Application of Phosphorus and Calcium to Improve Physiological Response of Groundnut 
\textit{(Arachis hypogaea L.)}

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Authors’ contributions
This work was carried out in collaboration among all authors. Authors HTN and CLR designed the study, performed the statistical analysis. Author HTN wrote the protocol and wrote the first draft of the manuscript. Authors CLR and JSF managed the analyses of the study. Author WSK managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT
Genotype, environment and agronomic practices are key determinants of crop growth and productivity which are important to provide food, feed, raw materials and income to small holder farmers in Africa. The study was conducted at Crop museum, Sokoine University of Agriculture, Morogoro, Tanzania to investigate physiological response of groundnut to calcium and phosphorus nutrition among three improved groundnut genotypes \textit{Mangaka} \textit{(G\textsubscript{1})}, \textit{Masasi} \textit{(G\textsubscript{2})} and \textit{Pendo} \textit{(G\textsubscript{3})} with three levels of phosphorus and at calcium control \textit{(T\textsubscript{0})}, 125 kg/ha \textit{(T\textsubscript{1})} and 55 kg/ha \textit{(T\textsubscript{2})} supplied from Diammonium Phosphate (DAP) and \textit{Minjingu mazao}, respectively. Significant (P<0.05) influence of calcium was observed on leaf area index (LAI) 4.03; crop biomass 88.79 g/plant; number of nodules 66.22, and crop growth rate (CGR) 15.05 g m\textsuperscript{-2}/day. Whereas phosphorus had
significant influence on net assimilation rate (NAR) 10.84 g m$^{-2}$/day. Similarly, groundnut genotype (Masasi) had significant influence on LAI (3.95); CGR (13.04 g m$^{-2}$/day); NAR (12.36 g m$^{-2}$/day) and number of nodes (64.93). However, there was no significant effect of genotype on crop biomass recorded from the investigation. Growth parameters were significant influenced by genotype and fertilizer interactions with Significant (P=0.2) interaction effect for crop biomass observed between G$_3$ x T$_1$, while G$_2$ x T$_2$ significantly affected number of nodules and G$_2$ x T$_1$ had Significant interaction effects on CGR and NAR. This is an indication that investment in improved genotype and appropriate application rate of fertilizers has the potential to enhance yield and income of smallholder farmers.

Keywords: Calcium; genotypes; groundnut; phosphorus; physiology.

1. INTRODUCTION

Groundnut (Arachis hypogaea L.) is an important global multi-purpose legume crop that reinforces livelihood strategies meeting food, nutrition, and income security [1]. It is an essential source of protein, calories, essential fatty acids, vitamins, and minerals associated with several health benefits for human, and raw materials for industrial use including products such as food, feed, fuel, paints, lubricants and insecticides and can be used as an ideal crop in rotational systems to improve soil fertility due to its natural ability to fix atmospheric nitrogen [1-6].

However, yield levels of the crop has remained relatively low (964 kg/ha) in Sub Saharan Africa (SSA) far less than potential yields of up to 3500 kg/ha reported elsewhere due to a range of biotic, abiotic and socio-economic constraints including lack of access to quality seeds, poor agronomic practices such as low use of fertilizers and limited access to extension and advisory services [4,7-11].

In Tanzania, groundnut serves as food, feed and raw material for industries, but farmers have limited access to improved genotypes and fertilizer resources due to cost and lack of education on new agronomic practices , thus major limiting production, especially where there are no recommended rates of fertilizers for groundnut whose production is characterized by low external input, limited extension and advisory services, erratic rainfall, hence fluctuating yields despite the economic importance of the crop [6,10,12].

Lack of access to improved groundnut genotype in Tanzania poses numerous challenges to farmers ranging from poor crop performance, crop quality and subsequently low income at household level. Selection of improved genotypes with superior characteristic under good agronomic practices has been highlighted as key to sustainable production Daudi et al. [10].

Sustainable groundnut production depends on proper selection of variety, fertilizer management and other management practices as optimal rates of fertilizer have positive effect on the performance of groundnut [13, 6]. Applications of micro and macronutrients are effective for increase of crop production. These had capability to mitigate negative effects of environmental stresses [14, 15]. Numerous crop production was improved by balance fertilization [16].

The objective of the study was to evaluate the effect of fertilizer on growth attributes of groundnut and provide information on optimal fertilizer rates for groundnut genotypes with superior performance, hence improved production.

2. MATERIALS AND METHODS

2.1 Experimental Site

Field experiment was laid out at Sokoine University of Agriculture (SUA), Crop Museum situated at latitude 6° 45’ South and longitude 37° 40’ East at 525 m.a.s.l in Morogoro municipality. The area is situated at the foot of Uluguru Mountain in Morogoro and has a bi-modal rainfall pattern, predominantly sub-humid with irregular and unreliable distribution characterized by kaolinitic clay soils, which are well drained and mostly clay [17,18].

2.2 Experimental Design

Three groundnut varieties (Mangaka, Masasi and Pendo) used in the experiment were obtained from Naliedendele Agricultural Research Institute.
(ARI), in Mtwara, Tanzania whereas fertilizer materials included Minjingu Mazao, source of calcium contained N (10%), 20% P$_2$O$_5$, 25% CaO, 5% S, 0.5% Zn, 1.5% MgO and 0.1% Boron [19] and Diammonium Phosphate (DAP), source of phosphorus (NH$_4$)$_2$ (HPO$_4$) was composed of 18% N and 46% P$_2$O$_5$.

The experiment was laid out in a split plot Randomized Complete Block Design (RCBD) with four replications and three groundnut genotypes, G$_1$ = (Mangaka), G$_2$ = (Masasi), G$_3$ = (Pendo) as main plot factor (factor A) whereas fertilizer types were applied as sub-plot factor (factor B). Fertilizer types were applied as follows: T$_0$ = (Control), T$_1$ = (55 kg P/ha) T$_2$ = (125 kg Ca/ha).

2.3 Soil Analyses

Soil samples were collected as recommended by Landon [20] at the depth of 0–30cm for physicochemical analyses. Soil pH was determined electrometrically in 1:2.5 soil–water suspensions as described by Thomas [21]. Available P was analyzed using Bray –1 [22]. Organic carbon determination was done by wet digestion method of Walkley and Black [23]. Total N was determined by the micro – Kjeldahl digestion–distillation method [24]. Exchangeable bases, K and Na were analyzed by flame photometer whereas Ca and Mg were determined by Atomic Absorption Spectrophotometer (AAS) as described by Petersen [25]. Available Cu, Zn, Fe and Mn were extracted by DTPA as described by Lindsay et al. [26].

Soil physicochemical properties were done by the Department of Soil and Geological Sciences at Sokoine University of Agriculture. Soil physicochemical properties are presented in Table 1. Agronomic and other management practices were carried out as described by Kanyeka et al. [27].

2.4 Data Collection

Data on growth characteristics of groundnut was done at 4, 6, 8 and 10 Weeks After Planting (WAP).

Crop biomass (g/plant): Crop Biomass (BM) was determined from three plants by harvesting a whole plant including roots from the penultimate rows of the plot. The harvested plants were oven dried at 70 °C for 48 hours and the dry weights were recorded using (Doran 7000, Doran Inc.) electronic weighing scale

Leaf area index: Leaf area index (LAI) was calculated as described by Brown [28].

\[
LAI = \text{leaf area per plant (cm}^2\text{)} / \text{Ground cover per plant (cm}^2\text{)}
\]

Crop growth rate (g.m$^2$/day): crop growth rate (CGR) at different growth stages was determined as recommended by Brown (1984) as follow:

\[
CGR = \frac{W_2 - W_1}{T_2 - T_1} \times \frac{1}{GA} (\text{g m}^{-2} \text{day}^{-1})
\]

Where:

\[
W_1 = \text{Weight at } T_1 \text{ of the period} \\
W_2 = \text{Weight at } T_2 \text{ of the Period} \\
T_1 = \text{Time in date at the start of the period} \\
T_2 = \text{Time in the date at the end of the period} \\
GA = \text{Ground area}
\]

Net assimilation rate (g m$^2$/day): NAR was determined by assessing the ratio of CGR and LAI at various growth stages. Net assimilation rate was calculated as described by Brown [28].

\[
NAR = \frac{1}{LAI} \times \frac{\text{dw/dt}}{\text{day}} = \text{g m}^2/\text{day}
\]

Where:

\[
NAR = \text{net assimilation rate dw/dt = the change in plant dry matter per unit time} \\
LAI = \text{leaf area index}
\]

Number of nodules per plant: number of nodules was determined by counting the number of nodes visible on the roots of three plants per plot and the average number of nodules was recorded.

2.5 Data Analysis

Data collected were subjected to analysis of variance (ANOVA) using GENSTAT released version 14th edition and declared significant at P < 0.05 using the following statistical model described by Gomez and Gomez [29]. Mean separation test was done using Duncan Multiple Range Test (DMRT) at P≤ 0.05.
3. RESULTS AND DISCUSSION

3.1 Soil Characteristics

Soil analyses in the current study revealed that soil textural (sandy clay loam); bulk density (1.2g/cm$^3$) and soil pH (5.9) were considered appropriate for root penetration, moisture retention, nodule formation, hence increased growth, development and production [30-32]. Nkot et al. [33] reported poor nodule formation in soil with pH between 3.5 and 5.0 had adverse effect on root penetration, nodule formation, nutrient availability.

A very low status of available P (0.048 mg/kg), as observed in the current study was evident that P was a major limiting factor for groundnut production in the study area. Phosphorus deficiency constitutes a serious limitation to crop production in weathered tropical soils containing high Fe and Al oxides that quickly fix added P. Moazed et al. [34] reported that low quantities of soluble P in Oxisols limit crop production and productivity.

Soil calcium level in the current study was rated very high according to Landon [20], but was inadequate to meet crop growth requirement. High level of soil Ca is affected by amounts of organic matter resulting in the inability of the soil to hold the amount required for crop growth. Calcium availability is crucial for pod formation Calcium application also influences shelling percent, seed and pod yields in groundnut [35]. Results also revealed that macronutrient deficiencies, especially P could be a major constraint to groundnut production because initial soil P was very low for optimum yield [36].

Crop Biomass: results from the current study showed no significant influence of genotype on crop biomass however; fertilizer had significant (P=0.001) influence on biomass. The highest biomass (88.79 g/plant) was observed with application of Minjingu mazao. Also, an exponential increase in biomass was observed from week 8 to 10 due to the influence of fertilizer Fig. 1.

Leaf Area Index (LAI): Genotypic variation on LAI was recorded during the current study with Masasi having significant influence on LAI. Significant influence of calcium was observed on LAI compared to other treatments. Similar findings were reported by Habib [37], Song et al. [38].

Crop growth rate: groundnut genotype had significant influence on CGR as the lowest CGR was observed in Mangaka (11.18 g m$^2$/day) whereas the highest was observed in Masasi (12.88 g m$^2$/day). Significant influence of fertilizer type on CGR was observed and recorded in the order of Minjingu mazao > DAP > control. Such findings are in conformity with Mahpara et al. [16] who reported increased ground growth with application of macronutrients.

Net assimilation rate: Net assimilation rate was significantly influenced by genotype as Masasi recorded the highest NAR (10.56 g m$^2$/day), whereas P significantly improved NAR from 7.79 to 10.84 g m$^2$/day. Nagaraj et al. [39] attributed increased NAR in groundnut with increase in the amount of P compared to control plots.

Number of nodules: number of nodules was significantly (P=0.001) affected by application of P. The current study recorded a 29.0% increase in number of nodules with the application of P

Table 1. Soil physio-chemical characteristics at experimental site

| A. Physical (%) | Chemical Macro Nutrients / Rating$^1$ | Exchangeable Cations cmolc$^{(+)}$ | Micronutrients (mg/kg) |
|----------------|-------------------------------------|-----------------------------------|------------------------|
| Sand 49.2      | pH 5.9* Organic Carbon 0.07 - VL   | Calcium 27.3 VH                   | Iron 31.7 VH           |
| Clay 42.72     |                                     | Magnesium 186.6 VH                | Manganese 92.0 VH      |
| Silt 8.08      | Total Nitrogen 0.18 -VL Organic Matter 0.12-VL | Potassium 2.16 H                | Cooper 13.0 VH         |
| Textural Class: Sandy clay | C : N ratio 1:1.25-VL Extractable P 0.048 - L | Sodium 5.4 VH                  | Zinc 24.6 VH           |

$^1$Soil nutrient status rating as described by Landon, 1991. L = low, H= high, VL= very low, VH= very high
compared to control plots, similar observations were reported by Mouri et al. [13], Dzomeku et al. [40] with the application of P. Similarly, number of nodules significantly (P=0.001) differed among genotypes with Masasi recording the highest number of nodules (64.93) compared to the other genotypes. An observed increase in the number of nodules was recorded for all genotypes between weeks 6 and 8 with steady decline at week 10 (Fig. 2).

Fig. 1. Response of crop biomass to fertilizer treatment

Fig. 2. Effect of fertilizer on number of nodules in groundnut at various growth stages
Table 2. Effect of genotype on growth characteristics of groundnut

| Genotype | BM (g/plant) | LAI | CGR (g m$^{-2}$/d) | NAR (g m$^{-2}$/d) | Number of Nodules |
|----------|--------------|-----|-------------------|-------------------|------------------|
| G$_1$    | 75.00a       | 3.71b | 11.18a           | 8.53a             | 42.14a           |
| G$_2$    | 79.25a       | 3.95b | 13.04c           | 12.36c            | 64.93b           |
| G$_3$    | 75.01a       | 2.86a | 12.88b           | 10.56b            | 63.25b           |
| SE +     | 3.33         | 0.14  | 1.67             | 0.16              | 3.24             |
| CV P value | 4.4 0.01   | 4.60 0.01 | 5.3 0.03 | 3.1 0.001 | 5.700.001 |

*Means in the same column and factor followed by the same letter are not significantly different at P (≤ 0.05) according to Duncan Multiple Range Test. Note: BM = plant biomass (g/plant), LAI = Leaf area index; CGR: Crop growth rate (g m$^{-2}$/d,) and NAR: Net assimilation rate (g m$^{-2}$/d)

Table 3. Effect of fertilizer on growth variables of groundnut

| Treatment | BM (g/plant) | LAI | CGR (g m$^{-2}$/d) | NAR (g m$^{-2}$/d) | Number of Nodules |
|-----------|--------------|-----|-------------------|-------------------|------------------|
| T$_0$     | 63.62a       | 3.51a | 8.48a            | 7.79a             | 47.01a           |
| T$_1$     | 76.85b       | 3.52a | 13.55b           | 10.84b            | 57.08b           |
| T$_2$     | 88.79c       | 4.03b | 15.05c           | 10.81b            | 66.22c           |
| SE +      | 3.17         | 0.02  | 3.35             | 0.076             | 2.76             |
| CV        | 4.1          | 1.8   | 4.9              | 3.0               | 4.9              |
| P value   | 0.001        | 0.001 | 0.001            | 0.001             | 0.001            |

*Means in the same column and factor followed by the same letter are not significantly different at P (≤ 0.05) according to Duncan Multiple Range Test. Note: BM = plant biomass (g/plant), LAI = Leaf area index; CGR: Crop growth rate (g m$^{-2}$/d,) and NAR: Net assimilation rate (g m$^{-2}$/d)

Table 4. Genotype × Fertilizer interaction effect on growth characteristics of groundnut

| Interaction (G xT) | BM (g/plant) | LAI | CGR (g m$^{-2}$/d) | NAR (g m$^{-2}$/d) | Number of Nodules /plant |
|--------------------|--------------|-----|-------------------|-------------------|---------------------------|
| G$_1$ x T$_0$      | 62.24a *     | 3.10 c | 12.00a           | 11.65a            | 27.74a                     |
| G$_1$ x T$_1$      | 62.51a       | 3.15 c | 13.77a           | 11.87a            | 54.79c                     |
| G$_1$ x T$_2$      | 66.12ab      | 3.26 c | 13.75a           | 11.74a            | 63.70d                     |
| G$_2$ x T$_0$      | 73.63bc      | 1.95a | 22.72c           | 16.30b            | 34.99b                     |
| G$_2$ x T$_1$      | 82.01cd      | 1.99a | 26.83c           | 16.35b            | 54.54c                     |
| G$_2$ x T$_2$      | 74.92bc      | 1.88a | 24.96c           | 17.32c            | 51.50c                     |
| G$_3$ x T$_0$      | 85.26de      | 2.31b | 13.76c           | 11.57a            | 85.46e                     |
| G$_3$ x T$_1$      | 93.24e       | 2.31b | 21.86c           | 11.57a            | 85.48e                     |
| G$_3$ x T$_2$      | 87.87de      | 2.36b | 16.36b           | 12.52a            | 85.48e                     |
| Mean              | 76.42        | 2.47  | 69.0             | 15.64             | 56.77                      |
| SE                | 6.13         | 6.06  | 6.13             | 0.30              | 4.46                       |
| CV(ab) %          | 7.8          | 13.3  | 8.9              | 2.3               | 7.9                        |
| P- value          | 0.2          | 0.09  | 0.04             | 0.003             | 0.001                      |

Interaction effects: Significant (P=0.2) interaction effect between G$_3$ x T$_1$ was observed on plant biomass; significant (P=0.09) interaction effect for G$_2$ x T$_2$ on LAI; Significant (P= 0.04) and (P= 0.003) interaction effect for CGR and NAR was observed with G$_2$ x T$_1$ and G$_2$ x T$_2$, respectively. Number of nodules was significantly (P=0.001) influenced by genotype x fertilizer interaction with G$_3$ x T$_2$ interaction showing positive response.

4. CONCLUSION

Results from the current study showed both genotype and fertilizer had significant (P≤ 0.05) influence on growth characteristics of groundnut with positive influence calcium observed on leaf area index (LAI) 4.03; crop biomass 88.79 g/plant; number of nodules 66.22, and crop growth rate (CGR) 15.05 g m$^{-2}$/day. Whereas phosphorus had significant influence on Net assimilation rate (NAR) 10.84 g m$^{-2}$/day.
Similarly, groundnut genotype (Masasi) had significant influence on LAI (3.95); CGR (13.04 g m⁻²/day); NAR (12.36 g m⁻²/day) and number of nodes (64.93). Given the significant interaction effect of genotype x fertilizer on growth parameters, genotype Pendo proved superior performance under both treatments were significantly influenced by genotype x fertilizer interactions with Significant (P=0.2) interaction effect for crop biomass observed between G₃ x T₁, while Gₓ T₁ significantly affected number of nodules and Gₓ T₁ had significant interaction effects on CGR and NAR. This is an indication that under appropriate agronomic management with 55 kg P/ha and 125 kg Ca/ha, Pendo has the potential to superior performance, hence yield compared to other genotypes used in the experiment.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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