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SOURDOUGH BREAD: PROCESSING, FLAVOR AND HEALTH BENEFITS

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ABSTRACT

Sourdough is an ingredient containing cereal components, liquids and active microorganisms which are lactic acid bacteria (LAB) and yeast. Traditional sour dough bread technology is based on a spontaneous fermentation process from LAB and yeast that occurring naturally in flour. Classic sourdough preparation is a multiple stage process that starts with a mixture of flour and water left for a specific period of time. Temperature, dough consistency, and dough resting time at each stage determine the development of active microflora. The modern biotechnology of baked goods largely uses sourdough as a natural leavening agent because of the many advantages it offers over baker's yeast especially in the development of the characteristic flavour of bread that resulting in a final product with high sensory quality. Organic acids together with alcohols, esters, carbonyl compounds and others, strongly affect the flavor of sourdough bread. Furthermore, sourdough fermentation has several health benefits which are reducing antinutritional compounds, enhancing nutritional values, converting toxic compounds and producing antimicrobial substances.

Keywords: *sourdough, bread, processing, flavor, health benefits*

INTRODUCTION

The modern biotechnology of baked goods largely uses sourdough as a natural leavening agent because of the many advantages it offers over baker's yeast especially in the development of the characteristic flavour of bread that resulting in a final product with high sensory quality.

Sourdough is an ingredient containing cereal components, liquids and active microorganisms which are lactic acid bacteria (LAB) and yeast. A general procedure of the entire manufacturing process used in sourdough bread production is shown in Figure 1 (Hansen & Schieberle, 2005). The generation of sufficient amounts of volatile

compounds during sourdough fermentation needs a multiple step process of about 12–24 h, while fermentation by bakers yeast alone is finished within a few hours. Sourdough bread has a higher content of volatiles and, also, achieves higher scores in sensory tests compared to bread chemically acidified with lactic and acetic acid (Hansen & Hansen, 1996). In recent years there has been a growing interest in sourdough bread production because of its health benefits (Diowksz & Ambroziak, 2006).

MICROORGANISMS IN SOURDOUGH

The traditional production of sourdough is based on spontaneous fermentation due to the development of microflora naturally present in

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the raw material. At the very beginning of the fermentation process, bacteria from the family Enterobacteriaceae are dominant. They produce mainly hydrogen (H₂) and carbon dioxide (CO₂) and small amounts of organic acids: lactic, acetic, formic, and succinic acids. After this stage, in which the preliminary acidification takes place, the number of gram-negative bacteria decreases, and gram positive bacteria and yeast start to dominate (Gobbetti, 1998).

Lactic acid bacteria

LAB isolated from traditional sourdough belong mainly to four genera: *Lactobacillus*, *Pediococcus*, *Leuconostoc*, and *Weissella*. In most cases lactobacilli are the dominating bacteria. They are gram positive, nonmotile rods, anaerobes and acid tolerant. These bacteria have complex nutritional requirements for amino acids, peptides, vitamins, minerals, fatty acids, and carbohydrates. Their proteolytic activity and the metabolic profile depend on growth temperature. The optimal temperature for growth is 30 -35 °C (Meuser et al, 1990).

On the basis of dominant carbohydrate metabolic pathway, lactic acid bacteria can be divided into two groups (Martinez-Anaya et al, 1994): (1) homofermentative, which produce mainly lactic acid; and (2) heterofermentative, which produce, apart from lactic acid, considerable amounts of ethanol, acetic acid, and carbon dioxide (Figure 2). Homofermentative lactic acid bacteria dominate spontaneously fermenting

sourdough. Homofermentative LAB strongly influence crumb porosity and elasticity but have limited influence on the sensory qualities of sourdough bread (Sugihara, 1985). Heterofermentative LAB plays the main role in creating bread flavor due to characteristic of their metabolites (Damiani et al, 1996). Sourdough lactobacilli, consisting of obligately and facultative heterofermentative, and obligately homofermentative can be seen in Table 1.

Yeast

In the course of fermentation, yeast plays several important roles. First, it produces carbon dioxide, which expands the dough, resulting in the proper porosity of the crumb and the proper volume of the bread. Besides, yeast produces a lot of side products such as aldehydes, aliphatic and isoaliphatic alcohols, acids, keto acids, and esters, which alone or in combination with other compounds can create specific and unique flavors of bread (Hansen & Hansen, 1996).

Several yeasts are found in sourdoughs but *Saccharomyces cerevisiae* is considered the dominant organism for leavening of bread (Corsetti et al., 2001). *Saccharomyces cerevisiae*, characteristic mainly for sourdough, is a top fermenting yeast with an optimum growth temperature of 26-32 °C. It is an aerobe or facultative aerobe. It utilizes glucose, maltose,

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galactose, sucrose, and partly raffinose (Gobbetti, 1998). The frequently dominant *S. cerevisiae* is often introduced through the addition of baker's yeast. Important yeasts in sourdough starters include *Saccharomyces exiguous* (physiologically similar to *Candida milleri*), *Candida krusei*, *Pichia norvegensis* and *Hansenula anomala*. The extensive variability in the number and type of species found depends on several factors, including degree of dough hydration [dough yield (DY) = weight of dough/weight of flour ·100], type of cereal used, leavening temperature, and sourdough maintenance temperature (Gobbetti et al., 1995).

Mixed cultures of LAB and yeasts

Mixed cultures of LAB and yeasts vary in composition in sourdough sponges. The use of mixed cultures has a number of important advantages, such as improved flavour and texture and retained freshness for longer compared to baker's yeast bread (Meignen et al., 2001). In such mixed cultures, yeasts act mainly as leavening agents, while LAB contribute mainly to the flavouring compounds of bread.

FLAVOR

Sourdough fermentation is essential to achieve an acceptable flavour, since a comparison between chemically acidified bread and sourdough bread showed that the latter possessed a superior sensory quality (Kirchhoff & Schieberle, 2002).

Types of flavouring compounds in sourdough breads

There are two categories of flavour compounds, produced during sourdough fermentation. Non-volatile compounds including organic acids produced by homo- and heterofermentative bacteria which acidify, decrease pH and contribute aroma to the bread dough. The second category is volatile compounds of sourdough bread - includes alcohols, aldehydes, ketones, esters and sulphur. All these compounds are produced by biological and biochemical actions during fermentation and contribute to flavour (Spicher, 1983).

Microbial metabolisms verify production of different volatile compounds for hetero- and homo-lactic LAB fermentations. Abundant products of yeast fermentation are 2-methyl-1-propanol, 2,3 methyl-1-butanol and other iso-alcohols. Heterofermentative LAB products are dominated by ethyl acetate and certain alcohols and aldehydes whereas major homofermentative LAB products are diacetyls and carbonyls.

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Effect of ingredients and processing on the sourdough flavor

The LAB strains of sourdough vary in metabolism and aroma compounds. Monoculture fermentation of dough for 15 h at 30 °C, followed by mixing and a further 10 h fermentation has shown increase in the production of sourdough volatile compounds (Merotha et al., 2004). Using only yeasts in wheat bread, seven volatiles were found abundant: acetaldehyde, acetone, ethyl acetate, ethanol, hexanal, isobutyl alcohol, and propanol. The quantity of volatile flavour compounds can be improved by the addition of glucose and sucrose less by maltose. Addition of enzymes to sourdough sponges can also enhance bread volatile compounds (Martinez-Anaya, 1996).

Low temperature (25°C) and sour dough firmness are appropriate for LAB souring activities but limited yeast metabolism. Raising the temperature to 30 °C and semi-fluid sour doughs gave more complete volatile profiles. At 3 h leavening time, the sour dough is mainly characterized by iso-alcohols. An increase of leavening time up to 9 h gave a total amount of volatiles about three times higher than that at 5 h and strengthened the LAB contribution. The additions of fructose and citrate to the dough enhanced the acetic acid and volatile synthesis by LAB, respectively. After baking, the ethanol disappeared, 2-methyl-1propanal is synthesized, lactic and acetic acids remained constant, the total amount of volatiles is

reduced to a level <12.5% of the initial and an increase in the relative percentage of iso-alcohols and aldehydes are detected (Gobbetti et al, 1995).

Contribution of lactic acid fermentation to bread flavor.

The ratio between lactic and acetic acid, defined as the fermentation quotient (FQ), is an important factor that might affect the aroma profile, although it is also relevant for the structure of final products. Acetic acid, produced by heterofermentative LAB, is responsible for a shorter and harder gluten while lactic acid can gradually account for a more elastic gluten structure (Lorenz, 1983). Compounds strongly affecting bread flavour are mainly organic acids, alcohols, esters and carbonyls (Kirchhoff & Schieberle, 2002). In order to generate sufficient amounts of volatile compounds, the generation process needs multiple steps of about 12–24 h; when baker's yeast is used the fermentation is completed within a few hours (Hansen & Schieberle, 2005).

Sourdoughs made with both LAB and yeasts resulted in more aroma compounds as compared to sourdoughs made from single starter based either on LAB (e.g. *Lb. brevis*) or yeast (e.g. *S. cerevisiae*). Sourdoughs with a higher relative percentage of yeast-derived fermentation products are produced if a combination of *S. cerevisiae* with *Lb. sanfranciscensis* and *Lb. plantarum* is used during the sourdough fermentation process (Gobbetti, Corsetti, & Rossi, 1995). This

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observed increased production of aroma compounds in a mixed-starter process appears to be related to the proteolytic activity of LAB.

HEALTH BENEFITS**Reduction of antinutritional compounds**

Cereal grains are important sources of minerals such as iron, potassium, magnesium and zinc, but also contain phytic acid or myo-inositol hexakiphosphate, which is considered to be an anti-nutritional factor for humans. Its anti-nutritional property is due to the central hexaphosphate ring that acts as a chelator of dietary minerals preventing their absorption and thus reducing their bioavailability (Lopez et al., 2001). Phytases are widely present in plant materials such as wheat and rye flours, whose level depends on variety and crop year, but generally reported to be insufficient to significantly decrease the amount of phytic acid. However, it has been recently published that a moderate decrease of pH by sourdough fermentation is sufficient to reduce phytate content of whole wheat flour through endogenous phytase activity (Leenhardt et al, 2005).

A study on the characterization of phytase activity of sourdough microorganisms showed that combining selected yeasts and LAB it is possible to reach high level of phytate biodegradation and the best combination are *S.cerevisiae/Lb. plantarum/Leuconostoc mesenteroides* (Chaoui, Fais, & Belhcen, 2003). These results emphasize microbial potential in improving the nutritional quality

of cereal-based products. Phytate degradation in sourdough, resulting from LAB and yeast development, effectively prevents deficiencies of zinc, calcium, iron, and other essential minerals (Lopez et al, 2001)

Enhancement of nutritional values

It is worth stressing that sourdough technology allows production of 100% rye and whole meal breads, which are considered to possess increase nutritive value, mainly due to the elevated level of dietary fiber. Regular consumption of such bread reduces the risk of variety of chronic diseases such as cardiovascular disease, diabetes, and some forms of cancer and prevents premature death (Linko et al, 1997).

Sourdough bread digestibility is enhanced by enzymatic processes that take place during fermentation and increase the content of easily available carbohydrates and proteins. Health benefit of LAB development in sourdough are based mainly on lactic acid production, which results in a decrease in pH below the point at which undesirable bacteria can grow. In the human digestive tract this factor controls the quality of intestinal microflora, ensuring the proper course of digestion and excretion. Consumption of lactic-acid containing products is advised in intestinal tract malfunctions such as chronic gastritis, hyperacidity, chronic colitis, or diarrhea of unknown etiology. Lactic acid is permitted in liver, kidney or pancreas malfunction (Hammes & Tichaczek, 1994).

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Sourdough can be effective in lowering serum triglyceride and cholesterol levels as well as improving the ratio between HDL and LDL fractions. This effect may result from exopolysaccharide production occurring during sourdough fermentation (Tieking et al, 2003). In addition, acetic acid in sourdough bread can enhance the salty taste, which reduces the amounts of salt used for bread production. This factor is for great importance, especially in the case of kidney problem and hypertension.

The use of bakery starter cultures enables nutrient enrichment of bread. LAB and yeast convert minerals from the culture medium into bioavailable organic forms. Selenium supplementation may serve as a good example of elevating the content of an element deficient in the diet via sourdough bread enrichment (Diowksz et al, 2000).

Conversion of toxic compounds

Sourdough bread owes its popularity to its naturalness and tradition, however, due to its gluten content, it is toxic to people affected by celiac sprue (CS), also known as gluten-sensitive enteropathy, an autoimmune disease of the small intestinal mucosa. Ingestion of gluten causes self-perpetuating mucosal inflammation and subsequent loss of absorptive villi and hyperplasia of the crypts. The list of proteins that liberate toxic peptides also includes high molecular weight glutenins. Further proteolysis of such toxic peptides is made difficult by the position and abundance of proline residues (Hausch et al, 2003). For

the above reasons, persons affected by CS cannot ingest gluten-containing products such as bread or pasta. Lactobacilli have been shown to possess an outstanding potential in decreasing the CS-inducing effects of gluten. Di Cagno et al. (2004) demonstrated active hydrolysis of various Pro-rich peptides, including the 33-merpeptide mentioned above, by some Lactobacillus species. Following this, the mixed starter composed of *Lb. alimentarius*, *Lb. brevis*, *Lb. sanfranciscensis* and *Lactobacillus hilgardii* was shown to almost completely hydrolyze gliadin fractions and consequently the resulting bread was tolerated by CS patients (Di Cagno et al., 2004).

Production of antimicrobial substances

In general, LAB play a crucial role in the preservation and microbial safety of fermented foods, thus promoting the microbial stability of the final products of fermentation (Caplice & Fitzgerald, 1999). Since LAB naturally occur in various food products, they have traditionally been used as natural food biopreservatives. Apart from lactic acid, other by products such as acetic acid, formic and propionic acids, ethanol and CO₂ inhibit the growth of spoilage microflora, preventing the growth of pathogens and putrid microorganisms. This effect is intensified by oxygen removal, which prevents the growth of acid-tolerant molds (De Vuyst & Vandamme, 1994). Protection of foods is also due to the production of antifungal compounds such as fatty acids or phenyllactic acid, bacteriocins

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and antibiotics such as reutericyclin (De Vuyst & Vandamme, 1994). Lactic acid bacteria substances of bacteriocin character effectively inhibit the development of pathogens such as *Listeria monocytogenes* or *Bacillus subtilis* that cause defects in bread during storage and are potentially dangerous to human health. This strong antimicrobial effect of sourdough eliminates the use of artificial preservatives (Caplice & Fitzgerald, 1999). Fundamental features of an antimicrobial substance to be active under food conditions is that it is produced at active concentrations and that the effect is not masked by food components.

CONCLUSION

The metabolic activities of sour dough lactic acid bacteria (LAB) and yeasts are involved in the development of the characteristic bread flavor. Organic acids together with alcohols, esters, carbonyl compounds and others, strongly affect bread flavour. Even though precursors are present in wheat flour and the largest amount of flavor substances are formed during baking, sourdough fermentation is essential to achieve an acceptable flavour, since chemically acidified bread failed in the sensory quality. Furthermore, sourdough fermentation has several advantages which are reducing antinutritional compounds, enhancing nutritional values, converting toxic compounds and producing antimicrobial substances.

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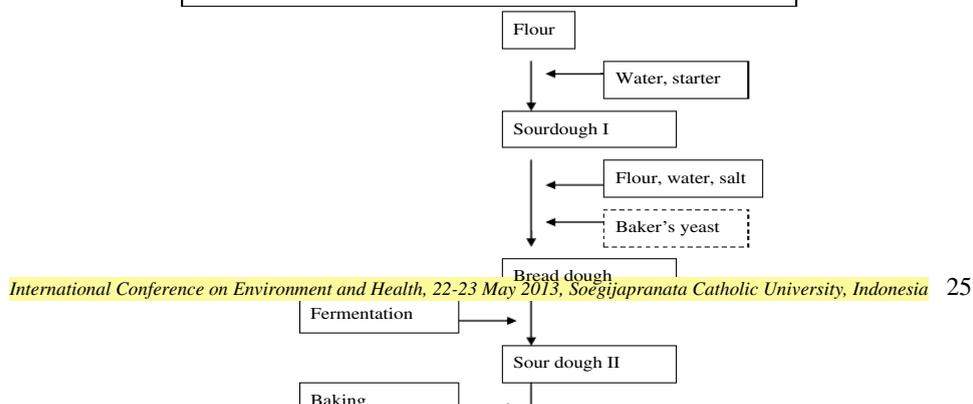
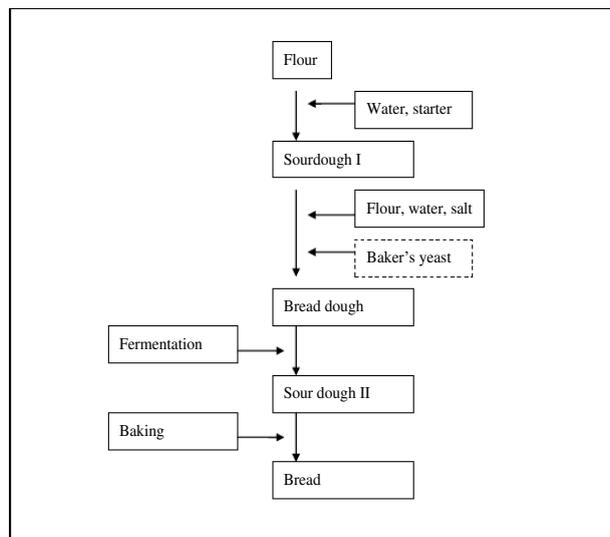
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Table 1. Lactobacillus species generally associated with sourdough fermentation or found in fermented sourdough (Corsetti & Settanni, 2007)

Obligately heterofermentative	Facultative heterofermentative	Obligately Homofermentative
<i>Lb. acidifarinae</i>	<i>Lb. plantarum</i>	<i>Lb. amylovorus</i>
<i>Lb. brevis</i>	<i>Lb. pentosus</i>	<i>Lb. acidophilus</i>
<i>Lb. buchneri</i>	<i>Lb. alimentarius</i>	<i>Lb. delbrueckii subsp. delbrueckii</i>
<i>Lb. fermentum</i>	<i>Lb. paralimentarius</i>	<i>Lb. farciminis</i>
<i>Lb. fructivorans</i>	<i>Lb. casei</i>	<i>Lb. mindensis</i>
<i>Lb. frumenti</i>		<i>Lb. crispatus</i>
<i>Lb. hilgardii</i>		<i>Lb. johnsonii</i>
<i>Lb. panis</i>		<i>Lb. amylolyticus</i>
<i>Lb. pontis</i>		
<i>Lb. reuteri</i>		
<i>Lb. rossiae</i>		
<i>Lb. sanfranciscensis</i>		
<i>Lb. siliginis</i>		
<i>Lb. spicheri</i>		
<i>Lb. zymae</i>		



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Figure 1. Scheme of sourdough bread production; baker’s yeast may be added.

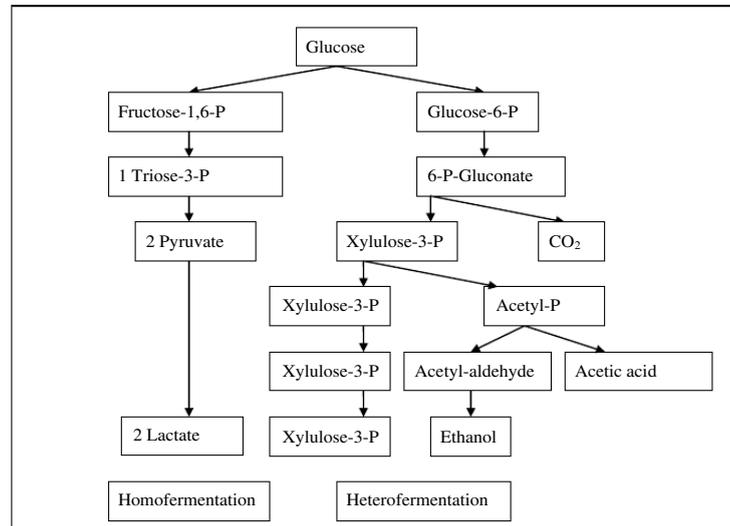


Figure 2. A simplified scheme of major pathways of glucose fermentation in LAB