EFFECTS OF NITROGEN LEVELS ON YIELD AND NITROGEN UTILIZATION EFFICIENCIES OF TWO RICE VARIETIES IN KIRINYAGA COUNTY, KENYA

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Abstract

Nitrogen is one of the most limiting elements in crop production and this is even worse in the tropics where the farmers do not have the capacity to purchase the inputs. Ammonium and urea forms of nitrogen have traditionally been applied in paddy rice. Despite the anaerobic conditions expected in flooded rice, conversion of ammonium to nitrate has been reported. Nitrate easily gets leached and lost. The other loss pathway is volatilization, leading to emissions of ammonia gas. There is need for judicious application of N to ensure proper utilization without incurring the losses. This can be assessed by determination of nitrogen use efficiencies. The current study evaluated the utilization efficiencies of two rice varieties (Nerica 4 and Nerica 10) supplied with four rates of nitrogen in form of urea. The experiment was laid in Randomized Complete Block Design (RCBD) with split plot arrangements, where two varieties represented the main plot while the nitrogen rates constituted the sub-plots. The experiment was replicated three times. The growth parameters determined were plant height, panicle length, shoot dry weight, root dry weight, Harvest index and grain weight. The grain and straw nitrogen contents were determined by Kjeldahl method and the following utilization efficiencies were computed: Nitrogen Use Efficiencies (NUE), Nitrogen Agronomic Efficiency (NAE), Nitrogen Harvesting Index and Nitrogen partitioning in grain and straw. Analysis of variance was performed using Statistical Analysis System version 9.00. Means were separated using least significance difference at a significance level of 5%. Associations between variables were determined by regression analyses. The results revealed significant differences in shoot and root dry weights, grain weight and plant height with Nerica 4 being superior in these characters compared to Nerica 10. The NHIs were not affected by nitrogen rates in both varieties though all of them were generally high (more than 0.6). The NAE ranged between 23-25% which is a fairly good indicators of better N utilization. The NUE ranged between 25-158, with majority being above 100 which is also an indication of low soil nitrogen. Moreso, the partitioning of the N to the grain was threefold or more in the grain compared to the straw. The association between N rates and grain yields were positive. Whereas the association was polynomial for Nerica 4, it was linear for Nerica 10. We conclude that more N can still be applied to Nerica 10 for increased yield since the N levels supplied were still sub-optimal. For Nerica 4 there could have been other factors such as low temperatures that contributed to low yields.

Key words: nitrogen partitioning, remobilization, N mining, N export, nitrogen agronomic efficiency

Introduction

Increase in fertilizer nutrient input, especially N fertilizer, has contributed significantly to the improvement of crop yields in the world (Cassman *et al*., 2003). Development of semi-dwarf rice (*Oryza sativa* L.) varieties in the 1960s was a rallying call during the grain revolution led to greater achievement towards high grain yield particularly under increased N fertilizer rates because of their lodging resistance at high N inputs (Chen *et al*., 2014). To maximize grain yield, farmers often apply a higher amount of N fertilizer than the minimum required for maximum crop growth (Lemaire and Gastal, 1997). According to previous work by Pham *et al*. (2004), over-application of nitrogenous fertilizer has been shown decrease grain yield. The reduction in yield is either by increasing susceptibility to lodging and/or damage from pests and diseases (Cu *et al*., 1996). The high rates of N fertilizer input and improper timing of N application in China and the world-over have led to low recovery efficiency as well as low agronomic N use efficiency. It is not surprising that Zhu (1985) reported a recovery efficiency of less than 30% for ammonium bicarbonate and 30–40% for urea in China. Furthermore, Li (1997) estimated that recovery efficiency (a very important component of NUE) for rice in China was around 30–35%. However, Li (2000) observed that the average recovery efficiency of rice in Jiangsu Province was only 20%. This was further confirmed by Wang *et al*. (2001), who reported that recovery efficiency of farmers' N fertilizer practice was 18% in an on-farm experiment conducted in Zhejiang. Peng *et al.* (2006) found that recovery efficiency of farmers' N fertilizer practice was 20 to 30% in four provinces in China.

Nitrogen is required in large quantities by rice and N deficiency frequently limits rice grain yields in traditional rice (*Oryza sativa* L.) farming systems (Vinod and Heuer, 2012). Nitrogen losses from fields result in N fertilizer recoveries of 20–40% where N fertilizer is used in lowland rice production. The majority of these losses are thought to be through NH_3 volatilization and full denitrification of NO_3- to N_2, as a result of the highly reduced soil conditions that occur in flooded rice systems (Mikkelsen, 1987; Cassman *et al*., 1998). Optimizing N uptake and minimizing potential environmental impacts by predicting the most favourable timing, rate and placement of N fertilizer application to lowland rice crops have thus been the subject of continued research as argued by Cassman *et al.* (1996). These therefore need to be prevented by proper N management through Nitrogen use efficiency and utilization.

Nitrogen use efficiency is separated into different component indices by agronomists using N-omission plots as demonstrated by Novoa and Loomis (1981). The yield increase that results from N application in comparison with no N application is defined as Agronomic N use efficiency (ANE, kg kg^{-1}). The previous results by Zhu (1985) estimated NAE to be less than 30% for ammonium bicarbonate and 30-40% for applied urea in China. A decade later, Li (1997) estimated NAE for China to be about 30-35, an indication of an upward application of nitrogen fertilizers.

Another important indicator crucial for nitrogen management, according to Fageria (2014), is nitrogen harvest index (NHI). The NHI is defined as the ratio between nitrogen (N) uptake in grain and N uptake in grain plus straw or shoot. Incidentally NHI ratio refers only to N in the aboveground parts of the plant (Rattunde and Frey, 1986), and this is due to the fact that N in the roots, particularly of field crops has little influence on the efficiency of N partitioning (Fawcett, 1980). This therefore implies that the NHI is an important index to measure re-translocation efficiency of absorbed N from vegetative plant parts to grain. According to the previous work of Fageria and Baligar (2003), the NHI is considered key in measurement of N partitioning in crop plants, and is indication of how efficiently the plant utilizes the nitrogen for grain production. Since grain nitrogen index (GHI) is positively correlated with grain yield, agronomists consider it necessary that more N be re-tanslocated to grain. Fageria (2014) further argues that the use of adequate rate and source of N is an important aspect in improving N uptake and use efficiency and consequently NHI in crop plants. This is because nitrogen is very dynamic in soil-plant systems and changes with time and space. It is this realization that necessitated the current experiment and the realization that the NUE has mostly been a concern of countries with oversupply of N, but undersupply of N can lead to mining of the minerals hence even a stronger need for estimation of NUE and its components.

Materials and Methods

Description of study site

The study was conducted at the Kenya Agricultural and Livestock Research Organization (KALRO-Mwea) Kirogo farm in Kirinyaga County. Kirogo farm is situated at a latitude of 00 37'S and longitude 370 20' E at an elevation of 1159 m above sea level. The area receives an average rainfall of about 850mm per annum which is divided into long rains (March to June) with an average of 450mm) and short rains (mid-October to December with an average of 350mm). The rain is characterized by uneven distribution in total amount, time and space. The soil of the experimental field is *Nitisol* containing 2.39% total carbon (C) 0.19% total N (Njinju *et al*., 2018).

Experiment design and layout

The experiment was laid out as Randomized Complete Block Design (RCBD) with a spilt-plot arrangement and the treatments were randomly assigned and it was replicated three times. The main plots were the rice genotypes and the sub- plots were the nitrogen rates. Plots measuring 2m × 1.5m were marked for planting each of the five upland rice genotypes. The seeds were directly planted in rows.

Yield and yield components

Harvesting was done at the appropriate time (40 days after heading). This was when 80% of the grain had reached hard dough stage. At maturity, all panicles per hill of the selected plants were harvested and threshed. Filled grains were separated from unfilled grains and counted. The filled grains were weighed in grams using weighing balance and one thousand grain weight was taken. The data on yield and yield related parameters were recorded during and after harvesting. Measurements of the main tiller from the ground to the neck and then to the tip of the panicle was taken using a ruler to obtain the culm and panicle lengths respectively. The number of hills and panicles harvested in each plot were counted, recorded and put together for processing. Panicle weight, thousand grain weight and grain yield were adjusted to 14% moisture content.

Analysis of Plant Tissue Nitrogen and Nitrogen Partitioning

Five plant samples from the experimental plots were collected at vegetative, reproductive and harvesting stages. Plants were thoroughly washed in running water free from soil. The samples were separated into shoots and grain at harvesting and taken to the laboratory for drying at temperature of 70^0 C for 48 hours. The N content in the plant tissue was determined by Kjeldahl (Bremner, 1996) digestion procedure since it converts N compounds into ammonium that can be analysed titrimetrically. The machine was rinsed six times before the start of the analysis procedure. Four (4) % of boric acid solution was pipetted into 20 ml test-tube containing bromocresol green and methyl red indicator. Ten (10) ml of the plant sample was pipetted into Gerhardt tubes, blanks being the first to start followed by quality control samples. The digestion tubes were injected with 10ml of 40% NAOH and 20 ml of water. Forty (40%) of aliquot was distilled against the boric acid and later boric was titrated against 0.05 of H_2SO_4. The residue was dissolved in 25ml pf of 1.00 M hydrochloric acid. A sample of 0.25g of the tissue was mixed with 2 g of Na_2SO_4 and 7ml of digestion mixture was concentrated into

Sulphuric acid, selenium and salicylic acid. Drops of Sodium Thiosulfate was added after 2 and 3-4 hours. The mixture was then allowed to cool for 45 minutes and 4 ml of hydrogen peroxide at 30 % was added. Mixture digestion was done at 410^0 C till a clear liquid was obtained and cooled with water. The % of nitrogen was determined calorimetrically by an auto-analyzer. To determine nitrogen partitioned to roots, stem, leaves and grain the N content obtained was divided by the total amount of N in the whole plant and later converted to a percentage through multiplying by 100.

Computation formulas for Nitrogen Use Efficiency

The NUE in rice was determined as per the Wang and Zhou (2014) method. Which was computed as follows: N use efficiency=N uptake from fertilized plants- Uptake in unfertilized crops /N rate of applications.

Computation of N-agronomic efficiency and Nitrogen Harvest Index

Agronomic use efficiency was determined through use of the following equation; Difference method by (Varvel and Peterson, 1990). NAE = (GY_1)-(GY_0) / R; GY_1 = Grain yield from fertilized plots; GY_0 = Grain yield from unfertilized plots; R = rate of fertilizer N applied- where NAE- Nitrogen Agronomic efficiency. NHI was computed according to Muchow (1988), as the ratio between N uptake in the grain and N uptake in the straw.

Data Analysis

Data collected in the field was arranged and compiled for statistical analysis. Analysis of variance (ANOVA) was performed using SAS statistical computer package version 9.00 TS Level 00M0 XP-PRO platform to test for levels of significance due to treatments and interactions Means were separated using least significance difference (LSD) test at a significance level of 5%. Associations between variables were determined by regression analyses.

Results

Results revealed significant differences in parameters between the Nerica 4 and Nerica 10 and N levels. Nerica 4 was superior to Nerica 10 in terms of plant height, panicle length, grain weight shoot and root dry weights as well as harvest indices. The differences were statistically significant ($p \le 0.05$) except for panicle lengths and harvest indices (Table 1). Different levels of N differentially affected these parameters, with increased levels of nitrogen leading to increased plant height. All the nitrogen rates led to significantly higher plant height ($p \le 0.05$).

The results also revealed lack of interactions between varieties and nitrogen rates. Meaning that the varieties did not have synergetic effects on nitrogen uptake and utilization due to incremental the nitrogen. A similar trend was observed with grain weight per plant, shoot and root weights. The results recorded generally low harvest indices (HI), which were no significantly differences for N as well as varieties. Furthermore, the interactions between N rates and Varieties and had no significant effect on HI.

Influence of nitrogen forms on Nitrogen Agronomic Efficiency (NAE), Nitrogen Harvest index (NHI) and Nitrogen use efficiency (NUE) in Nerica 4 and Nireca 10 varieties

There were significant ($p \le 0.05$) differences in Nitrogen Agronomic efficiencies (NAE) between the varieties due to increasing nitrogen levels (Table. 2). The NAEs varied from 23.37 to 24.57 for Nerica 4 variety, which was 5.1% in NAE values between lower (26 kg ha^{-1}) and highest application (68 kg ha^{-1}) rates while on the other hand, it varied from 25.68 to 26.22 for Nerica 10 (2.1% between the lowest and the highest N treatments).

Table 1: Effects of nitrogen rates on yield and yield components of Nerica 4 and Nerica 10

Variety	N rates	Plant height (cm)	Panicle Length (cm)	Grain wt/plant	1000 grain wt	Harvest Index (%)	Shoot Dry wt	Root dry wt
NERICA4		90.5b	20.6a	17.1a	27.5a	56.1a	13.3a	0.98a
NERICA10		84.0c	18.4a	11.8b	25.4b	55.5a	9.1b	0.73b
	0N	74.2d	17.3b	8.0c	29.7b	50.0a	7.7d	0.70c
	26N	85.9c	18.3b	13.2b	29.7b	52.6a	11.7c	0.82bc
	52N	93.4b	19.7a	16.0b	30.7a	51.2a	15.3b	0.94ab
	78N	103.9a	20.6a	21.3a	30.8a	52.6a	19.3a	1.04a
Var.		***	***	**	***	*	***	*
Trt.		***	***	***	*	ns	***	**
Var× Trt		ns	ns	ns	ns	ns	ns	ns
CV (%)		7.7	6.9	34.3	4.1	8.0	30.	23.3

[a]Means followed by same letter are not significantly different at p≤0.05

Table 2. Mean nitrogen harvest indices (NHI), nitrogen agronomic efficiency (NAE) and nitrogen use efficiencies (NUE) of rice varieties as affected by nitrogen rates.

	NERICA 4			NERICA 10		
N rates kgha^{-1}	NH1	NAE	NUE	NHI	NAE	NUE
0	0.69^{a}	-	-	0.68^{a}	-	-
26	0.69^{a}	23.37^{a}	107.13^{ab}	0.74^{a}	25.68^{a}	158.16 ^{a}
52	0.74^{a}	24.49^{a}	182.55^{ab}	0.69^{a}	25.50^{a}	45.90^{ab}
78	0.76^{a}	24.57^{a}	30.70^{a}	0.81^{a}	26.22^{a}	26.18^{b}
LSD	0.15	0.39		0.10	1.22	

[a]Means followed by same letter are not statistically different at p≤0.05

The results for Nitrogen Harvest Indices (NHI) in the two varieties had no statistical differences (p≤0.05). Despite lack of statistical differences, all the NHI were relatively high (0.68-0.81). The differences in NHI between the control and the highest rate was 10.1% for Nerica 4 while this difference was 19.1 % in Nerica 10. Furthermore, the NHI and NAE had an increasing trend with increase in N rate in both varieties. The contrary was observed with Nitrogen use efficiency (NUE) which showed a decline with increase in nitrogen rates (107 and 30.7 NUEs for 26 and 78 kg ha^{-1} N respectively) for Nerica 4. Similar trend was observed with Nerica 10 (158 and 26 NUEs for 26 and 78 kg ha^{-1} N respectively). Though the differences were not significant (p≤0.05) in Nerica 4, threefold increase in N rate led to fourfold reduction, while the reduction was fivefold in Nerica 10 and indeed there were significant differences between the lower and highest N rates (p≤0.05) (Table 2).

Effects of nitrogen rates in nitrogen partitioning between the shoot and grain in two rice varieties

The nitrogen rates had no clear effect on nitrogen partitioned to both straw and grains of the two varieties. However the two rice varieties accumulated more nitrogen in the grain (64-80%) for Nerica 4 and (68-72%) in Nerica 10 (Fig.1 a and c respectively).

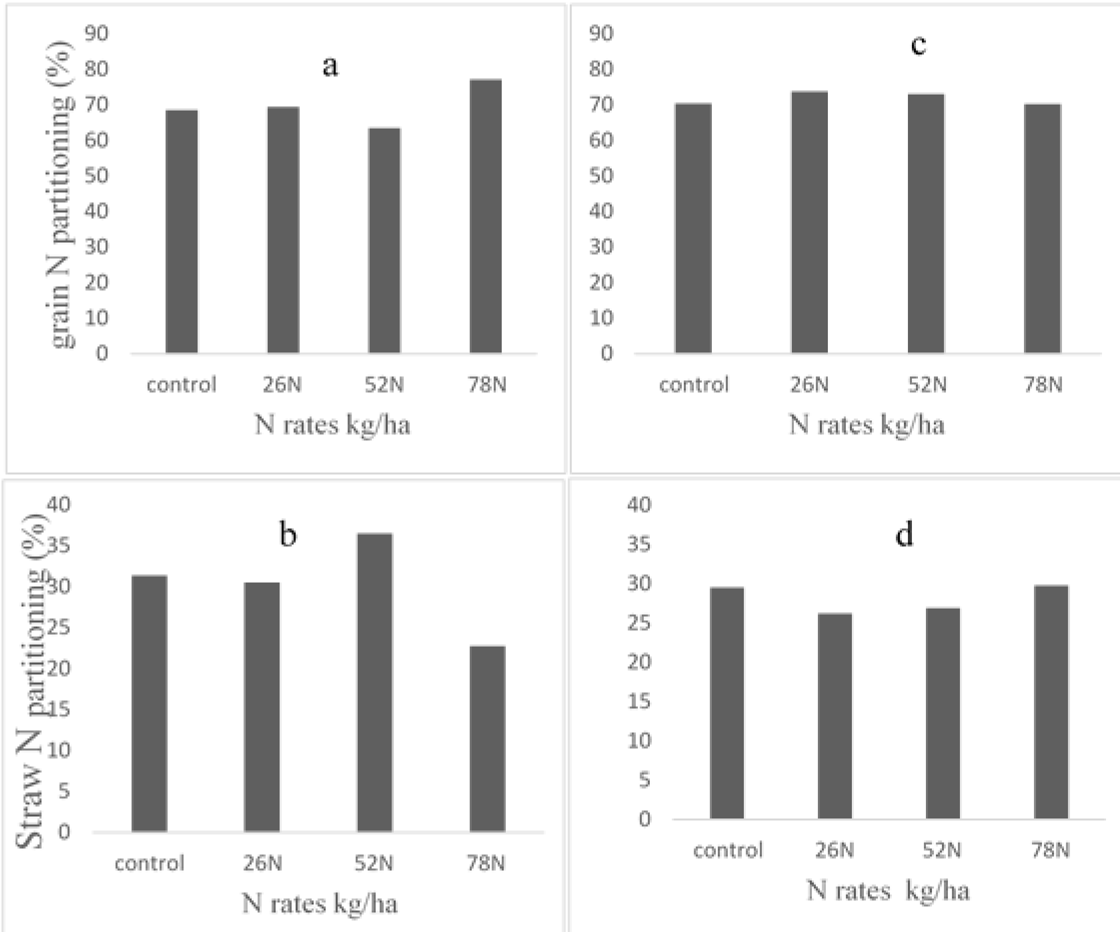

Figure 1: Nitrogen partitioning between straw and grain as affected by nitrogen rates (**a** and **c**- nitrogen partitioned to the grains of Nerica 4 and Nerica 10 respectively; while **b** and **d**- nitrogen partitioned to the straws of Nerica 4 and Nerica 10 respectively).

The nitrogen partitioned to the straw was much lower compared to the grains for both varieties. The amount of nitrogen partitioned to the grain was two to threefold higher than the straw in both varieties. This is an important characteristic in terms of efficiency and economy and health but has implication in nitrogen transport since the grain is always sold away unlike straw that is left in the field to decompose and enhance nitrogen cycle.

Association of nitrogen rates and grain yield in the two Nerica rice varieties

There were clear and positive associations between nitrogen levels and grain yields. The regression was polynomial between N rates and grain yield for Nerica 4 with R^2 value of 0.42 while this was linear in Nerica 10 (R value of R^2=0.55)

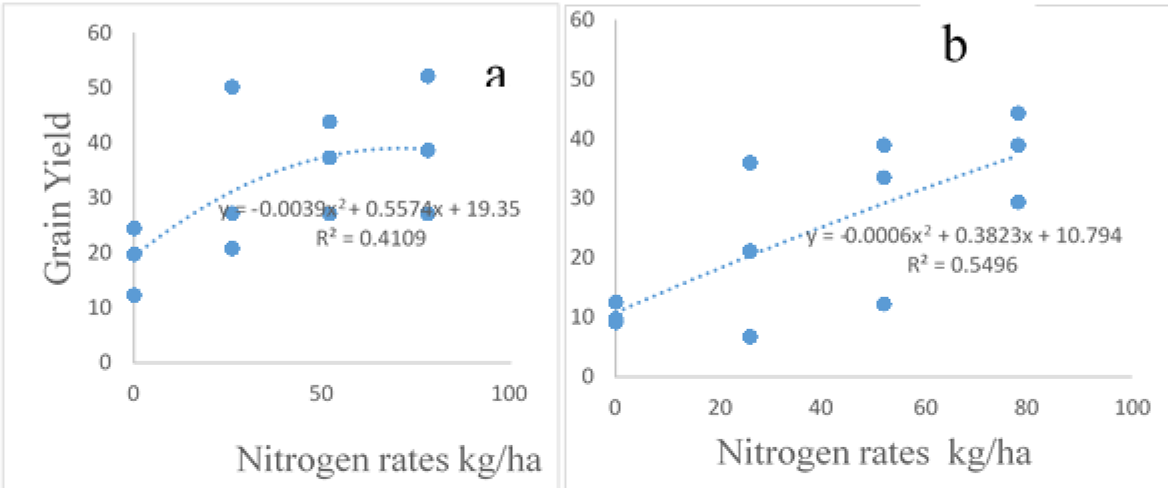

Figure 2: The association between nitrogen rates and grain yields (a: Nerica 4 while b: represents Nerica 10).

Whereas the associations may imply that any amount of N applied beyond 78 kg ha^{-1} may not elicit further yield increment in Nerica 4, further yield increment can still be achieved by applying N beyond 78 kg ha^{-1} (the highest amount in the experimental trial) for Nerica 10.

Discussion

The yield data reported in Table 1 are in concurrence with previous work by Njinju *et al* (2018) who showed that nitrogen and varieties contributed to differences in yield and yield components. The results are also in agreement with those of Fageria and Barbosa Filho (2001) that depicted an interaction effects of nitrogen and rice genotypes on their yields and yield components of rice crop. The work presented here showed a strong positive association between nitrogen rates and grain yields (Figure 2). For the Nerica 4, it would imply that N applied was already optimal and hence further addition would result into economic loss to the farmer and environmental pollution. This is not in tandem with NAE and NUE values reported in Table 2 which had values that border on "nutrient-mining scenarios". These type of results could be explained by other factors other than nitrogen and varieties. Njinju *et al.* (2018) working in the same locality demonstrated lack of grain filling at higher nitrogen and associated this with low growth temperature as a rate-limiting step. The results with Nerica 10 (Fig. 1) showed a linear association between nitrogen rates and grain yields. Similar results were reported by Ntinyari (2018) in terms of nitrogen application rates and IR rice variety. Wekha et *al* (2017) also reported such associations in finger millet varieties supplied with different rates of phosphorus.

The current results on NAE are within the ranges reported by previous workers. For instance, Rahman *et al*. (2014) reported an NAE of 23.6-17.7g g^{-1} for N rates ranging between 40-80 kg ha^{-1}. The results are also in concurrence with the work of Yoshida (1981) who estimated agronomic N use efficiency(NAE) to be between 15–25 kg rough rice per kg applied N in the tropics. These are in tandem with the current findings (Table 2). The results reported herein are in concurrence with those of Cassman *et al*. (1996) reported that agronomic N use efficiency was 15 to 18 kg kg^{-1} N in the dry season in the farmers' fields in the Philippines. In China, agronomic N use efficiency was 15–20 kg kg^{-1} N from 1958 to 1963 and declined to only 9.1 kg kg^{-1} N between 1981 and 1983 (Lin, 1991). Since then, agronomic N use efficiency has further decreased in China because of the increase in N rate (Peng *et al*., 2002). Wang *et al*. (2001) reported that agronomic N use efficiency of farmers' N fertilizer practice was 6.4 kg kg^{-1} in Zhejiang while Peng *et al*. (2006) reported that rice yield increased by only 5 to 10 kg for every kg of N fertilizer. However excessive applications of N fertilizer in soil in turn have drawn much attention to the environmental impact of N fertilization practices (Cassman *et al*., 2002). Therefore, emphasis has been given to N fertilizer management practices to the maintenance of safe environment and sustainable agriculture.

Nitrogen use efficiencies are within the ranges reported in different authors depending on soil N, ranges and crop varieties. Fageria and Barbosa Filho (2001) reported NUE ranging from 35-71 and these depended on N rates and varieties. Just like in the current the study, the lower NUE was observed with higher N rate. The current findings are also in agreements of those by Ntinyari (2018) who reported a much wider NUE range (18-134%) depending on method of application and stage of N. The report by Doberman indicate a higher NUE of about 123 Kg kg-1 for Africa while other regions between 32- 90 kg kg-1 (with 32 representing E Asia and 90 representing E Europe and E Asia). The N rate supply in the regions were higher in E Asia while Africa had the lowest (as low as 10 kg ha-1 while E Asia is an average of 155 kg ha-1). The low NUE in East Asia, which is dominated by China, is of particular concern for the global Nr budget because this region uses the greatest amount of N fertilizer. Declines in NUE on cereal production in developing countries will likely continue without greater investment in research and extension to reverse this trend. The above scenario is not new and has been vividly captured by an excellent review by Masso et al. (2017) who intimated that these processes continuously create the spatial paradox of 'too little' and 'too much' N respectively, perpetuating food insecurity quantitatively and qualitatively. These are likely to contribute to nutrient pollution and eutrophication (Marler and Wallin 2006). In case of "too little; scenario as in SSA case, Kenya included, then higher NUE can lead to nutrient mining and land degradation.

The results revealed quite high NHIs (more than 0.65), though the indices were not significantly different (Fig.2). Similar results were reported by Fageria and Barbosa Filho (2001) in different rice varieties supplied with different N rates (with NHI ranging between 0.45-0.67). The higher NHI is also supported by higher partitioning of N into the grain than to the straw (Fig.1), meaning that more of the N supplied were remobilized from vegetative to reproductive structures, grain included. The NHIs observed in this experiment may imply high efficiency of utilization of N applied to the plant. According to Yesuf and Balcha, (2014), a higher level of NHI is a measure of the nitrogen acquired and transformed in the grains due to high efficiency of the N rate applied. In the grain, N derived from vegetative parts after anthesis is remobilized and relocated in the grain hence contributing to high proportion on the applied N in the grain than any other part of the plant. Additionally, the higher concentration of N in the grain is as a result of decreases in the pools of N reserves soon after anthesis and thus a considerable amount is translocated to the grain (Fergusson, 1999).

Conclusion

The nitrogen agronomic efficiencies and nitrogen harvest indices were generally high and were within ranges of most developing countries, implying better utilization of N by the two varieties but at the same time may be an indicator of nutrient mining. It is also concluded that these efficiencies are inherently affected by the varieties and levels of nitrogen. The other strategies to consider improve the use efficiencies may include timing and split application of N fertilization.

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