An investigation of the effects of hydrochar application rate on soil amelioration and plant growth in three diverse soils

Megan de Jager¹ · Luise Giani¹

Received: 28 September 2020 / Accepted: 22 January 2021 / Published online: 7 April 2021
© The Author(s) 2021

Abstract

The hydrothermal carbonization (HTC) of biogas digestate alters the raw materials inherent characteristics to produce a carbon (C)-rich hydrochar (HC), with an improved suitability for soil amelioration. Numerous studies report conflicting impacts of various HC application rates on soil properties and plant growth. In this study, the influence of HC application rate on soil improvement and plant growth aspects was investigated in three diverse soils (Chernozem, Podzol, and Gleysol). Pot trials were conducted in which all soils were amended with 5, 10, 20 and 30% (w/w) HC in quintuplicate, with two controls of pure soil (with and without plants, respectively) also included. Prior to potting, soil samples were collected from all HC-amended soils and controls and analyzed for soil pH, plant available nutrients ($\text{PO}_4\text{-P}$ and K), and microbial activity using standard laboratory and statistical methods. Immediately after potting, a 6-week seed germination experiment using Chinese cabbage was conducted to determine germination success, followed by a plant growth experiment of equal duration and plant species to determine biomass success. At the end of the study (after a total plant growth period of 12 weeks), each pot was sampled and comparatively analyzed for the same soil properties as at the beginning of the study. Soil pH shifted toward the pH of the HC (6.6) in all soils over the course of the study, but was most expressed in the 20% and 30% application rates, confirming the well-documented liming effect of HC. The addition of HC increased the $\text{PO}_4\text{-P}$ and K contents, particularly with 20% and 30% HC amendments. These results are proposedly due to the large labile C fraction of the HC, which is easily degradable by microorganisms. The rapid decomposition of this C fraction prompted the quick release of the HCs inherently high $\text{PO}_4\text{-P}$ and K content into the soil, and in turn, further stimulated microbial activity, until this fraction was essentially depleted. HC addition did not inhibit seed germination at any rate, presumably due to a lack of phytotoxic compounds in the HC from aging and microbial processes, and furthermore, showed no significant impact (positive or negative) on plant growth in any soil, despite improved soil conditions. In conclusion, although less pronounced, soil improvements were still achievable and maintainable at lower application rates (5% and 10%), whereas higher rates did not ensure greater benefits for plant growth. While the addition of high rates of HC did not detrimentally effect soil quality or plant growth, it could lead to leaching if the nutrient supply exceeds plant requirements and the soil's nutrient retention capacity. Therefore, this study validates the previous study in the effectiveness of the biogas digestate HC for soil amelioration and suggests that smaller regularly repeated HC applications may be recommendable for soil improvement.

Keywords Biogas digestate · Hydrochar application rate · Soil improvement · Nutrient availability · Microbial activity

1 Introduction

Biogas production in Germany peaked at the beginning of the twenty-first century due to the country's objective to expand renewable energy sources to reduce greenhouse gas

(GHG) emissions. Accordingly, a large percentage of energy crops grown in Germany are purposed for biogas production, occupying almost 1.4 million hectares of land in 2017 (FNR 2018). A by-product of the biogas production process is digestate, which is commonly applied as a fertilizer to soils to improve soil quality and fertility. However, digestate is readily degraded by microorganisms and therefore relatively unstable in soils. Consequently, its application to soils has shown to provide a surplus of nutrients, as well as release a significant amount of carbon dioxide (CO_2), which

✉ Megan de Jager
megan.de.jager@uni-oldenburg.de

¹ Carl Von Ossietzky Universität, Institut Für Biologie Und Umweltwissenschaften, Oldenburg, Germany

has negative implications for soil quality and climate mitigation (Mukherjee et al. 2016). These detrimental effects can be compensated by the further processing of digestate by pyrolysis to produce a carbon (C)-rich charcoal-like material referred to as biochar (BC). Pyrolysis results in the retention of a substantial proportion of C content of the source material (~ 50%), as well as a higher recalcitrance of that C to mineralization and microbial degradation (Lehmann et al. 2006; Steiner et al. 2009). In addition, the use of BC as a soil amendment has received considerable attention and extensive research has been undertaken to explore its potential as such. The addition of BC to soils has shown to increase nutrient availability, namely phosphorous (P), potassium (K) and nitrogen (N); enhance crop yields; and improve soil pH, particularly in acidic soils (Biederman and Harpole 2013; Glaser and Lehr 2019). Despite the benefits of BC, the pyrolysis process from which it is produced has a number of technical drawbacks including the release of ~ 50% of the feedstocks original C content back into the atmosphere, as well as the restricted use of dry feedstocks and the consequent higher-energy consumption if pre-drying is required (Lehmann et al. 2006; Libra et al. 2011).

As a means to overcome these technical challenges, a growing focus has been placed on potential alternatives to pyrolysis, such as hydrothermal carbonization (HTC). HTC also produces a C-rich material called hydrochar (HC), but under water-saturated conditions, as well as lower temperatures (180–250 °C) and higher pressure (≤ 20 bar), relative to pyrolysis (Libra et al. 2011). The reduced HTC operating temperatures equate to drastically reduced C emissions and its capability to process wet feedstocks within a single system means that HTC may provide a more environmentally and economically feasible alternative to pyrolysis. Despite disproportionate research regarding HC's effectiveness for soil amelioration compared to BC, a number of studies have shown HC to have similarly positive impacts on the soil properties and nutrient dynamics that are essential for enhanced plant growth (Bargmann et al. 2013b; de Jager et al. 2020; Fang et al. 2015). A number of studies also highlight the stimulating effect BC and HC have on microbial biomass and activity when applied to soils (Lehmann et al. 2011; Prayogo et al. 2014). However, these results are largely dependent on various factors including production parameters (e.g. temperature and residence time), feedstock and soil properties, experimental conditions (e.g. ambient temperature and open-air vs. sealed apparatus), as well as the application rate at which the material is added to the soil (Bargmann et al. 2014a; Dieguez-Alonso et al. 2018; Puccini et al. 2018; Reza et al. 2014; Zhang et al. 2017).

Existing research offers conflicting suggestions regarding the optimal application rate of BC and HC to soils to achieve the greatest benefits while avoiding any potential harmful impacts. This discord is largely fueled by the contradictory

results that have been found regarding the influence of increasing application rates of BC and HC on soil properties and plant growth. In a comprehensive meta-analysis by Jeffrey et al. (2017), it was suggested that the average application rate of 30 t ha⁻¹ of BC in temperate regions of the world resulted in significantly reduced crop yields. Comparable application rates of HC in a study by Bargmann et al. (2013b) of 2, 4 and 10% (equating to 30, 60 and 150 t ha⁻¹, respectively, at a depth of 10 cm) found that the germination of spring barley was unaffected at rates below 4%, while at 10%, germination success decreased compared to the control. Contrarily, the addition of an aged HC at 5% and 10% by Puccini et al. (2018) showed a significant reduction in seed germination inhibition compared to fresh (non-pre-treated) and washed HC. Furthermore, Prayogo et al. (2014) found an increase in soil pH with a 2% (w/w) BC addition, as well as changes to the microbial community structures at application rate as low as 0.5%. Increases in soil pH are also reported by Sun et al. (2020) following the addition of 0.5% and 1.5% HC of variable wheat origins. Although the application of less than 1% HC in field trials by Reza et al. (2014) did not significantly impact the soil properties, it did reduce the sugar beet crop yield, while an even smaller application rate of 0.8% HC in Melo et al. (2018) increased the dry biomass of *Phaseolus* beans over two harvests, and also improved soil nutrient content and soil fertility.

A related study by de Jager et al. (2020) found that the addition of the same HC used in this study at a 5% application rate (w/w) induced a short-term fertilization effect by significantly increasing the phosphate (PO₄-P), K and ammonium (NH₄⁺) concentrations in three different soils. Furthermore, a compensatory shift in pH of all soil types toward the pH of the added HC was observed. However, the addition of the 5% HC was insufficient to improve other soil properties essential to plant growth, such as water holding capacity (WHC) and aggregate stability, and was unable to sustain a long-term fertilization effect. From the above information, it is evident that the recommended optimum application rate of HC for soil amelioration must consider a number of factors, including the potential consequences for soil pH, nutrient provision and cycling, and microbial activation and stimulus, which ultimately effect plant growth. As such, the primary aim of this study is to investigate the influence of the application rate of HC produced from biogas digestate on soil improvement, germination success and biomass production. The HC was added to three distinct soil types: Chernozem, Podzol and Gleysol, at application rates of 5, 10, 20 and 30% (w/w). This aim will be achieved through the specific objectives, which include to (1) identify whether an increase in HC application rate will positively or negatively affect the pH, PO₄-P and K contents, and the microbial activity of the soils; (2) assess the impacts of the different HC application rates on the seed germination and biomass

production of Chinese cabbage; (3) compare the impacts of HC addition between the three soil types used in the study (Chernozem, Podzol and Gleysol); (4) determine which of the three soil types used in the study is most affected by HC addition; and (5) evaluate which HC application rate(s) is most suitable for the purpose of soil amelioration.

These objectives will test the hypotheses that: (a) increasing application rates will result in a sustainable increase in the nutrient content ($\text{PO}_4\text{-P}$ and K) and microbial activity; (b) changes in soil pH will be greater at higher application rates; and (c) seed germination inhibition and reduced plant growth will occur with increasing application rates. This study builds on the previous work by de Jager et al. (2020), and provides an innovative contribution to the limited existing body of research focused on the effects of the application rate of HC derived from biogas digestate for soil amelioration and improved plant growth.

2 Materials and methods

2.1 Experimental set-up

2.1.1 Soil sampling and properties

The experimental design is described in a previous work by de Jager et al. (2020), with modifications concerning the amount of HC added. Pot trials were conducted using three soil types: Chernozem, Podzol and Gleysol. The Chernozem was sampled in November 2019 from a site located within the Hohe Börde Municipality in Saxony-Anhalt, Germany ($52^\circ 10' 35'' \text{ N}$, $11^\circ 30' 42'' \text{ E}$). The Podzol and Gleysol were sampled in October 2019 at sites situated within the Bruchhausen-Vilsen Municipality ($52^\circ 48' 35'' \text{ N}$, $8^\circ 59' 41'' \text{ E}$) and the Ovelgönne Municipality in Lower Saxony, Germany ($53^\circ 15' 45'' \text{ N}$, $8^\circ 19' 49'' \text{ E}$), respectively. All soils were collected from working farms, with the Chernozem and Podzol sampling sites subjected to long-term arable use, and the Gleysol sampling site is currently a grassland area. The soils were collected haphazardly at 0–30 cm depth and air-dried in a non-climate-controlled greenhouse. The Chernozem was classified as a silty clay, the Podzol a sandy loam and the Gleysol a clay (de Jager et al. 2020). The Podzol was treated with lime prior to sampling.

2.1.2 Hydrochar preparation and characterization

The HC was provided by the Grenol Company (Meiersberg (Ratingen), Germany) and was produced from biogas digestate derived from plant- and animal-based feedstocks, including corn, sugar beet, beef manure and liquid pig manure. The feedstock and resultant HC were not pre- or post-treated for any specifications. HTC process parameters included $\sim 200^\circ \text{C}$ temperature, 18–20 bar pressure and ca. 3 h processing time, with heating and cooling rates of 1.5 h each. As determined in de Jager et al. (2020), which used the same HC as this study, the elemental composition of the HC, namely the C, hydrogen (H), N and sulfur (S), was determined directly using a Euro Elemental Analyzer (Eurovector; HEKAtech GmbH) after oven-drying the HC at 105°C overnight. The ash content of the HC was also measured directly after dry oxidation at 550°C , according to the DIN EN 14775:2010-04 standard. The CHNS and ash contents were then used to determine the oxygen (O) content of the HC, by calculating the difference between these values and 100%, as demonstrated in the following equation:

$$\text{O}[\%] = 100\% - \text{C}[\%] - \text{H}[\%] - \text{N}[\%] - \text{S}[\%] - \text{Ash content} [\%]. \quad (1)$$

The elemental composition (CHNS and O) and ash content of the HC were then used to calculate the molar element ratios H/C, C/N, and O/C, the results of which are presented in Table 1. The HC was stored in an unsealed cardboard box in a non-climate-controlled greenhouse for approx. 20 months prior to application to soils. The soils and HC were ground to $< 2 \text{ mm}$ particle size by milling, and mixed homogenously at application rates of 5, 10, 20 and 30% (w/w) HC-to-soil. The high application rates of 20% and 30% HC, which correspond to $\sim 300 \text{ t ha}^{-1}$ and 450 t ha^{-1} , respectively, are unrealistic for practical application and were used solely for experimental purposes (Bargmann et al. 2013b). Two sets of un-amended soils (no HC addition) were used as controls, where one set was devoid of seeds and plant growth (referred to as “control” hereinafter), and the other set was sown with Chinese cabbage seeds (denoted as “control_pl” hereafter). Five replicate pots of $\sim 13 \text{ cm}$ diameter (ca. 1 L [l] volume), were used for each HC-amended soil at each application rate, as well as for the controls.

Table 1 The ash content, elemental composition (CHNS and O), and resultant molar element ratios of the hydrochar derived from biogas digestate

Ash content (wt %)	Elemental composition (wt %)					Molar element ratio		
	C	H	N	S	O	H/C	C/N	O/C
47.2	35.2	3.8	2.7	0.9	10.2	1.3	15.2	0.2

2.2 Germination and plant growth experiments

The pot experiments were carried out in a greenhouse under climate-controlled conditions, with an average room temperature of $\sim 23^\circ\text{C}$ and supplementary lighting for approx. 8 h per day. Shortly after (i.e. on the same day as) potting and wetting the soils, each pot with HC-amended soil and one control set were sown with 25 Chinese cabbage seeds (*Brassica rapa* ssp. *Pekinensis*). The germination experiment consisted of two rounds of three weeks each, from 19 November to 10 December 2019 and from 13 December 2019 to 03 January 2020, respectively. The second round of the germination experiment followed the same procedure as round one, in which 25 seeds were sown after wetting the soils. Throughout the experiment the pots were watered manually according to plant requirements. At the end of each round, the number of germinated seeds in each pot was counted and germination success calculated, where 25 seeds equated to 100% germination. Furthermore, after each round, the above-ground biomass of each pot was harvested, and weighed before and after oven-drying at 105°C to constant weight to determine the total biomass for each HC treatment and the control. Following the germination experiment, each pot was sown once more with 4 Chinese cabbage seeds. The number of plants per pot was thinned to 1 after a growth period of approx. 4 weeks. After a further 2-week growth period (6 weeks of growth in total), the above-ground biomass in each pot was harvested, and weighed before and after oven-drying to constant weight at 105°C to determine the total biomass for each HC-amended soil (soil plus HC at varied application rates) and the control.

2.3 Soil analyses

The control samples for soil property analyses were collected prior to the addition of HC at the beginning of the study, and are hereinafter referred to as time 0 (t_0) samples. Samples were also collected immediately after the HC was added at the respective application rates and homogenously

mixed with the soils prior to potting at the beginning of the study. These samples are hereinafter referred to as time 1 (t_1) samples. After 12 weeks, at the end of the plant growth experiment (i.e. end of study), each of the five replicate pots was sampled destructively, and are hereinafter referred to as time 2 (t_2) samples. All samples for soil property analyses were transported to the laboratory where they were air-dried and sieved $< 2\text{ mm}$.

The pH, $\text{PO}_4\text{-P}$, K and microbial activity of the controls and HC-amended soils were analyzed according to the standard methods of soil analysis. The pH of the HC, the HC-amended soils and controls was determined in a 1:2.5 (w/v) soil/ water suspension using 10 g dry soil in 25 mL distilled water, which was stirred manually every 15 min for 1 h. An extraction method using 0.6% calcium (Ca)-acetate-lactate was used to measure plant available P and K (VDLUF 2012). The $\text{PO}_4\text{-P}$ concentration was determined colorimetrically with a Shimadzu UVmin-1240 Spectrophotometer, using a 0.5% ascorbic acid and 1% ammonium heptamolybdate coloring agent. Plant available K was measured by atomic absorption spectrometry (AAS).

Microbial activity was measured as microbial respiration using an adapted method similar to that described by Santos et al. (2019). Due to the (air) dry nature of the soils, distilled water was added to the soils to achieve approximate field capacity of the soils, to stimulate microbial activity. The water content of the samples was determined after wetting, according to VDLUF 2012). A glass flask with 20 mL of 0.2 molar sodium hydroxide (NaOH) absorption solution and two drops of phenolphthalein indicator solution was placed in the center of mason jars, with 100 g of each soil type placed around the flask in the respective jars. The jars were hermetically sealed and incubated for 24 h at room temperature ($\sim 25^\circ\text{C}$). To serve as controls, three jars without soil material were also sealed and incubated as previously described. After the incubation period, the NaOH solution was titrated with 0.2 molar hydrochloric acid (HCl) to a pH of 7.0. The amount of CO_2 released by microbial respiration was determined as the difference in the volume of HCl consumed between the treatments with soil and the controls, and was calculated using the equation below:

$$\text{mgCO}_2\text{kg}^{-1}\text{h}^{-1} = \frac{(\text{mmolHCl}[\text{control}] - \text{mmolHCl}[\text{sample}]) \times 44\text{mg mmol}}{2 \times 24\text{h} \times \text{dry weight}(\text{kg})}, \quad (2)$$

where the factor 44 mg mmol^{-1} represents the molar mass of CO_2 .

2.4 Statistical analyses

The critical assumption of a normal distribution for statistical analyses could not be determined with confidence due to the low number of replicates used in this study ($n=3$ for t_0 and t_1 , and $n=5$ for t_2). Consequently, non-parametric statistical analyses were performed on all residuals remaining after tests for normal distribution by the Shapiro–Wilk test, and for homogeneity of variance by Levene’s test (Laerd Statistics 2015). The Kruskal–Wallis H test was employed to determine significant differences ($p < 0.05$) between the HC application rate treatments and controls (0, 5, 10, 20, 30% HC) at each time phase of the study (t_0 , t_1 and t_2). The Kruskal–Wallis H test utilizes the pairwise comparison post-hoc test, which uses the procedure by Dunn (1964) with a Bonferroni correction for multiple comparisons (Laerd Statistics 2015).

The Mann–Whitney U test was performed using the exact statistical significance level (Dinneen and Blakesley 1973) to determine significant differences ($p < 0.05$) between the HC application rate treatments and controls over the entire course of the study (from t_0 to t_2). The results were reported as the significant median difference where the data distributions were similarly shaped, based on visual inspection. Statistically significant differences were reported as mean ranks where the data distributions were dissimilarly shaped (Laerd Statistics 2015). Statistical analyses were performed using SPSS 26.

3 Results and discussion

3.1 Hydrochar physico-chemical properties

The physico-chemical properties of the HC provide insight into the degree of carbonization that took place during the HTC process, and subsequently the degradability and associated release of nutrients from the HC in soils (Bento et al. 2019; Eibisch et al. 2013; Santin et al. 2017). The elemental compositions, namely the C (35.2%), H (3.8%), N (2.7%), and O (10.2%), of the HC used in this study were lower than those of a comparable HC (C: 44.1%; H: 5.1%; N: 3.2%; O: 31.3%) produced from a similar feedstock and under similar HTC conditions, namely 200°C for 4.5 h (Parmar and Ross 2019). However, the S content was higher in this study than in

Parmar and Ross (2019) (0.9% and 0.3%, respectively). These disparities may be related to slight differences in the feedstock materials between the respective studies, whereby the feedstock used to produce the HC in this study additionally contained sugar beet and liquid pig manure. These findings are supported by Möller and Müller (2012), who stated a generally lower C content (total and organic) for digestate (raw) materials. The high ash content (47.2%) of the HC used in this study is likely due to the feedstock materials, and is in line with the ash content (49.4%) of a digestate-derived biochar reported by Marmiroli et al. (2018). According to Verheijen et al. (2009), the thermochemical conversion of agricultural materials, such as grass, straw residues and manures, result in a charred product containing a large proportion of ash, with consequential effects on the pH and O/C ratio of the char product (Dieguez-Alonso et al. 2018).

The C/N ratio is widely suggested as an indicator for nutrient retention and nutrient plant availability (de Jesus Duarte et al. 2019). A C/N ratio of < 20 is proposedly indicative of N mineralization, while a ratio > 30 suggests N immobilization (Dieguez-Alonso et al. 2018). The C/N ratio of the HC used in this study (15.2; Table 1) suggests that the addition of this HC to soils should result in net N mineralization. In a previous study using the same HC as in this study, with the same C/N ratio, a high NH_4^+ content was reported shortly after the HC was added to the soil, thus substantiating this assumption (de Jager et al. 2020). Associated with this mineralization is the release of other nutrients as well, including $\text{PO}_4\text{-P}$ and K.

The H/C (1.3) and O/C (0.2) ratios of the HC used in this study are in line with those of an HC produced from an agricultural digestate (1.4 and 0.36, respectively) by Parmar and Ross (2019). The H/C and O/C ratios are considered reliable indicators for the degradability of BC and HC, alike (Eibisch et al. 2013; Steiner et al. 2016). A high H/C and O/C ratio suggests a relatively low degree of carbonization during the HTC process (Eibisch et al. 2013; Gai et al. 2014; Santin et al. 2017), resulting in an increased degradability and thus, increased nutrient supply. The proposed H/C and O/C ratios of ≤ 0.6 and ≤ 0.4 , respectively, are considered indicative of a stable BC/HC (Dieguez-Alonso et al. 2018; Eibisch et al. 2013). Although, based on these values, the HC used in this study may exhibit a proclivity for degradation and therefore be potentially less suitable for the purpose of C sequestration, this degree of degradability could be highly beneficial for nutrient content and supply, and consequently, for soil amelioration.

3.2 Effect of hydrochar application rate on soil properties

3.2.1 Soil pH

Soil pH is considered one of the most important soil chemical parameters due to its influence on, inter alia, nutrient availability, microbial activity and plant growth (Blume et al. 2016; Dieguez-Alonso et al. 2018). Therefore, the effect of HC addition on soil pH is an important consideration in sustainable soil management practices. The pH buffering capacity and liming effect of BC and HC in soils are well documented (Bargmann et al. 2014a; Liao and Thomas

2019; Fang et al. 2015), however, little is known about the impact of their application rates on soil pH.

At the beginning of this study (t_0), the average pH of the Chernozem control (Chernozem_{con}) was slightly alkaline (7.9 ± 0.1), while the Podzol_{con} and Gleysol_{con} were weakly (6.4 ± 0.1) and moderately acidic (5.1 ± 0.1), respectively (Fig. 1). The addition of the weakly acidic HC (6.6 ± 0.0) at t_1 , decreased the pH of the Chernozem at each application rate, with the greatest decrease of 0.7 units occurring in the 30% HC-amended Chernozem (Chernozem₃₀) compared to the Chernozem_{con} at t_0 ($p=0.015$). The pH of the HC-amended Podzol at t_1 remained relatively unchanged compared to the Podzol_{con}, increasing slightly at the higher

Fig. 1 Average pH (H₂O) for the controls and hydrochar-amended Chernozem, Podzol and Gleysol over the course of the study. The solid line represents the pH of the HC (6.6). Error bars represent standard deviation of the means. Different letters indicate significant differences in means at $p < 0.05$ level between treatments at the respective time periods (lowercase letters = t_0 and t_1 , capital letters = t_2). n.s = nonsignificant. Patterned bars indicate significant differences in means ($p < 0.05$) between treatments at the beginning and end of the study. Solid (non-patterned) bars = nonsignificant. Control_pl = control with plants

application rates (20% and 30% HC) to the same pH as that of the HC (6.6). The pH of the HC-amended Gleysol increased at each application rate, with the largest increase of 0.9 units occurring in the Gleysol₃₀ compared to the Gleysol_{con} ($p=0.015$). At the end of the study (t_2), the pH values remained relatively unchanged from t_1 in all soils. Therefore, the results evince a rapid influence by HC addition in shifting soil pH, which is stronger with higher HC application rates. Furthermore, the shift is dependent on the initial pH of the soil, and the resultant pH values were maintained over the course of the study.

The results of this study reiterate those of de Jager et al. (2020), in which the addition of a digestate-derived HC shifted the soil pH toward the pH of the HC, and further testify the fact that the soil's response to HC addition is dependent on the initial soil and HC pH (Biederman and Harpole 2013). DeLuca et al. (2009) attribute the liming effect of BC to an enhanced concentration of major cations (namely K^+ , calcium (Ca^{2+}), and magnesium (Mg^{2+})) and a lower aluminum (Al^{3+}) and H^+ contents in the BC itself. Upon the addition of an alkaline BC to soil, the contribution of these cations and their subsequent exchange with Al^{3+} and H^+ on the negative charge sites in the soil result in an increase in soil pH (Hailegnaw et al. 2019). This liming effect can be inferred for HC as well, since numerous studies have found HC from various feedstocks to contain appreciable quantities of these (and other) macronutrients, depending on the HTC process conditions (Dieguez-Alonso et al. 2018; Ghanim et al. 2018; Puccini et al. 2018; Reza et al. 2013).

Contrarily to de Jager et al. (2020), where significant pH shifts occurred only in the finer grained 5% HC treatments toward the end of the study in the Podzol and Gleysol, this study evinced significant shifts in pH in all soils already immediately after HC addition. Furthermore, in this study, which used a grain size equivalent to the medium HC treatments used in de Jager et al. (2020), significant differences between the HC-amended soils and the controls only occurred with the higher HC application rates (20% and 30%). This finding, together with that of Bargmann et al. (2013a) who reported a significant increase in soil pH by the addition of a relatively fine grained HC (1 mm) at comparatively low application rates of 2% and 4%, suggests that HC grain size may be a more important factor than application rate for soil pH adjustment. However, regardless of grain size, the liming effect induced by the added HC in this study is clear. As such, the biogas digestate-derived HC used in this study would be suitable for the adjustment of soil pH; however, a sustainable influence may require repeated applications. Furthermore, cognizance regarding the required pH result and the initial pH of the soil and HC is essential to avoid limitations to plant growth that may arise from unwanted consequences of HC application.

3.2.2 Phosphate (PO_4 -P)

A number of studies have highlighted the importance of HC application rate for nutrient supply, namely PO_4 -P and K (Bargmann et al. 2014a; Bento et al. 2019). At an application rate of 5% HC, de Jager et al. (2020) found a significant increase in PO_4 -P content using the same HC as that in this study, which was suggested to be due to the relatively easily decomposable nature of HC compared to BC (Gronwald et al. 2015; Verheijen et al. 2009). However, the fertilizer effect was short-lived, and therefore, it is hypothesized that a higher application rate would result in a longer-term supply of essential nutrients for plant growth.

At the beginning of this study (t_0), the results showed an initial average PO_4 -P content of $199.9 \pm 27.3 \text{ mg kg}^{-1}$ for the Chernozem_{con}, $193.9 \pm 4.2 \text{ mg kg}^{-1}$ for the Podzol_{con}, and $29.1 \pm 1.2 \text{ mg kg}^{-1}$ for the Gleysol_{con} (Fig. 2). The applied HC had an average PO_4 -P content of $6544.3 \pm 192.0 \text{ mg kg}^{-1}$. After HC addition at t_1 , the PO_4 -P content increased exponentially with increasing application rate in all soils, compared to the control at t_0 . The largest and statistically significant increases occurred in the Podzol₃₀ and Gleysol₃₀, increasing by 378% ($p=0.015$) and 2135% ($p=0.015$), respectively, compared to the controls. At the end of the study (t_2), the PO_4 -P content continued to increase exponentially with increasing HC application rates in all soils, compared to the controls at t_2 . The highest HC application rates (20% and 30%) showed the highest PO_4 -P content in all soils, which were statistically significantly different to the controls at t_2 , with the exception of the Chernozem₂₀. The results indicated that the HC served as a direct source of PO_4 -P, which was released rapidly upon application to soils, and which was maintained over a period of months, as evidenced by the significant increase in the PO_4 -P content toward the end of the study (from t_0 to t_2 ; $p<0.05$).

The inherently high PO_4 -P content of the HC ($6544.3 \pm 192.0 \text{ mg kg}^{-1}$) due to its biogas digestate feedstock source (Gronwald et al. 2015) and its relatively low resistance to microbial degradation compared to BC (Bai et al. 2013; Bargmann et al. 2014b) are the likely factors responsible for the exponential increase in PO_4 -P content in the HC-amended soils. Similar findings were reported by Melo et al. (2018), using an HC produced from sewage sludge, whereby the highest application rate of 1.6% (w/w) showed the highest P content (25 mg/dm^3) compared to the lower rates. Of the HC-amended soils, the smallest increases occurred at the lower HC application rates (e.g. 95% increase in the Chernozem₅), and the largest increases at the highest application rates (e.g. 2135% in the Gleysol₃₀) at t_1 (see Fig. 2). The lower stability of the HC is suggested to be due to the relatively large labile C fraction of the HC (Bargmann et al. 2013b; Malghani et al. 2015), and is further indicated by the molar O/C ratio of the HC (0.2, Table 1),

Fig. 2 Average phosphate content ($\text{PO}_4\text{-P}$) for the controls and hydrochar-amended Chernozem, Podzol and Gleysol over the course of the study. Error bars represent standard deviation of the means. Different letters indicate significant differences in means at $p < 0.05$ level between treatments at the respective time periods (lowercase letters = t_0 and t_1 , capital letters = t_2). n.s = nonsignificant. Patterned bars indicate significant differences in means ($p < 0.05$) between treatments at the beginning and end of the study. Solid (non-patterned) bars = nonsignificant. Control_pl = control with plants. *Statistically significantly different (not visible)

which characterizes a higher degradability (Bamminger et al. 2014; Bargmann et al. 2013a; 2014b). The results clearly indicate that the higher application rates of HC in the soil translate to more material being available for microbial degradation and the consequent increased release of nutrients.

These results also correlate with the change in soil pH induced by the addition of HC. The increase in soil pH via HC addition may serve to increase the solubility of Al and iron (Fe) phosphates, thereby liberating the affixed P through their dissolution in the acidic Podzol and Gleysol, while a decrease in pH in the alkaline Chernozem increases the solubility of Ca phosphates, which also releases the associated P into the soil solution (Anderegg and Naylor 1988; Glaser and

Lehr 2019; Huang et al. 2005; Penn and Camberato 2019). This is further supported by Huang et al. (2005), who stated that at pH values between 5.5 and 6.5, Ca and Al phosphates could share an equal control of P solubility. These reaction processes may thus have contributed to the increased $\text{PO}_4\text{-P}$ content in all the soils, but likely at a smaller magnitude compared to the release of $\text{PO}_4\text{-P}$ directly from the HC.

At the end of the study (at t_2), the same trend was observed in all soils as described above, whereby the HC-amended soils had a higher $\text{PO}_4\text{-P}$ content than the controls, which was only statistically significant for the higher application rates (20% and 30%, except Chernozem₂₀). This high $\text{PO}_4\text{-P}$ content (at t_2) contradicts the corresponding

results of the previous study by de Jager et al. (2020), in which the $\text{PO}_4\text{-P}$ content decreased over the course of the study. The significantly higher $\text{PO}_4\text{-P}$ content in the HC-amended soils at t_2 (compared to the controls at t_2 , as well as the corresponding results of the previous study), may be a direct result of the high $\text{PO}_4\text{-P}$ content of the aged HC ($6544.3 \pm 192.0 \text{ mg kg}^{-1}$) compared to the HC of the previous study ($2034.6 \pm 38.3 \text{ mg kg}^{-1}$). It is plausible to assume that during the plant growth phase of this study, the plants were able to absorb the required amount of $\text{PO}_4\text{-P}$ essential for their growth; however, once this requirement was met, the unutilized $\text{PO}_4\text{-P}$ remained in the soil solution.

In general, the thermochemical conversion of biomass during pyrolysis and/or HTC results in a less mobile, more stable P form, which when introduced into the soil is relatively resistant to leaching and fixation by clay minerals, compared to P from raw agricultural residues for example (Dai et al. 2016). Related to this, the nature of the HC is an important consideration, and the results of this study indicate that $\text{PO}_4\text{-P}$ contributed by the biogas digestate HC has a significant fertilizer effect, and furthermore, is able to sustain an adequate supply of $\text{PO}_4\text{-P}$ to plants. However, the $\text{PO}_4\text{-P}$ sorption capacity of soils must also be taken into account when applying fertilizers, or HC in this case, as well as other factors, such as the soil physical properties and hydrological condition, which influence the fate of the excess $\text{PO}_4\text{-P}$ in the soil. The high input of $\text{PO}_4\text{-P}$ from such amendments could exceed the limited sorption capacity of soils for $\text{PO}_4\text{-P}$, particularly in sandy soils, thus resulting in leaching as an unintended consequence (Alterra 2011). Additionally, an excessive concentration of nutrients resulting from high HC application rates has also shown to reduce the biomass production of some plant crops, such as beans (Melo et al. 2019). Therefore, to reduce the potential for $\text{PO}_4\text{-P}$ loss by leaching and toxic soil conditions, a systematic application of HC in smaller doses is recommended, rather than a single large dose application (Lehmann and Schroth 2003).

3.2.3 Potassium (K)

Potassium (K) is an essential nutrient for plant growth, and is therefore critical for crop yield and enhanced crop quality (Wang et al. 2018). Numerous studies have shown BC and HC to directly contribute nutrients, including K, upon application to soils (Biederman and Harpole 2013; de Jager et al. 2020). However, the latter study revealed a short-lived fertilizer effect, and therefore, it is hypothesized that a higher application rate would result in a longer-term nutrient supply.

The initial K contents, for the controls at t_0 were $439.6 \pm 55.7 \text{ mg kg}^{-1}$ for the Chernozem, $179.2 \pm 4.1 \text{ mg kg}^{-1}$ for the Podzol, and $119.7 \pm 6.8 \text{ mg kg}^{-1}$

for the Gleysol (Fig. 3). The HC had a K content of $2384 \pm 302.5 \text{ mg kg}^{-1}$. The addition of HC at t_1 resulted in increasing K content with increasing application rates, with the 30% HC addition showing a statistically significant difference ($p < 0.05$) to the controls in all soils. At the end of the study (t_2), the 20% and 30% HC treatments had a significantly higher K content than the control with plants in all soils, whereas the lower HC application rates hardly differed from the controls. Over the course of the study, the K content of the Chernozem increased, while for the HC-amended Podzol and Gleysol, the K content decreased from t_0 to t_2 . Nonetheless, it is evident that the highest application rate of 30% HC provided and retained the most K in all soils over the entire course of the study.

The results of this study are consistent with those of the previous study by de Jager et al. (2020), and are further corroborated by Alling et al. (2014) and Hailegnaw et al. (2019). The initial increase in K content shortly after HC addition at the beginning of this study is likely contributed directly from the K-rich HC ($2384 \pm 302.5 \text{ mg kg}^{-1}$ K content). The consecutive increase in K content with increasing HC application rate (Fig. 3) is supported by Hailegnaw et al. (2019), who also reported a greater release of exchangeable K from the highest BC application rate used (8% w/w) already after one week. The rapid K release from the HC observed in this study is related to the more degradable structure of the HC (Bargmann et al. 2013a). Hailegnaw et al. (2019) also found a negative correlation between the percentage increase in K content after BC addition and the original soil K content, whereby soils with a lower original K content showed a higher K increase after BC addition. These results confirm this correlation in the case of HC, whereby the Podzol and Gleysol with lower initial K contents compared to the Chernozem, showed a greater increase in K content upon HC addition at all application rates (average increase of all application rates: 46%, 117%, and 219% for Chernozem, Podzol and Gleysol, respectively).

The sustained K contents at the higher application rates (20% and 30%) in all soils at t_2 suggest that the K input from the HC was more than the amount required by plants and microorganisms, thus resulting in this surplus to remain in the soil solution for an extended period of time. This is further verified by the smaller decrease that occurred over the course of the study in the 30% HC-amended Podzol and Gleysol (5% and 23%, respectively), compared to the lower application rates. Furthermore, the largest depletion of K from t_1 to t_2 occurred in the Gleysol, which corresponded to the highest average dry plant weights of all the soils, as shown in Fig. 4. Leaching of K from the sandy textured Podzol may also have contributed (in addition to plant uptake) to the reduced K content at t_2 (Biederman and Harpole 2013). The increased K content in the Chernozem from t_1 to t_2 may be a consequence of the high native K content of the soil,

Fig. 3 Average potassium (K) content for the controls and hydrochar-amended Chernozem, Podzol and Gleysol over the course of the study. Error bars represent standard deviation of the means. Different letters indicate significant differences in means at $p < 0.05$ level between treatments at the respective time periods (lowercase letters = t_0 and t_1 , capital letters = t_2). n.s = nonsignificant. Patterned bars indicate significant differences in means ($p < 0.05$) between treatments at the beginning and end of the study. Solid (non-patterned) bars = nonsignificant. Control_pl = control with plants

providing an already adequate supply of K for plant uptake and microbes. Regardless of the decreased K content over the course of the study in the Podzol and Gleysol, the higher application rate (30% HC) was still able to maintain a K supply well above 400 mg kg⁻¹ K after a twelve-week period. These findings, although varied depending on soil type, verify the fertilizer potential of the biogas digestate HC.

3.2.4 Microbial activity

Microbial activity has long been considered an essential process in the nutrient- and C- cycle in soils (Higashida and

Takao 1986; Lehmann et al. 2011). The microbial abundance and composition in soils effectively aid in regulating the soil organic carbon (SOC) pool through its decomposition of organic C inputs (Sun et al. 2020). The C input from BC and HC addition to soils has direct and indirect effects on soil microorganisms by its direct C contribution, its retention capacity for dissolved organic matter (DOM) and its influence on soil properties, such as pH, WHC and cation exchange capacity (CEC) (Biederman and Harpole 2013; Budai et al. 2016; Salem 2013). However, these effects are dependent on the characteristics of the HC, which are determined by the HC feedstock material, as well as the

Fig. 4 Average microbial activity presented as the weight of respired $\text{CO}_2\text{-C}$ (mg) per kg soil per hour for the controls and hydrochar-amended Chernozem, Podzol and Gleysol over the course of the study. Error bars represent standard deviation of the means. Different letters indicate significant differences in means at $p < 0.05$ level between treatments at the respective time periods (lowercase letters = t_0 and t_1 , capital letters = t_2). n.s = nonsignificant. Patterned bars indicate significant differences in means ($p < 0.05$) between treatments at the beginning and end of the study. Solid (non-patterned) bars = nonsignificant. Control_pl = control with plants

production conditions under which it is produced (Budai et al. 2016).

The controls at t_0 had an initial microbial respiration rate of 3.7 ± 0.4 mg $\text{CO}_2\text{-C}$ kg^{-1} h^{-1} for the Chernozem, 5.7 ± 0.2 mg $\text{CO}_2\text{-C}$ kg^{-1} h^{-1} for the Podzol, and 16.3 ± 0.7 mg $\text{CO}_2\text{-C}$ kg^{-1} h^{-1} for the Gleysol (Fig. 5). The addition of HC at t_1 exponentially increased the microbial respiration rate of all soils with increasing application rate. In the Chernozem and Podzol, the 30% HC treatment was significantly higher than the control ($p = 0.015$, respectively), while the Gleysol₂₀ was significantly higher than the control ($p = 0.038$). At the end of the study (t_2), the HC-amended soils had a higher microbial respiration rate than

the controls, most significantly at the higher HC application rates (20% and 30%). Over the course of the study (t_0 to t_2), the rate of microbial respiration decreased significantly in all soils for the controls as well as the HC-amended soils. The most significant decrease occurred in the Gleysol HC-amended soils, which decreased on average by ~75% from t_1 to t_2 . The results indicated a short-term stimulus of microbial activity by HC addition, however, regardless of application rate, the stimulus could not be sustained over a longer term.

According to a USDA classification for soil respiration activity (based on Doran and Brinton 2001), and following a unit conversion (from mg $\text{CO}_2\text{-C}$ kg^{-1} week^{-1} to mg $\text{CO}_2\text{-C}$ kg^{-1} h^{-1}), the Chernozem and Podzol controls at t_0 exhibited

a medium respiration rate ($2.98\text{--}5.95 \text{ mg CO}_2\text{-C kg}^{-1} \text{ h}^{-1}$), while the Gleysol_{con} was unusually high ($> 11.9 \text{ mg CO}_2\text{-C kg}^{-1} \text{ h}^{-1}$). The microbial respiration rate of the Gleysol_{con} was presumably the result of the greenhouse conditions, which improved the water state of the soil and thereby provided a more optimal environment for microbes. Upon the addition of HC at t_1 , the respiration rate improved in all soils, by an average of 142%, 108% and 38% in the Chernozem, Podzol and Gleysol, respectively. An increase in microbial activity following BC and HC addition to soils has been reported in numerous studies, which attribute this development to the degradation of the C components of these char materials (Bargmann et al. 2013a; Bamminger et al. 2014; Bento et al. 2019; Hardy et al. 2019; Mukherjee et al. 2016). Relative to BC, the high labile C content of HC makes it less stable in soils, thereby providing a more easily accessible and decomposable C source to soil microorganisms (Bargmann et al. 2013a). The level of degradability of the biogas digestate HC is also indicated by the H/C and O/C ratio, as discussed in Sect. 3.1.

Furthermore, it is postulated that the increased respiration may be caused by the increased soil pH induced by BC amendment (Hardy et al. 2019), as well as the decreased level of toxicity caused by Al (Alling et al. 2014). Considering that the shift in soil pH was most prevalent at the beginning of this study, it could be argued that a pH influence on microbial activity was also most prevalent at this stage. This is confirmed by the increased microbial respiration rate at t_1 (Fig. 5), particularly at the higher application rates, where the shift in pH was also most pronounced (Fig. 1). Concurrent to the shift in soil pH, the release of essential nutrients, namely $\text{PO}_4\text{-P}$ and K (as previously described), from the HC into the soil would create conditions under which microorganisms could thrive (Blume et al. 2016), which is effectively illustrated at the beginning of this study.

At the end of the experiment, the HC-amended soils continued to show a higher microbial respiration rate compared to the controls (with and without plants). However, over the course of the study and following the introduction of plants, the microbial activity decreased from t_0 to t_2 in the controls and all HC-amended soils. Similar findings are reported by Bargmann et al. (2013a) and Mukherjee et al. (2016), whereby the perceived increase in microbial activity following HC addition is only amenable for as long as the less resistant C fraction of the HC is available to microorganisms, which was found to be 14 days and 60 days, respectively. Once this C fraction is depleted, the microorganisms are not able to access and decompose the more resistant C forms, such as the recalcitrant aromatic C rings (Mukherjee et al. 2016).

The introduction of plants during the study, and thus the induced competition for nutrients, may also have contributed to the decreased microbial activity between t_1 and t_2 .

Evidence for this can be seen in the lower respiration rates of the controls with plants compared to the controls without plants in all soils at t_2 , thus suggesting nutrient competition from plants as an influencing factor. The results clearly demonstrate that the addition of the biogas digestate HC rapidly stimulated microbial activity even at relatively low application rates. Although the increase in microbial respiration following the application of HC in high quantities (20% and 30%) was only temporarily observed, the mediation of the respiration rate over the course of the study indicates an improved biological status of the soils, which could further translate to improved nutrient cycling and plant growth. Furthermore, the microbial respiration results of this study provide some insight into the degradability of the biogas digestate HC, which is important for the use of this material for soil amelioration. The relatively short-lived increase in microbial respiration hints to the potential of a longer-term more stable C fraction, which may serve a secondary purpose for C sequestration.

3.3 Effect of hydrochar application rate on the plant growth of Chinese cabbage

3.3.1 Seed germination

The average germination success of the controls over the two rounds of the germination experiment was $86.8 \pm 15.4\%$ for the Chernozem, $54.0 \pm 48.6\%$ for the Podzol, and $91.2 \pm 9.9\%$ for the Gleysol (Fig. 6). The response to the addition of HC was varied across the soil types, however, no significant differences in germination success were observed between the controls and HC-amended soils or between HC application rate treatments in all soils. It is therefore evident that, contrary to the original hypothesis, the higher application rates of HC were not detrimental to the germination of Chinese cabbage seeds.

The evident lack of significant differences between the germination rate of the controls and HC-amended soils in all soil types indicates that HC was not a driving factor in either the success or inhibition of the germination of Chinese cabbage seeds. The germination success observed in all the soils may be largely attributed to the energy reserves within the seed coatings and their mobilization following germination (Alencar et al. 2012; Pritchard et al. 2002). As such, soil parameters and organic amendments to soils are less important for seed germination and have more influence once the seedlings emerge from the seed and become photoautotrophic. However, the absence of seed germination inhibition following HC application to soils contradicts numerous studies, which attribute this hindrance to the phytotoxic nature of the HC, due to its content of, inter alia, volatile fatty acids, phenols and tannins (Bargmann et al. 2013b; 2014a; Puccini et al. 2018; Röhrdanz et al. 2019).

Fig. 5 Average percentage seed germination for the control with plants and hydrochar-amended Chernozem, Podzol and Gleysol over the course of the germination experiment. Error bars represent standard deviation of the means. *n.s* nonsignificant ($p \geq 0.05$)

Additionally, a negative correlation between the HC's dissolved organic carbon (DOC) content and germination success was found in Bargmann et al. (2013b), whereby the addition of DOC-rich HC severely limited seed germination at an application rate of 10%. However, in the same study, the inhibiting effect of the HC was diminished two weeks after soil incorporation, which the authors ascribe to the open-air conditions of the experiment, thus allowing for free gas exchange (release) of the potentially harmful substances into the atmosphere.

Another popular view on the reduced inhibition of seed germination is the microbial decomposition of the phytotoxic compounds inherent in the HC following its incorporation into the soil (Bargmann et al. 2013b; 2014b; Puccini et al. 2018). Interestingly, the process of aging has also shown to reduce the content of phytotoxic substances in the HC, as well as promote their microbial decomposition by stimulating microbial activity in the soil (Puccini et al. 2018). Since the HC used in this study was stored in open-air conditions for approx. 20 months prior to soil incorporation, it is suggested that gaseous exchange likely took place at the soil–air interface, resulting in the release of the potentially harmful organic compounds from the HC. Furthermore, the relatively high microbial activity recorded in all soils at the beginning of this study (see Sect. 3.2.4) may have further served to reduce the concentration of phytotoxic substances in the HC, thus providing favorable conditions for successful seed germination. Therefore, regardless of application rate, the addition of the biogas digestate HC did not produce any detrimental effects on the germination of Chinese cabbage seeds.

3.3.2 Biomass production

No statistically significant differences in biomass production were observed between the controls and HC-amended soils or between HC application rate treatments in any soils. Despite an evident variability in biomass production across the soil types, the results indicated that the addition of HC, even at high application rates, did not positively or negatively affect plant growth.

These findings are in line with those of the previous study (de Jager et al. 2020), which found no significant influence on the growth of Chinese cabbage using 5% HC. Few other studies have also reported a similar absence of seed germination- and plant growth inhibition, as well as contrasting effects on yield using various application rates (Bargmann et al. 2013b; 2014b; Puccini et al. 2018; Röhrdanz et al. 2019). Such findings have been attributed to a variety of mechanisms, including the removal of the potentially harmful substances inherent in HC via the HTC production parameters; by storage and/or aging the HC after production; by means of gaseous release prompted by open-air experimental conditions; as well as the decomposition of phytotoxic compounds by microorganisms (Bargmann et al. 2013b; 2014b; Puccini et al. 2018). These results must, however, also consider plant species as an influential factor, and cognizance must be given to the fact that the same application rate could have very different effects on different plant crops (Bargmann et al. 2014a). For example, Melo et al. (2019) found the highest yields for beans and rice were achieved by the addition of the same (sewage

Fig. 6 Average biomass production expressed in mean dry weight (g) of Chinese cabbage for the control with plants and hydrochar-amended Chernozem, Podzol and Gleysol at the end of the study. Error bars represent standard deviation of the means. *n.s* nonsignificant ($p \geq 0.05$)

sludge-derived) HC at different application rates, namely 0.5% and 3%, respectively. Although the results of this study do not satisfy the hypothesis that higher application rates would reduce plant growth, the addition of HC (at all concentrations) did not induce any positive effects either, despite the provision of conditions that typically promote plant growth (e.g. optimal pH for nutrient absorption, supply of plant available nutrients and the stimulation of microbial activity).

4 Conclusion

The findings of this study support the use of HC produced by the HTC of biogas digestate as an effective alternative to chemical fertilizers for the purpose of soil amelioration. The results of this study showed that the addition of HC at application rates of 5, 10, 20 and 30% improved the soil pH, supplied nutrients ($\text{PO}_4\text{-P}$ and K) and stimulated microbial activity in three soils of variable character. The rapid reaction processes and the associated quick release of nutrients following HC incorporation into the soils are likely due to the large fraction of the more easily degradable, labile C in the HC. Despite the fast-acting influence of the HC on the three diverse soils evidenced at the beginning of the study, the resultant positive effects proved persistent over its three-month duration. The rather neutral (neither positive nor negative) effect of the HC on the seed germination and plant growth of Chinese cabbage at all application rates, indicates an absence of phytotoxic compounds in the HC due to various aging and microbial processes. This study indicates that the resultant impacts of

HC addition in any soil type, regardless of application rate, are strongly influenced by the initial characteristics of the soil and the HC. As such, the most significant improvements were observed in the Gleysol soil, of which the controls had the lowest pH and $\text{PO}_4\text{-P}$ - and K content at the beginning of the study. Although the most significant increases in pH, nutrient contents and microbial activity occurred with the higher HC application rates of 20% and 30%, these results must take into consideration the increased potential for nutrient leaching or, on the other hand, increased toxicity where the nutrient supply exceeds that required by plants and microorganisms. Furthermore, although less pronounced, HC addition at rates of 5% and 10% still achieved and sustained improvements to pH, nutrient contents, microbial activity and the plant growth aspects, compared to the controls.

Taking into account the trade-off between economic feasibility and the optimum application rate for soil improvement, we recommend that HC be regularly and repeatedly incorporated into soils at smaller application rates of 5%, rather than by larger single applications. However, the importance of conducting preliminary investigations into the inherent characteristics of the HC and soils to assess the potential longevity of the desired outcomes of soil amendment is highlighted herein. Accordingly, future research should include investigating the influence of HC addition at variable application rates on other nutrients, such as mineral nitrogen (N_{min}), Ca, Mg, Al and H, particularly due to their impact on soil pH and the availability of $\text{PO}_4\text{-P}$ and K. Long-term field studies using a variety of plant crops of different nutritional requirements should also be the focus of future research, to validate the results

of controlled pot- and greenhouse experiments, such as the one conducted in this study.

Acknowledgements We would like to express our gratitude to the European Union Interreg North Sea Region Project for their sponsorship. Secondly, a sincere thank you to the students of the Bioenergy Practical and Ingeborg Eden for the analytical assistance provided, and to Thomas Pollmann for his continued support, guidance and encouragement.

Author contributions All authors had a role in the conceptualization and design of the study. Data collection and analyses were performed by MJ. All versions of the manuscript were written by MJ. Sample collection and the revision and editing of all manuscript versions were performed by Prof. Dr. LG. All authors have read and approved the final manuscript for submission.

Funding Open Access funding enabled and organized by Projekt DEAL. This study was conducted under the auspices of the ‘BIO-CAS, circular BIOMass CASCade to 100%’ project, which is funded by the European Union Interreg North Sea Region Project 38-2-4-17. The funders played no role in the design of the study, the collection and analyses of data or the preparation of the manuscript.

Compliance with ethical standards

Conflicts of interest The authors have no conflicts of interest to disclose, financial or otherwise.

Availability of data and material The data analyzed during this study which support its findings are available on request from the corresponding author.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

References

Alencar NLM, Innecco R, Gomes-filho E, Gallão MI, Alvarez-Pizarro JC, Prisco JT, De Oliveira AB (2012) Seed reserve composition and mobilization during germination and early seedling establishment of *Cereus jamacaru* D.C. ssp. *jamacaru* (Cactaceae). *An Acad Bras Cienc* 84(3):823–832

Alling V, Hale SE, Martinsen V, Mulder J, Smebye A, Breedveld GD, Cornelissen G (2014) The role of biochar in retaining nutrients in amended tropical soils. *J Plant Nutr Soil Sci* 177:671–680

Alterra (2011) Recommendations for establishing Action Programmes under Directive 91/676/EEC concerning the protection of waters against pollution caused by nitrates from agricultural sources: Part C: Analysis of the processes in soil that influence nutrient leaching and runoff. Final Report. Contract Number: 07

0307/2010/580551/ETU/B1. Alterra, Wageningen University and Research Centre, Wageningen. pp. 94. Available online: http://publications.europa.eu/resource/ellar/4ec63804-0cc9-4133-ad73-31b65ef584f3.0001.01/DOC_1. Accessed 28 Sept 2020

Anderegg JC, Naylor DV (1988) Phosphorus and pH relationships in an andic soil with surface and incorporated organic amendments. *Plant Soil* 107:273–278

Bai M, Wilske B, Buegger F, Esperschütz J, Kammann CI, Eckhardt C, Koestler M, Kraft P, Bach M, Frede H-G, Breuer L (2013) Degradation kinetics of biochar from pyrolysis and hydrothermal carbonization in temperate soils. *Plant Soil* 372:375–387

Bamminger C, Marschner B, Jüschke E (2014) An incubation study on the stability and biological effects of pyrogenic and hydrothermal biochar in two soils. *Eur J Soil Sci* 65:72–82

Bargmann I, Martens R, Rillig MC, Kruse A, Kücke M (2013a) Hydrochar amendment promotes microbial immobilization of mineral nitrogen. *J Plant Nutr Soil Sci* 177:59–67

Bargmann I, Rillig MC, Buss W, Kruse A, Kücke M (2013b) Hydrochar and biochar effects on germination of spring barley. *J Agron Crop Sci* 199:360–373

Bargmann I, Rillig MC, Kruse A, Greef J-M, Kücke M (2014a) Effects of hydrochar application on the dynamics of soluble nitrogen in soils and on plant availability. *J Plant Nutr Soil Sci* 177:48–58

Bargmann I, Rillig MC, Kruse A, Greef J-M, Kücke M (2014b) Initial and subsequent effects of hydrochar amendment on germination and nitrogen uptake of spring barley. *J Plant Nutr Soil Sci* 177:68–74

Bento LR, Castro AJR, Moreira AB, Ferreira OP, Bisinoti MC, Melo CA (2019) Release of nutrients and organic carbon in different soil types from hydrochar obtained using sugarcane bagasse and vinasse. *Geoderma* 334:24–32

Biederman LA, Harpole SW (2013) Biochar and its effects on plant productivity and nutrient cycling: a meta-analysis. *GCB Bioenergy* 5:202–214

Blume H-P, Brümmer GW, Fleige H, Horn R, Kandeler E, Kögel-Knabner I, Kretzschmar R, Stahr K, Wilke B-M (2016) Scheffer/Schachtschabel Soil Science. Springer-Verlag, Berlin Heidelberg, pp 123–353

Budai A, Rasse DP, Lagomarsino A, Lerch TZ, Paruch L (2016) Biochar persistence, priming and microbial responses to pyrolysis temperature series. *Biol Fertil Soils* 52:749–761

Dai L, Li H, Tan F, Zhu N, He M, Hu G (2016) Biochar: a potential route for recycling of phosphorus in agricultural residues. *GCB Bioenergy* 8:852–858

de Jager M, Röhrdanz M, Giani L (2020) The influence of hydrochar from biogas digestate on soil improvement and plant growth aspects. *Biochar* 2:177–194

de Jesus Duarte S, Glaser B, Cerri CEP (2019) Effect of biochar particle size on physical, hydrological and chemical properties of loamy and sandy tropical soils. *Agronomy* 9:165

DeLuca TH, MacKenzie MD, Gundale MJ (2009) Biochar effects on soil nutrient transformations. In: Lehmann J, Joseph S (eds) *Biochar for environmental management: science and technology*. Earthscan, London, pp 251–270

Dieguez-Alonso A, Funke A, Anca-Couce A, Rombolà AG, Ojeda G, Bachmann J, Behrendt F (2018) Towards biochar and hydrochar engineering-influence of process conditions on surface physical and chemical properties, thermal stability, nutrient availability, toxicity and wettability. *Energies* 11:496

Dinneen LC, Blakesley BC (1973) Algorithm AS 62 A generator for the sampling distribution of the Mann-Whitney *U* statistic. *Appl Statist* 22(2):269–273

Doran J, Brinton W (2001) Soil quality correlation of basal respiration and N mineralization over time using intact 3-inch field soil cores under optimal temperature and moisture in the lab. United

- States Department of Agriculture (USDA), Agricultural Research Service
- Dunn OJ (1964) Multiple comparisons using rank sums. *Technometrics* 6:241–252
- Eibisch N, Helfrich M, Don A, Mikutta R, Kruse A, Ellerbrock R, Flessa H (2013) Properties and degradability of hydrothermal carbonization products. *J Environ Qual* 42:1565–1573
- Fang J, Gao B, Chen J, Zimmerman AR (2015) Hydrochars derived from plant biomass under various conditions: characterization and potential applications and impacts. *Chem Eng J* 267:253–259
- Gai X, Wang H, Liu J, Zhai L, Liu S, Ren T, Liu H (2014) Effects of feedstock and pyrolysis temperature on biochar adsorption of ammonium and nitrate. *PLoS ONE* 9(12):e113888
- Ghanim BM, Kwapinski W, Leahy JJ (2018) Speciation of nutrients in hydrochar produced from hydrothermal carbonization of poultry litter under different treatment conditions. *ACS Sustain Chem Eng* 6:11265–11272
- Glaser B, Lehr V-I (2019) Biochar effects on phosphorus availability in agricultural soils: a meta-analysis. *Scientific Reports* 9:9338
- Greenberg I, Kaiser M, Gunina A, Ledesma P, Polifka S, Wiedner K, Mueller CW, Glaser B, Ludwig B (2019) Substitution of mineral fertilizers with biogas digestate plus biochar increases physically stabilized soil carbon but not crop biomass in a field trial. *Sci Total Environ* 680:181–189
- Gronwald M, Don A, Tiemeyer B, Halfrich M (2015) Effects of fresh and aged chars from pyrolysis and hydrothermal carbonization on nutrient sorption in agricultural soils. *SOIL* 1:475–489
- Fachagentur Nachwachsende Rohstoffe e.V. (FN R) 2018 Bioenergy in Germany: Facts and figures 2019. Gülzow-Prüzen, Germany. Available from: international.fnr.de http://www.fnr.de/fileadmin/allgemein/pdf/broschueren/broschuere_basisdaten_bioenergie_2018_engl_web_neu.pdf
- Hailegnaw NS, Mercl F, Pračke K, Száková J, Tlustoš P (2019) Mutual relationships of biochar and soil pH, CEC, and exchangeable base cations in a model laboratory experiment. *J Soils Sediments* 19:2405–2416
- Hardy B, Sleutel S, Dufey JE, Cornelis J-T (2019) The long-term effect of biochar on soil microbial abundance, activity and community structure is overwritten by land management. *Front Environ Sci* 7(110):1–14
- Higashida S, Takao K (1986) Relations between soil microbial activity and soil properties in grassland. *Soil Sci Plant Nutr* 32(4):587–597
- Huang Q, Wang Z, Wang C, Wang S, Jin X (2005) Phosphorus release in response to pH variation in the lake sediments with different ratios of iron-bound P to calcium-bound P. *Chem Speciation Bioavailability* 17:55–61
- Jeffery S, Abalos D, Prodana M, Bastos AC, Van Groenigen JW, Hungate BA, Verheijen F (2017) Biochar boosts tropical but not temperate crop yields. *Environ Res Lett* 12:053001
- Laerd Statistics (2015) Kruskal-Wallis H Test using SPSS Statistics. Statistical tutorials and software guides. Retrieved from: <http://statistics/laerd/com/>
- Lehmann J, Schroth G (2003) Nutrient leaching. In: Schroth G, Sinclair FL (eds) *Trees, crops and soil fertility—concepts and research methods*. CAB International, Wallingford, UK, pp 151–166
- Lehmann J, Gaunt J, Rondon M (2006) Bio-char sequestration in terrestrial ecosystems—a review. *Mitigation Adapt Strategies Global Change* 11:403–427
- Lehmann J, Rillig MC, Thies J, Masiello CA, Hockaday WC, Crowley D (2011) Biochar effects on soil biota—a review. *Soil Biol Biochem* 43(9):1812–1836
- Liao W, Thomas SC (2019) Biochar particle size and post-pyrolysis mechanical processing affect soil pH, water retention capacity, and plant performance. *Soil Syst* 3:14
- Libra JA, Ro KS, Kammann C, Funke A, Berge ND, Neubauer Y, Titirici M-M, Fühner C, Bens O, Kern J, Emmerich K-H (2011) Hydrothermal carbonisation of biomass residuals: a comparative review of the chemistry, processes and applications of wet and dry pyrolysis. *BioFuels* 2:71–106
- Malghani S, Jüschke E, Baumert J, Thuille A, Antonietti M, Trumbore S, Gleixner G (2015) Carbon sequestration potential of hydrothermal carbonization char (hydrochar) in two contrasting soils; results of a 1-year field study. *Biol Fertil Soils* 51:123–134
- Marmioli M, Bonas U, Imperiale D, Lencioni G, Mussi F, Marmioli N, Maestri E (2018) Structural and functional features of chars from different biomasses as potential plant amendments. *Front Plant Sci* 9:1119
- Melo TM, Bottlinger M, Schulz E, Leandro WM, de Aguiar Filho AM, Wang H, Ok YS, Rinklebe J (2018) Plant and soil responses to hydrothermally converted sewage sludge (sewchar). *Chemosphere* 206:338–348
- Melo TM, Bottlinger M, Schulz E, Leandro WM, de Oliveira SB, de Aguiar Filho AM, El-Naggar A, Bolan N, Wang H, Ok YS, Rinklebe J (2019) Management of biosolids-derived hydrochar (Sewchar): effect on plant germination, and farmers' acceptance. *J Environ Manag* 237:200–214
- Möller K, Müller T (2012) Effects of anaerobic digestion on digestate nutrient availability and crop growth: a review. *Eng Life Sci* 12(3):242–257
- Mukherjee S, Weihermueller L, Tappe W, Vereecken H, Buraue P (2016) Microbial respiration of biochar- and digestate-based mixtures. *Biol Fertil Soils* 52:151–164
- Parmar KR, Ross AB (2019) Integration of hydrothermal carbonisation with anaerobic digestion, opportunities for valorisation of digestate. *Energies* 12:1586
- Penn CJ, Camberato JJ (2019) A critical review on soil chemical processes that control how soil pH affects phosphorus availability to plants. *Agriculture* 9(6):120
- Prayogo C, Jones JE, Baeyens J, Bending GD (2014) Impact of biochar on mineralisation of C and N from soil and willow litter and its relationship with microbial community biomass and structure. *Biol Fertil Soils* 50:695–702
- Pritchard SL, Charlton WL, Baker A, Graham IA (2002) Germination and storage reserve mobilization are regulated independently in *Arabidopsis*. *Plant J* 31(5):639–647
- Puccini M, Ceccarini L, Antichi D, Seggiani M, Tavarini S, Latorre MH, Vitolo S (2018) Hydrothermal carbonization of municipal woody and herbaceous prunings: hydrochar valorisation as soil amendment and growth medium for horticulture. *Sustainability* 10:846
- Reza MT, Lynam JG, Uddin MH, Coronella CJ (2013) Hydrothermal carbonization: fate of inorganics. *Biomass Bioenergy* 49:86–94
- Reza MT, Andert J, Wirth B, Busch D, Pielert J, Lynam JG, Mumme J (2014) Hydrothermal carbonisation of biomass for energy and crop production. *Appl Bioenergy* 1:11–29
- Röhrdanz M, Greve T, de Jager M, Buchwald R, Wark R (2019) Cocomposted hydrochar substrates as growing media for horticultural crops. *Sci Hortic* 252:96–103
- Salem MSA (2013) Soil biota interactions with hydrochar. PhD thesis, Freie Universität Berlin, Germany
- Santín C, Doerr SH, Merino A, Bucheli TD, Bryant R, Ascough P, Gao X, Masiello CA (2017) Carbon sequestration potential and physicochemical properties differ between wildfire charcoals and slow-pyrolysis biochars. *Sci Rep* 7:11233
- Santos MOD, Barreto-Garcia PAB, Gama-Rodrigues EF (2019) Soil microbial biomass as an edge effect indicator in semi-deciduous seasonal forest fragments. *Floresta e Ambiente* 26(4):e20171037
- Steiner C, Teixeira WG, Woods WI, Zech W (2009) Indigenous knowledge about terra preta formation. In: Woods WI, Teixeira WG, Lehmann J, Steiner C, WinklerPrins A, Rebellato L (eds) *Amazonian dark earths: Wim Sombroek's vision*. Springer, Dordrecht, pp 193–204
- Steiner C, Bayode AO, Ralebitso-Senior TK (2016) Feedstock and production parameters: effects on biochar properties and microbial

- communities. In: Ralebitso-Senior TK, Orr CH (eds) *Biochar application: essential soil microbial ecology*. Elsevier, United Kingdom, pp 41–54
- Sun K, Han L, Yang Y, Xia X, Yang Z, Wu F, Li F, Feng Y, Xing B (2020) Application of hydrochar altered soil microbial community composition and the molecular structure of native soil organic carbon in a paddy soil. *Environ Sci Technol* 54:2715–2725
- VDLUFA (Verband Deutscher Landwirtschaftlicher Untersuchungs- und Forschungsanstalten (Hg) (2012) Bestimmung des Wassergehaltes (bzw. der Trockenmasse) durch Trocknen im Trockenschrank, Die Untersuchung von Böden, A 2.1.1, VDLUFA Methodenbuch I, Grundwerk, VDLUFA-Verlag, Darmstadt. https://www.vdlufa.de/Methodenbuch/index.php?option=com_content&view=article&id=2010&lang=de Accessed 13 Aug 2020
- Verheijen FGA, Jeffrey S, Bastos AC, van der Velde M, Dias I (2009) *Biochar application to soils: A critical scientific review of effects on soil properties, processes and functions*. EUR 24099 EN, Office for the Official Publications of the European Communities, Luxembourg
- Wang L, Xue C, Nie X, Liu Y, Chen F (2018) Effects of biochar application on soil potassium dynamics and crop uptake. *J Plant Nutr Soil Sci* 181(5):635–643
- Zhang X, Zhang L, Li A (2017) Hydrothermal co-carbonization of sewage sludge and pinewood sawdust for nutrient-rich hydrochar production: synergistic effects and products characterization. *J Environ Manag* 201:52–62