Food Research International 42 (2009) 963–975

Contents lists available at ScienceDirect

Food Research International

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

The role of corn starch, amaranth flour, pea isolate, and *Psyllium* flour on the rheological properties and the ultrastructure of gluten-free doughs

Manuela Mariotti [a,*], Mara Lucisano [a], M. Ambrogina Pagani [a], Perry K.W. Ng [b]

[a] *Department of Food Science and Microbiology (DiSTAM) – University of Milan, Via G. Celoria 2, 20133 Milan, Italy*
[b] *Department of Food Science and Human Nutrition – Michigan State University, 135 GM Trout FSHN Building, East Lansing, MI 48824-1224, USA*

ARTICLE INFO

Article history:
Received 9 January 2009
Accepted 22 April 2009

Keywords:
Gluten-free
Dough
Amaranth
Pea isolate
Psyllium
Rheology
CLSM
SEM

ABSTRACT

The removal of gluten from bakery products, in order to produce foods (mainly based on gluten-free cereal flours and starch) for people with celiac disease, impairs dough's capacity to properly develop during leavening and baking. The main aim of this research was to produce and evaluate some experimental gluten-free (GF) doughs containing different levels of corn starch, amaranth flour (to enhance the nutritional benefits), pea isolate (to increase the protein content) and *Psyllium* fiber (as thickening agent and fiber source) in order to study the influence of the different ingredients on the rheological properties and on the ultrastructure of the doughs. *Psyllium* fiber generally enhanced the physical properties of the doughs, due to the film-like structure that it was able to form, and the most complex among the experimental formulations looked promising in terms of final bread technological and nutritional quality even when compared to two different commercial GF mixtures.

1. Introduction

Gluten is the structure-forming complex in wheat, responsible for the viscoelastic properties needed to produce good quality baked products. Interactions of gliadins and glutenins through covalent and non-covalent bonds to form gluten complexes result in viscoelastic dough that has the ability to withstand stresses applied during mixing and to retain gas during fermentation and baking, producing a light baked product (Lindsay & Skerritt, 1999). Unfortunately, this complex can be harmful for people suffering from celiac disease (CD) or from other allergic reactions or intolerances to gluten consumption. CD, in particular, is a chronic malabsorption disorder of the small intestine caused by exposure to gluten in the genetically predisposed individual (Laurin, Wolving, & Falth-Magnusson, 2002): it is characterized by a strong immune response to certain amino acid sequences found in the protein fractions of wheat, barley, and rye (Fasano & Catassi, 2001; Thompson, 2001). The only effective treatment for celiac people is a strict adherence to a gluten-free diet throughout their lifetime.

The replacement of gluten in bakery products is a major technological challenge, as it is the essential structure-building protein. Its removal impairs dough's capacity to properly develop during kneading, leavening and baking. The absence of gluten often results in a liquid batter rather than a dough and can result in breads with a crumbling texture, poor color and other post-baking quality defects (Gallagher, Gormley, & Arendt, 2004). Thus, substances that imitate the viscoelastic properties of gluten, in order to provide structure and retain gas, are always required. Recently, there has been an increasing interest in gluten-free (GF) breads, whose formulations mainly involve the incorporation of starches of different origin, dairy proteins, other non-gluten proteins, gums, hydrocolloids, and their combinations, into a GF flour base (mostly rice and corn flour) that could simulate the viscoelastic properties of gluten and could result in maintaining structure, mouthfeel, acceptability and shelf-life of the finished products. However, currently, many GF breads available on the market are of low technological and nutritional quality, particularly when compared to their wheat counterparts, exhibit a dry crumb and have poor mouthfeel and flavor.

GF products are frequently produced with the addition of various proteins to a starchy base, to increase their nutritional value. The incorporation of dairy proteins has long been established in the baking industry, but legumes can also be a good supplement for cereal-based foods since they increase the protein content

Abbreviations: ANOVA, analysis of variance; BU, brabender units; CD, celiac disease; CLSM, confocal laser scanning microscopy; D, dough; FITC, fluorescein isothiocyanate; FWA, farinographic water absorption; G', storage modulus or elastic modulus; G'', loss modulus or viscous modulus; $|G^*|$, complex shear modulus; GF, gluten-free; HPMC, hydroxy-propyl-methyl-cellulose; LSD, least significant difference; LVE, region of linear viscoelasticity; SEM, scanning electron microscopy; tan δ, loss tangent or damping factor (DF).

* Corresponding author. Tel.: +39 02 50319186; fax: +39 02 50319190.
E-mail address: manuela.mariotti@unimi.it (M. Mariotti).

0963-9969/$ - see front matter © 2009 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodres.2009.04.017

and complement the nutritional value of cereal proteins. Cereals are deficient in lysine, one of the essential amino acids for the human diet, while legumes have a high level of this amino acid; simultaneously, cereal proteins are able to complement legume proteins in the essential amino acid methionine (Iqbal, Khalil, Ateeq, & Khan, 2006). Although the most-used legume protein is the soybean protein, due to its valuable functional properties (Ranhorta, Loewe, & Puyat, 1975; Ribottan et al., 2004), pea proteins can also be successfully used in bakery products, obtaining an enrichment in proteins and improving biological value of these products (Tömsközi, Lásztity, Haraszi, & Baticz, 2001).

Pseudocereals such as buckwheat and amaranth can also be useful for the above purpose. The protein content of amaranth (*Amaranthus* spp.) (11.7–18.4%) is generally higher than that of wheat (Berghofer & Schoenlechner, 2002, chap. 7) and contains acceptable levels of essential aminoacids (particularly lysine, tryptophan, and methionine), which are found in low concentrations in cereals and leguminous grains of common usage; structural characteristics of these proteins influence their functional properties (Avanza, Puppo, & Añón, 2005). The fat content of amaranth is also interesting and it is characterized by a high amount of unsaturated fatty acids, with a very high level of linoleic acid (Berghofer & Schoenlechner, 2002, chap. 7; Lucisano, Mariotti, Pagani, & Caramanico, 2006). Starch comprises the main component of the carbohydrates and significant amounts of calcium, iron, potassium, phosphorous, vitamins, and dietary fiber are present.

Gums and hydrocolloids are essential ingredients in GF bread formulations for improving the texture and the appearance of the final products. Due to their structure forming properties, their addition assures higher dough consistency, improved gas retaining capacity and longer shelf-life. In this respect, it has been proved that the most effective gum and hydrocolloid compounds are hydroxypropyl-methyl cellulose (HPMC), locust bean gum, guar gum, carrageenan, and xanthan gum (Christianson, Gardner, Warner, Boundy, & Inglett, 1974; Gallagher et al., 2004; Lazaridou, Duta, Papageorgiou, Belc, & Biliaderis, 2007). The association of HPMC (2 g/100 g rice flour) with *Psyllium* fiber (1 g/100 g rice flour) in the formulation of GF bread gave good results in terms of loaf volume due to the formation of a weak gel network, capable of trapping CO_2 by virtue of the gelling and water-absorbing abilities of *Psyllium* fiber, and of the heat-induced gelation of HPMC (Haque & Morris, 1994). Higher additions (5%, 7.5%, 10%) of *Psyllium* to a GF formulation determined an increased fiber content of the bread (190–450% higher than that of the control bread) and a softer crumb during a 4-day storage period. *Psyllium*, besides being an excellent source of natural soluble fiber, has been widely recognized for its cholesterol-lowering effect and insulin sensitivity improvement capacity (You, Perret, Parker, & Allen, 2003). The enrichment of GF baked products with dietary fiber has proved to be necessary, since celiac patients generally have a low intake of fiber attributed to their GF diet (Thompson, 2000).

The main aim of this research was to produce and evaluate some experimental GF doughs containing different levels of corn starch, amaranth flour (to enhance the nutritional benefits), pea isolate (to increase the protein content) and *Psyllium* flour (as thickening agent and fiber source), in order to: (1) study the influence of the different ingredients on the rheological properties and on the ultrastructure of the dough; and (2) develop healthier breads that contain higher protein and dietary fiber levels. For these purposes, the experimental GF doughs were compared to those obtained from two commercial GF bread mixes. Moreover, rheometry and microscopy approaches (limited up till now in studies of GF dough properties), were both used, with the microscopic images generated by scanning electron microscopy and confocal laser scanning microscopy, to develop a fundamental understanding of the ultrastructure of GF doughs and constituents as a good basis for further improvement.

2. Materials and methods

2.1. Raw materials

2.1.1. Origin

The raw materials used to produce the GF formulations were of commercial origin: the corn starch was supplied by Molino Quaglia SpA (Vighizzolo D'Este, PD, Italy), the *Psyllium* fiber by Giulio Gross Srl (Trezzano sul Naviglio, MI, Italy) and the pea isolate (Pisane F9) was provided by Prodotti Gianni SpA (Milan, Italy). Amaranth (*Amaranthus hypochondriacus*) was furnished as seeds by *Cooperativa Quali* (Tehuacàn, Mexico). Amaranth seeds were ground to flour at DiSTAM, just before use, by a laboratory mill (Labormill 4RB; BONA srl, Monza, MI, Italy), and three milling fractions (coarse, medium, fine) were obtained. On the basis of previous studies, a mixture of 30% fine fraction and 70% medium fraction was used for this research. This blend was characterized by the following particle size distribution: 48% ⩾ 200 μm, 125 μm ⩽ 36% < 200 μm, 16% < 125 μm.

2.1.2. Chemical characterization

The raw materials were characterized for their moisture (AACCN°44–15A; AACC, 1983), protein (AACCN°46–11; AACC, 1983), lipid (ICCN°136; ICC, 1999), fiber (Prosky, Asp, Schweizer, DeVries, & Furda, 1988), total starch and damaged starch ("Total Starch Assay Kit", "Starch Damage Assay Kit"; Megazyme International Ireland Ltd., Bray Business Park, Bray, Co., Wicklow, Ireland) contents. Data obtained from these characterizations are listed in Table 1 and are the average of at least three determinations.

2.1.3. Scanning electron microscopy

The ultrastructure of corn starch, amaranth flour, pea isolate, and *Psyllium* fiber was observed by means of scanning electron microscopy (SEM). Powders were mounted on aluminum stubs, sputter-coated with gold, and their ultrastructures were imaged in a LEO438 VP SEM (LEO Electron Microscopy Ltd., Cambridge, UK) under high vacuum conditions (10^{-4} Pa) at an accelerating voltage of 20 kV.

2.2. Gluten-free formulations

2.2.1. Experimental GF bread formulations

Corn starch, amaranth flour, pea isolate, and *Psyllium* fiber were used to produce six experimental GF formulations (Table 2). Two

Table 1
Composition of the raw materials involved in the experimental GF formulations.

	Moisture (g/100 g)	Protein (g/100 g db)	Lipids (g/100 g db)	Total starch (g/100 g db)	Damaged starch (g/100 g db)	Fiber (g/100 g db)
Corn starch	13.70 ± 0.01	–	0.69 ± 0.02	>95[a]	1.91 ± 0.06	–
Amaranth flour	12.01 ± 0.06	15.78 ± 0.05	8.16 ± 0.06	55.04 ± 0.70	9.31 ± 0.11	8
Pea isolate	5.93 ± 0.04	86.99 ± 0.01	8.55 ± 0.03	0.38 ± 0.01	–	–
Psyllium fiber	9.66 ± 0.01	2.14 ± 0.23	0.70 ± 0.05	–	–	>95[a]

[a] As reported on the product label.