

Dough and Baking Properties of High-Amylose and Waxy Wheat Flours

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ABSTRACT

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The dough properties and baking qualities of a novel high-amylose wheat flour (HAWF) and a waxy wheat flour (WWF) (both *Triticum aestivum* L.) were investigated by comparing them with common wheat flours. HAWF and WWF had more dietary fiber than Chinese Spring flour (CSF), a nonwaxy wheat flour. Also, HAWF contained larger amounts of lipids and proteins than WWF and CSF. There were significant differences in the amylose and amylopectin contents among all samples tested. Farinograph data showed water absorptions of HAWF and WWF were significantly higher than that of CSF, and both flours showed poorer flour qualities than CSF. The dough of WWF was weaker and less stable than that of CSF, whereas HAWF produced a harder and more viscous dough than CSF. Differential scanning calorimetry data showed that starch in HAWF dough gelatinized at a lower temperature in the baking process

than the starches in doughs of WWF and CSF. The starch in a WWF suspension had a larger enthalpy of gelatinization than those in HAWF and CSF suspensions. Amylograph data showed that the WWF starch gelatinized faster and had a higher viscosity than that in CSF. The loaves made from WWF and CSF were significantly larger than the loaves made from HAWF. However, the appearance of bread baked with WWF and HAWF was inferior to the appearance of bread baked with CSF. Bread made with WWF became softer than the bread made with CSF after storage, and reheating was more effective in refreshing WWF bread than CSF bread. Moreover, clear differences in dough and bread samples were revealed by scanning electron microscopy. These differences might have some effect on dough and baking qualities.

Wheat (*Triticum aestivum* L.) starch normally contains ≈20–30% amylose, and the rest is amylopectin. Recently, wheat grains containing various ratios of amylose and amylopectin in wheat starch have been developed by genetic recombination. In wheat, at least four kinds of proteins (waxy protein and three starch granule proteins SGP-1, -2, -3) tightly bind to starch granules and are responsible for starch synthesis (Yamamori et al 2000). The waxy protein is a granule-bound starch synthase (GBSS) and is responsible for amylose production (Yamamori et al 2000). Recently, waxy (amylose-free, glutinous) wheats have been produced in Japan and elsewhere (Nakamura et al 1995a, Kiribuchi-Otobe et al 1997, Yasui et al 1997). The characteristics of waxy type wheat flours have been reported (Miura and Sugawara 1996; Fujita et al 1998; Araki et al 1999, 2000; Yamamori and Quynh 2000). The starch granule protein SGP-1 is responsible for amylopectin synthesis (Yamamori et al 2000). Therefore, if the wheat grain genetically lacks SGP-1, the wheat can produce a novel high-amylose starch and be used to expand variation in wheat starch.

Amylose content plays an important role in the quality of wheat flour because it can affect the texture, stability, and viscosity of processed foods. Furthermore, the amylose-to-amylopectin ratio of wheat starch is an important factor in the production of various kinds of wheat-related processed foods. The white salted noodle *udon* is a popular food in Japan. Most Japanese prefer *udon* with a glutinous (sticky) texture that can be achieved by lowering the amylose content. Australian wheat flours, which have a lower amylose content (and are less brown) than Japanese wheat flours, are considered suitable for *udon* noodles. Australian Standard White (ASW) flour is considered especially suitable for this purpose (Oda et al 1980; Taira et al 1989; Toyokawa et al 1989). The quality of domestic wheat flour is much lower than that of imported wheat flours and, consequently, it is used only for noodle-making in Japan. Domestic wheat flour has been used recently for

a limited number of other processed foods. However, to be able to use it for a greater number of processed foods, much research is needed to understand the physicochemical properties of its starch. The purpose of the present study was to test the practical applications, especially for breadmaking, of novel wheat flours such as waxy (WWF) and high-amylose wheat flours (HAWF). In particular, we focused on the dough and baking properties of WWF and HAWF and compare them with those of common wheat flours.

MATERIALS AND METHODS

Materials

For the normal amylose content sample, nonwaxy wheat flour Chinese Spring (CSF) was test-milled from Okumoto Flour Milling, Osaka. High-amylose wheat was bred from Kanto 79/Turkey 116 F2 // Chousen 57 (Yamamori et al 2000). The variant high-amylose progeny F5 and F6 with no SGP-1 were selected, and F5 and F6 seeds were harvested at Tsukuba in 1999 and at Morioka in 2000, respectively. In contrast, waxy wheat was selected from the F2 progeny of CS (Kanto 107 7A)/CS (Kanto 107 4A) // CS (Bai Huo 7 D) and backcrossed twice to the normal CS (Miura et al 1999). This line was confirmed to be nearly isogenic to CS with respect to agronomic performance and chain-length distribution in amylopectin structure (Miura et al 2002). These high-amylose and waxy wheats, HAWF and WWF, respectively, were test-milled by Okumoto Flour Milling, Osaka. Chemicals were reagent-grade.

Wheat Flour

Flour characteristics were determined using standard methods: lipid (Approved Method 30-10, AACC 2000); dietary fiber (Nakamura et al 1995b); moisture content of breadcrumbs (Approved Method 44-15A). Sodium (Na) content of the flour sample (mg/g) was measured using atomic absorption spectrometry.

The breadmaking formula and procedures were performed with a slightly modified Approved Method 10-10B (AACC 2000). Flour (50 g), sodium chloride (0.75 g), sucrose (3 g), dry baker's yeast (1.00 g, Asahi Kasei Co., Tokyo), and the appropriate amounts of water were mixed for 15 min using an automatic home baker as described previously (Morita et al 1999). The optimum amount of water for the flour was determined from the farinograph water absorption ratio at 500 BU (Approved Method 54-21). The mixed dough fermented for 60 min in the cabinet at a constant temperature of 30°C and 85% rh. After fermentation, the punched and fermentation at 30°C for 30 min was repeated. Subsequently, the dough was divided into three pieces (15 g), rounded, and molded (mechanical molder SM-230, Baker's Production Co., Osaka), and

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TABLE I
General Analysis (%) of Three Wheat Flours^{a-c}

| Sample | Characteristics (%) | | | | | | | | | | |
|--------|---------------------|-------|-------|------------|---------|-------|-------|------|-------|---------------|----------------------|
| | AM | AP | Water | Wet Gluten | Protein | Ash | Lipid | DF | L | Na (mg/100 g) | <i>a_w</i> |
| CSF | 24.9b | 75.1b | 12.5b | 46.9b | 14.9a | 0.51a | 0.8a | 2.2a | 95.0b | 0.65a | 0.989b |
| HAWF | 37.5c | 62.5a | 11.3a | 43.4a | 17.5c | 1.08c | 1.4b | 6.7c | 90.8a | 0.65a | 0.984a |
| WWF | 2.3a | 97.7c | 13.0c | 51.3c | 16.0b | 0.54b | 1.0a | 3.3b | 95.2b | 0.70a | 0.983a |

^a CSF, test-milled soft-type wheat from Chinese Spring; HAWF, high-amylose wheat flour; WWF, waxy wheat flour.

^b AM, amylose content measured by colorimetry; AP, amylopectin; DF, dietary fiber; L, lightness value; *a_w*, water activity.

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

TABLE II
Differential Scanning Calorimetry (DSC) Data of Three Wheat Doughs^{a-c}

| Sample | 1st Peak | | | | 2nd Peak | | | | 3rd Peak | | | |
|--------|-----------------------|-----------------------|-----------------------|--------------|-----------------------|-----------------------|-----------------------|--------------|-----------------------|-----------------------|-----------------------|--------------|
| | <i>T_{i1}</i> | <i>T_{p1}</i> | <i>T_{r1}</i> | ΔH_1 | <i>T_{i2}</i> | <i>T_{p2}</i> | <i>T_{r2}</i> | ΔH_2 | <i>T_{i3}</i> | <i>T_{p3}</i> | <i>T_{r3}</i> | ΔH_3 |
| CSF | 53.4b | 63.0b | 67.3b | 0.67a | 73.9 | 83.0 | 91.6 | 0.62 | 98.4b | 112.1b | 120.8b | 1.28b |
| HAWF | 44.8a | 53.1a | 61.1a | 2.13b | ...d | ... | ... | ... | 88.9a | 103.8a | 110.6a | 2.21c |
| WWF | 58.3c | 75.1c | 86.1c | 5.12c | ... | ... | ... | ... | 95.6b | 110.1b | 119.8b | 0.61a |

^a CSF, test-milled soft-type wheat from Chinese Spring; HAWF, high-amylose wheat flour; WWF, waxy wheat flour.

^b *T_i*, *T_p*, and *T_r* = initial, peak, and recovery temperatures (°C); ΔH = enthalpy (J/g of dough).

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

^d No peaks observed.

TABLE III
Viscoelasticity of Doughs Containing Three Wheat Flours^{a-c}

| Sample | <i>g</i> | γ | η |
|--------|----------|----------|--------|
| CSF | 1.03a | 2.81b | 1.46a |
| HAWF | 2.19b | 5.24b | 2.99b |
| WWF | 0.89a | 2.37a | 1.15a |

^a Compression stress (*g*), modulus of elasticity (γ), and viscosity coefficient (η).

^b CSF, test-milled soft-type wheat from Chinese Spring; HAWF, high-amylose wheat flour; WWF, waxy wheat flour.

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

TABLE IV
Summary of Amylograph Data of Three Wheat Doughs^{a-c}

| Sample | GT | MVT | MV |
|--------|-------|-------|------|
| CSF | 57.7c | 90.3b | 481b |
| HAWF | 45.3a | 93.0c | 308a |
| WWF | 57.0b | 66.1a | 636c |

^a GT, gelatinization (°C); MVT, maximum viscosity (°C); MV, maximum viscosity (BU).

^b CSF, test-milled soft-type wheat from Chinese Spring; HAWF, high-amylose wheat flour; WWF, waxy wheat flour.

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

placed in a baking pan. The dough was subjected to a final proof in the cabinet, maintaining a constant temperature of 38°C and 90% rh, followed by baking at 200°C for 15 min. After baking, the loaves were removed and weighed immediately, and their volumes were measured by rapeseed displacement (Morita et al 1998). The loaves were sliced in half for visual examination.

The optimum amount of water for a flour sample was determined by farinograph mixing at 500 BU. The mixed dough was used for the subsequent tests.

Wheat Dough

Water activity (*a_w*) of dough was determined as described by Czuchajowska and Pomeranz (1989).

Farinograph data (arrival, development, and stability times) were obtained using Approved Method 54-21 with a 50-g stainless steel bowl. Mixing was done at the standard speed of 63 rpm at 30°C. The effect of amylase activity on the viscosity of flour samples was determined using an amylograph at 30–95°C at a rate of 1.5°C/min according to Approved Method 22-12. Viscoelastic properties of the dough after mixing in the farinograph were measured with a rheometer (Rheotech Co., Tokyo) (Zhang et al 1992). The plunger for viscoelastic measurement was 1 cm in diameter. The mixed dough sample was placed in a plastic vessel (2.5 cm, i.d. × 2.5 cm). After standing for 10 min, compression stress, modulus of elasticity, and viscosity coefficient were measured at 30°C. The speed of the plunger penetration into the dough sample was 30 cm/min and the depth was controlled at 1 cm. Data were processed using a computer program (Rheosoft TR-06, Rheotech Co., Tokyo).

Differential scanning calorimetry (DSC) was performed with a Shimadzu DSC instrument (model DSC-50, Tokyo) as reported previously (Morita et al 1998). An empty aluminum pan was used

for a reference. The heat of gelatinization (ΔH), initial temperature (*T_i*), peak temperature (*T_p*), and recovery temperature (*T_r*) were used to characterize the thermal transition of starch in the dough.

The bread and dough samples prepared with various flours were observed by scanning electron microscopy (SEM) (Hitachi model S-800) as reported previously (Morita et al 1998).

Moisture content of breadcrumbs after storage, with or without heating, was determined by Approved Method 44-15A (AACC 2000).

Staleness of breadcrumbs, compression stress, failure strength, and Young's modulus were determined with the rheometer using breadcrumbs of 2 × 2 × 1 cm³. The plunger size was 1 cm in diameter, and the compression depth was controlled at 3 mm.

The firmness of reheated bread was measured by the modified refreshing method of Ghiasi et al (1984). A slice of bread stored for three to seven days was wrapped with aluminum foil and placed for 15 min in a cabinet at 110°C. After heating, the bread was cooled to room temperature for 60 min, and the firmness of the breadcrumbs was determined.

Statistical Analyses

Data were independently analyzed as least in duplicate. Analysis of variance (ANOVA) was performed using Duncan's multiple-range test (Steel and Torrie 1960) to compare treatment means; differences were considered significant at $P < 0.05$.

RESULTS AND DISCUSSION

Wheat Flours

The characteristics of CSF, HAWF, and WWF are summarized in Table I. Each flour contained significantly different amounts of

TABLE V
Effect of Waxy Starch on Farinograph Mixing^{a-c}

| Sample | Water Absorption | Time (min) | | | Weakness (BU) | VV |
|--------|------------------|------------|-------------|-----------|---------------|-------|
| | | Arrival | Development | Stability | | |
| CSF | 65.8a | 1.58a | 2.50ab | 7.33b | 68.8a | 48.8b |
| HAWF | 106.1c | 1.63a | 2.13a | 1.00a | 197.5b | 31.0a |
| WWF | 79.3b | 2.33a | 3.70b | 5.83b | 103.8a | 49.5b |

^a CSF, test-milled soft-type wheat from Chinese Spring; HAWF, high-amylose wheat flour; WWF, waxy wheat flour.

^b VV, valorimeter value.

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

TABLE VI
Baking Results of Three Wheat Flours^{a-c}

| Sample | Specific Volume (cm ³ /g) | Firmness (10 ² N/m ²) After Days of Storage | | | | |
|--------|--------------------------------------|--|--------|--------|--------|----------|
| | | 0 | 1 | 3 | 5 | 7 |
| CSF | 5.32b | 37.8a | 277.0b | 798.5c | 925.1c | 1,050.7c |
| HAWF | 1.49a | 278.0b | 351.0c | 549.8b | 662.0b | 789.2b |
| WWF | 5.54b | nd | 52.9a | 356.4a | 409.2a | 593.3a |

^a CSF, test-milled soft-type wheat from Chinese Spring; HAWF, high-amylose wheat flour; WWF, waxy wheat flour.

^b nd, no determination. $n = 8$.

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

TABLE VII
Effect of Waxy Starch on Refreshing of Bread Crumbs by Reheating

| Sample | Firmness (10 ² N/m ²) | | | | | |
|--------|--|--------|----------|----------------------------|----------|----------|
| | Storage Days Before Heating | | | Storage Days After Heating | | |
| | 3 | 5 | 7 | 3 | 5 | 7 |
| CSF | 789.1a | 925.1a | 1,029.5a | 1,045.1b | 1,707.4b | 1,063.6a |
| HAWF | 524.2a | 662.0a | 773.2a | 628.9b | 717.0a | 789.8a |
| WWF | 356.4b | 409.2b | 632.7b | 171.0a | 262.2a | 181.5a |

^a CSF, test-milled soft-type wheat from Chinese Spring; HAWF, high-amylose wheat flour; WWF, waxy wheat flour.

^b $n = 10$.

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

amylose and amylopectin. WWF had considerably more water, wet gluten, protein, and dietary fiber than CSF. HAWF significantly showed the highest values of protein, ash, lipid, and dietary fiber among all the samples tested. An especially large amount of dietary fiber was characteristic of HAWF. Na content was not distinctly different among samples but it was slightly lower in HAWF than in the other samples. Water activity of all samples was ≈ 0.98 , but water activity of CSF was significantly different from the values of HAWF and WWF. The dough made from CSF contained a larger amount of free water than doughs made from HAWF and WWF.

Dough Properties

The gelatinization temperatures and enthalpies of the starch in doughs made with the three wheat flours are shown in Table II. The CSF dough had three peaks: the first and second peaks correspond to the gelatinization temperature of starch; the third peak corresponds to the melting temperature of a starch-lipid complex (Eliasson 1980). The gelatinization temperature (T_{p1}) of starch and the melting temperature of the starch-lipid complex (T_{p3}) of HAWF were significantly lower than those of CSF and WWF. Therefore, the starch in HAWF was gelatinized more quickly in the baking process than the starches in CSF or WWF. On the other hand, T_{p1} of WWF was clearly higher than the T_{p1} of the CSF and HAWF. The gelatinization enthalpies (ΔH_1) of the starches of HAWF and WWF were distinctly greater than the ΔH_1 of CSF. The ΔH_1 of WWF was especially high, resulting in increased gelatinized starch. Based on the third peak, the enthalpy of formation of starch-lipid conjugates (ΔH_3) of HAWF was greater than that of CSF, while ΔH_3 of WWF was less than that of CSF. In general, the formation of starch-lipid conjugates was more closely related to amylose content than to the amylopectin content. HAWF contains a larger amount of amylose than CSF and WWF and, therefore, the larger

amount of starch-lipid conjugates in HAWF may have formed more easily than those in CSF or WWF.

The compression stress (g), modulus of elasticity (γ), and viscosity coefficient (η) of the WWF dough were significantly lower than those of HAWF (Table III). In contrast, HAWF made significantly harder or significantly more viscous doughs than CSF, and the g and η values of its dough were higher than those of CSF. The hardness of dough resulting from low amylopectin content was probably responsible for low stickiness.

The amylograms of the HAWF and WWF pastes were quite different amylogram of the CSF paste (Table IV). The maximum viscosity temperature of WWF paste was significantly lower than that of CSF paste. The maximum viscosity of WWF paste was larger than that of CSF paste. This indicates that the starch in WWF gelatinizes quickly and has a high viscosity. In contrast, the gelatinizing temperature of HAWF starch was distinctly lower than the gelatinizing temperatures of CSF and WWF starches. This result was similar to the DSC results (Table II) and also similar to the results reported by Yamamori et al (2000). HAWF starch had a significantly higher maximum viscosity temperature than the other starches, but its maximum viscosity was significantly lower. Although HAWF made a harder and more viscous dough than CSF or WWF, no increase in viscosity was observed in the HAWF paste when it gelatinized during heating. In general, amylograph data are influenced by the properties of starch in wheat but not by gluten (Approved Method 22-12). Therefore, amylograph data in Table IV might be due to the differences in characteristics of starch in wheat flour.

To make clear whether the effect of WWF on the baking properties depends on the protein or amylose contents, the dough properties were measured using a farinograph (Table V). The water absorptions of HAWF and WWF were significantly higher than

that of CSF. Furthermore, from the values of stability and weakness, WWF dough was significantly stable compared with HAWF dough. However, no differences were observed between CSF and WWF. Therefore, HAWF tended to have inferior flour qualities to CSF. From these results, the high water absorption and unstable curve of HAWF might be caused by a large amount of dietary fiber rather than protein in HAWF. These properties of HAWF and WWF may have affected the baking results.

Baking Properties

The loaf volume of bread made from HAWF was significantly smaller than that of CSF, while the loaf volume of bread made from WWF was slightly larger than that of CSF (Table VI). Because breadcrumbs made from WWF became obviously soft, viscous, and glutinous immediately after baking, the breadcrumbs could not keep the original form. Furthermore, after seven days of storage, breadcrumbs made from WWF were significantly softer than those from CSF and HAWF.

CSF bread (Fig. 1A) had the best appearance and the gas cells in breadcrumbs seemed to be of a finer and more consistent size than those of the HAWF and WWF breads (Fig. 1B and C). The gas cells of HAWF bread were too small, resulting in inadequate expansion, while some of the gas cells of WWF bread were very large. Loaves made from WWF had flat tops and the tops were wider than the bottoms. Therefore, loaves of bread made from WWF and HAWF were inferior in appearance to those made from CSF and would not be acceptable to consumers.

The amylose content of HAWF is obviously different from other cultivars of wheat (Yamamori et al 2000). In this study, the dough and baking results of HAWF were also very different from those of common wheat flours as reported previously (Maeda et al 1999).

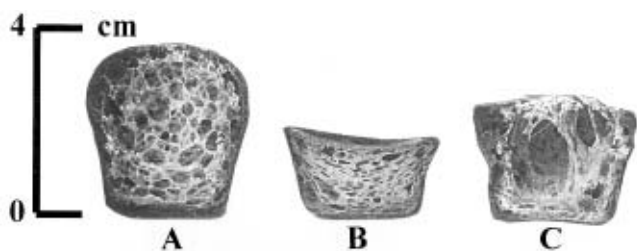


Fig. 1. Cross-sectional views of bread crumbs from three wheat flours. **A**, Chinese Spring flour (CSF); **B**, high-amylose wheat flour (HAWF); **C**, waxy wheat flour (WWF).

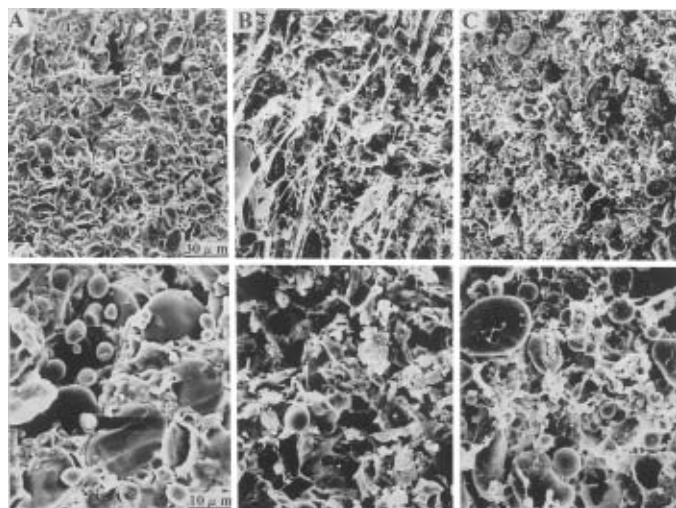


Fig. 2. Scanning electron microscopy of dough from three wheat flours. **A**, Chinese Spring flour (CSF); **B**, high-amylose wheat flour (HAWF); **C**, waxy wheat flour (WWF).

Thus, the present results of HAWF were principally caused by the amylose content.

After storage for three to seven days, WWF bread was obviously softer than CSF bread. After reheating, it became even softer than reheated CSF bread (Table VII). The firmness of CSF or HAWF breadcrumbs were distinctly increased by heating. However, WWF bread could significantly refresh the hard staled breadcrumbs more easily by heating than could CSF bread. When the staled bread of WWF was reheated, the firmness of breadcrumbs decreased distinctly. In general, after breadcrumbs are reheated, they become stale or firm much faster than non-reheated breadcrumbs (Ghiassi et al 1984). However, this was not the case for WWF bread. Reheated breadcrumbs retained their softness after reheating. Therefore, WWF bread was more favorable than CSF or HAWF breads when it was refreshed by reheating.

SEM Results

The gluten of CSF (Fig. 2A) was not extensible, as the dough was rigid and starch granules were mostly buried in the hard gluten matrix. HAWF dough (Fig. 2B) had a rougher structure than the doughs of CSF or WWF (Fig. 2C). The gluten sheet in the HAWF dough was irregular and discontinuous and did not cover some starch granules. This result might be caused by the presence of a larger amount of dietary fiber in HAWF. The gluten of WWF (Fig. 2C) was not as evenly dispersed as it was in CSF dough and did not cover the starch granules as uniformly or entirely as it did in CSF dough.

To study the effect of dough properties on the bread quality after baking, the bread structure immediately after baking was also observed by SEM. CSF bread (Fig. 3A) had more pore structures than WWF bread (Fig. 3B). CSF bread had an even and uniform pore structure, and it had a thin gluten sheet that evenly covered the starch granules. In contrast, the WWF bread had a thick sheet-like substance that was broken by pore structures with a rougher and more irregular appearance than those in CSF bread. Although the CSF dough seemed to be harder than the WWF dough (Fig. 2), it made a bread with an extensible and favorable structure. This

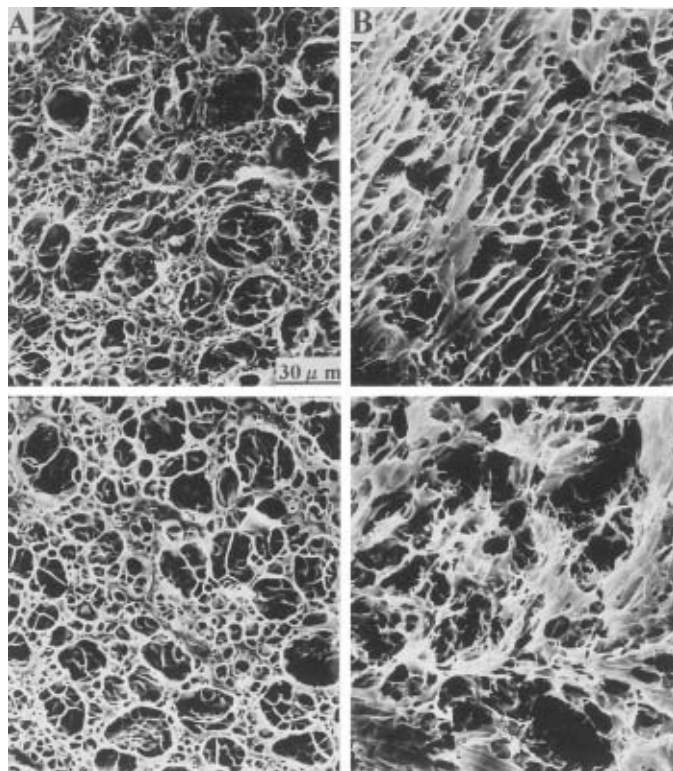


Fig. 3. Scanning electron microscopy of bread from two wheat flours. **A**, Chinese Spring flour (CSF); **B**, waxy wheat flour (WWF).

might explain the good appearance of the breadcrumbs in Fig. 1. In contrast, the discontinuous structure of WWF dough did not result in a favorable or extensible bread structure after baking, even though the specific volume of WWF was larger than that of CSF. This might explain the inferior appearance of breadcrumbs in Fig. 1. In general, gluten must have a high extensibility and it must sufficiently cover starch granules to obtain a good quality bread that is soft and has a large loaf volume. Therefore, it is likely that the clear differences in SEM images of the dough and bread samples affected the appearance of the breadcrumbs in Fig. 1.

CONCLUSIONS

The texture of bread is considered to be one of the most important factors determining taste. The breads that we baked with HAWF or WWF alone did not have good results. The WWF bread was considerably inferior to the CSF bread in appearance. However, the loaf volume and softness of WWF bread were improved over that of CSF. WWF breadcrumbs were also refreshed better than CSF or HAWF breads by reheating. Japanese consumers like glutinous or sticky foods such as glutinous rice and rice cake and eat them almost every day. They also eat bread as a staple food or snack. So it would be desirable for the bread also to have a glutinous or sticky texture. In addition to having a more glutinous texture, WWF bread has a high ability to be refreshed by heating, which is one of the most important and essential qualities for bread. Consequently, we expect that WWF, when added to other conventional flours, will improve the texture and storage ability of the bread. In contrast, HAWF was found unsuitable for breadmaking in the present study. However, because of its nonglutinous nature, HAWF could be used to replace durum wheat flours in pastamaking or as an added ingredient in steamed Chinese processed foods. The genetic background of the present HAWF is quite different from that of CSF. Further studies of HAWF and WWF are needed to develop practical applications of these flours in breadmaking.

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