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Note

Characteristics of Pentosan in Polished Wheat Flour and Its Improving Effects on Breadmaking

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Abstract: Eight fractions of polished flours were prepared by gradually polishing soft-type whole-wheat grains using a rice-polisher. The gluten matrix of doughs and breads made from polished flours was broken by some pericarp layers and the appearances were not sufficient for breadmaking. Polished flours contained water-soluble pentosan (WSP) with a significantly larger amount of xylose as a main chain, while water-insoluble pentosan (WISP) had a smaller amount of xylose than those from N61 and commercial flour (Hermes). The addition of WSP obtained from polished flours of the innermost fraction 30–0% to Hermes significantly improved the dough and baking properties, as compared with that from N61. The improvement of polished flours for breadmaking was due to the characteristics of WSP, namely its high content, high ratio of xylose to arabinose, large amounts of ferulic acid and excellent foaming stability.

Key words: pentosan, polished wheat flours, bread, microscopy, gas chromatography

Polysaccharides such as water-soluble (WSP) and -insoluble pentosans (WISP) contained in wheat flour or added externally to wheat flour have been reported to be important components for breadmaking and affect the baking properties.[1–7] The rheology of dough, such as development time, consistency, extensibility and resistance to breakdown, has also been found to be affected by pentosans.[8–12] When whole-wheat grain was stepwisely polished from the outer layer by 10% of the total weight using a rice-polisher, eight fractions of polished wheat flours were obtained until the 30% to core of the whole grain.[13–16] Polished flour alone could not provide sufficient dough or bread qualities for commercial products; however small amounts of substitution for common flours without additives reversely improved the baking properties.[14,15] On the other hand, pentosanase is one of important enzymes for breadmaking, but the detailed effects on dough or bread quality and mechanisms for their improvement have not been elucidated completely yet.[4,17–19] But the increase of WSP by enzymatic hydrolysis of WISP was found to improve the baking properties,[14,20] and characteristics of pentosans, such as molecular weight, degree of polymerization and solubility in water have been studied in relation to functional properties in the food system.[21] Since polished flours contained larger amounts of WSP and WISP than common wheat flours,[13] the relationships between the characteristics of pentosans in polished flours and their improving effects on breadmaking were studied. To clarify the functional property of pentosans, the effect of pentosans obtained from polished flours on breadmaking will be discussed in comparison with those from common flour.

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MATERIALS AND METHODS

Flour and chemicals. Soft-type polished flours of eight fractions from NA-1 (100–90%) to NA-8 (30–0%) were prepared from wheat grain of Norin 61 by the same method as reported previously.[14,15] In the following experiments, NA-1 (100–90%), NA-5 (60–50%) and NA-8 (30–0%) were used. For the control samples, a commercial hard-type wheat flour of Hermes and conventionally milled wheat flours of N61 from Norin 61 were used. All other chemicals of analytical grade were used without further purification.

Light microscopic observation of polished flour doughs and breads. Dough and breads samples for light microscopy (LM) were prepared by the previous method[15] and observation was conducted with the same apparatus and procedure as reported previously.[16,20] But some samples were embedded in an Epon mixture and sectioned with glass knives on a Porter-Blum Ultra-microtome MT1 (Sorvall Inc., USA), and stained with 0.05 M-phosphate buffer containing 1% toluidine blue or 0.1% basic fuchsin at pH 6.8.

Characteristics of pentosans isolated from flours.

Isolation of pentosans from flours. To determine the characteristics of pentosans, WSP and WISP were isolated from the polished flour of the innermost fraction of NA-8, N61 and Hermes. Namely, the WSP and WISP were isolated from the water-extracts and tailings fractions by the previous methods.[4,22] The procedures and enzymes used for the isolation were the same as reported previously.[16]

Sugar composition. The ratio of arabinose to xylose

(Ara/Xyl) in pentosans was tested by the earlier method[23] with a slight modification using gas-liquid chromatography (GC). Alditol acetate derivatives of the pentosan were analyzed by a glass column (4 mm I.D.×1.8 m) packed with 3% ECNSS-M on Gaschrom Q 100–120 Mesh (GL Science Co., Ltd., Tokyo) using a gas-chromatograph G3800 (Yanaco Co., Ltd., Osaka) with a flame-ionization detector. The column was programmed at a rate of 0.5°C/min from 190°C to 210°C, and temperatures of the injector and detector were 260°C.

Foaming stability. The effect of WSP on the stability of the foam prepared by the use of surface-active protein [bovine serum albumin (BSA)] was studied according to the previous procedure.[24] One mL of 2% (w/v) BSA and 0.25 mL of 1.0% (w/v) WSP solution were mixed in a test tube. Effects of acid and heating on the foaming stability were measured from the change of foam volume.

Ferulic acid content. The amount of ferulic acid in WSP and WISP obtained above were determined according to the method reported by Ciacco and D'appolonia,[25] using the absorbance at 320 nm.

Effects of isolated pentosans on dough and baking properties. Mixing properties of dough containing isolated pentosans were determined by Farinograph mixing.[26] Test baking was performed by the same method as described previously.[26] The WSP and WISP were added to Hermes by 1.0% on a flour weight basis and their effects on the dough and baking properties were determined. Furthermore, the flours with gluten, starch and pentosan at the same ratio as normal breadmaking described above were reconstructed and test baking was done to determine the direct effect of pentosan on the baking properties.

Statistical analysis. Values were obtained as the means ± standard deviation of 3 determinations, analyzed by ANOVA and Dunett's multiple-range test using SPSS (Version 11.0, SPSS Inc., Chicago, USA). Differences among samples were considered significant at $p<0.05$.

RESULTS AND DISCUSSION

LM results of polished flour doughs and breads.

The gluten matrix of doughs made from polished flours which was stained with toluidine blue was distinctly broken by an aleurone or pericarp layer (indicated by arrow), and polished flours could not form an extended gluten network (Fig. 1). For the case of breads baked with polished flours alone, a little portion of thick and discontinuous gluten protein, which was stained with green colour was not constantly dispersed around starch granules with pink color, as compared with Hermes and N61 (Fig. 2). This appearance was similar to that of dough images as shown in Fig. 1. As shown in these LM images, utilization of polished flour alone for breadmaking could not make the sufficient dough and bread to produce proper bread qualities as commercial products, as reported previously.[15]

Pentosan content and GC analysis.

All polished flours included significantly more amounts of WSP and WISP than N61 or Hermes as shown in Table 1.[16] The ratio of Ara/Xyl in WSP obtained from NA-8 significantly decreased, whereas that in WISP increased, as compared with that of N61 (Table 1). Therefore, the WSP from NA-8 of polished flour was found to have a larger amount of xylose as a main chain than that from N61, while the WISP had the smaller amount.

Ferulic acid content.

NA-8 contained significantly larger amounts of ferulic acid in the WSP and WISP, as compared with Hermes

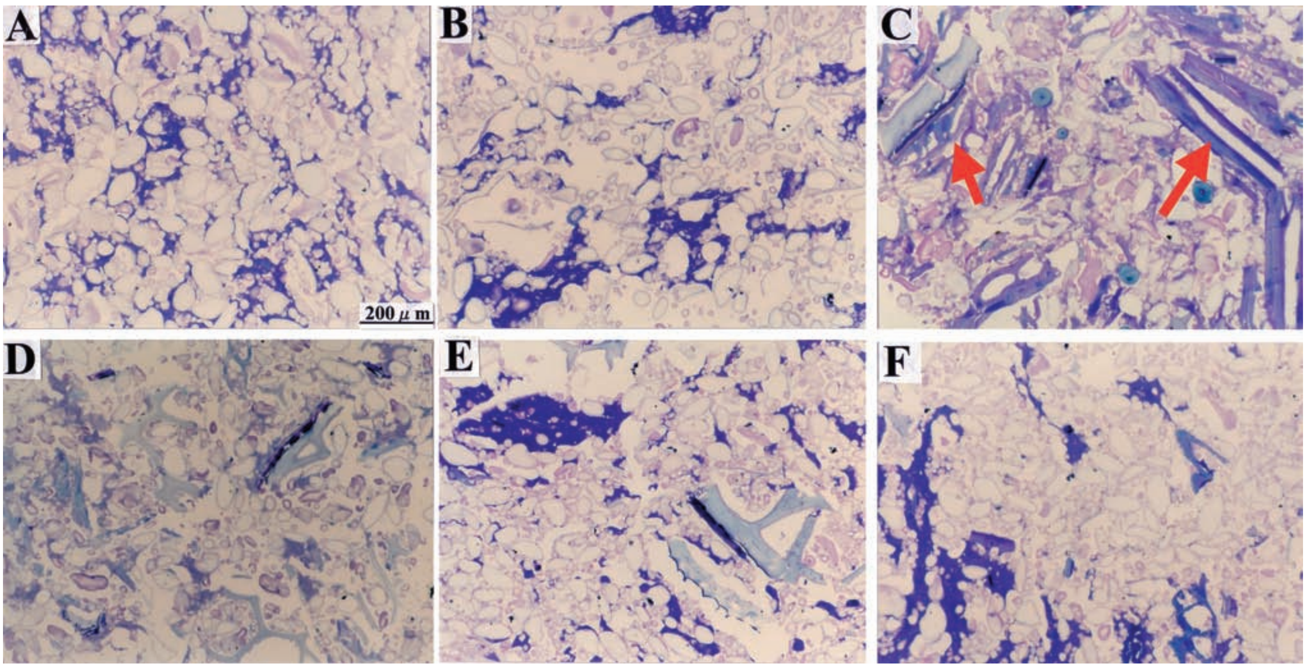

Fig. 1. Light microscopic observation of dough stained with toluidine blue.

A, Hermes; B, N61; C, NB-1; D, NA-1; E, NA-5; F, NA-8. Hermes and N61 are hard- and soft-type common wheat flours, respectively. Polished flours used: NA-1 and NB-1, 100–90%; NA-5, 60–50%; NA-8, 30–0% fractions of soft-type wheat grain. Polished flours were sieved through a pore size of 125 μm, except that NB-1 was sieved through one of 600 μm.

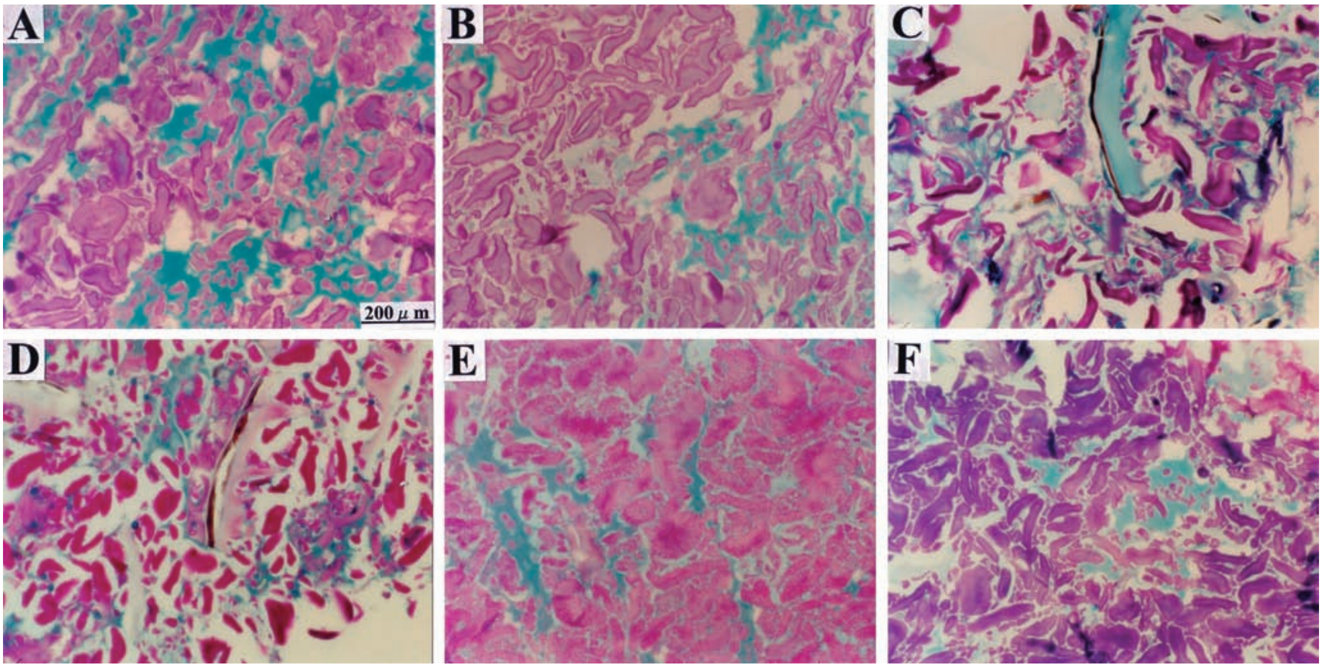

Fig. 2. Light microscopic observation of bread stained with Periodic Acid Schiff reaction and light green.

A, Hermes; B, N61; C, NB-1; D, NA-1; E, NA-5; F, NA-8. Abbreviations are the same as in Fig. 1.

Table 1. Summary of characteristics of pentosans in various polished wheat flours.

Sample	Pentosan content[16]		GC analysis		
	WSP	WISP	TP	WSP	WISP
	(%)	(%)	Ara/Xyl	Ara/Xyl	Ara/Xyl
Hermes	0.73[b]	1.08[b]	1.22[b]	0.99[b]	1.38[b]
N61	0.78[a]	0.89[a]	0.80[a]	0.90[a]	0.74[a]
NA-1	1.08[ab]	13.43[ab]	0.88[ab]	0.82[a]	0.88[a]
NA-5	1.15[ab]	2.34[ab]	0.83[ab]	0.77[ab]	0.86[a]
NA-8	1.28[ab]	1.84[ab]	0.76[ab]	0.67[ab]	0.79[a]

WSP and WISP are water-soluble and -insoluble pentosans, respectively. Polished flours used: NA-1, 100–90%; NA-5, 60–50%; NA-8, 30–0% fractions of soft-type wheat grain (N61). [a] and [b]: $p<0.05$ (*versus* Hermes and N61, respectively).

and N61 as shown in Table 2.

Foam stability.

Foam stability of WSP has been considered to be an important functional quality for breadmaking.[27] The good foaming property might retain the gas generated in the dough, and the pentosans with high foam stability could have the same role as gluten did.[28] WSP obtained from NA-8 did not have enough foam stability at just (0 min) or 10 min after standing at room temperature, whereas after heating for 2–3 min at 100°C at stages IV and V, the WSP showed higher stability than other samples (Table 2). These properties of WSP in NA-8 could regulate the extensibility and maturation of the dough, resulting in a good application to breadmaking.

Characteristics of pentosan and ferulic acid in polished flours.

The ratio of Ara/Xyl in arabinoxylan prepared from the outer portion of wheat grain is reported to be higher than that from the inner portion.[29,30] Molecular weights of water-soluble and -insoluble arabinoxylans in wheat flours are 50,000–100,000 and 132,000–148,000, respectively.[31] And those of arabinoxylans from bran and endosperm cell-walls are 40,000 and 800,000–5,000,000, respectively.[31] With regard to ferulic acid in wheat grains, its contents in germ, endosperm, aleurone cell-wall and pericarp have been reported to be 0.14, 0, 1.8, and 0.45–0.5% as phenolic acids, respectively.[31,32] Since the wheat grain has a hollow called a crease in the central portion, NA-5 or NA-8 is also contained the bran. The higher ratio of Ara/Xyl or the greater amount of ferulic acid in pentosans of polished flours might be due to the larger amounts of aleurone and

Table 2. Characteristics of pentosans isolated from various wheat flours.

Original flour	Ferulic acid content		Foam stability of WSP[*]				
	μg/mg WSP	μg/mg WISP	I	II	III	IV	V
Hermes	0.248[b]	0.500[b]	14.0	21.8	16.5	2.5	0.5
N61	0.310[a]	1.282[a]	15.6	26.2	18.2	4.6	2.0
NA-8	0.640[ab]	1.744[ab]	15.6	22.8	16.2[b]	23.0[ab]	4.6[ab]

Abbreviations are the same as in Table 1. [*]Bovine serum albumin and water-soluble pentosan were mixed for 30 s with a homogenizer. The height of foam in a constant test tube was measured: I, immediately after mixing; II, after reaction with citric acid for 30 s; III, after standing for 10 min at room temperature; IV, after heating for 2 min; V, after heating for 3 min. [a] and [b]: $p<0.05$ (*versus* Hermes and N61, respectively).

Table 3. Effect of pentosan isolated from various wheat flours on dough and baking properties.

Sample	Water absorption (%)	Arrival time (min)	Development time (min)	Stability time (min)	Weakness (BU)	V.V.	Specific volume (cm^3/g)	Firmness ($10^2 N/m^2$) Storage day			
								0	1	2	3
Hermes	65.6	8.4	24.3	19.3	40	98.8	4.85	41	347	1780	2339
N61	54.6[a]	1.3[a]	1.7[a]	1.3[a]	105[a]	42.5[a]	4.02[a]	95[a]	1077[a]	2481[a]	2528
NA-8	73.2[a]	1.4[a]	1.8[a]	1.2[a]	90[a]	44.0[a]	2.61[a]	177[a]	1508[a]	7346[a]	8533[a]
WSP (N61)	63.6[a]	2.0[a]	3.5[a]	3.1[a]	290[a]	37.0[a]	3.99[a]	61[a]	388	2141[a]	2405
WSP (NA-8)	64.4a[*]	3.5a[*]	23.0[*]	27.0[a*]	23[*]	98.0[*]	4.50[*]	37[*]	336	852[a*]	2242
WISP (N61)	67.8[a]	4.1[a]	10.3[a]	9.3[a]	108[a]	75.5[a]	5.55[a]	82[a]	440	735[a]	1572[a]
WISP (NA-8)	66.8a[**]	4.5[a]	11.5[a]	10.9[a]	90[a]	80.5[a]	4.63[**]	43[**]	418	639[a]	2064[**]

N61 or NA-8 in parentheses means the origin of flour used to obtain pentosan. Abbreviations are the same as in Table 1, except for V.V. (Valorimeter value). The amount of WSP or WISP added is 1.0% on a flour weight basis. [a], [*] and [**] $p < 0.05$ (*versus* Hermes, WSP (N61) and WISP (N61), respectively).

pericarp layers in the outer portion, as compared with N61. As the viscosity or polymerization (chain length) of WSP was not determined in the present experiments, the exact molecular weight of WSP can not be discussed in depth. Nevertheless, the high-molecular-weight arabinoxylan regulates the characteristics of the water-holding capacity, oxidative gelation and viscosity.[33] The high-molecular-weight arabinoxylan has been known to form more rigid gels rather than low-molecular-weight arabinoxylan, and ferulic acid content has also been reported to be an important determinant of gel network rigidity.[34] Because the original polished flours already included a large amount of pentosans and their isolated WSP contained a large amount of ferulic acid, lower Ara/Xyl and higher foam stability as shown in Tables 1 and 2, WSP obtained from NA-8 was assumed to include some high-molecular-weight arabinoxylan with apparently longer core xylose chains, as compared with N61.

Comparison of pentosans between N61 and NA-8 for breadmaking.

NA-8 of original flour needed a significantly larger amount of water for suitable dough consistency at 500 B.U., and showed much poorer baking qualities with insufficient specific volume and storage properties, as compared with Hermes or N61 (Table 3). However, the WSP obtained from NA-8 significantly increased the specific volume more than that from N61, while the addition of WISP from NA-8 decreased the specific volume. The WSP obtained from NA-8 significantly improved the softness of breadcrumbs after storage for 0 and 3 days, as compared with that from N61. As to the baking results with reconstructed ingredients composed of gluten, starch and pentosan, effects of various pentosans on the specific volume of bread and storage properties showed similar tendencies to the bread baked with original ingredients as described above (data not shown). The dough containing WSP from NA-8 significantly increased the stability time and valorimeter value (V.V.) with lower weakness, as compared with the dough containing WSP from N61. The addition of WSP from NA-8 to Hermes formed hard and favorable doughs for making bread of good quality, and significantly improved the dough properties, as compared with Hermes without additives. And whatever original flours were N61 or NA-8, additions of the WISPs weakened the stability of dough, as compared with Hermes. Characteristics of arabinoxylan in pentosans, such as the molecular weight, ratio of Ara/Xyl, amount of ferulic acid and gelling properties have been examined to make clear the involvement for the dough properties and baking qualities.[33−37] As shown in Table 3, the addition of WISP did not result in clear differences in the dough and baking qualities, regardless the kinds of flours used. However, WSP obtained from NA-8 improved the dough properties, probably caused by the lower ratio of Ara/Xyl, as compared with that from N61. Higher foaming stability of WSP increased the viscosity and extensibility of the dough, followed by the excellent dough and the storage properties of breadcrumbs. The gelling property of pentosan with ferulic acid is one of the most important characteristics known to affect dough properties and bread qualities[12] and related studies on ferulic acid have been reported.[38,39] Therefore, the ferulic acid in the bran of polished flours might affect the hydrated network structure of gelled pentosan, which suppressed the collapse of gas cells formed in the dough, as compared with common flours.

Through the present results, the improving mechanism of original polished flours on breadmaking would be correlated to pentosans in polished flours, especially their characteristics of WSP, such as high contents, low ratio of Ara/Xyl and excellent foaming stability and large amounts of ferulic acid. Flour quality of the innermost polished flour, NA-8 was considered to be relatively similar to commercial flours, but it showed significantly different properties in WSP or WISP from N61 or Hermes. Therefore, the characteristics of pentosan in NA-8 described above were assumed to be similar to those in other polished flours. Consequently, we expect that polished flours will become a new foodstuff making the best use of their gelling properties for the baking industry.

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分級粉中のペントサンの特性とその製パンにおける改善効果

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軟質小麦穀粒を酒米搗精機で段階的に削り製粉し，8 分画の分級小麦粉を得た。分級粉で調製したドウやパンのグルテンはいくつかの外皮成分により切断され，製パンには不十分な形態であった。分級粉の水溶性ペントサン（WSP）は主鎖成分であるキシロースを通常粉（N61）や市販粉（ヘルメス）より有意に多く含み，一方不溶性ペントサン（WISP）中にはキシロース量は少なかった。中心部（30−0%）の分級粉から得られた WSP をヘルメスに添加すると，N61 由来の WSP よりドウの物性と製パン性を有意に改善した。分級粉の製パン改善効果は，その WSP の性質，すなわち含有量の高さ，アラビノースに対するキシロースの高い比率，多量のフェルラ酸，ならびに優れた泡沫安定性に起因すると考えられた。