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Changes in chemical composition and starch structure in rice noodle cultivar influence Rapid Visco analysis and texture analysis profiles under shading

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ABSTRACT

GuichaoII, a rice variety with high amylose content widely used to make rice noodles, exhibits high hardness (631.07–729.43), gel consistency (8.47–9.47 mm), and hold viscosity/peak viscosity (HPV/PKV) (0.85–0.88); however, it has a low protein content (5.74–6.96%) and swelling factor (5.49–9.77). Herein, GuichaoII was subjected to low-light stress (53% reduction) during the grain filling stage. The amylose content and crystallinity of GuichaoII and the control variety Shuhui 498 decreased while the protein content, short-chain branch ratio, and degree of branching increased, which affected the ability of the rice flour to absorb water and expand during the gelatinization process. The PKV, HPV, breakdown viscosity, and final viscosity were significantly reduced, while the hardness was significantly increased, and the gel consistency and the gelatinization quality of the rice were reduced, severely limiting the processing and production of rice noodles.

1. Introduction

Rice is a key dietary source of carbohydrates. Rice noodle, also called rice vermicelli or rice pasta, is one of the most popular types of noodles, which is typically made from long grain rice with low or medium amylose content. The GuichaoII is a high-yield conventional rice variety with a high amylose content (approximately 20%). Owing to its special viscosity characteristics, GuichaoII is widely used throughout China, Vietnam, Thailand, and other Southeast Asian countries as a high-quality raw material for various types of rice noodles and other rice-based foods (Nip, 2007).

The quality of rice noodles depends primarily on the physical and chemical properties of rice starch, which determines its specific structural network (Wu, Meng, Yang, Tao & Xu, 2015). Starch is a semicrystalline granular substance composed of amylose and amylopectin. It is synthesized in plant cells, where it accumulates to form a layered structure with alternating crystalline and amorphous layers (Gallant, Bouchet, & Baldwin, 1997; Zeeman, Kossmann, & Smith, 2010). The crystal structure of starch consists of a regular arrangement of amylopectin side chains and amylose molecules that form a double helix structure (Cai et al., 2015). The swelling capacity of the starch and the ratio of amylose to amylopectin both affect the quality of rice flour (Sun, Zhang, Xiong, & Dai, 2013). The crystallization of amylose facilitates the formation of a continuous network and can affect its physicochemical properties, including its thermal properties, swelling factors, and digestive ability (Lee, Lee, & Chung, 2017). These, in turn, affect the quality of the rice flour, including its chewiness, springiness, and retrogradation rate. Rice with higher amylopectin content has higher viscosity and digestibility, while amylose content is related to the hardness, cohesiveness, adhesiveness, and resilience of the rice (Wu, Shu, & Xia, 2001). The amylopectin chain length distribution also affects the dynamic elasticity and recovery rate of rice gel (Lu, Sasaki, Li, Yoshihashi, Li & Kohyama, 2009). Rapid Visco Analysis (RVA) can reflect the quality of rice noodles; RVA profiles and textural parameters of gels formed in the analyzer strongly correlate with noodle texture (Xuan, Yi, Liang, Wei, & Li, 2020; Yoenyongbuddhagal & Noomhorm, 2002).

The development of rice grains during the grain filling stage is

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strongly affected by light. In recent years, low light has gradually become a global climate phenomenon, leading to a decline in crop yields and quality in the field (Shao et al., 2020). In the Sichuan Basin, light levels are typically poor during the grain filling stage of rice growth (approximately June-September) owing to adverse weather conditions such as cloud and rain. The reduced light intensity reduces both the amylose and total starch contents of the rice, while lowering the proportiont of short chain amylopectin and increasing that of long chain amylopectin (Deng et al., 2018). Such a change in starch composition affects the appearance and pasting properties of the grain, with such grains exhibiting a higher degree of chalkiness, reduced peak viscosity, and breakdown (Deng et al., 2021; Wang, Deng, Ren, & Yang, 2013). In corn, shading reduces the amylose content and increases both the size of amylose particles and the proportion of long chains. This enhances the relative crystallinity and amorphous ratio of the starch, increases its viscosity and susceptibility to degradation, and reduces its thermal stability (Shi, Gu, Lu, & Lu, 2018). During the wheat grain filling process, shading increases the starch's peak, trough, final, and breakdown viscosities by increasing its relative crystallinity and the proportion of large particles, while reducing the total starch content and gelatinization temperature of the flour (Liu et al., 2017).

In this study, we grew GuichaoII rice (the raw material for rice noodles) and Shuhui498 (as a control) under light levels reduced 53% below ambient. The effects of low light stress on starch formation and rice quality were determined by analyzing; 1) the structural properties of the rice starch granules; 2) the RVA and TPA; and 3) the relationship between the starch structure and the RVA and TPA characteristics. Our findings can improve the quality of rice noodles by controlling the starch properties under shading conditions to optimize the gelatinization characteristics.

2. Materials and methods

2.1. Materials

Two indica rice varieties of different phenotypes (GuichaoII with high amylose content and chalkiness, and Shuhui498 with the opposite characteristics as a control variety) were planted in 2019 in Wenjiang (30°43'N, 103°52'E), Chengdu, China. The soil was a medium loam with 29.92 g kg⁻¹ organic matter, 1.47 g kg⁻¹ total N, 1.04 g kg⁻¹ total P, 16.43 g kg⁻¹ total K, 72.65 mg kg⁻¹ alkali hydrolysable N, 43.83 mg kg⁻¹ Olsen-P, and 164.1 mg kg⁻¹ exchangeable K. The seedlings were raised in the seedbed for 30 days, after which time two seedlings from each hill were transplanted to the field on May 23, 2019, with a spacing of 33.3 cm × 20.0 cm. The plot area measured 3 m × 10 m, and a random complete block design was adopted with 3 replicates. After rice heading, the plants were covered with a screen made of white cotton yarn for 30 days, which blocked approximately 53% of the light intensity. Nets were placed 50 cm above the plants to provide good ventilation. Plants without shading were used as controls and grown under similar microclimates (i.e., similar temperature conditions).

2.2. Starch isolation

Rice starch was isolated from mature grains as previously described by Wei et al. (2010). Head rice (20 g) was immersed in sodium metabisulfite solution (100 ml) containing alkaline protease (10 mg/g) and maintained at 42 °C for 24 h. After homogenization, the mixed water sample was filtered through a 200-mesh sieve, and the filter residue was thoroughly cleaned with sodium sulfite aqueous solution. The collected filtrate was left to stand overnight, after which the supernatant was decanted and sodium sulfite solution was added. The above steps were repeated 5–8 times until the starch was white and the supernatant was transparent, after which the supernatant was removed and the starch was allowed to dry before being transferred to an incubator to assess water balance over one week.

2.3. Determination of total starch, amylose, and amylopectin contents

The total starch and amylose contents in the head rice were measured using the amyloglucosidase- α -amylase method (AOAC 996.11) and iodine colorimetry (ISO 6647-2-2011), respectively. The amylopectin content was calculated by subtracting the amylose content from the total starch content.

2.4. Determination of the degree of branching and amylopectin branch chain length distribution

The degree of branching and amylopectin branch chain length distribution of the starch samples were determined using NMR spectroscopy (Bruker BioSpin GmbH, Rheinstetten, Germany) and highperformance anion-exchange chromatography (ICS-5000, Thermo Fisher Scientific, CA, USA) with a Dionex[™] CarboPac[™] PA10 anionexchange column, respectively, following the procedure of Zou, Xu, Wen, and Yang (2020).

2.5. Measurements of starch granule morphology and size distribution

After sifting through a 100-mesh sieve, the starch sample (approximately 100 g) was analyzed by scanning electron microscopy (SEM, Zeiss Merlin Compact). The starch particle size was determined using a laser diffraction particle size analyzer (Mastersizer 2000, Malvern, England; measurement range: $0.1-2000 \ \mu$ m) whose accuracy was verified using Malvern standard glass particles. The size distribution was expressed according to the volume of the corresponding spheres.

2.6. X-ray diffraction

The starch was analyzed by X-ray diffraction (XRD) spectroscopy using an ADVANCE X-ray diffractometer (D8, Bruker, Germany). The relative crystallinity of the starch was measured as described by Wei et al. (2010).

2.7. Fourier transform infrared spectrum measurements

Starch samples (approximately 50 mg) were mixed with potassium bromide and IR spectra were measured using a Nicolet Iz-10 Fourier-transform infrared spectrometer (Thermo, USA). The spectra were recorded between 400 and 4000 cm⁻¹ at a spectral resolution of 4 cm⁻¹ over 32 scans.

2.8. Analysis of swelling factor and thermal properties

The swelling factor of the rice starch was determined following the method described by Zhu et al. (2019). The gelatinization properties of each sample were determined using differential scanning calorimetry (Q2000, DSC, TA Instruments, DE, USA) following the method described by Shi et al. (2018).

2.9. Determination of pasting properties of rice

The pasting properties were investigated by Rapid Visco-Analysis (RVA, Tech-master; Newport Scientific, Warriewood, Australia) according to the methods of Zhu, Liu, Sang, Gu, and Shi (2010).

A A single layer of grains of a subsample of cooked rice flour (1 g) was deposited on a flat glass dish. Texture profile analysis (TPA) was then conducted following the method of Li, Prakash, Nicholson, Fitz-gerald, and Gilbert (2016). The gel consistency was evaluated according to Cagampang, Perez, and Juliano (1973).

2.10. Statistical analysis

Sample characterization, measurements were performed in triplicate

unless otherwise specified. Analysis of variance (ANOVA) was used to examine the differences using the SPSS 19.0 statistical analysis software. Results with p < 0.05 were considered statistically significant.

3. Results and discussion

3.1. Difference in RVA profiles and TPA of rice flour

RVA is widely used to evaluate the pasting qualities of rice noodles (Tong et al., 2014), which are known to be affected to a certain extent by reduced light levels (Wang et al., 2013).

As an important raw material for rice noodle processing, GuichaoII has a significantly lower peak viscosity (PKV) and breakdown viscosity (BDV) than Shuhui498. The hold viscosity (HPV), final viscosity (CPV), setback (SBV), and Pasting temperature (PaT) of GuichaoII are significantly higher than those of Shuhui498 (Table 1). Such characteristics give the rice flour made with GuichaoII superior ductility and tensile strength (Bhattacharya, Zee, & Corke, 1999), which is more conducive to the production of rice noodles.

After shading, the PKV, HPV, and BDV of GuichaoII and Shuhui498 were significantly higher than those observed under the normal light. Interestingly, while the CPV of GuichaoII was also significantly higher after shading, no difference was observed in the CPV of shuhui498 (Table 1). Although the SBV, PaT, and HPV/PKV all decreased slightly after shading, these reductions can be considered insignificant. Shading stress primarily affects the adhesiveness of the rice by reducing the PKV, HPV, BDV, and CPV. The HPV/PKV (Tables 1 and 2) is also proportional to the hardness and cook weight of the noodles (Collado & Corke, 1997).

TPA showed that the hardness, springiness, adhesiveness, chewiness, cohesiveness, and restorative properties of GuichaoII rice flour were significantly higher than those of Shuhui498 (Table 2). The HPV, CPV, SBV and PaT all exhibited the same trend. The stickiness and extensibility of the rice noodles increase with the gelatinization temperature.

After shading, both varieties showed no notable difference in their eating characteristics except for a reduction in the hardness of GuichaoII. The gel consistency of GuichaoII was also significantly higher than that of Shuhui498, although it decreased slightly after shading. GuichaoII has a higher PKV, CPV, SBV, and PaT than Shuhui498, which improves the chewiness, springiness, and gel consistency of rice noodles made from it.

3.2. Amylose and protein contents

Starch obtained from GuichaoII has a higher pasting temperature that that from Shuhui498 owing to its higher amylose content (14.17% and 14.98% higher than Shuhui498 under normal and shaded conditions, respectively, Table 2). Similar results have been observed in maize (Ning et al., 2005) and rice (Chung, Liu, Lee, & Wei, 2011). At present,

 Table 1

 RVA and thermal properties of GuichaoII and Shuhui498.

the amylose content of the starch is the main influence on the production of rice noodles (Marti & Pagani, 2013). Amylose crystals in starch link short starch chains to form a three-dimensional network in rice noodles. These amylose-based structures exist in the rice noodle in a composite form (V-type) or in retrograde form (B-type). These crystals are highly resistant to mild acid hydrolysis and melt at high temperatures (Mestres, Colonna, & Buleon, 1988). Varieties with high direct starch content are more suitable for making rice noodles owing to their high hardness, tensile strength, and consistency (Lii, Tsai, & Tseng, 1996).

After shading, the protein content of GuichaoII and Shuhui498 increased significantly by 21.25% and; 5.57%, respectively (Table 2). The protein content of rise has a significant influence on the properties of rice starch. The protein content exhibited a negative correlation with setback in RVA (Chavez-Murillo, Wang, Quintero-Gutierrez, & Bello-Perez, 2011). In the process of gelatinization, starch gradually escapes frome the protein network with increasing temperature, adsorbs in the pores of the protein network, whereupon finally forms a gel, and the complete starch particles are destroyed (Li, Yeh, & Fan, 2007). Protein inhibits the swelling and lowers the water-holding capacity of rice starch gel, which directly affects the convenience of rice flour processing (Zhang, Chen, Chen, & Chen, 2019). In contrast to wheat protein, GuichaoII rice protein does not readily form a network structure upon absorbing water, which is not conducive to improving the elasticity and strength of rice glue, making it difficult to meet the requirements of noodle processing (Kim, Kee, Lee, & Yoo, 2014). Accordingly, the increasedprotein content reduces the quality of the rice noodles made from rice subjected to shading. The protein warps around the starch, forming a net structure inhibits starch swelling (Noisuwan, Bronlund, Wilkinson, & Hemar, 2006). After shading, the protein content in grains increased, while the solubility and swelling factor of rice flour gradually decreased. The more this net-like protein structure was filled with starch, the more compact it was, reducing the amount fo amylose exuded and thereby inhibiting, the gelatinization and swelling of rice flour.Protein also inhibits the aging process of rice starch, which can improve the shelf life of rice noodles (Kim et al., 2014).

3.3. Starch fine structure

3.3.1. Starch granules and their distribution

Rice starch mainly exists as compound granules in endosperm cells. Electron microscopy revealed differences in the morphologies of starch granules in GuichaoII and Shuhui498 developed under different light conditions(Fig. 1). Under normal light levels, the starch granules in GuichaoII are smooth and oval, a feature often found in poorly enriched, highly-chalky endosperm cells (Deng et al., 2018). At reduced light levels, angular, polyhedral granules were observed in GuichaoII, similar to those found in Shuhui498 under normal light levels. In addition, honeycomb-shaped erosion pores appeared on the surface of starch

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Treatment	PKV (RUV)	HPV (RUV)	BDV (RUV)	CPV (RUV)	SBV (RUV)	HPV/PKV	PaT (°C)	To (°C)	Tp (°C)	Tc (°C)	ΔH (J/g)
G-CK	$\textbf{243.22} \pm$	$214.86~\pm$	$28.36~\pm$	316.39 \pm	$\textbf{73.17} \pm$	$0.88~\pm$	$\textbf{86.72} \pm$	$60.19~\pm$	65.6 \pm	$\textbf{73.39} \pm$	10.99 \pm
	2.36c	1.53a	3.86c	6.61a	3.25a	0.01a	1.08a	0.12c	0.01b	0.22b	0.09ab
G-	$\textbf{225.39} \pm$	198.08 \pm	$27.31~\pm$	$301.03~\pm$	75.64 \pm	$0.85 \pm$	87.57 \pm	59.74 \pm	65.73 \pm	$\textbf{72.93} \pm$	10.29 \pm
Shading	6.72d	4.17b	1.19d	9.22b	3.24a	0.02a	1.12a	0.38c	0.09b	0.20b	0.96b
Mean	$241.14~\pm$	$210.00~\pm$	$\textbf{27.84} \pm$	$319.07~\pm$	$\textbf{77.93} \pm$	$\textbf{0.87}~\pm$	86.74 \pm	59.96 \pm	65.67 \pm	73.15 \pm	10.64 \pm
	1.59B	3.49A	2.48B	2.93A	3.42A	0.01A	0.27A	0.20B	0.05B	0.17B	0.46A
S-CK	$281.64~\pm$	148.50 \pm	133.14 \pm	$228.14~\pm$	$-53.50~\pm$	0.53 \pm	74.43 \pm	62.80 \pm	$68.47~\pm$	74.53 \pm	$12.12~\pm$
	1.84a	5.02c	7.15a	11.09c	4.12b	0.02b	0.81b	0.03b	0.05a	0.06a	0.01a
S-Shading	$\textbf{275.14} \pm$	136.55 \pm	125.92 \pm	$221.08~\pm$	$-42.42 \pm$	$0.50 \pm$	74.45 \pm	$63.59~\pm$	$68.68~\pm$	74.33 \pm	11.28 \pm
	2.98b	3.76d	3.21b	4.21c	3.12b	0.05b	0.01b	0.04a	0.10a	0.01a	0.10ab
Mean	$273.85~\pm$	142.53 \pm	$131.32~\pm$	224.61 \pm	-49.24 \pm	$0.52 \pm$	76.29 \pm	$63.19~\pm$	$68.57~\pm$	74.42 \pm	11.70 \pm
	4.14A	4.10B	3.54A	4.21B	4.80B	0.01B	2.02B	0.18A	0.07A	0.05A	0.19A

PKV: peak viscosity; HPV: hold viscosity; BDV: breakdown viscosity; CPV: final viscosity; SBV: setback; PaT: pasting temperature. T_{o} , T_{p} , and T_{c} represent the onset, peak, and conclusion temperatures, respectively; ΔH: the gelatinization enthalpies,; G-CK: GuichaoII under normal light; G-Shading: GuichaoII under shading; S-CK: Shuhui498 under normal light; S-Shading: Shuhui498 under shading. Values with different letters within a column are significantly different (p < 0.05).

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Chemical co	mposition, textur	e profile analysis (T.	PA), gel consistency	y and swelling fac	tor of Guichaoll	and Shuhui49	š.					
Treatment	Amylose content (%)	Amylopectin content (%)	Amylose/ Amylopectin	Protein content (%)	Swelling factor (g/g)	Hardness	Springiness	chewiness	adhesiveness	cohesiveness	restorative	Gel consistency (mm)
G-CK	$26.32\pm3.99a$	$56.57\pm8.10\mathrm{b}$	$0.48\pm0.07a$	$5.74\pm0.20d$	$9.77\pm0.44\mathrm{b}$	$\begin{array}{c} 729.43 \pm \\ 112.43a \end{array}$	0.88 ± 0.03a	$\begin{array}{c} 400.72 \pm \\ 101.52a \end{array}$	454.48 ± 99.53a	$0.71 \pm 0.09 \mathrm{ab}$	$0.66\pm 0.03a$	9.47 ± 0.80 a
G- Shading	$20.78\pm0.41\mathrm{b}$	$55.49\pm2.73\mathrm{b}$	$0.38\pm0.01\mathrm{b}$	$6.96\pm0.03\mathrm{c}$	$5.49\pm0.08\mathrm{c}$	631.07 ± 65 20a	0.89 ± 0.02	501.83 ± 88 55a	567.67 土 103 30a	$\textbf{0.78}\pm\textbf{0.02a}$	$0.68 \pm$	$8.47\pm0.32ab$
Mean	$23.55 \pm \mathbf{1.61A}$	$56.03\pm2.22B$	$0.43\pm0.04\mathrm{A}$	$6.32 \pm \mathbf{0.28B}$	$7.63\pm0.97B$	680.25 ± 40.12	0.024 $0.88 \pm$ $0.01A$	451.27 ± 41.49 A	511.08 ± 44.87B	$0.75\pm0.03A$	$0.67 \pm 0.01A$	$8.97\pm\mathbf{0.32A}$
S-CK	$15.96\pm1.12c$	$71.43 \pm 7.16a$	$0.23\pm0.02\mathrm{c}$	$8.79\pm\mathbf{0.76b}$	$13.15 \pm 3.84 \mathrm{ab}$	$189.67 \pm 20.02 \mathrm{b}$	$0.70 \pm 0.08b$	$\begin{array}{c} 80.55 \pm \\ \mathbf{21.02b} \end{array}$	$113.40 \pm 21.30\mathrm{b}$	$0.59\pm0.05c$	$0.49 \pm 0.02b$	$7.27\pm1.07\mathrm{b}$
S-Shading	$14.20\pm0.70\mathrm{d}$	$\textbf{75.44}\pm\textbf{8.06a}$	$0.19\pm0.01d$	$9.28\pm1.22a$	$14.53 \pm 1.41a$	$178.13 \pm 40.46 \mathrm{b}$	$\begin{array}{c} 0.66 \pm \\ \mathbf{0.06b} \end{array}$	$\textbf{72.44} \pm \textbf{17.08b}$	$110.86 \pm 26.69 \mathrm{b}$	$\begin{array}{c} 0.62 \pm \\ \mathbf{0.01bc} \end{array}$	$\begin{array}{c} \textbf{0.50} \pm \\ \textbf{0.05b} \end{array}$	$7.23\pm0.31\mathrm{b}$
Mean	$15.08\pm\mathbf{0.52B}$	$73.43\pm2.92\mathrm{A}$	$0.21\pm0.01B$	$9.04\pm0.39\mathrm{A}$	$13.84\pm1.10\mathrm{A}$	$\begin{array}{c} 183.90 \pm \\ 11.82 \end{array}$	$0.68 \pm 0.03B$	76.50 ± 7.22B	$112.13 \pm 8.83 \mathrm{A}$	$0.61 \pm 0.01B$	$0.50 \pm 0.01B$	$\textbf{7.25}\pm\textbf{0.29B}$
Violuos with	diffound lottons	ithin a column and	cionificantly diffound	mt (n < 0.0E)								

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granules in GuichaoII grown under normal light and Shuhui498 under shading.

Studies have suggested that the size of starch granules correlates strongly with the quality of the rice (Jia, Li, Dong, & Zhang, 2011). Under low light stress, the number-weighed mean diameter decreased and the volume-weighted mean diameter of both GuichaoII and Shuhui498 increased, although the latter change was only significant in Shuhui498 (Table 3). Meanwhile, the surface-area weighted mean diameter of GuichaoII was significantly lower under low light conditions compared to the control, while no significant change in that of Shuhui498 was observed. Analysis of the size distribution show that the ratios of smaller starch granules ($<5 \,\mu$ m) and large starch granules (>15µm) significantly increased under low light, while that that of mediumsized starch granules (5–15 µm) decreased (Table 3). Photodeprivation is known to reduce the number of endosperm cells and starch granules (Bechtel, Zayas, Dempster, & Wilson, 1993). The increased ratio of large granules may arise from substrate transfer to existing granules (Jia et al., 2011), while the change in the surface area-weighted mean diameter is likely related to the smoothness of the starch granules, since pores increase the surface area.

3.3.2. Branch chain length distribution

Both rice varieties grown under shaded conditions showed no significant change in amylopectin content during the grain filling stage, while the amylose content decreased significantly (Table 2). This is consistent with the trends observed in our previous study (Deng et al., 2021), and in corn and wheat under low light (Shi et al., 2018). A reduced amylose content is primarily responsible for the lower total starch content observed under shading conditions.

Shading also significantly changed the chain length distribution in rice starch (Table 4). Under shading conditions, the percentage of shortbranched chains (DP: 6-12) in both varieties decreased significantly, while no significant difference in the percentage of medium length chains (DP: 13–36) or long chains (DP > 36) was observed, which is consistent with previous results (Deng et al., 2021).

A significant negative correlation exists between the number of short-branched chains and viscosity (An, Li, Li, Obadi, & Xu, 2021). Although the percentage of short-branched chains was the same in both varieties, the total starch content of the short-branched chains was significantly lower in GuichaoII (0.16 g/g) than in Shuhui498 (0.20–0.21 g/g, Table 4). More short-branched starch lectins contribute to a more disordered double helix wrap in the granules, which facilitates expansion and hydration. The swelling capacity and water absorption increase with increasing content of short amylopectin chains and decreasing length of the internal amylopectin B-chain, which also lowers the amylose leaching value of starch during heating. Amylose may be entangled with the internal amylopectin chains in the lamella of the granules, thus inhibiting starch leaching (Li & Zhu, 2017). The proportion of short-branched chains decreased after shading, resulting in a deterioration in the quality of the rice flour and, consequently, that of the rice noodles.

3.3.3. Degree of branching and crystal structure

Under low light stress, the degree of branching in GuichaoII increased significantly while that in Shuhui498 decreased significantly (Table 4), resulting in a higher degree of branching. The crystallinity of the rice starch is dependent on the amylose content (Chung et al., 2011). The crystal type and crystallinity of the starch in the two rice varieties produced under different light conditions was analyzed by XRD. The diffraction patterns of the two varieties are similar (Fig. S1a), with strong peaks at 15° , 17° , 18° , and 23° ; the peaks at 17° and 18° are connected double peaks. These peaks are commonly found in the XRD spectra of the starch of grain crops (Cheetham & Tao, 1998). The crystallinity of the starch in both varieties ranges from 30.52 to 39.85 %, with GuichaoII having a lower crystallinity than Shuhui498. After shading, the crystallinity of the starch in GuichaoII decreased by 4.75%



Fig. 1. Scanning electron microscopy images of rice starch granules from (a and b) GuichaoII and (c and d) Shuhui498. (a and c): under normal light levels, (b and d): under shading. The red arrows indicate elliptic or near-elliptic starch granules, and the yellow arrows indicate eroded pores on the surface of the starch granules. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Table 3
Effect of low light stress on the diameter of rice starch granules.

Treatment	Number- weighted mean	Volume- weighted mean	Surface- area weighted	Distribu weighte (%)	Distribution of volume- weighted mean diameter (%)			
	diameter (µm)	diameter (µm)	mean diameter (μm)	<5 μm	5–15 μm	>15 µm		
G-CK	$4.34~\pm$	7.11 \pm	$6.00~\pm$	24.66	73.96	1.38		
	0.01a	0.02a	0.01a	\pm	±	±		
				0.12c	0.14a	0.01b		
G-	$4.01~\pm$	7.14 \pm	5.82 \pm	27.71	69.89	2.40		
Shading	0.00b	0.02a	0.01b	\pm	±	±		
				0.11b	0.20b	0.00a		
Mean	4.18 \pm	7.81 \pm	5.92 \pm	26.19	71.92	1.89		
	0.07A	0.25A	0.04A	\pm	±	±		
				0.68A	0.91A	0.23A		
S-CK	4.41 \pm	$6.77 \pm$	5.81 \pm	26.83	72.54	0.62		
	0.01a	0.01b	0.01b	\pm	±	±		
				0.11b	0.11a	0.00c		
S-Shading	4.15 \pm	7.03 \pm	5.82 \pm	29.32	67.82	2.86		
	0.01b	0.01a	0.01b	\pm	±	±		
				0.21a	0.13b	0.01a		
Mean	4.28 \pm	7.79 \pm	5.82 \pm	28.08	70.18	1.74		
	0.06A	0.39A	0.01A	±	±	±		
				0.56A	1.06A	0.50A		

G-CK: GuichaoII under normal light; G-Shading: GuichaoII under shading; S-CK: Shuhui498 under normal light; S-Shading: Shuhui498 under shading. Values with different letters within a column are significantly different (p < 0.05).

while that of the starch in Shuhui498 increased by 10.09%, possibly due to the increased degree of branching in GuichaoII starch after shading, which changes the stereohelix of the three-dimensional starch structure and reduces the degree of crystallization. This result was further

confirmed by Fourier transform infrared (FTIR) spectroscopy.

FTIR spectroscopy is widely used to study the crystalline and amorphous regions near the surface of starch granules (Zhang, Zhu, Shao, Gu, & Liu, 2013). The ratio of intensity of the spectral peaks at 1047 and 1022 cm^{-1} is used to measure the degree of order in starch, with higher 1047/1022 cm⁻¹ and lower 1022/995 cm⁻¹ ratios indicating a higher degree of crystallinity (Almeida, Batista, Di-Medeiros, Moraes, & Fernandes, 2019). The spectra of the starch samples in this study display no other obvious peaks (Fig. S1). The 1047/1022 cm⁻¹ and 1022/995 cm⁻¹ ratios in the spectra of GuichaoII and Shuhui498 are shown in Table 4. After shading, GuichaoII exhibits a lower crystallinity, lower 1047/1022 cm⁻¹ ratio, and higher 1022/995 cm⁻¹ ratio than the control. The lower 1047/1022 cm⁻¹ and 1022/995 cm⁻¹ ratios in the spectra of Shuhui498 suggest a reduced degree of order under shading conditions, which is consistent with the low relative crystallinity observed by XRD (Colussi et al., 2014), all indicating a lower degree of order. A significant correlation was also observed among 1042/1022 cm⁻¹, 1022/995 cm⁻¹, and the degree of branching in the starch (Table 4) in both GuichaoII and Shuhui498, with the two rice varieties displaying opposite trends under low light conditions. Previous studies found a significant positive correlation between the crystallization rate of starch and the 1042/1022 cm⁻¹ ratio, which may arise from coinciding changes in the degree of branchingand the crystallinity of the starch (Cai et al., 2015; Zou et al., 2020).

3.3.4. Thermal properties of rice flour

Gelatinization is an endothermic transformation of starch from a semi-crystalline structure to an amorphous conformation owing to the dissociation of the double helixof amylopectin.

The values of the onset(T_{o}), peak (T_{p}), conclusion (T_{c}) temperatures, and gelatinization enthalpies (Δ H) after shading are given in Table 1. The gelatinization temperature of rice flour is higher than T_{o} , T_{p} and T_{c}

Treatment	Crystallinity degree (%)	Branching degree (%)	Ratio of 1045/ 1022 cm^{-1}	Ratio of 1022/ 995 cm^{-1}	(DP 6–12)	(DP 13–24)	(DP 25-36)	(DP > 36)	DP 6–12 of amylopectin content
G-CK	$35.11 \pm 1.34 \text{ab}$	$3.29\pm0.33 \text{d}$	$\textbf{0.75} \pm \textbf{0.01a}$	$1.40\pm0.01a$	$\begin{array}{c} \textbf{28.66} \pm \\ \textbf{0.58a} \end{array}$	$47.39 \pm 0.27a$	$11.73 \pm 0.17b$	$\begin{array}{c} 12.22 \pm \\ 0.68a \end{array}$	$0.16\pm0.01b$
G- Shading	$\textbf{32.54} \pm \textbf{1.17b}$	$6.66\pm0.36b$	$\textbf{0.73} \pm \textbf{0.01a}$	$1.42\pm0.00\text{a}$	$\begin{array}{c} \textbf{27.98} \pm \\ \textbf{0.23b} \end{array}$	47.27 ± 0.25a	12.04 ± 0.05ab	$\begin{array}{c} 12.71 \pm \\ 0.42 \mathrm{a} \end{array}$	$\textbf{0.16} \pm \textbf{0.01b}$
Mean	$33.82 \pm \mathbf{0.98B}$	$\textbf{4.97} \pm \textbf{0.79A}$	$\textbf{0.74} \pm \textbf{0.03A}$	$1.41\pm0.03\text{A}$	$\begin{array}{c} \textbf{28.32} \pm \\ \textbf{0.32A} \end{array}$	47.33 ± 0.16A	11.89 ± 0.11A	$\begin{array}{c} 12.47 \pm \\ 0.37 \mathrm{A} \end{array}$	$0.16\pm0.01b$
S-CK	$38.21 \pm \mathbf{0.95a}$	$5.06\pm0.70c$	$\textbf{0.76} \pm \textbf{0.01a}$	$1.38\pm0.01\text{a}$	$\begin{array}{c} \textbf{28.53} \pm \\ \textbf{0.58a} \end{array}$	47.60 ± 0.25a	$11.68 \pm 0.01b$	$13.06 \pm 0.30a$	$0.20\pm0.00a$
S-Shading	$\textbf{37.44} \pm \textbf{0.93a}$	$\textbf{8.85} \pm \textbf{0.45a}$	$\textbf{0.75} \pm \textbf{0.00a}$	$1.37\pm0.00\text{a}$	27.76 ± 0.49b	$\begin{array}{c} 47.02 \pm \\ 0.40a \end{array}$	12.16 ± 0.18a	12.55 ± 0.72a	$0.21\pm0.01a$
Mean	$37.82 \pm \mathbf{0.62A}$	$6.95\pm0.92\text{A}$	$\textbf{0.74} \pm \textbf{0.03A}$	$1.38\pm0.03\text{A}$	$\begin{array}{c} \textbf{28.15} \pm \\ \textbf{0.28A} \end{array}$	47.31 ± 0.25A	11.92 ± 0.13A	$\begin{array}{c} 12.62 \pm \\ 0.40 \mathrm{A} \end{array}$	$0.21\pm0.01a$

G-CK: GuichaoII control (under normal light); G-Shading: GuichaoII under shading; S-CK: Shuhui498 control; S-Shading: Shuhui498 under shading. Values with different letters within a column are significantly different (p < 0.05).

of the phase transition in the crystallization zone (Table 1). Since the gelatinization temperature is the temperature at which the viscosity of the starch suspension begins to increase owing water absorption and expansion of the starch particles, the above observation indicates that the breakage of the crystallization zone occurs earlier than the particle expansion in the gelatinization process. In addition, the double helix structure of the amylopectin side chain is the main component of the starch crystallization zone (Fan et al., 2013). Starch gradually exudes from the protein network with increasing temperature, damaging the double helix structure of amylopectin. With further increases temperature, starch is gradually adsorbed in the pores of the protein network, and finally the gelatinization process proceeds to completion, is completed, leaving complete starch granules.

The DSC parameters of the two varieties changed slightly, but no significant difference was observed. The Δ H of GuichaoII was lower than that of Shuhui498 under both normal light and shading (6.37% and 6.93% lower, respectively). After shading, the A-chain (DP 6–12) ratio of

amylose and the amylopectin content decreased, thereby reducing the value of Δ H (Deng et al., 2021).

3.4. Relationship between the pasting, the structural, and physicochemical properties

After shading, the pasting properties of starch were reduced (Table 1 and Table 2), owing to an increase in the number of small particles (Liu et al., 2017), and a reduction in the amylose content (Colussi et al., 2014). Many factors affect the gel properties of rice flour, such as the content of amylose, amylopectin, proteins, and lipids (Yoenyongbuddhagal, & Noomhorm, 2013). Pearson correlation analysis was carried out to determine the correlations between the six pasting properties measured using RVA and the structural and physicochemical properties of the rice starch (Fig. 2). The amylose content, protein content, swelling factor and degree of crystallinity all showed a strong positive correlation with the six RVA profiles, while the distribution of the volume-weighted



Fig. 2. Relationship between the pasting, structural, and physicochemical properties. PKV: peak viscosity; HPV: hold viscosity; BDV: breakdown viscosity; CPV: final viscosity; SBV: setback; PaT: pasting temperature.

mean diameter ($<5 \mu m$) had a significant negative influence on HPV. Amylopectin and the content of short amylopectin chains were significantly correlated with five RVA profiles (all except PaT), which was consistent with previous studies (Wang et al., 2013). The SBV and PaT decreased and increased, respectively, with increasing amylose content. Some researchers found that increasing the number of short-branched chains in amylopectin reduced the peak viscosity (Srichuwong & Jane, 2007), while another study (Wang et al., 2013) measured a lower peak viscosity in rice with fewer short-branched chains. In the present study, the pasting properties of rice decreased with the ratio of short-branched under low light levels (Table 3). A significant correlation between the amylose content and the RVA properties of the rice has been observed (Lii et al., 1996). The protein content was negatively correlated with CPV, SBV and HPV, and positively correlated with PKV and BDV. In the gelatinization process, the protein absorbs water to reduce the amount of free water combined with starch, while forming a network through disulfide bonds, which reduces the concentration of the gelatinized heterogeneous system and the viscosity of RVA. The reduction in the amylose content and the proportion of short chains in amylopectin, along with the increased in the protein content after shading, may be the main causes of the deterioration in the pasting properties of the rice.

 T_o , T_p , and T_c were showed a significant positive with the amylopectin content, short-chain amylopectin content, protein content, and swelling factor, and were negatively correlated with the amylose content. The crystallinity of rice flour was positively correlated with the four thermodynamic properties. GuichaoII has lower crystallinity, lower gelatinization enthalpies (Δ H) and less energy than Shuhui498, which is more conducive to the processing of rice noodles.

The amylose content, short-chain amylopectin content, and protein content strongly correlated with the six TPA profiles. The gel consistency was significantly affected by 8 physiological indexes including amylose content, short-chain amylopectin content, and amylopectin content (Fig. 2). The strength and heat resistance of rice noodle gels are maintained by a network of amylose molecules (Marti & Pagani, 2013). The gel strength of rice flour increases with amylose content, while the chain length distribution of amylopectin also has an impact on the gel properties of rice (Yu, Ma, & Sun, 2009). The structure of amylopectin differs between varieties of rice, and the degree of amylopectin polymerization affects the aging properties of amylopectin and further affect the gel properties (Vandeputte & Vermeylen, 2003; Lu, Sasaki, Li, Yoshihashi, Li, & Kohyama, 2009). The increase in hardness during gel placement was mainly caused by the recrystallization of amylopectin (Biliaderis, 1998). In the rice flour system, protein can intertwine with amylose to form a network structure (Noisuwan et al., 2006). The lower the protein content, the more amylose can regroup and arrange with each other. The degree of intertwining between molecules is therefore enhanced, compacting the internal stucture of rice flour, thereby increasing its hardness, chewability, resilience and cohesion. Emulsified of protein acts as an emulsifier in the starch, which enhances the stickiness of the system, which correlates with its protein content. After shading, the protein content and short-chain amylopectin content increased, while the amylose content, hardness, ΔH , and glue consistency decreased, thereby affecting the reprocessing of rice into rice noodle products.

4. Conclusions

GuichaoII has a lower protein content, short chain amylopectin content, swelling factor, degree of crystallinity, and volume weighted mean diameter (<5 μ m) distribution than Shuhui498. It also has a higher amylose content, hardness, springiness, chewiness, and gel consistency than Shuhui498. These characteristics render GuichaoII more conducive to the production of rice noodles. Reduced light intensity during the grain filling stage affects the starch structure and the quality of the harvested rice. The size of the internal starch granule, crystallinity, and Δ H, decreased under shading, while the disorder in the structure increased. After shading, the amylose content and the ratio of

short chain amylopectin decreased, while the protein content and degree of branching increased, thus inhibiting the water absorption capacity of the rice starch gel. Such properties are not conducive to improving the elasticity and strength of rice glue, and thus directly affect the processing of the rice noodles. This study elucidates the effect of low light stress on the pasting properties of rice and rice noodles, and provides new insight into the synthesis of starch granules and their influence on the properties rice noodles under stress.

CRediT authorship contribution statement

Hong Chen: Writing – original draft, Conceptualization, Methodology, Software. Tao Wang: Data curation, Writing – original draft. Fei Deng: Visualization, Investigation. Fan Yang: Formal analysis, Investigation. Xiaoyuan Zhong: Investigation, Formal analysis. Qiuping Li: Data curation, Formal analysis. Wanjun Ren: Conceptualization, Methodology.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

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