

Evaluation of CIMMYT Maize Inbred Lines for Low-Phosphorus Tolerance under Field and Hydroponic Conditions in the North Eastern Hills of Meghalaya India

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Abstract

Phosphorus is a critical nutrient for plant growth, and its limited availability in acidic soils represents a major abiotic stress threatening agricultural sustainability. Developing cultivars tolerant to low-phosphorus conditions is an environmentally friendly strategy, as excessive phosphorus fertilizer use can be detrimental to the environment. This study employed both field and hydroponic screening to identify maize inbred lines with contrasting tolerance to low-phosphorus conditions, aiming to develop high-yielding, phosphorus-efficient cultivars. Hydroponic screening using modified Hoagland's solution (low phosphorus: $2.5 \times 10^{-6} \text{ mol L}^{-1} \text{ KH}_2\text{PO}_4$; normal phosphorus: $2.5 \times 10^{-4} \text{ mol L}^{-1} \text{ KH}_2\text{PO}_4$) was conducted to assess traits such as shoot and root length, fresh and dry weight, and root-to-shoot ratio. A significant increase in root fresh weight, despite no notable change in primary root length, suggests that the tolerant inbreds may adopt a strategy of lateral root proliferation rather than elongation of the primary root. Based on the low-phosphorus tolerance index (LPTI) and low-phosphorus performance index (LPPI), genotypes were classified into four categories. Genotypes VL181019, VL20213, VL175994, and VL236956 were identified as tolerant and good-performing, whereas VZL22777, VL1885, and VL109138 were categorized as sensitive and poor-performing. The overlap of tolerant and sensitive lines across field and hydroponic conditions emphasises the reliability of early-stage hydroponic screening as a cost-effective method for genotype selection. The identified inbred lines represent valuable genetic resources for future breeding programs aimed at developing high-yielding, low-phosphorus-tolerant maize cultivars, thereby contributing to sustainable agriculture and long-term food security.

Highlights

- Selection of tolerant and sensitive parental lines based overlapping performance in field and hydroponics
- Correlation of root biomass with low phosphorus tolerance instead of significant increase in root length
- Grouping of genotypes based on low phosphorus tolerance and performance index
- Integration of field and hydroponics for accurate selection of genotypes

Introduction

Agriculture is one of the most affected industries by global warming (Farah et al. 2025). Extreme weather events and temperature changes can directly and indirectly affect crop production in the form of biotic and abiotic stresses (Pandey et al. 2012; Tabinda et al. 2025; Zhang 2023). The study of mechanisms related to crop tolerance and susceptibility under specific stress conditions is becoming essential for achieving sustainable agriculture (Daszkiewicz 2022; Kumari et al. 2022). Availability of low phosphorus in the soil is one of the major abiotic factors that affect plant growth status, particularly by reducing crop yield (Muhammad Aslam et al. 2020; He et al. 2014).

Low available phosphorus content is one of the major problems in acidic soils, due to high iron (Fe), aluminium (Al), and manganese (Mn), which fix the phosphorus in the form of chelates (Regasa et al. 2025; Sharma et al. 2025). The soil mechanisms that make the phosphorus unavailable to crops are (i) adsorption and fixation in acidic soil (low pH), (ii) precipitation reactions such as at low soil pH, P reacts with Fe^{3+} and Al^{3+} to form insoluble Fe-P & Al-P and at high soil pH, P precipitates with Ca^{2+} to form calcium phosphates (Ca-P) (iii) P gets occluded with oxides of iron and aluminium, making it unavailable to plants (Adnan et al. 2025; Singh et al. 2021; Zhang et al. 2025a). The greater amounts of P are fixed in clay-rich soils, mostly due to the higher amounts of Fe and Al oxides, dominated by kaolinite clays (Srinivasarao et al. 2015).

Maize (*Zea mays* L.) is considered a major global commodity that plays a key and increasing role in global agri-food systems, which include consumption as direct food and indirect feed pathways for animal-sourced foods (Erenstein et al. 2022). It is a heavy nutrient-demanding cereal crop with a relatively high phosphorus requirement to support optimal growth and productivity (Bhagat et al. 2025; Ismail et al. 2025). Early-season P nonavailability reduced total grain yield and affected kernel numbers. As half of the total P uptake occurs after maize flowering, short P supply during grain fill significantly affects yield (Barry and Miller 1989; Sun et al. 2023; Wang et al. 2019).

For the effective intensification of agricultural systems, optimising the use of phosphorus (P) fertilisers is a key aspect (Gong et al. 2025; Ros et al. 2020; Sica et al. 2025). The use of phosphorus-based fertilisers in phosphorus-deficient soils is one of the most effective strategies to maximise yield and food quality, thereby contributing to the fight against hunger (Bindraban et al. 2020). It is expected that the worldwide phosphorus market will increase to 81 million metric tons in 2027 from 68.5 million metric tons in 2020, with a compound annual growth rate (CAGR) of 2.5% (Ahmad et al. 2023).

Phosphate-based fertiliser industries are a major contributor to environmental pollution; the excessive use of phosphorus fertilisers results in soil P accumulation, which accelerates eutrophication and disturbs aquatic ecosystems (Liang et al. 2013; Ahmad et al. 2023). Phosphate fertilisers are primarily derived from phosphorite and apatite rocks, which often contain toxic elements like cadmium (Cd), arsenic (As), and lead (Pb) (de Boer et al. 2019; Lottermoser 2010; Lian-kai et al. 2025). Cadmium, in particular, can be absorbed by plants and enter the food chain, posing significant health risks. Its accumulation in the human body has been linked to kidney and liver dysfunction, as well as bone demineralisation (Jiao et al. 2012; Suciú et al. 2022). Therefore, adopting alternative strategies to phosphate fertilisers is imperative in the present context.

However, securing maize as a resilient future crop requires the synergistic integration of innovative agronomic practices and targeted genetic improvements to enhance tolerance against multiple abiotic and biotic stresses (Li et al. 2025; Sidahmed et al. 2025; Singh et al. 2023). Veneklaas et al. (2012) emphasised the role of genetics and molecular biology in improving phosphorus use efficiency (PUE) in crops and noted that such improvements can mitigate the environmental impacts of P fertilisers and waste while enhancing the nutritional value of food grains. Strategic collaboration between physiologists

and plant breeders is pivotal for advancing fundamental research on phosphorus use efficiency (PUE), ultimately enabling the development of crop varieties with superior performance in phosphorus-deficient soils (Khare et al. 2025).

Therefore, a study was conducted to screen inbred lines collected from the International Maize and Wheat Improvement Centre (CIMMYT) and ICRISAT Campus, Patancheru, Hyderabad, India, under both field and hydroponic conditions for low-phosphorus tolerance. The two major objectives of the study were first, to select genotypes with contrasting performance under both field and hydroponic conditions, as heterosis breeding is the most successful method in maize breeding programs and depends on the genetic divergence between the parental inbred lines (Melchinger and Gumber 1998) and second, to minimize the gap between field and hydroponic screening.

Materials and Methodology

Plant Material and Experimental Site

A set of 20 homozygous maize inbred lines were obtained from the International Maize and Wheat Improvement Center (CIMMYT), ICRISAT Campus, Patancheru, Hyderabad, India. The screening of these inbred lines was conducted at the experimental field of the All India Coordinated Research Project on Maize (AICRP-Maize), College of Agriculture, Central Agriculture University (Imphal), Kyrdemkulai, Meghalaya. The site is located at latitude of 25.74°N, longitude of 91.81°E, and an elevation of 700 meters above mean sea level.

The analysis of the soil samples revealed that the soil was moderately acidic, with a pH of 4.9. The nutrient analysis revealed that the available nitrogen (N) was 250 kg/ha, phosphorus (P) was 11.2 kg/ha, and potassium (K) was 180 kg/ha. According to the Bray method, the available phosphorus content was categorised as low. All other essential nutrients were supplemented following standard agronomic practices, except for phosphorus.

Experimental design and morphological characteristics

An alpha lattice design with two replications was used for screening maize inbred lines. Data were recorded for plant height, days to 50% anthesis, days to 50% silking, number of kernels per row, number of rows per ear, hundred grain weight, and grain weight per plant.

Twenty maize inbred lines showing contrasting performance for grain weight per plant were selected for further screening under hydroponic conditions. The field results were compared with hydroponic screening to improve the accuracy of selecting tolerant and sensitive maize inbred lines.

Hydroponic Screening

The experiment was conducted using a completely randomised design (CRD), comprising two treatments with three replications per treatment. A modified Hoagland's nutrient solution was employed for hydroponic screening. The composition of the working solution (per liter) was as follows: 2.0 mmol $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, 0.75 mmol K_2SO_4 , 1.5 mmol MgSO_4 , 0.1 mmol KCl , 0.1 μmol H_3BO_3 , 0.1 μmol MnSO_4 , 0.01 μmol $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, 0.1 μmol $\text{ZnSO}_4 \cdot 5\text{H}_2\text{O}$, 0.0005 μmol $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24} \cdot 4\text{H}_2\text{O}$, and 0.1743 mmol Fe-EDTA. Two phosphorus treatments were applied: low phosphorus (LP) at $2.5 \times 10^{-5} \text{ mol L}^{-1}$ KH_2PO_4 , and normal phosphorus (NP) at $2.5 \times 10^{-4} \text{ mol L}^{-1}$ KH_2PO_4 . A total of 20 maize inbred lines were screened under these conditions, following the methodology described by Azam et al. (2022).

Traits recorded in hydroponics

During the hydroponic screening, observations were recorded for shoot length (SL), root length (RL), shoot fresh weight (SW), and root fresh weight (RFW). Following fresh weight measurements, shoot and root samples were dried in an air-forced oven at 70 °C for four days to determine shoot dry weight (SDW) and root dry weight (RDW). These values were subsequently used to calculate the shoot-to-root ratio.

Low-phosphorus tolerance index (LPTI) and low-phosphorus performance index (LPPI)

The low-P tolerance and performance under P stress of the maize inbred lines were calculated according to the methodology described by Schuster et al. (2024). The mean value of each trait under low phosphorus (LP) conditions was divided by the corresponding mean under normal phosphorus (NP) conditions to obtain the relative trait values. These relative values were then subjected to principal component analysis (PCA). The principal components (PCs) with eigenvalues equal to or greater than one were retained and subsequently used to calculate the LPTI as follows:

$$\text{LPTI} = \sum_{i=1}^n PC_i \times CR_i$$

Where n represents the number of principal components with eigenvalues greater than one, and CR denotes the contribution rate of each principal component to the total variation in the relative trait values. The LPPI was estimated using the standardized trait values under LP conditions. The mean values under LP conditions were subjected to principal component analysis, and PCs with eigenvalues greater than one were retained. These PCs were then used to calculate the LPPI as follows:

$$\text{LPPI} = \sum_{i=1}^n PC_i \times CR_i$$

Based on these two indices (LPTI and LPPI), the maize inbred lines were classified into four groups: tolerant and good-performance group (TG), sensitive and good-performance group (SG), tolerant and poor-performance group (TP), and sensitive and poor-performance group (SP).

Statistical Analysis

Statistical analyses, including analysis of variance (ANOVA), Pearson correlation coefficient, principal component analysis (PCA), and the calculation of LPPI and LPTI indices, were performed using RStudio (version 4.5). The graphical representations were prepared using Microsoft Excel 2021.

Results and Discussions

Field Screening

Field screening based on their grain weight per plant showed a significant difference among the genotypes under acidic soil conditions with low available phosphorus, indicating substantial variability within the plant material for further evaluation and breeding (Table 1). Of the twenty inbred lines, ten exhibited poor performance, while the remaining ten showed good performance under field conditions (Table 1). These contrasting genotypes were subsequently subjected to hydroponic screening for more accuracy and confirmation of their responses to low phosphorous conditions.

Table 1
Mean Field data in alpha lattice design

Genotype	PH	DT	DS	HGW	LA	NKPR	NRPE	GWPP
SNL2078	78.60	66.50	69.00	14.50	339.59	14.40	9.00	10.75
VL109138	111.95	64.00	67.50	12.50	283.78	13.80	11.80	12.10
VL143898	86.18	66.87	70.95	18.30	269.70	10.80	9.22	9.94
VL175994	116.45	66.00	68.50	15.0	535.06	22.60	13.00	26.36
VL181019	109.38	62.87	66.95	19.80	444.22	22.50	12.22	27.60
VL18262-1	106.73	63.13	65.55	9.70	282.67	14.60	9.98	8.38
VL18310	114.95	64.00	67.50	14.50	498.60	24.50	13.40	27.58
VL1885	83.85	62.00	65.00	13.00	210.30	15.20	11.40	11.93
VL18981	79.43	65.13	67.55	14.20	258.79	9.40	9.98	8.00
VL18994	103.65	67.50	70.50	19.00	483.91	26.20	14.40	27.58
VL19957	101.48	64.63	67.55	18.20	443.03	21.20	13.38	29.22
VL20213	113.45	66.00	69.00	14.50	441.86	23.30	13.80	26.53
VL21948	78.80	62.50	66.50	13.50	244.28	9.80	14.00	10.62
VL22347	103.10	62.00	65.00	24.00	448.51	21.20	14.00	26.69
VL234534	98.45	62.50	65.50	13.50	231.30	12.80	9.60	8.98
VL236956	110.45	64.50	67.50	16.50	440.83	21.30	12.40	26.14
VL236989	111.03	63.13	66.05	18.20	438.38	20.00	13.18	28.56
ZL19359	129.78	63.87	65.95	17.80	430.82	23.00	13.22	31.11
ZL2119	107.60	68.50	71.50	13.50	229.64	10.50	9.60	9.07
ZL22777	96.30	62.50	65.00	13.50	345.01	13.10	11.51	12.09
PH = Plant height; DT= Days to 50% tasseling; DS= Days to 50% silking; LA= Leaf area; NRPE=Number of rows per ear; NKPR= Number of kernels per row; HGW= Hundred grains weight; GWPP= Grain weight per plant								

Hydroponic screening

Analysis of variance (ANOVA)

In the hydroponic screening, the ANOVA revealed significant variation among the maize genotypes, indicating the presence of genetic diversity for shoot length (SL), root length (RL), shoot fresh weight (SW), root fresh weight (RW), shoot dry weight (SDW), root dry weight (RDW), total dry matter (TDM), and

root-to-shoot ratio (RSR) in Normal Phosphorus (NP) and (Low Phosphorus (LP) (Table 2; Fig. 1a-h & Figs. 4 & 5). Across the phosphorus treatments, significant differences were observed for RW, SDW, and TDM (Fig. 1d, e& g and Figs. 4 & 5). Moreover, the genotype × phosphorus (G × P) interaction was significant for all traits except SL, suggesting that the majority of traits were differentially influenced by phosphorus availability (Table 2).

Table 2
Analysis of variance for Hydroponics

Source of variation	Phosphorus	Genotypes	Genotype x Phosphorus	Error
df	1	19	19	80
SL	0.049	25.376**	2.326	2.16
RL	21.336	97.502**	26.326*	12.392
SW	0.063	1.774**	0.49**	0.064
RW	0.148*	0.203**	0.052**	0.023
SDW	0.0025**	0.0053**	0.0009**	0.0003
RDW	3.3×10^{-6}	0.0017**	0.0004**	0.0002
TDM	0.0024*	0.0086**	0.0016**	0.0004
RSR	0.0508	1.1504**	0.1997**	0.0858
**, *= Level of Significance at 1%, and 5% respectively; SL = Shoot length; RL= Root length; SW= Shoot fresh weight; RW= Root fresh weight; SDW= Shoot dry weight; RDW= Root dry weight;TDM= Total dry matter; RSR= Root shoot ratio				

Under low-phosphorus conditions, RDW was significantly higher compared to normal phosphorus conditions, indicating a tendency of the genotypes to allocate more biomass to the root system in response to phosphorus stress (Table 3& Fig. 1f and Figs. 4 & 5). A similar trend was observed for TDM (Fig. 1g), reflecting an adaptive response of the genotypes under limited phosphorus supply. In contrast, SDW was generally higher under NP conditions, with most genotypes exhibiting reduced SDW under phosphorus-deficient conditions (Fig. 1e), exhibiting the sensitivity of shoot growth to phosphorus stress (Table 3).

Table 3
Descriptive statistics for hydroponics

Traits	Mean	Minimum	Maximum	Standard Deviation	Standard Error
SL	9.99	4.00	16.30	2.60	0.34
RL	14.3	3.2	27.2	0.75	0.75
SW	1.52	0.02	3.1	0.64	0.08
RW	0.61	0.09	1.1	0.26	0.03
SDW	0.086	0.008	0.18	0.037	0.005
RDW	0.052	0.012	0.1	0.02	0.0026
TDW	0.138	0.025	0.23	0.047	0.0061
RSR	0.78	0.133	2.75	0.57	0.073
SL = Shoot length; RL= Root length; SW= Shoot fresh weight; RW= Root fresh weight; SDW= Shoot dry weight; RDW= Root dry weight; TDM= Total dry matter; RSR= Root shoot ratio					

Low phosphorus tolerance and performance index (LPTI and LPPI)

Among the screened maize inbred lines, VL18262-1 exhibited the highest LPTI, despite showing a negative value for the LPPI (Table 4). This indicates that while the genotype demonstrated strong tolerance to P stress, its overall performance under LP conditions was relatively poor. In contrast, VL236956 ranked second in LPTI but recorded the highest LPPI, suggesting that it not only tolerated phosphorus deficiency effectively but also maintained superior growth under stress conditions (Table 4). Similarly, genotypes VL20213, VL181019, VL18981, VL175994, and VL234534 demonstrated consistent performance, showing both strong tolerance and favourable growth under LP conditions (Table 4). In contrast, VL18994 showed the weakest response, recording the lowest values for both LPTI and LPPI, identifying it as the most sensitive genotype to phosphorus deficiency among those evaluated in this study (Table 4).

Table 4
LPPI and LPTI index with respective genotypes

Genotype	LPTI	LPPI
VL18262-1	2.051322	-0.24055
VL236956	1.404216	1.917272
VL18310	1.057136	-0.6169
VL20213	1.002988	1.485483
VL22347	0.934808	-0.73544
VL143898	0.619905	-0.0385
VL181019	0.593761	1.2348
VL18981	0.262888	1.144019
VL21948	0.099762	-1.06432
VL175994	0.04827	1.414722
VL234534	0.013728	1.698711
SNL2078	-0.03205	1.359226
VL236989	-0.11387	1.01029
ZL2119	-0.11438	0.543828
VL1885	-0.51958	-0.60478
VL19957	-0.74712	-0.28909
ZL193359	-0.88626	-0.73916
VL109138	-1.30092	-0.34722
ZL22777	-1.52419	-3.44402
VL18994	-2.85041	-3.68838
LPTI = Low Phosphorus Tolerance Index; LPPI = Low Phosphorus Performance Index		

Correlation analysis for hydroponic screening

Correlation analysis revealed that, under LP conditions, most traits exhibited stronger associations with each other compared to those under NP conditions (Fig. 2). For instance, shoot fresh weight (SFW) showed a strong positive correlation with root length (RL; $r = 0.80$) under LP conditions, whereas this relationship was only moderate ($r = 0.60$) under normal phosphorus availability (Fig. 2). A similar trend was observed for total dry matter (TDM), which displayed a strong correlation with root dry weight (RDW)

under LP conditions, but only a moderate correlation under normal conditions (Fig. 2). These findings suggest that P stress may enhance the functional integration between root and shoot traits.

Root length emerged as a highly influential trait under phosphorus deficiency, demonstrating strong positive correlations with SFD, SDW, and TDM (Fig. 2). In contrast, these relationships were only moderate under normal phosphorus availability. Additionally, RFW and RDW showed moderate associations with root length under low-phosphorus conditions (Fig. 2), further emphasising their central role in the plant's adaptive response to phosphorus stress.

SDW consistently showed a very strong positive correlation with TDM under both phosphorus regimes (Fig. 2). In contrast, RDW showed a strong correlation with TDM under LP conditions ($r = 0.77$) but only a moderate correlation under NP availability ($r = 0.54$) (Fig. 2). The RSR, however, showed a negative association with most growth traits across both conditions (Fig. 2), indicating a trade-off between biomass allocation and overall growth traits.

Furthermore, a strong positive correlation ($r = 0.62$) was recorded between the LPTI and the LPPI (Fig. 2). This suggests that genotypes with superior performance under phosphorus stress also tend to exhibit higher tolerance, thereby reinforcing the reliability of these indices as effective selection criteria.

Classification of genotypes based on the LPTI and LPPI indices

Based on the combined evaluation of LPTI and LPPI indices, clear distinctions were observed among the maize inbred lines. Genotypes VL143898, VL175994, VL181019, VL18981, VL20213, and VL236956 emerged as the most promising candidates, exhibiting both high tolerance and strong performance under LP conditions (Table 5&Fig. 3). Their superior values for both LPTI and LPPI suggest that these lines possess inherent adaptive mechanisms enabling them to withstand phosphorus deficiency while maintaining satisfactory growth and performance. As such, these lines represent valuable parental materials for breeding programs aimed at developing phosphorus-efficient maize varieties.

Table 5
Classification of genotypes based on LPPI and LPTI

Group	Meaning	LPTI	LPPI	Name Genotypes
TG	Tolerant + Good Performer	High	High	VL143898, VL175994, VL181019, VL18981, VL20213, VL236956
SG	Sensitive + Good Performer	Low	High	SNL2078, VL234534, VL236989, ZL2119
TP	Tolerant + Poor Performer	High	Low	VL22347, VL21948, VL18310, VL18262-1
SP	Sensitive + Poor Performer	Low	Low	VL109138, VL1885, VL18994, VL19957, ZL193359, ZL22777
LPTI = Low Phosphorus Tolerance Index; LPPI = Low Phosphorus Performance Index				

In contrast, genotypes VL109138, VL1885, VL18994, VL19957, ZL193359, and ZL22777 were identified as highly sensitive, consistently exhibiting low values for both LPTI and LPPI (Table 5& Fig. 3). These lines showed poor adaptability to phosphorus stress, reflecting limited capacity for efficient phosphorus uptake and utilisation. Although not suitable for direct cultivation in P-deficient soils, these genotypes may still hold value in genetic studies aimed at identifying traits and loci associated with phosphorus sensitivity.

A third group of genotypes, including VL22347, VL21948, VL18310, and VL18262-1, displayed high LPTI values but relatively low LPPI values (Table 5& Fig. 3). This pattern indicates that while these lines tolerate phosphorus deficiency reasonably well, they do not effectively convert this tolerance into improved performance. These genotypes may possess adaptive root traits, such as enhanced root biomass or altered root architecture, that confer stress tolerance; however, their shoot growth and overall productivity remain limited. With targeted breeding and selection, these genotypes could serve as valuable sources of tolerance traits for incorporation into high-yielding backgrounds.

Interestingly, SNL2078, VL234534, VL236989, and ZL2119 exhibited the opposite trend, characterised by high LPPI but low LPTI (Table 5& Fig. 3). These genotypes were categorised as phosphorus-sensitive yet good performers, as they maintained satisfactory productivity despite their relatively poor tolerance to P deficiency. Such performance may be attributed to efficient allocation of assimilates to above-ground biomass or compensatory physiological mechanisms that enable sustained growth under sub-optimal nutrient availability. These lines represent a unique category that could be exploited for identifying traits associated with phosphorus use efficiency, even within sensitive genetic backgrounds.

Overall, the classification of genotypes into these distinct groups features the complexity of plant responses to nutrient stress. While some genotypes exhibit both strong tolerance and high performance, others excel only in one dimension. For breeding programs, the most desirable candidates are those with both high LPTI and LPPI, as they represent an optimal balance between stress resilience and productivity. However, genotypes showing contrasting responses may also harbour valuable adaptive

traits, serving as potential donors for the development of resilient and high-yielding maize varieties adapted to phosphorus-deficient soils.

Discussion

In the present study, a modified Hoagland's solution was used for hydroponic screening under two contrasting phosphorus regimes: NP ($\text{KH}_2\text{PO}_4 \approx 0.25 \text{ mmol L}^{-1}$) and LP ($\text{KH}_2\text{PO}_4 \approx 0.0025 \text{ mmol L}^{-1}$). These treatment levels are consistent with those used in previous studies by Azam et al. (2022), Islam et al. (2024) and Uddin et al. (2023) for screening maize inbred lines, thereby ensuring comparability of results across experiments. Hydroponics offers a well-controlled environment that allows precise management of nutrient stress, making it a reliable method for identifying genetic variability in response to phosphorus deficiency.

The ANOVA revealed significant differences among genotypes for all measured traits, emphasising substantial genetic diversity for phosphorus stress tolerance. Notably, RFW, SDW, and TDM showed marked variation across P treatments. Furthermore, the genotype $\times$ phosphorus interaction was significant for nearly all traits except shoot length. These results suggest that most traits were strongly influenced by P availability, while shoot elongation remained relatively stable across treatments. This variability indicates that maize inbred lines employ diverse adaptive strategies under nutrient stress, which can be exploited in breeding programs aimed at improving P deficiency.

Azam et al. (2022) and Postma et al. (2014), Li et al. (2017), Jia et al. (2018) reported significant differences for treatments, genotypes, and their interactions, whereas our findings differ, potentially due to the smaller sample size of inbred lines used in this study. The non-significant effect on shoot length suggests that plants maintain shoot elongation under both LP and NP conditions, possibly employing effective strategies to mitigate phosphorus deficiency. Additionally, the lack of significant differences in root length, coupled with stable shoot length and biomass across P treatments, adds an intriguing dimension to the study, indicating a complex adaptive response to P stress.

Previous studies have stressed that plants often respond to P stress by increasing lateral root branching rather than elongating the primary root (Postma et al. 2014; Li et al. 2017; Jia et al. 2018). The present findings are consistent with this observation, as RFW showed significant variation, while root length remained unchanged. This pattern indicates that genotypes primarily enhance root biomass and volume, thereby increasing and expanding the absorptive surface area for phosphorus uptake without necessarily increasing primary root length. In maize, some genotypes were found to increase the number and length of lateral roots, while others showed the opposite effect under P deficiency (Zhu et al. 2004). Supporting this variability, our study observed significantly higher shoot dry weight under low phosphorous conditions compared to NP, suggesting that certain genotypes may allocate resources differently to maintain growth under nutrient stress. In the present study, P was the only variable nutrient, while all other nutrients were supplied in equal concentrations across treatments. The observed increase in root volume or surface area under low P conditions may have enhanced the uptake of other nutrients, such as nitrogen, potentially contributing to the significantly higher shoot dry weight recorded under P-

deficient conditions. The findings by Lynch (2005), Guo and York (2019), and Wang et al. (2022) specified the critical role of lateral roots in the acquisition of nitrogen and other nutrients, which in turn supports increased shoot length and biomass under different environmental conditions.

Different studies have reported significantly higher root-to-shoot ratios under low phosphorus conditions compared to normal phosphorus conditions in both wheat (Teng et al. 2013) and maize (Deng et al. 2014). Duan et al. (2020) concluded that the variations in the root-to-shoot ratio in rapeseed were dependent on the genotypic potential. Lopez et al. (2023) reported a significant difference in root biomass but a non-significant difference in the root-to-shoot ratio. Our findings were consistent with this, showing a significant difference in root fresh weight, while the root to shoot ratio remained non-significant. More precisely, under low-phosphorus conditions, root fresh weight reached levels comparable to those of normal phosphorus conditions, suggesting that increased biomass allocation to roots is a key adaptive response. The observed increase in root volume without a proportional increase in root length indicates that plants may rely more on root proliferation and lateral branching rather than on elongation or rapid root turnover. Such morphological adaptations have been widely reported as effective strategies for coping with phosphorus deficiency (Postma et al. 2014; Liu et al. 2017; Liang et al. 2023) and represent one of the major positive outcomes of this study.

Correlation analysis provided a deeper understanding of the interdependence among traits under low P stress. In this study, moderate to high correlations were observed among root- and shoot-related traits, particularly under low-phosphorus conditions. Notably, strong associations between root traits and shoot biomass traits emphasise the crucial role of below-ground traits in sustaining above-ground growth under nutrient-limited conditions. These findings are consistent with earlier reports in maize by Schuster et al. (2024) and Zhang et al. (2013), which emphasised root weight as a key selection criterion for phosphorus efficiency. In particular, the present study observed strong correlations between root fresh and dry weight and total dry matter under stress conditions, whereas these associations were comparatively weaker under normal conditions. This suggests that, in low-phosphorus environments, biomass partitioning toward roots plays a central role in determining overall plant performance.

Furthermore, previous studies by Jia et al. (2018), Liu et al. (2017), and Postma et al. (2014) confirmed that maize acquires the majority of its phosphorus through enhanced lateral root branching rather than increased elongation. These findings support the present results, in which changes in root weight occurred independently of root length. Such correlations highlight the significance of root system traits as reliable indicators of stress resilience under low P conditions.

The screening of genotypes using LPTI and LPPI provided a comprehensive assessment of their adaptability to P-deficient conditions. A positive, moderate correlation ($r = 0.62$) between LPTI and LPPI was observed, which is in agreement with the findings of Li et al. (2021), Schuster et al. (2024) and Yu et al. (2015). This relationship suggests that genotypes exhibiting higher tolerance tend to perform better under stress; however, exceptions to this trend were also noted.

Based on the indices, the maize inbreds were categorised into four distinct groups. (1) *Tolerant and Good-Performing (TG)*: Six genotypes exhibited both high tolerance and superior performance under low P conditions, making them the most desirable candidates for direct utilisation in breeding programs aimed at improving P use efficiency. (2) *Sensitive and Good-Performing (SG)*: Four genotypes that performed well despite exhibiting low tolerance, indicating the presence of efficient internal P utilisation mechanisms or compensatory traits that enable productivity under stress. (3) *Tolerant and Poor-Performing (TP)*: Four genotypes that tolerated low P stress but failed to translate this tolerance into productivity, likely due to limitations in shoot growth. (4) *Sensitive and Poor-Performing (SP)*: Six genotypes were highly susceptible to low phosphorus stress, exhibiting consistently low values for both LPTI and LPPI, indicating limited adaptability and poor potential for use under nutrient-deficient conditions. This classification provides a systematic framework for selecting parental lines with complementary traits, facilitating their use in hybrid development programs.

However, as noted by Schuster et al. (2024), environmental interactions play a major role in nutrient availability, often causing discrepancies between hydroponic and field evaluations. Therefore, validation of hydroponic screening under field conditions is essential to ensure practical applicability. Furthermore, Tuberosa et al. (2002) identified QTLs associated with root characteristics and yield, emphasising the genetic basis of stress adaptation. In the present study, combined screening under both field and hydroponic conditions led to the consistent identification of seven maize genotypes, strengthening their stability and potential for low P tolerance across environments. Among these, four genotypes were categorised as tolerant, while the remaining three were classified as sensitive. This overlap between field and hydroponic results strengthens the reliability of the identified lines, confirming the stability of their performance across both controlled and natural environments.

The consistency of genotypic performance across environments is a critical factor for identifying stable and reliable donor lines in breeding programs. In the present study, genotypes VL181019, VL20213, VL175994, and VL236956 performed well under field screening and were classified as good-performing lines. Importantly, these genotypes also exhibited tolerance and strong performance under hydroponic screening, suggesting that their adaptive mechanisms are stable across both controlled and natural conditions. This dual confirmation further strengthens their potential as promising candidates for the development of phosphorus-efficient maize hybrids.

In contrast, inbred lines ZL22777, VL1885, and VL109138 demonstrated poor performance in field screening and were similarly classified as sensitive and poor-performing under hydroponic evaluation. The consistency of their poor response across both screening methods suggests a lack of key physiological and morphological adaptations required to withstand phosphorus stress. Consequently, their direct use in breeding programs may be limited. However, these genotypes could still serve as valuable contrasting checks in genetic and physiological studies aimed at identifying traits linked with phosphorus sensitivity.

Similar approaches have been adopted in other crops, where comparative selection under both field and hydroponic conditions has been shown to improve accuracy in identifying stress-tolerant genotypes. For instance, Soumya et al. (2021) applied this strategy in wheat genotypes; Meena et al. (2021) screened 100 moong bean genotypes for low-phosphorus tolerance using both methods; and Sun et al. (2025) reported comparable findings in cotton. These studies are in agreement with the effectiveness of integrated screening approaches in improving selection efficiency across diverse crop species.

The findings emphasise the dual importance of root traits and performance indices in identifying phosphorus-efficient maize inbreds. Tolerant and high-performing genotypes are ideal candidates for direct utilisation, whereas tolerant but poor performers may serve as donors of root-related adaptive traits. Similarly, sensitive but good-performing genotypes could contribute traits related to efficient phosphorus utilisation when incorporated into hybrid combinations. This study demonstrates its uniqueness by integrating controlled hydroponic screening with field-level validation to identify phosphorus-efficient maize inbreds, thereby bridging a critical gap between the two conditions. Unlike many previous studies in maize that relied solely on either hydroponic or field conditions, our study validates the stability of genotypic performance across both environments, along with adaptive responses of the root and shoot under low-phosphorus stress, thereby strengthening the reliability of selection.

Furthermore, integrating hydroponic screening, field evaluations, and molecular approaches will improve the precision of genotype selection and accelerate the development of sustainable maize hybrids adapted to phosphorus-deficient soils. This study serves as a valuable bridge between the research gap in the field and hydroponic screening, demonstrating that early-stage hydroponic performance can effectively enhance the selection of maize inbreds for subsequent field evaluation and breeding efforts.

Conclusion

The integration of field and hydroponic screening is essential for the precise and accurate selection of maize genotypes, as responses observed under controlled conditions may differ from those in the field. This study addresses this gap by demonstrating a significant correlation between field and hydroponic screening results, highlighting the potential for early selection of low-phosphorus-tolerant inbred lines in hydroponic conditions. This approach can reduce the cost and resource demands associated with extensive field trials, thereby enhancing breeding efficiency. Genotypes VL181019, VL20213, VL175994, and VL236956 exhibited tolerant performance and high grain weight per plant, whereas VZL22777, VL1885, and VL109138 were susceptible, exhibiting lower grain weight. The overlap of tolerant and susceptible lines across both screening conditions emphasises the reliability of hydroponic screening for early identification of low-phosphorus tolerance. These inbreds represent valuable genetic resources for future breeding programs targeting the development of high-yielding, low-phosphorus-tolerant maize cultivars. However, further multi-environment field screening is necessary to strengthen their selection significance and confirm their stability across diverse conditions.

Declarations

Author Contributions

The initial draft was written developed and conceptualized by V.V.W., and P.K.P. The manuscript was supervised and financially supported by P.K.P. The P.K.P., A.K., N.K., N.S.K., C.L., and S.K. reviewed and provided critical feedback.

Data availability

All data generated or analyzed during this study are included in this published article and its Supplementary Information files.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could appear to have influenced the work reported in this paper.

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Figures

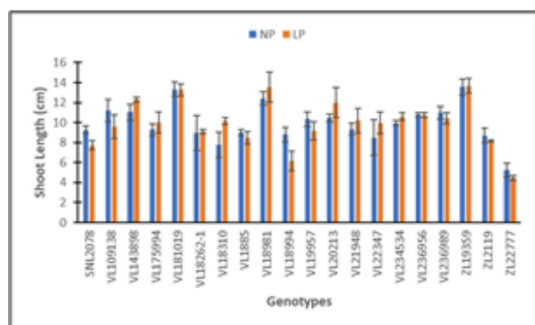


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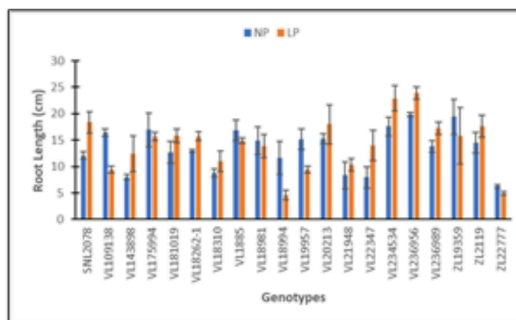


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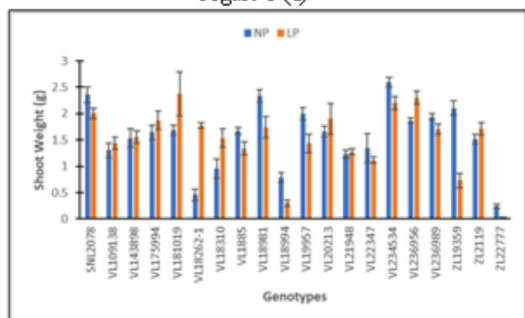


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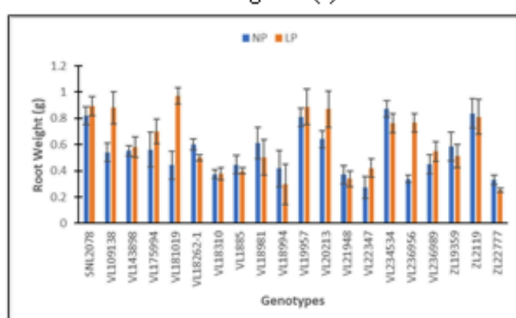


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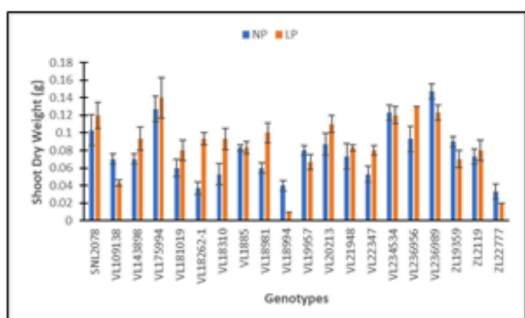


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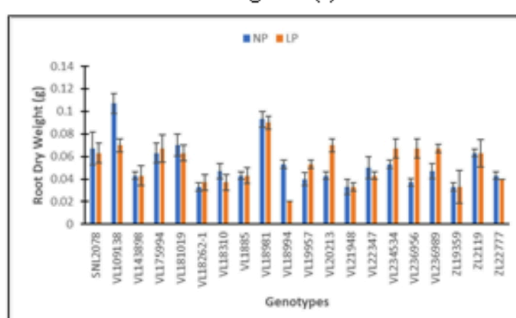


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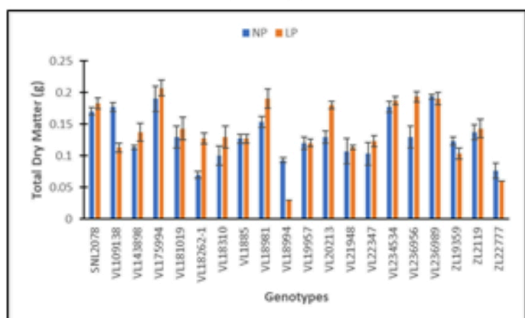


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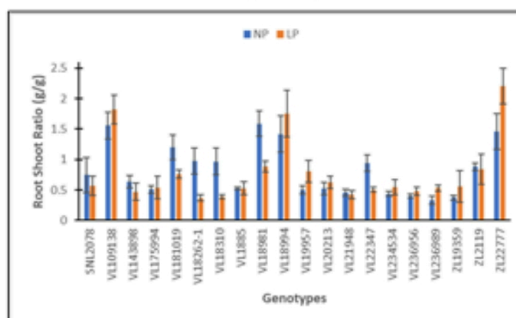


Figure 1 (h)

Figure 1

Bar plots depicting mean performance of maize genotypes (a): Shoot Length (cm); (b): Root length (cm); (c): Shoot fresh weight (g); (d): Root fresh weight (g); (e): Shoot dry weight (g); (f): Root dry weight (g); (g): Total dry matter (g); (h): Root to shoot ratio (g/g) under NP (Normal Phosphorus) and LP (Low Phosphorus) conditions

	SL	RL	SW	RW	SDW	RDW	TDM	RSR	LPTI	LPPI
SL	1.00	0.55	0.63	0.20	0.32	0.26	0.51	-0.16	0.13	0.51
RL	0.45	1.00	0.60	0.41	0.54	0.15	0.54	-0.04	-0.01	0.54
SW	0.52	0.80	1.00	0.55	0.71	0.22	0.72	-0.46	0.10	0.73
RW	0.32	0.52	0.72	1.00	0.36	0.21	0.41	-0.20	-0.02	0.44
SDW	0.48	0.83	0.81	0.39	1.00	-0.01	0.84	-0.71	-0.07	0.65
RDW	0.37	0.51	0.65	0.66	0.53	1.00	0.54	0.64	-0.23	0.22
TDM	0.50	0.81	0.85	0.54	0.95	0.77	1.00	-0.25	-0.07	0.68
RSR	-0.57	-0.66	-0.62	-0.16	-0.80	-0.07	-0.62	1.00	-0.31	-0.44
LPTI	0.48	0.61	0.68	0.21	0.70	0.23	0.61	-0.75	1.00	0.62
LPPI	0.60	0.86	0.94	0.72	0.88	0.78	0.95	-0.63	0.62	1.00

Figure 2

Pearson's Correlation Coefficient for evaluated traits of maize genotypes as influenced by conditions under Normal Phosphorus (above the diagonal) and Low Phosphorus (below the diagonal) (SL- Shoot length; RL- Root length; SW- Shoot fresh weight; RW- Root fresh weight; SDW- Shoot dry weight; RDW- Root dry weight; TDM- Total dry matter; RSR- Root shoot ratio; LPTI- Low phosphorus tolerance index; LPPI- Low phosphorus performance index)

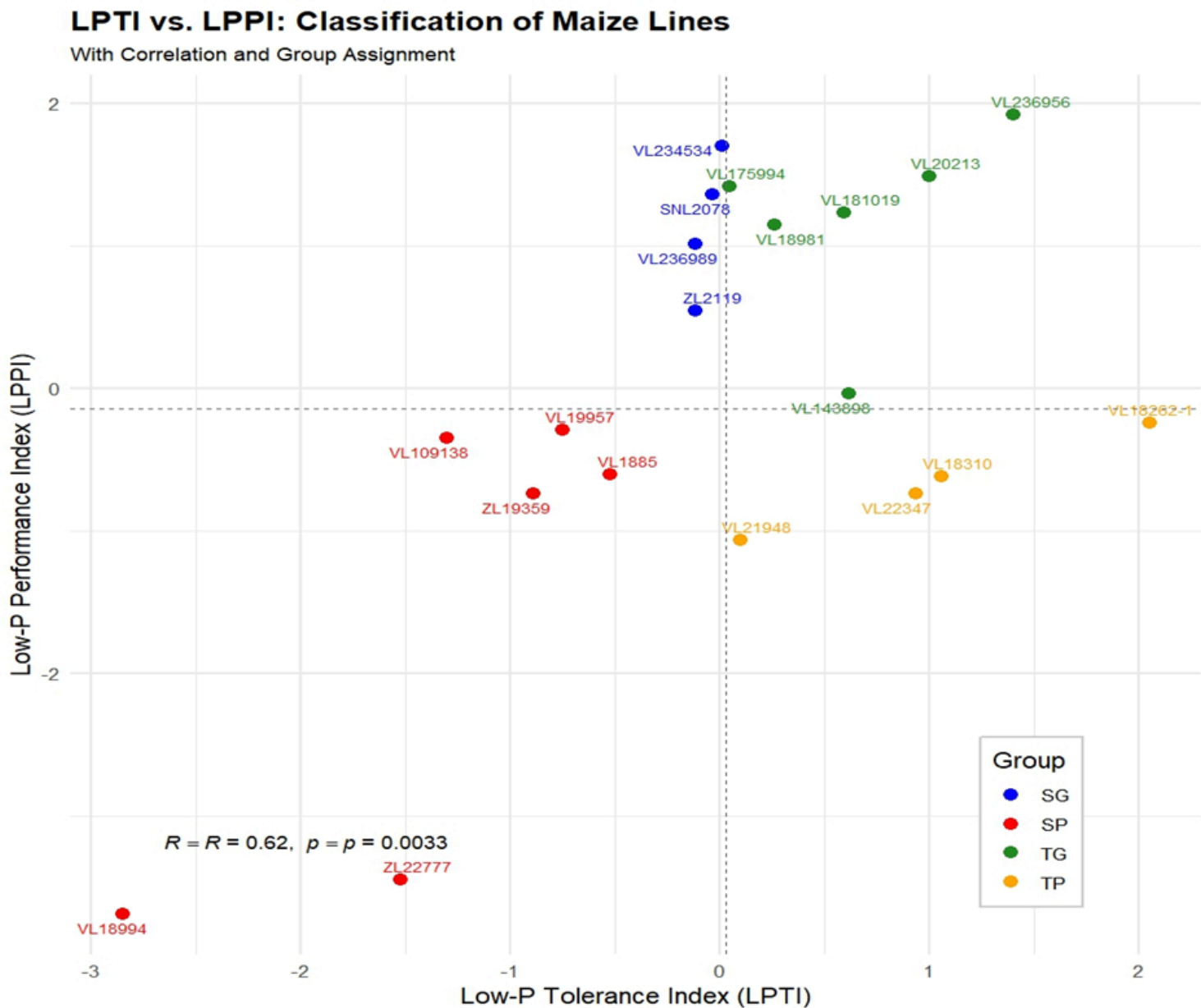


Figure 3

Classification of maize inbreds based on LPPI (Low phosphorus performance index) and LPTI (Low phosphorus tolerance index) indexes

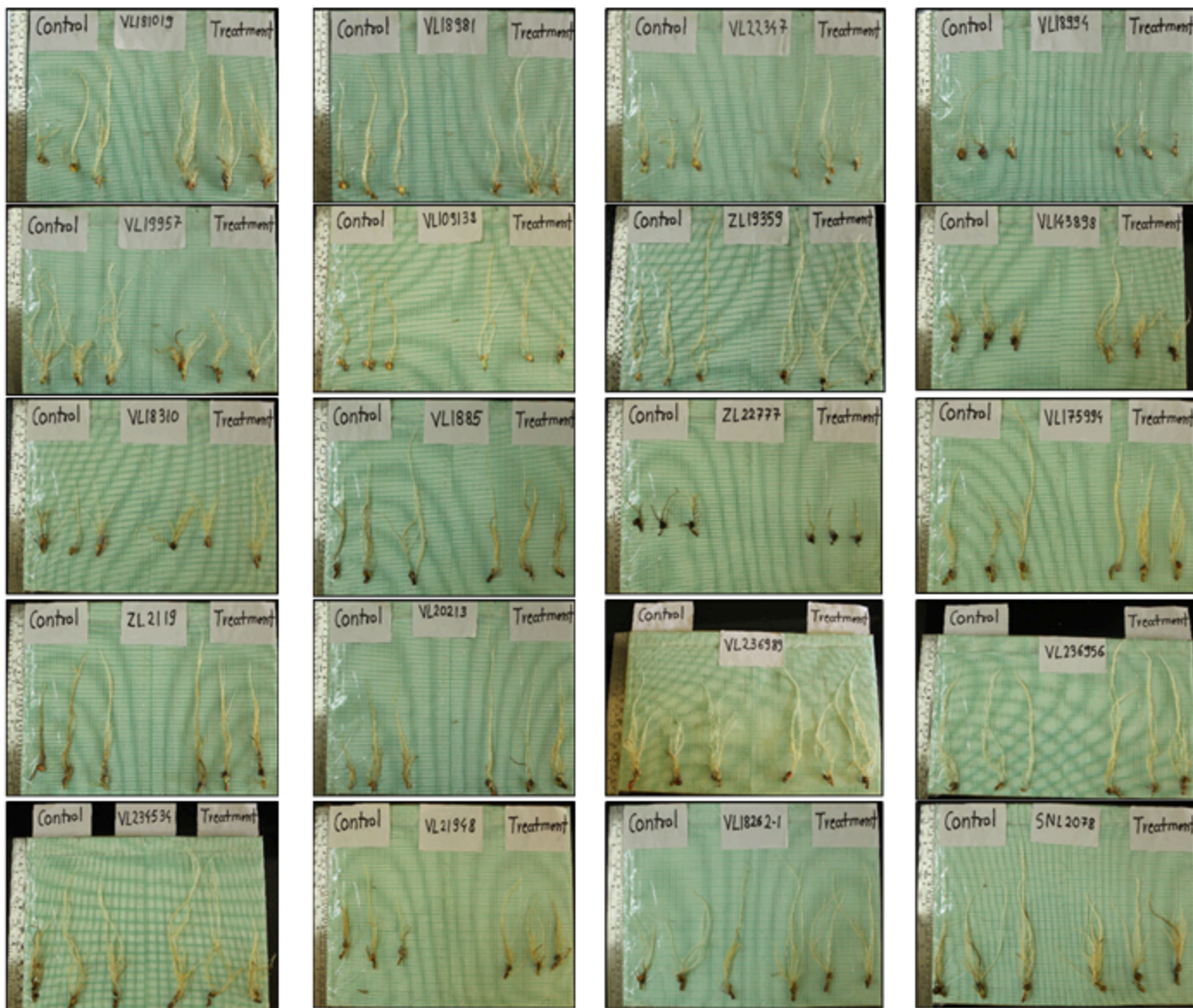


Figure 4

Root growth and proliferation under Control (Normal Phosphorus) and Treatment (Low Phosphorus) in various maize genotypes grown in hydroponic conditions

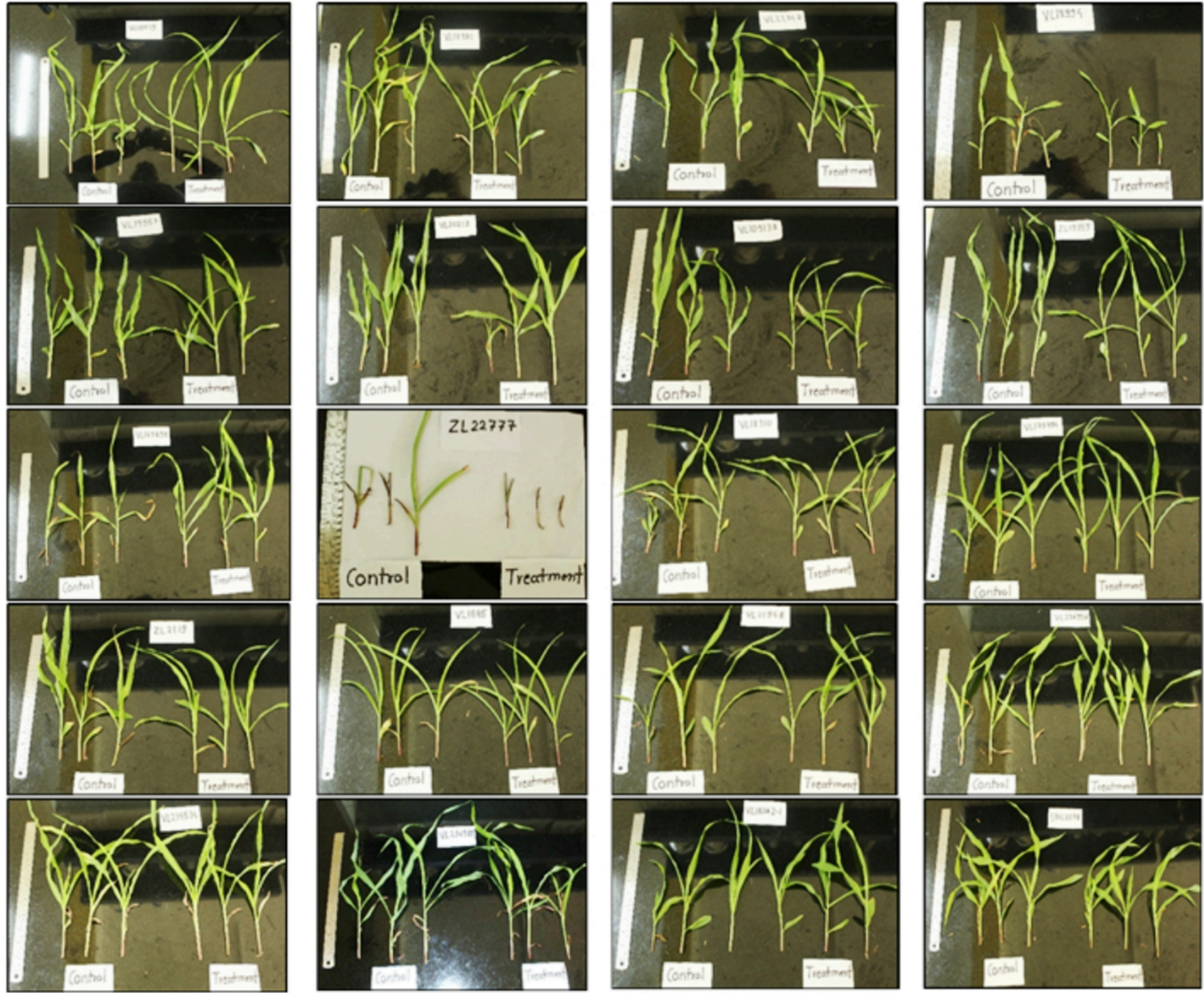


Figure 5

Shoot growth of various maize genotypes under Control (Normal Phosphorus) and Treatment (Low Phosphorus) grown in hydroponic conditions

Supplementary Files

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- [SupplementaryfileHydroponicsdataRawdata.xlsx](#)