**RESEARCH ARTICLE** 



# Physicochemical properties and freeze-thaw stability of rice flour blends among rice cultivars with different amylose contents

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**Abstract** The effectiveness of the rice flour blends (RFB) for improving the processing suitability of Dodamssal rice flour (DD), a functional rice variety with a relatively high amylose and resistance starch content, was investigated. Physicochemical properties and freeze-thaw stability of RFB composed of DD and four rice flour (RF) samples with different amylose contents were measured at different DD ratios. DD, which has low swelling power and low pasting viscosity properties, has improved some quality in terms of physicochemical properties by blending with other RF. Especially, non-additive behavior was observed in the blend with Geonyang No.2 RF (GY), a medium waxy variety, due to water competition caused by the difference in pasting temperature. The syneresis of DD was reduced by blending with 75% Hanareum No. 4 RF, with a gradual reduction effect observed following a repeated freeze-thaw

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cycle. GY significantly improved the low freeze-thaw stability of DD with only a 25% blend.

**Keywords** Rice flour · Dodamssal · Blending · Physicochemical property · Freeze–thaw stability

# Introduction

Rice (Oryza sativa L.) is a staple food for more than 60% of the global population and is a major source of calories (60–70%), particularly in the Asian diet. It is a hypoallergenic material that is used and studied in food processing as a gluten-free substitute for healthy food and convenience food worldwide (e.g. bread, pasta, noodles and pudding, and so forth), as well as being consumed as cooked rice at home (Lin et al., 2011; Sivaramakrishnan et al., 2004). Recently, functional rice varieties with various specific nutritional purposes have been developed to match changes in consumer demand due to increasing interest in health and food preferences (Jobling, 2004). The rice cv. Dodamssal is a functional rice developed in Korea that has a high amylose (>40%) and high resistance starch (> 12%) content (Sim et al., 2015). In addition, Dodamssal can be used as a functional ingredient as it contains more than twice the dietary fiber content (approx. 4.2%) compared to normal rice (Sim et al., 2015). Dodamssal has been reported to have anti-obesity effects and can help prevent cardiovascular disease as a source of resistant starch (Kim et al., 2019). Resistance starch is a starch fraction that is fermented in the large intestine without being hydrolyzed to D-glucose in the small intestine within 120 min of ingestion, and is a prebiotic that suppresses colon cancer (Fuentes-Zaragoza et al., 2011). For its application, various studies have been conducted on the functionality, and dyslipidemia and obesity improvement effects, of rice varieties containing high levels of resistance starch and dietary fiber (Kim et al., 2019; Rivera-Piza et al., 2020). However, high amylose rice has only a few potential uses due to the limitations in its processing, such as its low swelling and freeze-thaw stability and high gelatinization temperature (Suwannaporn et al., 2007; Varavinit et al., 2002). Even in the case of rice cv. Dodamssal, which has potential value for health functions, its use has not been fully determined due to a lack of research on its physicochemical properties and processing suitability (Park et al., 2019).

One possible way to obtain novel processing properties from starch-containing rice flour (RF) is to blend various starches with different properties. The starch blends can be used instead of natural starch to impart desired properties, such as improved paste and textural properties and freezethaw stability (Puncha-arnon et al., 2008). In addition, starch blends are expected to have non-additive effects as the different types of starch and the properties of the samples result in certain interactions during blending (Waterschoot et al., 2014). Since starch composition and cooking quality are mainly related to starch, which contains more than 90% of rice, the effect on the starch blend is also expected to be revealed in the rice flour blends (RFB) (Frei et al., 2003). In previous studies, RF has been blended with other cereal flours for preceding (Park et al., 2017; Sasaki et al., 2014). However, there has been little research on the blending of RF varieties with different amylose contents (Jeong et al., 2021). Moreover, few studies have attempted to improve the processing suitability of rice with a high resistant starch content, or assess the physicochemical properties and freeze-thaw stability of blends between RF with different amylose contents.

Therefore, the objective of this study was to investigate the effects of blending RF, with different amylose content, and Dodamssal rice flour (DD), with a high resistant starch and amylose content, on their physicochemical properties and freeze-thaw stability. This study is expected to improve the processing suitability of DD, a functional RF, through the blending technology, thereby contributing to the diversification and utilizability of RF based processed foods.

# Materials and methods

# Materials

National Institute of Crop Science (Rural Development Administration, Suwon, Republic of Korea). The RF in this study was named using their rice varieties abbreviations DD, SM, HA, GY, and MR, respectively. The amylose contents of RF were in the order of DD ( $\approx 43\%$ ) > SM ( $\approx 27\%$ ) > HA ( $\approx 18\%$ ) > GY ( $\approx 12\%$ ) > MR ( $\approx 5\%$ ), and their protein contents did not differ significantly (approx. 6.7–7.1%) (Rural Development Administration, 2021).

# **Preparation of rice flour blends**

Dry milled RF with 70–80 µm in a particle size were passed through a 100-mesh standard sieve (Chunggye Inc., Seoul, Republic of Korea) in advance. The rice flour blends (RFB) were prepared by mixing DD with other RF at blending ratios of 100:0, 75:25, 50:50, 25:75, and 0:100 (dry weight basis, db), respectively, and then manually stirred using a spatula.

## Swelling power and water solubility

The swelling power (SP, g/g) and water solubility (WS, %) of RFB were measured by the method of Schoch (1964), with a slight modification. RFB (0.2 g, db,  $W_0$ ) was dispersed in 10 mL of deionized water, followed by heating with stirring for 30 min in a water bath at 80 °C. The resultant hot paste was cooled to room temperature and centrifuged (15,000×g, 30 min). The supernatant was decanted carefully in an aluminum pan and dried overnight in a dry oven at 95 °C, followed by weighting the dried solid ( $W_s$ ). The dried precipitate was also weighed ( $W_r$ ). The SP and WS were calculated using following equations.

$$SP(g/g) = \frac{W_s}{W_0 \times \left(1 - \frac{WS}{100}\right)}$$
(1)

WS (%) = 
$$\left(\frac{W_r}{W_0}\right) \times 100$$
 (2)

# **Thermal properties**

The thermal properties of RFB were measured by a differential scanning calorimeter (DSC, Perkin-Elmer DSC 4000, Norwalk, CT, USA). DSC was calibrated by indium metal prior to measurement. The RFB samples (2.5 mg) with an excess of water (7.5 mg) were weighed in a 40  $\mu$ L aluminum pan and sealed. For gelatinization transition of RFB, the pan with the sample was heated from 30 to 120 °C at a rate of 10 °C/min using an empty sealed pan as reference. The onset temperature (T<sub>o</sub>), peak temperature (T<sub>p</sub>), conclusion temperature (T<sub>c</sub>), enthalpy of gelatinization ( $\Delta$ H), and temperature range of gelatinization ( $\Delta$ T, T<sub>c</sub> - T<sub>p</sub>) were calculated via the measured peaks.

# **Pasting properties**

The pasting properties of RFB were measured using a rapid visco analyzer (RVA-3, Newport Scientific Pty Ltd., Sydney, Australia) by the AACCI method (AACC, 2010). The RFB solution (12%, w/w) was mixed at 960 rpm for 10 s and held at 50 °C for 50 s, and then heated to 95 °C at the rate of 10 °C/min. After holding at 95 °C for 3.5 min, the paste samples were cooled to 50 °C at rate of 10 °C/min and also kept at this temperature for 2 min. The pasting temperature (PT), peak viscosity (PV), trough viscosity (TV) and final viscosity (FV) were measured by RVA curves and the parameter values of breakdown viscosity (BD, PV-TV) and setback viscosity (SB, FV-TV) were derived.

# **Textural properties**

To prepare RFB gels, RFB (10%, w/w) dissolved in deionized water was gelatinized in a water bath at 95 °C for 10 min, and then transferred into a petri-dish  $(30 \times 10 \text{ mm})$ . It was then sealed with parafilm to prevent moisture evaporation and cooled at room temperature. The sealed gel samples were kept at 4 °C for overnight and left at room temperature for 1 h to be used for texture experiments when the equilibrium temperature was reached. Gel texture analysis were measured using a TA-HDi texture analyzer (Stable Micro Systems Ltd., Surrey, UK) under a two-cycle compression test (TPA). Each gel was pressed to a 75% normal strain (trigger force = 0.05 N) with a 50 mm cylinder probe at a test speed of 2.0 mm/s, to obtain hardness (HD), adhesiveness (AD), and cohesiveness (COH).

# Freeze-thaw stability measurement

#### Freeze-thaw cycles

The freeze–thaw stability was determined by the modified method of Charoenrein et al. (2008), with slighly modification. RFB suspensions (10%, w/w, dry weight basis) dissolved in distilled water was stirred continuously at 300 rpm for 1 h, followed by heating in a water bath at 95 °C for 30 min with moderate mechanical agitation. The measured weight (wt<sub>s</sub>) of hot paste was then loaded into a custom-made sample holder (Fig. S1), steamed for 10 min, and finally placed in an incubator at 25 °C for 22 h and then thawed in an incubator at 25 °C for 4 h. This freeze–thaw cycle was repeated for up to five cycles.

#### Determination of syneresis

A 60 mesh standard sieve with a Whatman No. 1 filter paper was attached to the bottom of the cylindrical plastic tube, and then placed in a centrifuge tubes (Fig. S1). The empty centrifuge tube was weighed without cylindrical plastic tube (wt<sub>0</sub>). Thawed RFB gels via a freeze–thaw cycle inside the tube set was centrifuged at  $100 \times g$  for 15 min. and the final weighing (wt<sub>1</sub>) of the cylindrical plastic tube with the sample was weighted. The free water separated from RFB gel was weighed and the syneresis (%) was calculated using following equation.

Syneresis (%) = 
$$\frac{wt_1 - wt_0}{wt_s} \times 100$$
 (3)

# Statistical analysis

All experiments were repeated at least in triplicate, except of textural properties (n = 5). Statistical analyses were carried out using SPSS (IBM SPSS Statistics for windows, Version 25.0, Armonk, NY, USA). One-way ANOVA was used for comparison of group means, followed by Tukey's HSD multiple range test for determination of the significance of individual comparisons (p < 0.05).

# **Results and discussion**

# Swelling power and water solubility of rice flour blends

After blending DD with different varieties of RF, the SP and WS were measured at 80 °C to investigate the effects of blending on each parameter. As shown in Fig. 1, the SP of DD was lower than that of other RF due to very high amylose content (> 40%) and a relatively high resistant starch (> 12%), whereas MR had the highest SP (Park et al., 2017). Amylopectin is generally the main cause of starch swelling, whereas amylose inhibits its swelling and maintains the integrity of swollen granules (Tester and Morrison, 1990).

The SP of RFB significantly increased when other RF was blended with DD at the ratio of 75:25 (p < 0.05). This phenomenon could be caused by the blending of other RFs with a higher SP than DD. At the other ratio (25:75 and 50:50), SP was decreased, with a non-additive effect that could occur in starch blending. The non-additive effect meant that the predicted average value that was mathematically calculated from the characteristic values of individual materials before blending and the value measured in the actual experiment did not match (Waterschoot et al., 2015b). Differences in the swelling properties of RF

50

40

30

20

10

0

50

**DD0HA100** 

Water soulubility (%)



Fig. 1 Swelling power and water solubility (at 80 °C) of DD and SM blends (A), DD and HA blends (B), DD and GY blends (C), and DD and MR blends (D) at various blending ratios. Each data is the mean of three replicates. Bars with the different letter (a-d for swelling

with different amylose contents resulted in water competition in the RFB (Lin et al., 2013). In particular, in the DD50MR50, the non-additive effect was remarkable, and the SP value was much lower than that of the individual DD. This was similar to the results of a previous study, in which the SP was significantly reduced when high amylose and waxy varieties were blended in a similar ratio (Frei et al., 2003). The physicochemical properties of starch blends are affected by the blending ratio and tend to follow the characteristics of the high blending ratio samples (Puncha-arnon et al., 2008). In this study, when DD was blended in excess (more than 50%), the SP values of most of the RFB were dominated by the DD sample, although trend was not linear due to the non-additive effect. This suggests that to improve the low swelling capacity of DD, the optimal effect will be attained when the blended amount of the other RF is in excess of the proportion of DD.

Except for MR, there were no significant differences in WS among the blends of RF, and therefore there were no significant effects of blending on WS. The WS of MR was significantly higher than that of DD, and it increased with

■ Swelling power □ Water soulbility 14 12 Swelling power (g/g) Water soulubility (%) 10 30 ь 8 bc 20 6 10 2 0 DD0MR100 DD100MR0 DD75MR25 DD50MR50 DD25MR75

DD50HA50

DD75HA25

■ Swelling power □ Water soulbility

DD75HA25

14

12

10

8

6

4

2

0

DD100HA0

power; a'-d' for water solubility) are significantly different (p < 0.05). The dotted line in graph means predicted value of swelling power in theory calculated from those of individual rice flours, assuming additive behaviors.

an additive effect that was different to the pattern observed for the SP values.

# Thermal properties of rice flour blends

The thermal properties associated with gelatinization are summarized in Table 1. Among the individual RF, peak temperature  $(T_p)$  was highest in DD (approx. 76.1 °C) and lowest in GY ( $\sim 66.4$  °C). This is consistent with a previous study that found that s high amylose starch with a longer average chain length exhibited a higher transition temperature (Hagenimana et al., 2005). The same study reported that the T<sub>p</sub> had a strong correlation with the long chain length of amylopectin, and the linear amylose chains were entangled with each other, limiting the hydration in the amorphous region of the granule, thereby impeding the swelling and gelatinization. DD is a cultivar with a higher amylose and long chain length of amylopectin content than the other RF varieties (Sim et al., 2015). The gelatinization enthalpy value ( $\Delta H$ ) decreased as the amylose content of the RF increased. That is, DD had the lowest  $\Delta H$  of  $\sim 4.5$  J/g, which means that the energy required for

 Table 1
 Thermal properties of rice flour blends

Samples <sup>1</sup>	$T_o (^{\circ}C)^2$	T <sub>p</sub> (°C)	T <sub>c</sub> (°C)	$\Delta H (J/g)$	ΔT (°C)
DD	$69.6 \pm 0.3^{a,3}$	$76.1 \pm 0.3^{a}$	$85.5\pm0.2^{a}$	$4.5\pm0.2^{\mathrm{b}}$	$15.9\pm0.5^{a,b}$
DD75SM25	$68.6\pm0.4^{\rm b}$	$76.8\pm0.1^{\rm a}$	$85.3\pm0.1^{\rm a}$	$6.4\pm0.5^{a,b}$	$17.1 \pm 0.3^{a}$
DD50SM50	$68.1 \pm 0.1^{b}$	$76.1 \pm 0.2^{a,b}$	$85.1\pm1.2^a$	$7.0 \pm 0.1^{a,b}$	$17.0 \pm 1.3^{a}$
DD25SM75	$67.8\pm0.0^{\rm b}$	$75.4\pm0.4^{\rm b}$	$82.7\pm0.3^{\rm b}$	$9.5\pm2.0^{a}$	$14.9\pm0.4^{\rm a}$
SM	$66.9 \pm 0.1^{\circ}$	$73.9\pm0.2^{\rm c}$	$81.7\pm0.2^{\rm b}$	$7.1 \pm 0.1^{ab}$	$14.7\pm0.0^{\rm a}$
DD	$69.6\pm0.3^a$	$76.1\pm0.3^{\rm a}$	$85.5\pm0.2^a$	$4.5\pm0.2^{\rm b}$	$15.9\pm0.5^{\rm b}$
DD75HA25	$64.0 \pm 0.0^{b}$	$76.8\pm0.3^{\rm a}$	$84.4\pm1.7^{\rm a}$	$4.6\pm0.3^{ab}$	$20.5\pm1.7^a$
DD50HA50	$62.2\pm0.8^{\rm b}$	$67.9 \pm 0.3^{b}$	$75.0\pm0.3^{\rm b}$	$3.5\pm0.0^{b}$	$13.3 \pm 1.1^{b,c}$
DD25HA75	$63.1 \pm 0.3^{b}$	$68.8\pm0.1^{\rm b}$	$75.1\pm0.0^{\rm b}$	$4.1 \pm 0.2^{b}$	$11.9 \pm 0.2^{c}$
HA	$62.4\pm0.0^{\rm b}$	$68.3\pm0.3^{\rm b}$	$74.5\pm0.3^{b}$	$6.1 \pm 0.6^{a}$	$12.1 \pm 0.2^{c}$
DD	$69.6\pm0.3^a$	$76.1\pm0.3^{\rm a}$	$85.5\pm0.2^a$	$4.5\pm0.2^{\rm b}$	$15.9\pm0.5^{\rm b}$
DD75GY25	$62.6 \pm 1.0^{b}$	$66.7\pm0.8^{a}$	$84.3\pm0.3^a$	$2.6\pm0.3^{\rm c}$	$20.5\pm0.8^a$
DD50GY50	$60.7\pm0.5^{\rm c}$	$66.8\pm0.4^{\rm c}$	$73.5\pm0.5^{\text{b}}$	$2.9\pm0.2^{\rm c}$	$12.8 \pm 0.3^{b}$
DD25GY75	$61.7\pm0.4^{\rm c}$	$68.3\pm0.0^{\rm b}$	$75.6\pm0.3^{\rm b}$	$3.9\pm0.5^{\mathrm{b}}$	$13.8 \pm 0.0^{b}$
GY	$59.0\pm0.9^{\rm d}$	$66.4\pm0.5^{\rm c}$	$74.4 \pm 0.6^{b}$	$8.1\pm0.1^{\rm a}$	$15.4 \pm 0.3^{b}$
DD	$69.6\pm0.3^a$	$76.1\pm0.3^{a}$	$85.5\pm0.2^a$	$4.5\pm0.2^{b}$	$15.9\pm0.5^{\rm c}$
DD75MR25	$63.4\pm0.3^{\rm b}$	$76.2\pm0.1^{\rm a}$	$84.0\pm0.6^{\rm a}$	$6.8\pm0.1^{\rm a,b}$	$23.6\pm0.2^{a,b}$
DD50MR50	$62.7 \pm 0.9^{\rm b}$	$73.6\pm0.2^{a,b}$	$87.7\pm0.3^{\rm a}$	$9.2\pm0.5^{\rm a}$	$24.9\pm0.5^a$
DD25MR75	$62.6\pm0.6^{\rm b}$	$71.7\pm0.8^{\rm b}$	$81.9\pm0.4^{b}$	$9.1\pm0.5^{\rm a}$	$19.2 \pm 0.2^{\rm b,c}$
MR	$61.7\pm0.7^{\rm b}$	$71.5\pm0.3^{\text{b}}$	$79.4\pm0.2^{\rm c}$	$10.1 \pm 0.9^{a}$	$17.7 \pm 0.9^{c}$

<sup>1</sup>Rice flours used for blending consisted of *DD* Dodamssal, *SM* Saemimyeon, *HA* Hanareum No. 4, *GY* Geonyang No. 2, and *MR* Mirchal. The number following each abbreviation indicate the blending ratio  ${}^{2}T_{o}$  Onset temperature,  $T_{p}$  peak temperature,  $T_{c}$  conclusion temperature,  $\Delta H$  gelatinization enthalpy,  $\Delta T$  gelatinization range (T<sub>c</sub> - T<sub>o</sub>)

<sup>3</sup>All data represent the mean of triplicates and standard deviation, values in the same column of the same blend category with different letters (a–d) are significantly different (p < 0.05)

gelatinization was low due to the high amylose content and low crystallinity of DD. The  $\Delta$ H is mainly related to the loss of the double helix order in starch and decreases with amylose content (Gupta et al., 2009).

The  $T_o$ ,  $T_p$ , and  $T_c$  of the RFB were affected by the blending ratio, and had a tendency to decrease as the proportion of the other RF in the blend increased. Except for the  $\Delta H$  value, most of the thermal properties indicated a less interaction between the blending ratios and the temperature related to gelatinization. In the case of  $\Delta H$ , noticeable non-additive effects were observed between the RFB, with different patterns depending on the blended rice varieties. In particular, the  $\Delta H$  of the DD and GY blends was significantly lower than that of individual DD or GY. This may have resulted in lower  $\Delta H$  values by DD granules that were not completely gelatinized, as GY gelatinized earlier while impeding access to the water required for DD gelation. The gelatinization temperature range ( $\Delta T$ ) for the individual RF was 12.1-17.7 °C, whereas the range for the RFB was wider (11.9–24.9 °C). The wide  $\Delta T$  values indicates the presence of a large amount of crystals with varying stability, and therefore the wider  $\Delta T$  of RFB may be due to the lack of homogeneity of the ordered internal

structure (Gupta et al., 2009). Another possible mechanism for this phenomenon could be due to the relatively different  $T_p$  values between the two blended RF. In a previous study of the gelatinization of wheat-rice starch blends, it was reported that the gelatinization of rice starch was delayed due to a decrease in the amount of water available as consequence of the gelatinization of the wheat starch occurring earlier (Waterschoot et al., 2015a). The non-additive effects of this blend was similar to the swelling capacity results discussed earlier. These results suggest that the use of DD for blending could confer distinct properties that differed from those of individual RF, and could also result in unpredictable novel properties due to non-additive effects.

# Pasting properties of rice flour blends

The pasting parameters and RVA profile curve of RF and RFB are shown in Table 2 and Fig. S2. Pasting behavior can provide information on the physicochemical properties of RF and RFB and their potential to improve the processing suitability. In the comparison between varieties, the RVA curve of DD exhibited a gradual shape due to the

Samples <sup>1</sup>	PT (°C) <sup>3</sup>	PV (RVU)	TV (RVU)	BD (RVU)	SB (RVU)	FV (RVU)	(N) (H)	AD (N·s)	СОН
DD	$78.89\pm1.24^{\rm a,b,4}$	$167.00\pm1.18^{\rm d}$	$142.79 \pm 0.65^{d}$	$24.21 \pm 0.53^{c}$	$77.83 \pm 2.95^{\rm b}$	$220.63 \pm 2.30^{\rm d}$	$28.84\pm3.26^{\rm a}$	$2.93 \pm 1.13^{a}$	$0.16\pm0.03^{\rm a}$
DD75SM25	$80.63\pm0.03^{\rm a}$	$169.04\pm2.96^{\rm d}$	$133.92\pm2.00^{\rm d}$	$35.13\pm0.95^{\mathrm{c}}$	$106.5\pm0.42^{\mathrm{b}}$	$240.42 \pm 2.42^{\rm d}$	$18.15\pm0.94^{ m b}$	$2.00\pm0.14^{\rm a}$	$0.10\pm0.01^{\mathrm{a}}$
DD50SM50	$77.85\pm0.40^{\mathrm{b}}$	$404.21 \pm 6.71^{\rm b}$	$291.42 \pm 1.41^{\rm b}$	$112.79 \pm 5.29^{\rm b}$	$222.83 \pm 6.08^{a}$	$514.25 \pm 7.50^{\rm b}$	$16.36\pm0.07^{ m b}$	$1.63\pm0.33^{\mathrm{a}}$	$0.09\pm0.01^{\mathrm{a}}$
DD25SM75	$74.23\pm0.02^{ m c}$	$334.86\pm0.31^{\circ}$	$232.17 \pm 2.25^{\circ}$	$102.67 \pm 1.92^{\rm b}$	$201.21 \pm 1.79^{a}$	$433.38\pm0.45^{\rm c}$	$16.60\pm0.63^{\mathrm{b}}$	$2.15\pm0.38^{\rm a}$	$0.12\pm0.01^{\mathrm{a}}$
$SM^2$	$76.65\pm0.00^{\circ}$	$588.25 \pm 8.49^{a}$	$368.46\pm 6.89^{\mathrm{a}}$	$219.79 \pm 1.59^{a}$	$225.04 \pm 20.92^{a}$	$593.50 \pm 14.02^{a}$	$16.65\pm1.48^{\rm b}$	$1.98\pm0.68^{\rm a}$	$0.12\pm0.06^{\rm a}$
DD	$78.89 \pm 1.24^{\rm a}$	$167.00 \pm 1.18^{\circ}$	$142.79\pm0.65^{\rm c}$	$24.21\pm0.53^{\mathrm{e}}$	$77.83 \pm 2.95^{\circ}$	$220.63 \pm 2.30^{\circ}$	$28.84\pm3.26^{a}$	$2.93 \pm 1.13^{\mathrm{b}}$	$0.16\pm0.03^{ m b}$
DD75HA25	$80.58 \pm 0.02^{\rm a}$	$145.21 \pm 1.79^{\circ}$	$101.54 \pm 1.04^{ m d}$	$43.67\pm0.75^{\rm d}$	$88.04 \pm 1.79^{c}$	$189.59\pm2.83^{\rm c}$	$12.51\pm2.61^{\rm b}$	$3.04\pm0.82^{ m b}$	$0.18\pm0.06^{\rm b}$
DD50HA50	$75.10\pm0.00^{ m b}$	$316.17 \pm 12.75^{\rm b}$	$115.21 \pm 9.42^{\rm b}$	$112.58 \pm 3.33^{\circ}$	$178.38 \pm 1.79^{a}$	$381.96 \pm 21.21^{a}$	$7.07\pm0.07^{\rm c}$	$6.67\pm0.08^{\rm a}$	$0.52\pm0.03^{\mathrm{a}}$
DD25HA75	$70.60\pm0.07^{ m c}$	$321.67 \pm 2.41^{\rm b}$	$158.04\pm2.21^{\rm c}$	$163.63 \pm 4.63^{\rm b}$	$135.67 \pm 1.50^{\rm b}$	$293.71 \pm 0.71^{\rm b}$	$9.92\pm0.59^{ m bc}$	$7.24\pm0.72^{a}$	$0.54\pm0.06^{\rm a}$
$HA^2$	$70.13\pm0.04^{\circ}$	$526.96 \pm 3.48^{a}$	$285.83 \pm 1.18^{\rm a}$	$270.13 \pm 4.66^{a}$	$129.08 \pm 0.59^{b}$	$385.92 \pm 1.77^{\rm a}$	$12.58\pm0.36^{\rm bc}$	$7.62\pm0.65^{a}$	$0.49\pm0.09^{\mathrm{a}}$
DD	$78.89 \pm 1.24^{\rm a}$	$167.00 \pm 1.18^{\rm c}$	$142.79 \pm 0.65^{a}$	$24.21\pm0.53^{\mathrm{e}}$	$77.83 \pm 2.95^{\rm b}$	$220.63 \pm 2.30^{ m bc}$	$28.84\pm3.26^{\rm a}$	$2.93\pm1.13^{\rm b}$	$0.16\pm0.03^{ m c}$
DD75GY25	$80.25 \pm 0.40^{a}$	$117.38 \pm 1.38^{d}$	$81.79 \pm 0.21^{\rm b}$	$35.59\pm1.17^{ m d}$	$70.21 \pm 1.04^{\mathrm{b}}$	$152.00\pm1.25^{\rm d}$	$7.56\pm1.60^{\mathrm{b}}$	$3.75\pm0.93^{\mathrm{b}}$	$0.42\pm0.14^{ m b}$
DD50GY50	$71.43 \pm 0.42^{\mathrm{b}}$	$261.42 \pm 9.83^{\rm b}$	$152.38\pm 6.96^{a}$	$109.04\pm2.88^{\rm c}$	$120.88\pm 6.04^{\rm a}$	$273.25 \pm 13.00^{a}$	$4.44 \pm 0.47^{\mathrm{b}}$	$4.31\pm0.58^{\rm a}$	$0.61\pm0.05^{\rm a,b}$
DD25GY75	$68.60\pm0.00^{\rm c}$	$256.13 \pm 5.88^{\rm b}$	$107.04 \pm 6.04^{\rm b}$	$149.09\pm0.17^{\rm b}$	$79.00 \pm 2.17^{\mathrm{b}}$	$186.04\pm8.21^{\rm c}$	$5.47\pm0.85^{ m b}$	$3.13\pm0.56^{\rm b}$	$0.71\pm0.04^{\mathrm{a}}$
$GY^2$	$67.78\pm0.04^{\circ}$	$464.46 \pm 2.06^{a}$	$156.38 \pm 6.54^{\rm a}$	$308.08 \pm 4.48^{a}$	$81.04\pm0.18^{\mathrm{b}}$	$237.42 \pm 6.72^{\rm a.b}$	$8.15\pm0.15^{\rm b}$	$5.70\pm0.18^{a}$	$0.57 \pm 0.01^{\rm a,b}$
DD	$78.89 \pm 1.24^{\rm b}$	$167.00 \pm 1.18^{\rm b}$	$142.79 \pm 0.65^{a}$	$24.21\pm0.53^{\mathrm{c}}$	$77.83\pm2.95^{\rm a}$	$220.63 \pm 2.30^{a}$	$28.84\pm3.26$	$2.93\pm1.13$	$0.16\pm0.03$
DD75MR25	$88.35 \pm 0.40^{ m a}$	$72.96\pm0.88^{\mathrm{d}}$	$66.00 \pm 1.17^{c}$	$6.96\pm0.29^{\rm d}$	$35.71\pm0.46^{\mathrm{c}}$	$101.71 \pm 1.63^{\circ}$	$7.43 \pm 2.02$	$3.70\pm0.57$	$0.41 \pm 0.07$
DD50MR50	$73.10\pm0.40^{\mathrm{c}}$	$125.17 \pm 3.33^{c}$	$203.58 \pm 4.38^{\rm b}$	$9.96\pm1.04^{ m c,d}$	$49.79 \pm 0.71^{\rm b}$	$165.00\pm5.08^{\mathrm{b}}$	$ND^{5}$	ND	ND
DD25MR75	$68.53\pm0.02^{\rm d}$	$139.63 \pm 2.54^{\rm bc}$	$76.71\pm0.13^{\mathrm{c}}$	$62.92 \pm 2.42^{\rm b}$	$28.29\pm0.21^{ m d}$	$104.85 \pm 0.48^{\rm c}$	ND	ND	ND
$MR^{2)}$	$70.23 \pm 70.23^{d}$	$291.21 \pm 14.20^{a}$	$133.17 \pm 6.13^{a}$	$158.04 \pm 0.87^{\mathrm{a}}$	$37.75\pm0.94^{\mathrm{c}}$	$170.92 \pm 7.07^{\rm b}$	ND	ND	ND
<sup>1</sup> Rice flours us blending ratio	ed for blending cons.	isted of DD Dodamss	sal, <i>SM</i> Saemimyeon	n, HA Hanareum No.	4, GY Geonyang No.	. 2, and MR Mirchal.	The number follow	ing each abbrevia	tion indicate the

Data of pasting properties were adapted from our previous report (Hong et al., 2020)

<sup>3</sup> PT pasting temperature, PV peak viscosity, TV trough viscosity, BD break-down viscosity, SB setback viscosity, FV final viscosity, HD hardness, AD adhesiveness, COH cohesiveness

<sup>4</sup>All data represent the mean of triplicates and standard deviation except for textural properties (n = 5), values in the same column of the same blend category with different letters (a-d) are significantly different (p < 0.05) <sup>5</sup>ND means not determined

Table 2 Pasting and textural parameters of rice flour blends

overall significantly lower viscosity compared to the other RF varieties (Fig. S2). For the pasting temperature (PT), which indicated the gelatinization temperature, DD had the highest value ( $\sim 69.6$  °C) and GY had the lowest value ( $\sim 59.0$  °C), which was the same trend as observed for the thermal parameter T<sub>p</sub>. Despite the high SP values, MR, a glutinous rice variety with the lowest amylose content, had the lowest peak viscosity (PV) following the DD. This is similar to the results for glutinous RF reported in the previous studies, in which a tendency for a lower viscosity than normal RF was found (Yoon et al., 2015). The gelatinization properties are affected by various factors such as the amylopectin chain distribution, lipid content, and protein content as well as the amylose content contained in RF, and granule rigidity can also significant affect the viscosity (Chen et al., 2003). In the case of setback viscosity (SB), which indirectly indicates the retrogradation properties, MR had the lowest value, confirming that the retrogradation proceeded slowly. It was also found that GY, which is a medium waxy variety, had a low value, indicating that it was stable in terms of retrogradation stability (Zaidul et al., 2007). The final viscosity (FV) and SB had a strong positive correlation with the amylose content, except for in DD (Sivaramakrishnan et al., 2004). Overall, DD exhibited a low viscosity properties, with little change with respect to gelatinization and retrogradation as it did not swell well during temperature rise and maintenance, and the swollen starch granules also had a high resistance to heat and shear (Sim et al., 2015).

The low PV of DD was improved by blending with other RF, and increased as the blending ratio of them increased. The increased PV in the RFB suggests that an improved processability was imparted to DD by blending, supporting the improved SP that was attained by blending with other RF (Waterschoot et al., 2015a, b). The breakdown viscosity (BD) value had a tendency to predominantly increase as the blending ratio of other RF in DD increased. However, the BD of the DD75MR25 and DD50MR50 had lower than that of other individual RF, indicating a non-additive effect. In the case of SB, the non-additive effect was apparent in DD50HA50 and DD50GY50, which indicates unstable retrogradation properties.

The improvement effect by blending with other RF did not indicate as the SB value of the individual DD was significantly lower. The final viscosity (FV) of BRF displayed a similar trend under the influence of the SB values (Hormdok and Noomhorm, 2007). A 50:50 blending produced a non-additive effect, suggesting that it could create new gelatinization properties with a higher stability against heat, shear, and retrogradation.

#### Textural properties of rice flour blends

The textural properties of RFB gel are shown in Table 2. In general, stronger starch gel is associated with a higher amylose content and leaching of amylose molecules, as the amylose-based networks provide the elasticity and strength of the starch gel against deformation (Sandhu and Singh, 2007; Tang and Copeland, 2007). The hardness of the starch gel is not only influenced by the amylose content, but also by the content of amylopectin molecules and the content and rigidity of swollen granule fragment/remnant entrapped in amylose-based networks. DD had the highest gel strength (hardness, HD) and matched the highest amylose (Lu et al., 2009; Miles et al., 1985), whereas MR formed a very weak gel that could not be measured with TPA. The HD value was significantly lowered in all RFB samples compared to that of DD, indicating that non-linear blending effects occurred. It was found that when HA and GY blended by more than 50%, the resulting blend was effective for lowering the degree of HD. Previous studies have shown that the non-additive effect on the HD value through blending was decreased compared to the HD of individual samples (Waterschoot et al., 2015b). A possible mechanism for this result is the dilution of amylose contents following blending and limited leaching of amylose molecules from blends due to limitation of their water availability. The gel formation of an RF paste was mainly reliant on swollen granules holding water in the intragranular network. RF with a relatively low gelatinization temperature in RFB would swell earlier, reducing the amount of available water and consequently reducing the SP of DD and limiting amylose leaching, resulting in reduced retrogradation and weak gel formation (Punchaarnon et al., 2008). In the case of AD and COH, the values increased as the blending ratio of other RF increased except for the DD and GY blends. In the DD25GY75, AD showed a non-additively low value, and the COH showed the opposite trend.

# Freeze-thaw stability of rice flour blends

The freeze-thaw stability of RFB was investigated by measuring the amount of water separated over one, three, and five cycles of freezing and thawing (Fig. 2). To analyze the freeze-thaw stability, syneresis was measured using a centrifugation-filtration method. The degradation and storage stability of RFB gels can be evaluated by measuring syneresis, a phenomenon in which water escapes from the polymer network due to retrogradation in the process of repeated freezing and thawing (Xu et al., 2015). Unlike the centrifugation method that is used to measure syneresis, the centrifugation-filtration method has advantage that it does not show experimental errors and is



Fig. 2 Freeze-thaw stability as measured by syneresis (%) of DD and SM blends (A), DD and HA blends (B), DD and GY blends (C), and DD and MR blends (D) at various blending ratios. Each data is the

not subject to the inappropriate conclusions that may occur due to structural characteristics when high amylose varieties undergo multiple freeze-thaw cycles, In addition, the low centrifugal force  $(100 \times g)$  does not cause a significant distortion in the freeze-thaw gel (Charoenrein et al., 2008; Varavinit et al., 2002). DD had the lowest freezethaw stability, which was consistent with previous studies that have reported that flours with a high amylose content have a relatively low freeze-thaw stability (Puncha-arnon et al., 2008). In DD, a large amounts of amylose chains leached from the granules were associated with each other, leading to accelerated retrogradation and consequently decreased stability.

Overall, the syneresis values of all RF increased as the number of freeze-thaw cycles increased, freeze-thaw stability clearly improved by blending. The degree of improvement in freeze-thaw stability was positively correlated with the different in PT values of the two blended RF. Due to their small differences in PT values, the blending of DD and SM did not affect freeze-thaw stability by blending (Fig. 2A). The DD and GY blends with the



mean of three replicates. Bars with different letters (*a*–*d* for 1 cycle; a'-d' for 3 cycle; a''-d'' for 5 cycle) are significantly different (p < 0.05)

largest differences in PT values showed a significant decrease in syneresis compared to the single DD gel, even when only 25% GY was blended in the first cycle (Fig. 2C). In particular, DD25GY75 significantly maintained stability even as the number of freeze-thaw cycles increased. This result may be due to water competition due to the differences in PT between DD and GY. GY with a relatively lower PT swells first, thereby limiting the swelling of DD by reducing the amount of water available for DD gelatinization (Waterschoot et al., 2015b). It was considered that the limited swelling of DD by blending with GY impeded the leaching of a large amount of amylose from the granules and reduced the retrogradation phenomenon caused by the association with leached amylose, thereby enhancing the freeze-thaw stability (Charoenrein et al., 2008; Ma et al., 2020). HA and MR had slightly different effects on freeze-thaw stability as the difference in PT with DD was smaller than that of GY. The DD and HA blends effectively alleviated the syneresis in the first cycle, but the effect was gradually decreased as the cycle was repeated (Fig. 2B). Due to its excellent freezethaw stability, MR was expected to have the best effect when it was blended with DD, but no substantial effect was observed (Fig. 2C). The uneven water distribution of RF during gelatinization led to different behaviors in the blends than would be expected based on the behaviors of the individual RF (Hagenimana and Ding, 2005).

In conclusion, it was confirmed that the physicochemical properties and freeze-thaw stability of DD could be improved through the blending, and they were clearly affected by the addition and blending ratio of various RF blended with the DD. Therefore, considering the unique characteristics of the variety of RF used for blending, as well as the appropriate blending ratios due to their nonadditive effects, it is possible to obtain desirable properties for a wide range of industrial applications.

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#### Declarations

**Conflict of interest** The authors declare that they have no conflict of interest.

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