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EFFECT OF STORAGE TEMPERATURE ON THE EXTENT OF BALADY BREAD STALING

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ABSTRACT

The effect of storage temperature on freshness score, moisture content, alkaline water retention capacity (AWRC), the amount and composition of soluble starch as well as parting properties of balady bread were investigated. The data revealed that the temperature of storage has more important effect on the extent of bread freshness than its moisture content. The AWRC, the amount of soluble starch extracted and amylose decline sharply during the first 24hrs of storage, after which it leveled off.

The component of the soluble starch was predominantly amylopectin. The amount of amylose in the soluble starch extracted from fresh bread after withdrawal from oven was small, declining sharply during the first 24hrs.of storage and this decreased slowly afterwordes. After24hrs.of storage, the decrease in soluble starch was accompanied by decreasing in amylopectin content. The refrigerator temperature is more likely to increase starch crystallization in bread and thus acceralate the rate of staling occurred than ambient temperature. No considerable changes in freshness as well as staling parameters were observed in bread stored at freezing temperature up to 72 hrs.

Amylogram data showed slight decrease in the pasting temperature of bread slurries with longer storage time. The difference in peak viscosity, 15 min height, including a holding treatment at 95° C for 15 min; and after subsequent cooling to 50°C (setback) gave the simillar rankings. These changes are more extensive at refrigerator temperature than at ambient temperature.

Based on the data presented in this study, the AWRC, the amount of soluble starch, amylose and viscosity highly correlated with the sensory perception evaluation of bread staleness.

INTRODUCTION

Breads like most bakery foods, have limited shelf lives. They undergo staling, a complex phenomenon defined as a decrease of consumer acceptance caused by a set of physio-chemical changes other than those resulting from the action of micro-organisms (Herz, 1965). Because of greater economic importance of bread staling, it has received much more attention. Firming of bread is generally considered to be the most important index of staling. Consequently, most researchers agree that firming during storage is caused by gradual crystallization of the starch component in bread (Zobel & Senti, 1959; Axford, 1968; Mclver, *et al*., 1968; Colwell, *et al*, 1969; Willhoft, 1973; and Kim & D'Appolonia, 1977). However, it is not unequivocally established which fraction of starch in the bread is responsible for staling. Schoch & French (1947); Ghiasi *et al* (1979) and D'Appolonia & Morad (1981), reported that the amylopectin is primarily responsible for bread staling. However, the retrogradation of amylose fraction involved also in staling (Maga, 1975).

Several different methods have been reported for characterizing the properties of gelatinized starch because of its role in determining the distribution of moisture in the baked loaf. These methods included x-ray diffraction (Pence, *et al*., 1958; and Zobel, & Senti, 1959; Drags-dorf & Varriano-Marston, 1980), swelling capacity (Leach, *et al,* 1959); content of soluble material (Leach, *et al*., 1959; Kim & D'Appolonia, 1977), and rate and extent of amylolytic digestion (Fahmy & Mansour, 1982). Numerous studies have been done on the refreshening of starch and gluten gels (Colwell, *et al*., 1969 and Willhoft, 1973). Zobel and Senti (1959) noted that the x-rays diffraction pattern of stale bread reverts to that of a freshly baked bread, when the bread; in a moist state, is heated at 95C.

Taufeili, *et al*. (1993) showed no changes in starch susceptibility to α-amylase as staling progressed of Arabic bread. Sidhu, *et al*. (1997) stated that most of the physicochemical methods used for indirect measurement of staling in western pan bread have been used for measuring the extent of staling in Arabic bread. They found that the water activity, the amount of soluble starch and amylose contents decreased significantly as the bread aged during storage.

Rasmussen & Hansen (2001) demonstrated that storage of wheat bread in modified atmosphere packing containing 100% Co_2 and / or 50% Co_2 and 50% N_2 could be used to extent the microbial shelf-life without affecting the staling rate.

Shaikh, *et al*. (2007) reported that staling of chapatti (Indian unleavened flat bread) results in less in texture and eating quality. They found that moisture content, water soluble starch and in vitro starch digestibility decreased steadily during staling at both room temperature and refrigerated temperature over storage period of one month.

Little direct information concerns the effect of storage temperature on the balady bread; 80% of the total bread consumed in Egypt; staling. The changes in bread properties associated with staling are varied and include loss of flavour, increased crumbliness, firmness, opacity and starch crystallinety, decreased susceptibility to enzymes and degree of water binding capacity and lowered amounts of total water solubility (Kulp & Ponte, 1981 and D'Appolonia & Morad, 1981). A strong negative correlation between consumer acceptability and compressibility and /or firmness of the crumb has been reported in white pan bread (Axford, *et al*., 1968). However, crumb compressibility or firmness can not be used for studying the staling rate of Arabic bread due to its different physical structure.

Egyptian Arabic bread is double layered flat bread with no clear demarcation between the crumb and crust (Williams, *et al*., 1988).

Literature on the staling of Arabic bread and on the methods for the measurements of the extent of staling are scarce. Therefore, to understand more about balady bread staling more information is needed in regard to the role of storage temperature.

This work was carried out to investigate the application of an appropriate methods for the evaluation of extent of staling in balady bread stored at elevated temperature.

MATERIALS AND METHODS

Materials:

Hard wheat flour of 82% extraction was obtained from the Northern Alexandria Mills Company, Alexandria- Egypt.

Methods:

Preparation of Balady bread: Balady bread "Magar Type" dough was prepared by employing the straight dough method, which involved mixing all the ingredients at one step. The basic formula included ten Kg wheat flour, table salt (1%, flour basis), compressed baker's yeast (1%, flour basis) and about 5.5 liters of water. After 15 minutes mechanical mixing to optimum dough consistency. The resulting dough was covered with sack cloth and allowed to ferment at ambient temperature for 90 minutes. At the end of fermentation, the dough was moulded, divided into round pieces weighting 140g each. The divided pieces were flattened into familiar cylindrical shape of about 20 cm diameter and 0.5cm thickness. The shaped loaves were set on wooden boards covered with wheat flour and left in a warm place for 30 minutes proofing, followed by baking at 350°C for about 60-75 Sec. After baking the loaves were cooled at ambient temperature for 15 minutes and then wrapped in plastic bags, each containing four loaves which were sealed. Five bags were randomly assigned to place at one of the three storage temperature; ambient temperature (22±2°C), refrigeration (4±0.8°C) and freezing (-12±0.5°C), within two hours of production for 24, 48 and 72 hours. On each day of storage, the loaves were placed on a table at ambient temperature and allowed to equilibrate for one hour before physicochemical analysis and sensory evaluation.

Analytical procedure:

Moisture content of bread was determined as desculed by AACC (2000). The alkaline water retention capacity (AWRC) of Baladi bread samples were measured by the modified method of Kitterman & Rubenthaler (1971). The total water solubles were extracted from the bread sample as described by Kim & D'Appalina (1977) by shaked 25 g sample with 100ml of distilled water for 20 minutes. The slurry was centrifuged (2000g for 5 minutes) and filtered through whatman No.1 filter paper. The extraction procedure was repeated twice more. The soluble starch was isolated from the total water solubles according to the procedure of Schoch & French (1947). To the combined supernatants, three volumes of methanol was added and heated on a steam bath for one hour. After standing over-night at refrigeration temperature (4°C), the floculated soluble starch was collected and dried in a vaccum oven at 70°C. Amylose content in the collected soluble starch was determined by the procedure reported by Juliano (1971). The amylopectin content in the soluble starch was obtained by substracting the amount of amylose content from the total amount of soluble starch. Results are expressed as dry basis.

Pasting properties of bread:

After 24, 48 and 72 hrs storage, balady bread was removed and left at ambient temperature for one hr before drying in an air drier at 50°C. The dried sample was ground to pass through 60 mesh sieve. The pasting properties of ground bread sample were investigated with a Braliender Amylograph. Bread sample (55g) was suspended in 350 ml distilled water by agitation in a waring blender at low speed for one minute. The suspension was poured into the amylograph bowl and the blender was rinsed with 100 ml of additional distilled water. The bread suspension was heated uniformly to 95 °C, and held at 95°C for 15 minutes and then cooled uniformly to 50°C. The information obtained from the anylograph curve included pasting temperature, peak viscosity, 15 min height and set back (AACC, 2000). Bread samples after two hrs baking and cooling was provided as fresh bread (control).

Sensory perception evaluation of staleness:

Sampled loaves were cut into four quarters and ten panelists (males, and females) were permitted to form their judgment of the samples individually with respect to freshness/staleness, respectively using a hedonic scale described as fallows: 1 (very stale); 2 (stale); 3 (slightly stale); 4 (slightly fresh); 5 (fresh); 6 (very fresh) (AACC ,2000).

Statistical analysis:

The results were statistically analysed by analysis of variance as described by Steel & Torrie (1980).

RESULTS AND DISCUSSION

Results in Table (1) showed that as the storage period increased, freshness value of balady bread decreased with increased rigidity. Bread stored at ambient temperature in sealed polyethylene bag for 24 hrs was softer in texture than that stored in refrigerator. However, no considerable different in the freshness between the breads existed afterwards. On the other hand, minimal changes in freshness were detected for balady bread kept in Freezing.

Table 1: Sensory Judgment of freshness of balady bread during storage at different temperature.

Storage Condition	Storage time (hrs)			
	2	24	48	72
Ambient temperature (22±2°C)	5.64 a	3.43 b	2.40 b	1.72 b
Refrigeration temperature (4±0.8°C)	5.64 a	3.10 c	2.40 b	1.28 c
Freezing temperature (-12±0.5°C)	5.64 a	5.30 a	5.12 a	4.81 a

Values followed by the same letter within the same column were not significantly different ($P<0.05$)

Changes in moisture content of breads during storage at various temperatures are given in Table (2). Balady bread stored at ambient temperature showed a definite decrease in moisture content after 24 hrs of storage, reaching the highest loss after 72 hrs (13.1%). Consequently this water loss coincided with increased loaves hardness, and decreased freshness values thus the breads were scored between "Stale" and" very stale" categories .

The inverse relationship between the hardness of bread and moisture content has been previously reported (He & Hoseney, 1990). Schiraldi & Fessas (2001) proposed that water acts as a plasticizer in the bread; the decrease in the moisture content favors the formation of hydrogen bonds among the starch polymers or between the starch and the proteins yielding greater hardness.

Bread stored at refrigerator lost practically no moisture during the 72 hrs. However, it becames less acceptable and was scored as "very stale" as shown by the sensory results.

Table 2: Changes in moisture content (%) of balady bread during storage at different temperature.

Storage Condition	Storage time (hrs)			
	2	24	48	72
Ambient temperature(22±2°C)	32.44 a	30.76 b	28.42 c	28.20 c
Refrigeration temperature (4±0.8°C)	32.44 a	32.30 a	32.12 b	32.14 b
Freezing temperature (-12±0.5°C)	32.44 a	32.68 a	33.20 a	33.12 a

Values followed by the same letter within the same column were not significantly different (P<0.05)

Even though moisture changes for bread stored in Freezing was negligible, yet the bread maintained its freshness after 72 hrs of storage. Barcenas & Rosell (2006) observed that the hardness of bread was almost constant during the period of frozen storage. This behavior indicates that at frozen temperatures, the phenomena involved in the hardening was not present or they occurred at a very slow rate. The above results indicate that staling of balady bread is brobably not due to the loss of moisture by the drying-out process. Moreover, within certain limits, the temperature of storage has more important effect on bread freshness than its moisture content. Sidhu, *et al*. (1997) reported that bread may staled without appreciable loss of moisture.

The effects of storage temperature on the amount and composition of soluble starch as well as AWRC of balady bread are shown in Table (3). The extractable soluble starch content decreased progressively during storage except freezing temperature. The changes of the amporphous pattern of starch gel which is the characteristic of the fresh bread (2 hrs) to a discrete line or partially crystalline state accompanying by gradual increase in rigidity in stale balady bread (Morad & D'Appolonia, 1980), may explain the lowering of extractable soluble starch. Boyacioglu & D'Appolonia (1994) reported that starch granules in pastes or gels are known to associate on aging resulting in crystallite formation accompanied by gradual increase in rigidity and phase separation between polymers and solvent (synersis) (Morries, 1990). These crystallites are insoluble in water or their solubility is less than native starch gel. However, the difference between the amounts of soluble starch extracted from fresh bread (2 hr) and 72 hrs after baking was highest in bread stored in refrigerator (0.90%) and lowest in bread stored at ambient temperature (0.82%). This observation suggests that the refrigeration temperature is more likely to increase starch crystallization in bread than ambient temperature.

The results are in accordance with those of Sidhu, *et al*. (1997); Kelekci, *et al*. (2003) and Shaikh, *et al*. (2006). Otherwise, no significant changes in the amounts of soluble starch was occurred when balady bread was kept at freezing temperature. Hug-Item, *et al*. (2003) and Barcenas & Rosell (2006) found that baked bread did not show a retrogradation at any time of frozen storage. The water present in the system was in solid state, thus no possibilities that the starch polymers can forming the crystalline structures associated to the retrogradation phenomena that caused bread hardness.

The changes in the amylose and amylopectin contents of soluble starch extracted after different time from balady bread are also presented in Table (2). The component of the soluble starch extracted from fresh bread (2 hrs) was predominantly amylopectin. The amount of amylase, although small in comparison with the amount of amylapectin, decreased sharply during the first 24 hrs of storage, after which it leveled off. After 24 hrs of storage, the decrease in soluble starch content was accompanied by decreasing the amylopectin contents. These results support earlier observation of Kim & D'Appolonia (1977) and Sidlu *et al* (1997), that most amylose retrogradation takes place during baking and subsequent cooling of the loaf. Although breads stored at ambient and refrigeration temperature showed the same pattern of amylose decline. The decrease was highest in bread stored at refrigerator than those stored at ambient temperature. This may be due to the higher tendency of the linear amylose chains to crystallize into insoluble aggregates at low temperature (Toufeili, *et al*., 1993). The slight decline occurred in amylose content upon storage at freezing temperature confines earlier observation on extractable soluble starch changes. Schoch & French (1947) and Ghiasis *et al*. (1979) reported that amylose had no effect on bread staling and suggested that the staling was associated with the gradual and spontaneous aggregation of the amylpectin through-out the loaves.

The results, together with the sharp decline in the amylose content during the first 24 hrs storage period, imply that amylose contributes to bread staling even though the amylose content in the soluble starch of bread was initially small. Therefore, it can be postulated that the staling which occurs through the first 24 hrs of storage is related to the retrogradation of amylose content while that occurring thereafter, can be attributed to the changes in the amylopectin fraction.

Considerable and significant variations in AWRC values were observed as balady bread aged during the storage period. The AWRC of 2 hrs bread; after baking and cooling was 388. 34% which decreased sharply during the first 24 hrs of storage, thereafter, the changes were small. The greatest decrease in AWRC value was achieved for bread stored at refrigeration temperature as compared with bread stored at ambient temperature, while those stored in freezing tended a minimized decreased. The low AWRC of aged bread is due to the reduction in the proportion of free hyohaxyl groups as a result of intermolecular hydrogen bonding between the side chains of starch polymers and the formation of highly ordered micellor regions as retrogradation proceeds (Faridi,& Rubenthaler, 1984) and Toufeili, *et al*., 1993).

Table 3: Effect of storage temperature on soluble starch, amylose, amylopectin contents and Alkaline water retention capacity (AWRC) of balady bread.

Storage Condition	Storage time (hrs)	Soluble starch (%)	Amylose (%)	Amylopectin (%)	AWRC (%)
Ambient temperature	2	2.46 a	0.42 a	2.04 a	388.34 a
	24	1.98 c	0.22 c	1.76 c	274.28 c
	48	1.71 d	0.16 d	1.55 de	256.74 d
	72	1.64 de	0.14 d	1.50 e	233.61 e
Refrigeration temperature	2	2.46 a	0.42 a	2.04 a	388.34 a
	24	1.80 cd	0.17 d	1.63 d	263.66 d
	48	1.60 de	0.14 d	1.46 e	235.02 e
	72	1.56 e	0.13 d	1.43 e	229.60 e
Freezing temperature	2	2.46 a	0.42 a	2.04 a	388.34 a
	24	2.37 ab	0.41 a	1.96 ab	371.66 a
	48	2.28 b	0.37 a	1.91 b	357.84 b
	72	2.19 b	0.30 b	1.89 b	353.27 b

Values followed by the same letter within the same column were not significantly different ($P<0.05$)

Table (4) shows the values obtained for the pasting properties of balady bread after different storage period. Slight decrease in the pasting temperature of bread slurries occurred with longer storage time. Kim & D'Appolonia (1977) suggested that the retrograted starch in staled bread swells more readily when heated in excess of water than the starch granules in fresh bread, which would cause a lower pasting temperature. A sharp decrease in the peak viscosity of bread slurries was observed after the first 24 hrs of storage and then showed lesser changes up to 72 hrs. This change is more extensive at refrigeration temperature than at ambient temperature. These may be attributed to the increased extent of starch crystallization during bread storage at low temperature (Morries, 1990 and Shaikh, *et al*., 2006).

Table 4: Effect of storage time on the pasting properties of balady bread.

Storage time (hrs)	Bread stored at ambient temperature (22±2°C)				Bread stored at refrigerator (4±0.8°C)				Bread stored at freezer (-12±0.5°C)			
	Parting temperature (°C)	Peak viscosity (BU)	15min height (BU)	Set back (BU)	Parting temperature (°C)	Peak viscosity (BU)	15min height (BU)	Set back (BU)	Parting temperature (°C)	Peak viscosity (BU)	15min height (BU)	Set back (BU)
2	75.5	415	390	710	75.5	415	390	710	75.5	415	390	710
24	75	370	345	660	74	350	330	655	76.5	420	400	720
48	74.5	340	325	640	73.5	330	315	625	75	400	385	705
72	73.5	320	320	610	73.5	315	310	605	75.5	410	385	710

At 24 hrs of storage at ambient temperature and refrigeration temperature bread slurries exhibited a decrease of 45 B4 and 60 B4 after holding at 95°C for 15 min respectively, followed by viscosity remained partically constant up to 72 hrs of storage. During the cooling cycle of the balady bread amylogram, no peak was observed, but rather a uniform increase in viscosity (set back) throughout cooling period. The set back values of bread slurries decreased with the increased storage period.

No distinct differences in pasting properties were observed between fresh bread (2 hrs) and bread stored at freezing temperature up to 72 hrs.

Based on the data presented in this study, balady bread staled more rapidly when stored at refrigerator temperature than ambient temperature. At freezing temperature bread maintained its freshness without considerable changes in the other staling parameters over the storage period.

The linear correlation coefficients between some of the important staling parameters such as AWRC, soluble starch, amylose content, amylogram viscosity values and sensory perception evaluation of staleness of balady bread are presented in table (5). Of the different parameters studies, the AWRC, soluble starch and amylose contents correlated well with the freshness score and among themselves. The good positive correlation between staling and amylose content is consistent with the role of amylose as one of the main determinants of the textural properties of bread and starch gels (Hansen, *et al*, 1991).

The AWRC produced a highly significant positive correlation with the greatest number of parameters. In addition to sensory analysis, the AWRC is therefore, recommended as an appropriated method for assessing the extent of balady bread staling.

Table 5: Linear correlation coefficients between different parameters employed for the estimation of balady bread staling

Variable		Soluble starch content	Amylose content	Peak viscosity (BU)	15 min height (BU)	Setback (BU)	Freshness score
Alkaline water retention capacity	A	0.964	0.980	0.861	0.873	0.910	0.961
	B	0.972	0.984	0.870	0.862	0.884	0.974
Soluble starch	A	-	0.923	0.911	0.901	0.873	0.883
	B	-	0.908	0.922	0.914	0.857	0.900
Amylose	A	-	-	0.874	0.797	0.782	0.937
	B	-	-	0.856	0.802	0.806	0.912
Peak viscosity	A	-	-	-	0.827	0.803	0.851
	B	-	-	-	0.812	0.822	0.833
Viscosity at 95°C after 15 min (15 min height)	A	-	-	-	-	0.766	0.821
	B	-	-	-	-	0.784	0.826
Viscosity at 50°C (Setback)	A	-	-	-	-	-	0.766
	B	-	-	-	-	-	0.751
Freshness score	A	-	-	-	-	-	-
	B	-	-	-	-	-	-

A): Bread stored at ambient temperature
B): Bread stored at refrigerator temperature

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تأثيردرجه حرارة التخزين على معدل تجلد الخبز البلدي

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قسم علوم وتكنولوجيا الاغذيه- كليه الاقتصاد المنزلى- طنطا- جامعه الازهر

أجريت هذه الدراسه بغرض التعرف على تأثير درجة حرارة التخزين على معدل تجلد الخبز البلدى.وقد أظهرت النتائج أن درجه حرارة التخزين تلعب دورٍا هاما فى تجلد الخبز أكثر من فقد الرطوبه منه أثناء التخزين.

وقد تبين أن قيم القدره على الاحتفاظ بالماء وكميه النشا الذائب ومحتوى الاميلوزيحدث لها انخفاض حاد خلال٢٤ساعه الاولى من التخزين يلى ذلك أنخفاض بسيط وكان الخبز المخزن عند درجه حرارة الثلاجه أكثر تجلدا وأقل تقبلا لدى المستهلك أثناء التخزين مقارنه بالخبز المخزن عنددرجه حرارة الغرفه. ولم يظهر أى تغيرات محسوسه فى درجه طزاجه الخبزوقياسات التجلد الاخرى عند تخزين الخبز على درجه حرارة التجميد.وتبين أن الانخفاض في كميه النشا الذائب تتناسب مع الفقد فى خواص الطزاجه وأتضح أهميه دور الاميلوز فى تجلد الخبزخلال الساعات الاولى من التخزين.وبينت نتائج خواص الجلتنه لمعلق الخبزالبلدي بواسطه جهازالاميلوجراف حدوث انخفاض بسيط فى درجه حرارة الجلتنه وانخفاض كبيرفي قيم اللزوجه وقد تباينت هذه المتغيرات تبعا لدرجه حراره تخزين الرغيف وقدوجدت علاقه ارتباط قويه موجبه بين كلا من القدره على الاحتفاظ بالماء وكميه النشا الذائب ومحتوى الاميلوز وخواص اللزوجه مع درجه الطزاجه للرغيف.