SOURDOUGH YEASTS IN BREAD MAKING: A REVIEW[1*]

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ABSTRACT

Sourdough yeast is used as a starter culture in preparation of dough and bread and is basically a flour mixture with metabolically active yeast and lactic acid bacteria (LAB). Sour yeasts are grouped under three groups worldwide depending on production technologies such as; Traditionally produced sour yeast (Type I), Industrially produced semi-fluid sour yeast (Type II) and Industrially produced dried sour yeast (Type III). Each species of sour yeast has its own unique LAB microflora. During sour yeast fermentation, as a result of microorganism activities, the acetic acid, lactic acid and ethyl alcohol bread in which heterofermentative LAB create a unique aroma. In addition, added sour yeast to the bread dough supports less elastic and smoother dough formation and delays the stale of the bread. The microflora of the sourdough may also change due to the process parameters such as temperature, pH as well as starter culture (grain, yoghurt, kefir, fruit and vegetable). In this study, research has been conducted on sourdough bread, which is known as the oldest biotechnological method but has become more popular and consumed with the awareness of consumers in recent years.

Keywords: *Sourdough, Sour yeast, Lactic acid bacteria (LAB), Classification of sourdough yeast*

INTRODUCTION

Bread, which is among the first foods processed by humanity, is one of the most consumed food products all around the world **[1].** In general, bread is considered a delicious and digestible food substance obtained by kneading the mixture formed by adding wheat flour, water, yeast and salt

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in certain proportions as the main element and cooking the resulting dough at a certain time and temperature after fermentation **[2].** Bread; although its content, shape and technique have changed. Many different techniques are used in its production, and one of them is the production of sourdough bread. In the production of sourdough bread, sourdough which directly affects the quality of bread is added to the dough of bread.

Sourdough yeast is utilized as a starter culture in preparation of dough and bread and is basically a flour mixture with metabolically active yeast and lactic acid bacteria (LAB). **[3].** After a while, the dough, which is left to its own state without the addition of yeast, changes, and gas bubbles form in it, the self-releasing dough softens and changes in smell. These changes in the dough are caused by flour and water, as well as microorganisms in the environment. This dough, which is spontaneously fermented and has a sour taste, is called 'sourdough' or 'sour yeast' **[4].**

Sourdough yeast contains homo- and heterofermentative LAB and yeasts in varying proportions and compositions. As a result of the symbiotic life maintained by yeasts and labs in the fermentation of sourdough, while yeasts and heterofermentative LAB produce significant amounts of CO_2, ethyl alcohol, acetic acid and other volatile compounds, as well as lactic acid, and are responsible for the blistering of the dough, homofermentative LABs ferment sugar to form lactic acid, affecting the elasticity, acidity and flavor of bread **[5, 6].**

The use of sourdough in bread making improves the volume, structure and sensory quality of bread, and extends the shelf life of bread physically and microbiologically **[7].**

The basis of sourdough is grain fermentation. Grain fermentation has significant potential because it improves nutritional quality and therefore the health effects of foods **[8].** Rye and wheat are the most commonly used cereals in making sourdough. Production of acid during yeast fermentation increases the enzyme activity such as amylases and proteases. Microbial and chemical changes in sour yeast vary depending on the type of flour, temperature, time, amount of water and type/amount of starter **[9].** From this point of view, sourdough is considered one of the main methods used to increase the sensory properties and shelf life of bread. Sour yeast fermentation can also alter the nutritional quality of bread in several ways, such as increasing mineral intake, improving the content of bioactive compounds, and delaying the digestibility of starch **[10].**

Carbohydrates found in grain products, minerals, nitrogen sources, lipids, free fatty acids, enzyme activities and endogenous factors such as temperature, dough yield

(DY), oxygen, process parameters such as fermentation time, the yeast microflora significantly affect the properties of bakery products and sourdough **[11].** Because the use of sourdough in bread making contains various parameters, it is necessary to ensure precise control. The most important of these parameters are fermentation temperature, pH value and selection of the appropriate starter culture. For this purpose, various starter cultures are used in the production of sourdough bread such as *Lactobacillus plantartum, L. brevis*, *L. reuteri*, *L. casei*, *Lactococcus*, *Candida* and *Enterococcus* often combined *with Saccharomyces cerevisiae*, also known as traditional 'bread yeast' **[12].**

Properties of Sour Yeasts

Sourdough is a traditional product that has been used for thousands of years to improve the nutritional value, sensory properties and shelf life of bread. These traditional products, called sourdough or sour yeast, are generally described as a mixture of fermented flour and water with labs and yeasts or with the addition of starter culture **[13, 14].**

Sourdough fermentation affects dough rheology in two ways: in the sourdough itself and in the sourdough bread dough. In the dough, fermentation reduces elasticity and viscosity, while adding sourdough to the bread dough provides less elastic and softer dough formation. The level of rheological changes in this dough and its effects on bread quality may vary depending on fermentation time and the ash content of the preferred flour **[15].**

During fermentation, biochemical changes occur due to microbial and natural enzymes in the protein and carbohydrate-based components contained in the flour. Due to the metabolic activities of the LAB in the environment during fermentation, many characteristics of sour yeast occur, such as lactic fermentation, synthesis of volatile compounds, proteolysis, production of mold and rope inhibitor agents **[11].**

A sourdough contains different number of yeast and LAB. Although the literature shows that a wide variety of LAB have been isolated from sour, there are a limited number of Lactobacillus species that can be highly adapted to sour yeast environment. In particular; *L. sanfranciscensis*, *L. plantarum*, *L. brevis*, *L. pontis*, *L. rossiae* species are regarded as key organisms in the production of sourdough **[16].** Several yeast species have also been isolated from sourdough, but among them only *Saccharomyces exiguus*, *Candida humilis* and *Candida krusei* species are thought to be fundamental in the fermentation process **[17].**

The ratio between water and flour in the dough is stated as dough yield (DY).

Dough yield value significantly affects dough consistency and flavor profile **[11].** Because the absorption capacity of different flour waters is different, dough of various consistency with the same dough yield can be obtained. Usually, traditional sourdoughs are hard dough, characterized by a value of approximately 150-160 DY. In contrast, a liquid sourdough pulp is characterized by a value of approximately 200 DY **[18].** A low DY value means a firmer and harder sourdough. Therefore, acetic acid production is higher than lactic acid production. Another factor that affects the DY of sourdough is the rate of acidification. As the DY value increases, it leads to a faster rate of acidification, as the diffusion of organic acids produced into the outsides will be better **[19].**

Temperature is considered one of the most important factors. It significantly affects the aroma of sour yeast and particularly the molar ratio between lactic acid and acetic acid **[20].** Accordingly, more acetic acid production occurs in a fermented hard pulp at 25-30°C, while more lactic acid production occurs in a fermented soft pulp at 35-37 °C. Optimum temperatures for the growth of sourdough LAB, depending on the strain, are between 30 and 40°C, and 25 to 27°C for yeasts **[18].** Factors such as the use of whole wheat flour, excess water content and high temperature increase the production of acid occurring in wheat yeast. **[11].**

Titratable total acidity (TTA) and pH value are important in the fermentation process. The acidity of sour pulp is measured as a function of pH and TTA values. The pH values of traditional sourdough are between 3.5 and 4.3, which is the ideal pH range for the growth of dominant sourdough microorganisms. **[21].** In general, lactic acid bacteria dominate this ecosystem due to their adaptation to the low pH of *Lactobacilli*. However, lactic acid bacteria such as *Lactococcus, Enterococcus, Pediococcus, Leuconostoc* and *Weissella* found in grain kernels and flour dominate sourdough yeasts, which is characterized by higher pH.

L. sanfranciscensis shows a high maximum growth rate in sourdough fermentation under sub-acid conditions. But because the growth rate of *C. humilis* in the 3.5-5.5 pH range does not change significantly. Therefore the multipication factor of *L. sanfranciscensis* compared to *C. humilis*, *L. sanfranciscensis* is higher than *C. humilis*. **[18].**

The substrate used for sour yeast fermentation, especially flour, is a parameter that particularly affects sour yeast. It is important to know the ash content to determine the degree of flour and the extraction rate. Because the ratio of ash found in bran is approximately 20 times that of ash found in endospermran fraction is rich in mineral and micronutrient content, which is quite important for LAB growth. The proportion of endospermin found in bran is higher in small seeds. In addition, ash increases the buffering capacity of the sourdough system, allowing it to achieve a higher TTA **[11].** *L. brevis*, *Saccharomyces cerevisiae* or a combination of these starter cultures increase the hardness of fermented sourdough breads due to a high fiber content of flour. **[22].** The fall number is an indicator for the enzymatic activity of flour. For microflora to grow, the free sugar ratio must be high. A low falling number leads to increased amylase activity **[11].**

Classification of sour yeasts

Sour yeasts are grouped under three groups worldwide depending on production technologies (Table 1);

- Type I sour yeast: Traditionally produced sour yeast
- Type II sour yeast: Industrially produced semi-fluid sour yeast
- Type III sour yeast: Industrially produced dried sour yeast **[23].**

Type 0 group is a dough in which sourdough fermentation is not performed, but commercial bread yeasts are used to provide fermentation. It is possible to distinguish between Type I, II and III groups, and each type of sour yeast has its own unique LAB microflora **[24].**

In the Type I group, sour yeasts are produced using traditional methods. In this technique, refreshing is performed daily to keep microorganisms active. During the refreshing process, a piece of yeast that has completed the fermentation process is taken and used for the next fermentation **[25].** In this way, the purpose of the microorganisms have high metabolic activity and gas formation by providing a good way for raising the dough.

When preparing sour yeasts in type i group, the dough is generally left to fermentation at 20-30°C for 3-48 hours. During the fermentation period, flour and water are regularly used to freshen the process. Type I sour yeast is predominant in the LAB, and the pH values of this dough are approximately 4 **[26].**

Sour yeasts in the type II group have emerged to meet industrial demands. Sour yeasts in this group are faster, more effective, controllable and semi-fluid. In addition, Type II sour yeasts have a higher temperature (T>30°C), longer fermentation time (48-168 hours) and higher water content than Type I sour yeasts. Type II sour yeasts are often used as acidifiers in doughs because they have a high acid content (pH <3,5) **[27].** In addition, microorganisms present in Type II sourdough show restricted metabolic activity, suggesting that in this case, they are in the stationary phase.

Type III sour yeasts are powdery sour yeasts dried by spray dryers or drum dryers. Sour yeasts in this group are mainly used in industrial furnaces as acidifiers and flavorants. The amount of acetic acid in the dry doughs of drying is changed depending on the drying technique. Because the temperature of acetic acid in the evaporation process is 118°C, dried sourdough contains less acetic acid than fresh one. Type II method is applied when sour dough with high acidity is requested **[26, 28].**

In sour yeasts obtained by Type II and Type III methods other than Type I, yeast is added from outside as a blistering agent, and this addition S. bread yeast, also known as cerevisiae **[26, 29].**

Table 1. Classification and characteristic microflora of sour yeasts **[21]**

Type I	Type II	Type III
Obligately heterofermentative	**Facultatively heterofermentative**	**Obligately homofermentative**
L. sanfranciscensis	*L. brevis*	*L. brevis*
L. brevis	*L. fermentum*	
L. buchneri	*L. frumenti*	
L. fermentum	*L. pontis*	
L. fructivorans	*L. panis*	
L. pontis	*L. reuteri*	
L. reuteri	*L. sanfranciscensis*	
W. cibaria	*W. confuse*	
Facultatively heteroferentative		**Facultativelyheterofermentative**
L. alimentarius		*L. plantarum*
L. casei		*P. pentosaceus*
L. paralimentarius		
L. plantarum		
Obligately homofermentative	**Obligately homofermentative**	
L. acidophilus	*L. acidophilus*	
L. delbrueckii	*L. delbrueckii*	
L. farciminis	*L. amylovorus*	
L. mindensis	*L. farciminis*	
L. amylovorus	*L. johnsonii*	
Yeasts		
C. humilis		
C. krusei		

The effect of using sourdough in bread making

To obtain sourdough, a mixture of water, wheat/rye flour must be fermented with LAB and yeasts. As a result of the metabolic activities of yeasts and LAB, sour yeast develops sensory characteristics such as a distinctive aroma, but also plays a significant role in improving the nutritional and health quality of fermented products **[30, 23].** However, sourdough fermentation delays the stale of the bread during storage and protects against bacterial degradation due to the bio-transformation of the flour components in the dough stage of the bread **[31].**

The greatest contribution of sour yeast to bread quality is that acidification and metabolite production are carried out by LAB. The resulting acidification prevents the advanced degradation of starch during cooking and therefore inhibits endogenous amylase. Acidification is considered a prerequisite for ensuring an acceptable bread volume because it increases the water binding and gas holding capacities of existing pentose. **[32].**

Different metabolites such as exopolysaccharides (EPS), organic acids and/or enzymes that positively affect the texture of bread are produced by the LAB. EPS also known as hydrocollites, are produced by bacteria belonging to various food classes. EPS has a significant effect on different properties, such as increasing the viscoelasticity of the dough and the volume of bread, reducing the hardness of the bread and extending the shelf life. **[33, 34].** However, on-site production of EPS during yeast fermentation forces simultaneous acidification due to the metabolic activity of the bacteria, in which case it can greatly reduce the positive effects of EPS **[35].** In particular, acetate and lactate have been observed to significantly affect to dough rheology such as bread volume and hardness and it also offsets the benefits of EPS. **[36].** EPS created by the LAB are divided into homopolysaccharide (HoPS) and heteropolysaccharide (HePS). HoPS are made up of a kind of monosaccharide, while HePS are made up of three or eight monosaccharides. Examples of industrial EPS production are dextran from *Leuconostoc mesenteroides* and xanhtan from *Xanthomonas campestris* **[37].** Dextran is a HoPS composed of glucosyl. Due to its hydrokloid properties, they can bind a large amount of water, improve dough stability and gas retention, and maintain the freshness of the final product for longer **[36, 38].**

Acetic acid, lactic acid and ethyl alcohol are formed in the environment as a result of the activity of microorganisms during the fermentation of sour yeast, which leads to the bread gaining a unique aroma. It has been shown in studies that organic acids formed by heterofermentative LAB are more effective on aroma in the sourdough bread production. Lactic and acetic acids lead these compounds. Apart from these, minor acids such as propionic acid, isovaleric, n-butyric, α-methyl-n-valeric, valeric acid and so on are formed in very small amounts. Acetic acid ensures the formation of a strong aroma in bread, but also increases the effect of other aroma compounds **[6].**

Bread flavor varies depending on the raw materials used in general, the type of starter culture selected, the amount of sour yeast placed in it, fermentation time, fermentation temperature, pH, applied technology and cooking process **[39].** While the aroma of the bread interior is mainly due to enzymatic reactions during the fermentation process, the aroma of the bread crust is mainly due to thermal reactions during the cooking process **[40].**

Production of sour yeasts from different sources

In fermented food production, sources of microorganisms involved in fermentation may vary. The variety of LAB and yeast species that can be used as starter cultures in sour yeast batters is very wide. Given the known superiority of these microorganisms, the initial source of starter culture may have a major impact on the function of the initial organisms within a dough system **[41].**

The most important factors affecting the sourdough microbia are: process parameters such as water content and temperature; used grain flours microbiological, enzymatic, nutritional, and sensory properties; grain flour in the content of the interaction between LAB; other species of bacteria and yeast found in flour **[42, 27].**

When the microflora of natural sour yeast is formed, it is usually used in flour, the environment or things used as inoculum, such as fruit, yogurt, honey and etc **[17].**

Yogurt, kefir and/or kefir grain can be used as a starter in the production of sour yeast. In one study, sour yeast and sourdough breads were made using kefir grains developed from whey. As a result of the study, it was found that kefir acts on the pH and titration acidity (TTA) of sour yeast samples. However, commercial kefir sourdough bread made with yeast produced using the moisture retention of breads is compared with the capacity more than if a firmer texture, higher acidity (pH 4,9–5,5 and TTA 2.8–5.0 mg lactic acid/g) and have been identified they can maintain their freshness longer **[43].**

Sour yeast cultures used in bread production are mostly derived from flour or grain-derived foods. In addition, grapes, wet/dry apples, peaches, figs and such fruits with high sugar content are used. **[40].**

Fruits can be used to make sour yeast when in season. For example, breads prepared with sour yeasts from various fruits such as apples, grapes, pears, oranges have a unique taste and are preferred by consumers **[44].**

Fruits are an important part of a healthy diet. Due to its low energy content, high content of vitamins C and B complex, provitamin A, mineral substance and dietary fiber, it is considered a very important source for today's nutrition and human health.

Raw vegetables and fruits are subject to lactic acid fermentation when anaerobiose, water activity, salt concentration and suitable conditions such as temperature are provided **[45].** Lactic acid fermentation improves both the organoleptic and nutrient quality of fermented fruits and vegetables, as well as preserving the color pigments of such foods **[46].**

LAB forms a small part of the autochthonous (natural) microbiota (2.0-2.4 log CFU/g) of raw fruits and vegetables. The main LAB species isolated from self-fermenting vegetables and fruits are given in the Table 2. [**45].**

Table 2. LAB isolated from vegetables and fruits **[45]**

Lactic Acid Bacteria	**Source**
Lactobacillus plantarum	Tomato, Zucchini, Carrots, Cucumbers, Eggplant, Capers, Beetroot, Pineapple, Plum, Kiwi, Papaya, Fennel, Cherry, Cabbage
Lactobacillus pentosus	Capers, Papaya, Eggplant, Cucumber
Lactobacillus rossiae	Pineapple
Lactobacillus fermentum	Fresh Beans, Beets, Capers, Eggplant, Melon
Lactobacillus curvatus	Pepper
Lactobacillus brevis	Tomato, Capers, Eggplant, Cabbage, Cucumber, Melon
Lactobacillus paraplantarum	Cabbage, Capers
Leuconostoc mesenteroides subsp. mesenteroides	White Cabbage, Carrot, Pepper, Cucumber, Eggplant, Cherry, Lettuce
Weissella soli	Carrot
Weissella confusa/Weissella cibaria	Pepper, Tomato, BlackBerry, Papaya
Enterococcus faecalis/Enterococcus faecium	Fresh Beans, Tomatoes, Capers, Melons
Pediococcus pentosaceus	Fresh Beans, Tomatoes, Cucumbers, Capers, Cherries, Cabbage

In one study, it has been compared the fermentation activity of three sourdough breads containing sugarcane, apple and grape yeasts and concluded that yeast derived from grapefruit is the best **[47]**. In another study, the process of fermenting sourdough using apples and rye was compared and it was discovered that sour yeast prepared with apples produces more gas in total **[41].**

Studies have shown that strains of yeast and LAB derived from grapes, apples, peaches and other fruits with high sugar content differ from those associated with cereals. According to information from previous studies, it was observed that sour bread made with cultures whose source of production is fruit was less sour compared to bread made with cultures derived from cereals. **[48].**

CONCLUSION

Bread, which is constantly used from the past to the present and is not missing from our tables, can be produced by different methods. Sourdough yeast used in its production affects the physical and chemical properties of bread, so the consumer will choose which bread. As a result of the metabolic activities of LAB and yeasts, sourdough has different sensory characteristics, such as a distinctive pleasant aroma, as well as prolonging the shelf life of the product. The microbiota of sourdough yeast is home to many different organisms which are obtained from different foods such as cereals, milk and dairy products or even fruits and vegetables. In obtaining LAB, which are important for sour yeast microbiota, because they are less sour, especially compared to those obtained from grain, more fruit and vegetable sources can be used and studies can be done on this issue and new information can be added to the literature.

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