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Effect of Chlorophyll Meter and Leaf Colour Chart Based Nitrogen Management on Grain N Uptake for Rice in Iran

M. VALYNEJAD*, S. VASEGHI†, V.M. FALLAH‡, E. PAZIRA†,
A. FALLAH‡ and A. MOAMENI‡
Department of Soil Sciences, Islamic Azad University, Science and Research Branch, Tehran, Iran
Tel: (98)(911)3143817; E-mail: maryamvaly2000@yahoo.com; mvalinejad@rocketmail.com

Over application of N in rice leads to low N recovery efficiency and risk of NO_3 pollution of ground water. The chlorophyll meter also known as SPAD meter, is a simple, portable diagnostic tool for identifying crop N status. We used it to test need based N management approaches for rice (*Oryza sativa* L.) in two varieties on a clay loam texture in northern Iran. The overall objective is to develop optimal N-management for Neda and Shirudi. Two varieties were grown under eight treatments. Eight treatments included a zero-N control, chlorophyll meter 35, 37, 40, LCC 4 and 5. SPAD readings were taken weekly on the topmost fully expanded leaves. Grain yield, yield attributes, total N uptake and fertilizer - N use efficiency were measured in both varieties. Both LCC and SPAD can be used to improve N management for Neda and Shirudi, but the optimal SPAD threshold for determining the timing of N-application was 35 and 37 for Neda and Shirudi, respectively. LCC treatments indicated that N-management based on LCC shade 5 helped avoid over application of N to rice. Neda had higher grain yield than shirudi, but the difference in maximum grain yield among SPAD and LCC treatments was insignificant. The two varieties didn't show a clear difference in fertilizer-N use efficiency. It is suggested that when N-management technology such as real time N-management (SPAD and LCC) were used, would avoid to over application of N fertilizer by rice farmers.

Key Words: Chlorophyll meter, Leaf colour chart, N management, Rice.

INTRODUCTION

Nitrogen is very important nutrient that most limits rice production. It is typically required in greater quantities than any other nutrient, if rice farmers are to reap high yield and profits. Inappropriate N management also has the determinal-major effects on crop yield and environment and -agreevates disease and pest incidence and it leads to further lowering of N fertilizer recovery efficiency (RE), which is already not more than 50 %[1,2].

†Department of Soil Sciences, Islamic Azad University, Science and Research Branch, Tehran, Iran.
‡Department of Soil and Water, Rice Research Center, Amol, Iran.

Due to the predominantly alkaline reaction of the soil, urea top dressed in rice is preferentially lost *via* NH_3 volatilization. In highly permeable soil profiles with alternating aerobic and anaerobic soil conditions under rice, applied N release readily converted to NO_3 which is prone to loss *via* leaching, nitrification, denitrification or both[2,3]. Furture high levels of NO_3-N in the regions ground water have been reported recently.

Blanket fertilizer recommendation do not take into account the high field to field variability and within-season dynamic changes in indigenous N supply. Since the plant reflects the total N-supply from all sources, plant N status will be a good indicator of N availability to crops at any given time. The chlorophyll meter (SPAD) and leaf colour chart (LCC) are simple, portable diagnostic tools that can measure the crop N status *in situ* in rice fields to determine the timing of N top dressing. When SPAD value is less than the set critical reading, N fertilizer should be applied. The critical colour shade on the LCC needs to be determined to guide N applications. Such decision aids are useful to vary N application rates to rice crops based on crop demand and indigenous N supply[4].

In the Philippine dry season, application of 30 kg N ha^{-1} to rice cultivar IR72 when SPAD value was below the critical value of 35 resulted in higher agronomic efficiency (AE), compared with recommended splits. The threshold to be reduced to 32 for wet season[4]. Another study carried out in south India (IRRI-CREMENT, 1998), a value of 37 was found to be critical for obtaining high yields and N use efficiency of short-satured improved indica cultivars. In India, during the Kharif and Rabi season (2000-2001) application of 30 kg N ha^{-1} to rice cultivars when SPAD value was below 35 resulted in higher grain yield and N use efficiency compared with recommended splits (45, 90 and 135 kg N ha^{-1}). These studies indicate the need for determining chlorophyll meter threshold values of different rice-growing environments. In fixed time N management, one-third of 125 kg N ha^{-1} for rice has been recommended to be applied basally immediately before soil puddling and rice transplanting. Many farmers also apply a dose of N about 1 week after transplanting in lieu of basal application. As rice seedling take *ca.* 7 d to recover from transplanting shock[5], it is likely that most of the N applied around 7 d after transplanting (DAT) is not used by plants and is lost. As chlorophyll meter-guided N management in rice starts at 2 week after transplanting, the usefulness of applying a dose of N basally or at *ca.* 7 DAT needs to be examined.

A field experiment described in this study was carried out during 2007 through 2008 to refine the fixed critical reading approaches for N management using chlorophyll meter. It is also evaluated need-based N management strategies for rice using LCC. The objectives were (i) compare three SPAD values of 35, 37 and 40 as threshold values in rice in northern Iran. (ii) Compare the efficiency of two critical LCC values for guiding fertilizer application in rice. (iii) Evaluate the feasibility of decreasing N inputs to protect the environment without reducing crop yield.

EXPERIMENTAL

General site characteristics: Field experiments were conducted on Typic dysterudept in Mazandaran Rice Research center of IRAN in 2008. The experimental sites were located in Amol (28′36º N, 23′52º E, 29.8 maltitwe) the area receives on average 7432 mm year^{-1} rainfall. Mean maximum and minimum temperatures are 31.2 and 15.3 ºC during rice cropping (April to August). Soils are well drained Table-1 shows the physical and chemical properties of soil sample (0-15 cm) field site.

TABLE-1
SOIL (0-15 cm) PROPERTIES OF RICE EXPERIMENT SITE (2008)

Sand (%)	Clay (%)	pH	EC (ds/m)	Organic carbon (%)	TNV (%)	P olsen (mg kg^{-1})	K ammonium 1 M acetate (mg kg^{-1})
22	34.6	6.92	1.8	1.7	20	5.8	240

Nitrogen treatments: Experiment was laid out in a randomized complete block design with three replicates with rice varieties of Neda and Shirudi. Eight fertilizer N (as urea) management treatments are described in Table-2. In the recommended splits N was applied at transplanting and 7 days after transplanting, midtillering and panicle initiation. Chlorophyll meter reading, were taken weekly starting from 2 weeks after transplanting and 16 kg N ha^{-1} was applied whenever SPAD readings were below the critical value of 35, 37 and 40. Thershold SPAD values were picked up from studies carried out by peng and his colleagues[6].

Two treatment with LCC consisted of applying N (16 kg ha^{-1}) when rice leaf colour was less than shade 4 (LCC < 4 and 5). Control plot received a full dose of phosphorous, potassium and zinc but no N.

TABLE-2
TREATMENTS USED IN RICE EXPERIMENTS DURING 2008, MAZANDARAN, IRAN

Number	Treatment details	Time of N application
Zero-N control	Zero N control	–
Split 1	Fixed-N split with total N rate of 138 kg N ha^{-1}	Application of N in three equal splits at basal, early tillering and panicle initiation
Split 2	Fixed-N split with total N rate of 138 kg N ha^{-1}	Application of N in three equal splits at Seven days after transplanting, early tillering and panicle initiation
S35	16 kg N ha^{-1} with 30 kg ha^{-1} at basal	Whenever SPAD < 35
S37	16 kg N ha^{-1} with 30 kg ha^{-1} at basal	Whenever SPAD < 37
S40	16 kg N ha^{-1} with 30 kg ha^{-1} at basal	Whenever SPAD < 40
LCC4	16 kg N ha^{-1} with 30 kg ha^{-1} at basal	Whenever SPAD LCC< 4
LCC5	16 kg N ha^{-1} with 30 kg ha^{-1} at basal	Whenever SPAD LCC< 5

Crop establishment and management: Widely grown varieties of rice *i.e.*, Neda and Shirudi were used. Transplanting was done in April. Seedling age was 20-35 d. Transplanting spacing was 20 cm × 20 cm with two seedlings per hill. Plot size was 12 m^2. Phosphorus at 50 kg ha^{-1}, potassium at 100 kg ha^{-1} and zinc at 25 kg ha^{-1} were applied at basal. The plots were kept flooded throughout the growing season. Pests, disease and weeds were intensively controlled to avoid yield loss. Crops were harvested by hand at ground level at maturity on 26 August, 2008. Grain and straw yields were determined from an area (5 m^2) located at the center of each plot. Grains were separated from straw using a plot thresher, dried in a batch grain dryer and weighed. Grain moisture was determined, immediately after weighing and subsamples were dried in an oven at 65 ºC for 48 h. Grain weights for rice expressed at 140 g kg^{-1} water content. Straw weights were expressed on oven dry basis.

Plant sampling and analysis: Grain and straw subsamples were dried at 70 ºC and finely ground to pass through a 0.5 mm sieve. Nitrogen content in grain and straw was determined by digesting the samples in sulfuric acid (H_2SO_4), followed by analysis for total N by a micro-Kjeldahl method[7]. The N in grain plus that in straw was taken as a measure of total N uptake.

Chlorophyll meter and leaf colour chart measurements: Chlorophyll meter reading weekly with a Minolta SPAD-502 chlorophyll meter, starting 14 DAT. Ten hills of rice were chosen at random in each plot. From each hill three reading were taken from the uppermost fully expanded leaf. SPAD reading were taken up to 50 % flowering stage.

The leaf colour chart (LCC) developed by IRRI 1996 consisted of six green strips showing increasing greenness with increasing number. As with the chlorophyll meter, the chart was use to take weekly reading starting 15 DAT. Ten disease free rice plants were randomly selected in the plot and the colour of the youngest fully expanded leaf of selected plants was compared by placing its middle part on top of the colour strip in the chart. Like chlorophyll meter, reading were taken up to 50 % flowering stage. If more leaves read below a critical value (LCC 4 and 5) a dose of 16 kg N ha^{-1} was applied.

Data analysis: Analysis of variance was performed on yield parameters to determine effect of N management treatments using SAS. Duncan's multiple range tests were used at 0.05 level of probability to test difference, between treatment means. The N use efficiency measures, RE[8] and AE[9], were calculated as follows:

RE (%) = [(TNU in N-fertilized plot - TNU in zero - N plot)/
quantity of N fertilizer applied in N-fertilized plot] × 100

AE (kg grain/kg N applied) = (grain yield in N-fertilized plot- grain yield in
zero- N plot)/quantity of N fertilizer applied in N-fertilized plot

where, TNU is the total N uptake in grain and straw.

RESULTS AND DISCUSSION

Grain yield of two varieties responded to N application (Tables 3 and 4).

TABLE-3
GRAIN YIELD AND YIELD ATTRIBUTES OF NEDA AND SHIRUDI GROWN UNDER EIGHT TREATMENT

Treatments	N rate (kg/ha)	Grain yield (t/ha)	Panicle (m^{-2})	Grain filling (%)	Grain weight (mg)
Neda					
Zero-N control	0	6.48 ^{d}	348^{d}	84.4^{a}	30.5^{a}
Split 1	138	8.16 ^{c}	433^{bc}	84.3^{a}	31.6^{a}
Split 2	138	8.36 ^{b}	467.5^{ab}	82.9^{a}	29.5^{a}
S_{35}	62	8.77^{abc}	397^{cd}	84.9^{a}	30.4^{a}
S_{37}	78	8.81^{abc}	383^{cd}	84.3^{a}	31.2^{a}
S_{40}	94	8.84^{abc}	462^{ab}	84.3^{a}	30.6^{a}
LCC4	94	9.06^{ab}	422^{bc}	84.8^{a}	31.5^{a}
LCC5	94	9.59^{a}	493^{a}	87.4^{a}	29.9^{a}
Shirudi					
Zero-N control	0	5.67^{d}	308^{c}	84.8^{a}	26.9^{ab}
1	138	6.46^{c}	408^{ab}	84.9^{a}	26.1^{b}
2	138	6.93b^{c}	422.5^{ab}	84.5^{a}	26.5^{ab}
S_{35}	78	7.04^{abc}	379^{ab}	85.3^{a}	27.7^{a}
S_{37}	78	7.17^{ab}	372^{bc}	81.2^{ab}	26.9^{ab}
S_{40}	94	7.38^{ab}	420^{ab}	82.7^{ab}	26.7^{ab}
LCC4	78	6.98^{bc}	437^{ab}	81.2^{ab}	26.1^{b}
LCC5	78	8.71^{a}	441^{a}	80.5^{ab}	27.1^{ab}

Note: Within a column for each variety, means followed by the same letters are not significantly different according LSD (0.05).

A significant positive response of rice grain yield to N application relative to zero - N control was observed in all treatments for both cultivars. There was no significant difference in grain yield among S_{35}, S_{37}, S_{40} and LCC_5 for Shirudi. Grain yield of LCC treatments was greater than SPAD treatments in Neda. Yield loss was observed in shirudi when the LCC threshold was 4 and a significant positive response of grain yield relative to other treatment was observed in LCC 5. LCC 5 produced grain yield comparable with the LCC 4 and S40 at similar N rates. Grain yield of real time N management (LCC and SPAD treatments) was higher than that of the best splits with less N rate. Harvest index decreased as total N rates increased in LCC and SPAD treatments in both varieties. Panicle number responded significantly to N application (Table-3).

A significant increase in panicle number was observed when total N rates were 94 kg N ha^{-1} in Neda (S_{40}, LCC4 and LCC5). The same happened in shirudi but the increase was insignificant.Grain filling percentage showed no significant difference among N application treatments relative to the zero-N control in both varieties. The same was observed in grain weight. When N-fertilizer was not applied, N uptake from the indigenous sources ranged from 80 to 81 kg N ha^{-1} (Table-4).

TABLE-4
N UPTAKE, TOTAL FERTILIZER N APPLIED AND RECOVERY AND AGRONOMIC EFFICIENCIES (RE AND AE, RESPECTIVELY) OF TWO RICE CULTIVARS USING DIFFERENT NEED-BASED FERTILIZER N MANAGEMENT AT MAZANDARAN, IRAN

Treatment	N rate (kg/ha^{-1})	Total N uptake (kg ha^{-1})	RE (%)	AE (kg kg^{-1})
Neda				
Zero-N control	0	80^{c}	–	–
Split 1	138	157^{a}	58.6^{a}	12^{c}
Split 2	138	162^{a}	57.2^{a}	13.6^{c}
S_{35}	62	105^{b}	40.3^{b}	36.9^{a}
S_{37}	78	111^{b}	39.7^{b}	29.8^{b}
S_{40}	94	109^{b}	31.5^{b}	25.1^{b}
LCC4	94	111^{b}	33.6^{b}	27.4^{b}
LCC5	94	113^{b}	35.8^{b}	33.0^{a}
Shirudi				
Zero-N control	0	81^{d}	–	–
Split 1	138	160^{a}	57.2^{a}	5.72^{d}
Split 2	138	162^{a}	58.6^{a}	9.1^{c}
S_{35}	78	117^{c}	46.1^{ab}	17.5^{b}
S_{37}	78	111^{c}	38.5^{b}	19.2^{b}
S_{40}	94	125^{b}	47.8^{ab}	18.5^{b}
LCC4	94	130^{b}	56.3^{a}	32.3^{b}
LCC5	78	128^{b}	51.1^{a}	15^{b}

Note: Within a column for each variety, means followed by the same letters are not significantly different according LSD (0.05).

Total N uptake increased as N rates increased. Recovery efficiency ranged from 33.6 to 58.6 and 38.5 to 58.6 %. For Neda and Shirudi, respectively. Both N treatments and varieties did not show consistent and significant effect on RE. Large variation in AE was observed across N treatments. However, excessive N rates reduced AE to as 12 and 5.7 kg kg^{-1} in Neda and Shirudi, respectively. Neda generally had higher AE than shirudi at similar rates N input.

As a result, when SPAD value of 35, 37 and 40 was used as a critical value for applying 16 kg N ha^{-1}, a total of 62 to 94 kg N ha^{-1} was applied to both cultivars.

Accordingly, grain yield in these treatments showed a significant difference relative to two fixed time split in shirudi (Table-3). The same happened in Neda but grain yield increase was insignificant. As expected, AE was greater when less N fertilizer was used.

However SPAD treatments had the higher AE than two fixed time split and S_{35} and S_{37} had the highest AE among SPAD treatments for Neda and shirudi respectively. Based on this information, it is found that the SPAD threshold for optimizing N management is 35 and 37 for Neda and shirudi varieties, respectively.

These results are in agreement with the finding of peng and his colleagues[6], in the Philippine for indica varieties grown on the tropical conditions with the SPAD value of 35.

In south India a critical SPAD value of 37 was found to be appropriate for rice grown in the summer monsoon season[10]. It has also been suggested that different threshold SPAD values may have to be used for different varieties groups[11,12].

A total of 94 kg N ha^{-1} was applied following both LCC 4 and LCC 5. Grain yield showed increase in LCC 4 and 5 relative to other treatments for Neda. The same happened in LCC 5 for shirudi. Accordingly, AE in LCC 4 and 5 was significantly higher than two fixed time split for Neda. LCC 5 in shirudi had the highest AE among all treatments.

As a result, critical value of LCC 5 was more beneficial in enhancing the growth of Shirudi. Although, LCC treatments were significantly higher than split treatments, but there wasn't any significant difference between LCC 4 and 5 for Neda. Singh *et al.*[13] indicated that N management based on LCC shade 4 helped avoid over application of N to rice, in India. In Philippines, Islam *et al.*[14] found that application of N at 25 kg ha^{-1} based on biweekly LCC 5 observations was the best method of nitrogen management.

There was no clear difference in panicle m^{-2} between the two varieties. Real time N management (SPAD and LCC treatments) significantly increased panicle m^{-2} relative to zero-N control and two fixed-time splits. This was due to plant base N-management by SPAD and LCC.

There was no significant difference in grain filling and grain weight parameters among N treatments for both varieties, because of inefficient role of nitrogen in these parameters.

There was no clear difference in N uptake between the varieties. The two varieties had a similar ability to take up nitrogen from indigenous source. There was no significant difference in RE among N treatment or between the two varieties. High RE also observed in other N studies conducted in China[6]. This suggests that poor fertilizer-N use efficiency of the rice crop is not associated with N uptake ability. At similar N rate Neda generally had higher AE than shirudi. This difference was partially due to the zero-N control yield difference between Neda and Shirudi. The difference in grain yield between varieties might contribute to the difference in AE between them.

Conclusion

Both LCC and SPAD were effective in achieving high grain yield and agronomic efficiency in Neda and shirudi. The optimal SPAD threshold for guiding N management was 35 and 37 for Neda and shirudi. LCC shade 5 helped avoid over application of N to rice. Neda had higher grain yield but the difference on maximum grain yield among SPAD and LCC treatments was insignificant. The two varieties didn't show a clear difference in fertilizer use efficiency. We suggest that when N-management technology such as real time N-management (SPAD and LCC) were used, would avoid to over application of N fertilizer by rice farmers[14-18].

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