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Effects of arabinoxylan solubilization on wort viscosity and filtration when mashing with grist containing wheat and wheat malt

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Abstract

Arabinoxylans are partially water-extractable, high-molecular-weight polymers that contribute to the problems of viscosity and membrane filterability during beer brewing. These problems are more pronounced when wheat and wheat malt are used as adjuncts due to their higher arabinoxylan contents and higher molecular weights. This paper aimed at investigating the effects of mashing temperature, time, grist size and liquor:grist ratio on the solubilization of arabinoxylans. Results indicated that increasing the mashing temperature generally increased the amount of arabinoxylans released into the wort. When greater proportions of wheat or wheat malt were used as adjunct, higher arabinoxylan contents in the final wort were observed. The more finely ground the grist, the more were arabinoxylans released into the wort. When more diluted mashes were used, more efficient solubilization of arabinoxylans was observed. The effects of arabinoxylan content and β-glucan content on the wort viscosity were also examined using a General Linear Model (GLM). There was a good correlation ($R^2 = 0.98$) between wort viscosity and its arabinoxylan and β-glucan contents.

Keywords: Arabinoxylans; Mashing; Solubilization; Viscosity; Wheat

1. Introduction

The major constituents of walls of the starchy endosperm in barley grain are the β-glucan and arabinoxylans (AX). In under-modified malts, as arabinoxylans cannot be degraded sufficiently, those may be problems, such as low rates of wort separation, high wort viscosity (Li, Lu, & Gu, 2005; Li, Lu, Gu, Shi, & Mao, 2004), decrease of beer microfiltration rate (Sadosky, Schwarz, & Horsely, 2002; Stewart, Hawthorne, & Evans, 1998), and haze formation in the brewery (Coote & Kirsop, 1976). Furthermore, incomplete degradation of endosperm cell walls reduces the yield of extract which can be derived during mashing. These brewing problems were more pronounced when wheat and wheat malt were used as adjuncs due to their higher arabinoxylan content and higher molecular weight (Cleemput, Roels, Vanoort, Grobet, & Delcour, 1993).

It was reported that German wheat beer had the highest level of arabinoxylans (4200 mg/l) (Schwarz & Han, 1995). However, there are few reports of solubilization of arabinoxylans in mashing with grist containing wheat and wheat malt.

Historically, reduced beer filtration efficiency has been mainly attributed to β-glucan, another important non-starch polysaccharide in barley grain. Sadosky et al. (2002) reported that the effects of arabinoxylans on viscosity and filterability were at least as important as the effects of β-glucan. Stewart et al. (1998) found that pilot-brewed beer viscosity and membrane filter-

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ability were correlated with arabinoxylan content, whereas β-glucan was correlated only with viscosity.

In our pervious work, the behaviour of arabinoxylans during malting and brewing has been extensively investigated (Li et al., 2005) and the effects of mashing parameters on the solubilization and hydrolysis of arabinoxylans have also been examined (Li, Lu, Gu, Shi, & Mao, 2005). And a mathematical model predicting arabinoxylan concentration during the mashing process was also developed (Li et al., 2004). However, the effects of arabinoxylan solubilization on wort viscosity and filtration, when mashing with grist containing wheat and wheat malt, were not considered in these studies.

In this work, we aimed at investigating the solubilization of arabinoxylans during mashing when wheat and wheat malt were used as adjuncts. Effects of mashing parameters, such as temperature, ratio of liquor to grist, and grist size, on the solubilization of arabinoxylans were also studied. A General Linear Model (GLM) with SAS software (version 8.1, SAS Institute, Cary, NC, USA) was employed to investigate the relationship between wort viscosity and arabinoxylan and β- glucan contents.

2. Materials and methods

2.1. Raw materials

Three grist samples were obtained from commercial malt factories. Harrington variety of barley was from Canada and malted in Dalian malting (Liaoning Province, China). Hard red winter wheat was cultivated in Henan Province and malted in Jinxing Brewery (Henan Province, China).

2.2. Small scale mashing

Fifty grams of grist were milled in a Buhler Miag disc mill, using a fine grind (0.2 mm) and coarse grind (0.7 mm).

Mashes were carried out in stirred metal beakers, using EBC Congress mash. Two hundred ml of water, at 45 °C, were added to the ground grist. The mash was continually stirred. After 30 min at 45 °C, the temperature was increased at the rate of 1 °C/min for 25 min, to 70 °C. More water (100 ml at 70 °C) was added. The temperature was maintained at 70 °C for 1 h. The mash was then cooled to 20 °C, stirred and rinsed, the rinsings going into the mash. The mash volume was adjusted to 450 ml by the addition of water at 20 °C.

2.3. Wort filtration rate

The filtration rate of wort samples was determined by transferring the mashed samples into a fluted filter paper. The wort filtration volume represented the volume of wort collected in 30 min following recirculation of the initial turbid wort.

2.4. Wort viscosity

Viscosity of the wort was measured at 20 °C using a falling ball viscometer (Hoppler, Germany).

2.5. Hot water extract (HWE)

Hot water extract was determined by feeding the wort sample into a density meter at 20 °C and, after conversion to specific gravity, the hot water extract were calculated as recommended (IOB, 1989).

2.6. Determination of β-glucan in wort

Determination of β- glucan content with Congo red dye was modified from the method of Li, Yin, Gu, and Lu (1997). β-Glucan samples (100 μl) were mixed with 3.0 ml of 100 mg/l Congo red dissolved in 0.1 M (pH 9.0) glycine–NaOH buffer. Absorbance at 550 nm was measured with a spectrophometer (UV-2100 spectrophotometer, Unico Co., Ltd., Shanghai) and double deionized water (100 μl) was mixed with 3.0 ml of 100 mg/l Congo red as a blank. β-Glucan concentrations in the range of 0–1000 mg/l at intervals of 100 mg/l were used to prepare calibration curves.

2.7. Determination of arabinoxylans in wort

Arabinoxylan contents were determined by measuring total monosaccharide composition. Monosaccharide composition of biological samples is widely determined by acid hydrolysis of the polysaccharides, followed by conversion of the monosaccharides to alditol acetates and analysis by gas chromatography. Worts were centrifuged at $3000 \times g$ for 10 min and hydrolyzed for 90 min with 2.0 M trifluoroacetic acid (121 °C) (Debyser, Derdelinckx, & Delcour, 1997a). Alditol acetates were prepared by the method of Englyst and Cummings (1984). Separation of the alditol acetates was with a Finnigan (GC–MS) chromatograph, using a SP-2330 column (30 m × 0.25 mm). The temperatures of injection and detection (flame ionization detector) were 260 and 280 °C, respectively. Arabinoxylan contents were calculated as $0.88 \times (\%\ \text{arabinose} + \%\ \text{xylose})$ (Henry, 1986).

2.8. Statistical analysis

Statistical analyses were carried out using Proc GLM from SAS (version 8.1, SAS Institute, Cary, NC, USA). Linear effects of arabinoxylans and β-glucan on viscosity were evaluated using Proc GLM. Significance was declared at $P < 0.01$.

3. Results and discussion

3.1. Effects of mashing temperature on the solubilization of arabinoxylans

Isothermal mashings at 45, 55, 65 and 75 °C for 2 h were carried out to study the effects of mashing temperature on the solubilization of arabinoxylans. Table 1 shows that increasing the mashing temperature generally increased the amount of arabinoxylans that were released into wort. When mashing temperature increased from 45 to 75 °C, arabinoxylan content varied from 792 to 996 mg/l for BM_{100} wort, 988 to 1128 mg/l for $BM_{60}WM_{40}$ wort, and 821 to 1008 mg/l for $BM_{60}W_{40}$ wort. These results showed that increasing the mashing temperature generally increased the amount of arabinoxylans released into the wort. This can be explained in that more arabinoxylans in grist were converted into hot water-soluble arabinoxylans at high temperature (Home, Stenholm, Wilhelmson, & Autio, 1999; Suhasini, Muralikrishna, & Malleshi, 1997). This could also be explained by destruction of enzymes that degrade arabinoxylans or promotion of activity of enzymes that solubilize arabinoxylans.

When 20% and 40% wheat malt were used as part of the grist in 45 °C isothermal mashing, the arabinoxylan contents in final wort were 938 and 988 mg/l, respectively. However, when 40% wheat malt was used for isothermal mashing at 75 °C, the arabinoxylan contents were rapidly increased to 1128 mg/l. This content was 42.4% and 13.3% higher than arabinoxylans contents in 100% barley malt wort at 45 °C (792 mg/l) and 75 °C (996 mg/l), respectively. When wheat was used as adjunct, more arabinoxylans were released into wort with increasing proportion of wheat in the grist composition. The arabinoxylan contents of BMW wort were consistently greater than that of BM wort but somewhat lower than that of BMWM wort. This can be explained in that some water-insoluble arabinoxylans were solubilized during malting and released into the final wort (Dervilly et al., 2002; Leclerq et al., 1999). However, further increase of mashing temperature (above 55 °C) did not significantly accelerate the solubilization of arabinoxylans into wort. This may be because wheat is unmalted grist and cell walls are not extensively degraded, limiting the solubilization of arabinoxylans. So when a greater proportion of wheat or wheat malt was used as adjunct, a higher arabinoxylan content in the final wort was observed. This result was in agreement with the report that wheat endosperm contains more arabinoxylans than barley endosperm (Ducroo & Frelon, 1989; Henry, 1986).

When 100% barley malt was used for isothermal mashing, with increasing mashing temperatures, higher levels of arabinoxylans in wort were accompanied by increased wort viscosities and reduced filtration volumes. These problems were more pronounced when wheat and wheat malt were used as adjuncts. When 40% wheat malt was used for isothermal mashing at 75 °C, the filtration volume gave a very small value of 95 ml and the wort viscosity reached the maximum value of 2.089 mPa s, much higher than that at 45 °C (1.398 mPa s). These data are consistent with the finding that more solubilization of arabinoxylans would in-

Table 1
Effects of mashing temperature on the solubilization of arabinoxylans

Temperature	Grist composition	HWE (L°/kg)	Filtration volume (ml)	Arabinoxylans (mg/l)	Viscosity (mPa s)
45 °C	BM_{100}	131.4	140	792	1.325
	$BM_{80}WM_{20}$	136.5	138	938	1.336
	$BM_{60}WM_{40}$	133.1	115	988	1.398
	$BM_{80}W_{20}$	139.0	132	803	1.334
	$BM_{60}W_{40}$	112.6	111	821	1.303
55 °C	BM_{100}	252.2	135	826	1.498
	$BM_{80}WM_{20}$	253.0	120	1051	1.562
	$BM_{60}WM_{40}$	240.6	110	1123	1.625
	$BM_{80}W_{20}$	223.2	120	841	1.516
	$BM_{60}W_{40}$	211.5	100	876	1.553
65 °C	BM_{100}	278.6	110	984	1.694
	$BM_{80}WM_{20}$	275.3	98	1038	1.772
	$BM_{60}WM_{40}$	276.1	90	1104	1.782
	$BM_{80}W_{20}$	263.8	103	994	1.718
	$BM_{60}W_{40}$	269.6	75	1012	1.750
75 °C	BM_{100}	289.3	104	996	1.993
	$BM_{80}WM_{20}$	278.6	97	1044	2.068
	$BM_{60}WM_{40}$	291.8	95	1128	2.089
	$BM_{80}W_{20}$	273.7	90	1001	2.074
	$BM_{60}W_{40}$	278.6	70	1008	2.077

BM_{100}, 100% barley malt; $BM_{80}WM_{20}$, 80% barley malt + 20% wheat malt; $BM_{60}WM_{40}$, 60% barley malt + 40% wheat malt; $BM_{80}W_{20}$, 80% barley malt + 20% wheat; $BM_{60}W_{40}$, 60% barley malt + 40% wheat.

crease the wort viscosity and retard wort filtration. However, another important reason is that there may be a difference in molecular weight of the arabinoxylans as, the water-extractable arabinoxylans of barley malt (Debyser, Schooneveld-Bergmans, Derdelinckx, Grobet, & Delcour, 1997) have a much lower molecular weight than those from wheat (Cleemput et al., 1993) and, as the malt xylanolytic system is (to a certain degree) inactivated by wheat (Debyser, Derdelinckx, & Declour, 1997b). Highest HWE value was obtained at 75 °C with grist containing 40% wheat malt, which corresponded to the highest arabinoxylan content (1128 mg/l) in wort.

3.2. *Effects of grist size and liquor:grist ratio on the solubilization of arabinoxylans*

EBC congress mash was employed for investigating the effects of grist size and liquor-grist ratio on the solubilization of arabinoxylans. The effect of grist coarseness on arabinoxylan solubilization was studied using a fine grist (0.2 mm) and a coarse grist (0.7 mm) and the effects of liquor:grist ratio were also examined (Tables 2–4). These data show that the more finely ground the grist, the more arabinoxylan is released into wort, accompanied by increasing wort viscosity and decreasing filtration rate.

Table 2
Effects of grist particle size and grist:liquor ratio (1:3) on the solubilization of arabinoxylans

Grist composition	Liquor (ml)	Miag mill setting (mm)	HWE (L°/kg)	Filtration volume (ml)	Arabinoxylans (mg/l)	Viscosity (mPa s)
BM_{100}	150	0.2	289.3	110	890	1.621
$BM_{80}WM_{20}$	150	0.2	278.6	104	935	1.689
$BM_{60}WM_{40}$	150	0.2	291.8	105	944	1.768
$BM_{80}W_{20}$	150	0.2	273.7	80	737	1.676
$BM_{60}W_{40}$	150	0.2	278.6	63	926	1.744
BM_{100}	150	0.7	285.2	55	856	1.593
$BM_{80}WM_{20}$	150	0.7	250.6	59	904	1.651
$BM_{60}WM_{40}$	150	0.7	277.0	58	937	1.725
$BM_{80}W_{20}$	150	0.7	259.7	50	667	1.602
$BM_{60}W_{40}$	150	0.7	264.6	52	687	1.712

BM_{100}, 100% barley malt; $BM_{80}WM_{20}$, 80% barley malt + 20% wheat malt; $BM_{60}WM_{40}$, 60% barley malt + 40% wheat malt; $BM_{80}W_{20}$, 80% barley malt + 20% wheat; $BM_{60}W_{40}$, 60% barley malt + 40% wheat.

Table 3
Effects of grist particle size and grist:liquor ratio (1:4) on the solubilization of arabinoxylans

Grist composition	Liquor (ml)	Miag mill setting (mm)	HWE (L°/kg)	Filtration volume (ml)	Arabinoxylans (mg/l)	Viscosity (mPa s)
BM_{100}	200	0.2	283.5	170	1155	1.606
$BM_{80}WM_{20}$	200	0.2	286.0	150	1312	1.683
$BM_{60}WM_{40}$	200	0.2	290.1	90	1496	1.754
$BM_{80}W_{20}$	200	0.2	288.5	145	1106	1.704
$BM_{60}W_{40}$	200	0.2	277.8	115	1233	1.744
BM_{100}	200	0.7	284.4	66	1126	1.594
$BM_{80}WM_{20}$	200	0.7	260.5	55	1246	1.607
$BM_{60}WM_{40}$	200	0.7	287.7	58	1361	1.733
$BM_{80}W_{20}$	200	0.7	280.3	40	1048	1.646
$BM_{60}W_{40}$	200	0.7	255.5	55	1013	1.708

BM_{100}, 100% barley malt; $BM_{80}WM_{20}$, 80% barley malt + 20% wheat malt; $BM_{60}WM_{40}$, 60% barley malt + 40% wheat malt; $BM_{80}W_{20}$, 80% barley malt + 20% wheat; $BM_{60}W_{40}$, 60% barley malt + 40% wheat.

Table 4
Effects of grist particle size and grist:liquor ratio (1:5) on the solubilization of arabinoxylans

Grist Composition	Liquor (ml)	Miag mill setting (mm)	HWE (L°/kg)	Filtration Volume (ml)	Arabinoxylans (mg/l)	Viscosity (mPa s)
BM_{100}	250	0.2	278.6	70	1238	1.625
$BM_{80}WM_{20}$	250	0.2	292.6	85	1413	1.698
$BM_{60}WM_{40}$	250	0.2	290.1	106	1538	1.772
$BM_{80}W_{20}$	250	0.2	273.7	104	945	1.685
$BM_{60}W_{40}$	250	0.2	279.4	90	1194	1.719
BM_{100}	250	0.7	270.4	40	1142	1.573
$BM_{80}WM_{20}$	250	0.7	286.8	36	1038	1.635
$BM_{60}WM_{40}$	250	0.7	272.9	38	1080	1.713
$BM_{80}W_{20}$	250	0.7	273.7	47	791	1.607
$BM_{60}W_{40}$	250	0.7	255.5	41	797	1.673

BM_{100}, 100% barley malt; $BM_{80}WM_{20}$, 80% barley malt + 20% wheat malt; $BM_{60}WM_{40}$, 60% barley malt + 40% wheat malt; $BM_{80}W_{20}$, 80% barley malt + 20% wheat; $BM_{60}W_{40}$, 60% barley malt + 40% wheat.

100% barley malt, along with wheat and wheat malt used as adjuncts, was examined when grist:liquor ratio varied from 1:3 to 1:5. When mashes were thinner, lower arabinoxylan contents in final wort were observed. This can be explained in that, since a certain amount of water is bound by starch, the water phase in thick mashes becomes concentrated, limiting the solubilization of arabinoxylans. When 40% wheat malt was used for mashing and grist:liquor ratio was 1:5, using a fine grist, the arabinoxylan content in final wort reached a maximum value of 1538 mg/l, much higher than that content under the condition that grist: liquor ratio was 1:3 (944 mg/l). Generally, at the same grist coarseness and grist:liquor ratio, greter arabinoxylan contents were observed with the increase of wheat malt proportion in the grist composition, which is consistent with the results of Table 1.

Similarly to the isothermal mashings, wort viscosities and filtration volume are accompanied by changes of arabinoxylans contents. In Table 4, the highest wort viscosity value of 1.772 mPa s was found, which corresponded to the highest arabinoxylan content of 1538 mg/l.

3.3. *Effects of mashing time on the solubilization of arabinoxylans*

EBC congress mash was employed to evaluate the effect of mashing time on arabinoxylans solubilization with different grist compositions (Figs. 1–3). Samples were taken at intervals of 20 min and immediately cooled to 4 °C to prevent further sugar solubilization.

Fig. 1 shows the effects of mashing time on the solubilization of arabinoxylans and β- glucan when 100% barley malt was used. It is possible that endo-xylanase and β-glucanase in malt at low mashing temperature (45 °C) were still active, resulting in the degradation of arabinoxylans and β-glucan, accompanied by relatively low wort viscosity. So, during the first 20 min, there was no significant increase in the amount of arabinoxylans and β-glucan extracted into the wort. During the remainder of the mashing, arabinoxylan content increased continually and, consequently, reached a maximum value of 1154 mg/l in the final wort. However, the rapid release of β-glucan was observed only after mashing for 40 min (mashing temperature of 55 °C). The β-glucan content in final wort reached 539 mg/l, much lower than the arabinoxylan content (1154 mg/l). This is consistent with the report of Schwarz and Han (1995) that the amount of arabinoxylans in beer was greater than that of β-glucan. Wort viscosity increased from 1.286 to 1.685 mP as.

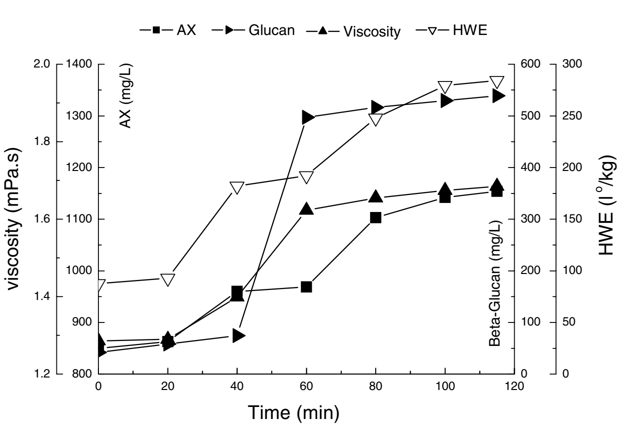

Fig. 1. Effect of mashing time on the solubilization of arabinoxylans and β-glucan when 100% barley malt was used.

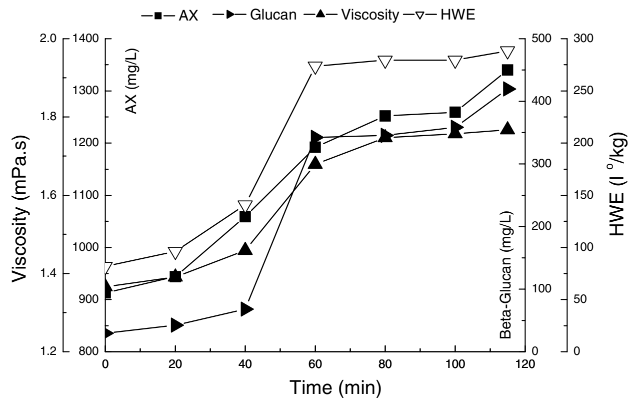

Fig. 2. Effect of mashing time on the solubilization of arabinoxylans and β-glucan when 60% barley malt and 40% wheat malt were used.

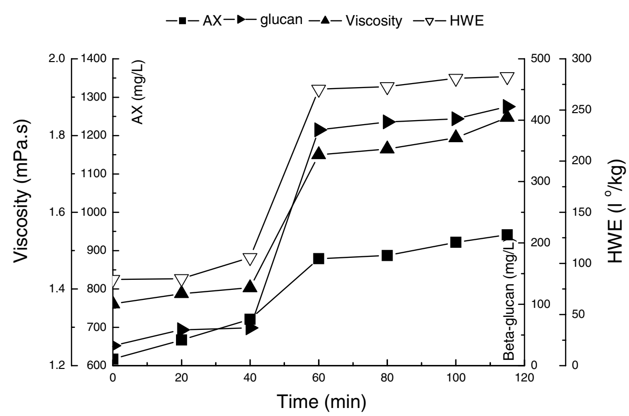

Fig. 3. Effect of mashing time on the solubilization of arabinoxylans and β-glucan when 60% barley malt and 40% wheat were used.

When 40% wheat malt was included, as part of the grist, the amount of arabinoxylans immediately dissolved was higher (Fig. 2). Again there was a minor increase in arabinoxylan content and β-glucan content over the first 20 min, but this was followed by a rapid solubilization of arabinoxylans and β-glucan over the remaining mashing period. In final wort, the contents of arabinoxylans and β-glucan were 1340 and 420 mg/l, respectively. The arabinoxylans content was higher than that when 100% barley malt was used for mashing, while β-glucan content was lower than the value in that condition. This can be explained in that non-starch polysaccharides in wheat malt mainly consisted of arabinoxylans but more β-glucan was found in barley malt (Henry, 1986). Wort viscosity in final wort reached the highest value of 1.767 mPa s, above that of 100% barley malt wort (1.685 mPa s).

When 40% wheat was used as adjunct (Fig. 3), the arabinoxylan content (941 mg/l) in wort was much lower than those contents of the above two grist compositions (1154 and 1340 mg/l, respectively). This indicated that arabinoxylans in wheat was not extensively degraded and most of the arabinoxylans were insoluble, resulting in relatively low arabinoxylan contents during mashing. The β-glucan content in wort (422 mg/l) was similar to the case of replacement by wheat malt (420 mg/l). However, wort viscosity (1.847 mPa s) was much higher than those of the other two grist compositions (1.685 and 1.767 mPa s, respectively). This result is probably explained by the finding that high-molecular-weight arabinoxylans in unmalted wheat were not extensively degraded during the mashing process. So, higher high-molecular-weight arabinoxylans proportion in $BM_{60}W_{40}$ wort could result in increase of wort viscosity (Leclerq et al., 1999).

3.4. Effects of arabinoxylans and β-glucan contents on the wort viscosity

Polynomial regression analysis showed a linear effect ($P < 0.01$) of arabinoxylan content and β-glucan content on the wort viscosity.

When 100% barley malt is used for mashing, the equation for estimating the linear relationship is as follows:

$$\text{Wort viscosity (mPa s)} = 0.955 + 3.79 \times 10^{-4}[\text{AX}] + 5.59 \times 10^{-4}[\text{BG}], \qquad (1)$$

where [AX] is arabinoxylan content during mashing, [BG] is β-glucan content during mashing, $R^2 = 0.98$, coefficient variance = 1.82, $F = 130.39$, and $P < 0.01$.

When 40% wheat malt is used as adjunct for mashing, the equation is as follows:

$$\text{Wort viscosity (mPa s)} = 0.947 + 4.44 \times 10^{-4}[\text{AX}] + 6.24 \times 10^{-4}[\text{BG}], \qquad (2)$$

where $R^2 = 0.98$, coefficient variance = 1.73, $F = 127.65$, and $P < 0.01$.

When 40% wheat is used as adjunct for mashing, the equation is as follows:

$$\text{Wort viscosity (mPa s)} = 1.089 + 3.71 \times 10^{-4}[\text{AX}] + 9.04 \times 10^{-4}[\text{BG}], \qquad (3)$$

where $R^2 = 0.99$, coefficient variance = 1.12, $F = 436.0$, and $P < 0.01$.

The close R^2 to 1, large F-values, and small values of coefficient of variance, implied that the models were strong and could well predict the linear relationship, and at least 98% of the experimental variation could be explained by them. The three model equations showed that wort viscosity was strongly influenced by arabinoxylan content and β-glucan content during mashing.

In the past, many workers have investigated the effect of β-glucan on wort viscosity and wort filtration. It is reported (Fincher & Stone, 1986) that arabinoxylans can form highly viscous solutions, especially in the presence of β-glucan. However, the evidence that these substituted arabinoxylans alone, or synergistically with β-glucan (Viëtor, Voragen, & Angelino, 1993) could result in filtration problems, is scarely reported. Further researches, such as the effects of arabinoxylan structure and molecular weight on wort viscosity and membrane filtration will be carried out in our laboratory.

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