agronomy

MDPI

Article

Combination of Polymer-Coated Urea and Rapid-Release Urea Increases Grain Yield and Nitrogen Use Efficiency of Rice by Improving Root and Shoot Activities

Rongyue Xu [1,2], Jiangyao Fu [1,2], Yajun Zhang [1,2], Zhiwei Sun [1,2], Yuemei Xu [1,2], Weiyang Zhang [1,2], Kuanyu Zhu [1,2,*], Junfei Gu [1,2], Zhiqin Wang [1,2] and Jianchang Yang [1,2,*]

[1] Jiangsu Key Laboratory of Crop Genetics and Physiology, Jiangsu Key Laboratory of Crop Cultivation and Physiology, Agricultural College, Yangzhou University, Yangzhou 225009, China; 211702119@stu.yzu.edu.cn (R.X.); mx201904529@stu.yzu.edu.cn (J.F.); mx202160112@stu.yzu.edu.cn (Y.Z.); mx202164988@stu.yzu.edu.cn (Z.S.); mx202262447@stu.yzu.edu.cn (Y.X.); wyz@yzu.edu.cn (W.Z.); gujf@yzu.edu.cn (J.G.); zqw@yzu.edu.cn (Z.W.)

[2] Jiangsu Co-Innovation Center for Modern Production Technology of Grain Crops, Yangzhou University, Yangzhou 225009, China

* Correspondence: kyzhu@yzu.edu.cn (K.Z.); jcyang@yzu.edu.cn (J.Y.)

Abstract: The use of polymer-coated urea (PCU) can improve nitrogen use efficiency (NUE), compared to the application of rapid-release urea (RU). However, the effect of PCU-based nitrogen management on grain yield and the NUE of rice and its underlying mechanism remain unclear. A japonica rice cultivar Jinxiangyu 1 was grown in the field with four treatments including N omission (0N), split application of RU (Control), one-time application of 100% PCU (T1), and one-time application of 70% PCU + 30% RU (T2). Results showed that, compared to the control, the grain yield was significantly increased in the T2 treatment, while it was comparable in the T1 treatment. This was mainly due to increased total spikelets in the T2 treatment. Root oxidation activity (ROA) and root zeatin (Z) + zeatin riboside (ZR) content during booting were the distinct advantages of the T2 treatment, compared to either the control or T1 treatment, exhibiting significant or highly significant correlations with leaf photosynthesis. This process contributed significantly to total spikelets and total N uptake. Additionally, the T2 treatment absorbed more N than the control without reducing the internal N use efficiency (IE_N), primarily due to its unchanged harvest index (HI) driven by comparable non-structural carbohydrate remobilization. In conclusion, combining PCU with RU can enhance the coordination of root and shoot traits during booting while maintaining a competitive HI at maturity, thereby significantly improving grain yield and achieving a balance in N uptake and utilization.

Keywords: rice (*Oryza sativa* L.); polymer-coated urea; grain yield; nitrogen use efficiency; root and shoot traits

check for updates

Citation: Xu, R.; Fu, J.; Zhang, Y.; Sun, Z.; Xu, Y.; Zhang, W.; Zhu, K.; Gu, J.; Wang, Z.; Yang, J. Combination of Polymer-Coated Urea and Rapid-Release Urea Increases Grain Yield and Nitrogen Use Efficiency of Rice by Improving Root and Shoot Activities. *Agronomy* **2024**, *14*, 1585. https://doi.org/10.3390/agronomy14071585

Academic Editor: Massimo Fagnano

Received: 12 June 2024
Revised: 17 July 2024
Accepted: 19 July 2024
Published: 20 July 2024

1. Introduction

Rice (*Oryza sativa* L.) is one of the world's major staple crops, providing nearly 70% of dietary calories for over 3 billion people [1,2]. With urbanization and environmental pollution accelerating, China's arable land is dwindling [3]. Therefore, enhancing the rice yield per unit area is pivotal for ensuring food security [4]. Nitrogen (N) is a key factor in rice production and a significant component of production costs [4,5]. Generally, rapid-release urea (RU), i.e., conventional urea, is the most widely used fertilizer in rice production [5,6]. However, in high-yielding regions such as Jiangsu Province, the current high rates of nitrogen fertilizer input, with average RU applications exceeding 300 kg ha^{-1}, have merely resulted in annual rice yields surpassing 19 Mt since 2015 [4]. Previous studies have shown that the overuse of RU not only results in N loss and low nitrogen use efficiency (NUE) in paddy fields but also leads to soil acidification, water pollution, and elevated emissions of greenhouse gas [4,6,7]. Given the trade-off between an increase in NUE and adverse effects on the environment, agronomists propose that a split application of RU, that is, applying RU with an appropriate ratio based on the plant nutrient requirements at different growth stages, can address both of these concerns [8,9]. Nevertheless, this approach increases labor input amidst urbanization-induced labor shortages [7,10]. Therefore, improving both grain yield and NUE while minimizing environmental pollution, as well as achieving labor/time-saving, are urgent issues in rice production.

Controlled-release N fertilizer (CRNF) is a novel approach in fertilization characterized by its slow nutrient release, fewer application times, and reduced environmental impact [7,11,12]. It is commonly reported that polymer-coated urea (PCU), as a widely used CRNF, can mitigate nitrogen losses in paddy fields and improve NUE in rice [11]. However, some recent studies argue that a one-time application of PCU fails to increase grain yield in both rice and wheat due to its slow nitrogen release rate during the early stages, whereas a subsequent study observes an increase of 14.8–18.2% in cotton yield [12–14]. For example, previous studies present conflicting results, with some showing a decrease in rice grain yield when using PCU compared to RU, while others report an increase of up to 10% [13,14]. Additionally, some studies indicate that combining RU with PCU does not significantly affect grain yield or NUE, while other observations extensively demonstrate its potential to increase both [15,16]. These results suggest that PCU application management exhibits instability in increasing both grain yield and NUE depending on crop species. Therefore, it is important to investigate the effects of PCU-based N management on grain yield and NUE in rice.

In addition, many studies demonstrate that there is a coordinated relationship between root and shoot traits in rice, which affects grain yield and NUE [17–20]. Specifically, it is reported that rice root's morpho-physiological traits, including root dry weight, root length, and root activity, are conducive to improving shoot photosynthetic production [18,21]. Correspondingly, improved shoot activity can maintain considerable root activity by delivering photo-assimilates [22,23]. There is a proposal that enhancing root and shoot activities during the key growth stages of rice, particularly during spikelet formation and grain filling, can significantly increase grain yield and NUE [18,24]. Results reported by different studies have confirmed that root and shoot traits during these key growth stages vary with N application management strategies [18]. For instance, site-specific nitrogen management (SSNM) is believed to increase the number of spikelets per panicle and the percentage of filled grains, primarily driven by an enhanced root activity [25,26]. Additionally, numerous studies suggest that postponing nitrogen application benefits photosynthetic production, thereby improving grain filling in cereal crops [27–29]. However, the differences in root and shoot traits under PCU-based N management still remain obscure, and the mechanisms underlying the biological process for grain yield and NUE are not well understood.

This study tested the hypothesis that a combination of PCU with RU could synergistically increase grain yield and NUE in rice. Furthermore, we also explored the differences in root and shoot traits across different N management treatments, aiming to uncover the mechanisms governing grain yield and NUE. We anticipate that the findings of this research will provide practical and theoretical insights for the advancement of high-yield and high-efficiency cultivation practices in rice.

2. Materials and Methods

The site for this study was Yangzhou University, situated in Jiangsu Province, China (latitude 32.35° N, longitude 119.55° E). The experiment took place over the rice-growing seasons, from May to October, in both 2021 and 2022, at fixed points. The physicochemical properties of the soil are listed in Table S1. The average temperatures during the 2021 and 2022 growing seasons were 27.2 °C and 26.9 °C, respectively. Monthly rainfall averaged 142 mm in 2021 and 83.4 mm in 2022, while average monthly sunshine hours were 116 h in 2021 and 168 h in 2022 (Figure S1).

2.1. Experimental Design

A japonica variety, Jinxiangyu 1, was grown in a paddy field under four N management treatments: N omission (0N), split application of rapid-release urea (Control), one-time application of polymer-coated urea (T1), and a combination of 70% polymer-coated urea + 30% rapid-release urea (T2). In the control treatment, 40% of the nitrogen was applied as basal fertilizer, followed by 20% during early tillering, another 20% at panicle initiation, and the remaining 20% at pistil and stamen differentiation. The N amount was 240 kg ha^{-1} for each treatment, with T1 and T2 treatments applied once as basal fertilizer. The PCU, coated with starch and montmorillonite-modified polyurethane, had a release period of 120 d (Moith, Co., Ltd., Hefei, China). Additionally, 30 kg ha^{-1} of phosphorus (as P_2O_5) and 40 kg ha^{-1} of potassium (as KCl) were incorporated into the soil with the basal fertilizer. The experiment was conducted using a randomized block design, with each treatment plot measuring 30 m^2 and replicated three times. Chemical and manual methods were employed to manage weeds, insects, and diseases, preventing yield loss. The field experienced alternating wetting and drying cycles from 7 days after transplanting until one week before harvest.

2.2. Sampling and Measurement

Plant samples were collected at distinct growth stages: mid-tillering (MT) at 20 days after transplanting (DAT), panicle initiation (PI) at 40–42 DAT, booting stage (BT) at 47–49 DAT, heading time (HT) at 62–64 DAT, mid grain filling (MGF) at 87–90 DAT, and maturity (MA) at 125–128 DAT. The root and shoot traits were mainly determined at MT, BT, HT, and MGF.

2.2.1. Shoot Dry Weight and Nitrogen Use Efficiency

Five hills of rice plants were collected and separated into leaves, stems (including culms and sheaths), panicles (only at the HT and MA stages), and roots. Leaf area was measured using a leaf area analyzer (Li-3000C, LI-COR, Tucson, AZ, USA). The plant tissues were subsequently placed in an oven at 75 °C until they reached a constant weight, after which they were weighed to determine the dry matter weight. Finally, all the plant tissues were ground into powder to determine the N content using the Kjeldahl method (Foss 8400, Hilleroed, Denmark). The nitrogen use efficiency was calculated according to the method described by Xue et al. [26]. The leaf area duration (LAD) and crop growth rate (CGR) were calculated using the following expressions, respectively:

$$\mathrm{LAD}\left(\mathrm{m}^2\ \mathrm{m}^{-2}\ \mathrm{d}^{-1}\right) = \frac{1}{2}(LAI_1 + LAI_2) \times (t_2 - t_1) \quad (1)$$

$$\mathrm{CGR}\left(\mathrm{g}\ \mathrm{m}^{-2}\ \mathrm{d}^{-1}\right) = \frac{W_2 - W_1}{t_2 - t_1} \quad (2)$$

where LAI_1 and LAI_2 are the first and second measurements of leaf area index, respectively; W_1 and W_2 are the first and second measurements of shoot dry weight, respectively; and t_1 and t_2 represent the first and second day of measurement, respectively.

2.2.2. Root Traits and Non-Structural Carbohydrates (NSC)

Rice plants from five hills were sampled, with a soil volume of 20 cm × 20 cm × 20 cm. The roots within these soil samples were meticulously cleaned using a high-pressure water device. Each plant was divided into shoots and roots. Three root samples were collected to measure root length, using a method outlined by Ju et al. [17]. These root samples were then dried in an oven at 70 °C until reaching a constant weight to determine root dry weight. The remaining roots were weighed at 1 g and placed in a mixture of 25 mL of a 40 mg L^{-1} alpha-naphthylamine (α-NA) solution and 25 mL of a phosphate-buffered solution (pH 7). After standing for 10 min, 2 mL of the solution was taken to determine the remaining amount of α-NA. The degree of reduction in α-NA indicates the level of root oxidation activity (ROA). The preparation of the α-NA solution and the calculation of ROA were carried out using a method described by Ramasamy et al. [30].

The shoot samples, consisting mainly of stems (including culms and sheaths), were gathered for NSC content. The NSC is the sum of starch and soluble sugars. The samples were weighed at 0.1 g and extracted three times with 80% ethanol at 80 °C in a water bath for 30 min each. After extraction, the final volume was adjusted to 50 mL. Subsequently, 2 mL of the resulting extract was combined with 0.5 mL of anthrone and 5 mL of concentrated sulfuric acid. The mixture was immediately placed in a boiling water bath for 1 min and then analyzed using spectrophotometry at 620 nm for soluble sugar measurement. For the extraction of starch content, 2 mL of 9.2 mol L^{-1} perchloric acid was added, followed by a 15-min incubation in a boiling water bath. The remaining procedures were conducted identically to those used for the determination of the soluble sugar content. The calculation method for NSC remobilization was based on the approach described by Zhu et al. [31] and Sun et al. [32].

2.2.3. Root Zeatin (Z) and Zeatin Riboside (ZR) Content

Root and shoot parts were separated from three hills of rice plants for sampling. The roots were meticulously cleaned, dried, rapidly frozen in liquid nitrogen, and subsequently ground into powder to extract Z + ZR. The sample was extracted with an isopropanol solution (composed of isopropanol, ultrapure water, and concentrated hydrochloric acid in a ratio of 2:1:0.002) at 4 °C for 4 h. Following extraction, 10 mL of dichloromethane solution was added, and the mixture was subjected to centrifugation for 5 min (4 °C, relative centrifugal force of 13,000× *g*). The lower layer of the centrifuged solution, which contained the cytokinins, was collected and concentrated by freeze-drying. The extraction method followed the procedure outlined by Zhu et al. [31]. The quantification of Z + ZR in roots was determined using triple quadrupole high-performance liquid chromatography–mass spectrometry (HPLC–MS), as described by Pan et al. [33].

2.2.4. Leaf Photosynthetic Rate

Ten rice leaves from central rows were used for photosynthetic rate measurement.

The photosynthetic rate of the leaves was measured with a gas exchange analyzer (Li-Cor 6800, Tucson, AZ, USA). The measurement was made from 9:00 to 11:00 a.m., when photosynthetically active radiation above the canopy was 1300~1500 μmol m^{-2} s^{-1}.

2.2.5. Ammonia Volatilization

Ammonia volatilization was measured using a vented-chamber method [11]. Each plot contained two chambers made of polyvinyl chloride tubes (30 cm in height and 16 cm in diameter), each fitted with two phosphoglycerol-soaked sponges. The samples were collected daily after fertilization, then at 2–3 day intervals for the following week, and subsequently on a weekly basis until rice harvest. Ammonia absorbed in the lower sponges was extracted with 300 mL of 1.0 mol L^{-1} potassium chloride solution after oscillation for 1 h. The quantity of ammonium in the extracted solutions was determined using the method outlined by Li et al. [11].

2.2.6. Final Harvest

Between October 18 and 20, ten randomly chosen plants (excluding border plants) from the central row of each plot were sampled for measuring the number of panicles, spikelets per panicle, the percentage of filled grains, and the grain weight. The grain yield was determined by harvesting and weighing all plants from a 5 m^2 area in each plot, with subsequent adjustment to a 14% moisture content.

2.3. Statistical Analysis

An analysis of variance (ANOVA) was performed using SAS/STAT software (version 9.2; SAS Institute, Cary, NC, USA). Data visualization was carried out using Origin software (version 2021; Origin Lab, Northampton, MA, USA). To determine statistical significance, the means were compared using the least significant difference (LSD) method at a significance level of $p < 0.05$. The statistical model considered factors such as year, treatment, and their interactions (year × treatment) to identify sources of variation. Additionally, a correlation analysis was conducted and visualized using the R package (version 4.1.1; https://cran.r-project.org, accessed on 9 May 2024). Given that the 0N treatment, which serves as the nitrogen blank treatment, was used solely for calculating NUE, this study primarily focuses on the control, T1, and T2 treatments.

3. Results

3.1. Grain Yield, NUE, and Ammonia Volatilization

As shown in Table 1, compared to that in the control, the grain yield was significantly higher in the T2 treatment, while it was comparable in the T1 treatment. The number of total spikelets was significantly increased in the T2 treatment, but no significant difference was observed between the T1 treatment and the control. The percentage of filled grains and grain weight did not exhibit significant differences among the control, T1, and T2 treatments. It is noteworthy that the percentage of filled grains in 2022 was exceptionally low under the 0N treatment, primarily due to the extremely high temperatures with respect to the stages of heading and early grain filling (Table 1; Figure S1).

Table 1. Grain yield and its components of rice under various N management treatments.

Year/Treatment	Grain Yield (t ha^{-1})	Panicles (m^2)	Spikelets per Panicle	Total Spikelets (10^3/m^2)	Filled Grains (%)	Grain Weight (mg)
2021						
0N	5.84 c	179 c	136 c	24.3 c	90.4 a	26.5 a
Control	9.02 b	284 a	146 b	41.4 b	85.8 b	26.6 a
T1	9.17 b	271 b	157 a	42.5 b	85.5 b	26.4 a
T2	9.76 a	282 a	160 a	45.1 a	85.4 b	26.4 a
2022						
0N	3.56 c	192 c	123 c	23.7 c	69.0 b	24.9 a
Control	7.21 b	297 a	135 b	40.9 b	78.9 a	24.1 b
T1	7.43 b	281 b	147 a	41.8 b	77.3 a	24.2 b
T2	7.96 a	294 a	149 a	45.7 a	77.8 a	24.2 b
Analysis of Variance						
Year (Y)	883 **	65.8 **	124 **	13.6 **	765 **	2646 **
Treatment (T)	21.3 **	1145 **	125 **	775 **	7.87 **	25.0 **
Y × T	4.11 *	NS	NS	NS	75.6 **	21.1 **

0N, N omission; Control, split application of RU; T1, one-time application of a single PCU; T2, one-time application of 70% PCU + 30% RU. Different letters indicate statistical significance at $p = 0.05$ level within the same column. *, significant at $p = 0.05$ level. **, significant at $p = 0.01$ level. NS means not significant at the $p = 0.05$ level.

At the HT and MA stages, the above-ground N accumulation was significantly higher in the T1 and T2 treatments compared to that in the control. At the MT stage, N accumulation showed no significant difference between the control and the T2 treatment, but was higher in the control or in the T2 than in the T1 treatment. Meanwhile, no significant differences were observed among the control, T1, or T2 treatments at the PI stage (Figure 1A,C). Furthermore, from PI to HT, the N accumulation in the T2 treatment was the highest, followed by T1 and then control. From HT to MA, the T1 treatment exhibited the highest N accumulation, followed by T2 or the control. The N accumulation from MT to PI was comparable among these three treatments (Figure 1B,D).

Total N uptake (TNU) was significantly higher in both the T1 and T2 treatments, compared to that in the control, and exhibited no significant difference between T1 and T2 treatments (Table 2). Similar results were observed for N recovery efficiency (RE_N). The T2 treatment exhibited the highest agronomic N use efficiency (AE_N), while the AE_N values for the T1 treatment and the control were comparable. Additionally, the T1 treatment had significantly lower internal N use efficiency (IE_N), whereas no significant difference in IE_N was observed between the control and the T2 treatment.

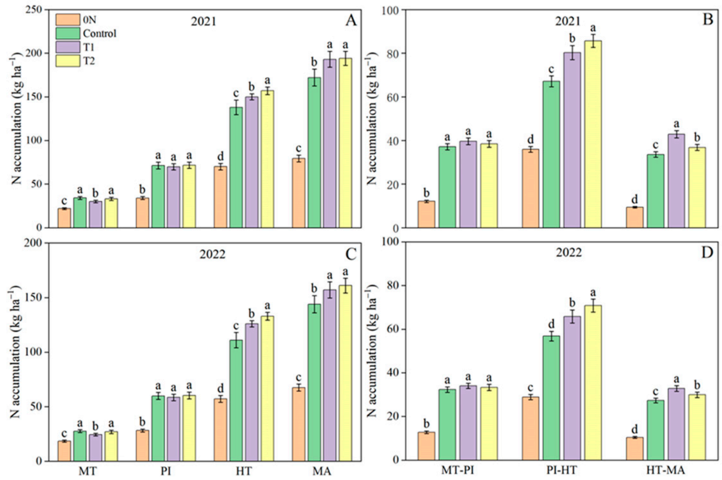

Figure 1. Nitrogen accumulation in 2021 (**A**,**B**) and 2022 (**C**,**D**) during each growth stage under various N management treatments. 0N, N omission; Control, split application of RU; T1, one-time application of a single PCU; T2, one-time application of 70% PCU + 30% RU. MT, mid-tillering; PI, panicle initiation; HT, heading time; MA, maturity. Different letters above the column indicate statistical significance at $p = 0.05$ level.

Table 2. Nitrogen use efficiency of rice under various N management treatments.

Year/Treatments	**TNU (kg hm^{-2})**	**AE_N (kg kg^{-1})**	**IE_N (kg kg^{-1})**	**RE_N (%)**
2021				
0N	79.4 c	-	73.6 a	-
Control	172 b	13.3 b	52.5 b	38.6 b
T1	193 a	13.8 b	47.5 c	47.3 a
T2	194 a	16.2 a	50.1 b	47.8 a
2022				
0N	67.5 c	-	52.7 a	-
Control	144 b	15.4 b	50.1 a	31.9 b
T1	157 a	16.1 b	46.1 b	37.3 a
T2	161 a	18.3 a	50.7 a	39.0 a
Analysis of Variance				
Year (Y)	586 **	47.1 **	66.3 **	137 **
Treatment (T)	1894 **	33.8 **	92.4 **	49.9 **
Y × T	22.9 **	NS	45.8 **	NS

0N, N omission; Control, split application of RU; T1, one-time application of a single PCU; T2, one-time application of 70% PCU + 30% RU. Different letters indicate statistical significance at $p = 0.05$ level within the same column. **, significant at $p = 0.01$ level. NS means not significant at the $p = 0.05$ level.

Throughout the entire growth stages averaged across two years, the total amount of ammonia volatilization from PCU-based N management treatments was significantly lower than that from the control, particularly during the period from PI to HT, which corresponds to 37 to 66 days after transplanting (Figure S2A,B).

3.2. Shoot Dry Matter Weight and Crop Growth Rate

At the MT and PI stages, the shoot dry matter weight was significantly lower in the T1 treatment compared to in the control or the T2 treatment, while no significant difference was observed between the control and the T2 treatment. At the HT stage, the dry matter weight was comparable between T1 and T2 treatments, and it was significantly higher in the T1 or in the T2 than in the control. At maturity, the dry matter weight was highest in the T2 treatment, followed by the T1 treatment, and showed the lowest in the control (Figure 2A,B).

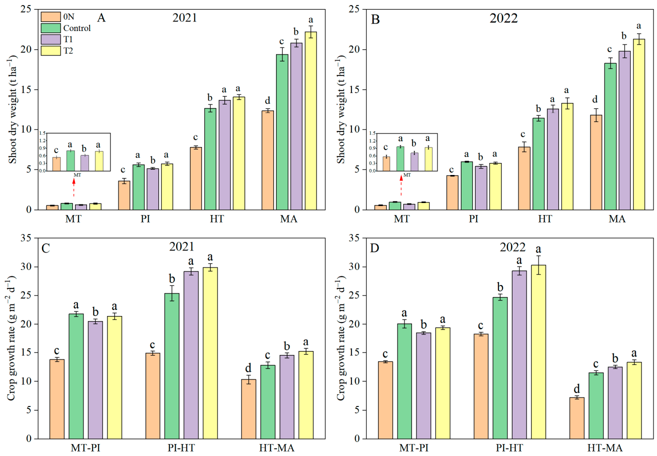

Figure 2. Shoot dry matter weight and crop growth rate during in 2021 (**A**,**C**) and 2022 (**B**,**D**) each growth stage under various N management treatments. 0N, N omission; Control, split application of RU; T1, one-time application of a single PCU; T2, one-time application of 70% PCU + 30% RU. MT, mid-tillering; PI, panicle initiation; HT, heading time; MA, maturity. Different letters above the column indicate statistical significance at p = 0.05 level. The red arrows indicate the magnification of the bar chart for the MT stage.

From MT to PI, the control and T2 treatments showed comparable crop growth rates (CGR), both exhibiting significantly higher CGRs than the T1 treatment. From PI to HT, there was no significant difference in the CGR between T1 and T2 treatments, and both had significantly higher CGRs than the control. From HT to MA, the CGR was significantly higher in either the T1 or the T2 treatments than in the control, with the T2 treatment showing the highest CGR (Figure 2C,D).

3.3. Leaf Area Duration and Photosynthetic Rate

The leaf area duration (LAD) was significantly lower in the T1 treatment than in the control or the T2 during the stage from MT to PI, while it showed similar values between the control and the T2. During the growth periods spanning from PI to HT and from HT to MA, the T1 and T2 treatments showed no significant difference in LAD, but both had remarkably higher LADs than the control (Figure 3A,B).

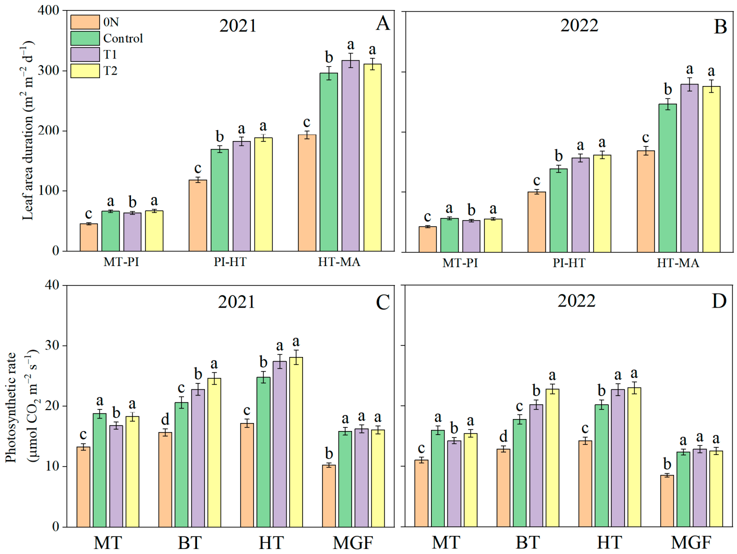

Figure 3. Leaf area duration and photosynthetic rate in 2021 (**A**,**C**) and 2022 (**B**,**D**) under various N management treatments. 0N, N omission; Control, split application of RU; T1, one-time application

of a single PCU; T2, one-time application of 70% PCU + 30% RU. MT, mid-tillering; BT, booting; HT, heading time; MGF, mid-grain filling. Different letters above the column indicate statistical significance at p = 0.05 level.

Compared to that in the control, the photosynthetic rate was significantly lower in the T1 treatment, while it was comparable in the T2 treatment at the MT stage. At the booting stage, both T1 and T2 treatments showed greater photosynthetic rate than the control, with the T2 showing the highest value. At the HT stage, the photosynthetic rate was comparable between the T1 and T2 and was significantly higher in the T1 or in the T2 than in the control. At maturity, there was no significant difference in the photosynthetic rate among the control, T1, or T2 treatments (Figure 3C,D).

MT, mid-tillering; BT, booting stage; HT, heading time; MGF, mid grain filling. The same is as bellow.

3.4. Root Morpho-Physiological Traits

At the MT stage, compared to that in the control, the root dry weight was significantly lower in the T1 treatment, while it was comparable in the T2 treatment. The root dry weight exhibited no significant difference between the T1 and T2 treatments, but both showed significantly higher root dry weight than the control from the BT to MGF. A similar trend was observed in the root length (Figure 4A–D).

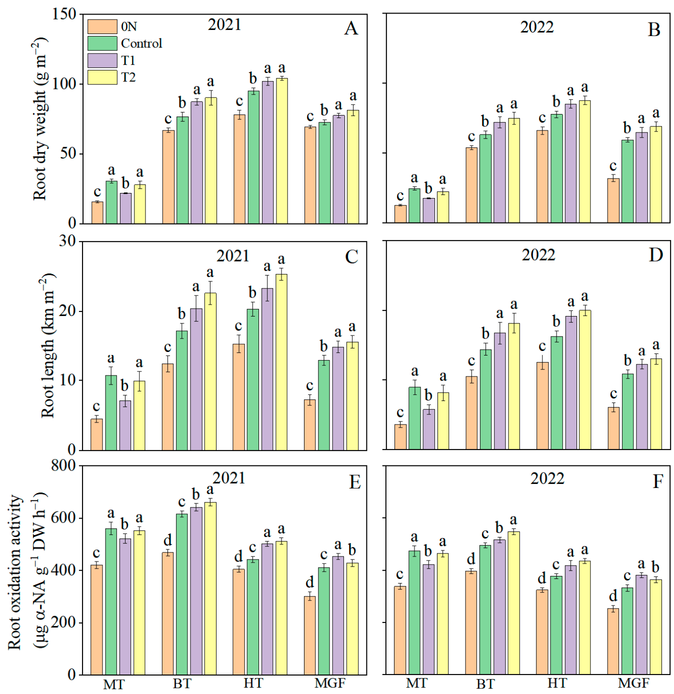

Figure 4. Root dry weight (**A**,**B**), root length (**C**,**D**), and root oxidation activity (**E**,**F**) in 2021 and 2022 under various N management treatments. 0N, N omission; Control, split application of RU; T1, one-time application of a single PCU; T2, one-time application of 70% PCU + 30% RU. MT, mid-tillering; BT, booting; HT, heading time; MGF, mid-grain filling. Different letters above the column indicate statistical significance at p = 0.05 level.

The root oxidation activity (ROA) showed no significant difference between the control and the T2 treatment, but was significantly higher in the control or in the T2 than the T1 treatment at the MT stage. At the BT, the T2 treatment showed a significantly higher ROA compared to the control, while the T1 treatment showed intermediate values. At the HT stage, there was no significant difference in the ROA between the T1 and T2 treatments, but both treatments had higher ROA than the control. At the MGF stage, the ROA was highest in the T1 treatment, followed by the T2 treatment or the control (Figure 4E,F). The root Z + ZR content showed a similar trend to ROA (Figure 5).

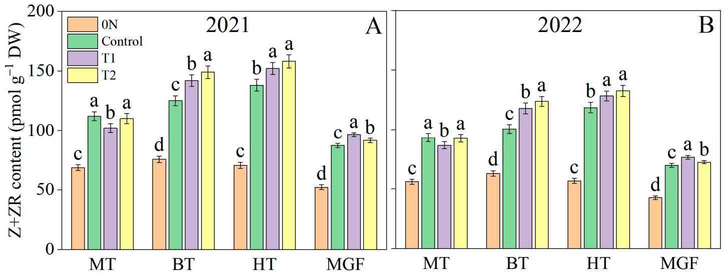

Figure 5. Z + ZR content in roots in 2021 (**A**) and 2022 (**B**) under various N management treatments. 0N, N omission; Control, split application of RU; T1, one-time application of a single PCU; T2, one-time application of 70% PCU + 30% RU. MT, mid-tillering; BT, booting; HT, heading time; MGF, mid-grain filling. Different letters above the column indicate statistical significance at $p = 0.05$ level.

3.5. NSC Remobilization and Harvest Index

The T2 treatment showed higher NSC accumulation at heading compared to the control, with the T1 treatment falling in between. At maturity, the NSC accumulation in the T1 treatment significantly surpassed that of the control or the T2 treatment, with the control exhibiting the lowest value. Compared to the control, the T1 treatment had significantly lower NSC remobilization, with the T2 treatment showing no significant difference. The harvest index followed a trend similar to that of NSC remobilization (Table 3).

Table 3. NSC remobilization and harvest index in rice under various N management treatments.

Year/Treatment	NSC Accumulation in the Stem (g/m^2)		NSC Remobilization (%)	Harvest Index
	Heading	Maturity		
2021				
0N	154 d	71.5 d	53.6 a	0.490 a
Control	210 c	104 c	50.5 b	0.465 b
T1	228 b	129 a	43.4 c	0.441 c
T2	240 a	115 b	52.1 b	0.460 b
2022				
0N	146 d	93.5 d	36.0 c	0.302 c
Control	198 c	105 c	47.0 a	0.416 a
T1	218 b	132 a	39.3 b	0.399 b
T2	232 a	124 b	46.6 a	0.413 a
Analysis of Variance				
Year (Y)	291 **	123 **	62 **	601 **
Treatment (T)	662 **	251 **	44.7 **	37 **
Y × T	NS	18 **	35.8 **	114 **

0N, N omission; Control, split application of RU; T1, one-time application of a single PCU; T2, one-time application of 70% PCU + 30% RU. Different letters indicate statistical significance at $p = 0.05$ level within the same column. **, significant at $p = 0.01$ level. NS means not significant at the $p = 0.05$ level.

3.6. Correlation Analysis

The root traits, particularly the ROA and root Z + ZR content during each growth stage exhibited significant or highly significant positive correlations with N accumulation, leaf area duration, crop growth rate, and leaf photosynthetic rate at the corresponding stages. Grain yield, N recovery efficiency, total N uptake, and total spikelets were highly correlated with ROA and root Z + ZR content during booting (Figure 6).

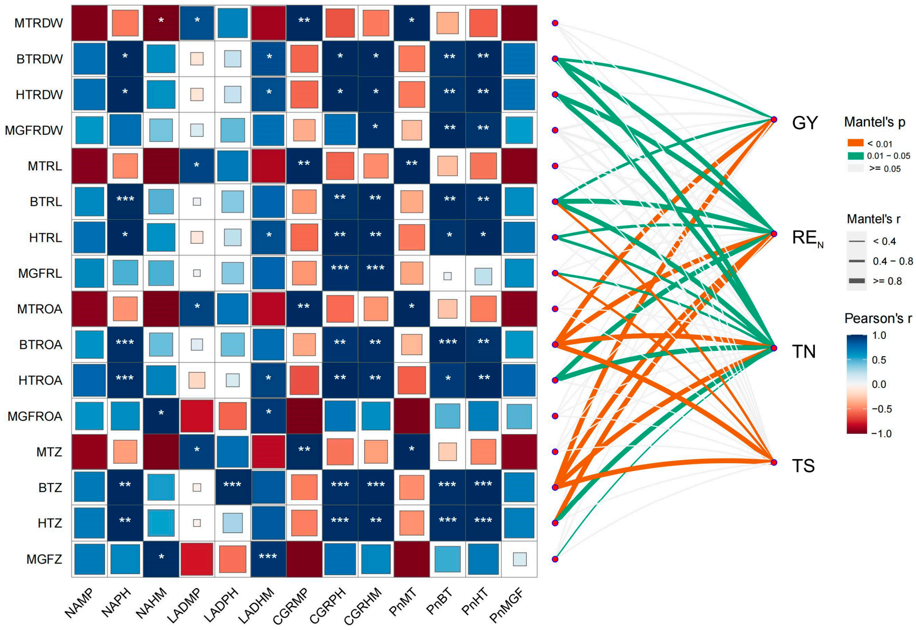

Figure 6. The correlations of main root and shoot traits with grain yield, N recovery efficiency, total N uptake, and total spikelets. GY, grain yield; RE_N, N recovery efficiency; TN, total N uptake; TS, total spikelets; MTRDW, root dry weight at mid-tillering; BTRDW, root dry weight at booting; HTRDW, root dry weight at heading; MGFRDW, root dry weight at mid grain filling; MTRL, root length at mid-tillering; BTRL, root length at booting; HTRL, root length at heading; MGFRL, root length at mid grain filling; MTROA, root oxidation activity at mid-tillering; BTROA, root oxidation activity at booting; MGFROA, root oxidation activity at mid grain filling; MTZ, root Z + ZR content at mid-tillering; BTZ, root Z + ZR content at booting; HTZ, root Z + ZR content at heading; MGFZ, root Z + ZR content at mid grain filling; NAMP, N accumulation from mid-tillering to panicle initiation; NAPH, N accumulation from panicle initiation to heading; NAHM, N accumulation from heading to maturity; LADMP, leaf area duration from mid-tillering to panicle initiation; LADPH, leaf area duration from panicle initiation to heading; LADHM, leaf area duration from heading to maturity; CGRMP, crop growth rate from mid-tillering to panicle initiation; CGRPH, crop growth rate from panicle initiation to heading; CGRHM, crop growth rate from heading to maturity; PnMT, leaf photosynthetic rate at mid-tillering; PnBT, leaf photosynthetic rate at heading; PnHT, leaf photosynthetic rate at booting; PnMGF, leaf photosynthetic rate at mid grain filling. *, **, ***, significant at p = 0.05, 0.01, and 0.001, respectively.

4. Discussion

In China's rice production, excessive N fertilizer application is a common issue, leading to a high yield but a low use efficiency [4,34,35]. To address this problem, agronomists have developed various N management strategies, particularly the introduction of PCU, which not only decreases N losses but also provides environmental and economic benefits [10–15]. However, there is still controversy regarding whether PCU could effectively increase rice grain yield and NUE, with this uncertainty primarily stemming from variations in application methods, rice varieties, and environmental factors [14]. In this study, we observed that the T2 treatment (one-time application of 70% PCU + 30% RU) significantly increased grain yield, while the T1 treatment (one-time application of a single PCU) achieved a comparable level, compared to the control (split application of RU) (Table 1). Some studies suggest that the yield increase from PCU-based N management is due to improved grain filling rate and grain weight [13,14,16]. However, in this study, the primary factor influencing yield variation was the total number of spikelets. Briefly, the T2 treatment demonstrated a greater total spikelet number, while the T1 treatment maintained an unchanged value, in relation to the control (Table 1). It is generally believed that the panicle number and the spikelets per panicle are the two key factors that determine the total spikelet number [36,37]. We observed that both the T1 and T2 treatments exhibited a higher number of spikelets per panicle compared to the control. However, the panicle number in the T2 treatment was comparable to the control, while it was lower in the T1 treatment (Table 1). These results suggest that a combination of PCU and RU can effectively overcome the inhibition of the tiller number observed in the sole application of PCU. This approach finally converts the advantage of a higher number of spikelets per panicle into the superiority in total spikelets. In addition, high temperatures during the heading and early grain-filling stages in 2022 resulted in significantly lower yields across all treatments, compared to 2021, especially under the 0N treatment (Table 1). This result also indicates that nitrogen application can mitigate the adverse effects of high temperatures. Nevertheless, the consistent trends observed over both years demonstrate the stability of the yield-enhancing effect of PCU combined with RU across different growing seasons.

Notably, both T1 and T2 treatments substantially improved the N recovery use efficiency (RE_N), compared to the control. Numerous studies consistently conclude that the N uptake during booting is crucial for determining the total N uptake and RE_N in rice [38,39]. In the present study, we also observed that the PCU management, especially the T2 treatment, possessed a distinct advantage in N accumulation from the PI to HT stages (Figure 2C,D). Moreover, compared to the PCU-based N treatments, the split application of RU resulted in higher ammonia volatilization, especially during the stages from PI to HT (Figure S2). Therefore, we propose that the enhanced nitrogen use efficiency (NUE) in PCU management treatments is primarily due to reduced nitrogen loss, which also mitigates environmental risks. Moreover, it is widely recognized that increased N uptake can have a negative impact on N utilization, leading to a trade-off between N uptake and N utilization [40,41]. The present study showed that the T1 treatment significantly decreased the internal N use efficiency (IE_N) in comparison with the control, while the T2 treatment showed similar values to the control (Table 2). Furthermore, both T1 and T2 absorbed more N than the control after heading (Figure 1C,D). There are reports showing that excessive N uptake, particular post-anthesis N uptake, may lead to unfavorable senescence, retarding the assimilate remobilization, thereby decreasing the IE_N [42–44]. It is further hypothesized that rice varieties with a larger sink size can mitigate the negative effect of high N uptake by enhancing the remobilization of photo-assimilates from vegetative organs, which are closely associated with IE_N [45,46]. In summary, we conclude that a reduced application rate of PCU, combined with RU substitution, is an effective strategy to obtain higher N uptake without sacrificing its utilization.

Prior to this study, little was known about the mechanism underlying the improvements in grain yield and NUE under a combination of PCU with RU. We observed that root traits, particularly the ROA and root Z + ZR content during the BT, contributed significantly to the grain yield and N uptake. Furthermore, the T2 treatment had a unique advantage in these traits, which were also highly correlated with the number of total spikelets (Figure 6). It is reported that the booting stage (BT) is critical for the maximum tiller number and essential for spikelet formation in rice [47,48]. Hence, reducing tiller mortality and promoting more spikelet production are fundamental to achieving a higher number of total spikelets [48–51]. However, this process requires more photo-assimilates to provide a sufficient energy basis [51]. The present results show that the T2 treatment exhibited a greater leaf photosynthetic rate, leaf area duration, and crop growth rate compared to the control at the BT (Figures 2 and 3). They were very positively correlated with the ROA and root Z + ZR content (Figure 6). There are reports suggesting that maintaining a higher root activity, e.g., ROA and root Z + ZR content, is conductive to shaping the N gradient in rice leaves, thereby obtaining greater photosynthetic production [42,52]. Our earlier work has also demonstrated that rice varieties with a greater ROA and higher root Z + ZR content can induce the expression of genes related to nitrogen transport [52,53]. Overall, we conclude that the combination of PCU and RU can significantly increase the content of Z + ZR in roots, particularly during booting. This enhancement can promote shoot photosynthetic production, providing the material basis for the formation of more spikelets. Additionally, a greater amount of photo-assimilates can feed back to the roots, enhancing root activity and promoting N uptake from the soil. This process is crucial for the formation of a higher total number of spikelets and greater N uptake.

It should be noted that, compared to the control, the T1 treatment exhibited a better performance in ROA and root Z + ZR content at the MGF stage, followed by the T2 and control. There is evidence suggesting that higher ROA and cytokinin content in rice roots during the mid or late grain-filling stages are conducive to promoting grain filling and consequently increasing the filled-grain percentage [23]. In the present study, the filled grain percentage show no significant difference among these three treatments. The results imply that, during the MGF stage, the root and shoot development in the PCU treatments may not be well-coordinated. In other words, the PCU treatments fail to translate the advantage of root activity into the superiority of the filled-grain percentage. Some observations report that the leaf photosynthetic rate is also constrained by the environmental temperature and light intensity [54–56]. Typically, as rice progresses into the MGF stage, there is a considerable decrease in these two parameters, limiting photosynthetic productivity [55,56]. We herein observed that the photosynthetic rate in PCU treatments was only higher than the control at HT, while no significant differences were observed among treatments at the MGF stage (Figure 3C,D). Our observation showed that NSC remobilization was pronounced lower in the T1 treatment, in comparison with the control. In contrast, the T2 treatment achieved an NSC remobilization comparable to the control (Table 3). Therefore, we argue that during the MGF stage, the NSC remobilization from vegetative organs to grains would be very important for improving grain filling. It is proposed that NSC remobilization is very positively correlated with the harvest index (HI), thus affecting the IE_N [57–59]. Moreover, the HI showed a similar trend to NSC remobilization (Table 3). Subsequent results showed that the T2 treatment, in conjunction with competitive NSC remobilization and higher photosynthesis at heading, led to a greater crop growth rate during grain filling, and, therefore, a higher dry matter weight at maturity (Figures 2 and 3; Table 3). This process combined with a competitive HI substantially increased grain yield.

However, previous studies have demonstrated that the HI of rice plants varies widely, ranging from 0.17 to 0.63, indicating a significant potential for enhancing the IE_N by improving the HI [59,60]. Our observations showed that the HI averaged 0.432 over two years among the control, T1, and T2 treatments (Table 3). Typically, rice plants can sustain an HI of around 0.5 or higher to achieve high yields without lodging. Consequently, there is substantial potential to enhance the HI in our study, especially regarding the T2 treatment. Further research is needed to explore cultivation practices that integrate the combination of PCU with RU, aiming to enhance the HI and achieve the triple goals of improving grain yield, N uptake, and N utilization efficiency.

5. Conclusions

The combination of PCU and RU applied one time as basal fertilizer could synergistically increase rice yield and nitrogen uptake. This was mainly due to the increased number of total spikelets, higher N accumulation during booting, and dry matter weight at maturity, while maintaining an unchanged HI. Higher root and shoot activities during booting and comparable NSC remobilization under this combination were responsible for these improvements. This combination not only allowed for more N uptake without decreasing the IE_N by maintaining a competitive HI but also mitigated environmental risks by reducing ammonia volatilization. Further research is needed to enhance the HI under this combination, thus achieving triple goals of improving the grain yield, N uptake, and NUE.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/agronomy14071585/s1, Table S1: The physicochemical properties of the soil in the paddy field before transplanting; Figure S1: Temperature (A), Precipitation (B), and sunshine hours (C) during the rice growing season in 2021 and 2022 at the experiment site; Figure S2: NH3 volatilization rate (A) and accumulative NH3 emission (B) during the whole growth stages averaged across two study years under various N management treatments.

Author Contributions: Conceptualization, W.Z., J.G., K.Z., Z.W. and J.Y.; validation, J.F.; investigation, Z.S. and Y.X.; data curation, J.F., Y.Z., Z.S. and Y.X.; investigation and writing—original draft preparation, R.X.; writing—review and editing, K.Z. and J.Y.; supervision, K.Z. and J.Y.; project administration, J.Y.; funding acquisition, K.Z. and J.Y. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the National Natural Science Foundation of China (32272198; 32071943; 32301930), the Yangzhou University Student Science and Technology Innovation Program (XCX20230645), and the Priority Academic Program Development of Jiangsu Higher Education Institutions (PAPD).

Data Availability Statement: The data presented in this study are available upon request from the corresponding author.

Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

AE_N: agronomic N use efficiency; BT: booting; CGR: crop growth rate; HI: harvest index; HT: heading time; IE_N: internal N use efficiency; LAD: leaf area duration; MA: maturity; MGF: mid-grain filling; MT: mid-tillering stage; NSC: non-structural carbohydrate remobilization; PCU: polymer-coated urea; PI: panicle initiation; RE_N: N recovery efficiency; ROA: root oxidation activity; RU: rapid-release urea; TNU, total N uptake; Z + ZR: zeatin + zeatin rboside.

References

1. Bin Rahman, A.R.; Zhang, J. Trends in rice research: 2030 and beyond. *Food. Energy Secur.* **2023**, *12*, e390. [CrossRef]
2. Ahmed, N.; Turchini, G.M. The evolution of the blue-green revolution of rice-fish cultivation for sustainable food production. *Sustain. Sci.* **2021**, *16*, 1375–1390. [CrossRef]
3. Liu, Y.S.; Wang, J.Y.; Long, H.L. Analysis of arable land loss and its impact on rural sustainability in Southern Jiangsu Province of China. *J. Environ. Manag.* **2010**, *91*, 646–653. [CrossRef] [PubMed]
4. Cai, S.; Zhao, X.; Pittelkow, C.M.; Fan, M.; Zhang, X.; Yan, X. Optimal nitrogen rate strategy for sustainable rice production in China. *Nature* **2023**, *615*, 73–79. [CrossRef] [PubMed]
5. Wang, Y.; Lu, Y. Evaluating the potential health and economic effects of nitrogen fertilizer application in grain production systems of China. *J. Clean. Prod.* **2020**, *264*, 121635. [CrossRef]
6. Ji, Y.; Liu, G.; Ma, J.; Zhang, G.; Xu, H.; Yagi, K. Effect of controlled-release fertilizer on mitigation of N_2O emission from paddy field in South China: A multi-year field observation. *Plant Soil* **2013**, *371*, 473–486. [CrossRef]
7. Sun, H.; Zhou, S.; Zhang, J.; Zhang, X.; Wang, C. Effects of controlled-release fertilizer on rice grain yield, nitrogen use efficiency, and greenhouse gas emissions in a paddy field with straw incorporation. *Field Crops Res.* **2020**, *253*, 107814. [CrossRef]
8. Ishfaq, M.; Akbar, N.; Zulfiqar, U.; Ali, N.; Jabran, K.; Nawaz, M.; Farooq, M. Influence of nitrogen fertilization pattern on productivity, nitrogen use efficiencies, and profitability in different rice production systems. *J. Soil Sci. Plant Nut.* **2021**, *21*, 145–161. [CrossRef]
9. Chen, Y.; Peng, J.; Wang, J.; Fu, P.; Hou, Y.; Zhang, C.; Fahad, S.; Peng, S.; Cui, K.; Nie, L.; et al. Crop management based on multi-split topdressing enhances grain yield and nitrogen use efficiency in irrigated rice in China. *Field Crops Res.* **2015**, *184*, 50–57. [CrossRef]
10. Xu, D.; Zhu, Y.; Zhu, H.; Hu, Q.; Liu, G.; Wei, H.; Zhang, H. Effects of a one-time application of controlled-release nitrogen fertilizer on yield and nitrogen accumulation and utilization of late japonica rice in China. *Agriculture* **2021**, *11*, 1041. [CrossRef]
11. Li, P.; Lu, J.; Wang, Y.; Wang, S.; Hussain, S.; Ren, T.; Cong, R.; Li, X. Nitrogen losses, use efficiency, and productivity of early rice under controlled-release urea. *Agric. Ecosyst. Environ.* **2018**, *251*, 78–87. [CrossRef]
12. Geng, J.; Ma, Q.; Zhang, M.; Li, C.; Liu, Z.; Lyu, X.; Zheng, W. Synchronized relationships between nitrogen release of controlled release nitrogen fertilizers and nitrogen requirements of cotton. *Field Crops Res.* **2015**, *184*, 9–16. [CrossRef]
13. Ma, Q.; Wang, M.; Zheng, G.; Yao, Y.; Tao, R.; Zhu, M.; Ding, J.; Li, C.; Guo, W.; Zhu, X. Twice-split application of controlled-release nitrogen fertilizer met the nitrogen demand of winter wheat. *Field Crops Res.* **2021**, *267*, 108163. [CrossRef]
14. Zhang, Y.; Ren, W.; Zhu, K.; Fu, J.; Wang, W.; Wang, Z.; Gu, J.; Yang, J. Substituting readily available nitrogen fertilizer with controlled-release nitrogen fertilizer improves crop yield and nitrogen uptake while mitigating environmental risks: A global meta-analysis. *Field Crops Res.* **2024**, *306*, 109221. [CrossRef]
15. Ke, J.; Xing, X.; Li, G.; Ding, Y.; Dou, F.; Wang, S.; Liu, Z.; Tang, S.; Ding, C.; Chen, L. Effects of different controlled-release nitrogen fertilisers on ammonia volatilisation, nitrogen use efficiency and yield of blanket-seedling machine-transplanted rice. *Field Crops Res.* **2017**, *205*, 147–156. [CrossRef]

16. Zhang, W.; Yu, J.; Xu, Y.; Wang, Z.; Liu, L.; Zhang, H.; Gu, J.; Zhang, J.; Yang, J. Alternate wetting and drying irrigation combined with the proportion of polymer-coated urea and conventional urea rates increases grain yield, water and nitrogen use efficiencies in rice. *Field Crops Res.* **2021**, *268*, 108165. [CrossRef]
17. Ju, C.; Buresh, R.J.; Wang, Z.; Zhang, H.; Liu, L.; Yang, J.; Zhang, J. Root and shoot traits for rice varieties with higher grain yield and higher nitrogen use efficiency at lower nitrogen rates application. *Field Crops Res.* **2015**, *175*, 47–55. [CrossRef]
18. Yang, J.C.; Zhang, H.; Zhang, J.H. Root morphology and physiology in relation to the yield formation of rice. *J. Integr. Agric.* **2012**, *11*, 920–926. [CrossRef]
19. Xu, G.W.; Lu, D.K.; Wang, H.Z.; Li, Y. Morphological and physiological traits of rice roots and their relationships to yield and nitrogen utilization as influenced by irrigation regime and nitrogen rate. *Agric. Water Manag.* **2018**, *203*, 385–394. [CrossRef]
20. Xin, W.; Liu, H.; Zhao, H.; Wang, J.; Zheng, H.; Jia, Y.; Yang, L.; Wang, X.; Li, J.; Li, X.; et al. The response of grain yield and root morphological and physiological traits to nitrogen levels in paddy rice. *Front. Plant Sci.* **2021**, *12*, 713814. [CrossRef]
21. Xu, G.W.; Jiang, M.M.; Lu, D.K.; Wang, H.Z.; Chen, M.C. Nitrogen forms affect the root characteristic, photosynthesis, grain yield, and nitrogen use efficiency of rice under different irrigation regimes. *Crop Sci.* **2020**, *60*, 2594–2610. [CrossRef]
22. Chen, L.; Wang, G.; Chen, P.; Zhu, H.; Wang, S.; Ding, Y. Shoot-root communication plays a key role in physiological alterations of rice (*Oryza sativa*) under iron deficiency. *Front. Plant Sci.* **2018**, *9*, 757. [CrossRef]
23. Yu, S.M.; Lo, S.F.; Ho, T.H.D. Source–sink communication: Regulated by hormone, nutrient, and stress cross-signaling. *Trends Plant Sci.* **2015**, *20*, 844–857. [CrossRef] [PubMed]
24. Zahir, Z.A.; Asghar, H.N.; Arshad, M. Cytokinin and its precursors for improving growth and yield of rice. *Soil Biol. Biochem.* **2021**, *33*, 405–408. [CrossRef]
25. Ping, J.L.; Ferguson, R.B.; Dobermann, A. Site-specific nitrogen and plant density management in irrigated maize. *Agron. J.* **2008**, *100*, 1193–1204. [CrossRef]
26. Baral, B.R.; Pande, K.R.; Gaihre, Y.K.; Baral, K.R.; Sah, S.K.; Thapa, Y.B.; Singh, U. Real-time nitrogen management using decision support-tools increases nitrogen use efficiency of rice. *Nutr. Cycl. Agroecosyst.* **2021**, *119*, 355–368. [CrossRef]
27. Zhou, W.; Lv, T.; Yang, Z.; Wang, T.; Fu, Y.; Chen, Y.; Hu, B.; Ren, W. Morphophysiological mechanism of rice yield increase in response to optimized nitrogen management. *Sci. Rep.* **2017**, *7*, 17226. [CrossRef] [PubMed]
28. Xiong, Q.; Tang, G.; Zhong, L.; He, H.; Chen, X. Response to nitrogen deficiency and compensation on physiological characteristics, yield formation, and nitrogen utilization of rice. *Front. Plant Sci.* **2018**, *9*, 1075. [CrossRef]
29. Beillouin, D.; Trépos, R.; Gauffreteau, A.; Jeuffroy, M.H. Delayed and reduced nitrogen fertilization strategies decrease nitrogen losses while still achieving high yields and high grain quality in malting barley. *Eur. J. Agron.* **2018**, *101*, 174–182. [CrossRef]
30. Ramasamy, S.; Ten Berge, H.F.M.; Purushothaman, S. Yield formation in rice in response to drainage and nitrogen application. *Field Crops Res.* **1997**, *51*, 65–82. [CrossRef]
31. Zhu, K.; Yan, J.; Shen, Y.; Zhang, W.; Xu, Y.; Wang, Z.; Yang, J. Deciphering the morpho–physiological traits for high yield potential in nitrogen efficient varieties (NEVs): A japonica rice case study. *J. Integr. Agric.* **2022**, *21*, 947–963. [CrossRef]
32. Li, G.; Hu, Q.; Shi, Y.; Cui, K.; Nie, L.; Huang, J.; Peng, S. Low nitrogen application enhances starch-metabolizing enzyme activity and improves accumulation and translocation of non-structural carbohydrates in rice stems. *Front. Plant Sci.* **2018**, *9*, 1128. [CrossRef]
33. Pan, X.; Welti, R.; Wang, X. Quantitative analysis of major plant hormones in crude plant extracts by high-performance liquid chromatography-mass spectrometry. *Nat. Protoc.* **2010**, *5*, 986–992. [CrossRef] [PubMed]
34. Ding, W.; Xu, X.; He, P.; Ullah, S.; Zhang, J.; Cui, Z.; Zhou, W. Improving yield and nitrogen use efficiency through alternative fertilization options for rice in China: A meta-analysis. *Field Crops Res.* **2018**, *227*, 11–18. [CrossRef]
35. Qiao, J.; Yang, L.; Yan, T.; Xue, F.; Zhao, D. Nitrogen fertilizer reduction in rice production for two consecutive years in the Taihu Lake area. *Agric. Ecosyst. Environ.* **2012**, *146*, 103–112. [CrossRef]
36. Kamiji, Y.; Yoshida, H.; Palta, J.A.; Sakuratani, T.; Shiraiwa, T. N applications that increase plant N during panicle development are highly effective in increasing spikelet number in rice. *Field Crops Res.* **2011**, *122*, 242–247. [CrossRef]
37. Sui, B.; Feng, X.; Tian, G.; Hu, X.; Shen, Q.; Guo, S. Optimizing nitrogen supply increases rice yield and nitrogen use efficiency by regulating yield formation factors. *Field Crops Res.* **2013**, *150*, 99–107. [CrossRef]
38. Inamura, T.; Hamada, H.; Iida, K.; Umeda, M. Correlation of the amount of nitrogen accumulated in the aboveground biomass at panicle initiation and nitrogen content of soil with the nitrogen uptake by lowland rice during the period from panicle initiation to heading. *Plant Prod. Sci.* **2003**, *6*, 302–308. [CrossRef]
39. Ye, C.; Ma, H.; Huang, X.; Xu, C.; Chen, S.; Chu, G.; Zhang, X.; Wang, D. Effects of increasing panicle-stage N on yield and N use efficiency of indica rice and its relationship with soil fertility. *Crop J.* **2022**, *10*, 1784–1797. [CrossRef]
40. Ogawa, T.; Oikawa, S.; Hirose, T. Nitrogen-utilization efficiency in rice: An analysis at leaf, shoot, and whole-plant level. *Plant Soil* **2016**, *404*, 321–344. [CrossRef]
41. Huang, M.; Fan, L.; Chen, J.; Jiang, L.; Zou, Y. Continuous applications of biochar to rice: Effects on nitrogen uptake and utilization. *Sci. Rep.* **2018**, *8*, 11461. [CrossRef] [PubMed]
42. Feller, U.; Fischer, A. Nitrogen metabolism in senescing leaves. *Crit. Rev. Plant Sci.* **1994**, *13*, 241–273. [CrossRef]
43. Gaju, O.; Allard, V.; Martre, P.; Snape, J.W.; Heumez, E.; LeGouis, J.; Moreau, D.; Bogard, M.; Griffiths, S.; Hubbart, S.; et al. Identification of traits to improve the nitrogen-use efficiency of wheat genotypes. *Field Crops Res.* **2011**, *123*, 139–152. [CrossRef]

44. Huang, L.; Yang, D.; Li, X.; Peng, S.; Wang, F. Coordination of high grain yield and high nitrogen use efficiency through large sink size and high post-heading source capacity in rice. *Field Crops Res.* **2019**, *233*, 49–58. [CrossRef]
45. Tegeder, M.; Masclaux-Daubresse, C. Source and sink mechanisms of nitrogen transport and use. *New Phytol.* **2018**, *217*, 35–53. [CrossRef] [PubMed]
46. Chen, K.E.; Chen, H.Y.; Tseng, C.S.; Tsay, Y.F. Improving nitrogen use efficiency by manipulating nitrate remobilization in plants. *Nat. Plants* **2020**, *6*, 1126–1135. [CrossRef] [PubMed]
47. Badshah, M.A.; Naimei, T.; Zou, Y.; Ibrahim, M.; Wang, K. Yield and tillering response of super hybrid rice Liangyoupeijiu to tillage and establishment methods. *Crop J.* **2014**, *2*, 79–86. [CrossRef]
48. Patel, R.; Mohapatra, P.K. Regulation of spikelet development in rice by hormones. *J. Exp. Bot.* **1992**, *43*, 257–262. [CrossRef]
49. Del Moral, M.G.; Del Moral, L.G. Tiller production and survival in relation to grain yield in winter and spring barley. *Field Crops Res.* **1995**, *44*, 85–93. [CrossRef]
50. Pasuquin, E.; Lafarge, T.; Tubana, B. Transplanting young seedlings in irrigated rice fields: Early and high tiller production enhanced grain yield. *Field Crops Res.* **2008**, *105*, 141–155. [CrossRef]
51. Liu, L.; Xia, Y.; Liu, B.; Chang, C.; Xiao, L.; Shen, J.; Tang, L.; Cao, W.; Zhu, Y. Individual and combined effects of jointing and booting low-temperature stress on wheat yield. *Eur. J. Agron.* **2020**, *113*, 125989. [CrossRef]
52. Zhu, K.; Xu, Y.; Sun, Z.; Zhang, Y.; Zhang, W.; Xu, Y.; Gu, J.; Zhang, H.; Wang, Z.; Liu, L.; et al. Post-anthesis dry matter production and leaf nitrogen distribution are associated with root-derived cytokinins gradient in rice. *J. Integr. Agric.* 2024; *in press*. [CrossRef]
53. Sakakibara, H. Cytokinin biosynthesis and transport for systemic nitrogen signaling. *Plant J.* **2021**, *105*, 421–430. [CrossRef] [PubMed]
54. Moore, C.E.; Meacham-Hensold, K.; Lemonnier, P.; Slattery, R.A.; Benjamin, C.; Bernacchi, C.J.; Lawson, T.; Cavanagh, A.P. The effect of increasing temperature on crop photosynthesis: From enzymes to ecosystems. *J. Exp. Bot.* **2021**, *72*, 2822–2844. [CrossRef] [PubMed]
55. Kläring, H.P.; Krumbein, A. The effect of constraining the intensity of solar radiation on the photosynthesis, growth, yield and product quality of tomato. *J. Agron. Crop Sci.* **2013**, *199*, 351–359. [CrossRef]
56. Zhou, J.; Li, P.; Wang, J. Effects of light intensity and temperature on the photosynthesis characteristics and yield of lettuce. *Horticulturae* **2022**, *8*, 178. [CrossRef]
57. Yang, J.; Zhang, J. Simultaneously improving grain yield and water and nutrient use efficiencies by enhancing the harvest index in rice. *Crop Environ.* **2023**, *3*, 157–164. [CrossRef]
58. Dwivedi, S.K.; Kumar, S.; Natividad, M.A.; Quintana, M.R.; Chinnusamy, V.; Henry, A. Disentangling the roles of plant water status and stem carbohydrate remobilization on rice harvest index under drought. *Rice* **2023**, *16*, 14. [CrossRef] [PubMed]
59. Onoda, K.; Ohta, R.; Peter, M.A.; Edifor, R.; Hamajima, M.; Mae, A.; Murai, A.; Kondo, M.; Sekiya, N. Post-heading accumulation of nonstructural carbohydrates and nitrogen in rice (*Oryza sativa* L.) roots. *Field Crops Res.* **2024**, *315*, 109478. [CrossRef]
60. Yuan, L. Progress in super-hybrid rice breeding. *Crop J.* **2017**, *5*, 100–102. [CrossRef]