

## Article

# The Effect of Untreated and Acidified Biochar on NH<sub>3</sub>-N Emissions from Slurry Digestate

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**Abstract:** The development of new options to reduce ammonia (NH<sub>3</sub>) emissions during slurry manure storage is still required due to the shortcomings of the current technologies. This study aimed to identify to what extent untreated and acid-treated biochar (BC) and pure acids could reduce ammonia nitrogen (NH<sub>3</sub>-N) volatilization and increase nitrogen retention in slurry digestate. The NH<sub>3</sub>-N emissions were effectively reduced by H<sub>2</sub>SO<sub>4</sub> and H<sub>3</sub>PO<sub>4</sub> acids, untreated BC when applied mixed into the digestate and acidified BC treatments applied on the surface of the digestate. Acidification increased the specific surface area and number of O-containing surface functional groups of the BC and decreased the pH, alkalinity and the hydrophobic property. Compared to untreated BC, the ability of BC to reduce NH<sub>3</sub>-N emissions was greater when it was acidified with H<sub>2</sub>SO<sub>4</sub> and applied to the digestate surface. The effect on digestate pH of acidified BC when applied mixed into the digestate was not different, except for H<sub>2</sub>O<sub>2</sub>, from that of the addition of the respective pure acid to digestate. The total N concentration in digestate was not significantly correlated with NH<sub>3</sub>-N emissions. These findings indicate that acidified BC could be an effective conditioner to reduce NH<sub>3</sub>-N emissions from slurry digestate storage.

**Keywords:** biochar; slurry digestate; NH<sub>3</sub>-N emissions reduction; biochar acidification; digestate N concentration



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## 1. Introduction

Approximately 80–90% of global ammonia (NH<sub>3</sub>) emissions originate from agriculture [1], and 64% are associated with livestock production [2,3]. The major sources of NH<sub>3</sub> emissions are livestock housing, manure storage [4] and application of manure to grassland or arable land [5].

Although NH<sub>3</sub> is not considered a greenhouse gas, it is an important environmental pollutant. NH<sub>3</sub> emissions are linked to several global environmental problems such as acidification of soils [6], eutrophication in terrestrial and aquatic ecosystems [7] and the formation of particulate matter in the atmosphere [8]. Deposition of NH<sub>3</sub> in soil, water and air can have repercussions for plant biodiversity and human and ecosystem health at both local and regional scales [9].

The gaseous loss of nitrogen (N) also reduces the value of animal manure as a fertilizer for crop production [10,11]. Depending on the storage period, NH<sub>3</sub> losses from dairy cow digestate can account for between 6% and 40% of the total N contained in the digestate [12], whereas from pig slurry they can reach 50–60% [13].

The mitigation of NH<sub>3</sub> emissions has become a major focus of research in many countries to mitigate environmental issues [14] and reduce N losses from slurry fertilization [15]. Several manure management technologies have the ability to reduce NH<sub>3</sub> emissions, but

only a few of these solutions are commercially available for use in livestock farming because of technical and economic barriers that hinder their implementation [16].

One of the best-known methods to decrease  $\text{NH}_3$  emissions is covering the slurry storage areas. Slurry covers can be permeable or impermeable, fixed or floating. Currently, livestock farmers use permeable covers such as natural crust, straw, geotextiles or clay minerals or impermeable covers such as plastic, concrete, wood or steel [17–19]. Impermeable covers tend to have higher life expectancy and are up to 100% more effective than permeable covers in reducing  $\text{NH}_3$  gases from manure storage [20]. However, impermeable covers are very expensive, require a rainwater handling system and make the agitation of the manure more difficult [21].

Acidification of animal slurry is another manure management practice developed in Denmark, which uses concentrated sulfuric acid ( $\text{H}_2\text{SO}_4$  96%) as the acidification material [10]. However, the main shortcomings related to this technology are increased nitrous oxide ( $\text{N}_2\text{O}$ ) emissions [22], sulfur (S) leaching [23], risk of foaming [24] and potential hazards related to acid handling [10].

The level of  $\text{NH}_3$  emissions is greatly dependent on the equilibrium between aqueous (aq) ammonium ions ( $\text{NH}_4^+$ ) and (aq)  $\text{NH}_3$  in manure [25]. The most important factors that influence the transformation rate of  $\text{NH}_4^+$  to gaseous  $\text{NH}_3$  include  $\text{NH}_4^+$  concentration, pH and exposure to the atmosphere [26]. However, pH is the decisive factor affecting  $\text{NH}_3$  formation at a constant temperature [20]. The greatest increase in  $\text{NH}_3$  emissions takes place between a pH of 7 and 10, while at pH levels below 7 only small quantities of  $\text{NH}_3$  are released, and there is almost no measurable free  $\text{NH}_3$  around a pH of 4.5 [27]. Therefore, the concept of reducing slurry pH to decrease  $\text{NH}_3$  emissions relies on shifting the balance between  $\text{NH}_4^+$  (aq) and  $\text{NH}_3$  (aq) [28].

In recent years, there has been an increase from farming communities in using slurry conditioners. These products are considered to mitigate  $\text{NH}_3$  emissions in a cost-efficient manner [29] and are relatively easily applied to manure compared with other technologies that require modifications to the existing infrastructure and/or the purchase of expensive equipment [30]. Biochar (BC) is a porous material produced through pyrolysis or gasification of biomass at different temperatures with no or very low oxygen (O) availability [31]. BC has received increasing attention in recent years because of its diverse functionality in the fields of climate change mitigation, sustainable agriculture and environmental control [32]. The BC properties of high surface area, high porosity and high cation or anion exchange capacity make it a promising slurry conditioner to enhance  $\text{NH}_4^+$  retention [33] and reduce nitrate ( $\text{NO}_3^-$ ) leaching [34]. These changes occur through the addition of BC in manure pits and/or to the soil together with the manure [35,36]. BC has been shown to decrease  $\text{NH}_3$  volatilization and improve N retention in poultry litter composting [37] and to adsorb ammonium nitrogen ( $\text{NH}_4^+\text{-N}$ ) from piggery manure anaerobic digestate slurry [38], thus enhancing the fertilizer value of manure. However, with the increasing quantity of BC addition, the alkalinity of BC is likely to increase the pH of the manure and shift the  $\text{NH}_4^+/\text{NH}_3$  equilibrium toward  $\text{NH}_3$  volatilization [39]. To address this issue, the acidic oxidation of BC can neutralize the alkaline pH and facilitate the adsorption of  $\text{NH}_4^+$  because of the increased amount of O-containing surface functional groups [40]. To date, only a few studies have examined the reduction of  $\text{NH}_3$  emissions using untreated BC as a digestate conditioner applied to manure and slurry storage tanks [35,41]. It was determined that untreated BC can effectively mitigate  $\text{NH}_3$  emissions from stored swine manure [35]. At the same time, acid modification can affect the physicochemical properties of the BC which in turn could alter the mitigation effect [42]. To the best of our knowledge, there are no published studies comparing surface and mixing application of acidified BC to reduce  $\text{NH}_3$  emissions.

The aim of the present study was to investigate (i) the ability of hay BC to reduce  $\text{NH}_3\text{-N}$  emissions from digestate, (ii) to what extent the suppression effect of  $\text{NH}_3\text{-N}$  emissions of BC can be enhanced through acidification, (iii) which method is more effective for the application of untreated and acidified BC (mixed or surface), and (iv) whether untreated

and acidified BC addition reduces total N ( $N_{\text{tot}}$ ),  $\text{NH}_4^+$ -N and nitrate-nitrogen ( $\text{NO}_3^-$ -N) losses in digestate. We hypothesized the following: (1) low-temperature hay BC can reduce  $\text{NH}_3$ -N emissions from slurry digestate, (2) BC acidification increases the capacity of BC to reduce  $\text{NH}_3$ -N emissions, (3) BC effectiveness in reducing N losses depends on its application method to the digestate, and (4) untreated and acidified BC addition mitigates N losses from the digestate. The results of our study will generate information for the development of alternatives to reduce N losses from slurry storage facilities.

## 2. Materials and Methods

### 2.1. Experimental Design

The research trial was carried out under controlled laboratory conditions at the Estonian University of Life Sciences (EULS) during a period of 240 days from October 2018 until May 2019. The experiment included a total of 12 treatments and consisted of two experimental factors: (i) digestate conditioner (untreated BC, acidified BC and acid) and (ii) application method of digestate conditioner (BC on the surface – BCs; BC mixed into the digestate – BCm). The pure acids were applied in pure form mixed into digestate only (Table 1).

**Table 1.** Experimental treatment abbreviations and description. BC: biochar.

Abbreviation	Treatment description
Control	Digestate
BCm	Untreated BC mixed into digestate
BCs	Untreated BC on the digestate surface
BCm + $\text{H}_2\text{SO}_4$	BC treated with sulfuric acid mixed into digestate
BCs + $\text{H}_2\text{SO}_4$	BC treated with sulfuric acid on the digestate surface
BCm + $\text{H}_3\text{PO}_4$	BC treated with phosphoric acid mixed into digestate
BCs + $\text{H}_3\text{PO}_4$	BC treated with phosphoric acid on the digestate surface
BCm + $\text{H}_2\text{O}_2$	BC treated with hydrogen peroxide mixed into digestate
BCs + $\text{H}_2\text{O}_2$	BC treated with hydrogen peroxide on the digestate surface
$\text{H}_2\text{SO}_4$	Sulfuric acid mixed into digestate
$\text{H}_3\text{PO}_4$	Phosphoric acid mixed into digestate
$\text{H}_2\text{O}_2$	Hydrogen peroxide mixed into digestate

For accuracy and logistical reasons, the experiment was conducted in two batches. The measurements of digestate pH can increase N emissions in BC treatments, which can bias the estimation of retained N concentrations in digestate. In addition, it was more convenient to measure the N concentrations in all treatments at the same time (and not as one replicate after another during the time span of 8 months). Therefore, in the first batch,  $\text{NH}_3$  emissions and digestate pH were measured and replicated three times from October 2018 until May 2019. In parallel, the second batch examined  $\text{NH}_4^+$ -N,  $\text{NO}_3^-$ -N, total nitrogen ( $N_{\text{tot}}$ ) and total carbon ( $C_{\text{tot}}$ ) concentrations, which were replicated four times from February 2019 until April 2019. In both batches, the replications were carried out in the same order and the digestate conditioners were applied in the same amounts, manner and laboratory conditions.

### 2.2. Materials

The BC was produced from reed canary (*Phalaris arundinacea* L.) hay pellets and torrefied at 300 °C. The quantities on a dry weight basis of total phosphorous ( $P_{\text{tot}}$ ), total potassium ( $K_{\text{tot}}$ ), total calcium ( $\text{Ca}_{\text{tot}}$ ) and total magnesium ( $\text{Mg}_{\text{tot}}$ ) were 2400 mg  $\text{kg}^{-1}$ , 22,800 mg  $\text{kg}^{-1}$ , 9300 mg  $\text{kg}^{-1}$  and 4700 mg  $\text{kg}^{-1}$ , respectively. The ash concentration was 10.4%, cumulative pore volume represented 0.0015  $\text{cm}^3 \text{g}^{-1}$ , and the concentration of volatile compounds was 62.9%. The nutrient concentrations, specific surface area (SSA), pH and acid neutralization capacity were determined in the EULS laboratory (Tartu, Estonia). The remaining physicochemical properties were determined at the Lithuanian Energy Institute (Kaunas, Lithuania).

The digestate originated from the Ilmatsalu biogas plant (Tartu county, Estonia) and consisted of 85% cattle slurry; approximately 10% was a mixture of pig slurry, manure (straw bedding) and green biomass, and less than 5% contained food industry by-products. Physicochemical details of the acidified BC conditioners are shown in Table 2.

**Table 2.** Characteristics of digestate, untreated and acid-treated BC.

Parameter	Digestate	BC	BC + H <sub>2</sub> SO <sub>4</sub>	BC + H <sub>3</sub> PO <sub>4</sub>	BC + H <sub>2</sub> O <sub>2</sub>
pH	8.2	6.40	1.55	2.65	4.80
Specific surface area, m <sup>2</sup> g <sup>-1</sup>	-	126.14	161.48	145.92	166.31
Acid neutralization capacity (CaCO <sub>3</sub> , %)	-	3.80	0.00	0.49	3.86
Dry matter (%)	4.06	100	100	100	100
C <sub>tot</sub> (%)	40.37	50.29	51.18	53.57	51.77
N <sub>tot</sub> (%)	6.87	2.79	2.81	2.96	2.85
C/N	5.88	18.03	18.22	18.10	18.16
NH <sub>4</sub> <sup>+</sup> -N (mg kg <sup>-1</sup> )	46,390	80	400	230	220
NO <sub>3</sub> <sup>-</sup> -N (mg kg <sup>-1</sup> )	3520	60	80	90	80

### 2.3. Experimental Set-Up

A schematic of the experimental set-up is shown in Figure 1. Digestate was stored in a closed 30-L plastic bucket at 4–6 °C during the entire experiment. Before each sampling, the digestate was mixed thoroughly with a rod to obtain a homogenous material. The digestate was then transferred with a graduated glass beaker to 125-mL Berzelius beakers for the control and BC treatments while 200-mL beakers were used for the acids to allow for the formation of foam. Precision balances PS 450/X (RADWAG, Radom, Poland) were used to weigh digestate and conditioner materials. The weight of digestate for each sample was 100 g, and the weight of untreated and acid-treated BC was 5 g. The weights of added acids were calculated based on the standard dosage rate of 5 kg of H<sub>2</sub>SO<sub>4</sub> (96% concentration) per 1 ton of slurry, which has been previously utilized in the slurry acidification technology [10]. The weights of the other two acids were calculated based on the molar mass and concentration of the acid:

$$m = nM$$

where  $m$  is the mass of the acid (g),  $n$  is 0.005 (mol), and  $M$  is the molar mass of the acid (g/mol). Therefore, the application rate of H<sub>2</sub>SO<sub>4</sub> at 96% concentration was 0.5 g, H<sub>3</sub>PO<sub>4</sub> of 85% was 0.59 g, and H<sub>2</sub>O<sub>2</sub> of 50% was 0.34 g.

The BC pellets were ground manually using a lab porcelain mortar and pestle until they had a particle size of less than 1.25 mm.

Ten grams of milled BC were weighed into a 200-mL plastic flask with a cap, to which 100 mL of 0.5 N H<sub>2</sub>SO<sub>4</sub> solution was added at 1:10 ( $w/v$ ) BC to acid ratio. The plastic flasks with BC and acid solution were sealed and placed in a reciprocating shaker GFL 3006 (GFL, Burgwedel, Germany) to be agitated for a minimum of 24 h at 100 rpm. The acidified BC particles were separated from the liquid via filtering. A cone-like shaped piece of filter paper 56 G (Lachner, Neratovice, Czechia) was placed within a glass funnel with a diameter of 9–11 cm, which in turn was placed into a conical glass flask of 300 mL. The suspension from the plastic flasks was poured through the funnel and left to filtrate for at least 24 h. Identical solution normality and preparation steps were followed to acidify BC with H<sub>3</sub>PO<sub>4</sub> and H<sub>2</sub>O<sub>2</sub>.

A multi-gas detection equipment X-am 7000 (Dräger, Lubeck, Germany) connected to an 860-CG acrylic desiccator chamber with a gas port (Plas-Labs, Lansing, MI, USA) was used to measure NH<sub>3</sub> concentrations. The volume of the chamber (Plas-Labs) was 24.9-L, equipped with two gas ports and built of transparent acrylic material. Recorded data were transferred to PC using an infrared interface and GasVision software version 5.8.2. (Dräger, Lubeck, Germany). The X-am 7000 apparatus was calibrated before each replication.

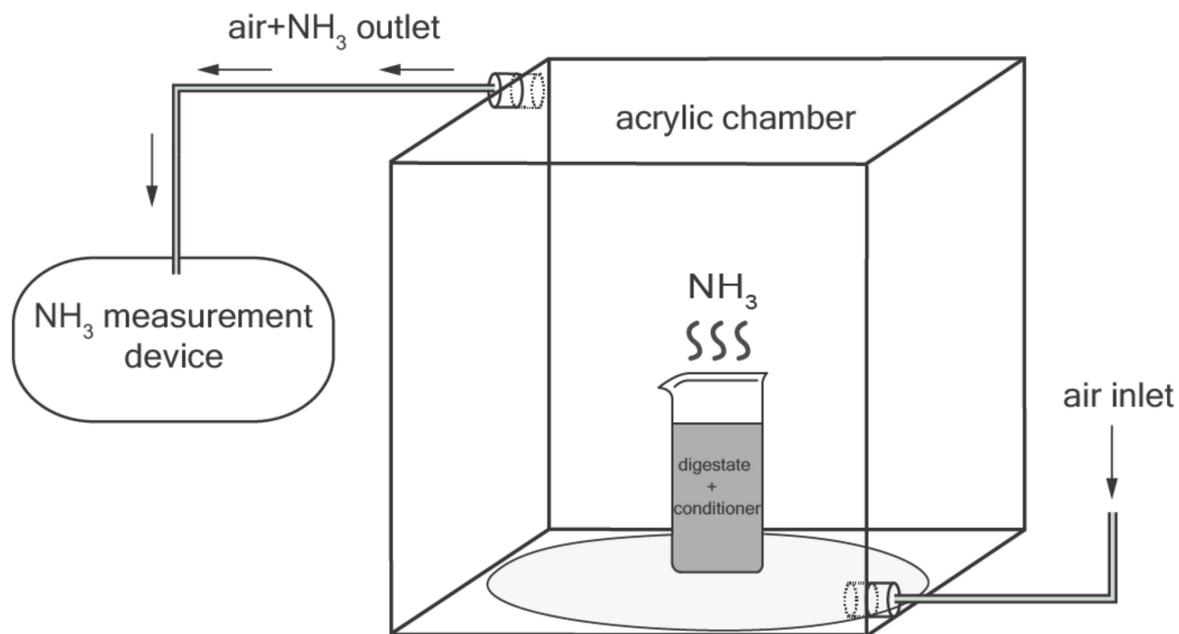


Figure 1. Schematic of the experimental set-up.

## 2.4. Measurements and Analyses

### 2.4.1. NH<sub>3</sub> Concentration and Digestate pH

The NH<sub>3</sub> concentrations were measured in two separate sessions. The first session lasted for 48 h (2880 min), immediately after the digestate was exposed to ambient conditions. In the second session, the gas concentrations were monitored for 24 h (1440 min) from the 7th to 8th day after the beginning of the experiment. Between the measurement sessions, all samples were stored in an unilluminated laboratory cabinet with similar environmental conditions to the location of the measurement sessions.

The pH of the digestate was measured with an HD 2156.2 pH meter (Delta OHM, Padua, Italy) at the beginning of the experiment and after 48, 72, 96 and 120 h. In the untreated and acidified BCs treatments, the pH measurements were recorded under the BC layer (approximately 3–5 cm deep) and in the middle of the vessel (approximately 7–9 cm deep). In the untreated and acidified BCm as well as in the control and acid treatments, the pH was measured only in the middle of the vessel.

### 2.4.2. Nutrient Concentration

The N<sub>tot</sub>, C<sub>tot</sub>, NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N concentrations were measured in the digestate at the beginning of the experiment and after 30 days. The same parameters were also examined separately in the BC conditioners before their application to the digestate. The N<sub>tot</sub> and C<sub>tot</sub> concentrations were determined after dry combustion with a varioMAX CNS elemental analyzer (Elementar Analysensysteme GmbH, Langenselbold, Germany). The NH<sub>4</sub><sup>+</sup>-N and NO<sub>3</sub><sup>-</sup>-N were determined after steam distillation [43] using a UDK 126D distillation unit (VELP Scientifica, Usmate Velate, Italy). The N-containing species were calculated by subtracting the concentration in the BC from the total concentration retained in the digestate.

### 2.4.3. BC Examination

The pH of BC conditioners was determined by a benchtop pH meter SevenCompact S210 (Mettler-Toledo, Schwerzenbach, Switzerland) in a suspension of 1 M KCl using a 1:2.5 (w/v) BC to solution ratio. CaCO<sub>3</sub> concentration of BC was established via the titrimetric method explained in detail in the standard operating procedure of Food and Agriculture

Organization (FAO) [44]. The acid neutralization capacity expressed as a percentage of  $\text{CaCO}_3$  was calculated based on the following formula:

$$\text{CaCO}_3, (\%) = \left( \frac{V_{\text{HCl}}N_{\text{HCl}} - V_{\text{NaOH}}N_{\text{NaOH}}}{m} \right) \times 0.05 \times 100$$

where  $V_{\text{HCl}}N_{\text{HCl}}$  and  $V_{\text{NaOH}}N_{\text{NaOH}}$  are the volume and normality of hydrogen chloride (HCl) and sodium hydroxide (NaOH), and  $m$  is the mass of BC material (g).

The SSA ( $\text{m}^2 \text{g}^{-1}$ ) was estimated by the water vapor adsorption method [45] using the relationship explained by Gámiz et al. [46]:

$$\text{SSA} = \frac{n_m N \sigma}{m} 100 = 36.16 \left( \frac{n_m}{m} 100 \right)$$

where  $n_m$  is  $\text{H}_2\text{O}$  in monolayer (mol),  $N$  is Avogadro's number ( $6.02 \times 10^{23}$ ),  $\sigma$  is surface area per  $\text{H}_2\text{O}$  molecule ( $10.8 \times 10^{-20} \text{m}^2$ ), and  $m$  is the mass of sorbent (g).

Fourier transform infrared spectroscopy (FTIR) analysis was performed to analyze the surface functional groups using a Thermo-Nicolet iS10 Fourier transformed infrared spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA). The spectral range was between 4000 to  $400 \text{cm}^{-1}$  with a resolution of  $4 \text{cm}^{-1}$ , and 32 scans were conducted. An automated baseline correction was applied to adjust the resulted spectra in OMNIC software version 8.3.103. (Thermo Fisher Scientific, Waltham, MA, USA).

## 2.5. Flux Calculation

The  $\text{NH}_3$  concentrations ( $\text{mg m}^{-3}$ ) were calculated in accordance with the ideal gas law:

$$C_{\text{NH}_3} = \frac{V_{\text{NH}_3} MW_{\text{NH}_3} P}{RT}$$

where  $V_{\text{NH}_3}$  is the volume of  $\text{NH}_3$  ( $\text{ppm min}^{-1}$ ),  $MW_{\text{NH}_3}$  is the molecular weight of  $\text{NH}_3$  (g),  $P$  is pressure (atm),  $R$  is the universal gas constant ( $\text{L atm mol}^{-1} \text{K}^{-1}$ ), and  $T$  is the ambient temperature (K).

The  $\text{NH}_3$  measurements were conducted in an open dynamic chamber system in which the sampled air was pumped outside from the chamber while at the same time the fresh ambient air was drawn into the chamber. The air from the chamber with  $\text{NH}_3$  concentration was pumped out of the measurement equipment after measurement to avoid contamination with the air pulled into the chamber. The measured  $\text{NH}_3$  concentrations had therefore to be corrected for the  $\text{NH}_3$  concentration loss of the gas flow. The corrected  $\text{NH}_3$  concentrations ( $\text{mg m}^{-3}$ ) were determined according to in- and outflow rates as follows:

$$C_{\text{corNH}_3} = C_{\text{NH}_3} + \frac{\{ [IR(Air_{\text{NH}_3} IR)] + [C_{\text{NH}_3}(V_{\text{chamber}} - OR)] \}}{V_{\text{chamber}}}$$

where  $C_{\text{NH}_3}$  is the measured  $\text{NH}_3$  concentration in the chamber ( $\text{mg m}^{-3}$ ),  $IR$  is inflow rate ( $\text{m}^3 \text{min}^{-1}$ ),  $Air_{\text{NH}_3}$  is the concentration of  $\text{NH}_3$  in the air outside the chamber ( $\text{mg m}^{-3}$ ),  $V_{\text{chamber}}$  is the volume of the chamber ( $\text{m}^3$ ), and  $OR$  is outflow rate ( $\text{m}^3 \text{min}^{-1}$ ).

The  $\text{NH}_3\text{-N}$  fluxes ( $\text{mg m}^{-2} \text{h}^{-1}$ ) were calculated from the slope of the linear regression of the corrected  $\text{NH}_3$  concentrations of a 10 min interval (10 measurements) using the formula [47]:

$$F_{\text{NH}_3\text{-N}} = \frac{(m_{\text{NH}_3} V_{\text{chamber}}) M_{\text{N}}}{(S_{\text{beaker}} t M p^{-1}) M_{\text{NH}_3}}$$

where  $m_{\text{NH}_3}$  is the slope of  $\text{NH}_3$  concentration changed in time ( $\text{mg m}^{-3}$ ),  $V_{\text{chamber}}$  is the volume of the chamber ( $\text{m}^3$ ),  $M_{\text{N}}$  is the molar mass of N ( $\text{g mol}^{-1}$ ),  $S_{\text{beaker}}$  is the surface of the beaker ( $\text{m}^2$ ),  $M p$  is the number of measurement points during measurement sessions,  $t$  is the time conversion factor to 1 h, and  $M_{\text{NH}_3}$  is the molar mass of  $\text{NH}_3$  ( $\text{g mol}^{-1}$ ).

## 2.6. Statistical Analysis

Statistical analysis was conducted using the R programming software [48]. Data were analyzed using analysis of variance (ANOVA) to study the effect of BC treatment (untreated and acidified), time and the interaction between them on NH<sub>3</sub> emissions; digestate pH and N concentrations were compared for the two application methods (mixed in or on the surface). The pure-form acids were also tested but using only the mixed application method. Moreover, the effect of the three acids on the capacity of BC to reduce digestate pH and mitigate N losses over time was investigated between the application methods. When significant differences existed, a post-hoc Tukey's honest significant difference (HSD) test was conducted to study the differences between groups [49]. The differences in pH were analyzed within and between time points. The NH<sub>3</sub> emissions were compared between 100 min (1.6 h) intervals and cumulatively at the end of the first 48 h and for the session between the 7th to 8th measurement days. The different N compounds were tested between treatments at the end of the 30-day period. In addition, the proportion of the variance explained by each factor was determined from ANOVA analysis. Linear regression and Pearson correlation coefficient were used to analyze the relationship between NH<sub>3</sub>-N emissions and pH or mineral N concentration. Principle component analysis (PCA) of the FTIR spectra was performed using the function "dudi.pca" from the package "ade4" [50].

## 3. Results

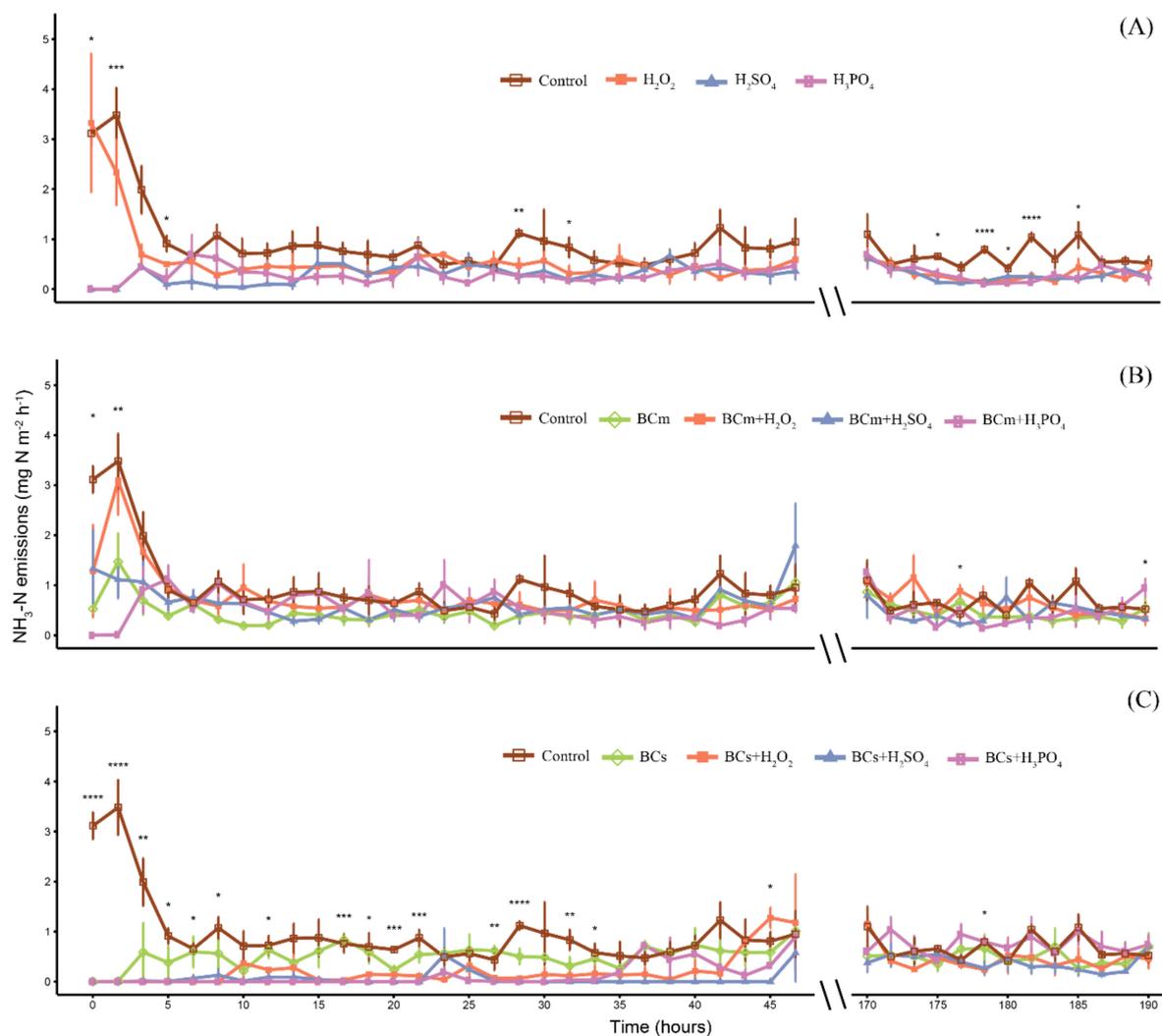
### 3.1. NH<sub>3</sub>-N Emissions

The NH<sub>3</sub>-N emissions were significantly influenced by the digestate conditioner, time and BC application method ( $p < 0.001$ ). Significant interactions were observed between BC, application method, acid and time ( $p < 0.001$ ) (Table 3).

**Table 3.** NH<sub>3</sub>-N emissions during 0–48 h as influenced by conditioner, application method and time passed since conditioner application (ANOVA analysis;  $n = 3$ ). Asterisks indicate the level of significance. \*\*\*\*  $p < 0.0001$ ; \*\*\*  $p < 0.001$ ; \*\*  $p < 0.01$ ; \*  $p < 0.05$ . A lack of \* symbols indicates significance at  $p < 0.1$ .

Factor	Df	Sum Sq.	Mean Sq.	F Value	Pr(>F)	R <sup>2</sup>	ω <sup>2</sup>	Sig
BC	1	48	47.70	46.127	$1.07 \times 10^{-11}$	0.0039	0.0038	***
BC application method	1	251	251.33	243.025	$<2 \times 10^{-16}$	0.0207	0.2066	***
Acids	3	206	68.68	66.411	$<2 \times 10^{-16}$	0.0170	0.0167	***
Time passed	287	446	1.55	1.502	$1.60 \times 10^{-7}$	0.0368	0.0122	***
BC:acids	3	101	33.80	32.686	$<2 \times 10^{-16}$	0.0083	0.0081	***
BC application method:acids	3	104	34.72	33.573	$<2 \times 10^{-16}$	0.0086	0.0083	***
BC:time passed	287	439	1.53	1.478	$4.85 \times 10^{-7}$	0.0362	0.0117	***
BC application method:time passed	287	374	1.30	1.262	0.002165	0.0309	0.0064	**
Acid:time passed	861	1087	1.26	1.221	$2.86 \times 10^{-5}$	0.0897	0.0162	***
BC:acid:time passed	861	1069	1.24	1.200	0.000121	0.0882	0.0147	***
BC application method:acids:time passed	861	838	0.97	0.941	0.879399	0.0691	0.0000	
Residuals	6912	7148	1.03					

All BC treatments, except for BC<sub>m</sub>+H<sub>2</sub>O<sub>2</sub>, significantly reduced NH<sub>3</sub>-N emissions relative to the control during the first 3.5 h, but the reducing effect of BC<sub>m</sub>+H<sub>2</sub>SO<sub>4</sub> became significant between 1.6 and 3.5 h. From 3.5 h until the end of the first measurement session, NH<sub>3</sub>-N emissions increased in untreated BC<sub>m</sub>, BCs and all acidified BC<sub>m</sub> treatments and were not significantly different from the control ( $p > 0.05$ ) (Figure 2). In the acidified BCs treatments, NH<sub>3</sub>-N emissions were altogether lower than the control during the first 33 h ( $p < 0.05$ ). From 33 h until 48 h, the NH<sub>3</sub>-N emissions from BC<sub>s</sub>+H<sub>2</sub>SO<sub>4</sub> continued to be lower than those of the control ( $p > 0.05$ ), but from the BC<sub>s</sub> + H<sub>3</sub>PO<sub>4</sub> and BC<sub>s</sub> + H<sub>2</sub>O<sub>2</sub> treatments, a few gaseous emissions were higher than the control.



**Figure 2.**  $\text{NH}_3\text{-N}$  emissions (mean  $\pm$  standard error) during 0–48 h and from the 7th to 8th day after the application of pure acids (A), untreated and acidified biochar mixed in the digestate (B) and untreated and acidified biochar applied on the digestate surface (C). Control without conditioner;  $\text{H}_2\text{O}_2$ ,  $\text{H}_2\text{SO}_4$  and  $\text{H}_3\text{PO}_4$  pure-form acids applied mixed to digestate; BC: biochar; BC +  $\text{H}_2\text{O}_2$ , BC +  $\text{H}_2\text{SO}_4$  and BC +  $\text{H}_3\text{PO}_4$  represent BC acidified with the respective acid; BCm indicates that the BC was mixed into the digestate, and BCs indicates that the BC was applied on the digestate surface. Asterisks indicate the level of significant differences between treatments. \*\*\*\*  $p < 0.0001$ ; \*\*\*  $p < 0.001$ ; \*\*  $p < 0.01$ ; \*  $p < 0.05$ . A lack of \* symbols indicates significance at  $p < 0.1$ .

The suppression effect of untreated BCs was not different from untreated BCm during the first 3.5 h ( $p = 0.80$ ). In the untreated BCm treatment, a sharp increase in  $\text{NH}_3\text{-N}$  emissions was detected during the first 1.5 h (Figure 2B), which was followed by a decrease between 1.5 and 3.5 h. However, in untreated BCs, no emissions were recorded during the first 1.5 h, and a steep increase in  $\text{NH}_3\text{-N}$  emissions was recorded from 1.5 until 3.5 h. From 3.5 h onward, the  $\text{NH}_3\text{-N}$  emissions in untreated BCs were higher than untreated BCm, but the differences were not significant ( $p > 0.05$ ).

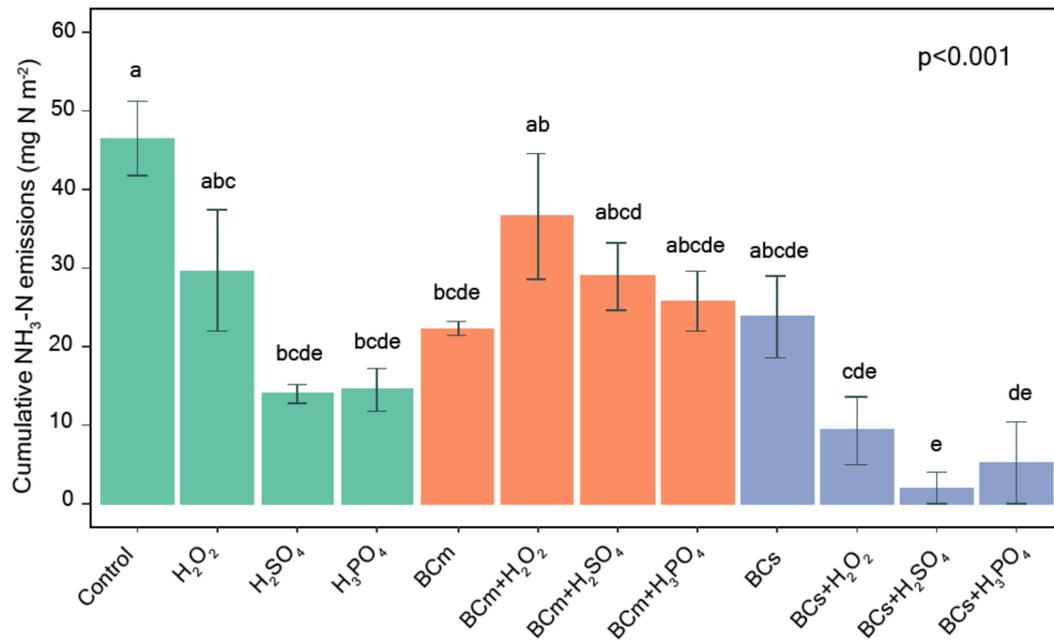
Of the pure acids, only  $\text{H}_2\text{SO}_4$  and  $\text{H}_3\text{PO}_4$  significantly reduced  $\text{NH}_3\text{-N}$  emissions. During the 48-h time period, the reduction effect of both acids was statistically significant during the first 5 h and from 28 to 33 h ( $p < 0.01$ ) (Figure 2A).

BC acidification improved the suppression effect of BCs but not of BCm (Figure 2B,C). The enhancement of BCs was dependent on the type of acid. Acidification with  $\text{H}_2\text{SO}_4$ ,  $\text{H}_2\text{O}_2$  and  $\text{H}_3\text{PO}_4$  prolonged the reduction effect of BCs by 41.5, 38.5 and 31.5 h, respectively.

After seven days, the emissions of  $\text{NH}_3\text{-N}$  in all BC treatments were not significantly different from those of the control ( $p > 0.05$ ) (Table S1), although the suppression effect of pure acids occasionally were strong (Figure 2A).

### 3.2. Cumulative $\text{NH}_3\text{-N}$ Emissions

Cumulative  $\text{NH}_3\text{-N}$  emissions in the control were  $46.45 \pm 4.73 \text{ mg N m}^{-2}$  after 48 h (Figure 3). Addition of untreated BCs and BCm reduced the cumulative  $\text{NH}_3\text{-N}$  emissions by 48% ( $p = 0.07$ ) and 52% ( $p = 0.04$ ) compared with the control. The suppression effect of BCs and BCm was not significantly different ( $p = 1.00$ ).



**Figure 3.** Cumulative  $\text{NH}_3\text{-N}$  emissions for 0–48 h from conditioner application (mean  $\pm$  standard error ( $n = 3$ )). Results marked with different lowercase letters are statistically different. Control without conditioner;  $\text{H}_2\text{O}_2$ ,  $\text{H}_2\text{SO}_4$  and  $\text{H}_3\text{PO}_4$  pure-form acids applied mixed to digestate; BC: biochar; BC +  $\text{H}_2\text{O}_2$ , BC +  $\text{H}_2\text{SO}_4$  and BC +  $\text{H}_3\text{PO}_4$  are BC acidified with the respective acid; BCm indicates that the BC was mixed into the digestate, and BCs indicates that the BC was applied on the digestate surface.

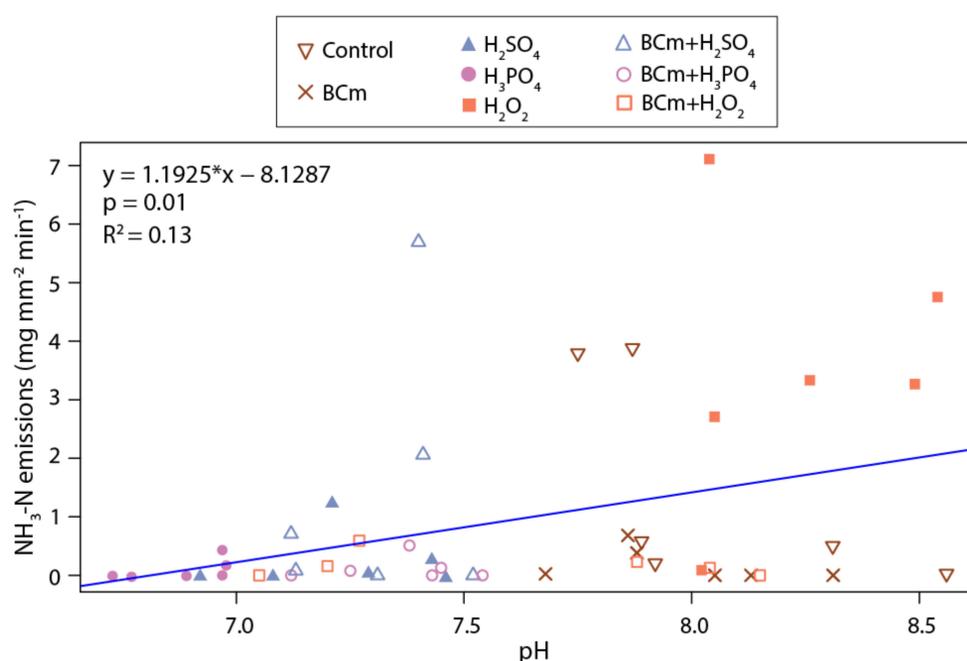
Relative to control, the  $\text{H}_2\text{O}_2$ ,  $\text{H}_3\text{PO}_4$  and  $\text{H}_2\text{SO}_4$  acids reduced  $\text{NH}_3\text{-N}$  emissions by 36.7% ( $p = 0.37$ ), 68% and 70% ( $p < 0.01$ ), respectively. In addition, the acidified BCs+ $\text{H}_2\text{O}_2$ , BCs+ $\text{H}_3\text{PO}_4$  and BCs+ $\text{H}_2\text{SO}_4$  conditioners reduced  $\text{NH}_3\text{-N}$  emissions compared to control by 80%, 88% and 95% ( $p < 0.001$ ). Meanwhile, acidified BCm+ $\text{H}_2\text{O}_2$ , BCm+ $\text{H}_2\text{SO}_4$  and BCm+ $\text{H}_3\text{PO}_4$  decreased cumulated emissions only by 21% ( $p = 0.93$ ), 38% ( $p = 0.31$ ) and 44% ( $p = 0.14$ ) in comparison to control.

Acidification improved the suppression effect of BCs while it decreased the effectiveness of BCm. The suppression effect in acidified BCs +  $\text{H}_2\text{O}_2$ , BCs +  $\text{H}_3\text{PO}_4$  and BCs +  $\text{H}_2\text{SO}_4$  treatments was improved by 61% ( $p = 0.57$ ), 78% ( $p = 0.24$ ) and 91% ( $p = 0.09$ ) compared to untreated BCs, respectively. In contrast, the suppression success in BCm +  $\text{H}_2\text{O}_2$ , BCm +  $\text{H}_2\text{SO}_4$  and BCm +  $\text{H}_3\text{PO}_4$  conditioners compared with the untreated BCm decreased by 64% ( $p = 0.59$ ), 30% ( $p = 0.99$ ) and 16% ( $p = 0.99$ ). Between the 7th and 8th day, the cumulative  $\text{NH}_3\text{-N}$  emissions were not significantly different between treatments ( $p > 0.05$ ) (Figure S1).

### 3.3. pH of Digestate

The rates of  $\text{NH}_3\text{-N}$  emissions were positively correlated with the pH values in digestate after the addition of the experiment conditioners. The majority of  $\text{NH}_3\text{-N}$  emissions occurred in the pH range from 6.80 to 8.50 and peaked between 7.75 and 8.50 (Figure 4). The

application location of untreated and acidified BC, type of acid and time had a significant effect on pH levels (Table S2). Untreated BCm significantly decreased ( $p = 0.01$ ) the average digestate pH by 0.40, whereas the average pH in the untreated BCs treatment was not different ( $p = 0.35$ ) from that of the control.  $H_2SO_4$  and  $H_3PO_4$  significantly decreased ( $p < 0.001$ ) the average pH in digestate by 0.80 and 1.15, whereas  $H_2O_2$  slightly increased it ( $p = 1.00$ ) (Table S3). The reduction effect on the digestate pH of BCm +  $H_2O_2$  ( $p = 0.003$ ), BCm +  $H_2SO_4$  ( $p = 0.004$ ) and BCm +  $H_3PO_4$  ( $p = 0.01$ ) was two times greater than that of the untreated BCm, and it was not different from the effect of the addition of  $H_2SO_4$  ( $p = 1.00$ ) and  $H_3PO_4$  ( $p = 0.07$ ) acids alone during the first five days of measurement. Acidified BCs did not influence digestate pH in the middle of the vessel ( $p > 0.05$ ), but the pH was 0.2–0.3 less beneath the surface cover than that under the untreated BCs cover. BCs +  $H_3PO_4$  was the only treatment to have an average pH significantly lower ( $p = 0.03$ ) than untreated BCs.

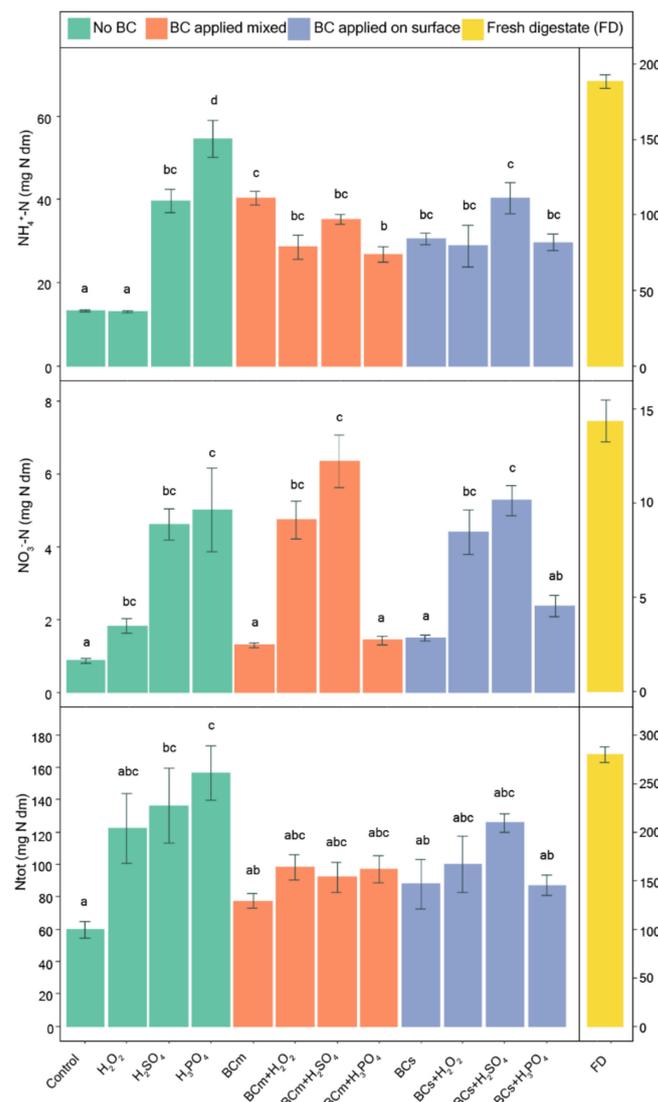


**Figure 4.**  $NH_3$ -N emissions from pure acids, untreated and acidified BCm treatments depending on digestate pH ( $n = 3$ ). Control without conditioner;  $H_2O_2$ ,  $H_2SO_4$  and  $H_3PO_4$  pure-form acids applied mixed with digestate; BC: biochar; BC +  $H_2O_2$ , BC +  $H_2SO_4$  and BC +  $H_3PO_4$  are BC acidified with the respective acid; BCm indicates that the BC was mixed with the digestate.

### 3.4. Concentration of $NH_4^+$ -N, $NO_3^-$ -N and $N_{tot}$

Thirty days from the beginning of the experiment, the  $NH_4^+$ -N concentration was significantly higher ( $p < 0.01$ ) than that of the control in all conditioner treatments, except  $H_2O_2$ .  $NO_3^-$ -N concentration in the BC +  $H_2O_2$ , BC +  $H_2SO_4$  and the  $H_2SO_4$  and  $H_3PO_4$  treatments ( $p < 0.001$ ) and  $N_{tot}$  in the  $H_2SO_4$  and  $H_3PO_4$  treatments were higher ( $p < 0.01$ ) than control (Figure 5).

The impact of BCs and BCm on the  $NO_3^-$ -N concentration increased significantly ( $p < 0.01$ ) after they were acidified with  $H_2O_2$  and  $H_2SO_4$ , whereas acidification with  $H_3PO_4$  decreased ( $p = 0.04$ ) the impact of BCm on the  $NH_4^+$ -N concentration. However, the effect of the BC on the  $N_{tot}$  concentration remained unchanged through the acidification process ( $p > 0.05$ ).



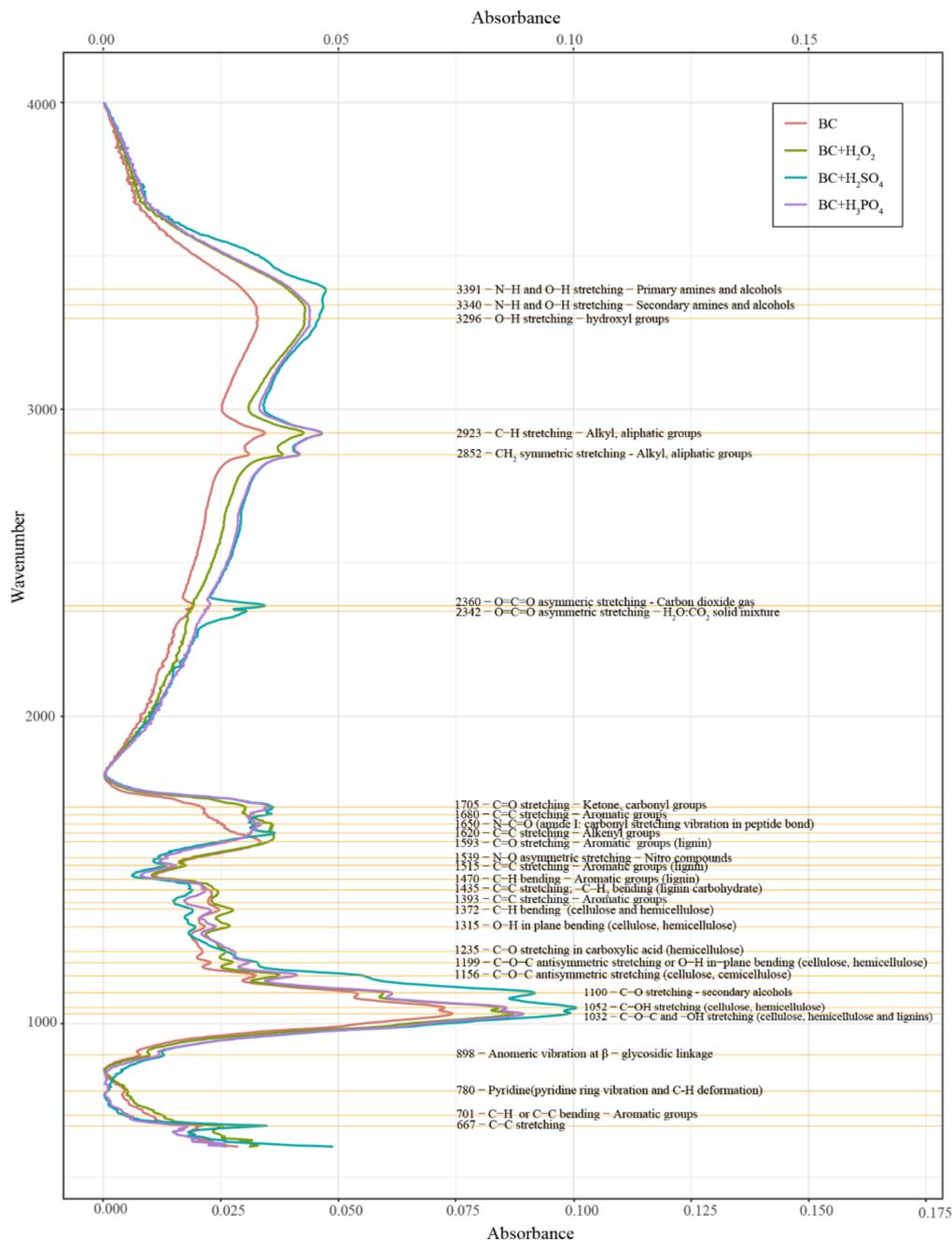
**Figure 5.** Concentration of  $\text{NH}_4^+\text{-N}$ ,  $\text{NO}_3^-\text{-N}$  and  $\text{N}_{\text{tot}}$  (mean  $\pm$  standard error;  $n = 3$ ) in the digestate 30 days after conditioner addition. Results marked with different lowercase letters are statistically different. Control without conditioner;  $\text{H}_2\text{O}_2$ ,  $\text{H}_2\text{SO}_4$  and  $\text{H}_3\text{PO}_4$  pure-form acids applied mixed to digestate; BC: biochar; BC +  $\text{H}_2\text{O}_2$ , BC +  $\text{H}_2\text{SO}_4$  and BC +  $\text{H}_3\text{PO}_4$  represent BC acidified with the respective acid; BCm indicates that the BC was mixed with the digestate, and BCs indicates that the BC was applied to the digestate surface.

### 3.5. Structural Characterization of BC

FTIR spectra showed increased amounts of O-containing functional groups in all three acidified BC conditioners compared with the untreated BC (Figure 6). The PCA of the FTIR spectroscopy indicated that the surface of untreated BC was different from BC +  $\text{H}_2\text{SO}_4$ , BC +  $\text{H}_3\text{PO}_4$  and BC +  $\text{H}_2\text{O}_2$  in the amounts of aromatic, carbonyl, aliphatic, hydroxyl, amine and alcohol groups. The first principal component (PC1) explained the greatest portion of the variance (77.5%) while the second (PC2) contributed 13.2% to the total variance (Figures S2 and S3).

The differences in infrared spectra between acidified BC materials showed also different effects in the studied acids. The asymmetric and symmetric stretching vibrations (amine  $-\text{NH}$  and  $-\text{NH}_2$ ) at  $3340$  and  $3391\text{ cm}^{-1}$  increased the most in BC acidified with  $\text{H}_2\text{SO}_4$ , indicating that the nitro groups ( $-\text{NO}_2$ ) at  $1539\text{ cm}^{-1}$  converted to the corresponding amino group [51]. The intensity of the hydroxyl ( $-\text{OH}$ ) peak at  $3296\text{ cm}^{-1}$  was higher also in BC +  $\text{H}_2\text{SO}_4$  than that of BC +  $\text{H}_3\text{PO}_4$  and BC +  $\text{H}_2\text{O}_2$ , possibly due to a greater degree of

acidification [52]. The asymmetric and symmetric stretching vibrations (aliphatic -C-H and -CH<sub>2</sub>) at 2923 and 2852 cm<sup>-1</sup> increased with acidification probably as a result of the aliphatic side chain oxidation [40,53]. Acidification enhanced the strength of the three acidified BC peaks at 1705 cm<sup>-1</sup> (-C=O stretching in the carboxyl group). The effect on the stretching vibrations of aromatic -C=C at 1515 and 1393 cm<sup>-1</sup> was weaker for BC + H<sub>2</sub>SO<sub>4</sub> and BC + H<sub>3</sub>PO<sub>4</sub> in comparison to BC + H<sub>2</sub>O<sub>2</sub> and untreated BC. Meanwhile, the vibration peak at 1620 cm<sup>-1</sup> (-C=C stretching vibration of alkenyl groups) was highest in BC + H<sub>2</sub>SO<sub>4</sub> and BC + H<sub>2</sub>O<sub>2</sub> [54]. Simultaneously, all three acidified BCs displayed a peak at 1235 cm<sup>-1</sup> that may represent -C-O stretching in carboxylic acid.



**Figure 6.** FTIR spectra of untreated and acidified BC with identified functional groups [55–62]. BC: biochar; BC + H<sub>2</sub>O<sub>2</sub>, BC + H<sub>2</sub>SO<sub>4</sub> and BC + H<sub>3</sub>PO<sub>4</sub> represent BC acidified with the respective acid.

## 4. Discussion

### 4.1. Effect of Untreated BC on $\text{NH}_3\text{-N}$ Emissions

The addition of untreated BCm into digestate caused a reduction of cumulative  $\text{NH}_3\text{-N}$  volatilization after 48 h compared with the control. In contrast, the untreated BCs did not produce a decrease in cumulative  $\text{NH}_3\text{-N}$  emissions relative to control. The suppression effect of BCm was delayed in comparison to BCs, but despite that delay, the cumulative  $\text{NH}_3\text{-N}$  emissions in BCm were not significantly different in comparison to BCs.

The short-term effectiveness (<1.5 h) of untreated BCs is likely to be related to the formation of a physical barrier on the digestate surface. The physical cover of untreated BCs particles prevented the digestate from being exposed to the surrounding environment and slowed down the transfer of  $\text{NH}_3\text{-N}$  gases at the digestate–air interface. The  $\text{NH}_3\text{-N}$  emissions were also slowed down in the control and pure acid treatments due to the digestate crusting. However, the surface crust in these treatments formed later than the BC cover and after a considerable gaseous loss had already occurred. The effectiveness of untreated BCs declined after 1.5 h possibly due to its strong water repellent characteristic. The hydrophobic property prevented the BCs particles from submerging deeper into the digestate, and the resulting BC cover was less resistant to  $\text{NH}_3\text{-N}$  emissions. The dry surface of untreated BCs observed 48 h after the application was an indicator of its durable hydrophobicity. This strong physical property was probably induced by the large quantity of aliphatic functional groups on the BC surface (Figure 6). Previous studies have shown a strong correlation between hydrophobicity and the presence of aliphatic functional groups (–C–H,  $\text{CH}_2$ ) on the BC surface [63,64]. Our findings are in accordance with those of Holly and Larson [41] in which BC, made from wood and maize cob pyrolyzed at 400 °C, effectively mitigated  $\text{NH}_3\text{-N}$  emissions from the dairy manure storage. The effectiveness of BC in their experiment was due to the action of the physical barrier on the manure surface provided by the BC cover thickness of 3.1 and 2.5 cm.

Untreated BC mixing increased the  $\text{NH}_3\text{-N}$  emissions at the very beginning of the experiment. The volatilization occurred probably because of the pH increase in the digestate caused by aeration when the BC was mixed with the digestate. The digestate pH also increased in the control treatment due to a higher concentration of  $\text{OH}^-$ , even though the digestate was not mixed. In the BCs treatment, the  $\text{NH}_3\text{-N}$  emissions and pH under the BC cover were notably lower than in control because the digestate was not in direct contact with ambient air. The reduced effect of the BCs treatment might also be because the relative change of digestate pH only occurred below the BC cover. According to García-González et al. [65], manure aeration stimulates  $\text{OH}^-$  groups release and increases manure pH, which in turn increases the conversion rate of  $\text{NH}_4^+\text{-N}$  to  $\text{NH}_3\text{-N}$ . In the BCm treatment, however, the aeration increased  $\text{NH}_3\text{-N}$  emissions in the short-term (<3.5 h). As the BCm particles started to migrate to the upper part of the vessel, they formed, like the BCs treatment, a physical barrier against  $\text{NH}_3$  loss. However, not all BCm particles floated to the digestate surface after mixing to provide resistance to  $\text{NH}_3\text{-N}$  emissions. The BCm particles located closer to the center of the vessel decreased the digestate pH in the middle of the vessel and diminished the formation of  $\text{NH}_3\text{-N}$ . The pH reduction was furthermore exacerbated by greater contact between BCm particles and digestate.

### 4.2. Effect of Acidification on BC Characteristics and $\text{NH}_3\text{-N}$ Emissions

The physicochemical characteristics of BC that affected  $\text{NH}_3\text{-N}$  emissions were positively influenced by acidification. Compared with the untreated BC, the SSA and number of surface functional groups were greater in acidified BC (Table 2, Figure 6), whereas pH and alkalinity levels were smaller. These findings indicate that the ability of acids to change BC properties is different.

SSA increased the most in BC +  $\text{H}_2\text{O}_2$  and BC +  $\text{H}_2\text{SO}_4$ , whereas the SSA increase was slightly less in BC +  $\text{H}_3\text{PO}_4$ . Maximum peaks of hydroxyl (–OH) and carbonyl (–C=O) functional groups were observed in the BC +  $\text{H}_2\text{SO}_4$ . The peak of aliphatic functional groups was highest in BC +  $\text{H}_3\text{PO}_4$ , but it was also high on the surface of BC +  $\text{H}_2\text{SO}_4$

(Figure 6). The amount of previously mentioned functional groups was a little lower in BC + H<sub>2</sub>O<sub>2</sub>. Vithanage et al. [66] and Cibati et al. [67] showed that BC treatment with H<sub>2</sub>SO<sub>4</sub> increased the O/C ratio twofold, whereas treatment with H<sub>2</sub>O<sub>2</sub> only increased it 1.5-fold. These findings indicate the capacity of different acids to generate O-containing functional groups on the BC surface [68]. The addition of O-H, C=O, C-O and N-H functional groups on the BC surface can decrease its hydrophobicity [69,70]. Shen et al. [71] found that acid treatment of BC with HNO<sub>3</sub> and H<sub>2</sub>SO<sub>4</sub> enhanced the hydrophilic surface of the BC. Recent study also found that acid-treated BC particles sank deeper into the digestate compared with untreated BC when the surface application method was applied, but they could not rise to the digestate surface in the mixed application method. These findings indicate that BC acidification is an effective method to cause the hydrophobic surface of the BC to become slightly more hydrophilic.

Acidification also remarkably reduced the pH and alkalinity of BC. The effect of acid-treated BC on digestate pH was comparable, except for H<sub>2</sub>O<sub>2</sub>, to the respective pure acid applied to the digestate alone. Our results contrast with those of Huff and Lee [72], who found that the pH of BC was only slightly reduced by H<sub>2</sub>O<sub>2</sub> because of the weak ability of this acid to create acidic functional groups on the BC surface.

Acidification improved the effect of BCs to suppress NH<sub>3</sub>-N emissions. As a result of the developed hydrophilic property, acidified BC particles could form a thicker cover on the digestate surface that withstood the pressure exerted by the gases in the digestate for a longer period than untreated BCs. The ability to reduce NH<sub>3</sub>-N emissions of BCs + H<sub>2</sub>SO<sub>4</sub> or BCs + H<sub>3</sub>PO<sub>4</sub> was slightly greater than that of both acids when applied in the pure form in the first 48 h only (Figure 3). BC acidification can also reduce the amount of BC needed to effectively decrease the NH<sub>3</sub>-N emissions. Maurer et al. [35] determined that NH<sub>3</sub> emission reduction from manure resulting from surface-applied untreated BC was attained at 4.56 kg m<sup>-2</sup> but not at 2.28 kg m<sup>-2</sup>. In the present study, the NH<sub>3</sub> emissions from slurry digestate were effectively reduced by applying acidified BC on the digestate surface at about 2.50 kg m<sup>-2</sup>.

However, the capacity of BCm to reduce NH<sub>3</sub>-N emissions was not improved by acidification because most of acidified BCm particles could not float to the digestate surface as they did in the untreated BCm treatment. Although the effect of acidified BCm to influence digestate pH was comparable to that of the pure acids, their impact on NH<sub>3</sub>-N emissions was smaller than that of the pure acids. Such results might be because of the slower capacity of acidified BCm to alter the digestate pH suggested mostly by the great differences in pH and NH<sub>3</sub>-N emissions between acid and acidified BC treatments observed in the early stage of the experiment (<3.5 h) (Table S3, Figure 2A,B). The NH<sub>3</sub>-N emission difference later diminished when digestate pH in acidified BC and acid treatments leveled off. Iriarte-Velasco et al. [73] showed that BC alkalinity was not removed entirely by acidification; BC treatment with H<sub>2</sub>SO<sub>4</sub> and H<sub>3</sub>PO<sub>4</sub> acids induced the formation of solid compounds such as calcium sulfate, calcium phosphate and other salts. The results of the current study show that the effect of acidified BC on digestate pH remained the same as that of pure acid for at least five days. The pH of digestate where acidified BC was added could increase at a certain moment in time due to the salts present in the ash of acidified BC. Thus, further studies are needed to analyze the effect on pH in longer periods.

Petit et al. [74] found that the capacity of BC to adsorb NH<sub>3</sub>-N can be increased through acidification. In the current experiment, the sorption capacity of BC was not studied, but the differences in the adsorbed amount of NH<sub>3</sub>-N among BC treatments can be inferred from the presence of O functional groups. NH<sub>3</sub>-N adsorption onto H<sub>2</sub>SO<sub>4</sub>-treated BC was markedly more effective than onto untreated BC because of the increased amount of O functional groups, particularly sulfonic groups (-SO<sub>3</sub>H) in the BC samples subjected to chemical treatment. Ro et al. [75] showed that H<sub>3</sub>PO<sub>4</sub> acid activation greatly increased the NH<sub>3</sub> adsorption capacity of BC. They suggested that exceptionally high NH<sub>3</sub> adsorption capacity was generated by H<sub>3</sub>PO<sub>4</sub> acid bound to the acid-activated BC.

Our findings indicate that acidification can alter BC physicochemical features, which may increase the suppression effect of BC on  $\text{NH}_3$  emissions.

#### 4.3. Effect of Conditioner Treatments on N Concentration in Digestate

The  $\text{N}_{\text{tot}}$  concentration in digestate after 30 days was not significantly correlated with  $\text{NH}_3$ -N emissions recorded in the first 48 h and between the 7th and 8th days. In addition to  $\text{NH}_3$ -N emissions, the  $\text{N}_{\text{tot}}$  concentration could be affected by other N-containing gas emissions. Previously, Chadwick et al. [76] showed that the crust or permeable cover formed on the slurry surface during the storage period reduced  $\text{NH}_3$  but increased  $\text{N}_2\text{O}$  emissions. In addition, in our experiment, the BC cover might have stimulated  $\text{N}_2\text{O}$  emissions because of the anaerobic conditions.

The conditioners used in the current experiment affected digestate pH and probably also gaseous exchange at the liquid–air interface by the cover formed on the digestate surface. The pH affects the equilibrium between aqueous (aq)  $\text{NH}_4^+$  ions and (aq)  $\text{NH}_3$  in manure [25]. As digestate pH increases, the concomitant increase of  $\text{NH}_3$  concentration may inhibit the activity of nitrifying bacteria. As a result,  $\text{NH}_4^+$  is converted into  $\text{N}_2\text{O}$  and dinitrogen ( $\text{N}_2$ ) gases bypassing the  $\text{NO}_3^-$ -N phase [77].

In the BCs treatments,  $\text{N}_2\text{O}$  emissions were probably not the main factor influencing the  $\text{NO}_3^-$ -N concentration in digestate. The  $\text{NO}_3^-$ -N concentration was higher in acidified BCs than in untreated BCs, even though a longer-lasting acidified BCs cover and slightly lower digestate pH should promote denitrifying bacteria activity [76,78,79]. It is possible that  $\text{NO}_3^-$ -N was adsorbed by acid-treated BC, and its degradation by microorganisms became more difficult. Lan et al. [80] noted that BC adsorption of  $\text{NO}_3^-$  in soil can decrease its availability for denitrifiers. The  $\text{NO}_3^-$ -N concentration between acidified BCs and BCm with  $\text{H}_2\text{SO}_4$  or  $\text{H}_2\text{O}_2$  was not different, although the cover was not formed in acidified BCm.  $\text{NO}_3^-$ -N sorption could be promoted by surface basic functional groups such as primary and secondary amine groups (Figure 6) via electrostatic interactions. This is in accordance with the findings of Wu et al. [81] that  $\text{NO}_3^-$  can interact with amine groups through electrostatic interactions during the adsorption process. In BC +  $\text{H}_3\text{PO}_4$ , the  $\text{NO}_3^-$ -N could be limited because of the presence of competitive phosphate anions ( $\text{PO}_4^{3-}$ ) bound by sorption sites during acidification, which could remain immobilized because of their highly negative charge. A small amount of  $\text{NO}_3^-$ -N could be adsorbed to positively charged cations ( $\text{K}^+$ ,  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$ ) present in the ash of acidified BC by bridge bonding. Fidel et al. [82] mentioned that some  $\text{NO}_3^-$ -N sorption to acidified BC can occur via cation bridging. However,  $\text{NO}_3^-$ -N could not be bound by untreated BC primarily because of the lower peaks of basic functional groups. Alsewaileh et al. [83] found that low-temperature BC (300 °C) exhibited minimal adsorption efficiency of  $\text{NO}_3^-$ -N because of the reduced total basicity (surface basic functional groups) and limited surface area.

## 5. Conclusions

Of the eleven treatments compared in the experiment,  $\text{H}_2\text{SO}_4$  and  $\text{H}_3\text{PO}_4$  acids, untreated BCm and acidified BCs treatments were found in reducing  $\text{NH}_3$ -N emissions from digestate. The  $\text{NH}_3$ -N emissions were reduced to a similar extent by all these treatments even though the suppression mechanism was different. Acids reduced the digestate pH and reduced the rate of conversion from  $\text{NH}_4^+$ -N to  $\text{NH}_3$ -N. Untreated BCm formed a physical barrier on the digestate surface that isolated the digestate from the atmosphere.

Acidification of BC increased its specific surface area and number of O-containing surface functional groups and decreased the pH, alkalinity and the hydrophobic property. The impact on  $\text{NH}_3$ -N emissions of acidified BC was dependent on the application method. Compared with untreated BC, the ability of BC to reduce  $\text{NH}_3$ -N emissions was greater when it was acidified with  $\text{H}_2\text{SO}_4$  and applied to the digestate surface because of the reduced hydrophobic property of BC particles that sank deeper into the digestate. The effect on digestate pH of acidified BCm was comparable, except for  $\text{H}_2\text{O}_2$ , to the respective pure acids applied individually. However, the reduction of pH of the digestate occurred

later in time, and thus the impact on  $\text{NH}_3\text{-N}$  emissions was weaker. The effect was also less strong compared to untreated BCm because most of the acidified BCm particles could not float to the digestate surface and form a physical cover. The novel finding of our study is that acidified BC applied on digestate surface could have an effective application potential to reduce  $\text{NH}_3$  emissions from slurry storage tanks.

These results suggest that BC cover might stimulate  $\text{N}_2\text{O}$  emissions because of the anaerobic conditions. In addition, BC +  $\text{H}_2\text{SO}_4$  and BC +  $\text{H}_2\text{O}_2$  might adsorb  $\text{NO}_3^-$ -N present in digestate and decrease its availability to denitrifying bacteria. These new hypotheses need to be tested in future research.

**Supplementary Materials:** The following are available online at <https://www.mdpi.com/2071-1050/13/2/837/s1>, Figure S1: Cumulative  $\text{NH}_3\text{-N}$  emissions from the 7th to 8th day (mean  $\pm$  standard error;  $n = 3$ ). Results marked with similar lowercase letters are not significantly different. Control is digestate without conditioner;  $\text{H}_2\text{O}_2$ ,  $\text{H}_2\text{SO}_4$  and  $\text{H}_3\text{PO}_4$  represent pure-form acids applied mixed into the digestate; BC is biochar; BC +  $\text{H}_2\text{O}_2$ , BC +  $\text{H}_2\text{SO}_4$  and BC +  $\text{H}_3\text{PO}_4$  represent BC acidified with the respective acid; BCm indicates that the BC was mixed with the digestate and BCs indicates that the BC was applied to the digestate surface. Figure S2: PCA1 and PCA2 of FTIR spectra for untreated and acidified biochar (BC) amendments. BC +  $\text{H}_2\text{O}_2$ , BC +  $\text{H}_2\text{SO}_4$  and BC +  $\text{H}_3\text{PO}_4$  represent BC acidified with the respective acid. Figure S3: Eigenvectors and wavenumber values for principal components 1 and 2 (PC1 and PC2, respectively) of the FTIR spectra. Table S1:  $\text{NH}_3\text{-N}$  emissions during 24 h on the 7th day after conditioner application as the effect of conditioner, application method and time passed since conditioner application (two-way ANOVA analysis;  $n = 3$ ). Asterisks indicate the level of significance. \*\*\*\*  $p < 0.0001$ ; \*\*\*  $p < 0.001$ ; \*\*  $p < 0.01$ ; \*  $p < 0.05$ . A lack of \* symbols indicates significance at  $p < 0.1$ . Table S2: Digestate pH during the first 5 days as an effect of conditioner, application method and time passed since conditioner application (two-way ANOVA analysis;  $n = 3$ ). Asterisks indicate the level of significance. \*\*\*\*  $p < 0.0001$ ; \*\*\*  $p < 0.001$ ; \*\*  $p < 0.01$ ; \*  $p < 0.05$ . A lack of \* symbols indicates significance at  $p < 0.1$ . Table S3: Digestate pH during the first 5 days (mean  $\pm$  standard error,  $n = 3$ ).

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