

STUDIES ON THE USE OF DIFERENT SOURCES OF ALPHA AMYLASE IN BREAD BACKING

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ABSTRACT

The effect of malt flour and α -amylase on wheat flour pan bread was studied. Wheat flour 72% extraction was mixed with 2,4 % and 6% malt flour and 40,60 and 80 ppm α -amylase to study the effect of their addition on dough quality and quality of pan bread prepared from treated dough. Farinograph parameters showed decrease in water absorption stability, development time and increase in dough weakening in treated samples compared with the control samples. Amylograph parameters as maximum viscosity, viscosity at 90°C, viscosity at 50°C and set back were decreased with increasing the different additions of malt flour and α -amylase. Sensory characteristics of pan bread and volume, specific volume, were increased in all treated samples. Also, alkaline water retention capacity was increased with increasing the storage periods. From these results it can be concluded that pan bread produced from wheat flour mixed with 4% malt flour and 80 ppm α -amylase gave the best results.

Keywords: α -amylase – maltflour- pan bread – farinograph – amylographi – bread baking.

INTRODUCTION

In Egypt, wheat is the most widely consumed crop for bread making. Wheat production is about 6.5 million tons annually (1999-2000) (Zakaria 2001). α -amylase is classified as an endoglucosidase that cleaves the α -1,4 glucosidic bond of the substrate at internal positions to yield dextrans and oligosaccharides with the CL-OH in the α configuration (Wong, 1995). The falling number of 72% extraction flour was 350 sec, liquefaction number 20 and sedimentation 59.7cm. (Bedeir 2004), the effects of α -amylase on farinograph results showed that all treatments of α -amylase significantly decreased the value of water absorption compared with the control sample, the mixing properties of dough significantly changed. All the treatments of α -amylase significantly shortened the development and stability times of dough. However, all the treatments of α -amylase significantly increased the values of the extensibility (R/E) after test time 135 min. All the treatments of α -amylase clearly weak the dough properties just after mixing but the extensograph data clearly show that α -amylase improved the dough properly during fermentation. (Maeda *et al.* 2003). Bread was characterized by compression tests, light microscopy (LM) and X-ray measurements. α -amylase reduced firming rate of bread on ageing. LM revealed that amylase and amylopectin phase separated within the starch granules and that freshly

baked control bread. Amylase containing bread exhibited strong birefringence in the amylase rich region of the granules directly after backing which did not significantly increase during ageing. The enzyme hindered the retrogradation of amylopectin as X-ray diffraction indicated that the enzyme induced low levels of starch crystallinity which did not change during ageing. It is hypothesized that the anti-staling effect of the amylase is based on the capacity to partially degrade amylopectin and, by this, to hinder its recrystallization. Conversely, the enzyme slightly degrades amylose by an endo-mechanism which, in turn, promotes rapid formation of a partly crystalline amylose network in fresh bread and hinders amylose rearrangements during ageing. (Hug-Iten *et al.* 2001). Addition of alpha-amylases and malt to bread produced low molecular weight dextrans from starch by hydrolysis. Its effect on starch retrogradation and considered anti-staling agents for retardation of bread staling (Duran *et al.*, 2001).

This study was carried out to investigate the effect of adding α -amylase and malt flour on the quality of pan bread and the rheological properties of wheat flour 72% extraction. Sensory properties of fresh resultant bread, storage period for 24, 48 and 72hrs were determined.

MATERIALS AND METHOD

Materials

- Hard wheat flour (*Triticum aestivum*) 72% extraction was obtained from the South Cairo Mills Company, Cairo, Egypt.
- Malt grains (two raw barely malt) (*Hordeum acstivum*): was obtained from Pyramids Brewery Company, Cairo. The samples were milled in a willy mill to pass through 60 mesh and stored at 5°C until used.
- Alpha amylase powder (Fungal Amylase): was obtained from EL-Gomhouriah Medicinal Company, Cairo, Egypt.
- Yeast (*Saccharomyces cerevisiae*): was used in dough preparation.
- Salt was obtained from local market.

Methods

• Blends:

Blends of wheat flour samples, supplemented and fortified with malt flour and added α -amylase were used to make pan bread as indicated in the following Table(1).

Table (1): Blends wheat flour 72% ext. with malt flour and α -amylase to make pan bread.

Samples	Blends composition
Control	Wheat flour of 72% extraction
1	98% wheat flour of 72% extraction + 2% malt flour
2	96% wheat flour of 72% extraction + 4% malt flour
3	94% wheat flour of 72% extraction + 6% malt flour
4	100% wheat flour of 72% extraction + 40ppm α -amylase
5	100% wheat flour of 72% extraction + 60ppm α -amylase
6	100% wheat flour of 72% extraction + 80ppm α -amylase

- **Pan bread preparation** : The straight dough method was carried out according to the method described by A.A.C.C (2002).
- **Physical properties for pan bread:**
 - ***Volume** : The volume of pan bread was determined by rape seeds displacement according to the method of A.A.C.C.(2002).
 - * **Specific volume:** specific volume was calculated according to the method of A.A.C.C.(2002) using the following equation:

$$\text{Specific volume} = \frac{\text{Weight (gm)}}{\text{Weight (gm)}}$$

- **Sensory evaluation of pan bread:** pan bread was evaluated for sensory characteristics by ten panelists from the staff of bread and pastry, Res. Dept. Food Tech. Res. Agric. Inst., Res. C enter. Giza, Egypt. The scoring scheme was established as mentioned by A.A.C.C.(2002) which tabulated in Table (2).

Table (2): Pan bread score card.

Property	Maximum score
Appearance	20
Color of crust	15
Color of crumb	15
Crumb texture	15
Flavor	15
Taste	20
Total scores	100

The average of total score was converted to a descriptive category as follows:

Very good	(V.G.)	90-100
Good	(G.)	80-89
Satisfactory	(S)	70-79
Questionable	(Q)	Less than 70

Chemical composition of materials (Moisture, crude protein, ether extract, ash content and crude fiber) were determined according to the methods of (A.O.A.C.2002) and total carbohydrates was calculated by difference.

***Determination of falling number (FN):** Falling number was determined according to the method described in (A.A.C.C 2002). The liquefaction number was calculated as follows:

$$\text{Liquefaction number} = \frac{6000}{\text{FN-50}}$$

- **Determination of alkaline water-retention capacity (AWRC) for flour:** It was determined according to A.A.C.C.(2002).

$$\%A.W.R.C.= \frac{\text{Tube, stopper get w.t} - \text{tuber stopper w.t}}{\text{Flour weight}} \times \frac{86}{100 - \text{Flour moisture}} - i \times 100$$

- **Farinograph test:** The parameters were taken from the farinograms as described in the A.A.C.C.(2002) .
- **Amylograph test:** A barabender visco-amylograph (Barabender type 2648813 OGH West Germany) according to the method described in the A.A.C.C.(2002) .
- **Alkaline water retention capacity:** The staling rate of bread was determined by measuring the Alkaline Water Retention Capacity (AWRC) according to the method described by A.A.C.C.(2002).
- **Struct-o-graph method (mechanical test):** Compressibility (softness): Represented the length of the curve in (mm) from the start until the top of the curve (Baker *et al.*,1988; Bedeir, 2004).
- **Percentage change in compressibility:** calculaters from following equation

$$\% \text{ change in compressibility} = \frac{C1 - C2}{C}$$

Where C1= Compressibility at zero time in (mm).
 C2= Compressibility after period of storage in (mm).

- **Statistical analysis:** was done according to Snedecor and Cochran (1980).

RESULT AND DISCUSSION

The results presented in Table (3) showed that the wheat flour 72% contained high value of protein, and low values in ether extract, ash and Crude fiber. Total carbohydrate was high in malt flour 87.11% and was low in 72% flour extraction 83.25%. The data are in the same line with that obtained by Mohamed (2000).

Table (3): Chemical composition of extraction flour 72% ext.and whole malt flour (on dry basis).

Sample	Protein	Ether extract	Ash	Crude fiber	Total carbohydrates
Wheat flour 72%	13.7	1.6	0.56	1.8	83.25
Malt flour	8.59	0.56	1.82	1.92	87.11

The obtained data in Table (4) showed that when the amount of α-amylase and malt flour increased, the values of sedimentation test, sedimentation value and alkaline water retention capacity of all tested samples decreased, this might indicate that the amount of granules starch binding with gluten to form network. The alkaline water retention capacity used for determining baking properties of wheat flour which is related to protein quality and quantity which reported by Miller (1988) and Sollars (1972).

Table (4) : Effect of malt flour on sedimentation test, sedimentation value and alkaline water retention capacity in wheat flour 72% extraction.

Sample	Sedimentation test%	Sedimentation value %	Alkaline water retention capacity (%)
Control (72% extraction flour)	59.70	5.30	87.30
72% extraction flour + 2% malt flour	59.30	5.28	87.10
72% extraction flour + 4% malt flour	56.70	5.10	86.20
72% extraction flour + 6% malt flour	57.20	4.80	84.80
72% extraction flour +40 ppm α - amylase	58.60	5.10	86.00
72% extraction flour +60 ppm α - amylase	57.00	4.70	84.50
72% extraction flour +80 ppm α - amylase	56.20	4.50	83.60

The present data reported in Table (5) showed the effect of adding α -amylase and malt flour on the Farinograph parameters of 72% extraction flour dough. It could be noticed that the water absorption of dough was clearly decreased with increasing the added amount of malt flour and α - amylase . The low water absorption of flour may be due to α - amylase and malt flour hydrolysis the starch which able absorp the water bond interaction with proteins to be network. This result are in agreement with those obtained by Mathewson (1998) . From these results, it could be observed that arrival time was increased in 72% flour extraction from 1.0 to 1.5 min for control and malt flour respectively. Also, addition α - amylase increased arrival time. While, development time decreased after adding 2.4 and 6% malt flour and 40,60,80 ppm α - amylase compared to control. These results in the same line with El-Farra *et al.*(1983).Weakening of dough was increased with increasing the added levels of malt flour and α - amylase. These results are close with the results reported by Maeda *et al.*(2003) concerning dough stability it was decreased from 10 to 2.5 min with 6% malt flour and 2.5min. with 80ppm α -amylase.

Table (5): Effect of adding malt flour and α - amylase to 72% flour extraction on Farinograph parameters.

Samples	Farinograph properties				
	Water absorpti on %	Arrival time (min)	Development time (min)	Stability (min)	Weaking of dough (B.U.)
Control 100% flour of 72% extraction	61.0	1.0	7.0	10.0	70.0
88% flour of 72% extraction + 2% malt flour	59.5	1.5	4.0	7.0	100.0
96% flour of 72% extraction + 4% malt flour	57.0	1.5	4.0	6.0	120.0
94% flour of 72% extraction + 6% malt flour	52.5	1.5	2.0	2.5	130.0
100% flour of 72% extraction + 40 ppm α - amylase	56.0	1.0	4.0	9.0	80.0
100% flour of 72% extraction + 60 ppm α - amylase	54.0	1.25	3.5	8.0	80.0
100% flour of 72% extraction + 80 ppm α - amylase	53.5	1.50	1.5	2.5	85.0

Effect of adding malt flour and α - amylase to 72% extraction on amylograph parameters:

The presented data in Table (6) explained that the obtained maximum viscosity (B.U.) for tested sample 72% extraction with malt flour and α -amylase with different levels were decreased with any increasement of adding malt flour and α - amylase to wheat flour samples. These may be due to form sugar bridges between starch chains which chang the shape and conformation of heliX of starch molecules. These results are in a parallel with those obtained by Tolstoguzov (1997).

Table (6): Effect of added α - amylase and malt flour on the set back of dough for 72% wheat flour extraction.

Samples	Maximum viscosity (B.U.)	Viscosity at 50°C(B.U.)	Set back (B.U.)
Control 100% flour of 72% extraction	485	440	45
88% flour of 72% extraction + 2% malt flour	50	15	35
96% flour of 72% extraction + 4% malt flour	25	---	25
94% flour of 72% extraction + 6% malt flour	25	---	25
100% flour of 72% extraction + 40 ppm α - amylase	470	440	30
100% flour of 72% extraction + 60 ppm α - amylase	455	440	15
100% flour of 72% extraction + 80 ppm α - amylase	430	440	10

- Effect of adding malt flour and α - amylase on the physical properties of pan bread:

Weight, volume and specific volume of pan bread prepared from wheat flour 72% extraction and different levels of malt flour and α - amylase were studies and the obtained results are illustrated in Fig.(1). From these results, it could be observed that volume and specific volume increased gradually with increasing the levels of malt flour and α - amylase. While weight decreased compare control with addition α - amylase. These result are in agreement with Bajwa (1990). Also, Maeda *et al.*(2003) mentioned, that addition mutant α - amylase to the ingredients increased the specific volume of pan bread.

- Effect of adding malt flour and α - amylase on the sensory evaluation of pan bread:

The data presented in table (7) showed the effect of adding malt flour and α - amylase with different levels to 72% extraction wheat flour on sensory characteristics of pan bread from the presented resulted could be noticed that adding 2,4 and 6% of malt flour show a significant difference between the control sample and treatments in texture of crumb and flavor, concerning crumb color crust general appearance and taste there were no significant difference between control sample and treatments. It could be concluded that the adding of 40, 60 and 80ppm of α - amylase could be recommended to produce pan bread.

Fig (1): Effect of adding malt flour and α - amylase on the pan bread of weight, volume and specific volume .

Table (7): Effect of adding malt flour and α - amylase on the sensory characteristics of pan bread.

Treatments	General appearance (15)	Crust color (15)	Crumb color (15)	Texture of crumb (15)	Taste (20)	Flavour (20)	Overall acceptability	Degree
Control	13.0 ^a	13.3 ^a	13.6 ^a	13.7 ^a	18.2 ^a	18.7 ^{ab}	90.5 ^a	(V)
2% malt flour	13.1 ^a	13.3 ^a	13.8 ^a	12.3 ^b	18.2 ^a	18.7 ^{ab}	89.4 ^{ab}	(G)
4% malt flour	13.2 ^a	13.4 ^a	13.4 ^a	12.6 ^b	17.8 ^a	17.9 ^{ab}	88.3 ^{ab}	(G)
6% malt flour	12.8 ^a	13.3 ^a	13.0 ^a	12.9 ^b	17.4 ^a	17.9 ^{ab}	87.3 ^{ab}	(G)
40ppm α amylase	13.6 ^a	13.5 ^a	13.2 ^a	13.5 ^a	18.7 ^a	18.8 ^a	91.4 ^S	(V)
60ppm α -amylase	13.9 ^a	13.5 ^a	13.9 ^a	14.2 ^a	18.5 ^a	18.8 ^a	92.8 ^a	(V)
80ppm α -amylase	13.2 ^a	13.4 ^a	13.4 ^a	14.3 ^a	18.4 ^a	18.6 ^a	91.3 ^a	(V)
L.S.D at 0.05	-	-	-	0.7414	-	07331	2.31	

Significant at 0.05 (properties with the same letter are not different significantly), 90-100 V. good (V), 80-89 Good (G), 70-79 Satisfactory (S), Less than 70 Questionable (Q).

Effect of adding malt flour and α - amylase on the alkaline water retention capacity of pan bread:

Alkaline water retention capacity (A.W.R.C) of different levels of malt flour and α - amylase used to prepare pan bread and A.W.R.C% was determined at different storage times zero, 12, 24, 48 and 72 hrs for pan bread stored in polyethylene bags at 24°C. It could be noticed that the (AWRC) was decreased with increasing the levels of malt flour. This may be due to crystallization of amylase after baking process during bread storage. These results agree with those of Stauffer (2000).

Table (8): Effect of adding malt flour and α - amylase on the alkaline water retention capacity of pan bread:

Additives	Alkaline water retention capacity (AWRC %)								
	Fresh%	After 12hrs %	*R.D%	After 24 hrs %	*R.D%	After 36hrs %	*R.D%	After 72hrs %	*R.D%
Control	400.10	390.00	2.52	376.00	6.02	365.33	8.69	315.60	21.12
2% malt flour	405.81	395.67	1.75	382.21	5.81	375.46	7.48	327.75	19.21
4% malt flour	407.20	400.00	1.76	385.19	5.41	320.06	6.67	332.30	18.39
6% malt flour	400.10	393.96	1.53	389.10	2.75	385.17	3.73	339.45	15.16
40ppm α -amylase	321.59	378.22	1.05	318.00	1.01	325.22	-3.10	326.31	-1.47
60ppm α -amylase	321.10	311.11	0.62	320.20	0.28	35.10	-1.25	329.60	-1.71
80ppm α -amylase	330.40	329.10	0.39	330.10	0.09	335.01	-1.39	336.40	-1.82

* R.D.= Rate of decrease.

- Compressibility softness of pan bread:

Compressibility as a measurement of mechanical test, i.e, texture softness of pan bread were evaluated using structograph after baking (zero time) and periodically during storage at room temperature. From the Tables (9,10), It could be noticed that compressibility of all samples with malt flour were decreased with increasing storage periods, it was clearly noticed that the optimum added malt flour ratio (4%) had the highest compressibility (120mm), and the lowest lost in its relative compressibility values with advancing storage period at 24, 48, and 72 hrs., 25.3 , 8.8 and 5.9% respectively, while the lowest compressibility (control) were 4.54, 2.72 and 2.50%, at 24,48 and 72 hrs respectively. These results are confirmed with Baker *et al.* (1988) who measured bread crumb firmness using universal testing machine.

Finally the foregoing results lead evidently to conclude that, the crumb of bread aged, and the amount of force require to compress the crumb increased, and the crumb firmness also increased the degree of compression. These results are in agreement with Bedery (2004).

Table (9): Compressibility of pan bread baked with different levels of malt flour and α -amylase during storage at room temperature ($25^{\circ}\text{C} \pm 5^{\circ}\text{C}$).

Additive	Compressibility (mm)		Storage period (hrs)	
	(Fresh) zero	24	48	72
Control	116	115	113	79
2% malt flour	118	108	95	88
4% malt flour	120	115	100	97
6% malt flour	124	124	102	95
Control	100	98	95	87
40 ppm α -amylase	105	103	98	90
60 ppm α -amylase	109	106	110	99
80 ppm α -amylase	108	110	97	94

Table(10):Compressibility of pan bread baked with different levels of malt flour and α -amylase during storage at room temperature ($25^{\circ}\text{C} \pm 5^{\circ}\text{C}$).

Additive	Storage period (hrs) Compressibility %			
	(Fresh) zero	24	48	72
Control	0.0	4.54	2.72	2.72
2% malt flour	0.0	23.80	6.80	5.70
4% malt flour	0.0	25.30	8.80	5.90
6% malt flour	0.0	---	---	---
Control	0.0	4.99	4.72	4.22
40 ppm α -amylase	0.0	6.79	6.99	7.85
60 ppm α -amylase	0.0	12.71	20.52	21.19
80 ppm α -amylase	0.0	1.81	2.72	3.64

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دراسات على استخدام مصادر مختلفة للآلفا أميليز في إنتاج الخبز
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أجريت هذه الدراسة بهدف تحسين خبز القوالب باستخدام مستويات مختلفة من دقيق المولت وأنزيم الألفا أميليز المضاف لإلى دقيق القمح أستخلاص ٧٢% وكانت النسب المستخدمة هي ٢،٤، ٦% دقيق مولت و ٤٠، ٦٠، ٨٠ جزء فى المليون من أنزيم الألفا أميليز وتم تقييم خبز القوالب كيميائيا وطبيعيا وحسيا وكذلك مدى أحتفاظ الخبز بالطزاجة. وأوضحت النتائج ان رقم السقوط كان مرتفعا أما قوة التشرىب للدقيق كانت تتناقص بزيادة نسبة الأضافة من دقيق المولت وانزيم الألفا أميليز. الأختبارات الريولوجية للخلطات مثل اختبار الفارينوجراف أظهرت القراءات انخفاضاً فى نسبة الأمتصاص ومدة العجن وثبات العجينة بينما أظهرت ارتفاع فى قيمة ضعف العجين أما الفسكواميلوجراف فكانت قراءة اللزوجة القصوى عند ٩٠م^٢ واللزوجة عند ٥٠ وال Set back منخفضة بزيادة نسبة الأضافة من دقيق المولت والألفا أميليز وسجلت التقديرات الحسية والحجم النوعى وقوة التشرىب للماء فى اختبار البيات مع زيادة نسب الأضافات المختلفة من المولت والألفا أميليز وكذلك تأخر ظاهرة التجلد فى خبز القوالب بزيادة نسبة الأضافات بالمقارنة بالكنترول وأوقضحت هذه النتائج أن إضافة ٤% من دقيق المولت وكذلك ٨٠ جزء فى المليون من الألفا أميليز إلى إعطاء أفضل النتائج.

