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## Research Article

# Nitrogen Stress on Maize Roots in Subtropical Condition of Nepal

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## Abstract

**Background and Objective:** Nitrogen (N) is one of the most important elements for plants. Moreover, N is the most dynamic and vulnerable element. Managing N is therefore very crucial in agriculture. Various studies have been carried on the influence of yield and yield attributes of maize at different N levels. Very few studies carried out on rooting architecture. So, this study aims to understand the rooting behavior of maize and to identify efficient cultivars that perform better under N stress condition. **Materials and Methods:** A pot culture experiment was conducted at Lamjung district of Nepal situated at subtropical condition. Two levels of N (0 and 80 kg ha<sup>-1</sup> N) were applied to five varieties (most promising variety) of maize viz., Deuti, Manakamana-3, QPM (Quality protein maize), Arun-2 and Rampur composite in randomized completely block design. Pot media was calculated on mass basis and required nutrients except N was placed at sufficient moisture level. Different root parameters were taken and two-way ANOVA was analyzed by using software MStat-C. The Ms-Excel 2007 was used to make graphs and figures. **Results:** Results showed that effect of nitrogen was significant for all the root traits except total root length and seminal root length. Interaction between N and variety was significant for root dry matter, number of seminal and crown roots and crown root length. At low N, QPM produced the maximum root dry matter, Deuti with the greatest number of seminal roots, Manakamana-3 with the maximum crown root length and Rampur composite with the highest number of crown roots. **Conclusion:** Under nitrogen stress condition, the physical parameters of roots perform better than sufficient level of nitrogen. It doesn't mean the increment in nutrient uptake efficiency in N stress condition. It requires further understanding. Rather, rooting behavior under N stressed condition varies with variety distinctly. The QPM producing the maximum root dry matter at low N and the minimum value at high N and is more tolerant and highly susceptible respectively than other variety. Rooting behavior not only influenced by soil aggregation and physical properties of soil but also with nutrient concentration. Further study is necessary to understand the nutrient uptake behavior in nutrient stress condition.

**Key words:** Nitrogen stress, nutrient use efficiency, pot culture, root architecture, root traits, root dry matter, rooting behavior, quality protein maize

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**Competing Interest:** The authors have declared that no competing interest exists.

**Data Availability:** All relevant data are within the paper and its supporting information files.

## INTRODUCTION

Maize (*Zea mays* L.) is a world's widely grown cereal and primary staple food of many developing countries. Its production process is highly dependent on mineral elements especially nitrogen. Nitrogen is a crucial and essential element for the crop growth and development. The optimum application of N not only sustains yield but also limit the loss through the process such as leaching and denitrification. However, the overuse of N fertilizer has caused serious damages to environment<sup>1</sup> and resulted in the emission of the greenhouse gases<sup>2</sup>. The yield of maize crop has been found to be affected by various factors. Ineffective utilization of the below ground resources by the roots is one of them. Roots and their architecture are the crucial factor that determines the below ground acquisition of resources<sup>3</sup>. The development of root at early seedling stage is very crucial for plant establishment and the overall grain yield<sup>4</sup>. Nitrogen is a highly mobile element and its concentration differs by 1000 folds over short distance and time period<sup>5</sup>. The heterogeneous distribution of N allows the plants to alter its root architecture to assimilate more nutrients<sup>6</sup>. This plasticity nature of roots supports plants to survive in non-ideal conditions and compete for resources<sup>7</sup>. The variation in the concentration of N in the soil has shown diverse type of response to overall performance of root growth. The reserve carbon (C) in roots required for the buildup of root system under nutrient deficiency<sup>8,9</sup> is conserved by the formation of aerenchyma in the root cortex which is induce by the deficiency of nutrient such as N, P and sulfur<sup>10</sup>. In the response of Low Nitrogen (LN), N should be the prominent limiting resources for the growth of root then C since it usually accumulates in the leaves and roots<sup>11</sup>. The research results reveals that with the uniformly high N supply under field conditions can significantly reduce the maize root growth<sup>12</sup>. Under low soil N the absolute amount of roots was found usually less than with high soil N<sup>13</sup>. There is documentation that reveals the enhancement of root elongation under LN stress condition<sup>14-16</sup>. Likewise, limited supply of N in the field condition increased the root size to assimilate more soluble nitrogen from the soil<sup>17</sup>. Plants have inherent capacity to adopt and modify its root to N-depleted soil. Root-shoot ratio, total root length and length of crown roots with reduction in the number was found to be increased under the application of low nitrogen<sup>18,19</sup>. The study of plant roots is one of the most promising but least explored areas of research related to plant growth<sup>20</sup>. The modeling approach used by Hammer *et al.*<sup>21</sup> has enlisted sufficient evidence to believe that the change in the root architecture have profound

effect then shoot architecture to increase maize yield with the availability of water at sowing. Further, for achieving high yield and green agriculture, the focus should be on the improvement on the overall function of the root system through key interest on improving the nutrient use efficiency<sup>22,23</sup>. Studies of effect of low N application on root traits characteristics such as root length, seminal and crown root number and length of maize are also lacking. This study aimed at exploring the rooting behavior of maize and selecting cultivars for better growth under stress N condition.

## MATERIALS AND METHODS

A pot experiment was conducted inside the plastic house during May-June at Sundarbazar, Lamjung, Nepal during 2015. Approximately 4 kg of soil mixed with 88 g of FYM was filled in 5 L pot. Nutrient solution containing  $\text{KH}_2\text{PO}_4$  (30 kg  $\text{ha}^{-1}$ ),  $\text{K}_2\text{SO}_4$  (20 kg  $\text{ha}^{-1}$ ), Ca (71 ppm), Mg (30 ppm), Cu (2 ppm), Mn (0.2 ppm), Mo (0.2 ppm), Zn (1 ppm), B (0.2 ppm) and Fe (2 ppm) of was then added to the working solution medium of 500 mL. Pots were treated with two levels of N (0 and 80 Kg  $\text{ha}^{-1}$ ) and then three pre-germinated seeds of five maize varieties viz., Deuti, Manakamana-3, Quality Protein Maize (QPM), Arun-2 and Rampur composite were sown at a depth of 2 cm. The treatments were replicated three times and were arranged in a Randomized completely Block Design (RCBD). At six days after sowing, only two plants were maintained in each pot. The amount of water removed through evapotranspiration was replaced at every 2 days interval. Other intercultural practices (weeding and insecticide spray) were done. Plants were harvested 30 days after sowing. Total root length, seminal and crown root length and root numbers were measured manually before oven drying to the constant weight at 65°C for 48 h. After oven drying, root biomass was measured. Data were analyzed using MStat-C and figures and graph were prepared through Ms-Excel 2007.

## RESULTS

**Root dry matter:** Result shows that level of N, variety and their interaction had significantly ( $p < 0.05$ ) influenced the root dry matter (Fig. 1). Increased level of Nitrogen from 0 to 80 kg  $\text{ha}^{-1}$  reduced the mean root biomass from  $2.8 \pm 0.16$  g to  $1.5 \pm 0.11$  g  $\text{plant}^{-1}$ , respectively. However, varietal responses at 0 kg soil N  $\text{ha}^{-1}$ , QPM accumulated the highest amount of root biomass ( $3.5 \pm 0.4$  g) but it did not differ with Deuti and Manakamana-3. Arun-2 and Rampur composite accumulated the lowest amount of root biomass at the same

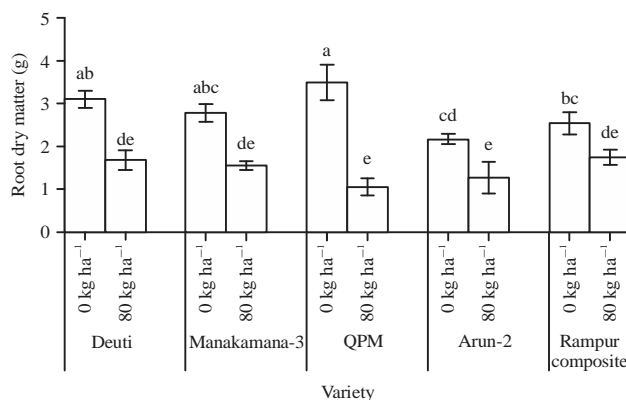


Fig. 1: Root dry matter of maize as affected by different nitrogen levels at 30 days of growth stage

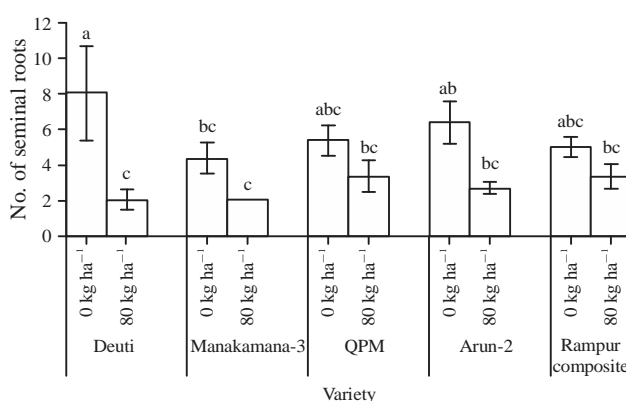


Fig. 2: Number of seminal roots of maize as affected by different nitrogen levels at 30 days growth stage

level of soil N. At 80 kg ha<sup>-1</sup> N varieties did not show significantly different level of root biomass accumulation.

**Number of seminal roots:** Data regarding number of seminal roots are shown in Fig. 2. Statistical analysis of the data indicated that different level of nitrogen and interaction effect of N and variety was significant at  $p < 0.05$ . Increased level of Nitrogen from 0-80 kg ha<sup>-1</sup> significantly reduced the mean seminal roots number from  $5.78 \pm 0.64$  to  $2.6 \pm 0.27$  plant<sup>-1</sup>, respectively. At 0 kg ha<sup>-1</sup> N, Deuti produced maximum number of seminal roots ( $8 \pm 2.6$ ) which was significantly different with Manakamana-3. Similarly, at 80 kg ha<sup>-1</sup> varieties did not show significant difference for number of seminal roots.

**Total root length:** Result shows that level of N, variety and their interaction did not show any variation for total root length (Table 1). Increased level of Nitrogen from 0-80 kg ha<sup>-1</sup> reduced the mean total root length from  $56 \pm 3.14$  to  $52.1 \pm 3.14$  cm plant<sup>-1</sup>, respectively. For variety response, maximum total root length was obtained for Manakamana-3 and minimum value for Arun-2.

Table 1: Total root length of maize as affected by different nitrogen levels at 30 days of growth stage

Variety	Fertilizer nitrogen rate (kg ha <sup>-1</sup> )	
	0	80
Deuti	54.8	57.8
Manakamana-3	65.8	50.7
Quality protein maize	50.1	52.8
Arun-2	46.0	46.0
Rampur composite	63.2	53.4
Mean	56.0	52.1

**Seminal root length:** Analysis of the data also revealed that the level of nitrogen, variety and their treatment combination had no significant ( $p < 0.05$ ) effect on the seminal root length of maize (Table 2). Increased level of Nitrogen from 0-80 kg ha<sup>-1</sup> reduced the mean seminal root length from  $32.2 \pm 2.17$  to  $25.6 \pm 4.3$  cm plant<sup>-1</sup>, respectively. In respect to variety, greatest seminal root length was observed for Manakamana-3 and minimum value for Deuti.

**Crown root length:** Result shows that level of N, variety and their interaction had significantly ( $p < 0.05$ ) influenced the

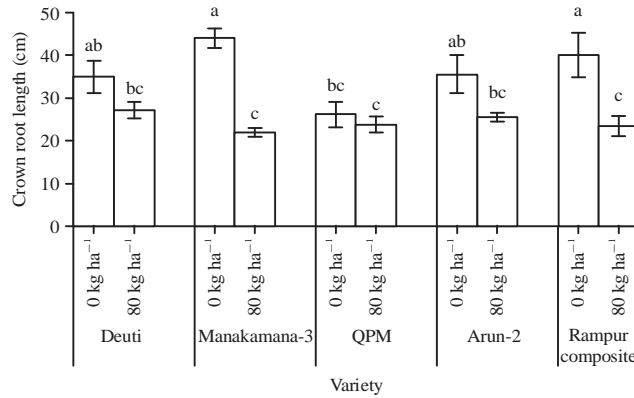


Fig. 3: Crown root length of maize as affected by different nitrogen levels at 30 days growth stage

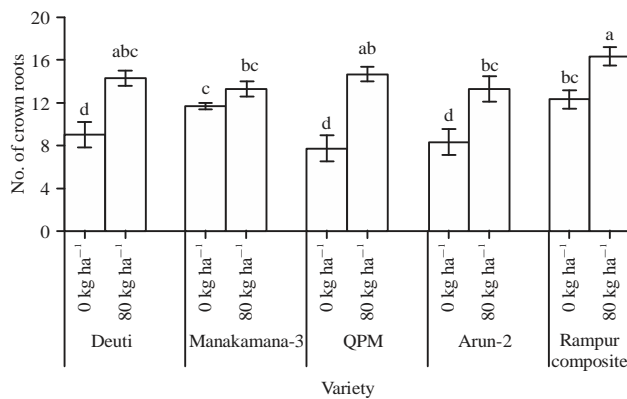


Fig. 4: Number of crown roots of maize as affected by different nitrogen levels at 30 days growth stage

crown root length (Fig. 3). Increased level of nitrogen from 0-80 kg ha<sup>-1</sup> significantly reduced the mean crown root length from 36.1 ± 2.16 to 24.3 ± 0.81 cm plant<sup>-1</sup>, respectively. However, for varietal responses at 0 kg ha<sup>-1</sup> N, Manakamana-3 produced the highest crown root length (43.9 ± 2.3 cm) which was nearly similar with Rampur Composite but it did not differ with Deuti and Arun-2. The QPM obtained the lowest value (26.2 ± 2.9 cm) at same level of soil N. At 80 kg ha<sup>-1</sup> N, varieties did not show significantly different level of crown root length.

**Number of crown roots:** Data regarding number of crown root is shown in Fig. 4. Statistical analysis of the data indicated that the different level of nitrogen, variety and their interaction was significant at p < 0.05. Level of Nitrogen from 0-80 kg ha<sup>-1</sup> significantly increased the mean crown root number from 9.8 ± 0.63 to 14.36 ± 0.43 per plant, respectively. For varietal response at 0 kg ha<sup>-1</sup> N, Rampur Composite produced maximum number of crown root (12.3 ± 0.9) which was similar with Manakamana-3 (11.6 ± 0.3). Similarly,

Table 2: Seminal root length of maize as affected by different nitrogen level at 30 days of growth stage

Variety	Fertilizer nitrogen rate (kg ha <sup>-1</sup> )	
	0	80
Deuti	32.4	16.6
Manakamana-3	37.4	32.7
Quality protein maize	31.5	36.4
Arun-2	32.4	18.8
Rampur composite	27.4	23.3
Mean	32.2	25.6

at 80 kg ha<sup>-1</sup> N, Rampur Composite produces the highest value (16.3 ± 0.9) which was similar with Deuti and QPM but significantly different with Manakamana-3 and Arun-2.

## DISCUSSION

Nitrogen being highly variable in its availability under soil may induce plants root to show their responses for nutrient assimilation. Comparing the two concentrations (0 and 80 kg N ha<sup>-1</sup>), highest root dry matter was obtained at low N (Fig. 1) suggesting that the photosynthetic carbon assimilation

was less at high N level as compared to low N level<sup>24</sup>. This was probably due to the stressed N soil causing the plant to increase the root size in order to assimilate more N. Similar results were observed for sweet-potato genotypes where amount of dry matter allocated to stored roots were greater under N-stressed<sup>12</sup>. Root mass was also found greater showing positive effect of low N on root dry matter<sup>18</sup>. Short-run high application of nitrogen fertilizer was found to decrease the root dry weight and root shoot ratio suggesting that there was limited allocation of photosynthetic carbon assimilates in the root<sup>25</sup>. The upkeep of root growth on low N allows maize with flexibility under this situation. Further, the enrichment of N lowered the soil pH by limiting the uptake of the other nutrients such as phosphorus<sup>26,27</sup>. Similarly, significant influence of nitrogen and varietal interaction implies that the response of root growth to different level differs among the varieties. In general it is believed that there will be increased root length with the application of N fertilizer. A study performed on maize RILs grown in solution culture reported an increase in axile root length with the two lowest N concentrations and finally increased N build-up in plants<sup>28</sup>. However, in the present study total root length of maize at 0 kg N ha<sup>-1</sup> was no greater than in the plants supplied with 80 kg N ha<sup>-1</sup> (Table 1). Also, different results were obtained with more efficient carbon-assimilation under high nitrogen with greater total root length and total lateral root length<sup>25</sup>. The non-significant effect of nitrogen on total root length might be due to inefficient cultivars unable to utilize the assimilated carbon. Similarly, there was no variation of N level and variety on length of seminal root but number of seminal roots increased under low N (Fig. 2). This indicates maize tend to increase its root branches instead of increasing its length under patchy soil N environment in order to assimilate more N uptake. In contrast there were reports that concluded the significant effect of different level of N on the number of seminal roots<sup>29</sup>. But there were reports with no any significant effect of different N levels on the number of seminal roots<sup>25,28</sup>. Low N soil also seems to increase the crown root length (Fig. 3) but inhibit the number of crown roots (Fig. 4). This result reveals the plant roots shows plasticity response to nitrate availability which finally results in fewer but longer laterals on low nitrate soils. These prefigured plasticity responses agree with the findings in the literature from artificial systems<sup>30,31</sup>. In hydroponics system, low nitrogen stress increases length of the crown root while reducing their number<sup>15,18,19,32</sup>. Similar kind of responses to low nitrogen was found in field research<sup>4,33</sup>. Tian *et al.*<sup>14</sup> reported the elongation of primary, seminal and crown roots inhibition of maize with the increasing level of external nitrate from 0.05-5 mM which was closely related to the reduction of IAA levels in the root

specifically at the region close to root tips. The significant varietal effect on root dry mass, number of crown and seminal roots and crown root length suggests that this better responding variety can be further studied under breeding programs to obtain efficient variety that make better use of N.

## CONCLUSION

Root growth and its response under stress N fertilization in soil was found positive for root traits such as root dry mass, number of seminal and crown roots and crown root length. Also, QPM producing maximum root dry mass at 0 kg N ha<sup>-1</sup> and minimum value at 80 kg N ha<sup>-1</sup> shows that the variety is tolerant at low N and highly susceptible at high N in comparisons to others. Further study is envisaged to increase nitrogen use efficiency under limited N supply root morphology.

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