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Water Hyacinth Biochar: A Sustainable Approach for Enhancing Soil Resistance to Acidification Stress and Nutrient Dynamics in an Acidic Nitisol of the Northwest Highlands of Ethiopia

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Abstract: Soil acidity impacted over 43% of Ethiopia's arable land and debilitated agricultural productivity. Due to reacidification susceptibility, high costs, and inadequate availability of lime, biochar has emerged as an alternative soil acidity ameliorator. However, biochar application, particularly from invasive plants such as water hyacinth, as a soil acidity amendment and waste management strategy has not fully expanded in Ethiopia. Therefore, this study investigated the potential of water hyacinth biochar (WHB) to enhance soil resistance to acidification stresses and nutrient dynamics in an acidic Nitisol of the northwest highlands of Ethiopia. An incubation study was conducted using nine treatments viz. control (soil only), biochar produced using a furnace (WHB_f), and a grounding method (WHB_g) each at 1% and 2% application rates, lime (L), fertilizer (F), 2% WHB_f combined with fertilizer ($2WHB_fF$), and lime combined with fertilizer (LF). Soil samples, except the control, underwent simulated acidification with HNO_3. The results showed that WHB decelerated nitrification, reduced H^+ ions released into the soil, and enhanced available phosphorus and nitrogen dynamics. After incubation, 1% and 2% WHB applications increased soil pH in the range of 0.30–0.35 and 0.72–0.86 units, respectively, compared to the limed soil. Conversely, exchangeable acidity decreased by 26.5% to 28.8% and 58.4% to 63%, respectively. The $2WHB_fF$ treatment led to soil pH increases of 0.71 and 0.90 units, with exchangeable acidity reductions of 49.8% and 64.7% compared to the LF and F treatments, respectively. Compared to lime, WHB treatments demonstrated more effective resistance against soil acidification from nitrification and simulated acidification with HNO_3. Therefore, WHB can be used as a sustainable approach to increase soil resistance against various acidification stresses while aiding in soil nutrient management. The study's findings can offer valuable insights to change environmental challenges into sustainable soil acidity management approaches.

Keywords: soil acidity stress; Lake Tana; water hyacinth biochar; lime; nitrification rate; proton release

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1. Introduction

Soil acidity impacted more than 50% of the world's cultivable lands and over 22% of Africa's landmass [1]. In Ethiopia, soil acidity and aluminum (Al) toxicity affected over 43% of the arable land, with 13% being highly acidic and 28% being moderately acidic [2]. Soil acidity stems from parent materials, acid rain, organic matter decomposition, and ammonium nitrification [3]. Natural soil acidification occurs gradually [4]. Conversely, anthropogenic causes such as an excessive application of NH_4^+-based fertilizers rapidly escalate soil acidity [5,6]. Nitrogen (N) fertilizers in ammonium form (NH_4^+-N) contribute to soil acidification with N cycling processes releasing 20 to 221 kmol H^+ ha^{-1} per year [4]. Dong et al. [7] stated that more than 68% of the acidic protons in agricultural soil originate from the nitrification of NH_4^+

into nitrate (NO_3^-). Additionally, agrochemicals such as herbicides and pesticides can induce soil acidity. For instance, in Sinana Woreda, Southeastern Ethiopia, applying glyphosate as a weed killer decreased soil pH by more than one unit during a growing season [8].

Soil acidity induces complex challenges to plant growth by impacting the soil's physical, chemical, and biological qualities. The deficit of important plant nutrients such as phosphorus (P), calcium (Ca), and magnesium (Mg), and the venomousness of Al and manganese (Mn), are the major reasons that make acidic soils unsuitable for crop cultivation [9,10]. Soil acidity destroys soil microbial activities, organic matter decomposition, nutrient captivation and utilization by plants, and it eventually depresses crop production unless proper management is taken [6]. Therefore, soil acidity needs to be addressed for better crop production.

Lime has been widely used as a soil acidity amendment. However, it was not effective in addressing soil acidity problems due to its reacidification susceptibility, inadequate transportation infrastructure, high costs for current farmers, and inadequate availability. Soil with pH below 4.5 may require lime application rates exceeding 0.8% per soil weight [11,12]. In Ethiopia, efforts to tackle soil acidity have mainly relied on lime application. However, this approach lacks long-term sustainability and economic feasibility, resulting in its limited success nationwide [13]. Thus, there is a need for alternative organic soil amendments to ameliorate soil acidity sustainably.

Biochar has emerged as an eco-friendly alternative soil conditioner to counter soil acidity [14]. Due to its high porosity, a wide range of phenolic and carboxylic compounds [15], as well as a high cation exchange capacity (CEC) [16], biochar can improve soil physicochemical properties, advance macro- and micro-nutrient retention [17], decrease exchangeable acidity [18], facilitate pesticide deactivation and biodegradation [19], enhance microbial activity and enzymatic processes in the soil [16], and boost crop productivity [14]. Biochar mitigates Al toxicity by binding it with organic hydroxyl and carboxyl groups or through external adsorption and co-precipitation as $SiAlO_8$ [20]. The nature of raw materials and preparation techniques significantly influence the physical and chemical qualities of biochar. Therefore, the selection of sustainable raw materials and appropriate preparation methods is very important. Research findings indicated improvement in soil properties and nutrient cycling using biochar produced from invasive plants [21]. Consequently, biochar production from invasive plants has emerged as an effective approach to mitigate ecological harm from biological invasion, offering waste management approaches and a cost-effective alternative to other feedstocks [22].

Water hyacinth (*Eichhornia crassipes*), which has been invading Ethiopia's largest lake, Lake Tana, since 2011, is among the most invasive aquatic plants found globally [23]. The physical, mechanical, chemical, and biological removal methods have been ineffective due to its rapid expansion surpassing removal rates [24]. Consequently, the overabundance of water hyacinth remains a pressing challenge, emphasizing the necessity for alternative methods to control the weed invasion. Water hyacinth biomass, with its abundant lignocellulosic composition, can be used as a sustainable feedstock for biochar production [25]. The conversion of water hyacinth into biochar represents a feasible solution for handling this invasive weed, contributing to sustainable waste management [26]. Moreover, the improvement of soil quality through biochar application promotes sustainable agriculture and contributes to a circular bioeconomy.

However, there are very few studies reporting on the potential of water hyacinth biochar (WHB) to alleviate soil acidity, improve soil nutrients, and enhance sustainable crop production, particularly in acidic soils of Ethiopian highlands. Additionally, the reported studies [8,27–34] regarding biochar's effects on soil acidification resistance lack clarity and contain gaps that necessitate further investigation. Former soil acidification studies were conducted after completing the incubation period. This can lead to a misinterpretation that soil acidification stresses occur post-harvest, as the incubation period primarily reflects the crop cultivation time. Furthermore, the reported soil acidification studies typically considered only one soil acidifying factor, such as chemical fertilizer, agrochemical, or acid rain. Therefore, studies that involve fertilizer as one soil acidifying factor and additional

soil acidification to mimic other soil acidity stresses would provide a more comprehensive understanding. Moreover, the split application of fertilizer (urea), which is typically applied in two or three splits, leading to different acidification stress levels in the soil, is still lacking. It is, therefore, very necessary to conduct further research to address the aforementioned gaps and investigate the effectiveness of biochar in resisting different soil acidity stresses.

Therefore, this study aimed to investigate the potential of water hyacinth biochar in enhancing soil resistance to acidification stress and nutrient dynamics in an acidic Nitisol of the northwest highlands of Ethiopia by hypothesizing that WHB displays greater resistance to soil acidification stress compared to lime.

The findings of this research can indeed represent a valuable strategy to deal with both the problem of the acidification of arable land and the disposal of invading plant biomass removed from waterways. The results may shed light on the benefits of using water hyacinth biochar to fortify soil against acidification stress from chemical fertilizers and agrochemicals. Ultimately, it can promote sustainable and productive agricultural practices with distinct environmental, economic, and social advantages.

2. Materials and Methods

2.1. Soil Sampling and Analyses

Soil samples (Red Nitisols), with basic physicochemical properties presented in Table 1, were collected from the surface layer (0–20 cm depth) from the Koga irrigation project site (11°10′ and 11°25′ N, and 37°02′ and 37°17′ E) in Northwestern Ethiopia. The samples were composited, air-dried, crushed, and sieved to <2 mm size and analyzed for selected soil physicochemical parameters.

Table 1. Basic physicochemical properties of the experimental soil and water hyacinth biochars.

Property	Soil	WHB_f	WHB_g
Sand (%)	10.2	—	—
Silt (%)	28.3	—	—
Clay (%)	61.5	—	—
Texture	Clay	—	—
Moisture content (%)	2.78 ± 0.1	4.64 ± 0.2	12.4 ± 0.1
Ash (%)	—	17.4 ± 0.0	23.2 ± 0.0
Volatile matter (%)	—	38.6 ± 0.0	59.7 ± 0.0
Fixed carbon (%)	—	44.0 ± 0.0	17.1 ± 0.0
Bulk density (g cm^{-3})	1.27	—	—
pH	4.85 ± 0.0	10.9 ± 0.0	10.4 ± 0.0
Liming potential (%$CaCO_3$ equivalent)	—	24.7 ± 0.1	20.4 ± 0.4
Exchangeable acidity (cmol (+) kg^{-1})	2.93 ± 0.2	—	—
Exchangeable Al^{3+} (cmol (+) kg^{-1})	1.52 ± 0.1	—	—
Exchangeable H^+ (cmol (+) kg^{-1})	1.41 ± 0.1	—	—
Electrical conductivity (µS cm^{-1})	42.1 ± 1.08	4.13×10^3 ± 0.01	2.77×10^4 ± 0.45
Cation exchange capacity (cmol (+) kg^{-1})	16.9 ± 0.5	42.6 ± 1.1	34.2 ± 0.2
Acid saturation (%)	17.3 ± 0.3	—	—
Total C (%)	2.00 ± 0.1	55.9 ± 1.3	34.8 ± 0.6
Total H (%)	2.08 ± 0.2	2.01 ± 0.4	0.700 ± 0.1
Total N (%)	0.180 ± 0.0	1.46 ± 0.0	0.720 ± 0.0
Total O (%)	—	23.3 ± 1.6	40.6 ± 0.6
C/N	11.1 ± 0.5	38.3 ± 0.9	48.6 ± 1.9
NH_4^+–N (mg kg^{-1})	10.7 ± 0.6	2.03 ± 0.2	0.760 ± 0.1
NO_3^-–N (mg kg^{-1})	10.3 ± 0.4	1.94 ± 0.0	2.54 ± 0.3
Available phosphorus (mg kg^{-1})	3.74 ± 0.26	4.44×10^3 ± 564	2.29×10^3 ± 73
Organic C (%)	1.76 ± 0.0	25.9 ± 0.1	21.6 ± 0.1
Organic matter (%)	3.02 ± 0.0	44.7 ± 0.1	37.3 ± 0.1
BET surface area (m^2 g^{-1})	—	61.4 ± 0.0	14.7± 0.0
Total pore volume (cm^3 g^{-1})	—	0.0700 ± 0.0	0.0400 ± 0.0
Average pore width (nm)	—	3.87 ± 0.0	9.77 ± 0.0
Phenolic content (mmol g^{-1})	—	1.80 ± 0.1	1.62 ± 0.0
Lactonic (mmol g^{-1})	—	0.400 ± 0.1	0.630 ± 0.1
Carboxylic (mmol g^{-1})	—	0.930 ± 0.0	0.800 ± 0.1

WHB_f: water hyacinth biochar produced using furnace; WHB_g: water hyacinth biochar produced by grounding method. Numbers after the symbol "±" are the standard deviations of triplicate measurements.

Soil texture was determined by the bouyoucous hydrometer method [35]. Bulk density (BD) was measured by the core sampler method [36]. pH and electrical conductivity (EC) were determined according to Robinson [37] using a pH meter (LAQUA F-71, Horiba Scientific, Kyoto, Japan) and EC meter (Mettler–Toledo, International Inc., Toledo, OH, USA), respectively. Exchangeable acidity (H^+ and Al^{3+}) was extracted with 1.0 mol L^{-1} KCl and titrated with 0.02 mol L^{-1} NaOH to pH 7.0 [38]. Cation exchange capacity (CEC) was determined by the ammonium acetate method using 1.0 mol L^{-1} KCl extraction [39]. Organic carbon (OC) and organic matter (OM) were determined by the Walkley–Black method [40]. Available phosphorus (P) was determined by Mehlich-3 extraction [41] using an autoanalyzer (FIAlyzer-1000, FIAlab Instruments, Inc., Seattle, WA, USA). Ammonium–nitrogen (NH_4^+–N) and nitrate–nitrogen (NO_3^-–N) were determined by a 2 mol L^{-1} KCl extraction method [42] using an autoanalyzer. The total C, H, and N were analyzed by the dry combustion method [43] using an elemental analyzer (Perkin Elmer, 2400 Series II, Waltham, MA, USA).

2.2. Biochar Preparation and Characterization

The biochar was produced from water hyacinth (*Eichhornia crassipes*) collected from Lake Tana, Ethiopia (10°45′54.1″ N, 36°10′24.9″ E, and 12°50′15.9″ N, 38°50′54.48″ E) in March 2023. Water hyacinth weeds were cleaned with distilled water, chopped into small pieces (3–5 cm), and dried in an oven for 48 h at 75 °C. The water content of the collected water hyacinth biomass was 94%. The dried water hyacinth biomass was then put into a cylindrical aluminum container containing four holes (3–4 mm diameter each) to allow a minimum gas release. It was then pyrolyzed in a muffle furnace (LT 40/12, Nabertherm GmbH, Lilienthal, Germany) at 450 °C for 2 h with a heating rate of 10 °C per min, resulting in the biochar designated as WHB_f. The other biochar type (WHB_g) was prepared by a grounding system, which is feasible for farmers who could not access electric furnace. To prepare the WHB_g, water hyacinth biomass was harvested from Lake Tana and subsequently sun-dried. A stack of cleaned and sun-dried water hyacinth biomass was arranged on a pristine ground surface. It was covered with teff (*Eragrostis tef*) straws and a layer of soil to prevent the ingress of oxygen. The progress of biomass conversion into biochar was consistently monitored throughout the preparation process, which took about 1 h from ignition of biomass and extinction of produced biochar. This grounding method does not permit us to know the actual pyrolysis temperature. After producing biochar, soil and residual teff straws were carefully removed to prevent inadvertent mixing with the biochar. Water was gently sprayed to cool down the biochar, which was then dried, packed, and transported to the laboratory, where it underwent the same subsequent processes as WHB_f. Both types of biochars were pulverized and sieved to <0.5 mm.

The potential of WHB_f produced using an electric furnace was evaluated for its effectiveness in resisting soil acidity stresses and enhancing soil nutrient dynamics because this method can be used to produce less volume of biochar, which can be used for pot experiments that require smaller quantities. On the other hand, the WHB_g's effectiveness in resisting soil acidity stresses and enhancing soil nutrient dynamics was evaluated in comparison with WHB_f, considering its suitability for field applications that require significant amounts of biochar. It is possible to produce large volumes of biochar at once on the ground employing this method.

The moisture content of the biochars was determined using the American Society for Testing Materials methodology [44]. The ash content, volatile matter (VM), and fixed carbon (FC) were determined by thermal gravimetric analysis (TGA) using simultaneous differential thermogravimetry (SDT Q600, TA Instruments, Lukens Drive, New Castle, DE, USA). The pH and EC were determined according to Singh et al. [45]. The liming potential was determined according to Rayment and Higginson [42]. The CEC was determined by the ammonium acetate method using 2.0 mol L^{-1} KCl extraction [39]. The OC and OM were determined by the Walkley and Black method [40]. The total C, H, and N were analyzed by the dry combustion method [43] employing an elemental analyzer (Perkin Elmer,

2400 series II, Waltham, MA, USA), and O was calculated by difference [46]. Available P was determined by a 2% formic acid extraction method [47], using a flow injection autoanalyzer (FIAlyzer-1000, FIAlab Instruments, Inc., Seattle, WA, USA). NH_4^+−N and NO_3^-–N were determined by a 2 mol L^{-1} KCl extraction method [42].

The surface functional groups were analyzed using a Fourier Transform Infrared Radiation (FTIR) spectrophotometer (IRAffinity-1S FTIR, Shimadzu, Kyoto, Japan) in 13 mm KBr pellets (1:100, *w*/*w*), pressed with a pressure of 7845 kPa, using a spectral resolution of 2 cm^{-1} and 64 scans in a spectral range of 4000–400 cm^{-1} [48]. The quantity of surface functional groups (phenolic, lactonic, and carboxylic) in the WHBs was estimated using the Boehm titration method [49]. The surface morphologies of the biochar samples were measured by mounting the biochar powders (<0.5 mm) onto aluminum stubs with conductive carbon double tape followed by structure imaging using a scanning electron microscope (SEM) (JSM-5600LV, USA). The specific surface area and porosity of the biochars were determined by Brunauer, Emmet, and Teller (BET) method [50] via N adsorption–desorption isotherms at 77 K, using accelerated surface area and porosimetery (ASAP) system (ASAP 2010, Micrometric, Lincoln, UK).

2.3. Incubation Experiment

A total of nine treatments, each in triplicate, were arranged in a completely randomized design (CRD), resulting in 27 samples (Table 2). The treatments were: control (soil only), two biochar types (WHB_f and WHB_g) each at 1% and 2% application rates represented as $1WHB_f$, $2WHB_f$, $1WHB_g$, and $2WHB_g$, respectively, one rate of lime (L) (3.72 t ha^{-1} in the form of powder $CaCO_3$ with a liming potential of 96.9% $CaCO_3$ equivalent), chemical fertilizer (F) (180 kg ha^{-1} of N in the form of urea and 138 kg ha^{-1} of P_2O_5 in the form of NPS, which contains N in ammonium form, P in phosphate form, and S in sulfate form), 2% WHB_f combined with chemical fertilizer ($2WHB_fF$), and lime combined with chemical fertilizer (LF).

Table 2. The details of the experimental treatments.

Abbreviation	Treatment Details
Control	No biochar + no lime + no fertilizer (soil only)
$1WHB_f$	WHB_f 1% + no lime + no fertilizer
$2WHB_f$	WHB_f 2% + no lime + no fertilizer
$1WHB_g$	WHB_g 1% + no lime + no fertilizer
$2WHB_g$	WHB_g 2% + no lime + no fertilizer
$2WHB_fF$	WHB_f 2% + no lime + fertilizer
LF	No biochar + lime + fertilizer
F	No biochar + no lime + fertilizer
L	No biochar + lime + no fertilizer

WHB_f: water hyacinth biochar produced using an electric furnace; WHB_g: water hyacinth biochar produced by grounding method.

Biochar application rates of 1% and 2% were used because 1%-to-2% rates are typical for field biochar application for crop production. The lime rate was determined based on the exchangeable acidity of the experimental soil (Equation (1)). Thus, the soil acidity resistance efficiency of WHB at common field biochar application rates was evaluated compared to lime at a rate based on the exchangeable acidity content of the experimental soil. Based on the biochar characterization results, WHB_f exhibited better liming potential with higher pH, greater surface area, and CEC compared to WHB_g (Table 1). Consequently, a treatment involving a higher application rate (2%) of WHB_f combined with fertilizer ($2WHB_fF$) was included to evaluate the potential of water hyacinth biochar in mitigating soil acidification from the applied fertilizers. This helps in estimating the extent to which WHB can slow down the nitrification of NH_4^+ from the applied fertilizer and reduce the rate and amount of H^+ ions released into soil. Fertilizer (F) was included in the experimental treatments because fertilizers are the primary soil acidity stressors, and lime and biochar

were used to assess their effectiveness in alleviating the acidity stress induced by these fertilizers.

The lime-requirement (LR) rate was determined using the following lime-rate-determining equation (Equation (1)) based on the exchangeable acidity of the experimental soil [51].

$$\mathrm{LR}\left(\mathrm{CaCO_3\ kg\ ha^{-1}}\right) = \frac{\mathrm{EA} * \mathrm{BD} * \mathrm{depth} * 10^4 \mathrm{m}^2 * 1000}{2000} \quad (1)$$

where EA is the exchangeable acidity of the experimental soil (2.93 cmol (+) kg^{-1}), BD is soil bulk density (1.27 g cm^{-3}), and depth = 0.2 m. Substituting the values results in 3721 kg ha^{-1}, or 3.72 t ha^{-1} of lime. The amount of chemical fertilizer was calculated based on the recommended fertilizer rate for maize production (a common crop in the soil sampling area) [52].

Before treatment application, 500 g of soil (<2 mm) was added into a 1000 mL plastic bottle, maintaining the moisture content at 55% of the soil field capacity. The bottles were covered by parafilm-containing holes to avoid moisture loss and permit gas exchange. The samples were then pre-incubated for 2 wk at the temperature of 30 ±1 °C to stabilize the soil microbes. Water loss was adjusted every 3 d by spraying deionized water. After two weeks, Day 0 samples (samples before treatment application) were taken and treatments were applied immediately. The basal dose of NPS fertilizer and half of the urea were applied just after Day 0 sampling, while the remaining half of urea was applied on Day 40 to mimic the split application of urea fertilizer.

On Days 35 and 63 of the incubation period, soil samples (except the control) were acidified with 30.7 and 18.425 mL of 260 mmol L^{-1} HNO_3, respectively. This was done to simulate soil acidity stressors such as herbicides, pesticides, and acid rain (concentration of HNO_3 was determined by a preliminary experiment). To determine the concentration of HNO_3 used for simulated soil acidification, a preliminary incubation experiment was conducted. In the preliminary experiment, a 2% application of WHB_f increased soil pH by 1.1 units. Based on this soil pH rising capacity of the WHB_f, we acidified non-amended soil samples using different concentrations of HNO_3 (1, 2.5, 5, and 200 mM) to determine the concentration of HNO_3 that can decrease soil pH by at least one unit. In this case, 200 mM of HNO_3 decreased soil pH by 0.65 units, while 5 mM decreased it by 0.20 units. Based on this preliminary experiment, the quantity of acid required to decrease soil pH by at least one unit was applied to evaluate the WHB's capacity to resist soil acidity stresses. Therefore, soil acidification with HNO_3 to decrease soil pH by at least one unit was based on (1) the WHB's capacity, which displayed 1.1 unit soil pH rise during the preliminary experiment, and (2) literature reports [8,34] which stated that soil acidity stresses, such as herbicides, and acid rain, can decrease soil pH by at least one unit during a cropping season. Soil acidification after 5 weeks (Day 35) suggests that for optimal soil pH, the recommended incubation period for WHB and lime should be 1 month before planting. Moreover, the design aimed to simulate soil acidity stresses caused by weed-control agrochemicals, typically applied 35–40 days after planting. The second soil acidification on Day 63 was intended to simulate soil acidity stresses resulting from the use of agrochemicals for weed, pest, and insect control, as well as from the third split application of urea. The control sample remained non-acidified to evaluate the extent to which WHB could resist soil acidity stresses, thereby maintaining soil pH at a comparable level to that of the non-acidified control.

HNO_3 was preferred for soil acidification, rather than acids such as HCl and H_2SO_4, due mainly to (1) when HNO_3 and H_2SO_4 enter the soil, the mechanism of adsorption between NO_3^- and SO_4^{2-} is different. NO_3^- is adsorbed only through electrostatic attraction, while SO_4^{2-} is adsorbed through electrostatic attraction and specific adsorption [53]. During this specific SO_4^{2-} adsorption, the OH^- will be released through a ligand exchange, and it can neutralize part of the acid, resulting in less acidification effect of the acid. The more the amount of SO_4^{2-} adsorption, the more the amount of OH^- is released; and (3) SO_4^{2-} from H_2SO_4 can form sparingly soluble $Al_2(SO_4)_3$ precipitates and can affect exchangeable Al^{3+} results.

Soil sampling was done on Days 0, 3, 7, 14, 28, 42, 56, 70, and 91 of the incubation period. These soil samples were dried overnight at 45 °C and analyzed for pH, exchangeable acidity (Al^{3+}, H^+), and available P. The NH_4^+-N and NO_3^-–N were analyzed using fresh soil equivalent to 2.0 g of dry soil.

The net nitrification rate and net proton (H^+) production from nitrification were determined by Equations (2) and (3), respectively [29].

$$\text{Net nitrification rate } \left(\text{mg kg}^{-1}\text{ d}^{-1}\right) = \frac{\Delta NO_3^-\text{–N}}{\text{t}} \quad (2)$$

where ΔNO_3^--N is the changes in NO_3^--N concentration (mg kg^{-1}) between Day 0 and each sampling date; t is the number of days between Day 0 and other sampling dates.

$$\text{Net } H^+ \text{ production } \left(\text{mmol } H^+ \text{ kg}^{-1}\right) = \frac{\Delta NO_3^-\text{–N} - \Delta NH_4^+\text{–N}}{14} \quad (3)$$

where $\Delta NO_3^- - N$ and $\Delta NH_4^+ - N$ are the changes of $NO_3^- - N$ and $NH_4^+ - N$ concentrations (mg kg^{-1}) before and after incubation, respectively; 14 is the atomic mass of N.

To avoid the impact of HNO_3 addition on the $NO_3^- - N$ concentration in the treatments, we deducted the added quantity of $NO_3^- - N$ from the values obtained after soil acidification. Thus, we deducted 446.992 mg kg^{-1} (amount added during the first soil acidification) from Days 42 and 56 soil $NO_3^- - N$ contents, and 894.105 mg kg^{-1} (amount added during the 1st and 2nd soil acidification) from Days 70 and 91 soil $NO_3^- - N$ contents.

2.4. Statistical Analysis

The software statistical package for social science (SPSS version 22, IBM Inc., Armonk, NY, USA) was used for statistical analysis of data. A one-way analysis of variance (ANOVA) was used to test the significant differences in soil pH, exchangeable acidity, exchangeable Al^{3+}, $NO_3^- - N$, $NH_4^+ - N$, and available P contents among the treatments within each soil sampling date. Significant effects of the applied treatments were determined using the least significant difference (LSD) with Tukey post hoc multiple comparisons test [54] at $p < 0.05$, following the one-way ANOVA. Pearson correlation analysis was done for soil exchangeable acidity and soil pH to see how soil exchangeable acidity in different treatments varies with a unit variation of soil pH. Microsoft Excel 2019 was utilized for manipulating analytical data and preparing graphs. Origin software (OriginPro 2018) was used to plot the FTIR curve of the surface functional groups in the WHBs. Data are displayed as mean ± SD (standard deviation) of triplicate measurements.

3. Results

3.1. Basic Physicochemical Properties of the Experimental Soil and Water Hyacinth Biochars

The experimental soil was a very strong acidic Nitisol [55] categorized under clay soil texture (Table 1). It exhibited a pH of 4.85, exchangeable acidity of 2.93 cmol (+) kg^{-1}, and CEC of 16.9 cmol (+) kg^{-1}. The available P, $NH_4^+ - N$, and $NO_3^- - N$ levels were 3.74, 10.7, and 10.3 mg kg^{-1}, respectively.

The WHB_f, with higher pH (10.9) and CEC (42.6 cmol (+) kg^{-1}), displayed better liming potential (24.7% $CaCO_3$ equivalent) than the WHB_g, which had a lower pH (10.4), CEC (34.2 cmol (+) kg^{-1}), and liming potential (20.4% $CaCO_3$ equivalent) (Table 1). The WHB_f also showed a higher BET surface area, total pore volume, and pore width compared to the WHB_g, which had higher ash and volatile matter contents.

The surface functional group spectra of the WHB_f and the WHB_g are presented in Figure 1. The broad spectral bands around 3421 cm^{-1} for both types of biochar correspond to the hydroxyl (O–H) stretching of alcohol functional groups [56]. The small band around 2366 cm^{-1} (only for WHB_f) might be attributed to the C≡N stretching vibration of the nitrile groups. The absence of this band in WHB_g corresponds with the low total N content in this biochar. The smaller bands around 1795 cm^{-1} for both types of biochar could be attributed to the C=O stretching. The bands around 1602 cm^{-1} could be attributed to the

C=C stretching of an aromatic group or vibration of aromatic groups overlapped with the C=O asymmetric stretching of the carboxylate group, while the band around 1450 cm^{-1} could be associated with the C=O stretching of carbonate overlapped with symmetric stretching of carboxylate groups [57,58]. The peak around 1051 cm^{-1} for WHB_g, which nearly disappeared for WHB_f, might be credited to the symmetric C–O stretching of alcohol and ether groups [59]. A very small appearance of this band for WHB_f agrees with the low O level in this biochar. The spectral bands around 881 cm^{-1} could be attributed to the C–C bending and the C–H out-of-plane bending of an aromatic group, while the smaller bands between 775–621 cm^{-1} could be ascribed to the C–H out-of-plane bending of an aromatic/polynuclear aromatics [59].

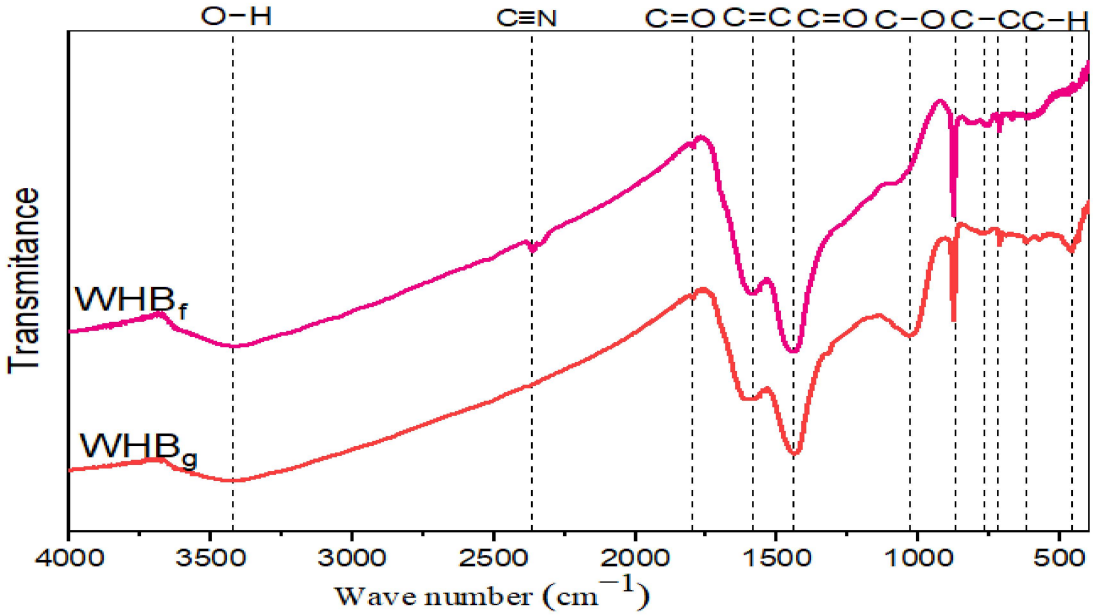

Figure 1. The FTIR bands illustrating surface functional groups in the two types of experimental biochar. (WHB_f: water hyacinth biochar produced using furnace; WHB_g: water hyacinth biochar produced by grounding method).

The relatively lower concentration of phenolic and carboxylic surface functional groups in WHB_g (1.62 and 0.80 mmol g^{-1}, respectively) compared to WHB_f (1.80, and 0.93 mmol g^{-1}, respectively) (Table 1) could be attributed to the entrance of oxygen during the preparation of WHB_g. The WHB_g was prepared onsite using the grounding method, wherein the control over oxygen entrance could not be entirely regulated. This could lead to the loss of easily volatile oxygen-containing surface functional groups. Conversely, WHB_f was prepared using an electric furnace by putting the biomass in a tightly closed aluminum tin where oxygen entrance was entirely regulated. Consequently, oxygen-containing surface functional groups could not be lost during the preparation process.

The scanning electron microscope (SEM) images indicated distinct differences in the morphology and surface structures between WHB_f and WHB_g (Figure 2). The WHB_f exhibited well-organized and porous structures, surpassing the WHB_g in terms of arrangement (Figure 2A,B). Conversely, the WHB_g displayed surface structures with underdeveloped pores, filled with volatile matter that remained within cell structures (Figure 2C,D). The BET surface area and total pore volume results aligned with the SEM observations. The WHB_f, with more developed pores, showed a higher BET surface area (61.4 m^2 g^{-1}) and total pore volume (0.07 cm^3 g^{-1}) compared to the WHB_g, which exhibited a lower BET surface area (14.7 m^2 g^{-1}) and total pore volume (0.04 cm^3 g^{-1}) (Table 1), indicating undeveloped pore structures. The SEM images showing underdeveloped pores in WHB_g were also consistent with its higher ash and volatile matter contents (Table 1).

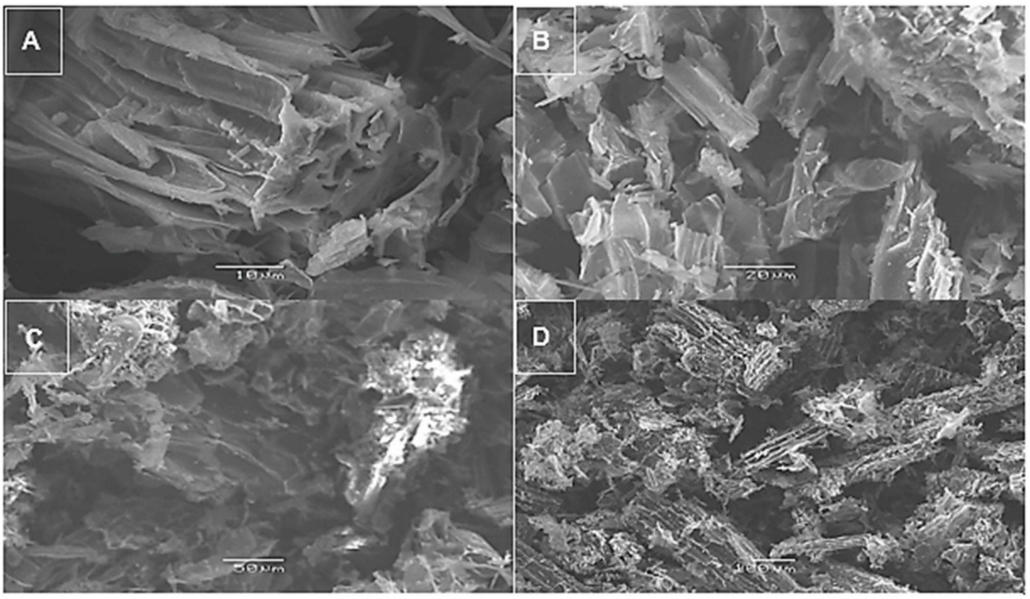

Figure 2. Surface morphologies/structures of (**A**) WHB_f magnified at 1000 times, (**B**) WHB_f magnified at 500 times, (**C**) WHB_g magnified at 1000 times, and (**D**) WHB_g magnified at 500 times as generated by scanning electron microscope (SEM). (WHB_f: water hyacinth biochar produced using furnace; and WHB_g: water hyacinth biochar produced by grounding method).

3.2. The effect of WHB on Soil Resistance to Acidification Stress

The soil pH significantly varied ($p < 0.05$) depending on the treatment types and application rates (Figure 3; Table 1). Following the application of WHB and lime, there was a notable increase in soil pH during the initial stages of incubation. Soil pH increases of 1.26, 1.25, and 1.00 units were noted on Day 3 in the $2WHB_fF$, $2WHB_f$, and $2WHB_g$ treatments, respectively, while the L treatment showed only a 0.46 unit increase. Thereafter, the pH decreased for all treatments, except the biochar treatments on Day 91, which displayed a rise in pH. After incubation, soil pH reduction due to acidification was significantly lower in biochar treatments (0.25, 0.40, 0.46, 0.78, and 0.82 units for $2WHB_f$, 2WHBg, $2WHB_fF$, $1WHB_f$, and $1WHB_g$ treatments, respectively) than in the LF, F, and L treatments (1.13, 1.33, and 1.12 units, respectively) (Figure 1).

The exchangeable acidity (Al^{3+} and H^+) exhibited significant variations ($p < 0.05$) throughout the incubation period depending on the treatment types and application rates (Figure 4; Table 1). A 1% application of WHB_f and WHB_g reduced exchangeable acidity by 1.68 and 1.36 cmol (+) kg^{-1}, while the 2% application reduced it by 2.75 and 2.53 cmol (+) kg^{-1}, respectively, during the first 3 incubation days. The WHB_f was more effective in hindering soil exchangeable acidity rise compared to the WHB_g. The LF and L treatments lessened it by 0.99 and 1.09 cmol (+) kg^{-1}, respectively. Conversely, F raised it by 0.43 cmol (+) kg^{-1} during this period. Thereafter, it increased over the entire incubation period, except for the biochar treatments on Day 91, which displayed a decrease in exchangeable acidity. However, the increase in exchangeable acidity, even after acidification, was lower in biochar treatments (0.43–0.50 cmol (+) kg^{-1} in 1%WHB, and 0.08–0.29 cmol (+) kg^{-1} in 2% WHB from Day 28 to Day 91) than in the L, LF, and F treatments (1.84, 2.08, and 2.69 cmol (+) kg^{-1} from Day 28 to Day 91, respectively) (Figure 2). Exchangeable acidity and soil pH in all treatments displayed negative correlations (Figure 5). After incubation, the exchangeable acidity level in different treatments followed the order: F > LF > L > $1WHB_g$ > $1WHB_f$ > $2WHB_fF$ > $2WHB_g$ > $2WHB_f$, which was reversed to the order of soil pH.

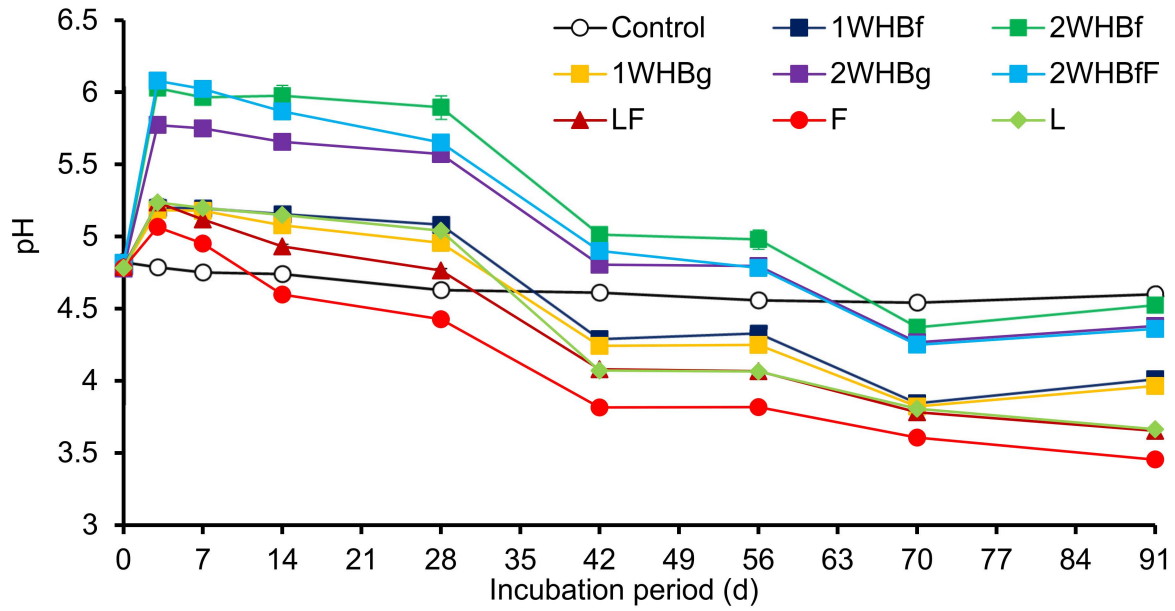

Figure 3. Changing trends of pH in acidic soil during the incubation period after being amended by water hyacinth biochar (WHB) or lime (L). (Control: soil only; $1WHB_f$ and $2WHB_f$: 1% and 2% applications of WHB produced using furnace; $1WHB_g$ and $2WHB_g$: 1% and 2% applications of WHB produced by grounding method; F: fertilizer at a recommended rate; $2WHB_fF$: $2WHB_f$ combined with fertilizer; L: lime at rate based on the exchangeable acidity of the experimental soil; and LF: lime combined with fertilizer). Error bars are the standard deviations of triplicate measurements.

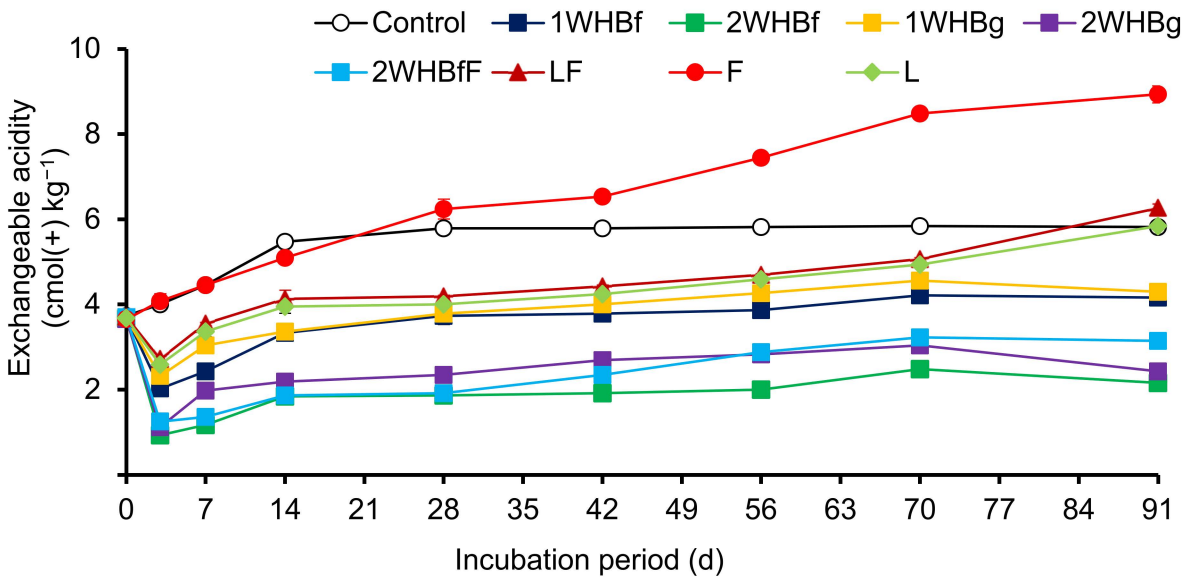

Figure 4. The dynamics of soil exchangeable acidity during the incubation period after being amended by water hyacinth biochar (WHB) or lime (L). (Control: soil only; $1WHB_f$ and $2WHB_f$: 1% and 2% applications of WHB produced using furnace; $1WHB_g$ and $2WHB_g$: 1% and 2% applications of WHB produced by grounding method; F: fertilizer at a recommended rate; $2WHB_fF$: $2WHB_f$ combined with fertilizer; L: lime at rate based on the exchangeable acidity of the experimental soil; and LF: lime combined with fertilizer). Error bars are the standard deviations of triplicate measurements.

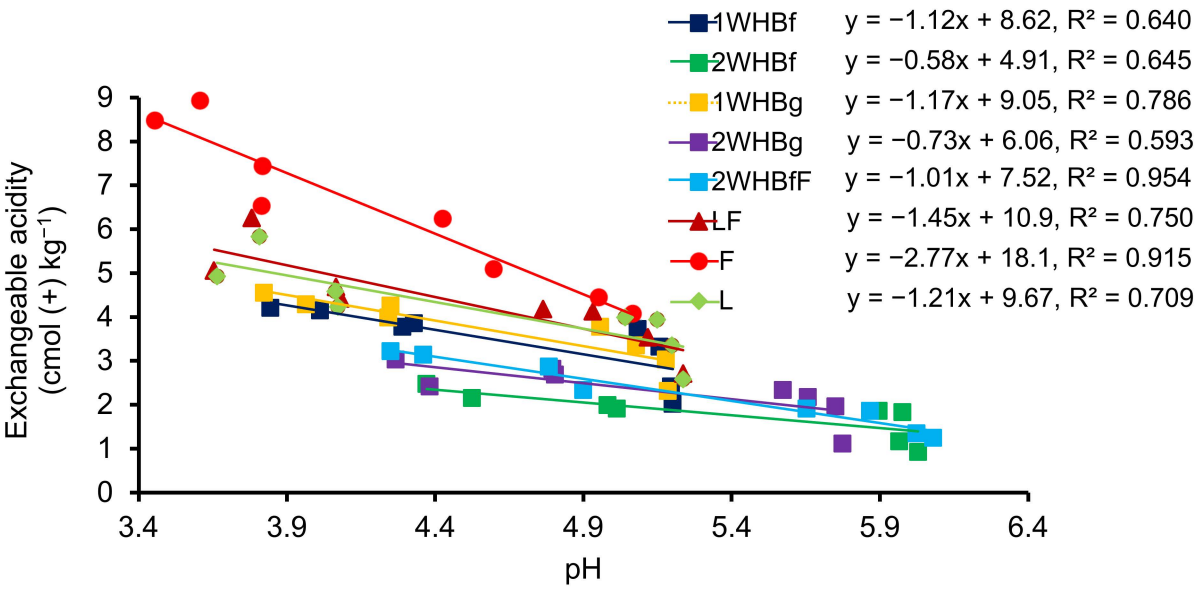

Figure 5. Correlation of exchangeable acidity and pH in soil amended by water hyacinth biochar (WHB) or lime (L). (1WHB$_f$ and 2WHB$_f$: 1% and 2% applications of WHB produced using furnace; 1WHB$_g$ and 2WHB$_g$: 1% and 2% applications of WHB produced by grounding method; F: fertilizer at a recommended rate; 2WHB$_f$F: 2WHB$_f$ combined with fertilizer; L: lime at rate based on the exchangeable acidity of the experimental soil; and LF: lime combined with fertilizer). Error bars are the standard deviations of triplicate measurements.

The exchangeable Al^{3+} underwent significant variations ($p < 0.05$) throughout the incubation period. No observable exchangeable Al^{3+} was recorded during the first 28 days of incubation in the 1% WHB application, nor during the 70 days of incubation in the 2% WHB application (Figure 2; Table 1). In the 1WHB$_f$ and 1WHB$_g$ treatments, exchangeable Al^{3+} increased from zero on Day 28 (before acidification) to 0.48 and 0.61 cmol (+) kg^{-1} on Day 56 (after first acidification), to 0.67 and 0.85 cmol (+) kg^{-1} on Day 70 (after second acidification), and to 1.36 and 1.39 cmol (+) kg^{-1} on Day 91 (after incubation), respectively. In the 2WHB$_f$, 2WHB$_g$, and 2WHB$_f$F treatments, it escalated from zero on Day 70 to 0.24, 0.37, and 0.35 cmol (+) kg^{-1} on Day 91, respectively. In the LF, F, and L treatments, it increased by 2.01, 2.16, and 1.84 cmol (+) kg^{-1} from Day 28 to Day 91, respectively. There was no exchangeable Al^{3+} rise in any of the biochar-treated soils compared to the initial value but in the limed soils (Figure 3). Compared to the exchangeable Al^{3+} content of the limed soil, a 1% application of WHB decreased exchangeable Al^{3+} by 30.5% to 32%, while the 2% application reduced it by 81.5% to 88% (Figure 3, Table 1). The 2WHB$_f$F treatment displayed 84.4% and 90% lower exchangeable Al^{3+} than the LF and F treatments, respectively.

3.3. The Effect of WHB on Soil Nutrient Dynamics

The application of WHB slowed the nitrification of NH_4^+–N into NO_3^-–N from the applied fertilizer. The 2WHB$_f$F, LF, and F treatments displayed higher NH_4^+–N levels throughout the incubation period, reaching maximum values of 75.2, 74.2, and 56.3 mg kg^{-1} on Day 3, and 84.3, 78.4, and 72.6 mg kg^{-1} on Day 42, respectively (Figure 6A). The NH_4^+−N content in the L treatment decreased from 8.33 mg kg^{-1} on Day 3 to 5.71 mg kg^{-1} on Day 14, increased to 19.5 mg kg^{-1} on Day 42, and finally increased to 20.1 mg kg^{-1}. After incubation, the NH_4^+−N content in the 2WHB$_f$, 2WHB$_g$, 1WHB$_f$, and 1WHB$_g$ treatments was higher by 13.8, 11.6, 11.6, and 11.3 mg kg^{-1}, respectively, compared to the control. Although the values exceeded the NH_4^+−N content in the control, there was no sharp increase or decrease in the NH_4^+−N content of biochar and lime treatments that did not contain fertilizer.

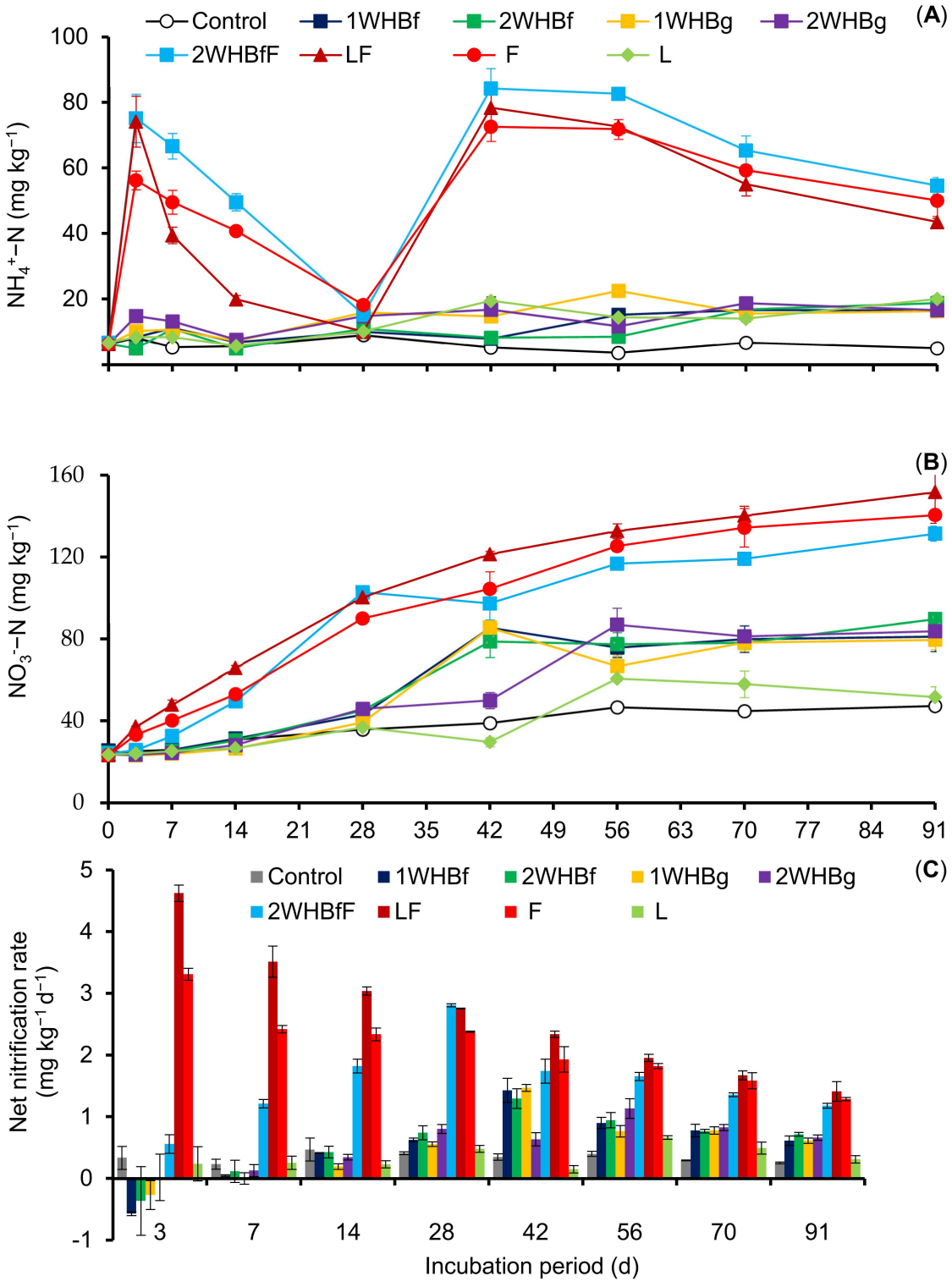

Figure 6. The dynamics of (**A**) NH_4^+–N, (**B**) NO_3^-–N, and (**C**) net nitrification rate in acidic soil during the incubation period after being amended by water hyacinth biochar (WHB) or lime (L). (Control: soil only; $1WHB_f$ and $2WHB_f$: 1% and 2% applications of WHB produced using furnace; $1WHB_g$ and $2WHB_g$: 1% and 2% applications of WHB produced by grounding method; F: fertilizer at a recommended rate; $2WHB_fF$: $2WHB_f$ combined with fertilizer; L: lime at rate based on the exchangeable acidity of the experimental soil; and LF: lime combined with fertilizer). Error bars are the standard deviations of triplicate measurements.

All biochar and lime treatments increased the NO_3^-−N content above the control, except for L on Day 42 (Figure 6B). The $2WHB_fF$, LF, and F treatments exhibited higher NO_3^-−N levels throughout the incubation period, peaking at 131, 151, and 141 mg kg^{-1} on Day 91, respectively. Their NO_3^-−N content was reversed to the order of their NH_4^+−N content. The L treatment showed the lowest NO_3^-−N content, rising from 24.1 mg kg^{-1} on Day 3 to 60.6 mg kg^{-1} on Day 56 and declining to 51.6 mg kg^{-1} on Day 91. The NO_3^-−N levels noted in soil samples with a 1% and 2% WHB application ranged from 23.1 to 23.4 mg kg^{-1} on Day 3 to 79.7 to 89.7 mg kg^{-1} on Day 91, respectively.

The $2WHB_fF$, LF, and F treatments exhibited higher net nitrification rates during the entire incubation period, with maximum rates of 2.81, 4.63, and 3.31 mg kg^{-1} d^{-1}noted on Day 3 for LF and F and on Day 28 for $2WHB_fF$, respectively (Figure 6C). After incubation, the net nitrification rates in the $2WHB_fF$, LF, and F treatments declined to 1.18, 1.41, and 1.29 mg kg^{-1} d^{-1}, respectively. Soil acidification decreased the net nitrification rate in the L treatment, which decreased from 0.48 mg kg^{-1} d^{-1} on Day 28 to 0.15 mg kg^{-1} d^{-1} on Day 42, and from 0.66 mg kg^{-1} d^{-1} on Day 56 to 0.31 mg kg^{-1} d^{-1} on Day 91. The $1WHB_f$, $1WHB_g$, and $2WHB_f$ treatments showed maximum net nitrification rates on Day 42 (1.43,1.47, and 1.29 mg kg^{-1} d^{-1}, respectively), and declined to 0.61, 0.61, and 0.72 mg kg^{-1} d^{-1} on Day 91, respectively. In the $2WHB_g$ treatment, the maximum net nitrification rate (1.13 mg kg^{-1} d^{-1}) was recorded on Day 56. The net nitrification rate and NO_3^-−N content exhibited similar trends across all treatments.

The type of treatments and application rates significantly influenced ($p < 0.05$) soil available P (Figure 7; Table 1). After 3 days of incubation, significant increases in available P were noted under different treatments with the most notable levels recorded for $2WHB_fF$ (17.1 mg kg^{-1}), $2WHB_f$ (15.2 mg kg^{-1}), $2WHB_g$ (7.04 mg kg^{-1}), and $1WHB_f$ (7.49 mg kg^{-1}), among others. The $2WHB_fF$ and $2WHB_f$ treatments displayed higher available P levels during the whole incubation period. Following the first acidification on Day 35, the available P decreased across all treatments. The lowest values were 3.83 mg kg^{-1} for $2WHB_fF$, 3.05 mg kg^{-1} for $2WHB_f$, and even lower values for other treatments, as recorded after incubation.

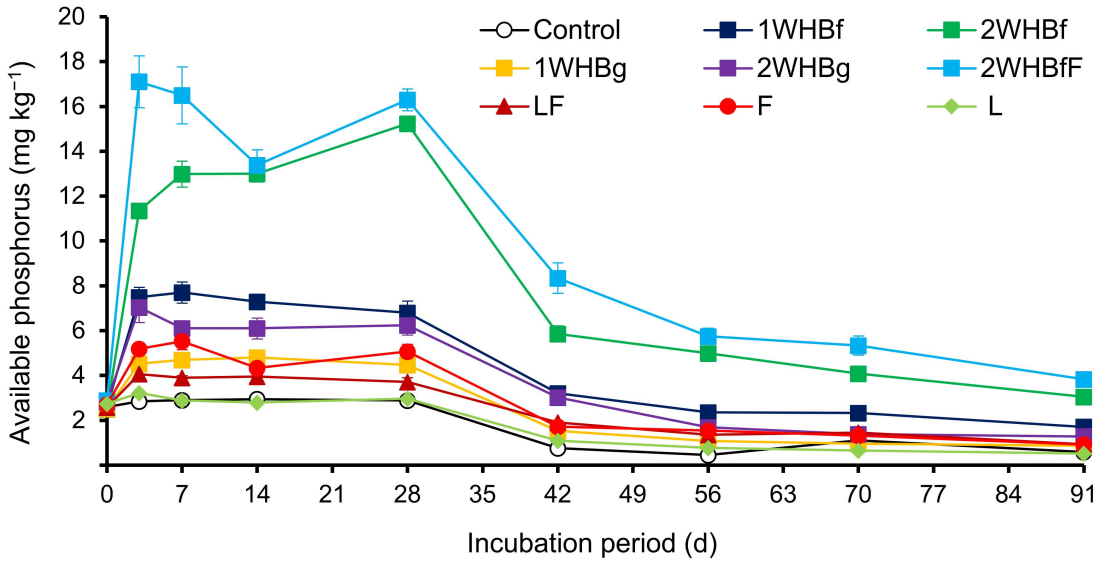

Figure 7. The dynamics of soil-available phosphorus during the incubation period after being amended by water hyacinth biochar (WHB) or lime (L). (Control: soil only; $1WHB_f$ and $2WHB_f$: 1% and 2% applications of WHB produced using furnace; $1WHB_g$ and $2WHB_g$: 1% and 2% applications of WHB produced by grounding method; F: fertilizer at a recommended rate; $2WHB_fF$: $2WHB_f$ combined with fertilizer; L: lime at rate based on the exchangeable acidity of the experimental soil; and LF: lime combined with fertilizer). Error bars are the standard deviations of triplicate measurements.

4. Discussion

4.1. WHB Effects on Soil Resistance to Acidification Stresses

The application of biochar as a soil-acidity ameliorator boosts soil pH primarily because of its alkaline composition [60]. Hence, the soil pH rises during the initial incubation period following the application of WHB, which could be attributed to the neutralization of soil acidity by the alkaline components in the biochar. The neutralization of H^+ ions in the soil solution by the OH^- from the hydrolysis of the first split urea (Equation (4)) resulted in a very slightly higher soil pH in the $2WHB_fF$ treatment (6.08) than that in $2WHB_f$ (6.03), as well as a minimal soil pH rise in the F treatment during the initial incubation stage. The higher soil pH in the $2WHB_f$ (6.03) on Day 3 (Figure 3), due to its higher alkaline content (24.7% $CaCO_3$ equivalent) than the pH in the $2WHB_g$ (5.77) with lower alkaline content (20.4% $CaCO_3$ equivalent) (Table 1), indicated that the WHBs' soil-acidity ameliorating potential was consistent with their chemical composition. A similar trend was found by Van Zwieten et al. [61], wherein biochar derived from papermill waste with a liming potential of 33% $CaCO_3$ equivalent raised the soil pH from 4.20 to 5.93, while another biochar with a lower liming potential of 29% $CaCO_3$ equivalent raised soil pH to 5.40. For the limed soil samples, the initial rise of pH was due to the neutralization of soil acidity (H^+) with the applied lime ($CaCO_3$) (Equation (5)).

$$CO(NH_2)_2 + 3H_2O \rightarrow 2NH_4^+ + 2OH^- + CO_2 \quad (4)$$

$$CaCO_3 + 2H^+ \rightarrow Ca^{2+} + H_2O + CO_2 \quad (5)$$

The deceleration of nitrification through NH_4^+ retention, and the neutralization or complexation of Al^{3+}, H^+, and Fe^{3+} by the phenolic, carboxylic, and hydroxyl groups on biochar's surfaces, could be the other mechanism by which biochar ameliorated soil acidity [31,62]. Accordingly, WHB_f with a higher porosity, as well as phenolic and carboxylic functional groups (Table 1), displayed a higher soil pH rise than WHB_g. Generally, the applied WHBs played dual roles in mitigating soil acidification: (1) They delayed NH_4^+ nitrification in soil, which otherwise would have accelerated soil acidification and increased soil pH; and (2) they increased soil pH buffer capacity. The results aligned with the findings by Shi et al. [29], where peanut and rice straw biochars played double roles in mitigating soil acidification, inhibited NH_4^+ nitrification in soil from the applied urea fertilizer, and increased soil pH buffer capacity.

During acidification, the soil-buffering capacity declined, leading to a decrease in soil pH. However, the reduction of soil pH in the WHB treatments was significantly lower than in the L, LF, and F treatments (Figure 1). After incubation, the soil pH in the $1WHB_f$, $1WHB_g$, $2WHB_f$, and $2WHB_g$ treatments was 0.35, 0.30, 0.86, and 0.72 units higher, respectively, than the pH in the limed soil. This disclosed that WHB increased soil's resistance against acidification stress more effectively than lime. Similarly, the $2WHB_fF$ treatment showed a soil pH that was 0.71 and 0.91 units higher than the LF and F treatments, respectively, demonstrating WHB_f's significant role in mitigating soil acidification. The non-significant difference in the soil pH reduction (ΔpH) between the non-acidified control and the twice acidified $2WHB_f$ treatment (Figure 1) further confirmed WHB's effectiveness in resisting soil acidification. WHB_f and WHB_g exhibited a longer-lasting increased soil pH than lime, suggesting WHB as a superior alternative for enhancing soil-buffering capacity and resisting acidification.

However, when using biochar for long-term applications, it is important to consider the possible accumulation of polycyclic aromatic hydrocarbons (PAHs) in soils. This negative effect can be minimized by employing optimum pyrolysis conditions to ensure that the benefits of biochar outweigh its harm. Buss et al. [63] found that optimal operating parameters, such as pyrolysis temperature and retention time, can minimize PAH concentration in biochar. Jose et al. [64] and Krzyszczak et al. [65] also found that sewage sludge, wood, rice husk, and wheat straw biochars produced by slow pyrolysis at 400–600 °C had PAH levels below international and European threshold limits, making

them safe for soil-amendment applications. Therefore, our WHBs produced by slow pyrolysis at 450 °C and by the grounding method are unlikely to contain PAH levels exceeding threshold limits.

The soil acidity-resistance capacity of the WHB observed in this study aligned with the reported research findings [29,31], wherein peanut and rice straw biochars improved soil's buffering capacity and resistance to reacidification more effectively than $Ca(OH)_2$. Similarly, in a simulated soil acidification experiment using HNO_3 [32], corn stover biochar heightened the soil's resistance to acidification more effectively than lime. Ding et al. [12] also reported that lime application was not effective in resisting the reacidification of the soil, unlike biochar. Furthermore, Li et al. [34] found that biochar derived from invasive plants ameliorated the detrimental impacts of acid rain on soil pH more effectively than lime.

The delayed nitrification in the $2WHB_fF$ treatment resulted in 35.1% and 19.2% lower net H^+ production from nitrification (4.24 mmol H^+ kg^{-1}) compared to the LF (6.53 mmol H^+ kg^{-1}) and F treatments (5.25 mmol H^+ kg^{-1}) (Figure 6C). Similarly, previous findings showed that slow nitrification after biochar application reduced H^+ ion production by 11% to 115% [29] and 16% to 18% [31]. Bolan et al. [66] found that biochar from different feedstocks increased soil resistance to acidity by neutralizing the H^+ ions generated from soil processes, such as CO_2 dissolution, ammonium assimilation, organic sulfur mineralization, and NH_4^+ nitrification. Additionally, Zhao et al. [67] stated that combining nitrogen fertilizer with rice straw biochar mitigated pomelo orchard soil acidification, raising soil pH more effectively than nitrogen fertilizer alone.

The content of exchangeable acidity (H^+ and Al^{3+}) primarily hinges on soil pH and rises as soil pH decreases [68]. In this study, exchangeable acidity, except in the control and F treatments, initially declined, coinciding with a rise in pH. Subsequently, it increased again, corresponding to a decrease in pH. (Figure 4). The extent of exchangeable acidity rise was significantly lower in the biochar treatments than that in the limed soil (28.8, 26.5, 58.4, and 63% lower exchangeable acidity after incubation in the $1WHB_f$, $1WHB_g$, $2WHB_g$, and $2WHB_f$ treatments, respectively). The replacement of H^+ and Al^{3+} from the soil exchange site by the basic cations (K^+, Ca^{2+}, and Mg^{2+}) from the WHBs led to lower exchangeable acidity in the biochar-treated soils compared to the limed soils. In the L and LF treatments, only Ca^{2+} can replace H^+ and Al^{3+} from soil colloids. High nitrification and increased availability of Al^{3+} at a lower soil pH resulted in 58.7, 69, and 145% rises in exchangeable acidity in the L, LF, and F treatments, respectively. Conversely, the exchangeable acidity in the $2WHB_f$, $2WHB_g$, and $2WHB_fF$ treatments decreased by 41.3, 33.4, and 15%, respectively (Figure 4). This underscores the superior efficacy of WHB over lime in enhancing soil resistance to acidification stress induced by chemical fertilizers and other soil acidity sources. Chintala et al. [69] demonstrated a comparable pattern; corn stover and switchgrass biochars applied to acidic clayey soil resulted in a greater increase in pH and a more substantial reduction in exchangeable acidity compared to the limed soils. Additionally, Zhang W. et al. [70] found that biochar derived from tobacco stalks completely neutralized soil exchangeable acidity in China's Qianxi region.

Soil pH and exchangeable acidity of all treatments showed inverse correlations (Figure 5). As indicated by the regression equations (Figure 5), soils amended with biochar exhibited smaller negative slopes than the limed soils. Based on the regression equations, when soil pH decreased by one unit, the exchangeable acidity of the soil treated by WHBs could increase by 0.58–1.17 folds, while it could rise by 1.21, 1.45, and 2.77 folds in the L, LF, and F treatments, respectively. This confirmed that WHB could be a better alternative to lime to boost soil's resistance against soil acidification stresses. Cai et al. [71] found that soil exchangeable Al^{3+} increased by 4.48 and 6.25 folds with a unit decrease in pH for maize straw and swine manure-treated soils, respectively, indicating that WHB-suppressed exchangeable acidity rises more effectively than these organic amendments.

Exchangeable Al^{3+} in acidic soils is a primary contributor to Al toxicity in plants. In this study, WHBs substantially mitigated exchangeable Al^{3+} rise during soil acidification. The

slower decrease in soil pH due to the enhanced soil-buffering capacity, following biochar application, restrained the escalation of exchangeable Al^{3+} during soil acidification. The availability of Al^{3+} in soil is highly dependent on soil pH. At a higher soil pH, Al^{3+} forms an insoluble precipitate of $Al(OH)_3$. Due to this, no exchangeable Al^{3+} was detected during the 28 days of incubation in the 1% WHB application, nor during the 70 days of incubation in the 2% WHB application, as soil pH increased following WHB application (Figure 2). When pH declined due to soil acidification, exchangeable Al^{3+} was recorded, including after the first soil acidification in the 1% WHB and after the second soil acidification in the 2% WHB applications. Conversely, in the L, LF, and F treatments, exchangeable Al^{3+} was recorded even before the acidification events. The better capacity displayed by WHB_f in reducing exchangeable Al^{3+}, compared to WHB_g, was consistent with its higher porosity, alkalinity, and CEC than that of WHB_g, which enabled it to trap and neutralize or precipitate more Al^{3+} than WHB_g. Generally, there was no rise in soil exchangeable Al^{3+} in the WHB-amended soils, unlike in the limed soils (Figure 3). This pointed out the greater potential of WHB over lime in enhancing soil resistance to various acidification stresses. Similarly, [29,31] found that crop residue biochars were more effective than $Ca(OH)_2$ in mitigating exchangeable Al^{3+} induced by fertilizer and simulated soil acidification with HNO_3. Becerra–Agudelo et al. [32] also found that exchangeable Al^{3+} remained below 2 cmol (+) kg^{-1} in the biochar-treated soil, while it increased to over 8 cmol (+) kg^{-1} in the limed soil when the soil was acidified with an equal amount of HNO_3. The interaction of Al^{3+} with the negatively charged species and its co-precipitation with silicate particles (such as $KAlSiO_8$) within the biochars may have resulted in the reduction of soil exchangeable Al^{3+} following the incorporation of WHB [72]. Jutakanoke et al. [73] demonstrated that the precipitation of toxic Al^{3+} by Alkaline oxides, carbonates, and silicates in water hyacinth biochar alleviated soil acidification.

4.2. WHB Effects on Soil Nutrient Dynamics

The change of NH_4^+-N concentration following treatment application indicated the occurrence of nitrogen transformation. The NH_4^+-N content reached maximum values on Day 3 (75.2, 74.2, and 56.3 mg kg^{-1}) and Day 42 (84.3, 78.4, and 72.6 mg kg^{-1}) in the $2WHB_fF$, F, and LF treatments, respectively (Figure 6A). This was due to the hydrolysis of urea (Equation (4)) applied on Days 0 and 40, respectively. Previous research findings [29,33] also indicated that soil pH and NH_4^+-N raised due to the hydrolysis of urea after 3 days of incubation. The NH_4^+-N content in these treatments declined, reaching minimum values (15.3, 10.1, and 18.2 mg kg^{-1}) on Day 28 due to the nitrification of NH_4^+ into NO_3^- as confirmed by a noteworthy increase in the NO_3^--N content (Figure 6B). The decline of NH_4^+-N in the $2WHB_fF$ treatment was slower than the reductions in the LF and F treatments (Figure 6A). This indicated the moderating effect of WHB on nitrification, resulting in lower soil acidification caused by H^+ ions released during nitrification. Conversely, the rapid decay of NH_4^+-N in the LF treatment implied that lime application could not provide a long-lasting solution to retain NH_4^+ and slow-down nitrification. Thus, lime could not provide an effective solution to resist soil acidification induced by fertilizers. The NH_4^+-N dynamics in our study agreed with the findings by Shi et al. [29], wherein the decrease in NH_4^+-N and the rise in NO_3^--N occurred notably slowly in biochar treatments compared to those using $Ca(OH)_2$.

The slower NH_4^+ nitrification due to the WHB_f in the $2WHB_fF$ treatment resulted in 20.3 and 9.06 mg kg^{-1} lower NO_3^--N content than those in the LF and F treatments, respectively (Figure 6B). The soil NO_3^--N content in these treatments followed the order $LF > F > 2WHB_fF$, which was reversed to the order of soil NH_4^+-N content. On the other hand, the slow rise of the NO_3^--N level in non-fertilized biochar treatments towards Day 42, even with soil acidification, revealed that WHB had enhanced soil's resistance to acidification stress and provided protection to ammonium oxidizing bacteria (AOB) against the detrimental effects of external acidification stress. Additionally, the increase in the NO_3^--N content in the biochar treatments that did not contain fertilizer revealed that

WHB could supply nitrogen to the soil, unlike lime. Conversely, the NO_3^-−N level in the L treatment declined on Days 42 and 70 (Figure 6B) due to acidification events occurring on Days 35 and 63, respectively. This signposted that lime could not protect soil microbes from acidification stress, and nitrifying microbes were demolished. Zhou et al. [33] observed inhibited nitrification below pH 4, while Bolan et al. [74] reported a significant decrease in nitrification below pH 4.5 in the limed soil.

In general, NH_4^+−N retention in biochar's porous structure [75], and the deactivation of AOB by toxic phenolic compounds and polycyclic aromatic hydrocarbons on biochar's surface [76], were the major reasons presumed for the slower nitrification of NH_4^+−N into NO_3^-−N when biochar was combined with fertilizer compared to the sole fertilizer application. Wang et al. [77] found that the toxic phenolic compounds in biochar decreased the population size of AOB by 2–4 orders of magnitude and inhibited nitrification, but the inhibitory impact vanished upon the removal of phenolic compounds from the biochar. Li et al. [78] also found that the use of biochar-based fertilizer reduced the nitrification rate, which was attributed to a decrease in the relative abundance of *Nitrosomonas* and *Nitrosospira* bacteria.

The lower NO_3^-−N level in the $2WHB_g$ treatment (50.0 mg kg^{-1}) than the remaining WHB treatments on Day 42 (Figure 6B) may be attributed to the N immobilization induced by the high application of WHB_g with a high C/N ratio. The WHB_g exhibited a higher C/N ratio (48.6) than the WHB_f (38.3), which could lead to N immobilization. The subsequent release of this immobilized NO_3^-−N might explain the peak NO_3^-−N level (86.9 mg kg^{-1}) in the $2WHB_g$ treatment recorded on Day 56. Deenik et al. [79] stated that biochar that owned a high C/N ratio led to competitiveness for heterotrophic nitrifiers and resulted in N immobilization. Soil micro-organisms typically thrive in environments with low C/N ratios, usually below 20 [80]. Thus, it is plausible that a higher C/N ratio of WHB_g might have led to temporal N immobilization in the soil. This result was analogous to the findings by Abera et al. [81] and Xiao et al. [82], who noted substantial immobilization of soil NO_3^-–N following the addition of crop straws with C/N ratios in the range of 36–42.

The slower nitrification in the $2WHB_fF$ treatment resulted in 15% to 88%, and 8% to 83% lower net nitrification rates compared to the net nitrification rates in the LF and F treatments, respectively (Figure 6C). This further underscores the intrinsic role of WHB in reducing soil acidification, preventing excessive NO_3^-−N loss, reducing nitrate pollution, and contributing to N cycling. Similarly, Agyarko–Mintah et al. [83] reported the significant role of biochar in nitrogen retention and its contribution to reducing nitrate and N_2O pollution. Additionally, Shi et al. [29] found that the net nitrification rate decreased by 55% and 19% due to the moderating effect of peanut and rice straw biochars, respectively. Moreover, Li et al. [84] found that biochar from hardwood slowed the hydrolysis of urea and lowered the nitrification rate compared to using urea alone.

Applying biochar into soil has been reported to increase P bioavailability [85]. In this study, soil available P was significantly ($p < 0.05$) increased after applying WHB (Figure 7, Table 1). The increase in available P after applying WHB could be explained by three mechanisms. Firstly, phosphate compounds with Al^{3+} and Fe^{3+} in acidic soils could be dissolved owing to an increase in pH induced by biochar application [86]. Secondly, the applied biochar could interact with soil components via the negatively charged alkaline functional groups on the biochar's surface, such as $–O^-$ and $–COO^-$, resulting in the release of phosphate, which was bound by acidic metal cations such as Al^{3+} and Fe^{3+} in the soil [47]. Thirdly, the applied biochars could have directly released P to the soil. Masud et al. [62] stated that biochar can directly release soluble P and enhance P availability.

The rise and fall of available P in biochar and lime-treated soil samples were related to the change in soil pH; it increased when pH was raised and declined when pH was subsequently lowered. Lower available P in the LF treatment with higher soil pH than that in the F treatment, except on Days 42 and 70, could be due to the microbial desorption of adsorbed P from clay minerals and soil organic matter by P solubilizing micro-organisms (PSM). The phenomenon of P desorption by PSM typically occurs alongside a drop in

pH. The P desorption results from the increased solubility of Fe and Al due to possible complexation with low molecular weight organic acids. At lower pH, P solubilizing micro-organisms can release various low-molecular-weight organic acids, such as citrate, oxalate, and malate. These organic acids and anions can displace P from adsorption sites through ligand exchange and temporarily block P adsorption sites, leading to an increase in available P [87]. Tian et al. [88] reported that 17%-to-34% of clay mineral absorbed P in Chinese red soils was desorbed and transformed to water-soluble and plant-available P by PSM. On the other hand, higher available P in the LF treatment than that in the F treatment on Days 42 and 70, following acidification events, indicated that P fixation by soil components, such as Al^{3+} and Fe^{3+}, during these acidification episodes overshadowed the P desorption by PSM in the F treatment. The consistent decrement of available P in all treatments after the first soil acidification could be due to the retention of available P by acidic metal cations. Zwetsloot et al. [89] pointed out the retention of available P in the second phase of the incubation period after it increased during the initial phase of incubation. Generally, it can be emphasized that the dynamic nature of P availability was influenced by microbial activity and the change in soil pH due to acidification events.

5. Conclusions

Water hyacinth biochar decelerated nitrification, suppressed soil pH reduction and curbed exchangeable acidity escalation more effectively than lime. WHB played two major roles: decelerated nitrification and increased soil pH-buffering capacity. As per our hypothesis, WHB resisted soil acidification more effectively than lime through (1) the reduction of H^+ ion generation by slowing nitrification, (2) the neutralization of H^+ by OH^- from alkaline groups in the biochar, and (3) the neutralization of acid by increasing soil pH buffer capacity as verified by slow soil pH reduction in biochar treatments during soil acidification. Additionally, WHB positively influenced available N and P dynamics. Biochar that was produced using an electric furnace (WHB_f) performed better than the biochar produced by the grounding method (WHB_g) in impeding pH reduction, suppressing exchangeable acidity escalation and promoting soil nutrient dynamics.

Therefore, WHB can be used as a better alternative to lime in reinforcing soil resistance against various acidification stresses while aiding in soil nutrient management. The study's findings can offer valuable insights into changing environmental challenges into sustainable soil quality management approaches. Nevertheless, conducting further studies at pot or field scales is necessary to delve deeper into the long-term effects of water hyacinth biochar on soil-acidity mitigation and its impact on crop production. The polycyclic aromatic hydrocarbon (PAH) content of WHB should be determined to minimize their accumulation in soils during broader field applications of biochar.

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Data Availability Statement: The data used to support the findings of this study are contained within the manuscript.

Conflicts of Interest: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A

Table 1. ANOVA results displaying statistically significant differences among the treatments of the soil parameters at the specified sampling dates.

Parameter	Sampling Date	Control	$1WHB_f$	$2WHB_f$	$1WHB_g$	$2WHB_g$	$2WHB_fF$	LF	F	L
pH	0	4.82 ± 0.00 ^{a}	4.80 ± 0.02 ^{a}	4.78 ± 0.00 ^{a}	4.78 ± 0.02 ^{a}	4.78 ± 0.02 ^{a}	4.82 ± 0.03 ^{a}	4.79 ± 0.01 ^{a}	4.78 ± 0.01 ^{a}	4.78 ± 0.01 ^{a}
	3	4.79 ± 0.02 ^{e}	5.20 ± 0.03 ^{c}	6.03 ± 0.05 ^{a}	5.18 ± 0.02 ^{c}	5.77 ± 0.05 ^{b}	6.08 ± 0.02 ^{a}	5.24 ± 0.03 ^{c}	5.07 ± 0.01 ^{d}	5.24 ± 0.03 ^{c}
	7	4.75 ± 0.01 ^{e}	5.20 ± 0.05 ^{c}	5.97 ± 0.01 ^{a}	5.18 ± 0.08 ^{c}	5.75 ± 0.02 ^{b}	6.02 ± 0.01 ^{a}	5.12 ± 0.01 ^{c}	4.95 ± 0.01 ^{d}	5.20 ± 0.00 ^{c}
	14	4.74 ± 0.00 ^{e}	5.16 ± 0.04 ^{c}	5.98 ± 0.09 ^{a}	5.08 ± 0.01 ^{c}	5.66 ± 0.06 ^{b}	5.87 ± 0.01 ^{a}	4.93 ± 0.02 ^{d}	4.60 ± 0.01 ^{f}	5.15 ± 0.01 ^{c}
	28	4.63 ± 0.00 ^{f}	5.08 ± 0.02 ^{c}	5.90 ± 0.10 ^{a}	4.96 ± 0.01 ^{d}	5.57 ± 0.04 ^{b}	5.65 ± 0.01 ^{b}	4.76 ± 0.02 ^{e}	4.43 ± 0.03 ^{g}	5.04 ± 0.01 ^{cd}
	42	4.61 ± 0.01 ^{d}	4.29 ± 0.03 ^{e}	5.01 ± 0.03 ^{a}	4.24 ± 0.02 ^{e}	4.80 ± 0.02 ^{c}	4.90 ± 0.02 ^{b}	4.08 ± 0.00 ^{f}	3.82 ± 0.00 ^{g}	4.07 ± 0.00 ^{f}
	56	4.56 ± 0.00 ^{c}	4.33 ± 0.04 ^{d}	4.98 ± 0.08 ^{a}	4.25 ± 0.02 ^{d}	4.80 ± 0.01 ^{b}	4.78 ± 0.03 ^{b}	4.07 ± 0.01 ^{e}	3.82 ± 0.00 ^{f}	4.06 ± 0.00 ^{e}
	70	4.54 ± 0.00 ^{a}	3.84 ± 0.02 ^{d}	4.37 ± 0.05 ^{b}	3.82 ± 0.00 ^{d}	4.27 ± 0.01 ^{c}	4.25 ± 0.02 ^{c}	3.78 ± 0.00 ^{e}	3.61 ± 0.00 ^{f}	3.81 ± 0.01 ^{e}
	91	4.60 ± 0.01 ^{a}	4.01 ± 0.03 ^{d}	4.52 ± 0.03 ^{b}	3.96 ± 0.01 ^{d}	4.38 ± 0.01 ^{c}	4.36 ± 0.02 ^{c}	3.65 ± 0.02 ^{e}	3.46 ± 0.00 ^{f}	3.66 ± 0.00 ^{e}
Exchangeable acidity (cmol (+) kg^{-1})	0	3.73 ± 0.05 ^{a}	3.71 ± 0.09 ^{a}	3.68 ± 0.08 ^{a}	3.68 ± 0.21 ^{a}	3.65 ± 0.05 ^{a}	3.71 ± 0.05 ^{a}	3.71 ± 0.05 ^{a}	3.65 ± 0.09 ^{a}	3.68 ± 0.00 ^{a}
	3	4.00 ± 0.00 ^{a}	2.03 ± 0.05 ^{d}	0.93 ± 0.05 ^{f}	2.32 ± 0.14 ^{c}	1.12 ± 0.08 ^{ef}	1.25 ± 0.05 ^{e}	2.72 ± 0.08 ^{b}	4.08 ± 0.21 ^{a}	2.59 ± 0.05 ^{bc}
	7	4.45 ± 0.05 ^{a}	2.43 ± 0.05 ^{d}	1.17 ± 0.05 ^{f}	3.04 ± 0.00 ^{c}	1.97 ± 0.05 ^{e}	1.36 ± 0.08 ^{f}	3.55 ± 0.05 ^{b}	4.45 ± 0.05 ^{a}	3.36 ± 0.14 ^{b}
	14	5.47 ± 0.09 ^{a}	3.33 ± 0.05 ^{c}	1.84 ± 0.08 ^{d}	3.36 ± 0.21 ^{c}	2.19 ± 0.12 ^{d}	1.87 ± 0.09 ^{d}	4.13 ± 0.26 ^{b}	5.09 ± 0.05 ^{a}	3.95 ± 0.18 ^{b}
	28	5.79 ± 0.09 ^{b}	3.73 ± 0.24 ^{d}	1.87 ± 0.05 ^{f}	3.79 ± 0.12 ^{cd}	2.35 ± 0.05 ^{e}	1.92 ± 0.14 ^{f}	4.19 ± 0.05 ^{c}	6.24 ± 0.29 ^{a}	4.00 ± 0.08 ^{cd}
	42	5.79 ± 0.05 ^{b}	3.79 ± 0.12 ^{e}	1.92 ± 0.14 ^{h}	4.00 ± 0.08 ^{de}	2.69 ± 0.05 ^{f}	2.35 ± 0.05 ^{g}	4.43 ± 0.05 ^{c}	6.53 ± 0.05 ^{a}	4.24 ± 0.14 ^{cd}
	56	5.81 ± 0.05 ^{b}	3.87 ± 0.05 ^{e}	2.00 ± 0.08 ^{g}	4.27 ± 0.05 ^{d}	2.83 ± 0.09 ^{f}	2.88 ± 0.08 ^{f}	4.69 ± 0.05 ^{c}	7.44 ± 0.08 ^{a}	4.59 ± 0.05 ^{c}
	70	5.84 ± 0.00 ^{b}	4.21 ± 0.12 ^{e}	2.48 ± 0.08 ^{g}	4.56 ± 0.08 ^{d}	3.04 ± 0.00 ^{f}	3.23 ± 0.12 ^{f}	5.07 ± 0.04 ^{c}	8.48 ± 0.08 ^{a}	4.93 ± 0.04 ^{c}
	91	5.81 ± 0.05 ^{c}	4.16 ± 0.14 ^{d}	2.16 ± 0.14 ^{f}	4.29 ± 0.09 ^{d}	2.43 ± 0.04 ^{f}	3.15 ± 0.05 ^{e}	6.27 ± 0.12 ^{b}	8.93 ± 0.24 ^{a}	5.84 ± 0.08 ^{c}
Exchangeable Al^{3+} (cmol (+) kg^{-1})	0	1.36 ± 0.08 ^{a}	1.36 ± 0.08 ^{a}	1.36 ± 0.08 ^{a}	1.36 ± 0.08 ^{a}	1.36 ± 0.00 ^{a}	1.36 ± 0.08 ^{a}	1.33 ± 0.05 ^{a}	1.36 ± 0.08 ^{a}	1.36 ± 0.08 ^{a}
	3	1.23 ± 0.05 ^{a}	0.00 ± 0.00 ^{d}	0.00 ± 0.00 ^{d}	0.00 ± 0.00 ^{d}	0.00 ± 0.00 ^{d}	0.00 ± 0.00 ^{d}	0.00 ± 0.00 ^{d}	0.83 ± 0.05 ^{b}	0.19 ± 0.05 ^{c}
	7	1.17 ± 0.09 ^{a}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.72 ± 0.08 ^{b}	0.00 ± 0.00 ^{c}
	14	1.15 ± 0.05 ^{a}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	1.04 ± 0.08 ^{b}	0.00 ± 0.00 ^{c}
	28	1.17 ± 0.09 ^{a}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.00 ± 0.00 ^{c}	0.21 ± 0.05 ^{b}	1.25 ± 0.05 ^{a}	0.16 ± 0.00 ^{b}
	42	1.65 ± 0.12 ^{b}	0.48 ± 0.00 ^{d}	0.00 ± 0.00 ^{e}	0.67 ± 0.05 ^{c}	0.00 ± 0.00 ^{e}	0.00 ± 0.00 ^{e}	1.65 ± 0.09 ^{b}	2.75 ± 0.05 ^{a}	1.68 ± 0.08 ^{b}
	56	1.68 ± 0.00 ^{b}	0.48 ± 0.00 ^{c}	0.00 ± 0.00 ^{d}	0.61 ± 0.05 ^{c}	0.00 ± 0.00 ^{d}	0.00 ± 0.00 ^{d}	1.76 ± 0.08 ^{b}	2.93 ± 0.05 ^{a}	1.87 ± 0.17 ^{b}
	70	1.71 ± 0.05 ^{d}	0.67 ± 0.04 ^{f}	0.00 ± 0.00 ^{g}	0.85 ± 0.05 ^{e}	0.00 ± 0.00 ^{g}	0.00 ± 0.00 ^{g}	2.21 ± 0.04 ^{b}	3.33 ± 0.05 ^{a}	1.97 ± 0.05 ^{c}
	91	1.81 ± 0.12 ^{c}	1.36 ± 0.08 ^{d}	0.24 ± 0.08 ^{e}	1.39 ± 0.04 ^{d}	0.37 ± 0.05 ^{e}	0.35 ± 0.05 ^{e}	2.24 ± 0.00 ^{b}	3.41 ± 0.05 ^{a}	2.00 ± 0.08 ^{c}
Exchangeable H^{+} (cmol (+) kg^{-1})	0	2.37 ± 0.12 ^{a}	2.35 ± 0.12 ^{a}	2.32 ± 0.16 ^{a}	2.32 ± 0.29 ^{a}	2.29 ± 0.05 ^{a}	2.35 ± 0.09 ^{a}	2.37 ± 0.05 ^{a}	2.29 ± 0.12 ^{a}	2.32 ± 0.08 ^{a}
	3	2.77 ± 0.05 ^{b}	2.03 ± 0.05 ^{d}	0.93 ± 0.05 ^{f}	2.32 ± 0.14 ^{c}	1.12 ± 0.08 ^{ef}	1.25 ± 0.05 ^{e}	2.72 ± 0.08 ^{b}	3.25 ± 0.20 ^{a}	2.40 ± 0.08 ^{c}
	7	3.28 ± 0.14 ^{c}	2.43 ± 0.05 ^{e}	1.17 ± 0.05 ^{g}	3.04 ± 0.00 ^{d}	1.97 ± 0.05 ^{f}	1.36 ± 0.08 ^{g}	3.55 ± 0.05 ^{ab}	3.73 ± 0.05 ^{a}	3.36 ± 0.14 ^{bc}
	14	4.32 ± 0.08 ^{a}	3.33 ± 0.05 ^{b}	1.84 ± 0.08 ^{c}	3.36 ± 0.21 ^{b}	2.19 ± 0.12 ^{c}	1.87 ± 0.09 ^{c}	4.13 ± 0.26 ^{a}	4.05 ± 0.09 ^{a}	3.95 ± 0.18 ^{a}
	28	4.61 ± 0.09 ^{a}	3.73 ± 0.24 ^{b}	1.87 ± 0.05 ^{d}	3.79 ± 0.12 ^{b}	2.35 ± 0.05 ^{c}	1.92 ± 0.14 ^{cd}	3.97 ± 0.09 ^{b}	4.99 ± 0.32 ^{a}	3.84 ± 0.08 ^{b}
	42	4.13 ± 0.09 ^{a}	3.31 ± 0.12 ^{c}	1.92 ± 0.14 ^{f}	3.33 ± 0.09 ^{c}	2.69 ± 0.05 ^{d}	2.35 ± 0.05 ^{e}	2.77 ± 0.05 ^{d}	3.79 ± 0.05 ^{b}	2.56 ± 0.08 ^{de}
	56	4.13 ± 0.05 ^{b}	3.39 ± 0.05 ^{c}	2.00 ± 0.08 ^{e}	3.65 ± 0.05 ^{c}	2.83 ± 0.09 ^{d}	2.88 ± 0.08 ^{d}	2.93 ± 0.12 ^{d}	4.51 ± 0.12 ^{a}	2.72 ± 0.20 ^{d}
	70	4.48 ± 0.00 ^{b}	3.55 ± 0.09 ^{c}	2.48 ± 0.08 ^{f}	3.71 ± 0.04 ^{c}	3.04 ± 0.00 ^{de}	3.23 ± 0.12 ^{d}	2.85 ± 0.09 ^{e}	5.15 ± 0.12 ^{a}	2.96 ± 0.08 ^{e}
	91	4.00 ± 0.16 ^{b}	2.80 ± 0.21 ^{c}	1.92 ± 0.21 ^{d}	2.91 ± 0.05 ^{c}	2.05 ± 0.05 ^{d}	2.80 ± 0.08 ^{c}	4.03 ± 0.12 ^{b}	5.52 ± 0.24 ^{a}	3.84 ± 0.14 ^{b}

Table 1. *Cont.*

Parameter	Sampling Date	Control	$1WHB_f$	$2WHB_f$	$1WHB_g$	$2WHB_g$	$2WHB_fF$	LF	F	L
NH_4^+–N (mg kg^{-1})	0	6.35 ± 0.21 ^{a}	6.45 ± 0.34 ^{a}	6.31 ± 0.08 ^{a}	6.36 ± 0.05 ^{a}	6.33 ± 0.17 ^{a}	6.74 ± 0.12 ^{a}	6.38 ± 0.24 ^{a}	6.35 ± 0.20 ^{a}	6.65 ± 0.06 ^{a}
	3	8.17 ± 0.29 ^{c}	8.41 ± 0.12 ^{c}	4.97 ± 0.09 ^{c}	10.3 ± 0.90 ^{c}	14.9 ± 0.95 ^{c}	75.2 ± 0.74 ^{a}	74.2 ± 0.52 ^{a}	56.3 ± 0.50 ^{b}	8.33 ± 0.31 ^{c}
	7	5.33 ± 0.13 ^{e}	11.2 ± 0.12 ^{de}	11.0 ± 0.95 ^{de}	10.7 ± 0.09 ^{de}	13.3 ± 0.34 ^{d}	66.7 ± 0.79 ^{a}	39.5 ± 0.29 ^{c}	49.6 ± 0.45 ^{b}	8.33 ± 0.47 ^{de}
	14	5.59 ± 0.46 ^{d}	6.83 ± 0.20 ^{d}	5.00 ± 0.24 ^{d}	7.23 ± 0.64 ^{d}	7.62 ± 0.08 ^{d}	49.6 ± 0.21 ^{a}	19.9 ± 0.54 ^{c}	40.8 ± 0.65 ^{b}	5.71 ± 0.72 ^{d}
	28	9.03 ± 0.73 ^{c}	10.1 ± 0.83 ^{c}	10.9 ± 0.58 ^{c}	16.0 ± 0.94 ^{b}	14.9 ± 0.66 ^{b}	15.3 ± 0.77 ^{b}	10.1 ± 0.45 ^{c}	18.2 ± 0.43 ^{a}	10.1 ± 0.22 ^{c}
	42	5.25 ± 0.50 ^{e}	7.96 ± 0.76 ^{de}	8.24 ± 0.63 ^{de}	14.8 ± 0.71 ^{cde}	16.8 ± 0.23 ^{cd}	84.3 ± 0.85 ^{a}	78.4 ± 0.94 ^{ab}	72.6 ± 0.74 ^{b}	19.5 ± 0.48 ^{c}
	56	3.60 ± 0.19 ^{f}	15.3 ± 0.81 ^{d}	8.61 ± 0.31 ^{e}	22.6 ± 0.76 ^{c}	11.8 ± 0.58 ^{de}	82.7 ± 0.96 ^{a}	72.6 ± 0.78 ^{b}	71.8 ± 0.67 ^{b}	14.5 ± 0.47 ^{d}
	70	6.71 ± 0.26 ^{d}	16.7 ± 0.68 ^{c}	16.9 ± 0.88 ^{c}	15.6 ± 0.77 ^{c}	18.8 ± 0.63 ^{c}	65.4 ± 0.99 ^{a}	55.0 ± 0.49 ^{b}	59.3 ± 0.53 ^{ab}	14.0 ± 0.50 ^{cd}
	91	5.02 ± 0.32 ^{d}	16.6 ± 0.62 ^{c}	18.8 ± 0.56 ^{c}	16.4 ± 0.37 ^{c}	16.7 ± 0.73 ^{c}	54.7 ± 0.83 ^{a}	43.5 ± 0.42 ^{b}	50.0 ± 0.85 ^{ab}	20.1 ± 0.58 ^{c}
NO_3^-–N (mg kg^{-1})	0	24.2 ± 0.37 ^{a}	25.6 ± 0.76 ^{a}	24.5 ± 0.45 ^{a}	23.9 ± 0.93 ^{a}	23.4 ± 0.92 ^{a}	24.1 ± 0.92 ^{a}	23.2 ± 0.86 ^{a}	23.3 ± 0.41 ^{a}	23.4 ± 0.65 ^{a}
	3	25.3 ± 0.71 ^{cd}	23.9 ± 0.69 ^{cd}	23.4 ± 0.21 ^{d}	23.1 ± 0.53 ^{d}	23.5 ± 0.54 ^{d}	25.8 ± 0.55 ^{c}	37.1 ± 0.51 ^{a}	33.3 ± 0.60 ^{b}	24.1 ± 0.58 ^{cd}
	7	25.9 ± 0.47 ^{d}	25.9 ± 0.74 ^{d}	25.3 ± 0.56 ^{d}	23.9 ± 0.20 ^{d}	24.3 ± 0.86 ^{d}	32.6 ± 0.37 ^{c}	47.8 ± 0.68 ^{a}	40.3 ± 0.47 ^{b}	25.2 ± 0.41 ^{d}
	14	30.8 ± 0.84 ^{d}	31.4 ± 0.71 ^{d}	30.5 ± 0.93 ^{de}	26.6 ± 0.20 ^{e}	28.2 ± 0.98 ^{de}	49.6 ± 0.59 ^{c}	65.7 ± 0.76 ^{a}	53.1 ± 0.65 ^{b}	26.6 ± 0.71 ^{e}
	28	35.7 ± 0.56 ^{e}	43.1 ± 0.59 ^{cd}	45.21 ± 0.40 ^{c}	39.4 ± 0.46 ^{de}	45.8 ± 0.97 ^{c}	102.8 ± 0.60 ^{a}	100.3 ± 0.69 ^{a}	90.0 ± 0.56 ^{b}	36.8 ± 0.71 ^{e}
	42	38.8 ± 0.43 ^{d}	85.6 ± 0.64 ^{bc}	78.9 ± 0.59 ^{c}	85.6 ± 0.71 ^{bc}	50.0 ± 0.72 ^{d}	97.3 ± 0.82 ^{bc}	121 ± 0.89 ^{a}	104 ± 0.80 ^{b}	29.7 ± 0.72 ^{d}
	56	46.6 ± 0.57 ^{f}	75.9 ± 0.67 ^{cde}	77.46 ± 0.97 ^{cd}	66.8 ± 0.62 ^{de}	86.9 ± 0.94 ^{c}	117 ± 0.52 ^{b}	133 ± 0.49 ^{a}	125 ± 0.87 ^{ab}	60.6 ± 0.45 ^{ef}
	70	44.8 ± 0.36 ^{d}	80.0 ± 0.79 ^{c}	78.24 ± 0.63 ^{c}	78.4 ± 0.65 ^{c}	81.2 ± 0.81 ^{c}	119 ± 0.91 ^{b}	140 ± 0.90 ^{a}	134 ± 0.78 ^{ab}	57.9 ± 0.54 ^{d}
	91	47.2 ± 0.39 ^{c}	81.3 ± 0.90 ^{b}	89.7 ± 0.80 ^{b}	79.7 ± 0.83 ^{b}	83.7 ± 0.84 ^{b}	132 ± 0.79 ^{a}	152 ± 0.95 ^{a}	141 ± 0.82 ^{a}	51.6 ± 0.64 ^{c}
Available Phosphorus (mg kg^{-1})	0	2.59 ± 0.00 ^{a}	2.61 ± 0.07 ^{a}	2.50 ± 0.12 ^{a}	2.52 ± 0.05 ^{a}	2.82 ± 0.13 ^{a}	2.89 ± 0.23 ^{a}	2.58 ± 0.19 ^{a}	2.69 ± 0.14 ^{a}	2.75 ± 0.29 ^{a}
	3	2.85 ± 0.31 ^{e}	7.49 ± 0.56 ^{c}	11.4 ± 0.20 ^{b}	4.53 ± 0.36 ^{de}	7.04 ± 0.83 ^{c}	17.1 ± 0.41 ^{a}	4.07 ± 0.34 ^{de}	5.19 ± 0.24 ^{d}	3.22 ± 0.03 ^{e}
	7	2.89 ± 0.24 ^{f}	7.70 ± 0.58 ^{c}	13.0 ± 0.72 ^{b}	4.69 ± 0.41 ^{def}	6.11 ± 0.32 ^{cd}	16.5 ± 0.56 ^{a}	3.90 ± 0.11 ^{ef}	5.53 ± 0.47 ^{de}	2.89 ± 0.17 ^{f}
	14	2.95 ± 0.24 ^{d}	7.29 ± 0.15 ^{b}	13.0 ± 0.44 ^{a}	4.81 ± 0.31 ^{c}	6.10 ± 0.57 ^{b}	13.4 ± 0.84 ^{a}	3.96 ± 0.24 ^{cd}	4.34 ± 0.39 ^{c}	2.79 ± 0.12 ^{d}
	28	2.89 ± 0.18 ^{e}	6.80 ± 0.65 ^{b}	15.2 ± 0.13 ^{a}	4.47 ± 0.32 ^{cd}	6.23 ± 0.53 ^{b}	16.3 ± 0.60 ^{a}	3.71 ± 0.24 ^{de}	5.07 ± 0.40 ^{c}	2.96 ± 0.20 ^{e}
	42	0.76 ± 0.04 ^{f}	3.21 ± 0.16 ^{c}	5.86 ± 0.44 ^{b}	1.53 ± 0.04 ^{ef}	3.03 ± 0.13 ^{cd}	8.35 ± 0.83 ^{a}	1.90 ± 0.04 ^{e}	1.71 ± 0.61 ^{de}	1.09 ± 0.03 ^{ef}
	56	0.46 ± 0.03 ^{f}	2.36 ± 0.22 ^{c}	4.98 ± 0.33 ^{b}	1.08 ± 0.08 ^{de}	1.69 ± 0.11 ^{d}	5.75 ± 0.44 ^{a}	1.35 ± 0.09 ^{de}	1.54 ± 0.18 ^{d}	0.77 ± 0.07 ^{ef}
	70	1.10 ± 0.05 ^{de}	2.33 ± 0.07 ^{c}	4.08 ± 0.42 ^{b}	0.95 ± 0.04 ^{de}	1.38 ± 0.08 ^{d}	5.34 ± 0.52 ^{a}	1.45 ± 0.11 ^{d}	1.31 ± 0.09 ^{de}	0.65 ± 0.05 ^{e}
	91	0.58 ± 0.06 ^{f}	1.71 ± 0.07 ^{c}	3.05 ± 0.10 ^{b}	0.87 ± 0.03 ^{e}	1.29 ± 0.06 ^{d}	3.83 ± 0.11 ^{a}	0.94 ± 0.09 ^{e}	0.93 ± 0.11 ^{e}	0.52 ± 0.04 ^{f}

Significant effects of the applied treatments on each parameter were determined within each soil sampling date using the least significant difference (LSD) with Tukey post hoc multiple comparisons test at $p < 0.05$. For each soil parameter, values across the rows (at each sampling date) with different lowercase letters indicate significant differences among the treatment means. Numbers after the symbol "±" are the standard deviations of triplicate measurements.

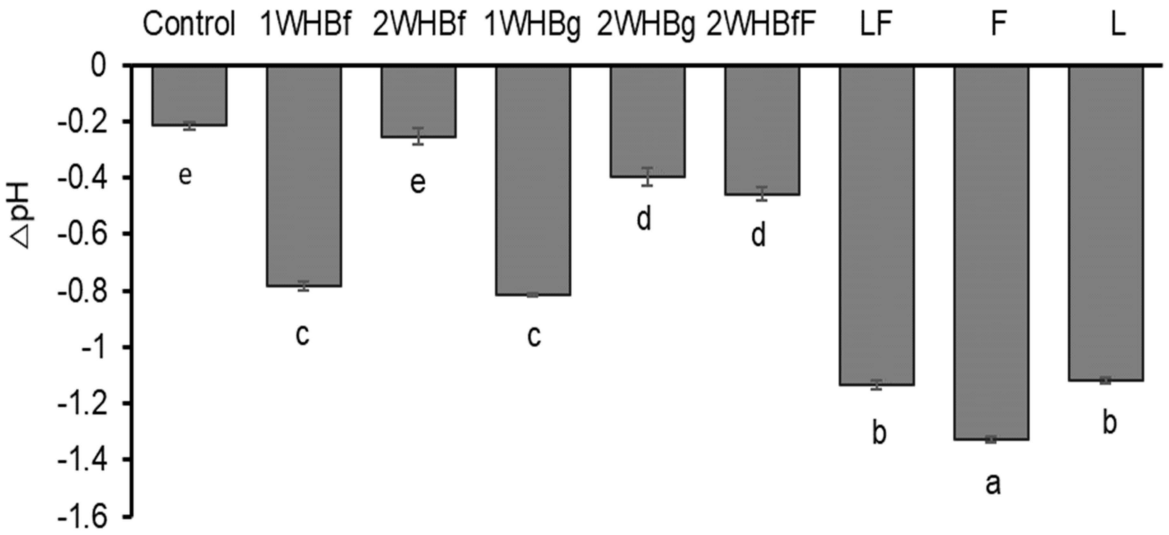

Figure 1. The magnitude of soil pH reduction during the 91 days of incubation due to soil acidification from nitrification of NH_4^+ from the applied fertilizer and/or simulated soil acidification with HNO_3. (Control: soil only; $1WHB_f$ and $2WHB_f$: 1% and 2% applications of WHB produced using furnace; $1WHB_g$ and $2WHB_g$: 1% and 2% applications of WHB produced by grounding method; F: fertilizer at a recommended rate; $2WHB_fF$: $2WHB_f$ combined with fertilizer; L: lime at rate based on the exchangeable acidity of the experimental soil; and LF: lime combined with fertilizer). Error bars are the standard deviations of triplicate measurements. Different letters on the pillars show significant differences among the treatments ($p < 0.05$).

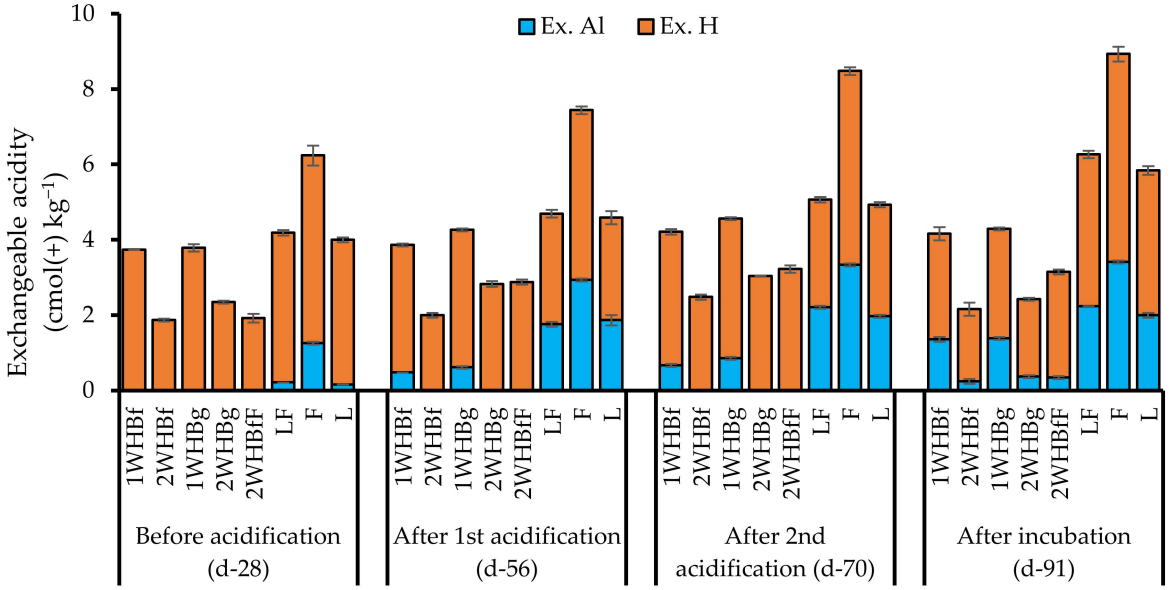

Figure 2. Soil exchangeable acidity (Al3+and H+) before and after acidification of soil amended by water hyacinth biochar (WHB) or lime (L). (1WHBf and 2WHBf: 1% and 2% applications of WHB produced using furnace; 1WHBg and 2WHBg: 1% and 2% applications of WHB produced by grounding method; F: fertilizer at a recommended rate; 2WHBfF: 2WHBf combined with fertilizer; L: lime at rate based on the exchangeable acidity of the experimental soil; and LF: lime combined with fertilizer). Error bars are the standard deviations of triplicate measurements.

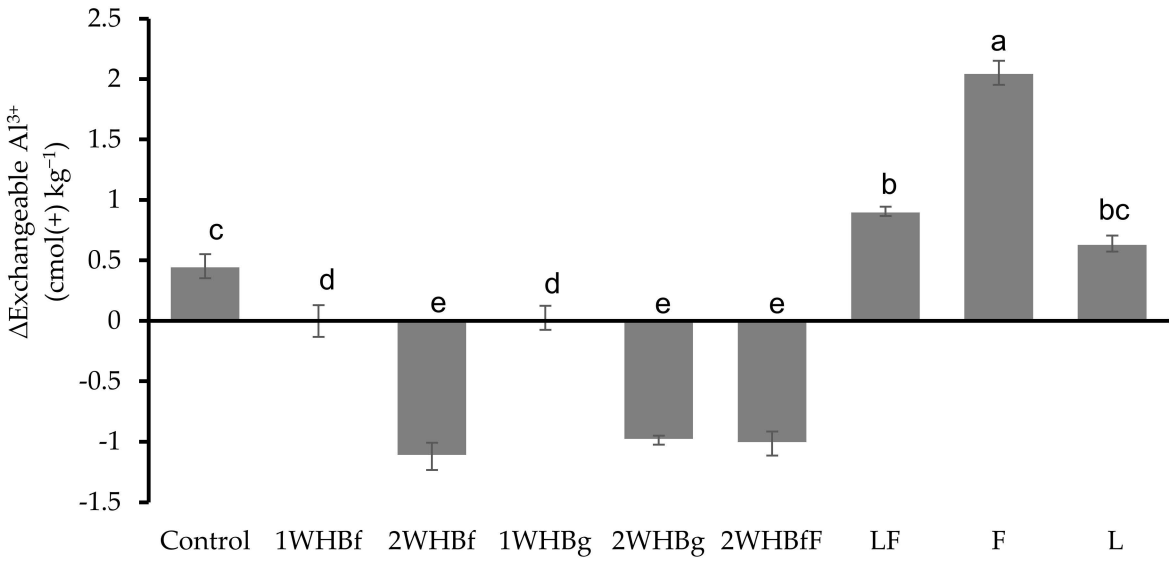

Figure 3. The magnitude of soil exchangeable Al3+ increased/decreased during the 91 days of incubation due to soil acidification from nitrification of NH4+ from the applied fertilizer and/or simulated soil acidification with HNO3. (Control: soil only; 1WHBf and 2WHBf: 1% and 2% applications of WHB produced using furnace; 1WHBg and 2WHBg: 1% and 2% applications of WHB produced by grounding method; F: fertilizer at a recommended rate; 2WHBfF: 2WHBf combined with fertilizer; L: lime at rate based on the exchangeable acidity of the experimental soil; and LF: lime combined with fertilizer). Error bars are the standard deviations of triplicate measurements. Different letters on the pillars show significant differences among the treatments ($p < 0.05$).

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