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Effect of environmental conditions on the genotypic difference in nitrogen use efficiency in maize

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Selection for nitrogen (N) efficient cultivars is typically conducted under favorable field conditions with only difference in soil N availability. However, in practical field conditions, variation in soil types and/or seasonal weather conditions may have a strong influence on plant growth and therefore, N use efficiency. In the present study, a set of 3 genotypes (JD209, JD180 and SM25) were compared for their response to N inputs in two locations with different soil types in 2004 and 2005. It was found that maize yield in Xin-Li-Cheng with black soils was significantly higher than that in Qian-an with light chernozem soil. At the same location, maize yield in 2005 was higher than in 2004 because there was more rainfall in 2005. With sufficient N supplies (150 to 300 kg/ha), no difference in yield potential was observed among the 3 hybrids under the favorable soil and weather conditions. Nevertheless, genotypic difference in maize yield in response to N inputs was observed under varied soil types and rainfall conditions. N-efficient JD209 only showed low-N tolerance under unfavorable soil (light chernozem) and water shortage condition (in 2004). It is concluded that, identification of N-efficient cultivars should be conducted under multiple environments.

Key words: Maize, N efficiency, soil, precipitation.

INTRODUCTION

Maize (*Zea mays* L.) is widely distributed from the sub-humid to semi-arid area in northeastern China Plain. Nitrogen is the major limiting mineral nutrient in maize production. Breeding for N-efficient cultivars, which can achieve a relative high yield at low N input, is considered a promising way to deal with the problem (Bolaños and Edmeades, 1993). Selection for N-efficient cultivars is typically conducted under favorable field conditions with only difference in soil N availability. However, in practical field conditions, variation in soil types and/or seasonal

weather conditions may have a strong influence on soil N dynamics and plant growth and therefore, N uptake and its subsequent utilization in plants. As a result, the response of a genotype to N inputs may be quite different under different soil and/or weather conditions (Shumway, 1992). A N-efficient genotype selected under favorable soil and weather conditions may not have superior performance in an adverse condition and vice versa. It is not clear if the adaptability to environment variation play an important role in efficient use of N fertilizer. Research in CIMMYT suggested that there is close relationship between low-N and drought tolerance (Bänziger et al., 2000). In the present study, the relationship between N-efficiency and environmental adaptability was further investigated by using 3 maize genotypes grown in two locations with different soil types in two years. The results suggest that the N-efficient trait of a genotype is closely related to its adaptability to soil characters and water supplies.

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Table 1. The major chemical characteristics of the soils.

Experimental location	pH (1:2.5)	Organic matter (g/kg)	Total nitrogen (g/kg)	Olsen-P (mg/kg)	NH ₄ COOH-K (mg/kg)	NH ₄ ⁺ N (0-30cm) mg/kg	NO ₃ ⁻ N (0-30cm) mg/kg
Xin-Li-Cheng	5.73	28.5	1.84	19.6	139.1	10.1	7.96
Qian-an	8.02	22.2	1.66	18.8	117.8	4.4	10.2

Table 2. Rates of N application in Xin-Li-Cheng and Qian-an in 2004 and 2005.

Experimental location	Rate of N fertilizer (kg N /ha)					
	2004			2005		
	N0	N1	N2	N0	N1	N2
Xin-Li-Cheng	0	190	300	0	150	200
Qian-an	0	190	300	0	190	300

MATERIALS AND METHODS

Locations

Maize *Zea mays* L. was grown in 2004 and 2005 in two locations of Jilin province of China, Xin-Li-Cheng and Qian-an, respectively. Xin-Li-Cheng is located in the central Northeast Plain of China with a semi-humid weather. During the growing season from April through September, the precipitation was 410.1 and 642.5 mm in 2004 and 2005, respectively. Qian-an is located about 200 km northwest of Xin-Li-Cheng. It has a semi-arid weather (Zhang et al., 2002; An, 2002). The precipitation from April through September was 316 and 543 mm, respectively. So rainfall in 2004 was less than in 2005 at both locations. In Xin-Li-Cheng, the soil type is a black soil with good nutrient buffer capacity (Wang and Liu, 1997). It has a pH of 5.73 (Table 1). In Qian-an, the soil types is a light chernozem which is much sandy in comparison to the black soil in Xin-Li-Cheng. The pH value is high (pH = 8.02) (Agricultural Planning Department of Qian-an, 1984). Both of the locations have been grown continuously for maize.

Genotypes and N treatments

Maize hybrids SM25, JD209 and JD180 were chosen according to their differential response to soil fertility in a previous study (Wu et al., 2001). The experiment was a split-plot design with N treatments as the main plot and genotypes as the sub-plot. Nitrogen fertilizer rates were shown in Table 2. Each treatment was repeated 3 times, resulting in a total of 72 plots. The plot size was 60 (in Xin-Li-Cheng) and 80 m² (in Qian-an), respectively. Plots were thinned at the seedling stage to a final stand of 60 000 plants ha⁻¹. No irrigation was applied. Phosphorus (P₂O₅ 69 kg ha⁻¹) and potassium (K₂O, 50 kg ha⁻¹) was applied as basal fertilizer before sowing. Thirty kg ha⁻¹ of the N fertilizer (as urea) was applied as basal fertilizer and the rest was applied at 9 leaf stage. Maize was sown in late April and harvested in early October in 2004 and 2005. The plot for each genotype and N treatment was fixed in the two years. In Xin-Li-Cheng in 2004, root samples were taken at anthesis stage by excavation method. A soil column surrounding a plant, with surface area of 0.25 m (1/2 distance between rows) × 0.17 m (1/2 distance between plants in a row), was excavated to a depth of 60 cm below the soil surface. The soil column containing the roots was washed by using a mesh sieve. All the plants from each plot were sampled for yield determination. Five plants from each plot were sampled for determining N content in plants. Nitrogen utilization

efficiency (NUE) was calculated as the grain yield divided by P accumulated in aboveground biomass at maturity. Data were statistically analyzed using SAS program and Microsoft excel.

RESULTS

Variance analysis

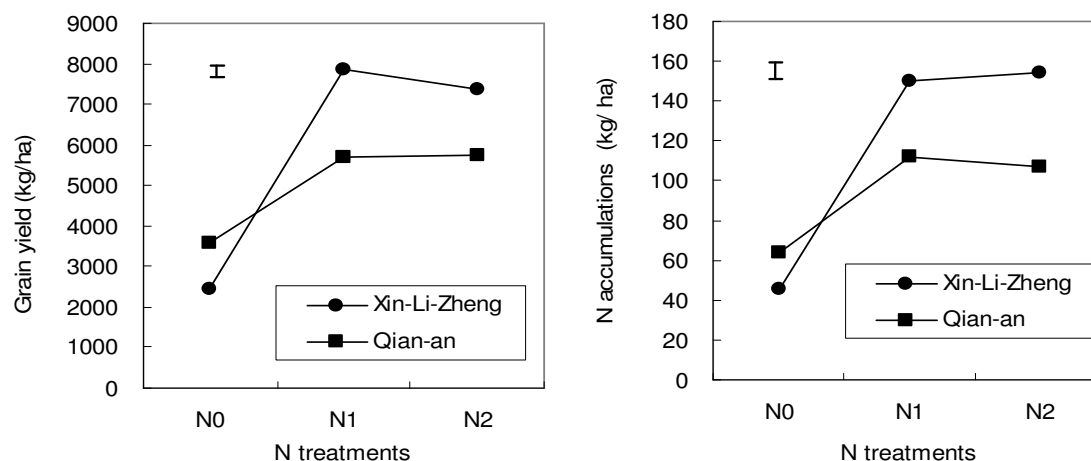
In general, there was a significant difference in maize yield and N accumulation among genotypes, N treatments, as well as between the two locations (Table 3). Across genotypes, N treatments and the two sites, grain yield was not different between 2004 and 2005. Except for year × N treatment, the interactions between any 2, among any 3 and 4 experimental factors were significant for grain and N accumulation, suggesting the special combination of genotype, location, weather (year), as well as N application play an important role in both N accumulation and grain yield. Nitrogen utilization efficiency (NUE) was significantly affected by years and N treatments and their interaction with sites, but was not different among genotypes.

Yield variation across soil types

At zero-N (N0) treatment, maize yield and N accumulation in Qian-an were higher than in Xin-Li-Cheng (Figure 1), reflecting that basic NO₃⁻-N content in the soil of Qian-an was higher (Table 1). Nevertheless, yield and N accumulation response to N application were stronger in Xin-Li-Cheng than that in Qian-an. This may result from two reasons. One reason is that soil structure in Xin-Li-Cheng was better. It is a black soil which is loamy with higher organic matter content and the soil pH was suitable for plant growth (pH = 5.73) (Table 1). While in Qian-an, the soil is a light Chernozem soil, which is much sandy with lower organic matter and its pH value is suboptimal (pH = 8.02) (Table 1). The second reason is

Table 3. Variance analysis of yield, N accumulations and N utilization efficiency of maize hybrids at different experimental areas.

Parameters	Yield			N accumulation			N utilization efficiency			
	Df	Mean square	F value	Pr > F	Mean square	F value	Pr > F	Mean square	F value	Pr > F
R (repeat)	2	614987	3.03	0.055	21.1	0.13	0.8825	54.1	0.87	0.4235
A (year)	1	3008	0.01	0.9035	11205	66.7	<.0001	1876	30.20	<.0001
B (sites)	1	21518622	106	<0.0001	13522	80.4	<.0001	2.08	0.03	0.8552
C (N treatments)	2	158858565	782	<0.0001	69690	414	<.0001	257	4.14	0.0199
D (cultivars)	2	4635729	22.8	<0.0001	1752	10.4	0.0001	0.04	0.00	0.9993
A*B	1	11321861	55.8	<0.0001	26080	155	<.0001	1544	24.9	<.0001
A*C	2	357511	1.76	0.1795	524	3.12	0.0504	9.46	0.15	0.8591
A*D	2	2305725	11.4	<0.0001	852	5.07	0.0088	18.1	0.29	0.7483
B*C	2	2821509	139	<0.0001	11425	68.0	<.0001	92.8	1.49	0.2316
B*D	2	837904	4.13	0.0202	565	3.36	0.0404	232	3.75	0.0285
C*D	4	1253663	6.17	0.0003	874	5.20	0.0010	35.3	0.57	0.6865
A*B*C	2	2684603	13.2	<0.0001	5400	32.1	<.0001	136	2.19	0.1195
A*B*D	2	1657505	8.16	0.0007	1276	7.59	0.0010	176	2.83	0.0659
B*C*D	4	549642	2.71	0.0371	761	4.53	0.0026	138	2.22	0.0757
A*B*C*D	8	437774	2.16	0.0416	338	2.01	0.0577	49.0	0.79	0.6138

**Figure 1.** Maize grain yield and N accumulation in two sites at 3 N input levels. Bars indicate the value of LSD_{0.05}.

that in comparison to Xin-Li-Chen, precipitation in Qian-an was lower and the crops are much subjected to seasonal aridity (see materials and methods).

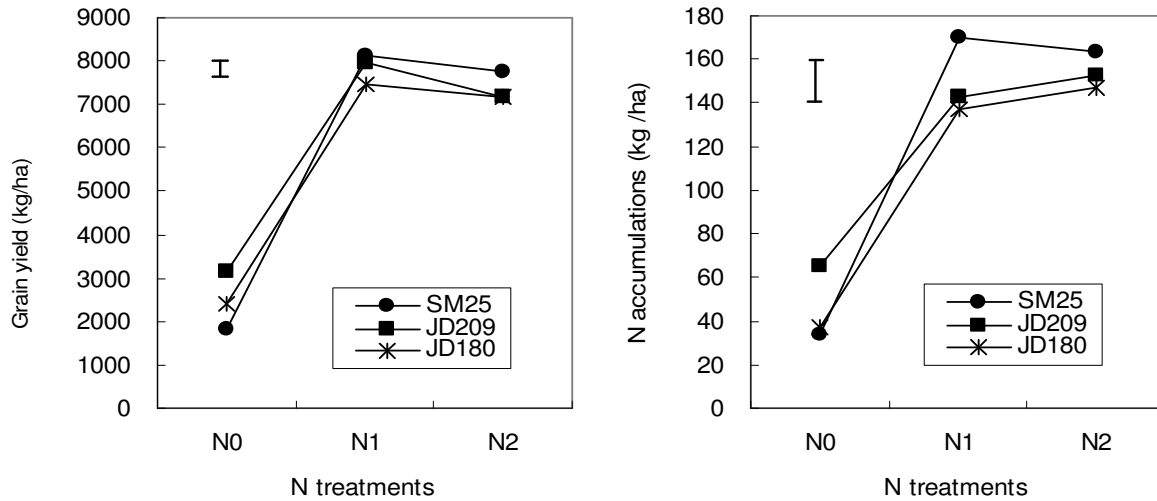
The yield level and plant N accumulation were similar at N1 and N2 treatments in both sites (Figure 1) suggesting that N amount at N1 treatments was enough to ensure the productivity of the genotypes. In Xin-Li-Cheng, no significant genotypic difference was found in yield at N1 and N2 treatments (Figure 2a), suggesting that the yield potential of these three cultivars are similar. SM25 accumulated more N than the other two genotypes, suggesting it has a high potential for N accumulation at suitable soil conditions. At N deficiency treatment (N0), JD209 performed the best in both sites (Figure 2a,b) and therefore, can be taken as N-efficient. In Qian-an, the yield of JD209 was the highest at all the 3

N treatments (Figure 2b), suggesting a high growth potential in the soil and climate conditions in Qian-an.

Genotypic difference in yield and N accumulation across years

When the data of all the three genotypes and two experimental sites were pooled, no yield difference was found between 2004 and 2005 at each N treatment (data not shown). During the growth seasons, the rainfall in Xin-Li-Cheng and Qian-an was 410.1 and 316 mm, respectively, in 2004, and 642.5 and 543 mm, respectively, in 2005. Therefore, the water supply in 2004 was much less than in 2005 in both sites. Interestingly, a significant genotypic difference in maize yield was found

A: Xin-Li-Cheng



B: Qian-an

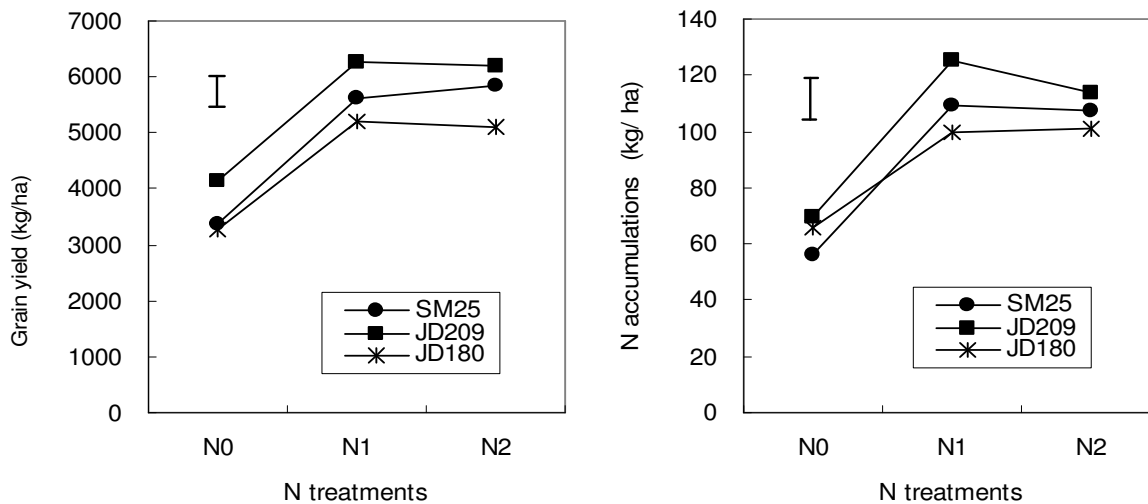


Figure 2. Genotypic difference in maize yield and N accumulation in Xin-Li-Cheng (A) in Qian-an (B). Bars indicate the value of $LSD_{0.05}$.

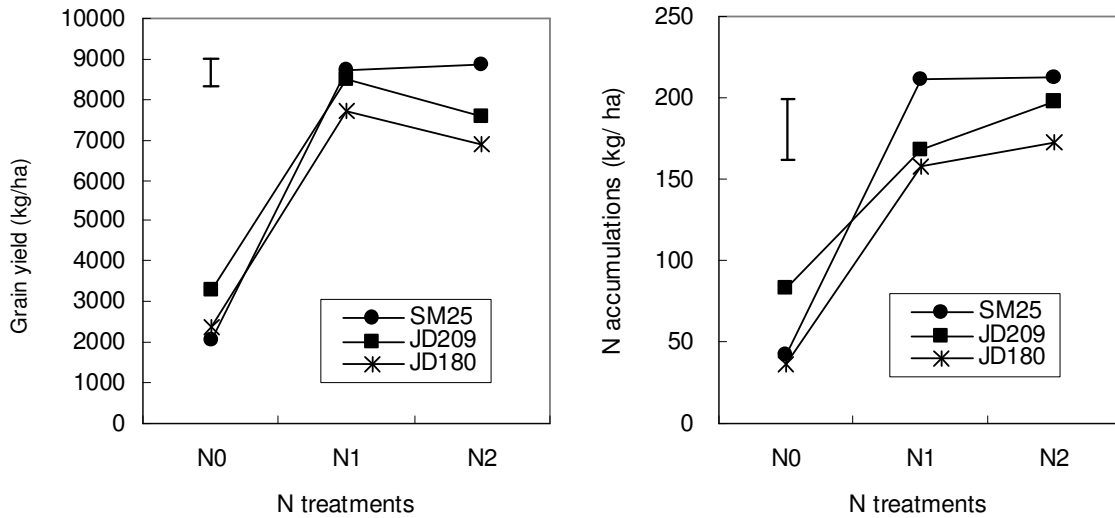
in 2004, but not in 2005 (Figures 3 and 4). In both sites, JD209 got higher yield at N0 and N1 treatments than the other two genotypes in 2004. At N2 treatment, the yield of JD209 was higher than the other two genotypes in Qian-an and was similar to that of SM25 in Xin-Li-Cheng. In general, JD209 accumulated more N at N0 treatment than the other two genotypes. JD180 got the lowest yield at N1 and N2 treatments in Qian-an in both years. These data suggest that precipitation may have a strong effect on the response of maize genotypes to N application. JD209 is adaptive to either N stress, drought or soil constraints. Therefore, it got higher yield across years, N

input and locations. SM25 seems adaptive to drought and constraints, but not low N stress. Therefore, it performed well at N1 and N2 treatments in 2004 only. JD180 was the least tolerant cultivar which yield was most susceptible to low N, water and soil constraints.

Root size of different genotypes

Root characters are a fundamental factor in adaptation to various abiotic stresses. To understand the mechanism for the genotypic difference of yield variation in different

2004



2005

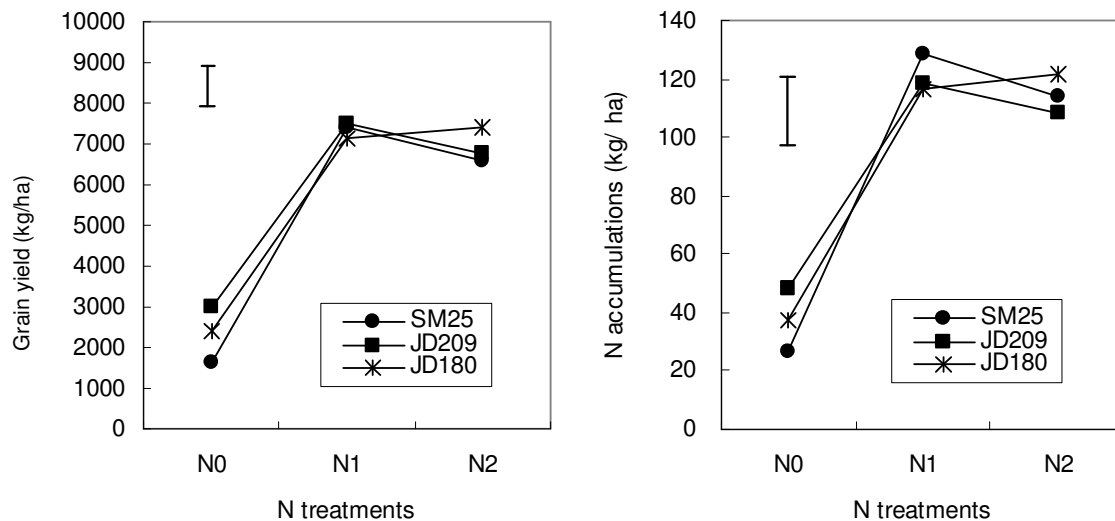


Figure 3. Genotypic difference in maize yield and N accumulation in 2004 and in 2005 in Xin-Li-Cheng. Bars indicate the value of LSD_{0.05}.

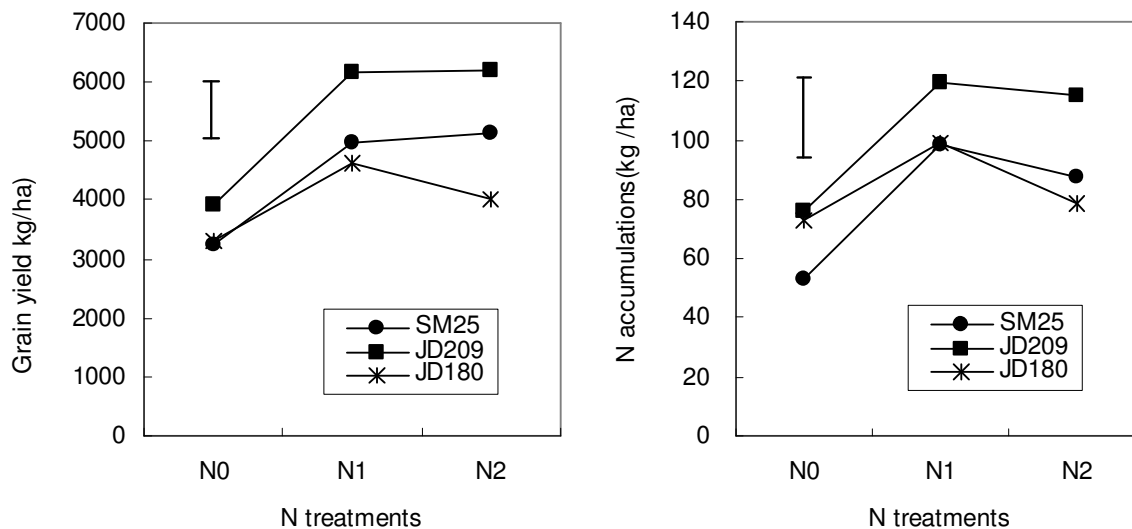
environments, the root size of the 3 three genotypes was investigated in Xin-Li-Chen in 2004. It was shown that the root size of JD209, as shown by the root dry weight at silking stage, was much larger than that of the other two genotypes, especially at the optimum N application (N2) treatment (Figure 5). Root size was reduced both at N0 and N2 in JD209 and SM25, suggesting that both N deficiency stress and overdose of N input had a negative effect on root growth. The root size of JD180 was small, but seemed not sensitive to variation in N applies. Across the N and genotype treatments, there is a significant

positive correlation between root dry weight and grain yield ($r = 0.7507$, $P < 0.01$).

Nitrogen utilization efficiency

Nitrogen utilization efficiency (NUE) is not significantly different among genotypes and sites (Table 3). As expected, there was significant difference in NUE among N treatments (Table 3), with higher NUE at low-N conditions (Table 4). In addition, NUE in 2004 was

2004



2005

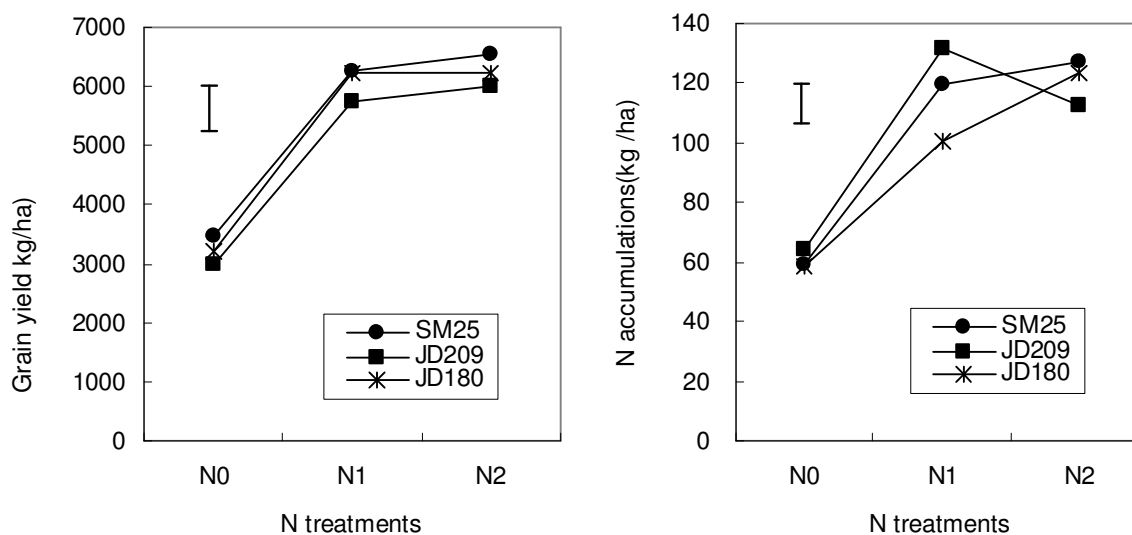


Figure 4. Genotypic difference in maize yield and N accumulation in Qian-an in 2004 and in 2005. Bars indicate the value of $LSD_{0.05}$.

significantly lower than that in 2005 (Table 4), suggesting that NUtE was much affected by weather conditions. Low NUtE in 2004 might be closely related to the less rainfall.

DISCUSSION

Using N-efficient genotypes has been suggested as one of the ways to increase N fertilizer use efficiency in crops. In theoretical research, evaluation of genotypes for N

efficiency was generally conducted under uniform experimental conditions where only N supply is a variant. In field conditions, however, there are strong interactions between N availability and other environmental constraints, such as soil characters, water supply etc. In this case, the efficiency for a genotype to use N fertilizer, which is largely determined by final grain yield, is unlikely to be only determined by its ability to take up N from the soil and subsequently utilize N efficiently in plant for grain production. In CIMMYT, breeding for drought and low-N

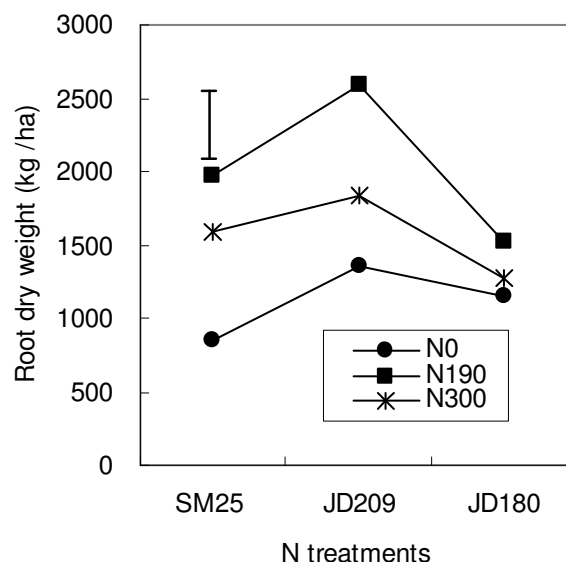


Figure 5. Root size of 3 maize genotypes in response to N inputs. Roots were sampled at anthesis stage in Xin-Li-Cheng, in 2004. Bars indicate the value of $LSD_{0.05}$.

Table 4. Genotypic difference in N utilization efficiency of 4 maize hybrids grown in Xin-Li-Cheng and Qian-an in 2004 and 2005.

Experimental location	Genotype	Rate of N fertilizer (kg N /ha)					
		2004			2005		
		N0	N1	N2	N0	N1	N2
Xin-Li-Cheng	SM25	49.7	41.5	41.8	61.7	59.5	60.7
	JD209	41.0	51.0	38.4	63.6	62.8	62.9
	JD180	64.5	49.2	40.3	66.0	62.6	60.7
Qian-an	SM25	61.2	50.6	60.1	58.9	52.3	52.0
	JD209	60.6	52.6	54.6	59.4	48.4	55.3
	JD180	45.0	46.6	51.6	55.6	57.4	50.6
Yearly effect	Average		50.0			58.4	
	$LSD_{0.05}$	3.0					

tolerance is closely related (Bänziger et al., 2000). In the present study, a strong genotype \times environment interaction was shown in controlling grain yield formation in response to N supplies. In comparison to 2005, the genotypic difference in response to N supplies was more significant in 2004 when the water supply was less (Figure 4). JD209 got higher yield at zero-N treatment (N0) than the other two genotypes. However, in 2005 no difference was found between the 3 genotypes. Although, JD209 had a high ability to accumulate N at N0 treatment (Figure 3 and 4), genotypic difference in yield could not be fully explained by N uptake. For example, JD180 accumulated the same amount of N as JD209 at N0 treatment in Qian-an in 2004, but the yield of JD180 was

much lower than that of JD209 (Figure 4). Therefore, the low-N tolerance character of JD209 is likely related to its ability in drought tolerance. It was found that, both soil water deficit and soil nitrate deficiency can induce stomatal closure and cause reductions in leaf growth rates in plants (McDonald and Davies, 1996). Wilkinson et al. (2004) found that nitrate signaling in soil had an effect on stomata and leaf growth through its interactions with soil drying, abscisic acid (ABA) and xylem sap pH in maize. This provides physiological evidence in explaining why low-N tolerance is closely related to drought tolerance in maize and can be selected simultaneously (Bänziger et al., 2000). In addition, genotypic difference was more profound under light chernozem soil conditions

(Figure 2) possibly because the soil was more sandy (Agricultural Programming Department of Qian'an country, 1984) and therefore, the possibility of N leaching is higher. Without water shortage, the yield advantage of JD209 was only shown under light chernozem soil conditions in Qian-an (Figure 2), suggesting that low-N tolerance in JD209 is also related to its ability to adapt to adverse soil conditions.

The yield stability of JD209 at varied soil and climate conditions in the present study may be explained by its large root system (Figure 6). Under field conditions, many studies highlight the essential role of root traits in N acquisition (Mackay and Barber, 1986; Wiesler and Horst, 1994), though there are different opinions (Robinson and Rorison, 1983; Robinson et al., 1991). To deal with a mid-season drought, Matthews et al. (1990) also suggested that a more intensive root growth and hence, the extraction of soil moisture from deeper layers is crucial. Water shortage and low-N limitation may happen at the same or different growth stage in field conditions. Therefore, a genotype with a large root system can be an insurance for yield formation at varied and hardly expected, environmental conditions.

In conclusion, environmental conditions like soil types and weather conditions have a strong effect on the genotypic difference in maize yield in response to N input. Identification of N-efficient cultivars should be conducted under multiple environments. Selection under favorable soil conditions with only difference in N supply may not result in the genotypes that would perform well under a variety of climate and/or soil conditions. Selection for drought tolerance may simultaneously improve N-efficiency.

ACKNOWLEDGEMENTS

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