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Keywords: broadcast; deep band; phosphorus accumulation; phosphorus use efficiency; side band; Zea mays



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Article

# Differential Effects of Nitrogen and Phosphorus Fertilization Rates and Fertilizer Placement Methods on P Accumulations in Maize

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**Abstract:** Crop production in Afghanistan suffers from limited phosphorus (P) availability, which severely hinders national agriculture sustainability. This present study hypothesized that deep fertilizer placement could significantly enhance uptake of immobile P and thus tissue P accumulation and crop yield. A two-year pot experiment growing two maize (*Zea mays*) hybrid cultivars (Xida-789 and Xida-211) was therefore conducted to test these hypotheses under three contrasting fertilizer placement methods (broadcast, side band, and deep band). In doing so, P concentrations in both maize tissues and soils were compared at 45, 60, and 115 days after sowing (DAS) under nine combinations of nitrogen (N) and P fertilizer rates (kg ha<sup>-1</sup>: N112P45, N112P60, N112P75, N150P45, N150P60, N150P75, N187P45, N187P60, N187P75). Results have shown that deep band placement significantly increased P uptake efficiency, leading to greater P concentration and accumulation in maize tissues compared to other two fertilization methods. This improved P uptake was attributed to several factors associated with deep placement, including reduced P fixation, enhanced root access to P, and moisture availability for P uptake. Additionally, deep band placement combined with higher N application rates (N187 and N150) further enhanced plant P uptake by promoting P availability and utilization mechanisms. Deep band placement also resulted in significantly higher total soil P, Olsen-P, and P use efficiency than broadcast and side band methods, indicating a more efficient P fertilization strategy for maize that can improve growth and yield. This present study also found positive correlations between P concentration in plant organs and soil Olsen-P, highlighting the importance of adequate soil P levels for optimal plant growth. Overall, our results have showed that deep band fertilizer placement emerged as a superior strategy for enhancing P uptake efficiency, utilization, and maize productivity compared to broadcast and side band placement. This approach can potentially optimize P fertilizer utilization, improve maize growth and yield, while minimizing environmental P losses associated with broadcast applications.

**Keywords:** broadcast; deep band; phosphorus accumulation; phosphorus use efficiency; side band; *Zea mays*

## 1. Introduction

Phosphorus (P) is essential for various physiological processes in plants, including the constitution of phospholipids, nucleic acid, adenosine triphosphate, enzymes, and plant dry matter [1,2]. The application of P-containing fertilizers has been instrumental in increasing global food production, with significant contributions to higher agricultural productivity and global food

security [3,4]. However, P has to be rationally managed due to its limited and nonrenewable nature [5,6]. Generally, plants can only use 10-25% of the P applied to soil, with the remainder becoming unavailable due to poor solubility, strong fixation to metal cation complexes and slow diffusion in soil [1,7]. All of these have resulted in low P use efficiency and crop output, along with waterbody eutrophication and soil degradation [1,8].

Maize (*Zea mays* L.) is an important staple crop in Afghanistan after wheat and rice, with a yearly production of ~0.27 million tons from  $0.14 \times 10^6$  ha of plantation [9]. However, its productivity lags significantly behind the global average, at a 49% lower rate [10] due to various factors, including high soil pH, low P and nitrogen (N) availability, organic matter, soil moisture, and fertilizer input, and poor field management [11-13]. While Afghanistan utilizes a total of 7282 tons of P fertilizer (mostly diammonium phosphate) in 2021 to increase crop productivity [10], current application methods like broadcasting and shallow placement increases the risk of P stratification in soil surface, and soil erosion and water runoff transport concentrated P to water bodies and then eutrophication [14-16].

To reduce P losses in soil surface while improve P use efficiency, deep band placement of P fertilizer is recommended as it increases P availability in root zone [15-17]. Applying fertilizer in moist soil also helps increase nutrient mobility and availability for plant uptake [18,19]. This is especially important in semi-arid environments, where high evaporation and temperature reduce soil moisture in the upper soil layer, resulting in restricted nutrient diffusion, lower P availability to root, reduced water use efficiency and consequently, lower crop growth rate and yield [20,21]. Additionally, placing chemical fertilizers in soil subsurface could ensure that they remain in moist conditions for a longer period during the crop growth cycle [20,21].

The present study hypothesized that deep fertilizer placement could (1) significantly enhance crop nutrient uptake, particularly immobile nutrients like P and consequently, and increase tissue P concentration and accumulation, and (2) increase the fertilizer's proximity to seeds and/or roots that could result in a better utilization of fertilizer than broadcast or side band. These hypotheses are based on the fact that deep fertilizer placement improves P fertilizer efficiency in soils with high P fixing capacity and provides nutrient access in the early growing season to crop roots for vigorous plant growth and development [22]. It also encourages root proliferation in nutrient-rich patches [20], while broadcast or shallow application reduces P availability to crop roots due to lower moisture in surface soil [18,23]. Importantly, Grant and Flaten [17] showed that deep fertilizer of  $\text{NH}_4$ , urea or  $(\text{NH}_4)_2\text{SO}_4$  along with P fertilizer increases P uptake compared to individual N or P application.

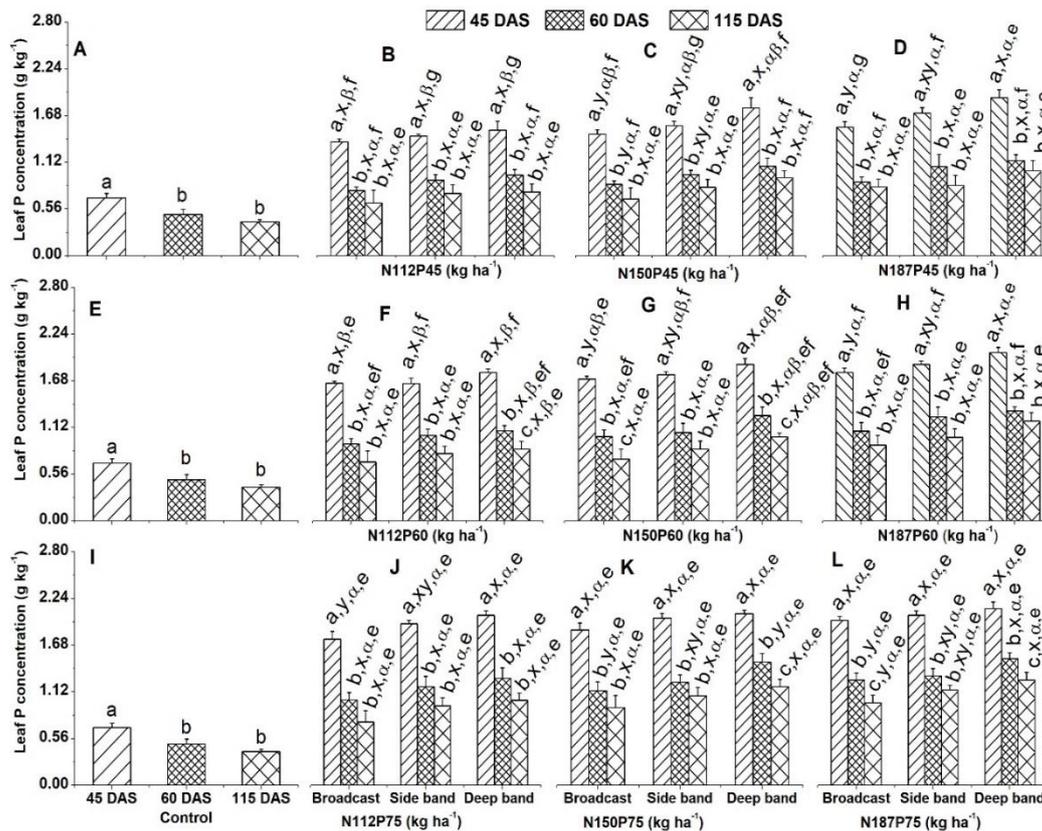
To test these assumptions, a two-year pot experiment was conducted using nine different combinations of N ( $\text{kg N ha}^{-1}$  as urea) and P ( $\text{kg P ha}^{-1}$  as superphosphate) fertilization rates (N112P45, N112P60, N112P75, N150P45, N150P60, N150P75, N187P45, N187P60, N187P75  $\text{kg ha}^{-1}$ ), with three different fertilizer placement methods (deep band, side band, and broadcast). The application corresponded to the growth and productivity of two maize hybrid cultivars of Xida-789 and Xida-211 in 2018 and 2019, respectively. Therefore, this study aimed to determine (1) the ideal  $\text{NH}_4^+$ -N and  $\text{H}_2\text{PO}_4$ -P fertilization rates, (2) a rational fertilizer placement method by comparing the three proposed fertilizer placements of broadcasting, side banding, and deep banding, and then (3) correlations between  $\text{NH}_4^+$ -N and  $\text{H}_2\text{PO}_4$  rates and placement methods for enhancing crop growth and productivity.

## 2. Results

### 2.1. Leaf P Concentration and Accumulation

#### 2.1.1. Leaf P Concentration

Leaf P concentration ( $\text{g kg}^{-1}$ ) in the no-fertilization control was significantly higher at the V8 (45 DAS) growth stage than at the VT (60 DAS) and R6 (115 DAS) growth stages (Figures 1A, E and I).



**Figure 1.** Effects of phosphorus fertilization rates and fertilizer placements on leaf P concentration of maize in the in the V8 (eight-leaf) growth stage at 45 days after sowing, VT (tasseling) stage at 60 days after sowing, and R6 (physiological maturity or harvest) stage at 115 days after sowing. Differences were showed in Figures (A, E, I) for no-fertilization control treatments between days after sowing; (B-D) between different N fertilization rates and fertilizer placements for the same P45 fertilization rate; (F-H) between different N fertilization rates and fertilizer placements for the same P60 fertilization rate; (J-L) between different N fertilization rates and fertilizer placements for the same P75 fertilization. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different growth days for the same N and P fertilization rate and same fertilizer placement or between 45 DAS, 60 DAS, and 115 DAS under the control (a, b, c), between different fertilizer placements for the same growth day and same N and P fertilization rate (x, y), between different N fertilization rates with the same P fertilization rate for the same fertilizer placement and plant growth day ( $\alpha$ ,  $\beta$ ), and between different P fertilization rates with the same N fertilization rate for the same fertilizer placement and plant growth day (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

Across various growth stages, leaf P concentration significantly differed for the same N and P fertilization rates and with the same fertilizer placement. Specifically, leaf P was significantly greater at 45 DAS than at 60 DAS and 115 DAS for N112P45, N150P45, N187P45, N187P60 and N112P75 under all placement methods (Figures 1B, C, D, H and J), N112P60, N150P75 under broadcast and side band, and N150P60 and N187P75 under side band (Figures 1F, G, K and L). Meanwhile, the highest significant leaf P concentration was observed at 45 DAS, followed by 60 DAS and the lowest at 115 DAS for N112P60 and N150P75 under deep band, as well as N150P60 and N187P75 under broadcast and deep band (Figures 1F, G, K and L).

Leaf P concentration was not significantly affected by fertilizer placement (broadcast, side band and deep band) when the same N and P fertilization rates (N112P45 and N112P60) were applied at 45 DAS, 60 DAS or 115 DAS. This was observed for N112P75 at 60 DAS and 115 DAS (Figures 1B, F and G), N150P45 at 115 DAS, N150P60 at 60 DAS and 115 DAS, N150P75 at 45 DAS and 115 DAS (Figures 1C, G and K), N187P45 and N187P60 at 60 DAS and 115 DAS, and N187P75 at 45 DAS (Figures 1D, H and L). However, deep band placement resulted in significantly higher leaf P concentration compared to broadcast placement under the combinations of N150P45, N187P45,

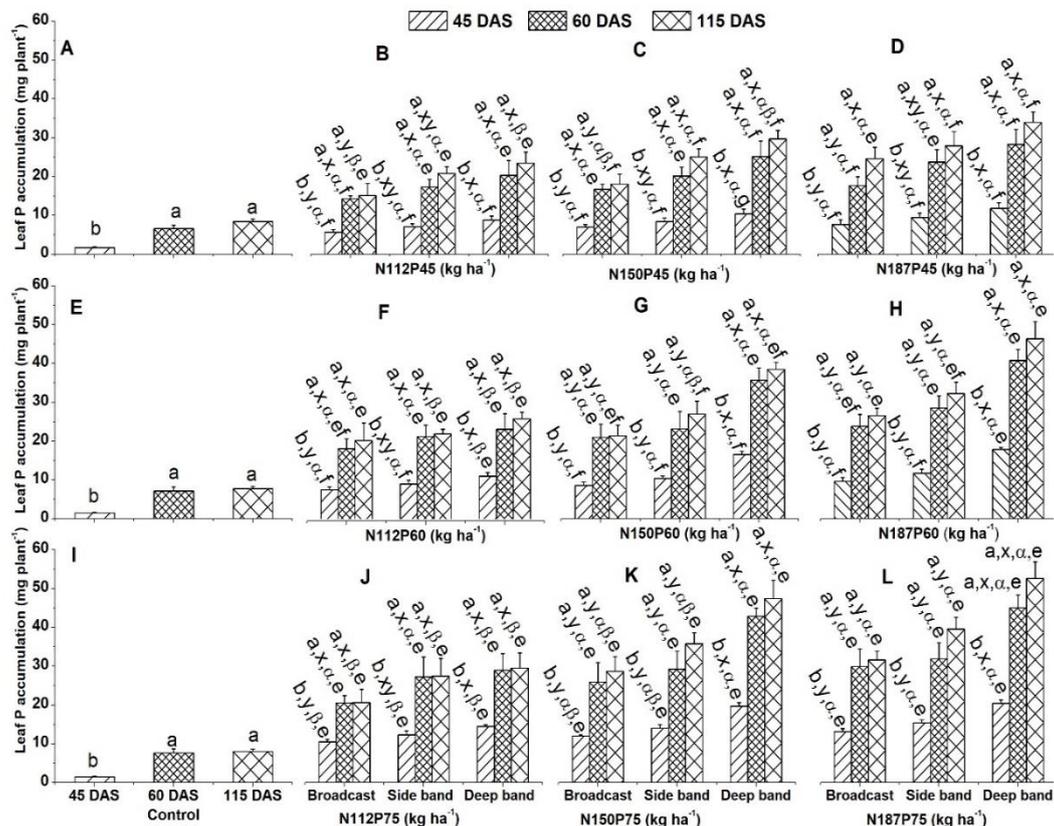
N150P60 and N187P60 at 45 DAS, N187P75 at 60 DAS and 115 DAS, N112P75 at 45 DAS, and N150P75 at 60 DAS (Figures 1C, D, H, L, J, K).

Across all three placement methods (deep band, side band, and broadcast) at 60 DAS and 115 DAS, leaf P concentration did not differ significantly between the different N fertilization rates (N112, N150 and N187) when combined with the same P fertilization rates (P45 and P60). However, at 45 DAS, leaf P concentration was significantly higher with N187 compared to N112 fertilization across all application methods (Figures 1B, C and D). Similar results were observed for the P60 fertilization rate. Leaf P concentration did not differ significantly among the N fertilization rates under side band and broadcast placements at 60 DAS and 115 DAS except at 45 DAS where N187 resulted in a significantly greater P concentration compared to N112 across all placement methods at 60 DAS and 115 DAS (Figures 1F, G and H). Meanwhile, leaf P concentration was unaffected by different N fertilization rates for the same P75 fertilization rate across all placement methods and all time points at 45 DAS, 60 DAS or 115 DAS (Figures 1J, K and L).

Leaf P concentration increased with increasing P fertilization rates (P45, P60, P75) applied at the same N rate (N112, N150, N187). This effect was observed across various application methods and timings (Figures 1B, F, J, C, G, K, D, H, L). At 45 DAS, P75 resulted in significantly higher leaf P compared to P45 for all N rates and application methods (broadcast, side band, and deep band) except N187 under deep band (Figures 1B, F, J, C, G, K). P60 also showed higher P concentration than P45 for N112 under broadcast, side and deep band at 45 DAS, N150 under side band, and N187 under side band and broadcast (Figures 1B, F, J, C, G, K). At 60 DAS, under broadcast application, P75 maintained a significantly higher P concentration than P45 for all N rates (Figures 1B, F, J). No significant differences in leaf P concentration were observed among P fertilization rates (P45, P60, P75) at later stages (115 DAS) for any N rate or application method (Figures 1B, G, J, C, G, K, D, H, L).

### 2.1.2. Leaf P Accumulation

The results showed a significant increase in leaf P accumulation ( $\text{mg plant}^{-1}$ ) over time across all fertilizer treatments (Figure 2A-L). Both the control group and P and N fertilized groups exhibited the highest leaf P accumulation at 115 DAS and 60 DAS compared to 45 DAS (Figures 2B-D, F-H and J-L).



**Figure 2.** Effects of phosphorus fertilization rates and fertilizer placements on leaf P accumulation of maize in the V8 (eight-leaf) growth stage at 45 days after sowing, VT (tasseling) stage at 60 days after sowing, and R6 (physiological maturity or harvest) stage at 115 days after sowing. Differences were shown in Figures (A, E, I) for no-fertilization control treatments between days after sowing; (B-D) between different N fertilization rates and fertilizer placements for the same P45 fertilization rate; (F-H) between different N fertilization rates and fertilizer placements for the same P60 fertilization rate; (J-L) between different N fertilization rates and fertilizer placements for the same P75 fertilization. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different growth days for the same N and P fertilization rate and same fertilizer placement or between 45 DAS, 60 DAS, and 115 DAS under the control (a, b, c), between different fertilizer placements for the same growth day and same N and P fertilization rate (x, y), between different N fertilization rates with the same P fertilization rate for the same fertilizer placement and plant growth day ( $\alpha$ ,  $\beta$ ), and between different P fertilization rates with the same N fertilization rate for the same fertilizer placement and plant growth day (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

Analysis of leaf P concentration revealed a clear pattern of higher accumulation with deep band placement compared to other methods. This was observed at 45 DAS for N112P45, N150P45, N187P45, N112P60 and N112P75 (Figures 2B, C, D, F and J), as was at 60 DAS for N187P45 and 115 DAS for N112P45 (Figures 2B, B and D). Similarly, deep band placement led to higher P accumulation than side band and broadcast methods for N150P60, N187P60, N150P75 and N187P75 at 45 DAS, 60 DAS or 115DAS (Figures 2G, H, K and L). At 115 DAS for N150P45, deep band and side band placements resulted in similar P levels (Figures 2C). In contrast, no significant differences were observed across all placement methods for N112P45, N150P45, N112P60 and N112P75 at 60 DAS (Figures 2B, C, F and J), and N187P45, N112P60 and N112P75 at 115 DAS (Figures 2D, F and J).

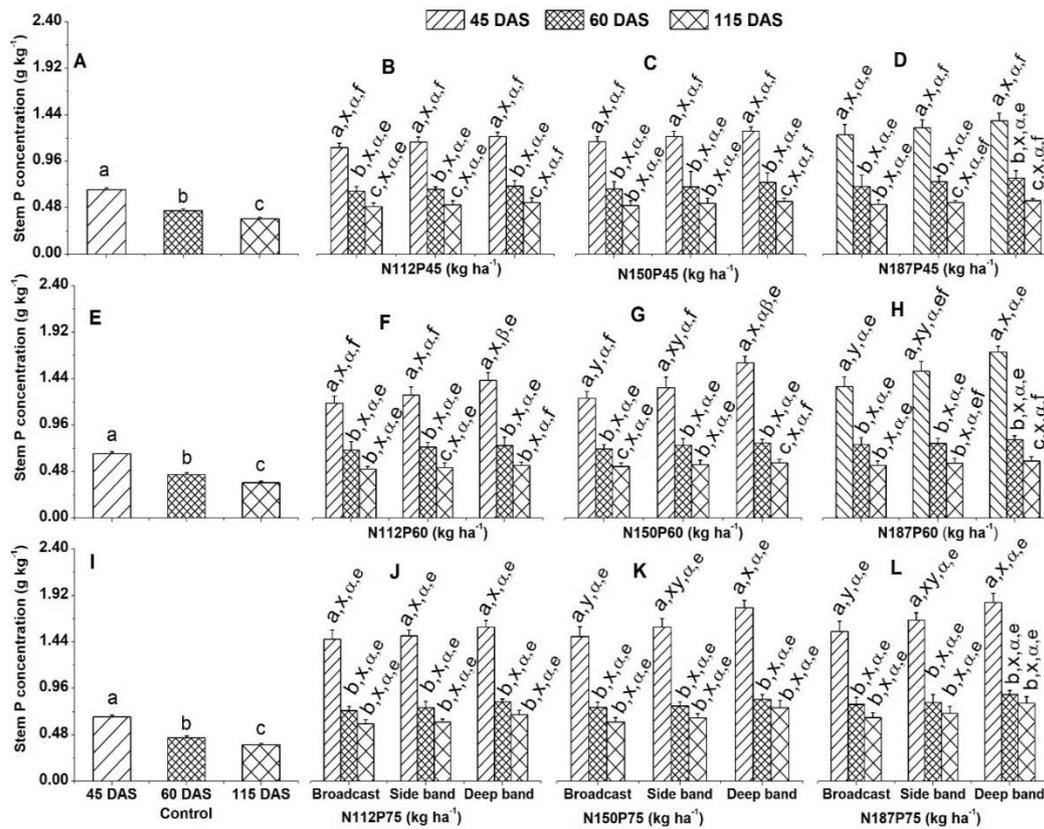
The rate of P accumulation in maize leaves was not significantly affected by the different N rates (N112, N150, and N187) when P application rate was fixed at P45, regardless of the fertilizer placement method (deep band, side band, or broadcast) and at the time of harvest (45, 60, or 115 DAS) (Figures 2B, C and D). Similar pattern was also observed for P60 fertilization rate under broadcast (at 45, 60, or 115 DAS) and side band (45 DAS and 60 DAS). However, increasing the N fertilization rates did significantly influence leaf P accumulation in N187 and N150 than in N112 under deep band at 45 DAS, 60 DAS and 115 DAS, and in N187 compared to N112 under side band at 115 DAS (Figures 2F, G, and H). At P75 rate, leaf P accumulation was significantly greater in N187 compared to N112 under broadcast and side band placement at 45 DAS and 115 DAS, and across all harvest times (45, 60, and 115 DAS) under deep placement. However, no significant difference in leaf P concentrations between N rates was observed under side band and broadcast at 60 DAS (Figures 2J, K and L). Leaf P accumulation increased significantly with increasing P fertilizer rate (P75) compared to lower rates (P45 and P60) at 45 DAS for N112, N150, and N187 under side band and broadcast, N150 under side band, and N112 under deep band (Figures 2B, F, J, C, G, K, D, H and L). The same trend was also observed at 60 DAS for N112 and N187 under broadcast, and at 115 DAS for N150 under broadcast and deep band, N187 under side band (Figures 2, B, J, C, K, D and L). However, there were no significant differences in P concentration observed between P fertilization rates for N112 at 115 DAS under broadcast and side band applications, nor for N150 at 60 DAS under both broadcast and side band, and for N187 under broadcast at 115 DAS and under side band at 60 DAS (Figures 2C, G, K, D, H, L).

## 2.2. Stem P Concentration and Accumulation

### 2.2.1. Stem P Concentration

Stem P concentration ( $\text{g kg}^{-1}$ ) showed significant decline over time in the no-fertilization control treatment (Figures 3A, E and I). A similar trend was observed for all N and P fertilizer treatments and placement. Stem P concentration was significantly higher at 45 DAS compared to both 60 and 115 DAS for N112P75, N150P75 and N187P75 under deep band, side band, and broadcast applications (Figures 3J, K and L), as were N150P45 and N187P60 under side band and broadcast (Figures 3C and H), N187P45 under broadcast, N112P60 under deep band and broadcast, N150P60 under side band (Figures 3D, F and G), N112P45 under all placement methods, N150P45 and N187P60 under deep

band (Figures 3B, C and H), N187P45 under side band and deep band, N112P60 under side band, and N187P60 under deep band (Figures 3D, F and H).



**Figure 3.** Effects of phosphorus fertilization rates and fertilizer placements on stem P concentration of maize in the V8 (eight-leaf) growth stage at 45 days after sowing, VT (tasseling) stage at 60 days after sowing, and R6 (physiological maturity or harvest) stage at 115 days after sowing. Differences were showed in Figures (A, E, I) for no-fertilization control treatments between days after sowing; (B-D) between different N fertilization rates and fertilizer placements for the same P45 fertilization rate; (F-H) between different N fertilization rates and fertilizer placements for the same P60 fertilization rate; (J-L) between different N fertilization rates and fertilizer placements for the same P75 fertilization. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different growth days for the same N and P fertilization rate and same fertilizer placement or between 45 DAS, 60 DAS, and 115 DAS under the control (a, b, c), between different fertilizer placements for the same growth day and same N and P fertilization rate (x, y), between different N fertilization rates with the same P fertilization rate for the same fertilizer placement and plant growth day ( $\alpha$ ,  $\beta$ ), and between different P fertilization rates with the same N fertilization rate for the same fertilizer placement and plant growth day (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

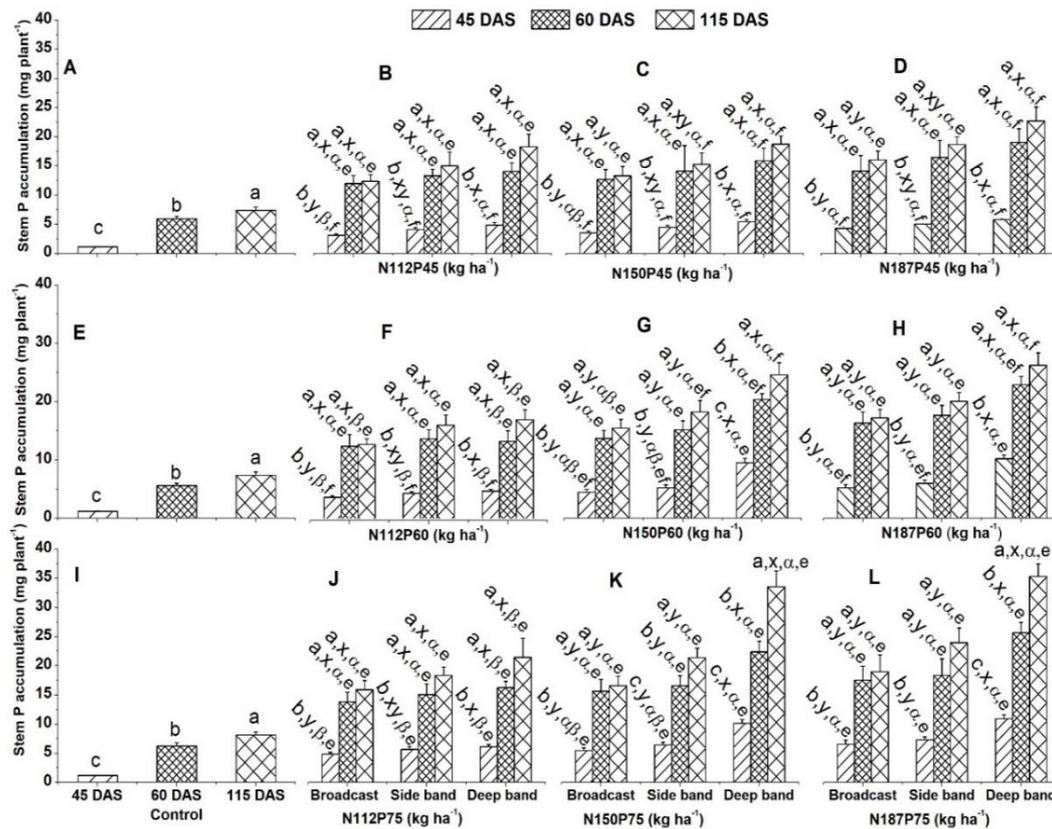
The effect of fertilizer placement (deep band, side band, and broadcast) on stem P concentration was minimal across all N (N112, N150, N187) and P (P45, P60, P75) fertilization rates at 45, 60, and 115 DAS, with no significant differences observed (Figures 3B-D, F-H and J-L). However, at 45 DAS, deep placement resulted in a significantly higher stem P concentration compared to broadcast placement for the N150P60, N187P60, N150P75 and N187P75 treatments (Figures 3G, H, K and L). Similarly, N fertilization rates (N112, N150, N187) did not significantly affect stem P concentration at any given P fertilization rate (P45, P60, P75) and placement (deep band, side band, broadcast) at 45 DAS, 60 DAS or 115 DAS, with one exception. Under deep placement at 45 DAS, maize fertilized with N187 showed a significantly greater stem P concentration compared to those receiving N112 for the same P60 treatment (Figures 3B-D, F-H and J-L).

Stem P concentration increased significantly with increasing P fertilization rates (P75 > P60 > P45) at specific N rates (N112 and N150) and placement methods. This effect was observed at 45 DAS

under broadcast and side band applications (Figures 3B, F, J, C, G, K), and 115 DAS under deep band application for N112, N150 and N187 (Figures 3D, H, and L). In contrast, no significant differences in stem P concentration were found among P rates at N112 and N150 under broadcast and side band applications at 60 DAS and 115 DAS (Figures 3B, C, F, G, J, and K), N187 under broadcast at 45 DAS, 60 DAS and 115 DAS (Figures 3D, H, L), side band and deep band at 60 DAS, and N112 and N150 under deep band at 60 DAS (Figures 3B, F, J, C, G, K, D, H and L).

### 2.2.2. Stem P Accumulation

Maize grown without fertilizer (control group) showed the highest stem P accumulation (mg plant<sup>-1</sup>) at 115 DAS, followed by 60 DAS and then 45 DAS (Figures 4A, E and I).



**Figure 4.** Effects of phosphorus fertilization rates and fertilizer placements on stem P accumulation of maize in the V8 (eight-leaf) growth stage at 45 days after sowing, VT (tasseling) stage at 60 days after sowing, and R6 (physiological maturity or harvest) stage at 115 days after sowing. Differences were shown in Figures (A, E, I) for no-fertilization control treatments between days after sowing; (B-D) between different N fertilization rates and fertilizer placements for the same P45 fertilization rate; (F-H) between different N fertilization rates and fertilizer placements for the same P60 fertilization rate; (J-L) between different N fertilization rates and fertilizer placements for the same P75 fertilization. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different growth days for the same N and P fertilization rate and same fertilizer placement or between 45 DAS, 60 DAS, and 115 DAS under the control (a, b, c), between different fertilizer placements for the same growth day and same N and P fertilization rate (x, y), between different N fertilization rates with the same P fertilization rate for the same fertilizer placement and plant growth day ( $\alpha$ ,  $\beta$ ), and between different P fertilization rates with the same N fertilization rate for the same fertilizer placement and plant growth day (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

Across all fertilization regimes (N and P rates and placement methods), stem P accumulation was significantly higher at 115 DAS and 60 DAS compared to 45 DAS. This pattern was observed for N112P45, N150P45, N187P45, N112P60, N187P60, N112P75 under deep band, side band and

broadcast (Figures 4B, C, D, F, H and J), N150P60 and N187P75 under broadcast and side band (Figures 4G and L), and N150P75 under broadcast (Figures 4K). For N150P60 and N187P75 fertilized maize, deep band application resulted in significantly higher stem P accumulation at 115 DAS compared to 60 DAS, which in turn was higher than 45 DAS (Figures 4G, L and K). A similar trend was also observed for N150P75 with side band and deep band placements.

Deep band placement resulted in significantly greater stem P accumulation compared to side band and broadcast applications under N150P60, N187P60, N150P75 and N187P75 at 45 DAS, 60 DAS or 115 DAS (Figures 4G, H, K and L). Similar trend was also observed for N112P45, N112P60 and N112P75 at 45 DAS (Figures 4B, F and J), N150P45 and N187P45 at 45 DAS and 115 DAS (Figures 4C and D). However, no significant difference was found between placement methods (deep band, side band, broadcast) for N112P45, N112P60 and N112P75 at 60 DAS and 115 DAS (Figures 4B, F and J), and N150P45 and N187P45 at 60 DAS (Figures 4C-D).

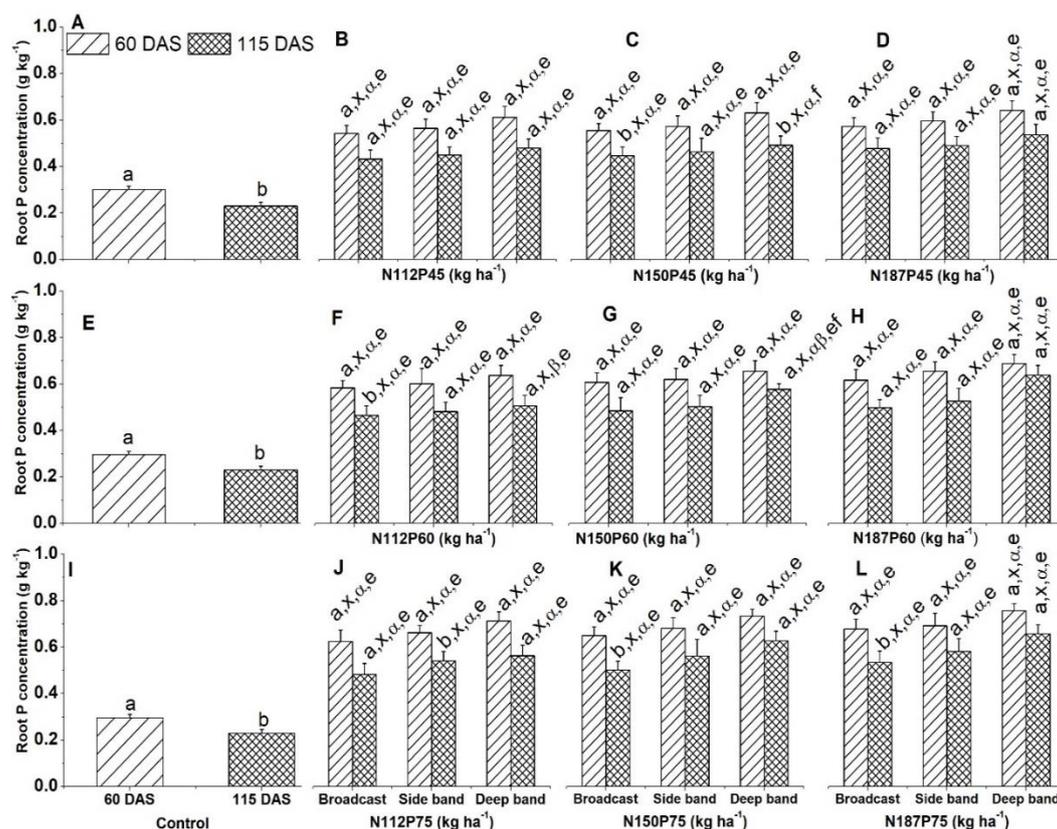
Among different N rates, stem P accumulation was significantly greater at N187 than N112 for the same P45 under broadcast at 45 DAS; with the same P60 under broadcast at 45 DAS and 115 DAS, under side band at 45 DAS; and with the same P75 under broadcast and side band at 45 DAS (Figures 4B and D, F and H, J and L). Moreover, stem P accumulation was significantly higher at N187 and N150 than N112 for the same P60 under Deep band at 45 DAS, 60 DAS or 115 DAS; and with the same P75 under deep band at 45 DAS, 60 DAS or 115 DAS (Figures 4B-D, F-H and J-L). However, no significant difference was observed among different N rates with the same P45 at 60 DAS and 115 DAS; with the same P60 under broadcast at 60 DAS, and under side band at 60 DAS and 115 DAS; and with the same P75 under broadcast and side band at 60 DAS and 115 DAS under all fertilizer placements (Figures 4B-D, F-H and J-L).

For maize receiving the same amount of N fertilizer, those that received the highest P rate (P75) accumulated more P in their stems compared to those receiving lower P rates (P45 and P60) at 45 DAS. This trend was observed across all fertilizer placement methods (broadcast, side band, and deep band) for N150 and N187 (Figures 4C, G, K, D, H and L). Similarly, for N112, P75 resulted in greater stem P accumulation compared to P45 across all placement methods at 45 DAS (Figures 4B, F, and J). However, at 60 and 115 DAS, the effect of P fertilization rate on stem P accumulation diminished as there were no significant differences between P45, P60, and P75 for any N rate or fertilizer placement method (Figures 4B, C, D, F, G, H, J, K, L).

### 2.3. Root P Concentration and Accumulation

#### 2.3.1. Root P Concentration

Maize grown without fertilizer (control) showed higher concentrations of P ( $\text{g kg}^{-1}$ ) in their roots at 60 DAS (Figures 5A, E and I). Likewise, higher root P concentration at 60 DAS was also observed for several combinations, namely N112P60, N150P75 and N187P75 under broadcast placement (Figures 5C, K and L), N150P45 under broadcast and deep band (Figures 5C), and N112P60 under broadcast (Figures 5F). However, root P concentration did not differ significantly between 60 DAS and 115 DAS for N112P45, N187P45, N150P60 and N187P60 under all three placement methods (Figures 5B, D, G and H); N112P60, N150P75 and N187P75 under side band and deep band (Figures 5F, K and L); N150P45 under side band, and N112P75 under broadcast and deep band (Figures 5C and J).



**Figure 5.** Effects phosphorus fertilization rates and fertilizer placements on root P concentration of maize in the VT (tasseling) stage at 60 days after sowing, and R6 (physiological maturity or harvest) stage at 115 days after sowing. Differences were showed in Figures (A, E, I) for no-fertilization control treatments between days after sowing; (B-D) between different N fertilization rates and fertilizer placements for the same P45 fertilization rate; (F-H) between different N fertilization rates and fertilizer placements for the same P60 fertilization rate; (J-L) between different N fertilization rates and fertilizer placements for the same P75 fertilization. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different growth days for the same N and P fertilization rate and same fertilizer placement or between 45 DAS, 60 DAS, and 115 DAS under the control (a, b, c), between different fertilizer placements for the same growth day and same N and P fertilization rate (x, y), between different N fertilization rates with the same P fertilization rate for the same fertilizer placement and plant growth day ( $\alpha$ ,  $\beta$ ), and between different P fertilization rates with the same N fertilization rate for the same fertilizer placement and plant growth day (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

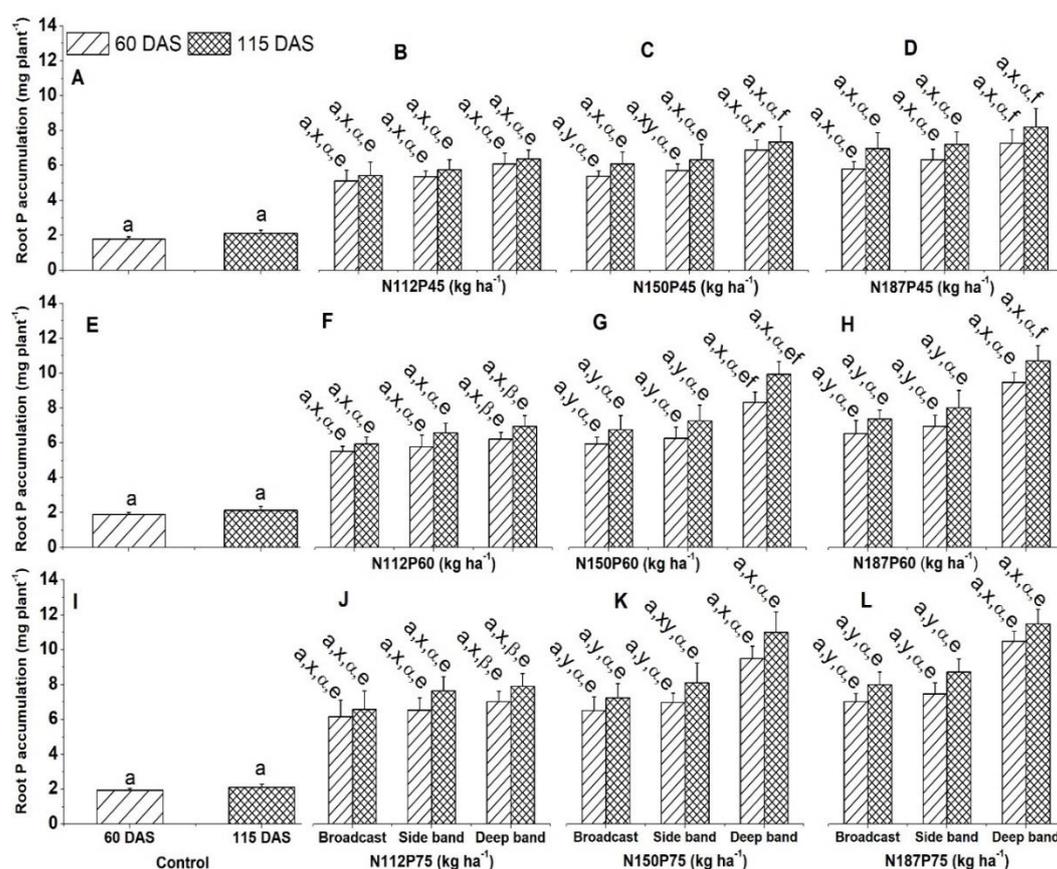
Fertilizer placement methods (deep band, side band, or broadcast) did not significantly affect root phosphorus concentration at either 60 DAS or 115 DAS for any fertilizer combination tested (N112P45, N150P45, N187P45, N112P60, N150P60, N187P60, N112P75, N150P75 and N187P75) (Figures 5B-D, F-H and J-L).

N fertilizer rate (N112, N150, or N187) also had no significant impact on root P concentration, regardless of the P rate (P45, P60 or P75) at both 60 and 115 DAS. The only exception was a higher concentration observed in N187 compared to N112 for the same P60 rate under deep band placement at 115 DAS (Figures 5F and H). P fertilizer rate (P45, P60, and P75) also did not affect root P concentration when applied with the same N rate (N112, N150 or N187) and placement method at 60 and 115 DAS. The sole exception was a significantly greater concentration in P75 compared to P45 for N150 under deep band at 115 DAS (Figures 5B, F, J, C, G, K and D, H and L).

### 2.3.2. Root P Accumulation

Maize grown without fertilizer (control) showed no difference in P accumulation ( $\text{g plant}^{-1}$ ) in their roots 60 DAS and 115 DAS (Figures 6A, E and I). Fertilizer placement had a significant effect, with deep band placement resulted in maize accumulating higher root P compared to those applied

under side band or broadcast placement for N150P60 and N187P60 at 60 DAS and 115 DAS, N150P75 at 60 DAS, N187P75 at 60 DAS and 115 DAS (Figures 6G, H, K and L), N150P45 at 60 DAS and N150P75 at 115 DAS (Figures 6C and K). For the combination of N fertilization rates (N112, N150, N187) with P45, no significant differences in root P accumulation across all placement methods at 60 and 115 DAS (Figures 6B, C, and D). Similarly, root P accumulation was not significantly different among different N fertilization rates for the same P fertilization rate either at P60 or P75 under side band and broadcast at 60 DAS and 115 DAS, but the combination of N187 and N150 rates with P75 and P60 resulted in higher root P accumulation under deep band placement at 60 and 115 DAS (Figures 6F, G, H, J, K and L).



**Figure 6.** Effects of phosphorus fertilization rates and fertilizer placements on root P accumulation of maize in the VT (tassling) stage at 60 days after sowing, and R6 (physiological maturity or harvest) stage at 115 days after sowing. Differences were showed in Figures (A, E, I) for no-fertilization control treatments between days after sowing; (B-D) between different N fertilization rates and fertilizer placements for the same P45 fertilization rate; (F-H) between different N fertilization rates and fertilizer placements for the same P60 fertilization rate; (J-L) between different N fertilization rates and fertilizer placements for the same P75 fertilization. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different growth days for the same N and P fertilization rate and same fertilizer placement or between 45 DAS, 60 DAS, and 115 DAS under the control (a, b, c), between different fertilizer placements for the same growth day and same N and P fertilization rate (x, y), between different N fertilization rates with the same P fertilization rate for the same fertilizer placement and plant growth day ( $\alpha$ ,  $\beta$ ), and between different P fertilization rates with the same N fertilization rate for the same fertilizer placement and plant growth day (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

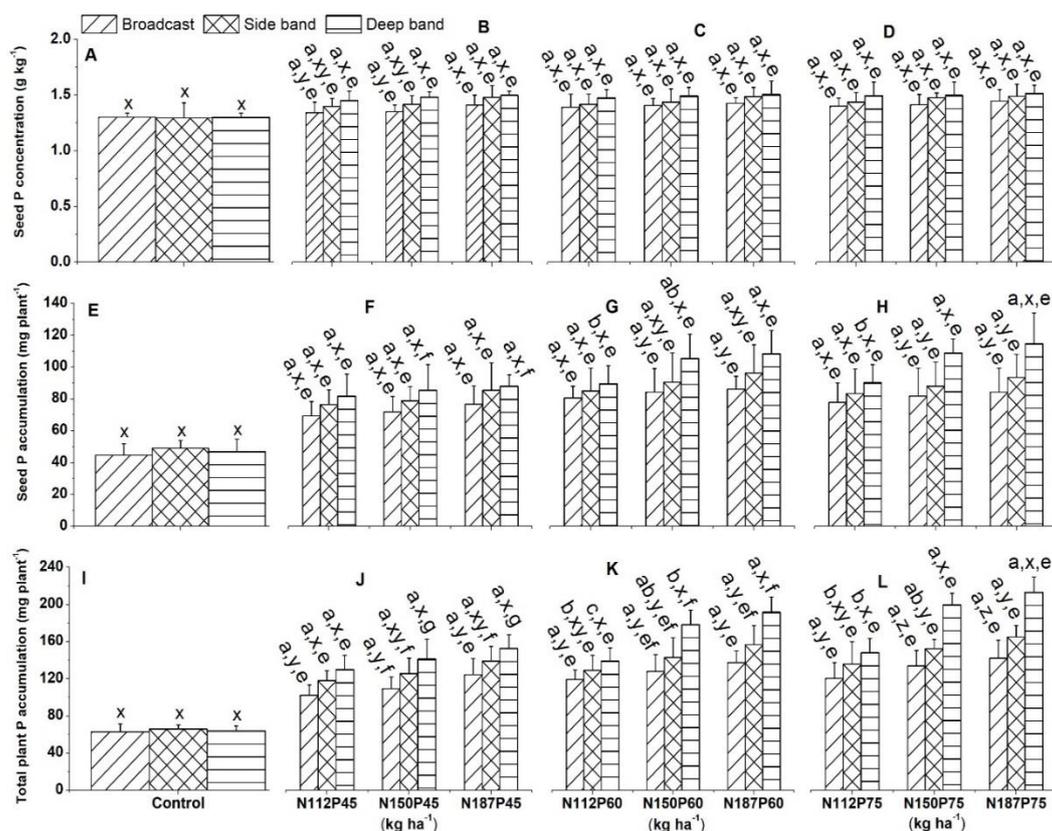
Meanwhile, increasing P fertilizer rates from P45 to P75 did not significantly affect root P accumulation at two different application methods (broadcast and side band) and for three N rates (N112, N150, N187) at 60 and 115 DAS. This was also observed for N112 with deep band application at 45 DAS (Figures 6B, F, J, C, J, K, D, H and L). However, under deep band application, significantly greater root P accumulation was observed in P75 compared to P45 for N150 at both 60 and 115 DAS,

and for N187 at 115 DAS (Figures 6C, K, D and L). For N187 at 60 DAS, both P75 and P60 resulted in higher root P accumulation compared to P45 (Figures 6D, H and L).

## 2.4. Seed P Concentration and Accumulation

### 2.4.1. Seed P concentration

In pots with no fertilizer was applied (control treatment), seed P concentration did not differ significantly between the deep band, side band, and broadcast placements at 115 DAS (Figures 7A). When fertilizer was applied, seed P concentration was significantly higher with deep band placement compared to broadcast placement for the lower N and P rates (N112P45 and N150P45) (Figure 7B). However, the effect of deep band placement diminished when the higher N and P rates (N187P45, N112P60, N150P60, N186P60, N112P75, N150P75, N187P75) were applied (Figures 7C and D).



**Figure 7.** Effects of phosphorus fertilization rates and fertilizer placement on seed P concentrations (A-D), seed P accumulations (E-H), and total plant (leaf + stem + seed + root) P accumulations (I-L) of maize crop at the R6 (physiological maturity or harvest) stage at 115 days after sowing. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different N fertilization rates for the same P fertilization and fertilizer placement (a, b), between different fertilizer placements for the same NP fertilization rate (x, y), and between different P fertilization rates for the same N fertilization and fertilizer placement (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

### 2.4.2. Seed P accumulation

In the control pots without fertilizer, seed P accumulation ( $\text{mg plant}^{-1}$ ) levels were similar at 115 DAS (Figures 7E). Fertilizer application did however affect seed P accumulation. When the same rates of N and P fertilizer were applied, deep band placement resulted in significantly higher seed P accumulation compared to side band and broadcast application for the higher N and P rates (N150P60, N187P60, N150P75, and N187P75) (Figures 7G and H). However, seed P levels were similar across all three placement methods (deep band, side band, broadcast) for the lower N and P rates (N112P45, N150P45, N187P45, N112P60, N112P75) (Figures 7F, G and H).

### 2.5. Total plant P accumulation

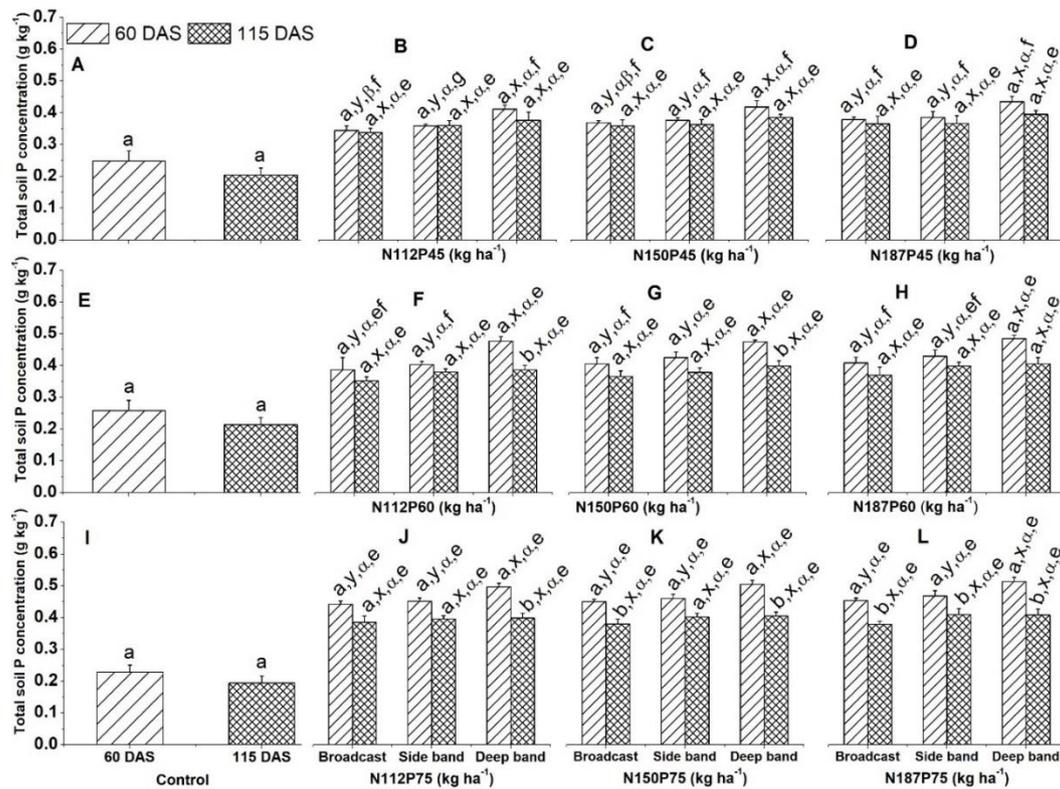
Maize grown in the control pots (no fertilizer) showed no significant difference in total plant P accumulation (sum of leaf, stem, seed, and root, in mg plant<sup>-1</sup>) with those fertilized under deep band, side band, and broadcast placement (Figures 7I). When fertilizer was applied, deep placement resulted in significantly greater total plant P accumulation compared to broadcast for most fertilizer combinations (N150P45, N187P45, N112P60, N112P75) (Figures 7J, K and L). Interestingly, total plant P accumulation was significantly higher under deep band placement compared to side band and broadcast for fertilizer combinations of N150P75 and N187P75 (Figures 7L).

Total plant P accumulation among N rates (N112, N150, N187) for the same P45 rate was not significantly produced under all placement methods, as well as for the same P60 and P75 under broadcast (Figures 7J). However, total plant P accumulation was significantly greater in N187 than N112 with the same P60 and P75 under side band (Figures 7K and L), while significantly greater total P accumulation was produced in N187 followed by N150 and lower at N112 for the P60 under deep band (Figure 7K).

Total plant P accumulation among P rates (P45, P60, P75) was significantly greater in P75 than P45 for the same N150 under broadcast and side band, as well as for the same N187 under side band (Figures 7J, K and L). However, significantly greater total P accumulation was produced in P75 followed by P60 and lower in P45 for the same N150 and N187 under deep band (Figures 7J, K and L). No significant difference was observed among different P rates for the same N112 under all placement methods (Figures 7J, K and L).

### 2.6. Total soil P concentration

Total soil P concentration (g kg<sup>-1</sup>) showed varied responses to fertilization rate, placement methods, and sampling day. In the control treatment with no fertilization, no significant differences in P concentration at 60 DAS and at 115 DAS (Figures 8A, E and I). However, total soil P concentration among different growth days for the same N and P fertilization rate and same fertilizer placement was similar at 60 DAS and 115 DAS for N112P45, N150P45, N187P45 and N187P60 under Deep band, Side band and Broadcast (Figures 8B-D and H), for N112P60, N150P60 and N112P75 under Broadcast and Side band (Figures 8F, G and J), also for N150P75 under Side band (Figures 8K), while total soil P concentration was significantly greater at 60 DAS than at 115 DAS for N112P60, N150P60 and N112P75 under Deep band, also for N187P75 under Deep band, Side band and Broadcast (Figures 8F, G, J, L).



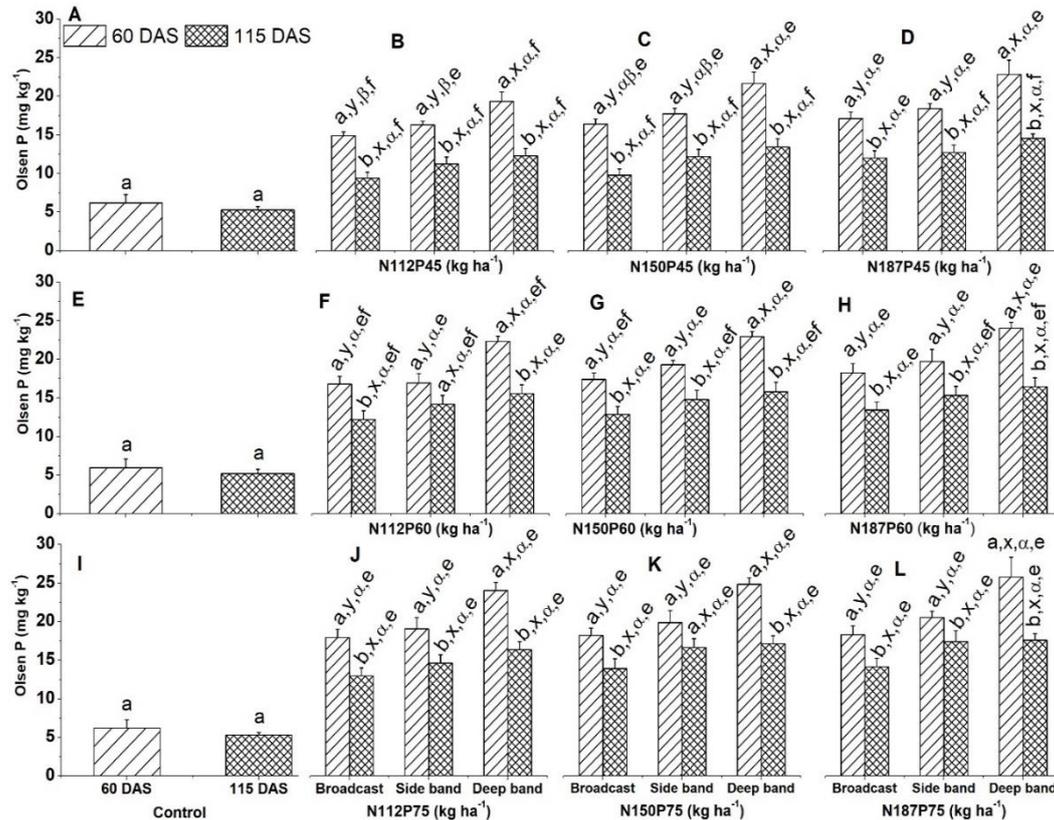
**Figure 8.** Effects of phosphorus fertilization rates and fertilizer placements on total soil P concentration of maize in the VT (tasseling) stage at 60 days after sowing, and R6 (physiological maturity or harvest) stage at 115 days after sowing. Differences were shown in Figures (A, E, I) for no-fertilization control treatments between days after sowing; (B-D) between different N fertilization rates and fertilizer placements for the same P45 fertilization rate; (F-H) between different N fertilization rates and fertilizer placements for the same P60 fertilization rate; (J-L) between different N fertilization rates and fertilizer placements for the same P75 fertilization. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different growth days for the same N and P fertilization rate and same fertilizer placement or between 45 DAS, 60 DAS, and 115 DAS under the control (a, b, c), between different fertilizer placements for the same growth day and same N and P fertilization rate (x, y), between different N fertilization rates with the same P fertilization rate for the same fertilizer placement and plant growth day ( $\alpha$ ,  $\beta$ ), and between different P fertilization rates with the same N fertilization rate for the same fertilizer placement and plant growth day (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

Among different fertilizer placement for the same N and P fertilization rate, total soil P concentration was significantly greater in deep band compared to side band and broadcast placement under all combination of fertilizer rates (N112P45, N150P45, N187P45, N112P60, N150P60, N187P60, N112P75, N150P75 and N187P75) at 60 DAS, while no significant difference was observed between deep band, side band and broadcast under the same N and P fertilization rate at 115 DAS (Figures 8C-D, F-H and J-L). Similarly, no significant difference in soil P concentration between N fertilization rates (N112, N150, N187) when combined with either P45, P60, or P75 at either 60 or 115 DAS (Figures 8C-D, F-H and J-L), regardless of placement method. The only exception was a higher concentration observed with N187 compared to N112 for the P45 rate under broadcast application at 60 DAS (Figures 8C and D).

Total soil P concentration among P rates (P45, P60, P75) was significantly increased at P75 and P60 than P45 for the same N112, N150 and N187 under deep band at 60 DAS (Figures 8B, F, J, C, G, K, D, H and L), as well as at P75 than P45 for the same for the same N112 and N187 under broadcast and side band at 60 DAS (Figures 8B, F, J, D, H and L), and at P75 than P60 and P45 for the same N150 and N187 under broadcast at 60 DAS (Figures 8 G, K, D, H and L). However, total soil P concentration was similar among P rates for the same N rates and same and same placement methods (Figures 8B, F, J, C, G, K, D, H and L).

## 2.7. Olsen P

In the control pots with no fertilization, Olsen P ( $\text{mg kg}^{-1}$ ) showed no significant difference at 60 DAS and 115 DAS (Figures 9A, E and I). Across all fertilization rates and placements, Olsen P was significantly higher at 60 DAS compared to 115 DAS, as observed in N112P45, N150P45, N187P45, N150P60, N187P60, N112P75 and N187P75 under deep band, side band and broadcast (Figures 9B, C, D, G, H, J and L). Similar trend was also observed for N112P60 and N150P75 under broadcast and deep band (Figures 9F and K). However, Olsen P levels at 60 DAS and at 115 DAS were not different for N112P60 and N150P75 under side band application (Figures 9F and K).



**Figure 9.** Effects of phosphorus fertilization rates and fertilizer placements on Olsen-P concentration of maize in the VT (tassling) stage at 60 days after sowing, and R6 (physiological maturity or harvest) stage at 115 days after sowing. Differences were showed in Figures (A, E, I) for no-fertilization control treatments between days after sowing; (B-D) between different N fertilization rates and fertilizer placements for the same P45 fertilization rate; (F-H) between different N fertilization rates and fertilizer placements for the same P60 fertilization rate; (J-L) between different N fertilization rates and fertilizer placements for the same P75 fertilization. Data (means  $\pm$  SE,  $n = 6$ ) are averaged under three fertilizer placements for the control treatments and under the same N and P fertilizer for the same fertilizer placement since no significant differences were observed between maize varieties and years (2018 and 2019), and followed by different letters indicate significant differences between different growth days for the same N and P fertilization rate and same fertilizer placement or between 45 DAS, 60 DAS, and 115 DAS under the control (a, b, c), between different fertilizer placements for the same growth day and same N and P fertilization rate (x, y), between different N fertilization rates with the same P fertilization rate for the same fertilizer placement and plant growth day ( $\alpha$ ,  $\beta$ ), and between different P fertilization rates with the same N fertilization rate for the same fertilizer placement and plant growth day (e, f) at  $P < 0.05$ . Abbreviations: DAS, days after sowing; N, nitrogen; P, phosphorus.

When comparing fertilizer placements with the same N and P rates, deep band application resulted in significantly higher Olsen P levels at 60 DAS compared to side band and broadcast applications. This was observed for N112P45, N150P45, N187P45, N112P60, N150P60, N187P60, N112P75, N150P75 and N187P75 at 60 DAS (Figures 9B-D, F-H and J-L). At 115 DAS, deep band placement showed higher Olsen P compared to broadcast application for N112P45, N150P45 and N112P75 (Figures 9B, C and J). However, there were no significant differences in Olsen P levels

between deep band, side band, and broadcast at 115 DAS for N187P45, N112P60, N150P60, N187P60, N150P75 and N187P75 (Figures 9D, F, G, H, K and L).

Olsen P levels in the soil were not significantly affected by the different N rates (N112, N150, N187) when applied with the same P rates (P45, P60, P75) and fertilizer placement method (deep band, side band, or broadcast) at 60 and 115 DAS (Figures 9B-D, F-H and J-L). However, at 60 DAS, N187 resulted in significantly higher Olsen P compared to N112 when P45 was applied with broadcast and side band placement (Figures 9B and D).

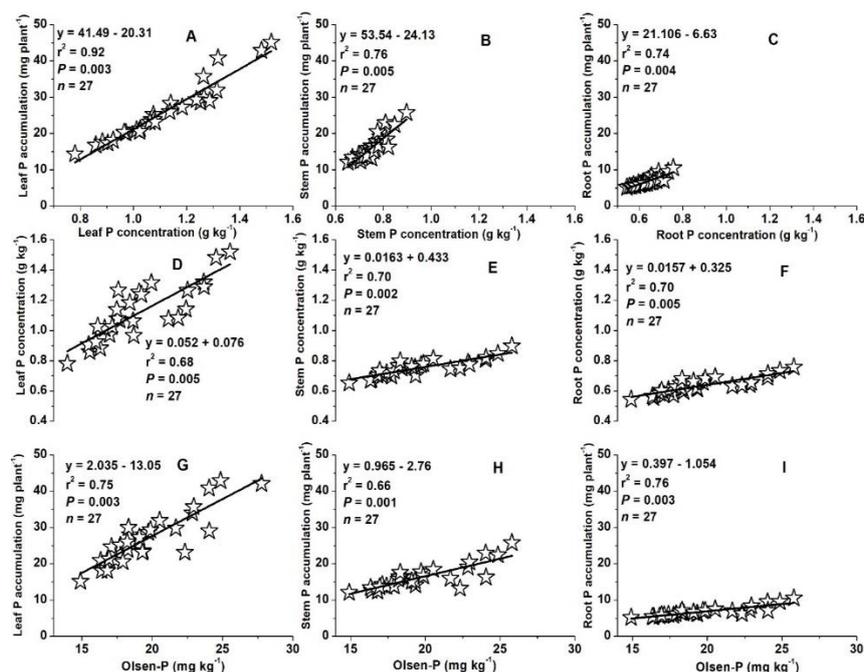
On the other hand, P fertilization rates significantly influenced Olsen P levels. When the N rate and placement method were constant, Olsen P was significantly higher with P75 application compared to P45 (Figures 9B, F, and J). This was observed for N112 under broadcast at both 60 and 115 DAS, under side band at 115 DAS, and under deep band at 60 DAS. Similarly, N150 and N187 also showed higher Olsen P with P75 compared to P45 under deep band, side band, and broadcast at 115 DAS (Figures 9C, D, F, G, H, and L). However, there were no significant differences in Olsen P among P fertilization rates (P45, P60, P75) for specific combinations of N rate and placement method (Figures 9B, C, F, G, H, J, and K).

### 2.8. Phosphorus Use Efficiency

Deep fertilizer placement significantly enhanced P uptake by the crop. This was evident in greater phosphorus agronomy efficiency (PAE) and partial factor productivity for applied P (PFP<sub>P</sub>) compared to broadcast and side band fertilization, particularly under N150P75 and N187P75, and phosphorus use efficiency (PUE) under N150P60, N187P60, N150P75, N187P75. Furthermore, for the same P rate and placement method, increasing N rates from N112 to N150 or N187 significantly enhanced both PUE and PFP<sub>P</sub> under deep band placement; this trend was not observed with broadcast or side band methods. Interestingly, at constant N rate i.e., N150 and N187, applying lower P rate (P45) resulted in a higher PFP<sub>P</sub> compared to P60 or P75 under all placement methods.

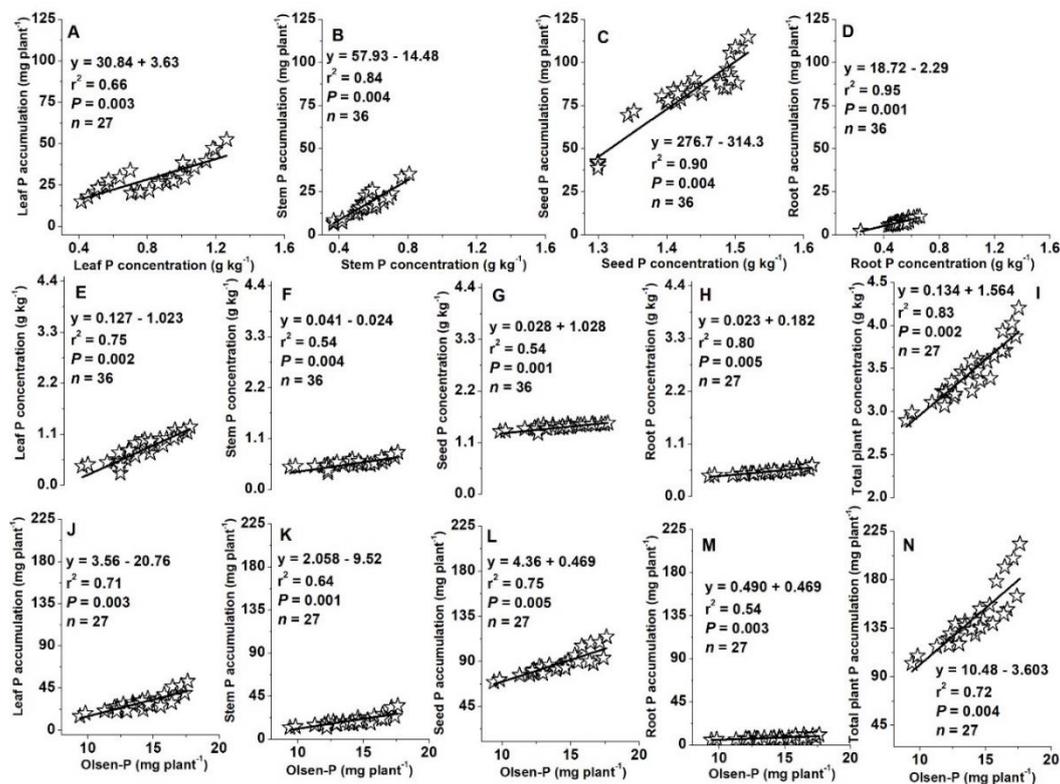
### 2.9. Relationships between plant tissue P accumulations and concentrations or between tissue P concentrations or accumulations and soil Olsen-P concentrations

Plant tissue phosphorus (P) concentration and accumulations showed strong positive correlations in leaves, stems, and roots ( $r^2 = 0.92 - 0.76 - 0.74$ ,  $P = 0.003 - 0.005 - 0.004$ ) at the VT (tasseling, at 60 DAS) growth stage (Figures 10A, B and C). Similarly, P concentrations and accumulations in each tissue (leaf, stem, and root P) were positively related to the available soil phosphorus (Olsen-P) at 60 DAS ( $r^2 = 0.68 - 0.70 - 0.70$ ,  $0.75 - 0.66 - 0.76$ ,  $P = 0.005 - 0.002 - 0.005$ ,  $0.003 - 0.001 - 0.003$ ) (Figures 10D - F, H - I).



**Figure 10.** Relationships between tissue P concentrations and accumulations in leaf (A), stem (B) or root (C) at the VT (tasseling, at 60 days after sowing) growth stage, also between tissue P concentrations or P accumulations in leaf (D, G), stem (E, H) and root (F, I) and Olsen-P concentrations at 60 days after sowing.

At harvest (115 DAS), positive correlations were also observed between P concentration and accumulation in leaves, stems, seeds, and roots ( $r^2 = 0.66 - 0.84 - 0.90 - 0.95$ ,  $P = 0.003 - 0.004 - 0.004 - 0.001$ , Figures 11A, B, C and D). Likewise, Olsen-P concentration in the soil showed positive relationships with P concentrations in all plant tissues (leaf, stem, seed, root) and total plant (leaf + stem + seed + root) at 60 DAS ( $r^2 = 0.75 - 0.54 - 0.54 - 0.80 - 0.83$ ,  $P = 0.002 - 0.004 - 0.001 - 0.005 - 0.002$ , Figures 11E-I). Finally, Olsen-P concentration also showed positive correlations with P accumulation in all plant tissues and total plant at 115 DAS ( $r^2 = 0.71 - 0.64 - 0.75 - 0.54 - 0.72$ ,  $P = 0.003 - 0.001 - 0.005 - 0.003 - 0.004$ , Figures 11J-N).



**Figure 11.** Relationships between tissues P concentration and accumulation in leaf (A), stem (B), seed (C), and root (D) at the R6 (physiological maturity or harvest at 115 days after sowing), between tissue P concentration in leaf (E), stem (F), seed (G), root (H) and total plant (leaf + stem + seed + root; I) and soil Olsen-P at 115 DAS, and between tissue P accumulation in leaf (J), stem (K), seed (L), root (M) and total plant (leaf + stem + seed + root; N) and soil Olsen-P at 115 days after sowing.

### 3. Discussions

Deep fertilizer placement fulfills the moisture requirement for nutrient dissolution process in the deeper soil layer, and in turn, increases nutrient availability for root uptake by maize plants and prevents from nutrient deficiencies [24]. Studies have shown that placing N and P fertilizers near maize seeds enhances their synergistic interaction, increases nutrient concentration and P uptake, promotes growth and development, nutrient use efficiency, and overall productivity, ultimately leading to higher seed yields [19,25]. To verify these potential growth benefits, a two-year (2018 and 2019) greenhouse pot experiment was conducted to examine nine different combinations of N and P fertilization rates (N112P45, N112P60, N112P75, N150P45, N150P60, N150P75, N187P45, N187P60, and N187P75 kg ha<sup>-1</sup>) applied using three placement methods (deep band, side band and broadcast) and the agronomic performance of two maize hybrid cultivars (Xida-789 and Xida-211) were investigated.

### 3.1. Greater P Concentration under Deep Band

Deep band fertilizer placement significantly enhanced phosphorus (P) uptake compared to broadcast placement. This was evident in increased leaf P concentration by 9.6% and 10.2% under N150P45 and N187P45, respectively, at 45 DAS, 13.7% and 9.9% under N150P75 and N187P75 at 60 DAS, and by 12.1% under N187P75 at 115 DAS (Figures 1C, D, K and L). Similarly, stem P concentration also showed a significant increase (12.9% and 11.6% under N150P60 and N187P60, respectively) with deep banding at 45 DAS (Figures 3G, H, K and L). Seed P concentration also increased by 3.4% and 4.8% under N112P45 and N150P45 (Figures 7B). These results indicate that deep banding of fertilizer, which is placed in close proximity to root systems, enhances the interaction between the fertilizer and roots, thereby optimizing P uptake efficiency [26,27]. Conversely, broadcasting exposes fertilizers to a larger soil surface area, leading to substantial losses through adsorption, leaching, and erosion [28]. This can cause P deficiency in the early growth cycle, which will have negative impact on both the crop's growth and its reproductive development [29]. Even if adequate P levels are reached later, they may not fully compensate for the initial deficiency. Deep placement, on the other hand, minimizes these losses and ensures a readily available P source within the root zone throughout the whole growth season [17,30].

The current study showed that higher P fertilization rates (P60 and P75) significantly increased leaf and stem P concentrations compared to the lower rate (P45) regardless of NP rate combination and fertilizer placement (Figures 1D, H and L; 3B, F, J, C, G and K). These results suggest that P fertilization rates at P75 or P60 are optimal for enhancing P uptake in maize plants under the given soil conditions. This aligns with established soil P recommendations for maize, where medium soil P levels (similar to those at 16.3 and 15.1 mg kg<sup>-1</sup> in 2018 and 2019 in our study, respectively) necessitate P application rates between 60 and 100 kg ha<sup>-1</sup> [28]. By meeting maize P demand throughout the growth season, these optimal P fertilization rates promote plant growth, seed yield, N and P concentration and accumulation in various maize organs.

The present findings corroborate previous studies demonstrating that higher P fertilization rates enhance maize P accumulation and biomass production. For example, Nadeem et al. [31] observed increased grain dry weight and P accumulation in maize fertilized with P92.50 kg ha<sup>-1</sup> compared to P5.60 kg P ha<sup>-1</sup>. In addition, Ahmad et al. [32] demonstrated that P fertilization at P90 kg ha<sup>-1</sup> significantly enhanced grain ear<sup>-1</sup>, 1,000-seed weight, and harvest index, seed yield and total biomass compared to P60 kg ha<sup>-1</sup>.

### 3.2. Greater Soil Total P and Olsen P Concentration under Deep band

In the present study, deep band placement resulted in significantly higher total soil phosphorus concentration compared to broadcast and side band placement across all N and P fertilization rates (N112P45, N150P45, N187P45, N112P60, N150P60, N187P60, N112P75, N150P75 and N187P75 (Figures 8B-C, F-H and J-L). This observation was mirrored by higher Olsen-P concentration at 60 DAS (Figures 9B-C, F-H and J-L).

Deep fertilizer placement offers several advantages that contribute to increased soil P concentration. Firstly, it minimizes fertilizer contact with soil particles, thereby reducing P fixation compared to broadcast application [33,34]. In calcareous soil specifically, banding P fertilizer reduces contact between fertilizer and soil, while maximizing root-fertilizer contact and P concentration compared to broadcasting, leading to less immobilization and greater P uptake [35]. Secondly, deep placement aligns fertilizer with the zone of adequate moisture in subsurface soil, which is crucial for P solubility. This promotes a significant increase in soil inorganic P fractions like dicalcium phosphate (Ca<sub>2</sub>-P), octacalcium phosphorus (Ca<sub>8</sub>-P), aluminum phosphorus (Al-P), and iron phosphorus (Fe-P) at various soil depths [18]. Conversely, P broadcasting induces P stratification near the soil surface. This increases P losses through mechanisms like soil and wind erosion, and surface runoff, ultimately reducing P availability for plant uptake. Compared to broadcasting, injecting fertilizers like monoammonium phosphate and poultry litter 1 cm below the soil surface have shown significantly reduce P loss by 98% and 84%, respectively [15].

Our findings showed that higher P fertilization rates (P60 and P75) significantly increased both total soil P and Olsen-P concentration compared to the lower rate (P45) at various N fertilization levels and fertilizer placement methods (deep band, side band and broadcast) at 60 and 115 days after sowing (DAS) (Figures 8 & 9). These results indicate that P availability in soil increases with higher P

application rates. Studies have shown a positive correlation between P fertilizer application rates and soil P availability. For instance, P100 and P200  $P_2O_5$  kg ha<sup>-1</sup> considerably increase soil available P by 100% and 200%, respectively, compared to no-P control [36]. A 21-year long-term study showed that P20, P39, P59, and P79 kg P ha<sup>-1</sup> considerably increased Olsen-P by 3.7, 5.2, 11.2 and 20.6 mg P kg<sup>-1</sup>, and total soil P concentration by 4.2%, 26.0%, 36.5% and 49.8%, respectively [37].

### 3.3. Greater P Accumulation and Yield Production under Deep Band

The present study found that deep band application significantly increased P accumulation in maize leaves, stems, and roots compared to sideband and broadcast application at all time points (45 DAS, 60 DAS, 115 DAS) and for N150P60, N187P60, N150P75 and N187P75 (Figures 2G, H, K and L; Figures 4G, H, K and L; Figures 6G, H, K and L). Deep band application also improved seed P accumulation under N150P75 and N187P75 (Figures 7H) and higher total plant P accumulation under N150P60 and N187P60 (Figures 7K). These findings can be explained by several factors. First, the broadcasting fertilizer mixes it with a larger soil volume, exposing it to a greater surface area. However, this can lead to P stratification near the soil surface, which increases the risk of immobilization or loss through runoff [28]. This, in turn, reduces the amount of P available for plant uptake and potentially P deficiency. Deep band placement, on the other hand, minimizes soil-fertilizer contact, thereby improving the availability of P for plant uptake [28,38,39, 40].

Second, P is relatively immobile in soil and relies on diffusion for uptake by plant roots. Placing fertilizer close to the roots provides them with easier access to this essential nutrient, leading to greater P use efficiency compared to broadcasting [28,41]. Third, for efficient P uptake, plants require a sufficient P supply during the early growth stages to develop a robust root system with enhanced P acquisition capabilities [42]. Studies by Hopkins (43) and Dhillon et al. (41) showed that applying the required P fertilizer near the seeds at sowing is a more efficient strategy than broadcasting or split applications. Deep band placement not only fulfills this requirement but also ensures access to moisture in deeper soil layers, which can further enhance P solubility and mobility in the root zone. This promotes root proliferation, N and P uptake, and P use efficiency [30,44,45].

Phosphorus accumulation in maize is also enhanced with higher N rates (N187 and N150) compared to a lower rate (N112) with the same P level (P60 or P75) under deep band placement. This was observed in leaves and stems at various points throughout the growing season (45 DAS, 60 DAS or 115 DAS) (Figures 2F-H and J-L; Figures 4F-H and J-L; Figures 6F-H and J-L). Root P accumulation also increased at 60 and 115 DAS (Figures 2F-H and J-L; Figures 4F-H and J-L; Figures 6F-H and J-L). Furthermore, total plant P accumulation at harvest (115 DAS) and seed P accumulation at the same P level (P75) under deep band placement were significantly greater with higher N rates (Figures 7K and L; Figures 7H). These findings suggest a synergistic effect of N fertilization on P uptake by maize. Studies have shown that N application enhances plant P availability and utilization, including stimulating the conversion of organic P forms into inorganic P forms that are more readily taken up by plants [46], reduce the amount of soil iron (Fe) and aluminum (Al) oxides, which are known to adsorb P and make it unavailable to plants [47], and improve P utilization efficiency, P translocation and P translocation efficiency [46]. A field experiment with maize in Zimbabwe demonstrated that increasing N application rates from 0, 40, 80 to 120 N kg ha<sup>-1</sup> led to higher total plant and grain P accumulation [48].

### 3.4. Greater P Use efficiency under Deep band

This present study found that deep fertilizer placement significantly increased phosphorus agronomy efficiency (PAE), partial factor productivity for the applied phosphorus (PFP<sub>P</sub>), and phosphorus use efficiency (PUE) compared to broadcast and side band fertilization (Table 1). This aligns with previous research demonstrating that placing P closer to the seed or root zone enhances PUE while reducing P application rates compared to broadcasting [49,50]. Phosphorus is a relatively immobile nutrient in soil and readily fixes to soil particles, especially when broadcast on soil surface. Deep fertilizer, however, reduces P fixation reactions due to the spatial availability, which enhances the acquisition of P and, consequently, P recovery efficiency [45,49]. Studies report that an average PUE is typically only around 5-10% under broadcast fertilization but considerably increased to 30-35% under deep placement [51].

**Table 1.** Effects of nitrogen (N) and phosphorus (P) fertilization rates and fertilizer placement on phosphorus agronomy efficiency (PAE, kg kg<sup>-1</sup>), P use efficiency (PUE, %), partial factor productivity of phosphorus (PFPP, kg kg<sup>-1</sup> P) of maize.

NP rate	Placement	PAE (kg kg <sup>-1</sup> )	PUE (%)	PFPP (kg kg <sup>-1</sup> P)
N112P4 5	Broadcast	35.4 ± 9.2 (a,x,e)	7.7 ± 1.3 (b,x,e)	76.4 ± 5.4 (a,x,e)
	Side band	34.3 ± 5.7 (a,x, e)	8.0 ± 0.6 (a,x,e)	63.9 ± 3.0 (a,y,e)
	Deep band	23.3 ± 6.0 (a,x, e)	6.2 ± 1.0 (a,x,e)	48.9 ± 2.8 (a,z,e)
N150P4 5	Broadcast	37.3 ± 4.8 (a,x,e)	8.9 ± 1.1 (a,x,e)	78.0 ± 4.4 (a,x,e)
	Side band	38.5 ± 7.5 (a,x,e)	11.3 ± 1.7 (a,x,e)	81.3 ± 3.0 (a,x,e)
	Deep band	43.5 ± 9.1 (a,x,e)	13.6 ± 1.9 (a,x,e)	84.1 ± 5.9 (a,x,e)
N187P4 5	Broadcast	39.3 ± 8.2 (a,x,e)	11.5 ± 1.7 (a,x,e)	79.8 ± 4.6 (a,x,e)
	Side band	42.3 ± 7.7 (a,x,e)	13.6 ± 1.6 (a,x,e)	84.5 ± 6.6 (a,x,e)
	Deep band	45.9 ± 7.5 (a,x,e)	15.6 ± 1.4 (a,x,e)	86.1 ± 3.2 (a,x,e)
N112P6 0	Broadcast	37.1 ± 7.2 (a,x,e)	10.0 ± 1.4 (ab,x,f)	80.1 ± 3.0 (a,x,e)
	Side band	34.4 ± 7.8 (a,x,e)	9.0 ± 1.4 (a,x,e)	65.6 ± 3.4 (a,y,e)
	Deep band	25.9 ± 6.0 (a,x,f)	7.8 ± 1.1 (a,x,f)	51.0 ± 3.6 (a,z,f)
N150P6 0	Broadcast	36.2 ± 4.4 (a,x,e)	9.1 ± 0.9 (a,y,ef)	65.5 ± 3.9 (b,y,e)
	Side band	38.3 ± 4.4 (a,x,e)	10.7 ± 1.0 (a,y,e)	69.0 ± 4.2 (b,xy,e)
	Deep band	49.5 ± 4.5 (a,x,ef)	15.2 ± 1.2 (a,x,e)	77.6 ± 3.2 (a,x,e)
N187P6 0	Broadcast	37.3 ± 3.5 (a,y,e)	10.3 ± 0.5 (a,y,e)	66.5 ± 3.2 (b,y,e)
	Side band	40.5 ± 5.4 (a,xy,e)	12.5 ± 1.0 (a,y,e)	70.9 ± 4.5 (a,xy,e)
	Deep band	51.5 ± 3.5 (a,x,e)	17.0 ± 0.9 (a,x,e)	79.2 ± 3.4 (a,x,e)
N112P7 5	Broadcast	41.7 ± 8.0 (a,x,e)	11.7 ± 1.2 (a,x,e)	82.5 ± 4.4 (a,x,e)
	Side band	36.9 ± 5.6 (a,x,e)	10.2 ± 1.2 (a,x,f)	66.7 ± 3.1 (a,y,e)
	Deep band	31.4 ± 6.6 (a,x,e)	9.1 ± 0.8 (a,x,f)	53.5 ± 3.2 (a,z,f)
N150P7 5	Broadcast	25.2 ± 6.0 (a,y,e)	7.6 ± 0.9 (a,z,e)	50.5 ± 3.5 (c,y,e)
	Side band	27.4 ± 2.5 (a,y,e)	9.5 ± 0.4 (a,y,ef)	52.3 ± 3.2 (c,y,e)
	Deep band	44.0 ± 3.0 (a,x,e)	14.5 ± 0.5 (a,x,e)	64.4 ± 3.2 (b,x,e)
N187P7 5	Broadcast	25.9 ± 4.1 (a,y,e)	8.5 ± 1.0 (a,z,e)	51.1 ± 2.8 (c,y,e)
	Side band	31.0 ± 2.2 (a,y,e)	10.8 ± 0.4 (a,y,e)	55.4 ± 3.5 (b,y,e)
	Deep band	46.3 ± 4.6 (a,x,e)	15.9 ± 0.4 (a,x,e)	66.4 ± 3.9 (b,x,e)

### 3.5. Variations in Relationships between Concentrations of Tissue P, Soil P, and Plant P Accumulations

In the present study, positive relationships were observed between P concentration and accumulation in leaves, stems, seeds, roots, and total plant biomass at 60 DAS and 115 DAS (Figure 10A-C, and Figure 11A-D). Similarly, P concentrations and accumulations in these plant parts also showed positive relationship with Olsen-P concentration at 60 DAS and 115 DAS (Figure 10D-F, 10G-I, 11E-I, and 11J-N). These results are consistent with previous studies that have documented positive relationships between plant organ P content and soil available P. For instance, [52] reported strong correlations between soil P and P concentration in leaves, stems, roots, and fruits of orange trees. They further suggested that fertilizer application strategies could be optimized based on relationships between soil and plant P concentrations, rather than relying solely on soil or plant P levels. Furthermore, Liu et al. [23] observed that increased P fertilization led to a higher proportion of inorganic P fractions (Ca2-P, Ca8-P, Al-P, and Fe-P) in winter wheat, which positively correlated with seed yield (ranging from 67% to 86%) and aboveground biomass (21% to 41%). Fan et al. [47] also observed significant correlation between various soil P fractions (available P, labile P, moderately labile P) and root biomass/length density in a subtropical forest ecosystem. The strongest correlations were found between available P and both root biomass (42%) and root length density (55%).

Maize takes up most P in its early growth between 3 and 6 weeks. This uptake generally happens alongside the accumulation of dry matter, though P uptake tends to be slightly ahead during this early stage but declines at later growth stages [53]. This pattern highlights that a positive correlation between seed yield or biomass production and P accumulation in plant organs indicates efficiently internal P utilization by wheat and lupin [54]. Ma et al. (55) further demonstrated a strong correlation between root P concentration and root biomass (84%) and leaf P concentration and leaf biomass (91%).

## 4. Materials and Methods

### 4.1. Study Site and Plant Materials

This 2-year pot study was conducted over two growing seasons from May 29 to September 15, 2018, and May 15 to August 28, 2019, in a glass greenhouse locating at the National Monitoring Base for Purple Soil Fertility and Fertilizer Efficiency (NMBPSFFE), Southwest University (29°48'N, 106°24'E, 266.3 m asl), Chongqing, China. The region experiences a humid subtropical climate, with mean summer temperature of 2018 and 2019 (26.0 and 25.0 °C) and winter temperatures (9.1 °C and 10.8 °C), and mean rainfall of 2018 (143.2 mm) and 2019 (157.1 mm), respectively (see Table at Sharifi et al. 2023 <https://www.mdpi.com/search?authors=Sharifullah+Sharifi&journal=plants>).

Pots at 57 cm × 23 cm × 27 cm (length × width × height) were filled with 26 kg purple soil that has developed from gray-brown-purple sand shale parent materials. This soil is classified as Eutric Regosol according to the FAO Soil Classification System. Soil samples at 0–20 cm depth were collected at the aforementioned NMBPSFFE. The soil was characterized by a texture of sand:silt:clay (14%:48%:38%), 1.37 g cm<sup>-3</sup> bulk density, pH (1:2.5; H<sub>2</sub>O) of 6.9, 7.40 g soil organic carbon kg<sup>-1</sup>, 0.70 g total N kg<sup>-1</sup>, 81.90 mg available N kg<sup>-1</sup>, 0.42 g total P kg<sup>-1</sup>, 15.70 mg available P kg<sup>-1</sup>, 10.16 g total K kg<sup>-1</sup> and 176 mg available K kg<sup>-1</sup>.

Two maize hybrid cultivars of Xida-789 and Xida-211 were respectively planted in 2018 and 2019. Maize seeds were surface sterilized using 10% H<sub>2</sub>O<sub>2</sub> for 20 min, followed by thorough rinsing with sterile water, and then pre-germinated on sterilized moist paper at 20/25 °C (night/day) for 1.5 days. Each pot was initially sown with five seeds, but after ten days of germination, only two healthy seedlings were retained per pot. Plants were daily observed throughout the growing season, and soil moisture was maintained at 70% field water-holding capacity. Weeding was carried out at 15 and 45 days after sowing (DAS). The Philippine downy mildew, which typically occurs during the maize's tasseling stage as identified by pale yellow to whitish discolorations, was controlled with a diluted 1/1000 solution of 25% Metalaxyl.

### 4.2. Experimental Design and Treatments

A randomized split-plot design was employed to the experimentation. The main factor was the fertilizer placement method: broadcasting, side banding, or deep banding. The sub-factor was a combination of N and P fertilizer rates: 112, 150, 187 kg N ha<sup>-1</sup> and 45, 60, 75 kg P ha<sup>-1</sup>, resulting in a total of nine combinations of NP fertilization treatments: N112P45, N112P60, N112P75, N150P45, N150P60, N150P75, N187P45, N187P60, and N187P75; while no-NP was the control treatment. Previous study suggests that NH<sub>4</sub><sup>+</sup> or PO<sub>4</sub><sup>3-</sup> are ideal forms of N and P, respectively, for fertilizer placement studies [56]. Therefore, the commercial urea (46% N) and calcium superphosphate (P<sub>2</sub>O<sub>5</sub> ≥ 12%) were utilized as basal fertilizers in a single application prior to seeding, in addition to potassium chloride (40 kg ha<sup>-1</sup>, 52% K) to ensure a balanced NPK supply. Each fertilization treatment was replicated thrice for a total of 30 pots or replicates (see figure at Sharifi et al. 2023, <https://www.mdpi.com/search?authors=Sharifullah+Sharifi&journal=plants>).

### 4.3. Harvesting and Phosphorus Variables Determination

Aboveground leaves and stems at three stages were harvested in the eight-leaf stage (V8 at 45 DAS), tasseling stage (VT at 60 DAS), and physiological maturity or harvest stage (R6 at 115 DAS). Roots were collected at VT and R6 and seeds were also harvested at R6. Harvested plant tissues were washed carefully with tap water, and then oven dried at 60 °C to a constant weight.

Plant samples were ground to powder, passing through 1 mm sieve, and then digested with 98% sulfuric acid and 30% hydrogen peroxide. After digestion tissue P concentrations were measured using the vanadium molybdate yellow colorimetric method [57].

The concentration of total P and Olsen-P in the soil were measured at 60 DAS and 115 DAS. To determine total soil P, 0.20 g of air-dried soil samples were weighted into a 30 mL nickel crucible and were heated in an electric furnace at 700-720 °C for 15 minutes after the addition of 2.0 g sodium hydroxide. After cooling, the digested mixture was dissolved with deionized water, and the total volume was brought up to 100 mL. Total soil P concentration was determined using the colorimetric molybdate and ascorbic acid method [58], with the absorbance measured at 660 nm. Soil Olsen-P was determined using the molybdovanado phosphatase method based on the extraction with 0.5 M

NaHCO<sub>3</sub> [59]. Plant tissue P accumulation was calculated by multiplying P concentrations in leaves, stems, seeds, and roots with their respective biomass. Total plant P accumulation was calculated as the sum of P accumulation in leaves, stems, seeds, and roots.

$$\text{Agronomy P use efficiency} = \frac{\text{seed yield at P treatment} - \text{seed yield at zero P treatment}}{\text{applied P at P treatment}} \quad (1)$$

$$\text{Phosphorus use efficiency (\%)} = \frac{(U_P - U_0)}{A_P} \times 100 \quad (2)$$

where,  $U_P$  or  $U_0$  is total P accumulation (leaf + stem + seed + root) under a specific P or no-P fertilization (control), and  $A_P$  is the total applied P from the specific P fertilization.

Partial factor productivity of P fertilizer (PFP<sub>P</sub>):

$$\text{PFP}_P (\text{kg grain kg}^{-1} \text{ P}) = \frac{Y_P}{A_P} \quad (2)$$

where,  $Y_P$  is the grain yield and  $A_P$  is the total P accumulation under a specific P fertilization.

#### 4.6. Statistical Analyses

Data (means  $\pm$  SE,  $n = 6$ ) were subjected to analysis of variance (ANOVA) using SPSS version 24.0 (SPSS Inc., Chicago, IL, USA). Data comparisons among treatments were examined by Duncan's multiple range test at  $P < 0.05$ . Correlations between fertilizer placement methods and fertilization rates were conducted using OriginPro2023b version 10.05 (Origin Lab Corp., Northampton, MA, USA).

## 5. Conclusions

A two-year greenhouse pot experiment was conducted to investigate the effects of deep band fertilizer placement on phosphorus (P) uptake and utilization by maize plants compared to broadcast and side band placement methods. The findings demonstrate that deep band placement significantly enhances P uptake efficiency, leading to greater P concentration and accumulation in maize leaves, stems, roots, and seeds compared to the other two methods. Deep placement offers several advantages that contribute to this improved P uptake. Firstly, it minimizes fertilizer contact with soil particles, reducing P fixation reactions and making P more available to plants. Secondly, placing fertilizer near the developing root system facilitates easier access to P for the roots. Thirdly, deep placement aligns fertilizer with zones of adequate moisture content in the subsurface soil, which is crucial for P solubility and promotes root proliferation. Higher N application rates (N187 and N150), when combined with deep band placement, further promote the mechanisms that enhance P availability and utilization by the plant. Deep band placement also led to significantly increased total soil P concentration, Olsen-P concentration, and P use efficiency compared to other placement methods. This suggests that deep band placement is a more efficient strategy for P fertilization in maize. It may allow for reduced P application rates while achieving similar or improved growth and yield outcomes. The study found positive relationships between P concentration and accumulation in various plant organs and soil Olsen-P concentration. This highlights the importance of maintaining adequate soil P levels for optimal plant growth. In conclusion, this study suggests that deep band fertilizer placement is a superior strategy for enhancing P uptake efficiency, utilization, and overall productivity in maize compared to broadcast and side band placement methods. This approach has the potential to optimize P fertilizer use and improve maize production while minimizing potential environmental P losses associated with broadcast application.

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