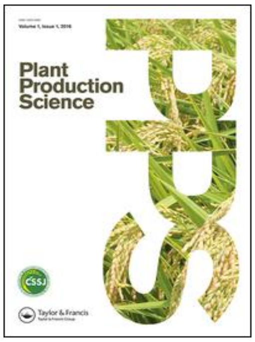

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Grain yield responses of lowland rice varieties to increased amount of nitrogen fertilizer under tropical highland conditions in central Kenya

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ABSTRACT
Tropical highland conditions in Mwea Kenya, ensure the high radiation and the large day–night temperature differences. Such conditions are generally believed to promote rice growth and yield, but the current grain yield is lower than the expectation. In the current standard N fertilizer practice in Mwea, 75 kg nitrogen (N) ha^{-1} is applied in three splits at fixed timing. The effects of increases in N fertilizer amount (125, 175, and 225 kg N ha^{-1}) on rice growth and yield were evaluated to test the hypothesis that unachieved high rice grain yield in Mwea is due to insufficient amount of N fertilizer. Two popular lowland varieties in Mwea (Basmati 370 and BW196) and two varieties reported as high yielding at other countries (Takanari and IR72) were used. Shoot dry weight (DW) increased with increases in the amount of N fertilizer applied in three splits at fixed timing, irrespective of variety. It reached approximately 20 t ha^{-1} under increased N conditions (>75 kg N ha^{-1}) in several cases, indicating that high biomass production could be achieved by increasing N application rate. However, the increased biomass did not increase grain yield, due to decreased grain filling under high N conditions in all varieties. Thus, N amounts above 75 kg ha^{-1} were ineffective for increasing grain yields in Mwea, where N fertilizer was applied in three splits at fixed timing. Increasing influence of low temperature under high N conditions may be one of the reasons for the decreased grain filling in Mwea.

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Excess N absorption; Kenya; low temperature influence; N application timing and dosage; N fertilizer amount; poor grain filling; rice

1. Introduction

The demand for rice in Kenya is increasing due to changes in eating habits and increasing urban population (Mati et al., 2011). In 2014, the area used for rice production in Kenya was 28,390 ha and the resulting production was 112,263 t (FAO, 2016a). However, this production level was below the expanding demand in the country. In 2013, Kenya imported 412,411 t of rice, costing the country 165 million USD (FAO, 2016b). To increase rice production, both planting area and productivity (grain yield) per unit area need to be improved. The Mwea Irrigation Scheme, which is located 100 km northeast of Nairobi at the southern foot of Mt. Kenya, is the largest rice-growing irrigation scheme in Kenya, covering 9000 ha and producing 80–88% of rice in the country (Mati et al., 2011; Onyango, 2014). Hence, increasing grain yield in Mwea would be one of the best ways to boost rice production in Kenya.

The location of Mwea, near to the equator and approximately 1200 m above sea level, ensures high radiation typical of a tropical region and large day–night temperature differences typical of a highland. The fact that high radiation and large differences in day–night temperatures promote rice growth and grain yield is widely accepted (Katsura et al., 2008). The weather conditions in Mwea may be favorable to achieve high rice grain yield. However, the advantage of weather conditions in Mwea has not been utilized to achieve high rice grain yield. In a study conducted among 325 farmers in Mwea, some achieved decent grain yield (>7.5 t ha^{-1}), but most of them obtained much lower yields ranging from 2.5 to 6.5 t ha^{-1} (Muhunyu, 2012). Furthermore, the average reported grain yield for two local popular lowland rice varieties in this region, Basmati 370 and BW196, is only approximately 5 t ha^{-1} (Kihoro et al., 2013; Kondo et al., 2001).

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The question is whether rice grain yield could be increased with increases in the amount of nitrogen (N) fertilizer or the current N fertilizer practice, which were determined based on the experiences of farmers and researchers, is sufficient for optimal rice production in this area. The amount of N fertilizer applied in Mwea was reported as 46 kg N ha^{-1} with unknown amount of animal manure (Kihoro et al., 2013), 75 kg N ha^{-1} (Muhunyu, 2012; Niki et al., 2014), and 110 kg N ha^{-1} (Ndiiri et al., 2012) which is considerably lower than those applied in high-yielding rice cultivation areas in other countries (Table 1). For example, in Los Baños, Philippines, the grain yield of a lowland rice variety IR72 was 9.7 t ha^{-1} with an application of 180 kg N ha^{-1} (Bueno et al., 2010). Rice N requirements are closely related to yield levels, which in turn are sensitive to weather conditions, supply of other nutrients, and crop management practices (Peng et al., 1996). Singh et al. (1998) reported that grain yield of several lowland rice varieties did not respond to increases in the amounts of N fertilizer and discussed that the grain yield of these varieties was likely influenced by factors other than N supply. Thus, it is worthwhile to validate the current N fertilizer practices and examine the possibility of yield increases through increasing the amount of N fertilizer.

In the present study, we hypothesized that unachieved high rice grain yield in Mwea was due to insufficient amount of N fertilizer. To test this hypothesis, field experiments were performed in irrigated lowland fields within an agricultural research farm in Mwea. The growth and grain yield of two local popular lowland rice varieties, Basmati 370 and BW196, and high-yielding varieties selected in other locations, Takanari and IR72, were examined under varying amounts of N fertilizer application. The effects of different N fertilizer practices on growth and grain yield were evaluated to assess the appropriateness of the current N fertilizer practice with the aim to improve rice grain yield in Mwea.

2. Materials and methods

Three field experiments were conducted at the Kenya Agricultural and Livestock Research Organization (KALRO) in Mwea, Kenya (0°39′ S lat., 37°22′ E long.), at an elevation of 1162 m above sea level, from 2013 to 2015. The soil of

Table 1. Yield and shoot dry weight of high yielding varieties, Takanari and IR72, under different growth conditions.

Variety	Location	Year	N fertilizer application (kg N ha^{-1})	Radiation (MJ m^{-2} d^{-1})	Temperature (°C)			Grain yield (t ha^{-1})	Shoot dry weight (t ha^{-1})
					Maximum	Mean	Minimum		
Takanari	Kyoto, Japan	2002^{a}	>120‡	12.6–16.3	27.1–31.4	22.6–27.4	18.3–23.7	10.6	15.4
		2003^{a}	0–230	11.5–12.0	26.0–29.7	21.8–25.3	18.2–21.7	7.3–9.8	11.1–14.9
		2003^{b}	140–280	8.0–17.0†	–	20.5–27.0†	–	9.6–9.8	14.8–14.9
		2004^{b}	140	10.0–17.5†	–	19.5–28.0†	–	11.4	15.7
	Osaka, Japan	2007^{c}	180	13.5–20.9	–	18.6–28.8	–	10.8–11.4	–
		2008^{c}	180	12.7–21.2	–	18.3–28.1	–	11.3–11.5	–
		2009^{d}	130	13.0–20.0	–	17.7–27.3	–	9.3–9.9	–
		2010^{d}	120	12.2–20.7	–	18.1–29.8	–	8.4–9.2	–
	Tokyo, Japan	2007^{c}	180	9.8–19.6	–	18.5–29.5	–	6.6–9.7	–
		2008^{c}	180	10.6–17.7	–	18.8–27.7	–	10.6–11.0	–
	Tsukubamirai, Japan	2009^{e}	90–180	14.8–18.5	–	20.6–25.3	–	11.6–12.8	18.5–20.8
		2010^{e}	90–180	16.2–19.8	–	20.9–28.2	–	11.5–12.1	18.3–21.1
	Yunnan, China	2002^{a}	>120‡	15.5–18.7	29.0–30.1	23.2–24.3	18.6–21.4	12.1	20.4
		2003^{a}	>0–230‡	16.3–19.1	29.7–31.4	25.2–25.4	18.7–21.5	10.6–15.1	15.5–22.0
IR72	Kyoto, Japan	2002^{a}	>120‡	12.6–16.3	27.1–31.4	22.6–27.4	18.3–23.7	9.8	17.0
	Los Baños, Philippines	2003^{f}	200	14.0–25.7†	–	24.7–29.4†	–	9.3	17.3
		2007^{g}	180	14.4–20.4	30.7–32.5	–	22.5–23.5	9.7	21.1
		2007^{h}	180	17.0	30.4	–	22.9	9.7	21.1
	Nueva Ecija, Philippines	2004^{h}	200	25.5	32.9	–	22.6	9.1	16.1
	Yunnan, China	1995^{i}	>200‡	15.0–22.5	29.5–37.5†	–	17.5–22.0†	9.8	24.0
		1996^{i}	>200‡	15.0–21.0	20.0–34.5†	–	18.0–23.5†	12.9	24.1
		2002^{a}	>120‡	15.5–18.7	29.0–30.1	23.2–24.3	18.6–21.4	11.4	19.4

[a] Katsura et al. (2008).
[b] Katsura et al. (2007).
[c] Kato et al. (2009).
[d] Katsura and Nakaide (2011).
[e] Yoshinaga et al. (2013).
[f] Yang et al. (2007).
[g] Bueno and Lafarge (2009).
[h] Bueno et al. (2010).
[i] Ying et al. (1998).
[†] Data were obtained from figures.
[‡] Manure was also applied.

the experimental field was Nitisol containing 2.39% total carbon (C) determined by wet oxidation chromic acid digestion method, 0.19% total N determined by micro-kjeldahl method, 618 mg available phosphorous pentoxide (P_2O_5 kg^{-1}; Bray II), and 0.20 cmol(+) kg^{-1} exchangeable potassium (K) determined after extraction with 1 M ammonium acetate (pH 7.0) using flame photometer, at the start of Experiment (Exp.) 1. Soil H_2O pH was 5.44. Temperature and radiation were recorded by a weather station (Weather Bucket; Agriweather Inc., Sapporo, Japan) located at the research farm.

A summary of the experiments conducted in the present study is shown in Table 2. The four lowland varieties used in this study were Basmati 370, BW196, Takanari, and IR72. The aromatic variety Basmati 370 and the non-aromatic high-yielding variety BW196 are popular among Mwea farmers. Takanari is a high-yielding variety that has been evaluated in Yunnan, China (Katsura et al., 2008) and Kyoto, Japan (Katsura et al., 2007 and 2008), and IR72 is a common high-yielding check variety grown in the tropics (Peng et al., 2006). In the preliminary experiments, Basmati 370 was highly susceptible to local rice-blast populations while BW196 was tolerant. The response of Takanari and IR72 to these blast populations are unknown. All varieties were sensitive to cold stress in the preliminary experiments conducted in Mwea during the cold season. In Exp. 1, Basmati 370, BW196, and Takanari were used, but in Exp. 2 and 3, Takanari was replaced with IR72, due to the former's unsuitability to growth conditions in Mwea.

The standard basal fertilization for rice cultivation in Mwea is 25 kg N, phosphorous (P), and K ha^{-1}, and was applied in the form of NPK (17:17:17) compound fertilizer. Additional ammonium sulfate was top-dressed at 25 kg N ha^{-1} at 21 and 45 days after transplanting (DAT), which is the standard practice (hereafter referred to as 75 N-3splits). The basal application was the same for all treatments. In all experiments, three N treatments, namely, N topdressing of 50, 75, or 100 kg N ha^{-1} applied twice at 21 and 45 DAT in the form of ammonium sulfate, were included in addition to the 75 N-3splits. The total amount of applied N in these three treatments was 125, 175, and 225 kg N ha^{-1}, respectively, and these are hereafter referred to as 125 N-, 175 N-, and 225 N-3splits, respectively.

All experiments had a 3 × 4 (three varieties and four N treatments) factorial design and each treatment included three replicated plots arranged in a randomized complete block design. The plot sizes were 2 m × 2 m. In all experiments, 21-day-old seedlings were transplanted at a spacing of 20 cm × 20 cm with one seedling per hill. In Exp. 1, 2, and 3, the transplanting was done on 4 October 2013, 17 March 2014, and 17 February 2015, respectively. Generally, rice transplanting in Mwea is carried out from July to August with a small transplanting peak in November (Kihoro et al., 2013). The timing of transplanting is determined by the irrigation distribution schedule within the Scheme. Preliminary experiments confirmed that rice cultivation is possible in Mwea if irrigation water is available, regardless of transplanting dates (with exceptions of April, May, and June when the cold damage often markedly decreases grain yield). All experiments were conducted under continuously flooded conditions using irrigation water allocated to the research farm, and hand weeding was performed as required. Oshothion 50EC and Diazol 60EC (Osho Chemical Industries Ltd, Nairobi, Kenya) were used for insect control when insects were noticed. Rodazim 50SC (Cooper K-Brand Ltd, Nairobi, Kenya) was applied when weather conditions were favorable for rice blast infection.

In all experiments, the number of tillers, plant length from ground to the tip of the uppermost leaf, and soil-plant analysis development (SPAD) value for chlorophyll concentration (SPAD-502 plus; Konica Minolta Inc. Tokyo, Japan) were periodically measured. At maturity, shoot parts were harvested at ground level from more than 24 hills, other than border hills. The harvested shoots were placed into paper bags and sun-dried for at least a month. The panicles were then removed from the stems, and hand threshed. Filled grains were separated from unfilled spikelets by submerging in tap water. Sunken and floating spikelets were considered as filled grain and unfilled spikelets, respectively. Using the panicles and spikelets, grain yield (filled grain weight at 14% moisture content) and yield components were determined. The straws, panicle rachis, and unfilled spikelets were oven dried at 70 °C for one day and weighed. Shoot dry weight (DW) was determined as the sum of grain yield and DW of straws, panicle rachis, and

Table 2. Summary of the experiments (Exp.) conducted in the present study.

Exp.	Variety used	Date of TP[a]	Harvest (DAT)	Application of N fertilizer
1	Basmati 370 BW196 Takanari	Oct. 4, 2013	130 for Basmati 370 145 for BW196 129 for Takanari	Three splits[b] of 75 kg N ha^{-1} (75N-3splits[c]) Three splits[b] of 125 kg N ha^{-1} (125N-3splits) Three splits[b] of 175 kg N ha^{-1} (175N-3splits) Three splits[b] of 225 kg N ha^{-1} (225N-3splits)
2	Basmati 370 BW196 IR72	Mar. 17, 2014	149–151 for three varieties	
3	Basmati 370 BW196 IR72	Feb. 17, 2015	129 for Basmati 370 133 for BW196 127 for IR72	

[a] Transplanting.
[b] Three nitrogen (N) splits application was performed at transplanting as well as at 21 and 45 days after transplanting (DAT).
[c] 75 N-3splits is the standard N fertilizer practice in Mwea.
TP: transplanting

unfilled spikelets. The harvest index (HI) was calculated as the grain yield divided by the shoot DW.

The statistical package R (Version 3.0.2, R Core Team, 2015) was used for statistical analyses. Three-way analysis of variance (ANOVA) was performed to identify significant effects of variety (V), N treatment (T), experiment (E), and their interactions on grain yield and yield-related parameters of Basmati 370 and BW196, which were consistently used in Exp. 1–3. In Exp. 1 and 3, two-way ANOVAs were performed to identify the significant effects of V, T, and their interaction on plant length and tillers per m^2. In Exp. 2, plant length and tillers per m^2 were measured in one replication only. The average values were separated using Tukey's honest significant difference (HSD) test. The SPAD values obtained in all experiments were compared using a one-way ANOVA among N treatments for each variety on each sampling day.

3. Results

3.1. Weather conditions during three experiments

Daily maximum, minimum, and mean temperatures ranged from 25.0 to 32.5 °C, 13.5 to 18.9 °C, and 20.1 to 23.2 °C, respectively, whereas daily average radiation ranged from 9.2 to 20.5 MJ m^{-2} d^{-1} throughout the three experiments (Figure 1). The average daily maximum temperatures during the growth periods were 29.4, 26.9, and 27.7 °C for Exp. 1, 2, and 3, respectively (Figure 1(A)). For Exp. 1, 2, and 3, the corresponding values for daily minimum and mean temperatures were 16.2, 17.2, and 16.9 °C and 21.9, 21.3, and 21.9 °C, respectively (Figure 1(B) and (C)). Average radiation during the growth periods of Exp. 1, 2, and 3 were 17.0, 13.6, and 15.7 MJ m^{-2} d^{-1}, respectively (Figure 1(D)). Variations in weather factors differed among the three experiments. The daily maximum temperature during the early growth stage in Exp. 3 was around 30 °C, which was higher than those in Exp. 1 and 2, and that in Exp. 1 during the middle and late growth stages was around or higher than 30 °C, which was higher than those in the other two experiments (Figure 1(A)). The daily minimum temperatures ranged from 13 to 16 °C in Exp. 3 and 1 during the early and middle growth stages, respectively (Figure 1(B)). The daily mean temperature during the early growth stage was around 23 °C in Exp. 3, the highest among the three experiments. During the middle growth stage, those in Exp. 2 and 3 started to decline and reached around 20 °C and then those were lower than that in Exp 1 during the late growth stage (Figure 1(C)). Radiation was relatively high in Exp. 3 during the early growth stage and relatively low in Exp. 2 and 3 during the middle to late growth stages (Figure 1(D)). The low temperature and radiation periods were prolonged in Exp. 2 than in Exp. 3 (Figure 1(A), (C) and (D)).

3.2. Growth and grain yield of rice varieties

Shoot DW tended to increase with increases in the amount of N fertilizer (Figure 2(A)–(C)). Shoot DW reached approximately 2000 g m^{-2} (equivalent to 20 t ha^{-1}) or more in BW196 under 225 N-3splits in Exp. 1 and in Basmati 370, BW196, and IR72 under 125 N-, 175 N-, and 225 N-3splits in Exp. 3 with the largest value of 2461 g m^{-2} (equivalent to 24.6 t ha^{-1}) in BW196 under 225 N-3splits in Exp. 3. In contrast, grain yield was less than 535, 783, 328, and 480 g m^{-2} (equivalent to 5.4, 7.8, 3.3, and 4.8 t ha^{-1}, respectively) in Basmati 370, BW196, Takanari, and IR72, respectively, throughout the three experiments. In Exp. 1 and 2, no significant differences in grain yield among N treatments were obtained ($p > 0.05$) (Figure 2(D) and (E)). In Exp. 3, the grain yield under the standard 75 N-3splits was similar to or significantly higher ($p < 0.05$) than that under 125 N-, 175 N-, and 225 N-3splits (Figure 2(F)). The grain yield of Takanari or IR72 did not exceed that of Basmati 370 or BW196 within each experiment, irrespective of N treatment (Figure 2(D)–(F)).

Three-way ANOVA was performed on grain yield and yield-related parameters of Basmati 370 and BW196 (Table 3). The effect of V was significant ($p < 0.001$) on all parameters except for grain filling and HI, while T had no significant effect ($p > 0.05$) only on grain yield and spikelets per panicle, and E significantly affected ($p < 0.001$) all parameters except for spikelets per panicle. The V × T and V × T × E interactions had no significant effect ($p > 0.05$) on all parameters, but either V × E or T × E interactions significantly affected ($p < 0.001$, 0.01, or 0.05) all parameters except 1000-grain weight and shoot DW. Thus, the effects of V and T on the various parameters were different depending on experiment.

Grain yield and yield-related parameters for each N treatment across Basmati 370 and BW196 are shown in Table 4. Grain yield, spikelets per panicle, and 1000-grain weight under the standard 75 N-3splits were not significantly different ($p > 0.05$) from those under the other N treatments, irrespective of experiment. Panicles and spikelets per m^2 and shoot DW increased with increases in the amount of N fertilizer in all experiments. In contrast, grain filling and HI decreased with increases in the amount of N fertilizer, irrespective of experiment.

In Exp. 1 and 3, the effects of V and T on plant length and tillers per m^2 were significant ($p < 0.001$), whereas under the standard 75 N-3splits these parameters tended to be inferior to the corresponding values under the other N treatments (Table 5). Similar tendencies were observed in Exp. 2.

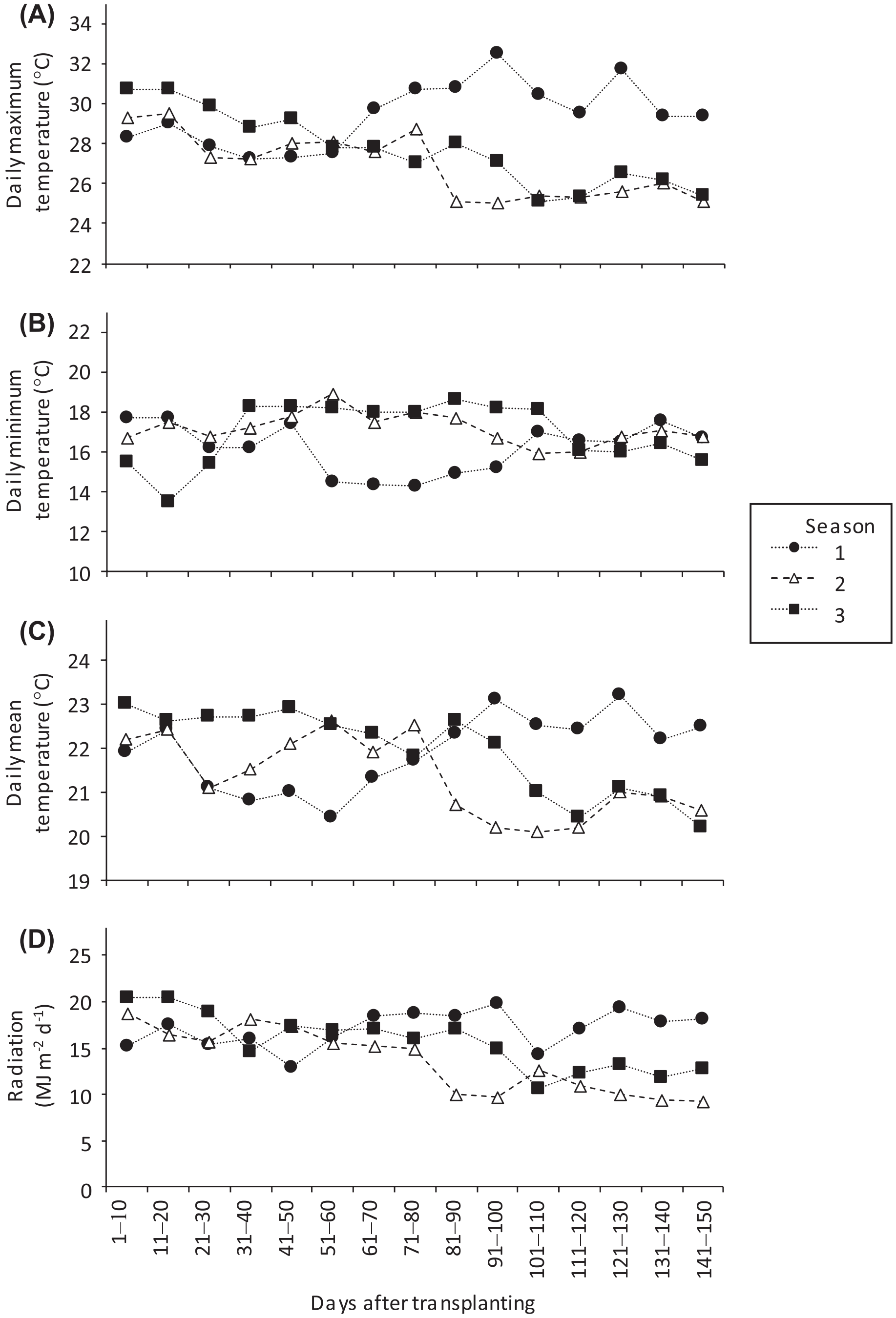

Figure 1. Changes in daily maximum (A), minimum (B), and mean (C) temperatures and daily average radiation (D) during the growth period in three experiments. Each point represents the mean of 10 days.

In Exp. 1 and 3, SPAD values tended to increase with increases in the amount of N fertilizer and ANOVA results indicated that the effect of T on SPAD values was significant on several sampling days (Figure 3). Basmati 370 and BW196 maintained high SPAD values until the late growth stage under increased amounts of N fertilizer. Similar tendencies were observed in Exp. 2.

4. Discussion

Our hypothesis that unachieved high rice grain yield in Mwea is due to insufficient amount of N fertilizer was not verified in the present study. The effects of N fertilizer application on biomass production and grain yield under the weather conditions in Mwea are discussed in the following two subsections. Moreover, in the third subsection, we deduce possible reasons for the poor responses

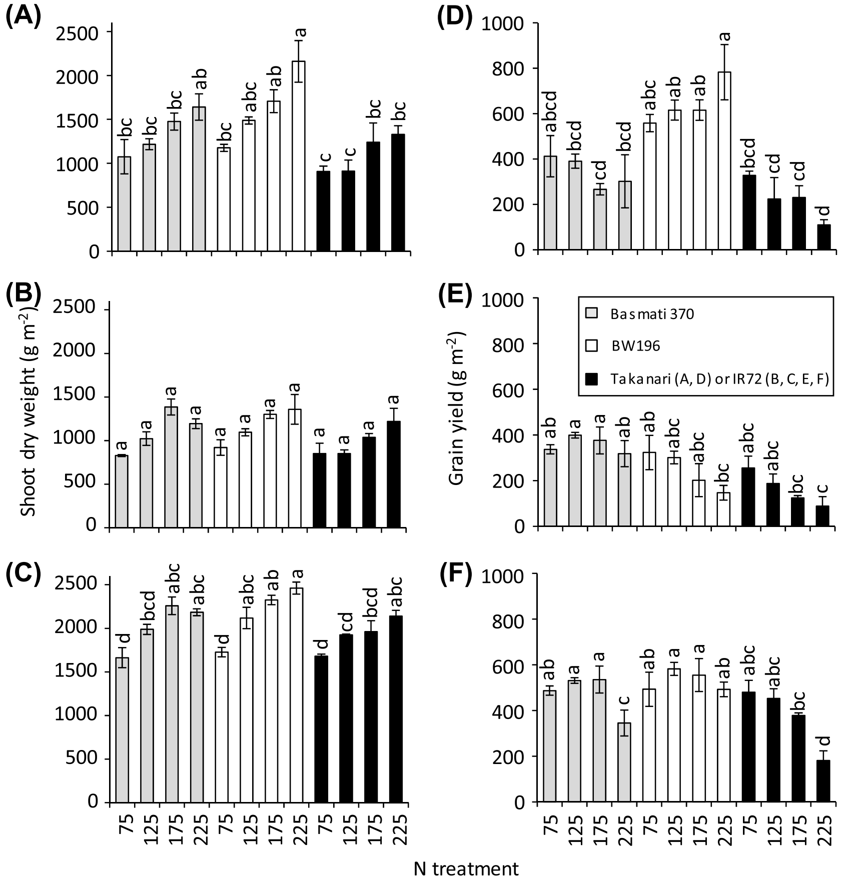

Figure 2. Shoot dry weight (A, B, C) and grain yield (D, E, F) under 75 N-, 125 N-, 175 N-, and 225 N-3splits (75, 125, 175, and 225, respectively) in Experiments 1 (A, D), 2 (B, E), and 3 (C, F). Bars with different lower case letters were significantly different according to Tukey's HSD test ($p < 0.05$).

Table 3. Three-way analyses of variance across Experiments 1–3 to determine the effects of variety, N treatment, and experiment on grain yield and yield-related parameters of Basmati 370 and BW196.

	df	Mean square	df	Mean square	df	Mean square	df	Mean square
	Grain yield (g m^{-2})		Panicles (m^{-2})		Spikelets ($panicle^{-1}$)		Spikelets (m^{-2})	
Variety (V)	1	116,818***	1	188,908***	1	6229***	1	238,918,314***
N treatment (T)	3	16,095	3	111,514***	3	73	3	264,600,267***
Experiment (E)	2	313,935***	2	266,651***	2	15	2	646,921,959***
V × T	3	11,244	3	2396	3	182	3	17,102,397
V × E	2	259,180***	2	43,597***	2	546**	2	26,613,129
T × E	6	17,535	6	15,695**	6	70	6	34,479,143*
V × T × E	6	17,332	6	5863	6	44	6	21,079,893
Residuals	47[a]	8445	48	4670	46[b]	75	46[b]	12,251,657
	1000-grain weight (g)		Grain filling (%)		Shoot dry weight (g m^{-2})		Harvest index	
Variety (V)	1	885***	1	15	1	382,259***	1	0.00954
N treatment (T)	3	11.2*	3	3452***	3	1,261,839***	3	0.08199***
Experiment (E)	2	96.4***	2	1700***	2	5,553,161***	2	0.03847***
V × T	3	4.4	3	225	3	58,714	3	0.00107
V × E	2	2.2	2	3677***	2	74,726	2	0.09089***
T × E	6	3.4	6	155	6	60,304	6	0.0047
V × T × E	6	1.6	6	194	6	8717	6	0.00289
Residuals	48	3.7	46[b]	107	47[a]	30,275	47[a]	0.00278

[a] One observation was deleted due to missing data.
[b] Two observations were deleted due to missing data.
***Indicates significance at 0.001 probability level;
**Indicates significance at 0.01 probability level;
*Indicates significance at 0.05 probability level.

Table 4. Grain yield and yield-related parameters for each N treatment (75 N-, 125 N-, 175 N-, or 225 N-3splits) across Basmati 370 and BW196 in experiments (Exp.) 1–3 and their average values across the three experiments.

N treatment	Exp. 1	Exp. 2	Exp. 3	Average	Exp. 1	Exp. 2	Exp. 3	Average	Exp. 1	Exp. 2	Exp. 3	Average	Exp. 1	Exp. 2	Exp. 3	Average
	Grain yield (g m^{-2})				Panicles (m^{-2})				Spikelets ($panicle^{-1}$)				Spikelets (m^{-2})			
75 N-3splits	500a	330a	490ab	440A	500b	330b	490a	440C	58.6a	50.0a	51.7a	53.4A	24576b	16452a	23765b	21597C
125 N-3splits	504a	350a	557a	470A	551b	357ab	524a	478B	51.9a	56.5a	54.9a	54.4A	28130b	20125a	27262ab	25172B
175 N-3splits	442a	109a	545a	365A	560b	370ab	550a	493B	53.5a	61.8a	59.7a	58.3A	29829b	21835a	31065a	27576B
225 N-3splits	542a	232a	419b	398A	750a	438a	602a	596A	54.0a	54.2a	53.9a	54.0A	39375a	23390a	31086a	31284A
	1000-grain weight (g)				Grain filling (%)				Shoot dry weight (g m^{-2})				Harvest index			
75 N-3splits	22.9a	26.3a	24.6a	24.6A	87.0a	79.0a	84.9a	83.7A	1135c	875c	1695c	1235C	0.44a	0.36a	0.30a	0.37A
125 N-3splits	22.2a	25.9a	24.9a	24.3AB	80.0a	69.4ab	83.1a	77.5A	1352bc	1060bc	2054b	1488B	0.37ab	0.32ab	0.28ab	0.32A
175 N-3splits	23.1a	28.4a	24.0a	25.2A	63.8a	48.0ab	74.4a	62.1B	1592ab	1344a	2291ab	1742A	0.27b	0.21bc	0.26b	0.25B
225 N-3splits	21.5a	25.2a	23.1a	23.3B	59.1a	41.2b	58.3b	52.9C	1900a	1276ab	2323a	1833A	0.27b	0.18c	0.20c	0.22B

Notes: Values followed by different lower case letters were significantly different among N treatments in each experiment, according to Tukey's HSD test ($p < 0.05$). Average values followed by different upper case letters were significantly different among N treatments across the three experiments, according to Tukey's HSD test ($p < 0.05$).

Table 5. Maximum values in plant length and tillers per m^2 identified during the growth period of each variety (Basmati 370 or BW196) under 75 N-, 125 N-, 175 N-, and 225 N-3splits and their average values for each N treatment across the two varieties in Experiments (Exp.) 1–3.

		Plant length (cm)			Tillers (m^{-2})		
Variety	N treatment	Exp. 1	Exp. 2†	Exp. 3	Exp. 1	Exp. 2†	Exp. 3
Basmati 370	75 N-3splits	78.0 ± 2.7	69.8	84 ± 2bc	588 ± 37	313	442 ± 68
	125 N-3splits	83.6 ± 2.8	76.5	96 ± 4b	800 ± 97	320	482 ± 46
	175 N-3splits	88.2 ± 2.9	82.2	114 ± 3a	830 ± 71	425	562 ± 38
	225 N-3splits	89.5 ± 5.3	85.9	119 ± 4a	1012 ± 127	455	747 ± 110
BW196	75 N-3splits	43.5 ± 3.5	48.6	50 ± 1g	868 ± 31	483	703 ± 36
	125 N-3splits	54.6 ± 3.0	55.7	54 ± 0fg	1058 ± 88	590	799 ± 44
	175 N-3splits	57.4 ± 1.6	58.3	60 ± 1efg	1266 ± 51	743	882 ± 43
	225 N-3splits	63.2 ± 1.7	59.3	66 ± 2def	1528 ± 20	710	1040 ± 73
Average	75 N-3splits	60.8B	59.2	67‡	728C	398	572B
	125 N-3splits	69.1AB	66.1	75‡	929BC	455	640B
	175 N-3splits	72.8A	70.3	87‡	1048AB	584	722AB
	225 N-3splits	76.4A	72.6	93‡	1270A	583	893A

ANOVA

	df	Mean square	df	Mean square	df	Mean square	df	Mean square	df	Mean square	df	Mean square
Replication	2	28			2	4	2	1227			2	2023
Variety (V)	1	54,637***			1	12,558***	1	832,537***			1	532,228***
N treatment (T)	3	269**			3	797***	3	307,991***			3	115,155***
V × T	3	18			3	134**	3	23,075			3	1136
Residuals	14	413			14	16	14	18,596			14	12,865

Notes: Values are average ± standard error for each variety under each N treatment. †In Experiment 2, measurements were conducted using only one replication. ‡Multiple comparisons were not performed, because the effect of the variety × N treatment interaction was significant according to ANOVA ($p < 0.05$). Average values followed by different uppercase letters were significantly different according to Tukey's HSD test ($p < 0.05$). Values within a column followed by different lower case letters were significantly different according to Tukey's HSD test ($p < 0.05$). *** and ** indicate significance at the 0.001 and 0.01 probability levels, respectively, according to ANOVA.

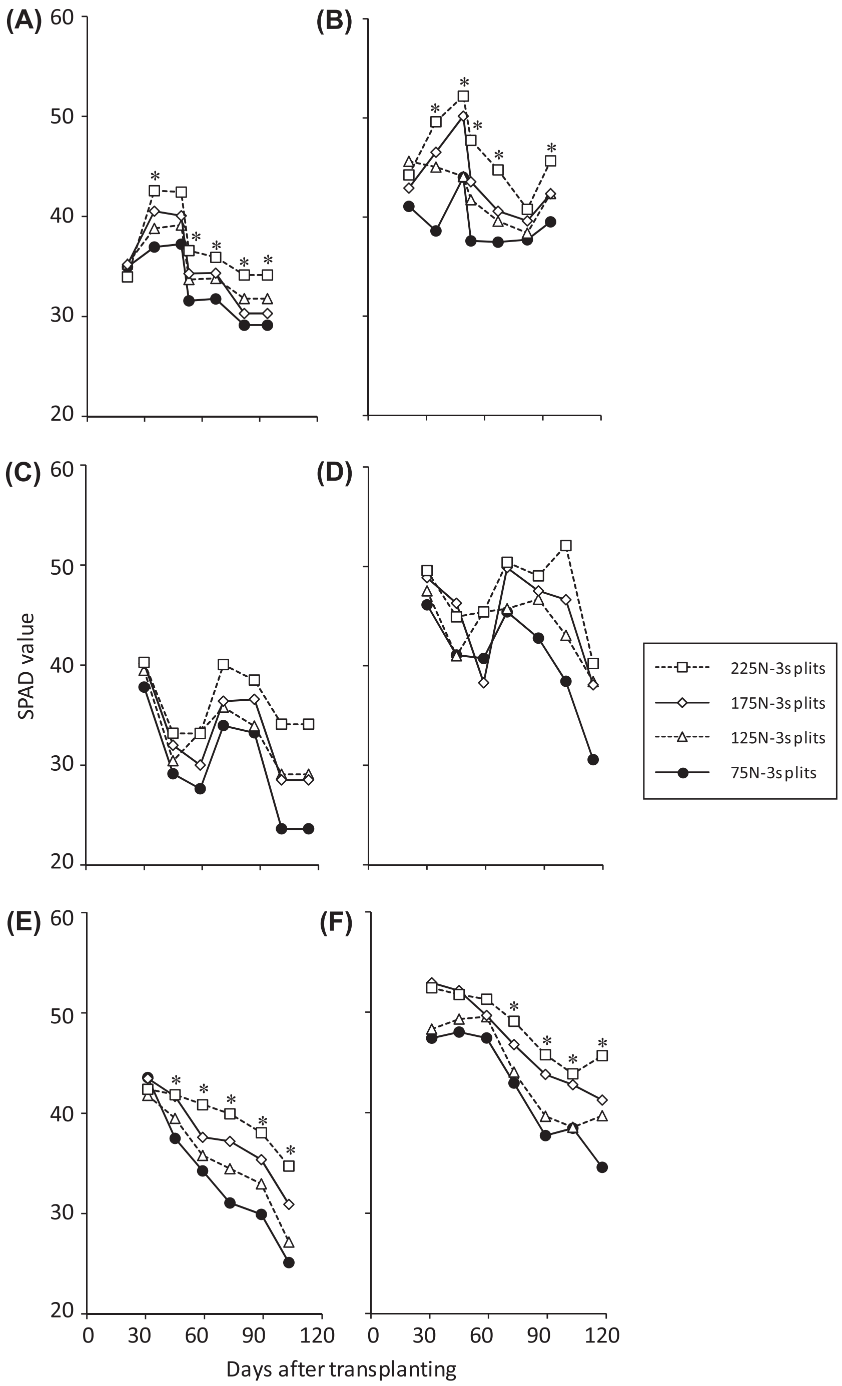

Figure 3. Changes in soil-plant analysis development (SPAD) values in Experiments 1 (A, B), 2 (C, D), and 3 (E, F) for Basmati 370 (A, C, E) and BW196 (B, D, F).

Notes: * Indicates significance at the 0.05 probability level, according to the analysis of variance performed among the four treatments on each sampling day. In Experiment 2, measurements were taken from a single replication.

of grain yield to N fertilizer and possible ways to improve grain yield in Mwea.

4.1. Response of biomass production to increases in the amount of N fertilizer

Biomass production increased with increases in the amount of N fertilizer in Mwea and the shoot DW sometimes reached approximately 20 t ha^{-1} or more in several varieties under high N conditions (Figure 2(A) and (C)). These shoot DW values (20 t ha^{-1} or more) are comparable to those of high-yielding varieties (Takanari and IR72) under favorable weather conditions in China, Japan, and Philippines, where grain yields of these varieties were approximately 10 t ha^{-1} or more (Table 1). In particular, the highest shoot DW in this study (24.6 t ha^{-1})

exceeded that in Table 1 (24.1 t ha^{-1}). The daily maximum temperature in Exp. 1 and 3 ranged from 27.2 to 32.5 °C and from 25.1 to 30.7 °C, respectively (Figure 1(A)). This was similar to that observed at several locations in China, Japan, and Philippines where higher Takanari and IR72 yields were recorded (Table 1). The daily minimum temperature in Exp. 1 and 3 ranged from 14.3 to 17.7 °C and 13.5 to 18.6 °C, respectively (Figure 1(B)). This was lower than that observed in other high-yielding conditions (Table 1). It is generally accepted that low night temperatures or large differences in day–night temperatures promote an increase in shoot DW (Katsura et al., 2008). The radiation in Exp. 1 and 3 ranged from 12.9 to 19.9 MJ m^{-2} d^{-1} and 10.7 to 20.5 MJ m^{-2} d^{-1}, respectively (Figure 1(D)). This was less than that recorded in the Philippines, but comparable to or higher than radiation at Yunnan, China, and several locations in Japan (Table 1). These findings suggest that potential biomass productivity of rice in Mwea is among the highest level in the world if sufficient amount of N fertilizer is applied.

The favorable weather conditions, however, do not always persist in Mwea. Shoot DW was lower in Exp. 2 than in Exp. 1 and 3, irrespective of variety (Figure 2(A)–(C)). The low shoot DW could have been attributed to, at least partly, low daily maximum and mean temperatures coupled with low radiation during the middle and late growth stages (Figure 1(A), (C) and (D)). Such non-ideal weather conditions might occur during the rice cultivation period in Mwea and the reasons for this seasonal variation in biomass production should be further studied.

4.2. Responses of rice grain yields to increases in the amount of N fertilizer

Although weather conditions were favorable for biomass production in Exp. 1 and 3, increasing N amounts above 75 kg ha^{-1} did not lead to increased grain yields since grain filling and HI decreased with increases in the amount of N fertilizer in Basmati 370 and BW196 (Tables 3 and 4; Figure 2(D)–(F)). These findings indicate that the current N fertilizer amount (75 kg N ha^{-1}) is sufficient for Basmati 370 and BW196 when applied in three splits at fixed timings, which is the common practice in Mwea. This information is not only important for economic reasons, but also important in order to avoid environmental hazards associated with over-application of N fertilizers. Moreover, diseases such as sheath blight and rice blast are generally prevalent in N-rich canopies (Peng et al., 1996; Tsujimoto et al., 2010). In our study, the blast-susceptible variety Basmati 370 showed severe blast damage under high N conditions in Exp. 1 (data not shown).

Although N uptake by rice plants was not determined in the present study, it is reasonable to conclude that increases in the amount of N fertilizer enhanced N uptake by Basmati 370 and BW196, based on the higher values obtained for shoot DW, tillers per m^2, panicles per m^2, spikelets per m^2, plant length, and SPAD values under higher N conditions in Exp. 1–3 (Tables 3–5; Figure 3). These results are similar to those reported in Singh et al. (1998) where grain yields of several rice genotypes did not respond to the amount of N fertilizer, although their N uptake and shoot DW continued to increase as the amount of N fertilizer increased. Furthermore, Ramasamy et al. (1997) reported that yield increase was not always associated with increased crop N uptake. This would be applicable for Basmati 370 and BW196 in this study.

4.3. Possible reasons for the poor responses of grain yield to N fertilizer in Mwea and future prospects

The no advantageous effect of increased N fertilizer application on grain yield in Exp. 1–3 might be caused by excess foliage at early growth stage and N shortage in the late growth stage. However, the SPAD values did not support the N shortage in the later growth stage under high N conditions (Figure 3). Mutual shading is another adverse effect of excess foliage growth. In further studies, shoot DW, leaf area index, and leaf canopy structure should be determined at each growth stage and growth analysis based on this data would help to determine if excess foliage is the reason of the poor responses of grain yield to N fertilizer in Mwea. In addition, limitation of nutrients other than N such as zinc (Kundu et al., 2017) and silicon (Tsujimoto et al., 2014) cannot be ruled out.

The increasing influence of low temperature under high N conditions may be the reason for the low grain yield with poor grain filling under higher N conditions in Mwea. Plants exposed to low temperature show lower fertility under high N conditions than under normal N conditions (Hayashi et al., 2000). In Exp. 1–3, grain filling was higher under the standard 75 N-3splits (79.0 to 84.9%) than under 225 N-3splits (41.2 to 59.1%; Table 4). Such large differences in grain filling might be due to low temperature since under cold conditions grain filling was above 80% when leaf N concentration at PI was less than 2.0% but below 40% when leaf N concentration at PI was more than 3.5% (Lee, 2001). Thus, the influence of low temperature in Mwea might become obvious under high N conditions for Basmati 370 and BW196, though no low fertility due to low temperature was observed under the standard 75 N-3splits. This concept is in line with the finding by Hayashi et al. (2000) that low temperature and high N conditions during reproductive growth stage cause severe spikelet sterility.

Daily minimum temperature was the lowest in Exp. 1 (around 14 °C) among three experiments during middle growth stage (Figure 1(B)). However, the negative effects of increased N application on grain yield was not necessarily conspicuous in Exp. 1 (Figure 2 and Table 3). This finding seems to contradict the concept that the influence of low temperature in Mwea might become obvious under high N conditions. However, daily maximum temperature was the largest in Exp. 1 (around or higher than 30 °C) among three experiments during the period (Figure 1(A)). Satake (1969) reported that the occurrence of spikelet sterility in rice was diminished if the daily maximum temperature is high enough even when the night temperature is considerably low. This would be applicable for Exp. 1 in this study.

It should be pointed out that the poor responses of grain yields to increase in the amount of N fertilizer were not limited to Basmati 370 and BW196. Under high N conditions, grain yields of Takanari and IR72 did not increase, but their shoot DW did (Figures 2). Such poor responses of these varieties' grain yield to increase in the amount of N fertilizer might be due to, at least partly, growth conditions in Mwea: in another location, for example, the grain yield of IR72 was significantly higher ($p < 0.05$) under 120 kg N ha^{-1} than under 90 kg N ha^{-1} (Peng et al., 1996). Validation of the concept that the influence of low temperature in Mwea becomes obvious under high N conditions would suggest that introduction of high-yielding varieties without paying attention to their sensitivity to low temperature is not effective in increasing rice production in the region. There are varietal differences in the response of grain yield to the amount of N fertilizer (e.g. Fageria & Barbosa Filho, 2001; Samonte et al., 2006) and in sensitivity to low temperature (e.g. Wainaina et al., 2015). It might be possible to increase grain yield under high N conditions by improving low-temperature tolerance of local popular varieties and/or by introducing new low-temperature tolerant varieties to Mwea. Increasing grain yield using these procedures should be tested in further studies, under Mwea's tropical highland conditions.

5. Conclusions

The results from this study indicated that high biomass production could be achieved by increasing N fertilizer application in Mwea. However, the increased biomass production did not increase grain yield due to decreased grain filling under increased N fertilizer applied in three splits at fixed timing. Excess applications of N fertilizer should be avoided until ways to avoid the poor responses of grain yield to N fertilizer are clarified in Mwea. It is necessary to clarify the factors causing the decreases in grain filling with increases in the amount of N fertilizer to promote rice production in Mwea. The increasing influence of low temperature may be one of the reasons for the poor grain filling under high N conditions in Mwea.

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Disclosure statement

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