

Preparation of resistant starch from starch–guar gum extrudates and their properties

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Abstract

Mixtures of starch, guar gum and citric acid were extruded at a temperature of 150 °C and screw speed of 180 rpm. Properties of the extrudates such as bulk density, expansion ratio, apparent viscosity and resistant starch content were measured with different concentrations of guar gum. Extrusion with citric acid influenced the physical properties and resistant starch content of the extrudate. Expansion ratios were lower for samples extruded with citric acid, and decreased as starch–gum concentration increased. The apparent viscosity of the extrudate increased with increasing guar gum concentration; however, citric acid at 2.0% concentration was found to lower the viscosity of the extrudate. Resistant starch content increased from 6.23% for extruded starch only to 14.21% for starch–gum extrudate, and further enhanced to 16.19% with the addition of 2.0% citric acid to the starch–gum extrudate. Resistant starch increased with increase of gum concentration and decreased with increase of starch–gum concentration from 7.5% to 12.5%.

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1. Introduction

As a new category of food ingredient, resistant starch is being examined for both its potential health benefits and functional properties in foods. Resistant starch has been defined by the European Flair Concerted Action on Resistant Starch (EURESTA) as the starch or products of starch degradation that escape digestion in the small intestine of healthy individuals and may be completely or partially fermented in the colon (Englyst, Kingman, & Cummings, 1992). Resistant starch (RS) was categorized into four classes, according to the mechanism that prevents its enzymatic digestion. Among these four types, RS type III seems to be of particular interest, because of its thermal stability, which allows it to be stable in most normal cooking operations, and enables its use as an ingredient in a wide variety of conventional foods (Faraj, Vasanthan, & Hoover, 2004; Haral-

ampu, 2000). Resistant starch formed during processing is associated with amylose retrogradation (Miles, Morris, Oxford, & Ring, 1985; Shamai, Bianco-Peled, & Shimoni, 2003; Sievert & Wursch, 1993).

It has been widely known in the food industry that non-starch hydrocolloids, especially native and modified polysaccharides, exhibit many functions, e.g., to control rheological and textural properties of foods, to improve moisture retention, and to maintain overall product quality during storage (Funami et al., 2005a). Galactomannans are water-soluble polysaccharides found in the seed endosperm of a variety of legumes and consist of a (1–4) linked β -D-mannopyranosyl backbone partially substituted with (1–6) linked α -D-galactopyranosyl substituents (Dea & Morrison, 1975). The most important galactomannans, industrially, are guar gum and locust bean gum (Goycoolea, Morros, & Gidley, 1995). Guar gum, a kind of cold water-soluble galactomannan, is widely used as a thickening and stabilizing agent, in a range of industrial applications (Wang, Ellis, & Ross-Murphy, 2000). The great advantage of galactomannans is their ability, at relatively

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low concentrations, to form very viscous solutions that are only slightly affected by pH, added ions, and/or heat processing (Sittikijyothin, Torres, & Goncalves, 2005). Therefore, galactomannans have been widely applied to food (Sudhakar, Singhal, & Kulkarni, 1996). Many investigations have tried to clarify the role and potential usefulness of galactomannans in controlling rheology and modifying the texture of starch-based food products. These investigations concluded that the addition of galactomannans enhance or modify the gelatinization and retrogradation behaviour of starch (Alloncle, Lefebvre, Llamas, & Doublier, 1989; Christianson, Hodge, Osborne, & Detroy, 1981; Funami et al., 2005b, 2005c; Kulicke, Eidam, Kath, Kix, & Kull, 1996; Rayment, Ross-Murphy, & Ellis, 1995; Sudhakar et al., 1996). Citric acid has been used before as a substituent of the hydroxyl group on starch, in order to modify different features during food production, and it is rated as nutritionally harmless, compared to other substances used for derivatization (Wepner, Berghofer, Miesener, Tifenbacher, & Ng, 1999).

To the best of our knowledge, there is no report on the interaction of a starch–guar gum combination in a twin screw extruder, in relation to resistant starch formation. The objective of this paper is to determine the changes that occur in physical and rheological properties and resistant starch formation when starch is extruded with guar gum, with or without the chemical additive, citric acid.

2. Materials and methods

2.1. Materials

A commercial corn starch (13.5% moisture, 23% amylose) was obtained from Wuxi starch factory, Wuxi, China. Guar gum was obtained from Shanghai Food Gum Factory Ltd. Food grade citric acid was obtained from Roche (China) Co. Ltd., Wuxi. Heat-stable α -amylase Termamyl 120 L (EC 3.2.1.1 from *Bacillus licheniformis*, 120 KNU/g), the protease Alcalase 2.4 L (EC 3.4.21.62, from *Bacillus licheniformis*, 2.4 AU/g), and the amyloglucosidase AMG 300 L (EC 3.2.1.3, from *Aspergillus niger*, 300 AGU/g) were from Novo Nordisk, Shanghai Co. Ltd., China.

2.2. Extrusion

Starch–guar gum mixtures were prepared by mixing corn starch (23% amylose) with various concentrations of guar gum (2%, 5%, or 10% db) in a Hobart blender for 2 min. The mixture was adjusted to a moisture content of 25% (db) during blending. Then citric acid was added at a level of 2% (w/w) and mixed for a further 5 min. The samples were put in a sealable plastic bag.

Extrusion was performed on a co-rotating twin screw extruder (model BC 45, Cletral, Firminy, France). The screws were 55.5 mm in diameter and had an overall active length of 500 mm. A module with a reverse flight was located on the terminal position of each screw, just before

the two dies, of 50 mm length and 5 mm diameter. The extruder was run at a temperature of 150 °C and screw speed of 180 rpm. The feeder was set at a controlled mass flow rate. The feeding section of the barrel was cooled with running water and the terminal section was heated by an induction heating belt. The extrudates were allowed to cool at room temperature and stored in a sealed plastic container until further analysis.

2.3. Measurement of apparent viscosity

Apparent viscosity was measured by the method of Christianson and Bagley (1983), with a slight modification, using a Haake Rotovisco viscometer with a coaxial cylinder design (Haake, Saddlebrook, NJ). A 200 g slurry was prepared by adding ground samples to distilled water at a 10% (w/w) concentration. The slurry was heated at 65 °C with continuous stirring, using an electrical stirrer for 15 min. After cooking, samples were removed from the water bath and poured into the viscometer cup and viscosity measurements were made at 60 °C. Readings were made progressively from 0 to 256 s⁻¹. All measurements were done using the MV-I cup. Data reported were the average of three measurements for each sample.

2.4. Expansion ratio (ER) and bulk density of extrudate

Expansion ratio of the extrudate was measured as the ratio of the cross-sectional area of the extrudate to that of the die (Yuryev, Zasytkin, Alexeev, & Bogatyryev, 1995), given by the following equation:

$$ER = \frac{D^2}{d^2},$$

where D is the diameter of extrudate (mm) and d is the diameter of die hole (mm). The diameter of extrudate was the average of 10 random measurements. The vernier caliper used for the measurements had a precision of 0.01 mm.

Bulk density was defined as the ratio of the mass of the sample to that of its volume, including the voids. Bulk density was determined by measuring the volume of extrudates by glass bead displacement (Hwang & Hayakawa, 1980). Bulk density (ρ_{ex}) of the extrudates was calculated as follows:

$$\rho_{ex} = \frac{W_{ex}}{W_{gb}} \times \rho_{gb},$$

where ρ_{ex} is the bulk density of extrudate (g/cm³); ρ_{gb} is the bulk density of glass beads (g/cm³); W_{gb} the weight of glass beads (g) and W_{ex} the weight of extrudate (g).

Five replications were measured for each sample.

2.5. Resistant starch preparation

2.5.1. Gel preparation

The extruded samples were mixed with distilled water at three different concentrations (7.5%, 10%, 12.5% (w/v)) to

form starch gels and kept at 4 °C overnight. After addition of a fourfold amount of 80% ethanol and homogenization at 10,000 rpm for 5 min with a homogenizer (high speed controllable homogenizer, FSH-2 Model), the homogenized samples were filtered through a glass filter and the resulting residues were dried overnight at 40 °C in an air oven and reground, to get dried retrograded starch gel powders (DRSG) (Vasanthan & Bhatt, 1998).

2.5.2. Isolation of resistant starch

Resistant starch was isolated, according to the method of Mangala, Malleshi, Mahadevamma, and Tharanathan (1999) with slight modification. One gram of DRSG was weighed and into a bioreactor containing 100 ml of distilled water, and the mixture was heated by circulating hot water from a water bath equipped with a pump. After the internal temperature of the bioreactor has reached 95 °C, 0.5 ml of thermostable α -amylase (Termamyl 120 L) was added. After the slurry incubated at the desired temperature for 30 min, the reaction mixture was cooled to room temperature and centrifuged at 5000g for 10 min. The resulting residue was suspended in 100 ml of phosphate buffer (0.08 M, pH 7.5) and treated with protease (0.5 ml) at 60 °C for 30 min. After the pH had been adjusted to 4.5 with diluted HCl, amyloglucosidase (0.5 ml) was added and the mixture was incubated at 60 °C for 30 min. The suspension was centrifuged at 5000g for 10 min. The insoluble residue was washed several times with distilled water. Finally, it was washed twice with 80% (v/v) ethanol and 95% (v/v) ethanol successively, and then dried at 40 °C overnight in a vacuum oven to get resistant starch.

2.6. Resistant starch determination

The resistant starch was analysed according to the methods of Goni, Garcia-Diz, Manas, and Saura-Calixto (1996). Sample (10 mg) was dispersed with 5 ml of 2 M KOH and stirred for 30 min at ambient temperature. After the pH had been adjusted to 4.5 with diluted HCl, amyloglucosidase (0.05 ml) was added and the mixture was incubated at 60 °C for 35 min. The released glucose was measured by the glucose oxidase method (Dahlqvist, 1964). The content of resistant starch was calculated as the product of free glucose from resistant starch hydrolysis with amyloglucosidase and a correction factor of 0.9 as follows: resistant starch = glucose \times 0.9 (Escarpa, González, Morales, & Saura-Calixto, 1997). Each representative experiment was performed in triplicate and the average value was taken.

3. Results and discussion

3.1. Effect of citric acid on viscosity of the guar gum–starch extrudate

The effect of citric acid at the level of 2% on the viscosity of the extruded starch with different concentrations of guar

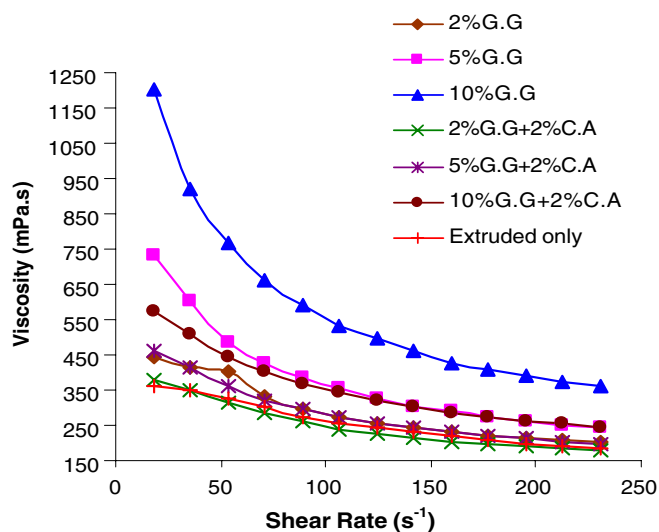


Fig. 1. Viscosity curves for extruded guar gum–starch mixture with or without citric acid. G.G, guar gum; C.A, citric acid, Extruded only means extruded starch only. Values reported were the means of three measurements.

gum was studied. Fig. 1 shows the viscosity curves (60 °C) of an extruded starch–gum system cooked at 65 °C. A trend was observed in which the viscosity of the mixture increased as concentration of guar gum increased from 2% to 10%, in the presence or absence of citric acid. The viscosity increase of extrudates might be due to the inherent viscosity of the guar gum and the synergistic interactions between guar gum and starch polymer (amylose and amylopectin) (Alloncle et al., 1989; Christianson & Bagley, 1983; Funami et al., 2005b). In addition, extrusion cooking, a high temperature short time process, has been shown to facilitate gelatinization (Wang, Klopfenstein, & Ponte, 1993). When starch–guar gum mixtures gelatinize, the swollen starch granules are disintegrated by gelatinization; the thickening effect of water-soluble guar gum may increase the effective concentration of amylose in the continuous phase, where the interactions between guar gum and swollen starch particles may rapidly take place (Funami et al., 2005a). All of these might be responsible for the increase of viscosity of the extruded system.

Although guar gum is reasonably stable in an acidic environment (Wang et al., 2000), with the addition of 2% citric acid, a high degree of reduction occurred in the viscosity of starch–gum mixtures for all samples. The addition of citric acid certainly reduced the pH of the system, and the acid hydrolysis of polysaccharides under extrusion at high temperature resulted, so that the viscosity of extrudates decreased.

3.2. Effect of citric acid on expansion ratio and bulk density of starch–guar gum extrudate

The expansion ratio is the most common descriptor for expanded materials (Guan & Hanna, 2004). Expansion ratios were measured to observe the physical changes that

occurred during extrusion cooking. The extent of physico-chemical changes during extrusion affects product expansion (Choudhury & Gautam, 1998). A lot of factors have been put forward to explain the increase or decrease in expansion ratios of extruded product, which includes starch types, amylose content, and structure of amylopectin (Chinnaswamy & Hanna, 1990).

Table 1 shows the physical properties of the extruded starch–guar gum combination, with or without addition of citric acid. The results show that extruded starch had a higher expansion volume than samples with guar gum, or guar gum and citric acid, and the expansion ratio decreased with increase in concentration of guar gum, which was probably due to viscosity changes that influenced expansion of starches. It is well known that the addition of guar gum increases the viscosity of starch dispersion (Lee, Baek, Cha, Park, & Lim, 2002; Yoshimura, Takaya, & Nishinari, 1999). An explanation for the guar gum–starch extrudates could be that as the viscosity increases with an increase of gum concentration, gelatinization is delayed, thereby hindering starch swelling and subsequent release of amylose and amylopectin. This tends to lower the ability of the material to expand. Furthermore, the high moisture content (25%) and the screw speed (180 rpm) at which the samples were extruded may have accelerated the movement through the extruder and thus decrease the residence time of the material in the extruder.

A similar trend of decrease in expansion ratio was also observed for samples in which citric acid was added before extrusion. This is similar to the result observed by Chinnaswamy and Hanna (1990). The addition of citric acid may have altered the pattern of macromolecular degradation as well as the viscoelastic (improvement of gel structure) properties.

Density, which generally is inversely proportional to expansion ratio, is another important characteristic of expanded materials. The bulk density of extrudate considers expansion in all directions. The bulk density values of extruded normal corn starch and samples containing added guar gum and/or citric acid are shown in Table 1, which range from 0.582 to 0.764 g/cm³. Bulk density increased with increase in guar gum concentration, as shown in Table 1. It may be that as guar gum concentration increases intermolecular interaction also increases, leading to a higher degree of entanglement, thereby limiting the extrudate expansion. This consequently resulted in intense rigidity of the extrudate. This same explanation could apply to cit-

ric acid treated samples as acidulants helps to improve the gel structure.

3.3. Effect of guar gum and citric acid on resistant starch formation

Fig. 2 shows the effect of different gum concentration, with or without citric acid, on resistant starch content during extrusion in a mixed system. A general trend was observed in which resistant starch content decreased with increase in concentration of starch–gum mixtures in water. During swelling of starch, water content tends to influence granule swelling and amylose leaching. Lower starch concentration facilitates swelling. In contrast, in higher starch concentration, limited amount of water would suppress swelling and amylose leaching, thus making amylose unavailable for recrystallization, an important aspect of resistant starch formation (Vasanthan & Bhatt, 1998).

Extrusion cooking is an important, versatile and very efficient food processing technology. Extrusion cooking of starchy material causes gelatinization of starch, among other physicochemical and functionality changes the material components undergo. Many investigators have demonstrated that retrogradation of gelatinized starch induces the formation of resistant starch and the retrogradation process is essential for resistant starch development (Berry, 1986; Eerlingen, Crombez, & Delcour, 1993; García-Alonso,

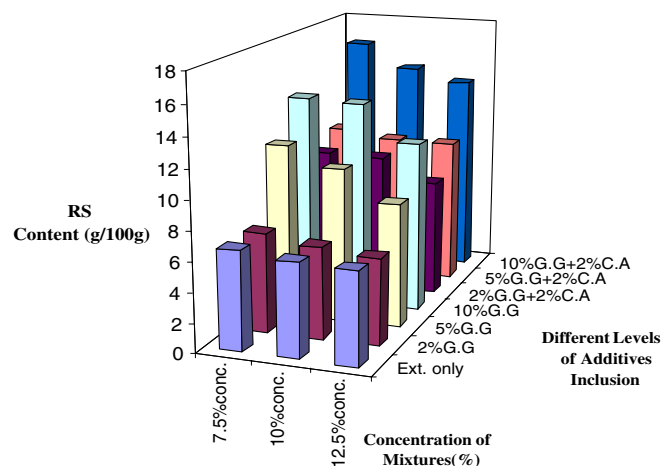


Fig. 2. Resistant starch content extruded guar gum–starch mixture with or without citric acid. G.G, guar gum, C.A, citric acid, Ext. only means extruded starch only; RS, resistant starch. Data were the means of three determinations.

Table 1
Physical properties of guar gum–starch extruded with or without citric acid

	Guar gum concentration (db)						
	Without citric acid				With citric acid		
	0	2%	5%	10%	2%	5%	10%
Expansion ratio ^a	8.42 ± 1.48	6.95 ± 1.12	6.02 ± 0.75	5.74 ± 0.39	5.79 ± 1.03	5.59 ± 0.67	5.18 ± 0.47
Bulk density (g/cm ³) ^b	0.582 ± 0.011	0.624 ± 0.008	0.669 ± 0.014	0.712 ± 0.006	0.695 ± 0.005	0.721 ± 0.009	0.764 ± 0.013

^{a,b} Data represented the means ± standard deviation ($n = 5$) of one representative experiment.

Jiménez-Escrig, Martín-Carrón, Bravo, & Saura-Calixto, 1999; Shamai et al., 2003). The literature on the effect of guar gum on starch retrogradation shows contrasting information. Some investigators (Davidou, Le Meste, Debever, & Bekaet, 1996; Lee et al., 2002) have reported that guar gum tends to retard retrogradation slightly. Funami et al. (2005a) reported that guar gum promoted corn starch retrogradation during the initial stage of storage and retarded retrogradation during longer storage. Rojas, Rosell, and de Barber (1999) reported that retrogradation was augmented by guar gum; however, our results further confirm that the addition of guar gum led to acceleration of retrogradation because the resistant starch content increased with increase of guar gum concentration. For example, when the concentration of guar gum–starch mixtures was 7.5%, the resistant starch contents were 6.71% for extruded starch only, 6.74–14.21% for guar gum ranging from 2% to 10% without citric acid, and 9.52–16.19% for guar gum ranging from 2% to 10% with citric acid, respectively. This probably could be related to the structure of the guar gum, which makes it a good thicker. Guar gum might interact with amylose and amylopectin with long exterior chains (amylose-like component) upon heating, leading to increase in the paste viscosity of the system. The high viscosity condition may have acted as a shield to the degraded macromolecule thereby limiting the activity of the enzyme. On cooling, guar gum increases the effective concentration of amylose and amylose-like components in the continuous phase, through their thermal thickening, leading to acceleration of retrogradation.

The addition of citric acid had a pronounced positive effect on resistant starch formation as shown in Fig. 2. Unlu and Faller (1998) also observed an increase in resistant starch content during extrusion of corn meal blended with high amylose maize starch in the presence of citric acid. During extrusion of the guar gum–starch mixture with citric acid, on the one hand, citric acid as an anhydride might react with corn starch, resulting in chemically modified starch (Xie & Liu, 2004). On the other hand, citric acid might hydrolyze starch polymers, to cause size reduction in amylose and amylopectin molecules. Meanwhile, the chain splitting of starch polymers took place during extrusion cooking, which was the result of shear (Della Valle, Kozlowski, Colonna, & Tated, 1989). As a result of this size reduction, smaller linear starch molecules might reassociate to increase the content of resistant starch.

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