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Shading at the Booting Stage Improved the Grain Quality of Hybrid Rice Due to Reduced Spikelet Production

Liyan Shang, Zichen Liu, Jiayu Ye, Tian Sheng, Ruijie Li, Jun Deng, Ke Liu, Xiaohai Tian, Yunbo Zhang and Liying Huang *

MARA Key Laboratory of Sustainable Crop Production in the Middle Reaches of the Yangtze River, College of Agriculture, Yangtze University, Jingzhou 434025, China; 2021710716@yangtzeu.edu.cn (L.S.); 2021710718@yangtzeu.edu.cn (Z.L.); 2021710697@yangtzeu.edu.cn (J.Y.); 2021710705@yangtzeu.edu.cn (T.S.); 2023710776@yangtzeu.edu.cn (R.L.); 2021730059@yangtzeu.edu.cn (J.D.); ke.liu@utas.edu.au (K.L.); 200564@yangtzeu.edu.cn (X.T.); yzhang@yangtzeu.edu.cn (Y.Z.)

* Correspondence: 519002@yangtzeu.edu.cn; Tel.: +86-716-8066541

Abstract: As a growing abiotic stress, light deficient conditions seriously affect the yield and quality of rice. However, few studies focus on the effects of shading on grain quality at the booting stage and the responses of different hybrid rice cultivars to shading. Field experiments involving four representative rice (*Oryza sativa* L.) cultivars across no shading (CK) and 40% shading at the booting (S) and grain filling stages (SS) were conducted in 2021 and 2022. Compared with CK, S reduced grain yield by 53.0% but increased the head rice rate by 11.4% averaged across varieties and years. The chalkiness degree (CD) and chalky grain percentage (CR) were reduced by 73.0% and 61.6% in S due to its 45.3% lower total spikelets m^{-2}, 44.0% lower grain–leaf ratio and 23.5% lower dry weight spikelet production efficiency, compared with CK. The CD and CR in SS were 49.5% and 41.0% higher and HR was 7.1% lower than that in CK. Shading significantly reduced amylose content, peak viscosity and breakdown value, but increased protein content and setback value, and the effects of SS were greater than S. Y-liangyou900 and Liangyoupeijiu showed better milling quality, while Y-liangyou900 and Chuanyou6203 obtained a better appearance and eating quality than the other varieties under both S and SS. In conclusion, shading at the booting stage significantly improved the milling, appearance and nutritional quality, and did not reduce the cooking and eating quality, but led to a significant decline in the grain yield of hybrid rice. Moreover, Y-liangyou900 exhibited better rice quality but lower yield under shading treatments. Therefore, more attention needs to be focused on screening shade-tolerant varieties using both yield and quality to cope with climate change in the future.

Keywords: booting stage; grain filling stage; grain quality; hybrid rice; shading

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1. Introduction

With the development of the national economy and the improvement of people's living standards, rice (*Oryza sativa* L.), with high quality, is becoming more and more popular among consumers [1]. The main goals of current rice breeding programs are high yield and high quality. High-quality rice usually has the characteristics of good appearance, a high head rice rate and excellent taste [2]. Rice quality and grain yield are easily affected not only by varieties, but also by climate, soil, cultivation techniques and other factors, among which solar radiation is one of the climatic factors that have great influence on the yield and quality of rice [3–7].

In recent decades, solar radiation has been decreasing globally, especially across China [8,9]. It was found that there was a decrease of 599 and 344 MJ m^{-2} per decade from the 1950s to the 1980s and from the 1990s to the 2010s in solar radiation in China, respectively [10]. Weak light during the growth of rice leads to a decrease in leaf photosynthesis, resulting in insufficient accumulation of dry matter, which in turn leads to a deficiency in grain fullness and consequently a decrease in rice quality [11]. Especially at the grain filling stage, weak light can lead to a significant reduction in the percentages of brown rice, milled rice and head rice, while increasing the chalky grain percentage and chalkiness degree [12]. Starch is the main component of the endosperm of rice, primarily consisting of amylose and amylopectin, in which amylose content has a greater impact on rice quality [13]. It is generally believed that rice varieties with good eating quality have a lower amylose content [14–16]. A significantly positive correlation between amylose content and light intensity in rice has been observed [17]. As the second largest storage substance in rice endosperm, protein content determines the nutritional quality of rice [18]. Significant and negative correlations between grain protein content and cooking and eating quality have been widely observed in rice [19–21]. Previous studies reported a decrease in the amylose content and an increase in the protein content of rice grains under weak light at the grain filling stage [22–24], which showed that insufficient solar radiation during the grain filling stage led to poorer milling, a poorer appearance, and a poorer cooking and eating quality in rice. However, shading during the early growth stage reduced the occurrence of chalkiness and had no effect on grain protein content, which improved the appearance and cooking and eating quality of rice to a certain extent [25].

As one of the main rice production areas, rice yield and quality in the middle and lower reaches of the Yangtze River are often affected by strong light and high temperature during the growth of rice, but are also affected by continuous overcast and rainy weather during the grain filling stage every 2–3 years. In recent years, the sunshine duration and total solar radiation in the middle and lower reaches of the Yangtze River have been decreasing, which will result in a reduction in light energy resources during the growth of rice, and ultimately impact rice yield and quality [26]. However, the studies on the effects of weak light on rice quality mainly focused on the grain filling stage, while there were few studies on weak light at the booting stage. To explore the effects of weak light at the booting stage on rice quality and the response characteristics of different rice varieties to shading, two field experiments were conducted with local widely planted hybrid rice varieties as the materials under three light conditions.

2. Materials and Methods

2.1. Experimental Site

Field experiments were carried out at the experimental farm of Yangtze University (32°21′ N, 112°31′ E, 34 m above sea level), Jingzhou City, Hubei Province, China, in 2021 and 2022. The soil in the experimental field was clayic Calcisol with the following properties: soil pH 6.8, organic matter 20.8 $g \cdot kg^{-1}$, total nitrogen 1.9 $g \cdot kg^{-1}$, readily available phosphorus 29.6 $mg \cdot kg^{-1}$, and available potassium 129.6 $mg \cdot kg^{-1}$ [27]. The experimental field is planted with one crop of medium-season rice every year and is free in winter. The daily minimum and maximum temperatures and daily solar radiation during the rice growing seasons were collected by the weather station near the experimental field. The average daily minimum and maximum temperature and solar radiation from transplanting to maturity were 24.4 and 32.1 °C and 16.9 $MJ \cdot m^{-2} \cdot d^{-1}$ in 2021, and 24.6 and 32.0 °C and 19.2 $MJ \cdot m^{-2} \cdot d^{-1}$ in 2022, respectively (Figure 1).

2.2. Experimental Design

The experiments were performed in a split-plot design with shading treatments as main plots and rice varieties as subplots. The experiment was replicated three times, and the subplot size was kept at 30 m^2. The three shading treatments were no shading (CK), 40% shading at the booting stage (S) and 40% shading at the grain filling stage (SS). Shading treatment was carried out separately according to the growth and development process of each variety. Black sunshade nets with 60% transmittance were used to simulate weak light stress in S and SS. Four hybrid local widely planted rice varieties, named Y-liangyou900 (YLY900), Liangyoupeijiu (LYPJ), Luoyou10 (LY10) and Chuanyou6203 (CY6203), were used in the study. Pre-germinated seeds were sown in seedbeds, after which 31 day-old seedlings were transplanted on 10 June 2021 and 2022. Transplanting was conducted at a hill spacing of 16.7 cm × 26.7 cm with two seedlings per hill in two experiment years. A total of 180 kg·N·ha^{-1} of N fertilizer as urea was applied at the basal (1 day before transplanting), early tillering (7 days after transplanting), and panicle initiation stages at a rate of 90, 36 and 54 kg·N·ha^{-1}, respectively. Phosphorus at a rate of 40 kg·P·ha^{-1} as calcium superphosphate was applied as the basal fertilizer, and potassium as potassium chloride at 100 kg·K·ha^{-1} was split equally between the basal and panicle initiation stages in each plot. The experimental field was kept flooded from transplanting until 10 d before maturity. Weeds, insects, and diseases were intensively controlled by chemicals to avoid yield loss.

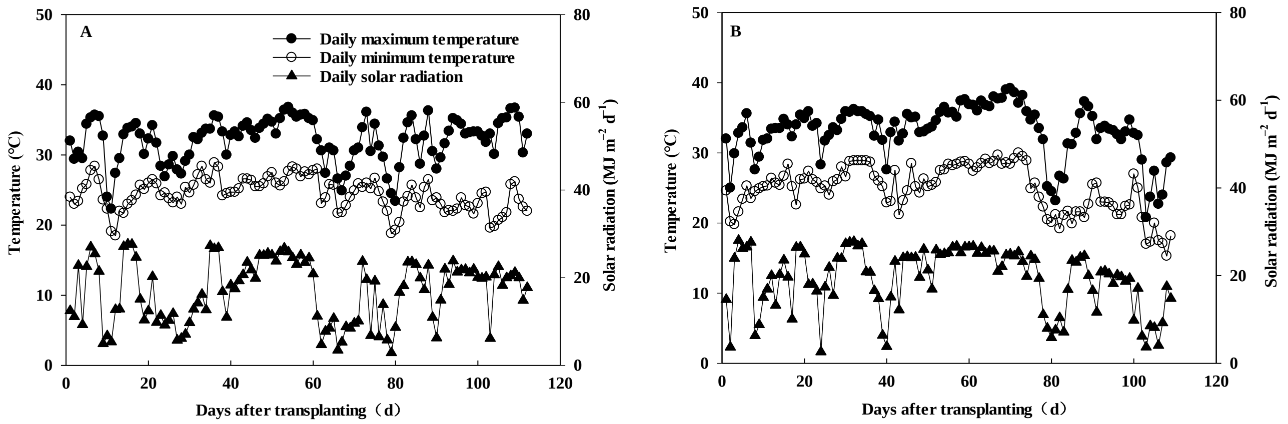

Figure 1. Daily maximum and minimum temperature and solar radiation during rice growing season from transplanting to maturity at the experimental farm of Yangtze University, Jingzhou, Hubei Province, China in 2021 (**A**) and 2022 (**B**).

2.3. Sampling and Measurements

2.3.1. Grain Yield and Other Growth Parameters

At the heading stage, the areas of the leaf subsamples were measured using a leaf area meter (LI-3100, LI-COR, Lincoln, NE, USA). At maturity, the grain yield was determined from a 5 m^2 sampling area in the center of each plot and adjusted to 14% moisture content. Plants from 12 hills in each plot were sampled to determine total dry weight and the number of total spikelets per m^2 at maturity [28]. The grain–leaf ratio (TS/LA) and dry weight spikelet production efficiency (TS/DW) were calculated as follows:

$$\text{TS/LA (cm}^{-2}\text{)} = \text{total spikelet number/leaf area at heading stage}$$

$$\text{TS/DW (g}^{-1}\text{)} = \text{total spikelet number/total dry weight at maturity}$$

2.3.2. Rice Milling and Appearance Quality

At maturity, rice grains were carefully harvested, threshed, cleaned, air-dried to a constant weight and then stored at an ambient temperature for three months. The national standard of the People's Republic of China (GB/T17891-2017) [29] was used to determine the brown rice rate (BR), milled rice rate (MR), head rice rate (HR), chalky grain percentage (CR), chalkiness degree (CD) and grain length/width ratio (L/W). The rice was milled using a rice polisher (Satake, Tokyo, Japan), and BR, MR and HR are the percentage of the weight of brown rice, milled rice and whole rice to the weight of sample rice, respectively. CR and CD were calculated via the following equations:

$$\text{Chalky grain percentage (\%)} = \text{Number of grains with chalkiness/Total number of grains observed} \times 100$$

$$\text{Chalkiness degree (\%)} = \text{Chalkiness square of chalky grains/Square of total number of grains observed} \times 100$$

2.3.3. Amylose and Protein Content

The head-rice sample was pulverized by a plant pulverizer (800A, Red sun, Yongkang, China) and then passed through a 100-mesh sieve. The amylose content (AC) was determined via iodine colorimetry [30], and the contents of albumin, glutenin, gliadin and globulin were determined by BCA method [31]. The total protein content of rice grains is the sum of the content of each protein component.

2.3.4. Rapid Viscosity Analysis (RVA) Profile Characteristics

The milled rice samples were oven-dried at a constant temperature of 60 °C, then ground into flour with a plant pulverizer, and passed through a 100-mesh sieve. The rapid viscosity-measuring instrument (RVA4500, PERTEN, Stockholm, SE, USA) was used for testing, and matching software thermocline for windows was used for analysis. The characteristic values of the RVA spectrum include peak viscosity (PV), hot viscosity (HV), final viscosity (FV), breakdown value (BKV), setback (SB), and gelatinization temperature (GT).

2.4. Statistical Analysis

Microsoft Excel 2019 (Microsoft, Redmond, WA, USA) was used for test data calculation and correlation analysis, and analysis of variance was performed with Statistix9 (Stat Soft Inc. Statistix, Tulsa, OK, USA, 1991) using the least significant difference (LSD) at 0.05 probability level. SigmaPlot (V14.0, Systat, San Jose, CA, USA) and OriginPro (2023b, Originlab, Northampton, MA, USA) were used for drawing all the figures.

3. Results

3.1. Grain Yield

Grain yield was significantly affected by shading treatment, variety and shading treatment × variety interaction in 2021 and 2022. Compared with no shading, shading significantly reduced the yields of the four varieties, and the yield loss caused by shading at the booting stage was greater than that at the grain filling stage. The yield was reduced by 53.0% under shading at the booting stage and 34.7% under shading at the grain filling stage, averaged across varieties and years, compared with no shading (Figure 2). The yield was comparable under no shading in 2021, but a significant difference was observed in 2022 among the four varieties. Under shading at the booting stage, LY10 had the highest yield followed by CY6203, LYPJ and YLY900 averaged across two years. The yield of CY6203 was significantly higher than that of the other three varieties under shading at the grain filling stage.

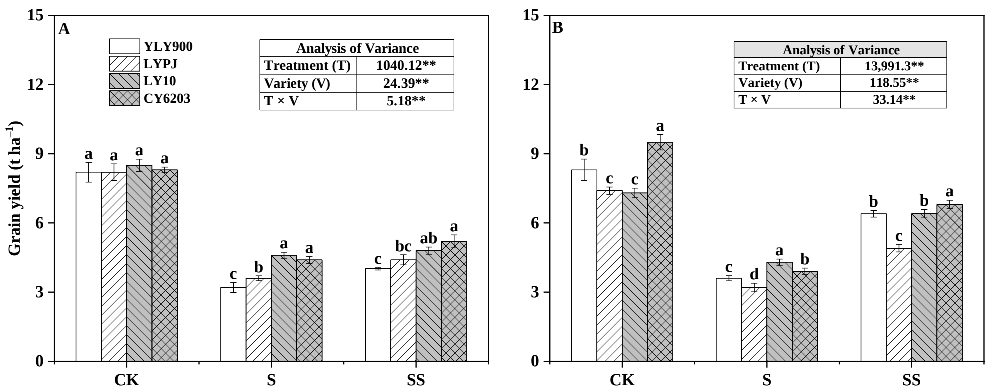

Figure 2. The yields of four varieties under different shading treatments in 2021 (**A**) and 2022 (**B**). Notes: Data are means of replicates, and the vertical bars represent the standard error. Different letters above the columns indicated statistical significance by LSD at the $p < 0.05$ level among four rice varieties within the same shading treatment. The numbers in the results of analysis of variance indicated F value. ** indicated significance at the $p < 0.01$ level. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage.

3.2. Milling Quality

Shading significantly affected HR in 2021 and 2022, but the effects on BR and MR varied in years. Compared with no shading, HR was increased by 11.4% under shading at the booting stage averaged across varieties and years, while HR was reduced by 7.2% under shading at the grain filling stage (Figure 3). BR, MR and HR were significantly affected by variety. In terms of HR, YLY900 was the highest, followed by LYPJ, and LY10 and CY6203 were lower under the three shading treatments. In addition, shading treatment × variety interaction was significant for BR, MR (except in 2022) and HR.

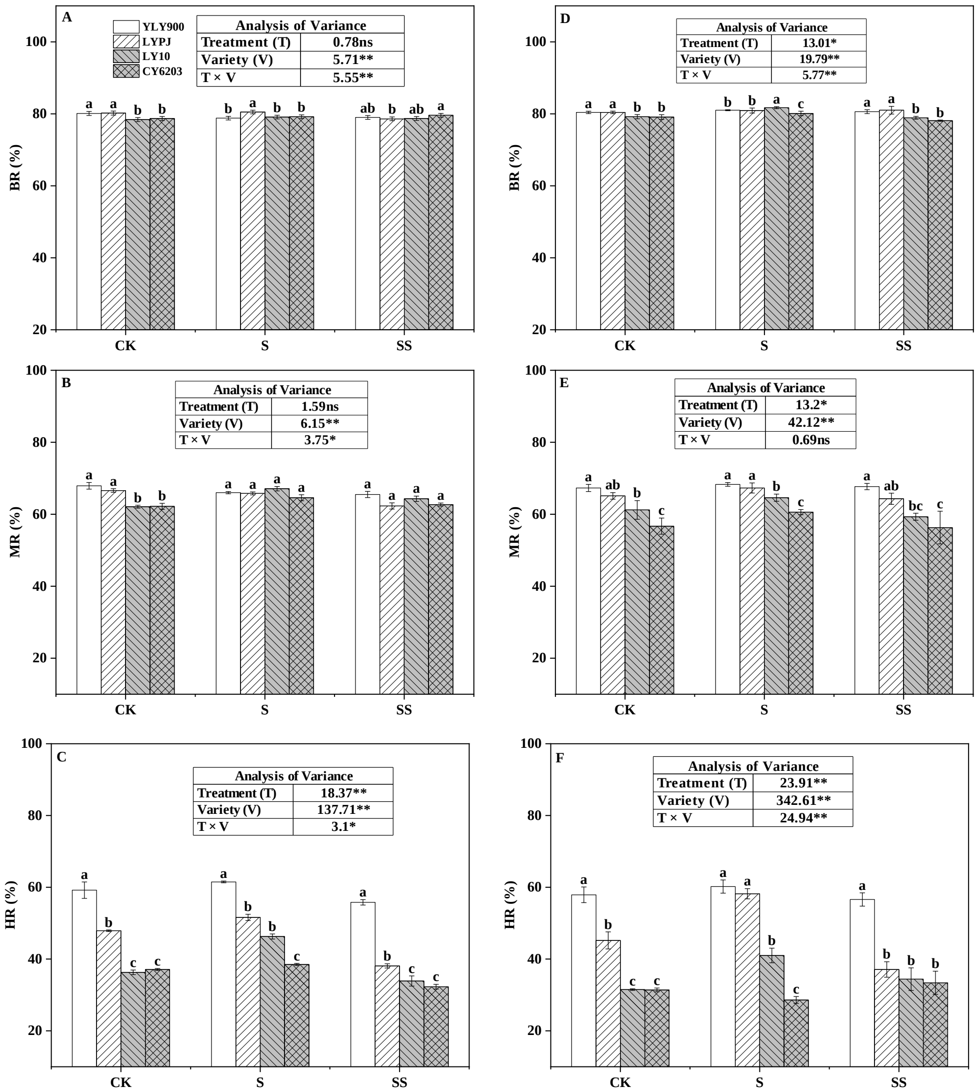

Figure 3. The rate of brown rice (**A**,**D**), milled rice (**B**,**E**) and head rice (**C**,**F**) of four varieties under different shading treatments in 2021 (**A**–**C**) and 2022 (**D**–**F**). Notes: Data are means of replicates, and the vertical bars represent the standard error. Different letters above the columns indicated statistical significance by LSD at the $p < 0.05$ level among four rice varieties within the same shading treatment. The numbers in the results of analysis of variance indicated F value. ns represented no significance at the $p < 0.05$ level. * and ** indicated significance at the $p < 0.05$, 0.01 levels, respectively. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage; BR, brown rice rate; MR, milled rice rate; HR, head rice rate.

3.3. Appearance Quality

Shading significantly affected CR and CD, compared with no shading, CR and CD were reduced by 61.6% and 73.0% under shading at the booting stage, while increased by

41.0% and 49.5% under shading at the grain filling stage averaged across four varieties and two years, respectively (Figure 4). In terms of varieties, YLY900 had lower CR and CD, followed by CY6203, while LY10 and LYPJ had higher CR and CD under the three shading treatments. L/W was only significantly affected by variety in both years and CY6203 had the highest L/W, followed by LY10, LYPJ and YLY900 averaged across two years.

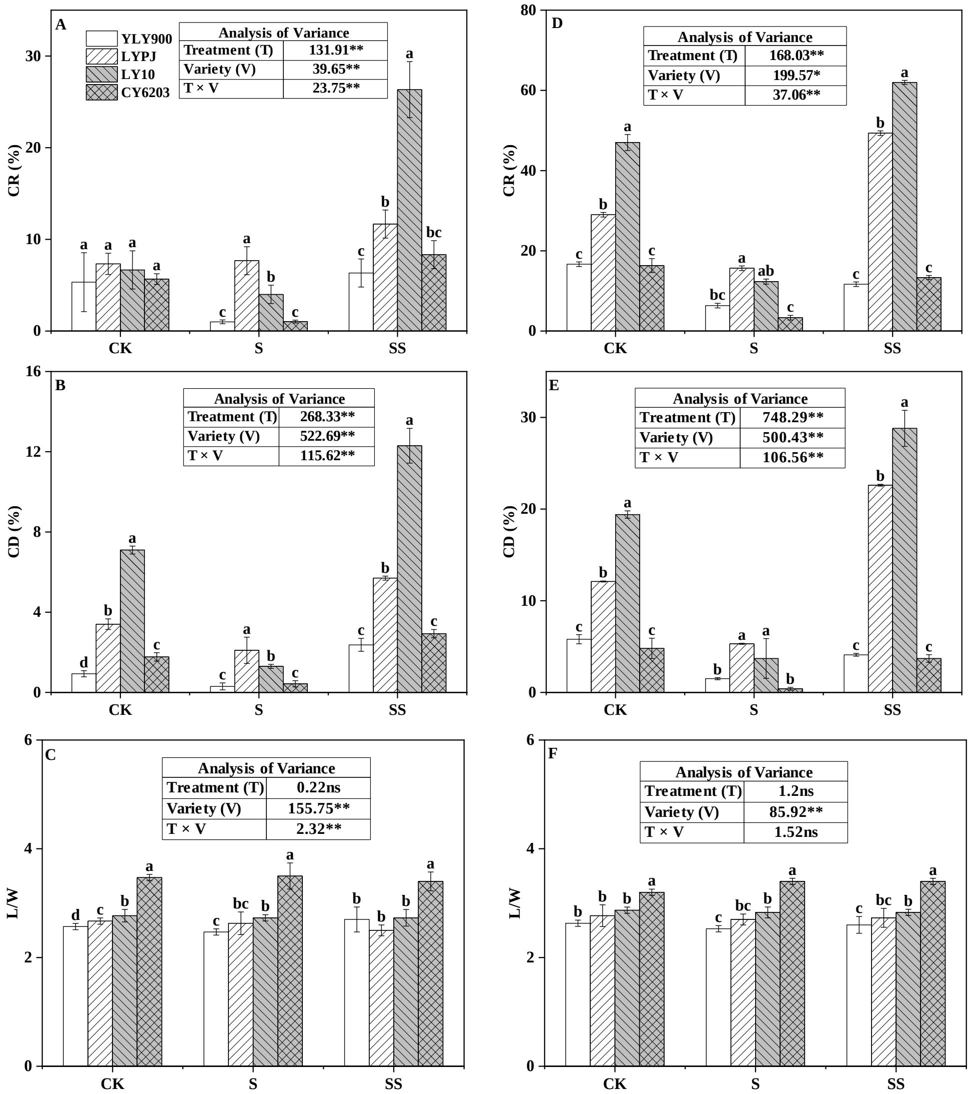

Figure 4. Chalky grain percentage (**A**,**D**), chalkiness degree (**B**,**E**) and grain length/width ratio (**C**,**F**) of four varieties under different shading treatments in 2021 (**A**–**C**) and 2022 (**D**–**F**). Notes: Data are means of replicates, and the vertical bars represent the standard error. Different letters above the columns indicated statistical significance by LSD at the $p < 0.05$ level among four rice varieties within the same shading treatment. The numbers in the results of analysis of variance indicated F value. ns represented no significance at the $p < 0.05$ level. * and ** indicated significance at the $p < 0.05$, 0.01 levels, respectively. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage; L/W, grain length/width; CR, chalkiness grain percentage; CD, chalkiness degree.

3.4. Amylose Content

AC was significantly affected by shading treatment, variety and shading treatment × variety interaction in 2021 and 2022 (Figure 5). The average amylose content of the four varieties under no shading, shading at the booting stage and shading at the grain filling stage was 24.3%, 16.6% and 19.3% averaged across both years, respectively (Figure 5). Compared with no shading, shading at the booting stage and shading at the grain filling stage decreased the AC by 31.7% and 20.6%, respectively. These results suggested shading significantly decreased AC, and the reduction was greater at the booting stage than that at the grain filling stage. The AC performance of the four varieties was inconsistent in shading treatments and experimental years.

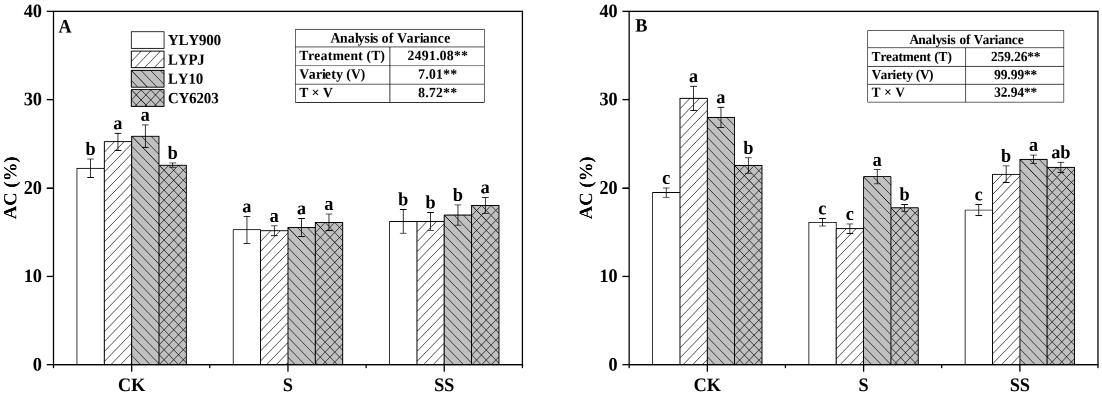

Figure 5. Amylose content of four varieties under different shading treatments in 2021 (**A**) and 2022 (**B**). Notes: Data are means of replicates, and the vertical bars represent the standard error. Different letters above the columns indicated statistical significance by LSD at the $p < 0.05$ level among four rice varieties within the same shading treatment. The numbers in the results of analysis of variance indicated F value. ** indicated significance at the $p < 0.01$ level. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage; AC, amylose content.

3.5. Protein Content

Protein content was significantly increased by shading, with a greater effect observed during the grain filling stage compared to the booting stage. Compared with no shading, shading at the booting stage increased the content of albumin, globulin, gliadin, glutenin and total protein by 17.0%, 8.5%, 37.0%, 2.9% and 5.0%, and shading at the grain filling stage increased by 34.0%, 27.0%, 50.0%, 15.7% and 18.5%, respectively, averaged across varieties and years. In addition, protein content was significantly affected by variety (except for the content of globulin in 2021) and shading treatment × variety interaction in both years (Table 1). The total protein contents of the four varieties were comparable under no shading in 2022, but in 2021, YLY900 had the highest total protein content, but difference was only significant between YLY900 and CY6203 (Figure 6). Under shading at the booting stage, total protein contents of YLY900 and LYPJ were significantly higher than the other two varieties. While under shading at the grain filling stage, the protein content of CY6203 was the highest among the four varieties, and the performance of the other three varieties varied in years.

Table 1. The contents of protein components of different rice varieties under different shading treatments in 2021 and 2022.

Treatment	Variety	2021				2022			
		Albumin	Globulin	Gliadin	Glutelin	Albumin	Globulin	Gliadin	Glutelin
		%				%			
CK	YLY900	0.17 c	0.82 b	0.25 a	6.08 a	0.18 b	0.55 bc	0.24 a	5.49 a
	LYPJ	0.32 a	0.79 b	0.24 a	5.63 b	0.18 b	0.51 c	0.23 a	5.03 a
	LY10	0.29 ab	0.80 b	0.21 b	5.81 ab	0.24 a	0.58 b	0.21 a	5.41 a
	CY6203	0.27 b	0.86 a	0.22 ab	5.48 b	0.25 a	0.72 a	0.22 a	5.08 a
S	YLY900	0.30 a	0.84 a	0.32 b	6.47 a	0.34 a	0.68 b	0.33 a	6.06 a
	LYPJ	0.28 a	0.84 a	0.30 b	6.59 a	0.23 d	0.64 b	0.29 a	6.09 a
	LY10	0.29 a	0.84 a	0.31 b	5.43 b	0.26 c	0.80 a	0.29 a	5.00 b
	CY6203	0.22 b	0.82 a	0.35 a	5.10 b	0.29 b	0.66 b	0.33 a	4.45 c

Table 1. *Cont.*

Treatment	Variety	2021				2022			
		Albumin	Globulin	Gliadin	Glutelin	Albumin	Globulin	Gliadin	Glutelin
		%				%			
SS	YLY900	0.29 ab	0.91 b	0.39 a	6.45 ab	0.47 a	0.87 a	0.40 a	6.49 a
	LYPJ	0.27 b	0.98 a	0.28 c	6.57 ab	0.37 b	0.86 a	0.34 b	6.16 b
	LY10	0.31 a	0.92 b	0.36 b	6.42 b	0.32 c	0.85 a	0.32 bc	6.03 b
	CY6203	0.30 a	0.91 b	0.37 ab	6.73 a	0.19 d	0.84 a	0.28 c	6.01 b
Analysis of Variance									
	Treatment (T)	43.68 **	214.74 **	652.71 **	104.86 **	505.18 **	331.42 **	284.81 **	258.81 **
	Variety (V)	19.19 **	2.39 ns	21.84 **	24.40 **	168.82 **	6.66 **	11.14 **	9.08 **
	T × V	30.15 **	13.49 **	13.45 **	21.88 **	130.28 **	9.32 **	4.71 **	18.09 **

Notes: Different letters indicated statistical significance by LSD at the $p < 0.05$ level among four rice varieties within the same shading treatment. The results of the comparison mean for each treatment independent from others. The numbers in the results of analysis of variance indicated F value. ns represented no significance at the $p < 0.05$ level. ** indicated significance at the $p < 0.01$ level. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage.

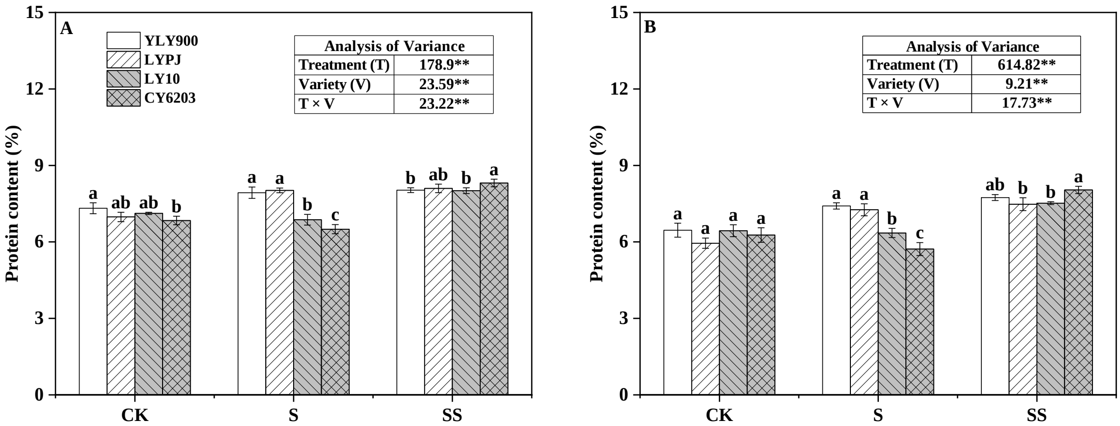

Figure 6. Protein content of different rice varieties under shading at different periods in 2021 (**A**) and 2022 (**B**). Notes: Data are means of replicates, and the vertical bars represent the standard error. Different letters above the columns indicated statistical significance by LSD at the $p < 0.05$ level among four rice varieties within the same shading treatment. The numbers in the results of analysis of variance indicated F value. ** indicated significance at the p 0.01 level. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage.

3.6. RVA Profile Characteristics

Shading led to a significant decrease in PV and BKV, and the effect was greater at the grain filling stage than that at the booting stage. Compared with no shading, PV and BKV were decreased by 9.7% and 19.0% under shading at the booting stage, and 16.2% and 35.6% under shading at the grain filling stage averaged across varieties and years, respectively (Tables 2 and 3). However, shading significantly increased SB, and 41.5% and 84.5% higher SB was observed under shading at the booting stage and at the grain filling stage than that under no shading, respectively. The effects of shading on HV and GT differed in 2021 and 2022, but significantly affected FV in both years. In terms of varieties, YLY900 exhibited the highest PV and BKV, followed by LY10, CY6203 and LYPJ in 2021 averaged across three shading treatments. However, YLY900 and CY6203 displayed higher PV and BKV than the other varieties in 2022. YLY900 and CY6203 showed consistently lower SB than the other two varieties in both years.

Table 2. RVA profile characteristics of four varieties under different shading treatments in 2021.

Treatment	Variety	PV	HV	BKV	FV	SB	GT
				cP			°C
CK	YLY900	3740.7 a	2266.0 ab	1474.7 a	3307.3 b	−433.3 d	81.4 a
	LYPJ	3348.0 c	2254.3 ab	1093.7 c	3796.7 a	448.7 a	81.1 a
	LY10	3553.3 b	2316.3 a	1237.0 b	3884.3 a	331.0 b	80.8 a
	CY6203	3298.7 c	2199.0 b	1099.7 c	3390.7 b	92.0 c	75.8 b
S	YLY900	3647.3 a	2436.7 b	1210.7 b	3475.7 b	−171.7 c	81.9 a
	LYPJ	2978.7 c	2079.0 c	899.7 c	3504.3 b	525.7 a	80.3 b
	LY10	3541.7 a	2576.3 a	965.3 c	3988.0 a	446.3 b	80.8 ab
	CY6203	3322.3 b	1995.7 c	1326.7 a	3163.3 c	−159.0 c	76.2 c
SS	YLY900	3318.7 a	2272.3 a	1046.3 a	3266.7 b	−52.0 d	81.9 a
	LYPJ	2769.3 d	2113.7 b	655.7 c	3294.3 b	525.0 b	81.5 a
	LY10	2910.3 c	2306.3 a	604.0 c	3514.3 a	604.0 a	81.9 a
	CY6203	3050.7 b	2187.3 ab	863.3 b	3245.7 b	195.0 c	80.7 a
Analysis of Variance							
	Treatment (T)	124.72 **	2.25 ns	705.53 **	43.91 **	1513.50 **	6.53 ns
	Variety (V)	85.28 **	38.82 **	132.60 **	89.77 **	3061.81 **	21.99 **
	T × V	9.22 **	12.51 **	27.50 **	13.57 **	127.40 **	3.00 *

Notes: Different letters indicated statistical significance by LSD at the $p < 0.05$ level among four rice varieties within the same shading treatment. The results of the comparison mean for each treatment independent from others. The numbers in the results of analysis of variance indicated F value. ns represented no significance at the $p < 0.05$ level. * and ** indicated significance at the $p < 0.05$, 0.01 levels, respectively. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage; PV, peak viscosity; HV, hot viscosity; FV, final viscosity, BKV, breakdown value; SB, setback; GT, gelatinization temperature.

Table 3. RVA profile characteristics of four varieties under different shading treatments in 2022.

Treatment	Variety	PV	HV	BKV	FV	SB	GT
				cP			°C
CK	YLY900	3228.0 a	2124.7 ab	1103.3 a	3374.0 a	146.0 c	85.2 b
	LYPJ	2331.3 d	1859.0 c	472.3 d	2974.7 b	643.3 b	89.9 ab
	LY10	2532.3 c	2014.3 bc	518.0 c	3355.7 a	823.3 a	91.0 a
	CY6203	2935.3 b	2186.3 a	749.0 b	3050.7 b	115.3 d	87.8 ab
S	YLY900	2936.7 a	2137.0 a	799.7 a	3133.7 a	197.0 d	89.1 a
	LYPJ	1702.0 c	1485.7 d	216.3 d	2591.0 b	889.0 b	90.7 a
	LY10	2226.7 b	1865.7 b	361.0 c	3167.0 a	940.3 a	92.3 a
	CY6203	2188.0 b	1688.0 c	500.0 b	2575.7 b	387.7 c	89.6 a
SS	YLY900	2888.3 a	2091.3 a	797.0 a	3188.3 a	300.0 d	88.9 a
	LYPJ	1709.0 d	1534.7 d	174.3 d	2739.7 c	1030.7 a	90.9 a
	LY10	2035.0 c	1654.3 c	380.7 c	2978.3 b	943.3 b	88.1 a
	CY6203	2238.7 b	1766.3 b	472.3 b	2690.0 c	451.3 c	89.6 a
Analysis of Variance							
	Treatment (T)	2730.16 **	708.01 **	1535.65 **	391.68 **	555.90 **	9.69 *
	Variety (V)	574.39 **	106.63 **	2344.58 **	150.21 **	4991.48 **	2.75 ns
	T × V	13.28 **	14.44 **	20.16 **	6.62 **	69.86 **	1.22 ns

Notes: Different letters indicated statistical significance by LSD at the $p < 0.05$ level among different rice varieties within the same shading treatment. The results of the comparison mean for each treatment independent from others. The numbers in the results of analysis of variance indicated F value. ns represented no significance at the $p < 0.05$ level. * and ** indicated significance at the $p < 0.05$, 0.01 levels, respectively. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage; PV, peak viscosity; HV, hot viscosity; FV, final viscosity, BKV, breakdown value; SB, setback; GT, gelatinization temperature.

3.7. Total Spikelets·m^{-2}, Grain–leaf Ratio and Dry Weight Spikelet Production Efficiency

The number of total spikelets·m^{-2}, grain–leaf ratio and dry weight spikelet production efficiency were significantly affected by variety and shading treatment in 2021 and 2022 (Figure 7). Compared with no shading, shading at the booting stage reduced the number of total spikelets·m^{-2}, grain–leaf ratio and dry weight spikelet production efficiency by 45.3%, 44.0% and 23.5% averaged across varieties and years, respectively. Shading at the grain filling stage led to a decrease of 9.8% and 19.8% in the number of total spikelets·m^{-2} and grain–leaf ratio, respectively, but an increase of 15.8% in dry weight spikelet production efficiency, compared with no shading. Correlation analysis showed that dry weight spikelet production efficiency has a significantly positive correlation with chalky grain percentage and chalkiness degree (Figure 8).

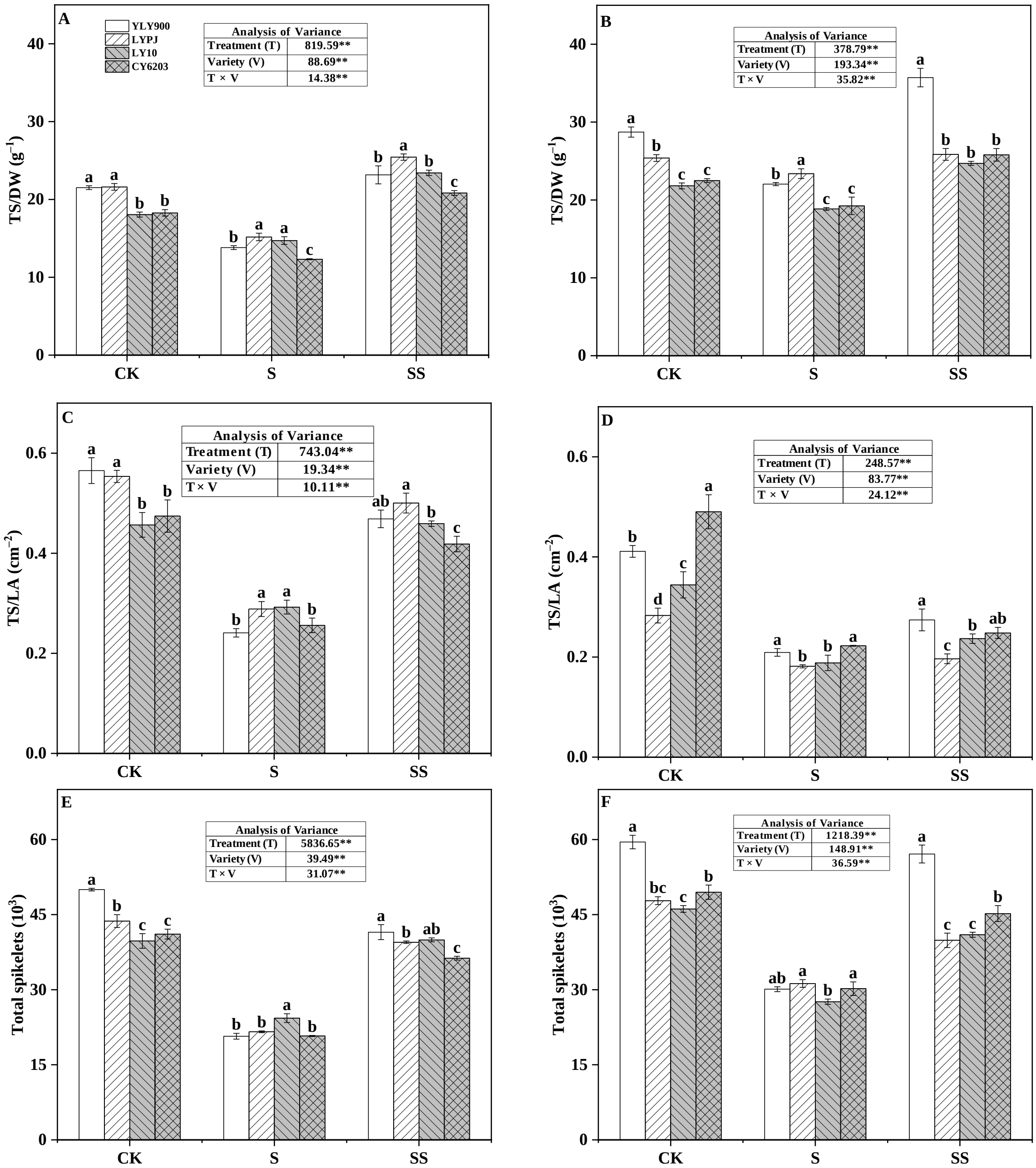

Figure 7. Dry weight spikelet production efficiency (**A**,**B**), grain–leaf ratio (**C**,**D**) and total spikelets (**E**,**F**) of four varieties under different shading treatments in 2021 (**A**–**E**) and 2022 (**B**–**F**). Notes: Data are means of replicates, and the vertical bars represent the standard error. Different letters above the columns indicated statistical significance by LSD at the $p < 0.05$ level among four rice varieties within the same shading treatment. The numbers in the results of analysis of variance indicated F value. ** indicated significance at the $p < 0.01$ levels. CK, no shading; S, shading at the booting stage; SS, shading at the grain filling stage; TS/DW, dry weight spikelet production efficiency; TS/LA, grain–leaf ratio.

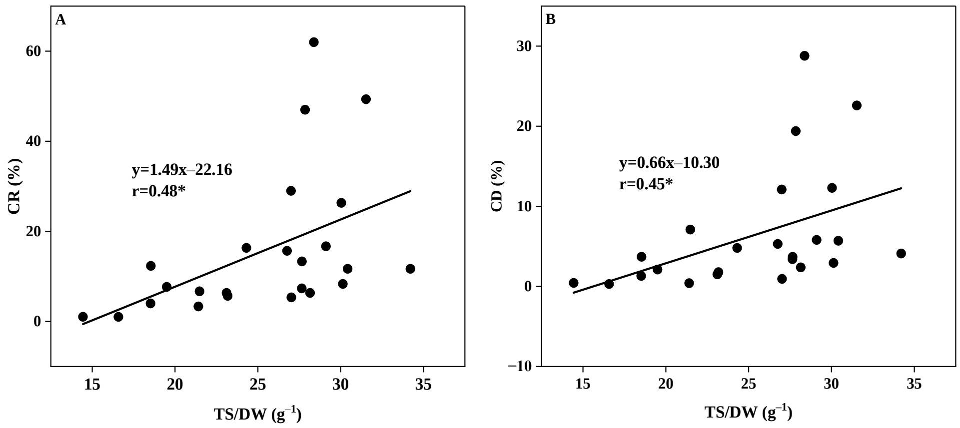

Figure 8. The relationships between dry weight spikelet production efficiency (TS/DW) and chalky grain percentage (CR) (**A**) and chalkiness degree (CD) (**B**) under different shading treatments in 2021 and 2022 (n = 24). Notes: * indicates significance by LSD at the $p < 0.05$ levels.

4. Discussion

4.1. Effects of Shading at the Booting and Grain Filling Stages on Rice Milling Quality

Regarding the effect of weak light stress on rice quality, previous studies mostly focused on shading at the grain filling stage [12,32,33]. Weak light at the grain filling stage significantly reduced the milling quality of rice, causing a decrease in BR, MR and HR [33]. The present study also showed shading at the grain filling stage resulted in an average decrease of 7.2% in HR in both years. As for the effect of shading at the booting stage on rice milling quality, the results of the few studies that did exist differed [11,25,34]. The study of Jiang et al. (2013) showed shading at the booting stage reduced HR [11], but Liu et al. (2006) pointed out it increased HR [25]. In the study, we found shading at the booting stage had no significant effect on BR and MR, but significantly increased HR, with an average increase of 11.4% in two years compared with no shading, which was in accordance with the previous results [25,34]. Weak light stress at the booting stage can affect canopy photosynthesis, and lead to insufficient carbon source for young panicles, thus hindering the differentiation of the spikelets, and resulting in more sterile spikelets. Therefore, the sink size (total spikelets·m^{-2}) of rice is decreased obviously, which leads to the increase in the source-sink ratio, so the grains are enriched well at the grain filling stage, thus improving HR under shading at the booting stage. In addition, the total protein content of rice under shading at the booting stage was higher than that under no shading, which might also result in an increase in HR. Higher protein content in endosperm could increase the hardness and toughness of rice grains, and thus reduce grain fracture and increase HR [35,36].

4.2. Effects of Shading at the Booting and Grain Filling Stages on Rice Appearance Quality

CR and CD are important indexes for evaluating appearance quality [37]. In this study, CR and CD were greatly affected by shading, but their responses to shading of different periods were different. Compared with no shading, CR and CD were increased significantly under shading at the grain filling stage but were reduced significantly under shading at the booting stage. These results showed shading at the booting stage was beneficial in improving the appearance quality of rice, which was consistent with a previous study [25]. The occurrence of rice chalkiness is mainly related to the level of source and sink during grain growth and development. Insufficient dry matter accumulation leads to poor grain filling and loose arrangement of starch particles in endosperm, which will lead to the increase in the chalkiness degree [38]. Grain–leaf ratio and dry weight spikelet production efficiency are both key indicators to reflect the coordination of source-sink relationship in rice. The higher the grain–leaf ratio and dry weight spikelet production efficiency, the less substance can be allocated to a single grain, the higher CR and CD of rice grains, and the worse the appearance quality [39]. In the study, grain–leaf ratio and dry weight spikelet production efficiency under shading at the booting stage were significantly lower than those of the other two shading treatments. Shading at the booting stage significantly affected the differentiation, growth and development of spikelets, although the ability of dry matter accumulation decreased after light recovery. However, the decrease in grain number led to the increase in assimilates allocated to individual spikelets, which resulted in better grain fullness and thus improved the appearance quality of rice [40].

4.3. Effects of Shading at the Booting and Grain Filling Stages on Rice Nutritional and Cooking and Eating Quality

The nutritional and cooking and eating quality of rice are two indicators that consumers generally pay close attention to. The cooking and eating quality of rice is a complex index, and generally can be quantified by the interrelationships among starch, protein and other chemicals such as amino acids in rice [41,42]. Amylose content is a key index for evaluating the cooking and eating quality; in general, amylose content was relatively low in rice varieties with good cooking and eating quality [15,16]. There was a significantly positive correlation between AC and light intensity in rice, and shading significantly reduced AC [33,43]. Our study also found that shading at the booting stage and at the grain filling stage caused AC to decline compared with no shading. Except for amylose content, protein content is another index affecting cooking and eating quality, because many studies have reported a significantly negative correlation between protein content and cooking and eating quality in rice [19,21]. Furthermore, protein content is an important index for evaluating the nutritional quality of rice [18]. In the present study, compared with no shading, shading at the booting stage and shading at the grain filling stage significantly increased the protein content of rice, which was in line with the results of previous studies [22,44]. The protein content in rice grains is 5–10% [45], and when it is higher than 7%, the eating quality will decline [19]. Total protein content under no shading, shading at the booting stage and shading at the grain filling stage was 6.7%, 7.0% and 7.9% averaged across two years in the study, respectively. This suggested that shading at the booting stage did not lead to a decrease in rice eating quality compared with no shading, while shading at the grain filling stage would lead to a significant decline, which might be due to the fact that the grain filling stage was the key period for the formation of grain quality and the light condition at this stage had a more important effect on grain quality. Thus, shading led to the increase in protein content, which affected the eating quality, but improved the nutritional quality of rice. In addition, shading had opposite effects on amylose and protein content, which might be related to an imbalance in carbon and nitrogen metabolism [46].

Pasting viscosity is another critical indicator for rice cooking and eating quality and is commonly measured with a rapid visco analyzer (RVA). It has been reported that rice varieties with high peak viscosity, large breakdown value, small setback and low gelatinization temperature have good cooking and eating quality [47]. In the study, regardless of the shading at booting or grain filling stage, we found the peak viscosity and breakdown values were decreased significantly, while the setback value was increased significantly, indicating that shading affected the RVA profile characters, and the effect of shading at the grain filling stage was greater than that at the booting stage.

4.4. Response of Grain Yield and Quality of Different Rice Varieties to Shading

The responses of different rice varieties to shading were diverse. Those varieties with higher shading tolerance have a higher light-harvesting and -use efficiency, grain filling rate [48], photosynthetic capacity and carbon and nitrogen metabolism stability [49], and thus have a lower reduction in grain yield [50] and a greater quality stability [51]. In this study, the appearance and eating quality of YLY900 and CY6203 were better under shading at the booting stage, while the milling quality of YLY900 and LYPJ was better than that of the other two varieties. Under shading at the grain filling stage, the appearance, nutritional and eating quality of YLY900 and CY6203 were better, but the milling quality of YLY900 and LYPJ was better than that of the other varieties. Thus, YLY900 and CY6203 especially YLY900 can obtain good grain quality even under weak light, which may be due to the stronger photosynthetic capacity of YLY900 [52]. However, YLY900 showed lower yield, but CY6203 obtained higher yield under shading. Therefore, CY6203 was beneficial to synergistically improve grain yield and quality under shading. CY6203 might be adapted to weak light condition, as it was bred and released in Sichuan province, China, a representative rice production region with low light intensity [43].

5. Conclusions

Rice quality was significantly influenced by shading, however, the effects varied in different growth stages. Compared to no shading, shading at the booting stage could significantly improve the milling, appearance and nutritional quality, and did not reduce the cooking and eating quality. The improved rice quality under shading at the booting stage was due to the increased allocation of dry matter caused by reduced spikelet production. Conversely, shading at the grain filling stage resulted in the deterioration of grain quality. Additionally, the study revealed that YLY900 could maintain good quality but display low yield under shading. Therefore, our next research will use the high-yield or high-quality varieties currently widely planted as materials to screen out both high-yield and high-quality varieties under shading condition for coping with climate change.

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