IJFAAS International Journal of Food and Allied Sciences
ISSN: 2415-0290 (Print)
ISSN: 2413-2543 (Online)
DOI:10.21620/ijfaas.201618-14

Research Article

Utilization of indigenously isolated single strain starter cultures for the production of sourdough bread

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History
Submitted: Aug 31, 2015
Revised: Oct 30, 2015
Accepted: Nov 20, 2015
Online: Aug 06, 2016

Keywords
Lactic acid bacteria
Starter cultures
Hetero-fermentative
Lactic acid

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Abstract

Sourdoughs were prepared with Saccharomyces cerevisiae (T_0) and indigenously isolated starter cultures i.e Lactobacillus brevis (T_1), Lactobacillus fermentum (T_2) and Lactobacillus plantarum (T_3). Breads were prepared from all sourdoughs samples in triplicate and analyzed for pH, Total Titratable Acidity (TTA), loaf volume, microbial characteristics (total plate count and fungal count) and sensory profile (internal and external). The breads prepared from Saccharomyces cerevisiae (T_0) exhibited the highest pH with the lowest TTA while T_1 showed the lowest pH with the highest TTA. The T_0 breads got the highest values for loaf volume followed by T_1. The breads produced with the addition of hetero-fermentative starter cultures (T_1 and T_2) showed resistance against the growth of the contaminating microorganisms. In the sensory evaluation, the breads produced with T_1 ranked the best for color (crust and crumb), taste, aroma, texture and overall acceptability by the panelists.

1. Introduction

Production of bread is a hydrothermal process. Baking temperature and flour constituents have significant impact on the quality of the end product (Banu et al., 2011). The mixture of ground cereals with water forms the dough, which has a sour aroma when left for some time. This is perhaps the first-time discovery of the fermented product (Lorenz & Brummer, 2003). Sourdough preparation is actually mid stage product between dough and bread preparation, which contains ingredients like water, flour, salt and microorganisms i.e lactic acid bacteria (LAB) and yeast (Rollan et al., 2010). The modern bakeries use sourdough as a leavening agent due to its advantages like production of flavor and high organoleptic characteristics of the final product (Katina et al., 2006). Leavening through sourdough is the oldest biotechnological technique in the fermented foods production. Sourdough was actually a small piece of dough, from previous successful fermentation which was incorporated in the final dough with other ingredients i.e flour, water and salt (Stolz, 2003).

Bacterial proteolysis during sourdough fermentation results in the production of particular sourdough bread aroma as compared to yeast fermented breads (Thiele, Ganzle & Vogel, 2002). In fermented products, LAB plays an important role in the production of flavor. It develops typical lactic acid taste and produces volatile compounds due to proteolytic and lipolytic activities (Yvon & Rijnen, 2001). LAB also increases the shelf life of sourdough bread (Banu, Vasilean, & Aprodu, 2011). LAB produces several natural antimicrobials compounds including organic acids (acetic acid, lactic acid, formic acid and caproic acid), hydrogen peroxide, carbon dioxide, diacetyl, ethanol and bacteriocins. The organic acid prevenst mold spoilage and improvse flavor of sourdough bread (Messens & De Vuyst, 2002). The improvement of microbial shelf life of sourdough bread is due to the antifungal effect of acetic acid instead of lactic acid (Corsetti & Settani, 2007).

Baked goods are spoiled often by fungal attack, which is also considered as public health concern due to the production of mycotoxins (Gerez et al., 2009). The fungi, which most commonly cause damage to the bakery products, belong to the genera of *Aspergillus*, *Fusarium*, *Mucor*, *Penicillium* and *Rhizopus* (Keshri, Voysey, & Magan, 2002). Sourdough is the most suitable technique

to preserve baked goods free from fungal spoilage among other techniques like chemical additives (such as sorbic, propionic, acetic acid and benzoic acid), irradiation and suitable packaging (Brock & Buckel, 2004). The sourdough addition with its additive-free image has been reported to be the best method of preservation for bread and other bakery products (Rollan et al., 2010). Given the benefits of sourdough technology, the present study was conducted to prepare sourdough bread using indigenous starter cultures (*Lactobacillus brevis*, *Lactobacillus fermentum* and *Lactobacillus plantarum*).

2. Materials and methods

2.1. Procurement of raw materials

The starter cultures of pure bacterial strains of the genus Lactobacillus i.e. *L. brevis*, *L. fermentum*, and *L. plantarum* were obtained from Microbiological Laboratory of National Institute of Food Science and Technology, University of Agriculture, Faisalabad. Commercial straight grade flour, baker's yeast (*S. cerevisiae)* and salt were purchased from the local market of Faisalabad.

2.2. Physicochemical analysis of flour

The flour was analyzed for moisture, crude protein, crude fibre, crude fat, ash and NFE according to their respective standard methods described in AACC (2000).

2.3. Preparation of starter culture for sourdough

The starter cultures of pure bacterial strains of the genus Lactobacillus i.e. *L. brevis*, *L. fermentum* (heterofermentative) and *L. plantarum* (homofermentative) were used for the preparation of sourdough samples (Table 1). Actively growing single cultures of lactic acid bacteria were inoculated (inoculum level 1.0% v/v) into Erlenmeyer flasks containing 100 mL of de Man Rogosa Sharpe (MRS) broth and incubated for 24 hr at 30°C. Biomass was collected by centrifugation (5000×g, 15 min, 4°C) and resuspended into 50 mL of sterile saline. This cell suspension that contained 10^8 CFU/mL of lactic acid bacteria was used as starter culture in sourdoughs preparation (Robert et al., 2005).

2.4. Sourdough preparation

A sponge was prepared using flour (200 g) and water (200 mL) and inoculated with each starter culture (to give CFU of 10^8 bacteria per gram of dough). This was mixed thoroughly and incubated for 20 hr at 30°C. The sourdoughs were prepared in triplicates from flour (200 g), water (200 mL) and fermented sponge (70 g) from each starter culture, incubated for 12, 18 and 24 hr at 30°C. Sourdoughs prepared from each starter culture were incorporated at 20 g/100 g (base flour) in corresponding breads dough. Leavening was ensured by addition of a small amount of baker's yeast (0.2 g/100 g). The bread's dough containing *S. cerevisiae* alone in the same amount were included in the test series as T_0 (Robert et al., 2005).

Table 1. Treatment plan for the preparation of sourdough breads

Treatments	Starter cultures	Fermentation time (hr)
T_0	*Saccharomyces cerevisiae*	12 18 24
T_1	*Lactobacillus brevis*	12 18 24
T_2	*Lactobacillus fermentum*	12 18 24
T_3	*Lactobacillus plantarum*	12 18 24

2.5. Preparation of sourdough bread

The bread baking potential was estimated for each sample by the sponge dough method 10-11 as described in AACC (2000). The ingredients (1000 g of flour, 640 mL tap water, 200 g sourdough prepared as described above, 2 g of baker's yeast and 44 g salt) were mixed homogenously for 5 min in a Hobart A-200 Mixer to form the sourdoughs and allowed to ferment at 30°C and 75% R.H. for 180 minutes. The sourdoughs were molded and panned into 100 g test pans and final proofing was done for 45 minutes at 35°C and 85% R.H. The breads were baked at 232°C for 25 minutes. Breads prepared in triplicate were kept at room temperature for further analysis.

2.6. Physicochemical analysis of breads

Breads were analyzed for pH, Total Titratable Acidity (TTA), loaf volume, microbial characteristics (total plate count and fungal count) and sensory profile (internal and external) in triplicate.

2.6.1. pH and TTA

Ten grams 10 g of bread crumb and 100 mL of distilled water were placed in a clean dry container, which was sealed and stirred until the bread dispersed into a semi-liquid mixture. The pH was recorded using pH-meter (inoLab pH 720, WTW 82362). The total titratable acidity (TTA) values were determined as the amount of 0.1 N NaOH solution (mL) used to neutralize 10 g sample (Banu, Vasilean, & Aprodu, 2011).

2.6.2. Loaf volume

The loaf volume, after cooling for 15 min, was measured using the rapeseed displacement method. Each loaf was put in a container and covered with rapeseed to totally fill the container. Then the loaf was removed and the volume of the rapeseed was recorded by following the method as described in AACC (2000).

2.7. Microbiological analysis

The bread samples were subjected to microbial analysis for total plate count (TPC) and fungal growth during the storage period according to the method as described by Yousef & Carlstrom (2003).

2.8. Sensory evaluation

Sensory characteristics of the bread samples were assessed by a panel of 10 judges in sensory evaluation laboratory, National Institute of Food Science and Technology, University of Agriculture, Faisalabad. The coded samples were served in clean white plastic plates at room temperature in individual booths with adequate florescent light. Sample presentation to the panelist was at random and one at a time. After the products were tested, the unconsumed product and plate were removed prior to the next product presentation. The panelists were given enough water to rinse their mouth between each sample. The panelists were asked to rate the bread samples for external properties (volume, crust color, symmetry of form, evenness of bake, crust character), internal characteristics (grain, crumb color, taste, aroma, texture) and overall acceptability on Pearson scale. The external and internal characteristics were evaluated over 30 and 70 total points, respectively (Land & Shepherd, 1988).

2.9. Statistical analysis

The samples for each parameter were run in triplicate and the data obtained was subjected to statistical analysis.

Table 2. Composition of commercial wheat flour

Constituent	Percent (%)
Moisture	12.87
Crude protein	10.34
Ash	0.60
Crude fiber	0.65
Crude fat	1.44
NFE	74.10

Completely Randomized Design (CRD) was applied and level of significance was defined as $p<0.05$. Means were further compared through Duncan's multiple range test (DMRt) following the procedure as described by Montgomery (2008).

3. Results and discussion

3.1. Commercial wheat flour compositions

The wheat flour was analyzed for moisture content, ash, protein, crude fat, crude fibre, NFE and values were found to be 74.30%, 12.56%, 0.60%, 9.65%, 1.4%, 0.64%, 74.30% respectively (Table 2). The results for the analysis of wheat flour are in line with the findings of Sameen, Niaz, & Anjum (2002) who analyzed different wheat varieties and observed 11.82 to 13.91 % moisture content, 10.54 to 13.00 % protein, 0.82 to 0.92 % fat, 0.11 to 0.15 % fiber and 0.36 to 0.48 % ash content.

3.2. pH and TTA of sourdough breads

pH varied significantly among the breads prepared from different starter cultures and maximum pH was found in the breads produced from *S. cerevisiae* (6.10) followed by T_3. The pH in breads decreased as a function of fermentation time in all starter cultures and the lowest pH (3.88) was observed in breads prepared from T_1 after 24 hours of fermentation time followed by the production of breads from the same culture after 18 hours of fermentation time. The breads prepared from the *S. cerevisiae* showed the lowest content of TTA (2.20%) as

compared to the breads prepared from other LAB starter cultures. The breads prepared from T_1 yielded significantly the highest content of TTA (8.90%) followed by T_2. There was a significant increase in the total titratable acidity with an increase in fermentation time. The breads from *S. cerevisiae* exhibited significantly the lowest TTA as shown in Table 3. The

Table 3. Effect of starter cultures and fermentation time on pH, TTA and loaf volume of sourdough breads (mean ± SD)

Treatments	pH			TTA (%)			Loaf volume		
	12 hr	18 hr	24 hr	12 hr	18 hr	24 hr	12 hr	18 hr	24 hr
T_0	6.10±0.23	5.90±0.19	5.65±0.18	2.20±0.07	2.60±0.10	3.10±0.11	472.88±15.13	514.00±15.42	507.84±18.28
T_1	4.20±0.16	3.95±0.13	3.88±0.12	7.00±0.22	8.40±0.32	8.90±0.31	476.66±15.25	480.66±14.42	478.66±17.23
T_2	4.43±0.16	4.16±0.14	4.02±0.13	6.20±0.19	7.10±0.27	7.70±0.27	470.00±15.04	472.66±14.18	473.00±17.03
T_3	4.67±0.17	4.29±0.14	3.96±0.13	6.10±0.19	6.60±0.25	6.90±0.24	456.66±14.61	466.66±14.00	467.66±16.84

Table 4. Total plate count (CFU/g) at different storage intervals in breads (mean of three replications)

Cultures	Fermentation Time (Hr)	0 Days	24 hr	48 hr	72 hr	96 hr	120 hr
T_0	12	3.2×10^2	5.2×10^3	3.4×10^4	3.7×10^5	4.6×10^7	4.9×10^7
	18	2.7×10^2	8.0×10^3	2.7×10^4	2.9×10^6	3.3×10^6	3.2×10^7
	24	4.0×10^2	2.6×10^4	6.7×10^5	6.4×10^6	7.0×10^7	7.1×10^7
T_1	12	-	6.2×10^2	4.0×10^2	7.5×10^3	8.1×10^3	8.0×10^3
	18	-	1.8×10^1	2.3×10^2	5.1×10^3	5.6×10^3	5.7×10^3
	24	-	5.4×10^1	1.1×10^2	3.4×10^3	3.9×10^3	4.1×10^3
T_2	12	-	6.2×10^2	4.8×10^3	7.2×10^3	3.9×10^4	4.3×10^4
	18	-	5.3×10^2	3.7×10^2	6.4×10^3	2.5×10^4	3.1×10^4
	24	-	4.0×10^1	2.6×10^2	5.5×10^3	2.1×10^4	2.8×10^4
T_3	12	6.9×10^1	4.2×10^2	6.3×10^3	8.6×10^4	8.9×10^5	9.0×10^5
	18	5.5×10^1	7.7×10^2	3.6×10^3	5.4×10^4	6.1×10^4	6.0×10^5
	24	5.4×10^1	3.8×10^2	1.4×10^3	3.9×10^4	4.4×10^4	5.0×10^5

Table 5. Total fungal count (CFU/g) at different storage intervals in breads (mean of three replications)

Cultures	Fermentation Time (Hr)	0 Days	24 hr	48 hr	72 hr	96 hr	120 hr
T_0	12	4.1×10^2	1.6×10^3	8.8×10^4	3.1×10^5	4.6×10^6	5.5×10^7
	18	9.6×10^2	2.9×10^3	2.7×10^4	5.7×10^5	3.8×10^6	4.3×10^7
	24	5.5×10^2	4.4×10^3	5.6×10^4	3.9×10^5	7.7×10^6	7.9×10^7
T_1	12	-	-	-	-	6.4×10^2	5.7×10^3
	18	-	-	-	-	4.1×10^2	4.2×10^3
	24	-	-	-	-	3.3×10^2	3.2×10^3
T_2	12	-	-	-	7.3×10^1	5.0×10^2	6.2×10^3
	18	-	-	-	4.4×10^1	3.5×10^2	4.5×10^3
	24	-	-	-	2.8×10^1	2.1×10^2	4.2×10^3
T_3	12	-	-	8.1×10^1	6.6×10^3	5.2×10^4	3.1×10^5
	18	-	-	6.4×10^1	4.5×10^3	3.4×10^4	2.7×10^5
	24	-	-	4.5×10^1	2.9×10^3	2.9×10^4	1.6×10^5

T_0 = *Saccharomyces cerevisiae,* T_1 = *Lactobacillus brevis,* T_2= *Lactobacillus fermentum,* T_3 = *Lactobacillus plantarum*

results of the present study are similar to the findings of Paramithiotis et al. (2005) who prepared sourdough bread by using different starter cultures. Drop in the pH and increase in acidity of sourdough bread is due to the production of acids produced by homofermentative and heterofermentative LAB. Production of lactic acid is always higher than acetic acid (Vuyst & Neysens, 2005).

3.3. Loaf volume of sourdough breads

The loaf volume of breads containing *S. cerevisiae* got significantly the highest loaf volume (514.00) followed by the T_1. However, the breads prepared from T_3 got significantly the lowest values for loaf volume. The results indicated that the loaf volume of breads increased with the increase in fermentation time (Table 3). LAB had positive effect on the volume of sourdough bread (Clarke, Schober, & Arendt, 2002). The positive effect of sourdough in bread volume is related to better gas holding capacity of gluten in sourdough (Ryan et al., 2011),

solubilization of pentosans during sourdough fermentation process (Corsetti et al., 2000), the activities of endogenous enzymes, subsequent low pH (Clarke et al., 2003), and faster yeast fermentation in the presence of LAB (Corsetti et al., 1995). The improvement in loaf volume of breads in T_1 and T_2 might be due to the ability of heterofermentative LAB for production of more CO_2 (Messens et al., 2002).

3.4. Microbiological analysis of sourdough breads

3.4.1. Total plate count (TPC)

The microbiological analysis of breads at different storage intervals showed that the microbial loads increased with the increase of time in all the starter cultures on overall basis. The total plate count was the highest ($7.1x10^7$ CFU/g) in breads prepared from *S. cerevisiae*. However, the total plate count was the lowest in breads prepared from T_1 ($4.1x10^3$ CFU/g) followed by the breads prepared from T_2 ($3.1x10^4$ CFU/g) after 120 hours of storage (Table 4). The breads produced with the addition of heterofermentative starter cultures (T_1 and T_2) exhibited resistance against the growth of the contaminating microorganisms. The results are in line with the finding of Rosenquist & Hansen (2000) who observed that during the storage of bread, microbial load increases with the passage of time. The present study suggested that the use of heterofermentative LAB starter cultures (T_1 and T_2) showed lower TPC indicating resistance against the growth of microorganisms.

3.4.2. Fungal count

The microbiological analysis of breads at different storage intervals showed that the fungal load increased by the increase in storage period in the breads produced with the addition of all starter cultures. The fungal count was the highest ($7.9x10^7$ CFU/g) in breads prepared from *S. cerevisiae*. However, the fungal count was the lowest in breads prepared with the addition of T_1 followed by breads prepared by the addition of T_2 after 120 hours of storage (Table 5). The breads produced by the addition of heterofermentative starter cultures (T_1 and T_2) showed more resistance against the growth of molds than homofermentative starter culture (T_3). The antifungal effect may be attributed to the acidification during sourdough fermentation process (Ryan et al., 2011).

3.5. Sensory evaluation of sourdough breads

Starter culture had significant effect on all the internal sensory characteristics (i.e crust colour, volume of bread, crust character) except evenness of bake and symmetry of form. The breads prepared from T_1 got significantly the highest scores for crust color followed by the breads produced from T_2 (Table 6), while T_2 achieved more scores for evenness of bake and symmetry of form followed by T_1 (Table 7). The results of the present study are comparable to that of Robert et al. (2005). The study found that the addition of LAB in sourdough has a good effect on the development of crust color in breads. The maximum scores were assigned to the volume of breads prepared from *S. cerevisiae* followed by the breads prepared from the T_1. Improved volume of the sourdough bread depends on the intensity and nature of the acids produced (Clarke et al., 2003). The sourdough breads prepared from T_1 and T_2 got the highest score for crust character. Crust character mainly depends on the maillard reaction (Paramithiotis et al., 2005).

Starter culture had significant effect on all the external sensory characteristics (i.e grain of bread, crumb colour, aroma, texture and taste). T_1 got significantly the highest scores for crumb color, aroma, texture and taste (Table 8). The improvement in aroma of breads prepared from T_1 might be due to the ability of LAB to produce flavoring compounds as reported by Onyango et al. (2000). Crowley et al. (2002) also observed that the breads containing 20% sourdough addition maintained superior textural properties. The breads prepared from control got significantly the highest scores for grain of bread followed by the breads produced from T_2. The crumb color and overall acceptability for the breads prepared from T_1 was the highest followed by T_0 (Table 9). Banu, Vasilean, & Aprodu (2011) also reported that the overall sensory characteristics depend on the type of starter culture used for the fermentation of sourdough bread. Clarke, Schober, & Arendt (2002) suggested that the addition of lactic acid bacteria in sourdough had a positive effect on overall bread quality including taste, smell and crumb color of bread.

4. Conclusions

The breads produced with the LAB starter cultures (*L. brevis, L. fermentum* and *L. plantarum*) were found acceptable with respect to physicochemical, microbiological and sensory characteristics as compared to breads produced from baker's yeast (*S. cerevisiae*).

Table 6. Effect of starter cultures and fermentation time on volume, crust color and crust character of sourdough breads

Treatments	Volume			Crust color			Crust character		
	12hrs	18hrs	24hrs	12hrs	18hrs	24hrs	12hrs	18hrs	24hrs
T_0	8.00±0.26	8.20±0.25	8.16±0.26	6.30±0.21	6.56±0.23	6.46±0.22	6.30±0.21	6.56±0.20	6.46±0.21
T_1	7.52±0.25	7.82±0.24	7.52±0.24	7.00±0.23	6.90±0.24	7.10±0.24	7.00±0.24	6.90±0.21	7.10±0.23
T_2	7.20±0.24	7.30±0.23	7.30±0.23	6.90±0.23	6.90±0.24	7.10±0.24	6.90±0.23	6.90±0.21	7.10±0.23
T_3	6.46±0.21	7.00±0.22	6.92±0.22	6.58±0.22	6.56±0.23	6.48±0.22	6.58±0.22	6.56±0.20	6.48±0.21

T_0 = *Saccharomyces cerevisiae*, T_1 = *Lactobacillus brevis*, T_2= *Lactobacillus fermentum*, T_3 = *Lactobacillus plantarum*

Table 7. Effect of starter cultures and fermentation time on symmetry and evenness of sourdough breads

Treatments	Symmetry of form			Evenness of bake		
	12hrs	18hrs	24hrs	12hrs	18hrs	24hrs
T_0	2.16±0.07	2.26±0.07	2.28±0.07	2.04±0.07	2.18±0.07	2.16±0.08
T_1	2.18±0.07	2.26±0.07	2.28±0.07	2.26±0.07	2.18±0.07	2.22±0.07
T_2	2.18±0.07	2.26±0.07	2.38±0.07	2.24±0.07	2.28±0.08	2.22±0.07
T_3	2.04±0.06	2.18±0.07	2.16±0.07	2.18±0.07	2.16±0.07	2.18±0.07

T_0 = *Saccharomyces cerevisiae*, T_1 = *Lactobacillus brevis*, T_2= *Lactobacillus fermentum*, T_3 = *Lactobacillus plantarum*

Table 8. Effect of starter cultures and fermentation time on taste, aroma and texture of sourdough breads

Treatments	Taste			Aroma			Texture		
	12hrs	18hrs	24hrs	12hrs	18hrs	24hrs	12hrs	18hrs	24hrs
T_0	9.66±0.31	10.50±0.35	10.36±0.36	6.66±0.22	7.20±0.22	7.12±0.23	10.10±0.33	11.00±0.39	10.88±0.37
T_1	10.26±0.33	10.50±0.35	10.32±0.36	7.38±0.24	7.48±0.23	7.28±0.23	11.28±0.37	11.18±0.39	10.94±0.37
T_2	7.20±0.23	7.38±0.24	7.24±0.25	6.68±0.22	6.78±0.21	6.00±0.19	10.76±0.36	10.50±0.37	10.60±0.36
T_3	6.50±0.21	6.58±0.22	6.68±0.23	6.26±0.21	6.58±0.20	6.40±0.20	9.24±0.30	9.40±0.33	9.00±0.31

T_0 = *Saccharomyces cerevisiae*, T_1 = *Lactobacillus brevis*, T_2= *Lactobacillus fermentum*, T_3 = *Lactobacillus plantarum*

Table 9. Effect of starter cultures and fermentation time on grain, crumb color and overall acceptability of sourdough breads

Treatments	Grain			Crumb color			Overall acceptability		
	12hrs	18hrs	24hrs	12hrs	18hrs	24hrs	12hrs	18hrs	24hrs
T_0	7.34±0.24	8.00±0.25	7.92±0.25	6.28±0.21	6.84±0.23	6.76±0.21	66.78±2.20	72.56±2.25	71.70±2.29
T_1	6.66±0.22	6.50±0.20	6.56±0.21	7.00±0.23	7.20±0.24	7.30±0.23	73.34±2.42	74.44±2.3	73.44±235
T_2	7.02±0.23	6.98±0.22	6.78±0.22	6.00±0.20	6.20±0.20	6.00±0.19	65.52±2.16	66.86±2.07	66.56±2.13
T_3	5.66±0.19	5.76±0.18	5.50±0.18	6.36±0.21	6.30±0.21	6.58±0.20	64.62±2.13	65.08±2.02	63.72±2.04

T_0 = *Saccharomyces cerevisiae* , T_1 = *Lactobacillus brevis*, T_2= *Lactobacillus fermentum*, T_3 = *Lactobacillus plantarum*

Among LAB starter cultures (*L. brevis, L. ferment* and *L. plantarum*) the breads prepared from T_1 (*L. brevis)* ranked the best for color (crust and crumb), taste, aroma, texture and overall acceptability by the panelists. LAB have very important role to increase microbial quality and shelf life of sourdough bread due to the production of natural antimicrobial compounds including organic acids. The sourdough breads produced from LAB inhibited microbial spoilage, extended shelf life and produced superior quality product with special reference to taste, aroma, flavor and texture as compared to Baker's yeast breads.

References

AACC. (2000). Approved Methods of American Association of Cereal Chemist. The American Association of Cereal Chemists, Inc. St. Paul. MN, USA.

Banu, I., Vasilean, I. Constantin, O.E., & Aprodu, I. (2011). Prediction of rye dough behaviour and bread quality using response surface methodology. *Irish Journal of Agricultural and Food Research,* 50: 239-247.

Banu, I., Vasilean, I., & Aprodu. I. (2011). Quality evaluation of the sourdough rye breads. *Food Technology*, 35: 94-105.

Brock, M., & Buckel, W. (2004). On the mechanism of action of the antifungal agent propionate propionyl-CoA inhibits glucose metabolism in *Aspergillus nidulans*. *European Journal of Biochemistry,* 271: 3227-3241

Clarke, C.I., Schober, T.J. Angst, E., & Arendt, E.K. (2003). Use of response surface methodology to investigate the effects of processing conditions on sourdough wheat bread quality. *European Food Research Technololgy,* 217: 23-33.

Clarke, C.I., Schober, T.J., & Arendt, E.K. (2002). Effect of single strain and traditional mixed strain starter culture on rheological properties of wheat dough and bread quality. *Cereal Chemistry,* 79: 640-647.

Corsetti, A., & Settani, L. (2007). *Lactobacilli* in sourdough fermentation. *Food Research International,* 40: 539-558.

Corsetti, A., Gobbetti, B. De Marco, B. Balestrieri, F. Paoletti, F., & Rossi, J. (2000). Combined effect of sourdough lactic acid bacteria and additives on bread firmness and staling. *Journal of Agriculture and Food Chemistry*, 48: 3044-3051.

Corsetti, A., Gobbetti, M., & Smacchi, E. (1995). Antibacterial activity of sourdough lactic acid bacteria: isolation of a bacteriocin- like inhibitory substance from *Lactobacillus sanfranciscensis* C57. *Food Microbiology*, 13: 447-456.

Crowley, P., Schober, T. Clarke, C. & Arendt, E. (2002). The effect of storage time on textural and crumb grain characteristics of sourdough wheat bread. *European Food Research Technology,* 214: 489-496.

Gerez, C.L., Torino, M.I. Rollan, G., & Font de Valdez, G. (2009). Prevention of bread mould spoilage by using lactic acid bacteria with antifungal properties. *Food Control,* 20:1 44-148.

Katina, K., Arendt, E. Liukkon, K.H. Autio, K. Flander, L., & Poutanen, K. (2006). Optimization of sourdough process for improved sensory profiles and texture of wheat bread. *LWT-Food Science and Technology,* 39: 1189-1202.

Keshri, G., Voysey, P., & Magan, N. (2002). Early detection of spoilage moulds in bread using volatile production patterns and quantitative enzyme assays. *Journal of Applied Microbiology,* 92: 165-172.

Land, D.G., & Shepherd, R. (1988). Scaling and Ranking Methods. In: "Sensory Analysis of Foods" (ed. J.R. Piggolt), Elsevier Applied Science, New York, USA, pages 155-185.

Lorenz, K., & Brummer, J.M. (2003). Preferments and sourdoughs in German breads. In: "Handbook of dough fermentations" (eds. K. Kulp and K. Lorenz), Marcel Dekker Inc., New York, pages 247-267.

Messens, W., & De Vuyst, L. (2002). Inhibitory substances produced by lactobacilli isolated from sourdoughs-a review. *International Journal of Food Microbiology,* 72: 31-43.

Messens, W., Neysens, P. Vansieleghem, W. Vanderhoeven, Johan, Vuyst, D., & Luc. (2002). Modeling growth and bacteriocin production by *Lactobacillus amylovorus* dce 471 in response to temperature and pH values used for sourdough fermentations. *Applied and Environmental Microbiology,* 68: 1431-1435.

Montgomery, D.C. (2008). Design and analysis of experiments. 7th edition. John Wiley and Sons. Inc. Hoboken, NJ, USA. 1: 650-656.

Onyango, C., Okoth, M.W., & Mbugua, S.K. (2000). Effect of drying lactic fermented uji (an East African sour porridge) on some carboxylic acids. *Journal of the Science of Food Agriculture*, 80: 1854-1858.

Paramithiotis, S., Chouliaras, Y. Tsakalidou, E., & Kalantzopoulos, G. (2005). Application of selected starter cultures for the production of wheat sourdough bread using a traditional three stage procedure. *Process Biochemistry,* 40: 2813-2819.

Robert, H., Gabriel, V. Lefebvre, D. Rabier, P. Vayssier, Y., & Faucher, C.F. (2005). Study of the behaviour of *Lactobacillus plantarum* and Leuconostoc starters during a complete wheat sourdough breadmaking process. *LWT-Food Science and Technology,* 39: 256-265.

Rollan, G., Gerez, C.L. Dallagnol, A. M. Torino, M.I., & Font, G. (2010). Update in bread fermentation by lactic acid bacteria. *Current Research and Education Topics in Applied Microbiology and Microbial Biotechnology,* 1168-1174.

Rosenquist, H., & Hansen, A. (2000). Themicrobial stability of two bakery sourdoughs made from conventionally and organically grown rye. *Food Microbiology,* 17: 241- 250.

Ryan, L.A.M. Zannini, E. Bello, F.D. Pawlowska, A. Koehler, P., & Arendt, E.K. (2011). *Lactobacillus amylovorus* DSM 19280 as a novel food grade antifungal agent for bakery products. *International Journal of Food Microbiology,* 146: 276-283.

Sameen, A., Niaz, A., & Anjum, F.M.. (2002). Chemical composition of three wheat (*triticum aestivum L.*) varieties as affected by npk doses. *International Journal of Agriculture Biology,* 4: 537-539.

Stolz, P. (2003). Biological fundamentals of yeast and lactobacilli fermentation in bread dough. In Kulp, K. and Lorenz, K. (Eds.), *Handbook of dough fermentations*, (pp. 23-43). New York: Marcel Dekker.

Thiele, C., Ganzle, M.G., & Vogel, R.F. (2002). Contribution of sourdough lactobacilli, yeast, and cereal enzymes to the generation of amino acids in dough relevant for bread flavor. *Cereal Chemistry,* 79: 45-51.

Vuyst, L., & Neysens, P. (2005). The sourdough microflora: biodiversity and metabolic interactions. *Trends in Food Science and Technology,* 16: 43-56.

Yousef, A.E., & Carlstrom, C. (2003). Food microbiology: a laboratory manual. Jhon Wiley and Sons, Pub.Co. Inc, Hoboken, New Jersey.

Yvon, M., & Rijnen, L. (2001). Cheese flavour formation by amino acid catabolism. *International Dairy Journal,* 11: 185-201.