Journal of Food Engineering 171 (2016) 102–110

Contents lists available at ScienceDirect

Journal of Food Engineering

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

Image analysis of dough development: Impact of mixing parameters and wheat cultivar on the gluten phase distribution

Adriaan Van der Mijnsbrugge [a, b], Frederic Auger [a, c], Sofie Frederix [a], Marie-Helene Morel [b, *]

[a] *Tereos Syral, Burchtstraat 10, 9300 Aalst, Belgium*
[b] *INRA, UMR 1208 Ingénierie des Agropolymères et des Technologies Emergentes (IATE), 2 place Pierre Viala, 34060 Montpellier cedex 1, France*
[c] *CSM Bakery Solutions, Mainzer Straße 152-160, 55411 Bingen, Germany*

ARTICLE INFO

Article history:
Received 22 May 2015
Received in revised form
14 August 2015
Accepted 1 October 2015
Available online 27 October 2015

Keywords:
Wheat flour
Dough mixing
Gluten agglomeration
Gluten-starch separation
Image analysis

ABSTRACT

Dough mixing is a key step in the wheat gluten-starch separation process promoting gluten agglomeration. The impact of the water/flour ratio (*W*/*F*), mixing speed (*N*), and wheat cultivar (Orvantis, Caphorn, Isengrain) on the protein phase distribution during mixing was studied by macroscopic image analysis. During dough mixing gluten agglomerates grew steadily and finally turned into a filamentous network at optimal dough development (t_{peak}). Prior to t_{peak}, neither *W*/*F* nor *N* impacted the average gluten lump diameter at a fixed stage of mixing. For Orvantis significantly larger gluten agglomerates (up to 272 μM) were observed as compared to Caphorn and Isengrain (up to 222 and 144 μM, respectively). Wheat flour cultivar was shown to have an important impact on gluten lump diameter, while mixing parameters (*N* and *W*/*F*) have no direct effect. Mixing parameters merely modulate the absolute gluten lump growth rate, just as they impact the optimal dough development time.

1. Introduction

Gluten-starch separation is a key step in the industrial processing of wheat flour for the co-production of vital wheat gluten and starch. Several types of fractionation processes exist, all containing a few common key steps. Flour is typically hydrated and processed into a dough mixer to allow gluten network development. Depending on the process used, the water/flour mixture is then either directly washed to separate starch from gluten (Martin process), or first diluted into a batter. Based on the typical gluten water insolubility, lump-forming capacity, and density, starch is then separated from the gluten phase by either centrifugation or sieving of the batter. A more detailed description of the different types of processes can be found in the following reviews entitled “Wet-milling of wheat flour: industrial processes and small-scale test methods” (Sayaslan, 2004), and “Fractionation of wheat and wheat flour into starch and gluten: overview of the main processes and the factors involved” (Van Der Borght et al., 2005).

Gluten development during mixing of water/flour blends is known to have an important impact on the efficiency of gluten-starch separation (Anderson et al., 1960; Dik et al., 2002; Frederix et al., 2004a; Johnston and Fellers, 1971; Kuktaite et al., 2007; Sayaslan et al., 2012). Mixing parameters such as mixing speed (*N*), water/flour ratio (*W*/*F*), temperature, and mixing time can significantly impact gluten formation. Auger et al. (2008) demonstrated that for highly hydrated doughs (*W*/*F* range 0.72–0.96) the instantaneous mixing power – which is impacted by *W*/*F* and *N* – is the mechanical parameter controlling the mixing time needed for optimal dough development for a fixed wheat cultivar dough. The impact of the *W*/*F* on gluten-starch separation has been incorporated in several studies (Anderson et al., 1960; Dik et al., 2002; Frederix et al., 2004a; Johnston and Fellers, 1971; Larsson and Eliasson, 1996a), however contrasting conclusions exist probably due to differences in mixing devices, mixing conditions, and separation methods used. Increased dough mixing speed at fixed mixing time was reported to enhance gluten extraction yields,

---

Abbreviations: *W*/*F*, water/flour ratio; *N*, mixing speed; t_{peak}, mixing time for optimal dough development; *AX*, arabinoxylans; *WEAX*, water-extractable arabinoxylan; *UPP*, SDS-unextractable polymeric protein; T_b, baseline torque; *RMS*-contrast, root mean square contrast; IU_{Abs}, intensity unit on absolute scale; $n_{detected}$, amount of detected gluten agglomerates per image; $D_{circ,\ avg}$, average circular diameter of gluten agglomerates; $D_{min,\ avg}$, minimal Feret diameter; $D_{max,\ avg}$, maximal Feret diameter.

[*] Corresponding author.
E-mail address: morel@supagro.inra.fr (M.-H. Morel).

http://dx.doi.org/10.1016/j.jfoodeng.2015.10.006
0260-8774/© 2015 Elsevier Ltd. All rights reserved.

explained by the higher energy input of the mixer (Frederix et al., 2004a). Another major factor impacting gluten-starch separation is the type of wheat cultivar which is processed (Dik et al., 2002; Larsson and Eliasson, 1996b; Roels et al., 1998). For the six different wheat varieties tested Roels et al. (1998) found very contrasting gluten protein recoveries within a range of 34.5–85.7 g extracted gluten protein/100 g flour protein.

The present study aims to further unravel the impact of the wheat cultivar and mixing parameters (W/F and N) on gluten development during dough mixing. Contrast between gluten protein and starch was enhanced by using Fast Green FCF – a specific protein stain. Protein phase distribution within dough was obtained from image analysis of standardized magnified pictures of dough samples obtained after variable mixing times. Image processing and analysis was used to allow a quantitative comparison of the results obtained at the different mixing conditions. The gluten phase distribution was studied on a macroscopic scale (2 × 1 cm images, 7 μm/pixel), rather than the microscopic scale more commonly used for bread dough studies (Bozkurt et al., 2014; Döring et al., 2015; Jekle and Becker, 2013; Létang et al., 1999; Peighambardoust et al., 2006). Indeed the high W/F ratio typically used in wheat starch processing allows larger gluten lumps to form, i.e. dilution increases the scale of spatial segregation. For such highly hydrated doughs macro-photography has proven to be more simple and more adapted than confocal microscopy to give an overview of the gluten phase distribution at a relevant scale (Auger et al., 2008). In addition, a macroscopic observation of the gluten lumps in dough would be more relevant for predicting the yield of sieving, one of the possible downstream fractionation steps in a wheat starch process. The three wheat bread-making cultivars used in the present study were selected based on their similar grain hardness (medium-hard) and protein content, while being contrasted in terms SDS-unextractable glutenin polymeric protein and arabinoxylan content.

2. Materials and methods

2.1. Raw materials and characteristics

French commercial wheat grains of certified cultivars (cvs. Orvantis, Caphorn, and Isengrain) were milled on a Bühler laboratory mill (MLU 202, Bühler, Switzerland) according to AACC (2000) method 26-31. Moisture and ash contents were determined according to AACC methods (44-15A and 08-01, respectively). The protein content ($N \times 5.7$) was determined using Kjeldahl method (AACC 46-10). Total arabinoxylan (AX) and water-extractable arabinoxylan (WEAX) contents were determined as described by Rouau and Surget (1994). The flour proteins were extracted and analyzed by SE-HPLC according to the method described by Morel et al. (2000), allowing to determine the fraction of SDS-unextractable polymeric protein (*UPP*). Table 1 displays an overview of the flour characteristics.

2.2. Dough formulation and mixing conditions

Flour and water were mixed together at 25 °C using a P600 planetary mixer (Brabender OHG, Germany), as described by Auger et al. (2008). Total dough mass was 550 g and all ingredients were equilibrated at 25 °C before mixing. Each mixing curve was characterized by its baseline torque (T_b) and t_{peak} (time needed to reach maximum torque). The different wheat variety flours were compared at the same mixing speed and consistency ($N = 90$ rpm and $T_b = 1.1$ N m), which allowed developing all flour doughs at the same instantaneous mixing power (Auger et al., 2008). Isengrain flour was used to test the impact of W/F ratio and mixing speed. The experimental setup is summarized in Table 2.

Table 1
Flour characteristics.

Wheat cultivar	Extraction rate (%)	Dry matter (%)	Ash (% db)	Protein (% db)	WEAX[a] (% db)	Total AX[a] (% db)	*UPP*[b] (%)
Orvantis	73.02	85.5	0.48	11.2	0.60	2.10	41
Caphorn	73.24	85.7	0.51	11.0	0.71	1.96	56
Isengrain	75.26	84.9	0.43	11.5	0.37	1.45	56

[a] WEAX = water-extractable arabinoxylan; Total AX = Total arabinoxylan content.
[b] *UPP* = SDS-unextractable polymeric protein.

2.3. In situ protein staining and dough observation

Evolution of the protein phase distribution was studied during mixing using Fast Green FCF as a protein specific stain. Fast Green FCF is a highly water soluble anionic dye which binds to positively charged proteins (*Mw*: 808.85 g/mol, water solubility: 150 g/l at 25 °C). For each mixing condition 0.896 mg of stain per gram of protein was added to dough water, according to Auger et al. (2008). Preliminary experiments were performed in order to verify that at the used dosage Fast Green FCF did not lead to any torque and development time change, while providing dough images with optimized contrast.

The optimal dough development times (t_{peak}) were determined for the seven different dough mixing conditions without stopping the mixer as means of a reference. The mixing experiments were repeated, with several stops (30 s) applied during mixing to sample the dough at fixed time over t_{peak} ratios (0.03–0.1–0.3–0.5–0.7–0.9–0.93–1.0–1.2–1.5 t_{peak}, see Fig. 1). Dough was sampled using a spoon while taking care not to disturb its surface. The dough sample was deposited on a rigid plastic sheet. Two spacers of 1.5 mm were placed on both sides of the dough layer and another rigid plastic sheet was tapped onto its surface with gentle pressure to smooth the surface. A Canon EOS 20D numeric camera (Canon, Japan) equipped with a 105 mm macro lens (Canon, Japan) was used to take pictures of dough samples. Natural lighting (5200 K) was provided by two fluorescent lamps (TC-L36W, Osram Dulux, Italy). The repeatability of the sampling was tested on the Isengrain dough at 0.745 W/F 90 rpm, for which three successive doughs were mixed and the different dough samples were obtained.

Table 2
Overview of dough mixing conditions and mixing curve characteristics.

Wheat cultivar	W/F^a (g/g_{wb})	N^a (rpm)	$t_{peak}{}^a$ (min)	$T_b{}^a$ (N m)
Isengrain	0.725	60	129	1.1
Isengrain	0.725	90	39	1.5
Isengrain	0.745	90	50	1.4
Isengrain	0.765	90	78	1.1
Isengrain	0.795	90	123	0.9
Caphorn	0.840	90	100	1.1
Orvantis	0.920	90	15	1.1

[a] W/F = water/flour ratio; N = mixing speed; t_{peak} = optimal dough development time; T_b = baseline torque.