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# Fate of <sup>15</sup>N fertilizer under different nitrogen split applications to plastic mulched maize in semiarid farmland

Shaojie Wang · Shasha Luo · Shanchao Yue · Yufang Shen . Shiqing Li

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Abstract A 2-year field microplot experiment was conducted to determine the effects of nitrogen (N) splits on grain yields and the fate of  $15N$ -labelled fertilizer applied to plastic mulched maize (Zea mays L.). Three N split applications at the same rate of 225 kg N  $\text{hm}^{-2}$  were performed. The N was applied at the day before sowing, the eight-leaf stage (V8), and the silking stage (R1) in the following ratios: 100  $\%$ -0–0 (N1), 40–60 %–0 (N2), and 40–30–30 % (N3). <sup>15</sup>N-labelled urea (10.14 atom%) was used to trace the fate of each N application in the microplots. The results showed that grain yields increased by 8.3 and 10.7 % in the treatments N2 and N3, respectively, compared with the N1 treatment. Plant N uptake

#### S. Li

College of Resources and Environmental Sciences, Northwest A&F University, Yangling 712100, China

## S. Luo

Lab of Soil Microbiology and Nutrient Cycle, Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun 130102, China

#### S. Wang

University of Chinese Academy of Sciences, Beijing 100049, China

derived from fertilizer (Ndff, %) averaged 26.8–32.4 % compared with 67.6–73.2 % derived from soils (Ndfs,  $\%$ ). Split applications of N significantly increased the Ndff in plant. The residual  $^{15}$ N in the 0–200 cm soil layer ranged from 48.3 to 51.3 % at maize harvest, approximately half of which remained in 0–20 cm soil layer. The  $15N$  recovery efficiency  $(^{15}$ NRE) was 37.5 and 39.1 % for treatments N2 and N3, respectively, and was significantly higher than that for N1 treatment (27.9 %). The potential N losses in the treatments N2 and N3 were 11.2 and 12.7 %, respectively, and were significantly lower than losses in treatment N1 (22.2 %). In conclusion, applying N with two splits could produce higher grain yields, higher NRE, and lower N losses in semiarid plastic mulched maize cropping system.

Keywords  $15N$  recovery  $\cdot$  Dryland  $\cdot$  N application  $\cdot$ Grain yield - Plastic mulching

## Introduction

Nitrogen (N) fertilizer has supported the rapidly expanding population of the world by increasing crop production during the last few decades (Tilman et al. [2011\)](#page-11-0), meanwhile the global demand for N fertilizer in agricultural systems has substantially increased (Schepers and Raun [2008](#page-10-0)). From 1960 to 2000, the use of N fertilizer increased by 800 %, with wheat,

S. Wang  $\cdot$  S. Yue  $\cdot$  Y. Shen  $\cdot$  S. Li ( $\boxtimes$ ) State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Institute of Soil and Water Conservation, Chinese Academy of Sciences and Ministry of Water Resource, Yangling 712100, China e-mail: sqli@ms.iswc.ac.cn

rice and maize accounting for about 50 % of current fertilizer use (Canfield et al. [2010\)](#page-9-0). China is the largest consumer of chemical N in the world, accounting for 32 % of the total consumption (Heffer [2009](#page-10-0)). However, excess N fertilizer and inappropriate application methods have led to low NRE and high N losses (Zhu and Chen [2002](#page-11-0)), which caused a great number of environmental problems, such as the surface and groundwater contamination, the release of greenhouse gases, and soil quality degradation (Davidson [2009](#page-10-0); Reay et al. [2012](#page-10-0)). Many researchers have reported a low NRE of 30–35 % in the 1990s (Zhu and Chen [2002\)](#page-11-0), and 26–28 % in 2001–2005 for major cereal crops in China (Zhang et al. [2007\)](#page-11-0), significantly lower than that in America  $(52 \%)$  and in Europe  $(68 \%)$ (Ladha et al. [2005](#page-10-0)). Thus, the fate of fertilizer N in farmland ecosystems has received broad attention because of its close relation with fertilizer efficiency and environmental problems (Xu et al. [2000\)](#page-11-0).

Numerous studies have been conducted on improving NRE, reducing N losses by adjusting the N application rate and the ratio of basal to topdressed fertilization. These studies showed that the average application rate to high-yielding maize in China was 237 kg N ha<sup>-1</sup> (Chen et al. [2011](#page-9-0)), and 183 kg N ha<sup>-1</sup> in Nebraska (Grassini et al. [2011](#page-10-0)). Under the same N application rate, a suitable ratio of basal to topdressed N can boost the efficient utilization of nitrogen fertilizers. Field experimental data showed that grain yields, plant N uptake, and NRE increased with the increases of topdressed N fertilizer (Cui et al. [2008b](#page-10-0); López-Bellido et al. [2005\)](#page-10-0). For example, the NRE was more than twice that when N was applied in splits of 1:1 compared with a 2:1 ratio of basal to topdressed N (Yi et al. [2008](#page-11-0)). Some researchers recommend applying fertilizer N with 3–4 splits applications according to the plant's N needs to reduce  $NO<sub>3</sub>$ -leaching (Kettering et al. [2013\)](#page-10-0). Scharf et al. [\(2002](#page-10-0)) found that yields was still responsive to N application until R1, but that the potential yields was not achieved when N applications were delayed until that stage. Liu et al. [\(2014a\)](#page-10-0) reported that consistent improvements in dry matter and N accumulation during the post-silking stage are essential for obtaining high yields from filmmulched maize. So we hypothesized that splitting moderate quantities of topdressed N fertilizer to R1 could increase grain yields and reduce fertilizer losses.

In the Loess Plateau of northwest China, limited precipitation and high evaporation are the primary limiting factors affecting crop production (Zhou et al. [2009\)](#page-11-0). Plastic mulching has been shown to increase crop yields, particularly in arid and semiarid regions (Liu et al. [2009;](#page-10-0) Sharma et al. [2011](#page-10-0)), and has been widely adopted in these areas (Li et al. [1999;](#page-10-0) Zhou et al. [2009](#page-11-0)). As a physical barrier, plastic mulching influences the exchanges of matter and energy between the soil and atmosphere and thus directly affects the soil microenvironment by redistributing moisture in the soil (Li et al. [2004](#page-10-0)), increasing soil moisture and temperature (Liu et al. [2014b](#page-10-0)), decreasing the leaching losses of fertilizers around the root zone (Anikwe et al. [2007](#page-9-0)), and improving soil nutrient availability (Li et al. [2004](#page-10-0)). As consequence, the behavior of fertilizer N may be changed by plastic mulching through these mechanisms. Therefore, a better understanding of N fate in mulched maize cropping system would help improving N management for high yield maize production. The isotopic ( 15N) tracer technique has been used to monitor the fate of applied N by mass balance since the 1970s (Hauck and Bremner [1976\)](#page-10-0), and is still widely used to evaluate fertilization management in agroecosystems (Lam et al. [2012](#page-10-0); Liang et al. [2013](#page-10-0); Liu et al. [2015c](#page-10-0); Stevens et al. [2005a](#page-11-0)). It provides a simple and effective method to trace the N fluxes in ecosystems, and ascertain the sources of N in different plant organs (Schindler and Knighton [1999\)](#page-10-0). However, little research has focused on the fate of fertilizer N under plastic mulching using  $15<sup>N</sup>$  tracing method. The objectives of this study were (1) to evaluate the fate of applied  $15N$  fertilizer under different N split applications to plastic mulched maize; and (2) optimize the N fertilizer management for plastic mulched maize production.

#### Materials and methods

#### Site description

A 2-year experiment was conducted at Changwu Agricultural and Ecological Experimental Station of Chinese Academy of Sciences (35.28°N, 107.88°E, 1200 m altitude), which has a semi-arid climate on the Loess Plateau of China. The annual mean air temperature is 9.7  $\degree$ C, and the average annual precipitation from 1960 to 2012 is 577 mm of which 73 % of this falls during the maize growth season, whereas the average potential evaporation is 1560 mm. Total precipitation was 579 mm and 573 mm in 2013 and 2014 (Fig. 1), respectively. The distribution of precipitation varied considerably between 2 years. The highest precipitation occurred in July in 2013 while August and September in 2014. The main cropping system in this area includes harvesting one crop of maize or wheat per year. According to the American system of soil classification, the soil at the experiment site belongs to Cumulic Haplustolls that developed from loess deposits. The soil properties at the top 20 cm were: bulk density 1.3  $\text{g cm}^{-3}$ , pH 8.3, organic C 8.1 g  $kg^{-1}$ , total N 1.0 g  $kg^{-1}$ , available phosphorus (Olsen-P) 21.5 mg  $kg^{-1}$ , available potassium  $(NH_4OAc-K)$  135.2 mg kg<sup>-1</sup>, and mineral N  $(NO<sub>3</sub>^- - N + NH<sub>4</sub><sup>+</sup> - N)$  28.3 mg kg<sup>-1</sup>.

## Experimental design

The N treatments consisted of three different N split applications at the same total rate of 225 kg N  $ha^{-1}$ . The N fertilizer was applied at the day before sowing, the eight-leaf stage (V8), and the silking stage (R1) in the following splits: 100 %–0–0 (N1), 40–60 %–0 (N2), and 40–30–30 % (N3). A no-N application treatment was also included as a control (N0). The experiment was arranged in a complete randomized block design with three replications; the area of each basic plot was 90  $m<sup>2</sup>$  $(6 \text{ m} \times 15 \text{ m})$ . N fertilizer was applied in the form of urea (prilled urea of 1–2 mm size, 46 % N), part of which was manually distributed over the soil surface prior to sowing and then plowed into the soil at a depth of 15–20 cm as a basal dressing; applications at V8 and R1 were made with a hole-sowing machine following

precipitation. Each plot was supplied with 40 kg P ha<sup>-1</sup> (calcium superphosphate,  $P_2O_5$ , 12 %) and 80 kg K ha<sup>-1</sup> (potassium sulfate, K<sub>2</sub>O, 45 %) at sowing. A high-yielding maize hybrid (Pioneer 335) was used in this study; the plant density was  $65,000$  plants ha<sup>-1</sup>. The maize seeds were sown to 5-cm depth using a handpowered hole-drilling machine on 21 and 28 April in 2013 and 2014, respectively, and maize plants were harvested on 5 and 19 September in 2013 and 2014, respectively. All treatments involved alternating a wide (60 cm wide, 10 cm high) and narrow (40 cm wide, 15 cm high) ridge and a furrow. Both ridges were manually mulched with the transparent polyethylene plastic film (0.008 mm thickness) after being constructed. The joint of the two pieces of film was in the midline of the broad ridge, and the joint was secured by placing soil on top of the film.

To monitor the fate of  $15$ N-labelled fertilizer, two sizes of microplots  $(1 \text{ m} \times 2 \text{ m} \text{ in } 2013 \text{ and}$  $1 m \times 1 m$  in 2014) were established within the fertilized plots, which were bordered by 0.5-m-high galvanized sheet iron inserted into the soil to a depth of 0.45 m and exposed for 0.05 m on the surface to prevent runoff and lateral contamination. In order to trace the fate of  $15N$  applied at every growth stage, there were one, two, and three microplots established in each plot of treatment N1, N2, and N3, respectively, in every year.  $^{15}$ N-labelled urea (10.14 atom%, provided by the Shanghai Chem-Industry Institute) was applied to the microplots using the same dose applied to the corresponding main plot. Detailed  $^{15}$ Nlabelled fertilizer application schemes are shown in Table [1](#page-3-0). For basal N, 5 kg of soil was taken from the corresponding microplot, sieved through a 2-mm

Fig. 1 Monthly precipitation and mean maximum and minimum temperatures in 2013 and 2014. MS and FS denote the maize growing season and fallow season, respectively



Treatment		Sowing $(\%)$	V8 $(\%)$	$R1(\%)$
N <sub>0</sub>		0	$\theta$	0
N <sub>1</sub>		$100*$	0	0
N <sub>2</sub>		$40*$	60	0
	П	40	$60*$	0
N <sub>3</sub>	I	40*	30	30
	П	40	$30*$	30
	Ш	40	30	$30*$

<span id="page-3-0"></span>Table 1 Nitrogen application stages and ratios of different treatments in 2013 and 2014

V8 and R1 denote applying N at the eight-leaf stage and the silking stage, respectively

\* Mean 15N-labelled urea

sieve, mixed with  $^{15}$ N-labelled fertilizer, and broadcast uniformly on the surface of the microplot, followed by mixed to a 15–20 cm depth. For topdressed N, the plastic film was removed prior to fertilization, and the  $15$ N-labelled fertilizer was dissolved in 4 L of distilled water and sprayed onto the soil surface of the microplot using a hand sprayer in order to apply it uniformly; the film was then replaced.

## Plant and soil sampling and analysis

Three <sup>15</sup>N labelled plants in each microplot were harvested close to the ground, and then were separated into leaf, stem, bract, ear axis, and grain. The remained roots in soil were broken up with shovels in situ and mixed into soil before sowing next year. The dry weight was determined after drying at  $70 °C$  to constant weight. An aliquot of each dry samples was ground  $(<0.15$  mm) with a ball mill for total N content and isotopic analysis. Total N in plant was analyzed using the Kjeldahl method. Soil samples were taken to a depth of 200 cm using a 4-cm-diameter soil auger at 20-cm interval. Three sites were selected and then mixed into one sample for each microplot. The soil sample was air-dried, then ground through 0.15-mm sieve. Total N in soil was determined by a permanganate-reduced iron modification of the Kjeldahl method, to include nitrate and nitrite (Bremner and Mulvaney  $1982$ ). The <sup>15</sup>N enrichment in soil and plant samples were determined using an isotope mass spectrometer (MAT-253, Thermo Fisher, America) at the State Key Laboratory of Soil Erosion and Dryland Farming on the Loess Plateau, Chinese Academy of Sciences and Ministry of Water Resource. The natural abundance of  $^{15}$ N in soil and plant were also measured.

Calculations and statistical analysis

The percentage of N derived from fertilizer N (Ndff, %) was calculated according to the following equation:

$$
Ndff(\%) = at\%^{15}N excess in plant or soil/at\%^{15}Nexcess in fertiliser × 100
$$
 (1)

N fertilizer accumulation and recovery by maize were calculated by the following equations:

Plant total N kg hm<sup>2</sup> - <sup>¼</sup> plant dry matter kg hm<sup>2</sup> - N concentration g kg<sup>1</sup> -=<sup>1000</sup> <sup>ð</sup>2<sup>Þ</sup>

Plant N from fertilizer $(kg \, \text{hm}^{-2}) = (2) \times \text{Ndfr}_{\text{plannt}}$  $(3)$ 

Residual N in soil kg hm<sup>2</sup> - <sup>¼</sup> soil thickness cmð Þ soil bulk density g cm<sup>3</sup> - N concentration g kg<sup>1</sup> -Ndffsoil <sup>100</sup> <sup>ð</sup>4<sup>Þ</sup>

N recovery efficiency $(\%)$ 

$$
= (3)/N \text{ application rates} (\text{kg hm}^{-2}) \times 100 \qquad (5)
$$

N residual efficiency $(\%)$ 

$$
= (4)/N \text{ application rates} (\text{kg} \text{ hm}^{-2}) \times 100 \qquad (6)
$$

Potential N losses $(\% ) = 100 - (5) - (6)$  (7)

N apparent recovery efficiency (ARE) was calculated by difference mothed:

$$
ARE(\%) = \frac{N \text{ uptake in fertilized plot} (kg \text{hm}^{-2}) - N \text{ uptake in unfertilized plot} (kg \text{hm}^{-2})}{N \text{ application rates} (kg \text{hm}^{-2})}
$$
(8)

Statistical analysis was conducted using the ANOVA procedure of SPSS 20.0. Treatments were compared by the least significant difference (LSD) at the 5 % level. Graphs were produced with SigmaPlot 12.5.

## Results

Yields, N uptake and ARE by maize

There were similar trends in both growing seasons, but grain yields, N uptake and ARE were higher with N-fertilized treatments in 2014 (Table 2). Compared with the N0 treatment, the application of N fertilizer significantly increased grain yields and plant N uptake. The grain yields for treatments N2 and N3 were significantly higher than those for treatment N1 by 8.6 and 11.7 %, respectively, in 2013, and by 8.0 and 9.6 % in 2014; there were no significant differences between the N2 and N3 treatments. The total N uptake increased with the times of split N applications in this study, ranging from 171 to 276 kg  $\text{hm}^{-2}$  in 2013 and from 136 to 282 kg  $\text{hm}^{-2}$  in 2014, and there were significant differences among all treatments. The ARE showed the same pattern as total N uptake, ranging from 27.1 to 46.4 % in 2013 and 44.2–64.9 % in 2014.

# Sources of plant N uptake and  $15N$  distribution in maize

Approximately 26.3–32.1 and 27.2–32.8 % of plant N uptake were derived from <sup>15</sup>N-labelled fertilizer in 2013 and 2014, respectively (Table [3](#page-5-0)). Compared with N1, split N applications significantly increased the proportion of fertilizer-derived N (N2 and N3 treatments), but there was no significant difference between N2 and N3 treatments. In the treatment N2, N uptake from 15N-labelled topdressed fertilization was approximately twice the uptake from  $15N$ -labelled basal fertilization. In the treatment N3, plant N uptake from the first topdressed N (topdressed at V8) was significantly higher than that from the second topdressed N (topdressed at R1), although the same  $^{15}N$ application rate was used at both stages. From 67.9–73.7 and 67.2–72.8 % of the N uptake by the maize plant was derived from the soil in 2013 and 2014, respectively. The proportion of N uptake from the soil in treatment N1 was significantly higher than in the treatments N2 and N3; however, the quantity of N uptake from the soil in treatment N1 was the lowest owing to its lower total N uptake. Applying N with three splits (N3) increased the quantity of N uptake from soil significantly in both years.

The distribution of  $15N$  among different organs was as follows: grain  $>$  leaf  $>$  stem  $>$  ear axis  $>$  bract (Fig. [2](#page-5-0)). In 2013 and 2014, 65.4–71.5 and 66.5–69.0 %, respectively, of total  $^{15}N$  uptake by the plant were partitioned to grain. The proportion of  $^{15}N$ partitioned to grain from topdressed N (topdressed at V8 and R1) was 69.9–73.3 % higher than that of basal N, whereas the opposite trend was observed for straw, indicating that basal N increased early N accumulation in straw, and topdressed N increased late N accumulation in grain.

# Distribution of residual  ${}^{15}N$  in soil

The pattern of residual  $15N$  distribution in the soil among treatments was similar for both growing seasons (Fig. [3](#page-6-0)). The total residual  $^{15}N$  in the 0–200 cm soil profile layer after maize harvest was 113.3–118.8 kg hm<sup>-2</sup> in the 2013 growing season,<br>accounting for 50.4–52.8 % of <sup>15</sup>N, and accounting for  $50.4-52.8$  % of  $^{15}N$ , and

	Grain yields (t $\text{hm}^{-2}$ )		N uptake (kg $\text{hm}^{-2}$ )		ARE $(\% )$	
	2013	2014	2013	2014	2013	2014
N <sub>0</sub>	10.6c	9.5c	171.2 d	136.3 d		
N <sub>1</sub>	12.8 <sub>b</sub>	13.5 <sub>b</sub>	232.1 c	235.7c	$27.1\text{ c}$	44.2 c
N <sub>2</sub>	13.9a	14.3a	255.0 <sub>b</sub>	265.1 b	37.2 <sub>b</sub>	57.3 b
N <sub>3</sub>	14.3a	14.8 a	275.5a	282.2 a	46.4a	64.9 a

Table 2 Effects of N split applications on maize grain yields, N uptake and apparent recovery efficiency (ARE) in 2013 and 2014

Different letters within a column indicate a significant difference ( $P < 0.05$ )

Treatment		N rate (kg $\text{hm}^{-2}$ )	Ndff $(\%)$		Ndff (kg $\text{hm}^{-2}$ )		Ndfs $(\%)$		Ndfs (kg $\text{hm}^{-2}$ )	
			2013	2014	2013	2014	2013	2014	2013	2014
N1	B	225	26.3a	27.2a	61.2 a	64.3 a				
N <sub>2</sub>	B	90	12.0c	9.8 <sub>e</sub>	30.5c	26.1 e				
	$T_{VS}$	135	20.1 <sub>b</sub>	22.9 <sub>b</sub>	51.5 <sub>b</sub>	60.8 <sub>b</sub>				
N <sub>3</sub>	B	90	10.5d	11.8 <sub>d</sub>	$29.1\text{ c}$	33.7d				
	$T_{\rm V8}$	67.5	$11.2 \text{ cd}$	13.2c	$30.8\text{ c}$	$37.2\text{ c}$				
	$T_{R1}$	67.5	8.2 e	7.8 f	22.5d	22.4 f				
N1		225	26.3 B	27.2 B	61.2 B	64.3 C	73.7 A	72.8 A	171.0 B	172.5 B
N <sub>2</sub>		225	32.1 A	32.7 A	82.1 A	86.9 B	67.9 C	67.3 B	173.1 B	178.5 AB
N <sub>3</sub>		225	29.9 A	32.8 A	82.4 A	93.4 A	70.1 B	67.2 B	193.1 A	191.9 A

<span id="page-5-0"></span>Table 3 Maize plant N derived from <sup>15</sup>N-labelled urea (Ndff) and from soils (Ndfs) in 2013 and 2014

Different lowercase letters within a column indicate a significant difference ( $P < 0.05$ )

Different capital letters within a column indicate a significant difference ( $P < 0.05$ )

B denote basal N;  $T_{VS}$  and  $T_{R1}$  denote topdressed N at the eight-leaf stage and the silking stage, respectively



Fig. 2 Effects of N application (basal vs. topdressed) on the distribution of <sup>15</sup>N among maize organs. The *different letters* on bars refer to significant differences at the 5 % level. B denotes basal N;  $T_{V8}$  and  $T_{RI}$  denote topdressed N at the eight-leaf stage and the silking stage, respectively

103.9–113.9 kg  $\text{hm}^{-2}$  in the 2014 growing season, accounting for 46.2–50.6 % of  $^{15}N$ , with about half of the residual  $15N$  remaining in the 0–20 cm soil layer. There was no difference among treatments. The 2-year average residual amount of basal N in the 0–200 cm soil profile was 44.0 and 40.3 kg  $\text{hm}^{-2}$  in the treatments N2 and N3, respectively, accounting for 48.9 and 44.8 % of the basal N applied. The average amount of residual N coming from topdressed N at V8 in the treatments N2 and N3 was 71.4 and 28.8 kg  $\text{hm}^{-2}$ , respectively, accounting for 52.9 and 42.6 % of the topdressed N applied. On average, the residual amount of topdressed N at R1 in the treatment N3 was 39.5 kg  $\text{hm}^{-2}$ , accounting for 58.6 % of the topdressed N applied.

# Fate of  $15$ N-urea in the maize cropping system

For all treatments, the mean recovery, residual, and potential losses were 33.4, 51.7, and 14.9 %, respectively, for the 2013 growing season, and 36.2, 48.0, and 15.8 %, respectively, for 2014 growing season (Table [4](#page-6-0)). The average  $15$ NRE over 2 years for the treatments N2 and N3 was 37.6 and 39.1 %, respectively, significantly higher than the  $15$ NRE for the N1 treatment (average 28 %), and there was no significant difference between the N2 and N3 treatments. The 2-year average potential N losses were lower for N2 and N3  $(11.2 \text{ and } 12.7 \%)$ , compared with N1

<span id="page-6-0"></span>

Fig. 3 The distributions of residual <sup>15</sup>N from basal and topdressed N in 2013 and 2014. The *bars* are the LSD at  $P < 0.05$ . B denotes basal N;  $T_{V8}$  and  $T_{R1}$  denote topdressed N at the eight-leaf stage and the silking stage, respectively

Treatments		N rate (kg $\text{hm}^{-2}$ )	Recovery $(\% )$		Residual $(\% )$		Potential losses $(\% )$	
			2013	2014	2013	2014	2013	2014
N1	B	225	27.2d	28.6e	52.8 b	$47.1$ bc	20.0a	24.3a
N2	B	90	33.9c	$28.9$ de	46.7c	51.0 b	19.4a	20.0a
	$T_{VS}$	135	38.2 <sub>b</sub>	45.1 <sub>b</sub>	55.4 b	50.3 <sub>b</sub>	6.4 <sub>b</sub>	4.6 <sub>b</sub>
N <sub>3</sub>	B	90	32.3c	37.5c	45.8c	43.8 cd	21.9a	18.7 a
	$T_{VS}$	67.5	45.7a	55.2 a	45.8c	39.4d	8.5 <sub>b</sub>	5.4 b
	$T_{R1}$	67.5	33.4c	33.2 cd	61.0a	56.1 a	5.6 <sub>b</sub>	10.7 <sub>b</sub>
N1		225	27.2 B	28.6 B	52.8 A	47.1A	$20.0\text{ A}$	24.3 A
N <sub>2</sub>		225	36.5A	38.6 A	51.9 A	50.6 A	11.6 B	10.8 B
N <sub>3</sub>		225	36.6 A	41.5A	50.4 A	46.2A	13.0 B	12.3 B

**Table 4** The fate of  $^{15}$ N-labelled urea in both maize growing seasons

Different lowercase letters within a column indicate a significant difference ( $P < 0.05$ )

Different capital letters within a column indicate a significant difference ( $P < 0.05$ )

B denote basal N;  $T_{V8}$  and  $T_{R1}$  denote topdressed N at jointing and silking stage, respectively

(22.2 %). However, no differences were observed among treatments for the residual N in the soil in either year.

The  $15$ NRE of topdressed N at V8 was significantly higher than that of basal N in the N2 and N3 treatments, whereas the opposite trend was observed for potential N losses (Table [4](#page-6-0)). The recovery, residual, and potential losses of basal N in the treatment N2 were 31.4, 48.9, and 19.7 %, respectively (mean of 2 years), whereas the values for topdressed N at this stage were 41.6, 52.9, and 5.5 %, respectively. In the N3 treatment, the recovery of topdressed N at V8 (50.4 % on average) was significantly higher than that of basal (34.9 %) and topdressed N (33.3 %) at R1; the opposite trend was observed for residual N. The recovery of topdressed N at R1 in the N3 treatment was similar to that of basal N, but there was more residual N in the soil and less potential N losses with topdressed N at this stage.

## Discussion

Effects of N split applications on N uptake and grain yields

It has been reported that higher grain yields and improved NRE are obtained when N fertilizer application is divided into an appropriate ratio of basal and topdressed N (Shi et al. [2012](#page-10-0)). In our study, applying N with two or three splits significantly increased grain yields, total N uptake, and ARE, compared with a single N application. However, there was no significant difference between treatments N2 and N3 in grain yields. In contrast to our results, Lü et al. ([2012](#page-10-0)) found that applying N in a 2:4:4 ratio at six leaves, ten leaves, and the grain filling stages significantly increased grain yields compared with a 4:6 ratio. The difference in results may be attributed to cultivation practice. In the present study, plastic mulching was applied in order to overcome the arid climatic conditions, which would retard N leaching to deep soil layers (Liu et al. [2015a](#page-10-0)); in addition, plastic mulching could increase soil moisture, temperature, and nutrient availability (Li et al. [2004\)](#page-10-0), making more available N being absorbed and assimilated by maize. Therefore, applying N with two splits may satisfy the maize crop growth period of nutritional requirements.

Approximately 26.8–32.4 % of plant N was derived from fertilizer, with  $67.6-73.2$  % of plant N coming from the soil in both years. This is consistent with previous studies by Rimski-Korsakov et al. ([2012](#page-10-0)) and Stevens et al. ([2005b](#page-11-0)), who found that 54–78 and 54–83 %, respectively, of plant N was derived from soil; Chen et al.  $(2010)$  $(2010)$  $(2010)$  also reported that the net N mineralization of the soil organic matter contributes 62–80 % of the total nitrogen in the maize crop. These results indicate that the mineralization of soil organic matter is an important source of N for crops. In our study, split N applications increased the quantity of plant N derived from fertilizer, as well as that from soils. This finding was in accordance with Stevens et al. [\(2005a](#page-11-0)), who found increases in plant uptake of soilderived N with increasing N application. In addition, N use efficiency under split N applications was investigated by two methods in this study: the  $15N$ -labelled method and the difference method. Split applications of N increased the ARE significantly, but no difference in <sup>15</sup>NRE was found between N2 and N3 treatments. These phenomenon could be explained by the so-called "priming effect" (Kuzyakov et al. [2000\)](#page-10-0), in which N already present in a given soil N pool may increase the effect of N addition (Jenkinson et al. [1985](#page-10-0)).

N application times affected the distribution of  $\rm ^{15}N$ in maize. Previous studies have reported that 61.0–78.0 % of the total <sup>15</sup>N taken up by each plant was partitioned to grain under different N-deficiency and timing of N supply (Subedi and Ma [2005\)](#page-11-0). In our study, of the total  $15N$  taken up by each plant, 66.9–69.0 % was partitioned to grain, followed by leaf, stem, ear axis and bract. Split N applications increased the proportion of  $^{15}N$  to total  $^{15}N$  uptake in grain in 2013. It was also observed that topdressedderived  $15N$  was more prevalent in grain than basalderived  $^{15}$ N, whereas the opposite trend was found in straw. These results are similar to those of Yang et al.  $(2011)$  $(2011)$ , who found that the quantity of  $15N$  derived from basal N in grain was lower than in straw, whereas the <sup>15</sup>N derived from topdressed N in grain was higher than in straw. Zhao et al. ([2012\)](#page-11-0) and Andersson et al. [\(2004](#page-9-0)) also found that a greater proportion of N uptake was allocated to the developing grain when supplementary N was provided during the late growth stage. It indicates that although the basal N is essential for maize vegetative growth at the early growth stage, topdressing timely at the middle or later growth stage plays a key role on obtaining high grain yields.

## N application timing and the fate of  $15N$

The <sup>15</sup>NRE of the plant significantly increased with split applications of N, whereas N losses decreased. In this study, the  $15$ NRE of treatments N2 (37.6 % on average for both years) and N3 (mean 39.1 %) were

significantly higher than treatment N1 (mean 27.9 %), but there was no significant difference between N2 and N3 treatments. It indicates that <sup>15</sup>NRE was not increased by three splits application of N compared with N two splits application, similar result was reported by Lü et al.  $(2012)$  $(2012)$  $(2012)$ . In the treatments N2 and N3, 60 and 30 % of total N fertilizer were topdressed at V8, when the maize plant needed massive N nutrient to support the rapid increase of biomass, thus leading to higher  $15$ NRE for topdressed N at this stage, especially in the N3 treatment (46–55 %). However, topdressed N at R1 showed a lower <sup>15</sup>NRE (33.3  $%$  on average) than topdressed N at V8 in the treatment N3, in spite of the same topdressed amount. It suggests that the peak of N uptake for maize appears at jointing stage; therefore, supplying relative sufficient N at jointing stage is a key strategy for obtaining higher yields and higher NRE.

In this study, the potential N losses (averaged 15.4 % among all treatments) were lower than some other reports. Shi et al.  $(2012)$  $(2012)$  found 21–32 % of N losses in a maize cropping system in China. The lower potential N losses in this study may be due to the lower potential ammonia  $(NH_3)$  volatilization and N leaching losses. The potential N losses with treatments N2 and N3 over both years averaged 11.2 and 12.7 %, respectively, which were significantly lower than the losses with the N1 treatment (average 22.2 %). This difference may be attributed to better synchronization between N supply and crop demand under N split applications. In other words, the application of more N near the time of peak crop N demand may increase grain yields and NRE, while decreasing the risk of soil N losses (Cui et al.  $2008a$ ). NH<sub>3</sub> volatilization losses of N fertilizer are closely related to the application technique and N rate (Sommer et al. [2003](#page-10-0); Zhao et al.  $2009$ ). N fertilizer deep placement could reduce NH<sub>3</sub> losses enormously, compared to surface application (Liu et al. [2015b\)](#page-10-0). In this study, topdressed N was injected into soil at a 10-cm depth with a hole-sowing machine in the main plots (in the microplots, topdressed applications were made with  $\mathrm{^{15}N}\text{-}$ labelled urea dissolved in water in order to apply it uniformly), which refrained  $NH<sub>3</sub>$  volatilization dramatically. Basal N was broadcast on the surface followed by being plowed into soil; however, parts of them were exposed on the soil surface after establishing the ridges, which increasing the risk of  $NH<sub>3</sub>$  losses compared to topdressed N. It is generally believed that the N losses through  $NH<sub>3</sub>$  volatilization increase with the increasing rate of N application in soils (Tian et al. [2001;](#page-11-0) Zhao et al. [2009\)](#page-11-0). In the present study, the whole N fertilizer was applied as basal dressing in treatment N1, whereas only 40 % of total N fertilizer was applied in the treatments N2 and N3. Therefore, the potential  $NH<sub>3</sub>$  losses with N1 treatment may be higher than that with N2 and N3 treatments. In this study, the potential N leaching losses may be low, due to the mulching practice in all treatments. Previous study has found that mulching (especially plastic mulching) is an effective measure for decreasing N leaching losses in dry farmland (Liu et al. [2015a](#page-10-0)). Liu et al. [\(2015c\)](#page-10-0) reported that plastic mulching increased total labelled-N remaining in the 0–170 cm depth in cropped soils, compared with no mulch.

The timing of N application resulted in significant differences in N fate. Topdressed N at V8 produced higher  $15$ NRE in plants than basal N, whereas the opposite trend was observed for potential N losses (Table [4](#page-6-0)). Similar results were reported by Yang et al. [\(2011](#page-11-0)), who found that the  $^{15}$ NRE of topdressed N were 50.9, 44 and 41.4 % at N application rates of 90, 180 and 270 kg  $\text{hm}^{-2}$ , respectively, higher than values from basal N (43.1, 36.2 and 33.7 %, respectively). Shi et al. ([2012\)](#page-10-0) also reported that the topdressed N showed a higher  $15$ NRE by maize (averaged 54 %), compared with basal N (averaged 32 %). The lower  $15$ NRE with basal N may be attributed to the less demand of the maize plant for N during the early growing stage (Abendroth [2011](#page-9-0)), when the maize biomass was relatively small. Therefore, delaying N application to later growth stage or applying N with multiple split applications is a better strategy of N management to improve the synchrony between soil N availability and crop N demand. Previous reports on N losses of basal and topdressed N have produced conflicting results. Yang et al. ([2011\)](#page-11-0) reported that total losses were lower with basal N  $(1.7–7.1 \%)$  than with topdressed N  $(3.3–12 \%)$ . In contrast, Shi et al. ([2012\)](#page-10-0) found lower N losses with topdressed N (averaged 11.5 %) than with basal N (averaged 37.5 %). In the present study, potential N losses with topdressed N ranged from 5.5 to 8.1 %, which was significantly lower than losses with basal N  $(19.7–22.2 \%)$ . This may be attributed to the more potential NH3 losses from basal N as explained above.

Split N application did not affect the total amount of residual  $^{15}N$  in the 0–200 cm soil layer. On average, <span id="page-9-0"></span>the residual amount of  $^{15}$ N-labelled fertilizer in the 0–200 cm soil layer was 112.1 kg  $\text{hm}^{-2}$ , accounting for 49.8 % of the total  $15N$  application. Similar results were reported by Yang et al. ([2011\)](#page-11-0) in the North China plain, who found that after the first maize crop was harvested, 45–60 % of the applied fertilizer N remained as residual N in the 0–150 cm layer of soil, with about half of that retained in the 0–20 cm layer. Pilbeam et al. ([2002\)](#page-10-0) also reported that 58 % of the applied N was recovered in the 0–60 cm soil layer at maize harvest, primarily in non-mineral N forms. In contrast to our results, Shi et al. [\(2012](#page-10-0)) reported a lower residual amount  $(33 \%)$  of N in the 0–100 cm layer of the Middle and Lower Yangtze River Basin. The higher residual N in the present study may be attributed to the plastic mulching practice and the fertilization methods, which could reduced the N leaching losses and gaseous losses (Liu et al. [2015b,](#page-10-0) [c](#page-10-0)). A previous study had found that the losses of N through  $NH<sub>3</sub>$  volatilization and N<sub>2</sub>O emission were not more than 1.5 %, when urea was mixed with soil (Li et al. [2008\)](#page-10-0). On average, the total residual amount of N for treatment N1 in the 0–60 cm was 91.5 kg  $\text{hm}^{-2}$ , which is higher than the 83.6 and 76.4  $kg \text{ hm}^{-2}$  for treatments N2 and N3; the opposite trend was found in the 60–200 cm layer. These values are inconsistent with some previous reports, which showed lower residual basal N in the upper soil layer (Shi et al. [2012\)](#page-10-0). The difference in results may be attributed to the high rate of microbial immobilization at the earlier growth stage in treatment N1. When large quantities of N fertilizer were applied as basal N at time of sowing, more available N was immobilized by the microbes at the upper soil layer during the early stage of maize growth, due to less competition for available N between the crop and microorganisms. For all the treatments in this study, 70–81 % of total residual N retained in 0–60 cm soil layer, which could be utilized more easily by the following crops. Therefore, it may be more reasonable to consider the total NRE by crops through several continuous growing seasons when evaluating an N fertilizer management.

## Conclusion

Split applications of N significantly increased grain yields, total N uptake, and ARE, but no significant difference in grain yields between two and three splits

applications. Split N applications significantly increased the plant N uptake derived from fertilizer; moreover, a three splits N application significantly increased the N uptake from the soil by plants. The <sup>15</sup>NRE was significant higher with two or three splits N applications than a single N application, whereas the opposite trend was observed for potential N losses. The residual  $15N$  in the 0–200 cm soil layer ranged from 48.3 to 51.3 % at maize harvest, about half of which remained in the 0–20 cm soil layer. Considering the large amount of labor required by topdressing fertilizer N under the plastic mulching, the two splits application of N is more suitable for the mulched maize in the semiarid region, which could produce higher grain yields, higher NRE, and lower N losses.

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