



Development of low-nitrogen adapted rice lines under low-nitrogen selection environment

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Abstract

A study was conducted on the materials derived from the crosses, Gampai/IR77674 and Progol/Asahan to evaluate the performance of lines selected through pedigree method of selection under adequate nitrogen (138 kg/ha) and low nitrogen (34.5 kg/ha) conditions. Similar doses of nitrogen were taken for evaluating (cultural environment) the selected lines. Selection was made from F₃ to F₅ generations under adequate and low nitrogen environments, then F₆ was evaluated at Muara Experimental Farm in dry season 2014, using augmented design, with checks replicated thrice. Results showed higher values of panicle weight, panicle length and plant height. The method followed under selection environment, similar to target environment was suitable enough to obtain adapted rice lines for the target environment. It is suggested that to obtain low nitrogen adapted rice lines, selection in breeding program should be conducted under sub-adequate nitrogen conditions in field.

Key word: Rice lines, nitrogen, low, adequate, selection environment

Introduction

As a staple food for almost people in the world, it is necessary to increase the rice production. To achieve this goal the essential plant nutrient should be fulfilled. Rice require as much as 14.7 kg nitrogen (N), 2.6 kg phosphorus (P) and 14.5 kg potash (K) ha⁻¹ for any ton of yield. Nitrogen (N) is an important nutrient for plant. The nutrients are able to be gained by plant from soil, water, fertilizer and organic material (Dobermann and Fairhurst 2000). Nitrogen is an essential crop nutrient for growth and high biomass for producing higher amount of grain yield. However, it is the most limiting element to increase rice productivity

(Zhong-cheng et al. 2012). The absence of N condition causes chlorosis, stunted growth (dwarf plant) and the decrease number of tillers, thus decrease the manifestation of grain weight and yield (Sui et al. 2013).

Rice breeding is mostly conducted in presence of high inputs, it has systematically missed the opportunity to exploit genetic differences at low N inputs (Ceccarelli 1996). However, in resource-poor areas, only a few farmers could fulfill the needs of N. Consequently, many resource-poor farmers with a little or without using fertilizer inputs face problems to fulfill the needs of fertilizer requirement and hence they get poor yield (Azwir and Ridwan 2009; Hossain et al. 2005). In contrast, the use of N fertilizer has increased 7-fold yield worldwide for at least four decades (Hirel et al. 2007). To reduce the losses at the farmers level under this limited condition, rice varieties that could have high production in sub-optimal area, especially low nitrogen content, is of paramount importance (Fess et al. 2011). Therefore, it is necessary to develop rice varieties that are tolerant to low fertilizing, through selecting and testing of promising lines in a location with low fertilizing to predict the genetic progress of the selection method used (Dawson et al. 2008).

There are several selection procedures, such as pedigree, single seed descent or bulk method (Kanbar et al. 2011) or honeycomb and panicle-to-row selections (Ntanos and Roupakias 2001) that have been proposed to improve grain yield in crops. Breeders need to consider carefully the method, which will be the most effective for selecting desirable and low input responding genotypes. The most commonly used

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selection method for rice is pedigree to obtain varieties with high grain yield with desired harvest index (Kanbar et al. 2011). El-Hissewy et al. (1999) reported the efficiency of pedigree selection method for drought tolerance and salinity stress tolerance. However, to determine the need of fertilizers, Ceccarelli (1996) assumed that it can only be identified if selection is conducted under the target level of inputs. Keeping in view the above and related aspects that mentioned before, the objective of this experiment was to identify the effect of low and adequate N selection environments on rice lines and evaluate the performance under low and adequate N cultural environments.

Materials and methods

Plant materials

Materials used the present study were derived from Gampai/IR77674 and Progol/Asahan crosses. The cross between two populations was done in 2011. Pedigree selection was made during three planting seasons from F_3 to F_5 segregating populations. The F_3 generation was selected under low N level (sub-optimal) in dry planting season 2012, followed by F_4 and F_5 generations which were selected under low and adequate (optimum) N level, planted in wet and dry seasons, respectively. The rate of N used was 34.5 kg/ha as low and 138 kg/ha as adequate condition. F_6 generation was evaluated in dry season 2014. Other fertilizers such as phosphorus and potassium were also applied at recommended dose.

Selection method

One hundred and fifty F_3 families each comprising of 20 plants separately from both the cross combinations were planted in field. Selections from each family were made on visual observation considering the short awns and higher panicle weight of healthy plants with 10% selection intensity. The top best performing families were assigned with pedigree number. The F_4 generations each comprising of 20 plants were planted under two different conditions creating selection environments with adequate and low nitrogen application. The best 5% plants were visually selected. Selection in F_5 generation was done on same principle as followed in F_4 . Finally, the top 43 lines each having four panicles were selected and planted as panicle-to-row method as F_6 lines. Thus, a total of 172 lines under adequate and low selection environment from the late season were evaluated in low and adequate nitrogen cultural environments. Six checks, namely, Ciherang, Inpari 6, Inpari 23, Inpari 33, IR77674 and

Asahan were used.

Experimental design and statistical analysis

The experiment was conducted in Latosol soil at Muara Experimental Farm Bogor during four growing seasons from 2012 to 2014. Chemical composition of soil is

Table 1. Chemical composition of soil samples taken from experimental site at Muara Experimental Farm Bogor, DS 2014

Chemical soil	Value	Criteria
C-org (%)	1.79	Low
N-total (%)	0.17	Low
C/N	11	Medium
P ₂ O ₅ HCl 25% (ppm)	222	High
P ₂ O ₅ Bray I (ppm)	8.8	Very Low
K (me/100g)	0.49	Very Low
Mg (me/100g)	2.02	Medium
Ca (me/100g)	11.11	High
KTK (me/100g)	15.18	Low
pH	5.4	Acid

presented in Table 1. The experiments were conducted in augmented design, with three replications of the control varieties. Each line was grown in a plot 5.5m² with spacing of 20cm x 20cm. Each hole consists of one 21-days-old seedling. Each rice line was planted in two separate experiments. In the first experiment 34.5 kg ha⁻¹ nitrogen, whilst in the second, 138 kg ha⁻¹ N were applied at three dates ($\frac{1}{3}$ three days after planting, $\frac{1}{3}$ four weeks after planting and $\frac{1}{3}$ seven weeks after planting). In both experiments, 36 kg of phosphorus (P₂O₅) and 60 kg of potassium (K₂O) were applied to the soil with the first nitrogen fertilizer application. Pest and disease control were done optimally in the vegetative to generative phases. The observations on days to flowering, plant height, number of productive tillers and yield were recorded on 15 plants from each plot. The measurements on number of grains per panicle and 100 grain weight were recorded on five plants only. Data were analyzed following augmented design using SAS 9.1, Microsoft Excel 2007 and Minitab 13. The mean for the treatments were determined using t-test at 0.05 % probability level.

Results

Variance analysis

Mean squares from the analysis of variance for all the

Table 2. Mean square of yield components and yield of lines derived from low (N-) and adequate (N+) environments, under low (N-) and adequate (N+) cultural environments.

Selection environment	Cultural environment	Block	Line	Check	Genotype
Number of productive tiller					
N-	N-	0.0	4.4	5.6	4.9*
	N+	1.1	3.8	5.5	4.2
N+	N-	0.0	5.0*	5.5	6.0*
	N+	1.8	4.9	5.3	5.3
Panicle weight					
N-	N-	0.4	0.7	0.6	0.7
	N+	0.1	0.6	1.1	0.6
N+	N-	0.4	0.5	0.6	0.5
	N+	0.5	9.1*	6.1	0.7
Number of filled grain perpanicle					
N-	N-	182	822*	204	790*
	N+	227	877*	723	883*
N+	N-	182	428*	204	444*
	N+	138	0.6	0.9	921*
Weight of 100 grains					
N-	N-	0.2	0.1	0.1	0.1
	N+	0.0	0.1	0.1	0.1
N+	N-	0.0	0.2	0.1	0.2
	N+	0.0	0.1	0.1	0.1
Yield					
N-	N-	3.9x10 ⁵	22x10 ⁵ *	25x10 ⁵ *	23x10 ⁵ *
	N+	2.3x10 ⁵	25x10 ⁵ *	47x10 ⁵	15x10 ⁵ *
N+	N-	3.9x10 ⁵	15x10 ⁵ *	26x10 ⁵ *	16x10 ⁵ *
	N+	2.5x10 ⁵	14x10 ⁵ *	40x10 ⁵	13x10 ⁵ *

*Significantly different at $P \leq 0.05$

traits measured are presented in Table 2. The results depicted highly significant differences among the rice genotypes for number of filled grain per panicle and grain yield. However, the crosses revealed the existence of non significant differences for panicle weight and weight of 100 grains. Analysis of variance for five characters showed that the blocks were not significantly different.

Effect of selection environment on the mean yield of components in population

Days to flowering in each population selected from low (N-) were less than adequate (N+) environments under low and adequate cultural environments (Table 3). However, the average of both the populations, selected in low and adequate nitrogen level was not significantly different between the two cultural (evaluation) environments, although significant difference was recorded between the populations in the same selection environment. Plant height showed a similar trend where the lines selected from low (N-) were shorter in height than lines selected under adequate environment. Under adequate conditions, the plants became ± 5 cm taller. Significant difference between the selection environment were reflected under evaluation environment.

The average number of productive tillers/plant in the two populations, selected under low environment were higher as compared to adequate cultural environment than low by 10.1 and 8.8 tillers/plant, respectively. A similar trend was noticed in adequate condition. The difference between the adequate and low environment were significant. However, the number of productive tillers were not significantly different between the lines that come from the adequate environment, although selection environments were different (Table 3).

Table 3. Mean of yield components of two populations, derived from low (N-) and adequate (N+) selection environments, evaluated at low (N-) and adequate (N+) environments

Selection environment	Cultural environment	Days to flowering (cm)	Plant height	No. of productive tillers	Panicle weight (g)	Panicle length (cm)	No. of filled grains/panicle	Weight of 100 grains (g)
N-	N-	79.2b	101.6b	8.8b	3.5b	26.1a	114.4a	2.74a
	N+	82.2b	107.9a	10.1a	3.7b	26.2a	117.4a	2.69a
N+	N-	79.6b	102.4b	7.7c	3.8a	25.5a	119.4a	2.81a
	N+	83.5a	109.0a	9.9a	4.1a	25.9a	120.3a	2.77a

*Character with the same letter at a column in each selection environment was not significantly different, with t-test $\alpha = 0.05$; *Yield with the same letter at a column was not significantly different, with t-test $\alpha = 0.05$

Mean of panicle weight, panicle length, number of filled grains and 100 grain weight in both the populations selected under low environment as well as in adequate environment did not differ significantly. The non-significant differences were reflected from the cultural (Evaluation) environment produced by low (N-) and adequate (N+) level of nitrogen.

Yield and the best 10% lines

Grain yield of all the lines under two conditions (selection environment) and evaluated under different nitrogen conditions is shown in Fig. 1. Lines selected

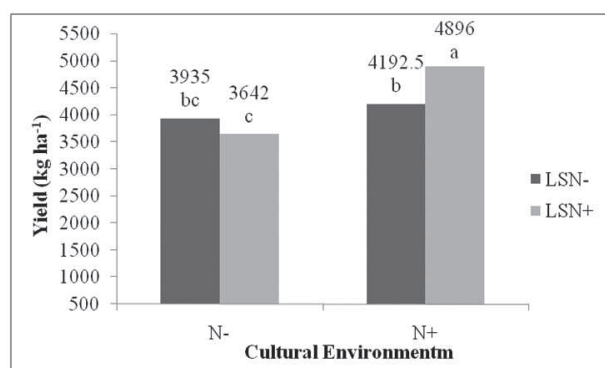


Fig. 1. Mean yield of two populations, derived from low (LSN-) and adequate (LSN+) selection environments and evaluated at low (N-) and adequate (N+) cultural environments

from low selection environment gave higher yield than lines that came from adequate N, evaluated under low condition, whereas the lines originating from the adequate conditions were significantly higher under adequate cultural environment. This suggests that in order to obtain the lines adapted to low N and kept maintaining yield levels at low N conditions, the selection could be done both under low and adequate conditions. However, it would be more efficient if the selection was done in only low conditions because it would reduce the cost involved in developing a suitable variety. As many as 10% of lines with the highest yield from each population, identified in selection environmental conditions and evaluated in cultural environment are given in Table 4. Lines were selected at low conditions had an average high yields if it were planted also on low condition (7517 kg ha⁻¹) compared to adequate (7240 kg ha⁻¹) selection environment.

Similar to yield of line selected in adequate environment were high under adequate cultural environment (7250 kg ha⁻¹) compared to low (6441 kg

ha⁻¹) selection environment, although the difference was not significant. These results indicated that in order to obtain the lines or variety conforming to an environmental condition the selection in a plant breeding program should be done in an environment that is similar to the target environment. A line of, B14250C-213-3-2 from the population 1 that has been selected under low conditions but showed high performance in adequate condition. Otherwise, line selected in adequate (N+) condition gave higher yield in low but less in adequate cultural environments was B14250C-169-4-2.

From population 2, the line B14262C-287-1-2 2, selected in the low environment displayed high yield also at low conditions and still belonged to the 10% of the best lines in the adequate cultural environment. The lines from population 1 showed high yield in low conditions and lines of population 2 showed high yield at the adequate condition. The highest differential selection occurs in the 10% best lines of population 1 derived from low selection environment, grown under low condition. It is suggested that to obtain high yields adapted lines to low N, the selection program should have been done in low and like wise conditions. Pedigree is a selection method that can be relied upon in rice breeding program.

Discussion

Nitrogen (N) is a crucial element in rice production. Low N affects rice growth and development, produces yellow leaves, dwarf plants that reduced grain yield and more loss can be accredited to lodging and panicle sterility (Dunn 2010; Lian et al. 2005). In South-Eastern China, the economically and ecologically adequate N rates were depending on rice subspecies, varieties and cropping systems practiced. The cutting one-third of N use would not reduce rice yield but is expected to mitigate negative environmental impact (Chen et al. 2011). However, using high level of N gave negative impact to environment. The agricultural and ecological environments have been gradually polluted because of the excessive use of nitrogenous fertilizer (Matson et al. 2002; Robertson and Vitousek 2009; Guo et al. 2010). Hence, varieties showed adaptation to low N and high Nitrogen Use Efficiency (NUE) are expected to do better. Rice varieties nowadays are derived from the breeding program where selections are conducted at adequate N conditions that resulted in high input varieties which were less tolerant to low (N-) environmental conditions. To reach high yield in poor environment or low input the use of a low-input

selection environment has been followed in maize (Ceccarelli 1996). It is concluded that the best avenue to a sustainable increase of agricultural production in low-input agricultural systems is through locally based breeding programs. The present experiment resulted in the selection of a line, B14250C-174-2-3 from population 1, identified under low input selection environment and reached the higher yield (8750 kg/ha) among other tested lines in low input N. The line B14250C-150-7-2, selected under adequate environment reached the higher yield 8458 kg/ha level among other tested lines, in adequate N condition. This result was more convincing than reported by Hach

and Nam (2006). They found that application of nitrogen fertilizer at 60 kg N/ha brought the higher net income in wet season while the best dose for rice in dry season was 80 kg N/ha.

Panicle number/plant showed the highest positive direct effect on grain yield/plant (Senapati et al. 2009). The present results showed that panicle weight of population 2 was heavier than population 1 under adequate condition. The panicle weight of population 2 was higher (4.2 g) as compared to population 1 which showed 3.8 g per panicle. However, the panicle length of both the populations was not

Table 4. Yield of the best 10% lines of population 1 (F₆ Gampai/IR77674) and population 2 (Progol/Asahan) always selected on low (N-) danadequate (N+), evaluated under low and adequate N environments

Selection environment		Cultural environment		
		N-	N+	
Line		Yield (kg ha ⁻¹)	Line	Yield (kg ha ⁻¹)
N-				
Population 1	B14250C-174-2-3	8750	B14250C-213-3-2	8223
	B14250C-285-5-2	8522	B14250C-174-2-2	6898
	B14250C-262-1-3	7007	B14250C-169-1-1	6666
	B14250C-87-4-2	6965	B14250C-258-3-1	5300
	B14250C-213-3-2	6610	B14250C-125-5-3	5070
Population 2	B14262C-287-1-2	8510	B14262C-256-2-3	7971
	B14262C-26-2-3	7642	B14262C-42-1-1	7080
	B14262C-148-1-3	7225	B14262C-287-1-2	5933
	B14262C-42-1-1	7370	B14262C-284-1-2	5835
	B14262C-105-1-2	6571	B14262C-155-3-2	5433
	Mean	7517a		6441a
	Diff. Selection	3595		2282
N+				
Population 1	B14250C-169-4-2	8133	B14250C-150-7-2	8458
	B14250C-231-2-3	8088	B14250C-239-2-1	7959
	B14250C-214-1-3	7273	B14250C-196-9-2	7735
	B14250C-236-5-1	6616	B14250C-231-2-1	7760
	B14250C-271-2-3	6607	B14250C-169-4-2	6280
Population 2	B14262C-246-1-1	8393	B14262C-229-3-2	7688
	B14262C-294-1-2	7831	B14262C-165-2-2	6874
	B14262C-84-3-1	6596	B14262C-295-1-1	6719
	B14262C-165-2-2	6550	B14262C-173-5-2	6640
	B14262C-155-2-2	6317	B14262C-295-1-3	6392
Mean	7240b			7250a
Diff. Selection	3369			2385

different from each other whether evaluated under low or adequate N conditions. Yoseftabar (2013) stated that application of 300 kg/ha N increased the panicle length and total number of grains. The number of productive tillers/plant in lines selected under low (N-) environment was significantly higher under adequate environment and there was no significant difference in the lines selected under adequate environment and evaluated under adequate cultural environment. Number of productive tillers were positively and directly correlated to grain yield (Senapati et al. 2009). Panicle number/plant and grain number/panicle should be considered as the most important characters during selection for yield improvement in segregating generations of rice. Rafii et al. (2014) showed that plant height and panicle length of F_1 indica rice could be successfully inherited to the next generations due to high genetic gain and heritability. According to Vanisree et al. (2013) the plant height, panicle length, panicle density and filled grains/panicle exhibited highly significant and positive correlation with grain yield/plant. Nitrogen application determined panicle structure such as number of panicles, spikelet density, panicle length, panicle curvature and the number of grains per panicle. The results showed that selections from population 2 had more grains than population 1, although it was not statistically different. Similarly, the 100 grain weight did not show significant differences between the two populations.

Thanh et al. (2006) used this method to attain field weathering resistance and better agronomic performance of soybean at different locations. The present experiment conducted under similar conditions following pedigree method of selection facilitated the identification of rice lines for the target environment. It appeared more effective in population 1 (Gampai/IR77674) than population 2 (Progol/Asahan). A similar situation was observed by Farag (2013) that pedigree method was not usually effective for the three populations of wheat for selection of desirable traits but he showed that for grain yield/plant and straw yield/plant, this method was comparatively superior only for two populations. Padi and Ehlers (2008) reported that the pedigree selection method for grain yield in cowpea was ineffective as compared to bulk method or Single Seed Descent (SSD). On the other hand, the pedigree selection method was effective in isolating drought tolerant lines of wheat with improved grain yield (Ali 2011). The present study showed that the pedigree method conducted in low N environment was effective to get low nitrogen input adapted rice lines.

Authors' contribution

Coconceptualization of research (APL, S, T, DS, HA); Designing of the experiments (S, APL); Contribution of experimental materials (G); Execution of field/lab experiments and data collection (APL); Analysis of data and interpretation (APL, S, T, DS, HA); Preparation of the manuscript (HA, APL).

Declaration

The authors declare no conflict of interest.

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