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Effects of Pretransplant Basal and Split Applications of Nitrogen on the Growth and Yield of Manawthukha Rice

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A field experiment was conducted to investigate the effects of pretransplant basal (surface application, BSF, and incorporation methods, BIC) and split applications of nitrogen (N) on the growth and yield parameters of rice. Using 120 kg N ha⁻¹ except (N0, control), different percentages of N rate were applied at basal, tillering, and panicle initiation in five N split treatments. Growth parameters and dry matter were greater in BIC than BSF until panicle initiation stages. Among N split applications, N2 (25:50:25) using low basal surface N was optimized for maximum dry matter and yield. With large incorporated basal N, N1 (50:25:25) obtained greater dry matter and yield but did not differ from N4 (50:50:0). With omitted N at tillering, N5 (50:0:50) did not increase rice yield or dry matter by either method. This study highlighted that N split-application patterns affect the growth and yield parameters of Manawthukha rice.

Keywords Basal methods, growth parameters, N split-application patterns, rice, yield

Introduction

Rice (*Oryza sativa* L.) is a staple food crop for half the world's population and is one of the most important global food crops. To assure food security in the rice-consuming countries of the world, farmers need to produce more rice with improved qualities to meet the demand of consumers in the coming years (Peng and Yang 2003).

Nitrogen (N) is a commonly limiting nutrient in crop production. Cereals including rice accounted for approximately 50% of the worldwide N fertilizer used in 2009 (IFA 2009). However, excessive application rates and inappropriate application methods have led to low N efficiency and high fertilizer losses through runoff, leaching, denitrification,

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and volatilization (Kirda, Derici, and Schepers 2001), resulting in a series of environmental problems. The efficiency of N recovery in rice plants is low. Based on a worldwide evaluation, N-recovery efficiency has been found to be approximately 30% in rice (Krupnik et al. 2004). Low N efficiency also increases production costs, results in crop lodging, increases pest and disease susceptibility, and leads to lower net returns for farmers (Wang et al. 2001). The development and promotion of more efficient N fertilizer management in rice consequently remains a high priority in terms of increasing profitability in rice farming while protecting the environment (Buresh et al. 2004).

Most Asian farmers broadcast urea directly into the floodwater 2–4 weeks after transplanting rice (De Datta et al. 1979). Vlek and Fillery (1984) reported that the major problem with broadcast application of N fertilizers is the development of high concentrations of urea and/or ammonium in the flood water and surface layer of soil where the major loss mechanisms, namely ammonia volatilization, nitrification–denitrification, and surface runoff, operate. They also suggested that the concentration of fertilizer-source N in the floodwater might be reduced by deep placement of the fertilizer; the use of slow-release fertilizer, nitrification inhibitors, or urease inhibitors; incorporation of the N into the soil; or the split application of the fertilizer. Several techniques have been devised in the past to reduce N losses and improve N-use efficiency in rice. Using urease inhibitors can delay the conversion of urea to ammonium (Vlek, Byrnes, and Craswell 1980a). Vlek, Stumpe, and Byrnes (1980b) also reported that the incorporation of 1% w/w of phenyl phosphor-diamidate (PPD) to urea was able to delay the appearance of ammonia N in floodwater by blocking the urease activity of the soil–water interface, which is the primary site for hydrolysis of urea in floodwater. Similar findings were reported by Chaiwanakupt et al. (1996). However, the anticipated cost of urease inhibitors restricts their use, and these techniques may have limited the success in the field.

In this study, basal application methods were conducted just before transplanting. The timing of basal N application was different from that used by farmers under field conditions. Normally, N application before transplanting can lead to N losses before rice plants take up nutrients. We sought to determine whether basal surface N application with tight water management could provide optimal N-recovery efficiency and growth of rice plants. Additionally, it was considered whether the incorporation or deep placement of urea into the anaerobic soil zone might be beneficial for improving N-use efficiency, growth, and yield of rice while protecting the environment. Deep placement of urea can prevent the rapid conversion of ammonium to nitrate and thereby prevent denitrification-based N losses. It has been shown that the deep placement of N fertilizer is an effective means of reducing ammonia concentrations in floodwater and drastically reducing ammonia volatilization (Vlek and Craswell 1981). High N-fertilizer efficiency in rice can be achieved through N-efficient rice varieties and improved timing and application methods of N fertilizers and improved incorporation of basal fertilizer without standing water (Ali et al. 2007).

The judicious use of N fertilizer in rice requires balancing N fertilizer application rates with the needs of plants. Split application is one strategy for the efficient use of N fertilizer throughout the growing season. By synchronizing N application with plant demands, denitrification losses can be reduced and N uptake improved, resulting in maximum straw and grain yield and an enhanced harvest index (Lampayan et al. 2010). Multiple split applications of mineral N fertilizer can reduce N losses and increase N-use efficiency (Cassman, Kropff, and Zhen-De 1994). In contrast, Singh et al. (1991) concluded that increasing the number of split doses of urea from 3 to 10 was not helpful in enhancing the efficiency of urea N in permeable soil under lowland rice cover in India. Increasing the number of splits

to six also had no effect (Belder et al. 2005). Nonetheless, N application has great impact on crop yield in rice when applied during the early and mid-tillering stages, producing a high number of panicles, optimum spikelets per panicle, and a high percentage of filled spikelets (Murty, Dey, and Jachuk 1992). To achieve high yields, N-application timing should be balanced with plant uptake to fulfil crop requirements before and after anthesis (Mahajan, Chauhan, and Gill 2011). Ha and Suh (1993) reported that an adequate and balanced supply of N promoted vigorous vegetative growth and a deep green color of the crop. They also found that by balancing N application with plant requirements, the utilization of phosphorus (P), potassium (K), and other plant nutrients was improved, resulting in enhanced crop growth.

The optimum split dose of N fertilization plays a vital role in the growth and development of rice plants (BRRRI 1990). Proper timing and rates of N application are also crucial to minimizing N losses (De Datta and Buresh 1989). Generally, nutrient absorption characteristics vary with the rice cultivar, fertilizer type, fertilization technology, soil type, and environmental factors (Huang et al. 2008). The amount of nutrient absorbed also varies with rice growth stage (Liu et al. 2007). Therefore, optimizing the split dose of N to different critical growth stages is essential for improving the growth and yield parameters of Manawthukha rice. It is also of interest to study whether basal N application methods affect the dry-matter accumulation, growth, and yield of rice.

This study was conducted (a) to determine the appropriate split-application pattern of N fertilizer to critical growth stages required to improve the growth and yield parameters of Manawthukha rice and (b) to examine the differences in growth and dry-matter accumulation of rice using basal surface application and incorporation methods.

Materials and Methods

Experimental Site

A field experiment was conducted at Kyushu University farm in Fukuoka Prefecture, Japan (33° 37' N, 130° 25' E) from June to October 2012 to evaluate the effects of split-application patterns and different basal application methods of N fertilizer on growth, yield, and yield parameters of Manawthukha rice.

Experimental Design and Treatments

The experiment was conducted in a split-plot design with three replications. Each experimental plot measured 4.5 × 1 m. Basal fertilizer application methods were used on “main plots” using the basal incorporation (BIC) and basal surface application (BSF) methods. Split-application patterns of N fertilizer were applied to “subplots,” where N fertilizer was allocated at the critical growth stages of rice as listed in Table 1.

The land was irrigated so as to be easily plowed and was subsequently harrowed and divided into two parts (as the main plot area), one using the BIC method and the other the BSF method. Bunds were created to prevent seepage between adjacent plots. A plastic lining was installed to a depth of 15 cm between the drains and the bunds toward the drains. Split N (as urea), 60 kg phosphorus pentoxide (P₂O₅) ha⁻¹ (as superphosphate), and 60 kg potassium oxide (K₂O) ha⁻¹ (as muriate of potash, potassium chloride; KCl) were basally incorporated at the last harrowing and levelling in the BIC method and broadcasted on the soil surface in the BSF method. The two basal applications were conducted 2 days before transplanting.

Table 1
Description of N split treatments in a split-plot design of field experiment

N treatment	Basal (%)	Active tillering (%)	Panicle initiation (%)
N0 (control)	0	0	0
N1	50	25	25
N2	25	50	25
N3	25	25	50
N4	50	50	0
N5	50	0	50

Note. The percentage of N application at each stage were based on 120 kg N ha⁻¹.

In the subplots, the split-application patterns of N fertilizer were applied at a rate of 120 kg N ha⁻¹, except for subplot N0 (N control), where N fertilizer was not applied. Different ratios in three split-application patterns were applied at basal, active tillering, and panicle initiation stages in N1 (50:25:25 at each stage, respectively), N2 (25:50:25), and N3 (25:25:50). Two split applications were conducted at basal and active tillering in N4 (50:50:0) and at basal and panicle initiation in N5 (50:0:50).

Soil Sampling and Analysis

Before conducting the field experiment, initial soil samples were collected from six locations in the experimental field at 0- to 20-cm depth using a 5-cm-diameter soil-sampling tube. Samples were spread, air dried at room temperature, crushed by hand, sifted through a 2-mm-mesh sieve, and stored for further analysis.

The soil texture was clay loam. The soil pH_{H2O} (1:2.5 soil/water) and soil pH_{KCl} (1:2.5 soil/KCl) were measured using a pH meter (Beckman ϕ 360 pH/Temp/mV Meter; Beckmann Coulter, Brea, Calif.). The nutrient contents of soil were extracted using the salicylic acid–sulfuric acid (H₂SO₄)–hydrogen peroxide (H₂O₂) digestion method (Ohyama et al. 1991); total N was determined using the indophenol method (Cataldo, Schrader, and Youngs 1974); total phosphorus (P) was analyzed using the ascorbic acid method (Murphy and Riley 1962); and total potassium (K) was measured using an atomic absorption spectrophotometer (Z-5300, Hitachi Ltd., Tokyo, Japan). Cation exchange capacity (CEC) and exchangeable cations were determined using the ammonium acetate shaking extraction method (Muramoto, Goto, and Ninaki 1992) followed by analysis using an atomic absorption spectrophotometer (Z-5300, Hitachi Ltd, Tokyo, Japan). Analysis of mineralizable N was performed with the soil incubation method (Sahrawat 1983) followed by the indophenol method (Cataldo, Schrader, and Youngs 1974). The available phosphorus (P) of soil samples was analyzed using Truog's method (Truog 1930) followed by the ascorbic acid method (Murphy and Riley 1962). The physicochemical properties of experimental soils are described in Table 2.

Crop Management

Rice (*Oryza sativa* L.) variety Manawthukha (Indica, Myanmar high-yielding variety) was cultivated for this study. This rice variety was already determined to be suitable for the Japanese environment by Dr. Umezaki, Mie University (Myint et al. 2011). The good seeds

Table 2
Physical and chemical properties of the surface (0–20 cm)
profile of soil at the experimental site

Physicochemical property	Value
Soil pH (soil/H ₂ O 1:2.5)	6.4
Soil pH (soil/KCl 1:2.5)	5.2
Total N (%)	0.13
Total P ₂ O ₅ (%)	0.24
Total K ₂ O (%)	0.48
Available N (mg N/100 g soil)	3.05
Available P (mg P ₂ O ₅ /100 g soil)	16.79
CEC (cmol _c kg ⁻¹)	13.18
Exc. Ca (cmol _c kg ⁻¹)	3.60
Exc. Mg (cmol _c kg ⁻¹)	0.80
Exc. K (cmol _c kg ⁻¹)	0.40
Exc. Na (cmol _c kg ⁻¹)	0.95

Notes. CEC, cation exchangeable capacity; Exc., exchangeable.

were chosen with a wind-blower machine. Then, the seeds were sterilized using hot water treatment. The seeds were immersed in hot water and shaken in hot water at 58 °C and 74 rpm for 15 min. The sterilized seeds were then washed with distilled water and stored in the incubator at 25 °C for 48 h without any light.

The incubated seeds were sown as two seeds per cell on the seedbeds using commercial seedbed soil (Kokuryu Baido, Seisin Sangyo Co., Kitakyushu, Japan). The seedbeds were placed in the pond, which was maintained at a shallow water level. The 21-day-old seedlings were transplanted at a hill spacing of 25 × 15 cm, with two seedlings per hill. Irrigation was conducted through a common management program with the surrounding area.

Plant Growth Characteristics

Three hills from each plot were marked for the determination of plant growth characteristics. Plant height (cm), tiller number, and soil and plant analysis development (SPAD) values were measured weekly from 10 days after transplantation (DAT) to the first flowering and at 2-week intervals after flowering using a chlorophyll meter (SPAD-502, Konica Minolta Sensing, Inc., Osaka, Japan). The top fully expanded leaf was chosen to measure the SPAD value before the panicle initiation stage, and the flag leaf was used after that stage.

Plant Sampling and Determination of Dry-Matter Accumulation

At active tillering, panicle initiation, and grain filling, two hills from each plot were harvested as destructive samples. The rice plants were cut 2–3 cm aboveground, oven dried at 70 °C for 48 hs, and then weighed immediately; values are expressed as (t ha⁻¹) for determination of dry-matter accumulation.

At harvesting time, the remaining 10 hills in each plot were harvested to determine total dry matter (t ha⁻¹), grain yield (t ha⁻¹), and yield components [(number of panicles

per hill, number of spikelets per panicle, filled grain (%), maximum panicle length (cm), and 1,000-grain weight (g)].

Statistical Analysis

The collected data were summarized and subjected to an analysis of variance (ANOVA). Comparison of means among treatments were performed using Tukey's honestly significant difference (HSD) test at a 5% probability level using STATISTIX 8 (Analytical Software, Tallahassee, FL, USA).

Results

Soil Analysis

Table 2 shows the physical and chemical properties of the surface (0–20 cm) soil at the experimental site.

Plant Growth Characteristics

Plant Height. Rice plant height was significantly affected by the two basal application methods at active tillering and panicle initiation stages (Table 3). The rice plants grown using the BIC method showed greater plant height than did those grown using the BSF method. However, at the time of harvest, no significant differences in plant height were observed between the two basal application methods. Regarding N split-application patterns, there were highly significant differences in plant height at each stage. The maximum plant height (96.93 cm) was recorded in group N4 (50:50:0) in which a large percentage of N was applied in two splits, at the basal and active tillering stages. However, these values were not statistically different from the height of plants in N2 (25:50:25) (95.65 cm). With heavy basal surface N, a lower plant height (94.65 cm) was observed in N1 (50:25:25). A large amount of panicle N fertilization also did not maximize the plant height (93.41 cm) in N3 (25:25:50). The plant height (88.78 cm) of N5 (50:0:50) was lower than that with the other treatments but greater than those of N0 (control).

Tiller Number. Before the panicle initiation stage, the tiller number per hill was significantly affected by basal application methods (Figure 1). A greater numbers of tillers were observed in plants grown using the BIC method. The differences in tiller number between the two basal application methods were large before panicle initiation, especially in N1 (50:25:25), N4 (50:50:0), and N5 (50:0:50), in which large amounts of basal N were applied. Less significant differences in tiller number between basal application methods were observed in N2 (25:50:25) and N3 (25:25:50), which had lower basal N applications. Throughout the crop period, the maximum tiller number (40 hill⁻¹) was recorded in N2 (25:50:25) at 52 DAT, which used N application synchronized with the rice plants' demand for N. A similar tiller number was obtained in N4 (50:50:0), in which a large amount of urea N was applied in two splits. Even though three split applications of N were applied, N3 (25:25:50) did not reach optimum tillering patterns throughout the crop period. At harvest, the greatest tiller number (19.33 hill⁻¹) was observed in N2 (25:50:25). Without N application at the active tillering stage in N5 (50:0:50), a lower tiller number (15.95 hill⁻¹) was obtained at harvest, but it was greater than the tiller numbers of N0 (control).

Table 3
Plant height (cm) of Manawthukha rice affected by N split-application patterns at the critical growth stages and harvest in two basal application methods

Treatment	Active tillering	Panicle initiation	Grain filling	Harvest
Basal application methods				
Surface application method	49.51 b	71.16 b	93.21 a	91.73 a
Incorporation method	51.69 a	73.23 a	93.31 a	92.50 a
Split-application patterns				
N0 (control)	46.73 c	62.73 c	87.31 c	83.24 c
N1 (50:25:25)	52.87 a	73.67 b	89.37 bc	94.65 a
N2 (25:50:25)	50.20 b	77.70 a	99.14 ab	95.65 a
N3 (25:25:50)	50.21 b	73.62 b	92.74 abc	93.41 a
N4 (50:50:0)	52.16 ab	79.14 a	99.54 a	96.93 a
N5 (50:0:50)	51.43 ab	66.31 c	91.46 abc	88.78 b
Source of variance				
Basal methods	0.0389	0.0426	ns	ns
N split	< 0.0001	< 0.0001	0.0031	< 0.0001
Methods × N split	ns	ns	ns	ns
CV (%)	2.74	2.87	5.80	2.24

Notes. Means followed by the same letter in each column and in each method are not significantly different by the Tukey HSD test ($P < 0.05$). The numbers in parentheses show the percentage of N applied based on 120 kg N ha⁻¹ at basal, active tillering, and panicle initiation stage. ns, nonsignificant difference.

SPAD Values (Chlorophyll Meter Reading). Prior to the panicle initiation stage, the SPAD values were significantly affected by basal application methods. The rice plants grown using the BIC method showed greater SPAD values than the same treatment using the BSF method (Figure 2). Large differences in SPAD values were observed between the BIC and BSF methods, particularly in N1 (50:25:25), N4 (50:50:0), and N5 (50:0:50), in which larger amount of basal N were applied. However, the application of lower basal N showed smaller differences in SPAD values between these two methods. Generally, the incorporation of urea N offered steady SPAD values over 35 throughout the growth of rice plants, especially in N4 (50:50:0). A similar pattern was observed in N2 (25:50:25), which showed SPAD values of approximately 35 throughout the crop period. With the lack of N application at the active tillering stage, the SPAD value was severely reduced, below 35, at the active tillering and panicle initiation stage in N5 (50:0:50). At the grain-filling stage, an increase in SPAD values occurred in each treatment including N0 (control).

Dry-Matter Accumulation. Significant differences in total dry-matter accumulation (TDM) were recorded between the two basal application methods at the active tillering stage. The incorporation of urea N into the soil provided the greatest TDM, while basal surface-applied N obtained a lower TDM (Figure 3). After this stage, no further differences in TDM were observed between the two methods. Even without split application, significant differences in TDM were observed between the two basal application methods in N5 (50:0:50) at the grain filling stage. However, N split-application patterns significantly

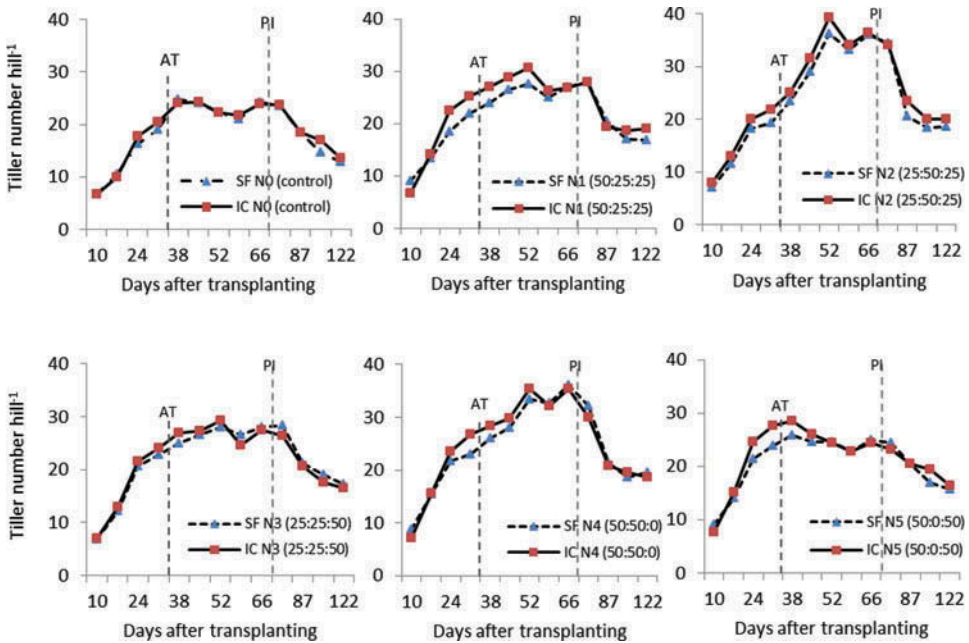


Figure 1. Tilling patterns of Manawthukha rice affected by different basal application methods and split application patterns of nitrogen. The numbers in parentheses show the percentage of N applied based on 120 kg N ha⁻¹ at basal, active tillering, and panicle initiation stage.

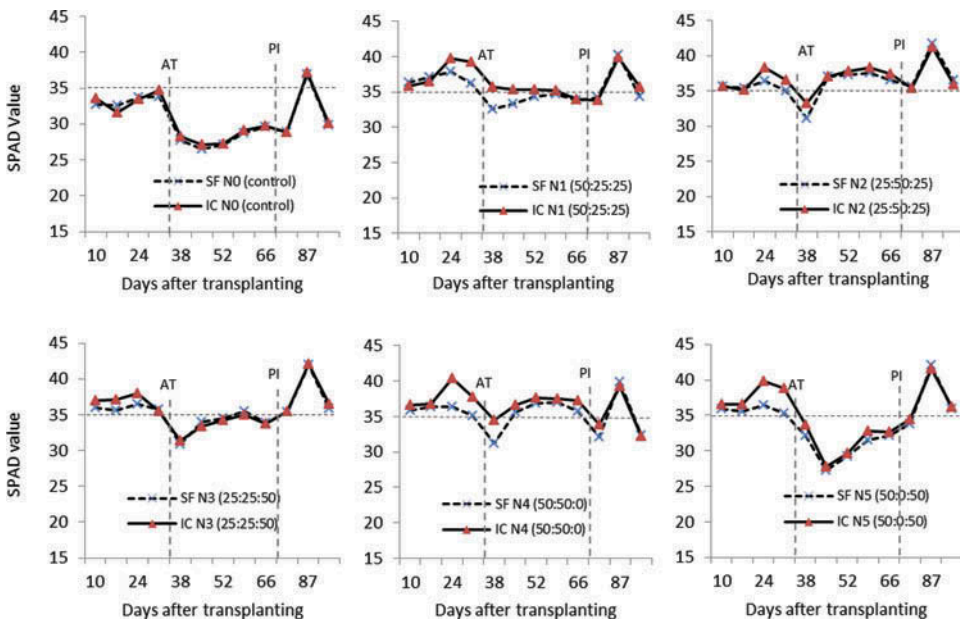


Figure 2. Changes in SPAD values affected by different basal application methods and split applications of nitrogen fertilizer in Manawthukha rice. The numbers in parentheses show the percentage of N applied based on 120 kg N ha⁻¹ at basal, active tillering, and panicle initiation stage.

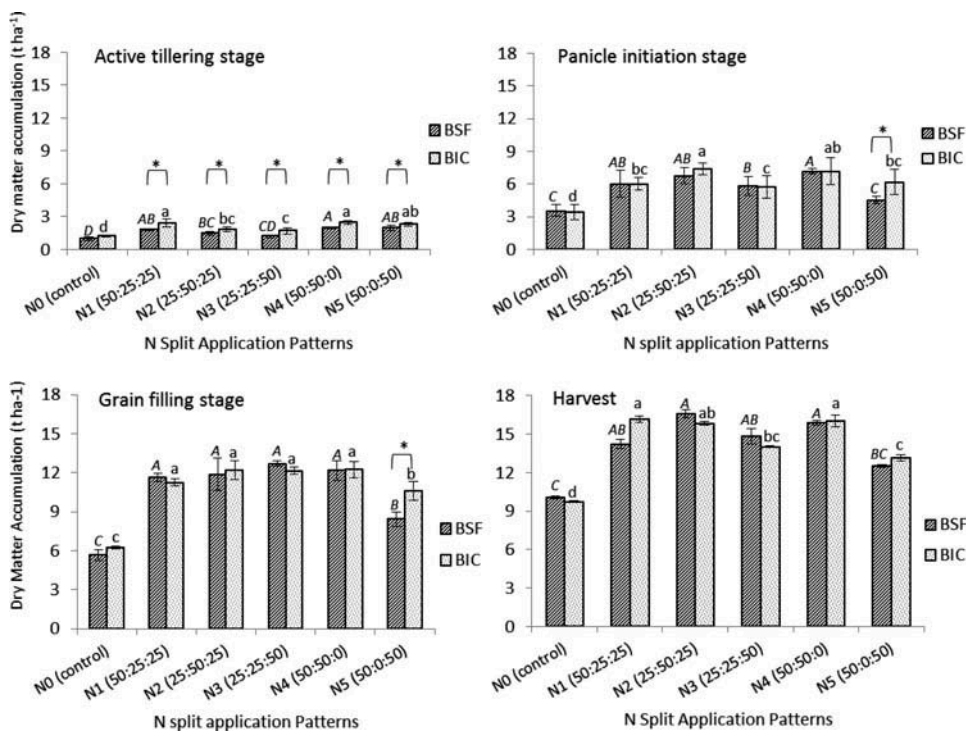


Figure 3. Dry-matter accumulation (t ha^{-1}) of Manawthukha rice affected by N split application patterns at the critical growth stages in basal surface application and incorporation method. The histograms with the same letter in the same case are not significantly different by the Tukey HSD test ($P < 0.05$). The bar on each histogram indicates standard deviation. The numbers in parentheses show the percentage of N applied based on 120 kg N ha^{-1} at basal, active tillering, and panicle initiation stage. Asterisk (*) denotes significant difference at 5% probability.

influenced TDM at each stage. The TDM of each treatment was positively related to the percentage of N application.

At harvest, the greatest TDM (16.59 t ha^{-1}) was obtained in N2 (25:50:25) using the BSF method, and it was similar in N4 (50:50:0). In terms of heavy basal surface N followed by low-level split application, the lowest TDM (14.22 t ha^{-1}) was observed in N1 (50:25:25). Without N application at the active tillering stage, optimum TDM was not achieved in N5 (50:0:50) from either basal application method.

In the BIC method, different TDM patterns were observed. Heavy basal incorporated N followed by split application with lower dose resulted in greatest TDM (16.14 t ha^{-1}) in N1 (50:25:25), with similar results in N4 (50:50:0). With lower amounts of incorporated basal N, N2 (25:50:50) and N3 (25:25:50) obtained the lower TDM scores than the other treatments, at 15.83 and 14.01 t ha^{-1} , respectively.

Harvest Index, Yield, and Yield Parameters

The harvest index (HI) ranged from 0.45 to 0.51 for both basal application methods. Although basal application methods did not influence the harvest index (HI), significant differences in HI were found among N split-application patterns (Table 4).

Table 4

Harvest index, yield, and yield components of Manawthukha rice affected by basal application methods and split applications of nitrogen

Treatment	Harvest index	No. panicle hill ⁻¹	No. spikelets panicle ⁻¹	Filled grain (%)	1,000-grain weight (g)	Maximum panicle length (cm)	Yield (t ha ⁻¹)
Basal surface application method							
N0 (control)	0.45 c	14.6 c	92.47 b	58.59 c	17.74 a	22.20 c	4.54 c
N1 (50:25:25)	0.51 a	17.5 a	106.47 a	63.55 bc	17.32 a	24.03 ab	7.30 a
N2 (25:50:25)	0.48 ab	18.5 a	117.03 a	64.84 bc	17.52 a	24.47 a	7.95 a
N3 (25:25:50)	0.50 a	16.8 a	111.82 a	72.53 abc	17.73 a	23.80 ab	7.48 a
N4 (50:50:0)	0.46 bc	18.0 a	110.63 a	66.02 bc	17.31 a	24.63 a	7.30 a
N5 (50:0:50)	0.51 a	15.7 b	101.42 b	77.36 a	17.44 a	23.13 b	6.41 b
Basal incorporation method							
N0 (control)	0.47 b	13.2 c	94.08 b	58.37 c	17.30 a	23.20 c	4.57 c
N1 (50:25:25)	0.46 c	18.7 a	111.47 a	65.34 b	17.28 a	24.63 ab	7.51 a
N2 (25:50:25)	0.47 b	18.5 a	109.38 a	65.23 b	17.28 a	25.10 a	7.42 a
N3 (25:25:50)	0.50 a	17.3 a	102.83 a	73.50 a	17.52 a	24.13 ab	7.03 a
N4 (50:50:0)	0.47 b	18.2 a	115.69 a	67.77 ab	16.87 a	24.23 ab	7.58 a
N5 (50:0:50)	0.51 a	17.0 b	99.64 b	73.33 a	17.74 a	23.47 c	6.76 b
Source of variance (<i>Pr</i> > <i>F</i>)							
Basal methods	ns	ns	ns	ns	ns	ns	ns
N split	0.0023	<0.0001	0.0014	0.0012	ns	0.0011	<0.0001
Methods × N split	ns	ns	ns	ns	ns	ns	ns
CV (%)	4.60	5.97	6.15	7.60	2.21	3.17	8.08

Panicle number was not affected by basal application methods. Although similar results were observed between the two methods, the panicle number resulting from the BIC method was slightly greater than that from the BSF method (Table 4). Different N split-application patterns significantly influenced panicle number. With the BSF method, greater panicle numbers (18.5 and 18.0 hill⁻¹) occurred in N2 (25:50:25) and N4 (50:50:0), respectively, through the application of high amounts of N at the tillering stage. With application of heavy basal surface N followed by lower split N, the lowest number of effective tillers (17.5 hill⁻¹) was found in N1 (50:25:25). Although a high N percentage was applied at panicle initiation, the panicle number did not increase in N3 (25:25:50). For N5 (50:0:50), omitting N application at the active tillering stage generated a limited number of tillers with panicles. Incorporation of high levels of basal N followed by split application gave the maximum panicle number (18.7 hill⁻¹) in N1 (50:25:25).

Applying N at the active tillering and panicle initiation stages greatly affected the number of spikelets per panicle. With the BSF method, greater numbers of spikelets (117.03 and 111.82 panicle⁻¹) were observed in N2 (25:50:25) and N3 (25:25:50), but there were no significant differences in spikelet number among N split applications other than N5 (50:0:50). By incorporating basal N into the soil, a greater number of spikelet was achieved in N1 (50:25:25) and N4 (50:50:0), with 111.47 and 115.69 spikelets per panicle, respectively. But in the cases of N2 (25:50:25) and N3 (25:25:50), low basal N followed by split application did not result in a greater spikelet number per panicle even though a large amount of N was applied at the active tillering and panicle initiation stages. A lower spikelet number (99.64 panicle⁻¹) was obtained in N5 (50:0:50), due to severe N deficiency at the active tillering stage.

Basal application methods had no significant influence on the percentage of filled grains on the panicle. Among N split-application patterns, significant differences in filled-grain percentage were observed. With the high N panicle fertilization, N3 (25:25:50) and N5 (50:0:50) had the greatest filled-grain percentages with the BSF (72.53 and 77.36%) method and the BIC method (73.50 and 73.33%). The N applied at the panicle initiation stage directly affected grain filling, as opposed to the N applied at the active tillering stage. This result was found in the case of N2 (25:50:25) and N4 (50:50:0), both of which resulted in a reduced filled-grain percentage.

Thousand-grain weight, one of the varietal characteristics, was not influenced by basal application methods and N split-application patterns. However, panicle length was affected by different N split-application patterns. The maximum panicle length (24.47 cm) was obtained in N2 (25:50:25) using the BSF method; however, there were no differences in panicle length among N split applications other than in N5 (50:0:50). With the incorporated basal N, N1 (50:25:25) displayed the maximum panicle length (24.63 cm), and similar results were found in the other treatments except for N5 (50:0:50) and N0 (control).

At the time of harvest, there were no differences in yield between the two basal application methods. However, different N split-application patterns significantly affected yield in both basal application methods (Table 6). Grain yield ranged from 4.54 to 7.95 t ha⁻¹ in the BSF method. The N supply pattern of N2 (25:50:25) was well synchronized with crop N demand throughout the cropping period and resulted in the greatest yield (7.95 t ha⁻¹). The other three-split applications, N1 (50:25:25) and N3 (25:25:50), showed similar yields to the two-split application N4 (50:50:0), which had no N application at the panicle initiation stage. No N was applied at the active tillering stage in N5 (50:0:50), and the optimum rice yield was not achieved. Similar results were found using the BIC method. Generally, the yields for all treatments with BIC were greater than those for treatments with BSF, with the exception of N2 (25:50:25) and N3 (25:25:50), in which low basal N amount

were incorporated into the soil. The greatest yields were obtained in N1 (50:25:25) and N4 (50:50:0), which had a large incorporated basal N supply. N5 (50:0:50) gave the lowest yield, but the yield was still greater than that (4.57 t ha^{-1}) in N0 (control).

Discussion

According to the data analysis, the experimental soil was slightly acidic (pH 6.4) under yearly rice cultivation. With moderate cation-exchange capacity ($13.18 \text{ cmol}_c \text{ kg}^{-1}$), the soil showed a low level of exchangeable calcium (Ca) and magnesium (Mg), a moderate level of K, and a high level of sodium (Na) (Metson 1961). Total N (0.13%) in the soil was initially low according to Enwezor et al. (1989). A low level of available N ($3.05 \text{ mg N}/100 \text{ g soil}$) and a moderate level of available P ($16.79 \text{ mg P}_2\text{O}_5/100 \text{ g soil}$) were observed based on previously report of Federal Ministry of Agriculture and Natural Resources (FPDD) (1990). Therefore, the soil (clay loam) had low fertility in terms of available nutrients.

In rice cultivation, N application synchronized to each critical growth stage is essential for the optimum growth and yield of rice. Most Asian farmers directly broadcast urea fertilizer into the flooded water of rice fields within 2 to 4 weeks after transplanting. The high concentrations of ammonia N in these floodwaters cause high N losses from the surface layer of soil. To overcome this problem, basal application methods were assigned as the main plot practice in this experiment. Until the panicle initiation stage, significant differences were observed between the BSF and BIC methods in terms of plant height (cm), tiller number (hill^{-1}), SPAD values, and dry-matter accumulation. These results showed that the BIC method can reduce ammonium N concentrations in flooded fields and increase N recovery in rice plants. In soil, urea is hydrolyzed by urease into ammonia (NH_3) and carbon dioxide (CO_2), causing a rise in pH and an accumulation of ammonium (NH_4^+). Gaseous losses of NH_3 , up to 50% of the N fertilizer applied, can occur with surface N application (Terman 1979). Incorporating urea fertilizer into the deep soil layer may resolve this problem and enhance nutrient uptake into rice plants. Craswell et al. (1981) reported that the deep application of N fertilizer is an effective means of reducing ammonia concentration in floodwaters and drastically reducing ammonia volatilization (Vlek and Craswell 1981).

Plant height reflects the vegetative growth of rice plants in response to applications of N. The plant height positively responded to the N application rate at each critical growth stage. At harvest, the maximum plant height was recorded at N2 (25:50:25), although no significant difference was found among treatments, except for N5 (50:0:50). The N applied at the active tillering stage enhanced the availability of N for the vegetative growth of rice plants and coincided with the crop demand. Despite heavy N application at panicle initiation, optimum plant height was not obtained in N5 (50:0:50) due to the unbalanced supply of N during the growth stages of the plants. An adequate and balanced supply of N promoted vigorous vegetative growth and deep green color of the rice plants and also influenced the utilization of P, K, and other plant nutrients, resulting in improved growth of the crop (Tahir et al. 2008). Ha and Suh (1993) reported increased plant height from applying N in split applications and lower plant heights when applying N in one dose. In this study, N split-application patterns influenced plant height at each growth stage even though all treatments showed the same statistical results at harvest time, except for N5 (50:0:50). This finding agreed with those of Islam, Hasanuzzaman, and Rokonuzzman (2009), who concluded that split applications of N fertilizer stimulated plant height at 65 and 90 DAT, although the effects were statistically negligible at maturity.

Generally, Manawthukha rice variety has high tillering capacity under favorable conditions. A steady increase in tiller number (hill^{-1}) occurred around 35 DAT and reached a maximum at 52 DAT before the panicle initiation stage in all treatments except N5 (50:0:50), in which no N application occurred at the active tillering stage. The tiller number gradually decreased when N was not applied at the tillering stage. This finding was highlighted by Irshad, Abbas, and Khaliq (2000), who reported that application of all N fertilizer at transplantation or delaying some of it until panicle emergence resulted in a very low number of tillers. The lack of N through the tillering period may result in the lowest number of tillers (Islam, Hasanuzzaman, and Rokonuzzman 2009). Among N split applications, the maximum tiller number was obtained with N2 (25:50:25), which had a large N application rate at the active tillering stage. Nitrogen absorbed during the vegetative period mainly promotes the early growth of plants and increases the number of tillers (Makino, Mae, and Ohira 1984). The optimum tillering pattern was observed in N4 (50:50:0), with two split applications at the basal and active tillering stage, followed by N2 (25:50:25). Shoo, Mishra, and Mohanty (1989) reported that N application at transplantation or in two equal split dressings at transplantation and tillering increased the total number of tillers per hill. Before the panicle initiation stage, significant differences in tiller number (per hill) were found between the two basal application methods. This result is thought to be associated with the N uptake of rice plants in each treatment. The BIC method acted to reduce N losses and enhance N recovery. In general, the dose of N fertilization at the tillering stage was essential for the optimum number of tillers in rice because N has a positive influence on the production of effective tillers per plant as well as on yield and yield attributes (BRRI 1990). The number of tillers per unit area is the most important measure of yield. As the number of tillers, particularly fertile tillers, is greater, the greater the rice yield will be. The number of tillers per square meter, which is dependent on greater availability of N, plays a vital role in cell division (Chaturvedi 2005). Therefore, the omission of N application at the active tillering stage reduces the optimum tiller number and subsequent rice yield.

In rice cultivation, the SPAD value showing the greenness of leaves is the most important plant growth characteristic. In the beginning of rice growth, the SPAD values of plants grown using the BIC method gradually increased in all treatments until 24 DAT. However, with the basal surface application SPAD values stalled around 35 DAT, and after that SPAD temporarily decreased, particularly in the treatments that applied a low basal N application. However, the incorporated basal N achieved greater SPAD values, compared with the basal surface applications, until the panicle initiation stage. This result reveals that incorporating basal N was effective in reducing N losses and in improving N uptake in rice plants. The urea N in the basal surface application may result in ammonia volatilization or surface runoff, resulting in lower N uptake. The deep placement of N might delay the rapid conversion of urea to ammonium and create a slow-release action to match the release of N to the difference between the N requirements of the plants and the N available from the soil. Craswell et al. (1981) stated that the deep placement of urea can prevent the rapid conversion of ammonium to nitrate and provide an effective means to reduce ammonia concentrations in floodwater. Basal surface N application methods resulted in decreased SPAD values, particularly in the treatments that entailing broadcasting large amounts of basal N. The results reported by Craswell et al. (1981) suggested that N losses through ammonia volatilization and surface runoff occurred on the soil surface, resulting in reduced N uptake by rice plants. Furthermore, broadcasting urea into floodwater resulted in an average 30% recovery of fertilizer N by the rice crop in both the dry and wet season.

In rice, an adequate amount of N is required for the growth of young panicles. The N absorbed at this time is efficiently used to increase spikelet number and panicle size

(Yoshida 1981). Throughout the crop period, a stable SPAD value of approximately 35 was maintained by both N2 (25:50:25) and N4 (50:50:0). These results reveal that the rice plant's requirements were adequately satisfied by the supply of N from the split-application pattern. In both treatments, a large percentage of N was top-dressed at the tillering stage. In N5 (50:0:50), the SPAD values gradually decreased below 35 at the active tillering and panicle initiation stages when N top dressing was not used at tillering. Peng et al. (1996) suggested the use of 35 as a critical SPAD value for an IRRI (International Rice Research Institute) high-yielding cultivar, IR72, in the dry season and recommended a top-dressing of 30 kg N ha⁻¹ whenever the SPAD value fell below 35. As an Indica variety, Manawthukha rice would be included in this critical value, although further studies are necessary to determine the optimum SPAD value for this variety. At the grain filling stage, SPAD values increased in all treatments. The N in the older leaves from the lower parts of the plant translocated to the upper part, the flag leaf, of the rice plant. This result may be a varietal characteristic of Manawthukha rice. Turner and Jund (1994) found that the SPAD values of low-N-applied rice increased (by approximately 35) at heading using different rates of N application. They explained that the senescence of the lower leaves of the plant with lower N application translocated N to the flag leaf, resulting in a greener flag leaf. Myint et al. (2011) also indicated that the SPAD value increased in the flag leaves of Manawthukha, which can also be assumed to be varietal character. During the crop's developmental stages, the SPAD values of N3 (25:25:50) and N5 (50:0:50) fell below 35. This result suggests that the N uptake of these two treatments may be lower than the crop demand due to the nonsynchronized application of N during each critical growth stage. However, knowledge of the optimum SPAD value for Manawthukha rice is essential for further research because the SPAD value may vary among plant types (semi-dwarf, tall local, hybrid varieties, etc.), systems of cultivation (transplanted vs. direct seeded), and environmental conditions (temperate, tropical, sunlight intensity, moisture regime, etc.) (WIN 2003).

Dry-matter accumulation, an important varietal character, was greatly influenced by different N split-application patterns in this study. Islam, Hasanuzzaman, and Rokonuzzaman (2009) showed that the effect of the split application of N fertilizer was statistically significant in terms of the dry matter of roots, stems, and leaves as well as total dry matter at the three growth stages. Prior to the panicle initiation stage, the BIC method resulted in greater TDM than did the BSF method. These results revealed that greater N recovery provides greater TDM with the incorporated basal N. The incorporation of N at sowing ensures a balanced supply of N for leaf growth, increased pre-anthesis crop growth, and greater dry-matter accumulation (Zhang et al. 2009). Generally, dry-matter accumulation is positively correlated with the total N uptake at each critical growth stage of rice plants. Chaturvedi (2005) reported that dry-matter accumulation increased significantly with N-fertilizer application in rice at all growth stages of the crop. According to the results of this study, N top-dressing at the active tillering stage was important for acquiring optimum TDM in rice plants, as shown by TDM values in N2 (25:50:25) and N4 (50:50:0). Generally, most of the N in the grain comes from remobilized and translocated N from the rice stems and leaves. Consequently, the uptake of fertilizer N early in the growing season affects the uptake of the native soil N later in the season, the dry-matter production of the rice plants, the harvest index or sink-source relationships of the rice plant, and ultimately the rice grain yield (Yusob et al. 2007). Therefore, adequate N application at the vegetative stage is crucial for the improved growth and crop performance.

The grain yield of rice was highly affected by different N split-application patterns. Although there were no significant differences in yield between the two basal application

methods, the results were varied in both methods due to differences in N-application patterns during plant growth. With low surface-applied basal N, the maximum yield was obtained in N2 (25:50:25), in which a large amount of N was top-dressed at tillering. The results indicated that with surface applications, a lower amount of basal N with a greater number of split applications may encourage optimum rice yield in clay loam soil. The N uptake in the vegetative stage was high and supported the spikelet differentiation of panicles. Therefore, the key period for nitrogen absorption by rice plants is from tillering to flowering. During this period the absorption of soil N is at its maximum rate. Most of the absorbed N is stored in leaves and may be transported to the grains during grain filling (Duan et al. 2005). Consequently, there will be a greater number of panicles and of spikelets per panicle, resulting in a greater yield. Murty, Dey, and Jachuk (1992) reported that N application has great impact on crop yield in rice when applied during the early and midtillering stages in producing a high number of panicles, optimum spikelets per panicle, and a high percentage of filled spikelets. Heavy basal-surface N application before transplanting is a serious problem in rice and is subject to N losses through ammonia volatilization and surface runoff.

Ammonia volatilization from rice field is an inevitable source of N loss that is closely related to the N application level during and after fertilizer application (Ye et al. 2011). However, incorporated basal N followed by split application provided greater yields in N1 (50:25:25) and N4 (50:50:0). Incorporation of basal N led to greater N recovery in rice and increased N available to rice on demand at the critical stages. The deep placement of urea reduced N loss by ammonia volatilization, which is an important consideration, as ammonia is known to be the main cause of urea-induced toxicities (Haden et al. 2011). Furthermore, the split application of N was effective for rice yield, except in N5 (50:0:50). Split application is one of the strategies for the efficient use of N fertilizer throughout the growing season. By synchronizing N application with plant demand, denitrification losses were reduced and N uptake improved, resulting in maximum straw and grain yield and an increased harvest index (Lampayan et al. 2010). Although high doses of N top-dressing were applied in a two-split pattern in N5 (50:0:50), the optimum yield did not result because the N application at tillering was omitted. Zhang et al. (2009) suggested that even high and nonsynchronously applied N may limit grain yield due to a limited grain filling rate resulting from decreased postanthesis assimilate translocation. The yields in this experiment were greater than the farmers' yield of Manawthukha rice in Myanmar except for N0 (control). In Myanmar, the urea fertilizer rate (50 kg ha⁻¹) was common in ordinary farmers' rice fields (Myint et al. 2011) and resulted in an average comparable to the Manawthukha yield (4 to 5 t ha⁻¹) (Tun et al. 2003).

Generally, N application before transplanting caused N losses such as surface runoff and ammonia volatilization. Additionally, the high concentration of ammonia can damage rice seedlings. In this study, the limited application of surface basal N before transplanting followed by a synchronized N split application maximized dry-matter accumulation and yield. The surface-applied N was covered by shallow water just after application, and tight water management was necessary to avoid losing N via surface runoff. Without water on the soil surface, the urea crystals were hydrolyzed and formed unstable carbonic acid, which rapidly decomposed into ammonia, which was released to the air as a gas. When covered by water or soil, urea reacts with water, changes to ammonium ions, and makes N available to rice seedlings. However, heavy basal surface N appeared to result in N loss. The heavy dose of incorporated basal N applied before transplanting minimized N losses, caused N to be released slowly, and made the nutrients available to rice plants throughout

the crop period. Therefore, the high basal incorporated N with split applications may prove beneficial for the optimum growth and yield of rice.

Nowadays, it is essential to produce a greater yield of rice with less impact on the environment to fulfil the demands of consumers, which are expected to increase in the future. A very high and unsynchronized application of urea was found to cause high N losses, which causes non-point-source pollution and other environmental pollution. Therefore, the split application of fertilizer N will remain an essential component of fertilizer management strategies, not only for improving N-use efficiency and rice yield but also for sustainable agriculture and the safety of the environment. However, it is necessary to an optimum split pattern of N fertilizer application patterned in line with the critical growth stages for specific varieties in various growing regions. Furthermore, the determination of specific application rates for a wide range of soil types is required as part of ongoing research on various application methods.

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