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Nitrogen Effects on Yield, Quality and Physiological Characteristics of Giant Rice

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Abstract: In China, the quantity of nitrogen fertilizer applied is large, but as a consequence of a high level of loss, its utilization rate is low. Compared to common rice, the new giant rice has interesting characteristics, namely high biological yield and good efficient use of fertilizer. However, it becomes urgent to further consider the appropriate rate of nitrogen fertilizer to be applied. The giant rice varieties Feng5 and Feng6 were set up in a pot experiment and a field experiment under five doses of nitrogen fertilizers, namely, 0 kg·ha^{-1} (CK), 75 kg·ha^{-1} (T1), 150 kg·ha^{-1} (T2), 225 kg·ha^{-1} (T3) and 300 kg·ha^{-1} (T4). Parameters such as leaf area index (LAI), lodging index (LI), nitrogen utilization rate, photosynthesis rate and grain yield were measured. The results showed that with the increase of nitrogen dose in a certain range, LAI, plant height, the number of tillers, net photosynthetic rate (NPn), the transpiration rate (Tr), and the grain yield increased while the lodging index (LI), the nitrogen agronomic utilization rate (AE) and nitrogen partial productivity (PFPN) decreased. Additionally, with the increase of nitrogen application, the grain yield index (HI) and nitrogen contribution rate (FCRN) of rice presented a parabolic trend.

Keywords: nitrogen fertilizer; production; quality; physiology; nitrogen efficiency

1. Introduction

Nitrogen fertilizer plays an important role in agricultural production and has the strongest effect on increasing agricultural production and income. As the cereal crop with the highest yield per unit area, rice occupies 20% of the global seeded area in China and has reached the largest in the world [1]. The use of nitrogen fertilizer accounted for 37% of the global fertilizer used in rice, and the average nitrogen fertilizer application rate in paddy fields was about 60% higher than the global level [2]. The application of nitrogen in rice in China has prominent problems such as a large amount of nitrogen fertilizer, low utilization rate and high wastage rate. Excessive application of nitrogen fertilizer aggravates soil degradation and environmental pollution [3–5]. Therefore, it is particularly necessary to explore the appropriate application amount of nitrogen fertilizer, which can not only reduce the loss of nitrogen fertilizer but also reduce the pollution of soil and the environment by nitrogen fertilizer. Nitrogen is a necessary nutrient element for plants, which is very important for photosynthesis, growth and development, yield, quality and biomass of rice. It is a component of amino acid in protein and an important component of chlorophyll in photosynthesis, and it exists in various organs of plants. There have been many studies on the effect of nitrogen fertilizer on rice.

A large number of studies have shown that rice yield increases with the increase of nitrogen application within a certain range, but the yield and nitrogen utilization rates also decrease when the nitrogen application is too high. The yield difference is mainly caused by the difference in seed setting rate and effective panicle number [6]. The formation of yield is closely related to photosynthesis. Studies have shown that increasing nitrogen fertilizer within a certain range can improve the net photosynthetic rate, while excessive nitrogen fertilizer will lead to a decline in photosynthetic capacity. The canopy population of leaves, as the main organ of photosynthesis, is affected by nitrogen. Meng and Du [7] showed that an appropriate nitrogen application rate could ensure that the rice canopy population reached a higher LAI. In addition, the formation of rice quality is also closely related to nitrogen. Bai [8] and Li et al. [9] research has shown that increasing nitrogen can help improve the nutritional quality and processing quality of rice, but the excessive nitrogen fertilization can increase rice chalkiness and rice appearance quality and cooking and eating quality becomes worse, with the increase of nitrogen fertilization rate of brown rice rate, milled rice rate, the head rice rate, high viscosity and breakdown values rise. At present, there is no unified nitrogen utilization efficiency evaluation system in rice in China and abroad, and there are many factors affecting nitrogen utilization. For this purpose, Lv et al. [10] and Li et al. [11] took the compact rice variety *Shennong 07425* and the loose rice variety *Qiu Guang* as the test materials to explore the differences in nitrogen metabolism between the different types of rice. The results showed that more nitrogen application would affect the growth of the plant. Also, less nitrogen will also affect the rice plants leaving them under nitrogen stress. In addition, their results showed that the efficiency of nitrogen use was related to varieties of rice. Hu et al. [12] Studies showed that Indica rice usually had higher nitrogen use efficiency than japonica rice. Ju [13] Using two nitrogen-sensitive and high-yield varieties *Huai Rice 5* and *Lianjing 7* and two nitrogen-insensitive varieties *Ningjing 1* and *Yangjing 4038* as materials, he found that nitrogen-sensitive varieties had higher nitrogen photosynthetic efficiency and nitrogen utilization rate. Li et al. [14] from the perspective of genes, it was found that a high expression level of *GRF4* could promote nitrogen uptake in plant roots, enhance leaf photosynthesis, promote the accumulation of dry matter in grout and endosperm, and thus increase crop yield. Kurai et al. [15] studies have shown that *Dof1* transcription factors are related to nitrogen utilization efficiency. There have been many previous studies on the effect of nitrogen on rice yield, quality and nitrogen utilization rate, but few reports on the effect of nitrogen on giant rice compared with common rice. The new variety of giant rice has the characteristics of a tall plant, high biological yield and requiring a large amount of fertilizer, so it is necessary to determine the optimal nitrogen application amount of giant rice. The purpose of this study was to explore the appropriate application amount of nitrogen fertilizer in giant rice so as to avoid unnecessary waste of nitrogen fertilizer and pollution of soil environment caused by excessive application of nitrogen, which was of great significance for large-scale popularization and specialized production of giant rice in South China.

2. Materials and Methods

2.1. Test Background Overview

This study was carried out at the farm of the South China Agricultural University (23°16′ N, 113°36′ E) in the field at the end of the crop season 2018 and in pots at the end of the season 2019. This area of China is typically recognized for the double rice cultivation system in which rice is grown and harvested twice a year in the same paddy field. The sandy loam soil of the field was tested, and the soil organic matter, total nitrogen, total phosphorus, available nitrogen, available phosphate, pH [16]. The results are shown in Table 1. Rainfall and temperature during the study are shown in Figure 1.

Table 1. Physical and chemical properties of the experiment soil.

Year	Organic Content ($g\ kg^{-1}$)	Total N ($g\ kg^{-1}$)	Total P ($g\ kg^{-1}$)	Total K ($g\ kg^{-1}$)	Available N ($mg\ kg^{-1}$)	Available P ($mg\ kg^{-1}$)	pH
2018	22.87	3.2	14.50	17.56	370.42	35.20	5.70
2019	18.82	2.97	12.23	19.60	352.21	30.22	5.64

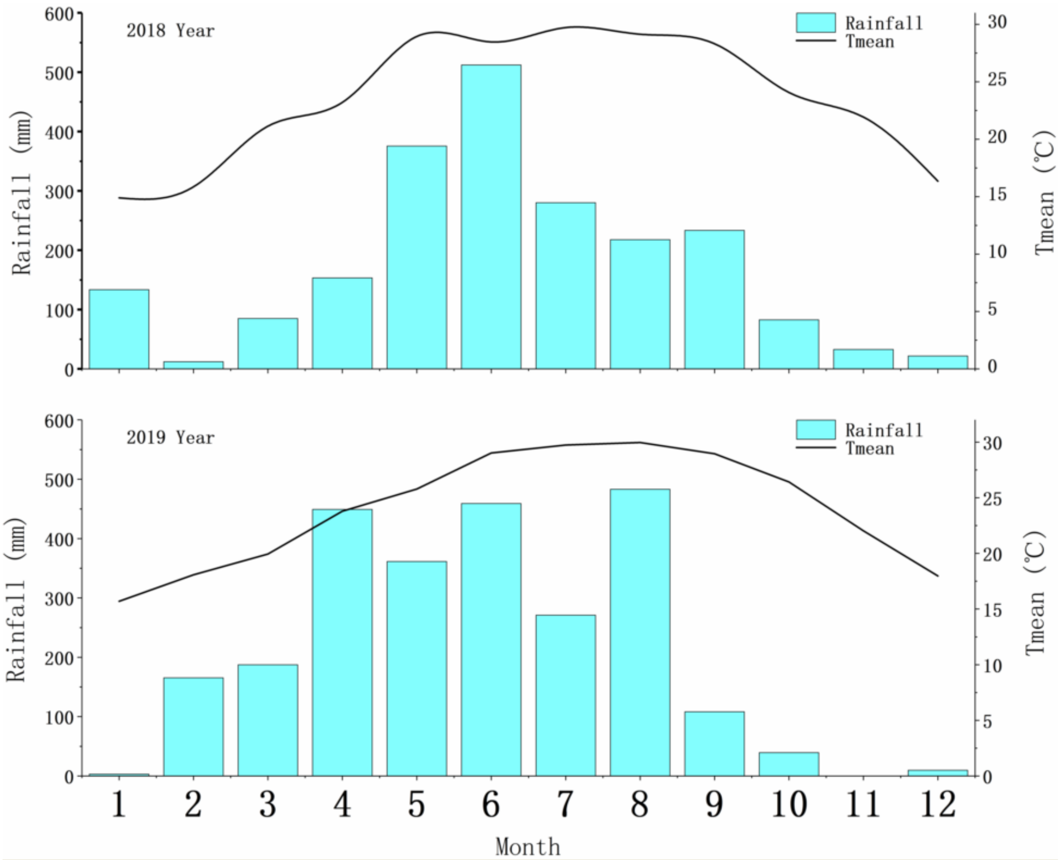


Figure 1. Temperature and rainfall during the experiment.

2.2. Experimental Design

In this experiment, five nitrogen level gradients were set. Two varieties, namely Feng5 (growth period 127 days) and Feng6 (growth period 117 days), were taken at each growth stage of rice to determine its yield quality, nitrogen utilization rate, leaf area index, lodging index, photosynthesis rate, plant height, rice tillers. A randomized block design with three replicates was used. The previous crop was rice. Nitrogen was applied as urea (CON_2H_4, total nitrogen ≥ 46%).

In the field experiment, the kinds of rice were first sown on July 18th and transplanted by machine on July 30th with an interrow distance of 30 cm at approximately 15 cm between the seedlings. Each plot area was 32.3 m^2, per hole was 2~3 seedlings. Five treatments of nitrogen including 0 $kg{\cdot}ha^{-1}$ (CK), 75 $kg{\cdot}ha^{-1}$ (T1), 150 $kg{\cdot}ha^{-1}$ (T2), 225 $kg{\cdot}ha^{-1}$ (T3) and 300 $kg{\cdot}ha^{-1}$ (T4). The other fertilizers were potassium chloride (256.8 $kg{\cdot}ha^{-1}$) and superphosphate (450 $kg{\cdot}ha^{-1}$). Apply all fertilizers after one week of transplanting.

The pot experiment started with preliminary sowing of the seeds on July 15th and transplanting on August 4th. The test plots were inverted conical plastic buckets with the top round diameter of 32 cm, the bottom round diameter of 26 cm and the height of 25 cm. Each pot received 5 seedlings and 5 different nitrogen fertilizer treatments 0 g (CK), 0.85 g (T1), 1.70 g (T2), 2.55 g (T3) and 3.40 g (T4). The other fertilizers were potassium chloride (1.71 g) and superphosphate (3.00 g). We applied all fertilizers after one week of transplanting.

2.3. Measurement Methods

2.3.1. Rice Quality

To calculate the rate of brown rice, approximately 120 g of rice samples were weighed and husked by the SDL-A rice huller to remove the glume and then weigh. In addition, to calculate the rate of milled rice, an SDJ−100 milled rice mill was used to remove the husk from the brown rice, which was then weighed. The milled rice was removed with a 1.0 mm round hole sieve, and the percent head rice was weighed out then calculated. The length and width of head rice were measured by Rice Appearance Quality Analyzer SC-E (The vendor: Wanshen Hangzhou, China). The specific operation was determined by referring to GB/T17891-2017 high-quality rice [17].

2.3.2. Lodging Index Determination Method

A plant lodging instrument was used to measure the lodging index at the maturity stage. Six plants with average effective panicle number were selected from each plot, and a lodging meter was used to uniformly push the base of the plant 20 cm from the ground to 45 degrees of plant inclination (the instrument was always perpendicular to the rice stem) to record the reading P and note down fresh weight G and measure plant height H [18,19].

$$\mathrm{F} = \mathrm{P} \times \mathrm{K}$$

(F for per plant rice setback force, the unit of N; P for the reading of lodging meter, the unit of N; K for different spring corresponding elastic coefficient.)

$$\mathrm{BM} = \mathrm{G} \times \mathrm{H}$$

(BM for bending moment; G for fresh plant weight, the unit is g; H for plant height, unit is cm.)

$$\mathrm{RM} = \mathrm{F/g} \times \mathrm{h}$$

(RM for setback torque; g as the gravitational constant, unit 9.8 N/1000 g; h = 20 cm, is the distance between the base of the plant and the site pushed by the lodging instrument.)

$$\mathrm{LI} = (\mathrm{BM/RM}) \times 100$$

(LI for Rice lodging index per plant. BM for bending moment. RM for setback torque.)

2.3.3. Photosynthetic Characteristics

The net photosynthetic rate (NPn), transpiration rate (Tr) and intercellular carbon dioxide concentration (Ci) parameters of rice leaves were determined by a portable photosynthetic system LI6400XT (LI-COR) in the morning (9:00–11:00) on a sunny day.

2.3.4. Leaf Area Index

$LAI = 0.75 \times \rho \times \frac{\sum_{j}^{m}\sum_{i}^{n}\left(L_{ij} \times B_{ij}\right)}{m}$ the formula, the leaf length (L_{ij}) and the maximum leaf width (B_{ij}) of each leaf of each plant were measured by ruler, and n was the total number of leaves of the j plant. m is the number of measured plants; 0.75 is the correction coefficient; ρ is the planting density [20].

2.3.5. Nitrogen Utilization Ratio Calculation

$$\mathrm{HI}\ (\%) = \mathrm{G/B} \times 100$$

$$AE\ (kg/kg) = (Y - Y_0)/F$$

$$PFP_N\ (kg/kg) = Y/F$$

$$FCR_N\ (\%) = (Y - Y_0)/Y \times 100$$

(HI for grain yield index; G for grain yield; B for biomass; AE for nitrogen agronomic utilization rate; PFP_N for partial nitrogen productivity; FCR_N for nitrogen contribution rate; Y is grain yield of crops treated with nitrogen fertilizer. Y_0 is the grain yield of crops not treated with nitrogen. F is the amount of nitrogen applied.) [21,22].

2.4. Data Analysis

The data collected from the experiment were analyzed with Microsoft Excel 2017 (Microsoft, Redmond, WA, USA). IBM SPSS Statistics 21 was used for significance analysis and interaction analysis. Multiple comparisons were conducted using the least significant difference method (LSD). The graphs were drawn using the Origin 2017 software. Multivariate analysis was analyzed by the MetaboAnalyst software (https://www.metaboanalyst.ca/).

3. Results

3.1. Effects of Nitrogen Treatment on Yield of Giant Rice

Nitrogen treatment affected the yield of giant rice and its components (Table 2). In terms of yields. In 2018, the highest yields of Feng5 and Feng6 were 8.78 t·ha^{-1} and 11.08 t·ha^{-1} at T2 and T3 treatments, respectively. Compared with CK, the yield increase was 38.27% and 55.84%, respectively. In 2019, the yield of Feng5 and Feng6 reached the maximum under T3 treatment. In terms of changes in effective panicle and grain-filling percentage. In 2018, the highest effective panicle and grain-filling percentage of Feng5 appeared in T3 treatment, which increased by 52.76% and 31.57%, respectively, compared with CK treatment. In 2019, the highest effective panicle and grain-filling percentage of Feng5 increased by 118.75% and 30.00%, respectively, compared with CK treatment. In 2018 and 2019, the maximum 1000 grain weight of Feng5 appeared in the T1 treatment, which increased by 0.80% and 4.51%, respectively, compared with the CK treatment. In 2018, the maximum 1000 grain weight of Feng6 was 2.81% higher in the T2 treatment than in the CK treatment. In 2019, the maximum 1000 grain weight of Feng6 appeared in T3 treatment with an increase of 5.61% compared with CK. The interaction between variety and treatment was analyzed (Table 3), and effective panicle, grain-filling percentage and yield were significantly affected by variety, while effective panicle, grain-filling percentage and yield were significantly affected by nitrogen treatment.

3.2. Effects of Nitrogen Treatment on Quality of Giant Rice

The grain length and width of head rice were significantly affected by nitrogen treatment, while the ear length was not significantly affected. The maximum ear length was mainly concentrated in T1 and T2 treatment (Table 4). In terms of brown rice rate, in 2018, Feng5 and Feng6 reached the maximum value of 79.71% and 80.66% under T3 treatment, which increased by 1.50% and 1.96%, respectively compared to CK. In 2019, the maximum value reached in T1 was 78.50% and 84.62%, which increased by 1.71% and 6.08%, respectively, compared to CK. In terms of milled rice rate and head rice rate, in 2018, Feng5 reached the maximum values of 68.89% and 56.82% under T4 treatment and Feng6 reached the maximum values of 70.20% and 60.59% under T3 treatment. In 2019, the maximum value of Feng5 at T2 was 67.40% and 59.13%, while the maximum value of Feng6 was 74.98% and 65.10% at T1 and T2, respectively. In terms of grain length and width of head rice, Feng5 reached 6.21 mm and 2.24 mm at T2 treatment in 2018, which were significantly increased by 1.64% and 2.28%

compared to CK. Feng6 reached 6.45 mm and 2.19 mm, with a significant increase of 4.54% and 3.79% compared to CK. In 2019, the grain length of head rice reached the maximum value under T4 treatment, with Feng5 and Feng6 reaching 5.70 mm and 5.77 mm, respectively. The maximum width of head rice was 2.26 mm and 2.30 mm at T2 and T1, respectively. The interaction between varieties and treatments was analyzed (Table 5). Varieties had a significant effect on the brown rice rate, milled rice rate and head rice rate. The grain length and width, brown rice, milled rice and head rice were significantly affected by nitrogen treatment.

Table 2. Changes of yield and components of giant rice under different nitrogen treatments.

Growing Season	Variety	Treatment	Number of Filled Grains Per Panicle	Effective Panicle Number (10^4/hm^2)	1000 Grain Weight (g)	Grain-Filling Percentage (%)	Yield (t·ha^{-1})
Late rice in 2018 field	Feng5	CK	192.87 ^{b}	156.00 ^{b}	27.45 ^{a}	41.84 ^{b}	6.35 ^{a}
		T1	211.21 ^{b}	186.33 ^{a,b}	27.67 ^{a}	48.39 ^{a,b}	7.73 ^{a}
		T2	274.81 ^{a}	195.00 ^{a,b}	25.40 ^{b}	54.07 ^{a}	8.78 ^{a}
		T3	243.96 ^{a,b}	238.33 ^{a}	26.57 ^{a,b}	55.05 ^{a}	7.70 ^{a}
		T4	267.60 ^{a}	203.67 ^{a,b}	26.90 ^{a}	44.70 ^{b}	8.33 ^{a}
	Feng6	CK	206.34 ^{a}	186.33 ^{a}	26.65 ^{a}	52.69 ^{a}	7.11 ^{b}
		T1	236.53 ^{a}	216.65 ^{a}	26.43 ^{a}	50.26 ^{a}	7.44 ^{b}
		T2	210.27 ^{a}	229.67 ^{a}	27.40 ^{a}	55.94 ^{a}	9.41 ^{a}
		T3	229.78 ^{a}	242.68 ^{a}	27.17 ^{a}	53.05 ^{a}	11.08 ^{a,b}
		T4	241.38 ^{a}	242.67 ^{a}	26.57 ^{a}	50.87 ^{a}	8.36 ^{a,b}
Late rice in 2019 potting	Feng5	CK	134.50 ^{a}	80.00 ^{c}	24.20 ^{a,b}	61.94 ^{b}	1.57 ^{b}
		T1	161.89 ^{a}	126.33 ^{b}	25.30 ^{a}	73.17 ^{a}	3.81 ^{a}
		T2	141.22 ^{a}	162.00 ^{a,b}	22.85 ^{b}	76.54 ^{a}	3.89 ^{a}
		T3	127.44 ^{a}	175.00 ^{a}	24.31 ^{a,b}	80.52 ^{a}	4.34 ^{a}
		T4	155.29 ^{a}	139.00 ^{a,b}	25.01 ^{a}	74.18 ^{a}	3.89 ^{a}
	Feng6	CK	85.11 ^{b}	74.67 ^{c}	25.29 ^{a}	74.97 ^{b}	1.21 ^{b}
		T1	106.00 ^{a,b}	84.33 ^{b,c}	25.50 ^{a}	76.00 ^{b}	1.76 ^{b}
		T2	128.33 ^{a,b}	151.00 ^{a}	26.10 ^{a}	75.02 ^{b}	3.68 ^{a}
		T3	142.73 ^{a}	138.67 ^{a}	26.71 ^{a}	74.76 ^{b}	3.77 ^{a}
		T4	130.51 ^{a,b}	128.00 ^{a,b}	26.23 ^{a}	84.35 ^{a}	3.58 ^{a}

Note: Different small letters mean significant at $p < 0.05$ level within a variety.

Table 3. Interaction of varieties and treatments on yield and its components in giant rice.

F Value		Number of Filled Grains Per Panicle	Effective Panicle Number (10^4/hm^2)	1000-Grain Weight (g)	Grain-Filling Percentage (%)	Yield (t·ha^{-1})
2018	Variety	2.68	5.34 *	0.15	5.51 *	4.49 *
	Treatment	2.08	3.74 **	1.16	4.06 **	4.91 **
	Variety × Treatment	2.16	0.26	5.24 **	1.89	1.00
2019	Variety	7.24 *	5.74 *	18.38 **	4.77 *	19.32 **
	Treatment	1.40	12.16 **	1.42	4.66 **	37.76 **
	Variety × Treatment	1.84	0.72	1.97	4.19 *	4.61 **

Note: ** and * mean significant difference at 1% and 5% level, respectively.

Table 4. Changes in the quality of giant rice under different nitrogen treatments.

Growing Season	Variety	Treatment	Ear Length (cm)	Length of Head Rice (mm)	Width of Head Rice (mm)	Brown Rice Rate (%)	Milled Rice Rate (%)	Head Rice Rate (%)
Late rice in 2018 field	Feng5	CK	21.46 ^{a}	6.11 ^{a,b}	2.19 ^{b}	78.53 ^{a}	67.02 ^{b}	49.97 ^{b}
		T1	21.59 ^{a}	6.14 ^{a,b}	2.22 ^{ab}	78.78 ^{a}	67.63 ^{a,b}	53.28 ^{a,b}
		T2	22.72 ^{a}	6.21 ^{a}	2.24 ^{a}	78.25 ^{a}	66.17 ^{b}	53.21 ^{a,b}
		T3	21.81 ^{a}	6.02 ^{b}	2.19 ^{b}	79.71 ^{a}	67.84 ^{a,b}	54.53 ^{a}
		T4	22.66 ^{a}	6.06 ^{a,b}	2.21 ^{a,b}	79.70 ^{a}	68.89 ^{a}	56.82 ^{a}
	Feng6	CK	23.92 ^{a}	6.17 ^{c}	2.11 ^{c}	79.11 ^{b}	69.06 ^{a}	57.61 ^{a}
		T1	25.08 ^{a}	6.33 ^{b}	2.16 ^{a,b}	79.97 ^{a,b}	68.85 ^{a}	59.06 ^{a}
		T2	24.63 ^{a}	6.45 ^{a}	2.19 ^{a}	79.38 ^{a,b}	68.77 ^{a}	58.04 ^{a}
		T3	24.87 ^{a}	6.24 ^{c}	2.14 ^{b,c}	80.66 ^{a}	70.20 ^{a}	60.59 ^{a}
		T4	24.19 ^{a}	6.16 ^{c}	2.12 ^{b,c}	80.30 ^{a,b}	69.81 ^{a}	59.03 ^{a}
Late rice in 2019 potting	Feng5	CK	21.88 ^{a}	5.58 ^{c}	2.22 ^{c}	77.18 ^{b}	60.13 ^{b}	51.02 ^{b}
		T1	22.98 ^{a}	5.64 ^{b}	2.31 ^{a}	78.50 ^{a}	60.85 ^{b}	53.88 ^{a,b}
		T2	21.90 ^{a}	5.67 ^{a,b}	2.26 ^{b}	78.07 ^{a,b}	67.40 ^{a}	59.13 ^{a}
		T3	21.98 ^{a}	5.49 ^{d}	2.22 ^{c}	77.48 ^{b}	66.11 ^{a}	58.91 ^{a}
		T4	22.54 ^{a}	5.70 ^{a}	2.22 ^{c}	77.35 ^{b}	60.82 ^{b}	52.20 ^{b}
	Feng6	CK	21.53 ^{a}	5.37 ^{b}	2.24 ^{b}	79.77 ^{b}	69.54 ^{b}	58.70 ^{b}
		T1	21.40 ^{a}	5.61 ^{a,b}	2.30 ^{a}	84.62 ^{a}	74.98 ^{a}	59.48 ^{b}
		T2	21.17 ^{a}	5.69 ^{a}	2.24 ^{b}	80.48 ^{b}	71.83 ^{a,b}	65.10 ^{a}
		T3	20.97 ^{a}	5.64 ^{a}	2.20 ^{c}	80.60 ^{b}	71.58 ^{a,b}	60.15 ^{b}
		T4	20.93 ^{a}	5.77 ^{a}	2.22 ^{b,c}	79.90 ^{b}	69.24 ^{b}	58.88 ^{b}

Note: Different small letters mean significant at $p < 0.05$ level within a variety.

Table 5. Interaction of varieties and treatments on the quality of giant rice.

F Value		Ear Length (cm)	Length of Head Rice (mm)	Width of Head Rice (mm)	Brown Rice Rate (%)	Milled Rice Rate (%)	Head Rice Rate (%)
2018	Variety	34.93 **	47.32 **	61.13 **	8.86 **	24.09 **	52.22 **
	Treatment	1.48	11.51 **	8.56 **	3.70 *	3.31 *	4.07 *
	Variety × Treatment	1.51	1.54	0.88	0.186	0.77	1.49
2019	Variety	1.72	0.02	1.31	83.21 **	143.91 **	24.87 **
	Treatment	0.10	6.04 **	40.93 **	9.24 **	7.95 **	6.16 **
	Variety × Treatment	0.09	2.67	1.20	3.62 *	5.97 **	1.03

Note: ** and * mean significant difference at 1% and 5% level, respectively.

3.3. Effects of Nitrogen Treatment on Photosynthetic Characteristics of Giant Rice

Nitrogen treatment affected the net photosynthetic rate and transpiration rate (Table 6). In a certain range, the net photosynthetic rate and transpiration rate increase with the increase of nitrogen application. In general, the net photosynthetic rate and transpiration rate of rice at the tillering stage were higher than those at the full panicle stage. The net photosynthetic rate and transpiration rate of rice in the field were higher than those of rice in the pot experiment. As for Feng5, the net photosynthetic rate and transpiration rate reached maximum values of 28.52 $\mu mol \cdot m^{-2} \cdot s^{-1}$ and 11.52 $g \cdot m^{-2} \cdot h^{-1}$ in the T2 treatment at the tillering stage in 2018, which were 26.19% and 3.97% higher than CK. The net photosynthetic rate and transpiration rate reached maximum values of 17.22 $\mu mol \cdot m^{-2} \cdot s^{-1}$ and 5.45 $g \cdot m^{-2} \cdot h^{-1}$ under the T3 treatment at the full heading stage, which were 81.45% and 31.64% higher than CK and 39.62% and 52.69% lower than maximum net photosynthetic rate and transpiration rate at tillering stage. In the T3 treatment for the tillering stage in 2019, the net photosynthetic rate and transpiration rate reached a maximum 12.00 $\mu mol \cdot m^{-2} \cdot s^{-1}$ and 4.89 $g \cdot m^{-2} \cdot h^{-1}$, 25.39% and 25.06% higher than CK, net photosynthetic rate and transpiration rate reached a maximum 10.30 $\mu mol \cdot m^{-2} \cdot s^{-1}$ and 4.92 $g \cdot m^{-2} \cdot h^{-1}$ in the T2 treatment for the heading stage, 43.45% and 20.59% higher than CK. As for Feng6, the net photosynthetic rate and transpiration rate at the tillering stage T3 in 2018 reached the maximum 25.09 $\mu mol\ m^{-2} \cdot s^{-1}$ and 8.07 $g \cdot m^{-2} \cdot h^{-1}$, which were up to 37.93% and 9.35% higher than CK; the net photosynthetic rate and transpiration rate at the full heading stage reached a maximum of 16.60 $\mu mol\ m^{-2} \cdot s^{-1}$ and 5.13 $g \cdot m^{-2} \cdot h^{-1}$ under the T2 treatment, which were 33.89% and 36.43% lower than the maximum net photosynthetic rate and transpiration rate at the tillering stage.

The net photosynthetic rate and transpiration rate reached maximum values of 7.24 μmol·m^{-2}·s^{-1} and 3.30 g·m^{-2}·h^{-1} at full heading stage T3 in 2019, which were 18.49% and 38.07% higher than CK and 44.61% and 34.91% lower than the maximum net photosynthetic rate of 13.07 μmol·m^{-2}·s^{-1} and transpiration rate of 5.70 g·m^{-2}·h^{-1} at tillering stage. In terms of 2018, in the field experiment and 2019 pot experiments, the maximum net photosynthetic rate and transpiration rate of Feng5 at the tillering stage in 2018 were 137.66% and 135.58% higher than that in 2019, and the maximum net photosynthetic rate and transpiration rate at full heading stage were 67.18% and 10.77% higher than that in 2019. In 2018, the maximum net photosynthetic rate and transpiration rate of Feng6 at the tillering stage were 91.97% and 41.58% higher than those in 2019, and the maximum net photosynthetic rate and transpiration rate at the full heading stage were 129.28% and 55.45% higher than those in 2019.

Table 6. Changes of photosynthetic characteristics under nitrogen treatment.

Growing Season	Variety	Treatment	Tillering			Full Heading		
			Ci (μmol)	Pn (μmol·m^{-2}·s^{-1})	Tr (g·m^{-2}·h^{-1})	Ci (μmol)	Pn (μmol·m^{-2}·s^{-1})	Tr (g·m^{-2}·h^{-1})
Late rice in 2018 field	Feng5	CK	343.00 ^{a,b}	22.60 ^{b}	11.08 ^{a}	89.56 ^{a}	9.49 ^{d}	4.14 ^{c}
		T1	340.67 ^{a}	22.98 ^{b}	10.33 ^{b}	83.56 ^{a}	14.98 ^{b}	4.87 ^{b}
		T2	332.56 ^{b}	28.52 ^{a}	11.52 ^{a}	60.44 ^{b}	11.13 ^{c}	3.77 ^{c}
		T3	313.44 ^{c}	28.18 ^{a}	10.19 ^{b}	94.89 ^{a}	17.22 ^{a}	5.45 ^{a}
		T4	309.11 ^{b}	22.82 ^{b}	8.66 ^{c}	79.33 ^{a}	10.93 ^{c,d}	4.10 ^{c}
	Feng6	CK	299.33 ^{a}	18.19 ^{c}	7.38 ^{b}	91.89 ^{a,b}	4.08 ^{e}	2.75 ^{c}
		T1	291.00 ^{a}	19.61 ^{c}	7.31 ^{b}	81.56 ^{b,c}	13.44 ^{b}	4.74 ^{a}
		T2	290.44 ^{a}	22.42 ^{b}	7.70 ^{a,b}	68.33 ^{c,d}	16.60 ^{a}	5.13 ^{a}
		T3	296.11 ^{a}	25.09 ^{a}	8.07 ^{a}	54.17 ^{d}	11.78 ^{c}	3.95 ^{b}
		T4	300.78 ^{a}	23.33 ^{b}	7.98 ^{a}	104.78 ^{a}	6.10 ^{d}	3.00 ^{c}
Late rice in 2019 potting	Feng5	CK	265.00 ^{a,b}	9.57 ^{b}	3.91 ^{a,b}	271.13 ^{a}	7.18 ^{b}	4.08 ^{a}
		T1	213.67 ^{d}	9.97 ^{b}	2.96 ^{b}	240.41 ^{a}	9.24 ^{a}	3.93 ^{a}
		T2	226.33 ^{c,d}	11.77 ^{a}	3.48 ^{b}	268.83 ^{a}	10.30 ^{a}	4.92 ^{a}
		T3	300.67 ^{a}	12.00 ^{a}	4.89 ^{a}	266.82 ^{a}	8.66 ^{a}	4.45 ^{a}
		T4	259.67 ^{b,c}	10.63 ^{a,b}	3.56 ^{b}	273.40 ^{a}	9.00 ^{a,b}	4.63 ^{a}
	Feng6	CK	288.00 ^{a,b}	11.70 ^{a}	4.4 ^{a,b}	191.33 ^{a}	6.11 ^{a}	2.39 ^{a}
		T1	310.33 ^{a}	12.20 ^{a}	5.70 ^{a}	210.00 ^{a}	6.34 ^{a}	2.44 ^{a}
		T2	308.67 ^{a}	13.07 ^{a}	5.67 ^{a}	214.08 ^{a}	6.52 ^{a}	2.74 ^{a}
		T3	278.00 ^{a,b}	12.53 ^{a}	5.02 ^{a,b}	234.54 ^{a}	7.24 ^{a}	3.30 ^{a}
		T4	244.67 ^{b}	9.34 ^{b}	3.45 ^{b}	249.62 ^{a}	7.03 ^{a}	3.19 ^{a}

Note: Different small letters mean significant at $p < 0.05$ level within a variety.

Nitrogen affected the LAI of giant rice (Figure 2). LAI increased with the increase of nitrogen application amount. In the rice-growing period, the maximum value of LAI was mainly concentrated in the booting stage, and the full heading stage and LAI increased significantly under the T4 treatment compared with CK treatment at the tillering stage and mature stage. In 2018. LAI under the T4 treatment significantly increased compared with CK at the tillering stage, heading, heading 15 days and mature stage. In 2019, in terms of Feng5, LAI in the T4 treatment was higher than that in CK treatment in all growth stages of giant rice. AS far as Feng6 is concerned, LAI under T4 treatment was significantly higher than under CK treatment at the tillering stage and maturation stage.

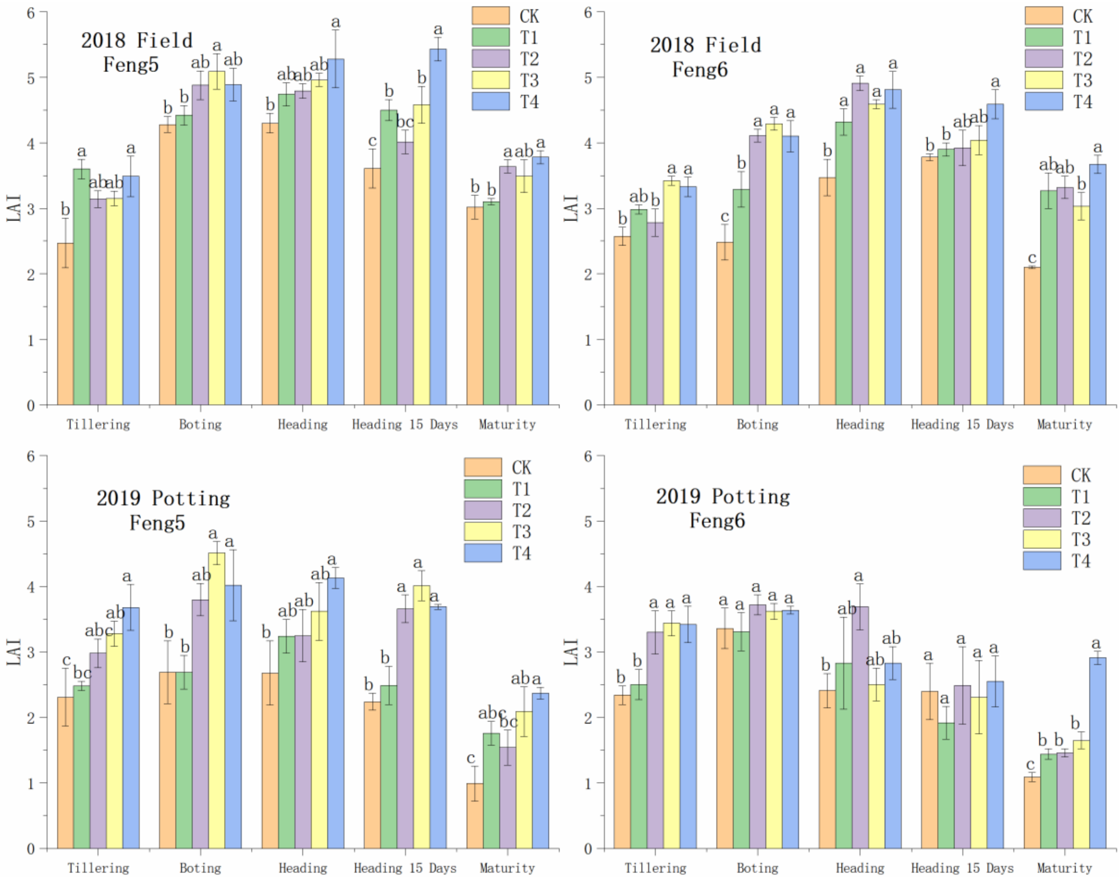


Figure 2. The leaf area index of giant rice under different treatments. Different small letters mean significant at $p < 0.05$ level within a variety.

3.4. Effects of Nitrogen Treatment on Physiological Characteristics of Giant Rice

Nitrogen affected plant height and the tillering of giant rice (Figure 3). With an increase in nitrogen application amount, the tiller number increased. The tiller basically maintained a dynamic balance after the booting stage. Plant height increased with the increase of nitrogen application amount and slightly decreased in the later stage. The highest value of plant height was mainly concentrated between the full-heading stage and 15 days after the full heading stage. In terms of tillers, Feng5 and Feng6 reached the highest number of tillers in the T4 and T3 treatments, respectively in 2018, among which, the number of tillers in T4 and T3 increased by 29.14% compared to CK, and that in T3 compared to CK increased by 34.97% compared to Feng6. In 2019, both Feng5 and Feng6 reached the highest tiller number in T4 treatment, among which, the highest tiller number of Feng5 in T4 increased by 54.50% compared with CK, and the highest tiller number of Feng6 in T3 increased by 64.24% compared with CK. In terms of plant height, the highest Feng5 and Feng6 plant height in 2018 were 171.67 cm and 181.20 cm under the T3 treatment, respectively, which were 37.70% and 38.32% higher than those of CK shortest plants of 124.67 cm and 131.00 cm. In 2019, the highest height of the Feng5 strain reached 148.07 cm under the T4 treatment, which was 21.04% higher than that of CK shortest strain 122.33 cm. The highest Feng6 plant height reached 151.30 cm under the T3 treatment, which was 21.92% higher than CK shortest plant height of 124.10 cm.

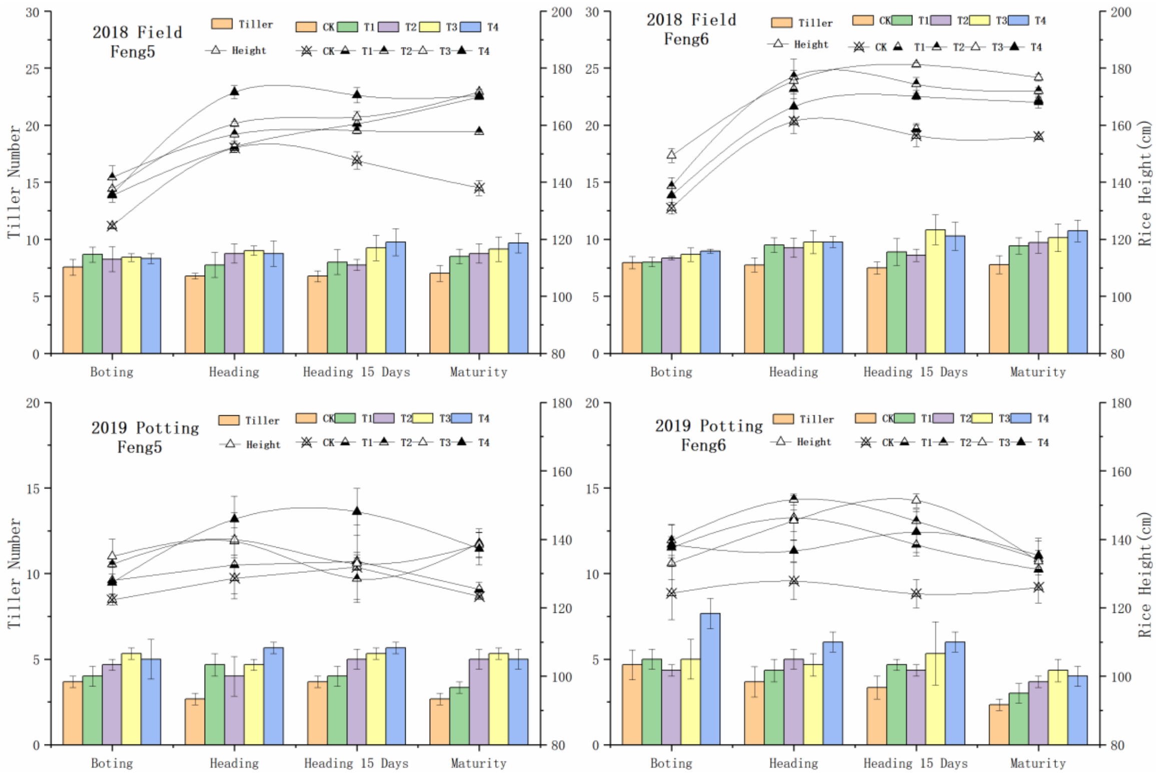


Figure 3. Changes in plant height and tillering of giant rice under nitrogen treatment.

Nitrogen affected the LI of giant rice (Figure 4). In the field test in 2018, the LI showed a decreasing trend after an increasing trend with the increase of nitrogen application amount. Among them, the LI of Feng5 under the T3 treatment reached the minimum value of 160.84, which was reduced by 13.98% compared with CK. The LI of Feng 6 at T2 reached a minimum of 96.51, which was 39.52% lower than that of CK. In the 2019 pot experiment, LI decreased with the increase of nitrogen application. Among them, the minimum value of Feng5 reached 122.00 in the T4 treatment, which decreased by 32.79% compared with CK. Feng6 reached a minimum value of 123.96 in the T3 treatment, which was 27.62% lower than CK.

3.5. Effects of Nitrogen Treatment on Nitrogen Efficiency in Giant Rice

The nitrogen application rate had an effect on the nitrogen utilization rate (Table 7). The nitrogen agronomic utilization rate (AE) and the nitrogen partial productivity (PFP_N) in rice decreased with the increase of nitrogen application rate. The grain yield index (HI) and nitrogen contribution rate (FCR_N) in rice first increased and then decreased with the increase of nitrogen application rate. For grain harvest index, in 2018, T2 > T1 > T3 > T4 > CK in each Feng5 treatment interval, T2 significantly increased by 32.77% compared with CK, while T1 > T2 > T3 > T4 > CK in each Feng6 treatment interval, T1 increased by 9.05% compared with CK. In 2019, Feng5 and Feng6 reached the maximum grain harvest index under T1 and T2 treatment, respectively. As for nitrogen contribution rate, in 2018, the maximum nitrogen contribution rate of Feng5 and Feng6 reached 27.27% and 35.74%, respectively, at T2 and T3, and in 2019, the maximum nitrogen contribution rate of Feng5 and Feng6 reached T3. For the nitrogen agronomic utilization rate (AE) and nitrogen partial productivity (PFP_N) of rice, the nitrogen partial productivity (PFP_N) of the two varieties treated with T1 was significantly higher than that treated with T4, and the nitrogen partial productivity (PFP_N) showed T1 > T2 > T3 > T4.

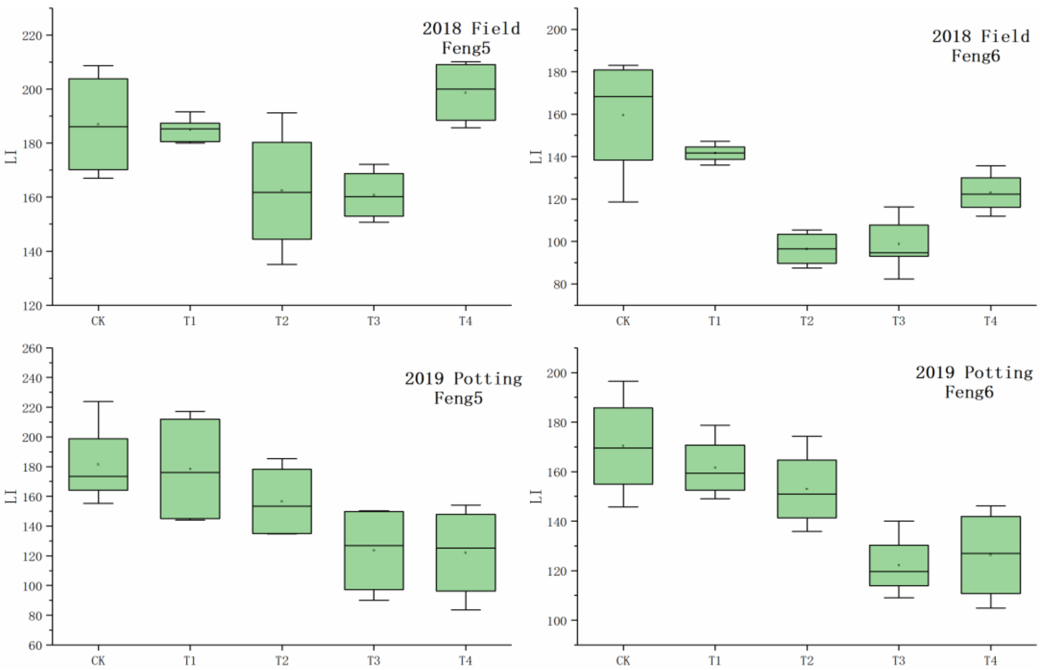


Figure 4. Lodging index (LI) change of giant rice under nitrogen treatment.

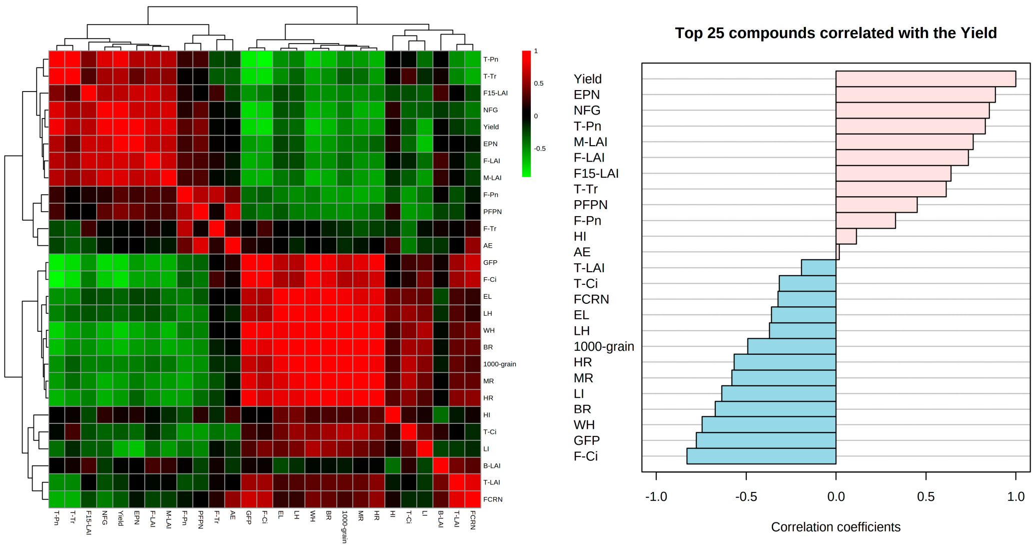
Table 7. Changes in nitrogen efficiency in giant rice under nitrogen treatment.

Growing Season	Variety	Treatment	HI (%)	AE (kg·kg^{-1})	PFP$_N$ (kg·kg^{-1})	FCR$_N$ (%)
Late rice in 2018 field	Feng5	CK	42.93 ± 4.17 ^{b}	-	-	-
		T1	46.57 ± 1.25 ^{b}	18.43 ± 5.08 ^{a}	61.83 ± 3.42 ^{a}	16.41 ± 7.44 ^{a}
		T2	57.00 ± 4.58 ^{a}	16.17 ± 2.89 ^{b}	58.54 ± 2.89 ^{a}	27.27 ± 3.68 ^{a}
		T3	46.48 ± 0.71 ^{b}	8.94 ± 1.11 ^{c}	34.22 ± 6.68 ^{b}	21.30 ± 9.95 ^{a}
		T4	43.42 ± 1.75 ^{b}	6.60 ± 1.29 ^{c}	27.78 ± 4.14 ^{b}	19.90 ± 13.0 ^{a}
	Feng6	CK	46.52 ± 1.30 ^{a}	-	-	-
		T1	50.73 ± 0.73 ^{a}	17.68 ± 2.24 ^{a}	69.2 ± 3.00 ^{a}	15.64 ± 1.71 ^{b}
		T2	47.65 ± 1.50 ^{a}	15.33 ± 3.60 ^{a}	62.75 ± 3.60 ^{a}	23.92 ± 4.39 ^{b}
		T3	46.75 ± 2.31 ^{a}	17.63 ± 1.02 ^{a}	49.25 ± 1.02 ^{b}	35.74 ± 1.36 ^{a}
		T4	46.67 ± 2.30 ^{a}	4.16 ± 1.00 ^{b}	27.87 ± 1.00 ^{c}	14.70 ± 3.06 ^{b}
Late rice in 2019 potting	Feng5	CK	40.02 ± 1.57 ^{a}	-	-	-
		T1	44.24 ± 0.46 ^{a}	29.86 ± 5.38 ^{a}	50.82 ± 5.38 ^{a}	57.71 ± 4.93 ^{a}
		T2	41.17 ± 1.75 ^{a}	15.49 ± 1.69 ^{b}	25.97 ± 1.69 ^{b}	59.32 ± 2.50 ^{a}
		T3	42.41 ± 4.58 ^{a}	12.34 ± 0.98 ^{b}	19.33 ± 0.98 ^{b,c}	63.65 ± 1.91 ^{a}
		T4	30.44 ± 1.38 ^{b}	7.75 ± 0.33 ^{b}	12.99 ± 0.33 ^{c}	59.59 ± 1.03 ^{a}
	Feng6	CK	29.38 ± 2.18 ^{c}	-	-	-
		T1	40.21 ± 2.06 ^{a,b}	16.47 ± 1.89 ^{a}	26.14 ± 0.92 ^{a}	37.95 ± 2.20 ^{b}
		T2	46.42 ± 5.69 ^{a}	9.96 ± 0.92 ^{b}	24.56 ± 1.89 ^{a}	66.69 ± 2.41 ^{a}
		T3	41.65 ± 1.13 ^{a,b}	11.39 ± 1.18 ^{b}	16.78 ± 1.18 ^{b}	67.54 ± 2.25 ^{a}
		T4	36.97 ± 1.30 ^{b,c}	7.90 ± 0.61 ^{b}	11.95 ± 0.61 ^{c}	65.98 ± 1.69 ^{a}

Note:Different small letters mean significant at $p < 0.05$ level within a variety. The same as below.

3.6. Multivariate Analysis

The investigated parameters were clustered for different treatments. For instance, brown rice rate and head rice rate were highly related to grain-filling percentage and width of head rice, whereas grain-filling percentage and the Ci contents at full heading stage were negatively related to yield, but effective panicle number and the LAI at full heading stage were highly related to yield. The length of head rice, ear length, milled rice rate were positively correlated with 1000 grain weight. Furthermore, effective panicle number, number of filled grains per panicle, net photosynthetic rate at tilling stage, leaf area index at mature stage, leaf area index at full heading stage as top five indexes, which positively correlated with yield, oppositely, it showed that intercellular carbon dioxide concentration at the full heading stage, grain-filling percentage, the width of head rice, negatively correlated with yield (Figure 5a,b). The PCA analysis of the investigated parameters revealed that group 1, group 2, group 3 accounted for 47.2%, 14.1% and 10.3%, respectively (Figure 6).



(a) (b)

Figure 5. Correlations of the investigated parameters. (**a**): Coordinate chart of correlation of each parameter. (**b**): Top 25 compounds correlations with the yield. T-LAI: leaf area index at the tillering stage. B-LAI: leaf area index at the booting stage. F-LAI: leaf area index at full heading stage. F15-LAI: leaf area index at 15 days after the full heading stage. M-LAI: leaf area index at the mature stage. NFG: number of filled grains per panicle. EPN: effective panicle number. GFP: grain-filling percentage. EL: ear length. LH: length of head rice. WH: width of head rice. BR: brown rice rate. MR: milled rice rate. HR: head rice rate. T-Ci: intercellular carbon dioxide concentration at the tilling stage. T-Pn: net photosynthetic rate at the tilling stage. T-Tr: transpiration rate at the tilling stage. F-Ci: intercellular carbon dioxide concentration at the full heading stage. F-Pn: net photosynthetic rate at full heading stage. F-Tr: transpiration rate at full heading stage.

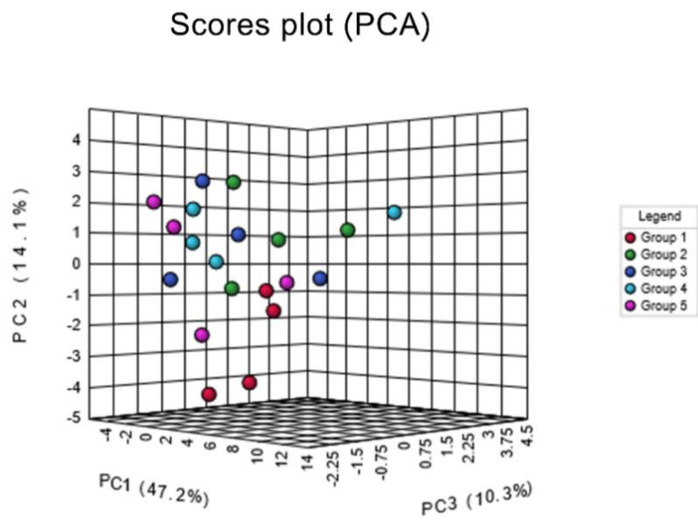


Figure 6. Principal component analyses (PCA) of 3D scores plot.

4. Discussion

Rice yield and quality are comprehensive traits controlled by internal genes and external growth environment and cultivation measures. In order to improve the yield and quality of rice, in terms of genetic breeding, rice has experienced a process of dwarfing, hybridizing and super rice. In terms of cultivation, there is no doubt that nitrogen fertilizer is very important to rice yield and quality. In recent years, with the rise of source-pool-flow theory, many scientists are exploring rice varieties with high biological yields in order to improve yield. Compared with common rice, the new type of giant rice has higher plant height and biological yield. Therefore, it is necessary to investigate the nitrogen fertilizer effects on giant rice.

The amount of nitrogen applied affects the photosynthesis of plants. Plants grow under nitrogen stress often with little nitrogen fertilizer. However, it should be noted that more nitrogen fertilizer also causes effects on the plant growth [10,11], and nitrogen could increase the leaf area index at the later stage and increase the canopy of rice. Meanwhile, nitrogen fertilizer would lead to the elongation of various internodes of rice stems, especially the basal internodes, thus increasing plant height [23]. The results of this study showed that appropriate nitrogen could improve photosynthesis, but excessive nitrogen was not conducive to photosynthesis, and photosynthesis was stronger before the full heading stage of rice. In addition, the leaf area index increased with the increase of nitrogen application amount, and the occurrence of maximum leaf area index showed a trend of delay in the rice growth period with the increase of nitrogen application amount. Meanwhile, nitrogen could slow down the plant height shortening degree in the later stage of rice, which was different under different treatments. With the increase of nitrogen fertilizer application, the shortening degree decreased, and the plant height even increased under high nitrogen treatment, which may be related to the transport of stem sheath materials in the later stage of rice and the extrusion of rice stem by the increase of panicle weight in the later stage.

There are many factors affecting the nitrogen utilization rate of rice, among which the nitrogen application quantity is one of them. Chen et al. [24], Pan et al. [25], and Rea et al. [26] found that the partial productivity of nitrogen fertilizer for grain crops was 40~70 $kg \cdot kg^{-1}$, the agronomic efficiency of nitrogen fertilizer was 10~30 $kg \cdot kg^{-1}$, and the absorption utilization rate of nitrogen fertilizer was within the range of 30~50%. The normal nitrogen application level in South China is 120 $kg \cdot ha^{-1}$~150 $kg \cdot ha^{-1}$ [27,28]. Chen [29] using conventional fragrant rice varieties *TY998* and *YXYZ* as materials and applied 150 $kg \cdot N \cdot ha^{-1}$ nitrogen fertilizer at different depths in South China, and the results showed that AE was between 8~25 $kg \cdot kg^{-1}$ and HI was between 39~63%. However, compared with conventional rice, giant rice has a higher biological yield and needs more fertilizer. The study results showed that under field conditions, medium nitrogen fertilizer could obtain higher HI and FCR_N, AE, and PFP_N significantly decreased with the increase of nitrogen application amount. HI ranged from 42–57%, FCR_N ranged from 14–35%, AE ranged from 4–18 $kg \cdot kg^{-1}$, PFP_N ranged from 27–69 $kg \cdot kg^{-1}$.

There is no doubt that nitrogen fertilizer is very important to rice yield and quality. Several reports have shown that the effective panicle number and yield increase with the increase of nitrogen application in a certain range [30], and the brown rice rate and head rice rate were higher at medium nitrogen levels [31]. The results of this study also confirm this opinion and found that the first two main factors affecting the yield are the effective panicle number and the number of filled grains per panicle. Moreover, this study found that nitrogen fertilizer significantly affected the grain length and grain width of head rice. Under field conditions, head rice had a longer grain length under medium nitrogen, while in pot plants, the grain length had longer grain length under high nitrogen, and the grain width reached its maximum at medium nitrogen level. In addition, the head rice rate increase trend in the field, but it showed the parabolic trend in the pot. There are several possible explanations for this result. A possible explanation for this may be that the nitrogen concentration gradients complement each other in the field environment. However, the environment of the potted plant is relatively closed, so nitrogen elements cannot complement each other. Another possible explanation for this is that there are more microbes in the field than in the pot. There are, however, other possible explanations. In the pot, the grains of rice under high nitrogen are too long and thin, leading to breakage of these grains during processing.

5. Conclusions

In summary, compared with conventional rice, giant rice has a higher biological yield and needs more fertilizer. In order to avoid unnecessary waste of nitrogen fertilizer and pollution of the soil environment by excessive nitrogen application, it is necessary to study the nitrogen requirement of giant rice. However, prior to this, there was no data on the optimal nitrogen application rate for giant rice, so the results of this study provide a theoretical basis for the widespread cultivation of giant rice in South China. The results of this experiment show that giant rice treated with T2 and T3 can achieve higher yield, brown rice rate and milled rice rate, better lodging resistance, higher net photosynthetic rate and leaf area index, as well as higher AE and PFP_N Compared with other treatments. Therefore, the optimal nitrogen application interval of giant rice in South China should be 150 kg·ha^{-1}~225 kg·ha^{-1}.

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