LWT - Food Science and Technology 66 (2016) 211–216

Contents lists available at ScienceDirect

LWT - Food Science and Technology

journal homepage: www.elsevier.com/locate/lwt

# Effects of added inulin and wheat gluten on structure of rye porridge

José L. Vázquez-Gutiérrez*, Daniel Johansson, Maud Langton

*Department of Food Science, SLU – Swedish University of Agricultural Sciences, PO Box 7051, SE-750 07 Uppsala, Sweden*

ARTICLE INFO

*Article history:*
Received 1 July 2015
Received in revised form
10 October 2015
Accepted 13 October 2015
Available online 19 October 2015

*Keywords:*
Rye
Starch
Microscopy
Fructan
Viscosity

ABSTRACT

The aim was to study the microstructure and distribution of components of rye porridge enriched with different inulin and gluten proportions (0:0, 3:9, 6:6, 9:3), and their relationship with texture. Inulin was labeled with fluorescein isothiocyanate (FITC) prior to its addition to the porridges, and multiple staining was applied to cryosections in order to also observe other components of the porridges. Porridge structure consisted of grain fragments and a continuous phase formed by released amylose, starch granules and protein. Addition of inulin and gluten to rye porridge partly hindered starch gelatinization due to their water binding capacity. The green fluorescence from FITC-labeled inulin was brighter in detached starch granules in the continuous phase, indicating greater interaction of inulin with starch than with protein. Viscosity was lower in those porridges with high inulin content and low gluten content. Solubilized inulin created a protective layer around starch granules limiting their swelling and amylose release, which may explain the differences in viscosity between the porridges and could have further influence in starch digestibility.



## 1. Introduction

Consumers are demanding healthier food products with improved functionalities and an increasing number of new food formulations are being developed to satisfy this need. The compatibility or incompatibility between ingredients in new food formulations can affect both texture and structure (Icoz, Moraru, & Kokini, 2005). These structural changes could also have later implications in the intended functionality of the product (McClements, Decker, Park, & Weiss, 2009). When it comes to porridge, the rheological properties are of great importance for quality control and consumer acceptance (Sai Manohar, Urmila Devi, Bhattacharya, & Venkateswara Rao, 2011). Moreover, it can also have influence on the satiating properties of the product (Mars, Hogenkamp, Gosses, Stafleu, & De Graaf, 2009). Rye foods, which are important elements in the healthy Nordic diet, have shown favorable effects on appetite (Isaksson et al., 2012), as well as beneficial effects on postprandial insulin responses and inflammatory biomarkers (Fung et al., 2002; Landberg et al., 2010; Rosén, Östman, & Björck, 2011). Addition of plant protein and fermentable dietary fiber could possibly enhance the appetite suppressing effect of whole-grain rye porridge. Such effects may in part be due to alterations in the microstructure of the product (Lundin, Golding, & Wooster, 2008).

Inulin is an oligo-fructose polymer of interest in human nutrition due to its ability to act as dietary fiber and prebiotic (Roberfroid, 2007). Due to its structure, inulin resists digestion in the human intestine and is fermented by bacteria in the colon, which has been suggested to affect appetite (Cani, Dewever, & Delzenne, 2004). Little work has been done to investigate the effects of inulin on food structure. Microstructural studies of inulin-enriched products have been carried out on cereal and dairy products (Aravind, Sissons, Fellows, Blazek, & Gilbert, 2012; Guardeño, Vázquez-Gutiérrez, Hernando, & Quiles, 2013; Guggisberg, Cuthbert-Steven, Piccinali, Bütikofer, & Eberhard, 2009; Rodríguez-García, Puig, Salvador, & Hernando, 2012; Sołowiej et al., 2015). However, the studies do not provide a detailed localization of solubilized inulin in the structure and only insolubilized inulin crystals have been detected by light microscopy (Guardeño et al., 2013). Interactions between inulin and the protein structural network in yogurt have been suggested (Guggisberg et al., 2009; Kip, Meyer, & Jellema, 2006), but such interactions have neither been properly described nor confirmed by microstructural observations.

Gluten is found in the endosperm of cereals such as wheat, barley, and rye and is an important by-product from wet milling of wheat flour. Wheat gluten is a common food ingredient in bakery products such as hamburger buns (Esteller, Pitombo, & Lannes,

* Corresponding author.
*E-mail address:* jose.luis.vazquez.gutierrez@slu.se (J.L. Vázquez-Gutiérrez).

http://dx.doi.org/10.1016/j.lwt.2015.10.034
0023-6438/

2005), meat products as binding and enriching ingredient (Zhang, Xiao, Samaraweera, Lee, & Ahn, 2010), breakfast cereals, and pasta (Day, 2011). Wheat gluten is marketed in two forms: 'non-vital' and 'vital'. Nonvital wheat gluten has undergone irreversible denaturation, while vital dry gluten in contact with water rehydrates rapidly and regains its intrinsic functionality (Esteller et al., 2005). Therefore, vital gluten constitutes a desired additive in baked and meat products due to its ability to form a viscoelastic mass through the interaction with water (Esteller et al., 2005; Zhang et al., 2010). Interactions between gluten and starch have been reported and supported by microscopy observation (Chen, Deng, Wu, Tian, & Xie, 2010). It has also been suggested that there could be interactions between gluten and inulin but this has not been confirmed by microstructural observation (Morris & Morris, 2012; Peressini & Sensidoni, 2009; Rubel, Pérez, Manrique, & Genovese, 2015; Wang, Rosell, & Benedito de Barber, 2002).

Labeling of inulin with fluorescein isothiocyanate (FITC) has been successfully used for studies of the phase behavior of inulin-waxy maize starch systems (Zimeri & Kokini, 2003a). To our knowledge, the method has so far only been used for model systems and this is the first time that FITC labeling and localization of inulin by confocal microscopy is performed in a complex food system. Previously only inulin crystals could be identified and the location of soluble inulin could only be suggested in such systems, not proven by fluorescence signal as in this study.

The aim of this study was to analyze the effect of partial substitution of rye flakes for inulin and gluten on the microstructure and texture of whole grain rye flake porridge to obtain a better understanding of the functionality of the product.

## 2. Materials and methods

### 2.1. Sample preparation

Rye porridge was made from whole grain rye flakes, produced by steaming, cutting and rolling rye kernels (Lantmännen Cerealia, Järna, Sweden). Four different samples were prepared, one with 40 g rye flakes and the rest contained 40 g rye flakes with different combinations of inulin (Orafti®GR inulin, purity 90%; Beneo, Mannheim, Germany) and gluten (Vital Wheat Gluten, purity 77%; Arrowhead Mills, Boulder, USA). The combined additions were recalculated to compensate for impurities to ensure ratios inulin/gluten of 1:3 (3 g inulin and 9 g gluten, 3I9G), 1:1 (6 g inulin and 6 g gluten, 6I6G) and 3:1 (9 g inulin and 3 g gluten, 9I3G), as well as similar total weight of all the porridges. Samples were prepared by adding boiling water (150 g) to the rye flakes/inulin/gluten mixtures and manually stirred for 30 s. The samples were then left to rest for 2 min and manually stirred again for another 30 s. The samples were left to rest for another 2 min, and then deposited in aluminum caps and frozen with liquid nitrogen. Short-chain inulin (degree of polymerization between 10 and 20) was chosen as it would have greater solubility than long-chain inulin (Tárrega, Torres, & Costell, 2011) and would be expected to have less effect on the viscosity of the product (Morris & Morris, 2012; Tárrega et al., 2011).

### 2.2. Labeling method

Inulin was covalently labeled with fluorescein isothiocyanate (FITC, Sigma–Aldrich Co. LLC., St Louis, MO) following the procedure described by Zimeri and Kokini (2003a) with modifications. Briefly, inulin (1 g) was dissolved in dimethyl sulfoxide (10 mL) containing two drops of pyridine. FITC (0.04 g) was added, followed by addition of the catalyst dibutylin dilaurate (20 mg). The mixture was heated for 3 h at 50 °C using a water bath. Several precipitations in ethanol were performed to remove the free dye. FITC-inulin was filtered using a filter paper No. 3 (Whatman, Wand R Balston Ltd, England), dried overnight in a vacuum oven at 85 °C, and stored in the dark under refrigeration to prevent loss of fluorescence. In order to prepare the porridges, an amount of FITC-labeled inulin (1% of the total inulin amounts described in Section 2.1) was added before the mixing with hot water and the sample preparation procedure outlined in Section 2.1 was followed.

### 2.3. Microscopy

The frozen samples were transferred to a cryostat, and 8 μm cryosections were obtained and placed in glass slides. Multiple staining was applied to cryosections, lugol's solution (0.05 g/L iodine) to detect starch and protein (Groves, 2006), 0.1 g/L Calcofluor White for β-glucan (Dornez et al., 2011), and 0.02 g/L Texas Red for protein (Johansson, Krona, & Stading, 2012). A Nikon Eclipse Ni–U research microscope coupled to a HGFI mercury lamp (Nikon, Tokyo, Japan) was used to visualize the microstructure of the porridges. Bright field and epifluorescence images were obtained using CFI Plan 4× objective (N.A. 0.20, W.D. 20 mm) and CFI Plan Fluor 10× (N.A. 0.30, W.D. 16 mm) and 20× (N.A. 0.75, W.D. 1 mm) objectives. Blue (Epi-FL Filterset DAPI, excitation wavelength 382–393 nm, emission 417–477), green (Epi-FL Filterset FITC, excitation wavelength 465–500 nm, emission 516–556 nm), and red (Epi-FL Filterset Texas Red, excitation 540–580 nm, emission 600–660 nm) light fluorescence filters were used to observe the fluorescence of Calcofluor, FITC-inulin, and Texas Red, respectively. Images were captured with a Nikon Digital Sight DS-Fi2-U3 digital camera.

### 2.4. Texture analysis

A RVA (Rapid Visco Analyzer, Newport Scientific Pvt. Ltd., Australia) with an impeller–cup combination was used to measure the viscosity of the porridges. Since rye porridge includes particles in the millimeter range it is impossible to use rheometry with gap distances which would give controlled shear rates and absolute measurements. For the RVA measurement the average temperature, as measured with a thermocouple connected to a digital readout during the preparation process described in Section 2.1, was used. The rate profile was set to simulate the stirring with an extra measurement period at the end of the run (Table 1). For each different formulation, approximately 35 g of the sample were introduced in a stainless steel cylinder and analyzed in the RVA in triplicate. The average viscosity during the last 15 s of each measurement period was used to derive a viscosity profile for each product. The first 15 s of the measurement periods were not included to avoid the initial instabilities.

### 2.5. Statistical analysis

Differences between viscosity profiles were evaluated using a mixed effect model suitable for repeated measurements with PROC mixed in SAS, version 9.4 (SAS Institute Inc, Cary, NC, USA). Time,

**Table 1**
Conditions for the RVA test on the porridge samples (total duration 330 s).

| Step | 1 | 2 | 3 | 4 | 5 |
|---|---|---|---|---|---|
| Temperature (°C) | 75 | 75 | 75 | 75 | 75 |
| Duration (s) | 30 | 120 | 30 | 120 | 30 |
| Agitation (rpm) | 30 | 0 | 30 | 0 | 30 |