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PHYSICO-CHEMICAL AND RHEOLOGICAL CHARACTERISTICS OF TOAST BREAD FORTIFIED WITH ALPHA-AMYLASE ENZYME AND CARBOXYMETHYLCELLULOSE

Mariam M.A. Abd El-Naby*, S.E. Elnemr, S.M. Aboelmaaty and Sabah M. Moner

Food Sci. Dept., Fac. Agric., Zagazig Univ., Egypt

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ABSTRACT: This study examined the effect of α -amylase and Carboxymethylcellulose on physicochemical and rheological characteristics of toast bread. Toast bread was fortified with α -amylase in ratios of 0, 0.25, 0.30, and 0.35 ppm and Carboxymethylcellulose in ratios of 0, 0.1, 0.3, and 0.5%. Physicochemical and rheological properties were evaluated. The results showed that protein value of toast bread fortified with Alpha amylase (AM) at higher ratio was higher than that of the other toast bread samples and the control. There was an increase in product moisture content by the addition of CMC. The highest ash value content of the prepared toast bread was obtained upon the addition of 0.50 % CMC. There are non-significant ($p < 0.05$) changes in the mean values of fat content among the prepared sample and the control. Addition of CMC and AM produced the greatest increase in loaf volume and specific volume. In the case of α -amylase, all three levels significantly increased volume compared to CMC. Supplementation with the high levels of α -amylase decreased crumb hardness and chewiness. While with the high level increased hardness. As the dosage level of AM increased in the bread dough formulation, reducing sugar formation was accelerated and the released sugars were utilized for the Maillard reaction. The resulting outcome displayed low L^* values and led to a much darker crust formation. While as the dosage level of CMC resulting outcome displayed high L^* and b values. Toast bread containing (AM) and CMC had higher significant ($p < 0.05$) scores for all the sensory characteristics as compared to the control. Moreover, toast bread fortified with (AM) had higher significant ($p < 0.05$) score for sensory characteristics compared with CMC treatments at the same concentrate.

Key words: Alpha amylase, Carboxymethylcellulose, toast bread, physicochemical, rheological properties.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is one of the most important crop and has been used worldwide as a main ingredient in bread making. The increasing mechanization of the baking industry and the demand for a wide range of bread types has determined the necessity to modulate structure and viscoelastic properties of dough (Dunnewing *et al.*, 2002; Shebl *et al.*, 2018). In modern baking industry with high demands for bread with superior nutritional and sensorial quality, using additives like enzymes and hydrocolloids are necessary to

modulate the rheological properties of dough (Paucean *et al.*, 2016).

Many attempts have been made for improving the bakery products properties quality. The enzymes addition to wheat flour is interesting replacement to generate changes in structure of the dough and in consequence, for improving functional properties of flour (Shebl *et al.*, 2018).

Alpha-amylase used in conjunction with Beta-amylase to increase the level of fermentable sugars to ensure adequate gas production during fermentation and to modify dough rheological

* Corresponding author: Tel.: +201123458937

E-mail address: mariam.mohamed00100@gmail.com

properties (Palacios, 1998). The addition of fungal Alpha-amylase to flour decreased arrival, stability, times, development, valorimeter value and water absorption (Maeda *et al.*, 2003; Kim *et al.*, 2006; Sundarram *et al.*, 2014).

Improvement of gluten properties by enzymes and hydrocolloids are widely used to improve bread quality in wheat bread (Eduard, 2014).

Hydrocolloids used in small quantities (< 1% on flour base) and are expected to increase water retention and loaf volume and decrease firmness and starch retrogradation (Collar *et al.*, 1999).

Carboxymethyl cellulose (CMC) is a sodium salt derivative of cellulose. Unlike cellulose, it is water soluble and can function as a suspending agent, stabilizer, film former or thickening agent. CMC finds use in gluten-free baking by providing dough with viscosity and bread with volume much like gluten proteins do. It also functions well in fillings as a thickener and in glazes as an agent to slow down sugar crystallization (Hoefler, 2019).

Carboxymethyl Cellulose CMC had a combined effect with enzymes and emulsifiers on textural properties of both dough and fresh bread, for example, high volume and retarding of staling (Collar *et al.*, 1999; Rosell *et al.*, 2001; Guarda *et al.*, 2004).

This study throws some light on the determine the optimum α -amylase and Carboxymethyl Cellulose concentrations can be used to prepare toast bred using weak flour and studying the effect of treatment on the physicochemical, rheological, textural and sensory characteristics of toast bread.

MATERIALS AND METHODS

Materials

Wheat flour extraction of 72% (semi hard) was obtained from El-Tayseer from Mills Company, kalubia, Egypt. Carboxymethyle Celledouse (CMC) was obtained from International Company for Backing Materials, Zagazig, Egypt. Alpha amylase (AM) enzyme was obtained from Copa comp Additive Food. Corn oil was obtained from Arma for Oils and

Soaps, 10th Ramadan city, Egypt. Dry yeast of *Saccharomyces cerevisiae* sucrose was Angel Yeast (Egypt) Co., Ltd, Beni Suef, Egypt.

Methods

Cakes Preparation

The method of made dough for toast breads production was carried out according to the method described by AACC (2002) as follows: Components consisted of wheat flour (1000 g), water (535 ml) for control sample, dry yeast (20 g), corn oil (20 g) and salt (5 g). The mixing of dough by adding powder components in the mixer for 4 min at slow speed (30 r.p.m) and adding water 2 parts, after that, adding oil for 6 (min) at fast speed (60 r.p.m) to 10 min from started of mixing. The dough rested for 20 (min) at 28-30°C (first proofing), after forming, left to ferment for 60 (min) at 36°C (final proofing). Then the baking process in electrically oven at 210-220°C for 7 (min) for toast breads, loaves were separated from the metal pan and allowed to cool at room temperature before evaluation and analyses.

Chemical Analyses

Moisture, ash, protein, fats and falling number (FN) were determined according to AOAC (2000). Total carbohydrates were calculated by the difference: Total carbohydrates = 100– (g moisture + g protein + g fat + g ash).

Rheological Properties

Farinograph and extensograph tests

The rheological analyses (farinograph and extensograph) of wheat flour supplemented with different levels of AM, CMC and control flour) were measured according to AACC (2000). Where, the AACC method 54-21 and AACC method 54-10 with Brabender equipment were used to determine the farinography (Farinograph / Resistograph FA/R-2, Germany) and extensography (Brabender Extensograph DM 90-40, Germany) properties of studied flours, respectively.

Falling number

Falling Number was performed according to AACCI Approved Method 56-81.03.

Table 1. Toast bread formula and toast bread treatments

Toast bread formula		Toast bread treatment	
Component	Weight (g)	Treatment	Component
Semi-hard wheat flour (72% ext)	1000	Control	Toast bread formula
Dry yeast	20	AM 25 ppm	Toast bread formula + 25 ppm AM
		AM 30 ppm	Toast bread formula + 30 ppm AM
		AM 35 ppm	Toast bread formula + 35 ppm AM
Corn oil	20	CMC 0.1%	Toast bread formula + 0.1% CMC
		CMC 0.3%	Toast bread formula + 0.3% CMC
		CMC 0.5%	Toast bread formula + 0.5% CMC

AM= Alpha Amylase

CMC= Carboxymethyle cellelouse

Physical Properties

Determination of specific volume

Specific volume was obtained by dividing the volume of sample by their weight according to ACCC 10-05 method (AACC, 2000).

Gluten index

Wet and dry gluten were determined according to Anon (1985).

Texture profile analysis (TPA)

TPA was conducted for control and treated samples as described by **Guadarrama-Lezama et al. (2016)** and **Soleimanifard et al. (2018)**. Texture Analyzer (Brookfield Texture Pro CT V1.6 Build, USA) was used for analyses the texture of control and treated cupcake samples. The apparatus equipped with a 10000 g load cell and cylindrical probe (36 mm diameter) at a test speed of 4.00 mm/s, this equipment was used for the double compression Texture Profile Analysis (TPA) tests. Cylindrical crumbs of cupcake with 50 mm diameter and 25 mm height were compressed to 50%. A number of textural parameters (hardness1, hardness 2, Cohesiveness, Springiness, Gumminess and Chewiness) were extracted from the resultant force-time curve. The experiment was performed in triplicate.

Determination of colour by Hunter Lab

The colour of samples was measured using Hunter lab (Model 45/0 Colour FelxEz, USA) based on three colour coordinates: L^* (luminosity), a^* (redness/greenness), b^* (yellowness/blueness). The measurement for each sample was replicated and the average value was recorded for each colour parameter.

Sensory assessments

The sensory assessments of pan breads (control and different blends) were done as described by **Kralmer and Twigg (1962)**. The quality scores of toast breads included colour (20), texture (20), taste (20), flavour (20), general appearance (20) and overall acceptability was calculated (100). Scores included: 100-90 Very good (V.G.), 89-80 Good (G.), 79-70 Satisfactory (S.), Less than 70 questionably (L).

Statistical Analysis

The obtained results were evaluated statistically using analysis of variance as reported by **McClave and Benson (1991)**. In addition the other reported values were expressed as mean \pm SD and \pm SE, two – tailed Student's t test was used to compare between different groups. P value less than 0.05 was considered statistically significant. SPSS (Chicago, IL, USA) software window Version 16 was used.

RESULTS AND DISCUSSION

Physical and Chemical Composition of Semi-Hard Wheat Flour

Physical and chemical composition of semi-hard wheat flour is shown in Table 2. The results show that flour contained 13.9% moisture, 11.4% protein, 0.52% ether extract, 0.65% crude fibers, 0.57% ash, and 86.93% total carbohydrates. Also, wet and dry gluten were 25.8% and 10% respectively. Falling number and gluten index were 612 sec. and 86.4%. Similar results were recorded by **Shebl et al. (2018)**.

Table 2. Chemical composition and properties of semi-hard wheat flour (72% ext.)

Component	Per cent
Moisture	13.9±0.60
Ash	0.57±0.04
Protein	11.4±0.36
Crude fat	0.52±0.02
Crude fiber	0.65±0.60
Carbohydrates	86.93±4.12
Wet gluten content	25.8±1.74
Dry gluten content	10±0.22
Gluten index	86.4±5.42
Falling number	612±52.2

Rheological Characteristics

Effect of α -amylase concentration on the farinograph parameters

Characteristics of wheat flour extraction (72%) unfortified (control) and fortification with different additives: AM according to farinograph parameters are presented in Table 3. It was found that water absorption (%), time development (min), dough stability (min), departure time (min) decreased in all the fortified samples with addition of AM at different concentrations than that of the control values.

Concerning arrival time (min), it can be noticed that the values of arrival time decreased by the addition of AM to wheat flour (0.25, 0.30 and 0.35 ppm, respectively) compared with control value. The results showed that mixing to tolerance index (B.U) decreased by addition of AM compared with the control value. The degree of weakening of the dough was decreased due to addition of AM. This may be due to that α -amylases depolymerise damaged starch and reduced its ability to bind moisture, thus allowing more moisture to be available for gluten hydration (Shebl *et al.*, 2018). Similar results were recorded by Sanz Penella *et al.* (2008), Yang *et al.* (2014) and Nasef *et al.* (2016).

Effect of carboxymethyl cellulose concentration on the farinograph parameters

The farinograph parameters of wheat flour dough containing CMC as hydrocolloids are presented in Table 3 and Figs. 1, 2, 3, 4, 5, 6 and 7. The development time, water absorption,

stability, and weakening of dough increased with higher amounts of CMC hydrocolloids. The highest water absorption was recorded in CMC 0,5 (61.50%), while the lowest value was obtained in the control samples. Furthermore, the addition CMC positively contributed to dough development time and dough stability compared to control sample.

The rheological properties of dough are primarily attributed to protein content and the types of additives present in the dough. Higher water absorption capacity, dough development time, and dough stability could be attributed to higher protein content and the ability of hydrocolloids to absorb water within the interrelated network and their interactions with starch granules (Al-Dalain and Morsy, 2018). These results are in agreement with previously reported findings (Elhassaneen *et al.*, 2014; Ferrero, 2017).

Extensograph Parameters

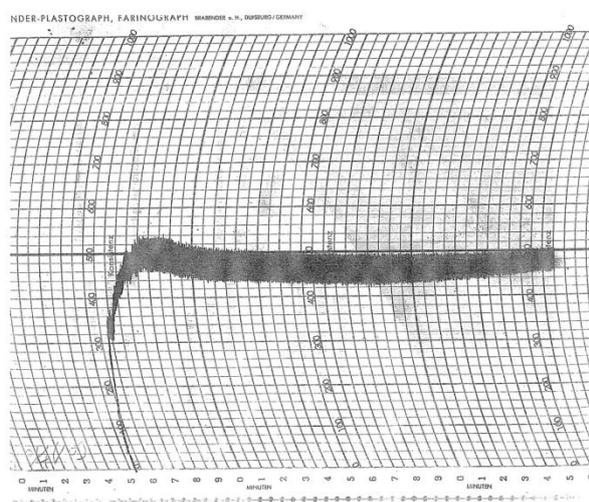
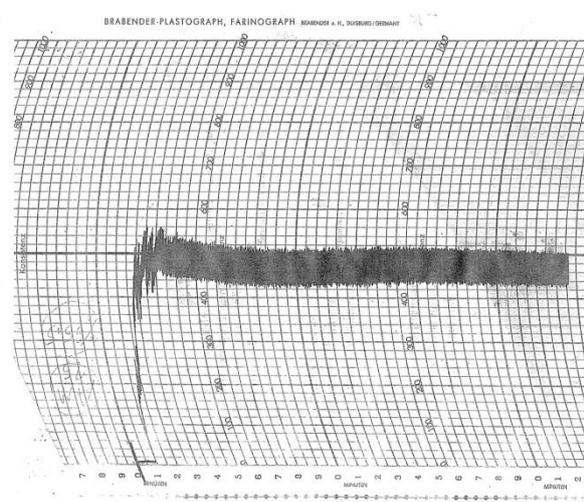
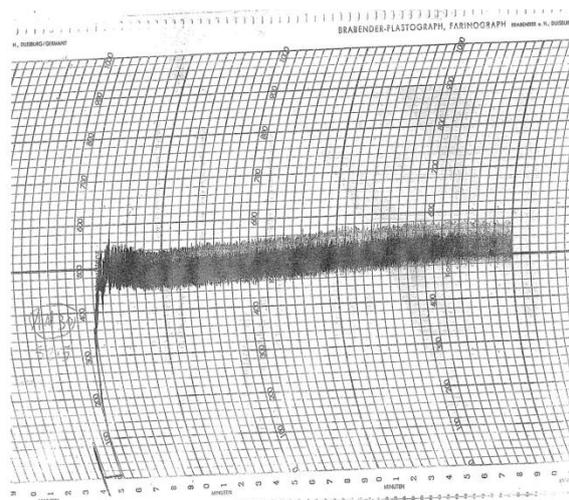
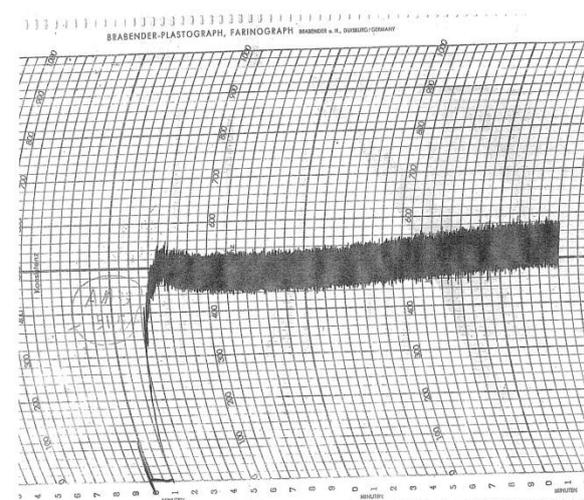
Effect of α -amylase concentration on the extensograph parameters

The effect of different concentrations α -amylase on the extensograph parameters of weak wheat flour is presented in Table 4 and Figs. 8, 9, 10, 11, 12, 13 and 14. The fungal α -amylase concentration was ranged between 0 to 0.35 ppm. The α -amylase concentration had a significant ($P \leq 0.05$) effect on the extensograph parameters. Resistance increased from 450 to 650 BU at a low concentrate AM 0.25 ppm and then decreased at higher concentrations, 0.30 and 0.35 ppm.

Table 3. Effect of α -amylase and carboxymethyl cellulose concentrations on the farinograph parameters of wheat flour (72% extraction)

Blend	Water absorption %	Arrival time (min)	Dough development time (min)	Stability time (min)	Weakening dough (B.U.)
Control	60.00±2.22 ^{ab}	1.50±0.11 ^a	2.50±0.18 ^a	8.00±0.14 ^d	35.0±1.18 ^b
AM 25 ppm	55.50±2.60 ^b	1.00±0.06 ^b	2.00±0.12 ^b	12.00±0.74 ^c	30.0±1.30 ^b
AM 30 ppm	52.50±2.72 ^c	1.00±0.05 ^b	2.00±0.16 ^b	12.00±0.66 ^c	30.0±1.58 ^b
AM 35 ppm	51.50±2.80 ^d	1.00±0.04 ^b	2.00±0.14 ^b	12.00±0.80 ^c	30.0±1.22 ^b
CMC 0.1%	60.00±2.04 ^{ab}	1.50±0.02 ^a	2.00±0.11 ^b	3.50±0.26 ^e	50.0±1.18 ^a
CMC 0.3%	60.50±2.02 ^{ab}	1.00±0.03 ^b	2.00±0.12 ^b	20.00±0.32 ^a	50.0±1.24 ^a
CMC 0.5%	61.50±2.06 ^a	1.00±0.05 ^b	1.50±0.14 ^c	15.00±0.16 ^b	50.0±1.28 ^a

* Values (means ±SD) with different superscript letters are statistically significantly different ($P \leq 0.05$). AM= Alpha Amylase CMC= Carboxymethyl cellulose

**Fig. 1. Farinograph parameters of wheat flour (control)****Fig. 2. Farinograph parameters of wheat flour containing AM 25 ppm****Fig. 3. Farinograph parameters of wheat flour containing AM 30 ppm****Fig. 4. Farinograph parameters of wheat flour containing AM 35 ppm**

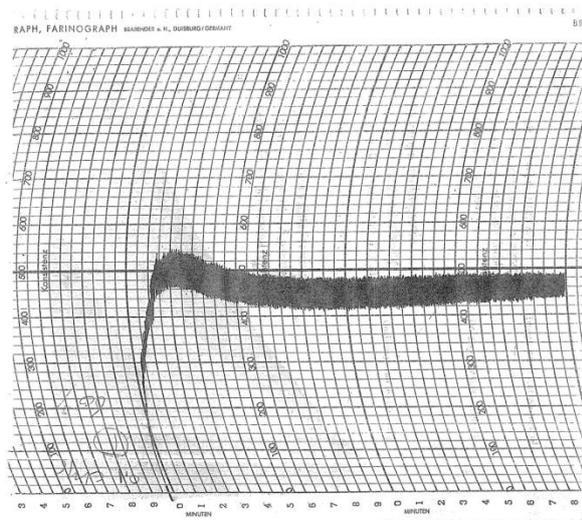


Fig. 5. Farinograph parameters of wheat flour containing 0.1% CMC

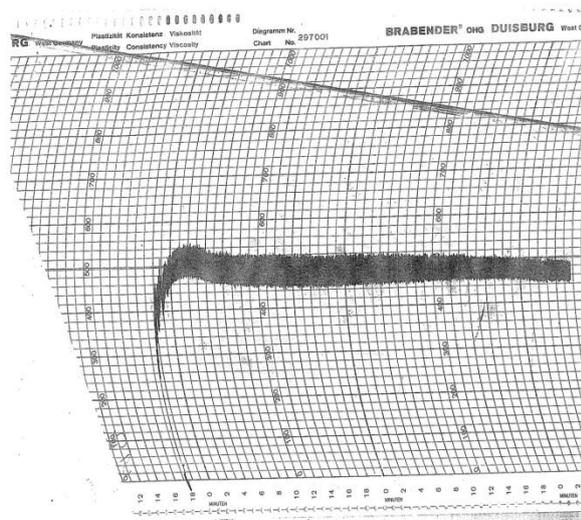


Fig. 6. Farinograph parameters of wheat flour containing 0.3% CMC

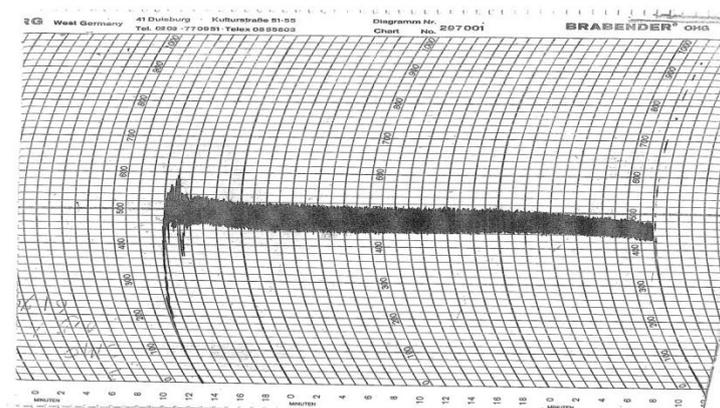


Fig. 7. Farinograph parameters of wheat flour containing 0.5% CMC

Table 4. Effect of α -amylase (AM) and carboxymethyl cellulose (CMC) concentrations on the extensograph parameters of wheat flour (72% extraction)

Blend	Resistance to extension (B.U)	Extensibility (mm)	Proportional number	Energy (cm ²)
Control	450±7.48 ^b	170±3.54 ^a	2.65±0.04 ^e	45±2.27 ^e
AM 25 ppm	650±9.20 ^a	80±2.87 ^f	8.12±0.66 ^a	55±3.12 ^d
AM 30 ppm	230±6.34 ^d	140±3.82 ^c	1.64±0.04 ^f	35±2.54 ^f
AM 35 ppm	210±8.12 ^a	130±3.90 ^a	1.61±0.08 ^f	45±3.06 ^e
CMC 0.1%	640±7.55 ^a	105±3.72 ^{dc}	6.09±1.26 ^b	70±2.52 ^b
CMC 0.3%	460±9.12 ^b	150±3.82 ^b	2.87±0.08 ^d	75±3.16 ^a
CMC 0.5%	420±8.20 ^c	120±4.32 ^d	3.50±0.80 ^c	65±3.50 ^c

* Values (means ±SD) with different superscript letters are statistically significantly different ($P \leq 0.05$). AM= Alpha Amylase CMC= Carboxymethyl cellulose

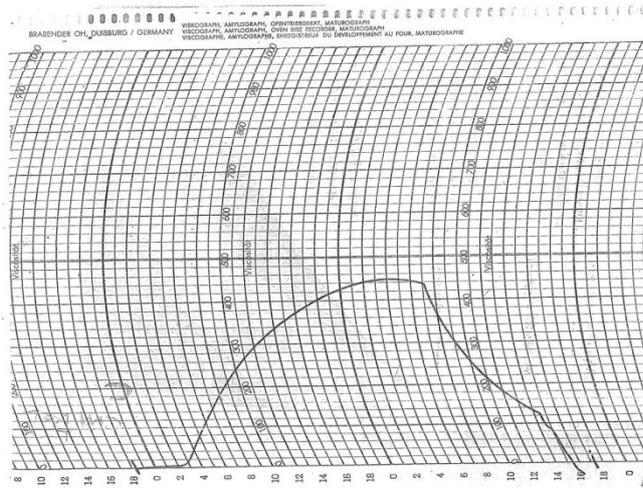


Fig. 8. Extensograph parameters of wheat flour (control)

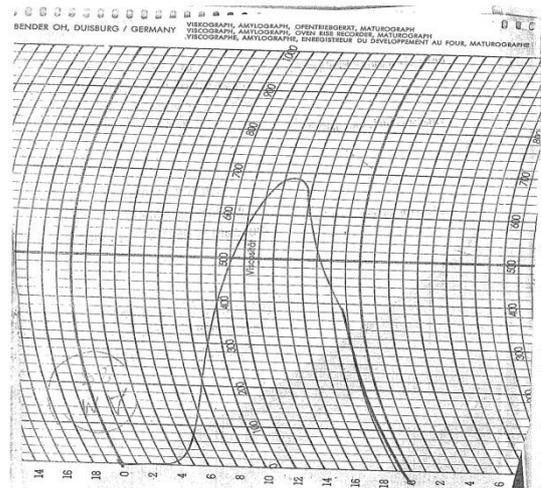


Fig. 9. Extensograph parameters of wheat flour containing AM 25 ppm

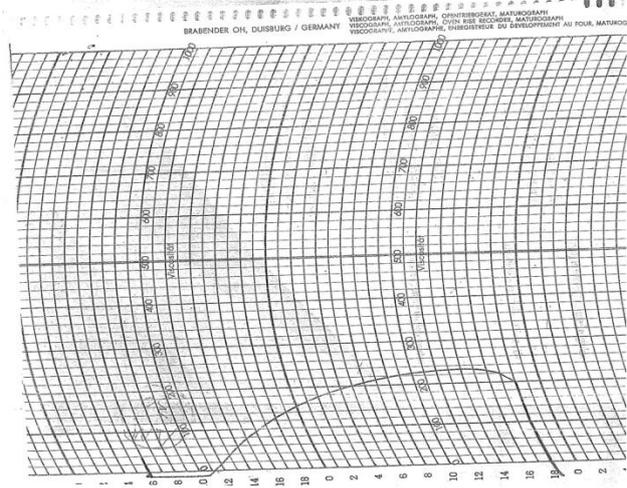


Fig. 10. Extensograph parameters of wheat flour containing AM 30 ppm

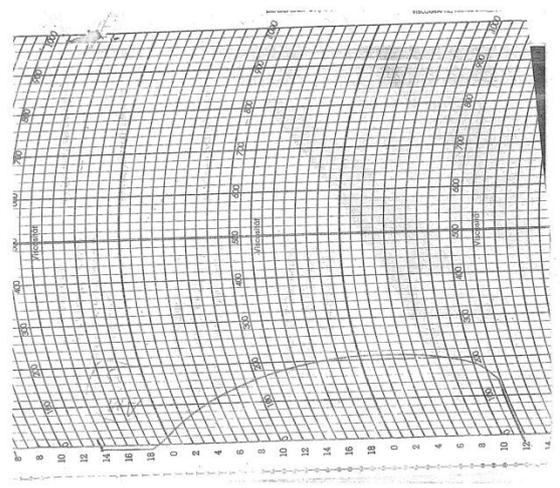


Fig. 11. Extensograph parameters of wheat flour containing AM 35 ppm

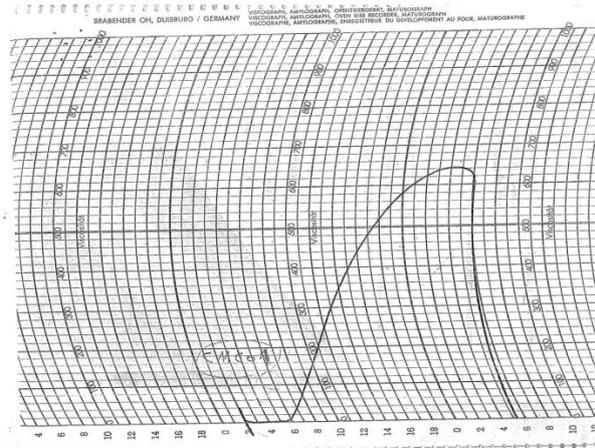


Fig. 12. Extensograph parameters of wheat flour containing 0.1% CMC

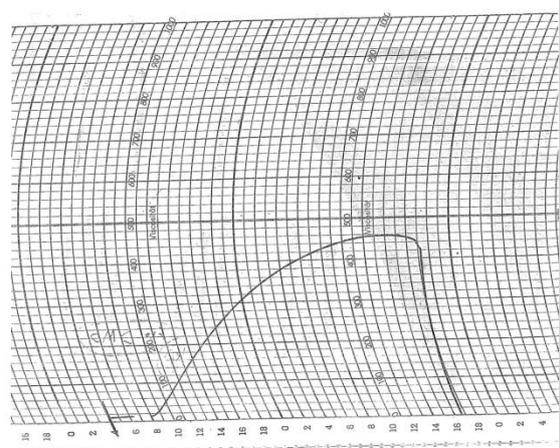


Fig. 13. Extensograph parameters of wheat flour containing 0.3% CMC

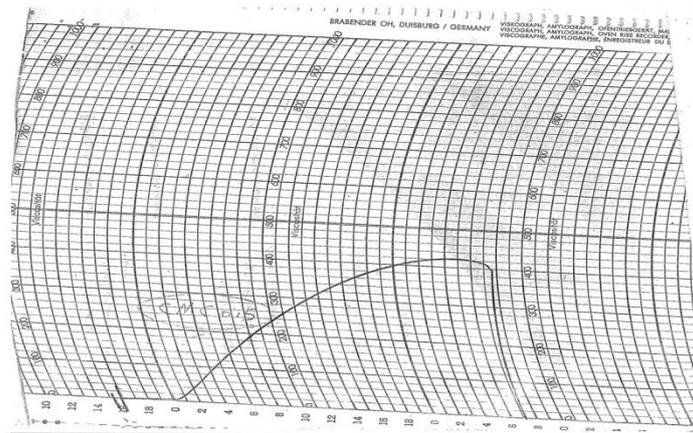


Fig. 14. Extensograph parameters of wheat flour containing 0.5% CMC

Effects of addition CMC and AM enzyme on chemical composition of produced toast bread

The different blends of toast bread were analyzed for their chemical composition and the results are presented in Table 5. The main values (%) of carbohydrate, protein, moisture, ash and fat of unfortified toast bread (control) were found to be 49.19, 12.82, 33.73, 1.20 and 3.06, respectively. Carbohydrate content among unfortified and fortified toast bread with different additives: α -amylase and CMC at different concentrations. Moreover, there was slight difference in protein content of unfortified and fortified toast bread with different additives: α -amylase and CMC at different concentrations compared with toast bread unfortified (control). However, protein value of toast bread fortified with Alpha amylase powder (AM) at higher ratio was higher than that of the other toast bread samples and the control.

There was an increase in product moisture content by the addition of CMC. Hydrocolloids commonly known as water soluble gum can bind as much as 100 times their weight of water and thus help reduce water migration and ice crystal formation and increase product stability. These facts explain the observation of the present study which showed that CMC caused an increase in bread moisture content when being added individually or in bread and other leavened bakery products, produces dough with constant functional properties and good water holding characteristics (Sanderson, 1996; Nasef *et al.*, 2016).

On the other hand, moisture content of the fortified samples with additives such as alpha amylase (AM), decreased. The highest ash content of the prepared toast bread was obtained upon the addition of 0.50 CMC individually (1.46), while the unfortified sample (control) showed to contain (1.20%) ash. Generally, the study revealed that toast bread fortified with CMC under the present experimental conditions had higher values of ash as compared to the other prepared samples.

Results presented in Table 5 also showed that there are non-significant ($p < 0.05$) differences in the mean values of fat content among the prepared sample and the control. These results are in the same line with Azizi *et al.* (2006) and Nasef *et al.* (2016).

Effects of addition CMC and AM enzyme on physical properties (Loaf volume and specific volume) of produced toast bread

Addition of CMC and AM enzyme produced the greatest increase in loaf volume and specific volume. AM enzyme treatment significantly increased the loaf volume and specific volume (Table 6). In the case of α -amylase, all three levels significantly increased volume compared to control. Maltogenic α -amylase produces primarily α -maltose through the hydrolysis of α -1,4 glycosidic bonds within the starch polymer. It is believed act as an endo-enzyme, but also has exo-action especially at higher temperatures in white pan bread, the effect of maltogenic α -amylase on loaf volume has been inconsistent (Goesaert *et al.*, 2009; Gomes-Ruffi *et al.*, 2012). The present studies showed an increase in volume due to maltogenic α -amylase, depending on the dose.

Table 5. Effects of addition CMC and AM enzyme on chemical composition of produced toast bread

Treatment	Moisture	Protein	Fat	Ash	Total carbohydrates
Control	33.73±1.02 ^{cd}	12.82±0.65 ^c	3.06±0.12 ^a	1.20±0.08 ^d	49.19±1.14 ^b
AM 25 ppm	33.16±1.13 ^d	12.96±0.42 ^c	3.02±0.09 ^a	1.12±0.12 ^e	49.74±1.12 ^b
AM 30 ppm	32.28±1.22 ^{de}	13.27±0.38 ^b	3.05±0.14 ^a	1.10±0.14 ^e	50.30±1.10 ^a
AM 35 ppm	32.43±1.18 ^e	13.79±0.32 ^a	3.02±0.11 ^a	1.20±0.11 ^d	49.56±1.22 ^b
CMC 0.1%	34.34±1.06 ^c	12.84±0.48 ^c	3.08±0.07 ^a	1.32±0.08 ^c	48.42±1.34 ^{bc}
CMC 0.3%	35.38±1.02 ^b	12.86±0.62 ^c	3.04±0.12 ^a	1.40±0.04 ^b	47.32±1.24 ^c
CMC 0.5%	36.30±1.04 ^a	12.90±0.53 ^c	3.02±0.08 ^a	1.46±0.02 ^a	46.32±1.42 ^{cd}

* Values (means ±SD) with different superscript letters are statistically significantly different ($P \leq 0.05$). AM=Alpha Amylase
CMC= Carboxymethyl cellulose

Yeast will preferentially ferment glucose followed by fructose, which are the two products of sucrose hydrolysis. Once the concentration of glucose and fructose is diminished, yeast activates its maltase and maltose permease enzymes, allowing the organism to hydrolyze and ferment maltose (Sluimer, 2005). Instead, the increase in loaf volume due to α -amylase may be related to a decrease in dough viscosity during starch gelatinization, hence prolonging oven rise (Goesaert *et al.*, 2009). This reasoning could explain the increased loaf volume observed for both conventional α -amylase, which generates low molecular weight α -dextrins and oligosaccharides of varying length, and for maltogenic α -amylase. The dextrins and oligosaccharides produced from the hydrolysis by conventional α -amylase are further hydrolyzed into maltose by endogenous β -amylase (Palacios, 1998).

With the exception of CMC hydrocolloids increased specific volume of the whole wheat bread for at least one of the levels evaluated (Table 6). The findings for CMC and hydrocolloids are in accordance with published works on whole wheat bread (Armero and Collar, 1996a).

The ability of hydrocolloids to improve loaf volume is often attributed to a strengthening effect on the gluten network and an improvement in gas retention (Bárcenas *et al.*,

2009; Linlaud *et al.*, 2011; Ribotta *et al.*, 2005). The interaction with especially gluten, the main structural component of bread, is of particular interest in explaining the effect of hydrocolloids on loaf volume. Such interactions include hydrogen bonding, in the case of neutral hydrocolloids like guar gum, and non-covalent linkages between amide groups of gluten and the hydroxyl groups of anionic hydrocolloids like xanthan gum and alginate (Linlaud *et al.*, 2011; Ribotta *et al.*, 2005). Hydrocolloids have been shown to alter the secondary structure of gluten proteins (Linlaud *et al.*, 2011), which affects the gluten network.

Effects of addition CMC and AM enzyme on texture profile of produced toast bread

Supplementation with the high levels of α -amylase decreased crumb hardness and chewiness as measured on the first day after baking (Table 7). The enzymes produced little effect on the texture profile analysis (TPA) parameters, except for α -amylase, which decreased crumb resilience, cohesion, springiness, and chewiness. Loaf volume is a major contributor to hardness, but the nature of the crumb material is also involved (Armero and Collar, 1996a).

The lowest hardness value was obtained with the highest dose of α -amylase produced compared to control bread. Several other researchers have reported significant reductions in crumb hardness for whole wheat bread

Table 6. Effects of addition CMC and AM on physical properties of produced toast bread

Treatment	Loaf Volume (cm ³)	Weight (g)	Specific Volume (cm ³ /gm)
Control	1314 ±8.32 ^f	231.33 ±2.40 ^b	5.68 ±0.24 ^c
AM 25 ppm	1356 ±8.14 ^c	226.38 ±2.58 ^d	5.98±0.16 ^b
AM 30 ppm	1372 ±8.04 ^b	228.43 ±2.74 ^c	6.00 ±0.14 ^b
AM 35 ppm	1398 ±7.58 ^a	224.00 ±2.88 ^e	6.24 ±0.18 ^a
CMC 0.1%	1335±8.33 ^e	233.0±2.14 ^a	5.72±0.33 ^{bc}
CMC 0.3%	1344±8.12 ^d	233.40±2.128 ^a	5.75±0.30 ^{bc}
CMC 0.5%	1350±8.24 ^{cd}	233.70±2.22 ^a	5.77±0.28 ^{bc}

* Values (means ±SD) with different superscript letters are statistically significantly different (P≤0.05). AM= Alpha Amylase
CMC= Carboxymethyl cellulose

Table 7. Effects of addition CMC and AM enzyme on texture profile of produced toast bread

Treatment	Hardness N1	Hardness N2	Adhesiveness	Resilience	Cohesiveness	Springiness	Gumminess	Chewiness
Control	20.91±1.44 ^d	19.39±1.58 ^d	0.40±0.22 ^c	0.42±0.04 ^{cd}	0.81±0.12 ^c	8.62±1.14 ^a	16.84±1.42 ^b	145.10±7.20 ^a
AM 25 ppm	39.12±2.28 ^b	35.24±1.14 ^b	3.30±0.14 ^b	0.13±0.06 ^d	0.66±0.23 ^d	0.48±1.22 ^c	26.00±1.08 ^a	125.30±8.25 ^b
AM 30 ppm	42.25±1.12 ^a	37.47±1.02 ^a	3.50±0.12 ^a	0.11±0.08 ^{de}	0.60±0.27 ^d	0.39±1.36 ^c	25.16±1.14 ^{ab}	97.60±8.16 ^{dc}
AM 35 ppm	27.18±2.40 ^e	24.42±1.66 ^c	2.30±0.18 ^c	0.13±0.08 ^d	0.64±0.20 ^d	0.42±1.28 ^c	17.47±1.52 ^b	73.60±8.28 ^c
CMC 0.1%	18.57±2.36 ^e	17.02±1.84 ^e	1.00±0.25 ^d	0.73±0.02 ^a	0.78±0.18 ^{cd}	7.47±1.13 ^b	14.47±1.68 ^c	108.10±7.40 ^c
CMC 0.3%	13.57±2.68 ^e	13.00±1.20 ^e	0.20±0.31 ^f	0.52±0.04 ^b	0.99±0.11 ^a	7.53±1.14 ^b	13.42±1.70 ^c	101.0±7.55 ^d
CMC 0.5%	15.35±2.22 ^f	14.28±1.32 ^f	-0.10±0.33 ^e	0.44±0.06 ^c	0.87±0.14 ^b	7.54±1.12 ^b	13.33±1.66 ^{cd}	100.50±7.52 ^d

* Values (means ±SD) with different superscript letters are statistically significantly different (P ≤ 0.05).

AM= Alpha Amylase

CMC= Carboxymethyl cellulose

supplemented with enzymes (**Driss *et al.*, 2013; Ghoshal *et al.*, 2013**). The textural change is most often attributed to the increase in volume. A reduction in starch crystallization and crystal growth, based on texture profile analysis (TPA) analysis, has also been suggested (**Ghoshal *et al.*, 2013**).

Conventional α -amylase showed a trend for decreasing hardness. Conventional α -amylase is commonly used to improve loaf volume, and a decrease in crumb hardness for whole wheat bread has been reported (**Armero and Collar, 1996a; Matsushita *et al.*, 2017**), and it has been shown to decrease hardness and firming in white bread (**Goesaert *et al.*, 2009**).

Conventional α -amylase is an endo-enzyme that acts on damaged starch and gelatinized starch. This action reduces the molecular weight of the polymers and weakens the starch networks present in the final loaf, which can contribute to a decrease in crumb firmness (**Goesaert *et al.*, 2009**).

Additionally, dextrans of intermediate size inhibit crumb firming by interfering with crosslinking between remnants of starch granules and protein fibrils (**Martin and Hosney, 1991**).

Although staling involves changes in several quality parameters including moisture migration and loss, loss of aroma, and textural changes (**Hug-Iten *et al.*, 2003**), perhaps the most important characteristic of staling is an increase in crumb hardness over time, which is also referred to as firming. Table 7 displays the rate of firming as defined by the slope of the increase in hardness during storage. The plot of this firming data a pronounced decrease in firming rate was obtained for α -amylase at the medium and high levels.

Maltogenic α -amylase is generally used in bread formulations for its anti-staling effect, which is mostly accomplished by the hydrolysis of amylopectin side chains, thus preventing retrogradation (**Goesaert *et al.*, 2009**). Amylopectin retrogradation may result in crumb firming due

to the immobilization of water within the crystal structure. That water is consequently unavailable to plasticize the gluten network (Goesaert *et al.*, 2009).

By limiting the formation of amylopectin crystallites, maltogenic α -amylase allows more water to remain available as a plasticizer, thus leading to a decrease in firming. The anti-firming effect of maltogenic α -amylase may also be attributed to modifications of the amylose fraction (Hug-Iten *et al.*, 2003). Overall, the exact mechanisms of bread staling and their impact on crumb firming remain unclear (Fadda *et al.*, 2014).

Texture profile analysis (TPA) of toast bread revealed mostly non-significant reductions in crumb hardness as a result of hydrocolloids, except for significant reductions due to the medium level of CMC (Tables 7). The initial softening effect of CMC in whole wheat bread has been previously reported (Armero and Collar, 1996a; Zannini *et al.*, 2014). The other textural parameters measured by TPA were also largely unaffected by hydrocolloid addition. The treatments that produced the largest loaf volume were not always the ones with the lowest values for crumb hardness, reinforcing the fact that although loaf volume is a major contributor to firmness (Collar *et al.*, 1998), gas retention capacity and increased water absorption of dough (Zannini *et al.*, 2014) and the specific nature of the crumb also play a role in the resistance of crumb to compression (Armero and Collar, 1996a).

The increase in hardness with the high level of CMC could be caused by the low loaf volume compared to the low and medium treatments (Collar *et al.*, 1998). The increased hardness can also be caused by a lack of water for plasticizing the gluten network (Goesaert *et al.*, 2009). CMC the low and medium levels showed a trend for decreasing the rate of staling based on the rate of increase in crumb hardness over time.

The anti-staling effect of CMC could result from its ability to hinder interactions among the other components in the crumb by enveloping them in a polymer network (Barcenas and Rosell, 2005) and by its preferential binding to starch, which influences the interactions among

lipid, starch, and gluten (Collar *et al.*, 1998). Water retention capacity and starch interactions have also been proposed to explain the softening effects of hydrocolloids (Guarda *et al.*, 2004).

Effects of addition CMC and AM enzyme on colour properties of produced toast bread

The colour of bread crust is one of the important quality factors affecting consumer preference expected from bread products. In general, bread crust is characterized as low moisture and dark brown in colour. The typical bread colour on the crust is generated by chemical reactions such as the Maillard reaction and caramelization (Kim and Yoo, 2020).

Table 8 shows that as the dosage level of AM increased in the bread dough formulation, reducing sugar formation was accelerated and the released sugars were utilized for the Maillard reaction. The resulting outcome displayed low L^* values and led to a much darker crust formation. The crust colour between light and dark brown was preferred by most consumers, and the flavour and taste of baked bread could be improved as the result of the Maillard reaction (Kim and Yoo, 2020). These results agree with Eugenia Steffolani *et al.* (2012) and Kim and Yoo (2020). While as the dosage level of CMC resulting outcome displayed high L^* and b values. The increased b values could be attributable to a more favorable water distribution due to the hydrocolloids, which affects Maillard browning reactions and caramelization (Sciarini *et al.*, 2010). These results agree with Eduardo *et al.* (2015).

Effects of addition CMC and AM enzyme on sensory characteristics of produced toast bread

Sensory evaluation results of toast bread containing each of AM and CMC individually and control sample after baked are shown in Table 9. It was found that toast bread containing (AM) and CMC had higher significant ($p < 0.05$) scores for all the evaluated characteristics: appearance, colour, texture, flavour, taste, and general acceptability as compared to the control. Moreover, toast bread fortified with (AM) had higher significant ($p < 0.05$) score for sensory characteristics compared with CMC treatments

Table 8. Effects of addition CMC and AM enzyme on colour properties of produced toast bread

Treatment	l	a	b
Control	36.85 ±1.76 ^d	12.31±0.44 ^b	17.89±0.74 ^d
AM 25 ppm	33.96 ±1.88 ^f	13.18±0.36 ^{bc}	16.36±0.86 ^e
AM 30 ppm	35.55±1.70 ^e	12.68±0.52 ^b	16.19±0.82 ^e
AM 35 ppm	31.88±1.96 ^g	12.12±0.46 ^{bc}	15.17±0.90 ^{ef}
CMC 0.1%	38.45±1.42 ^c	12.00±0.38 ^c	20.24±0.66 ^c
CMC 0.3%	40.28±1.36 ^b	12.24±0.36 ^{bc}	23.62±0.54 ^b
CMC 0.5%	42.36±1.40 ^a	13.04±0.30 ^a	24.51±0.50 ^a

* Values (means ±SD) with different superscript letters are statistically significantly different (P≤0.05). AM= Alpha Amylase
CMC= Carboxymethyle cellulouse

Table 9. Effects of addition CMC and AM enzyme on sensory characteristics of produced toast bread

Treatment	Appearance 20	Taste 20	Flovour 20	Texture 20	Colour 20	Overall Acceptability 100
Control	17.0±0.46 ^c	18.0±0.55 ^b	18.0±0.44 ^c	17.0±0.76 ^c	18.0±0.70 ^b	88.0±1.00 ^f
AM 25 ppm	17.0±0.50 ^c	19.0±0.46 ^a	19.0±0.36 ^b	19.0±0.44 ^a	18.0±0.72 ^b	92.0±0.86 ^c
AM 30 ppm	18.0±0.42 ^b	19.0±0.42 ^a	20.0±0.40 ^a	19.0±1.46 ^a	20.0±0.52 ^a	96.0±0.78 ^a
AM 35 ppm	18.0±0.44 ^b	19.0±0.46 ^a	19.0±0.50 ^b	19.0±0.46 ^a	18.0±0.66 ^b	93.0±0.96 ^b
CMC 0.1%	17.0±0.50 ^c	18.0±0.50 ^b	18.0±0.55 ^c	18.0±0.55 ^b	18.0±0.64 ^b	90.0±0.88 ^e
CMC 0.3%	19.0±0.40 ^a	19.0±0.44 ^a	18.0±0.46 ^c	19.0±0.52 ^a	18.0±0.66 ^b	93.0±0.82 ^b
CMC 0.5%	18.0±0.42 ^b	18.0±0.55 ^b	18.0±0.44 ^c	19.0±0.50 ^a	18.0±0.64 ^b	91.0±0.78 ^d

* Values (means ±SD) with different superscript letters are statistically significantly different (P≤0.05). AM= Alpha Amylase
CMC= Carboxymethyle cellulouse

at the same concentration. The present result agreed with the reported findings by **Hemalatha *et al.* (2011)** who stated that the appropriate levels of fungal α -amylase have been reported to improve the crumb structure and texture of the final bread. All additive treatments achieved significant ($p < 0.05$) higher scores for all characteristics as compared to control after being backed. As reported by **Nasef *et al.* (2016)** who found that addition of enzymes and galactomanan gum to wheat Balady bread enhanced the sensory characteristics of the final products. Also, **Aleid *et al.* (2015)** found that fortification of wheat bread with arabic gum, mongglycerides and alpha-amylase enhancing the sensory characteristics of Arabic bread.

Conclusion

This study demonstrated the specific application of enzymes and hydrocolloids in toast bread to increase loaf volume and decrease initial crumb hardness and bread staling, which may help improve the physicochemical, rheological and sensory properties of toast bread.

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الخواص الفيزيوكيميائية والريولوجية لخبز القالب المدعم بانزيم الالفا اميليز والكربوكسي ميثيل سليلوز

مريم محمد عباس عبد النبي - شريف عيد النمر - سامى محمد ابو المعاطى - صباح محمد منير

قسم علوم الأغذية - كلية الزراعة - جامعة الزقازيق - مصر

تم دراسة تأثير اضافة كلا من انزيم الالفا اميليز والكربوكسي ميثيل سليلوز علي الخواص الفيزيوكيميائية والريولوجية والتركيبية والحسية لخبز القالب، حيث تم اضافة الالفا اميليز الي الدقيق المستخدم بنسبة 25 ، 30 و35 جزء في المليون كما تم اضافة الكربوكسي ميثيل سليلوز بمعدلات 0.1، 0.3 و0.5% ووضحت النتائج ان عينات خبز القالب المحتوية علي انزيم الالفا اميليز بأعلى تركيز كان محتواها من البروتين مرتفع عن باقي المعاملات كما حدثت زيادة في محتوى الرطوبة في المعاملات المحتوية علي الكربوكسي ميثيل سليلوز وواظهرت المعاملة المحتوية علي 0.5% كربوكسي ميثيل سليلوز اعلي محتوى للرماد عن باقي المعاملات كما لم يلاحظ اي فروق معنوية في محتوى الدهن بين كل المعاملات، إضافة الالفا اميليز والكربوكسي ميثيل سليلوز زادت من الحجم النوعي لعينات خبز القالب واطهرت معاملات الالفا اميليز زيادة في الحجم النوعي مقارنة بعينات الكربوكسي ميثيل سليلوز. كما قلت صلابة العينات باضافة الالفا اميليز بينما حدثت زيادة للصلابة باضافة الكربوكسي ميثيل سليلوز خصوصا عند التركيز العالي منها. كما اظهرت العينات المحتوية علي الالفا اميليز قيم L منخفضة مقارنة بعينات الكربوكسي ميثيل سليلوز التي اظهرت قيم L, b مرتفعة. ومن حيث الخواص الحسية فان اضافة كلا من الالفا اميليز والكربوكسي ميثيل سليلوز حسنت من الخواص الحسية لخبز القالب الناتج وكانت المعاملات المحتوية علي الالفا اميليز لها تأثير أفضل علي الخواص الحسية مقارنة بالمعاملات المحتوية علي الكربوكسي ميثيل سليلوز. من هذه النتائج يمكن استنتاج انه يمكن اضافة بعض المحسنات للدقيق مثل الالفا اميليز بمعدل 30 جزء في المليون والكربوكسي ميثيل سليلوز بمعدل 0.30% عند صناعة خبز القالب حيث حسنت هذه الاضافات من الخواص الكيميائية والريولوجية لخبز القالب الناتج.

المحكمون :

- 1 - أ.د. يوسف ذكي أبو العزم
- 2- أ.د. كمال محفوظ الصاحي

أستاذ الصناعات الغذائية المتفرغ - معهد بحوث تكنولوجيا الأغذية - مركز البحوث الزراعية.
أستاذ الصناعات الغذائية المتفرغ - كلية الزراعة - جامعة الزقازيق.