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EFFECT OF NITROGEN APPLICATION ON NODAL ROOT CHARACTERISTICS AND ROOT LODGING RESISTANCE IN MAIZE

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Abstract

Increased plant density has a greater risk of lodging than decreased plant density because of the changes in the root system structure. The root architecture and nutrition uptake of high-density populations can be regulated by nitrogen regimes in maize. In this study, the effects of nitrogen application on root system structure and lodging resistance of a high-density maize population was evaluated. Using the maize XY335 as an experimental model, lodging-related root traits and shoot parameters were evaluated under various planting densities [4.50×10^4, 6.75×10^4,and $9.00 \times 10^4 \cdot hm^{-2}$ (LD, MD, and HD, respectively)] and nitrogen applications [0, 225, and 337.5 $kg \cdot N \cdot hm^{-2}$ (N0, N1, and N2, respectively)]. The root failure moment (*Rfm*), root plate diameter (*d*), and the diameter and number of nodal roots per maize plant were measured. Results showed that nitrogen application increased *Rfm*, root plate diameter, and the number and diameter of nodal roots on stem node P6–P8. A significant positive linear relationship between *Rfm* and root plate diameter was found. In addition, the root plate diameter showed a significant positive linear relationship with the weighted mean of nodal root diameters on stem node P5–P8. In conclusion, nitrogen application significantly enhanced the lodging resistance by regulating the number and diameter of nodal roots in maize.

Key words: Maize, Root lodging, Lodging resistance, Nodal root, Crown root, Root failure moment.

Introduction

As one of the major crops in China, maize (*Zea mays* L.) is widely grown in the Northeast China Plain, located in approximately 39–53°N (Wang *et al.*, 2012). Increasing the planting density is an important way to enhance the maize yield (Zhang, 2007). However, higher planting densities lead to higher risk of lodging. Lodging not only limits mechanical harvesting (Setter *et al.*, 1997) but also causes yield loss (Bian *et al.*, 2016; Tollenaar & Lee, 2002) and poor grain quality (Zhang *et al.*, 2014).

Lodging refers to the permanent displacement of aboveground portions of crop plants from their vertical stance, which can be classified as root and stem lodging (Pinthus, 1973). Normally, root lodging results from the failure of the root–soil anchorage system (Wu & Ma 2016), which usually occurs during rainy and windy days.

The root–soil anchorage system is important for root lodging resistance (Piñera-Chavez *et al.*, 2016), which is closely related to root traits (Liu *et al.*, 2012). The root system is composed of a primary root, several seminal roots, and nodal roots in maize (Hopper *et al.*, 1986). Nodal roots arise from crown and brace roots. The brace roots on higher stem nodes appear later in plant development and are critical for root anchorage in maize. The number, diameter, and growth angle of nodal roots and root plate spread are all related to root lodging resistance (Berry *et al.*, 2000; Piñera-Chavez *et al.*, 2016). The spread of the root plate is also one of the main factors for root lodging resistance (Berry *et al.*, 2004; Berry & Berry, 2015).

The increase in planting density results in intense competition between crop plants for light and nutrients, which may reduce the diameter and number of nodal roots (Huang *et al.*, 2014), leading to decreased lodging resistance (Liu *et al.*, 2012). Sposaro *et al*., (2008) found that root plate diameter is reduced by higher planting density in sunflower. Berry *et al*., (2000) reported that the spread of the root plate is decreased with higher planting density, hence depressing plant anchorage strength in wheat. In Northwest Mexico, Piñera-Chavez *et al*., (2016) have documented that the optimized root plant spread is 51 mm for root lodging resistance in wheat.

Nitrogen (N) application affects many plant traits in crops (Greef, 1994). In maize, higher length but lower dry weight of nodal roots was found under low N conditions (Reid & Smith, 2002), whereas larger shoot and root biomass, more tillers, and increased root branching were observed with excess N fertilization (Xue *et al.*, 2017). Thus, N application is critical for the formation of the root system structure (Zobel *et al.*, 2007). Optimized N supply can reduce the competition for nutrients in a high-density maize population by regulating the growth of roots and shoots. Mulder (1954) and Crook & Ennos (1995) reported that the root–soil anchorage system can be regulated by N application. However, the effects of nitrogen application on root system structure and lodging resistance in a high-density maize population are still unclear.

Normally, the N application rate is higher in a high-density maize population. In this case, lodging-related root traits can be regulated for enhanced root lodging resistance by N application. However, the relationship between root system structure and lodging resistance under varied N application rates in a high-density maize population is unclear. Therefore, the objectives of this study were to clarify (1) the response of lodging-related root traits to various N application rates; (2) the effect of N application on root lodging resistance of maize plants under different planting densities; and (3) the relationship between root system structure and lodging resistance.

Materials and Methods

Field experimental design: Maize cv. XY335 was sown in the field at the experimental station in the Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun (44°12′ N, 125°33′ E), in May 1, 2013. A split-plot design was used with three planting densities as the main plot [4.50×10^4, 6.75×10^4, and $9.00 \times 10^4 \cdot hm^{-2}$ (LD, MD, and HD, respectively)] and three N levels as the subplots [0, 225, and 337.5 $kg \cdot N \cdot hm^{-2}$ (N0, N1, and N2, respectively)]. One-third N and 75 $kg \cdot P_2O_5 \cdot hm^{-2}$ and 150 $kg \cdot K_2O \cdot hm^{-2}$ were applied as basal fertilizer, and the remaining N was top dressed at the jointing stage. The row space was 65 cm, and the length of the row was 10 m. The subplot size was 6.5 × 10 m. The rows were oriented in the south–north direction. The soil was a black soil clay containing 28.2 g kg^{-1} organic matter, 1.6 g kg^{-1} total N, 143 mg kg^{-1} available N, 66.5 mg kg^{-1} Olsen-P, and 150 mg kg^{-1} available K. The monthly mean temperature and average precipitation in 2013 are shown in Fig. 1.

Plant height, ear height, and height of the gravity center: Plant height, ear height, height of the gravity center, nodal root number, and diameter of nodal root and root plate were measured at 110 days after sowing (DAS). Plant height, ear height, and height of the gravity center were measured with a ruler.

Root failure moment *(Rfm)*: Similar equipment as reported by Sposaro *et al*., (2008) was used to measure the root lodging force at 110 DAS. Ten plants were selected for each plot. Two days before measurement, all plots were well irrigated. All leaves and sheaths were removed, and the stem was detopped at 40 cm from the stem base. Force was applied at 20 cm from the stem base. The plants were lodged to the west. The maximal force (F_{max}) was measured with a protractor (HF-500N, Haosa Inc., Shanghai, China), and the angle between the stem and the ground during F_{max} (α) was recorded.

The root failure moment (*Rfm*, in Nm) was calculated with the following equation:

$$Rfm = F_{max} \times \cos\alpha \times h$$

where F_{max} is the maximal force (Newton) of the push, α is the angle between the stem and the ground during F_{max}, and h is the distance between the point of force application and the ground, which is 0.20 m in this study.

Root plate diameter (d) and number and diameter of nodal roots: After measuring *Rfm*, all roots for each plant were cleaned with water, and then the root plate diameter (d) and number and diameter of nodal roots were measured.

In this study, the mesocotyl was considered as the first phytomer (P1), the lowest stem node with nodal roots was the second stem node (P2), and the rest of above stem nodes were numbered in order as P3 to P8 (Girardin *et al.*, 1986). The number of nodal roots arising from stem node P4–P8 was counted, where the number of nodal roots on the above-soil internodes was not included. The diameter of nodal root was measured 5 cm below the stem base with a Vernier calliper.

The root plate diameter (*d*) was calculated by averaging the highest and lowest root plate spread (Berry *et al.*, 2000).

Analysis: Statistical analyses were performed using SPSS 16.0 (SPSS Inc., Chicago, IL, USA). Two-way ANOVA was used to assess the effects of N application rates and planting densities and their interaction.

Results

Plant height, ear height, and height of the center of gravity: Compared with N0, N application significantly enhanced the shoot height, ear height, and height of the gravity center (Fig. 2a, b, and c). The interactive effect of N application and planting density was also found.

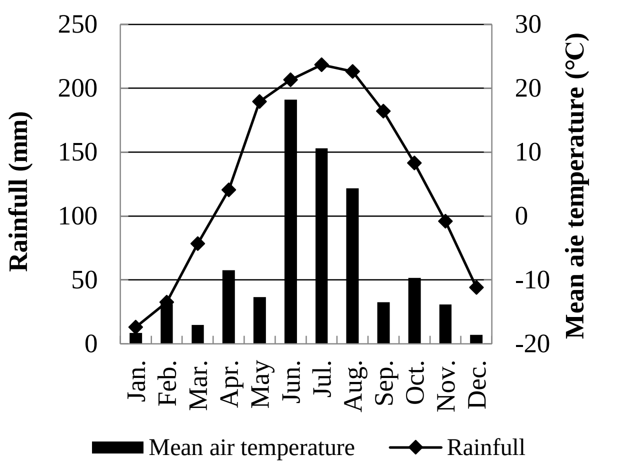

Fig. 1. Monthly mean temperature and average precipitation in 2013.

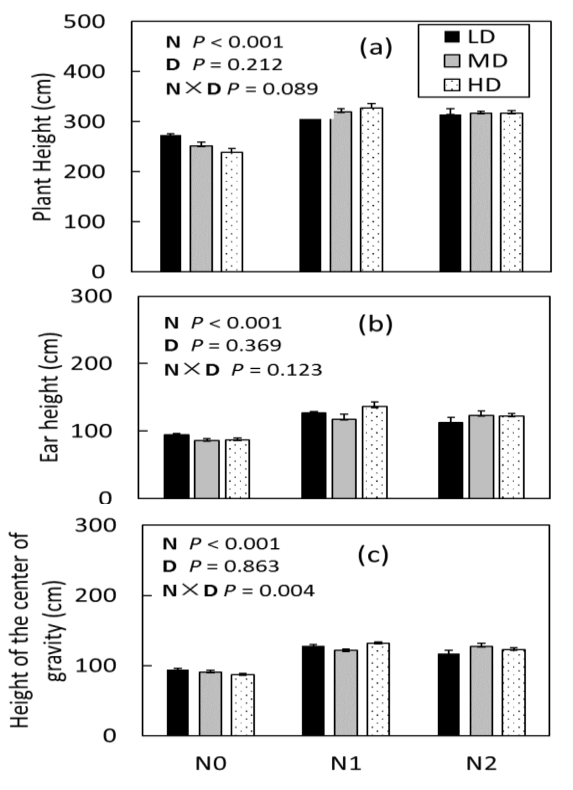

Fig. 2. Plant height (a), ear height (b), and height of gravity centre (c) in maize under different nitrogen levels and planting densities (N: nitrogen levels; D: planting density).

Table 1. Output of two-way ANOVA for the effect of nitrogen (N) and planting density (D) and their interactions (N×D) on the number and diameter of nodal roots on stem node P2–P8.

Factor	Root type				
	P4	P5	P6	P7	P8
The number of nodal roots					
N	ns	ns	,	,	,
D	ns	ns	,	,	,
N×D	,	ns	ns	ns	,
The diameter of nodal roots					
N	,	ns	*	,	,
D	,	ns	,	,	,
N×D	,	ns	,	,	,

* Indicates significance level at p<0.05; ns, No-significance

Number and diameter of nodal roots: The number of nodal roots on stem node P4 and P5 was not significantly affected by either planting density or N application, whereas the number of nodal roots on stem node P6–P8 was significantly affected by N application. Under LD and MD, the number of nodal roots on stem node P6 was the lowest in N0 followed by N1, and the highest value was found in N2. Under HD, the lowest and highest numbers of nodal roots on stem node P6 were in N1 and N2. Under LD and MD, the number of nodal roots on stem node P7 was lower in N0; however, under HD, it was lower in N1 and N0. The nodal roots arising from stem node P8 were only observed in N2 under LD (Fig. 3a and Table 1).

The diameter of nodal roots on stem node P4 and P5 was neither affected by planting densities nor by N levels. However, the diameter of nodal roots on stem node P6–P8 was significantly influenced by N levels. Under LD, the diameter of nodal roots on stem node P6 was the lowest in N2, whereas it was higher in N0 and N2 under MD. Under HD, the nodal root diameter on stem node P6 was the lowest in N0 and highest in N2. Under LD, the lowest diameter of nodal roots on stem node P7 was found in N1. However, under MD, the lowest value was in N0. The diameter of nodal roots on stem node P7 was the lowest in N0 and N1 under HD (Fig. 3b and Table 1). In addition, the interactive effects of planting density and N application were found on nodal root number on stem node P8 and nodal root diameter on stem nodes P6–P8 (Table 1).

Root failure moment *(Rfm)*: *Rfm* was significantly affected by N application (Fig. 4). Under each planting density, the lowest *Rfm* was found in N0, followed by N1, whereas the highest *Rfm* was found in N2.

Root plate diameter (d): Root plate diameter was significantly affected by N levels (Fig. 5). Under LD, the root plate diameter was the lowest in N0 and was the highest in N2. Under MD and HD, the lowest *d* was also found in N0, and no significant difference in *d* was found between N1 and N2.

Relationships of root plate diameter and number of nodal roots on the upper internodes: A significant positive relationship was found between root plate diameter and total number of nodal roots on stem node P5–P8 ($R^2 = 0.644^{**}$, p< 0.019). The root plate diameter was positively correlated to the total number of nodal roots on stem node P5–P8 (Fig. 6a). In addition, it had a close positive linear relationship with the weighted average of diameters of nodal roots on stem node P5-P8 (Fig. 6b) and *Rfm* (Fig. 6c).

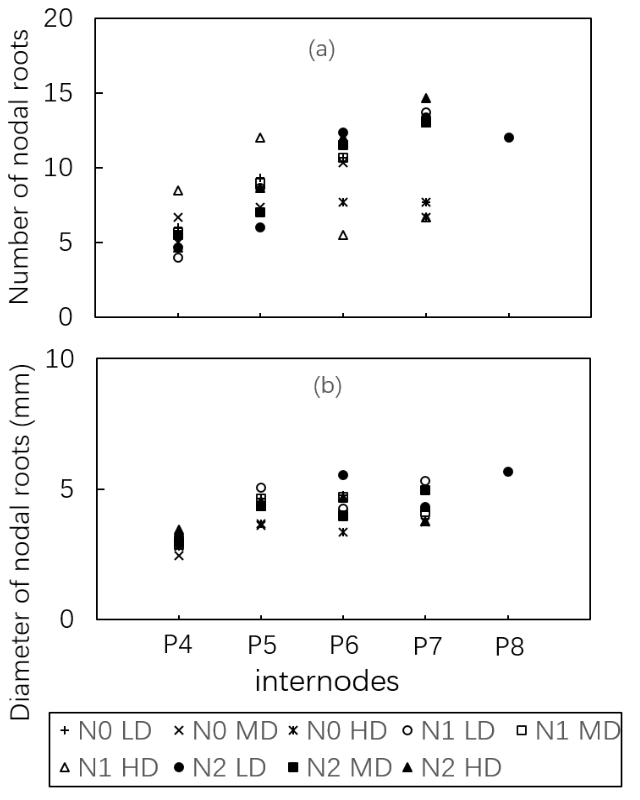

Fig. 3. Number and diameter of nodal roots in maize under different nitrogen levels and planting densities.

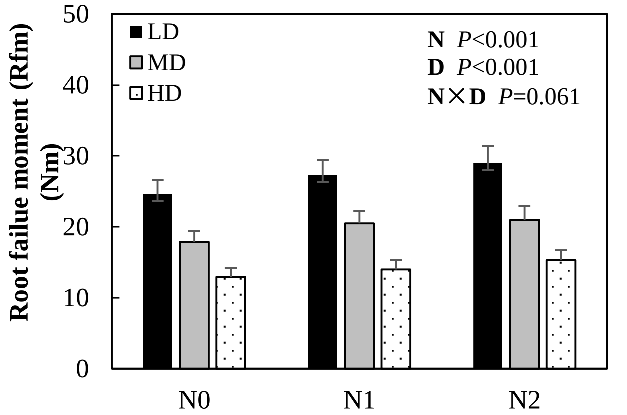

Fig. 4. Root failure moment (*Rfm*) in maize under different nitrogen levels and planting densities.

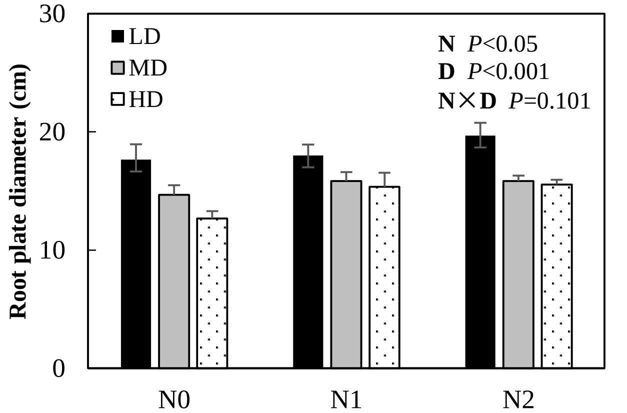

Fig. 5. Root plate diameter in maize under different nitrogen levels and planting densities.

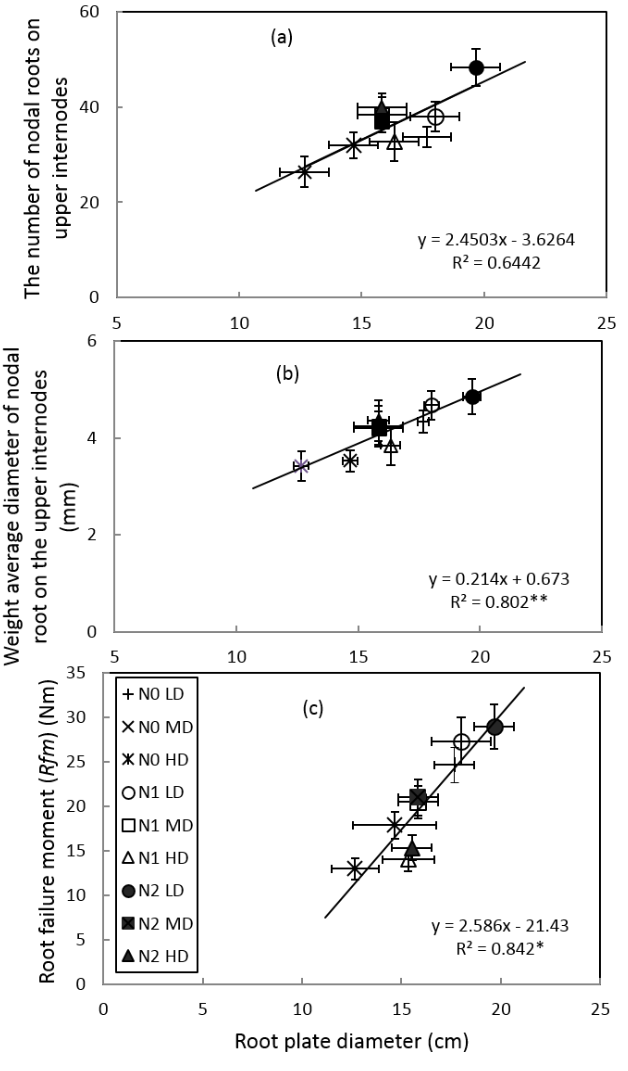

Fig. 6. Relationships between root plate diameter, the total number (a) and weighted average of diameters of nodal roots on stem node P5–P8 (b), and root failure moment (*Rfm*) (c) in maize under different nitrogen levels and planting densities.

Discussion

Lodging normally occurs 2 to 3 months before harvest in maize (Liu *et al.*, 2012). In wheat, it usually occurs during the grain filling stage because of the high shoot biomass. After grain filling, the root–soil anchorage system is stable, leading to higher lodging resistance. It is similar in maize wherein the lodging resistance is the lowest at grain filling due to enhanced ear weight and increased height of gravity center (Berry *et al.*, 2000).

The plant height, ear height, and height of gravity center are all important for lodging resistance (Sanguineti *et al.*, 1998). In the present study, N application significantly increased the height of plant, ear, and gravity center compared with N0 (Fig. 2). No significant relationship was found between *Rfm* and the height of the plant, ear, and gravity center. This result was consistent with the study of Troyer & Rosenbrook (1983), where no significant relationship was found between root lodging resistance and height of stem and ear. However, enhanced root anchorage is required when shoot height increases with higher N application.

The growth and development of maize roots are both dependent on genetics and the environment (Feix *et al.*, 2002). The number of primary and nodal roots on the first three stem nodes are not normally influenced by environmental changes, whereas the plasticity of nodal roots arising from higher stem nodes has been reported (Demotes-Mainard & Pellerin, 1992; Hochholdinger, 2009; Pellerin, 1994). In this study, the number of nodal roots on stem node P4 and P5 was not significantly affected either by planting density or N application. However, the number of nodal roots on stem node P6–P8 varied with different N application rates and planting densities. In addition, the diameter of nodal roots on stem node P6–P8 was also significantly affected by N application and planting density (Fig. 3 and Table 1). We also found the interactive effects of N levels and planting densities on the nodal root number on stem node P8 and the diameter of nodal roots from stem node P6–P8. This result is consistent with the report from Pellerin (1994), wherein the nodal root number from relatively higher stem nodes is affected by planting density and can be regulated by N supply. The increase in planting density results in intense competition among crop plants for light and nutrients, which may shift the nutrient distribution and translocation (Pagano & Maddonni, 2007). Demotes-Mainard &Pellerin (1992) suggested that higher planting density resulted in poor illumination and ventilation in internal canopy of maize, which reduced the synthesis and use efficiency of carbohydrates, hence decreasing the number of functional nodal roots. N supply could reduce the competition for nutrients and increase the growth of nodal roots on higher stem nodes, hence improving the total number and diameter of nodal roots in a high-density population.

Our results documented that *Rfm* and root plate diameter were significantly affected by planting density (Figs. 4 and 5). Higher planting density reduced *Rfm* and root plate diameter, which were consistent with the results in wheat and sunflower (Sposaro *et al.*, 2008; Berry, 2000). It also indicated that lodging resistance was critical for grain yield in a high-density maize population. Sposaro *et al.*, (2008) reported that the diameter of the root plate could be used for evaluating the root lodging resistance of sunflower on the R5.9 stage, and the diameter of the root plate was easier to measure in relation to *Rfm*. Our study showed that the root plate diameter was significantly positively related to *Rfm* (Fig. 6c).

In this study, the number of nodal roots on relatively higher stem nodes is closely correlated to the root plate diameter (Fig. 6a and b). In our previous study, the angle between nodal roots and stem node was not affected by planting density (Liu *et al.*, 2012). Here, we also obtained the same result (data not shown). Therefore, for a given cultivar, the diameter of the root plate is not related to the angle between nodal roots and stem node, but it is affected by the number and diameter of nodal roots under various planting densities and N levels.

Foerster *et al*., (2000) compared the diameter of nodal roots arising from higher stem nodes and the number and angle of aerial roots between lodging-resistant and sensitive maize cultivars. Their results showed no significant difference in the parameters between lodging-resistant and sensitive cultivars. The relationship between root traits and lodging resistance of four wheat cultivars were investigated in the study of Crook & Ennos (1993). They documented that the strategy of preventing lodging is cultivar dependent. Thus, the evolution of lodging resistance is limited depending on a single trait.

In the present study, higher planting density depressed the root anchorage, reduced the number of nodal roots arising from higher stem nodes, and decreased the diameter of the root plate, which are in line with the results of Berry *et al*., (2004). Optimized N application increased *Rfm*, indicating that the root anchorage and lodging resistance can be enhanced by optimizing the N application in high-density maize populations. Furthermore, the diameter of the root plate and the number and diameter of the nodal root arising from higher stem nodes can also be regulated by N application. Previous studies showed that having more nodal roots increases the diameter of the root plate, hence improving plant lodging resistance (Berry, *et al.*, 2000; Sposaro *et al.*, 2008). This is because the lodging resistance-related traits are improved with high-diameter root plate (Ennos & Cook, 1993). In summary, nitrogen application significantly enhanced the lodging resistance by regulating the number and diameter of nodal roots in maize.

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