

## Effect of soil incubation time on some soil parameters and soil quality indexes by using lime, powdered biochar, and biochar extract in acid soil improvement

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### ABSTRACT

Although acid soils with low pH are always of interest to farmers in the farming process, the use of amendments to control pH can affect some other components of the soil. Soil quality index (SQI) and soil parameters including pH(H<sub>2</sub>O), electrical conductivity (EC), total organic carbon content (TOC), available phosphorus (Pav) and ammonium nitrogen (NH<sub>4</sub><sup>+</sup>) were monitored during short-term incubation (60 days) with amendments (lime, solid biochar and liquid biochar extract derived from macadamia husk) under laboratory conditions. The SQI estimation was based on the simple addition method. The results showed that soil parameters such as pH, EC, TOC, available P, and NH<sub>4</sub><sup>+</sup> did not change in a clear trend over the incubation times. However, the results showed that the SQI of acid soil amended with biochar extract was evaluated as more optimal than the other two amendments at all incubation times and maintained at good and very good levels for 35 days of incubation. The findings of the study provide a basis for potential applications of biochar as well as for improving the efficient use of agricultural by-products.

**Keywords:** biochar extract, macadamia husk, incubation time, biochar, dissolved organic carbon.

### INTRODUCTION

The chemical and physical properties of soil vary over short and long periods. Some soil parameters cannot be easily replenished promptly during cultivation and have affected soil quality. This is a global concern for food security (Nasir et al., 2024). In many studies, organic matter, pH, electrical conductivity (EC), nitrogen, and phosphorus are often considered as the main parameters that need to be monitored and used to calculate the soil quality index (SQI) (Nasir et al., 2024).

Acid sulfate soils in Vinh Long province, Vietnam, account for 39.23% of the total area, which can release toxic Al and Fe to plants, reduce pH, and leach nutrients. Most soil samples in this area show low pH, ranging from 3.65–5.4, requiring the use of amendments (Minh et al., 2023; Phuong, 2024). Amendments are commonly applied to reduce soil acidity based on their

alkalinity, such as lime (traditional) and biochar (Addisu and Seyoum, 2021; Pouangam Ngalani et al., 2023). However, excessive or prolonged use of lime can cause precipitation of calcium phosphate (Pouangam Ngalani et al., 2023) and reduce soil available phosphorus.

Biochar is produced by the thermal decomposition of organic waste under oxygen-limited conditions at temperatures ranging from 300 to 700 °C (Liu et al., 2022). In Vietnam, macadamia husks is an agricultural by-product that is discarded, causing environmental pollution (Vu et al., 2023). This is considered raw materials for biochar production.

Recommendations suggest that using too high a dose of solid powdered biochar will reduce its positive effects and may even be harmful to plants. Furthermore, in the form of fine powders, there is also the potential for air pollution. Therefore, an alternative approach could be to use

biochar extracts whose representative component is dissolved organic carbon (DOC). They can directly change the physical and chemical properties of the soil and contribute to increased crop yields (Kujawska et al., 2024; Kumar et al., 2021; Liu et al., 2019).

The DOC components of great interest in the agricultural sector are humic compounds and humic acid-like substances (Liu et al., 2022; Tian et al., 2022). They can retain essential mineral nutrients in the soil, such as ammonium (Boriwal, 2023; Uddin et al., 2021), stabilize soil acidity, and help stabilize organic matter, reduce decomposition rates, and promote long-term carbon storage in the soil (Boriwal, 2023), increase available P (Xu et al., 2021), and reduce exchangeable acidity (Olowoboko et al., 2018).

According to previous research, the use of KOH to extract macca husk biochar pyrolyzed at 300 °C has a higher yield and humic acid-like DOC composition than biochar at pyrolysis temperature of 450 or 600 °C with the same extraction conditions (Van Phuong, 2025). This is considered the basis for selecting biochar 300 and its extract as an acid soil improver in the study.

The addition of lime (traditional solution) or solid powdered biochar has been shown to increase (or change): pH, EC, moisture content, soil organic C, and available N and P (Chaudhry et al., 2024); however, the impact of applying biochar extract solution is unclear (Gupta et al., 2024) and information is lacking. Furthermore, comparing only the changes in soil parameters during the soil incubation process (Van Phuong, 2024) did not show the interaction effects of the investigated parameters. Therefore, estimating the SQI would be more useful and would show more clearly the changes in soil quality during the composting process. Therefore, the objective of this study was to compare and evaluate the effects of using acid

soil amendments including lime, biochar (solid powder) and biochar extract on some parameters of acid soil (soil pH, soil electrical conductivity,  $\text{NH}_4^+$ , available P and TOC) and SQI over time of soil incubation under laboratory conditions. The study aims to improve the understanding and effectiveness of agricultural by-products use in sustainable management.

## MATERIALS AND METHODS

### Sample collection

Soil samples used for the study were collected at coordinates 10°01'45.2"N; 106°05'25.2"E. Depth from 0-30 cm, sampling area with diameter of 10 m, 5 samples were taken at the 4 corners with the center of the diagonal and mixed to get 1 composite sample. This composite sample was then used for both pre-analysis and incubation experiments. Soil samples were air-dried, ground, and sieved through 2 mm. Analysis determined: Bulk density, specific gravity, electrical conductivity (EC), carbon content (TOC), pH, ammonium nitrogen and available P (PAv) in soil. The results of these parameters were presented in Table 1.

Determine soil particle density according to TCVN 6863:2000, determine soil pH according to ISO 10390:1993, and soil EC is measured with an EC meter (Hanna) in soil-water mixture at a ratio of 1:5 (Nasir et al., 2024), bulk density of soil according to TCVN 8305:2009, organic carbon (TOC) of biochar and soil according to the Walkley Black method (Tan, 2005), available phosphorus according to Olsen method,  $\text{NH}_4^+$  determined according to TCVN 6179-1:1996, particle size distribution determined by the hydrometer method (Tan, 2005).  $\text{K}_2\text{Cr}_2\text{O}_7$ , NaOH,  $(\text{NH}_4)_2\text{Fe}(\text{SO}_4)_2$ ,  $\text{NH}_4\text{Cl}$ ,  $\text{HNO}_3$  were from Merck, and KCl, HCl,  $\text{NaH}_2\text{PO}_4$ ,  $\text{H}_2\text{O}_2$  from China

**Table 1.** Some soil properties

Parameters	Result	SD
Bulk density, $\text{g/cm}^3$	1.01	0.01
Particle density, $\text{g/cm}^3$	1.85	0.12
Soil type	Clay loam	
pH	4.50	0.03
EC (S/cm)	0.58	0.03
TOC, %	3.15	0.11
Available phosphorus content, mg $\text{P}_2\text{O}_5/\text{kg}$	60.9	25
$\text{NH}_4^+$ content, mg/kg	21.4	2.5

**Note:** SD: Standard deviation.

### Experimental setup

#### Preparation for biochar extract

Macadamia husks were pyrolyzed at 300 °C, heating rate 10 °C/min, holding time 2 h, cooled overnight, and ground through a 1 mm sieve. Some properties of biochar were detailed in a previous study (Van Phuong, 2025).

Preparation of 10% biochar extract solution: The extraction conditions were carried out according to previous studies. Specifically, accurately

**Table 2.** Formula of ingredients of the treatments

Treatments	Experimental materials
Lime	Soil + Lime 0.2%
Biochar, (Bio)	Soil + 0.2% biochar
Biochar extract, (Bio-Ex)	Soil + 2 g of extraction solution (equivalent to 0.2% biochar)

weigh 40 grams of biochar (at pyrolysis temperatures of 300 °C) and put it into a 500 mL glass beaker containing 400 mL of 1M KOH solution. The mixture was stirred and ground on a homogenizer at 6000 rpm for 2 hours. Centrifuge and quickly filter through a 45 µm filter (Van Phuong, 2025). Use 1N H<sub>2</sub>SO<sub>4</sub> acid to convert the extract to pH 8. Store the sample in dark polyethylene bottles and seal it for use in subsequent studies.

#### Soil incubation experiment in the laboratory

The incubation process was simulated according to Mosharrof et al. (2021). The treatments are described in details in Table 2.

There were 3 experimental treatments, each treatment had 4 soil incubation periods, 3 replications. The dosage of improving agents was the minimum drawn from preliminary experiments to ensure soil pH > 5, suitable for the growth characteristics of orange trees. Specifically, 50–60 mL of water containing improving agents (pre-mixed) was put into a polyethylene cup containing 100 g of air-dried soil, and mixed well. All treatments were incubated at 27 ± 2 °C for 60 days, Table 2. The water holding capacity was maintained at 60% constant throughout the incubation period by adding demineralized water every 3 days throughout the experiment based on weight loss. Observations were recorded at 7, 14, 21, 35, and 60 days. After incubation, the soil samples were air-dried and ground through a 2 mm sieve to determine pH, EC, TOC, ammonium, and available P. In the study, the lime treatment (traditional solution) can be considered as a control study.

#### Data processing

##### Calculating SQI

In this study we use a simple estimation technique to calculate SQI, Equations 1 and 2.

$$\sum SQI = \sum S_i \quad (1)$$

The SQI index is calculated according to Equation 2

$$SQI = \frac{\sum S_i - SQI_{min}}{SQI_{max} - SQI_{min}} \quad (2)$$

In which,  $S_i$  is calculated based on Equations 3, 4, and 5 (Bandyopadhyay and Maiti, 2021; Kahsay et al., 2025; Nasir et al., 2024).  $S_i$  is calculated in the case of “Less is more”

$$S_i = LSF = \frac{X}{X_{max}} \quad (3)$$

“More is more”

$$S_i = LSF = \frac{X_{min}}{X} \quad (4)$$

“Optimum Point”

$$S_i = NLSF = \frac{a}{\left[1 + \left(\frac{X_i}{X_{imean}}\right)^b\right]} \quad (5)$$

where:  $a$  – maximum value is 1,  $X_i$  – value of soil sample index  $i$ ,  $X_{imean}$  – mean value of soil sample index  $i$ ,  $b$  – The slope of the equation, and is -2.5 for the “more is better” curve and +2.5 for the “less is better” curve.

SQI is classified according to the scale: very low (0–0.19); low (0.20–0.39); average (0.4–0.59); good (0.6–0.79), and very good (0.8–0.99) (Damiba et al., 2024).

A one-way ANOVA was conducted to test the effect of different soil incubation times on the experimental treatments. Significant differences among the treatments (lime control, solid biochar powder, and biochar extract) were then identified using the Tukey post hoc test at a 95% significance level ( $p \leq 0.05$ ), with all analyses performed in SPSS 23.

## RESULTS AND DISCUSSION

### Impact on soil pH (H<sub>2</sub>O)

The results of the survey soil incubation time with improving substances on soil pH, Figure 1, showed that pH changed insignificantly from the 7th to the 14th day of incubation (Figure 1A) but increased rapidly on the 21st day, and lasted until the 35th day. Specifically, pH was 5.38, 5.38, 5.99, and 6.01, respectively at the survey times, and decreased sharply to 5.35 on the 60th day.

In the case of biochar amendment, Figure 1B shows that the soil sample pH (pH 4.55) increased on day 7 (pH 5.42) and remained stable until day 35, fluctuating between 5.37 and 5.41, a change that was insignificant according to One-way ANOVA analysis. Then, the pH decreased significantly to 5.19 on day 60.

With the biochar extract solution, (Figure 1C), the soil pH remained high from day 7 to day 21, corresponding to the pH value range of 5.84–5.88. Then, the pH decreased until the 60th day of incubation, to 5.40 and 5.26, respectively; the decrease was significant.

In general, the use of lime, biochar, or biochar extract increases soil pH. The soil pH value changes with the incubation time, depending on the type of improver used. However, the results obtained all show that on the 60th day, it is lower than on the previous incubation days. This can be explained, the long incubation time may have continued reactions with protons deep in the soil particles; moreover, it may be due to the nitrification process, releasing acid. The study by Mkhonza et al. (2020) also gave similar results.

With lime amendment, the alkalinity of lime neutralizes protons and exchangeable Al (Addisu and Seyoum, 2021). However, as the incubation time was extended, the pH decreased again, which could be explained by the continued reaction between lime and protons inside the soil particles. The study of Mosharrof et al. (2021) also found that lime addition increased the pH of the soil solution.

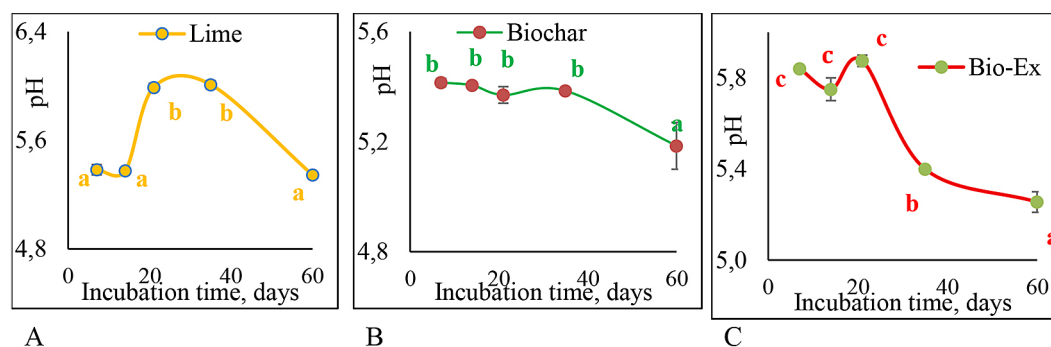
With the use of biochar and extract, the pH value remained stable for 7–35 days. This can be explained by the ability to form complexes with Fe, Al from organic sites in biochar or extract, which increases the ability to maintain pH (Pouangam Ngalani et al. 2023; Xu et al. 2021).

The study of Gupta et al., also suggested that biochar application maintained stable pH in acidic soils (Gupta et al., 2024)

### Impact on soil EC

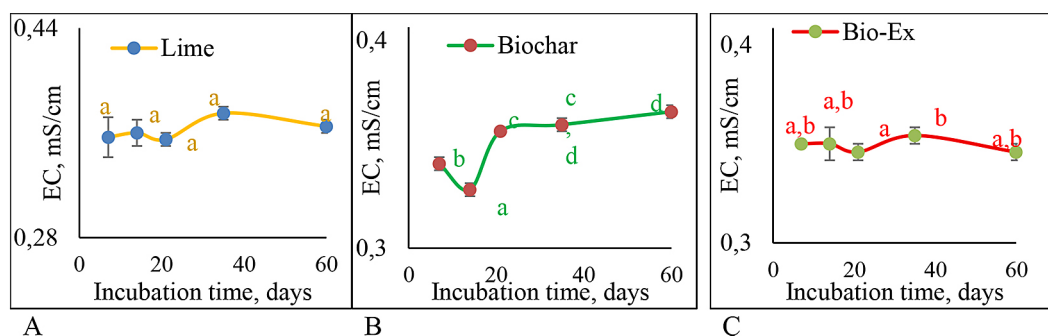
Using lime as an amendment to control EC of the studied soil samples showed that EC (from 0.58  $\text{mS cm}^{-1}$ ) decreased, the decrease was insignificant according to One-way ANOVA analysis and remained stable throughout the incubation process, fluctuating between 0.36 and 0.38  $\text{mS cm}^{-1}$ , (Figure 2A). This may be due to precipitation reactions that occurred due to alkalinity or Ca, Mg ions causing a reduction in the electrolytic properties of the soil solution (Addisu and Seyoum, 2021). There is also the assimilation of ions released by lime dissolution and the loss of protons from charged sites of the soil (Bramble et al., 2019). When using biochar to improve the soil, EC decreased sharply to 0.33  $\text{mS cm}^{-1}$  on the 14th day. Then it increased significantly on the 21st day (0.7  $\text{mS cm}^{-1}$ ) and remained stable until the 60th day of incubation (ranging from 0.37 to 0.39  $\text{mS cm}^{-1}$ ), (Figure 2B). The sharp decrease in EC may be due to some minerals adsorbed and adhered to the biochar surface. In case of an increase again, it may be due to organic matter on the biochar surface competing for mineralization sites in the soil. The results are consistent with the reports of Thite et al. (2022) and Van Phuong (2024).

In the case of using biochar extract, the EC value decreased and remained stable during the 60-day incubation period, (Figure 2C). This can be explained due to P mineral salts bind Al and Fe ions due to the formation of insoluble forms. In addition, organic substances in the extract can form complexes with Fe and Al, reducing the electrolysis (Mkhonza et al., 2020; Van Phuong., 2024).



**Figure 1.** Changes in soil pH over time following amendments application (the letters a, b, c indicate significant differences): (A) lime, (B) biochar, (C) biochar extract solution





**Figure 2.** Changes in soil EC over time following amendments application (the letters a, b, c, d indicate significant differences): (A) lime, (B) biochar, (C) biochar extract solution

### Impact on soil TOC

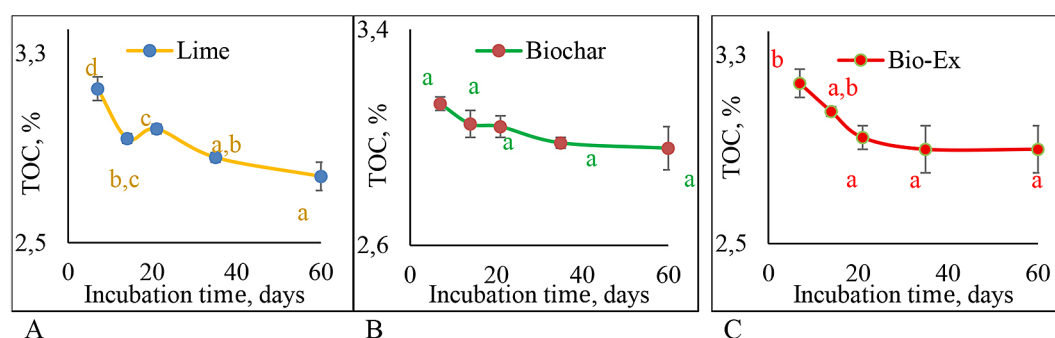
The effect of lime application on soil amendment is shown in Figure 3A. The results showed a significantly rapid decrease in TOC after 14 days of incubation, specifically, TOC from 3.15% (Table 1) decreased to 2.94% (7% loss). Then it continued to decrease to 2.78% after 60 days of incubation (another 5% loss). This can be explained due to when lime is added, the  $\text{CO}_2$  content in the soil decreases, shifting the equilibrium, increasing the ability to decompose organic matter. The study of Bramble et al., also found a similar argument (Bramble et al. 2019).

In the case of using biochar or biochar extract, the TOC of soil samples decreased slowly with a decrease ranging from (3.15–2.96%) to (3.15–2.90%), with a loss of about 6–8% of organic matter after 60 days of incubation, (Figure 3B and 3C). The decrease was lower than in the case of using lime, possibly due to a small amount of organic matter added from biochar and extract. Furthermore, as reported by Boriwal, it was suggested that biochar extract stabilized organic matter, reduced decomposition rate, and promoted long-term carbon storage in soil (Boriwal, 2023).

However, a reduction was noted (the reduction was not statistically significant), possibly due to mineralization by microbial activity (Pouangam Ngalani et al., 2023).

### Impact on soil available phosphorus

The investigation of the effects of lime, biochar, and biochar extract amendments on available phosphorus in soil samples is presented in Figure 3. In the case of lime application, it was found that available phosphorus increased rapidly from day 7 to day 14 corresponding to the available phosphorus content from 75 to 105 mg/kg, a significant increase. Then the available phosphorus content decreased to 96, 84, and 88 mg/kg, respectively at days 21, 35, and 60, (Figure 4A). The significant increase can be explained due to lime improves pH in acidic soils, weakens Al-P, Fe-P bonds, leading to increased P availability in soil. Similar results were also found in previous studies (Lima et al., 2021; Morales et al., 2021; Van Phuong, 2024). Then, it gradually decreased until day 60, possibly due to the mineralization processes of organic matter in the soil, reducing pH and soluble P re-precipitating with Al and Fe ions in the soil



**Figure 3.** Changes in soil TOC over time following amendments application (the letters a, b, c, d indicate significant differences): (A) lime, (B) biochar, (C) biochar extract solution

(Van Phuong, 2024). The study of Lima et al, also showed that biochar addition to soil significantly increased available P (Lima et al., 2021)

Figure 3B showed that when biochar was added to the soil sample, available P changed over the incubation time. Specifically, available P increased from 84 to 123 mg/kg, a significant increase corresponding to the incubation time from 7 to 14 days. Then, it decreased rapidly from 123 to 86 mg/kg, corresponding to the incubation time from 14 to 21 days, a statistically significant decrease. The available phosphorus content continued to remain stable until the 60th day of incubation. This can be explained due to the biochar surface contains anionic functional groups, such as carboxylic and hydroxide groups, that compete for phosphorus sites on the soil particle surface (Fink et al., 2016).

When using biochar extract to improve the research soil samples, it showed that the available phosphorus increased continuously until the 21st day, the increase was significant at the sampling times, (Figure 4C). Specifically, the available phosphorus increased by 75, 81, and 101 mg/kg respectively at the sampling times of 7, 14 and 21 days. After the 21st day, the change was insignificant but still maintained at a high content of available phosphorus, fluctuating at a value of 96–101 mg/kg. This showed that the organic anions in the extract replaced P bound to Al, Fe in the soil, increasing the available phosphorus. (Lima et al., 2021; Xu et al., 2021).

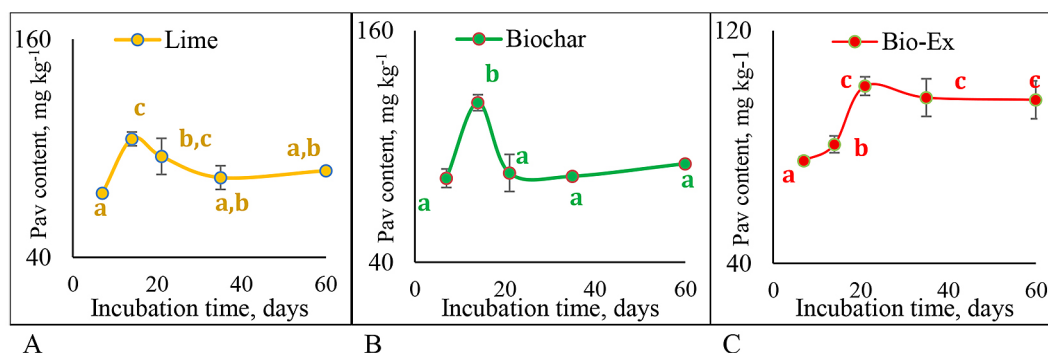
Compared with the initial soil sample's available phosphorus content (60.9 mg/kg), Table 1, the addition of lime, biochar, or biochar extract all improved the available phosphorus content (Figure 4) (Xu et al., 2021)

## Impact on $\text{NH}_4^+$

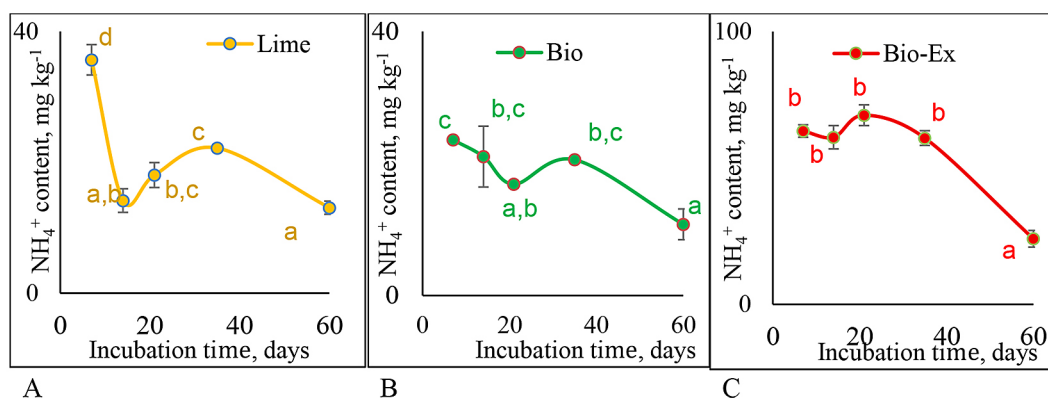
When using lime to improve acidic soil samples, the results showed that the exchangeable  $\text{NH}_4^+$  content increased in the first week of soil incubation (35.4 mg/kg), then decreased/increased without a clear trend, respectively 35.7, 14.2, 18.0, 22.2 and 13.1 mg/kg for the sampling days 7, 14, 21, 35 and 60, (Figure 5A). This can be explained due to the additional lime increased the pH, increased microbial activity, leading to increased organic nitrogen decomposition. In the case of a decrease in the exchangeable  $\text{NH}_4^+$  content, it could be due to the nitrification process. The increasing/decreasing trend was not clear, similar to Van Phuong's study (2024).

With biochar, a similar trend was also shown, specifically the  $\text{NH}_4^+$  content decreased/increased by 23.6, 21.0, 16.9, 20.6 respectively on survey days 7, 14, 21 and 35 and decreased deeply by 10.8 mg/kg on the 60th day, (Figure 5B). With the biochar extract, the  $\text{NH}_4^+$  exchange increased and remained for 35 days of incubation, fluctuating from 61.0–69.3 mg/kg, the fluctuation was insignificant. Then it decreased significantly on the 60th day (10.8 mg/kg), (Figure 5C). The reason may be that biochar possesses a large number of surface functional groups that can dissociate to create negatively charged ions, which can absorb  $\text{NH}_4^+$  cations (Xu et al., 2021)

The treatment using biochar extract (Figure 5C) showed that the  $\text{NH}_4^+$  content in the soil sample remained at a high level, ranging from 61.0 to 69.3 mg/kg, and the change was not statistically significant. Then, it decreased sharply on the 60th incubation day (24.0 mg/kg). This can be explained due to the organic anions in the extract strongly bind with  $\text{NH}_4^+$  on soil colloidal



**Figure 4.** Changes in soil available phosphorus over time following amendments application (the letters a, b, c, d indicate significant differences): (A) lime, (B) biochar, (C) biochar extract solution. Pav: available phosphorus content



**Figure 5.** Changes in soil  $\text{NH}_4^+$  over time following amendments application (the letters a, b, c, d indicate significant differences): (A) lime, (B) biochar, (C) biochar extract solution

particles, strongly increasing the dissolved  $\text{NH}_4^+$  and also increasing the stability of the exchangeable  $\text{NH}_4^+$  (Xu et al., 2021).

After 35 days of incubation,  $\text{NH}_4^+$  decreased sharply, possibly due to the dominance of nitrification during incubation (Bramble et al., 2019). Except, TOC decreased with the composting time when using amendments. Soil parameters such as pH, EC,  $\text{NH}_4^+$ , and available P, when examined with composting time, fluctuated without a clear trend. Therefore, estimating SQI will give a more comprehensive assessment result for soil improvement purposes.

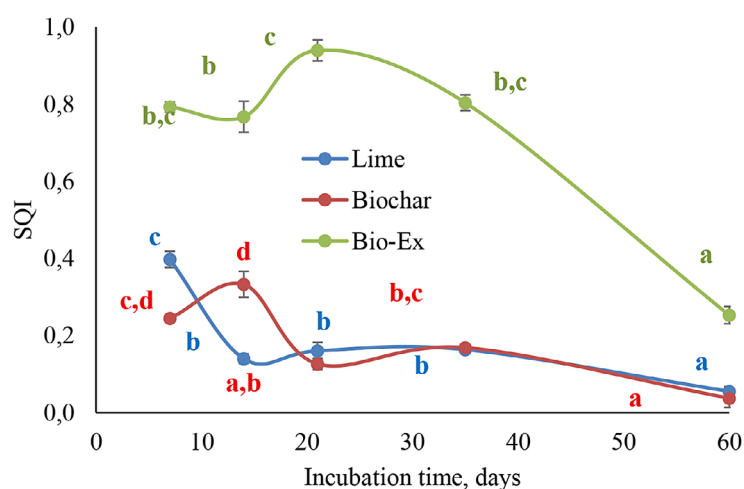
### Impact on soil quality index

In the study we use simple estimation techniques to calculate SQI, Equations 1 and 2 (Nasir et al. 2024). With the lime treatment SQI decreased rapidly on day 7 from 0.40 to 0.14 (day

14), the decrease was significant and remained stable, fluctuating from 0.14–0.17 to day 35. Then it decreased to 0.06 on day 60. The results showed that lime addition could only maintain high SQI after 7 days of incubation, then decreased rapidly (Figure 6).

With the biochar treatment, the SQI on the 14th day of incubation increased significantly (0.33) compared to day 7 (0.24). Then it decreased sharply on day 21 (0.13), the decrease was statistically significant and fluctuated stably until day 35 before decreasing to 0.04 on day 60. The results showed that the SQI reached its highest on the 14th day of incubation and then decreased similarly to the lime treatment, (Figure 6).

In contrast to the two treatments using lime and solid powdered biochar, the treatment supplemented with biochar extract maintained a very high SQI. The SQI value reached 0.8 from the 7th to the 14th day of incubation, continuing to



**Figure 6.** SQI values according to incubation time with amendments, letters a, b, c, d in the same color show that the average SQI values are significantly different

increase significantly from the 21st day of incubation (0.9). Then the SQI decreased significantly until the 60th day (0.3), (Figure 6).

The use of three amendments (lime, biochar, and biochar extract) had a very clear impact on soil quality based on SQI estimates. Compared to the use of lime or biochar, the use of biochar extract gave good to very good quality over a 35-day incubation period, while the use of lime and biochar showed low and medium quality indexes according to the scale of Damiba et al. (2024).

## CONCLUSIONS

Three amendments including lime, biochar, and biochar extract derived from macadamia husk, were used in the study. The survey of the effects of incubation time of acid soil samples in Vinh Long with the amendments showed that soil parameters such as pH, EC, TOC, ammonium nitrogen, and available phosphorus did not change according to a clear trend. However, when estimating SQI, the results showed that the use of biochar extract significantly increased the SQI at all incubation times compared to the other two amendments. Furthermore, SQI was also maintained at good and very good levels after 35 days of incubation. The study confirmed that the type of amendment used and the incubation time affected the changes in soil parameters (pH, EC, TOC, available phosphorus, and ammonium). Furthermore, the study also found that the advantage of using biochar extract improved the soil quality index, which is the basis for increasing the value of using agricultural by-products.

However, the results of the controlled laboratory experiments only focused on a few soil parameters. In the actual field conditions, there are also differences compared to the laboratory experiments. Therefore, field studies will be the key factor to more accurately assess the feasibility of the study.

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