

## **SHELF LIFE EXTENSION OF PITA BREAD BY MODIFIED ATMOSPHERE PACKAGING**

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### **ABSTRACT**

Shelf life of pita bread is limited by microbial growth and staling. In a storage trial pita bread were packed in atmosphere of carbon dioxide and nitrogen at three levels (15% : 85%; 40% : 60% and 85% : 15%). Some packs included an oxygen absorbent sachet. The results showed that all samples slightly changed in their water activity and pH at the end of 28 days storage period. Control samples in 15% carbon dioxide with no sachets showed visible mold growth after 2 days. Microbial spoilage was prevented until the end of storage period in packs flushed with 100% carbon dioxide and at 40% and 15% when sachets was included.

The results also revealed that pita breads packaged in air with oxygen absorbent showed lower rate of staling than pita breads packaged in CO<sub>2</sub>:N<sub>2</sub> alone.

### **INTRODUCTION**

Pita bread is a staple in the middle east and now is popular in Australia. Shelf life of pita bread is limited by microbial spoilage and staling (Quail et al.,1991). When Fresh breads emerges from oven it is free from viable microorganisms. During cooling and packaging, contamination can result from either air borne spores or contact with contaminated surface. Some bacterial spores can survive the baking process or contamination can occur before packaging is complete.

Avital and Mannheim (1988) and Quail et al., (1990) found that the water activity of pita bread can range from 0.90 to 0.96. At these levels, growth of yeasts and molds can occur. Under warm conditions, packaged bread can develop visible mold within 48 hours of baking (Almohiza et al., 1987).

Bread staling is a process of chemical and physical change which results in a loss of consumer acceptance. The major changes after baking immediately usually involves moisture loss or gain, starch retrogradation, increases firmness, increased fragility, and loss of aroma and flavour (Kulp, 1979; Hebeda et al. 1991; Fredriksson et al., 1998; Brody, 2001 and Gray and Bemiller, 2003). This problem can be prevented by the use of moisture impermeable wrapping materials such as low density polyethylene (L.D.P.E) which is a high barrier to moisture (Smith and Simpson, 1996).

There are three main factors which influence the rate of staling. These are product formulation, particularly fat or shortening content, the baking process, and storage conditions (Smith and Simpson 1996).

Various methods have been used to control the rate of staling of bakery products. Surfactants have been used to slow staling by complexing with the starch molecules and reducing the tendency for retrogradation (Hebeda et al. 1991; Gray and Bemiller, 2003).

The use of enzymes for retarding the staling of baked products has been researched by (Hebeda et al. 1991). This involves the use of  $\alpha$ -amylase from various sources to hydrolyze the starch molecules into smaller fragments, producing a softer product.

Modified Atmosphere Packaging (MAP) defined as the enclosure of food products in a high gas barrier film in which the gaseous environment has been changed or modified to slow microbial growth and retard enzymatic spoilage with the intent of extending shelf life (Young et al., 1988). Several methods can be used to modify the gas atmosphere within the packaged product including vacuum packaging, gas packaging, the use of  $O_2$  absorbents,  $CO_2$  generators and ethanol vapor generators (Day, 2000 and Shahat *et al.*; 2003).

MAP to extend the shelf life of food has been subject for many investigations in recent years. This has resulted not only from development in the packaging technology but also from the industry's need for a less energy intensive and more economical method of short term food preservation (Smith and Simpson, 1996). The effect of modified atmosphere packaging on the shelf-life of pita bread has been investigated by Black et al. (1993) and on the shelf-life of crusty rolls by Smith et al. (1986). A detailed report on modified atmosphere packaging has been given by Smith (1994).

The shelf-life of crusty rolls packaged in air was found to be 5-6 days, however this was extended to 16-18 days by packaging in an atmosphere of  $CO_2$  and  $N_2$  (60:40). Crusty rolls packaged with oxygen absorbent remained mold-free for >60 days (Smith et al.1986). This paper reviews the effect of such technology on the physical and microbiological spoilage which may present in pita bread and also on the organoleptic properties of the product.

Gas packaging has become available method for the extension of the mold-free shelf-life of bakery products. It involves packaging the product under an atmosphere of various gases, such as  $CO_2$ ,  $N_2$ ,  $CO$ ,  $SO_2$  etc., the most common being  $CO_2$  alone or in conjunction with other gases (Young et al., 1988). The aim of the present study was to observe the effect of modified atmospheres involving oxygen absorbent and gas packaging on the shelf life of pita bread stored at ambient temperature.

## **MATERIALS AND METHODS**

### **Sampling**

The experimented pita bread samples was obtained from bakery of El-Beida, Libya. The Pita bread samples were taken from the cooling racks of commercial pita bakery. The product was made according to a commercial recipe consisted of loaves approximately 160 mm in diameter and 20 mm thickness. Sterile tongs were used to transfer the loaves into sterile plastic bags.

### **Packaging process and shelf life conditions**

Once the samples were baked and aerated, three loaves of pita bread were bagged having an average volume of  $1600\text{ cm}^3$ . The permeability

to oxygen of the formed bags was about 4 ml/m<sup>2</sup>/24h at 25 °C and 0 % RH. They were stored under the following condition:

- a) Control packaged pita breads (air atmosphere headspace).
- b) Enclosing oxygen scavenger sachets (Ageless type FX, produced by Mitsubishi & Toppan Gas Chemical Co., Japan) with air atmospheric headspace.
- c) Inserting mixed gas of CO<sub>2</sub>:N<sub>2</sub> with ratio of 15: 85, 40 : 60 and 100: 0 to pack headspace instead of air.
- d) Inserting the same gas mixture with enclosing one sachets of Ageless FX

The packaged pita bread samples were double sealed using heat- seal packaging machine (Model A300). The packaged samples were stored at room temperature (23 °C) and examined on initial and end of shelf life (1 and 28 days ; respectively). One bread was used for microbial analysis while the other was used for pH, water activity and sensory analysis. Headspace gas analysis was performed on each bag prior to opening.

#### **Water activity**

Water activity was measured using a Decagon CX-1 Water Activity meter, previously calibrated with a saturated sodium chloride solution (Prior 1978).

#### **pH**

pH measurements were done by making 1:2 slurry of 10 g of bread with 20 g distilled water and immersing the electrode of pH meter (model 220, Corning, NY) directly in the mixture.

#### **Headspace analysis**

Product was analyzed for headspace gas composition at each sampling day. Samples of headspace gas were withdrawn using a 10 ml gas-tight Pressure-Lock syringe ( Precision Sampling Corp., Baton Rouge, LA) through silicone seals attached to the outside of each package, and injected into a Varian Gas Chromatograph (Model 3400), fitted with a Thermal conductivity Detector(TCD) and using Porapak Q (80-100 mesh) and Molecular Sieve 5A (80-100 mesh) column in series. Helium was used as carrier gas at a flow rate of mL / min. The column oven was held at 80 °C while the injector and the detector were maintained at 100 °C and 150 °C respectively. The gases (CO<sub>2</sub>:N<sub>2</sub>, and O<sub>2</sub>) were identified by using a Hewlett-Packard integrator (model 3390A).

#### **Microbial analysis**

Yeast and mold counts were performed on homogenates obtained by dicing the whole loaf and dispensing in 0.1 % sterile peptone at dilution of 10% using a stomacher; duplicate plates containing oxytetracyclin-glucose yeast extract agar were prepared, 1ml of the above dilution and further ten-fold dilutions to a total of 1:1000 were applied. Plates were incubated at 25 °C and colonies were counted and calculated to colonies/g. The method was based on AS 1766.2.2 (1980).

#### **Texture analysis**

Texture analysis was done using an Instron Universal Testing Machine. In this study a compressibility test was done by using a flat head. This was in accordance with the method of Gray and Bemiller (2003)

## RESULTS AND DISCUSSION

### Water activity

Changes in water activity ( $a_w$ ) of product throughout storage are shown in Table 1.  $A_w$  values were fairly constant and  $A_w$  changed very slightly from an initial  $a_w$  of 0.90 to 0.88 at the end of the 28 day storage period. The change in  $a_w$  can be attributed to moisture migration from the product into the package headspace until the product reaches its equilibrium relative humidity (ERH) of ~ 88.6%, i.e.,  $a_w = 0.886$ .

**Table 1: Changes in water activity ( $a_w$ ) of pita breads stored under various packaging condition.**

Treatment	Initial end of shelf life	
Air packaging (Control)	0.90	**
Low CO <sub>2</sub> ( No insert)	0.91	**
Low CO <sub>2</sub> (O <sub>2</sub> scavenger)	0.91	0.89
40% CO <sub>2</sub> (No Insert)	0.91	0.89
40% CO <sub>2</sub> (O <sub>2</sub> Scavenger)	0.91	0.88
100 % CO <sub>2</sub> (No Insert)	0.91	0.88
100% CO <sub>2</sub> (O <sub>2</sub> Scavenger)	0.91	0.88

Means of the measurements SD± 0.002

\*\* mold growth after 3 days

### pH

Changes in product pH (results not shown) were also minimal and changed from 5.85 at day 1 to 5.65 at day 28. This slight decrease in pH is due to either (i) dissolution of headspace CO<sub>2</sub> into the aqueous phase of the product and / or (ii) growth of lactic acid bacteria in the stored product (Gill, 1988).

### Headspace analysis

Changes in gas composition for bread packaged at the beginning and end of shelf life of various packs are shown in Table 2. For air packaged samples (control), headspace O<sub>2</sub> decreased from 20% to 15% with a corresponding increase in headspace CO<sub>2</sub> to 5% at the end of shelf life ( visible growth of mold), while headspace N<sub>2</sub> remained fairly constant ( i.e., 80%) throughout storage period. The changes in O<sub>2</sub> and CO<sub>2</sub> levels are due to mold growth which was visible on air packaged samples after 3 days storage at room temperature.

For low CO<sub>2</sub> packaged pita bread with no scavenger (Table. 2), O<sub>2</sub> in headspace O<sub>2</sub> decreased from 12.6% to 0.02 with a corresponding increase in headspace CO<sub>2</sub> from 17.9 to 32.1 after 28 days. Headspace N<sub>2</sub> remained slightly constant at 69.4% and 67.9%, throughout storage. The changes in O<sub>2</sub>-CO<sub>2</sub> levels are due to mold growth, which was visible on all products after 2 days storage ambient temperature (~23°C). For low CO<sub>2</sub> packaged pita breads with scavenger (Table. 2), CO<sub>2</sub> decreased from 15.3 % to 0.9 % after 28 days and O<sub>2</sub> decreased from 7.4 to 0.1 at the end of shelf life. This is due to its absorption or loss through packaging film.

For samples packaged in 40% CO<sub>2</sub> with no scavenger (Table 2), headspace CO<sub>2</sub> and N<sub>2</sub> remained fairly constant at 39% and 60%, respectively, throughout storage. Also O<sub>2</sub> decreased slightly from 0.12% to 0.02%. For samples packaged in 40% CO<sub>2</sub> with scavenger headspace CO<sub>2</sub> and O<sub>2</sub> decreased from 40% to 35.4% and from 0.4% to 0.1% respectively with increase in headspace N<sub>2</sub> from 59.4% to 64.5%.

These changes in headspace can be attributed to facultative aerobic microorganisms with utilize O<sub>2</sub> and produce CO<sub>2</sub> e.g., Lactic acid bacteria (Smith et al, 1983).

For samples packaged in 100% CO<sub>2</sub> with no scavenger headspace CO<sub>2</sub> decreased to 85% and N<sub>2</sub> increased to 16%. At the end of storage headspace O<sub>2</sub> decreased to 0.02%. The results also showed that headspace CO<sub>2</sub>, N<sub>2</sub>, and O<sub>2</sub> remained fairly constant at 98.5%, 1.2% and 0.02% for all samples packaged in 100% CO<sub>2</sub> with scavenger.

**Table 2:Head space composition of packaged pita bread at the beginning and the end of storage period.**

Treatment	Initial (Day 1)			End of shelf life ( Day 28)		
	CO <sub>2</sub> %	N <sub>2</sub> (%)	O <sub>2</sub> (%)	CO <sub>2</sub> %	N <sub>2</sub> (%)	O <sub>2</sub> (%)
Air ( Control)	0	80	20	5	80	15
Low CO <sub>2</sub> (No Insert)	17.9	69.4	12.6	32.1	67.9	0.02
Low CO <sub>2</sub> (Scavenger)	15.3	77.3	7.4	0.90	99.0	0.01
40% CO <sub>2</sub> (No Insert)	39.0	60.3	0.12	38.8	60.5	0.02
40% CO <sub>2</sub> (Scavenger)	40.6	59.4	0.40	35.4	64.5	0.01
100% CO <sub>2</sub> (No insert)	100	0	0	85.0	16.0	0.02
100% CO <sub>2</sub> (Scavenger)	100	0	0	98.5	1.20	0.02

### **Microbiological analysis**

Change in the initial counts and counts at the initial and the end of the shelf life of pita breads packaged under various gas atmospheres for mold and yeast are shown in Table 3. This study showed that mold and yeast counts of the packaged pita breads varied considerably over their storage period. For air and Low CO<sub>2</sub> packs with no insert had an increased in mold counts and reached 2\*10<sup>3</sup> after 28 days These results confirm previous studies in our laboratory (Shahat et al.; 2003) which showed that mold can grow in low levels of O<sub>2</sub> (<2%) even in the presence of elevated levels of CO<sub>2</sub> (~60%). The 40% pack with no scavenger had an increased in mold counts, while mold counts remained constant during the storage period in the 40% pack with scavenger. The 100% CO<sub>2</sub> pack with no scavenger had an increased in mold count, reaching 2\*10<sup>3</sup>, while the 100% CO<sub>2</sub> pack with scavenger had < 10 mold at the end of shelf life. This lack of a pattern for increasing mold and yeast counts over time was found among most treatment. It is believed to be due to random contamination of individual loaves. However, it is clear that reduced visible yeast and mold growth occurred in treatments which included the oxygen scavenger insert, hence the extended shelf life of these

treatments. These results may be due to the reduced level of O<sub>2</sub> in the package headspace to less than 1% which would enhance the antimicrobial effects of CO<sub>2</sub>. It has been demonstrated that the inhibitory effect of CO<sub>2</sub> is dependent on oxygen concentration in gas packaged products (Ooraikul, 1991).

**Table 3:** Yeast and mold counts of stored pita breads

Treatment	Initial (Day 1)		End of shelf life (Day 28)	
	Mold	Yeast	Mold	yeast
Low CO <sub>2</sub> (No Insert)	9*10 <sup>2</sup>	2*10 <sup>2</sup>	2*10 <sup>3</sup>	8*10 <sup>2</sup>
Low CO <sub>2</sub> (Scavenger)	9*10 <sup>2</sup>	2*10 <sup>2</sup>	2*10 <sup>3</sup>	8*10 <sup>2</sup>
40% CO <sub>2</sub> (No Insert)	9*10 <sup>2</sup>	2*10 <sup>2</sup>	4*10 <sup>3</sup>	7*10 <sup>2</sup>
40% CO <sub>2</sub> (Scavenger)	9*10 <sup>2</sup>	2*10 <sup>2</sup>	3*10 <sup>2</sup>	4*10 <sup>2</sup>
100% CO <sub>2</sub> (No insert)	9*10 <sup>2</sup>	2*10 <sup>2</sup>	2*10 <sup>3</sup>	2*10 <sup>2</sup>
100% CO <sub>2</sub> (Scavenger)	9*10 <sup>2</sup>	2*10 <sup>2</sup>	1*10 <sup>1</sup>	2*10 <sup>2</sup>

### Texture analysis

A summary of the textural changes (compression) in all pita breads are shown in Table 4. It is evident from the table that the crumb (compression) was becoming harder due to staling throughout storage period. All pita breads packaged in low CO<sub>2</sub> alone or with oxygen scavenger had an initial compression test measurement of ~0.013 MPa at day 1. This value increased steadily throughout storage to 0.023-0.028, as a result of crumb hardening i.e., staling. It is interesting to note that breads packaged in air with an oxygen absorbent were less hard than breads packaged in a CO<sub>2</sub>-N<sub>2</sub> atmosphere alone. The results also showed that products in 100 CO<sub>2</sub> alone or with oxygen scavenger exhibited less staling. However, staling does not just involve starch retrogradation and moisture migration but also loss of flavor components.

**Table 4:** Changes in texture of pita bread stored under various atmosphere.

Treatment	Days of storage					
	1	2	7	14	21	28
Low CO <sub>2</sub> (No Insert)	0.0127	0.0127	0.0127	0.0127	0.0127	0.0127
Low CO <sub>2</sub> (Scavenger)	0.0127	0.0123	0.0182	0.017	0.0237	0.0280
40% CO <sub>2</sub> (No Insert)	0.0127	0.0193	0.0198	0.0202	0.258	0.0260
40% CO <sub>2</sub> (Scavenger)	0.0127	0.0122	0.0182	0.0230	0.0237	0.0239
100% CO <sub>2</sub> (No insert)	0.0127	0.0133	0.0145	0.0170	0.0195	0.0205
100% CO <sub>2</sub> (Scavenger)	0.0127	0.0133	0.0140	0.0160	0.0175	0.0190

### Conclusion

This study has shown that MAP involving gas packaging alone or in combination with absorbent technology is an effective method for extending the shelf life of pita bread. Where as a long shelf life is possible for pita bread packaged under modified atmosphere if microbial contamination before packaging is minimized and if oxygen concentration in the packaged product

is maintained at a low level ( at least below 1%). The mold free shelf life could be extended to more than 3 weeks on method of atmosphere modification. It is also evident from this study that the textural and sensorial shelf life of pita bread is much less than the mold free shelf life.

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**اطاله فترة حفظ خبز (pita bread) بواسطة التعبئة في جو معدل  
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يعتبر النمو الميكروبي والبيات من العوامل المحددة لفترة حفظ خبز pita bread . في هذه الدراسة تم تعبئة عينات الخبز في جو معدل من ثاني أكسيد الكربون والنيتروجين على ثلاث مستويات ويحتوي كل مستوى على عبوات بعضها يحتوي على مواد ماصة للأكسجين والبعض الآخر لا يحتوي على مواد ماصة للأكسجين.

كذلك أظهرت النتائج حدوث تغير طفيف في النشاط المائي ورقم الحموضة بعد 28 يوم من التخزين بالنسبة لجميع العينات. لوحظ نمو للفطريات بعد يومين من التخزين في العينة الكنترول التي تم تعبئتها في جو معدل من ثاني أكسيد الكربون 15% والتي لا تحتوي مواد ماصة للأكسجين . من جهة أخرى لم يحدث فساد ميكروبي اثناء فترة التخزين (28 يوم) لعينات الخبز التي تم تعبئتها في جو معدل من ثاني أكسيد الكربون 100 % وكذلك للعينات المعبئة في جو معدل 15% و 40% المضاف لها مواد ماصة للأكسجين. أظهرت الدراسة أيضا أن معدل البيات كان أقل في عينات الخبز المعبئة والمحتوية على مواد ماصة للأكسجين بالمقارنة بالعينات الأخرى. جميع عينات الخبز اكتسبت نكهة غير مقبولة نتيجة لفقدان النكهة المميزة لها.

قام بتحكيم البحث

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