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Food Research International

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## Changes in dietary fiber fractions and gut microbial fermentation properties of wheat bran after extrusion and bread making



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#### ARTICLE INFO

Article history: Received 30 January 2015 Received in revised form 28 April 2015 Accepted 2 May 2015 Available online 8 May 2015

Keywords: Butyrate Short chain fatty acids Screw speed Moisture Arabinoxylans Non-starch polysaccharides

#### ABSTRACT

The dietary fiber in wheat bran, principally non-starch polysaccharides (NSP), is mostly water-unextractable and is poorly utilized by human gut microbiota. The purpose of this study was to determine the change in water-extractability of NSP in wheat bran upon extrusion and then to determine if extrusion impacts the availability of NSP for fermentation by the fecal microbiota during in vitro fecal fermentation. A secondary objective was to incorporate extruded bran into a product formulation to determine if changes in WE-NSP and NSP fermentation were maintained in a finished product. Bran was extruded using combinations of high or low moisture (15% and 30% wb) and high or low screw speed (120 and 250 rpm). All extrusion conditions resulted in increases in WE-NSP and fecal microbiota short chain fatty acid (SCFA) production upon fermentation compared with unextruded bran. Low screw speed and low moisture resulted in the greatest increase in WE-NSP (3-fold) as well as the highest production of SCFA during fermentation (1.4-fold) compared with unextruded bran. Whole wheat breads containing extruded bran did not show increases in either WE-NSP or SCFA production compared with the control. In conclusion, extrusion of wheat bran increased WE-NSP, which enabled greater fermentability by human fecal microbiota. However, once extruded bran was used in a whole wheat bread formulation the changes in fermentation outcomes were no longer evident.

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#### 1. Introduction

Dietary fibers can be classified based on their solubility. Dietary fiber solubility is related to its chemical and physical structure and can define its physiological functionality. For instance, although wheat bran is a rich source of fiber, its fermentation in the human colon is not complete; carbohydrate consumption after 24 h of fermentation ranges from 15% to 25% (Bourquin, Titgemeyer, & Fahey, 1996; Karppinen, Liukkonen, Aura, Forssell, & Poutanen, 2000). However, when the insoluble fiber from wheat is treated with enzymes to make it soluble, it becomes highly fermentable with prebiotic properties (Napolitano et al., 2009).

Final products of dietary fiber fermentation in the human intestine are short chain fatty acids (SCFA). These metabolites are the primary energy source for colonic cells, serve as modulators in the immune response, and act as signaling molecules for several hormones involved in metabolism and appetite (Russell, Hoyles, Flint, & Dumas, 2013).

Extrusion combines mechanical shear and temperature to disrupt cell wall structures (Robin, Dubois, Pineau, Schuchmann, & Palzer, 2011). Improvements in dietary fiber solubility during extrusion of

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wheat bran have been reported before with varying results. Ralet, Thibault, and Della Valle (1990) showed that processing wheat bran at 100 °C, 240 rpm and 6.1% moisture conditions increases soluble dietary fiber from 8% to 16%. Wang, Klopfenstein, and Ponte (1993) reported maximum fiber solubility increase of 2.5-fold after processing wheat bran at high moisture (28.8%) and high screw speed (400 rpm). Less marked effects were reported by Kahlon, Berrios, Smith, and Pan (2006), who obtained a maximum of 1% increase (from 2.1% to 3.1%) in soluble fiber in wheat bran using a twin-screw extruder operated at 400 rpm and barrel temperatures of 80 to 130 °C. Gualberto, Bergman, Kazemzadeh, and Weber (1997) obtained increases in soluble fiber of less than 1% (from 3.1% to 3.5%) when wheat bran was extruded using a twin-screw extruder at 200 rpm, 15% moisture and a barrel temperature of 122 °C.

Although increases in dietary fiber solubility upon extrusion have been reported, the possible positive impact on fiber fermentability by human fecal microbiota has not been described. Furthermore, additional changes in fiber fractions once extruded wheat bran is further processed into a food product have not been investigated. Thus, there were two objectives to this study. First, changes in extractability and in vitro fecal fermentation of non-starch polysaccharides (NSPs), the principle components of dietary fiber in wheat bran, following extrusion were determined. Second, differences in extractability of NSP and

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fermentation properties of bread made from reconstituted whole wheat flour containing extruded wheat bran were evaluated to determine if the changes observed on the bran alone were maintained in the final bread product.

#### 2. Materials and methods

#### 2.1. Wheat bran samples

Hard red winter wheat (*Triticum aestivum* 'Wesley') was obtained from Husker Genetics, the University of Nebraska–Lincoln's Foundation Seed Division. Wheat kernels were tempered overnight to 15% moisture followed by milling on a laboratory mill (Bühler model MLU-202, Uzwil, Switzerland) according to approved method 26-21.02 (AACC International, 2015). Dial settings for break rolls were 10 left, 8.5 right, and for reduction rolls were 7 left, 3 right. Milling fractions obtained were 74.7% straight-grade flour, 2.3% shorts and 23.0% coarse bran (Fig. 1). The shorts and coarse bran were combined and used as 'bran' in this study.

#### 2.2. Extrusion of wheat bran

A 2 × 2 factorial design was used to test the effect of screw speed and bran moisture on WE-NSP. High screw speed (250 rpm) and low screw speed (120 rpm) and high moisture (30% wet basis) and low moisture (15% wet basis) were chosen to generate different processing conditions (Fig. 1). Bran moisture was adjusted 24 h before the experiment by the addition of water using a manual trigger spray bottle while mixing by hand. Once all water was added, the bucket containing the sample was closed and shaken well. Samples were stored in closed containers at 4 °C until extrusion. The experiment was conducted in a single screw KE 19 Brabender laboratory extruder with a single stage mixing zone, 3:1 compression ratio, 3 mm die diameter, and a 20:1 L/D ratio (CW Brabender Instruments, NJ, USA). The extruder was operated by a direct current drive unit (Intelli-Torque, Pastic Corder Lab-station, C.W. Brabender) with a motor of 7.5 hp. The bran was fed into the extruder using a volumetric feeder (FW 40 Plus, C. W. Brabender) set at a constant flow rate of ~50 g/min. Barrel temperatures were set at 80 °C (zone 1; inlet), 120 °C (zone 2), and 120 °C (zone 3; die assembly). All treatment combinations were performed in duplicate. Following extrusion, the samples were dried in an oven at 70 °C overnight, and then ground in a cyclone mill (UDY Corporation, Co, USA) equipped with a 1 mm mesh screen. Control bran was also milled using the same equipment.

#### 2.3. Characterization of wheat bran

The milled extruded and unextruded bran were assayed for protein (approved method 46-30.01), lipids (approved method 30-25.01) and ash (approved method 08-01.01; AACC International, 2015). Total starch was measured using a total starch assay kit (K-TSTA, Megazyme, Bray, Ireland) following the procedure for samples that contain resistant starch but do not contain free glucose (KOH format). Total dietary fiber, was determined as the sum of neutral sugars residues, uronic acid residues and Klason lignin following approved method 32-25.01 (AACC International, 2015) with some modifications. Briefly, 300 mg of sample, 3 mL of acetate buffer (0.1 M, pH 5, containing 5 mM CaCl<sub>2</sub>) and 25  $\mu$ L thermostable  $\alpha$ -amylase (3000 U/mL, Megazyme) were combined in a 15 mL test tube and then incubated at 60 °C overnight. Tubes were then centrifuged at  $1000 \times g$ , for 10 min, the supernatant was transferred to a 50 mL tube and the pellet was washed by suspending and re-centrifuging with water (2 × 3 mL). The collected supernatants contained the water-extractable NSP (WE-NSP), which was precipitated by adding 35 mL absolute ethanol while mixing. The tubes were allowed to stand for 1 h in an ice bath, centrifuged at  $1000 \times g$  for 10 min, and the supernatant was discarded. The pellet

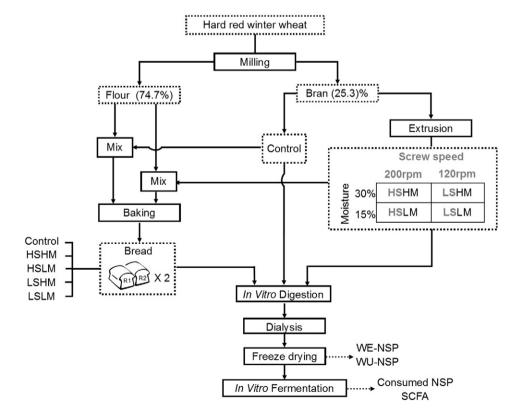


Fig. 1. Overview of the experiment; light gray color denotes high screw speed (HS) and low screw speed (LS); black color denotes high moisture (HM) and low moisture (LM); WE = water-extractable; WU = water-unextractable; NSP = non-starch polysaccharides; and SCFA = short chain fatty acids.

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