Factors driving carbon mineralization priming effect in a sandy-loam soil amended with different types of biochar

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Abstract

The effect of biochar on soil carbon mineralization priming effect depends on the characteristics of the raw materials, production method and pyrolysis conditions. The goal of the present study is to evaluate the impact of three different types of biochar ~~on soil CO_2 emissions and in different physicochemical properties~~ on physicochemical properties and CO_2 emissions of a sandy-loam soil. For this purpose, ~~a sandy-loam~~ selected soil was amended with ~~the~~ three different biochars (BI, BII and BIII) at a rate of 8 wt% and soil CO_2 emissions were measured for 45 days. BI is produced from a mixed wood sieving's from wood chip production, BII from a mixture of paper sludge and wheat husks and BIII from sewage sludge. Cumulative CO_2 emissions of biochars, soil and amended soil were well fit to a simple first-order kinetic model with correlation coefficients (r^2) greater than 0.97. Results shown a negative priming effect in the soil after addition of BI and a positive priming effect in the case of soil amended with BII and BIII. These results can be related with different biochar properties such as ~~ash~~ carbon content, carbon aromaticity, volatile matter, fixed carbon, easily oxidised organic carbon ~~oxidised with dichromate~~ or metal and phenolic substances content in addition to surface biochar properties. Three biochars increased the values of soil field capacity and wilting point, while effects over pH and cation exchange capacity were not observed.

Keywords: biochar; soil; carbon dioxide (CO_2); priming effect; physico-chemical properties, first-order kinetic model

1. Introduction

Biochar is a carbonaceous material obtained from biomass pyrolysis or gasification process. For many years now, it has been researched as a significant means to improve soil productivity, carbon storage, and filtration of soil's percolating water (Lehmann and Joseph, 2009). Biochar production emits carbon dioxide and other greenhouse gases, but combined with a proper waste disposal or biofuel production it offers a practical way to mitigate global warming (Barrow, 2012).

Nowadays, biochar production is attracting more attention because it is a safer method of organic waste management. Many types of biomass can be transformed into biochar including wood wastes, crop residues, switch grass, wastewater sludge or deinking sludges (Méndez et al, 2012; Paz-Ferreiro et al., 2014; Sohi et al., 2010). If enough farmers, larger agricultural enterprises, biofuel producers, and waste treatment plants established biochar production methods, it could reduce CO_2 emissions related to agriculture while improving soils productivity.

Biochar is a highly recalcitrant organic material, with a long-term stability in soil, which is in the scale of millennia or longer (Kuzyakov et al., 2014). The response that soil exhibits to biochar addition has global consequences for carbon cycling. Depending on the interaction between soil and biochar the ecosystem could become a sink or source of carbon.

The term priming effect refers to ~~alterations~~ increases or decreases in the mineralization of native soil organic matter due to the addition of substrates and has been observed in many studies, both in the field and under laboratory conditions (Paz-Ferreiro et al, 2012; Zavalloni et al, 2009; Zimmerman et al., 2011). While it is generally regarded that biochar addition results in a reduction in soil carbon emissions from the soil, the fact is that the results are biochar and soil specific. Indeed, previous works have shown that there is not a clear trend on CO_2 emissions after biochar application. For example, Zimmerman et al. (2011) found that carbon mineralization was generally less than expected (negative priming) for soils combined with biochars produced at high temperatures (525 and 650ºC) and form hard woods whereas carbon mineralization was greater than expected (positive priming) for soils combined with biochars produced at low temperatures (250 and 400ºC) and from grasses, particularly during the early incubation stage and in soils of lower organic carbon content.

~~that the soil application of biochars produced at temperatures between 500-600ºC had a negative priming effect due to the formation of stable aggregates and to the toxicity of biochar to soil microorganisms~~. Luo et al (2011) used biochar form plant residues and found during the first 13 days of incubation experiment, that biochar obtained at 350ºC cause a large positive priming effect, while biochar prepared at higher temperatures (700ºC) caused a relatively small positive priming effect. These authors hypothesised that priming effect was probably caused by labile organic matter remaining in the biochar after pyrolysis which in turn activated the soil microorganism. Jones et al. (2011) hypothesized that the increment in soil respiration is due to different mechanism as changes in soil physical properties (bulk density, porosity, moisture); biological breakdown of organic carbon released from the biochar; abiotic release of inorganic carbon contained in the biochar and a stimulation of decomposition of soil organic matter. Zavalloni et al. (2011) have showed that the amount of soil carbon respired was similar between the control and soil treated with biochar from coppiced woodlands pyrolysis in a short term incubation experiment. Also, Wardle et al. (2008) reported priming effect from a boreal soil after biochar addition, although the results of this experiment have been disputed by others (Lehmann and Sohi, 2008). If a strong positive priming effect occurs after biochar addition to the soil, then the beneficial effects attained by biochar addition to the soil becomes mitigated. Furthermore, although the use of biochar measuring soil respiration has been evaluated (Méndez et al, 2012; Zimmerman et al., 2011) fewer studies have studied the role of biochar addition of native soil organic matter (Zimmermann et al., 2011, Cross and Sohi, 2011, Gascó et al., 2012). For example, Gascó et al. (2012) observed using thermal methods that there is a degradation of more complex structures after application of a sewage sludge biochar to a Haplic Cambisol. The final chemical composition and physical properties of biochar, and thus, its potential for having a positive or negative priming effect depends on the characteristics of the raw materials, production method and pyrolysis conditions. Different studies has been performed in order to study the influence of feedstock, production method and pyrolisis temperature on biochar properties and uses (Calvelo Pereira et al, 2011; Méndez et al, 2012; Zimmermann et al., 2011; Paz-Ferreiro et al., 2014).

In the present work, three different biochars were used in order to study their influence on soil properties and CO_2 emissions. ~~Three~~ Biochars were obtained from pyrolysis of different types of biomass: mixed wood sieving's from wood chip production, paper sludge and wheat

husks and sewage sludge at temperatures between 500 and 620ºC using slow pyrolysis processes.

2. Materials and Methods

2.1 Soil selection and characterization

The selected soil was taken from the north-east of Toledo (Spain) and the soil was air-dried, crushed and sieved through a 2 mm mesh prior to analyses. The initial pH and electrical conductivity (EC) were determined with a soil:water ratio of 1:2.5 (g mL^{-1}) using a Crison micro-pH 2000 (Thomas et al., 1996) ~~in the case of pH~~ and a Crison 222 conductivimeter (Rhoades, 1996) ~~in the case of EC~~ respectively. CEC was determined by NH_4OAc/HOAc at pH 7.0 (Sumner and Miller, 1996). Total organic matter (TOM) was determined using the dry combustion method at 540 ºC (Nelson and Sommers, 1996). Soil metal content was determined using a Perkin Elmer 2280 atomic absorption spectrophotometer after sample extraction by digestion with concentrated HCl/HNO_3 following method 3051a (USEPA, 1997). Soil texture was determined following the methodology of Bouyoucos (1962). These analysis were performed by triplicate.

2.2 Biochar characterization

Three different biochar samples were selected and used for the present work: biochar I (BI) was produced by Swiss Biochar (Lausanne; Switzerland) from mixed wood sieving's from wood chip production at 620ºC; biochar II (BII) was produced by Sonnenerde (Austria) from a mixture of paper sludge and wheat husks at 500ºC; and biochar III (BIII) was produced by Pyreg (Germany) from sewage sludge at 600ºC. The pyrolisis duration was 20 minutes on all cases. All biochar samples were produced using Pyreg500-III pyrolysis (Germany) units which can work until 650ºC in a continuous process.

The pH, EC, CEC and metal content in biochars were performed as in Section 2.1. Proximate analysis was determined by thermogravimetry using a Labsys Setaram equipment. The sample was heated to a temperature of 600ºC under N_2 atmosphere and 30ºC min^{-1} heating rate. Humidity was calculated as the weight loss from the initial temperature to 150ºC. The volatile matter (VM) was determined as the weight loss from 150ºC to 600ºC under N_2 atmosphere. At this temperature, air atmosphere was introduced and fixed carbon (FC) was calculated as the weight produced when the final sample was burnt. The ashes were determined as the final weight of the samples. The content in C, H, N and S was analysed by an elemental microanalyzer LECO CHNS-932 and the oxygen content was

determined by difference. Biochar nitrogen adsorption analysis to determine BET surface area was carried out at 77 K in a Micromeritics Tristar 3000. Also, biochar CO_2 adsorption analysis to determine both CO_2 micropore surface area and monolayer capacity were performed at 273 K in a ASAP 2020 V3.01

Finally, biochar phenolic substances were determined using Folin-Ciocalteu´s reagent (Martín-Lara et al., 2009).

2.3 Treatments and soil respiration

The selected soil (S) was amended with the three biochar samples at 8 wt% (S+BI, S+BII, S+BIII) and mixtures were incubated at constant temperature (28 ± 2ºC) and humidity (60% FC) during 45 days. Additionally, it was studied if the application of the different amendments had an additive or synergistic effect in the soil (priming effect); in this way each biochar (BI, BII, BIII) was incubated individually in the experimental conditions.

Each sample (100 g) was introduced at 1L airtight jar and the CO_2 produced during incubation was collected in 50 mL of a 0.3N NaOH solution, which was then titrated using 0.3N HCl after the $BaCl_2$ precipitation of the carbonates. All treatments were performed by triplicate.

Organic carbon oxidised with dichromate from initial and final biochars were determined by the Walkley-Black method (Nelson and Sommers, 1996).

After incubation time, the next soil properties were determined: pH, EC, CEC, field capacity (FC), wilting point (WP) and available water (AW). pH, EC and CEC were determined as in section 2.1. Field capacity (FC) and wilting point (WP) were determined as the soil moisture content at 33 kPa (FC) and 1500 kPa (WP) (Richards, 1954). Available water (AW) was calculated as the difference between FC and WP. All analyses were performed by triplicate.

In addition, thermal analysis (TG, dTG and DTA) of soil was performed in a thermogravimetric equipment Labsys Setaram. About 50 mg of each sample were heated at 15 ºCmin^{-1} until 850 ºC in air atmosphere using a flow rate of 40 mL min^{-1}.

2.4. Mineralisation model

The cumulative mineralisation data were fitted to a first-order kinetic model, which is widely used to model soil respiration data (Méndez et al., 2013). The kinetic model used to calculate the evolved CO_2-C soil is described as follows:

$$Y = Ct^m \quad (1)$$

where Y is the cumulative CO_2-C (mg CO_2-C 100 g^{-1} soil), t is the cumulative time of incubation (d), and C and m are the mineralisation constants, with C·m representing the initial mineralisation rate. The convexity shape of Y in this model is defined mainly by m, with $m \leq 1$ and $C \geq 0$. This equation was fitted to describe the C mineralisation in S, the biochars (BI, BII and BIII) and the amended soils (S+BI, S+BII and S+BIII). The mineralisation rate parameters of Eq. 1 were estimated by a non-linear-model method, minimising RMSA.

To quantify the priming effect of the three raw materials, the model was fitted to the experimental data (Experiment) and to the respiration data with the addition of 92 g of soil with 8 g of biochars (Addition). Also, C_{10} was calculated as the evolved CO_2-C after 10 days according the model.

3. Results and Discussion

Table 1 shows main properties of the soil and three biochars. Soil texture was sandy loam, it had a slightly alkaline pH, the EC value indicated that soil has no risk of salinisation and soil organic matter content was 6.30%.

With respect to biochars, BI and BII showed basic pH whereas BIII had a pH value near 7. Proximate analysis of three biochar samples showed differences in their composition. The ash content of biochars followed the next sequence BIII > BII > BI depending on the feedstock, i.e., BI is prepared from woodchip, BII from paper sludge and wheat husk and BIII from sewage sludge presenting a higher mineral content. Indeed, BIII had the highest EC and metals content. Biochar metal content did not exceed the limit values for concentrations of metals in soil set up by the European Union (European Community, 1986) with BIII presenting the highest content, which can be explained according to its origin. All biochars presented a similar CEC which can be related with the comparable temperature of preparation. Volatile matter content of BI and BIII was similar and lower than that of BII. Fixed carbon of BI was significantly higher than that of BII and BIII. Combining VM and FC, the ratio FC/(FC+VM) could be indicative of the carbon stability. According to this, BI was a very recalcitrant carbon material, whereas BIII showed the lowest ratio. The molar H/C ratio was used as an indicator of the degree of aromatization. This ratio shows the sequence BI<BII<BIII. The O/C ratio was indicative of the degree of carbonization

following the same trend that H/C ratio, BI<BII<BIII. According to previous studies on biochars (Kuhbusch and Crutzen, 1995; Hammes et al., 2006) the H/C ratio of ≤ 0.3 (like BI) indicates a highly condensed aromatic ring system whereas H/C ratio of ≥ 0.7 (like BIII) represents a non-condensed structure.

Table 2 shows the changes of pH, EC and CEC after the 45 days of incubation experiment. Instead, biochar pHs were different (Table 1), pH did not change after biochar application though BI and BII presented pH 2 units higher than soil. Conversely, other studies have shown pH increments after biochar application. For example, Méndez et al (2012) observed an pH increment on an Haplic Cambisol after the addition of sewage sludge-derived biochar, Kloss et al. (2014) described a slightly increment of soil pH (0.3 units) in an acid soils after application of woodchip-derived biochar or Jien and Wang (2013) observed a significant increased in Ultisol pH from 3.9 to 5.1 after addition of biochar made from the waste wood of white lead trees. So, both biochar and soil composition influences the pH changes. ~~However, the electrical conductivity increased slightly depending on biochar electrical conductivity (Table 1)~~ Biochar addition slightly increased soil EC (Table 1) but the risk of salinisation was negligible at the applied dose (USDA, 1999). The increased in soil EC is very common in soils treated with biochar prepared from sludge, which is the case of BII and BIII, as reported in other studies Hossain et al. (2010) or Méndez et al. (2012). With respect to CEC, biochars did not increase soil CEC, a result according to previous works (Méndez et al, 2012) and which can be related with the low CEC of biochar with respect to soil OM (Lehmann, 2007).

Biochars increased the values of soil FC and WP following~~, respectively,~~ the ~~following~~ sequence S < S+BIII < S+BI ≈ S+BII for both properties ~~and S < S+BIII < S+BI ≈ S+BII~~. Also, there was an increment in the AW when the soil was treated by BI and BII. This improvement of water retention is in accordance with the results previously obtained by ~~of~~ Méndez et al. (2012) which found the same trend in a soil with a similar sand content treated with biochar prepared for sewage sludge at 600ºC. The higher increment of FC, WP and AW in S+BI and S+BII treatments could be related with the higher values of FC and WP of these biochar according to their high surface area and porosity (Table 1).

In the last years, thermal analysis has been proposed as an interesting technique in the characterization of organic matter stabilization processes. Additionally, it has been applied

to soil characterization to assess proportions of labile and recalcitrant organic matter (Plante et al., 2009) and to study the evolution of organic matter in amended soils (Barriga et al., 2010; Gascó et al., 2012). Thermal analysis has the advantage to provide information about the chemical characteristics of soil organic matter without any extraction step as all sample was analyzed. Figure 1 shows dTG (Figure 1.a) and DTA (Figure 1.b) of S, S+BI, S+BII and S+BIII samples after incubation period. Different peaks were observed in Figure 1, at temperatures lower than 150ºC, water releases was observed, then at temperatures from 200 to 650ºC, oxidation of organic matter takes place. Initially, weight loss corresponds to less humified matter (from 200 to 400ºC) whereas the peak observed at temperatures highest than 400ºC correspond to more humified organic matter. At temperatures higher than 550 ºC weight loss could be attributed to refractory carbon from biochars and clays decomposition (Gascó et al., 2012).

From DTA curve, it could be observed the first endothermic peak at temperatures lower than 150 ºC due to moisture release from soil sample. Then, two small exothermic peaks could be observed between 200 and 650ºC due to combustion reactions of soil organic matter. It is established that first peak was associated with combustion of less humified organic matter, whilst the second one was related to the more humified. Four samples show at 573 ºC, the characteristic small endothermic peak due to the quartz α-β inversion. Comparison of four samples in Figures 1.a and 1.b shows the influence of different biochars in soil organic matter composition. Biochar addition increases the amount of more humified or thermally stable organic matter following the sequence S+BI>S+BII>S+BIII. It was interesting to note that S+BIII shows a thermal behavior similar to that of unamended soil (S) indicating a similar organic matter composition that original soil.

With respect to biochar CO_2 emissions, these were higher in BI while significant differences between BII and BIII were not found. This fact can be attributed to the elevated ~~FC+VM ratio~~ carbon content of BI (82%) respect to BII (65.15%) and BIII (26.54%). In order to explain the similar CO_2 emissions of BII and BIII other factors needs to be account (Jones et al, 2011). Calvelo Pereira et al. (2011) found that dichromate oxidation reflect the degree of biochar carbonization and could therefore be used to estimate the labile fraction of carbon in biochar. Figure 2 shows as BIII with highest ash content and lowest C content and consequently, expected lowest CO_2 emissions, has the highest content of labile ~~organic~~ carbon ~~dichromate oxidised carbon and consequently, the highest labile carbon content.~~ So, the H/C and O/C ratios have showed that BIII has non-condensed organic structures. After

incubation, the labile carbon of BI decreases whereas that of BII and BIII slightly increases, indicating that some of the more stable organic structures were transformed into labile carbon. This result was according with that obtained previously by Gascó et al. (2012) using thermal analysis and biochar form sewage sludge. However, for BI the labile carbon slightly decreases after incubation.

Results shown that biochar addition increased CO_2 soil emissions approximately by 25%, but there were not differences between the different treatments (Figure 3). ~~Zavalloni et al. (2011) also found that the amount of soil carbon respired was similar between the control and the soil amended with biochar.~~ On the other hand, Zavalloni et al. (2011) found that respiration rate in soil with coppiced woodlands derived biochar were not significantly different from control soil. This matter can be attributed to combination of different factors not only to one. Méndez at al. (2013) found that higher CO_2 emissions can be related with higher content of VM ~~(BII)~~ and lower values of ratio FC/(FC+VM) from biochars. Also, the CO_2 evolved can be related with the ~~variation of oxidisable organic~~ labile carbon content of biochars (Figure 2). On the other hand, different authors (Méndez et al, 2013; Thies and Rillig, 2009) observed that the reduction of CO_2 emissions can be attributed to chemisorptions of the respired CO_2 on biochar surface. Indeed, BI had a CO_2 micropore surface area and CO_2 monolayer capacity more than 44% higher than BI and BII. So, their labile carbon content was lower. Also, H/C, O/C and FC/(FC+VM) ratios indicates that instead of their high carbon content it was a more stable carbon material. Finally, the electrical conductivity, ~~combination of~~ metal and phenolic substances of biochar can have negative effect on soil microbial activity reducing the respired CO_2. Table 4 summarizes the qualitative influence of different factors on CO_2 emissions and it shows an orientation about the influence of different biochar properties on the increment of soil CO_2 emissions after biochar application. pH limits have been fixed following the classes of soil pH of USDA (1998) and the guidelines to biochar production according (Schmidt et al, 2012). It must be pointed that pH of 6.6 to 7.3 is favorable for microbial activities that contribute to the availability of nitrogen, sulfur, and phosphorus in soils (USDA, 1998) and pH value exceeding 10 can have negative effects on soil pH but it must note that only the application of larger amounts of biochar will lead to changes in a soil's pH value (Schmidt et al, 2012). With respect to electrical conductivity, limits have been fixed according to the limits fixed by Richards (1954) where the high value (4 dS m^{-1}, 25 ºC) is the limit between normal and saline soils. The organic carbon limits have been fixed according to International Biochar

Initiative (2012) and the recommendations of Schmidt et al (2012) who described that organic carbon content of pyrolysed chars fluctuates between 10% and 95% of the dry mass dependent on the feedstock and process temperature used. With respect to volatile matter (VM) and fixed carbon (FC), values over 20% and 40% of VM and FC can be considered high according biochar prepared from different fedstocks as sewage sludge (Gascó et al, 2012; Méndez et al, 2012), rice husk (Kalderis et al, 2014), eucalyptus wood or poultry litter (Paz-Ferreiro, 2012; Lu et al, 2014). Finally, BET surface area values shuold be preferably higher than 150 m^2 g^{-1} (Schmidt et al, 2012) being values over 750 m^2 g^{-1} very high and of the same order that montmorillonite. It must stand out that the negative effects are usually due to a combination of different factors and not can be attributed to a unique factor.

Table 5 and Figure 3 show the parameters estimated according to simple first-order kinetic model to describe the C mineralization in soil (S), biochars (BI, BII, BIII) and amended soils (S+BI, S+BII, S+BIII). The kinetics of CO_2 evolved from biochars was well fit to the proposed model presenting r^2 values higher to 0.97. With respect to the amended soils, the fit presented a Root Mean Square Deviation (RSMD) lower than 2 and r^2 values higher than 0.99. In fact, this model of simple first-order kinetic model has been successfully used to estimate CO_2 emissions from biochar and biochar amended soil in short term incubation experiment (Méndez et al, 2013).

Also, results shown that the application of BI had a negative priming effect if data of the experiment (57.1 mg C-CO_2/100g**)** and addition (63.0 mg C-CO_2/100g) are compared (Table 4) according with the similar values of model parameters (m and C); ~~this fact probably can be due to the toxic effect of phenolic substances of BI on soil microorganism.~~ This result was according to that obtained by Zimmerman et al (2011) that found as biochar produced at high temperatures and from hard woods like BI show negative priming. With respect to the application of BII and BIII to soil, results showed a positive priming effect. It is interesting to note that both biochars increases their labile carbon content during individual incubation (Figure 2) whereas for BI, their content slightly decreases. ~~being~~ The initial organic matter mineralisation was very similar in all cases (C parameter ranged from 6.07 to 7.91) according to Méndez et al. (2012) which found an increment of CO_2 emissions after application at the same rate after application of biochar prepared from sludge to a similar sandy soil or results obtained by Smith (2010). Nevertheless, Paz-Ferreiro et al. (2012) found a negative priming effect after sewage sludge biochar application (prepared at 650ºC) to an Umbrisol. Indeed, Zimmerman et al. (2011) concluded that discrepancies in C

mineralization of biochar-treated soils are likely due to the type of both soil and biochar, the duration of the experiment and the dose of used biochar.

Finally, C_{10} parameter, i.e. evolved CO_2-C after 10 days according the model, is related with the labile fraction of biochar to be released by microbial activity. Results show that experimental data were very similar and the different between experiment and addition (Table 4) in the case of S+BI could suggest a toxic effect of biochar.

4. Conclusions

The effect of biochar on soil carbon mineralization priming effect depends on the characteristics of the raw materials, production method and pyrolysis conditions. Indeed, results shown a negative priming effect in the soil after addition of BI (prepared at 620ºC from a mixed wood sieving's from wood chip production) and a positive priming effect in the case of soil amended with BII (prepared at 500ºC from a mixture of paper sludge and wheat husks) and BIII (prepared at 600ºC from sewage sludge). These facts can be related with different biochar properties such as carbon content, carbon aromaticity, volatile matter, fixed carbon, easily oxidised organic carbon, metal and phenolic substances content and surface biochar properties. In addition, experimental results show that cumulative CO_2 emissions were well fit to a simple first-order kinetic model for the different biochar and amended soil. Also, biochars addition improved water soil retention. Finally, further research is required to determine the importance of the different biochar properties involved in soil CO_2 emissions.

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Table 1. Main properties of the soil (S) and biochars

	S	BI	BII	BIII
pH (1:2.5)	7.66±0.10	10.19±0.12	9.40±0.19	7.66±0.13
EC (1:2.5 (dS m^{-1}, 25 ºC)	70±10	1776±44	2330±50	3700±157
TOM ~~C~~(%)	6.30±0.15	87.71±0.71	59.90±0.89	25.15±0.40
CEC ($cmol_{(+)}$ kg^{-1})	15.87±0.25	23.77±0.36	20.97±0.24	24.19±0.30
Cd (mg kg^{-1})	-	0.43±0.05	0.72±0.08	4.98±0.01
Cr (mg kg^{-1})	-	21±2	32±4	76±8
Cu (mg kg^{-1})	-	61±9	37±8	406±25
Ni (mg kg^{-1})	-	18±1	30±1	78±10
Pb (mg kg^{-1})	-	4±1	24±3	141±10
Zn (mg kg^{-1})	-	47±5	134±9	1350±49
Phenolic substances (mg gallic acid g^{-1})		0.93±0.05	1.01±0.07	0.49±0.04
Sand (%)	77.78	-	-	-
Silt (%)	17.78	-	-	-
Clay (%)	4.44	-	-	-
Soil textural class (%)	Sandy loam	-	-	-
FC(%)		113±1	122±1	36±1
WP(%)		52±1	63±1	31±1
AW(%)		61±1	59±1	5±1
BET Surface Area (m^2 g^{-1})	-	332.138	92.6115	59.1572
Micropore area (m^2 g^{-1})	-	305.9972	66.9119	30.9545
Adsorption average pore width (Å)	-	21.2622	32.9697	77.1478
CO_2 micropore surface area (m^2 g^{-1})		414.206	229.399	86.329
CO_2 monolayer capacity (cm^3 g^{-1})		90.672	50.217	18.898
Proximate analysis				
VM (%)[a]	-	14.88	22.43	13.68
FC (%)[b]	-	77.25	42.72	12.77
Ash (%)	-	7.87	34.85	73.55
FC/(FC+VM)	-	0.84	0.66	0.48
Elemental analysis				
C (%)		82,00	50,75	18,45
H (%)		1,49	1,73	1,19
N (%)		0,33	1,36	2,10
O (%)		5,76	12,08	7,69
H/C atomic ratio		0,018	0,034	0,064
O/C atomic ratio		0,070	0,238	0,417

[a]VM: Volatile matter, [b]FC: Fixed carbon.

Table 2. pH, electrical conductivity (EC), cation exchange capacity of treated soils after the incubation experiment

	pH	EC ($\mu S\ cm^{-1}$)	CEC ($cmol_{(c)}\ kg^{-1}$)
S	7.45ab	496a	15.71a
S+BI	7.68b	535a	16.28a
S+BII	7.47ab	624b	16.08a
S+BIII	7.29a	764c	17.07a

Values in column followed by the same letter are not significantly different (P = 0.05) using Duncan test
The number of replicates were 3 for each determination.

Table 3. Field capacity (FC), wilting point (WP) and available water (AW) after the incubation experiment

	FC(%)	WP(%)	AW(%)
S	13.54a	11.04a	2.49a
S+BI	20.41c	13.79c	6.61b
S+BII	20.24c	13.91c	6.33b
S+BIII	16.31b	12.72b	3.60a

Values in column followed by the same letter are not significantly different (P = 0.05) using Duncan test
The number of replicates were 3 for each determination.

572 Table 4. Influence of different biochar properties on the increment of soil CO_2 emissions
573 after biochar application

Value	pH	Electrical conductivity	Organic carbon	Metal content	Phenolic substances	Volatile matter	Fixed carbon	BET surface area
High[b]	-[a]	-	+	-	-	+	+	-
Normal	+	+	+	+	+	+	+	-
Low	-	+	-	+	+	-	-	-

574 [a]+: positive effect; -: negative effect

575 [b]: **pH** (USDA, 1998; Schmidt et al, 2012): High: >10, Normal: 6-10, Low : <6. **Electrical Conductivity**
576 (Richards, 1958): High: >4 dS m^{-1}, Normal: 4-2 dS m^{-1} , Low < 2 dS m^{-1}. **Metal content** (European
577 Community, 1986): High: Cd >40 mg Kg^{-1}, Cu >1750 mg Kg^{-1}, Ni > 400 mg Kg^{-1} , Pb > 1200 mg Kg^{-1}, Zn >
578 4000 mg Kg^{-1}, Hg > 25 mg Kg^{-1}; Normal: Cd 20-40 mg Kg^{-1}, Cu >1000-1750 mg Kg^{-1}, Ni > 300-400 mg Kg^{-1}
579 , Pb > 750-1200 mg Kg^{-1}, Zn > 2500-4000 mg Kg^{-1}, Hg > 16-25 mg Kg^{-1}; Low: Cd <20 mg Kg^{-1}, Cu <1000 mg
580 Kg^{-1}, Ni < 300 mg Kg^{-1} , Pb < 750 mg Kg^{-1}, Zn < 2500 mg Kg^{-1}, Hg < 16mg Kg^{-1}. **Organic carbon**
581 (International Biochar Initiative, 2011): High: > 50%, Normal: 30-60% , Low < 10%. **Phenolic substances**
582 (Kuiters and Sarink, 1986): High: > 10 µg g^{-1}, Normal: 10-1 µg g^{-1} , Low : < 1 µg g^{-1} . Volatile matter: High:
583 > 20%, Normal: 20-10% , Low : < 10%. **Fixed carbon**: High: >40 , Normal: 40-20 , Low : <20. **BET surface**
584 **area** (Schmidt et al, 2012): High: > 750 m^2/g, Normal: 750-150 m^2/g , Low : < 150 m^2/g.
585

586

Table 5. CO_2-C evolved (mg CO_2 100 g^{-1} dry weight) during incubation experiment and parameters estimated according to simple first-order kinetic model to describe the C mineralization in soil (S), biochars (BI, BII, BIII) and amended soils (S+BI, S+BII, S+BIII). Mineralisation constants (C and m), Root Mean Square Deviation (RMSD), correlation coefficient (r^2) and coefficient of determination (R^2) of the fitted model are shown.

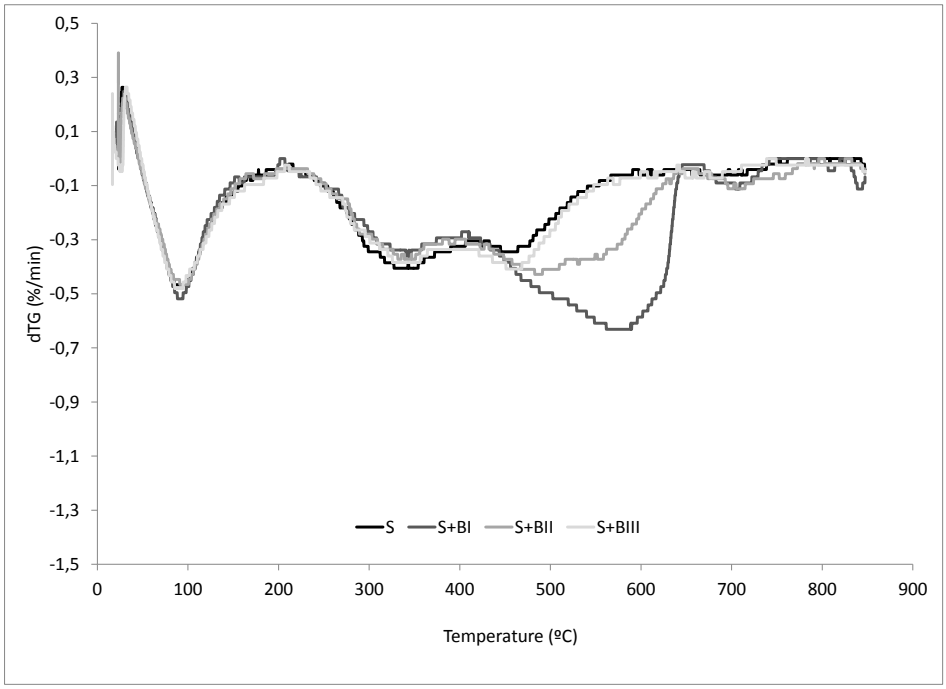
Substrate		CO_2 evolved (mg C-CO_2/100g)	m	C	RMSD	r^2	C_{10} [b] (mg C-CO_2/100g)
S		45.8	0.5524	5.81	1.23	0.996	20.72
BI		261.2	0.5513	32.15	10.94	0.989	114.41
BII		120.1	0.4092	25.51	6.69	0.975	65.46
BIII		125.6	0.5046	19.34	6.26	0.985	61.79
S+BI	Experiment	57.1	0.5606	6.83	0.94	0.998	24.83
	Addition[a]	63.0	0.5521	7.91	1.34	0.997	28.22
S+BII	Experiment	58.3	0.5987	6.07	0.86	0.999	24.10
	Addition	51.7	0.5262	7.22	1.22	0.997	24.25
S+BIII	Experiment	56.1	0.5872	6.08	0.82	0.999	23.50
	Addition	52.2	0.5434	6.87	1.40	0.996	23.99

[a]The addition of the experimental data has been made taking into account a dose of 8%

[b] C_{10} is the evolved CO_2-C after 10 days according the model

601 Figure 1. dTG (1.a) and DTA curves (1.b) of soil and soil amended with biochar

602 1.a) after incubation period



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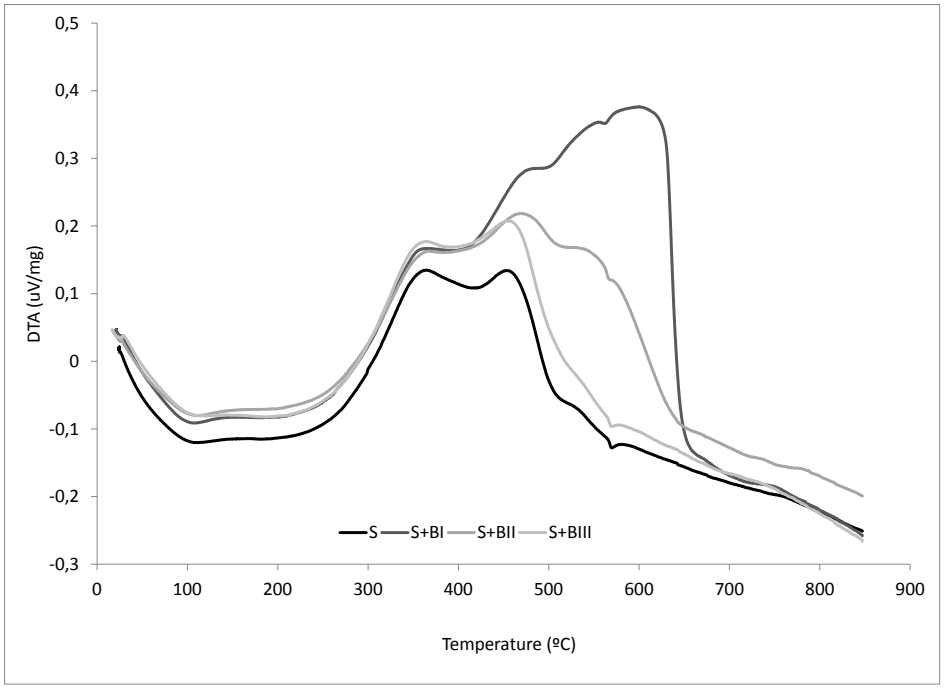
604 1.b)



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606 Figure 2. Evolution of organic carbon oxidised with dichromate. Values in column followed
607 by the same letter are not significantly different (P = 0.05) using Duncan test
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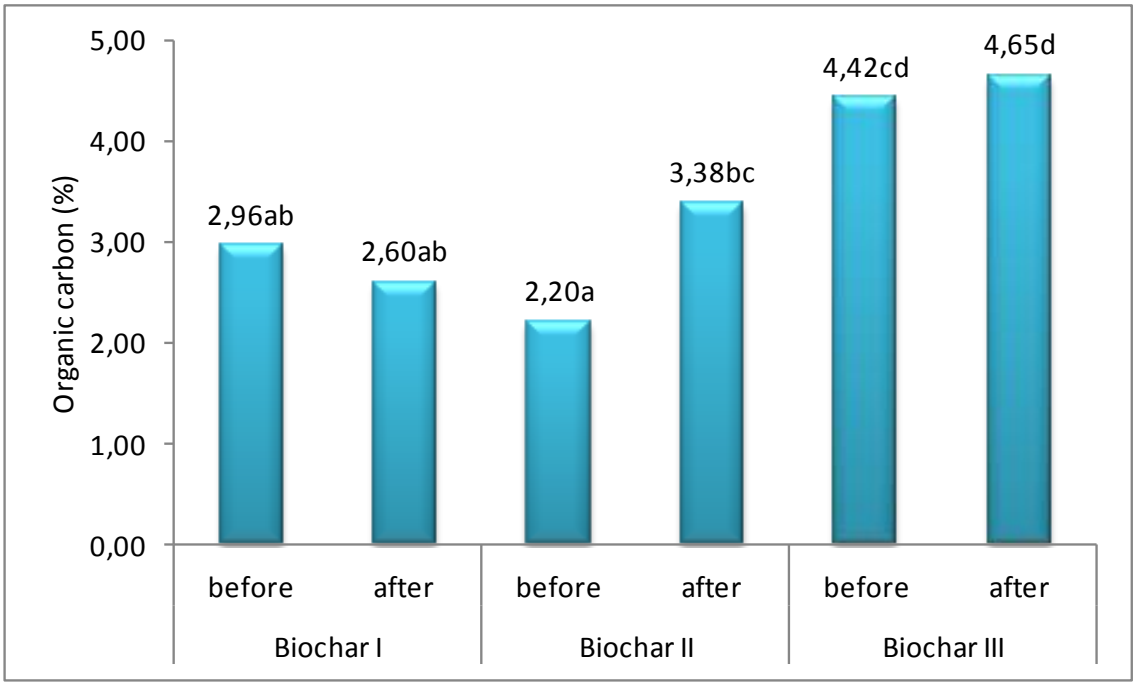

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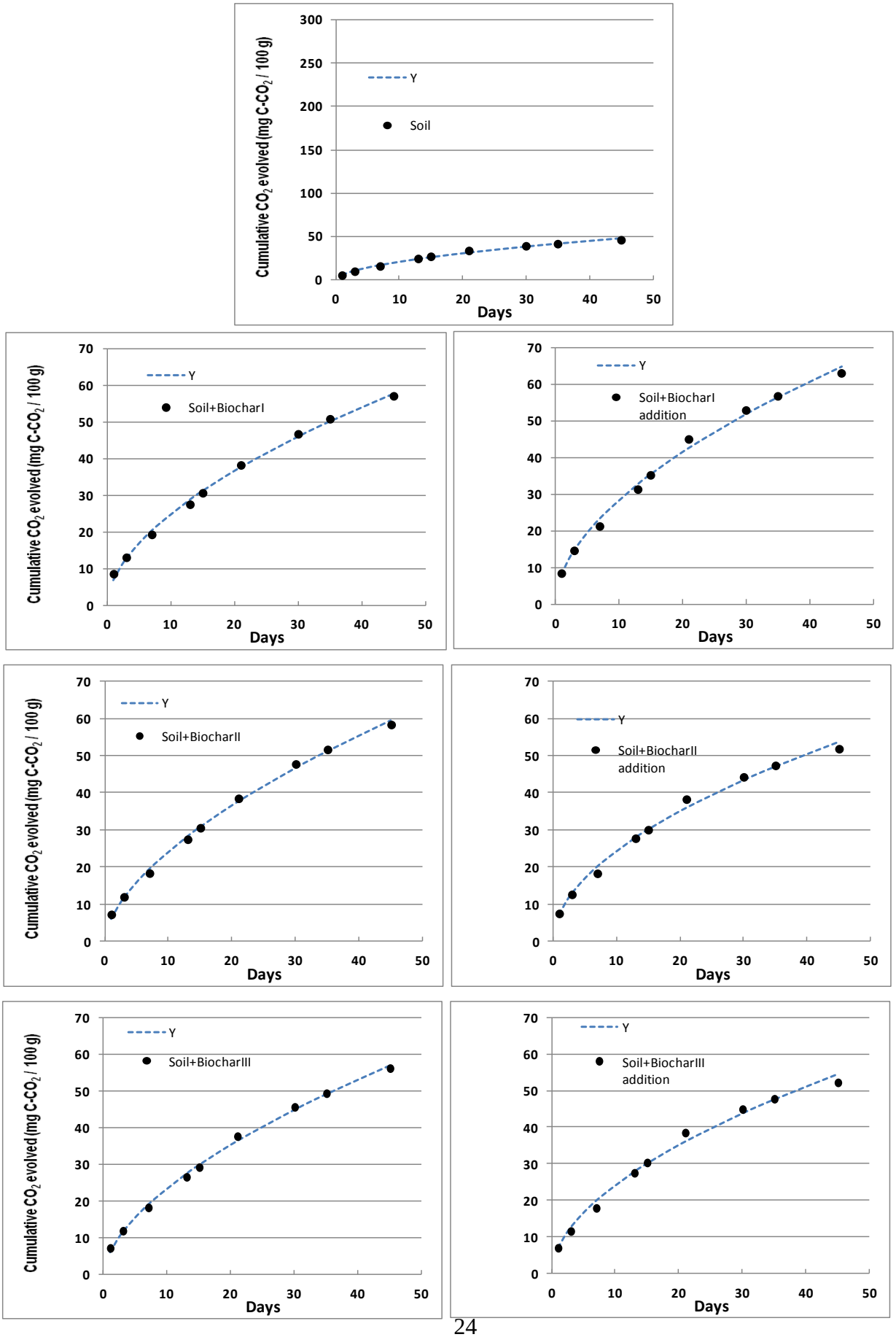
611 Figure 3. Exponential model of measured C mineralized (as CO_2) and that calculated by
612 addition of soil and BI, BII and BIII effects.



613 Figure 4. Exponential model of measured C mineralized (as CO_2) in BI, BII and BIII
614 biochars

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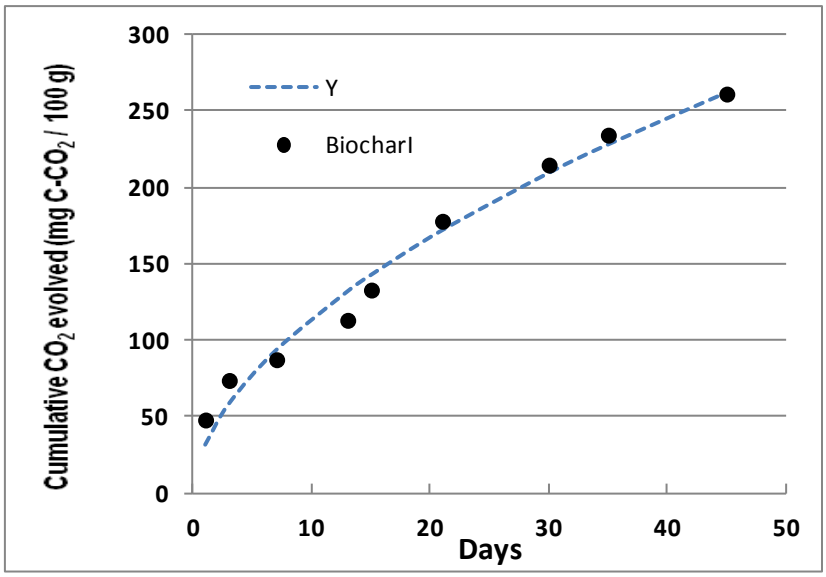

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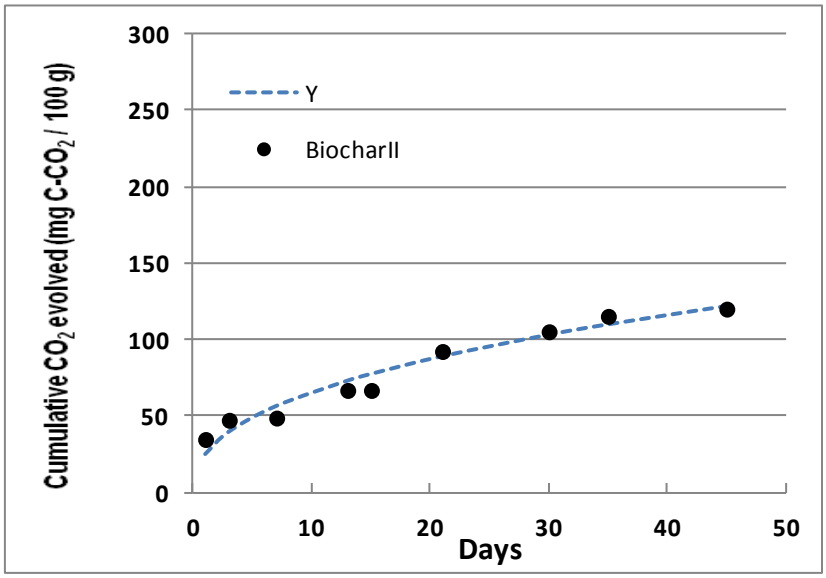

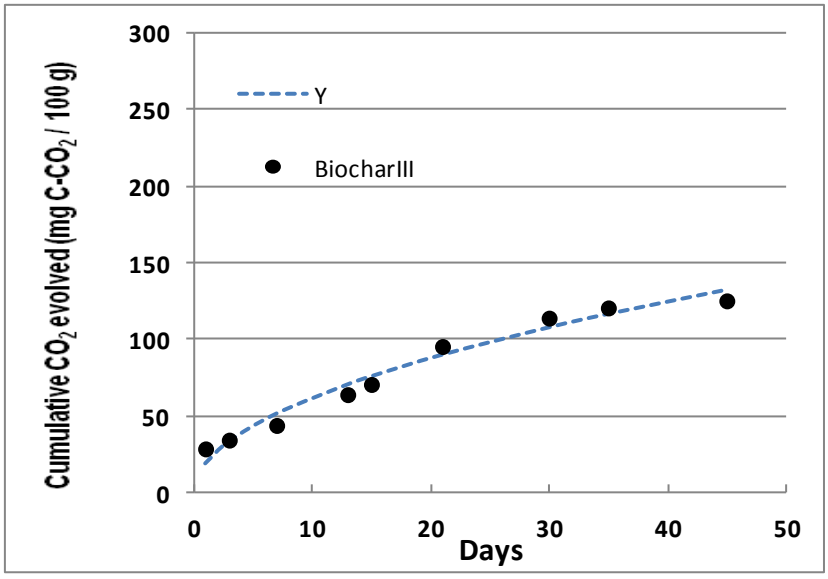

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