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Mechanical Stubble Righting after the Mechanical Harvest of Primary Rice Improves the Grain Yield of Ratooning Rice

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Abstract: Ratooning rice is an essential rice-planting method. However, mechanical harvesting of the primary rice crop, while increasing efficiency, can negatively affect the yield of ratooning rice. Therefore, it is crucial to find ways to improve the grain yield of ratooning rice after the mechanical harvest of the primary rice. A two-year field experiment was conducted; the grain yield of ratooning rice was assessed by stubble righting after mechanical harvesting of primary rice. The study used two popular rice cultivars, YLiangyou911 and Kenliangyou801, as experimental materials. The experimental treatments included three groups: one without righting after rolling rice stubble (CK), another with mechanized righting after rolling rice stubble (T1), and a third one without rolling rice stubble by the machine (T2). The results of the study demonstrate that stubble righting after the mechanical harvest of primary rice (T1) had a substantial impact on the grain yield of ratooning rice. It led to grain yields similar to ratooning rice without mechanical rolling of the rice stubble (T2) and significantly outperformed the treatment without stubble righting after the mechanical harvest of primary rice (CK). The study observed significant effects of the year of the experiment (Y), the treatment applied (T), and the interaction between year and treatment (Y×T) on grain yield. Additionally, the treatment showed a significant influence on the yield components. Specifically, in 2021, the T1 and T2 treatments showed remarkable grain yield increases in YLiangyou911 by 107.41% and 147.97%, respectively, compared to CK. For Kenliangyou801 in 2021, the T1 and T2 treatments also resulted in notable improvements in grain yield by 45.85% and 114.26%, respectively. Similarly, in 2022, the grain yield increased by 6.99% for T1 and 23.87% for T2 in YLiangyou911, and by 77.23% for T1 and 187.13% for T2 in Kenliangyou801, compared to CK. The study also detected enhancements in several aspects, including biomass accumulation, solar radiation and photosynthetic characteristics, antioxidant response and nitrogen metabolism, and bud-regeneration capacity due to T1 and T2 treatments. Furthermore, correlation analysis was conducted to assess the relationship between grain yield and the investigated parameters. In conclusion, stubble righting after the mechanical harvest of primary rice resulted in significantly improved grain yield for ratooning rice. This improvement can be attributed to enhanced biomass accumulation, solar radiation and photosynthetic characteristics, antioxidant response and nitrogen metabolism, and increased bud-regeneration capacity.

Keywords: mechanical harvest; ratooning rice; stubble righting; grain yield; biomass; regeneration capacity

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1. Introduction

The global demand for rice is enormous, and China is a major consumer and producer of rice. Rice production must be increased for society to maintain a stable food structure [1]. China's rice production has increased because of technological and economic advancements.

Still, it now confronts the challenges of rural labor shortages and inefficient cultivation practices that hinder its contribution to global rice production [2]. Reducing cultivation costs is essential for sustainable agricultural development on limited land. Increasing the frequency of rice harvesting on existing land, and thereby increasing the rice yields, is a viable strategy [3]. Ratooning rice permits two harvests in a single season, saves time and labor, and increases yield and quality, which is crucial for food security in China [4,5]. Ratooning rice is an ancient practice with the potential to contribute to the global rice supply substantially. Ratooning rice cultivation has become an essential extension in China [6,7].

Ratooning rice is more productive and profitable than conventional rice [8]. Ratooning rice grows from the sprouts of stubble and has distinct growth characteristics from the primary crop. During the ratoon season, rice has a shorter growing period than the primary season, making it suitable for replacing double cropping in resource-limited areas [9]. Ratooning rice minimizes land preparation, sowing, and transplanting, reducing labor, water, and seed expenses. Labor and seed inputs for ratooning rice cultivation decreased by 28.5% and 51.5%, respectively, compared to double-cropped rice. Ratooning rice is popular among farmers and its annual yield is greater than single-season rice [10].

Recent research indicates that the production of late rice exceeds 7 t ha^{-1} and that the total yield of double-cropped rice can surpass 16.5 t ha^{-1} [11]. The number of effective panicles per area, which is related to axillary bud-germination capacity, determines ratooning rice yield [12]. Previous studies indicated that a high yield of ratooning rice is contingent on increased panicles and significant dry matter accumulation [13]. The key to achieving high yield in ratooning rice is to germinate more tillers in the first season to increase leaf area index, promote dry matter accumulation, promote axillary bud differentiation in regeneration envy, and increase the number and weight of effective panicles, thereby increasing yield in the ratoon season [14,15]. Jiang et al. discovered that high stubble height (40 cm) enhanced the grain yield of ratooning rice by 96.4% compared to low stubble height due to an increase in panicle number and irrigation rate [16]. On the other hand, higher stubble height substantially increased crop rotation yields for varieties that rely primarily on panicles ratooning from the upper nodes [17]. Germination fertilizers increased soluble protein and POD activity in first-season rice stems and influenced axillary bud growth [18]. Wang et al. discovered that the application of seedling-promoting fertilizers improved nitrogen metabolism in regenerating-season rice, stimulated growth and development, increased the grain number, and ultimately increased the yield [19]. Dalliri et al. found that the timing of the first harvest and the stubble height significantly affected the rotational rice's productivity [15]. The chlorophyll content in rice leaves is essential for photosynthesis, light absorption, and ultimately crop yield [20]. Photosynthesis and light absorption are important for ratooning rice growth, biomass accumulation, and yield formation.

Mechanical harvesting of ratooning rice damages the stems, disrupting nutrient delivery and axillary bud growth, thereby decreasing yield, which studies have identified as a significant factor limiting high yields of ratooning rice [21]. After the first season's rice is harvested for yield using machinery, the stems are rolled by the operating tires, causing extensive stalk ambulation during the subsequent ratoon season. Axillary buds sprouting from rice stems that have been rolled suffer from inundation and allelopathy inhibition. The disruption of population structure caused by rolling, combined with reduced photosynthetic and biomass production capacity and internode underfilling, can directly reduce yield and quality [22]. The formation of regenerating rice panicles depends on the growth of axillary buds, which can be affected by the rolling of the harvest machine, causing changes in the transportation of photosynthetic products between nodes [10]. This phenomenon led to a significant decrease in the panicle number during the ratoon season, resulting in a one-third reduction in rice yield. In addition, buds regenerated from lower nodes are more likely to perish than buds regenerated from higher nodes, most likely due to the greater degree of shading in the lower nodes than in the higher nodes [23]. A report suggests that the border effect of ratooning rice cultivated in non-rolling areas has the potential to compensate for the 4–10% yield reduction caused by harvester damage during rolling [24].

China has developed mechanized cultivation of ratooning rice over the past decade and this technology entails mechanical harvesting of ratooning rice yields. Critical practices for achieving high yields in this system include selecting varieties, optimizing sowing dates, effectively managing water and fertilizer, and harvesting rice at the optimal height [25]. Paradoxically, the mechanical harvesting of rice yields will be rolling the rice stems and preventing axillary bud germination, resulting in lower yields. Our previous study showed that manual stubble righting during ratooning rice harvest could be a valuable management practice for reducing yield loss and promoting the growth of rotational rice in rice-production systems in China [26]. Nonetheless, how stubble righting affects grain yield of ratooning rice during the rice ratoon season remains unknown. To address the challenges of mechanically harvested ratooning rice yields, we hypothesized that mechanical stubble righting during the harvest of the main crop would be effective at increasing ratooning rice yields and enhancing yield components and biomass accumulation. This two-year field experiment examined yield formation, biomass accumulation, physiological and biochemical parameters, photosynthetic characteristics, and solar radiation interception rate. This study aimed to identify strategies to optimize ratooning rice cultivation for high ratooning rice yield.

2. Materials and Methods

2.1. Experimental Design

The two-year field experiments were conducted in Yangwan Village, Cailing Town, Duchang County, Jiujiang City, Jiangxi Province, in 2021 and 2022. The properties of the experimental soil were as follows: pH value of 5.20, organic matter content of 27.31 g kg^{-1}, alkaline nitrogen, fast-acting potassium, and the available phosphorus content was 86.18 mg kg^{-1}, 75.86 mg kg^{-1}, and 55.21 mg kg^{-1}. The varieties used for the field experiment were YLiangyou 911 and Ken Liangyou 801. The two cultivars are popular locally for ratooning rice production. The compound fertilizer (N:P:K = 15:15:15) was used. The nitrogen fertilizer applied to the primary season rice was 225 kg N hm^{-2} (with basal fertilizer:tillering fertilizer:booting fertilizer:grain-filling fertilizer = 4:2:3:1). The mean temperature of the experimental region was 24.4 °C during the experimental period. The primary rice was sown on 13 March, transplanted on 18 April with a planting density of 30 cm × 14 cm, and harvested on 8 August, while the ratooning was harvested on 29 October 2021. In 2022, the primary rice was sown on 11 March, transplanted on 10 April with a planting density of 30 cm × 14 cm, and harvested on 12 August, while the ratooning was harvested on November-6.

The field experiments were conducted with three treatments: stubble righting after the mechanical harvest of primary rice (T1), ratooning rice without mechanical rolling of the rice stubble (T2), and without stubble righting after the mechanical harvest of primary rice (CK). The Prestige Edition 4LZ-6.0 EK (Q) Combine Harvester was used for harvesting. The whole machine quality of no-load was 3680 kg, while with full load, it was about 4880 kg. The track width and length of harvesting machine was 500 mm and 1250 mm, respectively. The cutting width of the machine was 2.2 m. The harvest speed of the harvester during harvesting was 1.6 m/s. The soil moisture content during harvest was 17.15–23.54%. The experimental treatment with a plot size of 25 m × 20 m underwent three replications. The stubble-righting machine was 'the righting machine for crushed stubble after mechanical harvesting of ratooning rice'. The righting attachment was driven by an engine power machine (Figure 1).

2.2. Measurements

2.2.1. Determination of Bud Germination

A total of 30 representative plants were randomly selected to investigate bud germination. The number of buds sprouting at different nodes was recorded at 1, 3, 5, 7, 9, 11, 13, and 15 days after mechanical harvesting.

Figure 1. The righting machine for crushed stubble after mechanical harvest.

2.2.2. Determination of Grain Yield and Yield Components

At the maturity stage, a total of 30 represented plants were randomly selected, and the average of these was taken as the effective panicle number. Then, 12 represented rice plants at the experimental treatment were selected to investigate the grain number per panicle, filled grain percentage, and 1000-grain weight. Plot yield was determined at 2 m^2 per plot harvest with four replications, followed by threshing using a threshing machine to measure yield, which was converted to actual yield at 14% moisture content.

2.2.3. Determination of Dry Weight Accumulation

A total of 10 rice plant stubbles were randomly selected at the heading and maturity stage, undergoing four replications, and the plant height was measured. The rice plants were then divided into stem–sheath (stem), leaves, and panicles and oven-dried for 30 min at 105 °C and then oven-dried to a constant weight at 80 °C, and the biomass accumulation was then weighted.

2.2.4. Physiological and Biochemical Analysis

At the heading stage, 15 fresh leaves from each treatment were harvested and kept in liquid nitrogen for 1 min, then stored at −80 °C for physiological and biochemical analysis. The fresh plant tissue (0.3 g) was weighed and homogenized in 5 mL of 50 mmol·L^{-1} phosphate buffer solution (PBS, pH = 7.8), and then centrifuged at 8000 rpm for 15 min.

The content of malondialdehyde (MDA) was determined by Pan et al. [27]. For MDA content, the absorbance was recorded by a spectrophotometer at 450 nm, 600 nm, and 532 nm. The MDA content was defined as μmol g^{-1} FW. The activities of superoxide dismutase (SOD), catalase (CAT), peroxidase (POD), and hydrogen peroxide (H_2O_2) were determined according to Li et al. [28]. For SOD activity, the nitro-blue tetrazolium (NBT) method was employed. The absorbance was measured using a spectrophotometer at 560 nm. The SOD activity was defined in the unit U g^{-1} min^{-1}. For POD activity, a spectrophotometer was used to measure the absorbance at 470 nm with five replicates at an interval of 30 s, and the POD activity was expressed as U g^{-1} min^{-1}. For CAT activity, a spectrophotometer was used to measure the absorbance at 470 nm, and the absorbance was recorded every 30 s with four replicates. The CAT activity was defined as mmol min^{-1} mg^{-1}.

The proline content of the rice grains at the maturity stage was determined following Bates et al. [29]. Fresh plant tissue (0.3 g) was extracted in 3% sulfosalicylic acid (5 mL) and kept in boiling water for 10 min, then cooled. A total of 1 mL of supernatant was mixed successively with 1 mL of glacial acetic acid and 1 mL of 2.5% ninhydrin reagent, and then kept in boiling water for 30 min. The reaction mixture was extracted by 4 mL toluene, followed by standing stratification. The 1 mL extract was centrifuged at 4000 rpm for 5 min. The absorbance was recorded at 530 nm. The proline content was expressed as ug·g^{-1} fresh weight (FW). Ascorbic acid content in the plant tissues was determined according to Huang et al. [30]. For ASA content determination, the reaction system consisted of 0.4 mL of 150 mmol L^{-1} NaH_2PO_4 solution, 0.2 mL of enzyme extract, 0.2 mL of distilled water, and 0.4 mL of 10% TCA. After 30 s, the mixture was added to 0.4 mL of H_3PO_4, 0.4 mL of 4% 2,2-bipyridine, and 0.2 mL of 3% $FeCl_3$ solution. The amount of ASA in the sample was expressed as mg g^{-1}. The soluble protein content refers to the method determination by Yang et al. [31]. A total of 0.12 mL of the solution was measured with 0.6 mL of Kaomas Brilliant Blue thoroughly, and the absorbance value at 595 nm was measured and recorded after standing for 2 min. Under the same reaction conditions, bovine serum protein was used as the standard material to make the standard curve, and the soluble protein content was calculated according to the standard curve. The soluble protein content was expressed as mg g^{-1}.

The extraction and activity of nitrate reductase (NR), glutamine synthetase (GS), and glutamate synthase (GOGAT) were estimated according to the method described by Feng et al. [32]. The NR activity using a spectrophotometer and the absorbance at 540 nm was recorded, and the NR activity was expressed as μg g^{-1} h^{-1}. The absorbance was read at 540 nm using a spectrophotometer, and the GS activity was denoted as μg g^{-1} h^{-1}. The absorbance was recorded using a spectrophotometer at 340 nm internal 30 s for 5 min. The GOGAT activity was defined as $μg^{-1}$ min^{-1}.

2.2.5. Determination of Photosynthetic Characteristics

A total of 0.3 g fresh samples through light-avoidance grinding were added in the centrifuge tube to 10 mL of 95% ethanol-leaching fresh sample pigment. Each repetition was carried out 4 times, light-avoidance sealing was conducted for 24 h, and the fresh samples became white. They were shaken well after centrifugation at 25 °C and 5000 rpm conditions for 5 min, with the use of a BioTek Epoch microtiter plate spectrophotometer to determine the absorbance of supernatant at the two wavelengths of 665 nm and 649 nm [33].

$$\text{Chlorophyll a} = 12.7\text{D}663 - 2.69\text{D}645$$

$$\text{Chlorophyll b} = 22.9\text{D}645 - 4.68\text{D}663$$

$$\text{Total chlorophyll} = 20.2\text{D}645 + 8.02\text{D}663$$

2.2.6. Determination of Canopy Photosynthetically Active Radiation

The distribution of photosynthetically active radiation (PAR) within the canopy was measured at 9:00, 11:00, 13:00, and 15:00 on sunny days using a MQ306 long-rod photosynthetically active radiation meter. In the north–south direction, several sensors were evenly distributed to collect instantaneous photosynthetically active radiation, which was called one record. In the east–west direction, one record was collected in each of the five positions of 24 cm between rows, and one record was collected in each of the five positions near the east side of the rows at 6 cm, 12 cm, 18 cm, as well as 6 cm and 12 cm from the west side of the rows, which were collectively referred to as one horizontal sample. In addition, seven heights were measured vertically, from the ground level to the top of the canopy, which was referred to as one horizontal sample. In the vertical direction, seven heights were measured from the ground to the top of the canopy, namely 0 cm, 15 cm, 30 cm, 45 cm, 60 cm, and 75 cm from the ground and the top of the canopy (15 cm above the canopy), which were recorded separately, and one horizontal sample was measured at each height.

Interception rate of radiation of population (%) = (Radiation at 15 cm top of the canopy-Radiation at the ground)/Radiation at 15 cm top of the canopy × 100%

Interception rate of radiation of canopy (%) = (Radiation at 15 cm top of the canopy-Radiation at 20 cm down from the top of the canopy)/Radiation at 15 cm top of the canopy × 100%

2.3. Statistical Analysis

Microsoft Excel 2010 and Statistic 8.0 (Analytical Software, Tallahassee, FL, USA) was used for data collation and analysis, and the least significant difference (LSD) test was used for multiple comparisons ($p < 0.05$).

3. Results

3.1. Grain Yield and Yield Components

The righting treatment significantly influenced the grain yield and yield components. The T1 and T2 treatments produced higher grain yield than CK by 107.41% and 147.97% in YLiangyou911 in 2021, 45.85% and 114.26% in Kenliangyou801 in 2021, 6.99% and 23.87% in YLiangyou911 in 2022, and 77.23% and 187.13%in Kenliangyou801 in 2022, respectively. The T1 treatment harvested a lower grain yield than the T2 treatment. The T1 and T2 treatments showed higher panicle numbers per hill and filled grain per panicle than the CK treatment. Change in grain number per panicle was significant in cultivar and treatment. A higher 1000-grain weight was detected in T1 and T2 in YLiangyou911 in 2021 and Kenliangyou801 in 2022 (Table 1).

Table 1. Effects of stubble righting on grain yield and yield components of ratooning rice.

Treatment	Panicle Number Per Hill	Grain Number Per Panicle	Filled Grain Percentage (%)	1000-Grain Weight (g)	Grain Yield (g m^{-2})
2021					
YLiangyou911					
CK	12.62b	81.65a	60.71a	19.84b	189.94c
T1	21.03a	72.40a	70.73a	21.38a	392.85b
T2	20.35a	83.85a	71.14a	22.13a	471.43a
Kenliangyou801					
CK	14.68c	70.07a	60.20a	21.63a	253.49b
T1	18.03b	67.49a	75.64a	22.88a	369.09ab
T2	21.10a	70.19a	79.73a	23.18a	543.14a
2022					
YLiangyou911					
CK	20.48b	96.88a	47.79b	23.44a	243.24b
T1	27.73a	69.77c	53.37a	24.03a	260.28b
T2	26.45a	77.18b	55.25a	23.07a	301.62a
Kenliangyou801					
CK	16.68c	62.37a	34.97c	21.45b	101.72c
T1	24.23b	54.46b	45.27b	22.24ab	179.73b
T2	31.43a	59.67ab	61.22a	23.23a	290.76a
ANOVA					
Year (Y)	**	ns	**	ns	**
Cultivar(C)	ns	**	ns	ns	ns
Treatment (T)	**	**	**	**	**
Y×C	ns	**	*	**	ns
Y×T	*	**	ns	ns	*
C×T	**	**	ns	ns	ns
Y×C×T	**	*	ns	ns	ns

Y×C: Interaction between year and cultivar; Y×T: Interaction between year and treatment; C×T: Interaction between cultivar and treatment; Y×C×T: Interaction among year, cultivar, and treatment; * and ** represent a significant F-value at $p < 0.05$ and $p < 0.01$, respectively; ns represents a non-significant difference. CK, without stubble righting after the mechanical harvest of primary rice; T1, stubble righting after the mechanical harvest of primary rice; T2, ratooning rice without mechanical rolling of the rice stubble. Different lower-case letters followed by a cultivar in one year represents significance at $p < 0.05$.

3.2. Biomass Accumulation

Treatment significantly affected the biomass accumulation of ratooning rice. The biomass accumulation in stem and sheath, leaf, panicle, and the whole plant for T1 and T2 treatment was higher than those of CK at both the heading and maturity stages. The highest biomass accumulation was detected for the T2 treatment and then for the T1 treatment, while the CK treatment produced the lowest biomass at both the heading and maturity stages. The mean value of biomass accumulation at T1 for both cultivars in both years in stem and sheath, leaf, panicle, and the whole plant was 521.27 g m^{-2}, 415.96 g m^{-2}, 77.00 g m^{-2}, and 1014.23 g m^{-2} at the heading stage and 471.03 g m^{-2}, 409.25 g m^{-2}, 621.73 g m^{-2}, and 1502.02 g m^{-2} at the maturity stage, respectively. The mean value of biomass accumulation at T2 for both cultivars in both years in stem and sheath, leaf, panicle, and the whole plant was 717.50 g m^{-2}, 592.71 g m^{-2}, 150.19 g m^{-2}, and 1460.39 g m^{-2} at the heading stage and 616.24 g m^{-2}, 515.50 g m^{-2}, 1056.37 g m^{-2}, and 2188.11 g m^{-2} at the maturity stage, respectively (Table 2).

Table 2. Effects of stubble righting on biomass production of ratooning rice.

Treatment	Heading Stage Stem Dry Weight (g m^{-2})	Leaf Dry Weight (g m^{-2})	Panicle Dry Weight (g m^{-2})	Total Dry Weight (g m^{-2})	Maturity Stage Stem Dry Weight (g m^{-2})	Leaf Dry Weight (g m^{-2})	Panicle Dry Weight (g m^{-2})	Total Dry Weight (g m^{-2})
2021								
YLiangyou911								
CK	353.63b	414.36a	52.95b	820.94c	299.12b	434.22a	358.50c	1091.84b
T1	598.05a	423.38a	114.85a	1136.28b	493.72a	427.12a	762.84b	1683.68a
T2	674.63a	524.00a	162.12a	1360.75a	524.84a	465.26a	1018.91a	2009.01a
Kenliangyou801								
CK	348.99b	430.30b	66.25c	845.54c	278.08b	415.45a	413.23b	1106.76b
T1	547.36a	654.21a	144.52b	1346.09b	353.92b	484.58a	628.48b	1466.98b
T2	637.50a	740.80a	231.28a	1609.59a	561.67a	545.05a	1149.15a	2255.86a
2022								
YLiangyou911								
CK	353.63b	414.36a	52.95b	820.94c	455.20c	360.73b	390.31c	1206.24c
T1	598.05a	423.38a	114.85a	1136.28b	568.58b	404.70b	702.89b	1676.17b
T2	674.63a	524.00a	162.12a	1360.75a	695.60a	586.94a	1076.86a	2359.41a
Kenliangyou801								
CK	348.99b	430.30b	66.25c	845.54c	399.37b	308.89b	245.31b	953.57b
T1	547.36a	654.21a	144.52b	1346.09b	467.92b	320.59b	392.72b	1181.23b
T2	637.50a	740.80a	231.28a	1609.59a	682.84a	464.77a	980.56a	2128.17a
ANOVA								
Year (Y)	**	*	ns	ns	**	**	ns	ns
Cultivar(C)	ns	**	*	**	*	ns	ns	*
Treatment (T)	**	**	**	**	**	**	**	**
Y×C	ns	ns	ns	ns	ns	**	ns	*
Y×T	ns	ns	ns	ns	ns	ns	ns	ns
C×T	ns	**	ns	ns	*	ns	ns	ns
Y×C×T	ns	ns	ns	ns	ns	ns	ns	ns

Y×C: Interaction between year and cultivar; Y×T: Interaction between year and treatment; C×T: Interaction between cultivar and treatment; Y×C×T: Interaction among year, cultivar, and treatment; * and ** represent a significant F-value at $p < 0.05$ and $p < 0.01$, respectively; ns represents a non-significant difference. CK, without stubble righting after the mechanical harvest of primary rice; T1, stubble righting after the mechanical harvest of primary rice; T2, ratooning rice without mechanical rolling of the rice stubble. Different lower-case letters followed by a cultivar in one year represents significance at $p < 0.05$.

3.3. Interception Rate Solar Radiation and Photosynthetic Characteristics

The plant height of the ratooning rice was significantly affected by year, cultivar, and treatment. The plant height was about 48.22–65.60 cm. The interception rate of radiation after harvest was higher at the T1 and T2 treatments than at the CK treatment. The interception rate of radiation of the canopy and the interception rate of radiation of the population at the heading and maturity stage was higher at the T1 and T2 treatments than at the CK treatment (Table 3).

Table 3. Effects of stubble righting on solar radiation characteristics of the canopy of ratooning rice.

Treatment	Plant Height (cm)	Interception Rate of Radiation of Population (%)	Interception Rate of Radiation of Canopy (%)	Interception Rate of Radiation of Population (%)	Interception Rate Radiation of Canopy (%)	Interception Rate of Radiation of Population (%)
		after harvest	at heading stage		at maturity stage	
2021						
YLiangyou911						
CK	58.56b	61.40a	16.64b	61.80b	24.90b	69.34b
T1	61.88ab	67.67a	24.48ab	63.10b	47.71a	80.94a
T2	65.60a	69.62a	30.52a	80.01a	50.85a	84.62a
Kenliangyou801						
CK	57.27b	31.21c	14.90b	73.78b	49.37a	88.75a
T1	60.46ab	51.15b	38.60a	75.64ab	55.31a	88.15a
T2	64.96a	67.39a	38.94a	80.46a	55.48a	87.60a
2022						
YLiangyou911						
CK	58.36a	27.42b	59.90a	81.65a	63.16b	89.76a
T1	54.65b	38.58b	70.95a	84.41a	64.38b	90.83a
T2	61.51a	57.55a	66.18a	84.55a	76.23a	93.70a
Kenliangyou801						
CK	48.22c	21.09b	21.09c	80.17a	68.64ab	93.42a
T1	51.08b	53.76a	48.03b	83.24a	61.46b	94.41a
T2	60.56a	57.37a	59.78a	80.33a	75.33a	93.61a
ANOVA						
Year (Y)	**	**	**	**	**	**
Cultivar (C)	**	*	**	ns	*	**
Treatment (T)	**	**	**	*	**	**
Y×C	**	**	**	**	*	**
Y×T	ns	ns	ns	**	**	*
C×T	ns	**	**	*	*	**
Y×C×T	ns	*	*	ns	ns	**

Y×C: Interaction between year and cultivar; Y×T: Interaction between year and treatment; C×T: Interaction between cultivar and treatment; Y×C×T: Interaction among year, cultivar, and treatment; * and ** represent a significant F-value at $p < 0.05$ and $p < 0.01$, respectively; ns represents a non-significant difference. CK, without stubble righting after the mechanical harvest of primary rice; T1, stubble righting after the mechanical harvest of primary rice; T2, ratooning rice without mechanical rolling of the rice stubble. Different lower-case letters followed by a cultivar in one year represents significance at $p < 0.05$.

3.4. Antioxidant Response and Nitrogen Metabolism

Treatment significantly affected the SOD activity, POD activity, CAT activity, and proline and ascorbic acid content. Compared with CK, rice leaves under T1 treatment yield higher SOD activity, POD activity, and CAT activity in leaves. No significant difference in MDA and H_2O_2 content was detected for the T1 treatment compared to CK. Compared with CK, the T1 treatment significantly increased the proline content in YLiangyou911 and Kenliangyou801 in 2021 and significantly increased the ascorbic acid content in YLiangyou911 in 2022 (Table 4).

Table 4. Effects of stubble righting on antioxidant response in leaves of ratooning rice.

Treatment	SOD Activity (U g-1 min-1)	POD Activity (U g^{-1} min^{-1})	CAT Activity (mmol min^{-1} mg^{-1})	MDA Content (μmol g-1 FW)	PRO Content (μg g^{-1} FW)	Ascorbic Acid Content (mg g^{-1})	H_2O_2 Content (mmol g^{-1})
2021							
YLiangyou911							
CK	858.07b	172.48b	105.26c	11.55ab	8.76b	0.12a	17.25b
T1	930.21a	220.39a	146.72b	11.15b	23.24a	0.11a	19.99ab
T2	955.05a	228.03a	180.43a	12.4a	12.87b	0.08a	22.02a
Kenliangyou801							
CK	826.87a	184.54b	141.57b	12.86a	6.53c	0.09a	15.42b
T1	865.42a	208.03a	162.05a	12.88a	21.41a	0.08a	15.87b
T2	639.04b	199.43ab	149.98ab	12.69a	12.04b	0.04b	18.51a
2022							
YLiangyou911							
CK	1284.97a	263.87a	208.26a	17.08a	7.17a	0.17b	30.31a
T1	1255.51a	265.95a	215.92a	17.22a	5.68a	0.19a	36.89a
T2	1204.3a	285.26a	241.57a	15.39a	6.01a	0.17b	27.63a
Kenliangyou801							
CK	1123.63c	241.29a	214.8a	18.21a	6.69a	0.14a	12.82a
T1	1296.83a	277.17a	245.87a	17.78a	6.94a	0.15a	37.59a
T2	1197.13b	288.6a	234.1a	13.64a	7.76a	0.15a	34.37a
ANOVA							
Year (Y)	**	**	**	ns	**	**	ns
Cultivar(C)	**	ns	ns	ns	ns	**	ns
Treatment (T)	**	**	**	ns	**	**	ns

Table 4. *Cont.*

Treatment	SOD Activity (U g-1 min-1)	POD Activity (U g^{-1} min^{-1})	CAT Activity (mmol min^{-1} mg^{-1})	MDA Content (μmol g-1 FW)	PRO Content (μg g^{-1} FW)	Ascorbic Acid Content (mg g^{-1})	H_2O_2 Content (mmol g^{-1})
Y×C	*	ns	ns	ns	ns	ns	ns
Y×T	ns	ns	ns	ns	**	**	ns
C×T	*	ns	**	ns	ns	ns	ns
Y×C×T	**	ns	ns	ns	ns	ns	ns

Y×C: Interaction between year and cultivar; Y×T: Interaction between year and treatment; C×T: Interaction between cultivar and treatment; Y×C×T: Interaction among year, cultivar, and treatment; * and ** represent a significant F-value at $p < 0.05$ and $p < 0.01$, respectively; ns represents a non-significant difference.CK, without stubble righting after the mechanical harvest of primary rice; T1, stubble righting after the mechanical harvest of primary rice; T2, ratooning rice without mechanical rolling of the rice stubble. SOD, superoxide dismutase; POD, peroxidase; CAT, catalas; MDA, malondialdehído; PRO, prolina; H_2O_2, peróxido de hidrógeno. Different lower-case letters followed by a cultivar in one year represents significance at $p < 0.05$.

Treatment significantly affected the soluble protein content. Compared with CK, The T1 treatment significantly increased the GS activity in YLiangyou911 in 2022 and significantly increased the soluble protein content, NR activity, and GS activity in Kenliangyou801 in 2022 (Table 5).

Table 5. Effects of stubble righting on nitrogen metabolism in leaves of ratooning rice.

Treatment	Soluble Protein (mg g^{-1})	NR Activity (μg g^{-1} h^{-1})	GS Activity (μg g^{-1} h^{-1})	GOGAT Activity (μg^{-1} min^{-1})
2021				
YLiangyou911				
CK	8.86b	6.25a	10.42a	4.98a
T1	11ab	8.95a	14.92a	6.02a
T2	14.1a	7.88a	13.13a	6a
Kenliangyou801				
CK	7.36a	5.49a	8.41a	3.7a
T1	8.16a	5.39a	8.14a	6.34a
T2	8.59a	5.06a	8.43a	6.1a
2022				
YLiangyou911				
CK	9.66a	12.12a	8.09b	5.44a
T1	9.3a	12.14a	8.86a	5.5a
T2	9.89a	11.67a	7.77b	5.59a
Kenliangyou801				
CK	8.15b	10.68b	7.28a	5.83a
T1	9.44a	12.06a	7.63a	5.67a
T2	8.61b	11.36ab	7.5a	5.48a
ANOVA				
Year (Y)	ns	**	ns	ns
Cultivar(C)	**	**	*	ns
Treatment (T)	**	ns	ns	ns
Y×C	**	ns	*	ns
Y×T	*	ns	ns	ns
C×T	ns	ns	ns	ns
Y×C×T	ns	ns	ns	ns

Y×C: Interaction between year and cultivar; Y×T: Interaction between year and treatment; C×T: Interaction between cultivar and treatment; Y×C×T: Interaction among year, cultivar, and treatment; * and ** represent a significant F-value at $p < 0.05$ and $p < 0.01$, respectively; ns represents a non-significant difference. CK, without stubble righting after the mechanical harvest of primary rice; T1, stubble righting after the mechanical harvest of primary rice; T2, ratooning rice without mechanical rolling of the rice stubble. NR, nitrate reductase; GS, glutamine synthase; GOGAT, glutamate synthase. Different lower-case letters followed by a cultivar in one year represents significance at $p < 0.05$.

3.5. Bud-Regeneration Capacity

There was no significant difference between the T2 treatments on the final bud number of ratooning rice at the second node for all periods. A significantly higher bud number of ratooning rice at the third and fourth nodes was detected. Finally, a higher total bud number of ratooning rice was observed for the T1 and T2 treatment compared to the CK treatment (Figure 2).

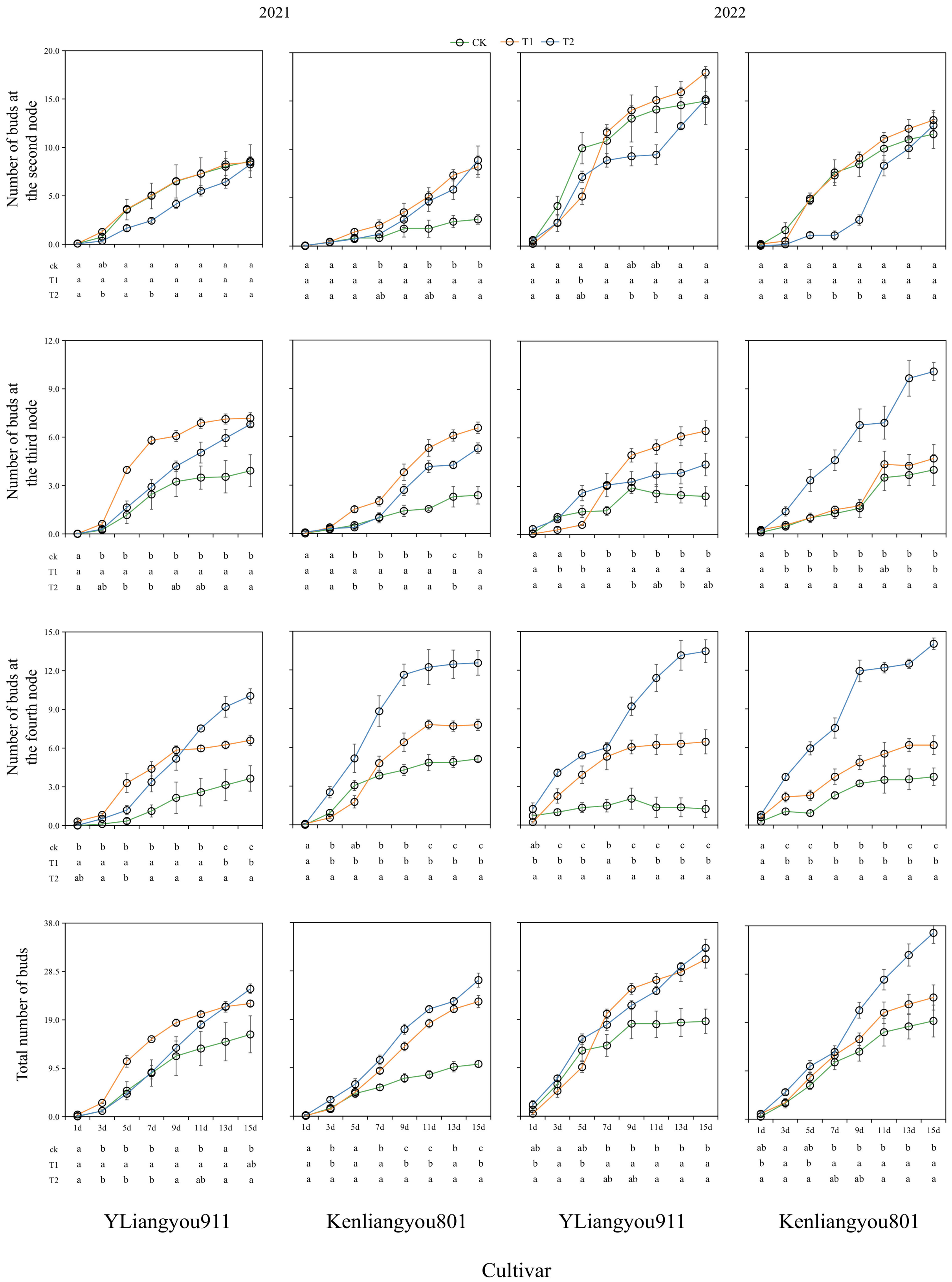

Figure 2. Effects of stubble righting on bud number of ratooning rice in 2021 and 2022. Within the same variety, different lower-case letters in the same column indicate significant differences at $p < 0.05$. Within the same variety, different lower-case letters in the same column indicate significant differences at $p < 0.05$. CK, without stubble righting after the mechanical harvest of primary rice; T1, stubble righting after the mechanical harvest of primary rice; T2, ratooning rice without mechanical rolling of the rice stubble.

3.6. Correlation Analysis

The grain yield was significantly correlated to the interception rate of radiation after harvest (Figure 3). A significant correlation between grain yield and bud numbers was detected (Figure 4). The filled grain percentage, plant height, and biomass accumulation were significantly positively correlated to grain yield (Figure 5). The grain yield was negatively correlated with the SOD activity, MDA content, and ascorbic acid content (Figure 6).

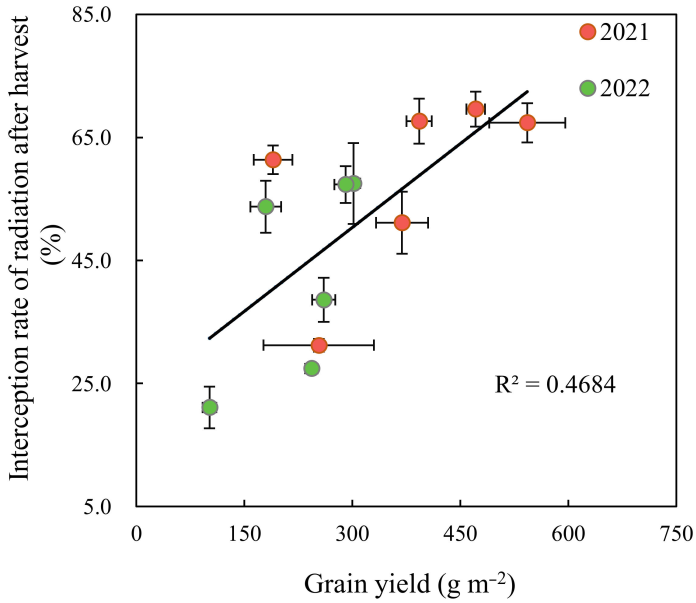

Figure 3. Correlation analysis between grain yield and radiation interception in 2021 and 2022.

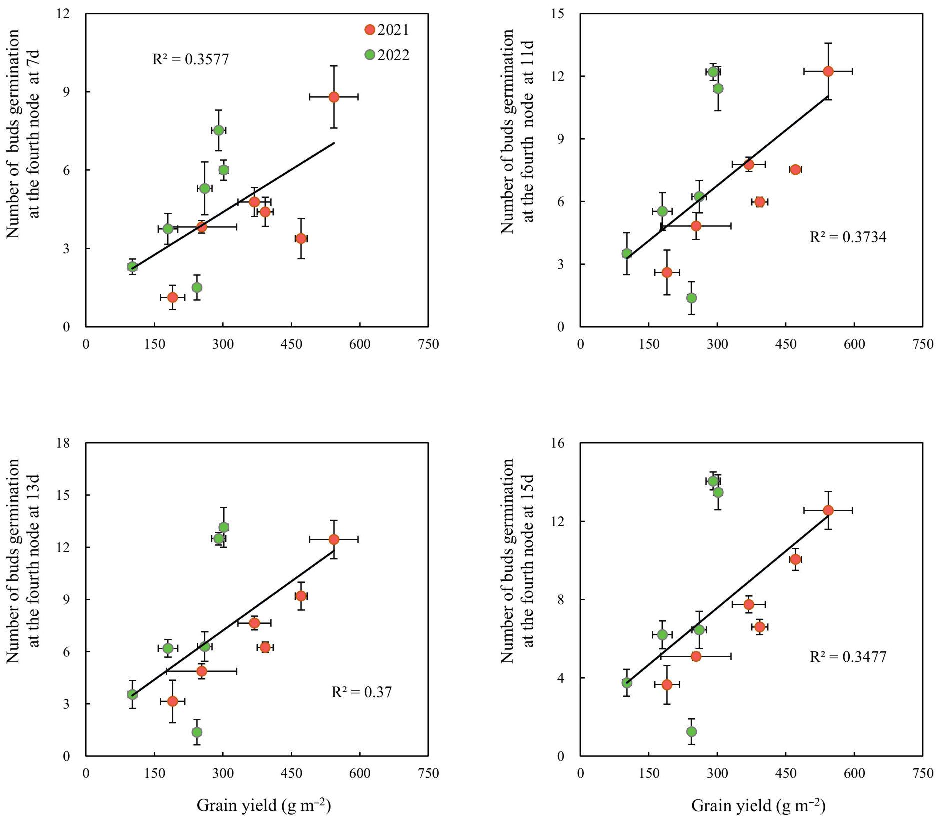

Figure 4. Correlation analysis between grain yield and the bud numbers in 2021 and 2022.

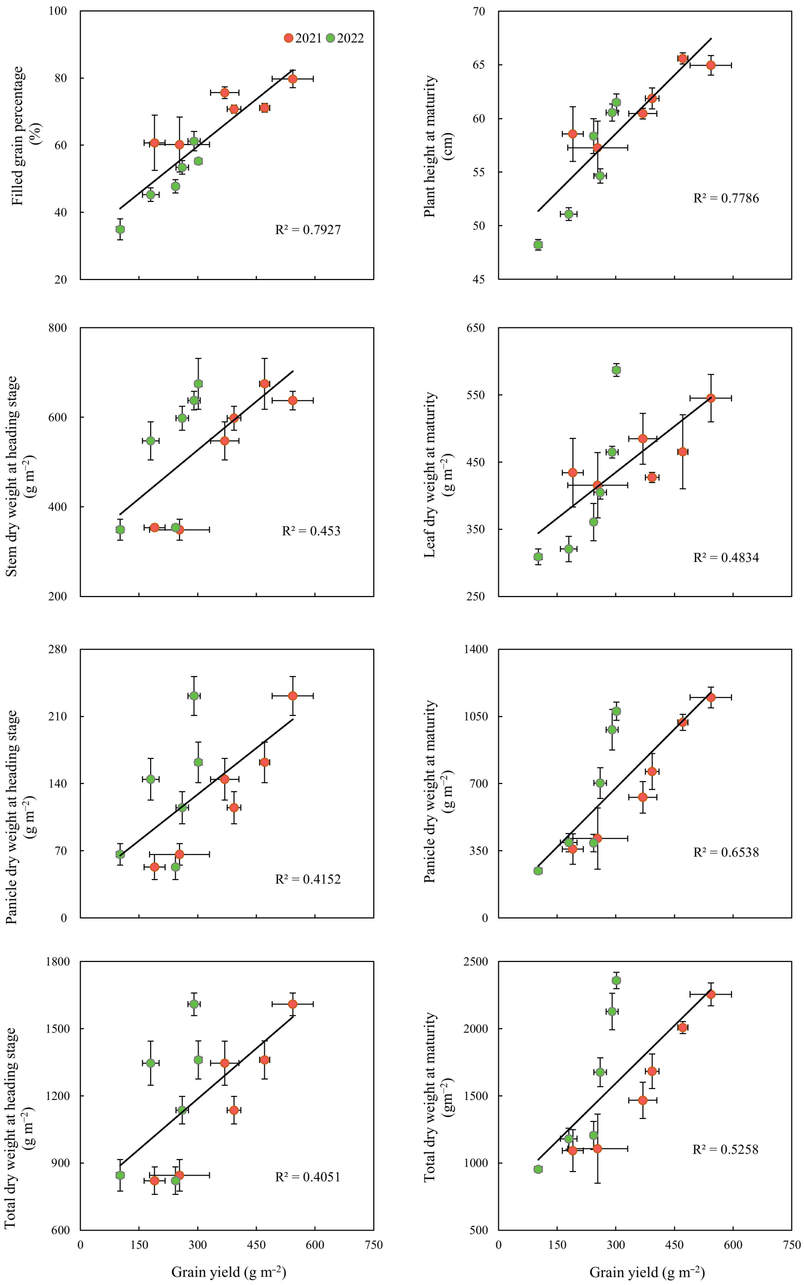

Figure 5. Correlation analysis between grain yield and plant height, filled grain percentage, radiation interception, and dry weight in 2021 and 2022.

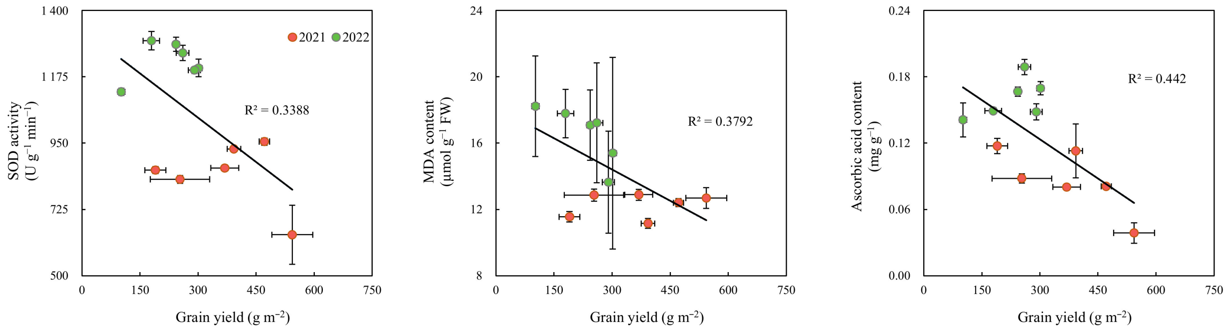

Figure 6. Correlation analysis between grain yield and antioxidant response in 2021 and 2022.

4. Discussion

In this study, the year (Y), treatment (T), and Y×T had significant effects on grain yield of ratooning rice (Table 1). There was a significant effect of stubble-righting measures on the yield of rolling ratooning rice. When harvesting the main crop, the stems of ratooning rice were rolled by machine, and after rolling, the buds of ratooning rice were reduced. After stem rolling, ratooning rice had fewer effective panicles, which ultimately leads to reduced yields, and the grain yield was higher in the T1 and T2 treatments than in the CK treatment in both years, with the T2 treatment yielding the highest grain yield (Table 1). This result is consistent with the findings of Chen et al. [26], who discovered that manual stubble righting reduces the yield loss of ratooning rice. The axillary-bud-germination capacity determines the number of effective panicles, which is a crucial yield component [12]. Increases in adequate panicle number and dry weight were positively associated with ratooning rice yield [14,15]. In the present investigation, the panicle number and filled grain rate was higher in T1 and T2 than in CK (Table 1). And there was a significant and positive correlation between filled grain rate and grain yield (Figure 5). After stubble righting, ratooning rice has higher yield components, resulting in higher yields.

Furthermore, a high yield of ratooning rice depends on higher dry matter accumulation [12]. Higher biomass is related to higher photosynthesis and light absorption. In the present study, the T1 treatment produced the highest content of photosynthetic pigments in the leaves of YLiangyou911 in 2022, whereas it produced the lowest content in the leaves of Kenliangyou801 in 2021 (Table 6). Furthermore, this study revealed a significantly positive correlation between ratoon grain yield and radiation interception rate after harvest (Figure 3). The results revealed that the plant height of ratooning rice ranged between 48.22 cm and 65.60 cm and that the radiation interception rate was substantially influenced by year, cultivar, and treatment. In particular, ratooning rice grown under the T1 and T2 treatments had higher radiation interception rates at the canopy and population levels during both the tassel and maturity phases (Table 3). Therefore, rice treated with T1 produced significantly more biomass than rice treated with CK (Table 2). Moreover, mechanical harvesting of main rice diminishes the photosynthetic pigment content and radiation interception rate of ratooning rice, resulting in diminished material production capacity and internode underfilling [21]. Concurrently, extensive areas of rolled rice stems disrupted the rice population structure, directly impacting the accumulation of dry matter in ratooning rice. He et al. found that high rice productivity during the ratoon season is closely associated with a substantial increase in dry matter accumulation [12]. A greater dry matter accumulation in ratooning rice indicates a higher tiller number and leaf area index, which is more advantageous in axillary bud differentiation and positively correlated with ratoon-season yield [14,15]. In this study, the biomass accumulation in stem and sheath, leaf, panicle, and whole plant for T1 and T2 treatments was higher than those of CK at both heading and maturity stages. The biomass accumulation positively correlated to grain yield (Figure 5).

Rolling affects the number and growth of axillary buds reduced the grain yield oof ratooning rice [21]. In this study, we further examined the number of buds germination at different nodes and confirmed that a substantially higher number of buds was observed on the third and fourth nodes (Figure 2). The correlation between grain yield and bud number was statistically significant (Figure 4). The stubble-righting treatment considerably increased the number of buds of ratooning rice in the upper nodes because the rolling in the first season prevented the transport of photosynthetic products to the second and fourth nodes (top–down). Axillary buds of rolled rice and panicles primarily regenerated at the inverted fifth and sixth nodes (lower nodes). The number of axillary buds regenerating normally from the inverted second and third nodes was drastically reduced [10]. In addition, buds regenerated from the lower nodes were more likely to perish than those regenerated from the upper nodes, likely due to the higher degree of shading in the lower nodes [23].

Table 6. Effects of stubble righting on chlorophyll content in leaves of ratooning rice.

Treatment	Chlorophyll a (mg g^{-1})	Chlorophyll b (mg g^{-1})	Carotenoid (mg g^{-1})	Total chlorophyll (mg g^{-1})
2021				
YLiangyou911				
CK	1.31a	0.47a	0.28a	1.78a
T1	1.36a	0.49a	0.29a	1.85a
T2	1.38a	0.49a	0.29a	1.87a
Kenliangyou801				
CK	1.08b	0.4a	0.23a	1.49a
T1	0.81c	0.29b	0.17b	1.09b
T2	1.32a	0.48a	0.26a	1.8a
2022				
YLiangyou911				
CK	1.13b	0.38b	0.24b	1.5b
T1	1.29a	0.43a	0.28a	1.72a
T2	1.08b	0.36b	0.24b	1.44b
Kenliangyou801				
CK	1.31a	0.45a	0.28a	1.76a
T1	1.32a	0.45a	0.29a	1.77a
T2	1.25a	0.41a	0.29a	1.66a
ANOVA				
Year (Y)	ns	ns	ns	ns
Cultivar(C)	ns	ns	ns	ns
Treatment (T)	ns	ns	*	ns
Y×C	**	**	**	**
Y×T	**	**	**	**
C×T	**	**	**	**
Y×C×T	*	*	ns	*

Y×C: Interaction between year and cultivar; Y×T: Interaction between year and treatment; C×T: Interaction between cultivar and treatment; Y×C×T: Interaction among year, cultivar, and treatment; * and ** represent a significant F-value at $p < 0.05$ and $p < 0.01$, respectively; ns represents a non-significant difference. CK, without stubble righting after the mechanical harvest of primary rice; T1, stubble righting after the mechanical harvest of primary rice; T2, ratooning rice without mechanical rolling of the rice stubble. Different lower-case letters followed by a cultivar in one year represents significance at $p < 0.05$.

The enhance of soluble protein and POD activity in main rice stems is related to the axillary bud growth of ratooning rice [18]. In this study, the T1 treatment significantly increased SOD activity, POD activity, and CAT activity in leaves as compared to CK. The T1 treatment significantly increased the proline content in YLiangyou911 and Kenliangyou801 in 2021 and the ascorbic acid content in YLiangyou911 in 2022 (Table 4). The grain yield was negatively correlated with the SOD activity, MDA content, and ascorbic acid content (Figure 6). Furthermore, the righting treatment significantly affected the soluble protein content, resulting in an increase in GS activity in YLiangyou911 in 2022 and an increase in soluble protein, NR activity, and GS activity in Kenliangyou801 in 2022 (Table 5). Consistent with our findings, Wang et al. also reported that the enhanced nitrogen metabolism is related to the grain yield of ratooning rice [19].

Overall, stubble righting following mechanical harvesting of main rice regulated solar radiation interception in the canopy, rice bud number, chlorophyll content; improved antioxidant responses and nitrogen metabolism; increased biomass production and yield components; and ultimately increased grain yields of ratooning rice (Figure 7).

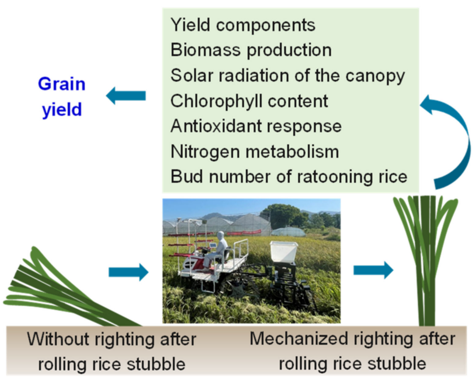

Figure 7. Mechanism for ratooning rice yield improvement under mechanical stubble righting after the mechanical harvest of primary rice.

5. Conclusions

The results show that stubble righting after the mechanical harvest of primary rice correction had a significant effect on yield increases in ratooning rice, but it was not as effective as without mechanical rolling of the rice stubble. The grain yield improvement can be attributed to the positive effects on biomass accumulation, solar radiation and photosynthetic characteristics, antioxidant response and nitrogen metabolism, and bud-regeneration capacity.

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