

## Research Article

# Impact of Emulsifiers Addition on the Retrogradation of Rice Gels during Low-Temperature Storage

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Rice and its products are widely consumed in Asian countries; however, starch retrogradation decreases the quality and shortens the shelf-life of rice foods particularly at low temperature. In this study sucrose ester (SE), glycerol monostearate (GMS), and sodium stearoyl lactylate (SSL) were added to rice flour and corresponding rice gels. Then, gelatinization properties, retrogradation characteristics, texture, and water content of these rice gels were investigated at 4°C and -20°C storage, respectively. The results demonstrated that the rice gels with 0.2% GMS had the lowest retrogradation index ( $\Delta H_r/\Delta H_g$ ) (11.84%) and hardness (1359 g) at 4°C for a 10 d period, which was significantly lower in comparison to control and the other two emulsifiers ( $P < 0.05$ ). Adhesiveness and water content were increased compared to the other samples. Furthermore, the retrogradation of rice gels stored at 4°C was comparatively rapid compared to gels stored at -20°C. Gel samples stored at -20°C were still acceptable for more than 15 days. Thus it was revealed that GMS has the potential to retard starch retrogradation and produce high-quality rice products in preservation.

## 1. Introduction

Rice sustains two-thirds of the world's population at present. Rice from northeast China possesses high nutritional value and good taste, based on consumers' preferences. However, during storage, several physicochemical and physiological changes occur in rice foods including loss of moisture and aroma, ultimately imparting firmness and cracking technically referred to as retrogradation [1]. Starch retrogradation is a process whereby when a gelatinized solution is cooled for a long time, it changes into a gel and rearranges itself into a crystalline structure. It is an unavoidable phenomenon affecting the texture and quality of many ready meals, including starchy rice foods [2, 3]. Starch retrogradation severely affects the nutritional properties and storage stability of these products, seriously limiting the development of the food industry. Therefore, understanding the factors required to curtail retrogradation is of prior concern.

Various methods have been used to control starch retrogradation, namely, physical techniques (temperature, pressure, humidity, and storage conditions), the addition of food additives (emulsifier, hydrocolloids, and nonstarch

polysaccharides), enzymolysis, and biotechnological modifications [4]. Among all food additives, emulsifiers have been widely used in delaying the retrogradation of bread and cake products [5]. Lai [6] reported that the addition of glycerol monostearate (GMS) and sucrose ester (SE) in rice starch formed a starch-emulsifier complex that stabilized the granule and delayed water penetration and swelling. Eduardo et al. [7] found that composite bread with a combination of hydrocolloids and emulsifiers had the lowest crumb firmness ( $P < 0.05$ ), significantly reduced melting enthalpy values compared to the reference bread, and delayed melting of recrystallized amylopectin enthalpy. The antiretrogradation effect of emulsifiers depends on the ratio of amylose and amylopectin in starch [2, 8]. In addition, the rice storage temperature, the water content, and other factors will also influence the result of emulsifiers.

Most of the researchers have tried controlling the temperature and water content along with other methods to inhibit the occurrence of rice retrogradation, but they are still unable to stop it. Research on the effect of emulsifiers on the retrogradation process of rice at low temperatures is also limited. The mechanism of how these surfactants

influenced starch structure is not completely understood. Therefore, the objective of this study was to investigate the changes in the retrogradation index, textural properties, and water content at low temperatures (4°C and -20°C) during the whole storage process and compare the effect of different emulsifiers on rice starch, helping to elucidate the mechanism of antiretrogradation of emulsifiers. These results could provide some theoretical guidance for retarding the retrogradation of rice during its storage.

## 2. Materials and Methods

**2.1. Sample Preparation.** Rice (Zhonggen Rice Industry Co., Ltd., Tianjin, China) was smashed through 100-mesh sieve. The starch content of rice was 67.10%, as determined by the butyl-alcohol sedimentation method. 30.00 g of rice flour was weighed in each beaker, with 0.3% sucrose ester (SE), 0.2% sodium stearoyl lactylate (SSL), and 0.2% glycerol monostearate (GMS) (w/w) and allowed to be mixed completely. Water was added to obtain a ratio of 1:2 (rice/water; w/v). A sample without an emulsifier was used as the control. These samples were then immersed in a hot bath (90°C) for 30 min with subsequent refrigeration and freezing at 4°C and -20°C for a 1, 3, 5, 7, 10, and 13 and 1, 3, 5, 10, 15, and 30 days of storage, respectively.

**2.2. Gelatinization Properties Determination.** Gelatinization properties were determined by DSC (Pyris 6 DSC, PerkinElmer, USA). It was performed according to Tao et al. [9] with some modification. Approximately 4 mg samples were weighed and distilled water was added to obtain a starch-to-water ratio of 1:2 (w/w), and the samples were hermetically sealed. Pans were kept for 10 h at 25°C and then heated from 20°C to 140°C at 10°C/min (empty pan as reference). From the thermograms, the gelatinization enthalpy ( $\Delta H_g$ ), onset temperature ( $T_o$ ), peak temperature ( $T_p$ ), and conclusion temperature ( $T_c$ ) were obtained after the first heating run. Then, the gelatinized rice samples (in the originally sealed pan) were stored at 4°C or -20°C for 1, 3, 5, 10, 15, and 20 days, respectively, for retrogradation research.

**2.3. Retrogradation Properties Determination.** The above stored samples were thawed at room temperature (25 ± 2°C) for 5 h before being scanned again using DSC with the same heating program above to obtain the retrogradation enthalpy ( $\Delta H_r$ ). The peak areas at 40–70°C were calculated as  $\Delta H_r$ . The retrogradation index ( $\Delta H_r/\Delta H_g$ ) was defined as the ratio between retrogradation ( $\Delta H_r$ ) and the gelatinization enthalpy ( $\Delta H_g$ ) [10].

**2.4. Textural Analysis.** A textural analysis of rice gels was performed according to Xia et al. [11] with some modification. Texture profile of the samples was determined by Texture Analyzer TA-XT 2 (Stable Micro Systems, Surrey GU7 1YL, UK). The rice gels (thickness of 2 cm, the diameter of 5 cm) were arranged on a platform. By compressing the samples with a probe (P/50) using a test speed of 0.5 mm/s and

posttest speed of 0.5 mm/s, the deformation level was 40% of the original sample height when the gels were compressed twice. Textural parameters (i.e., hardness and adhesiveness) were determined via Texture Expert software 3.2.

**2.5. Water Content Determination.** The rice gels were weighed (3–4 g) in a weighing pan having a glass cover and placed in a hot air oven at 100–105°C. After 2–3 h, samples were taken out and weighed again until constant weight was obtained. The water content is the ratio between the weight difference and initial weight.

**2.6. Statistical Analysis.** Statistical analyses were performed using SPSS 12.0 software (SPSS Inc.; Chicago, IL, USA). Significant differences among treatments were tested by ANOVA followed by Tukey's test ( $P < 0.05$ ). Data were expressed as the mean values ± standard error (SE). All the analyses were performed in a triplicate run. Data were expressed as the mean values ± standard error (SE).

## 3. Results

**3.1. Gelatinization Properties.** The gelatinization parameters of rice gels with different emulsifiers were shown in Table 1. The range of gelatinization temperatures (onset temperature ( $T_o$ ), peak temperature ( $T_p$ ), and conclusion temperature ( $T_c$ )) for all samples was approximately 60–80°C, and  $T_o$  was approximately 60°C. Compared to the control, values of  $T_p$  and  $T_c$  were significantly increased by the addition of 0.2% GMS ( $P < 0.05$ ).  $T_o$  and  $\Delta H_g$  were not significantly different among the rice gels with and without the addition of emulsifiers ( $P > 0.05$ ). Adding SE and SSL could increase  $\Delta H_g$  values while adding GMS decreased  $\Delta H_g$ . These results suggested that 0.2% GMS could reduce the gelatinization temperature ( $\Delta H_g$ ).

**3.2. Retrogradation Properties.** Figure 1 showed the retrogradation index ( $\Delta H_r/\Delta H_g$ ) of rice gels stored at 4°C and -20°C. With the advancement in storage time prolonged,  $\Delta H_r/\Delta H_g$  was increased gradually, which indicated retrogradation of rice gel was more obvious. Compared to the control, the  $\Delta H_r/\Delta H_g$  values of the samples with emulsifiers significantly decreased at 4°C in Figure 1(a). The rice gels with 0.2% added GMS exhibited no significant change in  $\Delta H_r/\Delta H_g$  during the first 10 days (11.84%) ( $P > 0.05$ ), whereas at 20th days of storage they declined by 30% compared to the control samples thus affirming GMS as an antiaging emulsifier inhibiting the rice starch retrogradation.

The results from Figure 1(b) showed that there was no significant difference in  $\Delta H_r/\Delta H_g$  value between the control and emulsifiers at -20°C ( $P > 0.05$ ). It was observed that adding of emulsifiers in rice gels certainly changed the retrogradation trend in these gels in comparison to the control samples. At the 20th day of storage the  $\Delta H_r/\Delta H_g$  values for GMS (32.19%) and SE (31.21%) were lower than control when stored at -20°C. Furthermore, regarding the retrogradation index, rice gels stored at 4°C had higher values

TABLE 1: Gelatinization properties of rice gels with different emulsifiers (means  $\pm$  SE).

Sample type	$T_o$ ( $^{\circ}$ C)	$T_p$ ( $^{\circ}$ C)	$T_c$ ( $^{\circ}$ C)	$\Delta H_g$ (J/g)
Control	$62.47 \pm 0.33^a$	$68.61 \pm 0.16^a$	$75.34 \pm 0.22^a$	$6.89 \pm 0.04^a$
0.3% SE	$62.54 \pm 0.50^a$	$69.08 \pm 0.10^{ab}$	$76.06 \pm 0.10^{ab}$	$7.91 \pm 0.35^a$
0.2% SSL	$62.60 \pm 0.16^a$	$69.52 \pm 0.16^{ab}$	$76.51 \pm 0.28^{bc}$	$7.55 \pm 0.30^a$
0.2% GMS	$63.31 \pm 0.41^a$	$70.13 \pm 0.57^b$	$76.99 \pm 0.19^c$	$6.70 \pm 0.76^a$

Significant differences in each column were expressed as different letters ( $P < 0.05$ ).  $T_o$ ,  $T_p$ , and  $T_c$  were the gelatinization temperatures of the onset, peak, and conclusion, respectively.  $\Delta H_g$  was the enthalpy change of gelatinization.

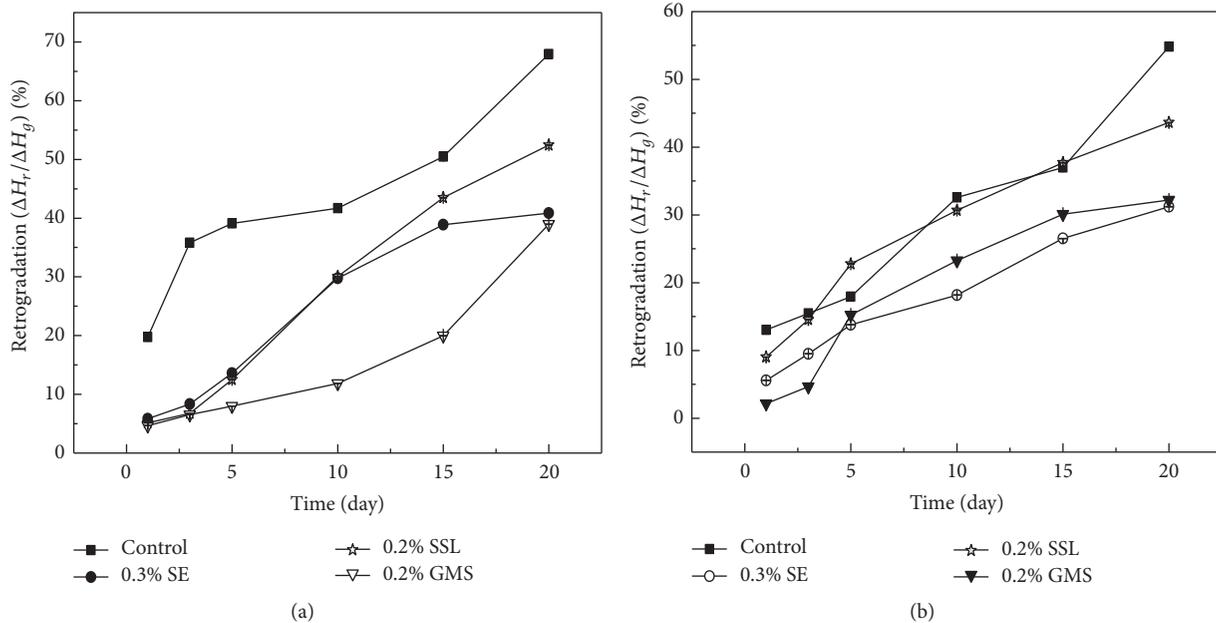


FIGURE 1: Retrogradation index ( $\Delta H_r/\Delta H_g$ ) of rice gels with different days of storage at 4°C (a) and -20°C (b). Error bars indicate standard error.

than that stored at -20°C. Thus, the temperature of -20°C could effectively retard the retrogradation of samples. In conclusion, 0.2% GMS showed a lower rate of retrogradation during the 10 days at 4°C, and the  $\Delta H_r/\Delta H_g$  values (11.84%) were within the satisfactory range.

**3.3. Textural Properties.** The hardness and adhesiveness of rice gels at different storage times were illustrated in Figure 2. During storage of samples at 4 and -20°C, the hardness was gradually increased, whereas the adhesiveness was gradually decreased. In combination with the retrogradation index, it was found that  $\Delta H_r/\Delta H_g$  values were positively correlated with hardness and negatively correlated with adhesiveness (Figures 1 and 2). Similar changes were also reported on waxy and normal corn starch gels retrogradation through controlled freezing rate [12]. In Figures 2(a) and 2(b), the changing rate of hardness and adhesiveness of 0.2% GMS was the lowest at 4°C. For hardness, a slight increase was observed during the first 10 days of 0.2% GMS (1359 g), followed by a noticeable increase throughout the storage period.

In Figure 2(c), the sample with the addition of 0.2% GMS had the lowest hardness values at the same storage time at -20°C. However, there was a sharp increase of almost 76%

in the first 15 days, with a further steady increase up to 3111.47 g at the 30 days of storage. Rice gels showed a steady increase in hardness with gradual decrease in adhesiveness during the first 12 days when stored at -20°C, reaching an asymptote after that. The adhesiveness of rice gels with 0.3% SE (-222.04 g·s) and 0.2% GMS (-204.23 g·s) was similar at the 30 days of storage at -20°C (Figure 2(d)). Therefore, compared to the control, rice gels with 0.2% GMS had lower hardness and higher adhesiveness during the 10 days at 4°C. GMS exhibited better textural properties than SE and SSL. From Figure 2, it can also be seen that -20°C can extend the storage time of rice gels by at least 15 days compared to 4°C.

**3.4. Water Content.** Changes in the water content of rice gels during storage were illustrated in Figure 3. As the storage time proceeds, the water content of samples was reduced gradually at 4°C and -20°C. Compared to the control, rice with the addition of emulsifier had greater water content when stored at 4°C. As shown in Figure 3(a), there was no significant difference among the three emulsifiers ( $P > 0.05$ ). During storage at 4°C, the sample with GMS addition has the highest water content than the other samples (Figure 3(a)). On the other hand, at -20°C, the sample with

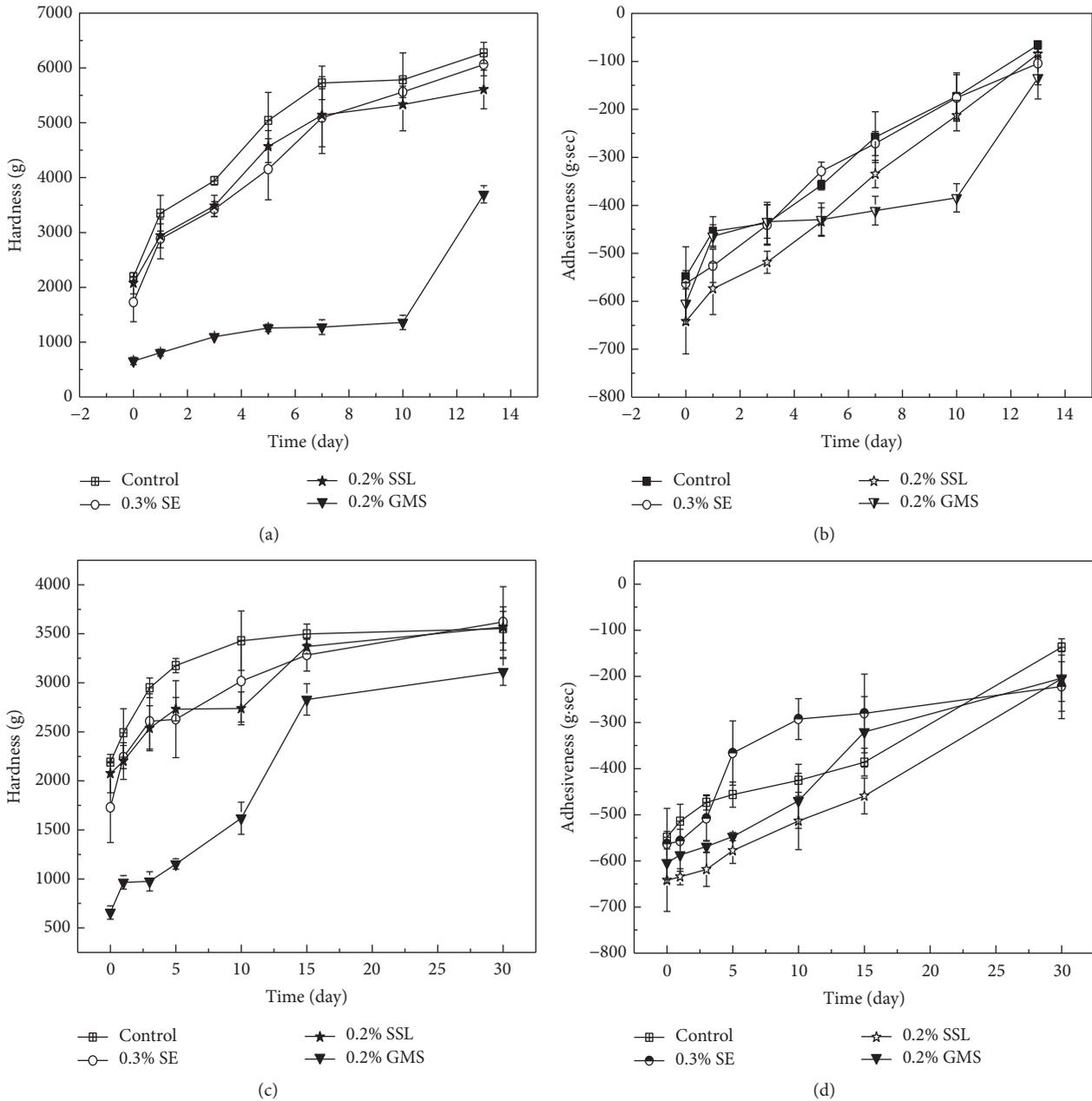


FIGURE 2: Hardness and adhesiveness of rice gels with different time storage at 4°C ((a), (b)) and -20°C ((c), (d)). Error bars indicate standard error.

SE addition had the highest water content (Figure 3(b)). Since retrogradation mainly occurred at 4°C and because the sample with GMS addition had lower hardness, adhesiveness, and retrogradation index than the samples with the other emulsifiers addition at 4°C storage, we choose GMS as a suitable antiretrogradation emulsifier.

#### 4. Discussion

0.2% GMS offered better antiretrogradation abilities than the control (Table 1 and Figure 1). GMS could be combined with starch molecules decreasing the starch crystallization

rate, resulting in prolongation of the retrogradation process at lower temperature [13]. Some researchers have reported that rapid associations of amylose molecules are considered to dominate starch retrogradation during the first days (several hours or more than ten hours) after gelatinization. Furthermore, GMS has the lipophilicity of an emulsifier with a nonpolar group. Hydrophobic groups on GMS were plugged into the alpha-helix structure of gelatinized amylose and combined with starch forming strong stable amylose-lipid complexes, thereby decreasing and preventing rapid retrogradation of amylose during the first 5 days of storage [4, 14].

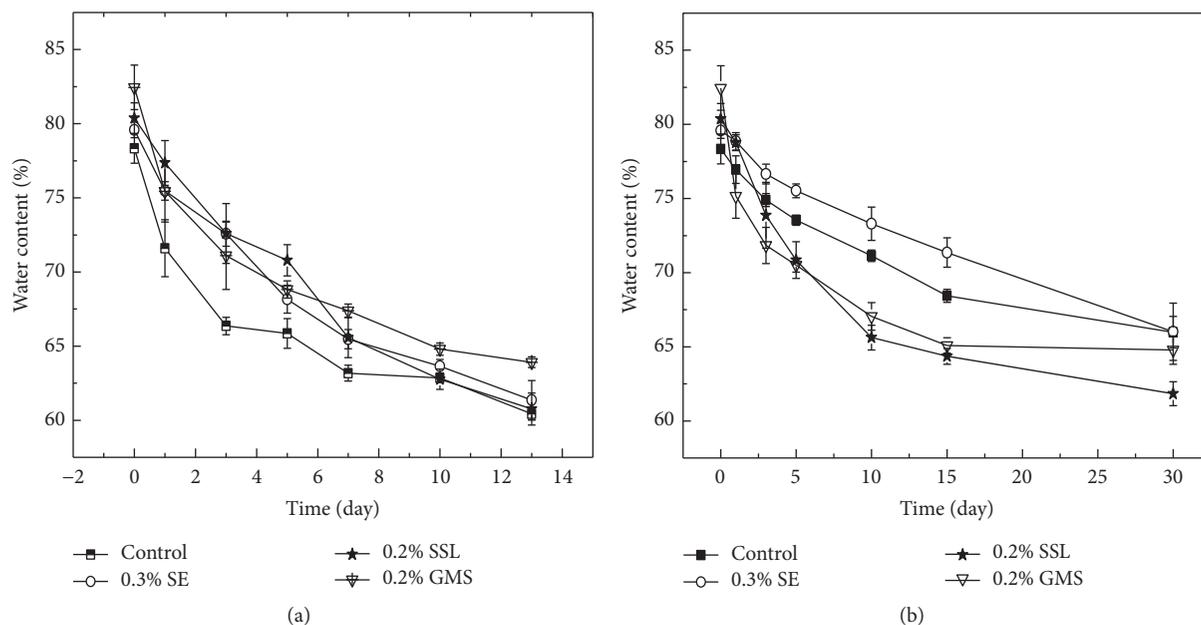


FIGURE 3: Water content of rice gels at different times of storage at 4°C (a) and -20°C (b). Error bars indicate standard error.

Al-Hajji et al. [14] reported that, during prolonged storage time (one to seven weeks), retrogradation mainly occurs in amylopectin. In our research the amylose content of rice was 10.43% and amylopectin was 89.57%. Thus, retrogradation may be mainly determined by amylopectin. As GMS could adhere to the surface of amylopectin and change the water distribution, interaction with the forked chain via hydrogen bonding was altered, reducing the water binding capacity of starch and ultimately indirectly delaying the long-term retrogradation [15]. In our research, the water content of adding GMS was higher than control (Figure 3). The other researchers showed that amylose-lipid complex resulted in the formation of single helical V-amylose complex. It had the capability to act as nuclei for retrogradation by cocrystallizing with amylopectin, affecting amylopectin recrystallization indirectly, and to some extent interfering with amylopectin retrogradation [13].

Retrogradation of rice starch during storage had a direct effect on the texture of rice gels. In our previous study, it was shown that the higher retrogradation index had higher hardness and lower adhesiveness. Adding emulsifier could improve the texture and decrease retrogradation index of rice gels. The texture of rice gels with SSL and SE addition was less better than GMS. As SSL and SE were all negatively charged emulsifiers, so they could repulse and prevent the strong association of amylose-lipid complexes [16]. Therefore, hardness level of samples in which SSL and SE were added changed more quickly than those in the presence of GMS (Figure 2(a)). Since GMS is the hydrophobic emulsifier its lipophilic moieties were attached to the nonpolar side chains of the protein complexes forming an intermolecular matrix via hydrogen bonding and proteins cross-linkages that resulted in reduced hardness [17].

The retrogradation index and texture of rice gels stored in -20°C were better than those stored in 4°C. That means the low temperatures and ultralow temperatures could delay starch retrogradation and maintain the textural properties of rice during the freezing process. The recrystallization of amylopectin was strongly dependent on the temperature, and the rate of nucleation was faster at 4°C [3].

Water plays a critical role in starch gels. The water content defines the extent of the granular form. Slade and Levine [18] reported the decrease in free water of bread with an increase in bound water content under storage. Free water can promote the migration of the starch molecules, while chemically bound water affected the formation of starch recrystallization crystals [19, 20]. In our research, the sample with GMS addition had the higher keeping water ability. It may be because GMS could complex with amylose and prevent the leaching of amylose and increased binding free water capacity [6]. Figures 1 and 3 showed that when water content (free water) decreased, the retrogradation index of rice gels increased. Zhou et al. [21] confirmed that, with water contents of 80% and 90%, no amylopectin crystallization was observed. However, with 60% and 70% water, there was amylopectin crystallization. This may be because when the water content was low, migration of starch molecules was difficult, and when the water content was high, it was easy for starch molecules to migrate due to the decrease of the starch concentration, and the chance of starch cross-linking and polymerization decreased. But when water content was below 70%, all samples' retrogradation index was increased greatly except the sample with GMS addition (Figure 1). It was reported that when the water content was 60%, the rate of long-term retrogradation was the highest [22]. Our research had verified these results. Therefore, it was necessary

to change the water activity of the starch-water system to control the starch retrogradation by adding surfactants and other substances [15].

## 5. Conclusions

Emulsifiers are known to influence the gelatinization and retrogradation of rice starch. In this study, the addition of 0.2% GMS could decrease the gelatinization enthalpy ( $\Delta H_g$ ), retrogradation index, and hardness and increase the adhesiveness and water content. In particular, in rice gels stored at 4°C for a period of 10 days,  $\Delta H_r/\Delta H_g$  values and hardness were reduced by 30% and 76%, respectively, compared to the control, while adhesiveness and water content increased to 120% and 3%, respectively. Furthermore, samples stored at -20°C had a lower degree of retrogradation compared to those stored at 4°C. These results implied that GMS could as a potential antiretrogradation agent retard starch retrogradation during low-temperature storage. This work could provide the theoretical guidance for slowing the retrogradation of starch.

## Additional Points

**Practical Applications.** Rice starch retrogradation phenomenon is accredited for lowering quality and shelf-stability of rice-based food products. Keeping mentioned concern in view, this study was intensive to evaluate the effect of different emulsifiers on antiaging and preservation of rice gels under low-temperature storage. The results showed 0.2% GMS had a good antiaging ability and can keep the texture of rice gel. Furthermore in 4°C storage the retrogradation of rice was faster than that at -20°C. A temperature of -20°C could extend the storage period for more than 15 days. Thus GMS has the potential to retard starch retrogradation and produce high-quality rice products in preservation.

## Conflicts of Interest

The authors have declared no conflicts of interest.

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