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Nitrogen Availability and Non-Aromatic Rice Quality under Leaf Color Chart Technique

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ABSTRACT

Leaf Color Chart (LCC) is applied at critical crop growth stage to avoid higher N application rates than standard recommended N rate whether it requires to be adjusted up or down considering leaf color; as it helps estimate plant N requirement for higher economic yields. This study investigated the impact of nitrogen (N) application rates using the Leaf Color Chart (LCC) method on non-aromatic rice yield and quality. Seven N levels, including the recommended dose, were applied to three rice varieties (DR-92, Shahkar, and DR-82) in a factorial experiment. Results showed significant effects of N levels on plant height, biological yield, paddy yield, harvest index, N uptake efficiency, amylose content, and gel consistency. Variety Shahkar was found to be more nutrient responsive and profitable. Optimal N application rates varied depending on the LCC reading, with 25 kg N ha-1 at LCC Col 3 showing the highest paddy yield. This study demonstrates the effectiveness of the LCC method in optimizing N application for improved rice yield and qualitive and can be grown in any rice tract more profitably as compared to DR-82 or DR-92 rice varieties.

Keywords: Rice, LCC, Nitrogen, varieties, Non-aromatic, growth, yield, grain quality

Introduction

Rice (Oryza sativa L.) is the most important staple food worldwide. Among the 23 known Oryza species, only Oryza sativa and Oryza glaberrima are widely cultivated (Reiter *et al.*, 2010). Rice can be stored for several months or even years as rough rice, which is then dehulled to produce brown rice. The bran layer of brown rice is removed to produce white rice (Choi *et al.*, 2015). Approximately 75% of the world's population and 60% of South Asians' food intake consists of rice. While white rice is the commonly consumed type, there is an increasing demand for brown rice due to its high nutritional value (Choi *et al.*, 2015). Basmati rice (Oryza sativa Linn.) ranks second after wheat in world cereal production (Lutfullah and Hussain, 2012; Reiter *et al.*, 2010). Freshly harvested rice is often aged to reduce cohesiveness, increase volume, and improve the texture of cooked rice (Butt *et al.*, 2008).

Recent studies have highlighted the effectiveness of LCC in optimizing N fertilizer use, improving yields, and reducing environmental impact (Bohara *et al.*, 2021). Additionally, research in 2021 demonstrated the potential of LCC to enhance N use efficiency in rice cultivation (Kumar *et al.*, 2021). A 2022 study found that LCC-based N management improved grain quality and reduced N losses (Liu *et al.*, 2022). Furthermore, a 2023 review emphasized the importance of precision N management in rice production, highlighting LCC as a valuable tool (Gao *et al.*, 2023).

In Pakistan, rice is a crucial dietary staple and a significant source of foreign exchange, contributing 20% of the nation's total. Of the rice produced, 34% is Basmati (fine type) and 66% is coarse type. Common cultivated rice varieties include Kangni-27, Sonhari Kangni, Kangni x Torhi, Dokri Basmati, Bangalo, Sugdasi, Lari, Motia Ratria, Irri-6, Irri-8, Lateefy, DR-82, DR-83, Shabab, and Shua (PARC, 2005; GoP, 2022). During 2021-22, rice was cultivated on 3,537 thousand hectares, producing 9,323 million tons, an increase of 10.7% compared to the previous year's 8,420 million tons. The area under rice cultivation has been increasing in recent years, often leading to an exportable surplus (GoP, 2022).

Nitrogen (N) is the most important macronutrient for plants, and its deficiency affects growth, yield, and quality. N is found in amino acids, auxins, cytokinins, proteins, alkaloids, and glucosinolates, all of which are crucial for plant development. It is essential for photosynthesis due to its role in chlorophyll's molecular structure (Havlin *et al.*, 2013; Singh *et al.*, 2010; Varsha Pandey, 2020). While some crops can meet their N needs from soil and organic sources, these are often insufficient to sustain crop productivity (Subedi and Panta, 2018; Varsha Pandey, 2020). Effective nutrient management requires monitoring the crop's N content and adding supplemental N as needed

(Witt *et al.*, 2004). The actual N status of the plant can be determined using tools like the SPAD or Leaf Color Chart (LCC) (Balasubramanian *et al.*, 1999).

LCC provides recommendations on when and how much N fertilizer to use, helping rice plants maintain and increase their N status, thus resulting in higher grain yields (Sathiya and Ramesh, 2009). Current guidelines for N fertilizer application often involve fixed rates at predetermined times, which can increase yields but may not optimize nutrient use efficiency. Consequently, farmers often apply higher N rates to ensure high yields, which can reduce N fertilizer efficiency further (Varinderpal-Singh *et al.*, 2007; Bohara *et al.*, 2021).

The color of rice leaves is a reliable indicator of N deficiency. The LCC, developed by IRRI in collaboration with Asian agricultural research systems, uses four- or six-color panels representing various green shades to assess leaf N content and manage N fertilizer (Varinderpal-Singh *et al.*, 2007). Monitoring leaf color every 7-10 days during the growing season allows for real-time N management based on LCC readings, with N fertilizer applied whenever the LCC value falls below a critical threshold (Varinderpal-Singh *et al.*, 2007). Some farmers prefer less frequent monitoring and apply moderate N rates at transplanting and 21 days after transplant (DAT), then use leaf color at panicle initiation around 42 DAT to guide further N application (Bohara *et al.*, 2021).

An LCC critical value of 3 is recommended for all high-density, direct-seeded rice varieties. This critical value is lower for plants transplanted to new sites. It is advisable to determine the LCC critical value based on local varieties and growing conditions (Bohara *et al.*, 2021). LCC helps boost productivity by optimizing N fertilizer use, making it both efficient and economical. This research aims to enhance N fertilizer use efficiency by using LCC for real-time N application. The hypothesis is that N application through the LCC method will result in optimal N availability, improving plant height, biological yield, and paddy yield in non-aromatic rice varieties. Specifically, it is hypothesized that different levels of N application based on LCC readings will significantly affect the growth, yield, and grain quality of the rice varieties DR-92, Shahkar, and DR-82.

This study introduces a comprehensive evaluation of the LCC technique for real-time N management in non-aromatic rice varieties. Unlike traditional methods with fixed N application rates and timings, the LCC technique dynamically applies N based on the plant's actual needs, as indicated by leaf color. This approach is expected to enhance N use efficiency, reduce the environmental impact of excessive N application, and improve economic yields. Additionally, this study will provide valuable insights into the N responsiveness of different non-aromatic rice varieties, which has not been extensively explored in previous research.

Materials and Methods

Experimental site: The field trials were performed to evaluate the impact of N based on leaf color chart (LCC) techniques on growth, productivity and quality of non-aromatic rice varieties for two consecutive years at the experimental fields of Rice Research Institute, Dokri, Pakistan during 2018 and 2019, the mean annual temperature in Dokri during these years was approximately 26°C, and the area received an average annual rainfall of about 150 mm and the agro ecological conditions of 26.949ºN latitude and 68.192ºE longitude. The proper land preparation operations were performed for equal distribution of irrigation and fertilizers as roots can absorb more nutrients from the soil after transplanting seedlings. Sowing was done with the help of marked hill during June 15-25 in both years. Seed for nursery raising was broadcasted in beds in morning at the rate of 17-18 kg ha^{-1}. When rice nursery plantation attained the age of 25-30 days, the seedlings were uprooted for transplanting; and the plant population was maintained at 250,000 plants ha^{-1}. At tillering stage water level was reduced to increase number of tillers hill^{-1}. Rice crop is most sensitive to moisture stress; therefore, upto the flowering stage subject to availability of irrigation water, the crop consumed NPK fertilizers at the rates of 135-67-25 kg ha^{-1}. All P and K fertilizers alongwith half dose of N fertilizers were applied as basal dose; while remaining N was applied according to the need of the crop as determined through LCC. The observations were made at different stages in experimental area and application of N at the rate of 20 kg ha^{-1} was applied with the matching of Col3 and Col4 of LCC; 25 kg N ha^{-1} was applied with the matching of Col3 and Col4 ofthe LCC; while 30 N kg ha^{-1} was applied matching of Col3 and Col4 of LCC.

The methods of (LCC) observations: Ten normal looking healthy rice plants were randomly selected from each replication of entire treatments. The topmost fully expanded leaf from each plant was selected to record the desired observations. Middle part of the leaf was placedon the chart and the leaf color was compared with the color panels of the LCC. The leaves selected for observation were neither detached nor destroyed; and the observations were recorded under the shade of the body, because direct sunlightmay affect leaf color readings. Average LCC reading was determined for the selected leaves. The LCC that was determined from the N parameter was utilized throughout both seasons of this study. The LCC consists of strips that varied in color from light to dark green, with each shade having a different number of strips to symbolize a higher level of color saturation. Readings for each plot's LCC were taken from 10 disease-free rice plants that were chosen at random fromthat plot. It was then placed on top of the LCC, and the middle area of the leaf was graded according to the color strip that was located on the LCC. During the process of measurement, the leaf that was being measured was kept in the shade ofthe body in order to prevent color fluctuation that could have been produced by sunlight. An application of 20, 25, and 30 kg of N ha^{-1} wasmade topically on the same day when the colors of six out of ten leaves went below the LCC's shade three criterion. We used a randomized complete block design (RCBD) with factorial layouts and N and LCC-based N management in three rice varieties to investigate crop production and N consumption efficiency.

Observations and their recording procedures

Meteorological observations & soil analysis: Meteorological data like minimum and maximum temperature, rainfall, humidity etc were recorded during the growing season of each experiment.Soil samples were drawn from the experimental site before transplanting and after harvesting of the experimental crop.

Agronomic and physiological traits: The plant height was measured in centimeters through measuring tape on the basis of five randomly selected plants from root to tip of plants in each replication of the entire treatments; while the panicle length was recorded on the basis of randomly selected 10 hills. The number of grain $panicle^{-1}$ was recorded from the same ten panicles and average calculated. Similarly, for recording observation on panicle weight, all grains were collected from each panicle and weighed using top loading digital balance in grams. In order to count the number of panicles m^{-2}, the one-meter crop area in each replication of entire treatments was selected and the all the panicles were counted and averaged for one-meter square area. For knowing the number of tillers $hill^{-1}$, 10 hills were selected in eachtreatment, the total number of tillers $hill^{-1}$ was averaged. For counting the hills, one-meter crop area in each replication of entire treatments was selected and the all the hills were counted and averaged for one-meter square area using 1 m^{-2} rows through 1 m^{-2} quadrate. However, the sterility was obtained by dividing the number ofunfilled grains by total number of panicles and multiplying with 100 in percentage.

The seed index was recorded on the basis of 1000 grains weight; where one thousand grains were taken from each plot and weighed on an electric balance after drying at 60ºC for 24 hours in an oven. Similarly, for recording the observationon the grain weight $panicle^{-1}$, all grains were collected from each panicle and weighed on top loading digital balance and data were recorded in grams. The grainweight $plant^{-1}$ was recorded in grams using selected ten plants in each replication of all the treatments and averaged. The biological yield ha^{-1} in kg was achieved byharvesting one-meter square area crop from central rows. The material was air- dried and weighed to record biological yield in kg ha^{-1}. Likewise, the grain yield ha^{-1} was recorded by threshing the harvested rice from an area of one-meter square.The threshed grains were cleaned, dried and weighed to record the average grain yield ha^{-1} in kg. However, the harvest index was recorded on the basis of percent grain yield ha^{-1} of the biological yield ha^{-1}. The leaf area is a physiological trait, and it was observed on the basis of five randomly selected plants from each plot and itwas determined through length and breadth method multiplied by factor i.e. 0.75. The N uptake efficiency was measured by using the following formula: N-uptake Efficiency = Plant N content /N application rate.

Grain quality characters: A simple and easy-to-use test that supplements the amylose content test was developed with the consistency of cold milled-rice paste that is 4.4 percent and dissolved in 0.20 percent N-KOH as described by Suh *et al.,* (2017). The length of the gel after it has been kept ina horizontal position for 0.5 or 1 hour in a test tube containing cold gel is used to determine the consistency of the gel. Even if the amylose concentration of the ricesamples is within the range of 24 percent to 30 percent, the amylograph setback viscosity may differentiate between low, medium, and high consistency rice samples. This is because the amylograph setback viscosity is related with consistency readings. A regression model that predicted the amylose concentration in rice was developed using the tridimensional CIE $L^*a^*b^*$ values that were found in solutions. In regression model, the L*a*b* values and the amylose content displayed a significant degree of connection (R^2=0.99). The $L^*a^*b^*$ values exhibited a significant and unfavorable relationship with the apparent amylose content. Using this method, one might potentially estimate the amount of amylose found in rice. The conversion of L*a*b* values into RGB values and color hexadecimal codes made it possible to reproduce the colors of the starch-iodine solution and create an explicit color board. This was made possible by the conversion of L*a*b* values into RGB values. We were able to classify the entriesand make an educated guess for the amount of apparent amylose present in each one with the use of this color board.

Statistical Analysis: The data recorded was analyzed using ANOVA in RCB factorial design using Statistix 8.1 software (Statistix, 2006). Comparison of means was performedby using the Duncan's Multiple Range (DMR) test. Moreover, R^2 (Regression model) was used as statistical measure of fit that indicates how much variation of a dependent variable is explained by the independent variable in a regression model (Steel *et al.,* 1997).

Results and discussion

Effect of nitrogen levels to Leaf Color Chart (LCC) columns: LCC is a tool used to assess the nitrogen status of crops. The LCC columns in our study refer to the different levels of nitrogen availability, as indicated by the Leaf Color Chart readings. The LCC readings range from 0 (yellow) to 5 (dark green), with higher values indicating higher nitrogen levels in the plant.

The results for nitrogen levels to LCC column showed significant response in relation to different growth, yield and quality traits (P<0.05). Maximum plant height (105.0) and (104.6cm) was recorded at 30 kg N ha^{-1} to column 3 and 4 recommended whereas, height (102.0, 101.6, 102,9 cm) was recorded at application of 135-67-25 NPK (kg ha^{-1}), 25 kg N ha^{-1} to column 3 and 4, respectively (Table 1); however minimum (96.94 cm) was observed at nitrogen application of 20 kg ha^{-1} to column 3 of LCC. Maximum panicle length (26.22 and 26.69) was observed under recommended N (135-67-25 NPK kg ha^{-1}) and 25 kg N ha^{-1} to column 3 of LCC; while panicle length slightly decreased (25.50, 25.50 and 25.41cm) under 25 kg N to column 4 of LCC and 30 kg N ha^{-1} applied to column 3 and 4, respectively. The higher number of panicles (396.3m^2) at 25 kg N ha^{-1} to column 3 of LCC; while, maximum tillers hill^{-1} with panicle (18.13 and 18.46) was recorded at application of 135-67-25 NPK kg ha^{-1} and 25 kg N ha^{-1} to column 3 of LCC, respectively; whereas, the minimum tillers hill^{-1} with panicle as (15.29, 15.75 and 15.63) were found at 20 and 30 kg N ha^{-1} to column 3 and 4 of LCC, respectively.

The maximum tillers without panicle (5.583) were recorded at 30 kg N to LCC Col 4; and minimum (1.25) at 25 kg N to LCC col 3. The number of filled grains panicle^{-1} (126.0) was highest at 25 kg N to LCC col 3; while the lowest (102.8) at 30 kg N to LCC col4. However, the unfilled grains panicle^{-1} (8.27) were recorded at 30 kg N to LCC col 4; and the lowest (4.361) at 25 kg N to LCC col 3.

The results showed that the maximum sterility (8.27%) was observed at 30 kg N ha^{-1} to column 4 of LCC; whereas the minimum sterility (4.361%) was recorded at 25 kg N ha^{-1} to column 4 of LLC. Maximum grain weight panicle^{-1} (38.10 g) was recorded at 25 kg N ha^{-1} to column 3 of LCC. As shown in Table 1, the maximum seed index value (24.14 g) was recorded at 25 kg N to LCC col 3, while the minimum seed index (23.28 g) was recorded at 20 kg N to LCC col 3. The highest grain weight panicle^{-1} (2.793 g) was recorded at 25 kg N to LCC col 4; while the least (2.178 g) was found at 20 kg N to LCC col 3. The grain weight plant^{-1} was highest (38.1 g) at 25 kg N to LCC col 3, closely followed by 36.84 g at 135-67-25 NPK kg ha^{-1}; while the lower grain weight plant (34.71, 33.51 and 32.99) was observed at 20 kg N ha^{-1} to column 3 of LCC, 30 kg N ha^{-1} to column 3 of LCC and 30 kg N ha^{-1} to column 4 of LCC, respectively. Enhanced biological yield of (17640 and 17690 kg ha^{-1}) was observed at 25 kg N ha^{-1} to column 4 of LCC and 30 kg N ha^{-1} to column 3 of LCC. The highest paddy yield (7165 kg ha^{-1}) was observed at 25 kg N ha^{-1} to column 3 of LCC, while the lower paddy yield of 6155, 6228 and 6269 kg ha^{-1} was recorded at 20 kg N ha^{-1} to column 3 and 4 of LCC and 30 kg N ha^{-1} to column 4 of LCC, respectively.

So far, the harvest index is concerned, highest (41.18 and 41.09%) was observed in recommended N level of 135-67-25 NPK kg ha^{-1} and 20 kg N ha^{-1} applied to column 3 of LCC. In case of straw yield, the greater values (10550, 10580, 10980 and 10510 kg ha^{-1}) were recorded at 25 and 30 kg N ha^{-1} to column 3 and 4 of LCC respectively. Maximum leaf area m^2 (181.844 and 187.567 m^2) was observed at 20 and 25 kg N ha^{-1} to LCC column 3 and 4, respectively. Similarly, the maximum nitrogen uptake efficiency (77.042 and 78.211 kg ha^{-1}) was observed at 20 kg N ha^{-1} to column 3 and 4 of LCC, respectively. However, the maximum amylase content (30.72) was recorded at 25 kg N ha^{-1} to column 30 of LCC, followed by 30 kg N ha^{-1} to column 3 and 4 of LCC and the lower amylase content (29.46) was recorded at recommended rate of 135-67-25 NPK kg ha^{-1}. The higher gel consistency values (31.57, 31.35, 31.30, 31.65 and 31.60) were recorded at 20 kg N ha^{-1} column 4 of LCC, 25 kg N ha^{-1} to column 3 and 4 of LCC and 30 kg N ha^{-1} to column 3 and 4 of LCC, respectively; while the bursting % remained unchanged statistically ($P<0.05$) with the values 6.533, 6.442, 6.496, 6.613, 6.513, 6.488 and 6.583 recorded at 135-67-25 NPK kg ha^{-1} to 30 kg N ha^{-1} to column 4 of LCC, respectively.

Response of varieties: The study showed that rice varieties responded differently for various traits (Table-2); and variety Shahkar showed better values for plant population (21.91 m^{-2}), plant height (107 cm), panicles (367.2 m^{-2}), tillers hill^{-1} with panicle (17.13), tillers hill^{-1} without panicle (3.125), filled grains panicle^{-1} (114.1), seed index (24.52 g), grain weight panicle^{-1} (2.473 g), grain weight plant^{-1} (35.24 g), biological yield (17420 kg ha^{-1}) and maximum paddy yield (6909 kg ha^{-1}), with higher harvest index (39.81%) and N uptake efficiency (69.877 kg ha^{-1}). However, variety DR-92 were relatively superior in panicle length (26.80 cm), amylose content (27.01 %), gel consistency (31.67 mm) and bursting percentage (6.536%); but at par with variety Shahkar in traits including plant population, panicles m^{-2}, tillers hill^{-1} without panicle, filled grains panicle^{-1}, grains weight panicle^{-1} and harvest index. However, DR-82 was parallel to variety Shahkar in producing higher values for plant population, tillers hill^{-1} without panicle, and straw yield; but produced highest values for unfilled grains panicle^{-1} and sterility, were at par for amylose content and gel consistency with variety DR-92. The variety DR-82 showed maximum values for rice traits. However maximum unfilled grains (9.116) and sterility (7.889%) percentage was found higher in DR-82 than other varieties

Table. 1. Effect of different N levels under leaf color chart (LCC) on agronomic traits physiological and quality characters of non-aromatic rice

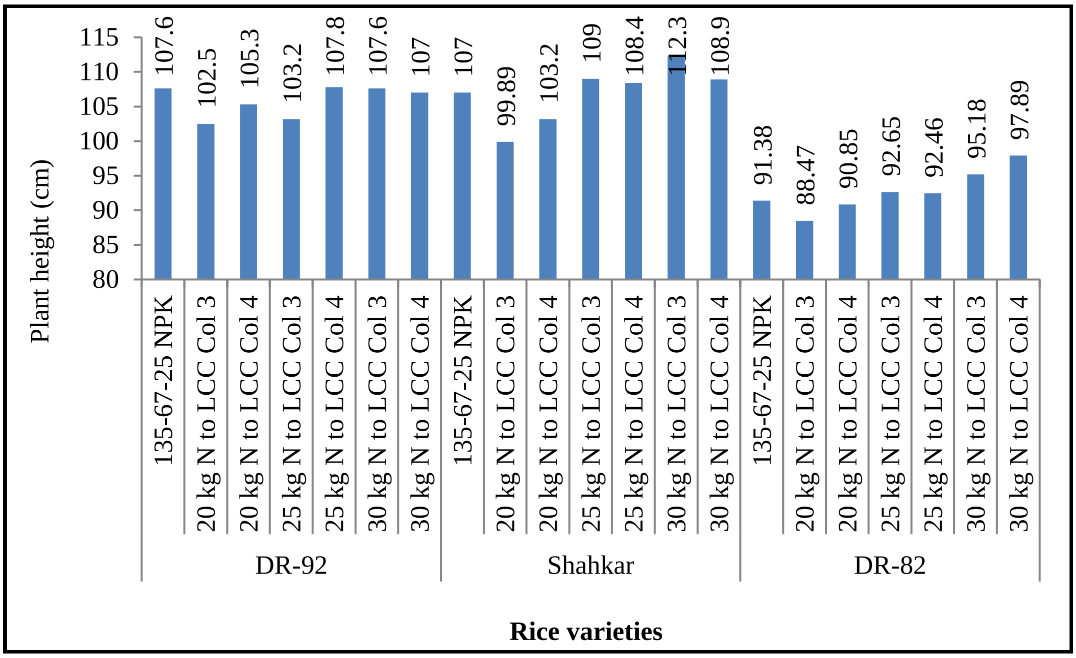
Plant traits	SE	LSD 0.05	TREATMENTS						
			135-67-25 NPK (Kg ha^{-1})	20 kg N ha^{-1} to column 3 of LCC	20 kg N ha^{-1} to column 4 of LCC	25 kg N ha^{-1} to column 3 of LCC	25 kg N to column 4 of LCC	30 kg N ha^{-1} to column 3 of LCC	30 kg N ha^{-1} to column 4 of LCC
Plant population	0.2426	0.6792	22.13 b	21.96 b	21.79 b	23.13 a	22.25 b	21.92 b	21.54 b
Plant height (cm)	0.4710	1.318	102.0 b	96.94 d	99.79 c	101.6 b	102.9 b	105.0 a	104.6 a
Panicle length (cm)	0.2240	0.6270	26.22 a	24.66 c	25.03 bc	26.69 a	25.50 b	25.37 b	25.41b
Panicles m^{-2}	5.882	16.47	385.5 ab	334.9 d	344.1 cd	396.3 a	372.5 b	354.4 c	338.2 cd
Tillers hill^{-1} with panicle	0.2607	0.7298	18.13 a	15.29 c	16.83 b	18.46 a	17.04 b	15.75 c	15.63 c
Tillers hill^{-1} without panicle	0.1951	0.5463	1.833 e	2.500 d	2.500 d	1.125 f	3.333 c	4.292 b	5.583 a
Filled grains panicle^{-1}	0.5621	1.574	121.6 b	105.7 e	108.8 d	126.0 a	111.3 c	106.9 e	102.8 f
Unfilled grains panicle^{-1}	0.2521	0.7056	6.197e	8.469 bc	7.857 cd	5.315 f	7.477 d	8.663 b	9.380 a
Sterility (%)	0.2043	0.5720	4.833 d	7.393 b	6.767 c	4.361 d	6.351 c	7.424 b	8.270 a
Seed index (1000 grain weight g)	0.1120	0.3135	23.50 bc	23.28 c	23.40 bc	24.14 a	23.51 bc	23.64 b	23.46 bc
Grain weight panicle^{-1} (g)	0.0403	0.1128	2.564b	2.178 d	2.301c	2.793a	2.547 b	2.360 c	2.370 c
Grain weight plant^{-1} (g)	0.3661	1.025	36.84 b	32.71d	34.57 c	38.10 a	34.71 c	33.51 d	32.99 d
Biological yield (kg ha^{-1})	93.28	261.1	16940 c	15470 d	15730 d	17290 b	17640 a	17690 a	17480.ab
Paddy yield (kg ha^{-1})	45.88	128.4	6970 b	6155 e	6228 e	7165. a	6768 c	6477 d	6269 e
Harvest index (%)	0.3574	1.000	41.18 a	41.09 a	39.86 b	40.65 ab	38.11 c	36.43 d	36.06 d
Straw yield ha^{-1}	233.2	652.8	9357 b	8318 c	9206 b	10550. a	10580 a	10980 a	10510 a
Leaf area (cm^{-2})	4.1014	8.92	177.93 b	177.451 b	181.844 a	187.567 a	165.564 c	179.59 ab	179.518 ab
Nitrogen uptake efficiency (kg ha^{-1})	1.1875	3.6764	51.354 fg	77.042 a	78.211 a	72.212 b	70.486 bc	58.306 d	56.729 de
Amylose content	0.2171	0.6077	29.46c	29.95 bc	29.88 bc	30.72a	30.34 ab	30.44 ab	30.13 ab
Gel consistency (mm)	0.2628	0.7358	30.50 b	31.16 ab	31.57 a	31.35a	31.30 a	31.65 a	31.60 a
Bursting (%)	0.0436	---	6.533	6.442	6.496	6.613	6.513	6.488	6.583

Table. 2. Comparison of non-aromatic rice varieties for growth, yield, physiological and quality response to different treatments (N levels under LCC)

Plant traits	**SE**	**LSD 0.05**	**Varieties**		
			DR-92	**Shahkar**	**DR-82**
Plant population	0.1588	0.4447	22.34 a	21.91 a	22.05 a
Plant height (cm)	0.3083	0.8631	105.9 b	107.0 a	92.70 c
Panicle length (cm)	0.1466	0.4105	26.80 a	25.85 b	24.02 c
Panicles m^{-2}	3.851	10.78	363.6 a	367.2 a	351.8 b
Tillers $hill^{-1}$ with panicle	0.1707	0.4777	16.59 b	17.13 a	16.48 b
Tillers $hill^{-1}$ without panicle	0.1278	0.3576	3.107 a	3.125 a	2.839 a
Filled grains $panicle^{-1}$	0.3680	1.030	113.4 a	114.1 a	108.0 b
Unfilled grains $panicle^{-1}$	0.1650	0.4620	6.842 b	6.909 b	9.116 a
Sterility (%)	0.1338	0.3745	5.802 b	5.765 b	7.889 a
Seed index (1000 grain weight g)	0.0733	0.2052	23.02 b	24.52 a	23.15 b
Grain weight $panicle^{-1}$ (g)	0.0246	0.0689	2.496 a	2.473 a	2.391 b
Grain weight $plant^{-1}$ (g)	0.2396	0.6708	34.82 ab	35.24 a	34.26 b
Biological yield (kg ha^{-1})	61.07	170.9	16330 c	17420 a	16920 b
Paddy yield (kg ha^{-1})	30.03	84.07	6453 b	6909 a	6365 c
Harvest index (%)	0.2339	0.6549	39.58 a	39.81a	37.77 b
Straw yield ha^{-1}	152.7	427.4	9621 b	10090 a	10070 a
Nitrogen uptake efficiency (kg ha^{-1})	0.8397	1.9570	65.705 b	69.877 a	63.421 bc
Amylose content	0.1421	0.3978	31.82 a	27.01 b	31.57 a
Gel consistency (mm)	0.1721	0.4817	31.67a	30.30 b	31.95 a
Bursting (%)	0.0286	--	6.536	6.611	6.425

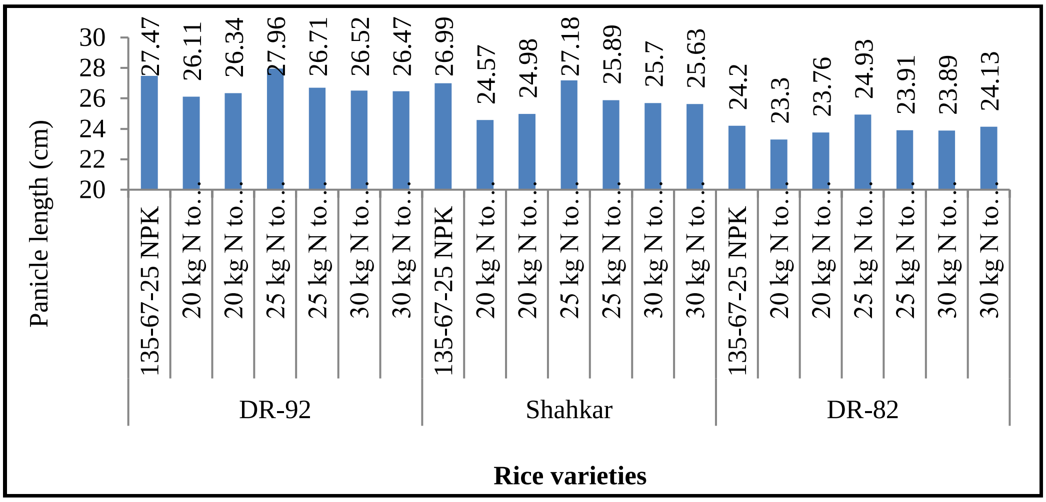
It was further observed that varieties DR-92 and Shahkar showed maximum values for panicles (367.2 and 351.8 /m^2), tillers $hill^{-1}$ without panicle (3.107) and (3.125); filled grains $panicle^{-1}$ (113.4) and (114.1), grain weight $panicle^{-1}$ (2.496 and 2.473 g), harvest index (39.58%). DR-92 and DR-82 varieties showed maximum values for amylase content (31.82 and 31.57) and gel consistency (31.67 and 31.95 mm). Relatively higher values for DR-92 were noted for plant height (105.9 cm), grain weight $plant^{-1}$ (34.82 g), paddy yield (6453 kg ha^{-1}), straw yield (9621 kg ha^{-1}) N uptake efficiency (65.705 kg ha^{-1}); whereas Shahkar produced large panicle length (25.85 cm), greater amylase gel consistency (30.30 mm) and DR-82 gave more panicles (351.8 m^2), filled grains $panicle^{-1}$ (108.0) grain weight $panicle^{-1}$ (2.391 g) grain weight $plant^{-1}$ (34.26g), biological yield (unit 16920), harvest index (unit 37.77%) percentage respectively. The overall results showed that the variety Shahkar ranked first for various rice growth, yield and quality traits of non-aromatic rice varieties followed by variety DR-92 and DR-82 ranked 3rd for observed rice traits.

Interactive effect of varieties × N levels to column LCC: The results of experiment revealed a significant growth, yield and quality response of rice to N levels to LCC chart and varieties. The maximum plant height (112.3 cm) was recorded at interaction of variety Shahkar × 30 kg N ha^{-1} to column 3 of LCC; while the minimum plant height (88.47 cm) was observed in variety DR-82 × 20 kg N ha^{-1} to column 3 of LCC (Figure 1). Greater panicle length on average (27.96 cm) was recorded in variety DR-92 × 25 kg N ha^{-1} to column 3 of LCC; whereas the least panicle length (23.3 cm) was recorded in variety DR-82 × 20 kg N ha^{-1} to column 3of LCCm (Figure. 2). Similarly, enhanced panicles number m^{-2} (419.8 m^2) was recorded in variety DR-92 × 25 kg N ha^{-1} to column 3 of LCC; whereas the lower panicle number (315.9, 315.8 and 309.1 m^2) was recorded in variety DR-92 x 20 kg N ha^{-1} to column 4, DR-82 x 30 kg N ha^{-1} to column 3 and 4 of LCC, respectively (Figure. 3).



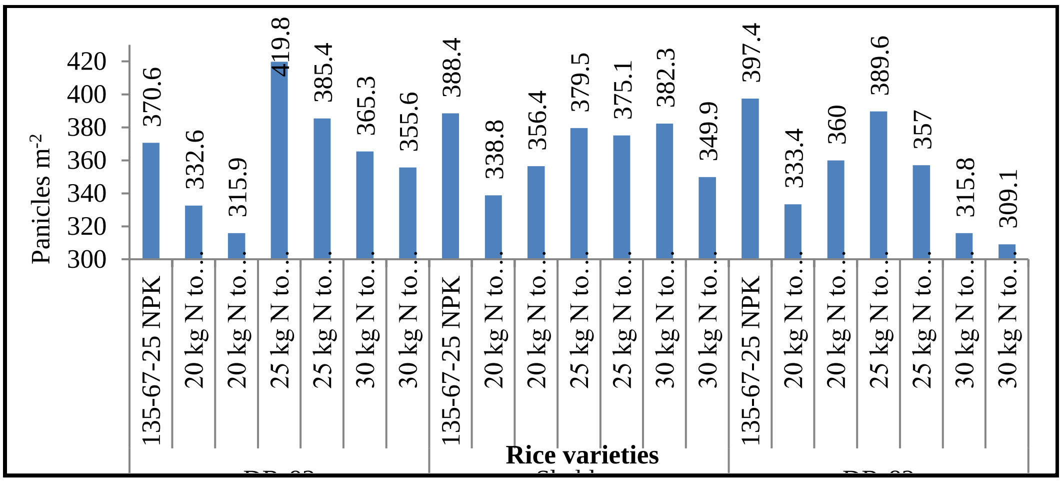
S.E.±0.8158 LSD 0.05 2.284

Figure. 1. Interactive effect of varieties × treatments under LCC on plant height (cm)



S.E.± 0.3879 LSD 0.05 1.086

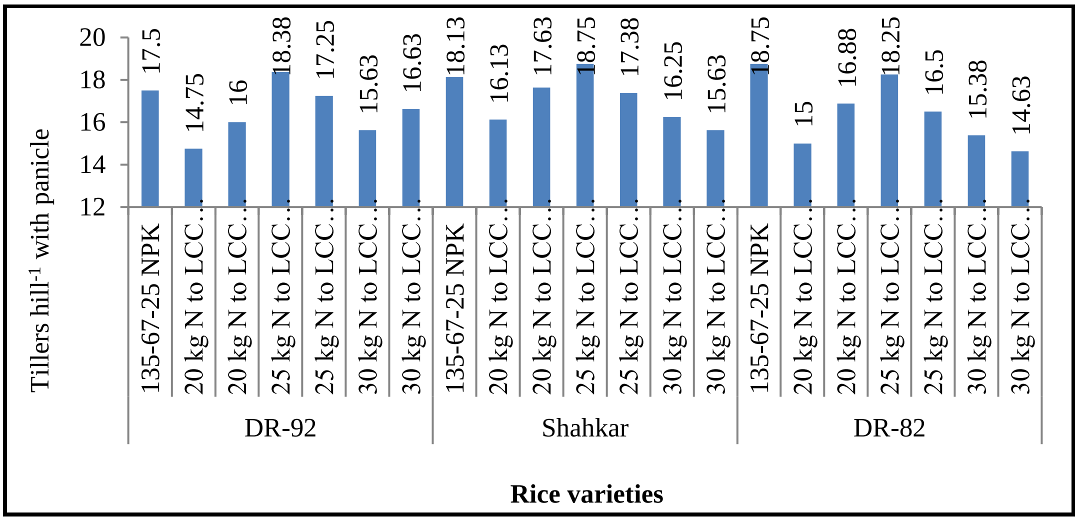
Figure. 2. Interactive effect of varieties × treatments under LCC on panicle length (cm)



S.E.±10.19 LSD 0.05 28.52

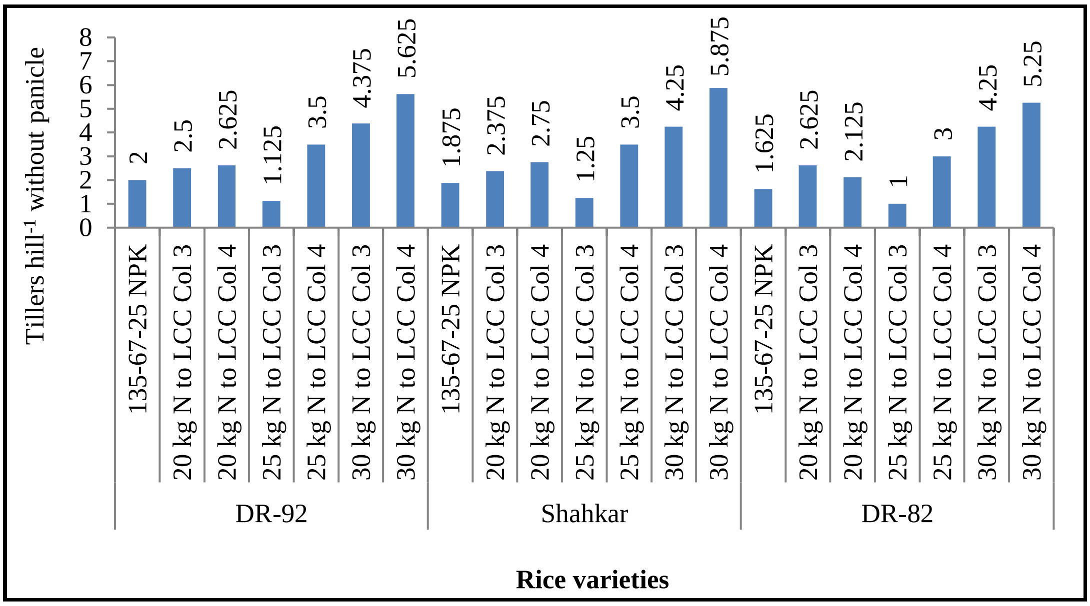
Figure. 3. Interactive effect of varieties × treatments under LCC on panicles m^{-2}

The results further revealed that the maximum number of tillers hill^{-1} with panicle (18.13 and 18.75) were recorded in variety Shahkar × 25 N kg ha^{-1} to column 3 of LCC and DR-82 × 135-67-25 NPK kg ha^{-1}, optimum number of tillers hill^{-1} with panicle (18.38) was observed in variety DR-92 × 25 kg N ha^{-1} to column 3 of LCC and minimum number of tillers hill^{-1} with panicle (14.75) was found in variety DR-92 × 20 N kg ha^{-1} to column 3 of LCC (. 4). More number of tillers hill^{-1} without panicle (5.625 and 5.875) were found in the interaction of variety DR-92 × 30 kg N ha^{-1} to column 4 of LCC (Figure . 5). However, the least number of tillers hill^{-1} without panicle was recorded in the interaction of variety DR-82× 25 kg N ha^{-1} to column 3 of leaf color chart (LCC). Likewise, the maximum number of filled grains panicle^{-1} (131.9) was recorded in rice variety Shahkar × 25 kg N ha^{-1} to column 3 of LCC; while the minimum number of filled grains panicle^{-1} (101.6) were in rice variety DR-82 × 20 kg N ha^{-1} to column 3 of LCC (Figure . 6).



S.E.±0.4515 LSD 0.05 1.264

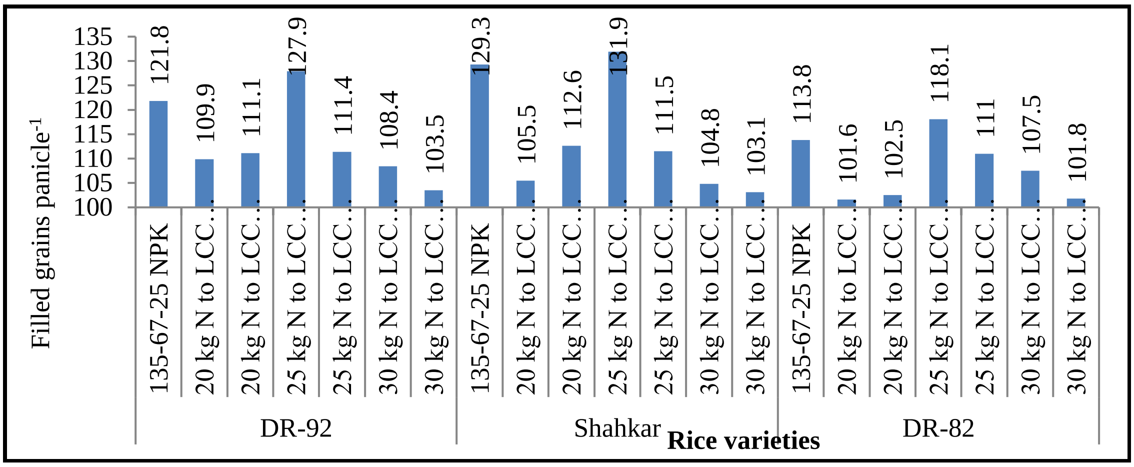
Figure. 4. Interactive effect of varieties × treatments under LCC on tillers hill^{-1} with panicle



S.E.± 0.3380 LSD 0.05 0.9462

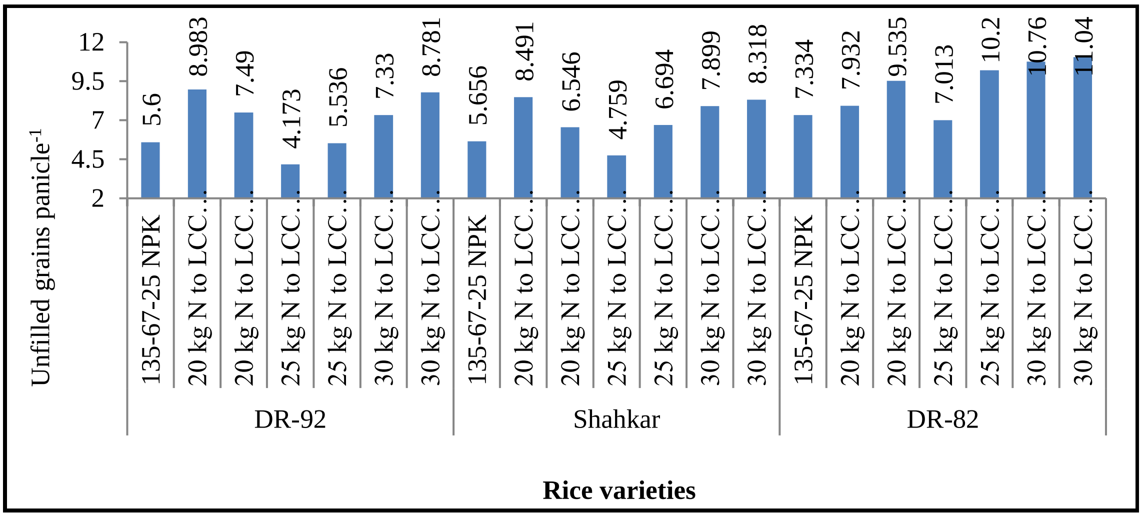
Figure.5. Interactive effect of varieties × treatments under LCC on tillers hill^{-1} without panicle

The study further showed that the maximum number of unfilled grains panicle^{-1} (11.04) was observed in interactive of variety DR-82 × 30 kg N ha^{-1} to columns 4 of LCC; while the minimum number of unfilled grains panicle^{-1} (4.173) was recorded in case of variety DR-92 × 25 kg N ha^{-1} to column 4 of LCC, respectively (Figure. 7). Higher sterility (9.68%) was observed under the interactive effect of variety DR-82 × 30 kg N ha^{-1} to column 4 of LCC; while the optimum sterility percentage (9.151) was observed in variety DR-82 × 30 kg N ha^{-1} to column 3 of LCC and the minimum sterility value of 3.278 % was found in the interaction of vafriety DR-92 × 25 kg N ha^{-1} to column 3 of LCC (Figure . 8). Similarly, maximum seed index (25.15 g) was observed in the interaction of variety Shahkar × 25 kg N ha^{-1} to column 3 of LCC; while the lesser seed index (22.60 and 22.59g) was observed in the interaction of DR-82 × 135-67-25 NPK kg ha^{-1} and 20 kg N ha^{-1} to column 30 of LCC, respectively (Figure. 9).



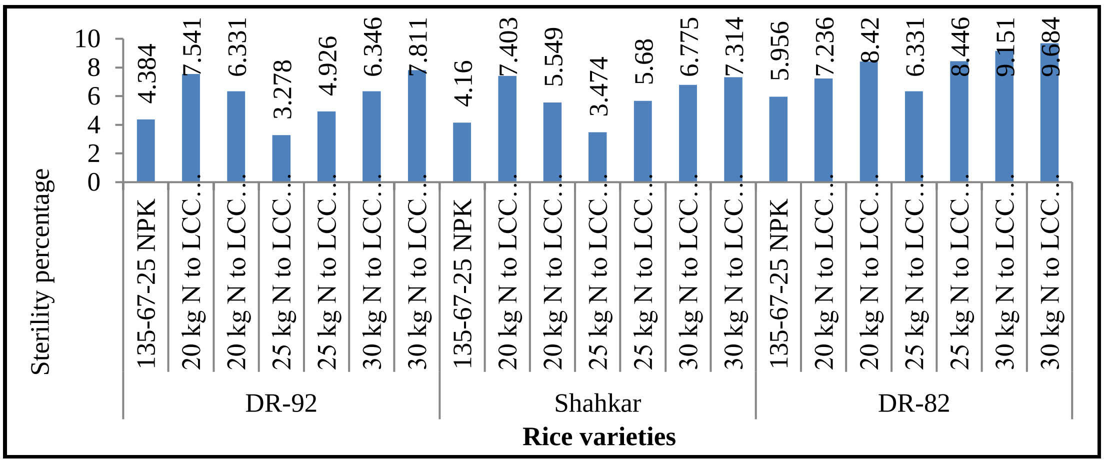
S.E.±0.9736 LSD 0.05 2.725

Figure. 6. Interactive effect of varieties × treatments under LCC on filled grains panicle^{-1}



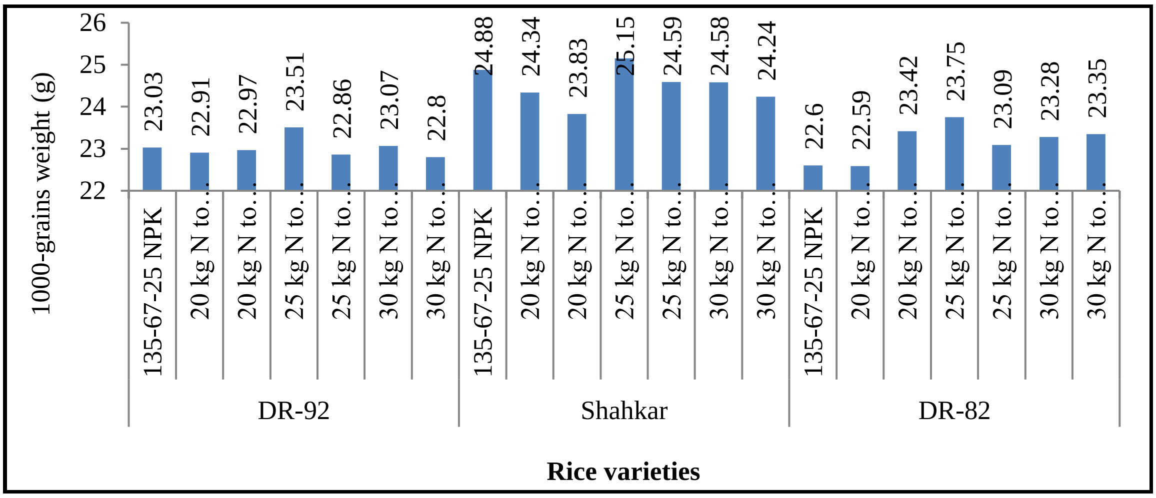
S.E.± 0.4366 LSD 0.05 1.222

Figure. 7. Interactive effect of varieties × treatments under LCC on unfilled grains panicle^{-1}



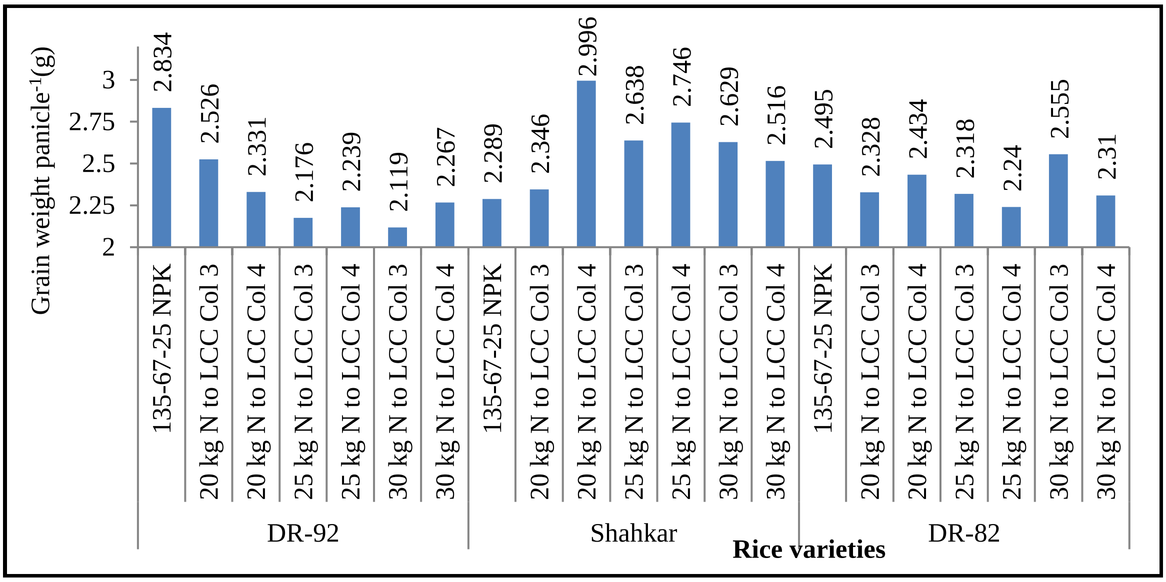
S.E.±0.3539 LSD 0.05 0.9907

Figure. 8. Interactive effect of varieties × treatments under LCC on sterility percentage



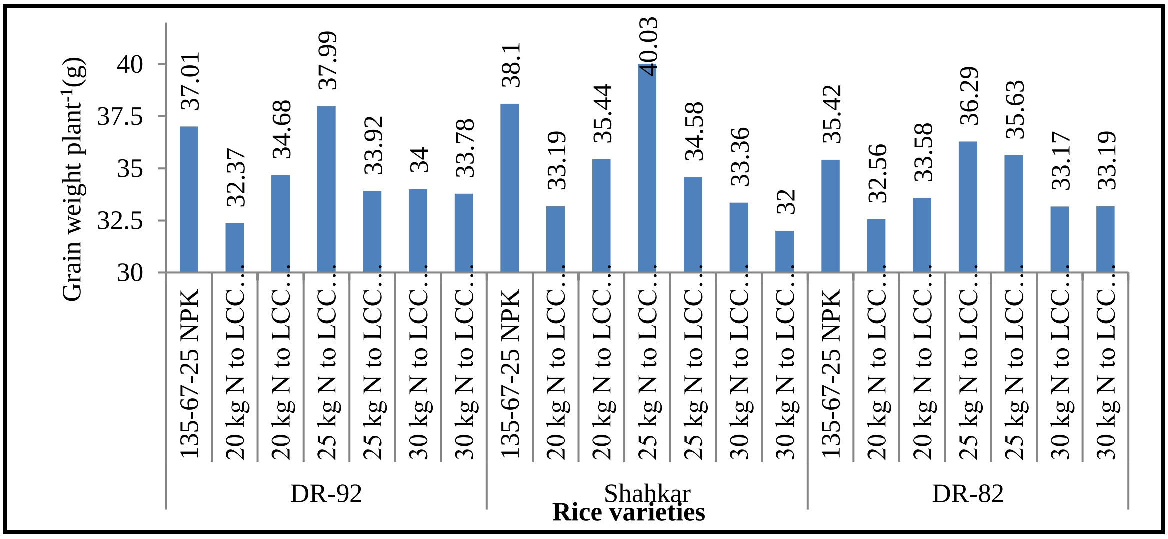
S.E.± 0.1940 LSD 0.05 0.5430

Figure. 9. Interactive effect of varieties × treatments under LCC on 1000-grains weight



S.E.± 0.06982 LSD 0.05 0.1955

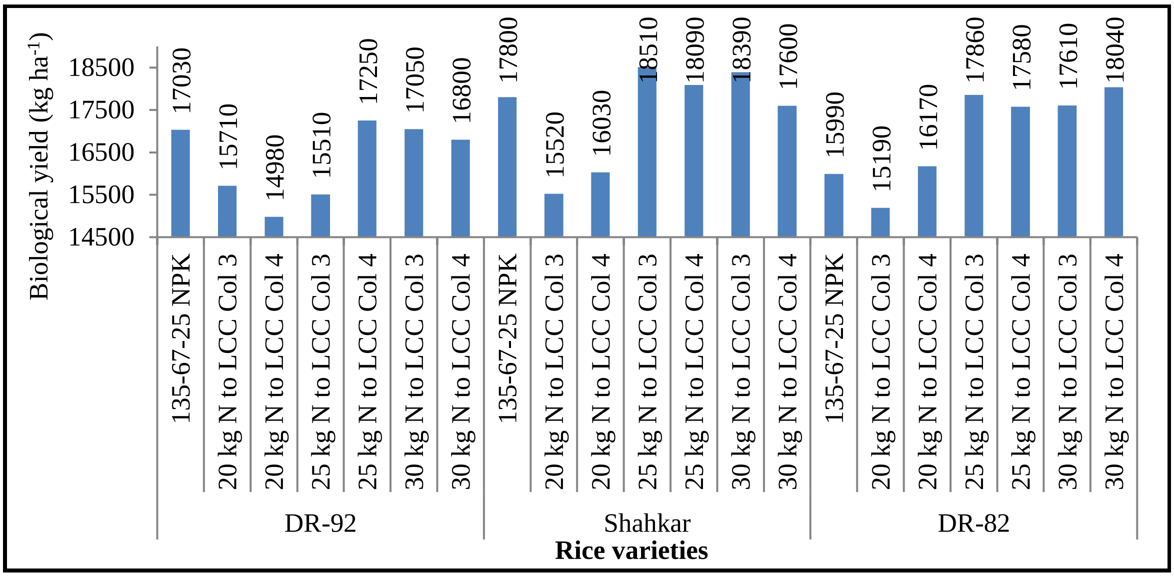
Figure. 10. Interactive effect of varieties × treatments under LCC on grain weight panicle^{-1} (g)



S.E.± 0.6340 LSD 0.05 1.775

Figure. 11. Interactive effect of varieties × treatments under LCC on grain weight plant^{-1} (g)

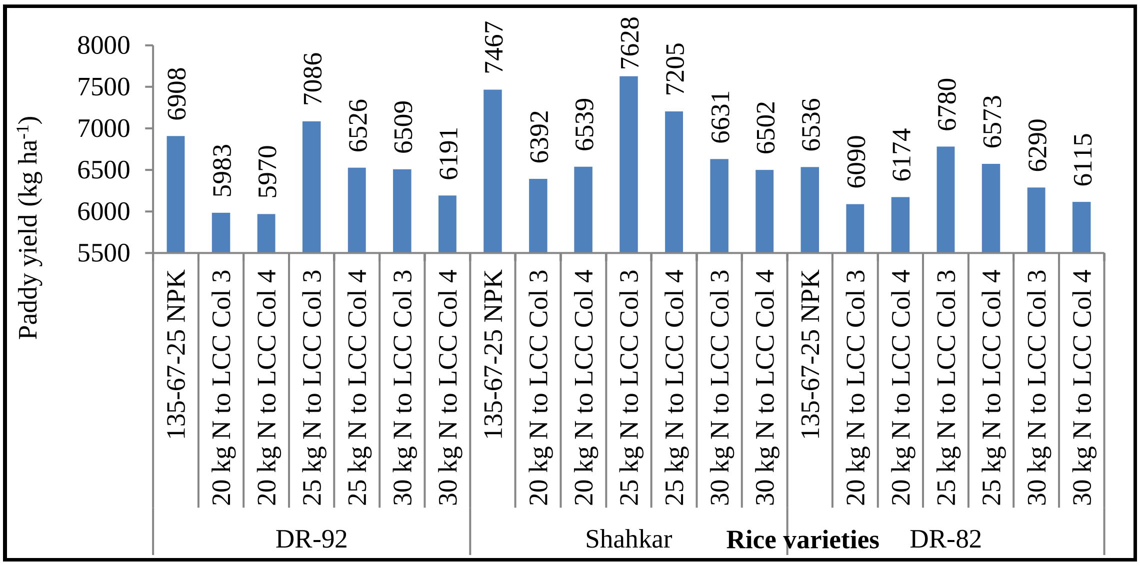
It is apparent from the experimental results that the maximum grain weight panicle^{-1} (2.996g) was recorded under the interactive effect of Shahkar × 20 kg N ha^{-1} to column 4 of LCC; while the least grain weight (2.119g) panicle^{-1} was observed under the interactive effect of variety DR-92 × 30 kg N ha^{-1} to column 3 of LCC (Figure. 10). Moreover, the highest grain weight plant^{-1} (40.03 g) was observed in the interaction of variety Shahkar × 25 kg N ha^{-1} to column 3 of LCC; while the lower values (32.37 and 32.00g) for this trait were recorded in the interaction of variety DR-92 × 20 kg ha^{-1} N ha^{-1} to column 3 of LCC and variety Shahkar × 30 kg N ha^{-1} to column 4 of LCC, respectively (Figure. 11). Similarly, the highest biological yield (18510 and 18390 kg ha^{-1}) was recorded in the interaction of variety Shahkar × 25 kg N ha^{-1} to column 3 of LCC and 30 kg N ha^{-1} to column 3 of LCC (Figure. 12).



S.E.± 452.3 LSD 0.05 1616.0

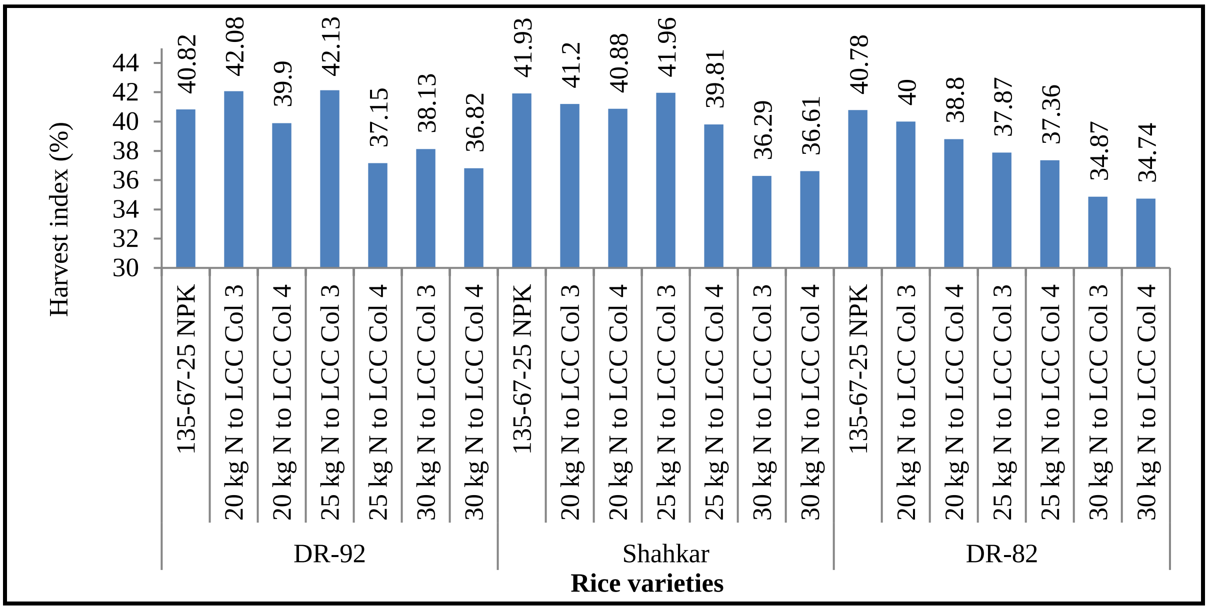
Figure. 12. Interactive effect of varieties × treatments under LCC on biological yield (kg ha^{-1})

In case of interactive effect on paddy yield, it was maximum (74.67 and 7628 kg ha^{-1}) under interaction of variety Shahkar × 135-67-25 NPK kg ha^{-1} and Shahkar × 25 kg N ha^{-1} to column 3 of LCC, respectively; while the minimum paddy yield (5970 kg ha^{-1}) was recorded in the interaction of variety DR-92 × 20 kg N ha^{-1} to column 3 of LCC (Figure.. 13). Similarly, the greater harvest index (42.08 and 42.13 unit) was recorded under the interaction of variety DR-92 × 20 kg N ha^{-1} to column 3 of LCC and 25 kg N ha^{-1} to column 3 of LCC, respectively; while the least harvest index (34.74%) was recorded in the interaction of variety DR-82 × 30 kg N ha^{-1} to column 4 of LCC (Figure. 14). Moreover, the straw yield (11580 kg ha^{-1}) was observed under interaction of Shahkar × 30 kg N ha^{-1} to column 3 of LCC; while the lowest straw yield (7446 kg ha^{-1}) was recorded in the interaction of DR-92 × 20 kg N ha^{-1} to column 3 of LCC (Figure. 15).



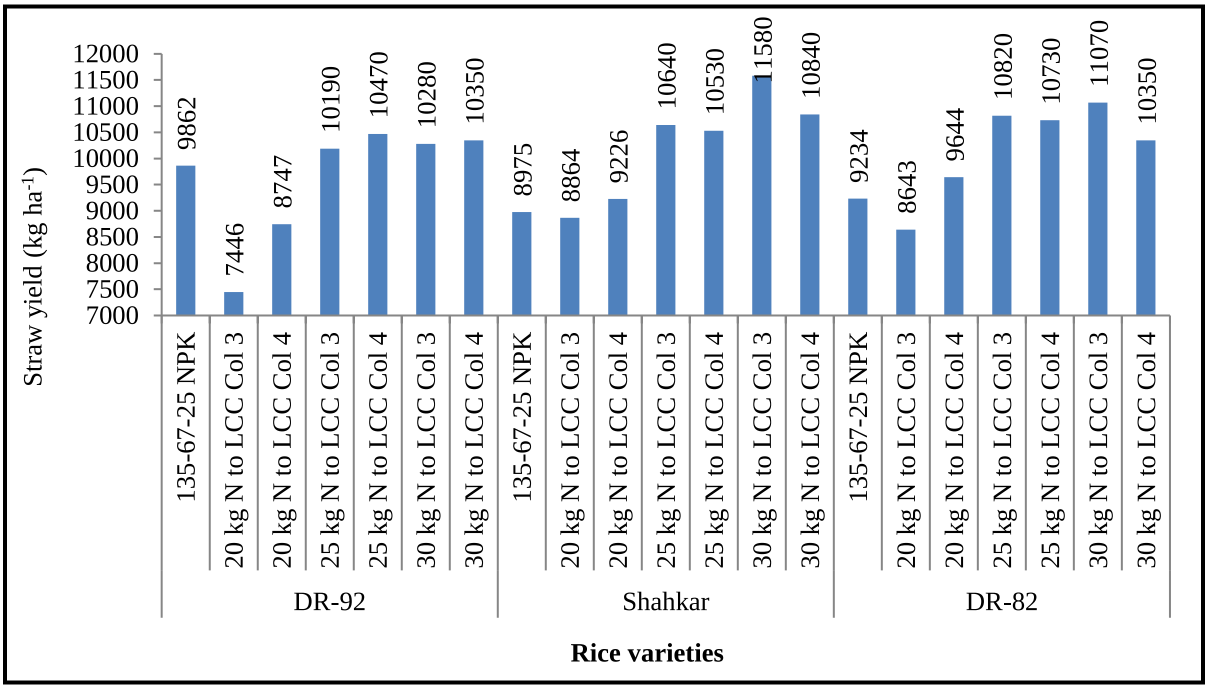
S.E.±79.46 LSD 0.05 222.4

Figure. 13. Interactive effect of varieties × treatments under LCC on paddy yield (kg ha^{-1})



S.E.±0.6190 LSD 0.05 1.733

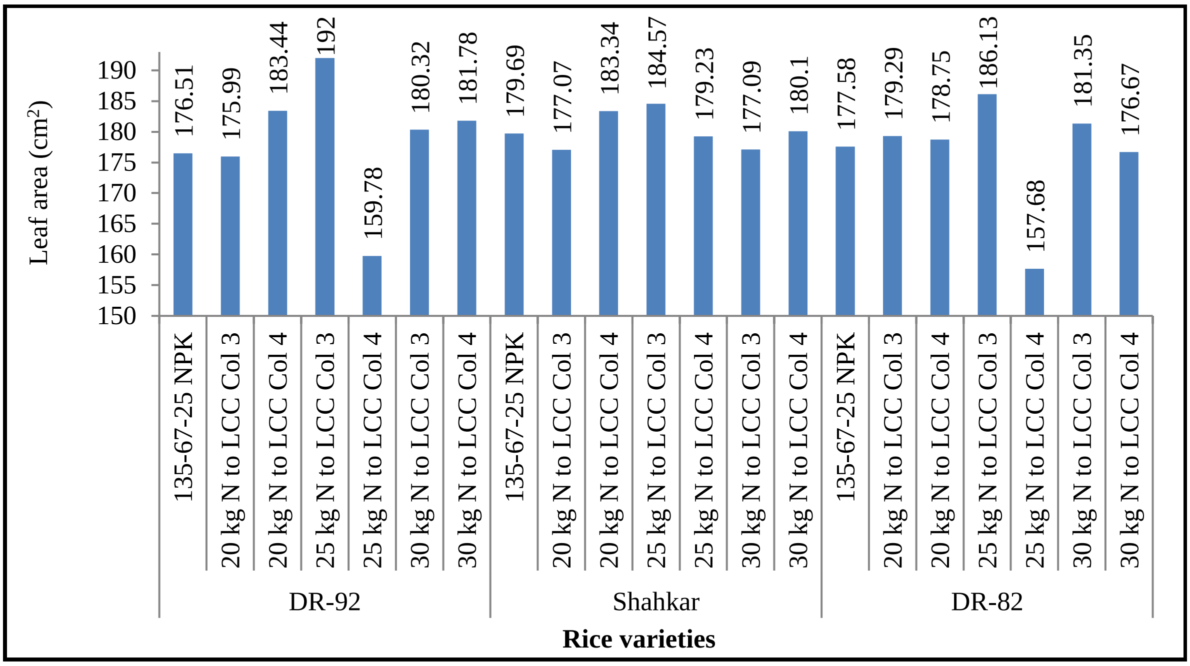
Figure. 14. Interactive effect of varieties × treatments under LCC on Harvest index (%)



S.E.±403.9 LSD 0.05 1131.0

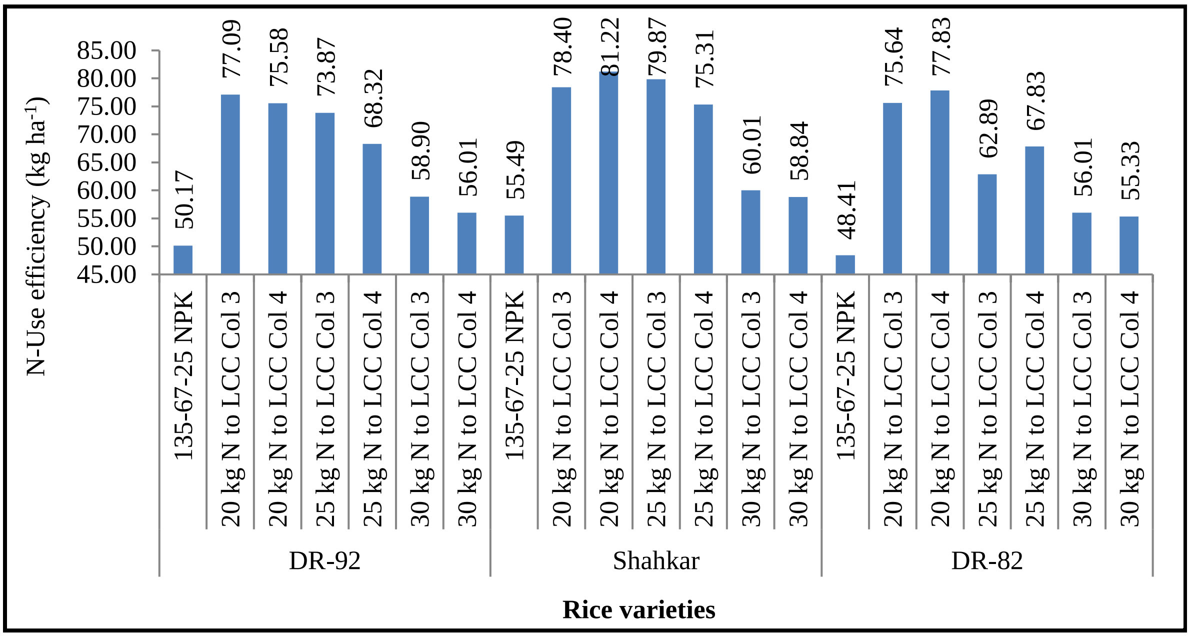
Figure. 15. Interactive effect of varieties × treatments under LCC on Straw yield (kg ha^{-1})

It is evident from the results as illustrated by Figure. 16, the maximum leaf area (192.20, 184.560 and 186.134) was recorded under the interaction of variety DR-92 × 25 kg N ha^{-1} to column 3 of LCC, Shahkar × 25 kg N ha^{-1} to column 3 of LCC and variety DR-82 × 25 kg N ha^{-1} to column 3 of LCC, respectively. However, the least leaf area (157.68 cm^2) was recorded in the interaction of variety DR-82 × 25 kg N ha^{-1} to column 4 of LCC (Figure. 16). Similarly, the higher nitrogen uptake efficiency (81.220 and 79.874 kg ha^{-1}) was observed under the interaction of variety Shahkar × 20 kg N ha^{-1} to column 4 and 25 kg N ha^{-1} to column 3 of LCC, respectively (Figure. 17).



S.E.± 7.1038 LSD 0.05 16.234

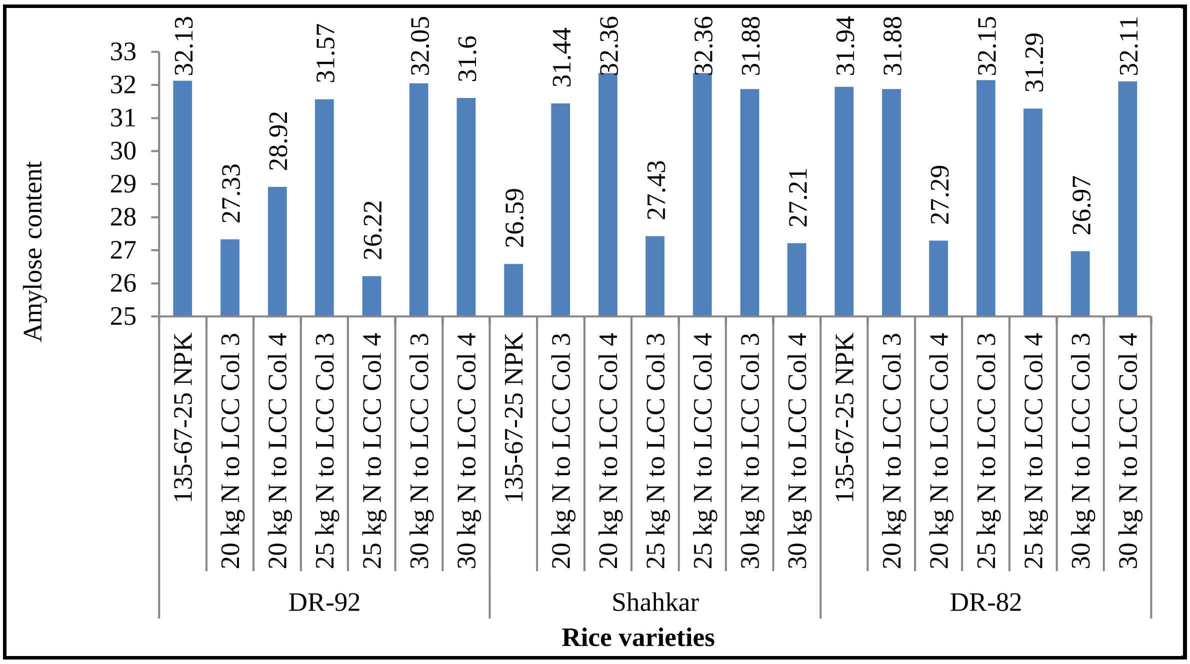
Figure. 16. Interactive effect of varieties × treatments under LCC on leaf area (cm^2)



S.E.±1.8139 LSD 0.05 5.432

Figure. 17. Interactive effect of varieties × treatments under LCC on N-Use efficiency (kg ha^{-1})

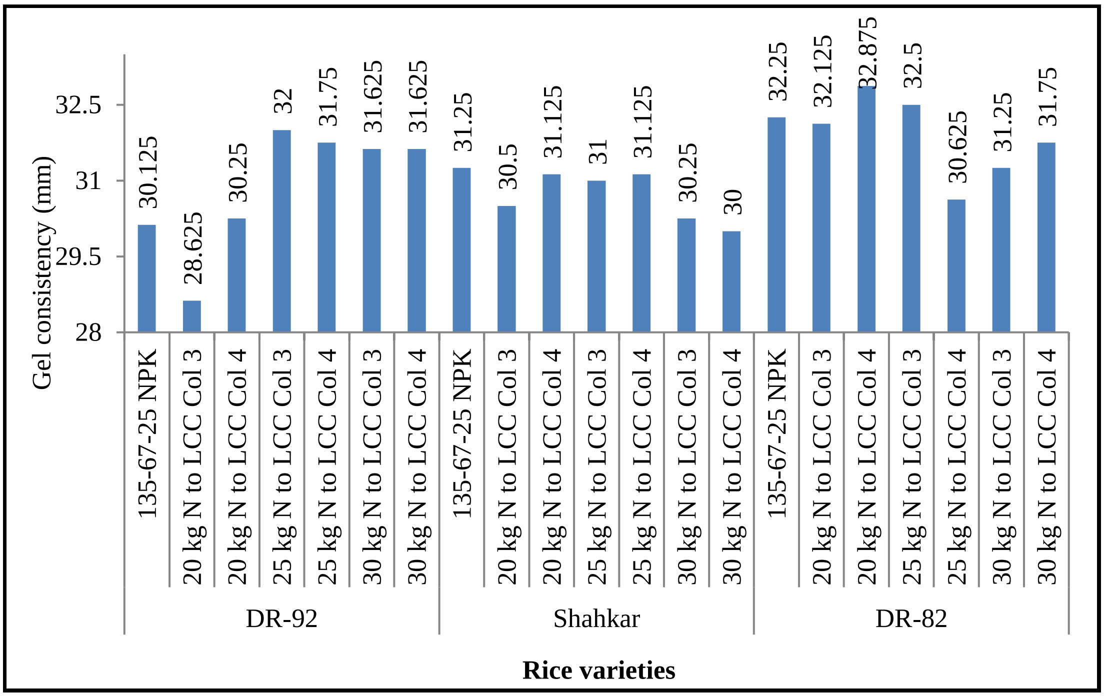
The data in relation to amylose content showed that the amylose content in average (32.13 31.57, 31.60 31.44, 32.36, 32.36, 31.88, 31.94, 31.88 32.15, 31.29 and 32.11) was recorded in the interaction of variety Shahkar × recommended N rate of 135-67-25 NPK kg ha^{-1}, 25 and 30 kg N ha^{-1} to column 3 and 4 of LCC, 25 kg N ha^{-1} to column 4 of LCC and 30 kg N ha^{-1} to column 3 of LCC, respectively. In variety DR-92 the maximum amylase content under the interaction of 135-67-25 kg N ha^{-1} × 20 and 25 kg N ha^{-1} to column 3 and 4 of LCC and 30 kg N ha^{-1} to column 3 of LCC × variety DR-92, respectively (Figure. 18).



S.E.±0.3760 LSD 0.05 1.053

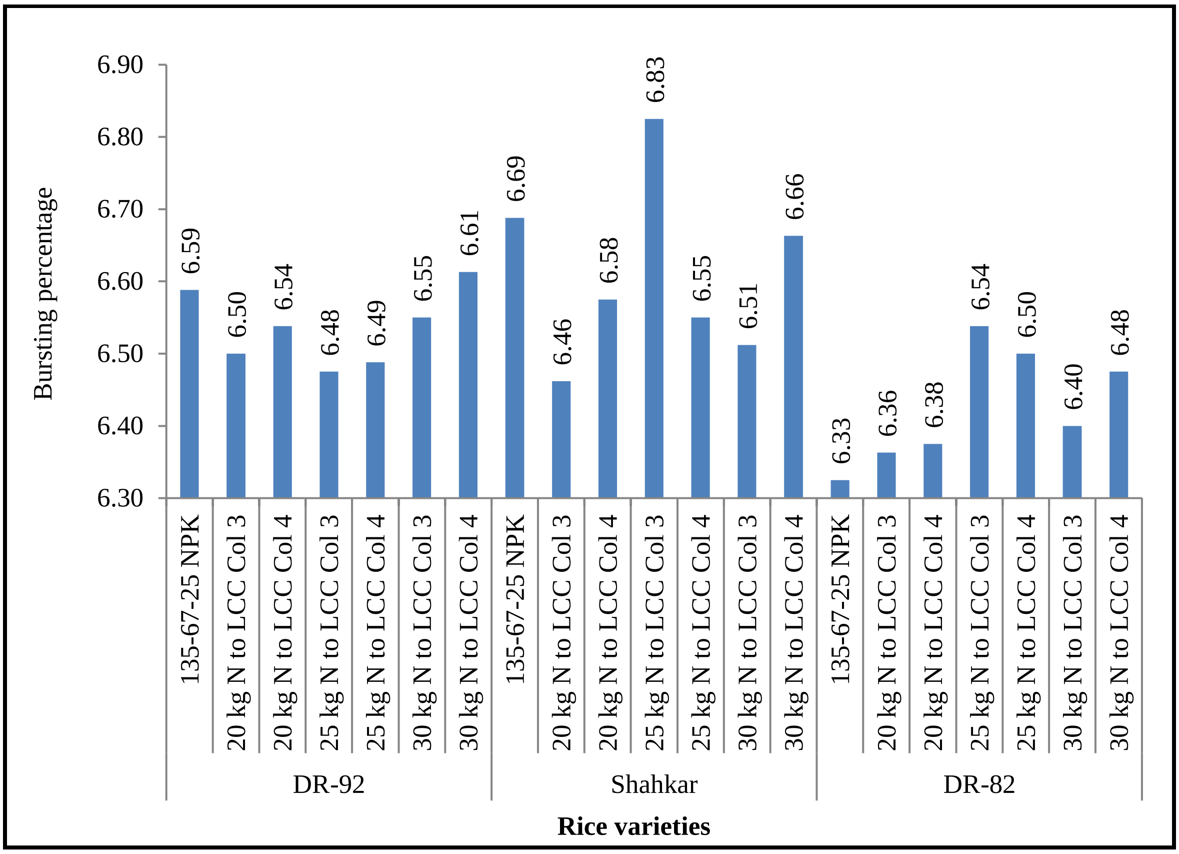
Figure. 18. Interactive effect of varieties × treatments under LCC on Amylose content

Similarly, the Gel consistency ratio was highest (33.875 mm) in the treatment interaction of rice variety DR-82 × 20 kg N ha^{-1} to column 4 of LCC; while the minimum gel consistency (28.625 mm) was recorded in the interaction of rice variety DR-92 × 20 kg N ha^{-1} to column 3 of LCC (Figure. 19). The bursting percentage was highest (6.83%) in the interaction of variety Shahkar × 25 kg N ha^{-1} to column 3 of LCC; while minimum (6.33 %) bursting percentage was noted in the interaction of variety DR-82 × recommended NPK rate of 135-67-25 kg N ha^{-1} (Figure. 20).



S.E.± 1.0660 LSD 0.05 4.132

Figure. 19. Interactive effect of varieties × treatments under LCC on gel consistency (mm)



S.E.±0.0755 LSD 0.05 ns

Figure. 20. Interactive effect of varieties × treatments under LCC on bursting percentage

Discussion

The LCC is inexpensive and a simple technology easy to understand and use. The color panels of the LCC are designed to indicate whether rice plants are hungry or over-fed by N fertilizer. By matching the color of the rice leaf to the color on the LCC, farmers can decide proper time and amount of N fertilizer for application. In this study the impact of N availability under LCC technique on growth, productivity and quality of non-aromatic rice varieties was evaluated; and the major findings are discussed to develop understanding on appropriate N requirements for sustainable higher rice yields. The plant N in rice is increased when yield is increased; which lead to assume that timely and more nutrients are needed for per unit biological and grain yield increase. Another way to improve N availability is to improve uptake efficiency of externally applied macro and micro nutrients; because the soils of Pakistan are not able to produce such level of yields that may ensure food security; hence, external application of inputs is essential (Khaliq *et al.,* 2005; Rehman *et al.,* 2020).

The study underscored the significant impact of nitrogen (N) application guided by Leaf Color Chart (LCC) technology on various growth and yield parameters of non-aromatic rice varieties. It was found that applying 30 kg N at LCC columns 3 and 4 enhanced plant height, corroborating findings from recent studies (Alam *et al.,* 2020; Rehman *et al.,* 2022; Singh *et al.,* 2020).

Panicle length notably increased with recommended doses of NPK (135-67-25 kg ha^-1) and 25 kg N at LCC column 3, consistent with current literature on nutrient management in rice (Farooq *et al.,* 2020; Fageria *et al.,* 2019; Singh *et al.,* 2021). Conversely, applying 30 kg N ha^-1 led to greater tillering without panicle development at LCC column 4, a phenomenon reported in previous research (Hussain *et al.,* 2021; Balasubramanian *et al.,* 2019).

Maximum biological yield was achieved at 25 and 30 kg N ha^-1 at LCC columns 4 and 3, aligning with studies emphasizing optimal N application rates (Alam *et al.,* 2020; Singh *et al.,* 2021). Paddy yield showed a significant increase with increasing N fertility, peaking under 25 N ha^-1 at LCC column 3, consistent with extensive literature on rice nutrition (Singh *et al.,* 2006; Hussain *et al.,* 2020).

Enhanced harvest index was observed at the recommended NPK dose, consistent with studies emphasizing balanced nutrient management (Farooq *et al.,* 2018; Maiti and Das, 2006). Straw yield peaked under 25 kg N ha^-1 at LCC columns 3 and 4, while leaf area was highest at 20 and 25 kg N ha^-1 at LCC columns 3 and 4, in line with studies on crop physiology and nutrient uptake (Rehman *et al.,* 2020; Singh *et al.,* 2021).

The study further indicated that N uptake efficiency was highest at 20 kg N ha^-1 at LCC columns 3 and 4, reflecting efficient nutrient utilization strategies (Hussain *et al.,* 2021; Fageria *et al.,* 2019). Quality traits of rice, such as amylose content, were influenced by N levels, with the highest amylose content observed at 25 kg N ha^-1 at LCC column 3, consistent with previous findings (Balasubramanian *et al.,* 2019; Singh *et al.,* 2021). Gel consistency was optimal under 20 kg N ha^-1 at LCC columns 3 and 4, while bursting percentage remained unaffected, as reported in similar studies (Hussain *et al.,* 2021; Singh *et al.,* 2020).

The interactive effects of varieties and N levels across LCC columns underscored a strong connection, as evidenced by treatment interactions (Rehman *et al.,* 2022; Alam *et al.,* 2020). Globally, studies have demonstrated that efficient N management practices, such as those facilitated by LCC, enable substantial N savings without compromising grain yield, thereby promoting widespread adoption among farmers (Singh *et al.,* 2021; Balasubramanian *et al.,* 2019).

Increasing the nitrogen (N) feeding to the crop resulted in higher paddy yields, particularly noted in the interaction of variety Shahkar with the recommended NPK dose of 135-67-25 kg ha-1. This combination also maximized the harvest index, influenced by N application as observed through the Leaf Color Chart (LCC) columns. The highest harvest index (HI) was recorded in the interaction of variety DR-92 with 20 and 25 kg N doses, observed in columns 3-4 of the LCC. However, the amylase content was found to be non-significant across all interactions and varieties. Other quality characteristics of rice, such as gel consistency and bursting percentage, were also found to be non-significant among different varieties and N applications to columns (Yogendra *et al.,* 2017; Singh *et al.,* 2006). Yogendra et al. (2017) demonstrated a significant increase in the grain yield of aerobic rice compared to the control by varying the amount of N applied from 90 kg ha-1 (Urea at 30 kg ha-1 as the basal, LCC-3) to 60 kg ha-1 (No basal, LCC-3). Despite the higher grain yield (5274 kg ha-1) in the first year with 90 kg N ha-1, the variation in N application still resulted in increased yield (Chen *et al.,* 2011). Singh et al. (2006) determined the optimal timing for N treatment in direct-seeded rice using a chlorophyll meter (SPAD meter) alongside an LCC. They found that only 70 kg N ha-1 was required to maintain the color intensity of the uppermost fully opened rice leaves at 90 percent or more of the color intensity in the over-fertilized reference plot. This represents a significant reduction in N fertilizer usage, as there was no yield difference between fields treated with 80 or 120 kg N ha-1 (Peng *et al.,* 1996; Dobermann and Fairhurst, 2000). According to Maiti and Das (2006), using SPAD and LCC can save up to 20 and 25 kg N ha-1, respectively, without reducing rice yields compared to treatments using 100 kg N ha-1. They recommend applying a basal dose of fifty percent of the recommended 100 kg N per hectare before planting rice. Given that rice seeds take five to seven days to germinate and two to three leaf seedlings to appear in fields, plants may not immediately utilize all the basal N provided. If more nutrients were available during various growth stages, grain digestion of photosynthates would be significantly enhanced (Witt *et al.,* 1999). Farooq et al. (2018) highlighted the importance of pre-anthesis assimilate accumulation and translocation due to high plant density. The positive impact on yield components led to increased grain yield, potentially due to the enhanced ability of the plant to absorb more N at various growth stages. The non-aromatic varieties DR-92 and DR-82 showed improved quality characteristics, such as amylase percentage and gel consistency, without significant variation due to N application (Kumar and Ladha, 2011).

Among the varieties studied, Shahkar and DR-82 proved superior in various agronomic, physiological, and nutritional traits compared to other non-aromatic varieties. The Leaf Color Chart (LCC)-based approach to nitrogen (N) management resulted in significant N savings while maintaining grain yield, irrespective of the crop's response to the applied N. The efficiency with which a crop absorbs N from the soil and uses it to produce grain is referred to as the crop's N use efficiency. This encompasses both N uptake efficiency and N utilization efficiency. Enhancing these efficiencies can lead to more effective N use, which is often highest when N input is minimized. N is accumulated during the vegetative growth stage of crops and is remobilized after flowering to be transferred to the grain. During grain filling, the N stored in the leaves before flowering is largely remobilized and contributes to the deposition of N-containing grain proteins (Hussain *et al.,* 2000; Bijay-Singh *et al.,* 2002; Singh *et al.,* 2021). Improving N recovery can be achieved by utilizing more N-efficient genotypes and optimizing fertilization rates, methods, and timing (Fageria *et al.,* 2009). In general, rice varieties selectively bred for alternating wetting and drying periods are used in the cultivation of aerobic rice under puddled planting conditions. For aerobic rice to achieve its full potential and for appropriate N management practices to be implemented, additional rice varieties and better-suited production technologies are needed.

Conclusions

The N levels corresponding to LCC columns significantly affected various traits. Higher plant height and biological yield were observed at 30 kg N ha^{-1} (LCC columns 3 and 4) and 25 kg N ha^{-1} (LCC column 4). Paddy yield was highest at 25 kg N ha^{-1} (LCC column 3), while N uptake efficiency was greatest at 20 kg N ha^{-1} (LCC columns 3 and 4). The highest amylase content and gel consistency were noted with 25 kg N ha^{-1} (LCC columns 3 and 4). Varieties Shahkar and DR-92 exhibited superior yield and nutrient uptake under specific N applications, with timely irrigation enhancing growth and quality.

Authors' Contribution

RAT: Study conception and design, Data Collection and analysis, Writing Original Draft, AWG: Study conception and design, Supervision, Data Analysis, Writing - Review & Editing. ST Study conception and design, Supervision, Data Interpretation, Review & Editing. NM, Conceptualization, Supervision, Data Interpretation, Review and Editing.

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Conflict of Interest

The authors declare no conflict of interest.

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