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Effect of Sourdough on Phytic Acid Content and Quality of Iranian Sangak Bread

Didar Z*

Assistant Professor of College of Food Science and Technology, Islamic Azad University, Neyshabur Branch,Iran

Abstract

Phytic acid is the principal storage form of phosphorus in many plant tissues and is able to bond with cations that cause mineral deficiency. Application of lactic acid bacteria sourdough for phytic acid reduction in loaf breads is proposed by some researchers, but it seems that there is not much published work on the application of sourdough lactic acid bacteria in flat breads. In this study, effect of several lactic acid bacteria sourdough on flat bread (Sangak) is investigated. Results showed that dough yield (DY), strain type and the percentage of sourdough addition affect pH, phytic acid content and quality of bread. Higher dough yield and higher sourdough addition decreased phytic acid content. Application of Lb. plantarum sourdough with dough yield=300 and 30% addition resulted in 45% decrease of phytic acid content (from 894.66 mg/100g in flour to 507.3 mg/100g in bread). As indicated by panelists, Lb. plantarum sourdough with dough yield=250 and 30% addition can present the greatest effect on overall quality score of the breads.

Keywords: Flat bread; Phytic acid; Sangak bread; Sourdough

Introduction

Bread is one of the most popular foods all over the world that provide a high quantity of daily energy and proteins. Nowadays, consumption of whole flours and flours with high extraction rate is recommended, because of their high amount of fibers, vitamins and minerals. Despite nutritional benefits of whole flour, concentration of some undesirable components such as phytic acid is higher than white flour [1]. Phytic acid is the principal storage form of phosphorus in many plant tissues that contains 50% to 80% of the total phosphorus in seeds. Phytate works in a broad pH-region as a highly negatively charged ion and therefore its presence in the diet has a negative impact on the bioavailability of divalent and trivalent mineral ions such as Zn^{2+}, $Fe^{2+/3+}$, Ca^{2+}, Mg^{2+}, Mn^{2+} and Cu^{2+} [2] Phytic acid content in flour increases with increasing the extraction rates [3]. It has been reported that 30% and 31% of Iranians are suffering from iron and zinc deficiency, respectively [4]. Sangak, as one of the most common Iranian flat breads, is made from high extraction rate flour; thus, obtaining Sangak with low phytate content has created the need for improving techniques.

There are several methods for phytic acid reduction in bread that one of these methods is use of sourdough. Lopez et al. [5] reported the decrease of phytate content in bread made with yeast is 41% and in bread made with sourdough is 71%. Screeramulu screened nineteen strains of lactic acid producing bacteria of the genera Lactobacillus and Streptococcus collected from different culture collections for the production of extracellular phytase. A number of them exhibited the enzyme activity in the fermentation medium but *Lactobacillus amylovorus* B4552 produced the maximum amounts of phytase. Lopez et al. [6] detected high phytase activity from *Lb. plantarum*, *Lb. amylovorus* and *Leu. mesenteriodes* in whole flour medium.

It seems that there is not much published work on the application of sourdough lactic acid bacteria in flat breads. Therefore, the objective of this study was to examine the effect of several sourdough lactic acid bacteria on the quality of Iranian Sangak bread, and their potential use in reducing the phytic acid content.

Materials and Methods

Materials

Alvand wheat was purchased from the Agricultural Research Center of Neyshabour and it was milled on the laboratory mill AQC 109 after being cleaned and conditioned to extraction rate of 98%. The strains used throughout this study were *Lactobacillus plantarum* (PTCC 1058) and *Lactobacillus reuteri* (PTCC 1655) that purchased from Iranian Research Organization for Science and Technology in a lyophilized form.

Methods

Moisture, ash, wet gluten and gluten index were determined according to the Standard Procedures 46–16A, 08–01 and 38–12 of AACC, respectively [7-9]. Protein content and phytic acid were determined by ISIRI 2863 and Garcia-Estepa methods, respectively [10,11]. PH of the samples was measured immediately after removal from the production by diluting 5 g samples with 30 ml water according to standard method [11].

Preferment preparation

Both lactic acid bacteria strains transferred to MRS broth medium in sterile condition and incubated at 37˚C for 18 hour, and then centrifuged (4000 rpm for 10 min) and microbial cells harvested. Different dilution (10^{-1}-10^{-7}) of mother culture prepared and transferred to MRS agar and cultured by pour plate method. The number of each bacterial strain was nearly 10^{7}cfu/g. Sour dough was prepared with

***Corresponding author:** Zohreh Didar, Assistant Professor of College of Food Science and Technology, Islamic Azad University, Neyshabur Branch, Iran, E-mail: z_didar57@yahoo.com

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dough yield (DY) 250 and 300. From each bacterial strain, 10 ml of mother culture was centrifuged and mixed for 1 min, transferred to a large beaker and covered with Aluminum foil, and then incubated at 37˚C for 20 h. Biomass was mixed with wheat flour until dough formation.

Bread production

The bread formula used for this kind of bread consisted of flour (80 kg); wet baking yeast (400g); dry baking yeast (100 g); salt (300g); water (about 60 liter based on water absorption). Sourdough was replaced in the ratio of 10, 20 and 30% instead of flour in dough formulation. A baking technique, similar in principle to that of commercial procedure, was used for baking experimental loaves having almost equal volumes. In this procedure, the ingredients were mixed to optimum dough development. The dough samples were fermented in sealed containers at 30˚C and 75–85% R.H. for 90 min, and then divided into 200 g pieces and rounded by hand. The dough pieces were rolled. The rectangular-shaped dough pieces were then punched. The dough pieces were then baked on a bed of hot tiny river stones for 15 min at 260˚C to obtain the proper thickness and acceptable color and texture.

Sensory evaluation

Sensory analysis was carried out using a 5-point hedonic scale, scoring 1 (lowest) to 5 (highest). Sensory evaluation was performed by 10 trained panelists. Six attributes of bread, i.e., bread form and shape, upper surface property, bottom surface property, chewing ability, odor, flavor and taste, and overall quality score were selected according to the Iranian traditional bread evaluation method described by Cereal Research Center of Iran. For each of the attributes, the average of the panelist scores was calculated [8].

Statistical analysis

In order to assess significant differences among samples, a completely randomized design was performed using the MSTATC program (version 1.41). Duncan's new multiple range test was used to describe means with 99% confidence.

Results and Discussion

Chemical characteristics of wheat flour

The chemical compositions of wheat flours are presented in Table 1. The characteristics of the wheat flour are in the range of typical values of medium strong flour, suitable for Iranian flat bread.

Phytic acid content of flours depends on several factors such as wheat cultivar, weather condition and milling parameters such as bran content and extraction rate [13]. The flour used in this study had high phytic acid amount, and this is only because of high extraction rate of flour (98%). Payan showed that most wheat varieties have 1000

Attribute	Value
Protein (g/100 g, d.b.)	11.54
Moisture (g/100g)	7.52
Ash (g/100 g, d.b.)	1.74
Phytic acid (mg/100g)	894.66
Wet gluten (g/100g)	30.1
Gluten index	73.42

Table 1: Quality characteristics of sangak flour.

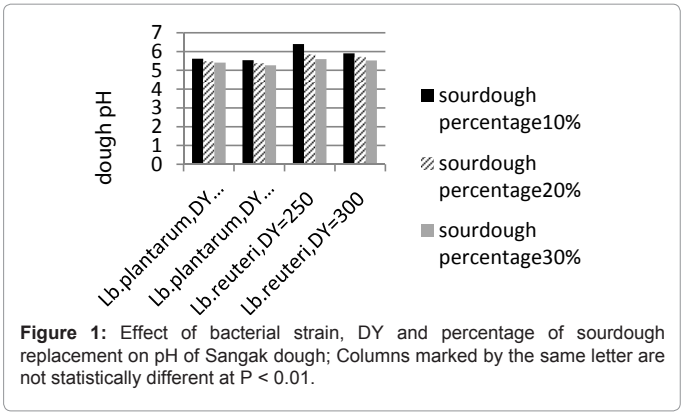

Figure 1: Effect of bacterial strain, DY and percentage of sourdough replacement on pH of Sangak dough; Columns marked by the same letter are not statistically different at P < 0.01.

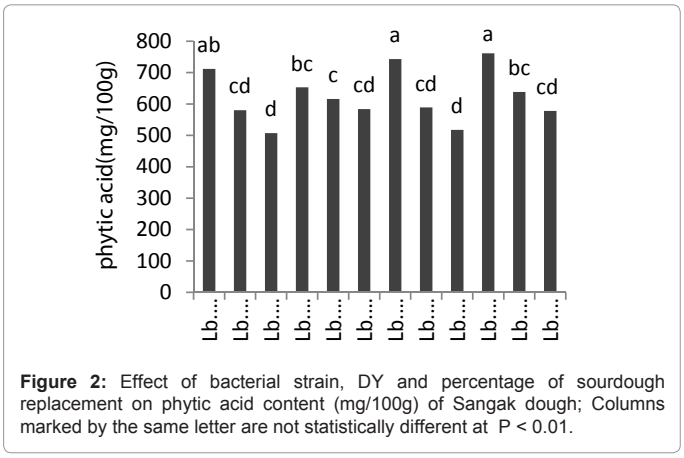

Figure 2: Effect of bacterial strain, DY and percentage of sourdough replacement on phytic acid content (mg/100g) of Sangak dough; Columns marked by the same letter are not statistically different at P < 0.01.

mg/100g (1%) phytic acid in Iran and the remained phytic acid in processed wheat vary according to extraction rate. The remained phytic acid in flours with high extraction rate (higher than 80%) is 600-700 mg/100g, while it decreases to 30% in flours with lower extraction rate (lower than 60%). According to Gargari et al. [14] the mean of phytic acid in flour and various breads is 262.75 mg/100g and 108.53 with AOAC method, respectively.

Measurement of dough pH

The pH of dough and bread is an important factor in degradation of phytic acid. Solubility of phytate that chaleted with cations depended on pH, type and extent of cations [5]. The pH of dough samples supplemented with sourdough is described in Figure 1.

According to Figure 1, sourdough from *Lb. plantarum* with DY=300 and 30% replacement in dough formulation showed the most marked effect on reducing pH of dough. Higher DY of sourdough resulted in higher decrease in pH of dough.

Phytic acid measurement

The phytic acid content in the samples from the three different bread preparations (Figure 2) followed nearly the same pattern as pH.

The phytic acid in bread samples supplemented with sourdough from *Lb. plantarum* was lower than samples treated with sourdough from *Lb. reuteri*. In addition, higher DY of sourdough resulted in higher decrease in phytic acid content of bread samples. Sangak bread made with sourdough from *Lb. plantarum* with DY=300 and 30% replacement in dough formulation has 507.3 mg/100g phytic acid. This is

Treatment	Characteristic					
	Odor, flavor and taste	Chewing ability	Form and shape	Bottom Surface properties	Upper surface properties	Overall Quality score
Lb. plantarum, DY=250, 10%	^{cd} 4.07	^{d} 4.09	^{ab} 3.50	^{ab} 4.31	^{abc} 4	3.54^{c}
Lb. plantarum, DY=250, 20%	^{bc} 4.18	^{b} 4.86	^{bc} 3.25	^{a} 4.57	^{ab} 4.15	4.31^{ab}
Lb. plantarum, DY=250, 30%	^{a} 4.51	^{bc} 4.80	^{cde} 2.86	^{bc} 3.86	^{cd} 3.64	4.17^{bc}
Lb. plantarum, DY=300, 10%	^{cde} 4.01	^{b} 4.86	^{bc} 3.29	^{abc} 4.14	^{abc} 3.93	4.15^{bc}
Lb. plantarum, DY=300, 20%	^{abc} 4.24	^{ab} 4.90	^{cd} 2.96	^{c} 3.79	^{bc} 3.82	4.18^{bc}
Lb. plantarum, DY=300, 30%	3.79^{de}	^{a} 4.97	^{g} 2.11	^{d} 2.88	^{e} 3.16	3.75^{d}
Lb. reuteri, DY=250, 10%	^{abc} 4.33	^{bc} 4.79	3.61^{ab}	^{a} 4.45	^{a} 4.32	4.39^{a}
Lb. reuteri, DY=250, 20%	^{ab} 4.51	^{b} 4.82	^{ab} 3.75	^{ab} 4.29	^{ab} 4.14	4.44 ^{a}
Lb. reuteri, DY=250, 30%	^{ab} 4.55	^{ab} 4.91	^{def} 2.79	^{bc} 3.98	^{bc} 3.86	4.29^{ab}
Lb. reuteri, DY=300, 10%	^{cd} 4.08	^{c} 4.49	^{ab} 3.75	3.95 ^{bc}	^{ab} 4.09	4.13 ^{bc}
Lb. reuteri, DY=300, 20%	^{abc} 4.33	^{ab} 4.91	^{fg} 2.36	^{d} 3	^{abc} 4.04	4.07^{c}
Lb. reuteri, DY=300, 30%	^{e} 3.67	^{ab} 4.87	^{efg} 2.43	^{d} 2.83	^{de} 3.36	3.71^{d}

Table 2: Sensory characteristics of fresh sangak bread containing selected sourdoughs.

probably because of microbial phytase enzyme and dough acidification that provided suitable condition for endogenic and microbial phytase activity and solubility increase of phytate complexes. Chaoui et al. [15] showed that bread making with sourdough from *Lb. plantarum* and *Leu. mesenteriodes* resulted in 76.5% and 67% decrease in phytic acid content, respectively. Lopez et al [6] reported high phytase activity of *Lb. plantarum*, *Lb. acidophilus* and *Leu. mesenterisoes* in whole flour medium. Palacios et al. [16] detected high phytase activity by *Lb. reuteri (LM-15)*. Bread from 24h-old sourdough of this strain has lower phytic acid than breads from other strains. According to this study, this bacterial strain is able to complete phytic acid degradation in bread. Angelis et al. [17] reported that 8 hour incubation of *Lb. sanfranciscensis* CB_1 cause 64-74% decrease in sodium phytate concentration.

Bread quality

Table 2 shows that all sensory attributes of Sangak bread are influenced by sourdough addition.

Results showed that form and shape, bottom and upper surface properties of bread samples decreased with addition of sourdough, especially in 30% replacement. These effects are because of acidification and proteolysis that occurred in dough with sourdough addition. Gluten proteins have an important role in rheological properties of wheat dough and bread texture. Proteolysis of gluten proteins influences gluten network forming and causes weak and sticky dough [18]. Cereal proteinases have optimum pH between 4 and 5 [19]. Fermentation with sourdough resulted in solubilization and depolymerization of macropolymers of gluten. Proteolysis during fermentation is dependent on the acid production. Lactic acid bacteria increase proteolytic activity by inducing optimum pH for cereal proteinase that catalyzes hydrolysis of wheat proteins dependent on type of strain [20]. Maher et al. [21] proposed that solubility of proteins increases in acidic conditions and repulsion increase of electrostatic forces leads to unfolding of gluten proteins. The exposure of hydrophobic groups increases the presence of strong intermolecular repulsion. Accordingly, electrostatic forces prevent the formation of new bonds that causes network weakening. In the most treatments, sourdough addition caused to increase in score of bread in the characteristic of odor, flavor and taste. Higher DY and 30% replacement caused negative effect on odor, flavor and taste of breads. This effect is probably because of acidification and enzyme activity and because of the various endogenous microbial and wheat flour proteolytic enzymes which could be active in the dough during acidification. Proteolytic enzymes in sourdough system cause the production of free amino acids that acts as flavor precursors [22,23]. Hansen and Hansen [24] showed that bread from sourdough has the highest aroma and taste components and the highest score in sensory evaluation in comparison with breads that are acidified with lactic or acetic acid. According to Carnevali et al. [25] liquid sourdough treatment that includes lactic acid bacteria and yeasts cause to increase the aroma and taste and acidity more than breads from baking yeast. According to this study, the micro-biota of sourdough markedly influences flavour and texture of bakery products.

Conclusion

In this study, significant effect of sourdough on phytic acid content and quality of Iranian Sangak bread was clarified. Dough yield (DY), strain type and the percentage of sourdough addition affected pH, phytic acid content and quality of bread. Based on these results, higher dough yield and higher sourdough addition decreased phytic acid content. Organoleptic analysis showed that *Lb. plantarum* sourdough with dough yield=250 and 30% addition can present the greatest effect on overall quality score of the breads.

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