Research Square

Preprints are preliminary reports that have not undergone peer review.
They should not be considered conclusive, used to inform clinical practice,
or referenced by the media as validated information.

Effect of Zn, P and rhizobial inoculant on plant growth and photosynthetic activity of chickpea (Cicer arietinum L.) genotypes planted in soils with different textures under glasshouse conditions

Nong Phinias Malesele
Tshwane University of Technology
Mmatladi Tesia Mataboge
University of South Africa
Titus Ngmenzuma
Tshwane University of Technology
Puffy Soundy
Tshwane University of Technology
Sipho Thulane Maseko
emasekts@unisa.ac.za

University of South Africa

Research Article

Keywords:

Posted Date: November 9th, 2026

DOI: https://doi.org/10.21203/rs.3.rs-11319658/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License.
Read Full License

Additional Declarations: No competing interests reported.

Effect of Zn, P and rhizobial inoculant on plant growth and photosynthetic activity of chickpea (*Cicer arietinum* L.) genotypes planted in soils with different textures under glasshouse conditions.

Nong Phinias Malesele [1,] Mmatladi Tesia Mataboge[2], Titus Yeliku-ang Ngmenzuma[1,2], Puffy Soundy[1] and Sipho Thulane Maseko[1,2*]

[1]Department of Crop Sciences, Tshwane University of Technology, South Africa; soundyp@tut.ac.za

[2]Indigenous Technological Knowledge Unit, Department of Agriculture and Animal Health, College of Agriculture and Environmental Sciences, University of South Africa, South Africa; emasekts@unisa.ac.za; matabmt@unisa.ac.za

*Correspondence: emasekts@unisa.ac.za; Tel.: +2712 433 9501

Abstract

This study evaluated the effect of phosphate and zinc applied alone and with rhizobia inoculants on plant growth and leaf gas-exchange parameters of chickpea genotypes grown in two contrasting soils. Four chickpea genotypes were subjected to six nutrient management treatments: control, Zn, P, Zn + P, Zn + P + R_p (peat-based inoculant), and Zn + P + R_l (liquid-based inoculant). Genotype ICCV8101 exhibited high root biomass and root: shoot ratio in both soils. ICCV3110 and ICCV92944 exhibited greater root, shoot, and whole-plant dry weight when grown in silty-clay loam soil, which resulted in a low root: shoot ratio. Applying P alone and in combination with Zn + P, Zn + P + rhizobia inoculant enhanced root and shoot biomass and leaf gas-exchange parameters when compared to sole Zn and control treatments when grown in both soils. However, the response to the fertilizer treatments was genotype- and soil-dependent. For example, in genotype ICCV97024, the highest shoot and whole-plant dry weight, photosynthetic rate, stomatal conductance, and transpiration rate were obtained following application of P alone and Zn + P when established in silty-clay-loam. Similarly, photosynthetic rate, stomatal conductance, and transpiration rate were enhanced by Zn + P + Rp in ICCV3110, while both the combination of Zn + P + Rp and Zn + P + RL improved the parameters in ICCV92944 and ICCV8101 grown in silty-clay loam soils. When established in silty-loam soil, genotypes ICCV3110, ICCV97024, and ICCV92944 exhibited greater root, shoot, and whole-plant dry weight, photosynthetic rate, stomatal conductance, and transpiration rate, and whole-plant dry weight with Zn + P + Rp application. Overall, silty-clay-loam provided more favourable conditions for chickpea growth and photosynthetic functioning.

1. Introduction

Chickpea (*Cicer arietinum* L.) is an important grain legume native to south-eastern Turkey and ranks third worldwide. It has since been adopted and is sold worldwide, including South Africa. Although Africa contributed 4.1% to the world production in 2024 (FAOSTAT, 2026), a large tonnage of the crop is contributed by Ethiopia; none is reportedly contributed by South Africa. The demand for chickpeas is increasing in South Africa due to its higher nutritional value; however, it is only cultivated by smallholder farmers in Mpumalanga and Limpopo provinces as a garden crop. Countries such as the United States of America, Australia, India, Portugal and Italy export chickpeas to South Africa to meet the demand.

A major constraint to chickpea production in South Africa is low soil fertility (Mpai & Maseko, 2018). As reported by Mohale *et al.* (2014), nitrogen (N), phosphorus (P), zinc (Zn), and boron (B) are among the most deficient nutrients in such cropping systems. Of these nutrients, the pool of N in soils can be renewed through biological N fixation, whereby nitrogen-fixing microorganisms convert atmospheric N_2 into ammonia (Wielbo, 2021). However, phosphate is produced from using a non-renewable source, rock phosphate, and is currently threatened by depletion due to economically exploitable rock phosphate reserves (Oloo & Asbon, 2020; Illakwahhi *et al.*, 2024). Recent studies conducted in South Africa demonstrated that application of 90 kg P/ha improved chickpea growth and yield (Madzivhandila *et al.*, 2012). However, the application of P, especially as a single fertilizer at the smallholder level, is hindered by its high cost, especially when bought from retailers (Mpai & Maseko, 2018). The lack of or insufficient fertilization of food crops leads to a significant reduction in their nutritional value. This background requires the need of alternative approaches that will result in improved solubility and uptake of native nutrients in soils cultivated by low-income farmers. However, when crops are grown without the application of phosphate fertilizers, they adopt to mechanisms that enhance the solubility and uptake of P, and these include mycorrhizal symbiosis, formation of cluster roots, and the production of organic acids (Lazali & Drevon, 2018). However, these mechanisms do not contribute enough P to balance that taken up through crop off-take or harvest.

Zinc, on the other hand, is involved in auxin metabolism, protein synthesis, formation of nucleic acid, promotes nodulation and nitrogen fixation in leguminous crops, and enhances

assimilation and utilization of N and P in plants. Plants supplied with enough Zn maintain higher activities of antioxidant enzymes (Balai *et al.*, 2017; Hamzah Saleem *et al.*, 2022; Dang *et al.*, 2024). In soils, Zn is contributed by the addition of livestock manures, fertilisers, sewage sludge, agrochemicals and Zn-containing fungicides (Luo *et al.*, 2009). In South African smallholder cropping systems, most applied chemical fertilizers, though not regularly, are NPK fertilizers. Although NPK fertilizers include Zn, the amounts are insufficient, and this practice has led to Zn deficiency in these cropping systems. Literature on Zn status of South African soils indicates that agricultural fields under commercial maize production showed decreased in Zn soil levels. For example, results on Zn levels in fields cultivated by smallholder farmers recorded a range between 0.77 and 4.31 mg/kg topsoil in selected fields (Habinshuti *et al.*, 2015). There is a link between the concentration of Zn in cropping fields and the Zn status of humans who eat such crops.

From this background, most cropping fields cultivated by smallholder farmers in South Africa experience inadequate replacement of P and Zn. To promote sustainable production of chickpea, environmentally friendly approaches that focus on reducing inputs without compromising yield or quality are required. This includes the use of biofertilizers. Biofertilizers contain substances and/or micro-organisms whose function, when applied to plants or the rhizosphere, is to stimulate natural processes to enhance/benefit nutrient uptake, nutrient efficiency, tolerance to abiotic stress, and crop quality (Fahde *et al.*, 2023; Jaiswal & Dakora, 2025). Several biofertilizer products, including rhizobial inoculants, are produced locally and readily available in South African markets, and their use in cropping systems has hardly been reported to lead to environmental pollution. Despite the availability of microbial inoculants, their adoption among smallholder farmers remains low, largely because many farmers are unaware of their benefits. The application of rhizobial inoculants promotes plant growth and productivity, particularly in soils dominated by ineffective indigenous rhizobia. At the smallholder level, researchers have shown that the use of rhizobium inoculants supplemented with fertilizers in producing grain enhanced legume production (Muthabi, 2020; Ogola *et al.*, 2023). A recent study showed that inoculating soybean and groundnut with rhizobial inoculants can reduce fertilizer input for smallholder farmers (Nemaembeni, 2023). This underscores the importance of rhizobial inoculants in the production of grain legumes and ensures sustainable and low-cost production by smallholder farmers. Sustainable production of N-fixing legumes such as chickpea requires regular application of, especially, the most deficient mineral nutrients such as P and Zn, alone and in combination with biofertilizers (van Vugt et al., 2018). Although

the application of P and Zn alone and rhizobia inoculant have been reported to improve crop performance, their interaction in enhancing chickpea productivity has not been adequately investigated. This study evaluated the effect of P and Zn fertilizer, and in combination with liquid and peat-based inoculants, on growth and photosynthetic performance of four chickpea genotypes grown under two contrasting soil textures. We hypothesized that the combined application would exert synergistic effects, resulting in greater improvements in plant growth and photosynthetic traits in different chickpea genotypes than either treatment alone. Moreover, we also expected that treatment response would be genotype- and soil- dependent. The findings are anticipated to provide insight into sustainable fertilizer strategies and soil-specific chickpea production systems in South Africa

2. Material and Methods

2.1. Description of the locations where the soil was collected, processing and analysis of soil

The experimental design, treatments, and crop management practices used in the present study have been described previously by (Maseko *et al.*, 2026). Briefly, about 3 kg of silty-clay-loam and silty-loam were added into clean and uncontaminated plastic pots, which were arranged on benches in a naturally ventilated glasshouse and laid out in a randomized complete block design with three replications. Treatments included 1) four desi-type chickpea genotypes (ICCV97024, ICCV92944, ICCV3110, and ICCV8101); 2) six levels of fertilizer, namely, no-fertilizer application, adding Zn (zinc sulphate heptahydrate) at 10 kg/ha, adding P (superphosphate) at 40 kg/ha, adding Zn and P; Zn plus P (P(peat-based inoculant)) denoted as [Zn + P + R_P]; and Zn plus P(L (liquid-based inoculant)) denoted as [Zn + P + R_L]; and 3) two soil texture (silty-clay-loam and silty-loam). Whereas the previous publication focused on mineral accumulation, this study evaluated biomass and leaf gas exchange parameters from the same experimental units.

2.2. Measurement of photosynthesis, transpiration and stomatal conductance

At the flowering growth stage, corresponding to 80 days after planting (DAP), gas-exchange measurements were performed on fully expanded trifoliate leaves from each replicated pot. The readings were taken on a sunny day between 09:00 a.m to 12:00 p.m, using a portable infrared gas analyzer (Li 6400XT, version 6.2) Lincoln, Nebraska. Photosynthetic rates were

measured in triplicates and expressed on a per-unit-mass basis (μmol CO_2 g^{-1} s^{-1}). Apart from the photosynthetic rates, stomatal conductance and transpiration rates were also measured from the same trifoliate leaf. The photosynthetic water-use efficiency (PWUE) was calculated as the photosynthetic rate over leaf transpiration rate (Lebogang et al. 206; Bunce, 2016).

2.3. Assessing plant biomass

To determine plant growth, immediately after gas exchange measurement, the aboveground part of each plant stand was cut off at the crown level, and the remaining roots were carefully dug out in each experimental pot. The soil attached to the root was shaken off from the root, and the soil attached to the roots was then washed off with tap water. Each shoot and root were kept into a pre-labelled paper bag and oven-dried at 65 °C for 72 h to constant weight. After the samples were oven-dried, each was weighed in order to determine the shoot dry weight.

2.4. Data analysis

The data generated from this study were subjected to a normality test before been subjected to analysis of variance using a STATISTICA software program version 10.0 (StatSoft, Inc., Tulsa, OK, USA). Plant growth and leaf gas-exchange parameters were compared between the test genotypes and fertilizer treatments using a two-way Analysis of Variance (ANOVA). Where treatment means showed significant differences, the Duncan's Multiple Range Test (DMRT) was used to separate the means at $p \leq 0.05$.

3. Results

3.1. Genotypic Variation in plant growth

Genotype had a significant effect ($p \leq 0.05$) on plant growth parameters in both soil textures (Table1). When chickpea was planted in silty-clay loam soil, the root, shoot, and whole-plant dry weights were markedly higher in ICCV3110 and ICCV92944, which resulted in a low root: shoot ratio. Genotype ICCV8101 exhibited higher root biomass and root: shoot ratio but low shoot dry matter and whole-plant dry matter in both soils. When ICCV92944 was planted in

the silty-loam soil, it exhibited the highest shoot and whole-plant biomass, while ICCV97024 produced the lowest (Table 1).

The application of P alone and in combination with other fertilizers (Zn + P, Zn + P + R_p, Zn +P + R_L) significantly increased root, shoot, and whole-plant dry weights compared to the control or sole Zn application in both soil textures. However, the zero control recorded an increased in root dry weight of chickpea plants grown in silty-clay loam soil (Table 1).

3.2. Interactive effect of plant growth on silty-clay-loam

There were significant effects of genotype x treatment interaction ($p \leq 0.001$) on root, shoot, whole-plant, and root: shoot dry weights of chickpea plants (Table 1). Genotype ICCV97024 treated with Zn + P + R_L (0.72 g/plant) and Zn + P + R_p (0.75 g/plant) recorded the highest root dry weight; this was followed by sole P (0.70 g/plant). As shown in Figure 1A. the application of Zn + P and Zn + P + R_L treatments led to greater root dry weight in genotype ICCV3110, while Zn + P and Zn + P + R_p treatments induced high root dry weight in genotype ICC8101. However, the control treatment only increased root dry weight in genotype ICCV929244. As shown in Figure 1B and C, the application of P either alone or in combination with other fertilizer treatments increased shoots and whole-plant dry weight among the chickpea genotypes. For example, sole P application revealed the highest shoot and whole-plant dry weight in genotypes ICCV97024 and ICCV3110, while Zn + P + R_p induced the highest in ICCV92944. With root: shoot, high values were recorded in the control plants and sole Zn treatment in all genotypes. With root: shoot ratio, the highest values were recorded in untreated plants in all genotypes, except for genotype ICCV3110, which recorded the highest root: shoot ratio value in Zn treatment (Figure 1D). However, the control plants in ICCV92944 genotype recorded the highest root: shoot ratio across the four genotypes.

3.3. Interactive effect of plant growth on silty-clay-loam

As shown in Figure 2A, the combination of Zn + P + R_p treatment of genotype ICCV97024 resulted in higher root dry weight, followed by P, Zn + P, and Zn + P + R_L. The application of Zn + P + R_L, Zn + P + R_p and Zn + P significantly increased root dry weight in genotypes ICCV3110 and ICCV8101 when compared to sole application of Zn and P as well as the control plants. Root dry weight was increased in the genotype ICCV92944 that was treated with P

(0.68 g/plant) alone, followed by the combination of Z + P (0.62 g/plant). However, root dry weight was lower in genotype ICCV92944 that was treated with Zn + P + R_L (0.45 g/plant).

Among all the genotypes, sole P and the combination of Zn and rhizobia inoculants resulted in significantly ($p \leq 0.001$) higher shoot dry weight when compared to the zero control and Zn alone. For example, the Zn + P + R_p treatment revealed a higher shoot dry weight in genotype ICCV97024 and ICCV3110 when compared to all other treatments (Figure 2 B and C). In genotype ICCV92944, the combination of Zn + P led to significantly increased in shoot dry weight when compared to sole application and combination with rhizobia inoculants. A similar trend was observed in genotype ICCV8101, with sole P and Zn + Zn promoting higher shoot dry weight. As shown in Figure 2C, the whole-plant dry weight mirrored the trend observed for shoot dry weight such that sole P and in combination treatment increased whole-plant dry weight in all genotypes, with values ranging from 1.36 g/plant to 3.37 g/plant when compared to control and sole Zn treatments. In all the genotypes, the control treatment significantly increased the root: shoot ratio (0.79 to 3.38) when compared to fertilizer application (Figure 2D). However, Zn + P also resulted in a higher root: shoot ratio.

Table 1: The combined effect of fertilizer and biofertilizer application on plant biomass of desi-type chickpea genotypes grown in silty-clay-loam and silt-loam soil. Zn + P + R_P represents Zinc plus P plus peat-based inoculant, while Zn + P + R_L represents Zinc plus P plus liquid-based inoculant. Mean values with dissimilar letters in a column are significantly different at ** $p \leq 0.01$. *** $p \leq 0.001$. ns = not significant

	Silty-clay loam				Silt loam			
Treatments	**Root dry weight**	**Shoot dry weight**	**Whole-plant dry weight**	**Root:shoot ratio**	**Root dry weight**	**Shoot dry weight**	**Whole-plant dry weight**	**Root:shoot ratio**
	g plant^{-1}							
Genotype								
ICCV97024	0.61±0.02b	1.40±0.17ab	2.01±0.19ab	0.74±0.10b	0.51±0.03b	1.90±0.20c	1.50±0.13c	0.58±0.04b
ICCV3110	0.80±0.05a	1.61±0.18a	2.41±0.21a	0.73±0.07b	0.53±0.03b	2.20±0.22bc	1.63±0.15bc	0.61±0.06b
ICCV92944	0.78±0.05a	1.71±0.15a	2.49±0.14a	0.37±0.02c	0.55±0.03b	2.69±0.24a	1.97±0.13a	0.46±0.05b
ICCV8101	0.80±0.03a	1.07±0.13b	1.87±0.14b	0.99±0.08a	0.64±0.02a	2.28±0.40b	1.73±0.15b	1.00±0.29a
Fertilizer								
Control	0.76±0.07a	0.73±0.12b	1.50±0.14b	1.35±0.24a	0.44±0.03b	0.96±0.11d	0.86±0.08d	1.48±0.38a
Zn	0.55±0.05b	0.75±0.11b	1.29±0.11b	0.82±0.08b	0.49±0.02b	1.46±0.16c	1.19±0.09c	0.74±0.05b
P	0.77±0.03a	1.87±0.17a	2.63±0.17a	0.48±0.07b	0.60±0.03a	2.93±0.16a	2.11±0.09a	0.42±0.03c
Zn + P	0.76±0.06a	1.93±0.12a	2.68±0.15a	0.41±0.03b	0.60±0.03a	2.89±0.26a	2.03±0.14ab	0.43±0.04c
Zn + P + Rp	0.85±0.05a	1.87±0.19a	2.71±0.20a	0.55±0.09b	0.61±0.04a	3.02±0.11a	2.18±0.08a	0.42±0.04c
Zn + P + RL	0.80±0.04a	1.56±0.15a	2.36±0.14a	0.79±0.29b	0.60±0.03a	2.39±0.19b	1.86±0.11b	0.48±0.04bc
F-statistics								
Genotypes	4.60***	3.05*	2.83*	14.19***	7.71***	9.02***	10.25***	8.81***
Fertilizer	3.93***	14.47***	15.85***	4.19**	8.96***	42.37***	52.31***	19.11***
G x F	6.88***	2.62**	3.03**	2.90**	2.59**	2.42*	2.43*	7.36***

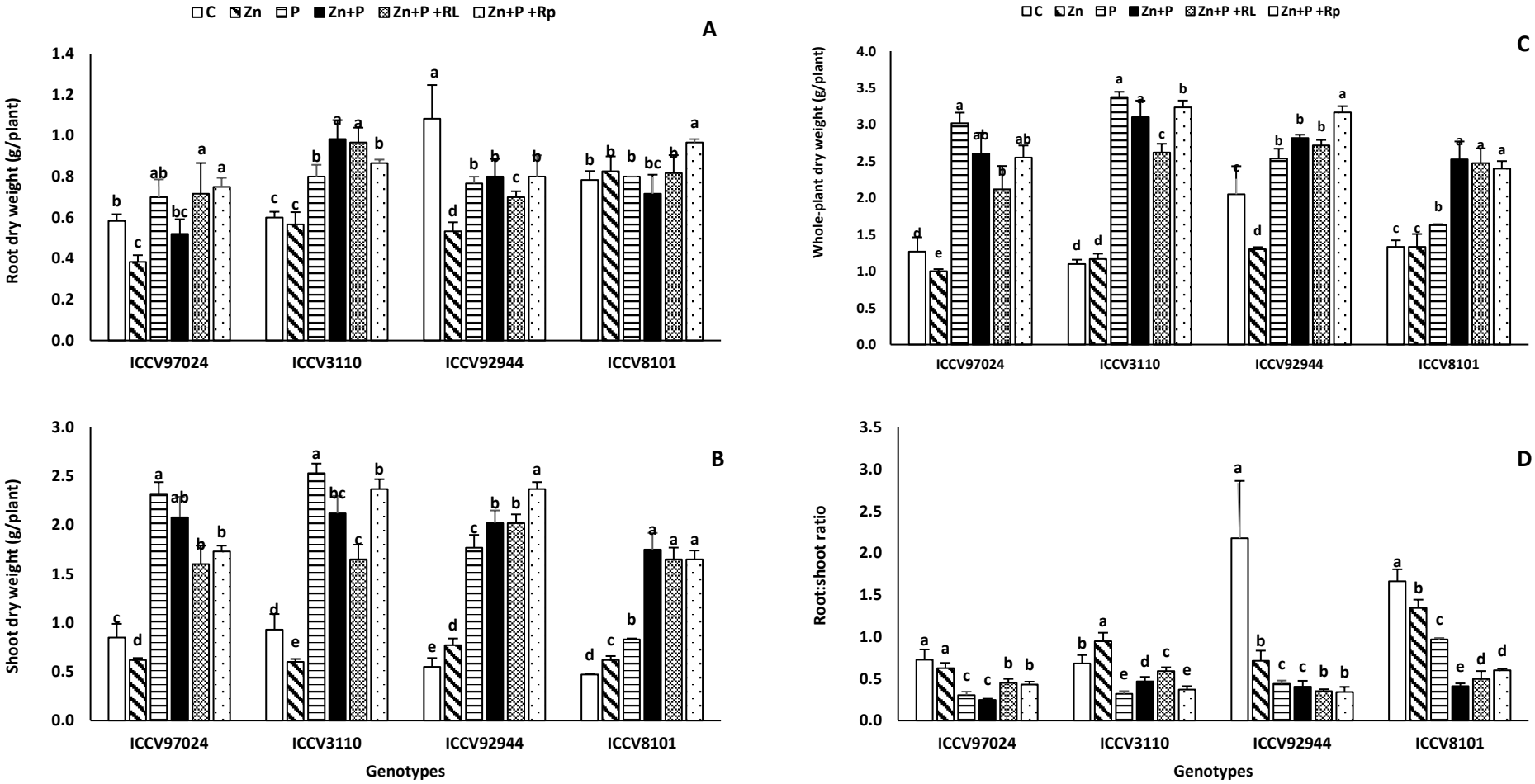


Figure 1: The interactive effects of genotype x fertilizer applications on A) root dry weight B) shoot dry weight C) whole plant dry weight and D) root:shoot weight of chickpea grown in silty-clay-loam soils collected from the field under glasshouse conditions. For each parameter, bars with dissimilar alphabets in a row are significantly different at $p \leq 0.05$. Error bars represent SE. Zn + P + R_P represents Zinc plus P plus peat-based inoculant, while Zn + P + R_L represents Zinc plus P plus liquid-based inoculant.

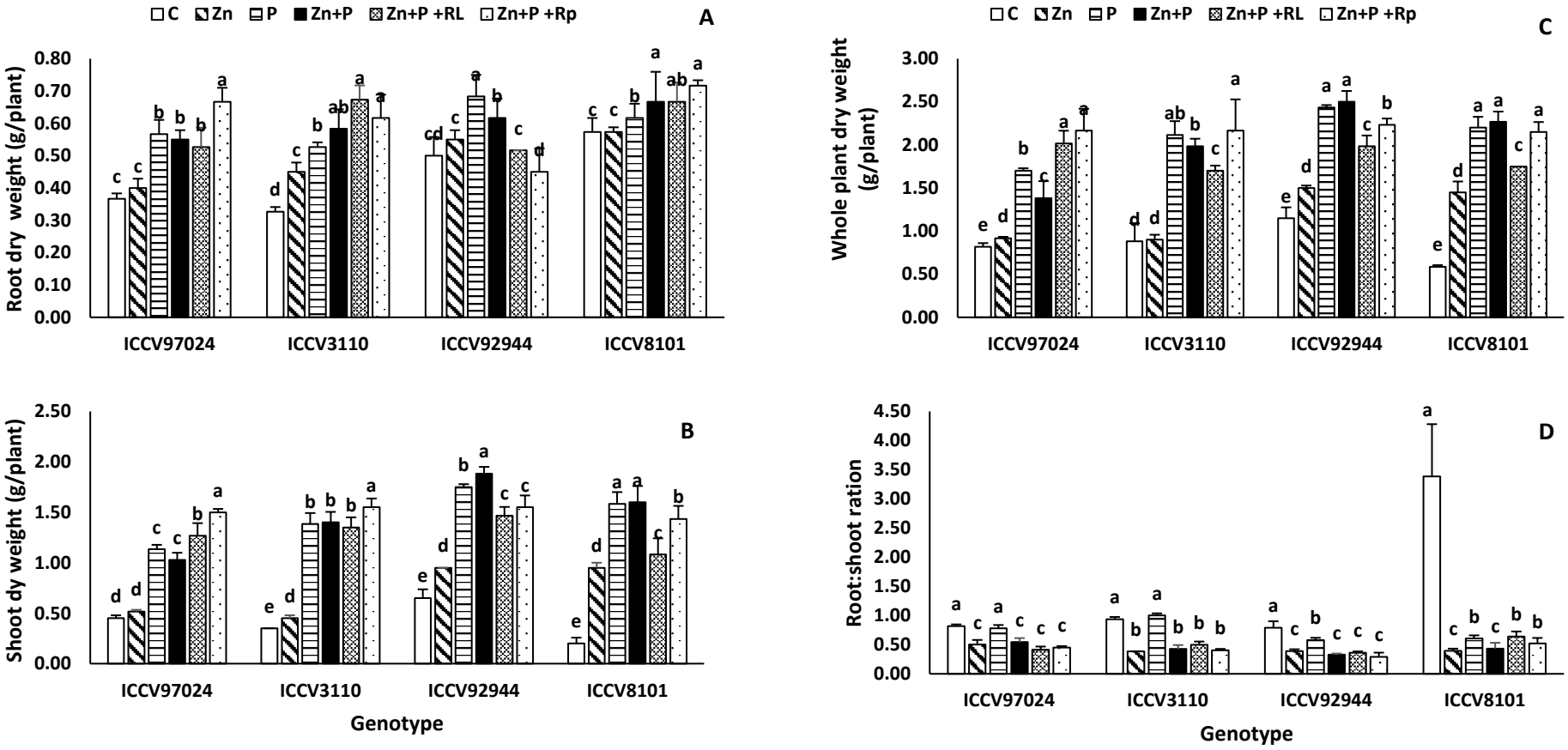


Figure 2: The interactive effects of genotype x fertilizer applications on A) root dry weight B) shoot dry weight C) whole plant dry weight and D) root: shoot weight of chickpea grown in silty-loam soils collected from the field under glasshouse conditions. For each parameter, bars with dissimilar alphabets in a row are significantly different at $p \leq 0.05$. Error bars represent SE. Zn + P + R_P represents Zinc plus P plus peat-based inoculant, while Zn + P + R_L represents Zinc plus P plus liquid-based inoculant.

3.4. Genotypic Variation in Photosynthetic Performance

When chickpea was established in silty-clay loam soil, ICCV97024 had a higher photosynthetic rate (10.70 µmol CO_2 m^{-2} s^{-1}), stomatal conductance (0.16 mol H_2O m^{-2} s^{-1}), leaf transpiration rate (2.92 mol H_2O m^{-2} s^{-1}), and intercellular CO_2 concentration (263.67 µmol CO_2 $molair^{-1}$), relative to other genotypes (Table 2). Interestingly, when the same genotype (ICCV97024) was grown in silt loam soil, it recorded the lowest photosynthetic rate (4.50 µmol CO_2 m^{-2} s^{-1}), stomatal conductance (0.06 mol H_2O m^{-2} s^{-1}), and intercellular CO_2 concentration (243.40 µmol CO_2 $molair^{-1}$) among the genotypes. However, its intrinsic water-use efficiency was among the lowest in silty-clay loam soil and higher in silt loam soil.

When chickpea was grown in silt loam soil, genotypes ICCV92944 and ICCV8101 exhibited the highest photosynthetic rate, stomatal conductance, and leaf transpiration rate, while ICCV8101 maintained the highest intercellular CO_2 concentration. On the other hand, ICCV3110 exhibited markedly an increased in photosynthetic rate (5.43µmol CO_2 m^{-2} s^{-1}) and leaf transpiration rate (1.91 mol H_2O m^{-2} s^{-1}), though it was not statistically different from ICCV92944 and ICCV8101. The lowest intrinsic water-use efficiency ($p \leq 0.001$) was recorded in genotype ICCV8101, followed by ICCV3110, and ICCV92944, with values ranging between 67.16 µmol (CO_2) mol^{-1}(H_2O) and 82.27 µmol (CO_2) mol^{-1}(H_2O)

3.5. Fertilizer Treatment Effects

The application of sole P and in combination with Zn + P, Zn + P + R_p, and Zn + P + R_L significantly improved photosynthetic rate, stomatal conductance, and leaf transpiration rate of chickpea plants grown in silty-clay loam and silty-loam soils (Table 2). Furthermore, the application of sole P and P + Zn reduced intercellular CO_2 concentration, while it markedly increased intrinsic water-use efficiency when grown in silty-clay loam soil. A similar trend was observed in silt loam soil, when adding sole P, Zn + P, Zn + P + R_p, and Zn + P + R_L improved intrinsic water-use efficiency with low intercellular CO_2 concentrations relative to sole Zn and the control chickpea plants.

.

Table 2: The combined effect of fertilizer and biofertilizer application on shoot dry matter and leaf gas exchange parameters on desi-type chickpea genotypes grown in silty-clay-loam soil and silt loam soil. Zn + P + R_P represents Zinc plus P plus peat-based inoculant, while Zn + P + R_L represents Zinc plus P plus liquid-based inoculant. Mean values with dissimilar letters in a column are significantly different at ** $p \leq 0.01$. *** $p \leq 0.001$. ns = not significant.

Treatment	Photosynthetic rate	Stomatal conductance	Transpiration rate	Intercellular CO_2 Concentration	Intrinsic water-use-efficiency	Photosynthetic Rate	Stomatal conductance	Transpiration rate	Intercellular CO_2 Concentration	Intrinsic water-use-efficiency
	µmol (CO_2) m^{-2} s^{-1}	mol (H_2O) m^{-2} s^{-1}		µmol(CO_2)mol air^{-1}	µmol(CO_2) mol $(H_2O)^{-1}$	µmol(CO_2)m^{-2} s^{-1}	mol (H_2O) m^{-2} s^{-1}		µmol (CO_2) mol air^{-1}	µmol (CO_2) mol $(H_2O)^{-1}$
	Silty-clay loam soil					Silty-loam soil				
Genotypes										
ICCV97024	10.70±0.60a	0.16±0.01a	2.92±0.15a	263.67±3.50a	70.82±2.30b	4.50±0.27b	0.06±0.003c	1.57±0.08b	243.40±3.70c	82.27±2.20a
ICCV3110	5.70±0.35b	0.07±0.004b	1.54±0.08b	249.13±4.70b	79.55±2.80b	5.43±0.29a	0.07±0.004b	1.91±0.07a	254.13±2.50b	74.86±1.50b
ICCV92944	5.14±0.38b	0.07±0.01b	1.57±0.1b	253.29±5.70b	76.95±3.50b	5.97±0.33a	0.08±0.01a	2.01±0.11a	250.64±3.00b	76.83±1.90b
ICCV8101	5.16±0.26b	0.06±0.004b	1.56±0.09b	227.74±6.20b	92.65±4.00a	5.79±0.23a	0.09±0.003a	1.94±0.05a	267.54±2.10a	67.16±1.20c
Fertilizer										
Control	3.96±0.35b	0.057±0.01b	1.28±0.09cg	270.69±5.1a	68.62±3.20c	3.94±0.19d	0.06±0.003d	1.63±0.07c	267.83±3.05a	67.83±1.92c
Zn	4.50±0.35b	0.06±0.01b	1.40±0.11c	262.96±5.3a	72.88±3.20bc	4.08±0.27d	0.06±0.01d	1.54±0.09c	262.06±3.35a	71.57±2.16c
P	7.30±0.94a	0.09±0.02a	1.83±0.24b	213.40±9.5c	102.35±6.20a	5.20±0.39c	0.07±0.01cd	1.72±0.11c	250.88±3.51b	77.41±2.09a
Zn + P	7.86±0.77a	0.10±0.01a	1.98±0.21ab	226.63±6.2c	93.04±4.10a	6.23±0.33ab	0.08±0.003bc	2.02±0.08ab	246.84±4.07b	78.86±2.34a
Zn + P + Rp	8.33±0.36a	0.11±0.01a	2.37±0.10a	244.67±4.3b	79.93±2.50b	5.97±0.32bc	0.08±0.01b	1.97±0.09b	248.01±3.94b	78.52±2.77a
Zn + P + RL	7.36±0.36a	0.11±0.04a	2.34±0.07a	265.54±4.1a	67.67±2.30c	7.01±0.33a	0.09±0.01a	2.23±0.12a	247.17±3.06b	78.05±1.99a
F-statistics										
Genotypes	8.13***	16.40**	7.37***	16.99***	16.83***	8.13**	16.40***	7.37***	16.99***	16.83***
Fertilizer	19.86***	10.88***	9.36***	9.19***	6.34***	19.86***	10.88***	9.36***	9.19***	6.34**
G x F	3.37***	15.64***	3.49***	5.16***	4.85***	3.37***	3.26***	3.49***	5.16***	4.85***

3.6.Comparison of soil texture on plant growth and photosynthetic activity

Of the two soil textures, chickpea grown in the silty-clay-loam soil exhibited greater root, shoot, and whole-plant dry weight (Table 3). Similarly, chickpea planted in the silty-clay loam soil revealed the highest photosynthetic rate (6.61 µmol (CO_2) $m^{-2}s^{-1}$), stomatal conductance (0.09 mol (H_2O) $m^{-2}s^{-1}$), and intrinsic water-use efficiency (80.49 µmol (CO_2) mol^{-1} (H_2O) when compared to silt loam. This translated into greater root, shoot, and whole-plant dry weight. However, the root: shoot ratio was not influenced by soil texture, while intercellular CO_2 concentration (253.81 µmol (CO_2)$molair^{-1}$)) and leaf transpiration (2.93 mol (H_2O) $m^{-2}s^{-1}$) were high in chickpea grown in silt loam soil.

Table 3: A 2-way comparison of soil texture on plant growth and photosynthetic parameters of chickpea genotypes planted under glasshouse conditions. Where Mean values with dissimilar letters in a column are significantly different at (* $p \leq 0.05$); (** $p \leq 0.01$) and (*** $p \leq 0.001$).

Treatment	**Root dry weight**	**Shoot dry weight**	**Whole-plant dry weight**	**Root: Shoot ratio**	**Photosynthetic rate**	**Stomatal conductance**	**Intercellular CO_2 concentration**	**Transpiration rate**	**Intrinsic water-use efficiency**
	g/plant	**g/plant**	**g/plant**		**µmol $(CO_2)m^{-2}s^{-1}$**	**mol$(H_2O)m^{-2}s^{-1}$**	**µmol(CO_2)molair^{-1}**	**mol$(H_2O)m^{-2}s^{-1}$**	**µmol(CO_2)mol$^{-1}(H_2O)$**
Silty-clay loam	0.75±0.02a	1.49±0.08a	2.23±0.09a	0.67±0.06a	6.61±0.25a	0.09±0.004a	248.42±2.84b	1.88±0.06b	80.49±1.7a
silty-loam	0.56±0.01b	1.14±0.06b	1.71±0.07b	0.66±0.08a	5.43±0.14b	0.07±0.002b	253.81±1.57a	2.93±0.14a	74.95±0.9b
F-statistics	18.66***	69.73***	11.17***	18.14***	25.60***	24.08**	5.75*	97.64**	97.64**

3.7. Interactive effect of silty-clay loam soil texture on photosynthetic parameters of chickpea genotypes planted under glasshouse conditions.

The data revealed a significant effects on genotype x treatment ($p \leq 0.001$) on the photosynthetic rate, stomatal conductance, leaf transpiration ratye, intercellular CO_2 concentration, and intrinsic water-use efficiency (Table 2). As shown in Figure 3A, B and C, the application of sole P and P + Zn in ICCV97024 recorded the highest photosynthetic rates, stomatal conductance, and leaf transpiration rate ($p \leq 0.001$) when compared to other genotypes and treatments. The photosynthetic parameters were generally higher in the Rhizobium-inoculated treatments. For example, ICCV92944 and ICCV8101 treated with Zn + P + R_p and Zn + P + R_L, as well as genotype ICCV3110 treated with Zn + P + Rp, recorded the highest photosynthetic rate, stomatal conductance, and leaf transpiration ($p \leq 0.001$). Moreover, application of P alone and in combination with other treatments resulted in higher photosynthetic rate, stomatal conductance, and leaf transpiration rate in ICCV97024, ICCV3110, and ICCV92944 genotypes when compared to sole Zn and control chickpea plants. However, photosynthetic rate was reduced in the plants of genotype ICCV8101 that were treated with sole P (3.22 µmol CO_2 m^{-2} s^{-1}) and P + Zn (3.47 µmol CO_2 m^{-2} s^{-1}) when compared to the other treatments.

Intercellular CO_2 concentration differed significantly ($p \leq 0.001$) among the tested genotypes. Application of P alone and P + Zn significantly decreased intercellular CO_2 concentrations in genotypes ICCV3110, ICCV92944, and ICCV8101 when compared to other treatments on the same genotypes. However, the control, Zn alone, and Zn + P + R_L promoted much higher intercellular CO_2 concentrations across the three genotypes. Interestingly, the control in chickpea plants resulted in lower intercellular CO_2 concentration in genotype ICCV97024 when compared to fertilizer treatments (Figure 3D).

With the intrinsic water-use efficiency, the highest values ($p \leq 0.001$) were recorded in the control plants by genotype ICCV97024, followed by those treated with sole Zn and Zn + P + RL of the same genotype (Figure 3E). In genotype ICCV3110, chickpea plants treated with the combination of Zn + P (104 µmol (CO_2) mol^{-1}(H_2O)), followed by sole P (91.72 µmol (CO_2) mol^{-1}(H_2O)), promoted higher intrinsic water-use efficiency. The highest intrinsic water-use efficiency was recorded with sole P application (114.85 µmol (CO_2)), followed by Zn + P (115 µmol (CO_2) mol^{-1}(H_2O) in genotype ICCV8101 and sole P (106.65 µmol (CO_2) mol^{-1}(H_2O)) in genotype ICCV92944 (Figure 3E).

3.8. Interactive effect of silt loam soil texture on the photosynthetic parameters of chickpea genotypes planted under glasshouse conditions

The photosynthetic activity of chickpea plants revealed a significant difference ($p \leq 0.001$) between genotypes and fertilizer treatments (Table 2). In genotypes ICCV97024, ICCV3110, and ICCV92944, the combination of Zn + P, Zn + P + R_p, and Zn + P + R_L led to significantly higher photosynthetic rate, stomatal conductance, and transpiration rate when compared to sole application and control treatments (Figures 4A, B, and C). However, sole P application resulted in the highest photosynthetic rate, stomatal conductance, and leaf transpiration rate for genotype ICCV8101.

For genotype ICCV97024, intercellular CO_2 concentration was greater ($p \leq 0.001$) in plants treated with Zn + P; however, inoculated plants (Zn + P + R_p, and Zn + P + R_L) recorded the lowest intercellular CO_2 concentration (Figure 4D). Intercellular CO_2 concentration decreased in genotype ICCV3110 that was treated with a combination of Zn + P, Zn + P + R_p, and Zn + P + R_L when compared to sole application and control treatments. Similarly, sole Zn application and control treatments increased intercellular CO_2 concentration for genotypes ICCV3110, ICCV92944, and ICCV8101, while Zn + P application reduced intercellular CO_2 concentration for the same genotypes.

The water-use efficiency among the genotypes showed an inverse trend with intercellular CO_2 concentration, with the addition of Zn + P + R_p and Zn + P + R_L improving intrinsic water-use efficiency in genotypes ICCV97024 and ICCV3110 (Figure 4E). Additionally, genotype ICCV92944 treated with Zn + P, as well as genotype ICCV8101 treated with sole P, Zn + P, and Zn + P + R_p recorded the highest intrinsic water-use efficiency.

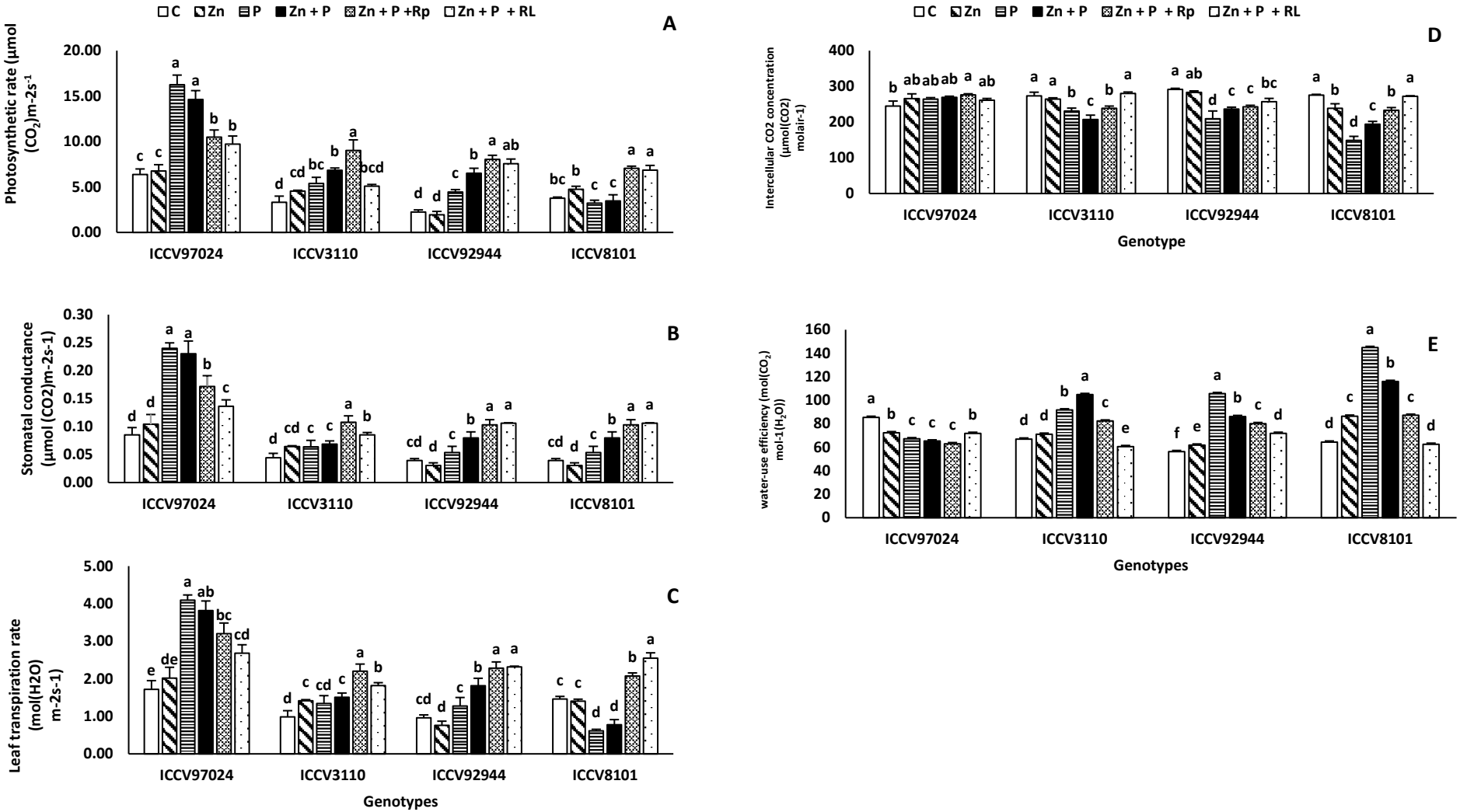


Figure 3: The interactive effects of genotype x fertilizer applications on A) photosynthetic rates B) stomatal conductance C) transpiration D) leaf internal CO_2 concentration and E) intrinsic water-use efficiency of chickpea grown in silty-clay-loam soils collected from the field under glasshouse conditions. For each parameter, bars with dissimilar alphabets in a row are significantly different at $p \leq 0.05$. Error bars represent SE. Zn + P + R_P represents Zinc plus P plus peat-based inoculant, while Zn + P + R_L represents Zinc plus P plus liquid-based inoculant.

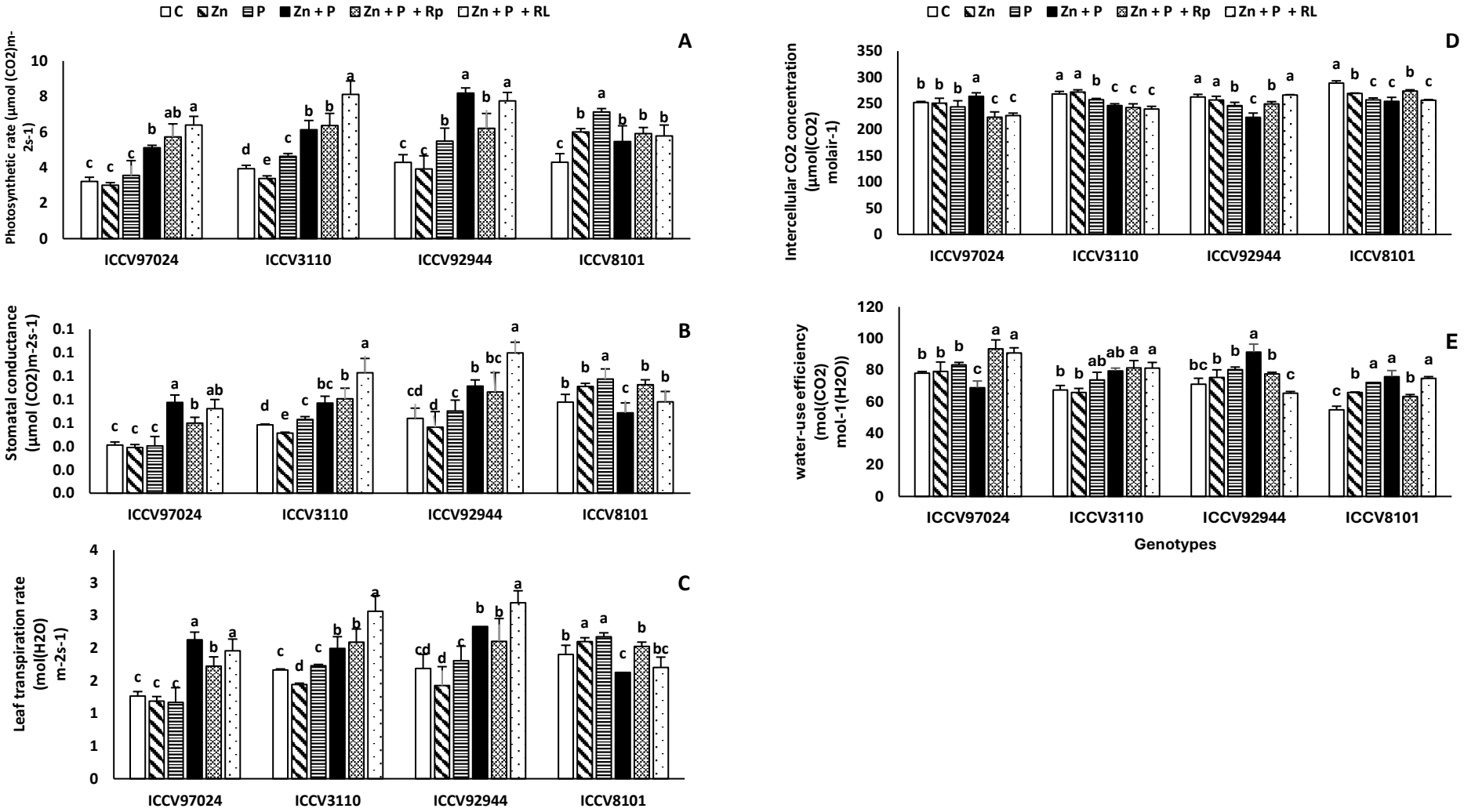


Figure 4: The interactive effects of genotype x fertilizer applications on A) photosynthetic rates B) stomatal conductance C) transpiration D) leaf internal CO_2 concentration and E) intrinsic water-use efficiency of chickpea grown in silty-loam soils collected from the field under glasshouse conditions. For each parameter, bars with dissimilar alphabets in a row are significantly different at $p \leq 0.05$. Error bars represent SE. Zn + P + R_P represents Zinc plus P plus peat-based inoculant, while Zn + P + R_L represents Zinc plus P plus liquid-based inoculant.

4. Discussion

4.1. Genotypic variation in biomass accumulation of chickpea

Of the four genotypes evaluated in this study, ICCV92944 exhibited the highest shoot and whole-plant dry weight, while ICCV97024 was among those that had the least (Table 1). A significantly higher shoot dry weight has been shown by ICCV92944 when evaluated in experiments that were conducted under field and controlled conditions (Meshram, 2014; Moloto, 2019; Motseo, 2019). Given that genotype ICCV92944 showed a larger root system, this indicates that the roots were able to absorb and supply sufficient quantities of water and nutrients to the aboveground organs of plants (shoots). This is the case because the literature background indicates that there is a direct and positive relationship between the biomass of roots and that of shoots (Bektas *et al.*, 2023; Ding *et al.*, 2025). In this study, it is possible that the larger root biomass of ICCV92944 created a conducive environment for absorption of sufficient nutrients which, when translocated to the aboveground organs, resulted in markedly greater shoot and whole-plant dry weights. In contrast, genotype ICCV8101 exhibited the highest root dry weight and root: shoot ratio but produced relatively low shoot dry weight when grown in both soils. In general, most plant species show a higher root: shoot ratio when grown or adapted to soils that contain lower concentrations of nutrients than those required for optimum growth and development (Bektas *et al.*, 2023; Cabeza *et al.*, 2024; Ding *et al.*, 2025). However, in this study, the soils had contrasting fertility, one with relatively lower levels of nutrients and the other with relatively higher concentrations of nutrients. Therefore, the high root: shoot ratio of ICCV8101 genotype across both soil types suggests that the genotype possesses an inherent biomass allocation strategy favouring root growth.

4.2. Effect of fertilizer and inoculant application on photosynthetic parameters of chickpea genotypes planted under glasshouse conditions

Applying P alone, Zn + P, and Zn + P + R, promoted the highest and second-highest root, shoot, and whole-plant dry weights of chickpea in silty-clay-loam and silty-loam soils (Table 1). Apart from the native or residual P in the soil, the chickpea plants supplied with P fertilizer had adequate soil solution P that ensured that their growth was supported largely through increased availability of Ca, Mg, and S, stimulating the exudation of organic acids, and enhancing the plant's ability to mobilize P in the rhizosphere (Zhao *et al.*, 2025). Furthermore, although rhizospheric soil pH was not determined in this study, the application of phosphate fertilizer alters (largely decreases) the soil pH in the rhizosphere (Nyoki & Ndakidemi, 2018). Through lowering rhizospheric pH, the solubility and desorption of P from substances in the rhizosphere are enhanced (Penn & Camberato, 2019). Therefore, with increased solubility and reduced adsorption, P concentration and accumulation would increase in the rhizosphere of soil supplied with P fertilizer.

As an important micronutrient, for optimal growth, plants require the application of Zn fertilizers, which, when supplied adequately, enhance the secretion and concentrations of the growth-promoting hormones, gibberellins (Hamzah Saleem *et al.*, 2022). The positive effect that the application of P, Zn, and Zn plus P has on the growth of chickpea is known (Balai *et al.*, 2017). On the other hand, the role of rhizobial inoculants not only promotes plant growth through symbiotic N_2 fixation but also produces plant growth regulators that stimulate root development, solubilize nutrients, and improve rhizosphere nutrient availability for better uptake by crops (Fahde *et al.*, 2023; Prasad *et al.*, 2026). Through the utilization of these and other mechanisms, inoculants exert positive effects on the growth and yield of crops, especially in soils containing ineffective rhizobia. Given the roles of P, Zn, and P plus rhizobial inoculation, their co-application would understandably increase the growth of the test chickpea genotypes, even when grown in soils with different textures.

4.3. Comparison of plant growth related parameters in chickpea genotypes grown in silt-clay-loam and loam-soil under glasshouse conditions

Of the two soil textures, chickpea planted in silty-clay-loam soil showed the largest root, shoot, and whole-plant biomass (Table 3). Silty-clay-loam soil contained higher concentrations of organic carbon and nitrate-N. Indeed, a high concentration of N, especially in the topsoil, increases the branching or proliferation and therefore increase in biomass production (Luo *et al.*, 2020). Generally, soils with high organic matter promote root growth through improved soil structure, microbial activity, and nutrient availability, which provide a favorable environment (Pisarčik *et al.*, 2024). Therefore, the markedly increased in biomass production could have been supported by the relatively greater concentrations of nitrate and organic carbon in the silt-clay-loam soil. Apart from the greater levels of organic carbon and nitrate-N, the silty-clay-loam soil also recorded higher concentrations of potassium, sodium, magnesium, calcium, sulphur, and boron, an indication that this soil had higher fertility. In contrast, the larger root biomass by chickpea in the silt-clay-loam soil could be a result of the fact that the soil did not contain sufficient concentrations of nutrients that were readily available for uptake by the chickpea. This could have been because, although the silty-clay-loam soil had high concentrations of nutrients, their movement and solubility could have been lower, which is typical of soils that have a higher cation exchange capacity. Typically, plants that grown in soils that contain low concentrations of plant-available nutrients reveal high root biomass. In fact, such plants prioritize the growth of roots (organs) that are involved in acquiring the scarce mineral nutrients) over aboveground organs (Marschner *et al.*, 1996).

4.4. Genotypic variation in photosynthetic performance and water-use efficiency of chickpea plants

The study showed that there are significant differences in the photosynthetic performance of the selected chickpea genotypes. Even with contrasts in trends, there are variations in the performance of genotypes. In fact, these are reportedly a result of genetic differences in their make-up, and other studies have shown that there exist differences in their morphology and accumulation of nutrients (Flood *et al*., 2011;Mpai & Maseko, 2018). Importantly, Devi *et al.* (2022) and Tiwari *et al.* (2023) showed that indeed chickpea genotypes do reveal significant differences in their photosynthetic performance.

The study revealed contrasting results on the leaf gas-exchange parameters. When grown in the silty-clay-loam soil, ICCV97024 recorded the highest photosynthetic rate, stomatal conductance, transpiration rate, and intercellular CO_2 concentration. However, when established in the silty-loam soil, both ICCV8101 and ICCV92944 recorded the highest photosynthetic rate, stomatal conductance, and transpiration rate, while ICCV97024 recorded the lowest (Table 2). About the contrasting photosynthetic rates between the types of soil, this is interesting because when using the same soil but supplied Kelpak and Bontera, (Moloto, 2019) found that ICCV92944 exhibited the highest photosynthetic rates when grown in both types of soil. Interestingly, ICCV8101 formed the largest root biomass in both soil textures, while ICCV92944 exhibited the highest in the silt-clay-loam soil (Table 1). Also, ICCV92944 exhibited the highest shoot and whole-plant dry weights when established in both types of soil, while ICCV97024 showed the least (Table 1). The fact that ICCV92944 exhibited larger growth and photosynthetic rate when grown in the silty-loam agrees with the background that an increase in the photosynthetic rate is associated with high growth or shoot biomass. Whatever the case, this study confirms the existence of factors that cause the differences in the photosynthetic performance of the desi genotypes.

4.5. Effect of fertilizer and inoculant on photosynthetic parameters of chickpea genotypes

When chickpea was planted in either soil type, the application of P alone, and in combination with Zn and rhizobia inoculant, significantly increased ($p \leq 0.05$) leaf gas exchange parameters compared to Zn alone and the control plants (Table 2, Figures 3 and 4). The external P supply could have stimulated net photosynthesis by changing the photosynthetic apparatus and CO_2 assimilation capacity, which in turn enhances the photosynthetic rate (carbon fixation) (Chtouki *et al.*, 2022). In addition, the application of P increases the availability and uptake of other macronutrients, including Ca, Mg, and S, by promoting root growth and facilitating nutrient interaction within the soil-plant system (Kumar *et al.*, 2021). Thus, the combination of P with all these nutrients enhances the photosynthetic rate and overall photosynthesis. Moreover, rhizobia inoculation, especially in soil with ineffective or less competitive native rhizobia, can maximize the solubility and uptake of plant nutrients (particularly N, P, Fe, and K) (Jaiswal & Dakora, 2025), These nutrients play a significant role in the photosynthetic rate of plants. Lastly, the application of optimal rates of Zn increases the chlorophyll content and photosynthetic rate of plants (Dang *et al.*, 2024). Taken together, it is not surprising that the combined application of the test nutrients coupled with rhizobial inoculation markedly increased the photosynthetic rate of chickpea grown in both silty-loam and silty-clay-loam soil in this study.

The application of P, as well as P plus Zn alone and with rhizobial inoculation, promoted higher intrinsic water-use efficiency in chickpea (Table 2). Concerning the role of P fertilizer, other studies have indicated that fertilization with P helps in osmotic adjustment, stomatal regulation, and enhances the photosynthetic rate, water retention in plants, and increases water-use efficiency (Khan *et al.*, 2023). Furthermore, it maintains net photosynthesis due to increased stomatal conductance promoted by P under conditions of water restriction. Rhizobial strains produce lumichrome, which decreases leaf stomatal conductance and reduces water loss via transpiration in the leaves, thereby increasing the water-use efficiency in plants (Phillips *et al.*, 1999; Dakora, 2003; Matiru & Dakora, 2005) . The decrease in stomatal conductance and increase in water-use efficiency from *Rhizobium* inoculation occur because of the effect of phytohormones (Matiru & Dakora, 2005).

4.6. **Comparison of photosynthetic performance among the two soil types**

Chickpea grown in the silty-loam soil exhibited the highest transpiration rate, while the other leaf gas exchange parameters were the lowest (Table 3). Given that transpirational water fluxes contribute to the movement of nutrients and/or their acquisition, especially by soil-grown plants, therefore, it is expected that the transpiration rate of plants established in low-nutrient soils is increased (Cramer *et al.*, 2009; Matimati *et al.*, 2014). For these soils, the silty-loam contained relatively lower concentrations of most of the selected nutrients, including nitrate-N, K, Na, Mg, Ca, S, and B. The reasons for these are multi-fold and include the reduction in the pool size of ribulose-1,5-bisphosphate and the decline in the content of photosynthetic pigments (Singh et al., 2018; Hantzis et al*.,* 2018). In contrast to the lower transpiration in plants established in the silty-loam soil, the markedly higher photosynthetic rate, stomatal conductance, and water-use efficiency in chickpea grown in the silty-clay-loam soil were supported by the higher organic matter, nitrate-N, K, S, and other nutrients (Xu *et al.*, 2024; Cao *et al.*, 2025).

5. Conclusion

There data revealed a significant difference in shoot biomass accumulation and photosynthetic performance in the four chickpea genotypes and fertilizer treatments, whether grown in silty-loam or silty-clay-loam soil. Genotype ICCV8101 exhibited the highest root biomass and root: shoot ratio in both soil types. For aboveground biomass, ICCV92944 had the highest shoot and whole-plant dry weight, while shoot and whole-plant biomass were greater when grown in the silty-clay-loam soil. Regarding photosynthetic performance, when chickpea was grown in silty-clay-loam soil, ICCV97024 recorded the highest photosynthetic rate, stomatal conductance, transpiration rate, and intercellular CO_2 concentration. However, when established in the silty-loam soil, both

ICCV8101 and ICCV92944 recorded the highest photosynthetic rate, stomatal conductance, and transpiration rate, while ICCV97024 recorded the lowest. Application of P, Zn + P alone, and with the rhizobial inoculants generally increased belowground and aboveground biomass, photosynthetic rate, transpiration rate, stomatal conductance, and intrinsic water-use efficiency Lastly, the selected growth-related parameters and gas-exchange parameters were markedly greater in chickpea grown in the silty-clay-loam soil. Our results show promise and we recommend further glasshouse and field experiments are required to explore the full potentials of the chickpea genotypes tested in the study.

6. Author Contributions

Conceptualization, S.T.M.; methodology, P.M.N. and S.T.M., software, S.T.M., validation, S.T.M. and P.S.; formal analysis, P.M.N. and S.T.M.and M.T., investigation, P.M.N. and S.T.M., resources, P.S., data curation, S.T.M., writing—original draft preparation, S.T.M., T.YN and M.T.M, writing—review and editing, S.T.M. and M.T.M., visualization, S.T.M. and P.S., supervision, S.T.M. and P.S., project administration, S.T.M. and P.S. All authors have read and agreed to the published version of the manuscript.

7. Data Availability Statement:

The data that supports the findings of this study are not publicly available. However, anonymized data supporting the findings of this study are available from the corresponding author upon reasonable request.

8. **Funding:** This research received no external funding.

9. Acknowledgments

The authors thank the Tshwane University of Technology for financial support towards this research.

10. Plant Ethics declaration

The crop species used in this study were identified and verified by qualified agronomists and researchers from the Department of Agriculture and Rural Development, Limpopo Province, South Africa who routinely manage and evaluate these crop species within national agricultural research programs. These chickpea genotypes were identified and supplied by Mr. David Lingwarti who is an agronomist specialist at the Department of Agriculture and Rural Development, Limpopo Province, South Africa.

10.1. Plant genotypes identified

The crop species used in the experimental trial in this study included four desi-type chickpea genotypes (*Cicer arietinum* L.), namely ICCV97024, ICCV92944, ICCV3110, and ICCV8101. These genotypes were identified and supplied by Mr. David Lingwarti who is an agronomist specialist at the Department of Agriculture and Rural Development, Limpopo Province, South Africa.

10.2. Permission

The soil sample used for the experiments were collected from the Tshwane University of Technology experimental farm, Pretoria, Permission was obtained from the farm manager and the department of crop sciences, Tshwane University of Technology, South Africa. The chickpea genotypes used in this study were obtained from the Department of Agriculture and Rural Development, Limpopo Province, South Africa and were supplied by Mr. David Lingwarti.

10.3. Source of plant materials

The study did not involve the collection of wild plants, protected species, endangered species, or herbarium specimens. All seeds used in the experiments were cultivated desi-type chickpea genotypes and were supplied by Mr. David Lingwarti (Agronomist) at the Department of Agriculture and Rural Development, Limpopo Province, South Africa.

Plant guidelines and compliance

The plant materials used in this study were cultivated agricultural crops commonly grown in South Africa and sourced through recognized national agricultural research programs. The study complied with national agricultural research regulations and guidelines in South Africa. No permits or licences were required because the research involved cultivated crop varieties used under normal agricultural research and extension activities. The study did not involve the collection of plant materials from protected areas or natural ecosystems.

11. Declaration of Computing.

This manuscript is original unpublished work, read and approved by all authors, and not being considered for publication elsewhere. All those listed as authors have contributed to the development of this paper and have approved of the manuscript and this submission. We have no conflicts of interest to declare.

12 Consent for publication

Not applicable.

13 Consent to Participate:

Consent to Participate

Informed consent was obtained from all individual participants involved in the study prior to data collection.

.

Reference

BALAI, K., JAJORIA, M., VERMA, R., DEEWAN, P. & BAIRWA, S. 2017. Nutrient content, uptake, quality of chickpea and fertility status of soil as influenced by fertilization of phosphorus and zinc. *Journal of Pharmacognosy and Phytochemistry,* 6**,** 392–398.

BEKTAS, H., HOHN, C. E., LUKASZEWSKI, A. J. & WAINES, J. G. 2023. On the possible trade-off between shoot and root biomass in wheat. *Plants,* 12**,** 2513.

BUNCE, J.A., 2016. Responses of soybeans and wheat to elevated CO2 in free-air and open top chamber systems. *Field crops research*, *186*, pp.78-85.

CABEZA, A., CASAS, A. M., LARRUY, B., COSTAR, M. A., MARTÍNEZ, V., CONTRERAS-MOREIRA, B. & IGARTUA, E. 2024. Genetic control of root/shoot biomass partitioning in barley seedlings. *Frontiers in Plant Science,* Volume 15 - 2024.

CAO, W., SUN, H., SHAO, C., WANG, Y., ZHU, J., LONG, H., GENG, X. & ZHANG, Y. 2025. Progress in the Study of Plant Nitrogen and Potassium Nutrition and Their Interaction Mechanisms. *Horticulturae,* 11**,** 930.

CHTOUKI, M., LAAZIZ, F., NACIRI, R., GARRÉ, S., NGUYEN, F. & OUKARROUM, A. 2022. Interactive effect of soil moisture content and phosphorus fertilizer form on chickpea growth, photosynthesis, and nutrient uptake. *Scientific Reports,* 12**,** 6671.

CRAMER, M. D., HAWKINS, H.-J. & VERBOOM, G. A. 2009. The importance of nutritional regulation of plant water flux. *Oecologia,* 161**,** 15–24.

DAKORA, F. D. 2003. Defining new roles for plant and rhizobial molecules in sole and mixed plant cultures involving symbiotic legumes. *New Phytologist,* 158**,** 39–49.

DANG, K., MU, J., TIAN, H., GAO, D., ZHOU, H., GUO, L., SHAO, X., GENG, Y. & ZHANG, Q. 2024. Zinc regulation of chlorophyll fluorescence and carbohydrate metabolism in saline-sodic stressed rice seedlings. *BMC Plant Biology,* 24**,** 464.

DEVI, P., JHA, U. C., PRAKASH, V., KUMAR, S., PARIDA, S. K., PAUL, P. J., PRASAD, P. V. V., SHARMA, K. D., SIDDIQUE, K. H. M. & NAYYAR, H. 2022. Response of Physiological, Reproductive Function and Yield Traits in Cultivated Chickpea (Cicer arietinum L.) Under Heat Stress. *Frontiers in Plant Science,* Volume 13 - 2022.

DING, R., NÓBREGA, R. L. B. & PRENTICE, I. C. 2025. Global Assessment of Environmental and Plant-Trait Influences on Root: Shoot Biomass Ratios. *Global Change Biology,* 31**,** e70543.

FAHDE, S., BOUGHRIBIL, S., SIJILMASSI, B. & AMRI, A. 2023. Rhizobia: A Promising Source of Plant Growth-Promoting Molecules and Their Non-Legume Interactions: Examining Applications and Mechanisms. *Agriculture,* 13**,** 1279.

HABINSHUTI, S., MASEKO, S. & DAKORA, F. 2015. Effect of N-fertilizer application on the symbiotic N2 fixation by field-grown common bean (Phaseolus vulgaris L.) in Eastern Cape Province.

HAMZAH SALEEM, M., USMAN, K., RIZWAN, M., AL JABRI, H. & ALSAFRAN, M. 2022. Functions and strategies for enhancing zinc availability in plants for sustainable agriculture. *Frontiers in Plant Science,* Volume 13 - 2022.

ILLAKWAHHI, D. T., VEGI, M. R. & SRIVASTAVA, B. B. L. 2024. Phosphorus' future insecurity, the horror of depletion, and sustainability measures. *International Journal of Environmental Science and Technology,* 21**,** 9265–9280.

JAISWAL, S. K. & DAKORA, F. D. 2025. Maximizing Photosynthesis and Plant Growth in African Legumes Through Rhizobial Partnerships: The Road Behind and Ahead. *Microorganisms,* 13**,** 581.

KHAN, F., SIDDIQUE, A. B., SHABALA, S., ZHOU, M. & ZHAO, C. 2023. Phosphorus Plays Key Roles in Regulating Plants' Physiological Responses to Abiotic Stresses. *Plants (Basel),* 12.

KUMAR, S., KUMAR, S. & MOHAPATRA, T. 2021. Interaction Between Macro- and Micro-Nutrients in Plants. *Frontiers in Plant Science,* Volume 12 - 2021.

LAZALI, M. & DREVON, J. J. 2018. Role of acid phosphatase in the tolerance of the rhizobial symbiosis with legumes to phosphorus deficiency: Lazali M., Drevon JJ. *Symbiosis,* 76**,** 221–228.

MSIZA, L.J., NGMENZUMA, T.Y., MOHAMMED, M. AND DAKORA, F.D., 2026. Water-Use Efficiency and Mineral Nutrition of Diverse Legume Species Nodulated by Different Native Rhizobial Isolates: Do Rhizobia Have a Say in the Mineral Nutrition of Their Host Plants?. *Plants*, *15*(10), p.1478.

LUO, L., MA, Y., ZHANG, S., WEI, D. & ZHU, Y.-G. 2009. An inventory of trace element inputs to agricultural soils in China. *Journal of Environmental Management,* 90**,** 2524–2530.
LUO, L., ZHANG, Y. & XU, G. 2020. How does nitrogen shape plant architecture? *Journal of experimental botany,* 71**,** 4415–4427.
MADZIVHANDILA, T., OGOLA, J. & ODHIAMBO, J. 2012. Growth and yield response of four chickpea cultivars to phosphorus fertilizer rates. *J Food Agric Environ,* 10**,** 451–455.
MARSCHNER, H., KIRKBY, E. & CAKMAK, I. 1996. Effect of mineral nutritional status on shoot-root partitioning of photoassimilates and cycling of mineral nutrients. *Journal of experimental botany,* 47**,** 1255–1263.
MASEKO, S. T., NONG, P. M. & SOUNDY, P. 2026. Assessing Nutrient Accumulation in Chickpea (Cicer arietinum L.) Genotypes Grown in Soils with Different Texture: Response to Application of P and Zn Fertilizers, and Rhizobial Inoculant. *Horticulturae,* 12**,** 553.
MATIMATI, I., VERBOOM, G. A. & CRAMER, M. D. 2014. Nitrogen regulation of transpiration controls mass-flow acquisition of nutrients. *Journal of Experimental Botany,* 65**,** 159–168.
MATIRU, V. & DAKORA, F. 2005. Xylem transport and shoot accumulation of lumichrome, a newly recognized rhizobial signal, alters root respiration, stomatal conductance, leaf transpiration and photosynthetic rates in legumes and cereals. *New phytologist,* 165**,** 847–855.
MESHRAM, K. 2014. *Evaluation of chickpea (Cicer arietinum L.) genotypes for late sown high temperature conditions.* Jawaharlal Nehru Krishi Vishwa Vidyalaya Jabalpur, India.
MOHALE, K. C., BELANE, A. K. & DAKORA, F. D. 2014. Symbiotic N nutrition, C assimilation, and plant water use efficiency in Bambara groundnut (Vigna subterranea L. Verdc) grown in farmers' fields in South Africa, measured using 15N and 13C natural abundance. *Biology and fertility of soils,* 50**,** 307–319.
MOLOTO, R. M. 2019. *Effect of biofertilizer and soil texture on the growth, photosynthesis, and phosphatase activity of chickpea genotypes.* M- Tech, Tshwane University of Technology.
MOTSEO, N. M. 2019. Effect of phosphorus and biofertilizers on the growth, grain yield and rhizosphere mineral accumulation in chickpea genotypes grown at Klipplaatdrift and Bon Accord, South Africa. *MTech diss., Tshwane University of Technology*.
MPAI, T. & MASEKO, S. T. 2018. Possible benefits and challenges associated with production of chickpea in inland South Africa. *Acta Agriculturae Scandinavica, Section B—Soil & Plant Science,* 68**,** 479–488.
MUTHABI, A. 2020. *The response of symbiotic performance, growth and yield of chickpea (Cicer arietinum L.) genotypes to phosphorus fertilizer rates and rhizobial inoculation.*
NEMAEMBENI, P. M. 2023. *Selection of Efficient Indigenous Rhizobia Inoculants for the Production of Selected Tropical Legumes in Limpopo Province (South Africa).*
NYOKI, D. & NDAKIDEMI, P. A. 2018. Selected chemical properties of soybean rhizosphere soil as influenced by cropping systems, rhizobium inoculation, and the supply of phosphorus and potassium after two consecutive cropping seasons. *International Journal of Agronomy,* 2018**,** 3426571.
OGOLA, J. B., MADZIVHANDILA, V., MASEKO, S. T. & LEBOHO, T. M. 2023. P-fertiliser and rhizobial inoculation increased the concentration of mineral nutrients in the rhizosphere of two chickpea genotypes. *Acta Agriculturae Scandinavica, Section B—Soil & Plant Science,* 73**,** 94–101.
OLOO, K. P. & ASBON, O. P. 2020. Depletion of phosphate rock reserves and world food crisis: Reality or hoax? *African Journal of Agricultural Research,* 16**,** 1223–1227.

PENN, C. J. & CAMBERATO, J. J. 2019. A critical review on soil chemical processes that control how soil pH affects phosphorus availability to plants. *Agriculture,* 9**,** 120.

PHILLIPS, D. A., JOSEPH, C. M., YANG, G.-P., MARTÍNEZ-ROMERO, E., SANBORN, J. R. & VOLPIN, H. 1999. Identification of lumichrome as a Sinorhizobium enhancer of alfalfa root respiration and shoot growth. *Proceedings of the national academy of sciences,* 96**,** 12275–12280.

PISARČIK, M., HAKL, J., TOLEIKIENE, M., FUKSA, P., RASMUSSEN, J. & HOOD-NOWOTNY, R. 2024. Role of cover crop roots in soil organic carbon accrual—A review. *European Journal of Soil Science,* 75**,** e13532.

PRASAD, B., KUMAR, P. & DUBEY, R. C. 2026. Advances in rhizobial technology: driving sustainable agriculture in the 21 st century. *Archives of Microbiology,* 208**,** 81.

TIWARI, P. N., TIWARI, S., SAPRE, S., BABBAR, A., TRIPATHI, N., TIWARI, S. & TRIPATHI, M. K. 2023. Screening and selection of drought-tolerant high-yielding chickpea genotypes based on physio-biochemical selection indices and yield trials. *Life,* 13**,** 1405.

WIELBO, J. 2021. Recent Advances in Biological Nitrogen Fixation. *Agronomy,* 11**,** 1941.

XU, X., ZHANG, X., NI, W., LIU, C., QIN, H., GUAN, Y., LIU, J., FENG, Z., XING, Y., TIAN, G., ZHU, Z., GE, S. & JIANG, Y. 2024. Nitrogen–potassium balance improves leaf photosynthetic capacity by regulating leaf nitrogen allocation in apple. *Horticulture Research,* 11.

ZHAO, W., ZHANG, Y., XIE, X., LI, H. & ZHANG, J. 2025. Organic acid-mediated phosphorus mobilization in black soils: differential effects of maize root exudates on alfisols and mollisols in Northeast China. *Plos one,* 20**,** e0333230.