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Determining response to nitrogen of promising rice line (O*ryza sativa* L.) in the low country wet zone of Sri Lanka

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Abstract

A field experiment was conducted during the *Maha* season from August 2019 to May 2020 at Regional Rice Research and Development Center, Bombuwala, Sri Lanka to investigate nitrogen response of promising rice line of Bw 12-574 compared at growth and flowering stages of that recommended rice variety (Ld 368). Growth parameters and yield components of both treatments were compared under five different nitrogen levels i.e., 0, 50, 100, 150, and 200% recommended by the Department of Agriculture. The experimental design was a Randomized Complete Block Design with three replicates. Two varieties were tested under 5 nitrogen levels. Data were analyzed by analysis variance (ANOVA) and mean separation procedure by LSD using appropriate SAS procedures. Plant height, number of productive tillers, panicle length, leaf colour

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intensity, and chlorophyll content of leaves in both Bw 12-574 and Ld 368 showed an increasing trend against added nitrogen. The higher nitrogen dosages were influencing the increase of flowering days. The yield parameters including length of panicle and number of grains per panicle have shown significant differences (p=0.05) between the tested line and recommended variety. However, no significant difference was observed between the tested line and recommended variety with respect to filled grain percentage and grain yield. Hence, the response to nitrogen of Bw 12 – 574 was found to be similar to the recommended variety, Ld 368. The study concludes that the same nitrogen recommendation applied for Ld 368 would be recommended for promising rice line Bw 12-574. However, further investigations are needed to confirm the findings.

Keywords: fertilizer, nitrogen response, paddy yield, parameters, rice

INTRODUCTION

The rice production and the use of fertilizer N in rice-growing countries have been increasing over the past decade. In order to produce enough food for the large Asian population, the use of N fertilizer combined with fertilizer reactive rice varieties and the areas under irrigation have been expanded. Since N fertilizer is a costly input, it is imperative that it has to be used effectively**.** The importance of nitrogen fertilization on the grain yield of the rice plant is necessary to know what is best for each variety as well as its influence on components of yield and other agronomic parameters such as tillering, plant height, lodging, and days to maturity. A taller plant is more susceptible to lodging and responds less to nitrogen (Albornoz, 2016). Panicles with a low percentage of sterile flowers permit the application of higher doses of nitrogen and produce better yields. Some factors, like early sowing, meet the twin objectives of producing higher yields and improving the grain quality. Other factors, like increased rates of fertilizer nitrogen, may increase the yield but reduce the quality of the grain (Albornoz, 2016). An adequate supply of nitrogen to the crop plants during their vegetative growth period is very important for the initiation of leaves and florets primordial (fundamental parts).

The type of nitrogen fertilizer can also affect the grain yield and quality (Albornoz, 2016). Some of these fertilizers, such as urea, are considerably cheaper than others, and their use may be justified on economic backgrounds so long as they do not adversely affect grain yield or performance. Chaturvedi (2005) reported a higher yield of calcium

ammonium nitrate is a more efficient source of nitrogen than urea, with a higher content of grain nitrogen.

Plants use both NO_3^- and NH_4^+ in the form of nitrogen (N) (Choudhury and Kennedy, 2004). Treatment of 200% of Department of Agriculture (DOA) recommended nitrogen amount (N4) has shown a significant difference with all other treatments on the chlorophyll content of leaves. It is the most essential factor for proper plant growth and development which increases and enhances yield and performance dramatically by playing a vital role in plant biochemical and physiological functions (Sirisena *et al*., 2006). Nitrogen deficiency is one of the most important nutritional disorders in areas cultivating lowland rice around the world. Recommendations for nitrogen fertilizer for lowland rice varieties (*Oryza sativa* L.) grown on Inceptisols are restricted. In order to achieve higher rice yields, nitrogen fertilization is important and is widely practiced in rice cultivation. This experiment is conducted to detect the nitrogen response of rice in the low country wet zone of Sri Lanka.

The healthy growth of rice plants depends on both internal and external factors; the external factors are light, water, temperature, and nutrients. These factors affect the plant's growth hormones, making the plant grow more quickly or slowly (Choudhury and Kennedy, 2004). The internal factors of plants are the viability of seeds and the performance of the variety (Sirisena *et al*., 2006). Establishment of the crop at the correct time according to the season, selection of the suitable variety for both cultivation area and season, using high quality seeds, proper land preparation practices, optimum water management, proper pest and disease management, and proper weed management are other important factors affecting to yield (Sirisena *et al.,* 2006).

Nitrogen is one of the major nutrients which directly affect the yield of rice. However, N requirement depends on the rice variety and soil condition of the cultivation area. Sometimes the selected rice variety has a higher nitrogen requirement than the DOA recommended level; hence higher rate of N is needed for better yield. On the other hand, over application of N fertilizer may create many environmental issues. In addition, excess application of fertilizer increases the cost of production as fertilizer is a costly input.

The yield of some recommended varieties is low compared to the yield of improved newly released rice varieties (Sirisena *et al*., 2006). There are

different experimental stages during the process of releasing a new variety of paddy. Assessing response to nitrogen is one of the experimental stages above mentioned processes. Therefore, the promising lines need to be tested for their nitrogen response level before release as a recommended variety. Hence, this study aimed to investigate the nitrogen response of a promising rice line of Bw 12-574.

MATERIALS AND METHODS

Location

This study was carried out at the Regional Rice Research and Development Center (RRDC), Bombuwala. The duration of the research was about 9 months from August 2019 to May 2020.

Rice line/ variety

In this experiment, promising line Bw 12-574 was compared with recommended variety: Ld 368 (Table 1). Bw 12-574 is a three and half month promising line (red samba) developed at RRDC, Bombuwala. Parental lines were Bw 361 x Bw 372. Bw 12- 574 showed good taste and higher yield (3.7 t/ha). The plant height of the Bw 12-574 is 80 cm - 95 cm (based on season and soil condition), 1000 grain weight is 15 g. It is moderately tolerant to Brown Plant Hopper (BPH), and iron toxicity. Ld 368 is DOA recommended variety (samba type) which gives a good yield, 1000 grain weight is 15 g. It shows similar characteristics compared to the tested line.

Experimental design and treatments

The experimental design was a Randomized Complete Block Design with three replicates. Two varieties were tested under 5 nitrogen levels as given below.

Nitrogen level (percentage based on DOA recommendation)

N_0 = zero nitrogen
N_1 = 50% of DOA recommended nitrogen fertilizer dosage
N_2 = 100% of DOA recommended nitrogen fertilizer dosage
N_3 = 150% of DOA recommended nitrogen fertilizer dosage
N_4 = 200% of DOA recommended nitrogen fertilizer dosage

V1 = Bw 12-574
V2 = Ld 368

Table 1: Different treatment combinations used for the study

Bw 12-574 (promising variety)	Ld 368 (comparing variety)
T_1 - V_1N_0	T_6 - V_2N_0
T_2 - V_1N_1	T_7 - V_2N_1
T_3 - V_1N_2	T_8 - V_2N_2
T_4 - V_1N_3	T_9 - V_2N_3
T_5 - V_1N_4	T_{10} - V_2N_4

There were thirty (30) plots in the field layout and the plot size was 18 m^2. The total area of the field was 540 m^2. The diameter of inter-channel was 0.30 m and the bund diameter is 0.50 m. Each plot included an inlet and outlet channel for irrigation and drainage practices. In addition to that, each plot was fully separated by the bunds from other plots to avoid mixing of irrigated water with drainage (Figure 1).

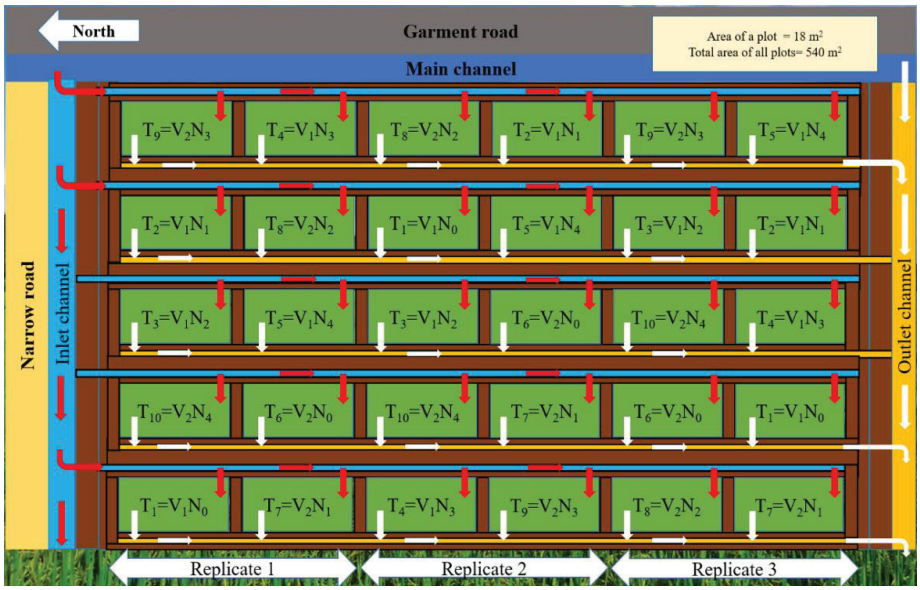

Figure 1: Experimental field layout

Land preparation

First, the field was kept with standing water layer, after the primary land preparation practices. After two weeks, the field was arranged according to the field layout. Also, the channel system and bunds were roughly prepared by mud according to the format. Then the secondary land preparation practice was done also each plot was inundated with water. After one week, the excess water was drained through the channels. Then, the field bunds were prepared sharply using a hoe. Also, each plot was done with puddling as the tertiary land preparation practice before the broadcasting of seeds.

Preparation of the planting materials

First, 160 g of seed paddy was put into the labeled polythene bags (15 bags of Bw 12-574 and 15 bags of Ld 368 separately (Figures 2 and 3)). Five hundred grams of Bw 12-574 seeds were weighed in another bag for border cultivation. Then, these seed bags were soaked in water during the 24 hours and kept for incubation with pressure for about 48 hours. The soaked seeds are wrapped tightly using by wet wool bag and some weight was put on it. That is important to germination of seeds before the broadcasting date.

Figure 2: Weighed Ld 368 egg seed bags

Figure 3: Weighed Bw 12-574 egg seed bags

Sampling

Soil samples (10 g) were collected into the labeled bags from each of the plots in the field to analyze chemical parameters. The sample was tested at the soil chemistry division of RRRDC at Bombuwala.

Figure 4: Sowing of seeds

The procedure of crop establishment

The manual seed broadcasting (sowing) method was used (Figure 4). Hundred grams of Tripple Super Phosphate (TSP) were applied into the field as an initial fertilizer before the sowing of seeds. The plots were labeled according to the field plan. All agronomic practices were conducted as recommended by DOA (Figure 5).

Figure 5: Weedicide application

Data collection

Soil pH and EC were measured using pH and EC meter after shaking the soil sample vigorously. Measurement of soil available N amount was conducted on digestion of samples using by Kjeldhal method. Measurement of available P amount was conducted using by spectrophotometer. Measurement of available K amount was conducted using by flame photometer. Measurement of organic matter percentage was conducted using by Walkey and Black method. Plant height from each replicate was recorded by measuring the height from ground level to the highest point of the plant and then three of the plant heights were taken from each treatment at marked points. The number of leaves of the plants, whose number of leaves was taken using the square feet frame and recorded on a two-week basis and the means determined. The number of tillers was

taken of the plants using the square feet frame and recorded. The number of the plants, whose plant count was taken using the square feet frame and recorded after and the means determined. The colour intensity of leaves was taken of the plants using by leaf colour chart and recorded (Figure 6). The chlorophyll content of leaves was taken from the plants and recorded using the SPAD meter (Figure 7).

Figure 6: Measuring colour intensity

Figure 7: Measuring chlorophyll content

Flowering index

The flowering index of the plants was taken and recorded, day count from sowing date to flowering date, its mean number of flowered plants per square feet as 5%, 50%, and 100% stages into each plot.

Initial soil parameters

According to the initial soil parameters (Table 2), N % is 0.58, Phosphorus content is 12.07 ppm and K is 142.7 ppm. Soil pH is approximately near to 4.87. EC of the initial soil sample is 0.092 µs/cm. The organic matter percentage of the initial soil sample is 10.5 %. Soil parameters of the initial soil sample consist of enough nutrients favorable for better plant growth.

Table 2: Properties of initial soil sample

Sample No.	Sample Name	Total N (%)	P content (ppm)	K content (ppm)	pH	EC (µs/cm)	Organic matter (%)
01	V_1N_0	0.60	12.24	138	4.88	0.07	10.202
02	V_1N_1	0.43	11.32	97	4.92	0.09	8.10
03	V_1N_2	0.57	12.59	103	4.80	0.08	10.53
04	V_1N_3	0.60	11.77	187	4.87	0.08	10.53
05	V_1N_4	0.66	11.57	136	4.91	0.10	11.34
06	V_2N_0	0.63	11.98	239	4.88	0.13	11.34
07	V_2N_1	0.53	11.11	121	4.94	0.08	11.34
08	V_2N_2	0.64	17.92	165	4.87	0.11	11.34
09	V_2N_3	0.53	11.43	102	4.83	0.08	9.72
10	V_2N_4	0.630	8.77	139	4.84	0.10	10.53
Mean		**0.58**	**12.07**	**142.7**	**4.87**	**0.09**	**10.49**

RESULTS AND DISCUSSION

Growth parameters

Height of the plant

Plant height showed an increasing trend from the first two weeks up to six weeks (Figure 8). Plant height has increased in a similar pattern every two weeks in both varieties. The treatment of 100% of DOA recommended nitrogen amount (V1N2), treatment of 150% of DOA recommended nitrogen amount (V1N3) and treatment of 200% of DOA recommended nitrogen amount (V1N4) were not significantly different from each other. Treatment of 150% of DOA recommended nitrogen amount (V1N3) was given the highest plant height (59.5 cm) in a promising line. The lowest plant heights were observed from the treatment of zero nitrogen (V2N0) after six weeks from the sowing date. There was a rapid increase of plant height after the six weeks and the reason may be due to the response of application of chemical fertilizer treatment, 3 weeks and 5 weeks after sowing date. Plant height has increased proportionally to the amount of

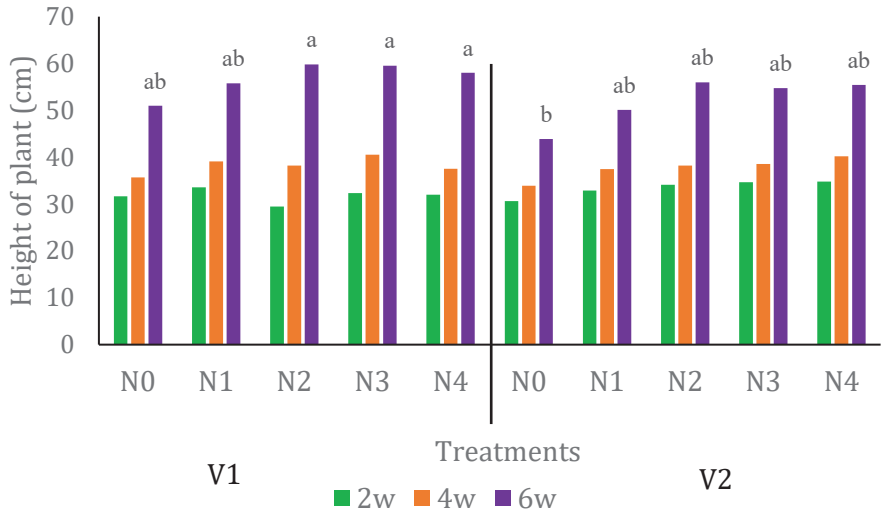

Figure 8: Plant height of the promising line and comparing variety under different nitrogen levels. V1- promising line, V2- comparing variety, N0- zero nitrogen, N1- 50% of DOA nitrogen recommendation, N2- 100% of DOA nitrogen recommendation, N3- 150% of DOA nitrogen recommendation, N4- 200% of DOA nitrogen recommendation. Means with the same letters are not significantly different among the treatments at 6th week.

urea contained in the treatments. Singh and Singh (2017) and Sirisena (2006) have reported plant height and all growth characters in rice crop significantly increased with nitrogen application. Major two factors, variety and nitrogen have significantly influenced the plant height variations and also interaction effects, such as variety and nitrogen interaction have shown a significant contribution on plant height.

Number of leaves

The number of leaves showed an increasing trend from the two weeks up to six weeks. Also, it has increased in a multiple pattern in the 2^{nd}, 4^{th}, and 6^{th} weeks. However, treatments are not significantly different from each other. Interaction effects, such as variety nitrogen have not shown a significant contribution to the number of leaves.

Number of tillers

The number of tillers showed an increasing trend after the six weeks (Figure 9). The number of tillers has increased in a similar pattern between the promising line (V1) and comparing variety (V2). The highest number of tillers was observed in the treatment of 200% of DOA recommended amount of nitrogen (N4) in both promising line (V1) and comparing variety (V2) after six weeks of sowing. However, these treatments were not significantly different from each other. The lowest numbers of tillers were observed in the treatment of zero nitrogen (N0) in both promising line (V1) and comparing variety (V2) after the six weeks of sowing date. Treatment of 200% of DOA recommended amount of nitrogen (N4) is significantly different from both varieties which were treated with 50% of DOA recommended nitrogen (N1) and the treatment which not treated with nitrogen (V1N0, V1N1, V2N0, V2N1). The reason for the above observation may be due to the quick response to the application of chemical fertilizer, in 3 weeks and 5 weeks. The number of tillers has increased proportionally to the amount of nitrogen (urea) contain in the treatments. Interaction effects (variety and nitrogen) have significantly influenced the variations. Variety has not shown a significant contribution to the number of tillers.

Colour intensity of leaves

According to the leaf color chart (LCC), low intensity (light green) to high intensity (dark green) is shown by colour series, these colours

are denoted by 1 to 6 values (Figure 10). The increase of the nitrogen value is proportional to increase in the intensity of leaf colour. Further, the increasing of LCC value means a high concentration of chlorophyll contained in the leaves. The Colour intensity of leaves showed an increasing trend after 4 weeks.

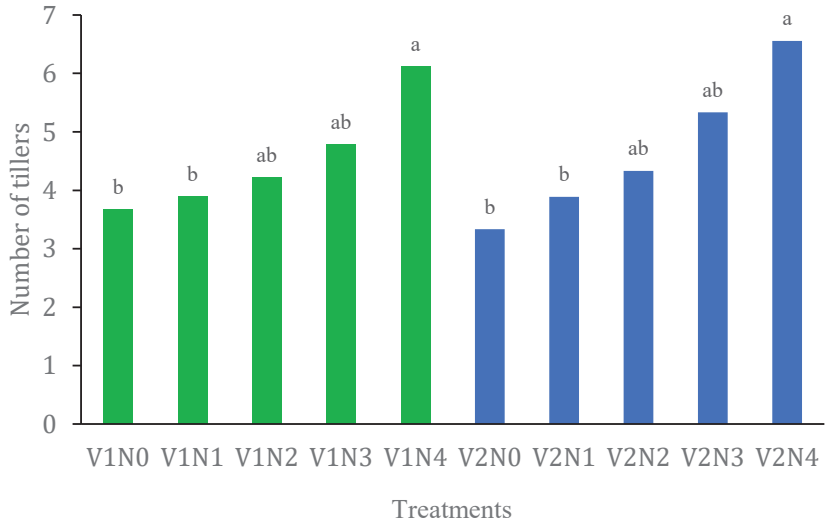

Figure 9: Number of tillers per plant of promising line and comparing variety under different nitrogen levels. V1- promising line, V2- comparing variety, N0- zero nitrogen, N1- 50% of DOA nitrogen recommendation, N2- 100% of DOA nitrogen recommendation, N3- 150% of DOA nitrogen recommendation, N4- 200% of DOA nitrogen recommendation. Means with the same letter are not significantly different.

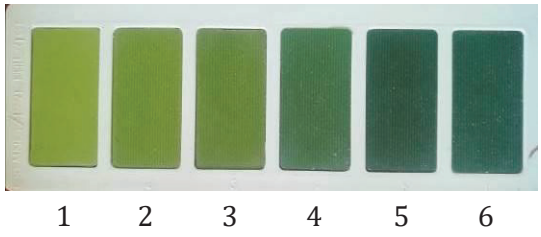

Figure 10: Leaf colour chart to measure colour intensity of leaves

The highest intensity of leaves was observed in the treatment of 200% of DOA recommended nitrogen amount (N4) and the lowest number of leaves was observed in the treatment of zero nitrogen (N0) in both

promising line (V1) and comparing variety (V2) regarding the intensity after the 4 weeks from sowing date (Figure 11). But, the variety and interaction effects of treatment combination have not shown a significant contribution to the colour intensity of leaves. There is a rapid increase of colour intensity after the 4 weeks and the reason may be due to the response of application of chemical fertilizer treatment, 3 weeks after sowing date. Colour intensity has increased proportionally to the amount of urea contained in the treatments. Sirisena *et al*. (2006) reported that leaf color chart (LCC) reading is a good indicator to determine the N requirement of rice varieties under Sri Lankan conditions.

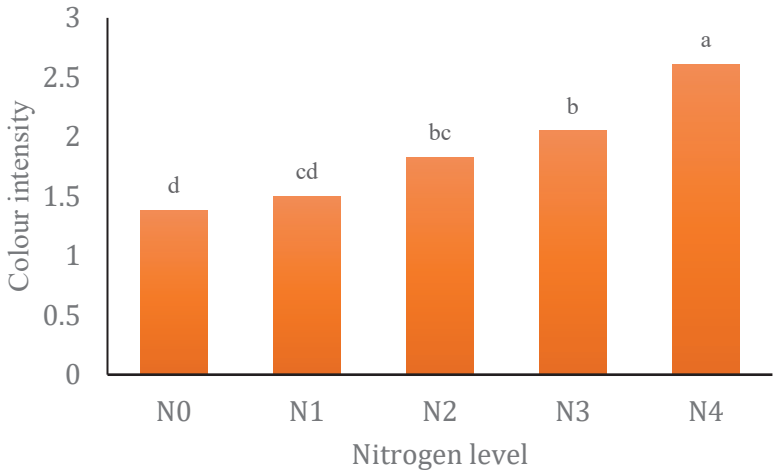

Figure 11: Colour intensity of leaves of promising line and comparing variety under different nitrogen levels. N0- zero nitrogen, N1- 50% of DOA nitrogen recommendation, N2- 100% of DOA nitrogen recommendation, N3- 150% of DOA nitrogen recommendation, N4- 200% of DOA nitrogen recommendation. Means with the same letter are not significantly different.

Chlorophyll content of leaves

Chlorophyll content of leaves had an increasing trend after 6 weeks (Figure 12). Significantly highest intensity (2.6) of leaves was observed in the treatment of 200% of DOA recommended nitrogen amount (N4) and the lowest number of leaves (1.38) was observed in the treatment of zero nitrogen (N0) in both promising line (V1) and comparing variety (V2) after the 6 weeks from sowing date. Also, the interaction effects of treatment

combination have not shown a significant difference in the chlorophyll content of leaves. There is a rapid increase of chlorophyll content after the 6 weeks and the reason may be due to the response of application of chemical fertilizer treatment, 3 weeks and 5 weeks after sowing date. Chlorophyll content has increased proportionally to the amount of urea contained in the treatments (Chaturvedi, 2005).

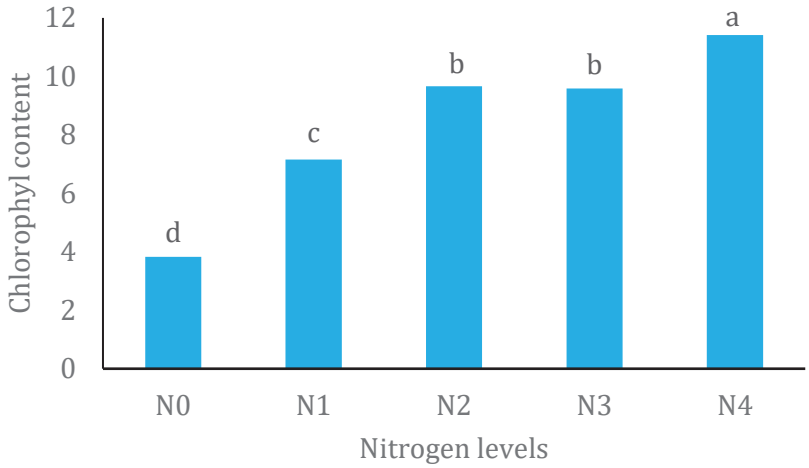

Figure 12: Nitrogen effects of chlorophyll content of leaves of promising line and comparing variety under different nitrogen levels. N0- zero nitrogen, N1- 50% of DOA nitrogen recommendation, N2- 100% of DOA nitrogen recommendation, N3- 150% of DOA nitrogen recommendation, N4- 200% of DOA nitrogen recommendation. Means with the same letter are not significantly different.

Yield parameters

Flowering percentage of plants

The flowering percentage of plants showed an increasing trend (Figure 13). The treatment of 150% of DOA recommended amount of nitrogen (V2N3) and treatment of 200% of DOA recommended amount of nitrogen (V2N4) have shown the highest number of days for flowering than promising line (V1). The promising line (V1) and comparing variety (V2) have shown a significant difference during the flowering. This implies that comparing variety (V2) has required more days for flowering than promising line (V1).

Also, treatments 100% of DOA recommended amount of nitrogen (N2), 150% of DOA recommended amount of nitrogen (N3), 200% of DOA recommended amount of nitrogen (N4) in promising line (V1), and zero nitrogen (N0) in comparing variety (N2) have shown same days for 100% flowering. Treatment of zero nitrogen (V1N0) has shown the lowest number of days for flowering and treatment of 200% of DOA recommended amount of nitrogen (V2N4) has shown the highest number of days (72) for flowering. This may be due to the higher nitrogen dosage and variety characteristics directly influencing on flowering days of paddy (Choudhury and Kennedy, 2004).

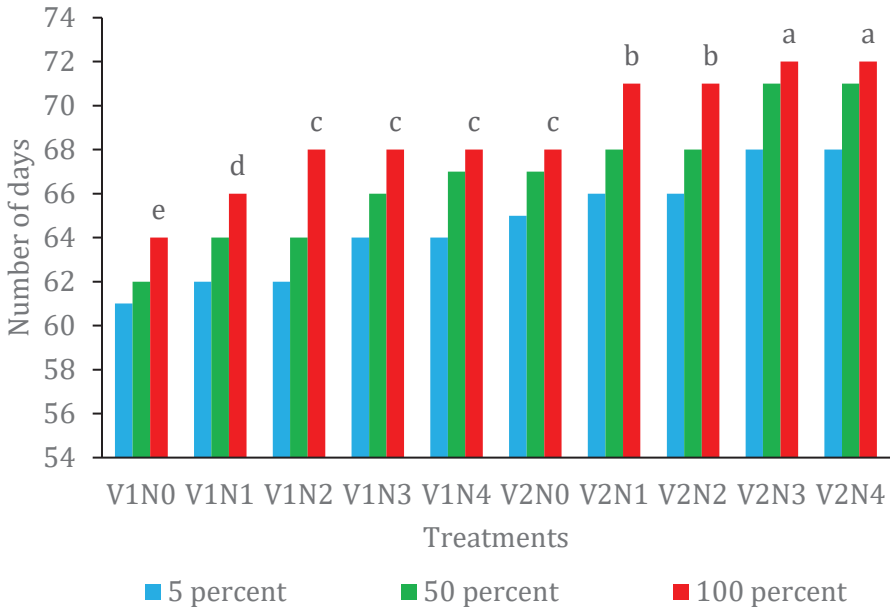

Figure 13: Flowering % stages of plants of promising line and comparing variety under different nitrogen levels. V1- promising line, V2- comparing variety, N0- zero nitrogen, N1- 50% of DOA nitrogen recommendation, N2- 100% of DOA nitrogen recommendation, N3- 150% of DOA nitrogen recommendation, N4- 200% of DOA nitrogen recommendation. Means with the same letters are not significantly different among the treatments at 100% flowering stage.

Length of panicles

The treatment of 150% of DOA recommended amount of nitrogen (V1N3) has shown the significantly highest length of panicle in promising line and different from others. Also, treatments 100% of DOA recommended amount of nitrogen (V1N2), 200% of DOA recommended amount of nitrogen (V1N4) in promising line, zero nitrogen (V2N0), and 100% of DOA recommended amount of nitrogen (V2N1) in comparing variety have shown the lower length of panicle, but not significantly different from others (Figure 14).

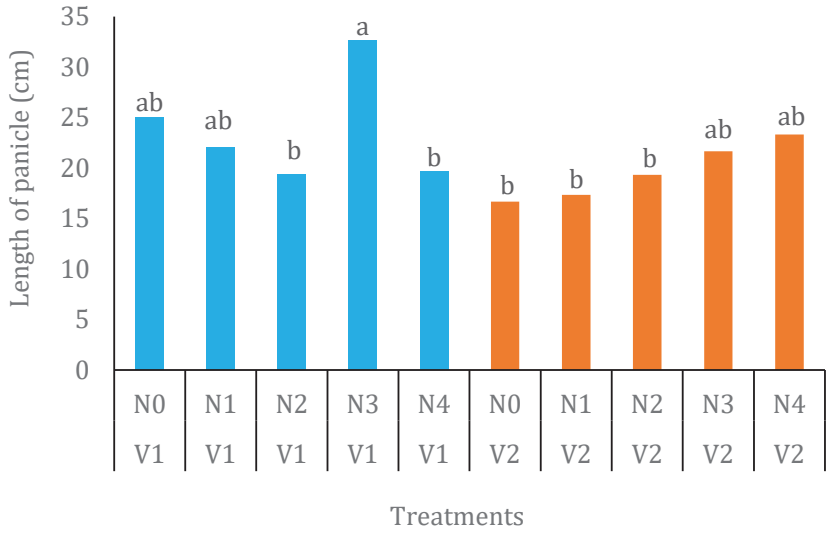

Figure 14: Length of panicle (cm) plants of promising line and comparing variety under different nitrogen levels. V1- promising line, V2- comparing variety, N0- zero nitrogen, N1- 50% of DOA nitrogen recommendation, N2- 100% of DOA nitrogen recommendation, N3- 150% of DOA nitrogen recommendation, N4- 200% of DOA nitrogen recommendation. Means with the same letter are not significantly different.

Number of grains per panicle

The treatment of 50% of DOA recommended amount of nitrogen in comparing variety (V2N1) has shown the highest number of grains per panicle (158) (Figure 15). Also, treatments zero nitrogen (V1N0),

100% of DOA recommended amount of nitrogen (V1N2), 200% of DOA recommended amount of nitrogen (V1N4) have shown a lower number of grains per panicle between the promising line (V1) and comparing variety (N2).

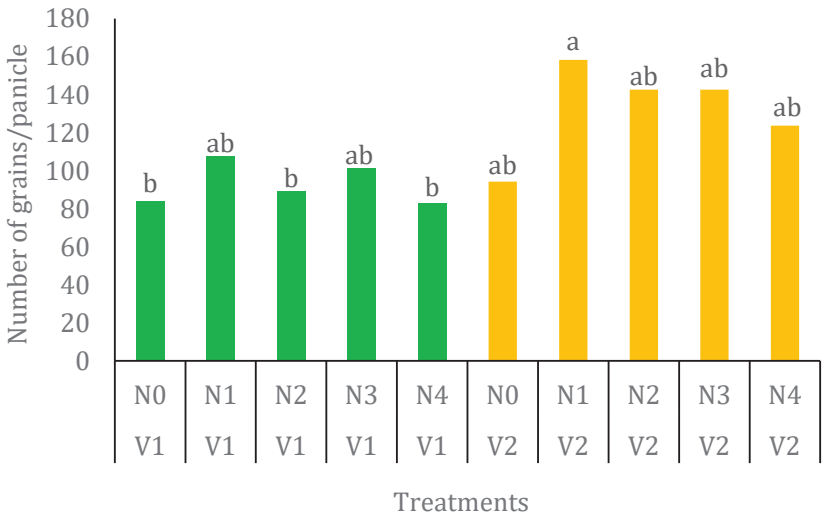

Figure 15: Number of grains per panicle of promising line and comparing variety under different nitrogen levels. V1- promising line, V2- comparing variety, N0- zero nitrogen, N1- 50% of DOA nitrogen recommendation, N2- 100% of DOA nitrogen recommendation, N3- 150% of DOA nitrogen recommendation, N4- 200% of DOA nitrogen recommendation. Means with the same letter are not significantly different.

Percentage of filled grains, thousand grains weight, and total grain weight per plot (kg)

The promising line (V1) and comparing variety (V2) have not shown a significant difference in the percentage of filled grains. Also, the highest dosage of nitrogen may adversely affect the increase of filled grains according to the bar chart data. Treatments have not shown any significant difference in thousand grains weight. Therefore, the percentage of DOA recommended nitrogen dosages are not affecting the thousand grains weight of both promising line (V1) and comparing variety (V2). Total grain

weight per plot (kg) is also not significantly different as the percentage of DOA recommended nitrogen dosages are not affecting the total grain weight per plot of both promising line (V1) and comparing variety (V2).

CONCLUSIONS

It could be concluded that promising line is not significantly different in most of the growth parameters except plant height. Treatment of 150% of DOA recommended amount of nitrogen (V1N3) has shown the highest height in promising rice line (V1) and treatment of zero nitrogen (V2N0) has shown lowest height in comparing variety (V2). Only nitrogen level has a significant difference in colour intensity of leaves and chlorophyll content of leaves. The number of tillers, nitrogen level, and their interaction effect has shown a significant difference with all treatment. Flowering percentage in plants, variety, and nitrogen level have shown a significant difference in both promising rice line (V1) and comparing variety (V2). During the flowering stage, the promising rice line (V1) has shown a lower number of days than comparing variety (V2). Further, the high nitrogen dosages influenced on increasing in flowering time.

Considering the yield parameters, lengths of panicle and number of grains per panicle have shown a significant difference. However, the percentage of filled grain, thousand grains weight, and total grain yield have not shown a significant difference. Therefore, it can be concluded the promising line of Bw 12 – 574 doesn't need special nitrogen requirements compared to the comparing variety Ld 368.

DECLARATION OF CONFLICT OF INTEREST

Authors have no conflict of interest to declare.

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