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Communications in Soil Science and Plant Analysis

ISSN: 0010-3624 (Print) 1532-2416 (Online) Journal homepage: http://www.tandfonline.com/loi/lcss20

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To cite this article: Lu Yan-Li, Bai You-Lu, Ma Da-Ling, Wang Lei & Yang Li-Ping (2018): Nitrogen Vertical Distribution and Status Estimation Using Spectral Data in Maize, Communications in Soil Science and Plant Analysis, DOI: 10.1080/00103624.2018.1424893

To link to this article: https://doi.org/10.1080/00103624.2018.1424893

COMMUNICATIONS IN SOIL SCIENCE AND PLANT ANALYSIS
https://doi.org/10.1080/00103624.2018.1424893

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Nitrogen Vertical Distribution and Status Estimation Using Spectral Data in Maize

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ABSTRACT

Nitrogen (N) is often distributed in a vertical gradient and the symptom of N deficiency usually appears earlier in the lower leaves than in the upper ones. As for hyperspectral technology, identifying the sensitive leaves and the effective index are the two key factors for timely N topdressing. The results of this paper showed that the N contents in the lower leaves produced significant difference among different N rate treatments ($a = 0.001$) and showed an extremely significant relationship with soil nitrate nitrogen at 30 days after emergence. Taking the optimal treat as a reference, Nitrogen Stress Index (NSI) and Nitrogen Spectral Stress Index (NSSI) of the lowest two leaves could indicate plant N deficiency and guide nitrogen topdressing. NSI or NSSI could change regularly with the nitrogen stress degree and indicate the necessary amount of N topdressing. NSSI is relatively a better indicator since it comes from a fast, timely, uncontaminated, and nondestructive method.

ARTICLE HISTORY
Received 17 March 2017
Accepted 8 August 2017

KEYWORDS
Lower leaves; nitrogen; spectral index

Introduction

Nitrogen (N) is one of the key factors in plant photosynthesis, ecosystem productivity, and leaf respiration. Low nitrogen-use efficiency (NUE) of crop has always been a concern in N management, resource saving, and environment protecting (Hou, Zhong, and Obrien 2002; Liu et al. 2015; Zhao et al. 2011). Unreasonable N input cannot be fully utilized by crop and is usually lost through ammonia volatilization and nitrate leaching in soil. Early fast and accurate N diagnosis is the precondition for instant and effective nitrogen regulation in crop production. The development of hyperspectral technology has provided an effective way for fast, nondestructive, and real-time monitoring of the N nutrition status (Alt, Kage, and Stützel 2000; Gebbers and Adamchuk 2010; Huang et al. 2004; Lu et al. 2009). Frels (2015) proposed that canopy spectral reflectance combined with genomic selection can be used in breeding for NUE in hard winter wheat.

Conventional method mainly uses canopy spectra to predict N status, which has proved more effective in estimating the plant N status and consequent grain quality or yield in late growth stage (Lu et al. 2009; Morier, Cambouris, and Chokmani 2015). However, canopy spectra are not practically available due to the complexity of canopy characteristics, variation in leaf internal structure, and soil background interference (Asner 1998; Matson et al. 1994). In addition, the canopy spectra vary with measurement time, direction, and environmental factors (Mistele and Schmidhalter 2008). Therefore, more research is needed in the critical stage and maximum efficient stage of N nutrition for highly efficient N topdressing.

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Nitrogen is also one of the most mobile constituents of the plant; the upper leaf tends to use remobilized N from lower leaves and stems for growth when N is deficient (Vouillot and Devienne-Barret 1999; Wang et al. 2005). Vertical distribution of nutrients and photosynthetic activity are not always consistent during crop development (Bertheloot, Martre, and Andrieu 2008) and are responsive to N status (Lötscher, Stroh, and Schnyder 2003). Winterhalter, Mistele, and Schmidhalter (2012) demonstrated that the changes in both leaf nitrogen uptake and leaf biomass with growing stages are in shape of a vertical bell, and that of the nitrogen content is descendant from top to bottom. Klem et al. (2014) also proposed that plants respond to N deficiency or other abiotic stress initially by changing vertical distribution of nutrients and pigments, aimed at optimization of photosynthetic efficiency and prevention of upper leaves from stress impact.

Most of the former studies focused on monitoring the crop canopy to diagnose N status by spectral technique (Inoue et al. 2012; Miphokasap et al. 2012; Xue et al. 2004). However, canopy spectral information is mixed with the plant, soil, and other unknown information. It produced less accuracy and lagging indicators because it is more difficult to abstract the accurate target information. Ciganda, Gitelson, and Schepers (2008) and Winterhalter, Mistele, and Schmidhalter (2012) had investigated nitrogen vertical profile and determined some spectral indices or functions to reflect chlorophyll or nitrogen status. Some limitations still exist in practical use of the hyperspectral techniques. An important limitation is that reflectance is scanned preferentially from upper leaf layers, while different vertical layers of crop canopy make different contributions to the spectral reflectance. The existing models of nitrogen prediction using spectroscopy are not satisfactory in predicting accuracy and universal applicability because of environmental difference. It normally leads to the delay and blindness in nitrogen recommendation.

Huang et al. (2014) showed that N deficiencies usually exhibit in the bottom layer leaves, while excessive nitrogen will affect the upper layer leaves first. Zhou and Wang (2003) suggested that the upper leaf was better indicative of N supplying status when using leaf biomass or growth rate as an indicator but the lower leaf was more suitable as a test sample for diagnosis of N status by leaf chemical analysis.

The ratio (relative value) of N content in the deficiency treatment to that of the N in the sufficiency treatment is relatively stable and can be used to guide topdressing. Nitrogen sufficiency treatment is taken as the optimal treatment that had highest yields under the seasonal conditions at that time. Nitrogen Nutrition Index is determined by dividing the actual N concentration by a critical N concentration and developed to determine the in-season N status of many species (Ziadi et al. 2010). The same goes for some spectral parameters. Zhao et al. (2011) proposed that the rates of topdressing N and total fertilizer N are increased with the increase of relative soil and plant analyzer development (SPAD) threshold values.

The objectives of this study were (1) to clarify the temporal-spatial distribution of N in different layers at different growth stages and at various N rates; (2) to analyze the relationship between leaf N and soil total N content (TN), nitrate nitrogen (NN), and soil organic matter (SOM), and then determine the most sensitive leaves to N supply; and (3) to verify the suitable indices that would be used as indicators of N status, and to make decision for fertilizer application.

Materials and method

Experimental site description

A field experiment was conducted in a long-term experimental area with different N rates initiated from October 2008, located at the International Agricultural Technology Demonstration Park of the Chinese Academy of Agricultural Sciences (N 39°35′47.03′′, E 116° 35′16.24′′) in Langfang City, Hebei Province. The region has warm and sub-humid continental monsoon climate with the average annual temperature 11.9 °C, annual average frost-free period 183 d, average annual precipitation 554.9 mm, and the mean annual sunshine 2660 h. The winter

wheat-summer maize rotation is the main cropping system in this area with conventional irrigation management.

The experimental data are from 2012 to 2013; during the experimental period, the wheat straw was completely returned into soil after wheat harvest and maize was sowed after the soil was rotary tilled in the early October. The tested summer maize cultivar was ND108 and the planting density was 7500 plants/hm^2. No plant diseases and insect pests occurred during the experiment seasons.

Experiment design

Seven N rates of 0, 60, 120, 180, 240, 300, and 360 kg·ha^{-1}, denoted as N0, N1, N2, N3, N4, N5, and N6, respectively, were set up in the long-term experiment. The N rates represent N supply levels of very deficient, deficient, medium, sufficient, and very sufficient in turn. The optimal rate is 180 kg·ha^{-1}, which was recommended by ASI method (Hunter 1984). The applied fertilizers were urea (46%N), calcium superphosphate (12% P_2O_5), and K_2SO_4 (50%K_2O). For all the treatments, P (P_2O_5) of 90 kg·ha^{-1} and K (K_2O) of 60 kg·ha^{-1} were all basal applied, 60% of the total N was basal applied, and 40% top dressed in elongation stage. The plot size was 100 m^2 (8 m × 12.5 m) and arranged in order of N rates.

Basic soil property

The field experiment was conducted in a fluvo-aquic soil with sandy texture. The fertility in top 20 cm of the soil is as follows: pH, 8.1 (soil:water = 1:2.5); organic matter, 11.7 g·kg^{-1}; alkali-hydrolyzable N, 56.74 mg·kg $^{-1}$; Olsen-phosphorus (P), 11.9 mg·kg $^{-1}$; and exchangeable potassium (K), 43.1 mg·kg $^{-1}$ (ISS and CAS 1980).

Sample collection, processing, and analysis

The basic soil samples were collected from 0 to 20 cm layer before sowing and other soil samples were collected in 30, 41, and 54 days after emergence (corresponding probably V6, V12, and VT, respectively). Leaf samples were collected synchronously from the uppermost fully expanded leaves to the bottom and every two leaves were taken as a sample, and denoted as L(1,2), L(3,4), L(5,6), and L(7,8), respectively. All samples were desiccated at 105 °C for 30 min and oven-dried at 80 °C in a forced air circulation oven until constant and then ground to pass a 2-mm sieve to determine the nutrient content. At harvest, each plot was harvested separately and yield in the plot was recorded.

Leaf samples were digested using the sulfuric acid (H_2SO_4)-hydrogen peroxide (H_2O_2) method and the N content was determined by distillation. The soil pH was determined by electrode method, alkaline hydrolysis of N by diffusion method, and available P by Olsen method (Olsen et al. 1954). Soil available K was extracted using 1 mol·L^{-1} ammonium acetate and determined by an anatomic absorption spectrophotometer. Soil organic matter was determined by Walkley and Black method (1934).

Measurement of spectral reflectance

The spectra of different leaf layers were measured with an ASD FieldSpec 1 Pro FR spectrometer with a high-intensity contact probe (PANalytical, B.V, Boulder, Colorado, USA, formerly Analytical Spectral Devices). This spectrometer was fitted with 5° field of view fiber optics, operating in the 350–2500 nm spectral region. The sampling interval between 350 and 1050 nm was 1.4 nm and that between 1050 and 2500 nm was 2 nm. The spectral resolution at 700 nm was 3 nm and 10 nm at 1400 nm. A 40 × 40 cm^2 $BaSO_4$ calibration panel was used for reflectance calculation, and the resampling interval was 1 nm.

After the samples were collected as described in the section "Sample collection, processing, and analysis", the spectra were detected with the high-intensity contact probe by clamping the leaf and avoiding veins. Five positions were measured from tip to basal of every leaf, and at each measurement the instrument made 10 internal scans for a satisfactory signal-to-noise ratio; 10 spectra were

averaged into one spectrum to represent the spectra of the position and the average of 5 positions was for one leaf, and finally the average of a leaf layer (2 leaves) was for a sample.

Results and discussion

Nitrogen vertical distribution in different growing stages

In N0 treatment, the leaf N contents were kept low in the three sampling stages, and those in treatments were increased significantly with the increased N fertilizer rate ($F = 23.15 > F_{0.001} = 10.39$,

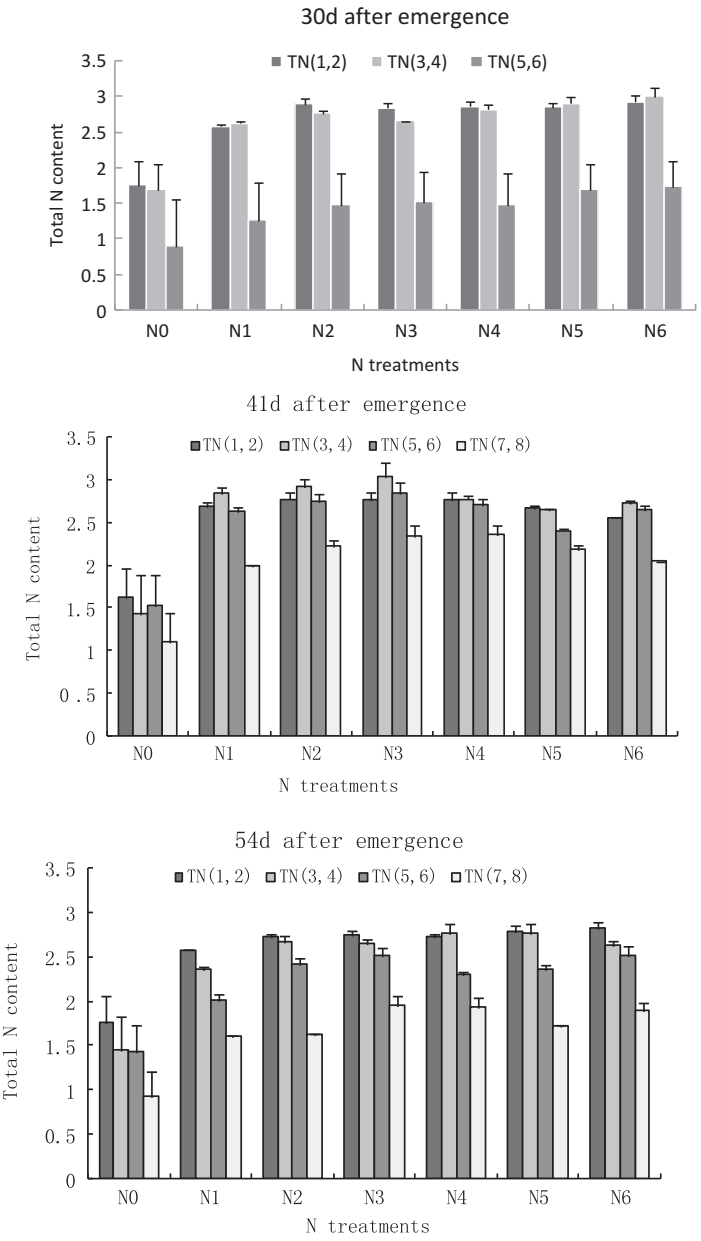

Figure 1. Total Nitrogen (TN) vertical distribution in maize.

P= 0.000001). However, with the increase in the amount of nitrogen, leaf nitrogen could not increase anymore and even presented a downward trend after the rate of 180 kg·ha^{-1}, especially in the middle and lower leaves (Figure 1). The above results indicate that the excessive nitrogen could not be absorbed by plant, but probably resulted in environmental contamination. The results also showed that the leaf N content was higher in upper and middle positions than in lower position, but the differences between any two treatments were bigger in lower leaves than in the upper leaves. So, it was initially concluded that the lower leaves were more sensitive to N rate.

Nitrogen correlation analysis between aboveground and underground

As can be seen in Table 1, the dynamic changes of N content in leaves depend on the TN in the soil to some degree, and there are different N correlations between soil and leaves in different stages. A statistically significant correlation existed between soil TN and leaf N in lower leaves. The relationship between N content of uppermost leaf group and soil TN was getting stronger after jointing stage; the N content of the lower leaves kept the strong relationship with soil TN in all three sampling stages. This result indicated that the lower leaves responded to the change in soil TN earlier than the upper leaves in the critical stage for fertilizing.

NN is the dominant form of available nitrogen in the experimental area. Basically, NN reflects more the ability of the soil N supply and total nitrogen represents more the N capacity (Bai, Yang, and Jin 2007). Compared with TN, NN had a similar relationship with N content in leaves, but stronger than soil TN did (Table 2), which indicates that NN decides leaf N status to a much larger extent.

Soil OM is another N indicator, which had a statistically significant relationship with N content in lower leaves in 30 days after emergence, but had no significant relationship in the following stages (Table 3). The tested soil was sandy fluvo-aquic soil with poor nutrient retention, and OM content was so low that there was even no significant difference between different treatments.

Compared to TN and OM, NN had a dominant effect on leaf N, and the significant correlation between NN and N in the lower leaves presented that the lower leaves can respond sensitively to the change in available N.

Spectral variation in response to the nitrogen in different layers

As can be seen in Figure 2, there was a statistically significant correlation between spectral reflectance in visible band (around green peak) and the N content of different leaf groups. The correlation analysis showed that N content in middle and lower leaves in 30 days after emergence had a significant correlation with spectral reflectance in 523–603 nm and 699–718 nm (α = 0.01), and with the elongation of growing process, the N content in all four groups showed statistically

Table 1. The correlation between leaf N and total soil N.

			Total soil N in the day after emergence		
Sampling time		Leaf group	30 d	41 d	54 d
Leaf N in the day after emergence	30 d	L(1,2)	0.434	0.485	0.511
		L(3,4)	0.441	0.260	0.543
		L(5,6)	0.729**	0.512	0.632*
	41 d	L(1,2)	0.559*	0.625*	0.425
		L(3,4)	0.522	0.435	0.391
		L(5,6)	0.354	0.538*	0.587*
		L(7,8)	0.521	0.582*	0.513
	54 d	L(1,2)	0.411	0.594*	0.495
		L(3,4)	0.474	0.421	0.519
		L(5,6)	0.493	0.388	0.485
		L(7,8)	0.552*	0.426	0.662**

*p = 0.05; **p = 0.01.

Table 2. The correlation between leaf N and oil nitrate nitrogen (NN).

			Soil nitrate N in the day after emergence		
Day after emergence		Leaf group	30 d	41 d	54 d
Leaf N in the day after emergence	30 d	L(1,2)	0.456	0.387	0.426
		L(3,4)	0.531	0.579*	0.513
		L(5,6)	0.742**	0.527	0.511
	41 d	L(1,2)	0.703**	0.590*	0.710**
		L(3,4)	0.627*	0.602*	0.669**
		L(5,6)	0.547*	0.534*	0.498
		L(7,8)	0.625*	0.845**	0.690**
	54 d	L(1,2)	0.573*	0.673**	0.794**
		L(3,4)	0.649*	0.586*	0.759**
		L(5,6)	0.574*	0.459	0.621*
		L(7,8)	0.679**	0.548*	0.611*

*p = 0.05; **p = 0.01.

Table 3. The correlation between leaf N and soil organic matter (SOM).

			Soil organic matter in the day after emergence		
		Leaf group	30 d	41 d	54 d
Leaf N in the day after emergence	30 d	L(1,2)	0.317	0.386	0.420
		L(3,4)	0.476	0.531	0.464
		L(5,6)	0.542*	0.617*	0.540*
	41 d	L(1,2)	0.485	0.320	0.305
		L(3,4)	0.435	0.183	0.258
		L(5,6)	0.209	0.456	0.522
		L(7,8)	0.351	0.467	0.600
	54 d	L(1,2)	0.450	0.411	0.398
		L(3,4)	0.234	0.164	0.431
		L(5,6)	0.126	0.171	0.251
		L(7,8)	0.380	0.452	0.446

*p = 0.05.

significant correlation with their reflectance in visible bands. The correlations were different in near-infrared bands among the different leaf groups, and got the maximum and significant coefficient in the layer of 5–6 leaf groups in 41 days after emergence. The correlation between spectral reflectance and N content of L(7-8) showed significantly negative in 521–587 nm and 699–737 nm, but significantly positive in 760–1363 nm in 54 days after emergence.Those determined bands lie in the strong absorption region of Chlorophyll and infrared region aroused by leaf tissues (Pu and Gong 2000). The significant correlation provided a feasible way to reflect N status using spectral data in maize.

Yield response to different nitrogen fertilizer rates

ANOVA showed that the yield differences were significant among the treatments (F = 8.67>F0.05 = 2.84, p= 0.0005), with the highest yield in N rate of 180 kg·ha^{-1} and the lowest in the control. Multiple comparison results showed that the application of nitrogen fertilization produced a great effect on yield, whatever it was, deficiency or excessive. In the experiment, N0 and N1 were the N deficiency treatments and showed significantly lower than N3 in yield. While N5 and N6 were N excessive treatments, the yields in both treatments were significantly lower than that in N3 either. Additionally, the yields in N2 and N4 were lower than in N3 but did not reach the significant level. So N rate of 180 kg·ha^{-1}(N3) was confirmed to be the optimal treatment in this experiment (Table 4).

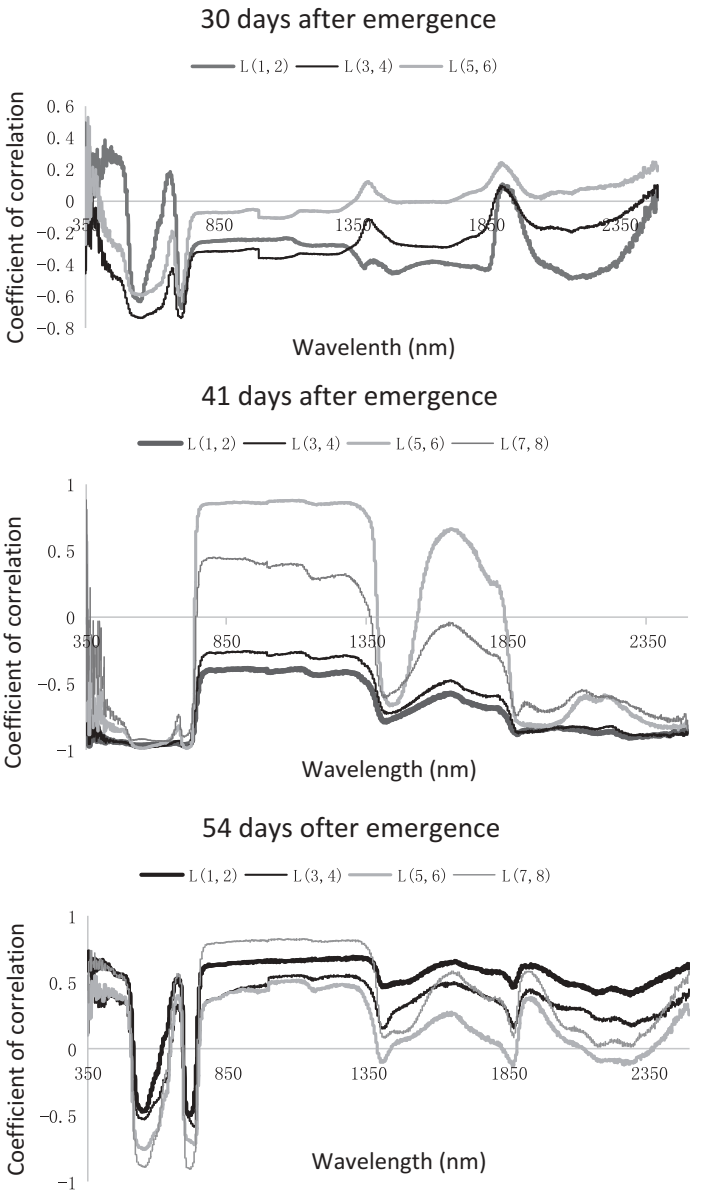

Figure 2. The correlation between reflectance and N content in different leaf groups.

Fertilization decision based on sensitive indices

Nitrogen stress index

Plant N ratio of the N deficiency treatment to the N sufficiency one has been proved effective for N diagnostics (Zhao et al. 2011; Ziadi et al. 2010). In this study, the N ratio of the optimal treatment N3 to other treatments (denoted as NSI, Nitrogen Stress Index) was calculated to determine the plant N status. Taking the optimal treatment NSI as 1, the NSI in lower leaves of the N deficiency treatments was less than 1, and the more deficient led to the greater difference between the N deficiency and N optimal treatments. As can be seen in Table 5, when the N applied rate was in deficient and serious

Table 4. Multiple comparison in different treatments.

Treat	Yield I (kg·ha^{-1})	Yield II (kg·ha^{-1})	Yield III (kg·ha^{-1})	Standard Error (SE)	Average (kg·ha^{-1})
N3	11640	10830	11700	26.45	11385a
N4	10665	11100	10215	24.09	10650ab
N2	10995	9975	10110	30.16	10350b
N5	10140	9930	10065	5.79	10050b
N6	10425	9855	9735	20.07	10005b
N1	10365	9870	9645	20.05	9960bc
N0	7980	7710	8835	31.97	8175c

Note: There is no significant difference between groups if they contain same letter, but it is opposite for different letters.

Table 5. Evaluation of the N deficiency diagnosed by NSI in different sampling time.

	30 d			41 d				54 d			
Treat	L(1,2)	L(3,4)	L(5,6)	L(1,2)	L(3,4)	L(5,6)	L(7,8)	L(1,2)	L(3,4)	L(5,6)	L(7,8)
N0	0.617	0.637	0.590	0.585	0.472	0.535	0.470	0.642	0.548	0.573	0.479
N1	0.908	0.990	0.834	0.969	0.933	0.924	0.844	0.942	0.895	0.804	0.818
N2	1.017	1.042	0.969	1.001	0.956	0.964	0.950	0.991	1.010	0.965	0.829
N3	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
N4	1.006	1.063	0.974	1.001	0.911	0.951	1.002	0.993	1.047	0.919	0.992
N5	1.001	1.093	1.111	0.963	0.872	0.845	0.934	1.018	1.046	0.940	0.980
N6	1.030	1.129	1.148	0.924	0.896	0.928	0.870	1.027	0.995	0.905	0.974

deficient treatments, the NSIs were less than 1 in the lowest two leaves denoted as L(5,6) in 30 d and L(7,8) in 41 d and 54 d. The NSIs in serious deficient treatment were lower than deficient group, which indicates that the NSI could indicate the N deficiency level. The result also indicated that the lower leaf group was feasible to diagnose N status in maize, and NSI also can guide topdressing quantitatively in the earlier growing stage in case of knowing the recommended amount for the whole stage and the basal rate.

Nitrogen spectral stress index

Some reflectances in single or combined bands and indices have been successfully generated to test N status in plants, such as Normalized Difference Vegetation Index and Ratio Vegetation Index (Ferwerda, Skidmore, and Mutanga 2005; Inoue et al. 2012; Miphokasap et al. 2012; Stroppiana et al. 2009). In this study, five spectral parameters based on the sensitive regions were selected and constructed to be as Nitrogen Spectral Stress Index (NSSI). They are reflectance in 550 nm which is sensitive band to chlorophyll, reflectance in 680 nm which is sensitive band to chlorophyll and nitrogen, normally named red edge, and reflectance in 1090 nm which is the important band of infrared area to N. It has been proved an effective method by making normalization to the reflectance in visible and near-infrared bands (Rouse et al. 1974). The formulas of other two indices based on above sensitive bands used in this study are as follows:

$$\mathrm{NVI}\ (680, 890) = (R_{890} - R_{680})/(R_{890} + R_{680})$$
$$\mathrm{NVI}\ (570, 670) = (R_{570} - R_{670})/(R_{570} + R_{670})$$

Corresponding to NSI, the NSSI is the ratio of spectral index of N deficiency treatment to that of N sufficiency one. Here only the lower leaves, which have already demonstrated to be the most sensitive to the plant N status, were used for NSSI. As can be seen in Table 6, higher NSSI came with stronger N stress in 550 and 680 nm, while it was opposite in 1090 nm which showed the lower NSSI under the more serious N stress. The NSSI from NVI(680,890) and NVI(570,670) also proved to be the better parameters to reflect plant N status. If the NSSI of N3 was 1, the NSSI (680,890) was less than 1, and the NSSI (570,670) was more than 1 when deficiency happened (Table 7). The values changed regularly with the stress degree in deficient plots, which showed that NSSI can indicate effectively the plant N status.

Table 6. NSSIs from reflectance in different days after emergence and different N rates.

	R550			R680			R1090		
	Days after emergence			Days after emergence			Days after emergence		
Treat	30 d	41 d	54 d	30 d	41 d	54 d	30 d	41 d	54 d
N0	1.574	1.743	1.347	1.108	1.304	1.089	0.823	0.913	0.936
N1	1.356	1.607	1.198	1.124	1.158	1.020	0.954	0.936	0.994
N2	1.223	1.439	1.133	1.052	1.087	0.972	1.015	0.961	1.005
N3	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000	1.000
N4	1.034	0.932	0.949	0.959	0.966	0.983	1.023	1.076	1.017
N5	0.973	0.922	0.993	0.895	0.973	0.939	1.008	1.128	1.017
N6	0.893	0.969	0.975	0.879	0.908	0.754	1.021	1.116	1.002

Table 7. NSSIs from normalized vegetation indices (NVI) in different days after emergence and different N rates.

	NVI[670,890]			NVI□570,670□		
	Days after emergence			Days after emergence		
Treat	30 d	41 d	54 d	30 d	41 d	54 d
N0	0.781	0.851	0.854	1.676	1.318	2.134
N1	0.881	0.916	0.923	1.409	1.230	1.222
N2	0.906	0.939	0.980	1.187	1.170	1.053
N3	1.000	1.000	1.000	1.000	1.000	1.000
N4	1.011	1.033	1.011	0.939	0.890	0.999
N5	1.021	1.020	1.097	0.971	0.845	0.972
N6	1.023	1.032	1.020	0.948	0.915	0.976

Conclusions

Nitrogen status of the lower leaves was proved to be more sensitive to spectral reflectance and significantly correlated with soil N level. Taking optimal treatment N3 (180 kg·ha^{-1}) as a reference, the ratio of other treatments to N3 treatment was a good indicator to N deficiency. The merit of the ratio method is its universal applicability; it is not affected by regional environment provided the sufficient treatment is set. In this paper, NSI and NSSI were calculated based on chemical and spectral analyses, respectively. The NSSI(670, 890) and NSSI(570,670) were better indicators, which, compared with NSI, provided a fast, timely, uncontaminated, and nondestructive method to detect N deficiency in the field.

Funding

This work was supported by the National Natural Science Foundation of China (Nos. 41371292 & 31572207).

References

Alt, C., H. Kage, and H. Stützel. 2000. Modelling nitrogen content and distribution in cauliflower (Brassica oleracea L. botrytis). *Annals of Botany* 86:963–73. doi:10.1006/anbo.2000.1252.

Asner, G. P. 1998. Biophysical and biochemical sources of variability in canopy reflectance. *Remote Sensing of Environment* 64:234–53. doi:10.1016/S0034-4257(98)00014-5.

Bai, Y. L., L. P. Yang, and J. Y. Jin. 2007. *Principles and practice of soil testing and formula fertilization*, pp. 47. Beijing: China Agricultural Press. (in Chinese).

Bertheloot, J., P. Martre, and B. Andrieu. 2008. Dynamics of light and nitrogen distribution during grain filling within wheat canopy. *Plant Physiology* 148:1707–20. doi:10.1104/pp.108.124156.

Ciganda, V., A. Gitelson, and J. Schepers. 2008. Vertical profile and temporal variation of chlorophyll in maize canopy: Quantitative crop vigor indicator by means of reflectance-based techniques. *Agronomy Journal* 100:1409–17. doi:10.2134/agronj2007.0322.

Ferwerda, J. G., A. K. Skidmore, and O. Mutanga. 2005. Nitrogen detection with hyperspectral normalized ratio indices across multiple plant species. *International Journal of Remote Sensing* 26:4083–95. doi:10.1080/01431160500181044.

Frels, K. 2015. *Breeding for nitrogen use efficiency in hard winter wheat using canopy spectral reflectance and genomic selection*. San Diego, CA: Plant & Animal Genome XXIII.

Gebbers, R., and V. I. Adamchuk. 2010. Precision agriculture and food security. *Science* 327:828–31. doi:10.1126/science.1183899.

Hou, Y. L., G. R. Zhong, and L. Obrien. 2002. Translocation of nitrogen and fructan in wheat nutritorium. *Scientia Agricultura Sinica* 35 (9):1066–70. (in Chinese with English abstract).

Huang, W. J., Q. Y. Yang, R. L. Pu, and S. Y. Yang. 2014. Estimation of nitrogen vertical distribution by bi-directional canopy reflectance in winter wheat. *Sensors* 14 (11):20347–59. doi:10.3390/s141120347.

Huang, Z., B. J. Turnera, S. J. Durya, I. R. Wallisb, and W. J. Foley. 2004. Estimating foliage nitrogen concentration from HYMAP data using continuum removal analysis. *Remote Sensing of Environment* 93:18–29. doi:10.1016/j.rse.2004.06.008.

Hunter, A. H. 1984. *Soil fertility analytical services in Bangladesh*. A consultancy report. Dhaka, Bangladesh: Bangladesh Agricultural Research Council.

Inoue, Y., E. Sakaiya, Y. Zhu, and W. Takahashi. 2012. Diagnostic mapping of canopy nitrogen content inrice based on hyperspectral measurements. *Remote Sensing of Environment* 126:210–21. doi:10.1016/j.rse.2012.08.026.

ISS and CAS (Institute of Soil Science, Chinese Academy of Science). 1980. *Analysis of physical and chemical characteristic*. China: Shanghai Science and Technical Publishers. (in Chinese).

Klem, K., P. Rajsnerová, K. Novotná, P. Míša, and J. Křen. 2014. Changes in vertical distribution of spectral reflectance within spring barley canopy as an indicator of nitrogen nutrition, canopy structure and yield parameters. *Agriculture (Polnohospodárstvo)* 60 (2):50–59. doi:10.2478/agri-2014-0006.

Liu, J., A. Zhan, H. Chen, S. Luo, L. Bu, X. Chen, and S. Q. Li. 2015. Response of nitrogen use efficiency and soil nitrate dynamics to soil mulching in dry land maize (zea mays, L.) fields. *Nutrient Cycling in Agroecosystems* 101 (2):271–83. doi:10.1007/s10705-015-9678-5.

Lötscher, M., K. Stroh, and H. Schnyder. 2003. Vertical leaf nitrogen distribution in relation to nitrogen status in grassland plants. *In Annals of Botany* 92:679–88. doi:10.1093/aob/mcg188.

Lu, Y.-L., S.-K. Li, Y.-L. Bai, C. L. Jones, and J.-H. Wang. 2009. Canopy spatial distribution and identification using hyperspectal data in winter wheat. *Communications in Soil Science and Plant Analysis* 40:1240–53. doi:10.1080/00103620902754598.

Matson, P., L. Johnson, C. Billow, J. Miller, and R. Pu. 1994. Seasonal patterns and remote spectral estimation of canopy chemistry across the oregon transect. *Ecological Applications* 4:280–98. doi:10.2307/1941934.

Miphokasap, P., K. Honda, C. Vaiphasa, M. Souris, and M. Nagai. 2012. Estimating canopy nitrogen concentration in sugarcane using field imaging spectroscopy. *Remote Sensing* 4:1651–70. doi:10.3390/rs4061651.

Mistele, B., and U. Schmidhalter. 2008. Spectral measurements of the total aerial N and biomass dry weight in maize using a quadrilateral-view optic. *Field Crops Research* 106:94–103. doi:10.1016/j.fcr.2007.11.002.

Morier, T., A. N. Cambouris, and K. Chokmani. 2015. In-season nitrogen status assessment and yield estimation using hyperspectral vegetation indices in a potato crop. *Agronomy Journal* 107 (4):1295–309. doi:10.2134/agronj14.0402.

Olsen, S. R., C. V. Cole, F. S. Watanabe, and L. A. Dean. 1954. *Estimation of available phosphorus in soils by extraction with sodium bicarbonate. U.S. Dep. of Agric. Circ. 939*. Washington, D.C.: U.S. Government Printing Office.

Pu, R. L., and P. Gong. 2000. *Hyperspectral remote sensing and its applications*. Beijing: Higher Education Press.

Rouse, J. W., R. H. Haas, J. A. Schell, D. W. Deering, and J. C. Harlan. 1974. Monitoring the Vernal advancement of retrogradation of natural vegetation. In *NASA/GSFC, Type, Final Report*, eds. Greenbelt, MD, USA: Nasa. pp. 1–371.

Stroppiana, D., M. Boschetti, P. A. Brivio, and S. Bocchi. 2009. Estimation of plant nitrogen concentration in paddy rice from field canopy spectra. *European Journal of Remote Sensing* 41:45–57.

Vouillot, M. O., and F. Devienne-Barret. 1999. Accumulation and remobilization of nitrogen in a vegetative winter wheat crop during or following nitrogen deficiency. *Annals of Botany* 83:569–75. doi:10.1006/anbo.1999.0861.

Walkley, A. J., and I. A. Black. 1934. An examination of the degtjareff method for determining soil organic matter, and a proposed modification of the chromic acid titration method. *Soil Science* 37 (1):29–38. doi:10.1097/00010694-193401000-00003.

Wang, Z. J., J. H. Wang, C. Zhao, M. Zhao, W. J. Huang, and C. Z. Wang. 2005. Vertical distribution of nitrogen in different layers of leaf and stem and their relationship with grain quality of winter wheat. *Journal of Plant Nutrition* 28 (1):73–91. doi:10.1081/PLN-200042175.

Winterhalter, L., B. Mistele, and U. Schmidhalter. 2012. Assessing the vertical footprint of reflectance measurements to characterize nitrogen uptake and biomass distribution in maize canopies. *Field Crops Research* 129:14–20. doi:10.1016/j.fcr.2012.01.007.

Xue, L., W. Cao, W. Luo, T. Dai, and Y. Zhu. 2004. Monitoring leaf nitrogen status in rice with canopy spectral reflectance. *Agronomy Journal* 96 (1):135–42. doi:10.2134/agronj2004.0135.

Zhao, S. C., P. He, S. J. Qiu, X. P. Xu, L. Chuan, and S. L. Xing. 2011. Application of relative SPAD values for nitrogen fertilizer management of different cultivars of summer maize. *Plant Nutrition and Fertilizer Sciences* 17 (5):1091–98. (in Chinese with English abstract).

Zhou, Q. F., and J. H. Wang. 2003. Comparison of upper leaf and lower leaf of rice plants in response to supplemental nitrogen levels. *Journal of Plant Nutrition* 26 (3):607–17. doi:10.1081/PLN-120017668.

Ziadi, N., G. Bélanger, A. Claessens, L. Lefebvre, N. Tremblay, A. N. Cambouris, M. C. Nolin, and L.-É. Parent. 2010. Plant-based diagnostic tools for evaluating wheat nitrogen status. *Crop Science* 50:2580–90. doi:10.2135/cropsci2010.01.0032.