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Comparative study on the structure and physicochemical of waxy rice starch by phosphorylation, lactylation and dual-modified

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Abstract

Waxy rice starch was modified by phosphorylation, lactylation and lactic acid-phosphorylation and its structure and thermodynamic properties were studied. Microscopy showed that phosphorylated and lactated starch granules maintained their integrity, while dual-modified starch granules exhibited partial adhesion. FTIR revealed minor spectral differences in phosphorylated starch, while a characteristic peak at 1738 cm^{-1} in lactated and dual-modified starch was due to C = O stretching vibration, indicating the presence of lactate groups. ^{31}P NMR spectroscopy revealed that monostich monophosphate and distich monophosphate were the primary forms of phosphate ester in phosphorylated and dual-modified starch. ^{13}C CP/MAS NMR spectroscopy showed the relative intensity of signals in the C_2, C_3, C_5 and C_6 regions reduced following modification. XRD showed that modification, especially dual modification, reduced starch crystallinity. The three modification methods did not change the non-Newtonian fluid properties of waxy rice starch, and they all belonged to shear-thinning systems and retained the A-type crystalline structure. The gelatinization temperature and enthalpy of modified starch decreased more than that of native waxy starch, especially dual-modified starch.

Keywords: waxy rice starch; phosphorylation; lactylation; dual-modified.

Practical Application: 1.Phosphorylated, lactated, and lactate-phosphorylated glutinous rice starch were prepared. 2.Structural properties of phosphorylated, lactated, and lactate-phosphorylated glutinous rice starch were verified by 31P NMR and 13C CP/MAS NMR spectroscopy. 3.Modified starch reduced crystallinity of starch, especially dual-modified starch. 4.Modified starch reduced gelatinization temperature and gelatinization enthalpy.

1 Introduction

Starch is a natural polymer with the advantages of cost-effectiveness, biodegradability and biocompatibility (Andini et al., 2012). However, water solubility, the loss of glucoside bond viscosity and the lack of some essential functional groups (such as carboxyl, ether and ester groups), limit the effective utilization of native starch (Atrous et al., 2015). Therefore, starch is usually modified to enhance its properties and expand its applications.

With the increasing diversification of food market and the need to continuously improve the added value of waxy rice products (Thomas et al., 2021). The development and application of waxy rice starch requires new methods and new technologies. Furthermore, in order to overcome the disadvantage that gelatinized waxy rice starch loses viscosity during cooling and storage, forming an opaque rigid gel (Butt et al., 2019), waxy rice starch needs be modified to improve its applications in the food industry.

The methods of starch modification include physical, chemical and enzymatic reactions. Physical modifications include hydrothermal treatment, microwave treatment, extrusion, high-pressure homogenization, and ionizing irradiation (Andini et al., 2012). Chemical modification is achieved mainly by cross-linking, oxidation, etherification, esterification or other means of adding functional groups to the hydroxyl group of starch to improve its processing performance and make it suitable for various production and processing techniques (Xu et al., 2021) (Bakouri & Guemra, 2019). Compared with physical processes, chemical modification well-defined goals and proven efficacy; however, it is prone to producing environmental pollutants. Enzymatic modification improves the physical and chemical properties of native starch by changing its molecular structure. However, the requirements were often strict and may not have the desired effect.

The hydroxyl group in starch can react with a variety of water-soluble phosphates, such as sodium orthophosphate (NaH_2PO_4, Na_2HPO_4), sodium pyrophosphate ($Na_4P_2O_7$), sodium trimetaphosphate [$(NaPO_3)_3$] and sodium tripolyphosphate ($Na_5P_3O_{10}$) (Hu et al., 2020). Monophosphate starch was prepared using NaH_2PO_4 and $Na_5P_3O_{10}$, and starch phosphodiester was prepared using $(NaPO_3)_3$ (Deetae et al., 2008). Compared with native starch, phosphorylated starch has lower gelatinization temperature, superior clarity, higher viscosity, lower retrogradation, lower coagulation, less aging, enhanced thermal stability and

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better water retention capacity (Wongsagonsup et al., 2005). The properties of foods containing phosphorylated starch remain unchanged after repeated freezing and thawing, and were widely used in the food industry.

Lactic acid has antimicrobial and moisturizing properties, can be used to adjust pH, while being considered safe and nontoxic by the US FDA (Zambelli et al., 2018). However, limited research has been published on lactated starch, which is the esterification of starch hydroxyl groups using lactic acid instead of hydrogen. Lactated starch displays improved hydrophobicity, reduced gelatinisation temperature and gelatinisation enthalpy, and increased thermoplasticity (Li et al., 2020). Except for the single modification of rice starch, there is no information on the comparative studies of phosphorylated, lactated, and dual-modified starch. In order to obtain a starch that combines the advantages of two kinds of modification, dual-modified starch was prepared by compound modification method of phosphoric acid and lactic acid and the physicochemical properties of the three modified starches were compared.

In this study, waxy rice starch was modified by phosphorylation, lactylation and lactate-phosphorylated, which broadened the application of modified waxy rice starch in food and non-food industries. The purpose of this study was (i) to investigate the structural characteristics of waxy rice starch after modification by phosphorylation, lactic acid and lactate-phosphorylation, (ii) Influence on the thermodynamic properties of starch before and after modification, and (iii) to lay the foundations for further study of the rheological properties of modified starch and broaden its applications within production industries.

2 Materials and methods

2.1 Materials

Waxy rice was supplied by Anhui Guangming Huai Xiang Co., Ltd (Anhui province, China), and starch was isolated by the alkaline method of Yang et al. (2017). The composition of waxy rice starch were 6.02% moisture, 0.045% fat, 0.33% ash, 0.49% protein and 93.6% starch. All chemicals used were analytical grade, including lactic acid ($C_3H_6O_3$), anhydrous disodium phosphate (Na_2HPO_4), and sodium dihydrogen orthophosphate dihydrate ($NaH_2PO_4{\cdot}2H_2O$) and were purchased from Chinese Medicines Group Chemical Co., Ltd (Shanghai, China).

2.2 Preparation of modified starch

Lactic acid modification

The lactic acid modification was produced according to a previously reported method (Butt et al., 2019). The slurry was made from 200 mL of 40% lactic acid solution (pH 3.5) and 100 g of waxy rice starch (dry basis). The slurry was stirred at 25 °C for 12 h, then filtered through a Brinell funnel under vacuum, and the residue placed in a 60 °C until the starch water content had reduced to 5-10%. Subsequently, cooled to 25 °C, then washed with deionized water to remove unreacted lactic acid, and oven dried at 45 °C .

Phosphorylation modification

Phosphorylated waxy rice starch was based on the Yang et al. (2017) described procedure with some modifications. 4.5 g (Na_2HPO_4), 2.5 g ($NaH_2PO_4{\cdot}2H_2O$), 100 g waxy rice starch and 200 mL distilled water were added into a 500 mL Erlenmeyer flask at 35 °C (pH 5.5-6.0), and stirred for 30 min. Following vacuum filtration through a Brinell funnel, the residue was dried at 55-60 °C for 10-12 h, then ground, dried at 60-65 °C for 90 min, and incubated in a 150 °C oil bath for 150 min. Subsequently, the mixture was cooled and 500 mL of 50% (v/v) methanol was added and the slurry was stirred for 30 min. Then, the mixture was vacuum filtered through a Buchner funnel, and the modified starch was washed sequentially with absolute ethanol and deionized water to obtain phosphoric acid modified starch.

Lactate-phosphorylation modification

The lactic acid glutinous rice starch (100 g) suspension solution prepared as described in Section Lactic acid modification, followed the steps prepared in Phosphorylation modification, replacing the native waxy rice starch, and adding phosphoric acid groups to prepare dual-modified starch.

2.3 DS

DS for lactated waxy rice starch

The DS of lactated starch was determined by the method of Liu et al. (2014) and the result was 0.0231.

The formula for calculating DS was as follows (Equation 1):

$$DS = \frac{162W}{100M - (M - 1)\,W} \tag{1}$$

W (% by weight of substituent) = [conjugated lactic acid (g)/sample (g)-conjugated lactic acid (g)] * 100;

M = The molecular weight of the lactic acid substituent is 73.08.

Determination of DS for phosphorylated and dual-modified waxy rice starch

The determination of phosphorus content in modified waxy rice starch was based on the method of Jiang et al. (1999). DS was determined by Manoi & Rizvi (2010) reported method. The Ds of phosphorylated starch and dual-modified starch were 0.0194 and 0.01975, respectively.

Calculated as follows (Equation 2):

$$DS = \frac{162P}{3100\text{-}102P} \tag{2}$$

P (w/w, dry basis) is the phosphorus content (%) in the starch sample.

2.4 Structural analysis

SEM

The particle morphology of the samples was determined by a Hitachi S-4800 cold field emission SEM (SEM; Tokyo, Japan).

FTIR spectroscopy

The FTIR spectra of the samples were measured by Nicolet iS50 FTIR spectrometer (Thermo Nicolet Corp., USA). Mix a fixed mass of sample with 100 times the mass of anhydrous potassium bromide before grinding. The resulting powder was compressed into thin discs.

^{31}P NMR

^{31}P NMR was based on the method of Sang et al. (2007) with some modifications. Phosphorylated starch was digested and converted to phosphorylated dextrin for ^{31}P NMR analysis. The suspension consists of starch (1 g) and sodium acetate buffer (0.1 M, 20 mL), boiled and stirred for 20 min, cooled to 25 °C, then add α -amylase (500 units) a from porcine pancreas to the above mixture and incubate at 25 °C for 10 h, and adjusted to pH 4.5, add glucoamylase (200 μL), incubate at 60 °C for 90 min, the enzymatic solution was centrifuged at 1500 g for 15 min . Subsequently, the supernatant was vacuum dried, and dissolved in 700 μ L D2O. 31P NMR spectra were acquired on a Bruker AVANCE III liquid nuclear magnetic resonance spectrometer (Bruker Wissembourg, France) at 500 and 202.34 MHz. Phosphoric acid (85%) was chosen as the external standard. Spectra were processed and analyzed with MestReNova 14.1.0 software.

^{13}C CP/MAS NMR

Samples were analyzed at a ^{13}C frequency of 100.63 MHz on a solid-state Bruker AVANCE III 400 WB NMR spectrometer (Bruker, Swiss, France). Experimental conditions were as follows: 4 mm CP/MAS detection probe, contact time 2 ms, acquisition time 34 ms, 2400 scans obtained for each spectrum.

XRD

XRD analysis of native and modified starches was carried out using a TTR-III X-ray diffractometer (Rigaku Co. Ltd., Tokyo, Japan) operated at 40 kV and 80 mA.

2.5 DSC

Thermodynamic properties of native and modified starches were determined using a DSC8000 calorimeter (Perkin Elmer Instruments Co. Ltd., Shanghai, China). Starch (2.0 mg) and 6 μL of distilled water were added to an aluminum container, mixed and and hermetically sealed. Subsequently, equilibration to ambient temperature for 12 h.

2.6 Determination of rheological properties

The rheological properties of native and modified starches were evaluated using a rotational rheometer (DHR-1, TA, USA).

Determination of static rheological properties

The starch slurry (3%) was completely gelatinized, the slurry was incubated to 25 °C, and then the shear rate was maintained at 0-1000 (1/s), taking 18 points each time. DHR-1

Determination of dynamic rheological properties

The starch slurry (3%) was completely gelatinized, and select the oscillation measurement mode. The test conditions are as follows: the measurement temperature is 25 °C, the shear strain is 1%, and the frequency sweep range is 0.1~10 Hz.

2.7 Statistical analyses

Significant differences ($P < 0.05$) were calculated by SPSS version 17.0 (SPSS Inc., Chicago, IL, USA).

3 Results and discussion

3.1 Structural properties

SEM

Figure 1 shows the surface appearance of the native and modified starches under SEM. Native starch displayed polyhedral particles with smooth surfaces, no cracks and discreet, separated particles. Phosphorylated waxy rice starch showed similar morphological characteristics. The surface of lactated waxy rice starch granuleswas slightly changed, showing a small degree of roughening and some adhesion. The separation of granules of lactate-phosphorylated waxy rice starch was incomplete, with several large granules being evident. Such granules are formed by the cross-linking of starch molecules.

Structural properties of native and modified starches

The functional group changes of waxy rice native starch and modified starch were shown in Figure 2A. In native starch, the absorption peak at 3385 cm^{-1} corresponds to the stretching vibration of O-H, and the absorption peak at 1250 cm^{-1} by the bending vibration of O-H. The peak at 2931 cm^{-1} is attributed to the stretching vibration of C-H. The peak was corresponding to the bending vibration of water molecules appeared near 1640 cm^{-1}, confirming the existence of bound water in the starch molecules (Seow et al., 2019) The peak near 1152 cm^{-1} was caused by the stretching vibration of C-O-C. With the introduction of phosphate groups in phosphorylated starch, the stretching vibration band of O-H increased, appearing at 3399 cm^{-1} (Hu et al., 2020). It has been reported that the stretching vibration of P-O and the asymmetric stretching vibration of P-O-C correspond to peaks around 1250 cm^{-1} and 1060 cm^{-1}, respectively. In this study, however, the absorption peaks of P-O and P-O-C did not appear due to the low DS of phosphorylation (Qian et al., 2020). Compared with native starch, a new absorption peak appeared at 1738 cm^{-1} in the lactated and lactate-phosphorylated starches. This characteristic peak is due to C = O stretching vibration, a bond created by the reaction of lactic acid and starch.

The ^{31}P NMR patterns of native and modified starch were shown in Figure 2B. The ^{31}P NMR spectrum of phosphorylated starch shows a group of three strong peaks at 2.73, 1.53 and 0.54 ppm. The spectrum of dual-modified starch shows a group of five strong peaks at 2.77, 1.53, 0.77, 0.66 and 0.37 ppm. No significant peaks were seen in native starch or lactated starch. It has been reported that specteal signals at 1 to 3 ppm, -5 to -10 ppm, 15.9 ppm, and -1 to 1 ppm correspond to

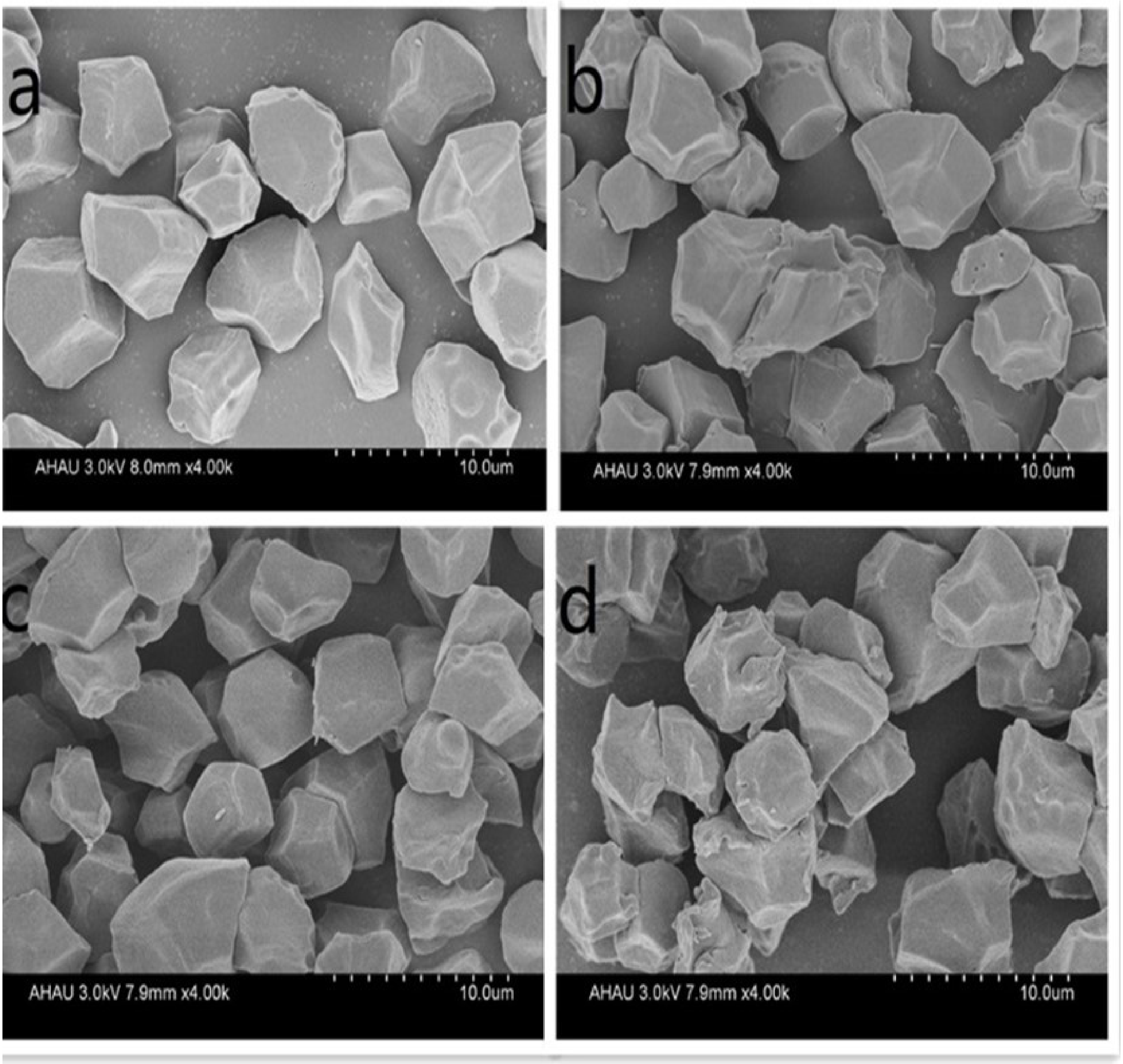

Figure 1. SEM images of native and modified starches. (A) Native starch; (B) phosphorylation; (C) lactylation; (D) dual-modified.

MSMP, MSDP, cyclic-MSMP and DSMP, respectively (Shukri & Shi, 2017). The major types of phosphate ester evident in Figure 2B are DSMP and MSMP, showing peaks at 0 to 1 ppm and 1 to 3 ppm, respectively. Compared with phosphorylated starch, the multi-component peak of dual-modified starch at 0-1 ppm may be due to the access of lactic acid group in the early stage which affects the reaction of phosphorylated group. In addition, the corresponding peak position of Na_2HPO_4 in the ^{31}P NMR spectrum was 0.06 ppm, and that of $NaH_2PO_4 \cdot 2H_2O$ was 2.83 ppm. There were no peaks at these positions in the native and modified starches. Combined with the results of FTIR analysis, this indicates that phosphate groups were still connected after the waxy rice starch had been modified by lactic acid in the dual-modified starch.

Figure 2C shows the ^{13}C NMR spectra of native and modified waxy rice starches. Yang et al. (2017) reported that C_6, $C_{4'}$., C_1 and $C_{2',3,5}$ in glucose displayed chemical shifts at 58-65 ppm, 77-83 ppm, 94-105 ppm and 68-78 ppm, respectively. The C1 signal region of waxy rice starch and three modified starches were obvious triplet peaks with chemical shifts of 90.42 ppm, 92.75 ppm, and 94.88 ppm. Peaks in C4 was expressed as the signal of the starch amorphous region (Fan et al., 2013), and the C4 chemical shift of the modified starch was shifted to the high field, indicating the increase of the starch amorphous

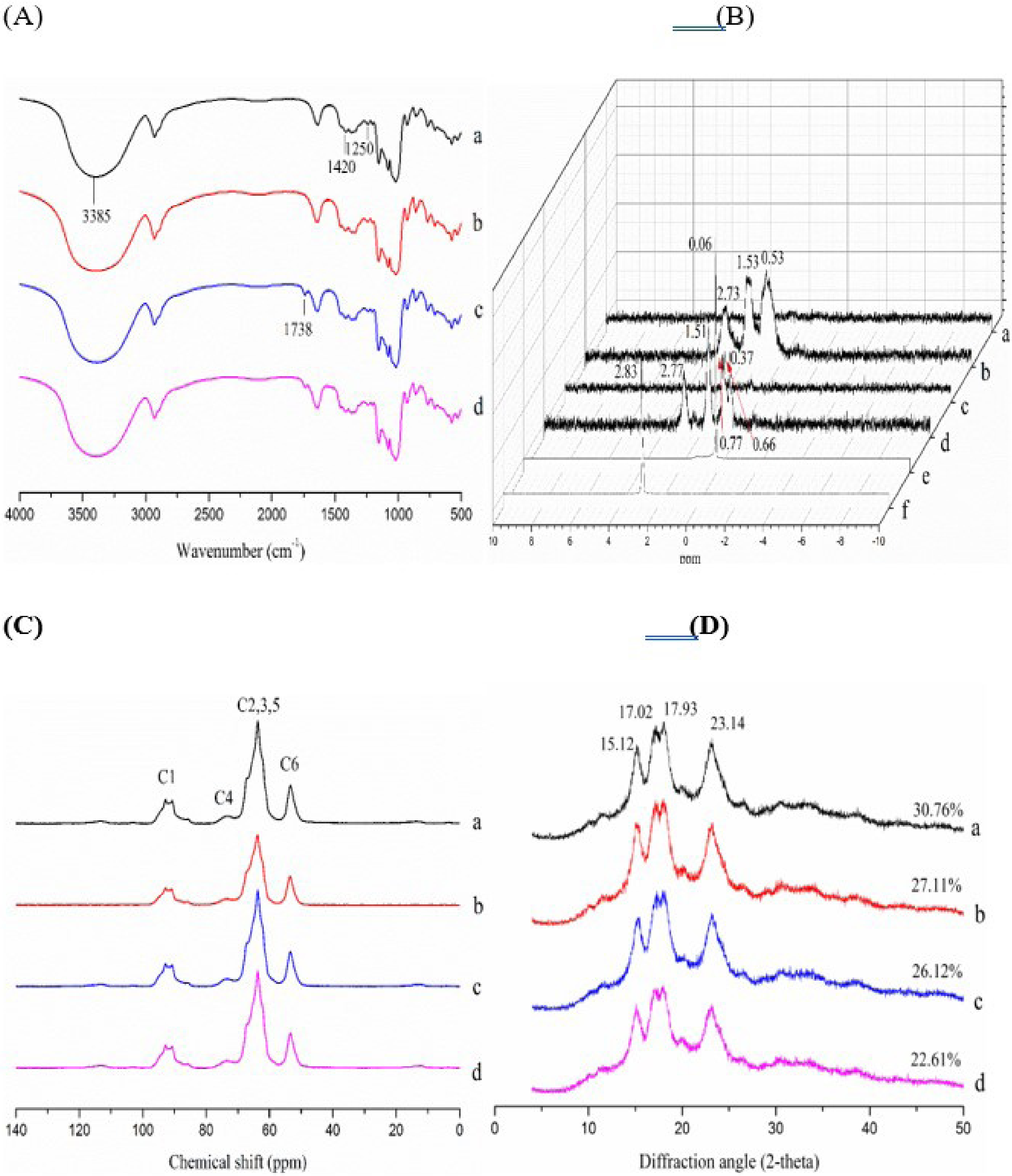

Figure 2. Structural properties of native and modified starches. (A) FTIR; (B) ^{31}P NMR, (a) native starch, (b) phosphorylation, (c) lactylation, (d) dual-modified, (e) Na_2HPO_4, (f) $NaH_2PO_4 \cdot 2H_2O$; (C) ^{13}C CP/MAS NMR; (D) XRD.

region. There were hydroxyl groups in C2, C3 and C6 in starch molecules. It can be noticed that the chemical shifts of C6 in phosphoric acid-modified starch and lactic acid-modified starch were shifted to the downfield by 0.13 ppm and 0.17 ppm, and the chemical shifts of C2, 3, and 5 were shifted to the downfield by 0.22 ppm. In the dual-modified starch, the chemical shift of C6 was shifted downfield by 0.22 ppm, while that of C2, 3, 5 was shifted downfield by 0.25 ppm. As reported by Zhang et al. (2020), the chemical shift shifts downfield, indicating that the reaction occurs at the hydroxyl group at this position. It can be speculated that the changes of functional groups of phosphoric acid-modified starch may be mainly in the C6 region, while the lactic acid-modified starch and dual-modified starch may be in the C2, C3 and C6 regions. In addition, new signal peaks appeared around 13.65 ppm, 113.61 ppm and 13.67 ppm, 113.49 ppm for lactic acid-modified starch and dual-modified starch, respectively. The peaks at 113.61 ppm and 113.49 ppm correspond to the carbon-carbon double bond of C9, while the signal peaks between 0 and 50 ppm were usually attributed to the methyl and methylene groups in alkanes. The appearance of new signal peaks was related to the introduction of lactate groups. The relative intensity of the signal in the C2, C3, C5 and C6 regions was clearly reduced in phosphorylated starch. Combined with results from ^{31}P NMR spectra, it can be seen that phosphate groups may react with hydroxyl groups at C6 regions. There was no significant difference between lactated starch and native starch in the range 0-140 ppm, but the region corresponding to the three active hydroxyl groups was weakened due to the substitution of hydrogen on the hydroxyl groups by lactic acid. However, signals in the C2, C3, C5 and C6 regions of the dual-modified starches were weakened due to the introduction of phosphate groups.

XRD patterns were used to analyse long-range ordered structural changes of starch molecules. XRD patterns of native starch, phosphorylated starch, lactated starch and lactate-phosphorylated starch were illustrated in Figure 2D. Native and modified starches exhibited strong diffraction peaks (2) at 15.12 o, 17.04 o, 17.93 o and 23.14 o. The peaks (2θ) at 17.04 o and 17.93 o were connected dual peaks. There was also a small diffraction peak at 20 o. This is characteristic of A-type crystalline starch structure, similar to other cereal seed starches. There were no significant differences between the XRD patterns of the three modified starches and native starch. This indicates that the long-range ordered structure of the starch granules did not change following modification and that the modification occurred in the amorphous regions of starch granules. The starch crystallization pattern did not change significantly after modification.

The crystallinity of natural starch was 30.76%. After phosphorylation, crystallinity decreased to 27.11%, indicating the presence of more amorphous regions than in native starch. Diffraction intensity also decreased slightly, possibly due to the substitution of some hydroxyl groups by phosphate groups. This is consistent with the findings of Singh & Nath (2012). However, compared with native starch, the crystallinity of lactated starch and lactate-phosphorylated starch decreased further to 26.12% and 22.61%, respectively. Furthermore, diffraction peak intensity was diminished, due to lactic acid molecules entering the starch granules and forming ester bonds with the starch, some even infiltrating the molecular crystalline regions, breaking hydrogen bonds between the molecules and causing starch crystallinity to decrease (Liu et al., 2020). Notably, the crystallinity of dual-modified starch was the lowest, indicating that the greater the modification, the greater the damage to crystallised regions (Wang et al., 2020). However, it has been reported that esterification damages starch granules to a certain extent, but does not change the crystalline structure (Li et al., 2020). The results showed that the three modifications mainly occurred in the non-crystalline region, and the reaction destroyed the original compact structure of the crystalline region of starch granules at a low level, while the starch structure became more and more sparse, resulting in a decrease in crystallinity without changing the crystalline form of starch.

3.2 Analysis of DSC

DSC was used to determine starch transition temperature and enthalpy of gelatinization, which are related crystallinity and other properties of starch particles. Onset, peak and conclusion gelatinisation temperatures, and gelatinisation enthalpy are shown in Table 1. Crystallisation provides structural stability and makes starch particles more resistant to gelatinization (Zeng et al., 2015). Compared with native starch, gelatinisation temperatures and enthalpy of gelatinisation of modified starch both decreased, so modified starch was more easily gelatinised. During the initial stages of gelation, the energy demand was largest in native starch. Gelatinisation temperature ranged from 63.23 °C to 73.15 °C, T_p being 68.52 °C. The gelatinisation temperature of phosphorylated starch decreased from 60.70 °C to 72.26 °C, T_p also decreasing to 66.93 °C, due to substitution by phosphate groups which destroyed the structure of the starch, leading to it being more easily destroyed by elevated temperatures. This is consistent with the findings of Gao et al. (2020). T_o, T_p, Tc and ∆H all decreased after phosphate modification. The infiltration of phosphate groups reduced hydrogen bonding between starch molecules and improved structural flexibility and, hence, the swelling properties of the starch particles. The gelatinisation temperature was, consequently, reduced, in keeping with the finding by XRD of decreased crystallinity, and the phosphorylated starch became more easily gelatinised.

Gelatinisation temperature and ΔH decreased in both lactated starch and lactate-phosphorylated starch. The enthalpy of gelatinisation represents the energy required to destroy the double helix in the crystalline and amorphous regions during the gelatinisation process (Sindhu & Khatkar, 2018). The gelatinisation temperature is related to the unwinding

Table1. DSC of native and modified waxy rice starches.

sample	Onset (°C)	Peak (°C)	End (°C)	∆H (J/g)
Native	63.23 ± 0.05^{a}	68.52 ± 0.10^{a}	73.15 ± 0.22^{a}	11.03 ± 0.46^{a}
phosphorylation	60.70 ± 0.55^{b}	66.93 ± 0.05^{b}	72.76 ± 0.82^{a}	7.06 ± 0.02^{b}
lactylation	59.73 ± 0.19^{c}	65.59 ± 0.05^{c}	70.53 ± 0.53^{b}	8.00 ± 0.77^{b}
dual-modified	57.67 ± 0.17^{d}	63.80 ± 0.04^{d}	69.54 ± 0.12^{b}	7.50 ± 0.42^{b}

Values are expressed as mean ± SD of three determinations. a-d: Data are means of triplicate analyses with standard deviation. Means in the same column with different uppercase letters were significantly different ($p < 0.05$).

and melting of the double helix of amylopectin agglomerates (Zheng et al., 2017). Decreased gelation temperature and ∆*H*, and early breaking of the amylopectin double helix structure were due to the introduction of new functional groups. Lactic acid esterification replaced hydrogen in the starch hydroxyl groups and hindered the linking of linear molecular chains. These ester groups increased the free volume within the starch molecular chains, reducing the strength of hydrogen bonding between molecules, thus lowering the strength of the crystal structure, It is consistent with the results of gelatinization temperature and enthalpy (Li et al., 2020). Lactate-phosphorylated starch contained both lactic and phosphoric acid groups, and its gelatinisation temperature of 57.67 °C was lower than that of both phosphorylated starch and lactate starch. According to Zhang et al. (2017), the introduction of a large number of functional groups into the framework of biopolymers enhanced structural flexibility (thermoplasticity) and contributed to the reduced gelation temperature of modified starch.

3.3 Rheological properties analysis

Analysis of static rheological properties

The static rheological results of waxyrice starch and three modified starches are shown in Figure 3A-B. The results show

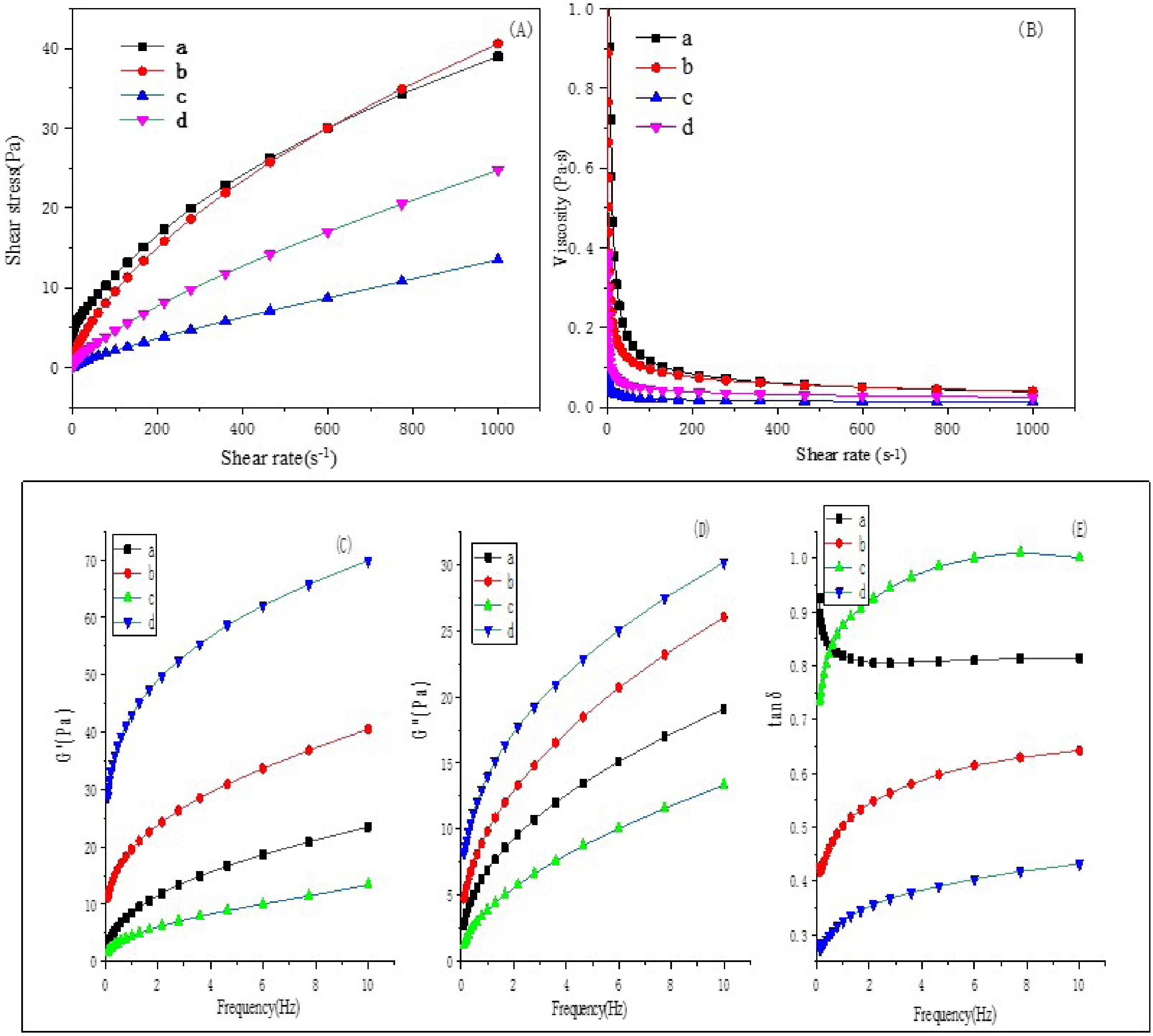

Figure 3. Static and dynamic shear curves of waxy rice starch and modified starch.(A)shear srtess of shear Rate Curve; (B) viscosity of shear Rate Curve;(C)G' of frequency Variation Curve; (D)G"of frequency Variation Curve(E)tan of frequency Variation Curve; (a) Native starch; (b) phosphorylation; (c) lactylation; (d) dual-modified.

that with the increase of shear rate, the shear stress of native starch and modified starch increases,and the curves exhibit varying degrees of tending to the shear stress axis, exhibiting the properties of non-Newtonian fluids (Yu et al., 2019). In addition, with the increase of shear rate, the apparent viscosities of the four starches all showed a downward trend, and the curves showed a convex trend toward the origin, indicating that the four starches belonged to the shear-thinning system. Our results indicated that the modification did not change the rheological model of starch. At the same shear rate, the apparent viscosity of starch showed as follows: waxy rice starch > phosphorylated modified starch > double modified starch > lactic acid modified starch.

Analysis of dynamic rheological properties

Figure 3C-E are the dynamic rheological model diagrams of waxy rice starch and three modified starches. It can be seen from the figure that both G' and G" of starch before and after modification increase with the increase of frequency, showing the dynamic rheological characteristics of weak gel. During dynamic rheological scanning, the G' of waxy rice starch, phosphorylated and dual-modified starch was greater than G", and these three starch gels exhibited solid properties, indicating that the gel deformation of the three starches was elastic and recoverability (Yousefi & Razavi, 2015). The ratio of loss modulus to storage modulus was expressed as loss tangent (tanδ), which was used to describe the viscoelastic behaviour of starch gels. The tanδ < 1 indicates that starch gels mainly exhibit elastic behaviours, and vice versa. It can be seen from Figure 3E that the tanδ of waxy rice starch, phosphate-modified starch and dual-modified starch are all less than 1 in the entire frequency sweep range, which illustrates the weak elastic gel properties of the three starch samples. However, the G' and G" of the lactic acid-modified starch among the three modified starches were different from those of the other three starches during the frequency sweep, which were all lower than their native starches, which may may be attributed to the fact that lactic acid-modified starch reduced the viscosity of waxy rice starch, and the friction between the samples was smaller, and the moving speed was faster.

3.4 Pasting properties

The pasting properties of waxy rice starch and three modified starches were shown in Figure 4. It can be observed from the figure that the peak viscosity and gelatinization temperature of the three modified starches were lower than those of the native waxy rice starch. This may be attributed to the fact that the potential steric hindrance effect of the access group, the binding ability between starch granules decreases, and gelatinization was more likely to occur. The breakdownvalue was the difference between the peak viscosity and the minimum viscosity, which can reflect the heat resistance and shear resistance of starch (Li et al., 2019).

According to the results in Table 2, the breakdown values of waxy rice starch and three modified starches were 553.00, 344.33, 368.33, and 465.33, respectively. The breakdown value was inversely proportional to the thermal stability of starch paste, and the results showed that the stability of modified starch was higher than that of waxy rice starch. According to Qian & Kuhn (1999), the setback value was related to starch retrogradation and was the difference between the final viscosity and the minimum viscosity. The retrogradation values of waxy rice starch and three modified starches were 134.33, 55.67, 9.67, and 46.33, respectively, indicating that the retrogradation value of modified starch was smaller than that of waxy rice starch, showing better anti-aging properties. It is attributed to the fact that the connection of linear molecular chains was effectively hindered by the introduction of bulky groups, which increases the free volume within the starch molecular chain and reduces the tension caused by intermolecular hydrogen bonds, thereby reducing the setback value. Lactic acid modified starch showed the lowest retrogradation value, which may be related to the structure of modified starch granules, pH value and other

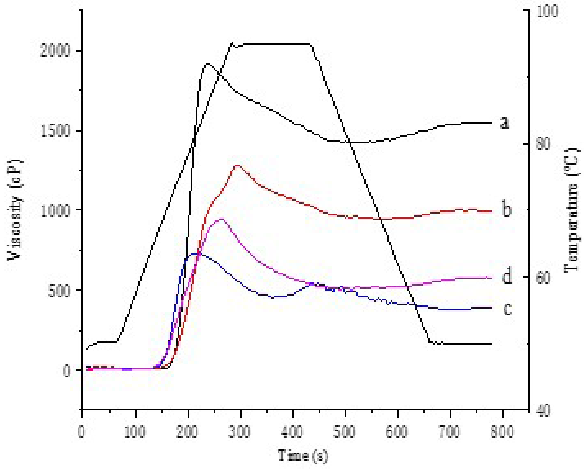

Figure 4. Pasting properties of waxy rice starch and modified starch. (a) Native starch; (b) phosphorylation; (c) lactylation; (d) dual-modified.

Table 2. Pasting properties of waxy rice starch and modified starch.

sample	Peak Viscosity (cP)	Minimum Viscosity (cP)	Breakdown (cP)	Final Viscosity (cP)	Setback (cP)	Pasting Temperature (°C)
Native	1,918.33 ± 0.88^{a}	1,366.33 ± 5.66^{a}	553.00 ± 6.00^{a}	1,500.00 ± 6.00^{a}	134.33 ± 9.33^{a}	70.06 ± 1.08^{a}
phosphorylation	1,273.67 ± 6.83^{b}	930.67 ± 8.83^{b}	344.33 ± 1.33^{d}	986.00 ± 1.00^{b}	55.67 ± 2.33^{b}	68.13 ± 0.24^{b}
lactylation	761.67 ± 8.16^{d}	395.00 ± 11.13^{d}	368.33 ± 6.33^{c}	402.33 ± 2.33^{d}	9.67 ± 0.33^{d}	67.65 ± 0.01^{c}
dual-modified	998.67 ± 7.83^{c}	534.00 ± 9.50^{c}	465.33 ± 8.33^{b}	579.33 ± 0.33^{c}	46.33 ± 5.33^{c}	66.95 ± 0.50^{d}

The experiment was repeated three times. The data in the same column represent the mean ± standard deviation (SD), and different superscript letters indicate that the data is significant ($P < 0.05$).

factors. The viscosity of dual-modified starch was higher than that of lactic acid-modified starch because of the increase of phosphoric acid groups.

4 Conclusion

Phosphorylated, lactated and lactate-phosphorylated starches were prepared from waxy rice starch. The presence of phosphate and lactate groups was confirmed by FTIR, ^{31}P NMR and ^{13}C CP/MAS NMR analyses. Modification reduced starch crystallinity but did not change the A-type crystalline patterns. Gelatinisation temperature and gelatinisation enthalpy of starch was reduced by chemical modification, and the thermoplasticity was increased. The G' > G" values of phosphoric acid-modified starch and dual-modified starch were both smaller than those of waxy rice starch, and the G' and G" values of lactic acid-modified starch were not significantly different. The thermal properties of waxy rice starch were successfully improved by chemical modification. The RVA results showed that the gelatinization temperature of the modified rice starch decreased, and the peak gelatinization viscosity, breakdownvalue and setback value increased. In the comparative study of the three modified starches, the research results help to reduce the viscosity, gelatinization temperature and gelatinization enthalpy of waxy rice starch, and the gelatinization temperature and gelatinization enthalpy of dual-modified starch were the lowest. Based on the above results, the comparative study on the structure and properties of three modified waxy rice starches provides a theoretical basis for the development of starch products, and expanding the application of starch in the food industry.

Abbreviations

DS: degree of substitution. DSC: differential scanning calorimeter. Gelatinization temperature at onset: (To); peak: (Tp); end (Tc). ΔH: transition enthalpy. FTIR: Fourier transform infrared spectroscopy. NMR: nuclear magnetic resonance. XRD: X-ray diffraction. SEM: Scanning electron microscopy. MSMP: monostarch monophosphate. MSDP: monostarch diphosphate. cyclic-MSMP: cyclic-monostarch monophosphate. DSMP: distarch monophosphate. G': storage modulus. G": loss modulus. RVA: Rapid Visco Analyser.

Conflict of interest

None.

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