلفة. تقييم الفسفور الميسر والكربون العضوي في التربة الجيرية المعاملة بمصادر الأسمدة الفوسفاتية التقليدية والغير تقليدية (كمقارنة) وكذلك بإضافة الأسمدة النيتروجينة والبوتاسية (N-K). حيث تم إجراء تجربة تحضين معملياً (١٢٠ يومًا، ٢٩ م^٥) بأستخدام التجارب العاملية ذات الثلاث عوامل للتصميم العشوائي الكامل. أظهرت النتائج أن الخواص الفيزيوكيميائية لفحم ورماد العظام إلى وجود الهيدروكسي أباتيت. كان لرماد العظام BA-600 أعلى قيمة للفسفور الذائب والميسر يليه BC-600 ثم PR وذلك مقارنة SSP. كانت قيم الفوسفورالذائب

في الماء وحامض الكبريتيك المخفف (M 0.005) قريبة جداً في كل من المصادر الفوسفاتية المذكورة. كما لوحظت وجود فروق معنوية (P<0.05) بين مستويات زمن التحضين ومصادر الأسمدة الفوسفاتية المختلفة وإضافة N-K على قيم الأس الهيدروجيني (pH) والكالسيوم الذائب والفوسفور الميسر. أدى إضافة N-K إلى التربة المعاملة بمصادر الأسمدة الفوسفاتية المختلفة إلى زيادة تركيز الكالسيوم الذائب معنوياً فى جميع المعاملات نتيجة انخفاض قيم pH. كان ترتيب الفوسفورالمتاح في جميع المعاملات على النحو التالي:SSP > PR < BA-600 < BC-600 > الكنترول ؛ وكانت أعلى اتاحية بعد ٨٠ يومًا من التحضين. تم تسجيل أعلى نسبة للكربون العضوي في التربة للمعاملة BC-600 وكانت الأعلى بعد ١٠٠ يوم من التحضين مع عدم وجود تأثير واضح لإضافة الأسمدة N-K. تدل النتائج على أن كلا من فحم ورماد العظام من مصادر الأسمدة الفوسفاتية المحتملة ذات الطبيعة المستدامة ، مع تفضيل فحم العظام كمصدر للكربون العضوي غير القابل للتحلل الميكروبي.