

# Determination of the critical soil mineral nitrogen concentration for maximizing maize grain yield

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## Abstract

**Background and aims** A critical soil mineral nitrogen concentration ( $N_{\min}$ ) for guiding fertilizer application and maximizing maize grain yield is needed.

**Methods** A three-year field experiment with three N regimes, unfertilized (N0), optimized N management (Opt.) and conventional N practice (Con.) was performed in maize.

**Results** The mean soil  $N_{\min}$  in 0–60 cm soil profile for N0, Opt. and Con. treatments was 2.0, 6.7 and 8.9 mg kg<sup>-1</sup> at V8–VT growth stages and 2.2, 6.1 and 11.2 mg kg<sup>-1</sup> on average over the whole growth season, respectively. Correspondingly, the soil N supplying capacity (soil  $N_{\min}$  content+fertilizer N) of the three N treatments was smaller, identical or greater than the plant N accumulation at different growth stages. The Opt. treatment had significantly higher N use efficiency, N recovery efficiency and N partial factor productivity compared with the Con. treatment, while it did not cause maize yield loss.

**Conclusions** Compared with the insensitivity of the critical shoot N dilution curve to excessive N application,

soil  $N_{\min}$  showed strong response to all treatments. We propose a minimum of soil  $N_{\min}$  of 6.1 mg kg<sup>-1</sup> at the sowing–V8, 6.7 mg kg<sup>-1</sup> at the V8–VT, and 5.5 mg kg<sup>-1</sup> at the VT–R6 growing stages with an average of about 6 mg kg<sup>-1</sup> of soil  $N_{\min}$  in the 0–60 soil depth for maximizing maize yield and N use efficiency in northern China. To maintain this critical  $N_{\min}$  value over the whole growth period, N topdressing at V8 and V12 stages was recommended.

**Keywords** N use efficiency · Critical soil mineral N concentration · N dilution curve · Soil N supplying capacity · *Zea mays*

## Introduction

Maize, one of the most important crops that devote to global cereal production, reached a total production of 820 million tons worldwide in 2009 (Food and Agricultural Organization 2012). Maize is generally considered to have a high soil fertility requirement to attain maximal yield (Paponov and Engels 2003). Efficient utilization of applied nitrogen (N) fertilizer in maize is necessary to maximize producers' economic returns and to reduce negative soil and environmental effects (Ma et al. 1999). Nitrogen deficiency usually causes reduction of leaf area, leaf photosynthetic rates and thus final biomass and grain yield (Boomsma et al. 2009; Uribebarrea et al. 2009; Ciampitti and Vyn 2011). However, overuse of N fertilizers does not significantly increase grain yield but decreases nitrogen use efficiency (NUE), and adversely affects environmental quality and human well-being (Ju et al. 2009; Vitousek et al

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2009). In China, a 71 % increase in total annual grain production from 1977 to 2005 was achieved at the cost of 271 % increase in N fertilizer application over the same period (Ju et al. 2009). The yearly total input of N fertilizer in a wheat-corn double-cropping system in northern China was three times more than that for maize production in the mid-west of the United States in recent years (Vitousek et al. 2009). In addition, farmers in China get used to apply all N fertilizers before sowing without top dressing in later developmental stages (Li and Wang 2006). This is, however, insufficient to supply adequate N for maize growth due to N volatilization and leaching to the deeper soil profile, as well as the varied N demand at different growth stages. Numerous studies have been undertaken to optimize N fertilizing time and rate to reduce N input while maintaining crop yield in Chinese farming systems (Chen et al. 2006; Zhao et al. 2006; Cui et al. 2008; Ju et al. 2009; Peng et al. 2012a).

A critical N dilution curve (defined as the minimum %N in shoots required to produce the maximum aerial biomass at a given time) for maize plants has been proposed to assess plant N status during the whole growth season (Greenwood et al. 1990; Plénet and Lemaire 2000). The question was whether there existed a critical soil mineral N concentration ( $N_{\min}$ ) during the maize whole growing period and what parameter could serve as an indicator for guiding N fertilizer application. In a previous study (Peng et al. 2012a), we proposed an optimized N supply strategy controlling total amount of the applied N fertilizer and synchronizing maize N demand and soil N supply, in order to maintain a superior root growth for efficient nutrient uptake and maximal grain yield. Soil N distribution is temporally and spatially heterogeneous in the soil profile and is influenced by fertilization, plant uptake and precipitation (Addiscott and Darby 1991). Since whole-plant N status prior to silking has a predominant effect on final grain yield (Ciampitti and Vyn 2011, 2012), sufficient soil  $N_{\min}$  supply during the late vegetative growth stage is important for robust plant growth and development, while post-silking N top dressing is unnecessary (Subedi and Ma 2005a, b; Peng et al. 2012a).

In the present work, a three-year field experiment with three N regimes was performed and soil  $N_{\min}$  in top soil (0–30 cm) and sub-top soil (30–60 cm) at critical developmental stages of maize plants was determined. In addition, due to the heterogeneous N distribution in the soil profiles (Addiscott and Darby 1991), the reliability and representativeness of estimates of  $N_{\min}$  values using

auger method depend on the number of samples and their locations. In comparison, whole soil block excavation is a better method to avoid over or under estimation of soil  $N_{\min}$  values. The influence of two methodologies of soil sampling in the field on soil  $N_{\min}$  values was studied. The purpose of this study was to obtain a critical soil  $N_{\min}$  value, and to provide a theoretical base for improvement of N management in Chinese intensive cropping systems.

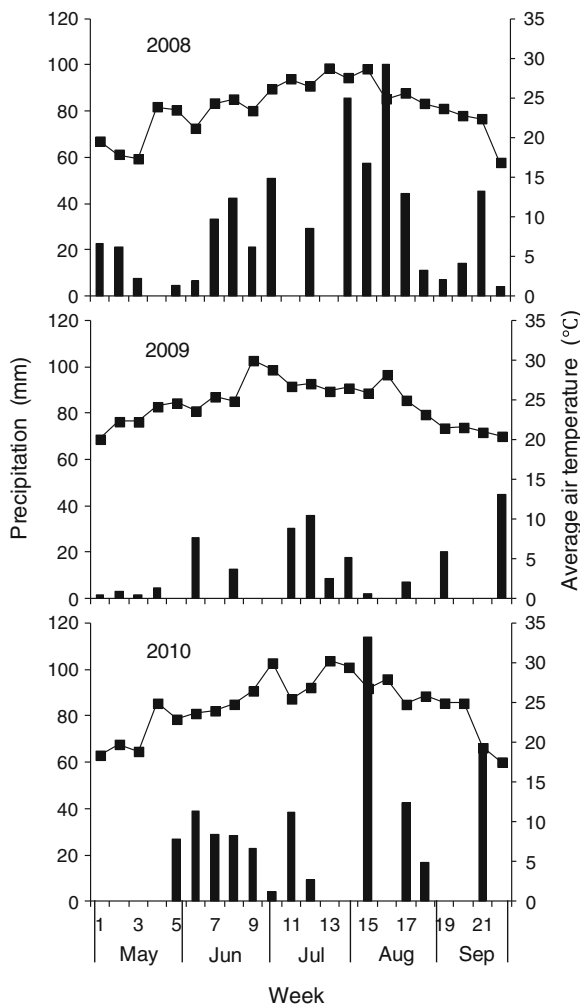
## Materials and methods

### Experimental design

A three-year field experiment (2008–2010) was conducted in three adjacent plots at the Shangzhuang Experimental Station of the China Agricultural University, Beijing (40° N; 116° E). The soil type at the study site is a calcareous alluvial soil with a loamy and silt texture (FAO classification) typical of the region. The chemical properties of the 0–30 cm soil layer of the study site in 2008, 2009 and 2010 were the same as described by Peng et al. (2012b). Briefly, the top soil layer (0–30 cm) contained 11.5, 7.3 and 14.8 g kg<sup>-1</sup> OM, and 0.83, 0.71 and 0.94 g kg<sup>-1</sup> total N with soil pH of 8.0, 7.86 and 7.74 in water during the 3 years. Maize hybrid DH 3719 was sown on 27 April 2008, 27 April 2009 and 29 April 2010, and harvested on 19 September 2008, 21 September 2009 and 4 October 2010, respectively. Maize was over-seeded with hand planters and was thinned at the seedling stage to a stand of 100,000 plants ha<sup>-1</sup>. The intra-row distance was 28 cm and the inter-row distance was 50 cm (wide row) alternating with 20 cm (narrow row). The seeds were sown inter-laced between the rows. The plot sizes were 40 m<sup>2</sup>, 56 m<sup>2</sup>, and 48 m<sup>2</sup> in 2008, 2009 and 2010, respectively. The study was arranged as a randomized complete block design with four replications. Border plots and weed control were the same as described by Peng et al. (2012a, b). The weekly precipitation during the maize growing season was shown in Fig. 1.

### Fertilization and treatments

There were three (2008 and 2009) or two (2010) N treatments: 1) N0 as control, which had no N fertilizer application in 3 years; 2) Optimized N management (250 kg N ha<sup>-1</sup>, Opt.); the N rate and time was



**Fig. 1** Weekly precipitation during maize growing season in three years

determined according to the results of N accumulation in plants and soil  $N_{\min}$  after the last harvest in the preliminary experiment in 2007.  $60 \text{ kgNha}^{-1}$  was applied as base fertilizer. The remaining N was applied before tasseling at V8 (120 kg) and V12 (70 kg), respectively. 3) Conventional N practice in 2008 and 2009 ( $450 \text{ kgNha}^{-1}$ , Con.), which was derived from numerous high-yield studies in China.  $175 \text{ kgNha}^{-1}$  was applied as base fertilizer, 50, 170, and  $55 \text{ kgNha}^{-1}$  in 2008, and 120, 70,  $85 \text{ kgNha}^{-1}$  in 2009 were applied in wide interrows by hand as topdressings at the V8, V12 and VT, respectively.

The rate and timing of phosphorus and potassium fertilization in each year were the same. In addition, zinc (Zn) was applied in each year as base fertilizer

because of the slight Zn deficiency in the experimental region. A total of  $135 \text{ kgha}^{-1}$  of  $\text{P}_2\text{O}_5$  as triple superphosphate [ $\text{Ca}(\text{H}_2\text{PO}_4)_2 \cdot \text{H}_2\text{O}$ ],  $120 \text{ kgha}^{-1}$  of  $\text{K}_2\text{O}$  as potassium sulfate [ $\text{K}_2\text{SO}_4$ ], and  $30 \text{ kgha}^{-1}$  of  $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$  were applied. Before sowing,  $90 \text{ kgha}^{-1}$   $\text{P}_2\text{O}_5$ ,  $80 \text{ kgha}^{-1}$  of  $\text{K}_2\text{O}$  and  $30 \text{ kgha}^{-1}$  of  $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$  were broadcasted and incorporated into the upper 0–15 cm of the soil by rotary tillage. Another  $45 \text{ kgha}^{-1}$  of  $\text{P}_2\text{O}_5$  at V12 and  $40 \text{ kgha}^{-1}$  of  $\text{K}_2\text{O}$  at VT were applied in wide interrows by hand as topdressings. Each topdressing (NPK) was applied after plant sampling.

#### Plant and soil sampling and N measurement

Plants were harvested on 53 (V8), 71 (V12), 86 (tasseling, VT) and 111 (blister stage, R2) days after sowing (DAS) before fertilization and 130 and 145 (physiological maturity, R6, when 50 % of the plants showed black layer formation in the grains from the mid-portion of the ears) DAS in 2008, on 33, 45 (V8), 61 (V12), 80 (VT), 110 (R2) and 147 (R6) DAS in 2009, and on 50 (V8), 78 (VT), 105 (R2) and 154 (R6) DAS in 2010. Harvest procedures and N content analyses in each plant sample were the same as described by Peng et al. (2012a, b). To estimate grain yield, ears in the central area of  $14 \text{ m}^2$  (2008),  $21 \text{ m}^2$  (2009) and  $19 \text{ m}^2$  (2010) within each experimental plot were hand-harvested at physiological maturity to get fresh weight. Six randomly selected ears from each harvesting area were shelled. All kernels were dried at  $60^\circ \text{C}$  in an oven to constant weight, and then the ratio of kernels to ears was determined and kernel moisture was calculated. The final grain yield was adjusted to 15.5 % moisture content.

After aboveground plant excision at each harvest, a soil volume of  $28 \text{ cm} \times 35 \text{ cm}$  and a total depth of 60 cm with 30 cm increment in each plot was dug out, thus two soil blocks with dimensions  $28 \text{ cm} \times 35 \text{ cm} \times 30 \text{ cm}$  were obtained per plot (soil excavation method). The area of  $28 \text{ cm} \times 35 \text{ cm}$  was the soil surface occupied by each plant at the plant density of  $100,000 \text{ plants ha}^{-1}$ . In addition, at each harvest in 2008, five 2-cm-diameter soil cores per plot were collected and mixed to measure soil  $N_{\min}$  (auger method, Böhm 1979). Samples were collected from the 0–60 cm soil layers (in 30 cm increments) in the interrow area (Cui et al. 2008). All visible roots in each soil sample obtained by the two methods were picked out by hand,

and then the soil sample was ground and passed through a 3 mm sieve in the field. A representative sample of the mixed soil was placed in a marked plastic bag, and extracted immediately after transfer to the laboratory with 0.01 molL<sup>-1</sup> CaCl<sub>2</sub> solution and analyzed for soil N<sub>min</sub> (NH<sub>4</sub><sup>+</sup>-N+NO<sub>3</sub><sup>-</sup>-N) by continuous

flow analysis (TRAACS 2000, Bran and Luebbe, Norderstedt, Germany).

Since the bulk density of 0–30 cm and 30–60 cm soil layer is not identical, 1.44 gcm<sup>-3</sup> and 1.51 gcm<sup>-3</sup>, respectively, the soil N<sub>min</sub> concentrations of 0–60 cm soil was calculated as following:

$$N_{\min(0-60)} = \frac{N_{\min(0-30)} \times 1.44 \times V_{0-30} + N_{\min(30-60)} \times 1.51 \times V_{30-60}}{\frac{1.44+1.51}{2} \times V_{0-60}}$$

Where N<sub>min (0–30)</sub>, N<sub>min (30–60)</sub> and N<sub>min (0–60)</sub> represent soil N<sub>min</sub> concentrations of 0–30 cm, 30–60 cm and 0–60 cm soil depth, respectively. V<sub>0–30</sub>, V<sub>30–60</sub> and V<sub>0–60</sub> represent the soil volumes of 0–30 cm, 30–60 cm and 0–60 cm soil depth, respectively.

The NUE, N recovery efficiency (NRE) and N internal efficiency (NIE) was calculated according to Ciampitti and Vyn (2011) as follows:

$$NUE = (GY_{\text{fert.}} - GY_{\text{unfert.}}) / \Delta N_{\text{applied}}$$

where GY<sub>fert.</sub> and GY<sub>unfert.</sub> are the per-unit-area grain yield in N applied treatment and no N treatment, ΔN<sub>applied</sub> is the total amount of N applied in Opt. and Con. treatments. Similarly, the NRE was calculated as:

$$NRE = (Nupt_{\text{fert.}} - Nupt_{\text{unfert.}}) / \Delta N_{\text{applied}}$$

where Nupt<sub>fert.</sub> is N uptake in the fertilized plot and Nupt<sub>unfert.</sub> is N uptake in unfertilized plot. The NIE was calculated as:

$$NIE = (GY_{\text{fert.}} - GY_{\text{unfert.}}) / (Nupt_{\text{fert.}} - Nupt_{\text{unfert.}})$$

The N partial factor productivity (PFP<sub>N</sub>) was followed to Cui et al. (2008):

$$PFP_N = GY_{\text{fert.}} / \Delta N_{\text{applied}}$$

### Statistical analysis

Data of each year were analyzed separately using analysis of variance with the SAS package (SAS

Institute, 1996). Differences between data in all tables were tested with PROC ANOVA. Nitrogen treatments were treated as fixed effects and least significant difference (LSD) test at P<sub>0.05</sub> was used to separate treatment means exhibiting significant differences.

### Results

Aboveground plant dry weight, total N, grain yield, and N efficiency indices

Nitrogen deficiency occurred in N0 and significantly reduced maize aboveground plant dry weight (DW), total N, and grain yield at the final harvest in all 3 years. However, conventional N practice (Con.) failed to further increase the above three measures compared with the optimized N treatment (Opt.) in 2008 and 2009. The Opt. treatment significantly increased NUE, NRE and PFP<sub>N</sub>, while it had no influence on NIE, compared with the Con. treatment (Table 1). During the whole growth period the highest N uptake rate (kgNha<sup>-1</sup> d<sup>-1</sup>) occurred in robust vegetative growth stage (V8–VT) regardless of N applications. Significant difference in N uptake rate between N0 and the N applied treatments was observed only in vegetative growth stage (Table 2).

Critical soil N<sub>min</sub> concentration, soil N supplying capacity and aboveground plant N accumulation during different growth periods, and N dilution curves

Soil N supplying capacity in Fig. 2 was the sum of the measured soil N<sub>min</sub> content plus the applied N fertilizer at the beginning of a certain growth interval (soil

**Table 1** Above ground plant dry weight and total N, grain yield, NUE, NRE, NIE and PFP<sub>N</sub> of maize, and N fertilizer applied in different N treatments in 3 years

Year	Treatment	Above ground plant dry weight (Mgha <sup>-2</sup> )	Above ground plant total N (Mgha <sup>-2</sup> )	Grain yield (Mgha <sup>-2</sup> )	NUE <sup>a</sup>	NRE <sup>b</sup>	NIE <sup>c</sup>	PFP <sub>N</sub> <sup>d</sup>	Total fertilizer N (kg ha <sup>-1</sup> )
2008	N0	20.6 b	0.18 b	11.0 b	–	–	–	–	0
	Opt.	27.2 a	0.27 a	13.8 a	11.1 a	0.39 a	34.9 a	55.2 a	250
	Con.	25.6 ab	0.25 ab	13.1 a	4.7 b	0.17 b	30.8 a	29.2 b	450
2009	N0	14.9 b	0.11 b	6.3 b	–	–	–	–	0
	Opt.	20.6 a	0.20 a	10.7 a	17.5 a	0.37 a	49.0 a	42.8 a	250
	Con.	21.2 a	0.21 a	11.0 a	10.3 b	0.22 b	47.4 a	24.4 b	450
2010	N0	15.6 b	0.17 b	6.2 b	–	–	–	–	0
	Opt.	23.8 a	0.23 a	9.0 a	11.5	0.23	40.1	36.2	250

Values within a column in each year followed by a different letter represent a significant difference between N treatments ( $P < 0.05$ )

<sup>a</sup>N use efficiency (kg grain kg<sup>-1</sup> N applied)

<sup>b</sup>N recovery efficiency (kg N uptake kg<sup>-1</sup> N applied)

<sup>c</sup>N internal efficiency (kg grain kg<sup>-1</sup> N uptake)

<sup>d</sup>N partial factor productivity (kg grain kg<sup>-1</sup> N applied)

N<sub>min</sub> content + fertilizer N); and shoot N accumulation was N currently taken up during this growth interval. Soil N supplying capacity over the growing season decreased with the prolonged growth period, regardless of N applications; while plant N accumulation during different growth periods increased from the first harvest, reached the highest value between V12 and R2 stages, and then decreased (Fig. 2). For N0

**Table 2** Aboveground plant N uptake rate (kg ha<sup>-1</sup> d<sup>-1</sup>) during different growth periods of maize supplied with different N rates in 3 years

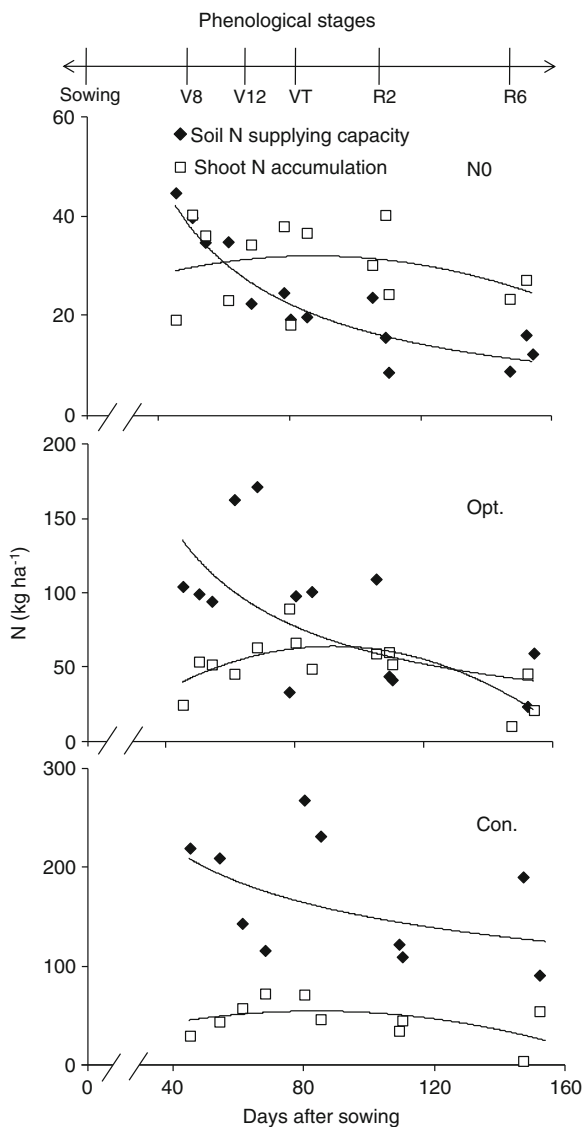
Year	Treatment	Growth period				
		Sowing-V8	V8-V12	V12-VT	VT-R2	R2-R6
2008	N0	0.69 b	1.92 b	2.46 b	1.62 a	0.81 a
	Opt.	0.99 a	3.55 a	3.3 a	2.43 a	1.36 a
	Con.	0.84 ab	4.04 a	3.12 a	1.9 a	1.12 a
2009	N0	0.43 b	1.46 c	0.97 b	0.82 a	0.64 a
	Opt.	0.56 a	2.88 b	3.53 a	1.75 a	0.29 a
	Con.	0.67 a	3.63 a	3.78 a	1.52 a	0.12 a
2010	N0	0.82 b	2.23 b		1.15 a	0.45 a
	Opt.	1.08 a	3.22 a		2.22 a	0.44 a

Values within a column in each year followed by a different letter represent a significant difference between N treatments ( $P < 0.05$ )

treatment, soil N supplying capacity was below the aboveground plant N accumulation curve after V8 stage owing to soil N<sub>min</sub> depletion by plants. The Opt. treatment had an overlap of soil N supplying capacity and aboveground plant N accumulation, indicating that soil N supply met plant demand for N over the whole growth period. In comparison, soil N supplying capacity for Con. treatment was farther above the aboveground plant N accumulation during the whole growth period (Fig. 2), due to the excessive N fertilizer application. The results were also confirmed by the calculated N balance after harvest (Table 3). Negative N balance of the N0 treatment in 3 years indicated a strong soil N depletion during the growth season; while a large amount of N surplus of the Con. treatment in 2008 and 2009 revealed an excessive N application.

Soil N<sub>min</sub> value of the N0 treatment was the lowest among three N treatments and showed little change in 0–30 cm soil layer during the whole growth period in 3 years. In comparison, Opt. N treatment had a larger N<sub>min</sub> change only in 0–30 cm soil layer; whereas Con. N treatment had the highest N<sub>min</sub> value and change even in 30–60 cm soil layer in 2008 (Fig. 3) owing to heavy rainfall (Fig. 1). Because soil N<sub>min</sub> is easily influenced by precipitation, it is reasonable to use the average soil N<sub>min</sub> of 0–60 cm soil profile for calculating





**Fig. 2** Changes in soil N supplying capacity (soil  $N_{\min}$  content + fertilizer N) and aboveground plant N accumulation during different growth periods at three N levels. Soil N supplying capacity was the sum of the measured soil  $N_{\min}$  content plus the applied N fertilizer at the beginning of a certain growth interval; and shoot N accumulation was N currently taken up during this growth interval. Sowing time and different phenological stages during the maize growing season were indicated on the top of the figure

the soil  $N_{\min}$  concentrations. The average soil  $N_{\min}$  concentrations for 0 N, Opt. and Con. treatments of the 3 years were 2.2, 6.1 and 11.2  $\text{mg kg}^{-1}$ , respectively. In addition, the 0–60 cm soil  $N_{\min}$  was 3.4, 2.0 and 1.6  $\text{mg kg}^{-1}$  for N0 treatment, 6.1, 6.7, and 5.5  $\text{mg kg}^{-1}$  for Opt. treatment, and 10.9, 8.9, 13.1  $\text{mg kg}^{-1}$  for Con. treatment during the three different growth intervals,

**Table 3** The calculated N balances ( $\text{kg ha}^{-1}$ ) for N0, Opt. and Con. treatments across the 3 years

		N0	Opt.	Con.
2008	N fertilizer input	0	250	450
	$N_{\min}$ before sowing	35 a	35 a	35 a
	$N_{\min}$ after harvest	17 b	25 b	57 a
	Plant N uptake	176 b	273 a	254 ab
N balance		-158	-13	174
2009	N fertilizer input	0	250	450
	$N_{\min}$ before sowing	45 a	45 a	45 a
	$N_{\min}$ after harvest	4 c	63 b	127 a
	Plant N uptake	109 b	202 a	210 a
N balance		-68	30	158
2010	N fertilizer input	0	250	450
	$N_{\min}$ before sowing	40 a	40 a	40 a
	$N_{\min}$ after harvest	10 b	35 a	35 a
	Plant N uptake	165 b	226 a	226 a
N balance		-135	29	29

Values within a row followed by a different letter represent a significant difference between N treatments ( $P < 0.05$ )

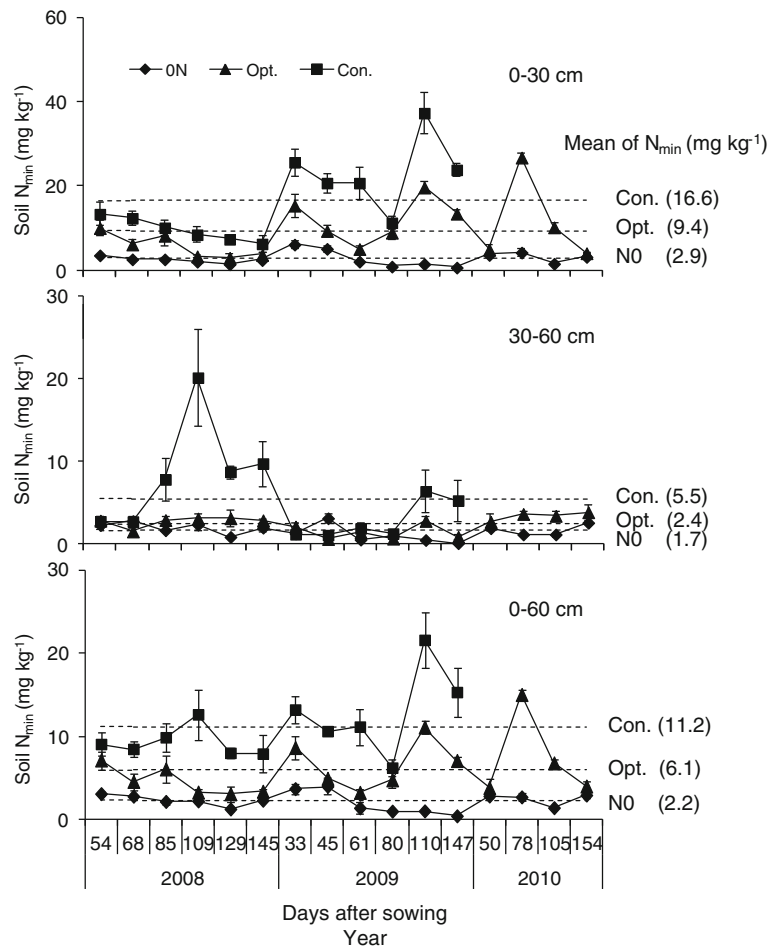
sowing to V8, V8 to VT and VT to R6, respectively (calculated according to the results of Fig. 3).

Aboveground plant N concentration of N0 plants was lower than that of the critical N dilution curve for maize reported by Plénet and Lemaire (2000); while that of both Opt. and Con. treated plants was close to the maize critical N dilution curve and overlapped across the 3 years (Fig. 4).

#### Comparison of $N_{\min}$ value of soils obtained by excavation and auger methods

The  $N_{\min}$  values of soils obtained by soil excavation and auger method in the same plots in 2008 were compared. The differences of the soil  $N_{\min}$  of both 0–30 and 30–60 cm soil layers obtained by the two methods for N0 treatment were very small among different sampling times. In comparison with the stable soil  $N_{\min}$  of 0–30 and 30–60 cm soil layer obtained by soil excavation method for Opt. and Con. treatments, the  $N_{\min}$  value of the soil obtained by auger method was obviously higher and very variable, especially in 0–30 cm soil layer after N topdressing. The differences of the soil  $N_{\min}$  between the two methods decreased in 0–30 cm soil layer but remained large in 30–60 cm soil layer in late growth stages (Fig. 5).

**Fig. 3** Changes in soil  $N_{min}$  of 0–30, 30–60 and 0–60 cm soil profile at different growth stages with three N levels in 3 years. The bars represented the standard error of the mean,  $n=4$ . Dash lines and numbers in the bracket represented the average soil  $N_{min}$  values of all data points of the same N treatments in 3 years



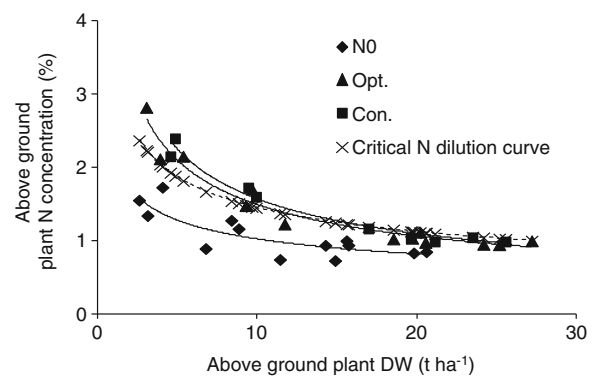
**Discussion**

Optimized N management significantly increased NUE, NRE and  $PPF_N$  compared with conventional N application

In China, farmers and even some agronomists believe that amounts of N greatly exceeding N rates of the Opt treatment can further increase crop yield. One reason for this misunderstanding is the lack of related knowledge for the relationship between soil fertility and plant N uptake (Ju et al. 2009). In fact, excessive N application not only failed to increase above ground plant total biomass and grain yield of maize plants, but also significantly reduced N efficiency and  $PPF_N$  (Table 1; Boomsma et al. 2009).

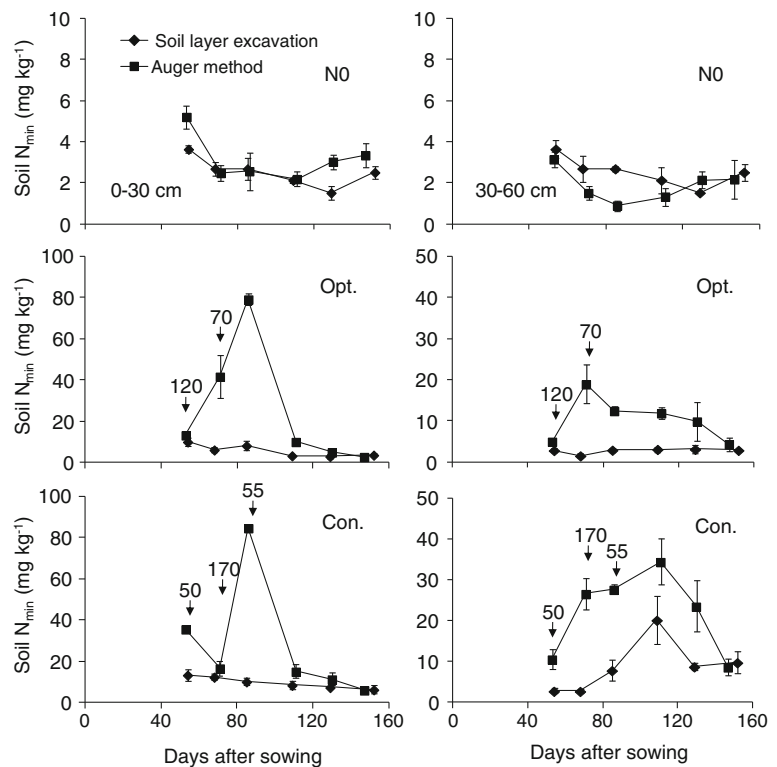
The NUE is related to both soil and plant processes. To better understand the grain NUE variability, it is necessary to re-examine the two main components of

NUE: NIE (which is associated with plant N conversion or utilization efficiency to final grain yield) and NRE (which is connected to plant N uptake efficiency) (Salviaggiotti et al. 2009; Ciampitti and Vyn 2011). The



**Fig. 4** N dilution curves of the three N treatments across the 3 years. Dashed lines in the figures represented the critical N dilution curves proposed by Plénet and Lemaire (2000)

**Fig. 5** Comparison of  $N_{\min}$  values of the soils collected by soil excavation and auger methods in 2008. The *left* and *right* panel showed the results of 0–30 and 30–60 cm soil layer, respectively. The *bars* represented the standard error of the mean,  $n=4$ . *Numbers* and *arrows* indicated the rates ( $\text{kgNha}^{-1}$ ) and times of N topdressings



NUE declined with the increased N application (Table 1), which was consistent with the previous studies (Ladha et al. 2005; Uribelarrea et al. 2007; Ciampitti and Vyn 2011). Ladha et al. (2005) summarized 61 sets of data of maize experiments across different regions of the world and showed that NUE averaged approximately  $24.2 \text{ kg grain kg}^{-1} \text{ N applied}$ , which was about twice the value found in the current study ( $11 \text{ kg grain kg}^{-1} \text{ N applied}$  as shown in Table 1). Ciampitti and Vyn (2011) also observed a low NUE from their two-location field research in Indiana of the United States, with average NUE of  $12 \text{ kg grain kg}^{-1} \text{ N applied}$ . The amount of the applied N fertilizer in Opt. treatment in the present study was  $250 \text{ kgNha}^{-1}$ , which was considerably higher than that in Illinois of the United States, only  $155 \text{ kgNha}^{-1}$  input (Vitousek et al 2009). However, the whole maize aboveground plant was removed from the field after grain harvest in our study in contrast to stalk return to the field in Illinois (Vitousek et al 2009). This could largely explain the reduced N application rate and significantly increased NUE in Illinois. Moreover, maize NUE declines with N fertilizer rate exceeding  $200 \text{ kgha}^{-1}$  (Dobermann and Cassman 2004). More work needs to be done in China to teach and train the farmers for saving

N fertilizer although there has been large progresses in the improvement of gain yield and reduction of fertilizer application in the recent decade (Chen et al. 2006; Zhao et al. 2006; Cui et al. 2008; Ju et al. 2009; Peng et al. 2012a).

The Opt. treatment also increased the NRE but did not significantly influenced NIE compared to Con. treatment (Table 1). The NRE is a reflection of root growth and high NIE is closely linked to the grain components (Ciampitti and Vyn 2011). Our recent work demonstrated that Opt. treatment was beneficial for root growth (Peng et al. 2012a) and thus led to a high NRE. Nevertheless, Opt. treated plants had similar total N uptake (Table 1), grain kernel number and weight compared with the Con. treated plants (data not shown), thus these two treatments resulted in similar NIE. The  $\text{PFP}_N$  reflects yield production per-unit-N fertilizer application, which integrates the use efficiency of both soil N and applied fertilizer N resources (Ladha et al. 2005). The Opt. N treatment decreased N fertilization application by 44 % and caused 75–89 % increase in  $\text{PFP}_N$  compared with Con. N treatment (Table 1). This is particularly important for improving NUE in China's cropping systems.



## Soil $N_{\min}$ responded sensitively to both N deficiency and excessive N application

Considering the diverse distribution of root and soil  $N_{\min}$  in the soil profile and the influence of precipitation (Fig. 3), using average  $N_{\min}$  value in the 0–60 cm soil layer was more appropriate to calculate the critical soil  $N_{\min}$ . This critical soil  $N_{\min}$  value for achieving maximal maize grain yield, averaged across the three experimental years, was about  $6 \text{ mg kg}^{-1}$ . Moreover, the critical soil  $N_{\min}$  values were 6.1, 6.7, and  $5.5 \text{ mg kg}^{-1}$  for the growth intervals, sowing to V8, V8 to VT and VT to R6, respectively. Here we used soil  $N_{\min}$  value of the optimized N management as the critical soil  $N_{\min}$  value because maize grown under this N management received a sufficient but not excessive N supply. This was supported by the overlapped soil N supplying capacity and the aboveground plant N accumulation of the Opt. treatment during the whole growth period (Fig. 2). The calculated N recovery after harvest also indicated a balanced soil N supply and plant N uptake for the Opt. treatment in 3 years (Table 3). Currently available critical shoot N dilution curve, an important indicator for plant growth, has been proposed as a diagnostic tool of shoot N status in  $C_3$  and  $C_4$  plants (Greenwood et al. 1990) such as winter wheat (Justes et al. 1994), winter oil seed rape (Colnenne et al. 1998), and maize (Plénet and Lemaire 2000). It seemed, however, that critical shoot N dilution curve could only reflect N deficiency but not excessive N supply. As shown in Fig. 4, N dilution curves for both Opt. and Con. treatments were near the critical N dilution curve but overlapped, since Con. treatment could not stimulate more N uptake than optimized N management (Table 1). However, soil N supplying capacity of the Con. treatment was far above the plant N accumulation curve, indicating excessive N application (Fig. 2; Table 3) that would increase the risk of N leaching and potential environmental pollution. In comparison, critical soil  $N_{\min}$  value proposed in this study was sensitive and capable to indicate both N deficiency and excessive N application.

In comparison with a series of the decreasing values of the shoot N dilution curve along with the prolonged growth period, there was only one value for critical soil  $N_{\min}$ . However, it should be pointed out that this was the minimum value for achieving the maximal maize grain yield. We also proposed a minimum of soil  $N_{\min}$  of  $6.1 \text{ mg kg}^{-1}$  at the sowing–V8,  $6.7 \text{ mg kg}^{-1}$

at the V8–V12, and  $5.5 \text{ mg kg}^{-1}$  at the VT–R6 growing stages. To maintain these minimum  $N_{\min}$  values during the whole growth period, adequate and timely N topdressing according to plant demand for N was necessary. For instance, maize N uptake rate changed over the whole growth period and reached the highest values during V8–VT (Table 2). To increase the soil N supplying capacity (Fig. 2) and maintain the minimum soil  $N_{\min}$  value of  $6 \text{ mg kg}^{-1}$  (Fig. 3) during this period, two N topdressings at V8 and V12 stage for the Opt. N management were performed in the present study. No N was topdressed after silking for the Opt. N management, since the decreased plant N uptake rate and increased soil N mineralization surrounding roots during reproductive growth stage allowed a good match between soil N supplying capacity and plant demand for N. Root mortality of maize plant takes place dramatically during the reproductive growth stage due to the decreased carbon supply (Wiesler and Horst 1994; Peng et al. 2010, 2012a). On the other hand, plant roots stimulate N mineralization by changing soil microorganism activities, and altered  $N_{\min}$  concentration near the root zones (Wang and Bakken 1997). In a wheat-maize rotation system in North China Plain, the average N mineralization in no N applied treatment was  $63.5 \text{ kg ha}^{-1}$  across the eight successive cropping seasons (Zhao et al. 2006). This could also explain why the soil  $N_{\min}$  value of N0 treatment remained relatively stable during the whole growth period, in spite of the continuous N uptake by plants (Fig. 3).

The critical soil  $N_{\min}$  value proposed in the present work was lower than that of the previous studies (Fox et al. 1989; Magdoff et al. 1990; Binford et al. 1992; Klausner et al. 1993; Heckman et al. 1995; Sainz Rozas et al. 2000), which ranged from approximately 16 to  $30 \text{ mg N kg}^{-1}$ . There were two possible reasons for this difference: Firstly, N fertilizer was split-supplied in Opt. and Con. treatments. The soil samples were collected after a period of plant growth and one or 2 days before each N topdressing. It was no doubt that after N topdressing, the soil  $N_{\min}$  value would be increased. In the previous studies, however, N fertilizer was applied only once before planting as base fertilizer, and soil sample was collected only once either at 5–6 leaf stage (Fox et al. 1989; Sainz Rozas et al. 2000), or when maize plant was 15–30 cm tall (Magdoff et al. 1990; Binford et al. 1992; Klausner et al. 1993; Heckman et al. 1995). Hence, the measured

$N_{\min}$  in these experiments was the value after application of all fertilizer. Therefore these values must be higher than that proposed in our study and the  $N_{\min}$  value presumably decreased continuously during the growth period, because there was no N topdressing afterwards. Secondly, in the previous studies, all these authors used an auger method to collect soil samples, while in the present study soil samples were obtained by soil excavation method. The results in Fig. 5 demonstrated that under the condition without N supply (N0),  $N_{\min}$  value of soils obtained by the two methods was similar; while under the condition with N supply (Opt. and Con.),  $N_{\min}$  value of the soil collected by auger method was significantly higher than that collected by soil excavation method in whole 0–60 cm soil profile. Maize root and soil  $N_{\min}$  were heterogeneously distributed in the soil profile. Root length density was obviously higher in maize intrarow than that in interrow (Peng et al. 2010), which caused strong depletion of soil  $N_{\min}$  near the root rhizosphere (Peng et al. 2012b). On the other hand, N topdressing in practice was banded in interrow area. As a result,  $N_{\min}$  value of the soil located in interrow was higher than that in intrarow. Analyzing soil samples in interrow area by auger method with 2-cm-diameter soil cores overestimated  $N_{\min}$ , as indicated by Fig. 5. For example, the applied total N fertilizer at VT for Opt. and Con. treatments was 250 and 395 kg ha<sup>-1</sup>, respectively; while the total amount of N of 0–60 cm soil depth calculated based on auger method was 398 kg ha<sup>-1</sup> and 491 kg ha<sup>-1</sup>, respectively. However, soil samples obtained by soil excavation method in this study included soils distributed vertically and horizontally in the soil block occupied by each individual root in the field.

In conclusion, optimal N management maximized maize growth and grain yield via controlling the amount of N application and synchronizing crop N demand and soil N supply by split-N applications. Correspondingly, soil N supplying capacity matched plant N uptake very well over the whole growth period. In comparison, luxurious N application failed to increase maize growth and grain yield. Soil N supplying capacity was much greater than plant N uptake, resulted in a significant decrease in NUE, NRE and PFP<sub>N</sub>, and potential risk of environment pollution. Soil  $N_{\min}$  value showed strong response to both N deficiency and excessive N application. We suggest a minimum soil  $N_{\min}$  value for achieving the maximum maize grain yield as 6 mg kg<sup>-1</sup> for the whole growth stage. For different growth intervals, the critical soil  $N_{\min}$  values were 6.1, 6.7, and 5.5 mg kg<sup>-1</sup>

for sowing to V8, V8 to VT and VT to R6, respectively. Because of the dramatic changes in N uptake rate at different growth stages, N topdressing during the rapid N uptake period was recommended to maintain the minimum soil  $N_{\min}$  value in the growing season. Due to the overestimation of soil  $N_{\min}$  by auger method, all recommended soil  $N_{\min}$  values were derived from the excavation method.

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