

Low-temperature produced peanut-waste biochar reduced nitrogen and phosphorous fertilizer dependency of maize hybrid Syngenta-6621 in alkaline soil

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ABSTRACT

Slow pyrolyzed, low-temperature (300-350°C) produced biochar can be used for improving chemical characteristics of nutrient-deficient alkaline soil, however, the effect of such type of biochar on crop fertilizer demand is poorly understood. Thus, a pot experiment was carried out to determine the optimal nitrogen (N) and phosphorus (P) fertilizer requirements for maize grown in peanut-factory waste biochar (PBC) amended calcareous soil. The experiment used a completely randomized design, where two treatments were applied without PBC: one with no N or P fertilizer (control) and one with the recommended doses of NP fertilizer (N3P3). In addition, sixteen treatments were applied with 1.5% PBC, incorporating all possible combinations of four N fertilizer levels (0%, 33%, 66%, and 100% of the recommended dose, labelled as N0, N1, N2, and N3) and four P levels (0%, 33%, 66%, and 100%, labelled as P0, P1, P2, and P3). The N3P3 treatment served as the baseline for evaluating maize growth stages. Three replicates for each treatment were harvested randomly at the V8 growth stage (after 25 days of germination based on N3P3), while the remaining three replicates were harvested at the V14 stage (after 50 days of germination based on N3P3). At V8, the PBC + N3P2 treatment resulted in a significant increase (24%) in shoot fresh weight compared to N3P3. The highest increases in shoot P concentration (25%) and uptake (20%) were observed in the PBC + N1P2 and PBC + N3P3 treatments, respectively, compared to N3P3. By the V14 stage, the PBC + N2P2 treatment yielded maximum value of dry matter (17.7 g pot⁻¹) and had 18% more shoot dry matter than N3P3. Additionally, this treatment showed the largest improvement (10%) in chlorophyll content. In terms of N, the PBC + N3P0 and PBC + N3P2 treatments had the highest concentration (19%) and uptake (37%), respectively, compared to N3P3. Treatment PBC + N2P2 also recorded the highest P concentration and uptake, with increases of 16% and 34%, respectively, over N3P3. Growth rates were significantly higher when PBC was used without additional N or P fertilizer, compared to the N3P3 treatment. The results at V14 suggest that applying 1.5% slow pyrolyzed, low-temperature PBC can reduce N and P fertilizer requirements by up to 34% without negatively affecting the nutrient content or uptake in fodder maize.

Keywords: Nitrogen, phosphorus, peanut waste biochar, maize.

INTRODUCTION

Biochar is widely recognized for its stability in soil, offering better nutrient retention than other organic soil amendments. This unique property makes it a

valuable tool for carbon (C) sequestration and improving soil quality.

Research shows that biochar can enhance soil's chemical and physical properties, improve plant nutrient availability, and promote healthier plant growth [1, 2, 3]. Among biochar's primary advantages is its capacity to enhance the soil's cation exchange capacity (CEC), which leads to improved nutrient retention and availability for plants [2, 4]. This directly contributes to enhanced crop productivity. Additionally, biochar itself contains nutrients that can act as a slow-release fertilizer, further benefiting plant growth [3, 5]. Biochar's potential to revitalize nutrient-depleted soils is particularly promising for expanding arable land and boosting agricultural yields. By improving soil health, it supports sustainable farming practices and can help meet growing food demands in a changing climate [6, 7].

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In Pakistan, the effectiveness of nitrogen (N) fertilizers in agriculture is reduced due to the country's harsh climatic conditions, which differ significantly from temperate zones. After applying nitrogen-based fertilizers, a large portion of the nitrogen is lost through processes like denitrification, ammonia volatilization, and nitrate leaching. Urea, the most commonly used fertilizer in Pakistan, suffers from these losses, resulting in a low nitrogen use efficiency i.e. only 30-50% [8]. Biochar offers a potential solution to this challenge by either directly adding nutrients or indirectly improving the soil's CEC, which helps adsorb more ammonium (NH_4^+) on its surface [3, 9, 10]. Biochar N content can range from 0.1% to 9.5%, influenced by the feedstock type and pyrolysis temperature [11]. Research suggests that biochar can enhance the efficiency of nitrogen fertilizers, leading to improved plant growth and yield [12, 13]. However, the effectiveness of biochar depends on several factors, including the type and amount applied, as well as the specific characteristics of the soil [14]. During the pyrolysis of plant-based materials, nitrogenous compounds like amino sugars and amino acids are converted into more stable N structures with heterocyclic forms [15, 16]. This nitrogen is released slowly over time, making biochar function like a fertilizer that releases nutrients gradually over time [17]. Additionally, biochar's high CEC helps retain NH_4^+ from applied N fertilizers, further boosting its effectiveness [18, 19].

Deficiency of phosphorus (P) is a common issue in nearly 90% soils of Pakistan [20], primarily due to its tendency to form dicalcium phosphate, which is not readily available to plants. Biochar can help to address this problem by acting as a direct source of both exchangeable P and soluble P salts. In addition to directly releasing P, biochar can improve its availability in the soil due to its high ion exchange capacity [21, 22]. This property allows biochar to influence P availability through anion exchange. Fresh biochar, especially in acidic soils, has been shown to have a significant anion exchange capacity [23], which can improve P uptake by plants. Moreover, biochar can neutralize metals like calcium in Pakistani soils, improving P availability. It can also modify soil pH, making P more accessible to plants. The P content in biochar can vary significantly, from less than 1.0% to 13.2% (as P_2O_5), depending on the feedstock and pyrolysis conditions [24, 25]. This makes biochar a flexible and valuable amendment for improving P availability in deficient soils.

Alkaline properties of biochar are often highlighted in research, particularly for its benefits in improving acidic soils [26]. However, its impact on crop growth in high-pH, calcareous soils, such as those found in Pakistan, is less understood. It was reported that peanut factory waste biochar (PBC) application at a rate of 15 g kg^{-1} (1.5% on w/w basis) of soil can be beneficial for improving the nutrient dynamics of alkaline soil, potentially enhancing their fertility [3]. This study aimed to explore this area by focusing on several key objectives i.e. to evaluate the sole effects of PBC on maize growth [without adding N and P (NP) fertilizers]; to determine the optimal NP fertilizer doses for maize grown in PBC-amended alkaline soil, based on dry matter yield at V14 stage; to compare the relative growth rate (RGR) of maize in control conditions versus PBC-amended treatments, and to assess NP uptake in PBC-amended treatments compared to those without PBC, at both the V8 and V14 growth stages.

MATERIALS AND METHODS

Experimental design and treatment details

Peanut waste from a processing facility on Rajbah Road, Faisalabad, was used to produce PBC. This biochar was prepared in a muffle furnace at 300°C , following the procedure described by Sanchez et al. [27]. The specific properties of the PBC utilized in this study were detailed by Aon et al. [28]. The composition of the PBC was as follows: 5.0% hydrogen, 55.1% C, 23.5% oxygen, 2.6% N, 0.6% sulfur, 1.2% potassium, 0.7% P, 2.1% calcium, 1.4% magnesium, along with 418 mg of iron, 261 mg of zinc, 442 mg of manganese, 51 mg of copper, 0.62 mg of chromium, 0.07 mg of cadmium, and 0.11 mg of lead per kg of PBC. In terms of physicochemical properties, the PBC had CEC of 49 cmolc kg^{-1} , surface area of $39 \text{ m}^2 \text{ gram}^{-1}$, and a pH of 6.91. Proximate analysis of PBC revealed 343 g kg^{-1} volatile matter, 138 g kg^{-1} ash content, and 642 g kg^{-1} conversion efficiency. Peanut-waste biochar also contained oxygen-rich functional groups i.e. $0.14 \text{ molc kg}^{-1}$ of carboxylic groups, $0.16 \text{ molc kg}^{-1}$ of phenolic groups, and $0.13 \text{ molc kg}^{-1}$ of lactonic groups.

A pot experiment was carried out in a greenhouse at Soil and Environmental Sciences Institute, University of Agriculture, Faisalabad, using alkaline soil with similar physicochemical properties to the one studied by Aon et al. [28]. The soil, a sandy clay loam, consisted of 21.2% clay, 22.5% silt, and 56.3% sand. It had a pH of 8.23 and an electrical conductivity of

1.13 dS m⁻¹. The soil's organic matter was 0.53%, with a CEC of 6.75 cmolc kg⁻¹. The total N content was 0.7 g kg, with 127 mg of extractable K and 4.3 mg of available P kg⁻¹ of soil. The soil also contained 34.3 g of calcium carbonate kg⁻¹, along with 3.18 g of NH₄⁺-N and 2.40 mg of NO₃⁻-N per kg.

A completely randomized design was employed for the experiment, involving two treatments without PBC and sixteen treatments with PBC. The two treatments without PBC included one with no NP fertilizer (control) and another with the recommended doses of NP fertilizers (N3P3). The 16 treatments with PBC consisted of 1.5% (w/w) addition of PBC to the soil and all possible combinations of four N fertilizer rates (0, 33, 66, and 100% of the recommended dose, respectively represented as N0, N1, N2, and N3) with four P fertilizer rates (0, 33, 66, and 100% of the recommended dose, respectively represented as P0, P1, P2, and P3). Each pot contained 6 kg of soil, and all treatments were six times replicated.

Prior to sowing, potassium was uniformly applied to all pots at a rate of 60 kg ha⁻¹ using sulfate of potash. The recommended fertilizer rates were 240 kg of N ha⁻¹ and 90 kg of P ha⁻¹. Urea and diammonium phosphate were used as NP fertilizer sources. The required amounts of NP fertilizers were added to the soil based on the treatment plan. Respective doses of N were applied in two equal splits: the first half before sowing and the second half one week after seed germination, combined with irrigation. After the fertilizers were applied (half doses of N and full doses of P and K fertilizers), the soil in each pot was thoroughly mixed to ensure even distribution.

Crop Establishment

Five pre-soaked, healthy seeds of the maize hybrid Syngenta-6621 were planted in each pot. Once the seedlings were established, they were thinned to leave two plants pot⁻¹. To maintain soil moisture in all pots, tubewell water (filtered by reverse osmosis process) was applied as needed throughout the experiment. Plants from three replicates were harvested at V8 stage (after 25 days of germination based on N3P3), while those from the remaining three replicates were harvested at V14 stage (after 50 days of germination based on N3P3).

Plant Measurements

Agronomical Measurements

Before harvesting, plant height was measured using a measuring scale. Right after harvest, the fresh weight of the shoots from each pot was measured using an analytical balance. The shoot samples were then air-

dried before being placed in a forced-air driven oven (Rikakikai-Eyela WFO-600ND, Japan) at 65-67°C until they reached a constant weight. After reaching this weight, the dry weight of the shoot samples was measured.

The RGR of the shoots (in mg g⁻¹ day⁻¹) was calculated using the following formula:

$$RGR = (\ln SDW_2 - \ln SDW_1) / \Delta T$$

In this equation, SDW 1 and SDW 2 represent the shoot dry weight (in grams) at V8 and V14 harvests, respectively, and ΔT represents the number of days between the two harvests.

Physiological measurements

Immediately before each harvest, the fully expanded second leaf from the top of each plant was chosen to measure chlorophyll content using a SPAD-502 meter (Konica Minolta-Japan).

Nutrient Measurements

Plant samples were ground in a Wiley mill fitted with stainless steel blades. For digestion, 0.2 g of each sample was processed according to the method outlined by Wolf [29]. Following digestion, the final volume was brought up to 50 mL with deionized water. The impact of treatments on NP concentrations in plant tissues was then assessed. Nitrogen content was determined via the Kjeldahl method [30], and P concentration was measured using a UV-visible spectrophotometer. The yellow coloration for phosphorus was developed using the vanadate-molybdate method [31] and measured at 410 nm wavelength, with a standard curve used for reference. Following formula was used to calculate total uptake of NP (separately for each nutrient) in maize:

$$[Uptake\ of\ N\ or\ P\ (mg\ pot)]^{(-1)} = ([Percentage\ of\ N\ or\ P \times\ Dry\ matter\ (mg\ pot)]^{(-1)}\ of\ maize) / 100$$

Statistical Analysis

Data calculations and statistical analysis were conducted using Microsoft Excel 365® (Microsoft Corporation-USA) and Statistix 8.1®. To determine the treatment means that were significantly different, Tukey's multiple comparison test was applied, setting the significance level at P ≤ 0.05.

RESULTS

Growth Parameters

The data from the V8 stage, as shown in Figure 1a, highlights that the addition of PBC resulted in a 54%

increase in fresh biomass weight compared to its control treatment. When PBC was combined with different levels of NP fertilizers i.e. 33%, 66%, and 100% of the recommended doses, fresh biomass was increased by 37%, 94%, and 95%, respectively, relative to the PBC + N0P0 treatment. Notably, the PBC + N3P2 treatment yielded the highest fresh biomass weight at the V8 stage (18.2 g), which was significantly ($P \leq 0.05$) greater than the PBC + N3P3 treatment.

As depicted in Figure 1b, at the V14 stage, the maximum fresh biomass weight (128 g) was recorded in the PBC + N2P2 treatment. Applying 100% of the recommended NP fertilizers without PBC led to a fresh biomass weight 2.2 times higher than the control. Furthermore, the PBC + N0P0 treatment yielded 65% more fresh biomass at the V14 stage, than the control.

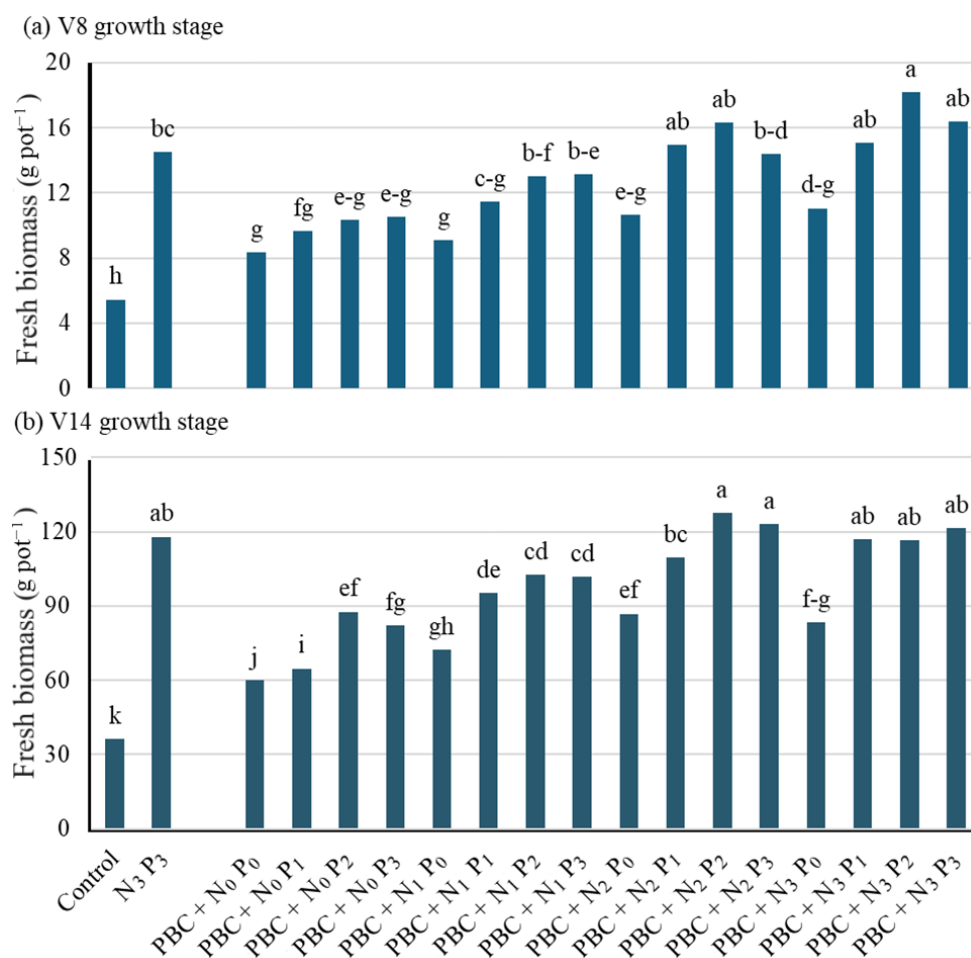


Figure 1. Effect of PBC at varying NP fertilizer rates on maize fresh biomass at the V8 and V14 growth stage. PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N₀ at 0%, N₁ at 33%, N₂ at 66%, and N₃ at 100%; treatments for P fertilizer are represented as follows: P₀ at 0%, P₁ at 33%, P₂ at 66%, and P₃ at 100% of recommended P dose. At $P \leq 0.05$, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

The data summarized in Figure 2a clearly shows that all treatments amended with PBC significantly ($P \leq 0.05$) increased plant height compared to the control. Without PBC, the addition of N3P3 fertilizers led to a notable 1.6-fold increase in plant height over the control. When NP fertilizers were absent, the

introduction of PBC still resulted in a significant 54% increase in fresh weight compared to the control. The highest plant height (74 cm) was observed in the PBC + N3P2 treatment.

Figure 2b illustrates the combined effects of PBC and varying rates of NP fertilizers on maize plant height at

the V14 stage. The PBC + N3P2 treatment showed the highest significant improvement, with a 1.2-fold increase in plant height relative to the control. However, within the PBC-amended treatments, no

significant difference ($P \leq 0.05$) was observed between the treatments N2P2 and N3P3 in terms of plant height.

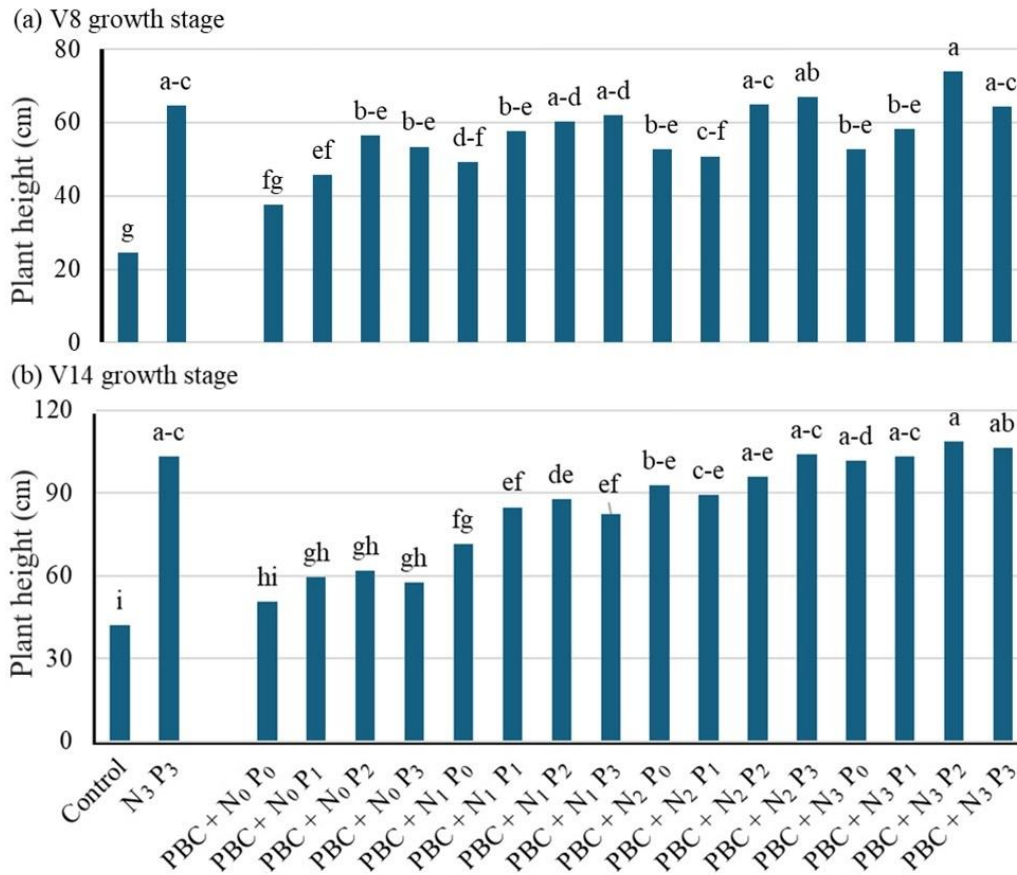


Figure 2. Effect of PBC at varying NP fertilizer rates on maize plant height at the V8 and V14 growth stage

PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N₀ at 0%, N₁ at 33%, N₂ at 66%, and N₃ at 100%; treatments for P fertilizer are represented as follows: P₀ at 0%, P₁ at 33%, P₂ at 66%, and P₃ at 100% of recommended P dose. At $P \leq 0.05$, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

As shown in Figure 3a, the highest dry biomass (3.41 g pot⁻¹) was achieved with the treatment PBC + N3P2, closely followed by 3.29 g pot⁻¹ from the treatment PBC + N3P3. Compared to the control, the N3P3 treatment yielded 1.9 times more dry matter at the V8 stage. Additionally, the dry matter yields from the N3P3 and PBC + N2P2 treatments were statistically comparable.

Figure 3b highlights the impact of various treatments at the V14 stage. As compared with control, all PBC-

amended treatments produced significantly higher dry biomass at $P \leq 0.05$. The PBC + N3P2 treatment provided the maximum dry matter yield (17.7 g per pot) at the V14 stage. The PBC + N2P2 treatment yielded twice as much dry biomass as the control. While there was no statistically significant ($P \leq 0.05$) difference in dry biomass between the PBC + N2P2 and PBC + N3P3 treatments, both produced significantly higher yields than the N3P3 treatment without PBC.

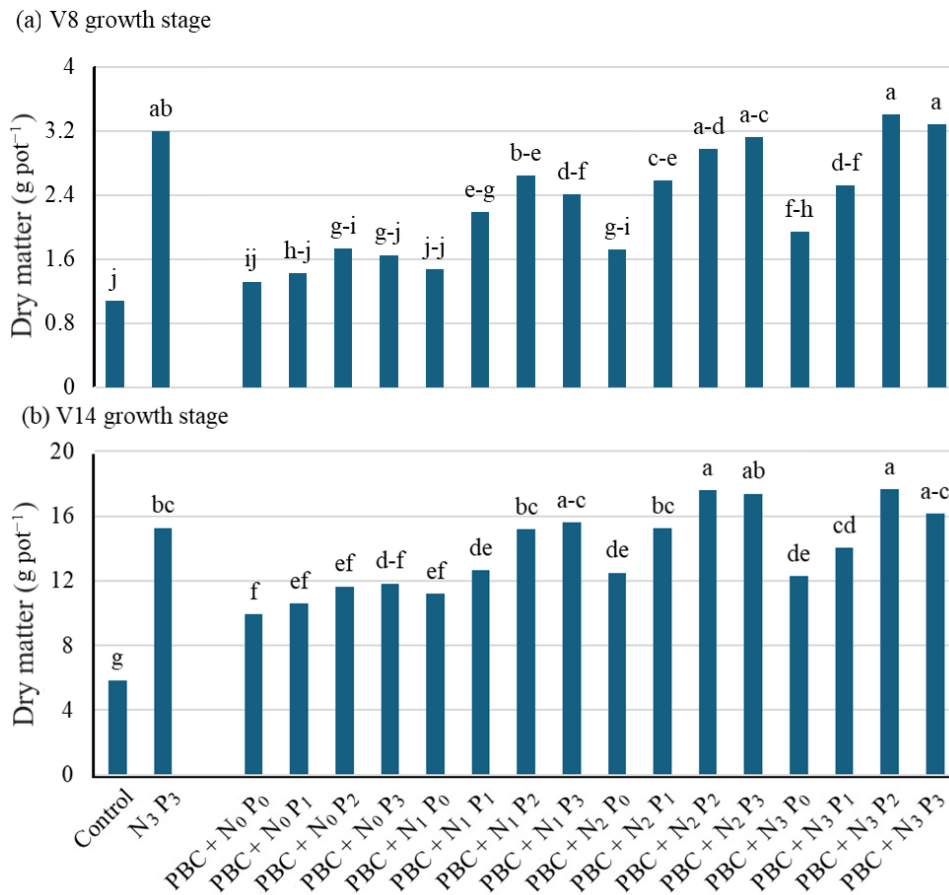


Figure 3. Effect of PBC at varying NP fertilizer rates on maize dry matter at the V8 and V14 growth stage. PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N₀ at 0%, N₁ at 33%, N₂ at 66%, and N₃ at 100%; treatments for P fertilizer are represented as follows: P₀ at 0%, P₁ at 33%, P₂ at 66%, and P₃ at 100% of recommended P dose. At P ≤ 0.05, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

Figure 4 clearly demonstrates that the PBC-amended treatments, even with reduced rates of NP fertilizers, exhibited higher relative growth rates (RGR) compared to the N₃P₃ treatment. The highest RGR value (81 mg g⁻¹ day⁻¹) was recorded in the PBC + N₁P₀ treatment. Additionally, the treatments PBC +

N₀P₀, PBC + N₁P₀, and PBC + N₀P₁ showed 29%, 30%, and 28% higher RGR, respectively, compared to N₃P₃. Specifically, the PBC + N₀P₀ treatment demonstrated a 27% improvement in RGR relative to PBC + N₃P₃.

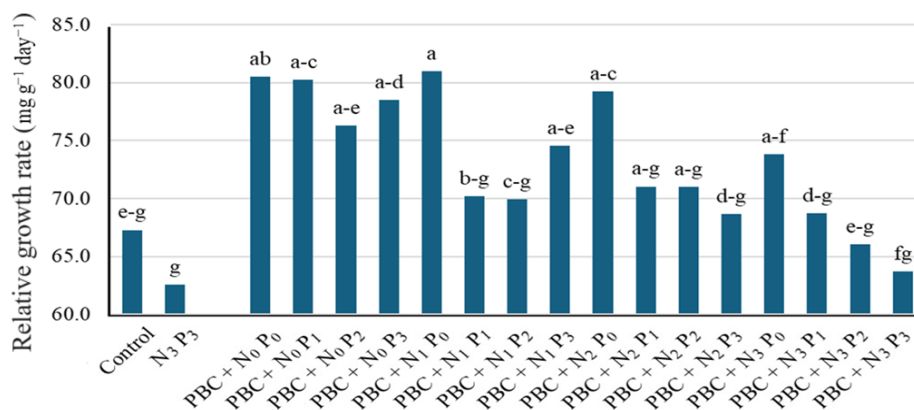


Figure 4. Effect of PBC at varying NP fertilizer rates on relative growth rate of maize

PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N_0 at 0%, N_1 at 33%, N_2 at 66%, and N_3 at 100%; treatments for P fertilizer are represented as follows: P_0 at 0%, P_1 at 33%, P_2 at 66%, and P_3 at 100% of recommended P dose. At $P \leq 0.05$, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

Chlorophyll content

The data presented in Table 1 highlights the impact of PBC and varying rates of NP fertilizers on the chlorophyll content in maize. At the V14 stage, all PBC-amended treatments, except for PBC + N_0P_0 , showed significantly higher ($P \leq 0.05$) chlorophyll content over control. The highest value of chlorophyll content (34.9) was recorded in the treatment PBC + N_3P_2 . There was no statistically significant difference ($P \leq 0.05$) in chlorophyll content between treatments

with the full recommended dose of NP fertilizers (N_3P_3), whether PBC was added or not.

At V14 stage, the PBC + N_2P_2 treatment exhibited the largest increase in chlorophyll content, with gains of 128% and 64% over the control and PBC + N_0P_0 treatments, respectively. Without PBC, the N_3P_3 treatment showed a 1.1-fold increase in chlorophyll content compared to the control. Meanwhile, the PBC + N_2P_2 treatment had 10% higher chlorophyll content than N_3P_3 .

Table 1. Effect of PBC at varying NP fertilizer rates on chlorophyll content in maize at V8 and V14 growth stage

PBC rate	Phosphorus dose	Nitrogen dose			
		N_0	N_1	N_2	N_3
		V8 growth stage			
Control	P_0	16.3±0.6h			
	P_3				35.3±2.6 a
1.5%	P_0	21.0±1.3 gh	26.8±1.6 d-f	28.2±2.3 c-e	33.5±1.2 ab
	P_1	23.0±1.4 fg	32.5±1.0 a-c	34.8±1.9 a	34.5±0.7 a
	P_2	21.9±2.0 g	29.5±1.7 b-d	33.2±2.0 ab	34.9±1.2 a
	P_3	23.3±1.0 e-g	27.7±1.8 c-f	35.4±1.2 a	34.7±1.8 a
		V14 growth stage			
Control	P_0	16.5± 0.75 h			
	P_3				34.2±1.8 bc
1.5%	P_0	23.0±1.1 g	27.3±0.7 f	30.8±0.7 de	31.9±1.2 cd
	P_1	25.3±0.6 fg	27.2±0.5 f	34.1±0.5 b-d	35.3±0.4 ab
	P_2	28.5±1.3 ef	31.3±1.5 c-e	37.7±1.9 a	33.8±1.2 b-d
	P_3	26.2±0.7 fg	32.2±1.4 b-d	35.3±1.2 ab	37.6±0.6 a

PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N_0 at 0%, N_1 at 33%, N_2 at 66%, and N_3 at 100%; treatments for P fertilizer are represented as follows: P_0 at 0%, P_1 at 33%, P_2 at 66%, and P_3 at 100% of recommended P dose. At $P \leq 0.05$, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

Nutrient Concentration

The data in Table 2 outlines the N concentration in maize shoots. At V8 stage, the highest N concentration (17.5 g kg⁻¹) was observed in the PBC + N_3P_1 treatment, closely followed by 17.1 g kg⁻¹ in the PBC

+ N_3P_2 treatment. The N_3P_3 treatment exhibited a 112% higher N concentration compared to control treatment. However, the PBC + N_3P_3 treatment had 10% lower shoot N concentration than the N_3P_3 treatment alone.

At V14 stage, the addition of PBC significantly ($P \leq 0.05$) enhanced shoot N concentration in the N_0P_0 and N_3P_3 fertilizer treatments. The PBC + N_3P_2 treatment showed the greatest increase, with an 18% higher N concentration than N_3P_3 . There was no statistically significant ($P \leq 0.05$) difference in shoot N concentration among treatments with N_2 and N_3

fertilizer rates. Additionally, the PBC + N_0P_0 treatment resulted in a 50% higher N concentration compared to the control, while the N_3P_3 treatment without PBC led to a 76% increase in N concentration over the control.

Table 2. Effect of PBC at varying NP fertilizer rates on N concentration (g kg^{-1}) in maize at the V8 and V14 growth stage

PBC rate	Phosphorus dose	Nitrogen dose			
		N_0	N_1	N_2	N_3
V8 growth stage					
Control	P_0	08.1±0.08 h			
	P_3				17.2±0.1 ab
1.5%	P_0	8.08±0.7 h	12.2±0.4 ef	15.4±0.8 bc	15.6±0.9 a-c
	P_1	9.68±0.3 gh	11.5±0.4 f-g	14.6±0.6 cd	17.5±1.0 a
	P_2	8.40±0.2 h	11.8±0.1 ef	14.9±0.7 cd	17.1±0.4 ab
	P_3	8.16±0.2 h	11.1±0.5 fg	13.4±0.6 de	15.4±0.6 bc
V14 growth stage					
Control	P_0	07.4±0.8 h			
	P_3				13.0±0.8 b-e
1.5%	P_0	11.1±1.0 d-g	12.8±0.9 c-f	14.5±1.0 a-c	15.5±0.4 a
	P_1	10.5±0.8 fg	12.7±0.8 c-f	15.0±1.1 a-c	14.8±0.7 a-c
	P_2	10.5±0.8 fg	11.1±0.6 d-g	13.8±0.8 a-c	15.4±0.4 ab
	P_3	09.2±0.6 gh	10.8±0.5 e-g	13.5±1.2 a-d	14.7±0.7 a-c

PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N_0 at 0%, N_1 at 33%, N_2 at 66%, and N_3 at 100%; treatments for P fertilizer are represented as follows: P_0 at 0%, P_1 at 33%, P_2 at 66%, and P_3 at 100% of recommended P dose. At $P \leq 0.05$, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

Table 3 summarizes the P concentration in maize shoots. At the V14 stage, the highest P concentration (3.46 g kg^{-1}) was observed in the PBC + N_1P_2 treatment, followed closely by 3.42 g kg^{-1} in the PBC + N_1P_3 treatment. The N_3P_3 treatment demonstrated a 42% increase in P concentration compared to the control. When PBC was added to the full recommended doses of NP fertilizers, it enhanced shoot P concentration by up to 17% over the N_3P_3 treatment without PBC.

At V14 stage, the PBC + N_2P_2 treatment exhibited the largest increase in shoot P concentration, with a 73% improvement relative to the control. Additionally, the N_3P_3 treatment resulted in a 48% increase in shoot P concentration compared to the control. The PBC + N_1P_2 and PBC + N_1P_3 treatments had significantly ($P \leq 0.05$) higher shoot P concentrations than the N_3P_3 treatment.

Table 3. Effect of PBC at varying NP fertilizer rates on P concentration (g kg^{-1}) in maize at V8 and V14 growth stage

PBC rate	Phosphorus dose	Nitrogen dose			
		N ₀	N ₁	N ₂	N ₃
		V8 growth stage			
Control	P ₀	1.94±0.3 g-i			
	P ₃	2.76±0.19 b-e			
1.5%	P ₀	2.04±0.17 f-i	1.94±0.13 g-i	1.55±0.14 i	1.67±0.13 hi
	P ₁	2.64±0.21 c-f	2.46±0.27 d-g	2.25±0.15 f-h	2.26±0.11 e-h
	P ₂	3.19±0.18 a-c	3.46±0.22 a	3.07±0.23 a-d	3.08±0.21 a-c
	P ₃	3.33±0.29 ab	3.42±0.22 a	2.77±0.14 b-e	3.23±0.22 a-c
		V14 growth stage			
Control	P ₀	1.61±0.29 fg			
	P ₃	2.39±0.04 a-c			
1.5%	P ₀	1.51±0.10 g	1.72±0.15 e-g	1.69±0.05 fg	1.83±0.10 d-g
	P ₁	2.05±0.14 c-f	2.22±0.30 b-e	2.57±0.13 a-c	2.28±0.10 a-d
	P ₂	2.68±0.15 ab	2.54±0.10 a-c	2.79±0.26 a	2.54±0.07 a-c
	P ₃	2.74±0.19 ab	2.37±0.20 a-c	2.67±0.22 ab	2.51±0.21 a-c

PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N₀ at 0%, N₁ at 33%, N₂ at 66%, and N₃ at 100%; treatments for P fertilizer are represented as follows: P₀ at 0%, P₁ at 33%, P₂ at 66%, and P₃ at 100% of recommended P dose. At $P \leq 0.05$, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

Nutrient Uptake

At the V8 growth stage, Figure 5a clearly demonstrates that N uptake in the PBC + N₃P₂ treatment was 5.5 times higher than in the control. The PBC + N₀P₀ treatment resulted in a 26% increase in P uptake compared to the control. No additional significant increase in N uptake was observed with the addition of PBC in the PBC + N₃P₃ treatment when compared to the N₃P₃ treatment alone. The N₃P₃ treatment exhibited a 5.1-fold higher N uptake compared to the control.

As shown in Figure 5b, at V14 stage, the highest N uptake (272 mg pot^{-1}) was observed in the PBC + N₃P₂ treatment. The N₃P₃ treatment demonstrated a 3.6-fold increase in N uptake compared to the control. Nitrogen uptake in the PBC + N₃P₂ treatment was significantly higher ($P \leq 0.05$) than in the N₃P₃ treatment. Additionally, the PBC + N₀P₀ treatment resulted in a 1.6-fold increase in N uptake compared to the control, while the PBC + N₃P₂ treatment showed 1.6 times higher N uptake than the PBC + N₀P₀ treatment.

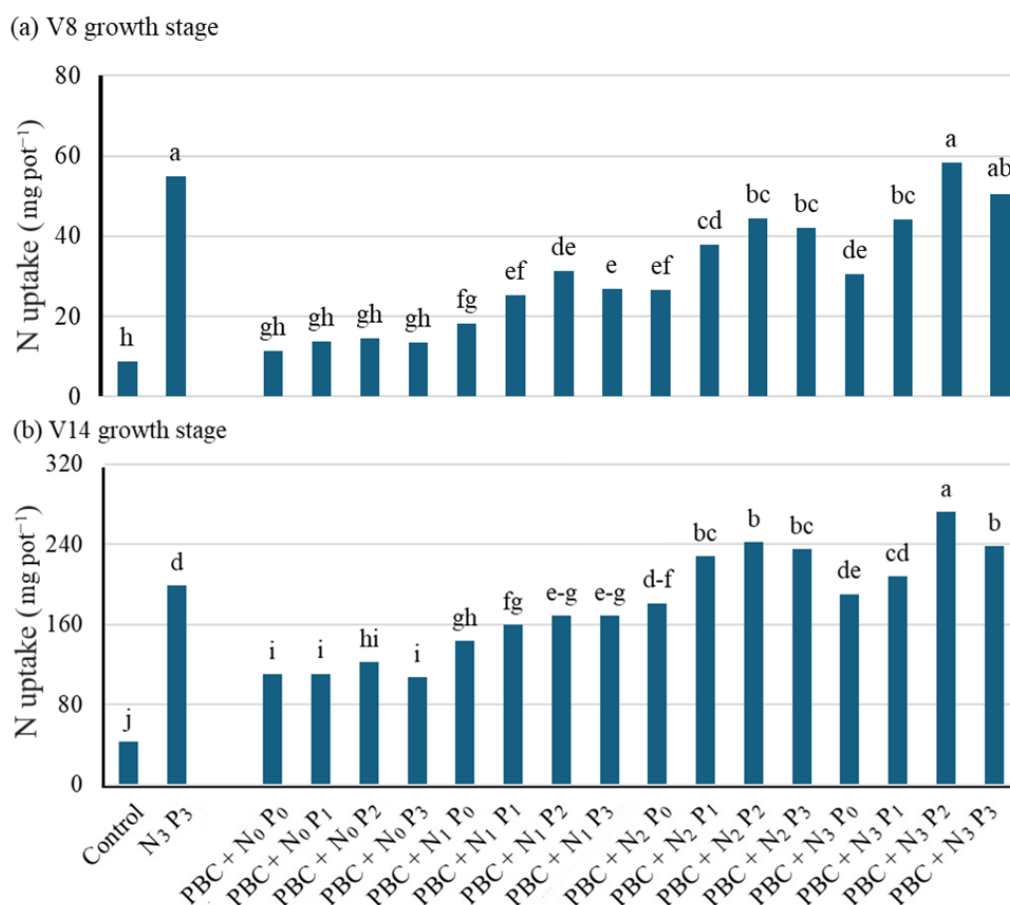


Figure 5. Effect of PBC at varying NP fertilizer rates on N uptake in maize at V8 and V14 growth stage

PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N₀ at 0%, N₁ at 33%, N₂ at 66%, and N₃ at 100%; treatments for P fertilizer are represented as follows: P₀ at 0%, P₁ at 33%, P₂ at 66%, and P₃ at 100% of recommended P dose. At $P \leq 0.05$, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

At the V8 growth stage, the PBC + N₃P₃ treatment had the highest P uptake (10.6 mg pot⁻¹) among all treatments (Figure 6a). This P uptake was significantly higher ($P \leq 0.05$) than that of the N₃P₃ treatment. The PBC + N₀P₀ treatment showed a 27% increase in P uptake compared to the control. Phosphorus uptake in the N₃P₃ treatment was 3.1 times greater than in the control. No significant difference ($P \leq 0.05$) was observed in P uptake between the N₃P₃ and PBC + N₂P₂ treatments.

Figure 6b illustrates the effect of treatments on P uptake at V14 stage. The PBC + N₂P₂ treatment had the highest P uptake (128 mg pot⁻¹), followed by the PBC + N₂P₃ treatment, with 123 mg pot⁻¹. Both treatments significantly outperformed ($P \leq 0.05$) the N₃P₃ treatment in P uptake. However, the N₃P₃ treatment still showed 2.9 times more P uptake than the control. In comparison to the control, the PBC + N₀P₀ treatment led to a 60% increase in P uptake.

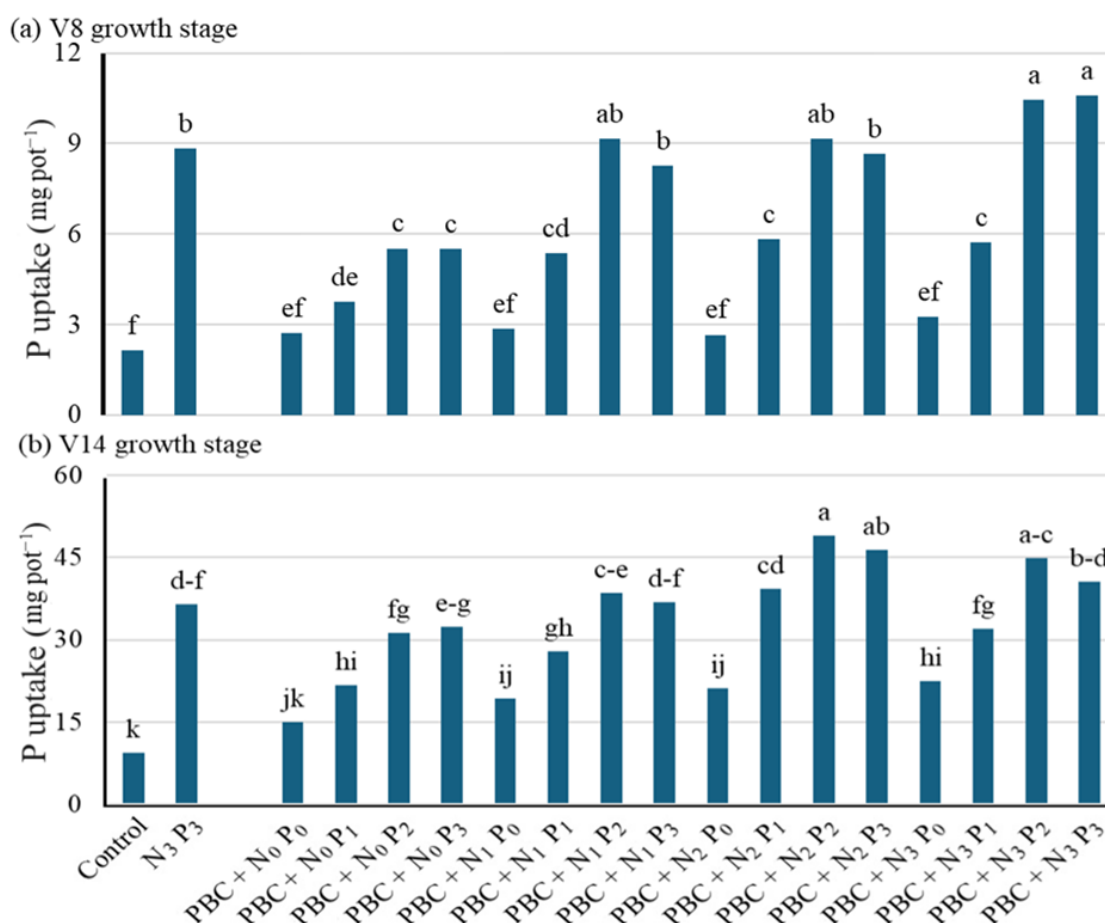


Figure 6. Effect of PBC at varying NP fertilizer rates on P uptake in maize at V8 and V14 growth stage

PBC represents peanut-factory waste biochar; treatments indicate the percentage of the recommended N fertilizer dose: N₀ at 0%, N₁ at 33%, N₂ at 66%, and N₃ at 100%; treatments for P fertilizer are represented as follows: P₀ at 0%, P₁ at 33%, P₂ at 66%, and P₃ at 100% of recommended P dose. At $P \leq 0.05$, the significance of treatments was compared by Tukey-HSD test, treatments marked with the same letter(s) indicate no significant difference; and each treatment was replicated thrice.

DISCUSSION

This study demonstrated that adding PBC to alkaline soil, even with reduced NP fertilizer doses, resulted in similar or, in some cases, better outcomes compared to using the full recommended dose of NP fertilizers (N₃P₃) without PBC (Figure 1-3). Traditionally, to achieve maximum production, farmers tend to increase the use of fertilizers, particularly N [32, 33]. However, with PBC, plant growth and nutrient uptake improved significantly, even at lower NP levels, suggesting that the PBC played a key role in enhancing these outcomes (Figure 4-6).

In this experiment, biochar was produced using waste from a peanut processing (PP) factory. Biochars produced at lower temperatures from manures and bio-solids typically contain organic nitrogen in hydrolysable forms, such as amino acids. Hossain et al. [34] previously reported that manure-based biochars, which are rich in N, can enhance N availability in the soil. The proportion of hydrolysable

N in biochar typically decreases as pyrolysis temperatures rise [35]. Thus, to address the deficiency of N in alkaline Pakistani soils, low-temperature (300°C) pyrolyzed PBC was used.

At V14 stage, the results showed that maize growth (Figures 1 and 3), chlorophyll content (Table 1), and N uptake (Table 2) improved significantly ($P \leq 0.05$) with PBC application, even without NP fertilizers, compared to the control. This growth improvement could be attributed to the nutrients provided by PBC. Previous work has also suggested that biochar may improve soil nutrient availability and enhance maize growth by supplying essential nutrients that may be limited in alkaline soil [7, 28].

Regarding RGR, at $P \leq 0.05$, a significant increase was observed in the PBC + N₀P₀ treatment, over control (Figure 4). Similarly, when compared to the N₃P₃ treatment, the significantly higher RGR at reduced NP fertilizer levels indicates that the PBC-amended treatments promoted greater shoot growth. The

mineralization rate and availability of organic N in biochar, applied to the soil, suggest that biochar can act as a slow-release nitrogenous fertilizer [17]. Consequently, the treatments amended with PBC likely increased nutrient availability, resulting in the enhanced relative growth rate observed in this study. The treatments PBC + N0P0 and PBC + N3P3 exhibited significantly better chlorophyll content ($P \leq 0.05$), respectively, over the control and N3P3 treatments (Table 1). The rise in chlorophyll content indicates improved nutrient availability and more robust plant growth in the treatments amended with PBC. Given that chlorophyll content is a crucial indicator of photosynthetic activity and is closely linked to N levels in plants, it also provides valuable insight into how crops react to soil nitrogen status and fertilizer applications [36, 37]. The improved N status in the plants from the PBC-amended treatments (as shown in Table 5 and Figure 5) corresponded to higher content of chlorophyll in maize plant tissue. Similarly, Minhas et al. [38] reported that the addition of bamboo biochar resulted in 37% increase in chlorophyll content in maize.

With respect to N concentration and uptake, Uzoma et al. [39] noted that cow manure biochar, produced at 500°C, improved maize yield and N uptake as its application rate increased, suggesting that N was released from the biochar. In this study, the PBC-amended treatments showed a more significant enhancement in shoot N concentration, even with reduced NP fertilizer rates, compared to treatments that received the full recommended dosage of NP fertilizer. Miller et al. [40] discovered that while the application of biochar reduced N uptake in roots, it did not impact plant biomass or N concentration. In a more recent pot experiment, O'Toole et al. [41] investigated ryegrass growth under various N fertilizer rates combined with wheat-straw biochar produced at temperatures between 500-600°C. They found that foliar N concentrations decreased in the biochar-amended treatments, likely due to biochar's capacity to adsorb or immobilize N, even though crop yields remained unchanged. Similarly, Kammann et al. [42] noted lower foliar N concentrations in a pot trial using nutrient-rich peanut hull biochar, but attributed this reduction to improved N use efficiency, reporting up to a 60% increase in biomass. In contrast, this experiment demonstrated that both N concentration and uptake improved with the addition of PBC, likely due to N release from the PBC, which contained 2.61% N. Generally, the hydrolysable N content in biochar tends to increase as temperatures during the pyrolysis process decrease [43]. Low pyrolysis temperature (300°C) produced biochar was used in this study to maximize the benefits of the N present in PP waste. An additional reason for the increased

nitrogen concentration and uptake in maize shoots may be the enhanced nutrient retention capacity of soil amended with PBC. Numerous studies have demonstrated that applying biochar can markedly improve soil CEC [4, 2]. In this study, the CEC of the soil and PBC were 6.75 and 49.1 cmolc kg⁻¹, respectively, suggesting that PBC may have improved the soil's CEC. With an increased CEC, the retention of NH₄⁺ during the cropping period likely improved, providing a steady supply of NH₄⁺-N for plant uptake.

Following the analysis of plant samples from each harvest (at V8 and V14 stage), it became evident that no statistically significant difference was observed ($P \leq 0.05$) in P concentration between the control and the PBC + N0P0 amended treatment. However, the PBC + N0P0 treatment demonstrated significantly higher uptake of P ($P \leq 0.05$) compared to the control, as reflected in dry biomass yield (Figure 3b). A marked increase in P uptake ($P \leq 0.05$) was also noted in the PBC + N3P3 treatment compared to the N3P3 treatment. Nigussie et al. [44] found increased P uptake in plants grown in soil amended with maize stalk biochar.

In acidic and neutral soil, a significant increase in P availability is reported due to biochar addition, however, biochar did not respond when soil conditions were alkaline [45]. In current study, the increase in P uptake in the PBC-treated soil may be attributed to the high P content found in waste from PP factory. Additionally, the notable increase ($P \leq 0.05$) in shoot growth observed in the PBC-amended soil could be attributed to the improved availability of P. Phosphorus deficiency, as determined by Olsen's method, is prevalent in nearly 90% of Pakistani soils [20]. A substantial amount of P in PP factory waste feedstock is bound within organic molecules through ester or pyrophosphate bonds [46]. Upon heating plant material, organic C begins to volatilize at around 100°C, while P remains stable until the pyrolysis process reaches roughly 700°C [47]. The burning or charring of organic materials greatly enhances P availability by releasing C and breaking organic P bonds, resulting in soluble P salts within the char residue. Additionally, biochars formed at lower pyrolysis temperatures generally contain higher levels of extractable phosphate (PO₄⁻³) than those produced at elevated temperatures [35]. In Pakistani soils, P deficiency is primarily caused by its precipitation as dicalcium phosphate [48]. Biochar may help reduce this issue by acting as an ameliorant for P-complexing metals such as Ca²⁺, Fe³⁺, and Al³⁺. This could explain the comparable P uptake between treatments using the full recommended NP fertilizer dose (100%) and those using a reduced dose (66%) supplemented with PBC.

CONCLUSIONS

Without the addition of NP fertilizers, PBC alone improved plant growth and the uptake of NP compared to the control treatment, with the effects being most noticeable at the V14 growth stage. A notable improvement ($P \leq 0.05$) in shoot dry matter, chlorophyll content, and NP concentration and uptake was observed in treatments combining reduced NP fertilizers with PBC, compared to the N3P3 treatment. The RGR was significantly higher ($P \leq 0.05$) in treatments that received reduced NP fertilizer rates, particularly those with N0 and/or P0, when PBC was applied, compared to the treatments without PBC. Based on the results at the V14 stage, the treatment combining PBC with N2P2 showed statistically better outcomes than the N3P3 treatment. In conclusion, addition of 1.5% low temperature produced PBC may reduce NP fertilizer demand of maize up to 33% for optimal maize growth. However, further research is needed to identify economically viable methods for producing biochars that are effective for alkaline soils.

CONFLICTS OF INTEREST

The author declares no conflict of interest

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