

Effect of Using Different Types of Yeasts on the Quality of Egyptian Balady Bread

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Abstract: Balady bread dough is usually fermented with baker's yeast (*Saccharomyces cerevisiae*) but in the present study soltani starter and sourdough which obtained by using the mixed culture of *S. cerevisiae*, *Lb. plantarum* and *B. longum* in combination form were used to determine their effect on balady bread quality. Microbiological contents (lactic acid bacteria, total bacteria count and yeasts), acidification characteristics (pH and total titratable acidity), and fermentative end-products (Organic acids and folic acid) contents and the degradation of phytic acid were evaluated during both soltani starter and sourdough corresponding bread dough fermentation. The lowest pH and highest acidity and organic acid content were recorded when sourdough was used in dough fermentation. The leavening ability of yeast was enhanced when used sourdough in fermentation. LAB used in sourdough balady bread increased shelf life and delayed staling. This point was considered very important because of the major economic losses that stale bakery products may entail. In sensory analyses, the soltani starter and sourdough application were preferred well by the panel.

[Yousif, M.R.G and Safaa, M. Faïd. **Effect of Using Different Types of Yeasts on the Quality of Egyptian Balady Bread.** *J Am Sci* 2014;10(2):100-109]. (ISSN: 1545-1003). <http://www.jofamericanscience.org>. 16

Key words: Egyptian balady bread - Baker's yeast - Soltani starter - Sourdough - Organic acid - Folic acid - Phytic acid - Staling - Sensory evaluation.

1.Introduction

Yeasts are actually microbial eukaryotes which belong to ascomycetes that are good source of vitamin B and protein. Yeasts are plant-like unicellular fungi thriving on every living organism. Being living organism fungi require warmth, water, albumen or nitrogenous material and sugars to remain alive (Ali *et al.*, 2012). The conditions required for growth yeast are warmth (optimum 25-30°C), moisture and food (starch plus a small amount of sugar). Refrigeration slows down the growth so that yeast can be kept for a limited period of time. When the yeast is used, the conditions and the utensils should be kept lukewarm to obtain the best results. As soon as the yeast has been added to the dough or batter, the yeast begins to feed on the starch in the mixture, forming sugar, alcohol and carbon dioxide. The bubbles of CO₂ cause the dough to expand. The dough must be "kneaded" thoroughly to distribute the bubbles evenly and then left to rise again, usually to about double its original volume. If the mixture is left too long, acid produced by the oxidation of the alcohol results in taste sour of the product (Ali *et al.*, 2012).

The yeast used for bread manufacturing is *Saccharomyces cerevisiae*, often referred to as simply baker's yeast. It converts the fermentable sugars present in the dough into carbon dioxide and ethanol as the main products. The fermentation intensity depends on the form of the yeast and the availability of fermentable sugars in the flour, including maltose

produced by starch hydrolysis (Hutkins, 2006 and Noroul Asyikeen *et al.*, 2013).

There is no doubt that folate (vitamin B₉) has a vital role in primary cell processes, such as nucleic acid and amino acid biosynthesis. Inadequate folate intake may lead to the typical folate insufficiency disease megaloblastic anaemia and greater risks for neural tube defects (Wald *et al.*, 1996 and Berry *et al.*, 1999). In addition, the useful role of folate for more than a few other diseases such as cardiovascular diseases (Brouwer *et al.*, 1999) and some forms of cancer (Choi and Mason, 2000).

Baker's yeast, *Saccharomyces cerevisiae*, has been found to generally contain a relatively high amount of folate per weight (Witthöft *et al.*, 2006). Seyoum and Selhub (1998) described a total folate content of 24.5 µg/g of dry matter of yeast while Patring and Jastrebova (2007) reported 35.2 µg/g. Folates from yeast obviously add to the finishing folate content in yeast fermented foodstuffs, such as bread (Kariluoto *et al.*, 2004). In wheat bread folate levels were improved 2.5 times when using yeast, in place of baking powder, as leavening agent (Kariluoto *et al.*, 2004).

Bread is an important folate source which, according to a Swedish survey, is estimated to provide 13% (i.e., about 26 mg folate) of the daily folate intake of the Swedish people (Becker and Pearson, 2003). On average, Swedish women consume 84 g bread/day and men 116 g bread/day (Becker and Pearson, 2003). Nevertheless, the Swedish National Food

Administration recommends a 40% higher intake, especially of wholegrain meal bread (Enghardt *et al.*, 2003). Clearly, baker's yeast contributes significantly to the final folate content in bread (Kariluoto *et al.*, 2004; 2006 and Hjortmo *et al.*, 2008), with 65–72% found to be of yeast origin (Osseyi *et al.*, 2001). Despite some folate losses during baking, the contribution from the yeast folates is still substantial in bread baked with yeast. Kariluoto *et al.* (2004) reported that wheat bread baked with yeast contained twice as much folate as wheat bread baked without yeast.

Sourdough is a mixture of flour and water fermented with lactic acid bacteria (LAB) and yeasts, which determine its characteristics in terms of acid production, aroma and leavening (Hammes and Gänzle, 1998; Rosenquist and Hansen, 1998 and De Vuyst and Neysens, 2005). Sourdough is a unique food ecosystem: it selects for LAB and yeasts which are adapted to the environment, and hosts highly specific microbial communities (De Vuyst and Vancanneyt, 2007).

One reason for admixing sourdough into bread could be that the content of phytate would decrease, as it did in rye sourdough bread (Salovaara, 1998). A well known problem concerning grain products is the inhibition of mineral absorption by phytate (Brune *et al.*, 1992), even in small amounts. The level of phytate should not exceed 0.5 $\mu\text{mol/g}$ ingested food (Sandberg and Svanberg, 1991).

The aim of the present study was to using different types of yeasts on the quality of Egyptian balady bread and investigate the influence of these types on the quality properties of bread loaves. Also, to investigate the effects of instant active dry yeast, soltani starter and sourdough on the rheological properties of bread doughs, the final quality of the resulting breads, the amounts of organic acids, increase the folate content, the degradation of phytate content of balady bread.

2. Material And Methods

Material:

Wheat flour 82% extraction rate was obtained from South Cario Mills Company, Fysal, Giza, Egypt. Instant active dry yeast (*Saccharomyces cerevisiae*) was obtained from AKMAYA Co., Turkey. Two pure cultures of *Lactobacillus plantarum* ATCC 14917 and *Bifidobacterium longum* ATCC 15707 were obtained from Microbiological Resource Center (Cairo-MIRCEN), Faculty of Agriculture, Ain Shams University, Cairo, Egypt.

Methods:

Preparation of yeast starters:

Soltani starter:

Soltani starter has been prepared by using part of the bread dough from the previous day.

Sourdough starters:

Lactic acid bacteria biomass:

Two Lactic acid bacteria strains (*Lactobacillus plantarum* and *Bifidobacterium longum*) were used in preparation of sourdough. *Lactobacillus plantarum* was grown on MRS broth at 30°C for 24 hrs. while, *Bifidobacterium longum* was grown on modified MRS broth supplemented with 0.05% L-cystein-HCl (was added to decrease the redox potential of the medium) and 0.3% lithium chloride according to the method described by (Dinaker and Mistry, 1994 and Dave and Shah 1996) than incubated at 37°C for 24 hrs. The cells were harvested by centrifugation (5000 xg, 10 min) and washed two times with sterilized distilled water. Under these conditions 1 g of LAB pellet contained $\sim 10^8$ cfu. These pellets were used for preparation of sourdough starters.

Sourdough fermentation:

From the preliminary experimental 2% commercial instant active dry yeast (*Saccharomyces cerevisiae*) was selected as optimal inoculation percentage for bread making. Sourdough was prepared according to the method described by Plessas *et al.* (2008). 400 g of flour and various amounts of microbial biomass (w/w based on flour weight) were mixed with 200 ml tap water. The dough was mixed for 5-10 min until the correct consistency was obtained. Sourdough fermentation was carried out at 30°C for 16 hrs.

Dough fermented analysis:

Soltani starter and sourdough samples were taken and analyzed immediately after mixing and after every 8 hrs of fermentation.

Measurement of acidification power:

Potentiometric measurements of pH and total titratable acidity measurements (TTA) during dough fermentation were followed. For pH measurement a pin electrode of a pH meter (Jenway-3305, UK) was used. For TTA ten grams of fermented dough sample were diluted in 90 ml distilled water. The solution was titrated with 0.1N NaOH at pH 8.5, under shaking. The total acidity was expressed as ml NaOH (0.1N) used. Three independent measurements of pH and TTA were performed on each sample and means were calculated according to the method described by Iacumin *et al.* (2009).

Microbiological analysis:

Microbiological analysis was carried out as described by Gül *et al.* (2005). Briefly, ten grams of fermented dough sample were homogenized with 90 ml of 0.85% (w/v) sterilized NaCl. Total bacteria count (TBC) were enumerated on plate count agar (Oxoid) following the pour-plate method and incubated at 30°C for 72 hrs. Lactic acid bacteria (LAB) were counted on

modified MRS agar (Oxoid) using the pour-plate method after incubating at 30°C for 72 hrs. Yeast count was determined on Malt Extract Agar (MEA) (Oxoid). The pH of MEA was adjusted at 3.5 using sterilized lactic acid (10%). TBC, LAB and yeast counts were calculated and expressed as cfu/g dough.

Leavening ability of fermented dough:

The leavening ability of baker's yeast in soltani starter and sourdough were determined by measuring the gas volume evolved from standard dough at 30°C for 2 hr as described by **Oda and Tonomura (1993)**.

Determination of organic acids:

Organic acids (Lactic, Acetic, Citric, Pyrovic, Formic and Succinic) were determined and quantified by HPLC apparatus according to the method of **Robert et al. (2006)**.

Determination of Folic acid:

Folic acid was determined and quantified by HPLC apparatus according to the method of **Hefni et al. (2010)**.

Determination of phytic acid:

Phytic acid was determined according to the method of **Mohamed et al. (1986)**.

Rheological properties:

Rheological properties were determined by Barbender Farinograph and Extensograph according to **A.A.C.C (2000)**.

Bread making process:

Balady bread was prepared according to the method described by (**Faridi and Rubenthaler, 1984**). Bread making involved mixing 100 g wheat flour (82% extraction rate), salt (1.5% w/w), yeast (1% w/w) and 75 ml water. Ingredients were mixed until the dough was developed, then the resulted dough was let to rest for 10 min then divided. The pieces of dough were placed on a tray sprinkled with a thin layer of wheat bran and let to ferment for 40 min. at $30 \pm 2^\circ\text{C}$ and 85% relative humidity (final proofing). The pieces were then flattened to about 20 cm diameters and baked directly at 450 - 500°C for 1-2 min in a pilot plant oven (Food Technology Research Institute, Agricultural Research Center, Giza, Egypt). After baking, loaves were allowed to cool at room temperature before sealed in polyethylene bags to prevent moisture loss then storage at room temperature ($18 \pm 2^\circ\text{C}$).

Concerning to the prepared of balady bread with soltani and sourdough starters the abovementioned method used with some modification by using 20% w/w of soltani and sourdough starters.

Evaluation of balady bread qualities:

Measurement of staling rate:

The staling rates of balady bread were determined by alkaline water retention capacity (AWRC %) as described by **Kitterman and Rubenthaler (1971)**.

Organoleptic evaluation:

Fresh samples of balady bread loaves were organoleptically evaluated according to **El-Farra et al. (1982)**. The fresh samples were delivered to the panelists within 2 hr after baking. Balady bread loaves were organoleptically evaluated for general appearance, crust color, taste, odour, roundness, crumb distribution, separation layer and overall acceptability.

Statistical analysis:

Data were analysis by Analysis of Variance using General Liner Model (GLM) procedure according to the procedure reported by **Sendecor and Cochran (1997)**. Means were separated using Duncan's test at a degree of significance ($P \leq 0.05$). Statistical analyses were made using the producer of the SAS software system program (**SAS, 1997**).

3. Results and Discussion

Physicochemical and microbiological characteristics of dough samples during fermentation:

From the data presented in Table (1), it could be observed that, there was a gradual decrease in pH values in balady bread dough which fermented with using the mixed culture of *S. cerevisiae*, *Lb. plantarum* and *B. longum* during fermentation period. While, the rate of reduction of pH value was greater than that observed in soltani starter and yeast fermentation. This may be due to the great production of organic acids during fermentation by LAB strains as present in the same table. This result is in agreement with **Arendt et al. (2007)** and **Iacumin et al. (2009)**.

For yeast fermentation it could be observed that, the rate of TTA increasing was relatively lower than that observed in other doughs. These observations are in agreement with those obtained by **Robert et al. (2006)** and **Bello et al. (2007)**. After 16 hr of fermentation the highest yeast counts was obtained when used sourdough which contain both of *Lb. plantarum* and *B. longum* in combination form with *S. cerevisiae*. On the other hand, total bacteria counts were dramatically reduced during sourdough fermentation when LAB was used in combination with *S. cerevisiae*. Although LAB counts were recorded as normal flora in the control treatment (prepared using instant active dry yeast), significant increasing in these counts was obtained by the other treatments (soltani and sourdough starters). Where, LAB counts were gradually increased during sourdough fermentation. At the end of sourdough fermentation considerable increasing in LAB counts were detected comparing with control treatments and soltani starter. These results are in agreement with **Vuyst and Neysens (2005)**, **Bello et al. (2007)** and **Sadeghi (2008)**.

From the obtained data, it could be observed that, there was a gradual increase in all organic acids concentration during soltani and sourdough starters fermentation. The development of organic acid

production was higher when using the mixed cultures of *S. cerevisiae* with *Lb. plantarum* and *B. longum* in combination form for production of sourdough starters. These results are in accordance with **Hammes and Gänzle (1998)**, **Gélinas et al. (1999)** and **Robert et al. (2006)**. For folic acid content, the obtained results indicated that, there were significant changes in folic acid content after 16 hr of fermentation between soltani and sourdough starters. These observations are in agreement with those obtained by **Lin and Young (2000)**, **Crittenden et al. (2002)**, **Sybesma et al. (2003)** and **Kariluoto et al. (2004 and 2006)**. Concerning to, the degradation of phytic acid, it could be observed that, there was a gradually decrease in

phytic acid content in balady bread doughs which fermented with using the mixed culture of *S. cerevisiae*, *Lb. plantarum* and *B. longum* during fermentation period. Also, the rate of phytate degradation was greater than that observed in yeast fermentation. This may be due to the faster reduction in pH as a result to great production of organic acids during fermentation which activated the flour phytase this beside the ability of yeast and LAB to production phytase which led to completely hydrolyzed of phytic acid in bread doughs during fermentation. These observations are in agreement with those obtained by **Lopez et al. (2000, 2001 and 2002)**, **Reale et al. (2004)** and **Shatta et al. (2004)**.

Table (1): Physicochemical and microbiological characteristics of Soltani and sourdough starters during fermentation

| Type of starter | FT (hr) | *pH | *TTA | *Counts of microorganisms (log 10 cfug ⁻¹) | | | Organic acids mg/100g | | | | | | Folic acid (mg/100g) | *Phytic acid (g/100g) |
|-----------------|---------|-------------|--------------|--|-------------|-------------|-----------------------|-----------|---------|---------|----------|---------|----------------------|-----------------------|
| | | | | Y | LAB | TBC | Citri c | Succi nic | Form ic | Aceti c | Pyro vic | Lacti c | | |
| Baker's yeast | 0 | 5.72 ± 0.03 | 1.30 ± 0.10 | 6.38 ± 0.03 | 5.54 ± 0.10 | 9.48 ± 0.02 | ND | ND | ND | ND | ND | ND | 13.482 | 0.650 ± 0.03 |
| | 8 | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- |
| | 16 | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- | ---- |
| Soltani starter | 0 | 5.72 ± 0.03 | 1.30 ± 0.10 | 6.38 ± 0.03 | 5.54 ± 0.10 | 9.48 ± 0.02 | ND | ND | ND | ND | ND | ND | 13.482 | 0.650 ± 0.03 |
| | 8 | 5.45 ± 0.01 | 2.60 ± 0.17 | 6.72 ± 0.24 | 5.87 ± 0.15 | 8.89 ± 0.02 | 1.509 | 1.707 | 2.578 | 3.627 | 0.830 | 8.313 | 134.725 | 0.423 ± 0.03 |
| | 16 | 5.39 ± 0.01 | 3.17 ± 0.06 | 7.10 ± 0.13 | 6.23 ± 0.08 | 8.32 ± 0.01 | 30.767 | 9.659 | 17.199 | 12.826 | 4.423 | 27.097 | 272.453 | 0.285 ± 0.02 |
| Sourdough | 0 | 5.76 ± 0.04 | 1.13 ± 0.12 | 6.39 ± 0.02 | 7.19 ± 0.01 | 9.48 ± 0.03 | ND | ND | ND | ND | ND | ND | 13.482 | 0.650 ± 0.03 |
| | 8 | 4.90 ± 0.02 | 5.60 ± 0.20 | 7.12 ± 0.04 | 8.10 ± 0.01 | 8.22 ± 0.03 | 4.377 | 13.184 | 3.129 | 48.910 | 3.768 | 107.188 | 152.884 | 0.276 ± 0.03 |
| | 16 | 4.25 ± 0.02 | 10.40 ± 0.10 | 7.82 ± 0.02 | 9.16 ± 0.01 | 6.90 ± 0.05 | 80.056 | 71.204 | 43.113 | 233.393 | 14.426 | 571.368 | 287.139 | ND |

* Means of triplicate ± SD. ND: Not detected. FT: Fermentation time. Y: Yeast. LAB: Lactic acid bacteria. TBC: Total bacteria count.

Leaving ability of Baker's yeast, soltani and sourdough starters:

Leavening ability of yeast in soltani and sourdough starters were determined after 16 hr of fermentation and expressed as ml CO₂ per gram starter as presented in Figure (1). The production of CO₂ was gradual increased with increasing the fermentation period for all treatment. Also, the ability of *S. cerevisiae* to produce CO₂ was enhanced in the presence of either soltani or sourdough starters. However, the increasing rate of CO₂ production in the presence of sourdough starter was higher than those obtained when soltani starter was used. These results verify that the LAB played an important role for improving the leavening ability of yeast in the production of sourdough starter. These results are in good accordance with those reported by **Martinez-Anaya et al. (1990)**, **Gobbetti (1998)** and **Häggman and Salovaara (2008)**.

Farinograph and extensograph parameters of dough behaviour of wheat flour with addition 20% of soltani and sourdough starters:

The farinogram and extensogram parameters of dough behaviour of wheat flour with addition 20% of soltani and sourdough starters are presented in Table (2). From the obtained data, it could be noticed that the water absorption of the wheat flour blended with addition 20% of soltani and sourdough starters were lower than control sample. The decreased in water absorption may be due to adding 20% of soltani and sourdough starters which contained the optimum level of water absorption preceding. The stability time of wheat flour dough was recorded 7.5 min. while, when soltani or sourdough starters were added it could be observed that the dough stability time was decreased by different levels according to the results presented in the Table (2). These results are in agreement with those obtained by **Bleux et al. (1997)**, **Di Cagno et al. (2002)**, **Thiele et al. (2002 and 2003)**.

Concerning to the mixing tolerance index, it could be noticed that the samples which recorded low dough stability, had higher tolerance index values. Also, the results in the above mentioned Table proved proportional correlation between the degree of softening and tolerance index. However, the degree of softening of the dough is a result of the breakdown of gluten net work after elapsing an appropriate mixing time.

In the same time, similar observation was noticed for the degree of softening. These results are in good accordance with those reported by **Kaur and Singh (1999)** study the effects of addition of 0.05, 0.1, 0.2 and 0.4% acetic acid on dough rheological properties. They found that, both of water absorption and dough stability were decreased, while degree of softening was increased with increase acetic acid concentration. Also, **Clarke et al. (2002)** reported that the bread dough which prepared using chemically acidified by the addition of lactic acid alone decreased both of dough stability time and degree of softening compared with control sample.

Also, from the obtained data, it could be noticed that the extensibility of wheat flour dough was recorded 127 mm. while, when soltani or sourdough starters were added it could be observed that the dough extensibility was decreased by different levels as present in the same Table. These results agree well with those reported by **Bleux et al. (1997)**, **Di Cagno et al. (2002)** and **Thiele et al. (2002)** mentioned that the hydrolysis of proteins in sourdoughs is attributable to cereal proteases, and sourdough lactic acid bacteria.

On the other hand, data in the same Table show that addition 20% of soltani or sourdough starter caused decrease in the values of resistance to extension by different levels as present in the above mentioned Table. These results are in agreement with those obtained by **Clarke et al. (2002)** and **Thiele et al. (2003)** reported that the addition of sourdough reduced the elasticity of the dough. Concerning to the proportional number. It could be observed that, the values of proportional number were decreased by different levels for all dough samples which prepared using 20% of soltani or sourdough starter when compared to the control sample (Without using any starter). This effect may be due to the decreased of resistance to extension and extensibility values.

For the energy, the control sample which prepared without using any starter recorded the highest value, while other treatments were lower in the energy values.

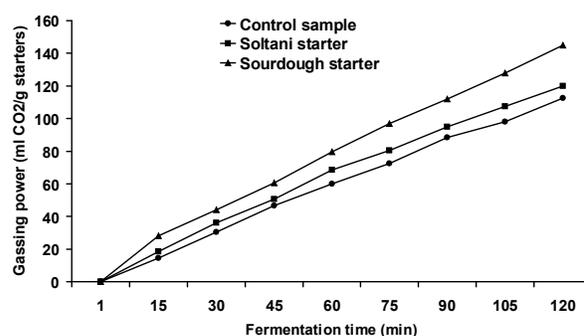


Figure (1): Leaving ability of baker's yeast, soltani and sourdough starters

Table (2): Farinograph and extensograph parameters of dough behaviour of wheat flour with addition 20% of prepared soltani and sourdough starters

| Dough samples | Farinograph parameters | | | | | | Extensograph parameters | | | |
|---------------------------------------|------------------------|--------------------|------------------------|-----------------------|-----------------------------|--------------------------|--|------------------------|---------------------------|---------------------------|
| | Water absorption (%) | Arrival time (min) | Development time (min) | Dough stability (min) | Mixing tolerance index (BU) | Degree of softening (BU) | Maximum resistance to extension "R" (BU) | Extensibility "E" (mm) | Proportional number (R/E) | Energy (Cm ²) |
| Control sample (without any starters) | 56.8 | 1.0 | 2.0 | 7.5 | 60 | 80 | 640 | 127 | 5.04 | 135 |
| Baker's yeast | 57.4 | 1.5 | 2.5 | 7.0 | 70 | 100 | 610 | 125 | 4.88 | 123 |
| Soltani starter | 39.3 | 1.5 | 3.0 | 6.0 | 80 | 110 | 520 | 115 | 4.75 | 98 |
| Sourdough | 39.8 | 2.0 | 4.0 | 5.5 | 100 | 130 | 480 | 102 | 4.71 | 75 |

Physicochemical characteristics of fresh balady bread samples:

From the data presented in Table (3), it could be noticed that the control sample which prepared using baker's yeast was recorded the highest value of pH,

followed by bread sample which prepared by using soltani starter being 5.87 and 5.42, respectively. Meanwhile, bread sample which prepared by using sourdough starter had the lowest pH value being 5.04. On the contrary, the control bread sample which

prepared using baker's yeast was recorded the lowest value of TTA, followed by bread sample which prepared by using soltani starter being 0.97 and 1.83, respectively. Meanwhile, bread sample which prepared by using sourdough had the highest TTA value being 3.07. These results are in agreement with those obtained by **Martinez-Anaya *et al.* (1990)** and **Clarke *et al.* (2002)**.

Concerning to the organic acids content of balady bread, it could be observed that, the control bread sample was recorded the lowest value of organic acids content, followed by bread sample which prepared by using soltani starter. Meanwhile, bread sample which prepared by using sourdough starter had the highest organic acids content. These observations are in agreement with those obtained by **Damiani *et al.* (1996)**, **Hammes and Gänzle (1998)**, **Gélinas *et al.* (1999)** and **Robert *et al.* (2006)**.

For folic acid content, the obtained results indicated that, there were no significant changes in

folic acid content between bread samples which prepared by using soltani or sourdough starter. While, there was significant change in folic acid content between bread samples which prepared by using soltani or sourdough starter and control sample which prepared using baker's yeast. These results are in agreement with **Osseyi *et al.* (2001)**, **Crittenden *et al.* (2002)** and **Kariluoto *et al.* (2004)**.

From the same Table, it could be observed that, the control bread sample which prepared using baker's yeast was recorded the highest value of phytic acid being 0.461 g/100g phytic acid, followed by bread sample which prepared by using soltani starter and bread sample which prepared by using sourdough starter being 0.280 and 0.143 g/100g, respectively. These results agree well with those reported by **Lopez *et al.* (2001)**, **Reale *et al.* (2004)** and **Shatta *et al.* (2004)**.

Table (3): The pH value, TTA, organic acids, folic acid and phytic acid content of fresh balady bread samples

| Type of starter | *pH | *TTA | Organic acids mg/100g | | | | | | Folic acid (mg/100g) | *Phytic acid (g/100g) |
|-----------------|-------------|-------------|-----------------------|----------|--------|--------|---------|---------|----------------------|-----------------------|
| | | | Citric | Succinic | Formic | Acetic | Pyrovic | Lactic | | |
| Baker's yeast | 5.87 ± 0.01 | 0.97 ± 0.15 | 0.329 | ND | ND | 0.726 | 0.286 | 0.986 | 27.859 | 0.461 ± 0.01 |
| Soltani starter | 5.42 ± 0.01 | 1.83 ± 0.06 | 11.740 | 2.324 | 3.771 | 4.847 | 0.844 | 9.568 | 63.141 | 0.280 ± 0.03 |
| Sourdough | 5.04 ± 0.02 | 3.07 ± 0.15 | 27.130 | 22.383 | 22.712 | 63.815 | 3.605 | 158.465 | 69.322 | 0.143 ± 0.01 |

* Means of triplicate ± SD.

ND: Not detected.

Staling of balady bread samples:

Staling of balady bread, prepared using baker's yeast, soltani and sourdough starters, was followed using AWRC method after bread production and during six days of storage at room temperature as presented in Figure (2). From the obtained results, it could be observed that, there was a gradual decrease in AWRC% (low freshness) for all different balady bread samples during storage periods. The lower reduction in staling value (high freshness) was noticed in balady bread sample which prepared using sourdough starters in comparison to control sample, where it was faster decreased with increased the storage periods.

Remarkably, mold spoilage of traditional bread (prepared using baker's yeast) and bread sample which prepared using soltani starter were noticed on the third day of storage, while the bread which produced by using sourdough starter pre-fermented with mixed culture from *Lb. plantarum* and *B. longum* with *S. cerevisiae* was remained till the sixth day of storage without any indication for beginning attack of mold spoilage. This may be due to production of some antibacterial and antifungal agents by LAB during sourdough fermentation. These observations are in

agreement with those obtained by **Corsetti *et al.* (1998)**, **Lavermicocca *et al.* (2000)** and **Gänzle and Vuyst (2004)**.

From the abovementioned results, it could be observed that, the incorporation of LAB with yeast during preparation of sourdough starters led to decrease staling rate of balady bread as measured by the AWRC ratio. This may be due to the ability of sourdough LAB to produce some metabolites which have a positive effect on the staling of bread such as organic acids, exopolysaccharides (EPS) and enzymes (e.g. α -amylase and protease). These results are in agreement with those obtained by **Corsetti *et al.* (1998 and 2000)**, **Korakli *et al.* (2003)** and **Sadeghi (2008)** mentioned that, during sourdough fermentation, lactic acid bacteria produce a number of metabolites which have been shown to have a positive effect on the texture and staling of bread (e.g. organic acids, exopolysaccharides (EPS) and enzymes).

Sensory evaluation of fresh balady bread:

The effect of using 20% of soltani and sourdough starters on sensory properties of balady bread samples are presented in Table (4). From the obtained data, it could be observed that, there was no significant

differences ($p>0.05$) in both of general appearance, crust color and roundness between the control sample (prepared using baker's yeast) and other bread samples which prepared using soltani starter or sourdough starter. Concerning to the taste and odor, no significant difference was recorded between bread sample which prepared using soltani or sourdough starters, but there were significant differences between control sample (prepared using baker's yeast) and the other which produced by using soltani or sourdough starters.

For bread crumb distribution and separation of layer, the obtained results indicated that there were significant differences ($p<0.05$) between control sample and bread samples which produced by using soltani or sourdough starters. These results are in agreement with those obtained by **Marklinder et al. (1996)** and **Rehman et al. (2006)**.

The over all acceptability values were a reflection of all the tested quality attributes and acceptability of the studied balady bread. These values were calculated from 100 as a sum of received sensory score. The results demonstrated that, the mean over all

acceptability values of control bread sample which produced by using baker's yeast was lower than those of other sample which produced by using soltani or sourdough starters.

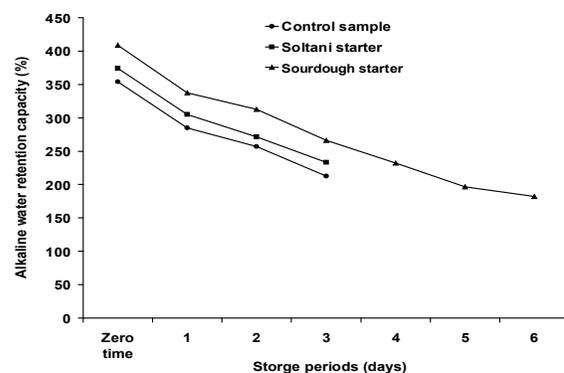


Figure (2): Alkaline water retention capacity (AWRC %) of balady bread samples during storage at room temperature.

Table (4): Sensory evaluation of fresh balady bread samples prepared by using baker's yeast, soltani and sourdough starters.

| Treatments | General appearance (15) | Crust color (15) | Taste (15) | Odor (15) | Roundness (15) | Crumb distribution (15) | Separation of layer (10) | Over all acceptability (100) |
|-----------------|-------------------------|-------------------|-------------------|-------------------|-------------------|-------------------------|--------------------------|------------------------------|
| Baker's yeast | 13.5 ^a | 14.0 ^a | 12.3 ^b | 10.9 ^c | 14.0 ^a | 12.4 ^b | 9.5 ^b | 86.6 ^b |
| Soltani starter | 14.0 ^a | 14.5 ^a | 14.5 ^a | 13.8 ^b | 14.2 ^a | 14.5 ^a | 9.8 ^{ab} | 95.3 ^a |
| Sourdough | 14.0 ^a | 14.5 ^a | 15.0 ^a | 15.0 ^a | 14.5 ^a | 14.7 ^a | 10.0 ^a | 97.7 ^a |

* Means followed by different letters in the same column are significantly different by Duncan's multiple test ($p<0.05$).

In general, the obtained results indicated that there is a positive effect on the sensory evaluation of sourdough bread due to combination of yeast (*S. cerevisiae*) and studied LAB strains (*Lb. plantarum* and *B. longum*). These results are in agreement with those obtained by **Martinez-Anaya et al. (1990)**, **Gobbetti (1998)**, **Messens and De Vuyst (2002)** and **Hansen and Schieberle (2005)**.

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