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Effect of water-soluble and insoluble non-starch polysaccharides isolated from wheat flour on the rheological properties of wheat starch gel

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Abstract

Water-soluble (WSP) and insoluble non-starch polysaccharides (WIP) were isolated from wheat flour to evaluate the effects of WSP and WIP on starch gel properties. Isolated WSP and WIP were added to two types of isolated wheat starch with different amylose content at a concentration of 3% based on the dry weight of starch. 30% starch gels were prepared and stored at 5 °C for 1, 8, or 24 h. The dynamic viscoelasticity of 30% starch gels mixed with WSP and WIP was measured using parallel plate geometry, showing that WSP and WIP affected the elastic component of starch gels in opposite ways. Adding WIP increased the storage shear modulus (G') of starch gels, while adding WSP decreased G' and dramatically increased the loss tangent ($\tan\delta = G''/G'$).
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1. Introduction

Non-starch polysaccharide (NSP) is a minor component of wheat flour and located in the cell walls of wheat grain. NSP is assumed to interact with other components and influence the physical properties and end-product quality of wheat flour. NSP significantly influences the properties of dough and final baking performance due to its high water binding capacity and high viscosity (Cleemput, Roels, Van Oort, Grobet, & Delcour, 1993; Jelaca & Hlynka, 1972; Kim & D'Appolonia, 1977a; Michniewicz, Biliaderis, & Bushuk, 1991). NSP is classfied into water-soluble (WSP) and water-insoluble fraction (WIP) by solubility. The WIP/WSP ratio in NSP widely differs among wheat cultivars (Sasaki, Yasui, & Matsuki, 2000). WSP and WIP have a different chemical structure and function in the physical properties of wheat flour products. Water-soluble arabinoxylan, which is the main component of NSP in wheat flour, reportedly impacts positively on dough and bread quality, while the insoluble fraction impacts negatively (Courtin Gelders, & Delcour, 2001; Primo-Martín & Martínez-Anaya, 2003). Kim and D'Appolonia (1977a), however, showed that adding WSP did not affect loaf volume. A negative relationship between WSP content and dough properties has also been shown (Roels, Cleemput, Vandewalle, Nys, & Delcour, 1993). This disagreement may be due to differences in isolation method and NSP composition, and the functions of WSP and WIP remain to be clarified.

Starch is the most abundant component in wheat endosperm. Physical properties of starch, such as gelatinization, retrogradation, and gelation, strongly influence the quality of wheat flour products. Starch and hydrocolloid mixtures are often used to modify the texture of food products. Much work has focused on blending cereal starch and other polysaccharide molecules for their rheological behavior to clarify the interactions between starch and polysaccharide (Alloncle, Lefebvre, Llamas, & Doublier, 1989; Freitas et al., 2003; Gudmundsson, Eliasson, Bengtsson, & Aman, 1991; Rayment, Ross-Murphy, & Ellis, 1995; Tecante & Doublier, 1999; Tester & Sommerville, 2003). It is well known that hydrocolloid addition strongly influences the gelatinization, retrogradation and gelation of starch. Few studies, however, have dealt with the interaction between NSP and starch in the same wheat flour, although this interaction is important in the final properties of wheat flour products. We previously evaluated the influence

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of NSP isolated from wheat flour on the gelatinization and gelation of wheat starch and found that the WIP:WSP ratio in NSP influences the physical properties of starch and NSP mixtures (Sasaki, Yasui, & Mastuki, 2000). It has been shown that NSP of wheat flour plays an important role in the rheological response of wheat starch. In this study, WSP and WIP were separately isolated from the same wheat flour. The isolated WSP and WIP were respectively added to two types of isolated starch with different amylose content to evaluate WSP and WIP function in the rheological properties of concentrated starch gels. An evaluation of the effects on concentrated starch system provides useful information on food quality, because the starch concentration is usually high in wheat flour products. Dynamic oscillatory rheometer has been proved useful in monitoring the mechanical properties of starch gels (Biliaderis & Tonogai, 1991; Biliaderis & Zawistowski, 1990; Gudmundsson et al., 1991; Karim, Norziah, & Seow, 2000; Perez, Villareal, Juliano, & Biliaderis, 1993; Rosalina & Bhattacharya, 2002; Won, Choi, Lim, Cho, & Lim, 2000; Yuan & Thompson, 1998). We investigated the dynamic viscoelasticity of starch gels mixed with WSP and WIP to determine their functions in the rheological properties of wheat starch.

2. Materials and methods

2.1. Materials

Two wheat cultivars, Norin 61 and Norin 31 were used to isolate WSP and WIP. Starch was isolated from the low amylose line, Kanto 107, and the normal amylose cultivar, Norin 61. These cultivars were grown at the National Agriculture Research Center, Tsukuba, Japan. Wheat grains were tempered overnight to 14% moisture, then milled using a Quadrumat Junior test mill (Brabender, Duisburg, Germany) with a 70GG mesh sieve.

2.2. Starch isolation

Starch was isolated from wheat flour using the dough-ball method of Wolf (1964). After removing tailing starches, prime starch was washed with 0.1 M NaCl and freeze-dried. The amylose content of isolated starches was determined by the method of Gibson, Solah, and McCleary (1997) using an amylopectin/amylose assay kit (Megazyme International Ireland Ltd, Ireland).

2.3. WSP and WIP isolation

WSP and WIP were isolated from wheat flour by a modified method of Sasaki et al. (2000). Wheat flour was incubated in 0.05 M of MES-TRIS buffer (pH 8.2) with Termamyl 120L (Novo-Nordisk., Bagsraerd, Denmark) at 95–100 °C for 30 min with continuous shaking. After being cooled to 60 °C, the mixture was incubated with protease (P3910, Sigma, USA) at 60 °C for 30 min. After pH was adjusted to 4.0 by adding 1 N HCl, amyloglucosidase (A9913, Sigma, USA) was added and the mixture incubated at 60 °C for 30 min. After centrifugation at 17,880*g* for 15 min at 20 °C, the supernatant was decanted. The supernatant was used to isolate WSP and the precipitate to isolate WIP.

In WSP isolation, the supernatant was heated at 100 °C for 15 min to inactivate the enzyme. After centrifugation at 17,880*g* for 15 min at 20 °C, the precipitate was removed and 4 volumes of 95% ethanol (60 °C) added to the supernatant. WSP was precipitated at 25 °C for 60 min. After centrifugation at 17,880*g* for 15 min at 20 °C, the supernatant was decanted and the residue washed twice each with 78% ethanol and 95% ethanol followed by once with acetone. After being air-dried, the WSP fraction was freeze-dried.

In WIP isolation, the precipitate was heated with 0.05 M of MES-TRIS buffer (pH4.0) at 100 °C for 15 min to inactivate the enzyme. After centrifugation and decantation, distilled water was added to the precipitate and incubated at 60 °C for 30 min with continuous shaking. After centrifugation, the WIP fraction was washed with 1.5% SDS to remove the protein, and then washed three times in distilled water, two times in 78% ethanol, two times in 95% ethanol, and once in acetone. After being air-dried, the WIP fraction was freeze-dried.

2.4. NSP content and composition

NSP content in the isolated fraction was calculated from the sum of neutral sugars by gas chromatography (GLC). After trifluoroacetic acid hydrolysis (Olson, Gray, Mei-Chen, Betschart, & Turnlund, 1988) and derivatization (Blakeney, Harris, Henry, & Stone, 1983), alditol acetates were separated on a fused silica capillary column (DB-225, J and W Scientific, CA, USA). The column and detector were maintained at 230 °C. Protein content (N × 5.7) of each isolated fraction was determined using a FP-528 Nitrogen/Protein Determinator (LECO Co., MI, USA). α-Amylase activity in the isolated NSP and starch was determined by the method of McCleary and Sheehan (1987) using an α-amylase assay kit (Megazyme).

2.5. Swelling power and solubilized starch measurement

Swelling power and solubilized starch of the mixture were measured as detailed elsewhere (Sasaki et al., 2000). The WSP or WIP isolated fraction was added to the mixture of starch and water (0.1:4.9) to adjust the NSP concentration calculated from the sum of neutral sugars at 3% based on starch dry weight. The mixture was heated at 70 °C for 10 min followed by boiling for 10 min. After centrifugation, swelling power was determined as sediment weight (g/g), while the supernatant was used for measuring solubilized starch. After the supernatant was freeze-dried, total

carbohydrate and amylose content in the soluble fraction was determined using an amylopectin/amylose assay kit (Megazyme).

2.6. Rheological properties of starch gels

Isolated WSP or WIP was added to a 30% starch suspension to adjust the NSP concentration calculated from the sum of neutral sugars at 3% based on starch dry weight. The suspension was stirred continuously in a glass tube with a Teflon-lined screw cap at 500 rpm by a magnetic stirrer for 30 min at room temperature, and heated at 55 °C for 2.5–4.5 min with continuous stirring at 500 rpm until the suspension became thick enough to keep starch from settling. Pastes were transferred between two glass plates with a 1.0 mm spacer and sealed in airtight bags, which were heated at 100 °C for 15 min. After cooling at 25 °C for 5 min, starch gels were stored at 5 °C for 1, 8, or 24 h. The dynamic viscoelasticity of starch gel was measured using a rheometer (RheoStress RS75, Haake, Germany) with parallel plate (35 mm in diameter, gap 1.0 mm). A 35 mm diameter disk was cut from the center of the gel and transferred to the rheometer plate. After the upper plate was lowered onto the gel, silicone oil was applied to the exposed edge of samples to prevent evaporation of water during the experiment. Dynamic viscoelasticity was measured in a frequency range of 0.01–10 Hz at 25 °C and under constant stress (50 Pa). At this stress, all samples showed linear viscoelasticity in the preliminary stress sweep test.

2.7. Statistical analysis

All samples were analyzed in duplicate. Dynamic viscoelasticity was measured at least in triplicate. The general linear model (SAS Institute, Cary, NC) was used to analyze data. Analysis of variance was conducted using Tukey's studentized range test at 5% significance.

3. Results and discussion

3.1. Chemical analysis of isolated WSP and WIP

Table 1 shows the neutral sugar composition in WSP and WIP isolated from the cultivars, Norin 31 and Norin 61. The neutral sugar composition varied between WSP and WIP. The sum of arabinose and xylose represented 71–77% of total sugars in WSP and 45–53% in WIP, meaning that arabinoxylan is the main polysaccharide of both isolated WSP and WIP. A much higher proportion of galactose, the constituent of arabinogalactan, was found in WSP than WIP. A high proportion of glucose was observed in WIP and mannose content was similar in WSP and WIP. The arabinose to xylose ratio is a very important parameter for representing the degree of substitution in arabinoxylan. The ratio of arabinose to xylose ranged from 0.57 to 0.76 (Table 1). The removal of arabinose residues from arabinoxylan was reported to lower their solubility (Courtin & Delcour, 2002), but no distinct difference in the ratio between WSP and WIP was observed in these samples. A small difference in the arabinose to xylose ratio was found between Norin 31 and Norin 61, with Norin 61 showing higher ratio in both WSP and WIP than Norin 31. This means that the arabinoxylan of Norin 61 has a more branched structure in both WSP and WIP than that of Norin 31.

The sum of neutral sugars accounted for 68.3–75.0%, on a dry basis, of fraction isolated by enzymatic treatment. The protein content of the isolated fraction was 7.7–10.0% in WSP and 6.7–9.6% in WIP. A similar degree of protein contamination was seen in other isolation methods of WSP or WIP (Cleemput, Roels, Van Oort, Grobet, & Delcour, 1993; Faurot et al, 1995; Girhammar & Nair, 1992; Izydorczyk, Biliaderis, & Bushuk, 1991). After protein digestion by protease, the WIP fraction contained an appreciable amount of protein and showed a higher ratio of mannose in neutral sugars. WIP is thought to interact with other cell wall constituents such as proteins in the cell wall, whereas WSP is loosely bound at the cell wall surface (Courtin & Delcour, 2002). Gluppen, Marseille, Voragen, Hamer, and Pilnik (1989) indicated that the presence of intracellular protein inhibited the isolation of pure WIP. Tilley, Lookhart, Hoseney, and Mawhinney (1993) found that mannose covalently linked with glutenin subunits and the presence of mannose in the isolated glutenin was significant. This indicates that the high ratio of mannose in neutral sugars resulted from isolated WIP fraction contaminated with considerable protein. The step of dissolving protein by SDS solution significantly decreased both protein and mannose content in isolated WIP.

The amylose content of starches isolated from Norin 61 was 27.3% and that from Kanto 107 was 23.6%. The protein content of isolated starches was less than 0.5%. α-Amylase activity in isolated WSP, WIP, and starches was analyzed, since α-amylase activity strongly influences starch gel

Table 1
Neutral sugar composition in isolated WSP and WIP

Sample	Arabinose (%)	Xylose (%)	Mannose (%)	Galactose (%)	Glucose (%)	Ara/Xyl
Norin 31 WSP	27.8	43.8	10.7	8.9	8.9	0.64
Norin 61 WSP	33.4	43.8	10.0	8.3	4.5	0.76
Norin 31 WIP	19.4	34.0	8.8	1.4	36.4	0.57
Norin 61 WIP	19.3	25.7	11.0	1.9	42.2	0.75

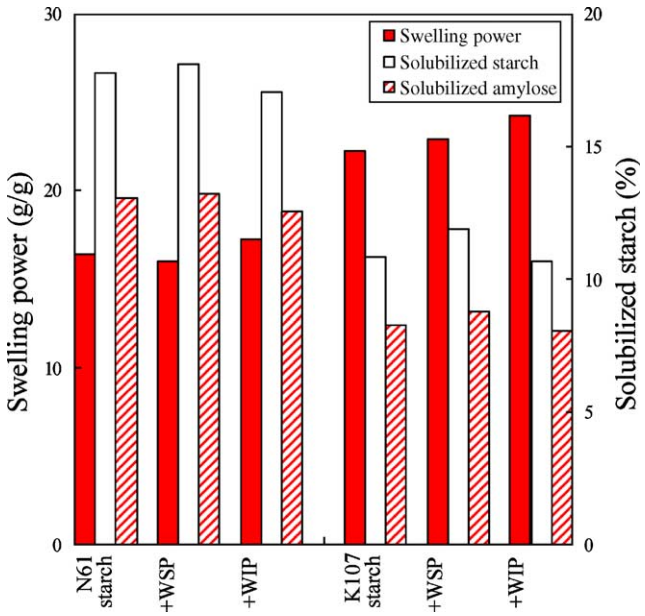

Fig. 1. Swelling power and solubilized starch for starch mixed with WSP or WIP.

properties. No samples showed high amylase activity in each isolated fraction.

3.2. Swelling power and solubilized starch

The swelling power of isolated starches and NSP mixtures was 16.4–17.2 g/g (Norin 61) and 22.3–24.2 g/g (Kanto 107), respectively (Fig. 1). The difference in swelling power among starch and starch mixed with WSP and WIP was smaller than the difference between Norin 61 and Kanto 107 starch. Starch swelling is a property of amylopectin, and amylose acts as a diluent (Tester & Morrison, 1990, 1992). The difference in amylose content had a strong impact on swelling power. When the swelling power of starch alone and of NSP mixtures is compared, adding WIP significantly increased swelling power, while no significant difference was observed in adding WSP. WIP and WSP contribute to water absorption of wheat flour, whereas WIP tends to absorb water and swell more strongly than WSP (Jelaca & Hlynca, 1971; Kim & D'Appolonia, 1977a). WIP is considered to significantly increase swelling power by only a 3% addition to starches due to its great water-holding capacity.

The solubilized starch content for starch mixed with WSP and WIP was 17.1–18.1% (Norin 61) and 10.6–11.9% (Kanto 107). Solubilized amylose content was 12.6–13.1% (Norin 61) and 8.1–8.8% (Kanto 107) (Fig. 1). Starch with lower amylose showed significantly lower solubilized starch and amylose content. Solubilized starch consisted mostly of amylose, which leads to higher solubilized starch content in starch with higher amylose content. For Norin 61 and Kanto 107 starches, starch mixed with WSP tended to show higher solubilized starch and starch mixed with WIP lower values than starch alone. The significant difference in the same starch was not found except in Norin 61 starch and the WIP mixture. No significant difference in solubilized amylose content was found, either, between starch alone and starch mixed with WSP or WIP.

3.3. Dynamic viscoelasticity of starch gels mixed with WSP and WIP

The dynamic viscoelasticity of 30% starch gels mixed with WSP and WIP at a concentration of 3% was compared to 30% starch gel without NSP to evaluate the effect of WSP and WIP on starch gel properties. Starch gels were prepared with starches isolated from the cultivars, Norin 61 and Kanto 107 and stored at 5 °C for 1, 8, or 24 h to determine the effects of isolated WSP and WIP on the retrogradation of starch gels. Fig. 2 shows the storage shear modulus (G') at a frequency of

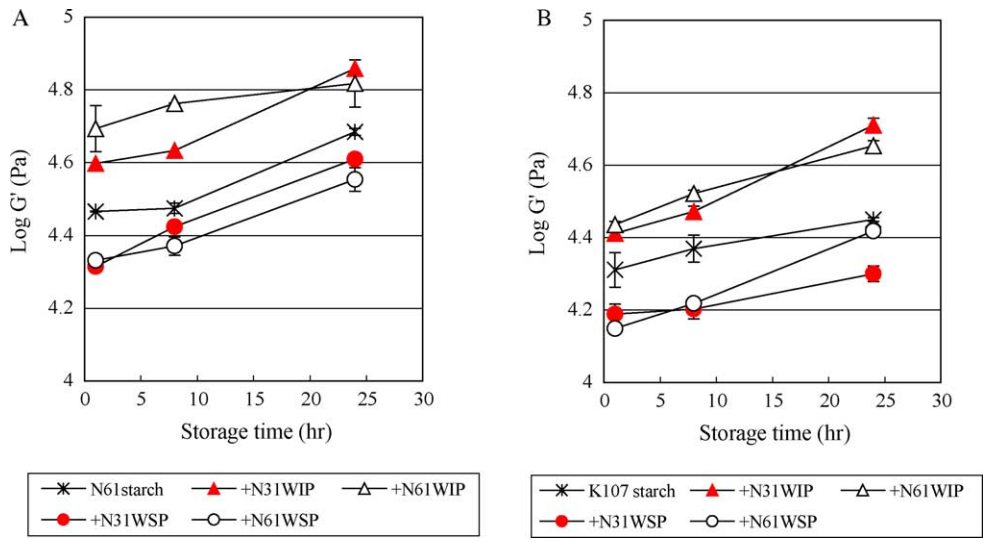

Fig. 2. Changes in storage shear modulus (G') at 1 Hz of 30% wheat starch gel mixed with WSP or WIP with storage at 5 ° C. A = Norin 61 starch; B = Kanto 107 starch.

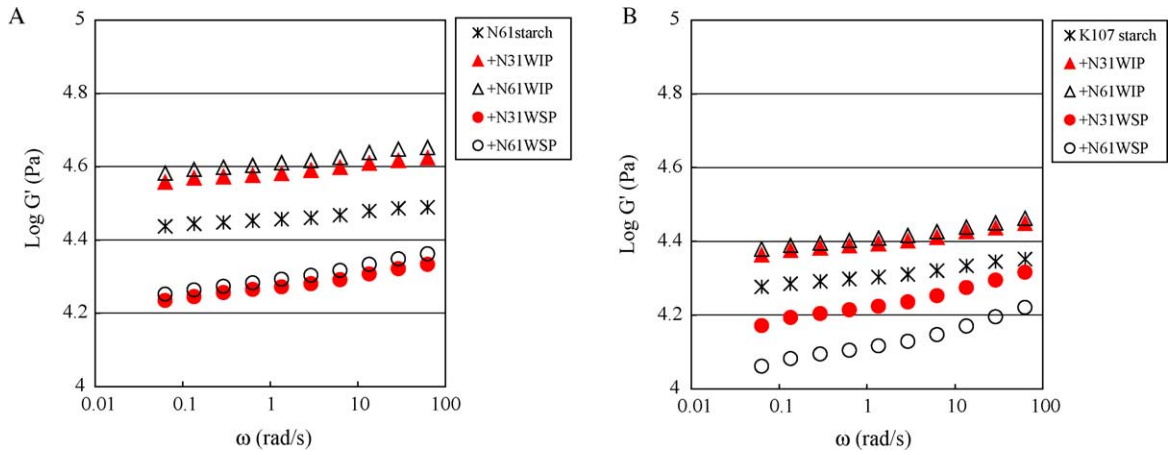

Fig. 3. Frequency dependence of storage shear modulus (G') of starch gels mixed with WSP or WIP stored at 5 °C for 1 h. A = Norin 61 starch; B = Kanto 107 starch.

1 Hz with the storage period at 5 °C. In comparison for different starches, starch gels prepared from the low amylose line, Kanto 107, showed significantly lower G' than Norin 61 throughout 24 h storage. Amylose restrains starch granule swelling and helps reduce loss in granular rigidity (Tsai, Li, and Lii, 1997). The rigidity of swollen starch granules is a major factor in determining the formation of gel, which suggests that a slight difference in amylose content contributes to the elastic component of starch gels (Parovuori, Manelius, Suortti, Bertoft, & Autio, 1997; Sasaki, Yasui, Matsuki, & Satake, 2002). For both Norin 61 and Kanto 107 starch, starch gel mixed with WSP showed lower G' than starch alone during storage. WSP had softening effect on starch gels. A significant difference ($P < 0.05$) was found between gels made from starch alone and starch mixed with WSP, when using Norin 61 starch gels stored for 8 h, and Kanto 107 starch gels stored for 1 and 24 h. In contrast, adding WIP to starch gels increased G' of starch gels throughout storage, suggesting that WIP strengthens the elasticity of starch gel. A significant difference ($P < 0.05$) was observed between starch alone and starch mixed with WIP, except for Norin 61 starch gels stored for 24 h. The influence of adding WSP and WIP fractions on G' of starch gels was similar between the two cultivars used to isolate WSP and WIP, i.e. Norin 31 and Norin 61. As shown in Fig. 2, G' of all starch gels continued to grow during 24 h at 5 °C. The increase in G' results from rearrangement involving amylose and amylopectin (Keetles, van Vilet, & Walstra, 1996). For Norin 61 starch, the rate of increasing G' was similar among samples judging from the slopes of lines (Fig. 2). For Kanto 107 starch, G' of starch gel mixed with WIP developed more rapidly than starch alone or with WSP. As stated above, starch mixed with WIP showed significantly higher swelling power than starch alone, indicating that WIP has a high capability to hold water around starch granules, accelerating the reassociation of starch molecules.

Figs. 3 and 4 showed the frequency dependence of G' for starch gels stored for 1 and 24 h. Starch gels mixed with WSP showed higher frequency dependence than starch alone or starch mixed with WIP. Starch alone and starch with WIP exhibited very low frequency dependence. The frequency dependence decreases with the formation of cross-links in a starch gel (Biliaderis & Tonogai, 1991) and the modulus of a rigid gel is frequency-independent,

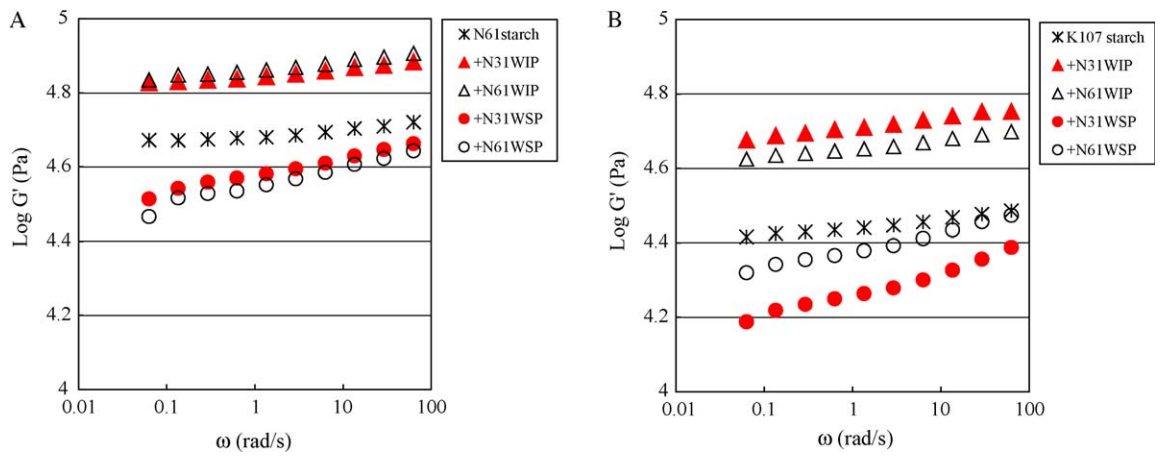

Fig. 4. Frequency dependence of storage shear modulus (G') of starch gels mixed with WSP or WIP stored at 5 °C for 24 h. A = Norin 61 starch; B = Kanto 107 starch.

Table 2
Loss shear modulus (G'') and loss tangent (tan δ) at 1 Hz of 30% wheat starch gel mixed with WSP or WIP stored at 5 °C for 1 and 24 h

Sample	G'' (kPa)		Tan δ	
	1 h	24 h	1 h	24 h
+Norin 61 starch				
Control	1.01 a	1.47 a	0.034 a	0.031 a
Norin 31 WSP	1.29 ab	2.93 b	0.063 c	0.072 b
Norin 61 WSP	1.39 ab	2.85 b	0.065 c	0.080 b
Norin 31 WIP	1.68 ab	2.61 b	0.042 ab	0.036 a
Norin 61 WIP	1.90 b	2.82 b	0.045 b	0.044 a
+Kanto 107 starch				
Control	0.96 a	1.31 a	0.047 a	0.047 a
Norin 31 WSP	1.36 b	2.23 b	0.095 b	0.112 b
Norin 61 WSP	1.47 b	2.59 b	0.105 b	0.099 b
Norin 31 WIP	1.32 b	2.44 b	0.051 a	0.048 a
Norin 61 WIP	1.61 b	2.06 b	0.060 a	0.046 a

Values followed by the same letter in the same column are not significantly different ($P < 0.05$).

suggesting that WSP obstructed the formation of cross-links between starch molecules and weakened the gel network. For Kanto 107 starch, the increase in G' due to adding WIP was pronounced after 24 h storage. Amylose content in starch greatly influences the properties of starch gels as stated above, and an obvious difference in G' was recognized between Kanto 107 and Norin 61 starch. Norin 61 starch mixed with WSP, however, showed a lower G' than Kanto 107 starch mixed with WIP (Figs. 3 and 4). Only 3% addition of WSP or WIP impacted strongly on starch gel properties and counteracted the effect of amylose content difference between Norin 61 and Kanto 107.

The addition of WSP and WIP had a similar effect on the loss shear modulus (G'') of starch gels (Table 2). Starch gels mixed with WSP and WIP showed higher G'' than gel with starch alone. A significant difference in G'' was observed between starch alone and starch with WSP or WIP for Norin 61 starch gels stored for 24 h, and Kanto 107 starch gels stored for 1 and 24 h. G'' of most starch gels continued to increase during 24 h storage at 5 °C as well as G'. For starch gel stored for 24 h, Kanto 107 starch showed a slightly lower G'' than Norin 61 starch. However, the difference in amylose content of starch between Norin 61 and Kanto 107 did not considerably affect the value of G''. The addition of WSP and WIP at 3% represented a pronounced contribution to increasing G''. The loss tangent ($\tan\delta = G''/G$) showed no marked change during storage unlike G' and G''. Tan δ of starch gel with WIP was similar to that with starch alone, while starch gel with WSP showed significantly higher tan δ than starch alone (Table 2). The variation in tan δ among samples reflects differences in the relative contribution of elastic and viscous components in starch gels. Low tan δ indicates that the elastic modulus predominates over the viscous modulus, meaning strong gel. The addition of WSP to starch decreased the elastic component and increased the viscous component of starch gel, resulting in an extremely high tan δ. WSP immobilizes water by hydrogen bonds and forms viscous solutions or gel (Wang, Hamer, Vliet, & Oudgenoeg, 2002). The pronounced increase of tan δ may reflect the physical properties of WSP itself. According to Alloncle and Doublier (1991), the reinforcement of the elastic character of the system is expressed by the higher G' and G'', and the lower G''/G'. The addition of WIP to starch increased G' and G'' of starch gel, while it had no effect on the value of tan δ, suggesting that WIP equally contributes to the increase of both elastic and viscous components in mixture gel. For two types of wheat starch, Kanto 107 starch gel with lower amylose content showed higher tan δ than Norin 61 starch. This indicates that amylose content contributes greatly to the elastic properties rather than the viscous properties, resulting in the lower tan δ of starch gel with higher amylose content.

Starch gels consist of swollen granules embedded in a continuous phases (Biliaderis & Izydorczyk, 1992). These swollen granules are mainly composed of amylopectin, while the continuous phase consists mainly of amylose. When different polymers are mixed, chemically dissimilar polymers tend to be incompatible for thermodynamic reasons. Phase separation between amylose and added hydrocolloids occurs in the continuous phase due to thermodynamic incompatibility (Alloncle & Doublier, 1991; Kulicke, Eidam, Kath, Kix, & Kull, 1996). Phase separation resulting from incompatibility between unlike polymers influences the firmness of the mixture gel. Yoshimura, Takaya, and Nishinari (1996) indicated that adding hydrocolloids increased the effective starch concentration by immobilizing water molecules, leading to a more stable structure of the mixture gel. Alloncle and Doublier (1991) demonstrated that the more rapid initial increase in starch gel firmness was induced by incompatibility between amylose and hydrocolloids. Added WSP or WIP to starch exists in the continuous phase of starch gels and their effects on the rheological properties of starch gel are considered largely due to phase separation. The initial development in the firmness of starch gel is dominated by the formation of a gel network from leached-out amylose, and long-term changes in firmness are attributed to the recrystallization of amylopectin (Gudmundsson, 1994; Orford, Ring, Carroll, Miles, & Morris, 1987). The effects of WSP weakening starch gel and WIP increasing the rigidity of starch gel were found throughout storage, indicating that WSP and WIP influence gel formation of leached-out amylose and the recrystallization of amylopectin. Mixing of dissimilar macromolecules causes phase separation and influences the water partition between the two phases. Effective concentration after phase separation is higher than the initial concentration of the two polymer constituents (Kasapis, 1995). In starch and polysaccharide mixtures, added polysaccharide are located within the continuous phase and causes an increase in its effective concentration of amylose in the continuous phase due to a reduction

of accessible volume, resulting in a higher viscoelasticity (Sudhakar, Singhal, & Kulkarni, 1996; Tecante & Doublier, 2002). Alloncle and Doublier (1991) suggested that the continuous phase containing added polysaccharide plays an increasing role in the viscoelastsic behavior of the composite due to the thickening properties of polysaccharides. Starches mixed with WIP had significantly higher swelling power than starch alone as stated above, suggesting that WIP immobilized water molecules and reduced the water availability of starch. Starch granule swelling during gelatinization furthermore increases the hydrocolloid concentration within the continuous phase. This implies that the added WIP was concentrated within the continuous phase during gelatinization and increased the effective concentration of amylose in the continuous phase, resulting in a higher viscoelasticity of starch gel mixed with WIP than starch alone. Moreover, WIP is thought to accelerate the formation of intermolecular covalent bonds between inter-amylopectin, or amylopectin and amylose by immobilizing water molecules around starch molecules. A strong interaction between gel matrix and the dispersed particles results in an increase of storage modulus with increasing volume fraction of the disperse particles (Van Vilet, 1988). In a concentrated system, as in this study, the continuous phase was considered to be suppressed by the tightly packed swollen granules, suggesting that the vicinity of starch molecules and WIP may influence the interaction between two components. In contrast, the measurement of dynamic viscoelasticity showed that WSP inhibited the formation of a gel network. If the main effect of added WSP is to increase the effective concentration in the continuous phase, adding WSP should increase the storage shear modulus of starch gel as WIP did. Alloncle et al. (1989) indicated that the depression in the final gel properties was due to incompatibility between the other polysaccharide and amylose in the continuous phase. The incompatibility between two dissimilar polymers can enrich the respective component in separate phases (Piculell, Bergfeldt, & Nilsson, 1995). The effect of WSP was considered to be attributed to incompatibility phenomena by thermodynamic reasons between amylose and WSP in the continuous phase and the phase enriched with WSP may weaken a gel network in the mixture. Kim and D'Appolonia (1977b) stated that NSP could reduce the extent of starch gel retrogradation by forming hydrogen bonds with amylopectin, which prevents hydrogen bond formation between amylose and amylopectin or inter-amylopectin. WSP retarded the reassociation of amylose and amylopectin, even though WSP presumably reduced the availability of water for starch due to its water-holding capacity. Added WSP may have interacted with starch molecules and alter the kinetics of network formation in starch gels. The measurement of dynamic viscoelasticity demonstrated the distinction between WSP and WIP functions in the rheological properties of concentrated starch gels.

4. Conclusion

The investigation showed the effects of WSP and WIP isolated from wheat flour on starch gel properties. WSP and WIP affected the elastic element of starch gel significantly. Adding WIP increased the elastic and viscous component of starch gels, implying that WIP has great capability to hold water and increases the starch concentration in the continuous phase, resulting in increased starch molecule reassociation. The addition of WIP to starch increased starch gel rigidity. In contrast to WIP, the addition of WSP decreased the elastic component of starch gels and markedly increased the loss tangent ($\tan\delta = G''/G'$). Although both WIP and WSP presumably increase the starch concentration in the continuous phase, WSP had a softening effect on starch gels and depressed the formation of a gel network. The polymer incompatibility between amylose and WSP in the continuous phase was considered to decrease the rigidity of mixture gel compared to the gel of starch alone and with WIP. Adding WSP or WIP showed the expectation to change starch gel properties significantly, even if the amount added was small.

References

Alloncle, M., & Doublier, J. L. (1991). Viscoelastic properties of maize starch/hydrocolloid pastes and gels. *Food Hydrocolloids*, *5*, 455–467.

Alloncle, M., Lefebvre, J., Llamas, G., & Doublier, J. L. (1989). A rheological characterization of cereal starch–galactomannan mixtures. *Cereal Chemistry*, *66*, 90–93.

Biliaderis, C. G., & Izydorczyk, M. S. (1992). Observation on retrogradation of starch polymers in the presence of wheat and rye arabinoxylans. In G. O. Phillips, P. A. Williams, & D. J. Wedlock (Eds.), (*Vol. 6*) (pp. 227–230). *Gums and stabilisers for the food industry*, Oxford: IRL Press.

Biliaderis, C. G., & Tonogai, J. R. (1991). Influence of lipids on the thermal and mechanical properties of concentrated starch gels. *Journal of Agriculture and Food Chemistry*, *39*, 833–840.

Biliaderis, C. G., & Zawistowski, J. (1990). Viscoelastic behavior of aging starch gels: effects of concentration, temperature, and starch hydrolysates on network properties. *Cereal Chemistry*, *67*, 240–246.

Blakeney, A. B., Harris, P. J., Henry, R. J., & Stone, B. A. (1983). A simple and rapid preparation of alditol acetates for monosaccharide analysis. *Carbohydrate Research*, *113*, 291–299.

Cleemput, G., Roels, S. P., Van Oort, M., Grobet, P. J., & Delcour, J. A. (1993). Heterogeneity in the structure of water-soluble arabinoxylans in European wheat flours of variable bread-making quality. *Cereal Chemistry*, *70*, 324–329.

Courtin, C. M., & Delcour, J. A. (2002). Arabinoxylans and endoxylanases in wheat flour bread-making. *Journal of Cereal Science*, *35*, 225–243.

Courtin, C. M., Gelders, G. G., & Delcour, J. A. (2001). Use of two endoxylanases with different substrate selectivity fro understanding arabinoxylan functionality in wheat flour breadmaking. *Cereal Chemistry*, *78*, 564–571.

Faurot, A. L., Saulnier, L., Bérot, S., Popineau, Y., Petit, M. D., Rouau, X., & Thibault, J. F. (1995). Large scale isolation of water-soluble and water-insoluble pentosans from wheat flour. *Lebensmittel-Wissenschaft und Technologie*, *28*, 436–441.

Freitas, R. A., Gorin, P. A. J., Neves, M.-R., & Sierakowski, M.-R. (2003). A rheological description of mixtures of galactoxyloglucan with high amylose and waxy corn starches. *Carbohydrate Polymers*, *51*, 25–32.

Gibson, T. S., Solah, V. A., & MacCleary, B. V. (1997). A procedure to measure amylose in cereal starches and flours with concanavalin A. *Journal of Cereal Science*, *25*, 11–119.

Girhammar, U., & Nair, B. M. (1992). Isolation, separation and characterization of water soluble non-starch polysaccharides from wheat and rye. *Food Hydrocolloids*, *6*, 285–299.

Gruppen, H., Marseille, J. P., Voragen, A. G. J., Hamer, R. J., & Pilnik, W. (1989). Mild isolation of water-insoluble cell wall material from wheat flour: composition of fractions obtained with emphasis on non-starch polysaccharides. *Journal of Cereal Science*, *9*, 247–260.

Gudmundsson, M. (1994). Retrogradation of starch and the role of its components. *Thermochimica Acta*, *246*, 329–341.

Gudmundsson, M., Eliasson, A. C., Bengtsson, S., & Aman, P. (1991). The effects of water soluble arabinoxylan on gelatinization and retrogradation of starch. *Starch*, *43*, 5–10.

Izydorczyk, M., Biliaderis, C. G., & Bushuk, W. (1991). Comparison of the structure and composition of water-soluble pentosans from different wheat varieties. *Cereal Chemistry*, *68*, 139–144.

Jelaca, S. L., & Hlynka, I. (1971). Water binding capacity of wheat flour crude pentosans and their relation to mixing characteristics of dough. *Cereal Chemistry*, *48*, 211–222.

Jelaca, S. L., & Hlynka, I. (1972). Effect of wheat-flour pentosans in dough, gluten, and bread. *Cereal Chemistry*, *49*, 489–495.

Karim, A. A., Norziah, M. H., & Seow, C. C. (2000). Methods for the study of starch retrogradation. *Food Chemistry*, *71*, 9–36.

Kasapis, S. (1995). Phase separation in hydrocolloid gels. In S. E. Harding, S. E. Hill, & J. R. Mitchell (Eds.), *Biopolymer mixtures* (pp. 193–224). Nottingham, UK: Nottingham University Press.

Keetles, C. J. A. M., van Vilet, T., & Walstra, P. (1996). Gelation and retrogradation of concentrated starch systems: 1. Gelation. *Food Hydrocolloids*, *10*, 343–353.

Kim, S. K., & D'Appolonia, B. L. (1977a). Bread staling studies. III. Effect of pentosans on dough, bread, and bread staling rate. *Cereal Chemistry*, *54*, 225–229.

Kim, S. K., & D'Appolonia, B. L. (1977b). Effect of pentosans on the retrogradation of wheat starch gels. *Cereal Chemistry*, *54*, 150–160.

Kulicke, W. M., Eidam, D., Kath, F., Kix, M., & Kull, A. H. (1996). Hydrocolloids and rheology: regulation of viscoelastic characteristics of waxy rice starch in mixtures with galactomannans. *Starch*, *48*, 105–114.

McCleary, B. V., & Sheehan, H. (1987). Measurement of cereal α-amylase: a new assay procedure. *Journal of Cereal Science*, *6*, 237–251.

Michniewicz, C. G., Biliaderis, C. G., & Bushuk, W. (1991). Effect of added pentosans on some physical and technological characteristics of dough and gluten. *Cereal Chemistry*, *68*, 252–258.

Olson, A. C., Gray, G. M., Mei-Chen, C., Betschart, A., & Turnlund, J. R. (1988). Monosaccharides produced by acid hydrolysis of selected foods, dietary fibers, and fecal residues from white and whole wheat bread consumed by humans. *Journal of Agriculture and Food Chemistry*, *36*, 300–304.

Orford, P. D., Ring, S. G., Carroll, V., Miles, M. J., & Morris, V. J. (1987). The effect of concentration and botanical source on the gelation and retrogradation of starch. *Journal of Science of Food and Agriculture*, *39*, 169–177.

Parovuori, P., Manelius, R., Suortti, T., Bertoft, E., & Autio, K. (1997). Effects of enzymically modified amylopectin on rheological properties of amylose–amylopectin mixed gels. *Food Hydrocolloids*, *11*, 471–477.

Perez, C. M., Villareal, C. P., Juliano, B. O., & Biliaderis, C. G. (1993). Amylopectin-staling of cooked nonwaxy milled rices and starch gels. *Cereal Chemistry*, *70*, 567–571.

Piculell, L., Bergfeldt, K., & Nilsson, S. (1995). Factors determining phase behaviour of multi component polymer systems. In S. E. Harding, S. E. Hill, & J. R. Mitchell (Eds.), *Biopolymer mixtures* (pp. 13–35). Nottingham, UK: Nottingham University Press.

Primo-Martín, C., & Martínez-Anaya, M. A. (2003). Influence of pentosanase and oxidases on water-extractable pentosans during straight breadmaking process. *Food chemistry and Toxicology*, *68*, 31–41.

Rayment, P., Ross-Murphy, S. B., & Ellis, P. R. (1995). Rheological properties of guar galactomannan and rice starch mixtures I. Steady shear measurements. *Carbohydrate Polymers*, *28*, 121–130.

Roels, S. P., Cleemput, G., Vandewalle, M., Nys, M., & Delcour, J. A. (1993). Bread volume potential of variable quality flours with constant protein level as determined by factors governing mixing time and baking absorption levels. *Cereal Chemistry*, *70*, 318–323.

Rosalina, I., & Bhattacharya, M. (2002). Dynamic rheological measurements and analysis of starch gels. *Carbohydrate Polymers*, *48*, 191–202.

Sasaki, T., Yasui, T., & Mastuki, J. (2000). Influence of non-starch polysaccharides isolated from wheat flour on the gelatinization and gelation of wheat starches. *Food Hydrocolloids*, *14*, 295–303.

Sasaki, T., Yasui, T., Matsuki, J., & Satake, T. (2002). Comparison of physical properties of wheat starch gels with different amylose content. *Cereal Chemistry*, *79*, 861–866.

Sudhakar, V., Singhal, R. S., & Kulkarni, P. R. (1996). Starch–galactomannan interactions: functionality and rheological aspects. *Food Chemistry*, *55*, 259–264.

Tecante, A., & Doublier, J. L. (1999). Steady flow and viscoelastic behavior of crosslinked waxy corn starh-κ-carrageenan pastes and gels. *Carbohydrate Polymers*, *40*, 221–231.

Tecante, A., & Doublier, J. L. (2002). Rheological investigation of the interaction between amylose and κ-carrageenan. *Carbohydrate Polymers*, *49*, 177–183.

Tester, R. F., & Morrison, W. R. (1990). Swelling and gelatinization of cereal starches. I. Effects of amylopectin, amylose, and lipids. *Cereal Chem*, *67*, 551–557.

Tester, R. F., & Morrison, W. R. (1992). Swelling and gelatinization of cereal starches. III. Some properties of waxy and normal non-waxy barley starches. *Cereal Chem*, *69*, 654–658.

Tester, R. F., & Sommerville, M. D. (2003). The effects of non-starch polysaccharides on the extent of gelatinization, swelling and α-amylase hydrolysis of maize and wheat starches. *Food Hydrocolloids*, *17*, 41–54.

Tilley, K. A., Lookhart, G. L., Hoseney, R. C., & Mawhinney, T. P. (1993). Evidence for glycosylation on the high molecular weight glutenin subunits 2, 7, 8, and 12 from Chinese Spring and TAM 105 wheats. *Cereal Chemistry*, *70*, 602–606.

Tsai, M. L., Li, C. F., & Lii, C. Y. (1997). Effects of granular structure on the pasting behaviors of starches. *Cereal Chemistry*, *74*, 750–757.

Van Vilet, T. (1988). Rheological properties of filled gels. Influence of filler matrix interaction. *Colloid and Polymer Science*, *266*, 518–524.

Wang, M., Hamer, R. J., Vliet, T. V., & Oudgenoeg, G. (2002). Interaction of water extractable pentosans with gluten protein: effect on dough properties and gluten quality. *Journal of Cereal Science*, *36*, 25–37.

Wolf, M. J. (1964). *Wheat starch isolation. Methods in carbohydrate chemistry IV*. New York: Academic Press, pp. 6–9.

Won, S.-Y., Choi, W. S., Lim, H. S., Cho, K.-Y., & Lim, S.-T. (2000). Viscoelasticity of cowpea starch gels. *Cereal Chemistry*, *77*, 309–314.

Yoshimura, M., Takaya, T., & Nishinari, K. (1996). Effects of konjac-glucomannan on the gelatinization and retrogradation of corn starch as determined by rheology and differential scanning calorimetry. *Journal of Agriculture and Food Chemistry*, *44*, 2970–2976.

Yuan, R. C., & Thompson, D. B. (1998). Rheological and thermal properties of aged starch pastes from three waxy maize genotypes. *Cereal Chemistry*, *75*, 117–123.