Production and Characterisation of Alcohol-Insoluble Dietary Fibre as a Potential Source for Functional Carbohydrates Produced by Enzymatic Depolymerisation of Buckwheat Hulls

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Abstract


We examined the potential use of buckwheat hulls as a raw material for producing soluble dietary fibre. The insoluble fibre fraction obtained from buckwheat hulls was hydrolysed by two commercial enzymes (Celluclast 1.5L for the cellulose fraction and Viscozyme L for the hemicellulose fraction) to obtain soluble fibre hydrolysates. Alcohol-insoluble dietary fibre (AIF) was separated from the freeze-dried soluble hydrolysate by treatment with 85% ethanol. The water-holding, oil-binding, and swelling capacities of AIF were increased by enzymatic hydrolysis. AIF had significantly (P < 0.05) higher functional properties than the control. AIF from the hemicellulose fraction effectively hindered the diffusion of glucose and bile acid from dialysis membranes, and had a significantly (P < 0.05) greater bile acid inhibitory effect than carboxymethylcellulose or pectin. It can be concluded that AIF from buckwheat hulls by enzymatic hydrolysis can be used as dietary supplement and additive in the food industry.

Keywords: agricultural by-products; enzymatic hydrolysis; hypoglycaemic effect; hypolipidaemic effect; soluble dietary fibre

Dietary fibre is resistant to hydrolysis by human digestive enzymes and consists of polysaccharides, oligosaccharides, and lignin. The potential health benefits of dietary fibre include reduction of blood cholesterol, blood glucose level regulation, and anti-carcinogenic effects (Kim 2000; Dongowski 2007; Zacherl et al. 2011). Epidemiological studies have revealed that diets containing high levels of dietary fibre have many health-related benefits such as the maintenance of gastrointestinal functional integrity along with reduced risks of colon cancer and coronary heart disease (Davidson & McDonald 1998; Cui et al. 2011). Dietary fibre is frequently used to develop functional foods (Puupponen-Pimia et al. 2002) due to its importance in nutrition and health. Dietary fibre is also added to various food products as a functional ingredient or acceptable factor (Yangilar 2013). The addition of fibre helps modify and improve the texture, sensory characteristics, and shelf life of foods due to its water-binding capacity and gel-forming ability as well as fat mimetic, anti-adhesive, texturising, and thickening effects (Thebaudin & Lefebvre 1997; Staffolo et al. 2004). The addition of dietary fibre to beverages increases viscosity and stability. Soluble fibre is the most frequently used for this purpose because it is more dispersible in water than insoluble fibre. Some examples of soluble fibres are those derived from grains, fruit, pectins, cellulose beetroot fibre, and polydextrose (Bollinger 2001; Rodríguez et al. 2006). The by-products of agricultural industries are of interest because they are inexpensive potential sources of dietary fibre that are available in large quantities (Chantaroro et al. 2008). Agri-food residues, mainly consisting of peels, pulps, stems, cores, and seeds, are lignocellulosic-rich materials.

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Buckwheat has been cultivated for food and medicinal applications, and has recently received increasing attention for its antioxidant, hypocholesterolaemic, and anti-diabetic activities (Qin et al. 2010). Buckwheat tea is a commercially available buckwheat-based food product, and has gained popularity in China, Japan, Republic of Korea, and Europe (Wang et al. 2013). Buckwheat is used in combination with wheat, rice, or maize in the food industry to produce cakes, noodles, breakfast cereals, soups, and porridges (Hromádková & Ebringerová 2003). For this, whole buckwheat seeds are subjected to dehulling which separates the seeds into about 70% of dehulled groats and 30% of hull (Wang et al. 2013). Most of the buckwheat bran is discarded even though the hull is rich in flavonoids (rutin and quercetin) and dietary fibre (cellulose and hemicellulose). As one kind of agricultural waste, buckwheat hulls have a low economic value and create serious disposal problems. We conducted the present study to investigate the possibility of using buckwheat hulls to produce water-soluble dietary fibre by enzymatic hydrolysis. In addition, we also evaluated the physical and health-related properties of the resulting soluble fibre hydrolysate.

MATERIAL AND METHODS

Material. Buckwheat hulls were dried at 60°C for 24 h, finely ground to 0.05-mm in size using a food mixer (FM-681C; Hanil, Incheon, Korea), and stored in a deep freezer at -40°C prior to use. The cellulose and hemicellulose fractions were obtained using a method described by Xu et al. (2006) with slight modifications. Buckwheat hull powder (30 g dry weight) was blended with 300 ml of 1 M NaOH for 3 min, shaken at 250 rpm for 3 h at 25°C, and centrifuged at 16 270 g for 20 minute. The residue was washed with distilled water until the wash water pH became neutral, freeze-dried, and used as the cellulose fraction. The supernatant was acidified to pH 5.0 with concentrated HCl and used as the hemicellulose fraction for the enzymatic hydrolysis experiments. Two commercial enzymes, Celluclast 1.5L and Viscozyme L (both from Novozymes A/S, Bagsvaerd, Denmark), were used to hydrolyse the cellulose and hemicellulose fractions, respectively. Enzyme assays were performed using the dinitrosalicylic acid (DNS) method previously described by Miller (1959). The cellulase activity of Celluclast 1.5L was 396.8 units per ml, and the xylanase activity of Viscozyme L was 639.8 units/ml. One unit of cellulase activity was defined as the amount of enzyme that produced 1 mmol glucose from 0.1% carboxymethylcellulose (Sigma Chemical Co., St. Louis, USA) solution per min at 50°C and pH 5.0. One unit of xylanase activity was defined as the amount of enzyme that produced 1 mmol of xylose from 0.1% birch wood xylan (Sigma Chemical Co.) solution per min at 40°C and pH 5.0.

Enzymatic hydrolysis. The experiments were conducted in 500-ml Erlenmeyer flasks containing the cellulose or hemicellulose fraction to produce soluble fibre. For enzymatic hydrolysis of the cellulose fraction, 10 g of the dried fraction were added to 250 ml of 50 mM sodium acetate buffer (pH 5.0), and then 0.1 ml (8.4 units) of 0.1% Celluclast 1.5L was added to the mixture. The solution was agitated at 200 rpm in a shaking incubator (KMC-8480SF; Vision Scientific Co., Seoul, Republic of Korea) for 74 h at 40°C. For hydrolysis of the hemicellulose fraction, 250 ml of the fraction (pH 5.0) was mixed with 30 units of Viscozyme L. The flasks were agitated at 150 rpm on a shaking incubator at 40°C and pH 5.0 for a total of 72 hours. Samples were taken at 24-h intervals and boiled to inactivate the enzyme activity at 90°C for 5 min, filtered, and freeze-dried.

Separation of AIF from enzymatic hydrolysate. The AIF was separated from the enzymatic hydrolysate as previously described by Yoon et al. (2005) with slight modification. The freeze-dried sample was dissolved in 85% ethanol at 80°C, incubated for 40 min at 60°C, and filtered. The residue was dissolved in distilled water and used as AIF for further experiments. AIF obtained from the cellulose and hemicellulose fractions was designated AIFC and AIFH, respectively. The yields of AIFC and AIFH were determined based on dry weight.

Water-holding capacity (WHC) and oil-binding capacity (OBC) of AIF. The WHC and OBC were determined according to Bencini (1986) with slight modifications. Freeze-dried AIFs (2 g) were mixed with 20 ml distilled water or maize oil in 50-ml centrifuge tubes. Each slurry was vortexed for 1 min, allowed to stand at room temperature for 30 min, and then centrifuged at 3000 rpm for 15 minutes. The results were expressed as ml of liquid retained per g of sample.

Swelling capacity of AIF. Swelling is defined as the ratio of the volume occupied when the sample is immersed in an excess of water after equilibra-
tion to the actual weight. Freeze-dried AIF (0.1 g) was hydrated in a known volume of distilled water (10 ml) containing 0.02% azide as a bacteriostat in a calibrated cylinder at room temperature. After equilibration (18 h) at room temperature, the bed volume was recorded and expressed as volume/g original sample dry weight (Robertson et al. 2000).

**The effect of AIF on glucose transport.** Glucose retardation indices (GRIs) are used to predict the inhibitory effects of AIF on glucose absorption in the gastrointestinal tract. GRIs were measured as previously described by Adiotomre et al. (1990). Ten-centimetre lengths of dialysis bags (Sigma, D7884: MW cut-off ≤ 1200) were soaked in 0.1% sodium azide solution for 24 h and filled with 6 ml of 0.1% sodium azide solution containing 36 mg of glucose with or without (control) 0.2 g of AIF; each fibre type had been previously hydrated in an aqueous solution of 0.1% sodium azide solution for 14 hours. Each bag was tied, suspended in 100 ml of 0.1% sodium azide, and placed in a stirring bath at 37°C for 4 hours. The glucose content in 1 ml of the dialysate was analysed using the DNS method at 30 min as well as 1, 2, and 4 hours. The GRI values were then calculated using the following equation:

\[
\text{GRI value} \% = 100 - \left[ \frac{\text{Total glucose diffused from bag containing fibre}}{\text{Total glucose diffused from bag without fibre}} \right] \times 100
\]

**The effect of AIF on bile acid transport.** Bile acid retardation indices (BRIs) are used to monitor the effect of fibre on sterol metabolism (Adiotomre et al. 1990). The BRI values were determined in the same manner as GRIs except that 50 mM phosphate buffer at pH 7.0 with 0.1% sodium azide and 15 mM taurocholic acid was used alone (control) or with the addition of 0.2 g AIF that had been hydrated for 14 h in the buffered taurocholate-containing solution. Each bag was then placed in 100 ml of phosphate buffer containing 0.1% sodium azide, and dialysis was performed at 37°C for 8 hours. A 2-ml dialysate sample was removed for analysis at 1, 2, 4, and 8 hours. The bile acid content in the dialysate was measured by calculating the taurocholic acid content as previously described by Boyd et al. (1966). BRIs were used to assess the effect of AIF on cholesterol metabolism. BRI values were calculated using the following equation:

\[
\text{BRI value} \% = 100 - \left[ \frac{\text{Total bile acid diffused from bag containing fibre}}{\text{Total bile acid diffused from bag without fibre}} \right] \times 100
\]

The AIF effect on glucose or bile acid diffusion was compared with that of standard dietary fibre samples such as pectin (Fluka-Biochemika, Buchs, Switzerland) or carboxymethylcellulose (CMC) (Sigma Chemical Co.).

**α-Glucosidase inhibitory activity.** α-Glucosidase inhibitory activity was evaluated as previously described by Phan et al. (2013) with modifications. For this, 50 µl of test sample (in 50 g/100 ml DMSO) was mixed with 50 µl of potassium phosphate buffer (50 mM, pH 6.8) prior to the addition of 100 µl of yeast α-glucosidase (0.2 U/ml; Sigma Chemical Co.). One unit of enzyme activity was defined as the amount of enzyme required to recover 1 µmol of ρ-nitrophenol from α- D-glucopyranoside (ρ-NPG; Sigma Chemical Co.) per min under the assay conditions. The mixture was pre-incubated at 37°C for 15 min before the addition of 100 µl of ρ-NPG (3 mM) to start the enzyme reaction with further incubation at 37°C for 20 minutes. The reaction was terminated by the addition of 100 µl of 0.1 M NaOH, and absorbance was measured at 405 nm using a microplate reader (EPOCH; BioTek Instrument Inc., Winooski, USA). Acarbose (Sigma Chemical Co.) was used as the positive control for the assay. Change of absorbance was monitored before and after incubation. Percent inhibitory activity was calculated with the following formula:

\[
\% \text{ inhibition} = \frac{\left[ \frac{\text{Absorbance of control}}{\text{Absorbance of sample}} \right] - 1 }{\text{Absorbance of control}} \times 100
\]

**Statistical analysis.** Statistical analyses were performed using SPSS v. 18.0 (Chicago, USA). All data are expressed as the mean ± standard deviation (SD) of triplicate experiments. An analysis of variance (ANOVA) was performed. Significant differences (P < 0.05) between mean values were identified using Duncan’s multiple range test.

**RESULTS AND DISCUSSION**

**Soluble fibre yield.** The AIFC and AIFH yields from buckwheat hulls for each commercial cellulase or xylanase preparation are presented in Table 1. Enzymatic hydrolysis was conducted for 72 h under the optimal conditions. AIFC recovery significantly (P < 0.05) increased from 2.41 g/kg to 5.36 g/kg after 24 h of reaction, but there was no significant increase after 48 h of hydrolysis. The AIFH yield increased significantly (P < 0.05) with reaction time. For exam-
Table 1. Yields of water-soluble dietary fibre from buckwheat hulls by enzymatic hydrolysis (g/kg dry matter)

<table>
<thead>
<tr>
<th>Hydrolysis time (h)</th>
<th>AIFC</th>
<th>AIFH</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>2.41 ± 0.12&lt;sup&gt;b&lt;/sup&gt;</td>
<td>84.30 ± 2.19&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td>24</td>
<td>5.36 ± 0.41&lt;sup&gt;a&lt;/sup&gt;</td>
<td>104.44 ± 3.56&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>48</td>
<td>5.70 ± 0.52&lt;sup&gt;a&lt;/sup&gt;</td>
<td>112.89 ± 1.78&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>72</td>
<td>6.05 ± 0.24&lt;sup&gt;a&lt;/sup&gt;</td>
<td>123.70 ± 2.89&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

AIFC – alcohol-insoluble fibre produced from the cellulose fraction by enzymatic hydrolysis; AIFH – alcohol-insoluble fibre produced from the hemicellulose fraction by enzymatic hydrolysis; each value represents the mean ± standard deviation of three experiments; values with different superscript letters in the same column are significantly different at P < 0.05.

Table 2. Physical properties of alcohol-insoluble dietary fibre (AIF) produced from buckwheat hulls by enzymatic hydrolysis (ml/g)

<table>
<thead>
<tr>
<th>Hydrolysis time (h)</th>
<th>Water-holding capacity</th>
<th>Oil-binding capacity</th>
<th>Swelling capacity</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>AIFC</td>
<td>AIFH</td>
<td>AIFC</td>
</tr>
<tr>
<td>0</td>
<td>1.67 ± 0.31&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.70 ± 0.10&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.67 ± 0.31&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>24</td>
<td>2.43 ± 0.25&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.97 ± 0.21&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.03 ± 0.21&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>48</td>
<td>3.03 ± 0.25&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.50 ± 0.17&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.50 ± 0.10&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>72</td>
<td>3.43 ± 0.15&lt;sup&gt;c&lt;/sup&gt;</td>
<td>2.33 ± 0.23&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.70 ± 0.26&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

AIFC – alcohol-insoluble fibre produced from the cellulose fraction by enzymatic hydrolysis; AIFH – alcohol-insoluble fibre produced from the hemicellulose fraction by enzymatic hydrolysis; each value represents the mean ± standard deviation of three experiments; values with different superscript letters in the same column are significantly different at P < 0.05.
form viscous solutions. They also adsorb and retain other substances like minerals as well as non-polar molecules (such as fats and bile acid) or glucose (Lecumberri et al. 2007).

**The effect of AIF on glucose diffusion.** GRI values were used to compare the inhibitory effects of dietary fibre on glucose diffusion, which are most likely due to increased viscosity (López et al. 1996; Peerajit et al. 2012). In Table 3, variations in glucose diffusion rates and GRI upon the addition of AIF or commercial dietary fibre preparations versus a control are shown. As the dialysis time increased from 0.5 h to 4 h, glucose contents in the dialysates containing different fibre samples increased from 6.49–6.68 mg/100 ml (30 min of dialysis) to 21.68–26.75 mg/100 ml (4 h of dialysis). All fibres had greater inhibitory effects on the flow of glucose across the dialysis bag compared to the control. The GRIs of fibres were 15.8–18.7% after 0.5 h of dialysis but there was not a significant variation between the samples. After 1 h of dialysis, CMC had the greatest GRI with 24.1% followed by AIFC (16.0%), pectin (14.6%), and AIFH (11.5%).

Afterward, the inhibitory effects were increased with longer dialysis time, and the maximum GRI values of all samples except for AIFH were obtained after 2 h of dialysis. GRI of the samples decreased thereafter. In contrast to our results, most previous studies have shown that dietary fibre has maximum glucose retardation index values after 30 min of dialysis and these values diminish after an extended dialysis time (Chau et al. 2003; Peerajit et al. 2012). We believe that this discrepancy is most likely due to differences in viscosity. Compared to the control, AIF decreased the amount of diffused glucose in the dialysate although AIFC hindered glucose diffusion less than CMC or pectin. A previous study indicated that the inhibition of glucose diffusion and absorption by fibre is affected by viscosity of the intestinal contents (Edwards et al. 1987), and the inhibitory effects of AIF might be due to the glucose adsorption capacity of AIF. Based on these results, it is conceivable that AIF could delay glucose diffusion and postpone glucose absorption in the gastrointestinal tract.

**The inhibitory effect of AIF on bile acid.** The BRI was used to assess the effect of fibre on sterol metabolism. Variations in bile acid diffusion after adding AIF compared to CMC, pectin, and the control were also evaluated, and the results are presented in Table 2. Taurocholic acid contents in the dialysate containing various fibre samples ranged from 83.4 to 93.8 µmol/l (1 h) to 281.3–345.0 µmol/l (at 8 h). Taurocholic acid levels in the dialysate of the control were 109.2 µmol/l in 1 h and 482.3 µmol/l at 8 hours. Compared to the control, AIF decreased the amount of taurocholic acid that diffused into the dialysate. In particular, AIF more effectively prevented bile acid from diffusing out of the dialysis membrane compared to CMC or pectin. AIFC had the greatest inhibitory effect (23.5%) on taurocholic acid diffusion after dialysis for 1 h followed

### Table 3. Inhibitory effect of AIF produced from buckwheat hulls by enzymatic hydrolysis on the dialysis membrane transport of glucose

<table>
<thead>
<tr>
<th>Sample</th>
<th>Dialysis time (h)</th>
<th>0.5</th>
<th></th>
<th>1</th>
<th></th>
<th>2</th>
<th></th>
<th>4</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>glucose in dialysate (mg/100 ml)</td>
<td>GRI (%)</td>
<td>glucose in dialysate (mg/100 ml)</td>
<td>GRI (%)</td>
<td>glucose in dialysate (mg/100 ml)</td>
<td>GRI (%)</td>
<td>glucose in dialysate (mg/100 ml)</td>
<td>GRI (%)</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>7.97 ± 0.17b</td>
<td>0</td>
<td>16.16 ± 0.50a</td>
<td>0</td>
<td>23.09 ± 0.79a</td>
<td>0</td>
<td>28.27 ± 0.50a</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>CMC</td>
<td>6.49 ± 0.23b</td>
<td>18.6</td>
<td>12.27 ± 0.61c</td>
<td>24.1</td>
<td>17.31 ± 0.28d</td>
<td>25.0</td>
<td>21.68 ± 0.11c</td>
<td>23.3</td>
<td></td>
</tr>
<tr>
<td>Pectin</td>
<td>6.71 ± 0.17b</td>
<td>15.8</td>
<td>13.79 ± 0.11b</td>
<td>14.6</td>
<td>19.01 ± 0.40c</td>
<td>17.6</td>
<td>24.57 ± 0.55d</td>
<td>14.2</td>
<td></td>
</tr>
<tr>
<td>AIFC</td>
<td>6.65 ± 0.23b</td>
<td>16.7</td>
<td>13.57 ± 0.40b</td>
<td>16.0</td>
<td>19.23 ± 0.78c</td>
<td>16.7</td>
<td>24.97 ± 0.28c</td>
<td>11.6</td>
<td></td>
</tr>
<tr>
<td>AIFH</td>
<td>6.68 ± 0.11b</td>
<td>16.3</td>
<td>14.31 ± 0.34b</td>
<td>11.5</td>
<td>20.57 ± 0.68b</td>
<td>10.9</td>
<td>26.75 ± 0.57b</td>
<td>5.4</td>
<td></td>
</tr>
</tbody>
</table>

AIFC – alcohol-insoluble fibre produced from the cellulose fraction by enzymatic hydrolysis; AIFH – alcohol-insoluble fibre produced from the hemicellulose fraction by enzymatic hydrolysis; CMC – carboxymethylcellulose; each value represents the mean ± standard deviation of three experiments; values with different superscript letters in the same column are significantly different at P < 0.05; % – ratio of glucose in dialysate to total glucose added; GRI – glucose retardation index = 100 – [(Glucose content in dialysate with fibre addition/Glucose content in dialysate in the absence of fibre) ×100]
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Table 4. Inhibitory effect of AIF produced from buckwheat hulls by enzymatic hydrolysis on the dialysis membrane transport of bile acid

<table>
<thead>
<tr>
<th>Sample</th>
<th>Dialysis time (h)</th>
<th>1 (µmol/l)</th>
<th>BRI (%)</th>
<th>2 (µmol/l)</th>
<th>BRI (%)</th>
<th>4 (µmol/l)</th>
<th>BRI (%)</th>
<th>8 (µmol/l)</th>
<th>BRI (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td></td>
<td>109.2 ± 6.0&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.0</td>
<td>177.7 ± 7.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.0</td>
<td>293.5 ± 3.5&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.0</td>
<td>482.3 ± 8.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.0</td>
</tr>
<tr>
<td>CMC</td>
<td></td>
<td>87.3 ± 6.8&lt;sup&gt;b&lt;/sup&gt;</td>
<td>20.0</td>
<td>142.5 ± 10.1&lt;sup&gt;b&lt;/sup&gt;</td>
<td>19.9</td>
<td>222.0 ± 6.3&lt;sup&gt;b&lt;/sup&gt;</td>
<td>24.4</td>
<td>345.0 ± 16.0&lt;sup&gt;b&lt;/sup&gt;</td>
<td>28.3</td>
</tr>
<tr>
<td>Pectin</td>
<td></td>
<td>93.8 ± 7.6&lt;sup&gt;b&lt;/sup&gt;</td>
<td>14.1</td>
<td>129.8 ± 9.1&lt;sup&gt;b&lt;/sup&gt;</td>
<td>27.0</td>
<td>209.9 ± 0.2&lt;sup&gt;c&lt;/sup&gt;</td>
<td>28.5</td>
<td>328.3 ± 2.6&lt;sup&gt;b&lt;/sup&gt;</td>
<td>31.9</td>
</tr>
<tr>
<td>AIFC</td>
<td></td>
<td>83.4 ± 3.6&lt;sup&gt;c&lt;/sup&gt;</td>
<td>23.5</td>
<td>122.0 ± 6.0&lt;sup&gt;c&lt;/sup&gt;</td>
<td>36.9</td>
<td>205.3 ± 8.9&lt;sup&gt;c&lt;/sup&gt;</td>
<td>30.1</td>
<td>305.1 ± 3.0&lt;sup&gt;c&lt;/sup&gt;</td>
<td>36.7</td>
</tr>
<tr>
<td>AIFH</td>
<td></td>
<td>87.8 ± 2.6&lt;sup&gt;b&lt;/sup&gt;</td>
<td>19.4</td>
<td>99.0 ± 4.9&lt;sup&gt;c&lt;/sup&gt;</td>
<td>44.3</td>
<td>181.0 ± 1.5&lt;sup&gt;d&lt;/sup&gt;</td>
<td>38.3</td>
<td>281.3 ± 4.5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>41.7</td>
</tr>
</tbody>
</table>

AIFC – alcohol-insoluble fibre produced from the cellulose fraction by enzymatic hydrolysis; AIFH – alcohol-insoluble fibre produced from the hemicellulose fraction by enzymatic hydrolysis; CMC – carboxymethylcellulose; each value represents the mean ± standard deviation of three experiments; values with different superscript letters in the same column are significantly different at P < 0.05; % – ratio of bile acid in dialysate to total bile acid added; BRI – bile acid retardation index = 100 – [(Taurocholic acid content in dialysate with fibre addition/Taurocholic acid content in dialysate without fibre) × 100]

by CMC (20.0%), AIFH (19.4%), and pectin (14.1%). The maximum BRI value of AIF was reached after 2 h of dialysis. The BRIs of AIFC and AIFH were 44.3% and 36.9%, respectively, at 2 hours. In contrast, the BRIs of CMC and pectin gradually increased during dialysis. After 8 h of dialysis, the BRIs of CMC and pectin were 28.3 and 31.9%, respectively. The BRI of AIF from buckwheat hull is higher than that of standard dietary fibre during dialysis. Specifically, AIFC had the highest BRI value among the samples. The AIF derived from apples, strawberries, rowan berries, carrots, white cabbage, red beets, and sugar beet pulp has a high binding capacity for three types of bile acids whereas cellulose has less of an effect (Dongowski 2007). These findings are consistent with our present results. Many studies have demonstrated that various types of soluble dietary fibres reduce the total and LDL cholesterol concentrations in blood, and that the cholesterol-reducing effect of water-soluble dietary fibre is greater than that of water-insoluble dietary fibre (Wood 1994; Kritchevsky 1995; Zacherl et al. 2011). Water-soluble fibre is believed to act predominantly by binding the water in chyme and increasing the viscosity (Zacherl et al. 2011). These findings suggest that AIF consumption might reduce the total and LDL cholesterol levels in serum.

**α-Glucosidase inhibitory activity.** α-Glucosidase is a key enzyme involved in the release of glucose from starch for intestinal glucose absorption. Inhibition of this enzyme decreases blood glucose levels and represents an important strategy for managing type 2 diabetes (Plus et al. 1977). α-Glucosidase inhibitors derived from natural food sources are an attractive strategy for controlling postprandial hyperglycaemia (Girish et al. 2012). As shown in Table 5, the α-glucosidase inhibitory activity of AIF was elevated according to increased hydrolysis time and concentration. For example, the inhibitory activities of AIFC and AIFH increased from 61.92 and 69.68% (before hydrolysis) to 89.14 and 84.74% (72 h of hydrolysis), respectively, at a concentration of 100 µg/ml. After 24 h of hydrolysis, the inhibitory effects of AIFC and AIFH increased from 34.67 and 40.80% (at 30 µg/ml) to 92.16 and 93.21% (at 500 µg/ml), respectively. AIF hydrolysed for 48 h had high activity (more than 90%) at a concentration of 300 µg/ml; this was significantly (P < 0.05) greater than that (19.54%) of acarbose used as the positive control. Findings from Ou et al. (2001) showed that fibres could lower postprandial serum glucose levels through several mechanisms such as hindering glucose diffusion, adsorbing glucose to decrease the concentration of glucose available in the small intestine, retarding carbohydrate action, and directly inhibiting the enzyme. Natural enzyme inhibitors are likely to offer an attractive therapeutic approach to treating postprandial hyperglycaemia due to fewer abdominal side effects arising from excessive inhibition of α-glucosidase by synthetic drugs such as acarbose, miglitol, and metformin (Sangeethapiyya & Sidduraju 2014). Therefore, results from our investigation indicated that AIF rich in soluble dietary fibre might
Table 5. α-Glucosidase inhibitory activity of AIF produced from buckwheat hulls by enzymatic hydrolysis

<table>
<thead>
<tr>
<th>Sample</th>
<th>Concentration (µg/ml)</th>
<th>Time (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>30</td>
</tr>
<tr>
<td>AIFC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>20.23 ± 0.74d</td>
<td>24.88 ± 0.37d</td>
</tr>
<tr>
<td>24</td>
<td>34.67 ± 0.52e</td>
<td>52.76 ± 0.65e</td>
</tr>
<tr>
<td>48</td>
<td>37.14 ± 0.63d</td>
<td>68.46 ± 0.50b</td>
</tr>
<tr>
<td>72</td>
<td>39.04 ± 0.75d</td>
<td>70.18 ± 0.41a</td>
</tr>
<tr>
<td>AIFH</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>28.91 ± 0.66f</td>
<td>48.25 ± 0.68f</td>
</tr>
<tr>
<td>24</td>
<td>40.80 ± 0.30c</td>
<td>57.8 ± 0.95d</td>
</tr>
<tr>
<td>48</td>
<td>45.27 ± 0.56b</td>
<td>65.32 ± 0.54c</td>
</tr>
<tr>
<td>72</td>
<td>55.41 ± 3.65a</td>
<td>70.69 ± 0.49a</td>
</tr>
<tr>
<td>Acarbose</td>
<td>11.73 ± 1.15b</td>
<td>15.55 ± 0.31b</td>
</tr>
</tbody>
</table>

AIFC – alcohol-insoluble fibre produced from the cellulose fraction by enzymatic hydrolysis; AIFH – alcohol-insoluble fibre produced from the hemicellulose fraction by enzymatic hydrolysis; each value represents the mean ± standard deviation of three experiments; values with different superscript letters in the same column are significantly different at P < 0.05.

decrease the rate of glucose absorption as well as the concentration of postprandial serum glucose. AIF might also help manage type 2 diabetes.

**CONCLUSIONS**

Data from this study demonstrated the feasibility of producing soluble dietary fibre via enzymatic hydrolysis of buckwheat hulls recovered during processing. AIF has high water-binding, oil-binding, and swelling capacities. This fibre effectively hindered the diffusion of glucose and bile acid from the dialysis membrane. Our results indicated that the soluble fibre hydrolysate from buckwheat hulls is a practical new material with hypoglycaemic and hypolipidemic effects that may be used in the food industry to prepare functional drinks and nutraceutical products. The use of agro-industrial waste to produce bioactive ingredients could improve the economic value of such waste. It is undoubtedly comprehensible that a more detailed investigation for producing on a larger scale is needed to prove its values.

**References**


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