

Caraka Tani: Journal of Sustainable Agriculture, 39(1), 117-139, 2024

URL: https://jurnal.uns.ac.id/carakatani/article/view/80217

DOI: http://dx.doi.org/10.20961/carakatani.v39i1.80217



Effect of Different Types of Biochar Applications and Phosphate Fertilizer on the Quality and Yield of Edamame Soybeans on Andisols

Ramdhana Karimah, Benito Heru Purwanto*, Eko Hanudin, Sri Nuryani Hidayah Utami and Margi Asih Maimunah

Department of Soil Science, Faculty of Agriculture, Universitas Gadjah Mada, Yogyakarta, Indonesia *Corresponding author: benito@ugm.ac.id

Abstract

Edamame soybean (Glycine max (L.) Merr.) cultivation in phosphorus-limited Andisols presents a formidable challenge due to restricted phosphorus availability despite high phosphorus retention. Unlocking the full potential of this crop demands innovative solutions. This study delves into the transformative effects of biochar and phosphorus fertilizer, individually and synergistically, on edamame soybean growth in Andisols. Employing a randomized complete block design, researchers investigate three types of biochar (B0: control, B1: biochar pellets, B2: biochar powder) and four phosphorus fertilizer rates (P0: control, P1: 27 kg ha⁻¹ P₂O₅, P2: 54 kg ha⁻¹ P₂O₅, P3: 81 kg ha⁻¹ P₂O₅). The bamboo-derived biochar was produced using the Kon-tiki method at ±500 °C. The study reveals no significant interaction between biochar and phosphorus fertilizer. Individually, treatments with B1, B2, and phosphorus fertilizers significantly enhance ammonium, nitrate, and phosphorus availability compared to B0 and P0. Biochar-induced modifications improve phosphorus and nitrogen absorption by roots, resulting in increased shoot dry weight and the root/shoot ratio. However, the number of leaves is solely influenced by phosphorus fertilizer treatment. Additionally, both biochar and phosphorus fertilizers contribute to nitrate reductase activity, root volume, an increased number of pods per plant and higher protein content in edamame soybeans. B2 outperforms B1 and high P3 intensifies this effect, improving nutrient uptake and yield. In summary, biochar and phosphorus fertilizers demonstrate significant potential to revolutionize edamame soybean cultivation in phosphorus-limited Andisols, optimizing pod number per plant and enhancing quality with elevated protein content.

Keywords: Andisols; biochar; edamame; nitrate reductase activity; phosphorus fertilizer

Cite this as: Karimah, R., Purwanto, B. H., Hanudin, E., Utami, S. N. H., & Maimunah, M. A. (2024). Effect of Different Types of Biochar Applications and Phosphate Fertilizer on the Quality and Yield of Edamame Soybeans on Andisols. *Caraka Tani: Journal of Sustainable Agriculture*, *39*(1), 117-139. doi: http://dx.doi.org/10.20961/carakatani.v39i1.80217

INTRODUCTION

Vegetable soybean (*Glycine max* (L.) Merr.), also known as "edamame soybean", has been widely consumed in Asia, particularly in Japan, for many years (Mozzoni and Chen, 2019). This versatile crop can thrive and yield good harvests in various Asian countries such as Thailand, India, China and Indonesia. However, with the expanding demand for edamame soybean in the food market, there is a need for greater

efforts to increase both the production area and product quality (Rohmawati and Ulfah, 2018). Edamame is a highly nutritious vegetable rich in high-quality protein, isoflavones, vitamins (C and E), monounsaturated fatty acids, minerals and dietary fiber (Yu et al., 2022).

The nutrient availability in the soil significantly influences the quality and protein composition of soybeans. Nitrogen, a crucial

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^{*} Received for publication November 6, 2023 Accepted after corrections December 30, 2023

element, plays a vital role in chlorophyll and enzyme formation, regulating physiological processes in soybean plants. It is essential for amino acid synthesis, the building blocks of and contributes to carbohydrate proteins metabolism. Nitrogen stimulates root growth and enhances nutrient absorption (Bagale, 2021). Soybean plants typically obtain nitrogen from inorganic sources like ammonium and nitrate in the soil. Nitrate reductase is a key enzyme in nitrate assimilation, catalyzing the reduction of nitrate to nitrite (Vans and Nason, 1953; Mohn et al., 2019). Plants utilize ammonium (NH₄⁺–N) and nitrate (NO₃-N) as nitrogen sources. Nitrate must be reduced to ammonium in plant tissue before it is synthesized to amino acids, while immediately ammonium is directly and synthesized to amino acids after its uptake. Plant deficient of phosphorus decreased nitrate uptake through roots (Rufty et al., 1990), resulting in decreased radicular adenosine triphosphate (ATP) availability and limitations in the synthesis of membrane transport systems for nitrate (Rufty et al., 1993), on the other hand, the increase in nitrate reductase activity (NRA) has improved the efficiency of crop nutrient uptake (Purwanto et al., 2021). NRA directly mirrors plant nitrogen uptake and utilization (Chen and Huang, 2020). Frequently showing correlation with growth and yield (Srivastava, 1980).

Phosphorus, a vital nutrient for soybean crops, is essential for their optimal growth and development. It plays a crucial role in storing and transporting energy generated through photosynthesis, supporting key processes like development growth, and reproduction. Phosphorus is a fundamental component of ATPs, energy carriers that enhance the effectiveness of other nutrients. The application of phosphorus fertilization affects various aspects of soybean crops, including yield, protein and oil content, nitrogen fixation, root expansion, leaf area and stress tolerance (Kakar et al., 2002; Tanwar and Shaktawat, 2003; Adeli et al., 2005; Schulze et al., 2006; Chaudhary et al., 2008; Okunade et al., 2023). Insufficient phosphorus availability poses a significant challenge for legume plants in symbiotic relationships with rhizobia, impacting nodules and nitrogen fixation (Cassman et al., 1981; Tang et al., 2001; Olivera et al., 2004). Nodules use energy-intensive metabolic steps to fix atmospheric nitrogen, maintaining phosphate homeostasis. However, phosphorus deficiency, especially in the plant cell fraction, leads to reduced ATP concentration and energy charge,

causing significant declines in nitrogenase activity (Sa and Israel, 1991; Olivera et al., 2004; Le Roux et al., 2006). Optimizing edamame growth relies on precise soil nutrient management. Conversely, phosphorus shortage harms soybean quality and nutrient absorption (Win et al., 2010). Thus, edamame's growth, output and quality hinge on effective phosphorus uptake, while inadequate soil phosphorus availability significantly affects soybean output and quality.

Andisols. widely distributed throughout exhibit Indonesia. remarkable fertility characteristics that make them promising soils for cultivating edamame soybeans. However, these soils face a fertility challenge due to the limited accessibility of phosphorus, caused by the high fixation capacity of reactive amorphous minerals such as allophane, imogolite, proto-imogolite and ferrihydrite (Neall, 2006). Research by Takahashi (2007) has shown that long-term wheat experiments on low-humic Andisols achieved only about 17% efficiency in using phosphorus fertilizer. Soybeans, being a P-dependent crop, respond to the application of phosphorus fertilizer by increasing phosphorus content in their leaves, stems and seeds. Different soybean cultivars exhibit varying levels of efficiency in phosphorus uptake under low-P soil conditions (Ao et al., 2014). Adding phosphorus fertilizer is crucial for edamame soybeans in Andisols with limited phosphorus. However, it is important to note that conventional fertilizer use poses environmental and economic challenges (Reijnders, 2014). Global demand for phosphorus fertilizer is projected to reach 49.10 million tons by 2022 (FAO, 2019), with depleting non-renewable phosphorus rock and high-quality phosphate reserves expected to be exhausted in 50 to 400 years (van Dijk et al., 2016). Excessive phosphorus fertilizer application increases the risk of phosphorus loss through leaching, runoff and erosion (Li et al., 2019), contributing to eutrophication that harms aquatic ecosystems globally (Chen et al., 2018).

Biochar, produced through biomass thermal processing with limited oxygen, enhances agronomic and environmental outcomes (Lehmann and Joseph, 2009). The porous structure of biochar, including macropores and channels, facilitates nutrient and water storage, benefiting soil microorganisms and promoting nutrient availability and fertilizer efficiency for plants (Joseph and Munroe, 2017; Kim et al., 2017; El Sharkawi et al., 2018; Gwenzi et al.,

2018; Zhu et al., 2019). Biochar, initially low in nutrients, requires nutrient incorporation to enhance its effectiveness as a fertilizer. Recent studies explore slow-release fertilizers utilizing biochar as a carrier (Cen et al., 2021; Sepúlveda-Cadavid et al., 2021; Zhou et al., 2021), offering a promising approach to address economic and environmental challenges linked to conventional fertilizer overuse. Utilizing biochar enriched with phosphorus emerges as an effective method to decrease soil phosphorus retention, boost phosphorus availability for plants (Wali et al., 2020) and enhance the production, growth, and development of soybeans (Zhu et al., 2019; Imran et al., 2022). Consequently, the adoption of slowrelease fertilizers containing biochar presents a viable strategy for promoting sustainable agriculture.

Challenges arise in applying biochar powder to soil due to the vulnerability of its fine and lightweight particles to wind dispersion. This dispersion not only raises concerns about air pollution but also poses a risk of unintentional loss of biochar intended for soil improvement. The associated risks involve the airborne transport of particles, potential inhalation hazards and the inadvertent reduction of the benefits of soil enhancement. In Québec field trials, it was reported that a substantial 30% loss of biochar powder occurred when applied with a lime spreader, resulting in wind-blown dust (Husk and Major, 2010). To address this issue, modifying biochar into pellets, which are larger and heavier compared to the powder form, has proven effective in reducing losses during soil application (Kim et al., 2014). Therefore, it is crucial to develop improved biochar materials that provide long-term nutrient supply with minimal loss. Recent research has focused on utilizing polyvinyl alcohols (PVA) as coating materials to achieve slow-release nutrient characteristic. The polymer network structure of PVA regulates water absorption into the biochar pores and facilitates nutrient release into the soil (Gwenzi et al., 2018; Liu et al., 2019). This polymer network structure holds the potential to control water absorption into the biochar pores and facilitate nutrient release into the soil (Liu et al.,

While several studies have investigated the effects of fertilization on grain yield in edamame soybeans, there is still a need for research specifically focusing on yield and protein concentration. This study hypothesized that phosphorus-enriched biochar could effectively

enhance plant phosphorus uptake, leading to improved crop yields and protein content in edamame soybeans. Therefore, the objective of this study was to examine the effects of phosphorus-enriched biochar pellets and powder on plant growth performance, yield characteristics, and protein content in edamame soybeans cultivated in Andisols with low phosphorus availability.

MATERIALS AND METHOD

Study area

The research was conducted in Ngablak, Magelang Regency, Indonesia (110°23'13.4" E. 7°22'44.7" S), at an altitude of 1,205 meters above sea level, from August 2020 to January 2021. The research area experiences a mean annual temperature of 25.62 °C and annual rainfall of 3,922 mm year⁻¹. The soil was classified as Andisols and had a loam texture, comprising 35.0% silt, 48.0% sand and 20.2% clay. It exhibited a gentle slope and maintained a relatively uniform level of soil fertility. The main soil chemical properties include a pH H₂O of 6.47, pH NaF of 11.10, cation exchange capacity (CEC) of 23.86 cmol(+) kg⁻¹, organic carbon content of 118.23 g kg⁻¹, total nitrogen content of 2.45 g kg⁻¹, ammonium content of 22.28 mg kg⁻¹, nitrate content of 46.67 mg kg⁻¹ and available phosphorus content of 0.013 mg kg⁻¹.

Experimental design

The experiment was planned using a factorial randomized complete block design (RCBD) with two treatment factors and three replications. The first factor was three types of biochar (B): 1) control without biochar amendment (B0), 2) phosphorus-enriched biochar pellets (B1), and 3) biochar powder (B2), all applied at a rate of 20 tons ha⁻¹. The second factor (P) represented the phosphorus fertilizer rate: 1) control with no phosphorus fertilizer (P0), 2) 27 kg ha⁻¹ P₂O₅ (P1), 3) 54 kg ha⁻¹ P_2O_5 (P2) and 4) 81 kg ha⁻¹ P_2O_5 (P3). For the treatment involving biochar powder (B2), the phosphorus fertilizer solution was applied simultaneously with the application. This resulted in a total of 12 treatment combinations, each replicated three times, resulting in 36 experimental units, including the control.

The experimental plots measured 1 m x 1.8 m with a bed height of 20 to 25 cm. Edamame soybean var. Ryoko 75 was planted with a spacing of 20 cm x 20 cm. Ten days prior to planting, the biochar was manually incorporated into

the soil layer at a depth of 0 to 10 cm. In the case of the biochar powder treatment (B2), the soil was sprayed with a phosphorus fertilizer solution, followed by the application of mulch to cover the entire treatment. To mitigate the potential dispersal and cross-contamination of biochar caused by wind into adjacent research plots, it is recommended to apply biochar in close proximity to the soil surface. This strategy is designed to enhance the adherence of biochar particles to the soil, thereby reducing the risk of wind-driven dispersion and ensuring the effectiveness of the application in the specified area.

Mineral fertilizer was applied twice, at 3 weeks after planting (WAP) with 50 kg ha⁻¹ urea and 50 kg ha⁻¹ ZK, and at 6 WAP with 25 kg ha⁻¹ urea, 50 kg ha⁻¹ ZA and 50 kg ha⁻¹ ZK. No additional phosphorus fertilizer was added during the plant growth stage. The plants were harvested 85 days after planting according to the respective treatment combinations. Pesticide application and hand weeding were performed as necessary during the plant growth period.

Biochar powder production and pelletization

Biochar prepared from bamboo was method (Gigantochloa apus) by Kon-tiki following the procedure of Schmidt and Taylor (2014), at a temperature of ± 500 °C. The biochar used in this study was prepared by pulverizing it through a milling machine and passing it through a ± 2 mm, resulting in biochar powder. For the production of phosphorus-enriched biochar pellets, a modified method based on Liu et al. (2019) was employed. In this process, different amounts of KH₂PO₄ (9.35 g for P1, 18.70 g for P2 and 28.04 g for P3, equivalent to 27, 54 and 81 kg ha⁻¹ P₂O₅, respectively) were mixed with 9 1 water. This solution was then added to bamboo biochar (3,600 g equivalent to 20 tons ha⁻¹), bentonite (270 g) and PVA (180 g). After an incubation period of approximately 24 hours, the phosphorus-enriched biochar pellet was sun-dried. During the pelletization process using a pelletizer machine, cassava starch (198 g) was added. The resulting biochar pellets were air-dried, sealed and stored for future use. The biochar pellets exhibit a uniform cylindrical shape, measuring 8 mm in diameter and 27 mm in length. In terms of chemical composition, different biochar samples (B2, B1P0, B1P1, B1P2. and B1P3) demonstrate distinct characteristics. These include pH H₂O values of 10.15, 9.90, 9.44, 9.37 and 7.60, respectively. Moreover, the biochar samples exhibit varying total phosphorus content ranging from 5.31, 5.36, 6.19, 7.19 and 8.39 g kg⁻¹, total carbon of 747.62 g kg⁻¹ and total nitrogen of 6.77 g kg⁻¹.

Soil properties

Soil properties were assessed at the beginning and end of the experiment by sampling the topsoil (0 to 15 cm). The collected soil samples were subsequently air-dried, homogenized and filtered through a 2-mm sieve. Detailed information on the soil's chemical properties is presented in Table 1. Soil pH was determined using a pH meter with a 1:5 ratio of soil to distilled water suspension. Additionally, soil pH NaF was measured in a 50 ml suspension of 1 M NaF using a pH meter. The CEC of the soil was extracted using the 1 M ammonium acetate method at pH 7.0. Soil available nitrogen (NH₄⁺-N and NO₃-N) was extracted using 1 N KCl, followed by steam distillation. The soil available phosphorus content was determined using the Bray I method, as described by Carter and Gregorich (2007). Organic carbon content was determined by subjecting oven-dried samples to loss-on-ignition in a muffle furnace at 550 °C (Ben-Dor and Banin, 1989; Nelson and Sommers, 1996).

Biochar properties

The carbon and nitrogen contents of the biochar were analyzed using the analyzer (Leco CN628) as described by Bird et al. (2017). The pH of the biochar was measured using a 1:5 ratio of solid to solution, with agitation for 30 minutes in deionized water, following the procedure specified by Singh et al. (2010). Total phosphorus content was determined using the dry-ashing method based on the procedure by Enders et al. (2017). The chemical composition of biochar is reported in Table 1. Scanning electron microscopy (SEM-EDX JEOL JSM-6510LA) was employed to examine the surface morphology and elemental distribution of the biochar pellets, revealing the presence of carbon, potassium and phosphorus on their surfaces (Figure 1).

Growth, yield and yield components

Soybean plant data were collected at the R7 stage, which marks the onset of maturity. Plant height was measured using a ruler, while the fresh and dry weights were determined through precise gravimetric measurements using an electronic scale. To assess root volume, researchers employed the water displacement technique introduced by Bohm (1979). The edamame plants were harvested with great care. After uprooting

them from the soil, researchers meticulously separated the stems and pods, and thoroughly washed the roots. For the determination of dry matter content, plant samples were subjected to a 48-hour drying process in an oven set at 65 °C.

Nitrate reductase activity (NRA)

The physiological characteristics of the plants were evaluated by measuring NRA in the leaves and protein content in the beans. Samples were collected at 5 and 9 WAP for NRA analysis, while protein content was determined at the time of harvest. The methodology described by Sudhakar et al. (2016) was followed to ensure accurate and consistent measurements of NRA and protein content.

Plant nutrients uptake

The uptake of nitrogen and phosphorus by the roots and beans was assessed at the time of harvest. The crop nitrogen and phosphorus uptake in root and bean was calculated as follows: phosphorus uptake (g plant⁻¹) = phosphorus concentration in plant part × dry weight. The concentration of phosphorus in plant tissue was determined using the ammonium molybdate method after plant tissue was destroyed by nitric-perchloric acid wet digestion, following the procedure described by Mills and Jones (1996).

Statistical analysis

The effect of biochar form, phosphorus fertilizer rate, and their interaction on soil and plant parameters was evaluated using analysis of variance (ANOVA). A two-way ANOVA was employed to assess the significance of these factors. To determine differences among treatments, a Duncan test was conducted at a significance level of 0.05. The statistical analysis was performed using the software package "R", version 3.4.4 (R-Core-Team, 2018).

RESULTS AND DISCUSSION

Scanning electron microscopy (SEM) and X-ray energy dispersive spectroscopy (EDS)

SEM and EDS have become indispensable techniques for investigating the surface morphology and elemental composition of biochar. SEM allows for detailed examination of biochar's intricate features, such as macropores and channels that distinguish it from other materials. Analysis of B1P2 (Figure 1a) and B1P0 (Figure 1d) using SEM reveals a highly porous structure with a smooth, compact and intricate

micro-tube bundle morphology (Liu et al., 2013). SEM analysis unveils phosphorus particles filling the biochar's pores and cavities, exhibiting uneven crystallization on the surface. These irregular crystal formations contribute to the development of numerous fissures, cracks and collapsed regions, emphasizing the complex structure of biochar. This unique structure positions biochar as a potential reservoir for nutrients and microbes, facilitating the retention of nutrients and water during irrigation or rainfall. The surface of B1P2 appears coarse and undulating. This surface texture results from the graft polymerization of PVA with biochar and bentonite, accompanied by the incorporation of phosphorus and bentonite into the biochar's cavities and channels. The formation of polymeric network structure effectively regulates the water permeability of porous biochar and controls the release of nutrients into the soil. Consequently, in a moist environment, a significant water-potential gradient can be established between the inner and outer parts of the pore channels (Spokas et al., 2014).

To provide additional insights into the elemental composition of the samples and their surface distribution, EDS analysis was conducted in conjunction with SEM imaging. Simultaneous surface element analysis was conducted using EDS alongside SEM at identical surface locations, enabling rapid semi-quantitative analysis of the elemental composition. The distribution of carbon, potassium and phosphorus exhibits heterogeneity (Figure 1b and 1e). This combined technique offers valuable information about the elemental composition and surface characteristics of the samples, facilitating the identification of potential applications.

The EDS technique was used to analyze the elemental composition of in-house produced biochar, as depicted in Figure 1c and Figure 1f. Carbon content, notably higher in B1P0 at 83.29%, was lower in B1P2 at 55.88%, indicating a greater presence of inorganic compounds. Further analysis of B1P2 revealed a higher phosphorus content of 0.15%, compared to the 0.07% in B1P0. These findings suggest that an interaction between phosphorus and biochar occurred, resulting in a modified composition. The combined utilization of SEM and EDS techniques offers comprehensive insights into the surface morphology, elemental composition and interactions within biochar, thus opening up various possibilities for its diverse applications.

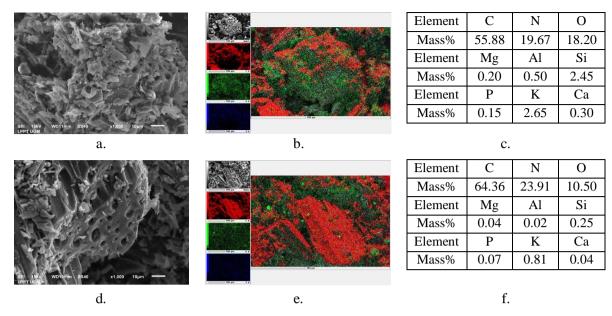


Figure 1. SEM of B1P2 (a) and B1P0 (d), with a scale bar of 10 μ m. The map overlay of the selected region of B1P2 (a) and B1P0 (d) reveals the presence of heterogeneous distribution in terms of carbon (C), potassium (K), and phosphorus (P) in (b) and (e), respectively, with a scale bar of 100 μ m. The corresponding EDS of the selected region for B1P2 (c) and B1P0 (f) are displayed in the right column

Soil and biochar properties

The soil utilized in this study is slightly alkaline, with a pH of 6.47 and an available phosphorus content of 0.013 mg kg⁻¹. Throughout the investigation, the soil exhibited a NaF-pH value of 11.10, surpassing the threshold of 9.4. This heightened pH indicates the presence of amorphous minerals, resulting in significant phosphorus fixation in the soil and consequently leading to low phosphorus availability (Poudel and West, 1999). These findings align with the results of the research conducted by Hidayanto et al. (2020) in the same location, revealing the presence of amorphous materials such as allophane, imogolite and ferrihydrite.

Upon the introduction of the KH₂PO₄ solution to the biochar, there was an observed increase in the overall phosphorus content within the biochar. The recorded values were 5.31, 5.36, 6.19, 7.19 and 8.39 g kg⁻¹ for biochars B2, B1P0, B1P1, B1P2 and B1P3, respectively. However, soaking biochar in KH₂PO₄ solution not only resulted in an increase in phosphorus content but also led to a reduction in the pH of the biochar. The pH values were measured at 10.15, 9.90, 9.44, 9.37 and 7.60 for biochars B2, B1P0, B1P1, B1P2 and B1P3, respectively. This pH decrease is attributed to the presence of KH₂PO₄, which contains hydrogen phosphate ions (HPO₄²⁻). These ions react with water, forming phosphoric acid

(H₃PO₄) and releasing hydroxide ions (OH⁻) into the solution. Consequently, this chemical reaction leads to an increased concentration of hydroxide ions, directly causing a decline in the pH of the solution. Studies suggest that immersing biochar in a KH₂PO₄ solution transforms alkaline biochar into biochar containing a notable quantity of acidic phosphorus (Li et al., 2020; Sepúlveda-Cadavid et al., 2021; Yang et al., 2021). The total carbon and nitrogen content in biochar exhibited a slight decline in the percentage content within biochar pellets when contrasted with biochar powder. This difference can be attributed to the absence of supplemental phosphorus fertilizer in the biochar powder (Table 1).

Influence of biochar powder and biochar pellets on the chemical characteristics of soil Soil pH

The soil reaction serves as a straightforward indicator of soil dynamics. Several studies have reported that the addition of biochar as a soil amendment can potentially elevate soil pH (Chintala et al., 2014; Yao et al., 2019). This pH increase can be attributed to the inherent characteristics of biochar, such as its elevated pH level, base cation content, calcium carbonate equivalent (CCE) and calcium carbonate (CaCO₃) content (Gaskin et al., 2010; Chintala et al., 2014). In this study, the application of both fertilizer and

Table 1. Elemental analysis of biochar

Parameters	B2	B1P0	B1P1	B1P2	B1P3
pH H ₂ O (1:5)	10.15 ± 0.02	9.90 ± 0.01	9.44 ± 0.02	9.37 ± 0.02	7.60±0.01
C-total (%)	79.86	77.42	72.70	70.88	71.26
N-total (%)	0.72	0.65	0.69	0.65	0.70
P-total (g kg ⁻¹)	5.31±0.13	5.36±0.11	6.19±0.19	7.19±0.29	8.39±0.23

Note: B2 = biochar powder, B1 = biochar pellet, P0 = phosphorus fertilizer 0%, P1 = phosphorus fertilizer 50%, P2 = phosphorus fertilizer 100% and P3 = phosphorus fertilizer 150%

biochar did not exhibit a significant influence on soil pH (Table 2). This research observed a slight increase in soil pH both before and after the harvest. The initial soil pH value before the harvest was 6.47 and there was a post-harvest rise of 0.12, 0.11, and 0.11 units in the biochar treatments B0, B1, and B3, respectively. The phosphorus fertilizer treatment showed increases of 0.11, 0.13, 0.15 and 0.13 units. It is noteworthy to mention that Zhang et al. (2019) demonstrated that biochar exhibited a noticeable impact on acidic soil pH but had minimal influence on alkaline soils.

Soil organic carbon

The quantity of soil organic carbon stored in the soil is contingent upon the equilibrium between the inflow of carbon and the outflow caused by the respiration of carbon-based gases resulting from microbial mineralization. Additionally, there is a potential loss through leaching in the form of dissolved organic carbon (FAO, 2017). The application of biochar can directly augment soil organic carbon content by introducing external organic material, thereby impacting the overall influx of organic carbon into the soil (Yang et al., 2020).

The analysis findings suggest that the application of biochar and phosphorus fertilizer does not significantly impact soil organic carbon. The initial soil organic carbon content measured 118.23 g kg⁻¹ before treatment. When compared with the post-harvest soil organic carbon content, it revealed a value that was relatively consistent with the organic carbon content of the soil prior to any treatment (Table 2). Madari et al. (2017) observed that the introduction of wood biochar to the soil did not alter the soil organic carbon content until the third year following application. However, a linear upswing in organic carbon content occurred in the fourth and fifth years, correlating with higher biochar doses. Similar findings were reported by Jien and Wang (2013), where the application of wood biochar incubated in the soil for 105 days did not result in a noteworthy increase in soil organic carbon

levels, despite the biochar having a high total carbon content. This outcome might be associated with the carbon content of the applied biochar and the duration of its application.

Cation exchange capacity (CEC)

The enhanced CEC of soils results from the increased surface area, the presence of hydroxyl and carboxy functional groups, and the variable charges facilitated by biochar (van Zwieten et al., 2010). The research results indicated that using biochar powder or pellets with phosphorus fertilizer simultaneously did not significantly alter the soil's CEC. However, a comparison between the post-harvest soil value and the initial soil value (23.86 cmol(+) kg⁻¹) showed an increase in soil CEC (Table 2). This finding aligns with the results of Butnan et al. (2018) research on eucalyptus forest biochar that no significant differences in the CEC of soil based on soil type, biochar type and dosage were identified. Lehmann (2007) further explained that the initial CEC of biochar is relatively low and increases as the biochar ages. However, Zhao et al. (2015) reported that applying 4-month-old wood biochar resulted in a 10% reduction in CEC during a 70-day incubation compared to fresh biochar. This validates the ever-changing influence of applying biochar to soil characteristics.

Nitrogen

Nitrogen is a crucial element found in organic compounds within plants and is indispensable for all living organisms. Most of the nitrogen in the soil is typically bound in organic forms. Assessing total nitrogen is a common method to assess the fundamental fertility of soil nitrogen and the availability of nitrogen in the soil is linked to plant growth (Gao et al., 2018). There were no significant differences in total soil nitrogen between treatments with biochar and phosphorus fertilizer. Likewise, the total nitrogen content in the initial soil before treatment (2.45 g kg⁻¹) did not exhibit a significant difference from the total nitrogen content in the soil after harvesting (Table 2). The study conducted by Gao et al. (2018) indicated that there was no significant

Table 2. Chemical properties of post-harvest soil under different treatments

Treatments	Hd	C-organic (g kg ⁻¹)	$C-organic \ (g \ kg^{-1}) CEC \ (cmol(+) \ kg^{-1}) N-total \ (g \ kg^{-1}) NH_4^+ \ (mg \ kg^{-1}) NO_3^- \ (mg \ kg^{-1}) P_2O_5 \ (mg \ kg^{-1}) NO_3^- \ (mg$	N-total (g kg ⁻¹)	NH ₄ ⁺ (mg kg ⁻¹)	NO_3^- (mg kg ⁻¹)	$P_2O_5 \text{ (mg kg}^{-1})$
Biochar (B)	su	su	ns	ns	*	*	ns
B0	6.59 ± 0.09	117.37 ± 6.76	25.49 ± 1.79	2.54 ± 0.30	30.09 ± 5.16^{b}	68.97 ± 9.76^{b}	0.032 ± 0.00
B1	6.58 ± 0.12	118.31 ± 6.61	25.29 ± 0.76	2.50 ± 0.13	40.84 ± 6.02^{a}	77.68 ± 11.39^{a}	0.033 ± 0.00
B2	6.58 ± 0.23	116.69 ± 3.59	26.18 ± 1.97	2.42 ± 0.20	44.69 ± 11.41^{a}	82.99 ± 6.03^{a}	0.034 ± 0.01
Phosphorus fertilizer (P)	su	su	su	su	*	*	*
P0	6.51 ± 0.11	116.74 ± 6.26	24.64 ± 1.24	2.43 ± 0.20	$31.98\pm4.72^{\circ}$	$67.12\pm10.40^{\circ}$	$0.025\pm0.00^{\circ}$
P1	6.60 ± 0.10	117.08 ± 7.06	25.97 ± 1.66	2.54 ± 0.20	35.95 ± 9.17^{bc}	71.14 ± 6.13^{bc}	$0.030\pm0.01^{\rm bc}$
P2	6.62 ± 0.17	118.69 ± 4.52	25.98 ± 1.76	2.47 ± 0.21	41.33 ± 5.80^{ab}	80.89 ± 13.71^{ab}	0.033 ± 0.01^{b}
P3	6.60 ± 0.19	117.32 ± 4.78	26.03 ± 1.37	2.52 ± 0.22	44.90 ± 10.42^{a}	87.03 ± 6.00^{a}	0.045 ± 0.00^{a}
ВхР	ns	su	ns	ns	ns	ns	ns
CV (%)	2.41	4.50	6.02	8.90	28.30	17.05	27.86
Note: The numbers followed by different letters in the same column are significantly different at the DMRT test level of $\alpha = 0.05$; * = significant; ns = not significant; CV =	y different letters	in the same column are	significantly different at	t the DMRT test lev	el of $\alpha = 0.05$; * = si	ignificant; ns = not	significant; CV =

impact on the total nitrogen content in the soil when treated with different amounts of biochar and NPK fertilizer.

Under specific soil composition conditions, plants take up nitrogen in the form of nitrate or ammonium, serving as sources of organic nitrogen and amino acids (Masclaux-Daubresse et al., 2010). The leaching of nitrogen has become a notable constraint in improving the efficiency of nitrogen utilization in agricultural production (Ding et al., 2016). The initial levels of NH₄⁺ and NO_3^- were 22.28 mg kg⁻¹ and 46.67 mg kg⁻¹, respectively. Subsequent to the harvest, there was a rise in the availability of nitrogen in the soil. The analysis of post-harvest soil conditions (Table 2) showed no interaction effect between biochar and the rate of phosphorus fertilizer. However, when considered separately, both B2 and B1 resulted in increased soil ammonium levels compared to the control group (B0). Although there was no statistically significant distinction between B1 and B2, the soil ammonium content rose by 14.60% and 10.75% under B2 and B1, respectively, in comparison to B0. Similarly, the P3 and P2 treatments exhibited an increased soil ammonium content of 12.92% and 9.35%, respectively, in contrast to P0. In accordance with soil ammonium content, biochar and phosphorus fertilizer, as individual treatments, also impacted soil nitrate content. B2 demonstrated a 14.02% increase in soil nitrate content compared to B0, whereas B1 showed increase. However, there was an 8.71% no statistically significant difference between B1 and B2. Notably, the P3 treatment recorded the highest soil nitrate content, with a 19.91% increase compared to P0.

These results align with the reports of research conducted by Thomazini et al. (2015), observing elevated NH_4^+ levels in forest soil during their incubation study. Laird et al. (2010) suggest that the application of biochar reduces NO_3^- leaching, attributed to the hydroxyl and alkyl functional groups in biochar. The presence of pores in biochar significantly contributed to the augmentation of the surface area, thereby providing a higher adsorption capacity and enabling the absorption of small molecules, including gases and solvents (Chia et al., 2015). This porous structure also enhanced nutrient retention and mitigated the leaching potential (Hua et al., 2009).

The increase in available nitrogen in the soil and total nitrogen in the roots of edamame soybeans treated with phosphorus fertilization

covariance; CEC = cation exchange capacity

may also be attributed to the improvement of symbiotic dinitrogen fixation, as reported by Graham and Rosas (1979). They further suggested that the stimulation of host plant growth, rather than a direct effect on the initiation, growth, development and function of root nodules, contributed to the enhanced symbiotic N₂ fixation response to phosphorus fertilization. Conversely, Chaudhary et al. (2008) conducted research indicating that phosphorus deficiency in legumes led to a reduction in the total weight of nodules in soybeans (Glycine max) and mung bean (Vigna radiata), with nitrogen fixation levels decreasing as the severity of phosphorus deficiency increased. Leidi and Rodriguez-Navarro (2000) discovered that under low nitrate concentrations (1 mM), an increase in phosphorus resulted in enhanced development of root nodules and N₂ fixation. Conversely, at high nitrate concentrations (10 mM), additional phosphorus supply did not stimulate the formation of root nodules and increased N2 fixation. Several studies (Jeffery et al., 2011; Jones et al., 2012; Abbruzzini et al., 2019) emphasize that the incorporation of biochar improves soil nitrogen availability, nitrogen uptake and nitrogen use efficiency by plants.

Phosphorus

The initial phosphorus content in the soil is 0.013 mg kg⁻¹ before harvest and subsequently rises after harvest. In evaluating post-harvest soil phosphorus availability, only the application of phosphorus fertilizer demonstrated a noteworthy increase compared to the application of biochar. The inclusion of phosphorus fertilizer leads to a higher soil phosphorus content in comparison to conditions without phosphorus fertilizer. Specifically, treatment P3 exhibited a 79.84% higher soil phosphorus content than treatment P0. Tryon (1948) documented a notable increase in phosphorus presence after the application of biochar to sandy or loamy soils. However, it is crucial to emphasize that the consistent enhancement of soil phosphorus availability was not always observed with the introduction of biochar. In a soil column experiment carried out by Novak et al. (2009), the use of biochar resulted in a significant upturn in phosphorus retention within soils, simultaneously causing a decrease in phosphorus levels in leachate solutions. The introduction of biochar led to a transient reduction in the presence of accessible phosphorus in two soil types. While one soil witnessed an improvement in phosphorus availability, the

other soil, which received additional phosphorus, did not demonstrate a notable effect on phosphorus availability (Nelson et al., 2011; Kumar et al., 2013).

Based on research findings, it is evident that the simultaneous application of biochar and phosphorus fertilizer does not have a significant impact on the overall chemical characteristics of the soil. However, through separate analysis, the use of biochar (both in powder and pellet form), along with the application of phosphorus fertilizer, demonstrated a substantial influence on ammonium and nitrate levels in the soil. Although effect of biochar on soil phosphorus availability is limited, it is advisable to consider its use as a potential supplement to enhance plant nutritional efficiency and improve soil conditions. Additionally, adjusting phosphorus fertilizer doses is acknowledged as a more effective approach to maximizing crop yields.

Nutrient uptake in the roots and seeds of edamame

The impact of biochar and phosphorus fertilizer on nitrogen and phosphorus uptake in the roots and seeds of edamame soybeans, along with the protein content in edamame soybean seeds, is presented in Table 3. The results showed that biochar treatment had a significant effect on root nitrogen uptake. Both B1 and B2 treatments exhibited a significant increase in root nitrogen uptake compared to the control group (B0), with B1 showing a 29.51% increase and B2 showing a remarkable 51.41% increase.

Similar to nitrogen uptake, biochar treatment had a significant effect on root phosphorus uptake. Specifically, B2 demonstrated a 26.25% higher phosphorus uptake than B0 and a 19.75% higher uptake than B1. The uptake of phosphorus by roots under the B2 treatment was significantly higher than that under the B1 treatment (Table 3). The application of biochar improved the morphological development of roots by alleviating nutrient deficiencies and significantly enhancing the concentration of phosphorus in the roots (Xiang et al., 2017). Biochar, with its porous structure, can enhance phosphorus content through adsorption, as indicated by studies on biochar from different sources such as sugar cane, Miscanthus (Trazzi et al., 2016), pine and maize straw (Zhao et al., 2017). The gradual release of phosphorus in biochar primarily occurs when soil solution phosphorus is depleted by crop roots or constrained by soil constituents. This slow release involves processes like phosphorus depletion by

Table 3. Biochar and phosphorus fertilizer effects on nitrogen and phosphorus uptake in edamame and protein content

Treatments	Root P uptake	Seed P uptake	Root N uptake	Seed N uptake	Protein
	(g plant ⁻¹)	$(g\ 100\ g^{-1})$			
Biochar (B)	*	ns	*	ns	*
B0	0.29 ± 0.06^{b}	1.34 ± 0.27	1.51 ± 0.26^{b}	18.73 ± 3.05	15.61±0.40 ^b
B1	0.30 ± 0.05^{b}	1.29 ± 0.28	1.96 ± 0.29^{a}	19.04 ± 2.89	15.45 ± 0.55^{b}
B2	0.36 ± 0.03^{a}	1.48 ± 0.20	2.29 ± 0.54^{a}	21.88 ± 3.65	16.35±0.45 ^a
Phosphorus	ns	ns	ns	ns	*
fertilizer (P)					
P0	0.29 ± 0.04	1.26±0.22	1.61±0.43	18.59±3.74	15.36±0.47 ^b
P1	0.31 ± 0.04	1.37 ± 0.26	1.90 ± 0.26	19.75±3.17	15.52±0.51 ^b
P2	0.32 ± 0.07	1.38 ± 0.26	1.99 ± 0.37	20.40 ± 2.67	16.13 ± 0.60^{a}
P3	0.35 ± 0.04	1.46 ± 0.27	2.18±0.39	20.80 ± 3.21	16.21±0.29 ^a
BxP	ns	ns	ns	ns	ns
CV (%)	20.33	19.14	28.43	17.71	4.80

Note: The numbers followed by different letters in the same column are significantly different at the DMRT test level of $\alpha = 0.05$; * = significant; ns = not significant

crop roots or soil constituents, as well as a significant amount of phosphorus loading through precipitation on biochar. Furthermore, when phosphorus is stored in biochar pores, its release into the soil solution experiences a slow intra-biochar diffusion (Koopmans et al., 2004). This delayed diffusion within biochar pores likely contributes to the gradual release of phosphorus from P-laden biochars, aligning with the adsorption-desorption equilibrium that governs the release mechanism. Additionally, a considerable amount of phosphorus was adsorbed onto biochar powder through precipitation and this portion of phosphorus could also be released slowly (Li et al., 2019). In comparison to biochar powder, biochar pellets have a higher density due to the pelletization process, resulting in good hydrophobicity (Xing et al., 2018). Moreover, the pores of biochars enriched with phosphorus were either occupied or obstructed by crystallized KH₂PO₄, potentially leading to a decrease in the specific surface area and porosity of the biochar (Li et al., 2019). The extremely slow intra-biochar diffusion of phosphorus within biochar pores likely contributes to the very slow release of phosphorus from phosphorus-laden biochars.

The combined application of biochar and phosphorus fertilizer did not show significant effects on the uptake of nitrogen and phosphorus by the seeds. The combination of biochar and phosphorus fertilizer did not significantly affect nitrogen and phosphorus uptake by seeds. However, when analyzed separately, the phosphorus fertilizer treatment demonstrated a non-significant increasing trend in seed

phosphorus uptake with higher doses. Compared to the control (P0), each phosphorus fertilizer treatment (P1, P2, P3) showed enhancements of 8.73%, 9.52% and 15.87%, respectively. In the biochar treatments, B2 increased seed phosphorus uptake by 10.45% compared to the control (B0), while B1 showed a 3.73% decrease. This phenomenon is possibly due to the hydrophobic (Xing et al., 2018) nature of biochar and a reduction in its specific surface area and porosity caused by crystallized KH₂PO₄ (Li et al., 2019) during biochar pellet production.

Regarding seed nitrogen uptake, although the increase was not significant, the application of biochar and phosphorus fertilizer led to enhanced seed nitrogen uptake. Compared to B0, the increase in seed nitrogen uptake was around 1.66% and 16.82% for the B1 and B2 treatments, respectively. For phosphorus fertilizer treatments, each treatment (P1, P2, P3) exhibited increases of 6.24%, 9.74% and 11.89%, respectively, compared to the control (P0). The uptake of phosphorus and nitrogen in seeds surpasses that in roots. According to Hammond et al. (1951), soybeans grown in Clarion soil accumulated larger quantities of overall nutrients, nitrogen and phosphorus in their beans in comparison to stems, roots, leaves and pods. The proportion of nitrogen and phosphorus in beans constitutes approximately 80.29% and 85.51%, respectively.

The results of protein analysis in soybean seeds (Table 3) did not reveal any interaction effect between biochar and the dosage of phosphorus fertilizer. However, when considered individually, both biochar and phosphorus fertilizer had a significant impact on seed protein

content. Among the biochar treatments, B2 exhibited the highest level of seed protein, surpassing B0 by 4.69%. Moreover, under P2 and P3 treatments, seed protein content increased by 5.53% and 5.03%, respectively, compared to the control (P0). Thus, the higher nitrogen uptake in the bean due to B2 and P3 would eventually contribute to increase the protein content of the seed, which is an important quality component of edamame soybean. The protein content in this study is higher compared to the protein content of edamame soybeans grown in Japan and Colorado, which is 11.4 and 12.4 g 100 g⁻¹, respectively (Johnson et al., 1999). These findings suggest that biochar application can enhance root nitrogen and phosphorus uptake in edamame soybean, while biochar and phosphorus fertilizer have distinct effects on seed protein content.

Nitrate reductase activity (NRA)

The NRA content in soybean leaves increases during the growth stages, particularly during flowering and seed development (Figure 2a). The impact of biochar on NRA showed contrasting patterns during different growth the experiment. At 5 of WAP, the introduction of biochar, both in B1 and B2, did not significantly affect NRA. In contrast, the control group without biochar showed a significant influence. The addition of biochar (B1 and B2) resulted in a significantly lower NRA at 5 WAP compared to the control group without biochar (B0), with NRA showing a reduction of 41.48% for B1 and 36.18% for B2. However, at 9 WAP, plots treated with biochar showed an increase in NRA. Yet, upon closer analysis, this increase is found to be statistically insignificant compared to the control group without biochar (Figure 2a). NRA displays a noticeable recovery, with an increase of 16.02% for B1 and 20.31% for B2. Another possibility is that the NO₃ absorbed by biochar may not be readily available to plants (Haider et al., 2016; Joseph et al., 2018). This physical retention of NO₃ within the small biochar pores, as suggested by Kammann et al. (2015), becomes apparent at 5 WAP, where the NRA in the biochar-treated group is lower compared to the group without biochar. Joseph et al. (2018) explain that the nitrate loading occurs through a combination of physicochemical mechanisms, possibly involving alternating cycles of soil/substrate moisture, leading to changes in redox potential (Eh), poreclogging, and the influence of electrostatic and H-bonding forces. Biochar possesses the ability

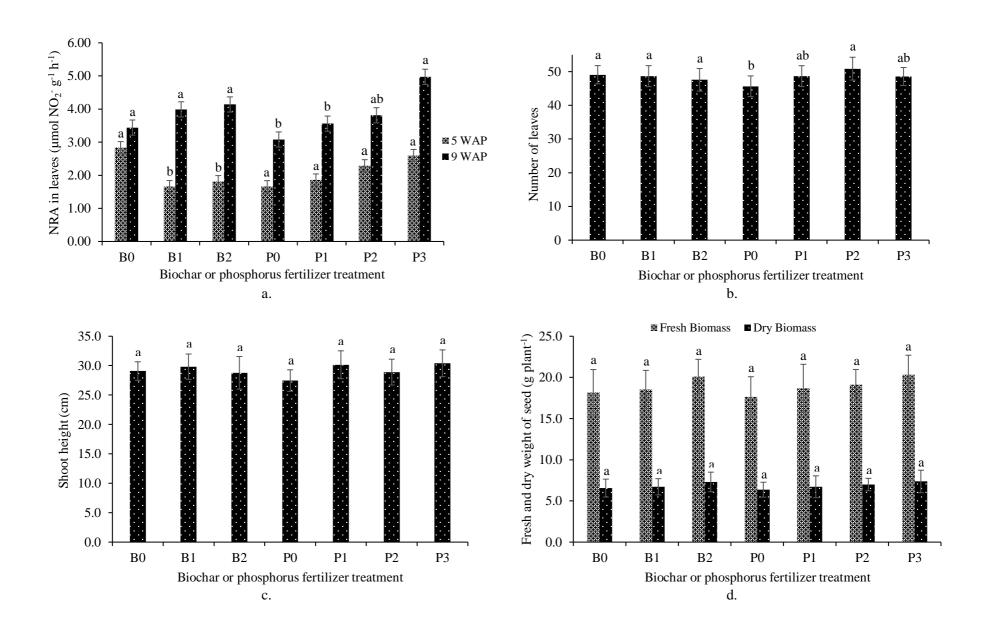
to store nutrients and act as a slow-release fertilizer, which is related to its specific properties such as pore structure and functional groups. Excess nutrients, including ammonium, nitrate and phosphate, can be deposited onto the biochar surface, as reported in studies by Ding et al. (2010) and Gwenzi et al. (2018).

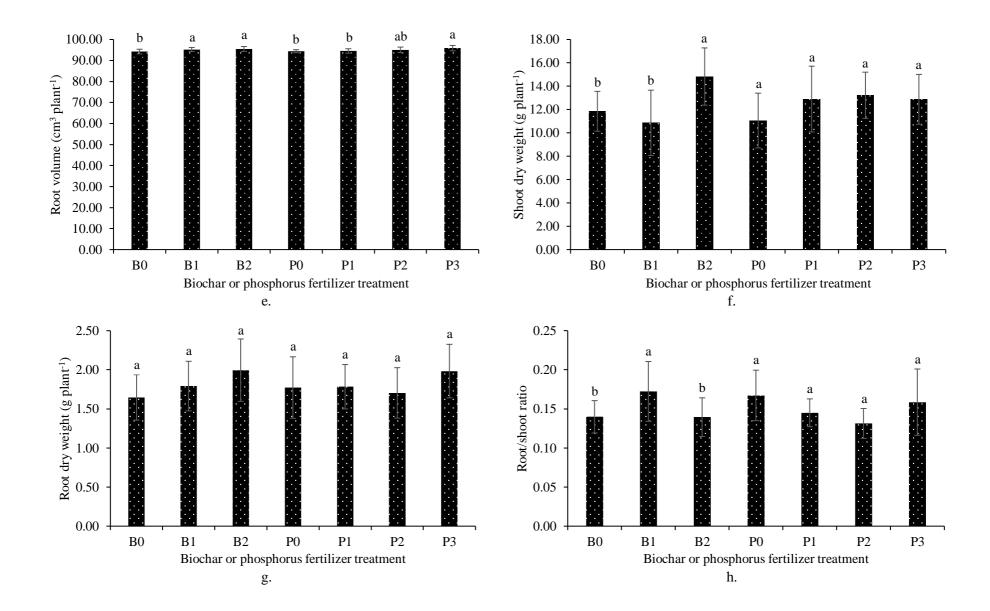
Regarding the application of phosphorus fertilizer, no significant effect on NRA was observed at 5 WAP. However, at 9 WAP, a significant difference was observed, with P3 and P2 treatments demonstrating significantly higher NRA values compared to other treatments. Remarkably, the P3 treatment had a substantial impact, significantly enhancing NRA by 56.88% at 5 WAP and 61.54% at 9 WAP compared to the P0 treatment. In this case, there may have been an interaction between phosphorus and nitrogen absorption by plants. Phosphorus deficiency affects the absorption and assimilation of nitrates in maize (Huang et al., 2008) and soybeans (Rufty et al., 1993). Availability of soil phosphorus might impact nitrate or ammonium uptake by plant through mediating phosphorylation levels of nitrate or ammonium transporters. NRA was identified as a differential phosphoprotein in soybean roots as a response to phosphorus starvation (Jiang et al., 2021).

Furthermore, the primary consequence of phosphorus deficiency in plants is a reduction in NO₃ absorption through the roots (Rufty et al., 1990). This reduction can be attributed to the decreased availability of radicular ATP and limitations in the synthesis of membrane transport systems for NO₃ (Rufty et al., 1993). Roots absorb phosphorus for cellular energy transport in the form of ATP (Scheerer et al., 2019). Despite the association of phosphorus deficiency with a substantial accumulation of amino acids in shoots, this increased amino acid content is clearly linked to enhanced protein degradation rather than simultaneous inhibition of protein synthesis (Rufty et al., 1990). Research conducted by Pilbeam et al. (1993) indicated that the rate of nitrate reductase activity in both leaf and root parts of tomato plants was higher in treatments with added phosphorus compared to plants not supplied with phosphorus.

Growth, yield and yield components

Figure 2 shows how using biochar and phosphorus fertilizer affects the growth of edamame soybeans. Interestingly, there was no significant connection between the amounts of biochar and phosphorus fertilizer in terms of





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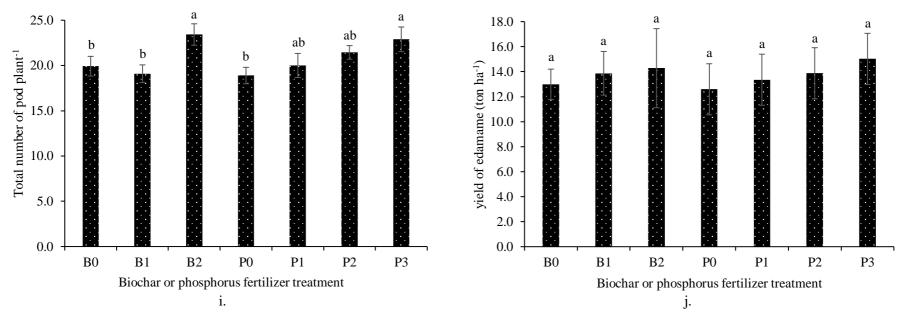


Figure 2. (a) Effect of biochar and phosphorus fertilizer on NRA in edamame soybean leaves at 5 and 9 WAP; (b) number of leave; (c) shoot height; (d) fresh and dry weight of edamame seed per plant; (e) root volume; (f) shoot dry weight; (g) root dry weight; (h) root/shoot ratio; (i) total number of pods per plant and (j) yield of edamame. Significant differences ($p \le 0.05$) between the treatments were tested and marked with different letters. Vertical bar indicates standard error

edamame growth, yield and related factors, as shown in Figure 2c, 2d, 2g and 2j. Even though adding biochar and phosphorus fertilizer did not notably affect shoot height (Figure 2c), treatment B1 produced the best results, while B2 had the lowest value. The B1 treatment increased shoot height by 3.62% compared to B2. As for phosphorus fertilizer, P3 significantly boosted shoot height, leading to a 10.70% increase compared to P0. Regarding the treatment of fresh and dry seed weight of seed (Figure 2d), B2 showed the highest values, with a 10.41% increase in fresh seed weight and an 11.40% increase in dry seed weight compared to B0. Similarly, P3 treatment had a notable impact, demonstrating a 15.08% increase in seed fresh weight and a 15.68% increase in seed dry weight compared to P0. In terms of root dry weight (Figure 2g), B2 treatment recorded the highest value, with a 21.15% increase compared to B0, while P3 treatment increased root dry weight by 11.84% compared to P0. In Figure 2j, P3 treatment exhibited the highest yield, indicating a 19.31% increase compared to P0 and applying B2 increased crop yields by 10.00% compared to B0. The edamame soybean yields in this study were relatively high, averaging from 12 to 15 tons ha⁻¹, surpassing those reported by Shanmugasundaram et al. (1991) in Taiwan, who obtained a total weight of 11.45 tons ha⁻¹ for nine varieties of edamame soybean including the Ryoko variety. However, it was comparable to the yield reported by Mozzoni and Chen (2019), which ranged from 10.8 to 14.9 tons ha⁻¹ for edamame soybean.

Figure 2b presents the impact of phosphorus fertilizer on leaf number, especially in treatment P2, where the effect is most pronounced, surpassing P0 by 10.87%. Although P1 and P3 showed increases of 6.52% compared to P0, these changes are not statistically significant. Moving on to root volume (Figure 2e), both biochar and phosphorus fertilizer play a significant role. B1 and B2 increased root volume by 1.06% and 1.33%, respectively, and P3 resulted in a 1.53% increase compared to P0. Figure 2f underscores the notable influence of biochar on shoot dry weight, especially in B2, which shows a substantial 25.13% increase compared to B0. Additionally, Figure 2h depicts a significant difference in the root/shoot ratio caused by biochar, with a considerable 23.21% increase in B1 compared to B0.

Proceeding to the total number of pod (Figure 2i), a significant contrast becomes apparent between biochar and phosphorus fertilizer. Treatment B2 exhibits a noteworthy rise of 17.57% in comparison to B0, whereas treatment P3 generates a substantial increase of 21.18% compared to P0. This investigation provides valuable perspectives on the influence of phosphorus-enriched biochar pellets and biochar powder on the growth, yield and yield components of edamame soybeans in Andisols with limited phosphorus availability. Previous studies on soybeans (Zhu et al., 2018; Liu et al., 2022) have reported similar findings, highlighting the significant impact of biochar on various agronomic traits. The potential of biochar to enhance plant growth, particularly by stimulating robust root systems, holds great promise. By increasing both the space occupied by roots and their absorption area in the soil, biochar effectively improves the availability of less mobile elements like phosphorus, crucial for optimal plant development. Moreover, the augmentation of root volume contributes to enhanced nitrogen uptake capacity and increased nitrogen use efficiency (Ran et al., 1994).

Applying phosphorus not only encourages root development but also improves the absorption of phosphorus (Jin et al., 2013), leading to better yields and quality in edamame soybeans. The observed improvement in growth indicators resulting from the use of biochar (either B1 or B2) suggests a positive influence on plant growth and yield. Specifically, B2 seems to be more effective than B1 in boosting the growth, yield and yield components of edamame soybeans. The distinction between B1 and B2 could be due to the compaction process and increased hydrophobicity related to the production of biochar pellets (B1), thus requiring further investigation to optimize the appropriate size and slow-release pattern of nutrients in biochar pellets within the soil. The application of B2 and P3 not only contributes to enhancing the growth, yield and yield components of edamame soybeans, as indicated by the increased number of leaves, root volume, shoot dry weight, root/shoot ratio and total number of pods. Biochar itself has unique physical and chemical characteristics, such as high porosity, enabling it to act as an adsorbent (Ahmad et al., 2014).

CONCLUSIONS

 T_0 soybean cultivation enhance in phosphorus-limited Andisols and improve both quality and yield, farmers are recommended to adopt a comprehensive agronomic strategy. This strategy includes the utilization of biochar, particularly biochar powder (B2), which has demonstrated superior performance compared to biochar pellets (B1). Additionally, the application of P3 phosphorus fertilizer (81 kg ha⁻¹ P₂O₅) is suggested to significantly enhance ammonium, nitrate and phosphorus availability. The combined treatment of biochar (B2) and high phosphorus fertilizer (P3) has proven to be the most effective in increasing nutrient uptake, root development, shoot dry weight and root/shoot ratio. This holistic approach not only optimizes overall plant performance but also increases the number of pods per plant and improves the protein content of edamame soybeans. Future research should focus on evaluating nutrient release and bioavailability, along with a prolonged assessment of the field application of both biochar pellets and biochar powder. This approach aims to comprehend their extended effectiveness in enhancing nutrient availability, growth and vield in edamame sovbeans.

ACKNOWLEDGEMENT

The authors would like to express their sincere gratitude for the support and assistance received during the course of this research. The authors would acknowledge the funding provided by Universitas Gadjah Mada through the Final **Project** Recognition Grant (RTA/Rekognisi Tugas Akhir) with grant 3190/UN1/DITLIT/DIT-LIT/PT/2021. which greatly supported this study. The authors are also deeply grateful to the participants who willingly volunteered to take part in this research and the experts who generously provided valuable insights and feedback. Their contributions played a pivotal role in the successful completion of this study.

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