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Effects of nitrogen application rate and topdressing times on yield and quality of Chinese cabbage and soil nitrogen dynamics

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ABSTRACT

A good understanding of the relationship between vegetable quality and soil N balance is very important for proper nitrogen (N) management for crop productions. In this study, a field experiment was carried out to investigate the N application rate and times on Chinese cabbage yield and quality, N use efficiency, soil nitrate-N concentration, and soil pH. The experiment was implemented in a two-way factorial design and the two factors comprises of number of N applications (once, twice and three times, denoted as T1, T2 and T3) and N rates (15%, 30% and 45% less than conventional rates (CF), denoted as F1, F2 and F3, respectively). The treatments were also compared with a no-fertilizer blank and a control (CF) with a conventional N management practice. The results showed that the highest yield of cabbage (164.65 t hm^{-2}), Vitamine C content (14.80 g 100 g⁻¹ fresh mass), soluble sugar content (2.33 mg kg⁻¹), plant N uptake (119.2 kg hm⁻²) were obtained under T3F1 treatment. Compared with CF treatment, T3F1 treatment significantly increased vegetable yield, vitamin C and soluble sugar content in fruit, and nitrogen use efficiency by 10.97%, 13.76% and 17.68%, and 18.76%, respectively. Nitrate-N content in cabbage was reduced by 7.55% in T3F1 treatment. With the reduced N application rate, soil pH gradually changed from 6.25 to 7.26. T3F1 treatment is a most suitable N management practice for vegetable production, in terms of higher vegetable yield and quality, soil N content, depressed soil acidification and nutrient uptake by Chinese cabbage.

Chinese cabbage (Brassica campestris spp. pekinensis) is one of the important winter vegetables in China, even in East Asia, due to high yield and rich nutrition [1,2]. In China, vegetable production plays an important role in the supply of vegetable market and people's daily life [3]. Excessive chemical fertilizers are applied to assure a high yield. However, too much fertilizers could cause decline of Chinese cabbage yield and quality, low nitrogen use efficiency (NUE), soil degradation [4] and increase accumulation of heavy metal in soil [5], and result in water pollution by soil nitrate leaching and runoff losses [6-8]. Therefore, the suitable fertilization management of Chinese cabbage is urgent in the production.

Many studies showed that residual levels of nitrate in soil increased sharply when N application rates exceed the crop requirements. Besides, excessive N application at rates above the 'saturation point' could reduce NUE and even decrease crop yield [9]. However, inadequate N supply would inevitably compromise crop yield. Thus, the effective N fertilizer management was comprehensively studied [10], in order to maintain an appropriate supply N fertilizer. It was found that proper application of urea was one of the most important management methods.

ARTICLE HISTORY

Received 26 September 2018 Accepted 5 November 2018

KEYWORDS

Vegetable; quality; NUE; soil pН

Urea is the most common source of N fertilizer for farmers [11]. Low-cost, high N content and rapid N release make urea a frequently-used base N fertilizer for Chinese cabbage production [12]. However, N release from urea is relatively rapid, and soluble N is susceptible to loss, which would therefore cause plant N deficiency in long term. For this sake, multiple N applications are recommended to meet the N demand of Chinese cabbage, especially at later growth stages when more N is required for plant growth. By contrast, there is less N requirement in early growth stage of cabbage [13]. Base application of excessive urea may thus lead to N accumulation, which may not only increase leaching downward in the soil profile, but result in soil consolidation and salinization, soil aggregates reduction [14-16]. Therefore, a better management strategy is needed for employing conventional urea in crop production systems.

Seeking a rational protocol of urea application is of great significance for promoting the sustainable development of agriculture. Previous studies showed that compared with base application of urea, urea topdressing could increase NUE and thus crop yields [17,18]. The hypothesis of this study was that urea topdressing with appropriate application rates and times will improve the yield and quality of Chinese

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cabbage and reduce N loss, compared to basal urea application. In this study, a two-factor and three-level orthogonal fertilization scheme was designed for Chinese cabbage cultivation. The two factors were N application rate and topdressing times. The objectives of this research were to investigate the influence of N application rate, topdressing times on Chinese cabbage growth, NUE, soil pH and soil nitrate content under green chamber.

1. Materials and methods

1.1. Experimental sites and materials

The study was carried out from September to November, 2017 in Tai'an, Shandong, China. The average annual rainfall was 702 mm and air temperature was 13°C. Physicochemical properties of experimental soil were analyzed following standard soil testing methods [19]: pH 7.38 (soil to water ratio of 1:2.5), total N content in soil 0.81 g kg⁻¹, soil nitrate concentration 35.36 mg kg⁻¹, ammonium concentration 6.33 mg kg⁻¹, available phosphorus content 60.58 mg kg⁻¹, and available potassium content 103.37 mg kg⁻¹.

The tested fertilizers included in conventional urea (N 46%), triple superphosphate (P_2O_5 46%) and potassium sulfate (K_2O 50%), which were purchased from commercial stores. All of fertilizers were freely produced by Kingenta Ecological Engineering Group Co.

1.2. Experimental design

The experiment was implemented in a two-way factorial design and the two factors include number of N applications (once, twice and three times, denoted as T1, T2 and T3) and N doses (15%, 30% and 45% less than conventional rates (CF), denoted as F1, F2 and F3, respectively). The treatments were also compared with a control (CF) and a no-fertilizer blank. In CF treatment, urea was applied as basal N fertilizer at rate of 170 kg hm⁻² (N). In other fertilizer treatments, 50% of total urea was applied as basal fertilizer, and the remaining 50% were then split into one, two and three times. The phosphorus and potassium fertilizer was used as base fertilizer at rate of 75 kg hm⁻² (P₂O₅) and 98 kg hm⁻² (K₂O) respectively. Each treatment was repeated 4 times and randomly arranged in 44 plots, each plot 4.76 m². The treatments were arranged in an orthogonal experimental design.

Thus, the experiment included eleven fertilizer treatments, including: CK (no N fertilizer), CF (with only basal N application at 170 kg hm⁻²), T1F1 (urea was applied at one time, and the N application rate was reduced by 15% than CF), T1F2 (once topdressing and reduced urea by 30%), T1F3(once topdressing and reduced urea by 45%), T2F1(twice topdressing and reduced urea by 15%), T2F2 (twice topdressing and reduced urea by 30%), T2F3 (twice



topdressing and reduced urea by 45%), T3F1 (three times topdressing and reduced urea by 15%), T3F2 (three times topdressing and reduced urea by 30%), T3F3 (three times topdressing and reduced urea by 15%).

Chinese cabbage was seeded on 1 September 2017, and harvested on 24 November 2017. The large and small row spacing were 80 cm and 60 cm, respectively, and seeds in each row were planted 40 cm wide. The cultivating density was 84,033 plant hm⁻². The irrigation was implemented as per local farmers' traditional irrigation methods. The water was irrigated between narrow rows for four times in the whole growth season. At the time of seeding on 1st Sep., the base fertilizer was applied and watered, and other fertilizer application were carried out for equivalently topdressing at 21th Sep., 5th Oct. and 25th Oct. prior to the irrigation. The disease control practices were implemented according to traditional methods of local farmers.

1.3. Sampling and measurement

After harvesting, three soil samples in each plot were randomly collected to a depth of 100 cm and sectioned in an interval of 20 cm. The three soil samples in each plot were homogenized, and stored in the sealed plastic bag and brought back to the laboratory.

The soil moisture content was determined by drying method and soil mineral N (NH_4^+ -N and NO_3^-N , extraction with 0.01 mol L^{-1} CaCl₂) concentration were determined on an AA3-A001-02E Auto-analyzer (Bran-Luebbe, Germany) within 24 hours after collecting. The other remaining soil samples were air dried, ground and sieved through a 2 mm sieve.

Chinese cabbage was harvested and measuring total yield of the plot, unit area was calculated. In the harvest, three plot Chinese cabbage samples taken. After the ending on 24 November 2017, 2 normal plants were randomly taken, separated and washed with water. The samples were then oven-dried for 30 min at 105°C to kill enzyme, then dried to constant weight at 85°C, followed by recording dry weights. A small mill was used to crush the dry plants and the powder was used for determination total N content.

Vitamin C (Vc) content, nitrate concentration, sugar concentration in Chinese cabbage were measured after mixed samples was homogeneously smashed with a juice machine. Vc content was determined using the spectrophotometer method at the wavelength of 230 nm, nitrate concentration was determined using the spectrophotometer method at the wavelength of 260 nm. Soluble sugar concentration was measured by abbe refractometer, dripped tomato pulp onto the instrument to determination [20]. Total N contents in the cabbage plants were measured by digesting samples with concentrated H_2SO_4 and H_2O_2 and measuring N using distillation method [21].

1.4. Statistical analyses

The nitrogen use efficiency (NUE) (%) = (accumulation N uptake with N treatment- accumulation amount without N treatment)/N application amount \times 100%

Statistical analyses using standard analyses of variance (ANOVAs) and Duncan tests (P < 0.05) were conducted using SAS software (SAS 2010, SAS Institute, Cary, NC, USA). Data were processed with Microsoft Excel 2016, figures were drawn out using Excel 2016 software.

2. Results and discussion

2.1. Effects of different treatments on nitrate distribution and ph in soil profile

Fertilization increased NO₃⁻N concentrations in the soil during the cabbage harvest stage, and different fertilization treatments had significantly affected the NO₃⁻N residue in the soil (Table 1). NO₃⁻N content in the 0-100 cm soil profile of post-harvest soil was significantly affected by N application rate, topdressing times and their interaction. Contents of soil NO₃⁻ N in the 0-100 cm soil profile increased with increase in N application rate (Table 1). Under the same nitrogen application rate, with the increased of topdressing times, the content of NO₃⁻N in 0–20 cm soil layer increased gradually, and the content of NO₃-N in 20-100 cm soil layer decreased gradually. Especially in T3F1 treatment, the content of NO₃⁻N in 0–20 cm soil layer was 46.19%, 21.24% and 56.39% higher than that in T1F1, T2F1and CF treatments, respectively. The content of NO₃⁻N in 20–100 cm soil layer was 32.07%,

Table 1	 Soil 	NO₃ [−] N	contents	with	different	treatments.
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23.16% and 44.04% lower than that of T1F1, T2F1, and CF, respectively. The results showed that topdressing significantly reduced the leaching of NO3⁻N to deep soil, and residual NO3-N in deep soil was more reduced with more topdressing times. Besides, reduced urea rate decreased downward leaching of urea to underlying soil, and thereby significantly decreased the NO3⁻N concentrations. Chinese cabbage was a shallow root crop, the lower urea application rate decreased soil N content in the early growing of Chinese cabbage, however, multiple urea applications could provide adequate N supply for plant growth [22]. Topdressing also improved the absorption of N to plants, reduced the leaching of N to the deep soil caused by irrigation, and slowed down the pollution of soil and groundwater by N application [23,24].

The effect of urea application on soil pH was significant (Figure 1). The pH values in CK treatment ranges between 6.25-7.30. Under the same N application rate, soil pH increased gradually by 0.15-0.31 units with the increased of topdressing times. With the same topdressing times, soil pH decreased gradually by 0.12-0.23 units with the increase of nitrogen application rate. The possible reason was that urea in the soil was hydrolyzed to NH_4^+ . Under aerobic conditions, the NH_4^+ was nitrified into NO₃⁻, in which formation of one mol NO₃⁻ was accompanied by the release of two mol H^+ (NH₄⁺+ 3/2 $O_2 = NO_2^- + 2H^+ + H_2O$). This resulted in soil acidification, reduced N absorption and NUE of Chinese cabbage, which affected the normal growth of Chinese cabbage [25].

	NO_3^-N concentration in 0–100 cm soil profile (mg kg ⁻¹)					
Treatment	0–20 cm	20–40 cm	40–60 cm	60–80 cm	80–100 cm	
F×T						
СК	14.54g	7.33i	5.59i	4.42h	3.71h	
CF	25.66de	21.63b	20.00a	19.20a	18.28a	
T1F1	27.45d	22.45a	16.99b	14.37b	12.11b	
T1F2	25.03e	17.60d	14.76c	12.01c	10.84d	
T1F3	23.20f	15.51e	12.46e	10.62e	7.10g	
T2F1	33.10b	20.22c	14.49c	12.11c	11.57c	
T2F2	29.68c	15.44e	10.30g	9.67f	8.58f	
T2F3	25.81de	12.40h	9.29h	8.47g	7.40g	
T3F1	40.13a	17.51d	13.48d	11.20d	9.62e	
T3F2	31.90b	13.46f	11.29f	9.32f	7.07g	
T3F3	27.14d	12.89g	10.37g	8.63g	7.42g	
Nitrogen applicatio	on rate					
F1	33.56a	20.06a	14.98a	12.56a	11.10a	
F2	28.87b	15.50b	12.12b	10.33b	8.83b	
F3	25.38c	13.60c	10.70c	9.24c	7.30c	
Topdressing times						
T1	25.23c	18.52a	14.74a	12.33a	10.01a	
T2	29.53b	16.02b	11.36c	10.08b	9.18b	
T3	33.06a	14.62c	11.71b	9.71c	8.03c	
Source of variance						
F	<0.0001	<0.0001	<0.0001	<0.0001	< 0.0001	
Т	<0.0001	<0.0001	<0.0001	<0.0001	< 0.0001	
$F \times T$	<0.0001	<0.0001	<0.0001	0.0118	< 0.0001	

Note: Means followed by the similar lowercase letters within same column of each item were not significant in difference at 5% level, and the P < 0.01 indicated the very significant level of 1%, '-'no valid value.



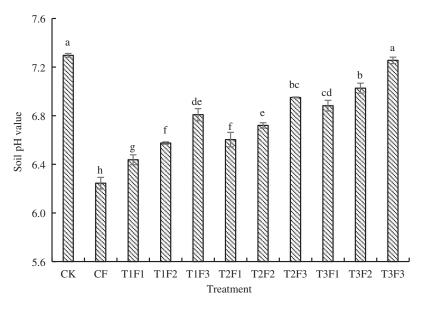


Figure 1. Soil pH value with different treatments.

2.2. Effects of different treatments on NUE of chinese cabbage

The dry matter of Chinese cabbage was the basis of yield formation. The dry matter, N accumulation and NUE of Chinese cabbage under different treatments were determined (Table 2). Under the same of top-dressing times, NUE increased with the decrease of N application rate. The NUE in F3 treatments was the highest (51.98%), which significantly increased by 7.27%-14.62% and 2.38%-11.37% compared with F1 and F2 treatments, respectively. With the equal N rate, NUE increased with the increase of number of top-dressing events. The highest NUE was observed in T3

treatments with averaged NUE as 53.82%, which was 12.66%-21.65% and 5.46%-12.97% significantly greater than compared with T1 and T2 treatments, respectively. The NUE in topdressing treatments increased by 4.46–33.38% compared with CF treatment. Statistical results showed that N application rate, topdressing times, topdressing times × N application rate interaction pronouncedly influenced dry weight and N accumulation of Chinese cabbage. Each factor's contribution to the total variation was ranked as: topdressing times > N application rate > their interaction.

Previous studies found that NUE was pronouncedly affected by fertilizer application rate, which decreased

Treatment	Dry weight (kg hm ⁻²)	N accumulation(kg hm ⁻²)	Nitrogen application rate(kg hm ⁻²)	Nitrogen use efficiency (%)
F × T				
CK	5824.48g	45.56g	0	-
CF	6973.87ef	96.80d	170	30.14
T1F1	7022.23ef	95.57de	144.5	34.60
T1F2	7012.12ef	92.55e	119	39.49
T1F3	6955.84f	84.71f	93.5	41.87
T2F1	7643.41b	107.40b	144.5	42.79
T2F2	7430.97c	101.13c	119	46.69
T2F3	7119.14e	92.82e	93.5	50.55
T3F1	7881.47a	116.22 a	144.5	48.90
T3F2	7530.99bc	107.63b	119	52.15
T3F3	7275.07d	104.95b	93.5	63.52
Nitrogen app	lication rate			
F1	7515.71a	106.40a	-	42.10
F2	7324.69b	100.43b	-	46.13
F3	7116.68c	94.16c	-	51.98
Topdressing t	times			
T1	6996.73c	90.94c	-	38.13
T2	7397.84b	100.45b	-	46.13
T3	7562.51a	109.60 a	-	53.82
Source of var	iance			
F	<0.0001	<0.0001	-	-
Т	<0.0001	<0.0001	-	-
$F \times T$	<0.0001	0.0030	-	-

Table 2. Nitrogen fertilizer use efficiency of Chinese cabbage with different treatments.

Note: Means followed by the similar lowercase letters within same column of each item were not significant in difference at 5% level, and the P < 0.01 indicated the very significant level of 1%, '-'no valid value.

with increasing rate of applied N fertilizer [26–28]. In the present study, the same tendency was found that there was also a negative relationship between NUE and N application rate (Table 2). NUE in CF treatment was lower than in F1 treatment because the higher N application rate in CF treatment released more N into deep soil, which led to higher N losses than from F1 treatment. Similarly, NUE increased with increase topdressing times, three times topdressing prolonged the period of N supply, promoted the uptake of N by plants, and reduced N release into deep soil. Our observation was consistent with the result of Ghaley [29].

2.3. Effects of different treatments on chinese cabbage yield

Chinese cabbage has a great demand for N, which promoted the growth of leaf bulbs and increased Chinese cabbage yield (Table 3). Statistical results showed that N application rate, topdressing times, topdressing times × N application rate interaction significantly influenced Chinese cabbage yield. Regarding yield, the contribution of each factor to the total variation of was ranked in the following order: N application rate > topdressing times > their interaction. Compared with CK treatments, the yield in other fertilization treatments were significantly increased by 17.65%-31.72%. With 15% urea reduction, the yield in T1F1, T2F1 and T3F1 treatments increased by 20.38%, 27.52%, 31.72% compared to CK treatment, respectively, by 1.42%, 7.43% and 10.97% as compared with CF treatment, respectively. Under the condition of 30% urea reduction, compared with CK treatment, the yield of T2F2 and T3F2 treatments increased by 22.49% and 25.00%

Table 3. Yield of Chinese cabbage with different treatments.

respectively, and compared with CF treatment, the yield increased by 3.19% and 5.31% respectively. Under the condition of urea reduction 45%, F3 treatments was not significantly increased compared with CF treatment. Under the condition of once topdressing, F3 treatments cabbage yield was not significantly increased than CF treatment. Under the condition of twice topdressing, Chinese cabbage yield of T2F1 and T2F2 treatments were 27.52% and 22.49% higher than CK treatment, 7.43% and 3.19% higher than CF treatments, respectively. Under the condition of three times topdressing, the yield of T3F1, T3F2 and T3F3 increased by 31.72%, 25% and 20.59 respectively compared with CK treatment, and increased by 10.97%, 5.31% and 1.59% respectively compared with CF treatment. The yield of T3F1 was the highest in all treatments, which was 31.72% higher than CK treatment, and 10.97% higher than CF treatment.

As for the multiple topdressing protocol, the soil N availability maintains at reasonable abundant levels in later stages, in which greatest amount of N is required by plants [30–32]. This is more advantageous than basal N application alone, in which N is very susceptible to loss [33]. Topdressing was enough to provide N for the maximum N use efficiency of Chinese cabbage.

2.4. Effects of different treatments on the quality of chinese cabbage

The concentration of Vc, nitrate, soluble sugar in cabbage were important indexes to prove the quality of Chinese cabbage, which directly affects the nutritive value of Chinese cabbage (Table 4). Statistical results showed that N application rate, topdressing times, topdressing times \times N application rate interaction significantly

Treatment Yield(t hm ⁻²)		Increase yield vs CK (%)	Increase yield vs CF (%)	
F × T				
СК	125.00h	-	-	
CF	148.37g	18.70	-	
T1F1	150.47ef	20.38	1.42	
T1F2	148.57e	18.86	0.13	
T1F3	147.06ef	17.65	-0.88	
T2F1	159.40f	27.52	7.43	
T2F2	153.11b	22.49	3.19	
T2F3	148.11d	18.49	-0.18	
T3F1	164.65ef	31.72	10.97	
T3F2	156.25a	25.00	5.31	
T3F3	150.74c	20.59	1.59	
Nitrogen application ra	ate			
F1	158.18a	26.54	6.61	
F2	152.64b	22.12	2.88	
F3	148.63c	18.91	0.18	
Topdressing times				
T1	148.70b	18.96	0.22	
T2	153.54b	22.83	3.48	
T3	157.21a	25.77	5.96	
Source of variance				
F	<0.0001	-	-	
Т	<0.0001	-	-	
F×T	<0.0001	-	-	

Note: Means followed by the similar lowercase letters within same column of each item were not significant in difference at 5% level, and the P < 0.01 indicated the very significant level of 1%, '-'no valid value.



Table 4. Chinese cabbage quality under different treatments.

Treatment	Vc concentration (g 100g ⁻¹)	Nitrate concentration (mg kg^{-1})	Soluble sugar concentration (mg kg^{-1})
F × T			
CK	10.71g	140.45g	1.42g
CF	13.01de	242.18a	1.98cd
T1F1	13.82bc	206.13d	1.99cd
T1F2	12.59e	191.00e	1.92de
T1F3	10.92g	171.88f	1.81f
T2F1	14.16b	217.60c	2.09b
T2F2	12.87e	206.98d	1.97d
T2F3	12.10f	189.88e	1.87ef
T3F1	14.80a	223.90b	2.33a
T3F2	13.44cd	214.08c	2.06bc
T3F3	12.77e	201.90d	1.91de
Nitrogen applicat	ion rate		
F1	14.26a	215.87a	2.14a
F2	12.97b	204.02b	1.98b
F3	11.93c	187.90c	1.86c
Topdressing time	S		
T1	12.44c	189.67c	1.91c
T2	13.04b	204.81b	1.98b
T3	13.67a	213.30a	2.10a
Source of varianc	e		
F	<0.0001	<0.0001	<0.0001
Т	<0.0001	<0.0001	<0.0001
$F \times T$	0.0008	0.0181	0.0002

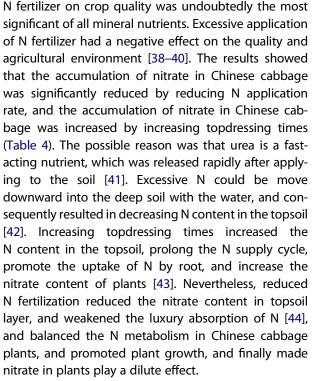
Note: Means followed by the similar lowercase letters within same column of each item were not significant in difference at 5% level, and the P < 0.01 indicated the very significant level of 1%, '-'no valid value.

influenced Vc and soluble sugar content of Chinese cabbage. Each factor's contribution to the total variation was ranked according to the following sequence: N application rate > topdressing times > their interaction. Both N application rate and topdressing times significantly influenced nitrate content, there was no significant influence of their interaction observed.

Vc content of cabbage in T3F1 treatment was the highest among all treatments and significantly increased by 13.69% compared with CF treatment. Nitrate content was related to the safety of Chinese cabbage. In China, the safety limit of nitrate content in fresh vegetable was less than or equal to 432 mg kg⁻¹ [34]. The nitrate content of different fertilizer treatments were 171.88–242.20 mg kg^{-1} , which was within the safety limits. The content of nitrate in cabbage increased linearly with the increase of N application. Compared with CF treatment, T3F1 treatment significantly reduced nitrate content by 7.55%. It indicated that combination of reduced N application rate by 15% and three times topdressing could not only significantly increase the yield of Chinese cabbage but also effectively reduce the accumulation of nitrate to the Chinese cabbage. Soluble sugar concentration of cabbage was an important index to determine the taste of vegetables. High soluble sugar content means good quality. Soluble sugar content in M3F1 treatment was the highest, reaching 2.33 mg kg^{-1} , and increased 17.97% compared with CF treatment.

N nutrient is the most important mineral element required by plants, and it is an important basic condition for biological construction [35,36]. Meanwhile, it also plays a key role in plant growth and development and material transformation [37]. Therefore, the effect of

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Under the condition of reduced N application rate, Vc and soluble sugar content of Chinese cabbage increased by increasing topdressing times. The possible reason was that topdressing with urea promoted the absorption of N and increased the absorption of other elements [45], thus enhanced the content of Vc and soluble sugar in Chinese cabbage.

3. Conclusion

Under the experimental conditions, the highest yield $(164.65 \text{ t } \text{hm}^{-2})$ of Chinese cabbage was observed in

T3F1 treatment which was 10.97% higher than CF treatment. Compared with CF treatment, 30% or 45% lower N still maintained stable yield. Reduced N application rate significantly decreased the nitrate content in edible part of Chinese cabbage, and correspondingly reduced the supply of nitrate N in soil. Increased topdressing times significantly improved the content of Vc and soluble sugar in Chinese cabbage and enhanced Chinese cabbage yield. Moreover, it effectively increased NUE and maintained higher nitrate content in the topsoil due to prolonging N supply period. These results showed that increased topdressing times and reduced N fertilizer could not only keep or even improve Chinese cabbage yield and guality but also cut down environmental risk. Vegetables were a large quantity of agricultural products with high demand for fertilizers, facing severe environmental pressure in production. In future, more research should be undertaken under different environmental conditions, as well as in different soil types.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

The research was funded by National Key R&D Program of China (2016YFD0201202).

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