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Bioprocessing of bran with exopolysaccharide producing microorganisms as a tool to improve expansion and textural properties of extruded cereal foams with high dietary fibre content

Markus Nikinmaa [a, *], Syed Ariful Alam [a], Mari Raulio [a], Kati Katina [a, b], Ilkka Kajala [a], Emilia Nordlund [a], Nesli Sozer [a]

[a] *VTT Technical Research Centre of Finland, PO Box 1000, FI-02044 Espoo, Finland*
[b] *Department of Food and Environmental Sciences, University of Helsinki, PO Box 66, FI-00014 Helsinki, Finland*

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ABSTRACT

High dietary fibre levels, especially insoluble that is typical for cereal bran, have been associated with poor structural, textural and sensory properties in extruded products. The effect of fermentation with baker's yeast, fermentation with a mixture of *Kazachstania exigua* and *Lactobacillus brevis* with and without added hydrolytic enzymes, as well as with *Weissella confusa*, on the structural and textural properties of high dietary fibre extrudates (ca. 6–14 g/100 g added fibre) in extrusion was studied. Superior structural and textural extrudate characteristics were achieved by fermentation of bran with dextran producing *W. confusa*. At 40 g/100 g addition of *W. confusa* –treated bran (12 g/100 g fibre content) radial expansion was the same as for the pure rye endosperm flour control, while density was 35% lower. Hardness (54.1 → 16.3 N) and crispiness work (4.11 → 0.45 Nmm) were reduced ($P < 0.05$), while the crispiness index was significantly higher (0.002 → 0.05) than that of the control extrudate. Bioprocessing with mixed fermentation, containing *K. exigua and L. brevis*, together with hydrolytic enzymes also improved the structural and textural characteristics of the bran in extrusion, while baker's yeast fermentation did not significantly affect these characteristics.

1. Introduction

Dietary fibre (DF) intake has been associated with several positive health effects, including improved cardiovascular health, lower risk of certain cancers, as well as prevention and treatment of obesity and type 2 diabetes (Murphy et al., 2012; Weickert & Pfeiffer, 2008). Despite this, intake of DF remains below recommended levels (25–38 g/day) (EFSA Panel on Dietetic Products Nutrition and Allergies (NDA), 2010). However, increased health awareness has created a demand for palatable high DF food products (Van der Kamp & Lupton, 2013). Extruded snacks and breakfast cereals is a food category where refined ingredients (e.g. endosperm cereal flours, corn and potatoes) with low DF content are mostly utilized (Brennan, Derbyshire, Tiwari, & Brennan, 2013), and thus a promising food category for increasing DF content, e.g. by addition of cereal brans that have naturally high DF content (Nordlund et al., 2012). Unfortunately, high DF and especially insoluble DF that is typical for cereal bran has been associated with poor structural and textural properties in extruded products (Sozer & Poutanen, 2013). Radial expansion decreased with addition of wheat bran fibre (5–24.4 g/100 g) (Brennan, Monro, & Brennan, 2008; Robin, Dubois, Pineau, Schuchmann, & Palzer, 2011; Yanniotis, Petraki, & Soumpasi, 2007), soy fibre (10–30 g/100 g) (Jin, Hsieh, & Huff, 1995), sugar beet fibre (10–30 g/100 g) (Lue, Hsieh, & Huff, 1991), corn bran (11–20 g/100 g DF) (Mendonça, Grossmann, & Verhé, 2000) and oat bran (20 g/100 g bran and 10 g/100 g DF) (Chassagne-Berces et al., 2011; Sibakov et al., 2014), whereas longitudinal expansion increased with sugar beet (30 g/100 g) and wheat fibre (12.6 and 24.4 g/100 g DF) (Lue et al., 1991; Robin et al., 2011) addition. Inferior textural characteristics, e.g. increased hardness (Brennan et al., 2008; Chassagne-Berces et al., 2011; Yanniotis et al., 2007) and reduced crispiness (Chassagne-Berces et al., 2011; Mendonça et al., 2000) were linked to increased bubble rupture (Yanniotis et al., 2007) and impaired

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* Corresponding author.
E-mail address: markus.nikinmaa@vtt.fi (M. Nikinmaa).

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visco-elastic properties of the melt (Robin et al., 2012).

Research focusing on improving structural and textural properties of extruded matrices with high DF content and especially using cereal bran as DF source is scarce. Particle size reduction of rye bran (440 → 28 μm) was reported to improve the expansion of extruded rye bran (Alam et al., 2014), and particle size reduction (702 → 84 μm) and hydrolytic enzyme treatment of wheat bran improved density and texture of high DF (15.8–16.4 g/100 g) extrudates (Santala, Kiran, Sozer, Poutanen, & Nordlund, 2014). In contrast, no effect on expansion of wheat bran by particle size reduction (317 → 224 μm) was observed in extrudates where the DF content was 12.6–24.4 g/100 g (Robin et al., 2011) possibly due to the small variation in particle size of bran as compared to the studies of Alam et al. (2014) and Santala et al. (2014). Soluble DF, e.g. inulin (5–15 g/100 g), guar gum (5–15 g/100 g) or pectin (5–10 g/100 g), was reported to exhibit better structural (e.g. expansion) and textural (e.g. hardness) properties in extrusion than insoluble DF (Brennan et al., 2008; Yanniotis et al., 2007). Moreover, improvement in the expansion of corn bran added to corn meal (26 g/100 g DF) was observed after treatment with NaOH due to solubilisation of bran DF (Pai, Blake, Hamaker, & Campanella, 2009).

Fermentation with starter cultures is a tool to improve the processability of bran. Fermentation-induced modification of bran has been linked to acidification and solubilisation of bran DF components (Coda et al., 2014; Katina et al., 2007, 2012; Salmenkallio-Marttila, Katina, & Autio, 2001). Certain lactic acid bacteria (LAB) also secrete EPS into the environment. EPS have been reported to function as hydrocolloids in baking and to improve rheological properties of dough and texture of bread (Galle & Arendt, 2014; Katina et al., 2009). Despite marked technological improvements of cereal raw material by fermentation in baking applications, thus far no studies on behaviour of fermented bran in extrusion have been reported. Therefore, the aim of this study was to elucidate how fermentation affects the structural (expansion, specific length and density) and textural (hardness and crispiness) properties of rye bran in extrusion. To achieve this, bran was bioprocessed with yeast, mixed fermentation with yeast and lactic acid bacteria with and without added hydrolytic enzymes, as well as EPS producing LAB, after which the behaviour of bran in cereal high DF extrusion was examined.

2. Materials and methods

2.1. Microbial strains and enzymes

Microbial strains used were *Lactobacillus brevis* (VTT E-95612), *Kazachstania exigua* (VTT *C-81116)* and dextran producing *Weissella confusa* (VTT E-133279) from VTT Culture Collection (VTT Technical Research Centre of Finland), as well as baker's yeast *Saccharomyces cerevisiae* (Suomen Hiiva, Finland). Starter cultures of *K. exigua*, *L. brevis* and *W. confusa* were prepared for bioprocessing. Frozen *K. exigua* was revitalized in YM (yeast mold) medium (3 g/L malt extract, 3 g/L peptone, 10 g/L dextrose), before being subcultured in wort broth. Frozen *L. brevis* and *W. confusa* were revitalized in MRS (de Man, Rogosa and Sharpe) broth (Oxoid LTD, Basingstoke, Hampshire, United Kingdom), before subculturing in GEM (general edible medium, containing 2 g/100 mL glucose and sucrose, 3 g/100 mL soy peptone, 0.7 g/100 mL yeast extract, 0.1 g/100 mL MgSO4 in 0.01 mol/L pH 6.3 potassium phosphate buffer). Enzyme preparations used were Depol 740L (Biocatalysts, Pontypridd, UK) and Grindamyl A1000 (Danisco, Copenhagen, Denmark). Depol 740L was analysed to have xylanase activity of 10464 nkat/mL (as described by Bailey, Biely, & Poutanen, 1992), but it also contained other hydrolase activities as reported by Hartikainen, Poutanen, and Katina (2014). Grindamyl A1000 was amylase with α-amylase activity of 120673 nkat/g (Hartikainen et al., 2014).

2.2. Extrusion raw materials

High DF rye bran material was prepared by milling and air classification. Native and polished rye kernels (Jalon Mylly, Kouvola, Finland) were milled with a 100 UPZ-lb Fine impact mill (Hosokawa Alpine AG, Germany) with stainless steel pin discs (17,800 rpm), followed by air classification (speed 3500 rpm, airflow 220 m^3/h, feed rate 50 kg/h) (British Rema Minisplit, Chesterfield, UK) to remove starch from the bran. The procedure was repeated for the coarse material (34%). The coarse fraction after the second air classification (23%) was classified as bran. Rye flour (starch base for extrusion) was from Helsingin Mylly (Järvenpää, Finland) and exogenous dextran was from Pharmacosmos (T2000, Denmark). This dextran was produced by fermentation with *Leuconostoc mesenteroides* and had an average molecular weight of $1.5 \times 10^6 – 2 \times 10^6$ g/mol. The degree of alpha-1,3-branching was approximately 5% and the branches were mostly 1–2 glucose units long.

2.3. Bioprocessing of bran

Four different bran bioprocessing treatments, fermentation with baker's yeast (Y), mixed fermentation with *K. exigua* and *L. brevis* with and without added enzymes (M and ME respectively), as well as fermentation with EPS producing *W. confusa (W)*, were performed (Table 1). Bioprocessing was performed with a bran:water ratio of 22:78. In the *W. confusa* treatments 10 g/100 g of the bran was substituted for sucrose as substrate for dextran production. Inoculum size for all microbes was 10^{6-7} cfu/g. Enzyme dosages were 1.688 g/100 g of bran weight for Depol 740L and 0.062 g/100 g of bran weight for Grindamyl A1000. Samples were manually mixed at the beginning with no further mixing during fermentation. Fermentation times and temperatures are shown in Table 1. Samples for microbial, pH and TTA-analysis were taken at the beginning and the end of fermentation. Fermented bran was dried in a Christ Epsilon 2–25 freeze drier (Martin Christ Gefriertrocknungsanlagen GmbH, Osterode am Harz, Germany). Freeze-dried samples were ground in 100 UPZ-lb Fine impact mill (Hosokawa Alpine AG, Germany) with stainless steel pin discs (17,800 rpm).

2.4. Microbiological and chemical analysis of bran

Total DF content was measured according to AOAC method no. 985.29 (Association of Official Analytical Chemists (AOAC) (1990)). Insoluble and soluble pentosan content was determined as described by Santala et al. (2014). Microbiological analysis, as well as pH and TTA analyses were carried out at the beginning and the end of fermentation as described by Kajala et al. (2016). Dextran content was analysed as described by Katina et al. (2009).

2.5. Extrusion

Two bran addition levels were used in extrusion, 20 g/100 g and 40 g/100 g. Untreated native and polished bran, yeast fermented native and polished bran, as well as *K. exigua* and *L. brevis* fermented polished bran (with and without enzymes), were added at both 20 g/100 g and 40 g/100 g concentration. *W. confusa* fermented polished bran and 5 g/100 g exogenous dextran supplemented polished bran were added at only 20 g/100 g concentration, while *W. confusa* fermented native bran was added only at 40 g/100 g concentration. Rye flour was used as starch matrix. A rye flour control sample was also included in the experiments as a reference