

## Maize (*Zea mays* L.) Root Architectural Plasticity in Response to Critical Nitrogen Stress using an Aeroponics System

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### ABSTRACT

Nitrogen (N) is a crucial macronutrient that influences maize yield and development both above and below ground. Previous studies on maize root architecture have focused on seedling roots. Since adult, post-embryonic adventitious roots are the major root organs in maize responsible for N capture, this study examined root plasticity at post-embryonic developmental stages in response to low N. To dynamically quantify the maize post-embryonic root system, which is extensive and developmentally complex, we engineered an aeroponics system, where the roots are suspended in the air and misted at regular intervals with a nutrient solution. We detected that adult maize, upon low N stress exposure, ceases the elongation of metabolically expensive, pre-existing crown roots and/or makes shorter *de novo* crown roots, in favour of allocating more energy to support less expensive lateral root growth and branching. Our results reveal a novel adaptation to low N in adult maize: induced root architecture plasticity to maintain soil exploration while reducing the internal metabolic resources required.

**KEYWORDS:** *Zea mays*, Root Architecture, Nitrogen, Aeroponics

### INTRODUCTION

Due to technical challenges, the effect of nitrogen on the architecture of the extensive post-embryonic maize root system has been poorly studied. The objective of this study was to use aeroponics, in which roots are suspended in the air, misted by a nutrient solution, as a new method to monitor root growth and architectural plasticity in response to N stress in adult maize.

### METHODS

Plants were grown using two independent aeroponic systems under high N (16mM total Nitrogen) for 20 days and then either maintained at high N or switched to low N (6mM total Nitrogen) for 15 days (8-9 visible leaves) in a complete randomized design with three replicates of two plants. The experiment was repeated three times. The customized aeroponics system consisted of two 5-inch diameter pots fitted onto each 75L black aeroponics chamber. The nutrient solution was pumped under pressure from a 100L container using a submersible utility pump to two 180°-range microjets per barrel. A timer regulated root spray 10 sec every 50 sec. Chambers were placed onto a sloping floor to promote gravity return flow to a 5L black container and then pressurized back to the 100L nutrient solution container. The nutrient solution was changed weekly; pH and conductivity were measured daily. Maize commercial hybrid seeds from Syngenta (SRG200) were germinated and transferred to the aeroponics growth system in a glass greenhouse at  $800 \mu\text{Mol m}^{-2} \text{s}^{-1}$  at pot level, 16h photoperiod, and 28°C day /20°C night. Roots were scanned and analyzed using WinRhizo software (Version PRO, Regent instruments Inc, Canada). Embryonic and post-embryonic axial roots along with their respective laterals were separately analyzed for total root length per plant (RL), total surface area (SA) and total volume (RV). Shoot and root tissues were dried and weighed. Statistical analyses were performed using

the MIXED procedure of the SAS statistical software package (Version 9.1, Statistical Analysis System, SAS Institute, USA) with a Type I error rate set at 0.05.

## RESULTS

Compared to High N, Low N plants showed a 29% reduction in total dry weight (TDW) and an increased root to shoot ratio (R:S) reflecting a larger proportion of the total biomass allocated to the roots (24%). The total root length (RL) was unaffected by the switch in N treatment; however the crown root length (CRL) showed a significant decrease under N stress while the lateral root length remained stable. The total root surface area (SA) and root volume (RV) were also significantly decreased, as were all the crown root traits (Table 1).

Table 1. Variation in root traits 15 days after exposure to constant or low nitrogen concentrations.

	Root Length (RL) (cm )		Surface area (SA) (cm <sup>2</sup> )		Root volume (RV) (cm <sup>3</sup> )	
High Nitrogen (16mM)	<b>Total RL</b>	65679 ± 9540	<b>Total SA</b>	7331 ± 482.6**	<b>Total RV</b>	159.6 ± 4.6**
	<b>Lateral RL</b>	48165 ± 4331	<b>Lateral SA</b>	2051 ± 323	<b>Lateral RV</b>	8.6 ± 1.5
	<b>Seminal RL</b>	14142 ± 1100	<b>Seminal SA</b>	2324 ± 320	<b>Seminal RV</b>	31.4 ± 4.5
	<b>Crown RL</b>	3371 ± 179*	<b>Crown SA</b>	2955 ± 63*	<b>Crown RV</b>	119.5 ± 5.8**
Low Nitrogen (6mM)	<b>Total RL</b>	58882 ± 7782	<b>Total SA</b>	5901 ± 439.4**	<b>Total RV</b>	100.2 ± 3.7**
	<b>Lateral RL</b>	45225 ± 5987	<b>Lateral SA</b>	1947 ± 264	<b>Lateral RV</b>	8.8 ± 1.2
	<b>Seminal RL</b>	11367 ± 937	<b>Seminal SA</b>	1762 ± 261	<b>Seminal RV</b>	24.6 ± 3.7
	<b>Crown RL</b>	2291 ± 146*	<b>Crown SA</b>	2192 ± 56*	<b>Crown RV</b>	66.7 ± 4.8**

Values are least square means from ANOVA ± standard error (n=9). (\*) (\*\*) indicate significant differences between High N and Low N treatments at p=0.05 and p=0.01 respectively.

The length ratio of laterals / axial roots (seminal and crown roots) was significantly increased under stress. Since LRL was not affected by the switch to low N, the decline in this ratio was due to a decrease in CRL suggesting that more or longer lateral roots grew from a smaller crown root attributes. The specific root length (SRL) (TRL/RDW) was unaffected by N stress showing no variation in root length per unit of mass invested in the root system. However, the root length/volume also increased under N stress due to a significant increase in lateral root length per volume of roots (p=0.032). Assuming roots are cylindrical, then the root diameter determines the ratio of RL to RV (Eissenstat, 1991); in this study, N stress caused a decrease in lateral root diameter whereas the diameters of seminal and crown roots remained unchanged.

## DISCUSSION

Aeroponics was able to overcome the challenge of dynamically monitoring a large post-embryonic roots system. Upon low N stress exposure, adult maize maintained TRL but invested less energy in metabolically expensive crown roots in favour of less expensive lateral roots, demonstrating the importance of quantifying post embryonic root architecture and not only root mass total length.

## REFERENCES

Eissenstat DM. 1991. On the relationship between specific root length and rate of root proliferation: a field study using citrus rootstocks. *New Phytologist* 118: 63-68.