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# Structural characterisation of pentosans from hemicellulose B of wheat varieties with varying chapati-making quality

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# ABSTRACT

Wheat varieties, such as DWR-162 and GW-322 (good chapati-making quality), and MACS-2496 and HD-2189 (poor chapati-making quality), were used to study the structural features of pentosans. Structural features of the purified pentosans from hemicellulose B were elucidated by a combination of methods, such as methylation analysis, <sup>1</sup>H NMR, FT-IR, periodate oxidation, Smith degradation and optical rotation measurements. Pentosans from hemicellulose B were mainly arabinoxylan type polysaccharides with xylan backbone in  $\beta$ -(1  $\rightarrow$  4) linkages. Mono, and di-substituted xylosyl residues were present in these polysaccharides. Variations in structural features of pentosans could be responsible for the differences in chapati-making qualities of wheat.

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# 1. Introduction

In wheat, pentosans are the major components of hemicellulosic polysaccharides and are associated in the cell wall fragments with other components, including cellulose,  $\beta$ -glucan, glycoprotein and polyphenols (Fincher & Stone, 1986; Izydorczyk & Biliaderis, 1995). Pentosans play an important role in dough rheology and bread quality because of their remarkable functional properties. They exhibit a high affinity for water and are partly responsible for the high water absorption and viscosity of dough (Courtin & Delcour, 2002). They are also used as adhesive, thickeners, stabilisers and as emulsifiers (Fang, Sun, Salisbury, Fowler, & Tomkinson, 1999). Depending on the sequence of extraction, hemicelluloses are categorised as hemicelluloses A, B and C, which vary in their sugar composition, physical and functional properties (O'Neill and Selvendran, 1985; Subba Rao & Muralikrishna, 2006).

The hemicelluloses are estimated to account for one third of all components available in plants (Oraphin, Pawadee, & Krisda, 2004). Structural features of the pentosans, also known as arabin-oxylans, vary significantly in different cereals and have been a topic of great academic interest (Izydorczyk & Biliaderis, 1995; Saulnier, Sado, Branlard, Charmet, & Guillon, 2007). They consist of a backbone of  $1 \rightarrow 4$ -linked  $\beta$ -D-xylopyranosyl residues to which  $\alpha$ -L-arabinofuranose units are linked as side branches. As one of

the major constituents of dietary fibre, they are known to reduce the incidence of various diseases, such as colon cancer, atherosclerosis and diabetes (Lu, Walker, Muir, Mascara, & Kerin, 2000).

Although arabinoxylans from various cereals share the same basic chemical structure, they differ in the way of substitution of arabinose (Ara) residues to the xylose (Xyl) backbone (Schooneveld, Beldman, & Voragen, 1999). The main differences are found in the ratio of arabinose to xylose, in the relative proportions and sequence of various linkages between these two sugars and in the presence of other substituents, such as glucuronic acid and ferulic acid (Andrewartha, Phillips, & Stone, 1979; Izydorczyk & Biliaderis, 1995).

Arabinoxylans from rice (Yui, Imada, Shibuya, & Ogawa, 1995) and sorghum (Nandini & Salimath, 2002; Verbruggen, Beldman, & Voragen, 1995) are more highly branched than those from wheat, rye and barley (Dervilly, Rimstein, Saulnier, Andersson, & Aman, 2001). Previously, we have reported on the isolation and carbohydrate composition of pentosans from wheat varieties, in relation to chapati-making quality (Revanappa, Bhagwat, & Salimath, 2007).

In the present investigation, pentosans from wheat varieties having differences in chapati-making qualities were purified from hemicelluloses B and their fine structural features were elucidated by a combination of methods, such as methylation, GC–MS, <sup>1</sup>H NMR, FT-IR and periodate oxidation studies. Methylation analysis was performed to determine the nature of glycosidic linkages between monosaccharide residues of arabinoxylans and give information on the nature of sugars present, either in pyranose or





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furanose forms and degree of branching. <sup>1</sup>H NMR was used for investigation of anomeric protons of arabinoxylans and their substitution pattern. FT-IR spectroscopy was used to detect functional groups, configuration of sugar residues and to discover the substitution pattern in arabinoxylans (Kacurakova, Ebringerova, Hirsch, & Hromadkova, 1994). Periodate oxidation and Smith degradation studies were carried out to confirm the degree of branching and the arrangement of the branched residues.

### 2. Materials and methods

# 2.1. Materials

DWR-162 variety of wheat (*Triticum aestivum* L.) was obtained from University of Agricultural Sciences, Dharwad, and MACS-2496, GW-322 and HD-2189 varieties were procured from Agharkar Research Institute, Pune, India. Termamyl (EC 3.21.1. from *Bacillus licheniformis*) was procured from Novo Nordisk, (Copenhagen, Denmark). Glucoamylase and dialysis bags (Cellulose membranes, 12,000 MW cut off) were procured from Sigma, St. Louis, MO. All other chemicals and reagents used were of analytical grade.

### 2.2. Isolation and purification of pentosans from hemicelluloses B

Hemicelluloses were extracted by following the previously described method (Nandini & Salimath, 2001). Briefly, flour was treated with 70% alcohol to remove free sugars. The residue was cooked with water, to gelatinise starch, and then subjected to glucoamylase digestion. The starch-free residue was then taken for the extraction of hemicelluloses, using 10% NaOH under nitrogen atmosphere. The extract was acidified with acetic acid (50%) at ice-cold temperatures to pH 4.5, to get a precipitate and the supernatant was dialysed and lyophilised (hemicellulose B). It was further purified by alcohol precipitation under acidic (pH 3.0) conditions followed by glucoamylase digestion at 60 °C for 4 h. After the digestion, the polysaccharides were precipitated by adding two volumes of alcohol; the residue was collected by centrifugation and uniformly dispersed in water and lyophilised (Nandini & Salimath, 2003).

### 2.3. Sugar composition analysis by GC

Sugar composition in the flour and water-insoluble fractions was analysed after solubilisation with 72% sulphuric acid (at icecold temperatures), followed by hydrolysis in 10% sulphuric acid at boiling water bath temperatures for 6–8 h. The water-soluble fractions were hydrolysed with 2 N trifluoroacetic acid in sealed tubes at 100 °C for 5–6 h. The sugars were analysed by gas chromatography as alditol acetates (Sawardekar, Slonekar, & Jeanes, 1967) on an OV-225 column at temperature of 200 °C using a Shimadzu GC (Shimadzu, Kyoto, Japan). The column was calibrated and the response factors were calculated by injecting a sugar standard mixture containing rhamnose, arabinose, xylose, mannose, galactose, glucose and inositol (used as internal standard). Sugars present in the samples were identified by comparing their relative retention times with standards and were quantified by using response factors. The sugar contents are expressed in mole percent.

# 2.4. Methylation analysis

Polysaccharides (10 mg) were methylated, following the method of Hakomori (1964). Permethylated polysaccharides were hydrolysed with formic acid and sulphuric acid, successively, acetylated and the partially permethylated alditol acetates were analysed by GC–MS. GC–MS analysis was performed on a Shimadzu (Model QP 5000) GC–MS using an SP-2330 capillary column (30 m  $\times$  0.31 mm; Supelco, Bellefonte, PA), operating at an ionisation potential of 70 eV, with a temperature programme of 180–200 °C, with a 4 °C rise per min. Chloroform was used as solvent and the carrier gas was helium. Permethylated sugars were identified based on their relative retention times, with 2,3,4,6-tetramethyl glucose used as a reference standard.

# 2.5. <sup>1</sup>H NMR Spectroscopy

Purified polysaccharide (10 mg/ml in D<sub>2</sub>O) was taken in sample probe ( $\phi$  5 mm × 15 cm) and a resonance spectrum was recorded in a Bruker 500-MHz NMR (Bruker Biospin, GmbH, Rheinstetten, Germany) and the shifts were recorded with reference to D<sub>2</sub>O (Hoffmann, Kamerling, & Vliegenthart, 1992).

# 2.6. Periodate oxidation

Polysaccharide solution (10 mg in 5 ml) was mixed with sodium metaperiodate (5 ml, 20 mM) and kept at 4 °C in the dark for 48 h. Aliquots were withdrawn from the sample at regular intervals (4 h) and the amount of periodate remaining was determined by following an earlier described method (Avigad, 1969). Formic acid liberated from the polysaccharide sample on oxidation was estimated by a titrimetric method, using sodium hydroxide.

#### 2.7. Smith degradation

Polysaccharide sample (10 mg in 5 ml water) was oxidised with sodium metaperiodate (5 ml, 20 mM) at 4 °C in the dark for 48 h, treated with ethylene glycol (0.1 ml to stop the reaction) and then reduced with sodium borohydride (100 mg) at room temperature for 16 h. The sample was hydrolysed with sulphuric acid (0.5 N) at room temperature for 48 h, acetylated and the resultant Smith degradation products were analysed by GC (Abdel-Akher, Hamilton, Montogomery, & Smith, 1952).

#### 2.8. Analytical methods

Estimation of total sugars was done by phenol-sulphuric acid method (Dubois, Gilles, Hamilton, Rebers, & Smith, 1956) and uronic acids by carbazole method (Dische, 1947). Ferulic acid was quantified by HPLC upon alkaline hydrolysis (Shyamprasad Rao & Muralikrishna, 2007). Optical rotation of polysaccharides was determined at 20 °C, using a Perkin–Elmer (model 243) polarimeter (Perkin–Elmer, Waltham, MA). For FT-IR measurements, polysaccharides were blended with KBr to form pellets and the spectra were recorded between 4000 and 400 cm<sup>-1</sup> (Kacurakova et al., 1994). All results are given as an average of triplicate determinations.

# 3. Results and discussion

# 3.1. Carbohydrate composition of purified pentosans from hemicellulose B

The carbohydrate composition of native and purified pentosans from hemicellulose B is given in Table 1. The carbohydrate content was higher in all the fractions of hemicellulose B compared to native samples. Uronic acid content was highest in DWR-162 followed by MACS-2496. Sugar composition analysis indicated that these fractions are rich in pentosans (arabinose and xylose) and glucose was the major contaminating sugar observed in all the fractions studied. Native fractions had a substantial amount of

Carbonydrate composition (%) of native and purified pentosans from hemicellulose B of wholewheat flour of different varieties.								
Samples	Total sugar	Uronic acid	Ara	Xyl	Man	Gal	Glc	Ara/Xyl
DWR-162								
Native	80.0	2.8	26.5	39.0	2.5	-	32.0	0.67
Purified	90.2	3.2	34.2	61.2	-	-	4.6	0.55
GW-322								
Native	90.0	4.5	13.5	63.0	-	-	23.5	0.21
Purified	94.7	2.6	35.0	58.6	1.4	1.0	4.0	0.59
MACS-2496								
Native	80.0	4.0	21.6	48.0	-	-	30.4	0.44
Purified	88.5	2.8	30.2	65.6	-	-	4.2	0.46
HD-2189								
Native	84.0	1.9	30.6	24.6	2.0	0.4	42.4	1.24
Purified	90.6	2.3	25.6	64.2	1.8	2.4	6.0	0.39

Table 1
Carbohydrate composition (%) of native and purified pentosans from hemicellulose B of wholewheat flour of different varieties.

Ara: Arabinose, Xyl: Xylose, Man: Mannose, Gal: Galactose, Glc: Glucose.

glucose, a part of which may be coming from undigested starch molecules and other associated polysaccharides, such as xyloglucans and glucans (Dupont & Selvendran, 1987). Galactose and mannose were observed in GW-322 and HD-2189 in small amounts. Galactose might be due to galactoarabinoxylans and arabinogalactans, the presence of which has been reported in the members of graminae (Izydorczyk & Biliaderis, 1995; Nandini & Salimath, 2001). Mannose may be due to glucomannans, which has been found in various cell walls. The lower arabinose to xylose ratio in all these fractions might be due to the presence of mixture of xylan and arabinoxylan type of polysaccharides (Nandini & Salimath, 2003). The presence of glucose in small amounts along with arabinose and xylose, indicate them to be glucuronoarabinoxylans and a xyloglucan type of polysaccharide (Dupont & Selvendran, 1987). Arabinoxylans may be more branched in GW-322 and DWR-162 wheat varieties having good chapati-making quality, as indicated by a higher Ara / Xyl ratio.

# 3.2. Methylation analysis of purified pentosans

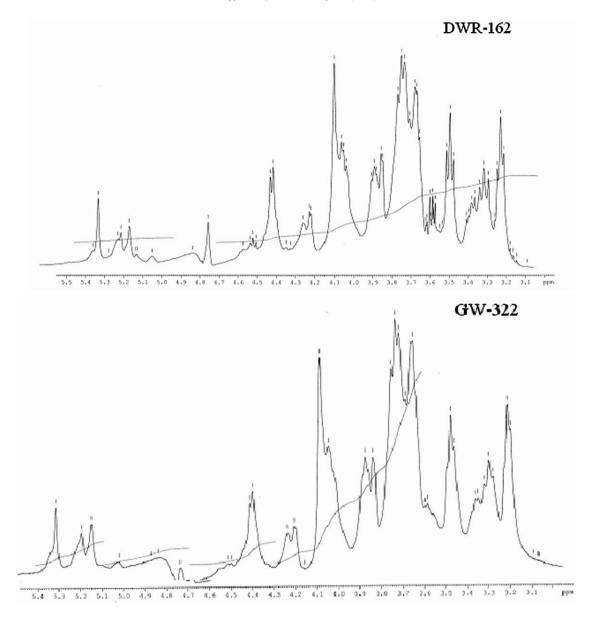
The pentosans from hemicellulose B were methylated in order to study the nature of glycosidic linkages and substitution pattern. Methylation and subsequent hydrolysis indicated that the xylose residues were present in three forms: unsubstituted, monosubstituted and disubstituted (Table 2). Methylation analysis revealed the xylose residues in the main chain with  $1 \rightarrow 4$  linkages. Most of the xylose residues were unsubstituted, as evidenced by the presence of large amounts of 2,3-Me<sub>2</sub>-Xyl. This was at higher levels in MACS-2496 and HD-2189 varieties which have poor chapatimaking characteristics. Small amounts of terminal xylose resides were present in the pyranose form, as indicated by the presence of 2,3,4-Me<sub>3</sub>-Xyl. The xylan backbone was substituted mainly by arabinofuranosyl residues at the O-3 position, as indicated by the presence of higher amounts of 2-Me-Xyl in the GW-322 variety, followed by DWR-162. Doubly substituted xylose residues were also found in all the wheat varieties, as indicated by the presence of xylitol among the methylated species. This was present in higher amounts in arabinoxylans from hemicellulose B of DWR-162 followed GW-322. Disubstituted xylose residues have been reported in wheat (Nandini & Salimath 2003), rice (Yui et al. 1995), and sorghum (Nandini & Salimath, 2002). Most of the arabinosyl residues were present as terminal sugars, indicated by the presence of 2,3,5-Me<sub>3</sub>-Ara. Main chain arabinosyl residues (2,3-Me<sub>2</sub>-Ara) were also observed in minor amounts in all the wheat varieties. Short arabinosyl chains have been reported in branched arabinoxylans (Izydorczyk & Biliaderis, 1995). Variations were observed in the degree of branching, as indicated by the ratio of unbranched to branched xyloses. The ratio was higher in arabinoxylans from hemicellulose B of MACS-2496 and HD-2189, indicating that arabinoxylans in these varieties are less branched, compared to DWR-162 and GW-322 wheat varieties. However, the ratio of disubstituted to mono-substituted xylose was higher in HD-2189 and lower in MACS-2496. The hemicellulosic polymers from the cell walls of wheat bran sequentially extracted with alkali showed both high and low degree of branching (Nandini & Salimath, 2003; Schooneveld et al., 1999).

# 3.3. <sup>1</sup>H NMR spectroscopy of pentosans

Nuclear magnetic resonance (NMR) spectroscopy was used to obtain more structural information on high molecular weight polysaccharides and their building blocks. <sup>1</sup>H NMR spectra of the hemicellulose B from wheat varieties are shown in Fig. 1. Chemical shifts were assigned by comparison with previously reported literature data (Hoffmann et al., 1992; Gruppen et al., 1992). Proton NMR analysis of hemicelluloses B fractions from wheat varieties revealed signals for anomeric protons of terminal  $\alpha$ -D-arabinofuranosyl residues at 5.2–5.4 ppm and of  $\beta$ -D-xylopyranosyl residues at 4.4–4.7 ppm (Hoffmann et al., 1992). Signals at 5.35, 5.29 and 5.26 ppm are due to anomeric protons of arabinose residues substituted at O-3 (mono-substituted) and at both O-3 and O-2 (di-substituted) of xylose residues, respectively. Signals obtained

Table 2

Peak no.	Mode of linkage	Alditol acetates	DWR-162	GW-322	MACS-2496	HD-2189
1	$Ara_f \rightarrow 1$	2,3,5-Me <sub>3</sub> -Ara	16.7	18.2	14.7	16.0
3	$5 \rightarrow Ara_f \rightarrow 1$	2,3-Me <sub>2</sub> -Ara	1.5	1.0	2.7	1.2
6	$2,3,5 \rightarrow Ara_f \rightarrow 1$	Ara	9.8	10.0	9.6	7.6
2	$Xyl_p \rightarrow 1$	2,3,4 -Me <sub>3</sub> -Xyl	3.5	3.2	3.8	3.2
4	$4 \rightarrow Xyl_p \rightarrow 1$	2, 3- Me <sub>2</sub> -Xyl	42.6	40.6	50.4	49.0
5	$3,4 \rightarrow Xyl_p \rightarrow 1$	2- Me-Xyl	16.2	17.6	12.8	14.0
7	$2,3,4 \rightarrow Xyl_p \rightarrow 1$	Xyl	9.7	9.4	6.0	9.0
_	-	$\frac{2,3-Me_2-Xyl}{2,Me_2-Xyl}$	1.64	1.50	2.6	2.0
-	-	2-Me-Xyl+Xyl Xylose 2-Me-Xyl	0.60	0.53	0.47	0.64

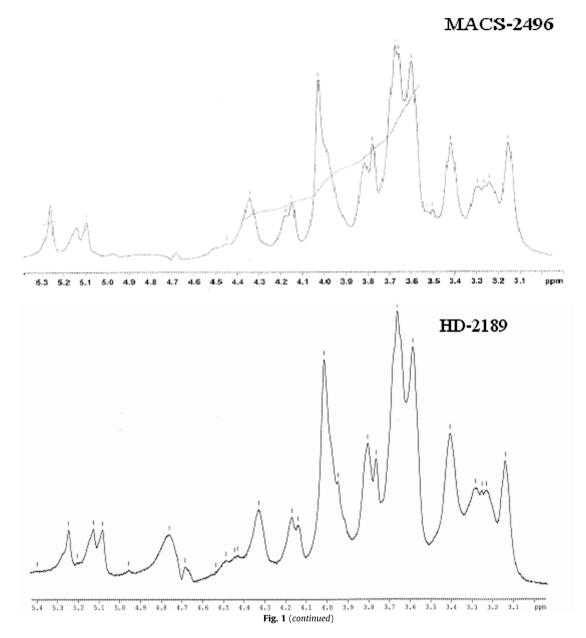


**Fig. 1.** Details of <sup>1</sup>H NMR spectra of arabinoxylans of hemicellulose B fractions from wheat varieties. Signals at 5.2–5.4 ppm is the anomeric region of arabinose residues. Signals at 4.4–4.7 ppm is the anomeric region of xylose residues. 4.8 ppm: signals from β-glucan (Hoffmann et al., 1992).

at 4.58, 4.53 and 4.42 ppm are due to the anomeric protons of  $\beta$ -Dxyloses substituted at C-2 and C-3 (di substituted), C-3 (monosubstituted) and unsubstituted residues, respectively. The signals for other protons of arabinose and xylose were observed in the region of 3.2 to 4.3 ppm and were in close proximity as reported for other cereal arabinoxylans (Gruppen et al., 1992). The unresolved signals at the left side of the peaks at 5.30 ppm (DWR-162 and GW-322) are probably the result of two neighbouring di-substituted Xyl residues in the chain (Cleemput, Roels, Vanoort, Grobet, & Delcour, 1993). There were low levels of mono-substituted xylose residues ( $\delta$  5.38 ppm) in HD-2189, as revealed by low intensity of peak. The small unresolved peaks at 5.38 ppm can be attributed to mono-substituted xylose adjacent to di-substituted xyloses and were observed in DWR-162 and GW-322. Apart from known arabinose peaks, the spectra of DWR-162 and GW-322 contained a variety of unidentified peaks, which revealed the complexity of the polysaccharides. Mono-substituted xylose residues adjacent to di-substituted xylose residues, as suggested by Cleemput et al. (1993), were probably not present in MACS-2496 and HD-2189, because the shoulder downfield of the signal of the arabinose (5.36 ppm) on the mono-substituted xylose was absent. The signals at 4.53 ppm for a mono-substituted xylose, at 4.66 ppm for an isolated di-substituted xylose and at 4.65 and 4.59 ppm for paired di-substituted xylose residues were observed in these spectra. The signal at 4.16 ppm was assigned to arabinose (H-2) attached at O-2 of xylose and the signal at 4.17 ppm was assigned to arabinose attached at O-3 of xylose (Hoffmann et al., 1992). The proton NMR spectra from these wheat varieties showed the complex structure of the polysaccharides and further substantiated methylation analysis.

# 3.4. FT-IR study of pentosans

IR spectra of arabinoxylans (pentosans) from different wheat varieties are illustrated in Fig. 2. All four spectra were characteristic of arabinoxylans (Kacurakova et al., 1994). Not much difference



in the spectra between good and poor varieties could be observed. The absorption at 1635 cm<sup>-1</sup> was principally associated with absorbed water, since the hemicelluloses have a strong affinity for water and in the solid state these macromolecules may have disordered structures, which can be easily hydrated (Kacurakova et al., 1994; Oraphin et al., 2004). Bands due to -CH<sub>2</sub> stretching vibrations were observed around 1420 cm<sup>-1</sup>. The prominent band around 1048 cm<sup>-1</sup> was attributed to C-O, C-C stretching or C-OH bending in arabinoxylans. The sharp band at 897 cm<sup>-1</sup> corresponding to the C-1 group frequency or ring frequency was characteristics of  $\beta$ -glycosidic linkages between the sugar units (Robert, Marquis, Barron, Guillon, & Saulnier, 2005). The bands which appeared between 3000 and 2500 cm<sup>-1</sup> represented the C-H stretching modes. The prominent band around 3380 cm<sup>-1</sup> is attributed to the hydroxyl stretching vibrations of the polysaccharides and water involved in hydrogen bonding (Nandini & Salimath, 2003). The band observed at 1045  $\text{cm}^{-1}$  was attributed to C–O, C-C and C-O-H bending vibrations. This band shows variation in spectral shape depending on the branches at O-2 and O-3 positions (Kacurakova et al., 1994).

#### 3.5. Periodate oxidation and Smith degradation

Periodate oxidation studies were carried out to determine the degree of substitution in arabinoxylans from different wheat varieties. Periodate consumed per mole of anhydrosugar was lowest in DWR-162 (0.64) followed by GW-322 (0.66, Table 3) indicating a higher degree of branching in these polysaccharides, which is also evident from their higher arabinose content (Table 1). Similar to this, highly branched arabinoxylans obtained from sorghum were shown to consume about 0.64 mol of periodate over 24 h of oxidation (Nandini & Salimath, 2002). Trace amounts of formic acid were released. Glycerol. arabinose and xylose were the major Smith degradation products (Table 4) identified. The products obtained by the Smith degradation further substantiated periodate oxidation results. Smith degradation analysis of the glucurono-arabinoxylans from sorghum showed high amounts of glycerol and xylose (Woolard, Rathbone, & Novellie, 1976). Similarly, arabinoxylans from native and malted ragi showed high amounts of glycerol and xylose (Shyamprasad Rao & Muralikrishna, 2007). Based on periodate oxidation and Smith degradation study of wheat arabinoxy-

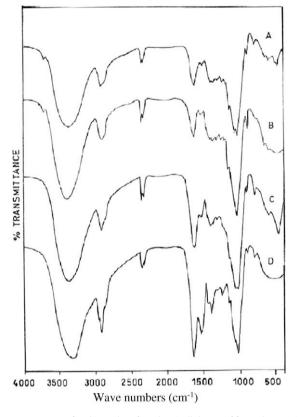


Fig. 2. FT-IR spectra of arabinoxylans from hemicellulose B of four wheat varieties (A: DWR-162, B: GW-322, C: MACS-2496, D: HD-2189).

#### Table 3

Moles of periodate consumed, optical rotation and ferulic acid contents of purified pentosans from hemicellulose B.

Wheat variety	Moles of periodate consumed / molecule of anhydrosugar	[α] <sub>D</sub>	Ferulic acid (µg/g)
DWR-162	0.64	-70.23	596.0
GW-322	0.66	-76.52	640.0
MACS-2496	0.80	-67.33	566.5
HD-2189	0.76	-65.42	545.4

 Table 4

 Analysis of Smith degradation products (%) obtained from purified pentosans from hemicellulose B.

Wheat variety	Glycerol	Threitol	Arabinose	Xylose
DWR-162	38.0	2.4	11.0	48.6
GW-322	33.2	2.8	12.5	51.5
MACS-2496	43.8	5.8	08.4	42.0
HD-2189	45.8	4.5	10.2	39.5

lans, Gruppen et al. (1992) reported that most of the branched residues were present as isolated units of blocks of two contiguous substituted xylose residues. Here, Smith degradation analysis of hemicellulose B showed a high amount of xylose for DWR-162 and GW-322, indicating their higher substitution.

# 3.6. Optical rotation measurements

Optical rotation values were  $-70.2^{\circ}$  and  $-76.5^{\circ}$  for DWR-162 and GW-322 respectively, while values for MACS-2496 and HD-

2189 were  $-67.4^{\circ}$  and  $-65.5^{\circ}$ , respectively (Table 3). A negative optical rotation value indicates a preponderance of  $\beta$ -linkages in the xylan backbone. These results are in agreement with the optical rotation values reported in the literature for arabinoxylans (Nandini & Salimath, 2002; Shyamprasad Rao & Muralikrishna, 2007).

#### 3.7. Ferulic acid content

The ferulic acid content  $(\mu g/g)$  of purified arabinoxylans is given in Table 3. GW-322 had the highest ferulic acid content (640  $\mu g/g$ ) followed by DWR-162 (596  $\mu g/g$ ). In wheat, ferulic acid is esterified to arabinose residues of cell wall arabinoxylans. Ferulic acid may be involved in cross-linking of cell wall polysaccharides in wheat, through ester and ether bonds. Apart from cross-linking, ferulic acid may also be involved in plant defence mechanisms and may have beneficial role as potent antioxidants (Izydorczyk & Biliaderis, 1995).

Chapati, the flat unleavened baked product prepared from wholewheat flour, is the main traditional staple food in the Indian sub-continent (Srivastava, Meyer, Haridas Rao, & Seibel, 2002). Wheat varieties, GW-322 and DWR-162 revealed good chapatimaking characteristics, while, MACS-2496 and HD-2189 had poor chapati-making quality, as reported earlier (Revanappa et al., 2007). Pentosans in different varieties of wheat are known to vary and relate to bread/chapati-making quality. The degree of branching of arabinoxylans (pentosans) may play an important role in the physicochemical properties of these constituents and branching is known to affect confirmation of these biopolymers in solutions (Courtin & Delcour, 2002).

Pentosans from hemicellulose B of wheat varieties showed the basic structure of xylan backbone in  $\beta$ -(1  $\rightarrow$  4) linkages, to which arabinose residues were attached at O-3 or O-2 and O-3 positions by α-linkages. Arabinoxylans differ not only among different cereals but also in different varieties. The variation was basically in the extent of branching by arabinosyl residues and their arrangement on the xylan backbone, resulting in differences in branching patterns (Izydorczyk & Biliaderis, 1995). Since the arabinoxylans have the capacity to retain water, flat breads made out of these doughs would have better palatability and pliability (Nandini & Salimath, 2003). Water absorption capacity of wheat dough becomes important during chapati making, because the dough is left for at least half an hour to swell. Water acts as a plasticiser of the glutenstarch composite matrix and lowers the rigidity of the products. Arabinoxylans which are low in substitution or unsubstituted form an insoluble complex, resulting in a low amount of water absorption (Izydorczyk & Biliaderis, 1995), which might lead to poor chapati-making characteristics (e.g., MACS-2496 and HD-2189), while, higher branched arabinoxylans (e.g., DWR-162 and GW-322) retain gas and water, roll well and may provide soft texture and pliability (Nandini & Salimath, 2003). Ferulic acid association with pentosan components has been known to be involved in oxidative gelation reaction (Neukom & Markwalder, 1978). Ferulic acid content was higher in good chapati varieties compared to poor varieties.

#### 4. Conclusions

Results presented in this investigation indicate that pentosans from DWR-162 and GW-322 (good chapati-making) are more branched compared to MACS-2496 and HD-2189 (poor chapatimaking) wheat varieties. A higher degree of ferulic acid substitution in arabinoxylans from GW-322 and DWR-162 wheat varieties may also be one of the contributing factors to the changes in functional properties, in terms of chapati-making quality.

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