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Effect of α-amylase enzyme on rheological properties and quality of Betifore-type cookies

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ABSTRACT

Wheat flour was treated with α-amylase at 5, 7.5 and 10 ppm levels to prepare betifore-type cookies. The rheological properties of dough and sensory properties of betifore were evaluated. Physical properties of betifore prepared with 7.5 ppm α-amylase were also evaluated. Water absorption of dough was not affected by treating wheat flour with α-amylase. Dough stability, (P) value, (P/L) ratio and (W) value were decreased by treating wheat flour with α-amylase, whereas the (L) value showed an opposite trend. Treating wheat flour with 7.5 ppm α-amylase reduced (W) value from 229×10-4 to 149×10-4 joule. This value indicated that the wheat flour is more suitable for betifore making. Betifores prepared with 7.5 ppm α-amylase had higher mean scores for all sensory properties, spread ratio and spread factor than the control.

Key words: Wheat flour, α-amylase, Mixolab, Alveograph, Farinograph, Betifore.

INTRODUCTION

Betifore-type cookies are Egyptian type cookies, which are highly consumed by most of the Egyptian people, especially during festivals. This type of product contains about 30% butter oil (Mansour *et al.*, 2003). Many attempts have been made to improve the biscuit quality. The enzymatic treatment of wheat flours is an interesting alternative to generate changes in the structure of the dough and in consequence, for improving functional properties of flours. They are generally recognized as safe (GRAS) and do not remain active in the final product after baking. Therefore, enzymes do not have to appear on the label, which is an additional commercial advantage (Caballero *et al.*, 2007; Hassan *et al.*, 2014).

Fungal α-amylases are usually added to optimize the amylase activity of the flour, initially aiming to increase the levels of fermentable and reducing sugars. The supplemented α-amylases break down damaged starch particles into low molecular weight dextrins during the dough stage, while endogenous β-amylase converts these oligosaccharides into maltose which is used as fermentable sugar by the yeast or sourdough microorganisms (Goesaert *et al.*, 2005; Synowiecki, 2007). The increased levels of reducing the sugars lead to the formation of Maillard reaction products, intensifying the bread flavour and crust colour. In addition, these enzymes can improve the gas-retention properties of fermented dough and reduce dough viscosity during starch gelatinization, with consequent improvements in product volume and softness (Cauvain and Young 2006; Goesaert *et al.*, 2009).

The addition of 1% malt to the wheat-cassava composite fours improved α-amylase activity and rheological properties. This improvement was translated into good bread making potential

(Khalil *et al.,* 2000). This study aimed to evaluate the effect of different levels of α-amylase enzyme on the rheological properties of dough and betifore-type cookies quality.

MATERIALS AND METHODS

Wheat flour, 72% extraction was obtained from El-Tahanoon El-Masrion, number 16 in the third industrial zone, 6 of October, Egypt. Powdered sugar, shortening, salt and vanilla flavouring were purchased from local markets, Shibin El-Kom, Egypt. Baking powder (corn flour 45%, sodium pyrophosphate; E450a; 31% and sodium hydrogen carbonate 24%) was purchased from Pearce Duff, England. Fresh whole eggs were obtained from Poultry Farm, Minufiya University, Shibin El-Kom, Egypt. Fungal α-amylase (Fungamyl SG) containing 2500 fungal amylase units/g was obtained from Novozymes, Denmark.

Physical properties of wheat flour:
Zeleny sedimentation and falling number values of the flour were determined according to AACC approved methods No: 56–60 and 56–81B, respectively (AACC, 2000).

Analytical methods:
Ash, wet gluten and its index of flour were determined according to AACC approved methods No: 08-03, 38-12 AACC Standard Methods (2000). The damaged starch content of the flour samples was determined by the SDMatic (Chopin Technologies, Villeneuve la Garenne, France) which provides results in AACC units.

Rheological properties:

Mixolab properties
Dough mixing and pasting behaviours of the wheat flour samples were studied using the mixolab (Chopin Technologies, Villeneuve La Garenne, France). Mixolab analysis was carried out using the water absorption level determined by the consistograph according the method of AACC 54-50 (2000). For the analysis of the mixing and pasting behaviour, the standard ‘‘Chopin +’’ protocol was followed: initial equilibrium at 30°C for 8 min, heating to 90°C over 15 min (at a rate of 4°C/min), holding at 90°C for 7 min, cooling to 50°C over 5 min (at a rate of 4°C/min) and holding at 50°C for 5 min. The mixing speed was kept constant at 80 rpm. The following parameters were recorded: water absorption, development time, stability at mixing, C1, C2, C3, C4 and C5 values.

Alveograph properties
Alveograph test was carried out in an Alveograph MA 82 (Chopin, Tripette et Renaud, France) following the AACC Approved Method No: 54-30 (2000). The following Alveograph parameters were automatically recorded by a computer software program: the maximum over pressure (P) expressed the resistance of the dough to deformation, and is related to the dough's tensile strength and stability, the average abscissa (L) represented the extensibility of the dough or its ability to rise, P/L indicating the ratio of elasticity to the extensibility of the dough and the deformation energy (W) an index of dough strength.

Farinograph Properties
Farinograph properties (water absorption, arrival time, dough stability, dough tolerance, dough weakening and degree of softening) were carried out according to the method of AACC (2000) No: 54-21 using a Barbender Farinograph (mixer bowl 300 g, Barbender OHG, Duisburg, Germany). The equivalent of 300 g of flour (14% moisture basis) is placed in the farinograph bowl. The instrument is turned on, and water is added from a burette. As the flour hydrates and the dough forms, the resistance on the mixing blades increases, and the pen on the chart recorder and/or the curve on the computer screen rises. The mixing curve obtained generally rises to a maximum and then slowly falls from that point. To ensure that farinograms from different samples can be compared, the midpoint of the farinograph bandwidth at the maximum resistance is always centered on the 500-Barbender Units (BU) line.

Preparation of betifores:
Betifores were prepared according to Mansour *et al.* (2003). Betifores formula consisted of 37.3% wheat flour, 29.9% shorting, 14.9% powdered

sugar, 13.1% whole egg, 0.59% baking powder, 0.47% salt, 0.04% vanilla and 3.7% water. To prepare the control treatment, the shorting and sugar were creamed for two min at low speed (speed 4) in an Oster Kitchen Center mixer (Model 972-26 H, Sunbeam Corporation, Milwaukee, Wisconsin, USA). The whole eggs and vanilla were added and mixed for another 2 min at the same speed. The blended dry ingredients (flour, baking powder and salt) and water were added to the mixture and beaten for 4 min at medium speed (speed 6). A manual scoop (about 5 g weight, 5 cm diameter and 0.2 cm thick) was used to drop betifores onto trays. The betifores were baked at 190°C for 12 min. Betifores were allowed to cool for 30 min before wrapping in polyethylene film. The α-amylase enzyme was added to the flour at the levels of 5, 7.5 and 10 ppm. The same order of mixing described for the control was followed.

Sensory properties of betifores:

Sensory properties of betifores were performed by ten trained graduate students. The randomly coded samples were served to the panellists individually. Panellists were supplied with cold tap water for cleaning the palate between samples. Sensory quality attributes were evaluated using a 10-point rating scale with 1 for dislike extremely and 10 for like extremely for each attribute is evaluated. Betifores were evaluated for appearance, texture, crispness, colour, flavour, mouth feel and overall acceptability.

Physical properties of betifores:

Betifores were evaluated for weight (g), width (cm), thickness (cm), spread ratio and spread factor. Spread ratio and spread factor were calculated according to Elkhalifa and Tinay (2002) using the following equations:

Spread ratio = width / thickness

Spread factor = (Spread ratio of the sample / speed ratio of the control) × 100

Statistical analysis:

The sensory and physical properties data of betifore were subjected to an analysis of variance (ANOVA) for a completely random design using a Statistical Analysis System (SAS Institute, Inc., Cary, NC, 2008). Duncan's multiple range tests were used to determine the difference among means at the level of 0.05.

RESULTS AND DISCUSSION

Physical and chemical properties of wheat flour:

Physical and chemical properties of wheat flour are presented in Table (1). The Zeleny sedimentation value of wheat flour was 25 ml, thus indicating that the wheat flour was medium suitable for bread making. Belderok (2000) reported that wheat flour with a Zeleny sedimentation value 20-30 ml is medium suitable for bread making and has medium protein content. Sedimentation values can be in the range of 20 or less for low-protein wheat with weak gluten to as high as 70 or more for high-protein wheat with strong gluten (AACC, 2000).

The falling number is indicative of the α-amylase activity and the fermentation process taking place in wheat flour dough. The falling number value in wheat flour was 454 Sec. Barrera *et al.* (2007) reported that, a falling number value of 350 seconds or longer indicates a low enzyme activity and very sound wheat quality (quite suitable for bread making). Values below 200 Sec indicate high levels of enzyme activity.

Zeleny sedimentation (ml)	Falling number (sec)	Ash (%)	Damage starch (%)	Gluten index		
				Dry gluten (%)	Wet gluten (%)	Gluten index (%)
25	454	0.53	6.0	8.6	23.6	89.3

Table 1: Physical and Chemical properties of wheat flour[1]

[1]14% moisture content basis

Mixolab properties	Levels of α-amylase (ppm)			
	0	5	7.5	10
Hydration[1] (%)	62.3	62.3	62.3	62.3
Development time (min)	1.13	1.10	1.00	1.13
Stability (min)	9.07	3.73	2.90	2.80
C1 (Nm)	1.08	1.08	1.08	1.08
C2 (Nm)	0.45	0.38	0.30	0.30
C3 (Nm)	1.70	1.56	1.51	1.48
C4 (Nm)	1.53	1.39	1.32	1.26
C5 (Nm)	2.20	1.93	1.86	1.78

Table 2: Mixolab properties of wheat flour treated with different levels of α-amylase enzyme.
[1]14% moisture content basis

The ash content in wheat flour has significance for milling. The ash content of wheat flour was 0.53%. Most flours will have ash content below 0.8%, cake flours can go as low as 0.5% (Al-Dmoor, 2013).

Some starch granules of flour are damaged during milling of wheat, and the degree of damage affects water absorption and dough mixing characteristics of the resulting flour. Desired level of damaged starch for cake making was < 7%, as higher percent of starch damage makes the starch more susceptible to enzyme attack that results in smaller cookies (Al-Dmoor, 2013). The damage starch of wheat flour was 6%.

The gluten content of wheat flour was 23.6% this indicated that the flour was within the low range, 18 – 23% (Pedersen *et al.,* 2004). The gluten index of wheat flour was 89.3%. Curik *et al.* (2001) reported that flours with the gluten index exceeding 95 are too strong and those with the index value less than 60 are too weak for bread production. The results indicated that the wheat flour generally represented medium strong flour. Similar results were reported by Hassan *et al.* (2014).

Rheological properties:

Mixolab properties

Mixolab properties of wheat flour treated with different levels of α-amylase enzyme are presented in Table (2). Mixolab performs mixing and heating to determine starch properties and α-amylase activity as well as protein quality (Koksel *et al.,* 2009). Water absorption was not affected by α-amylase enzyme levels. Development time decreased by treating wheat flour with α-amylase enzyme at 5 and 7.5 ppm levels whereas, at 10 ppm level development time did not affect. Dough stability decreased with increasing α-amylase enzyme levels.

The C1 and C2 values are related to protein quality whereas C3, C4 and C5 are related to the starch characteristics (Koksel *et al.,* 2009). The C1 values were not affected by α-amylase enzyme levels whereas C2 values were decreased by treating wheat flour with α-amylase enzyme. After the dough stability period a couple decrease is registered, represented by the dough mechanical soak, and explained by unfolding of the protein chain under the mechanical pressure force. Some changes of the protein during heating can be hidden by the modifying of the starch physico-chemical properties (Haros *et al.,* 2006).

Mixolab indices	Levels of α-amylase (ppm)			
	0	5	7.5	10
Water absorption index	8	8	8	8
Mixing index	2	1	1	1
Gluten index	4	4	3	3
Viscosity index	3	2	2	3
Amylolysis index	7	6	6	6
Retrogradation index	4	3	3	3

Table 3: Mixolab indices of wheat flour treated with different levels of α-amylase enzyme

The parameters C2 to C5 represent the end point of the corresponding mixing stages (Kahraman *et al.*, 2008). As the temperature increases, the quantity of water in the dough also increases, because the water released from the proteins which coagulate. The released water determines the swelling and gelling starch, which leads to an increase in the dough consistence. As the starch gelling power increases and the α-amylase activity decreases, the maximum consistency of the dough will be higher. The C3 values were decreased by increasing the level of α-amylase enzyme. These results agree well with those reported by Iuliana *et al.* (2008) and Hassan *et al.* (2014) they reported that the viscosity of dough (C3) decreased with increasing the level of protease enzyme which may be linked to a change in water distribution.

At constant temperature, the dough consistence decreases as the flour has an increased α-amylase activity. The C4 values were decreased by increasing the level of α-amylase enzyme. Similar results were reported in the Chopin Applications Laboratory (2009) and Hassan *et al.* (2014).

The last stage registered at the mixolab takes place during the dough cooling. The starch gelling and retrogradation begins, that leads to the starch increase in consistence. The starch retrogradation is marked with mixolab by C5 value. The C5 values decreased with increasing the level of α-amylase. Similar results were reported by Hassan *et al.* (2014) for treating wheat flour with protease enzyme.

Mixolab indices

The mixolab indices of wheat flour treated with different levels of α-amylase enzyme are shown in Table (3). The water absorption index was not affected by treating wheat flour with α-amylase enzyme. Mixing, amylolysis and retrogradation indices decreased by treating wheat flour with α-amylase enzyme. These results indicated that α-amylase activity was high in dough (Chopin Applications Laboratory, 2009). Iuliana *et al.* (2008) noticed that adding α-amylase allows decreasing the retrogradation. Viscosity index decreased by treating wheat flour with α-amylase enzyme until the 7.5 ppm level, however, at 10 ppm level viscosity index did not affect. Gluten index was not affected by treating wheat flour with 5 ppm α-amylase enzyme however, at higher levels (7.5-10 ppm) gluten index was decreased. Similar results were reported by Hassan *et al.* (2014) for treating wheat flour with protease enzyme.

Alveograph properties

Alveograph properties of wheat flour treated with different levels of α-amylase enzyme are presented in Table (4). The alveograph decides the gluten strength of dough by measuring the force required to blow and break a bubble of dough. The results comprise (P), (L) and (W) values (Al-Dmoor, 2013).

Alveograph properties	Levels of α-amylase (ppm)			
	0	5	7.5	10
P (mm)	167	120	106	105
L (mm)	31	38	36	40
P/L Ratio	5.4	3.2	2.9	2.6
W ($J \times 10^{-4}$)	229	188	149	169

Table 4: Alveograph properties of wheat flour treated with different levels of α-amylase enzyme. *P: The dough strength; L: The dough extensibility; P/L: The ratio between dough strength and extensibility; W: deformation energy of dough.*

Farinograph characteristics	Levels of α-amylase enzyme (ppm)			
	0	5	7.5	10
Water absorption (%)	62.4	62.4	62.4	62.4
Arrival time (min)	1.0	1.25	1.25	1.25
Dough stability (min)	2.0	1.7	1.5	1.5
Dough tolerance index (BU)[1]	100	110	110	120
Dough weakening (BU)	130	150	150	180
Degree of softening (BU)	90	100	120	120

Table 5: Farinograph characteristics of wheat flour treated with different levels of α-amylase enzyme. *[1]Barbender Units*

The (P) value and the (P/L) ratio were decreased by increasing the levels of α-amylase enzyme whereas the (L) value showed an opposite trend. Treating wheat flour with 7.5 and 10 ppm α-amylase enzyme reduced (P) value by 36.5% and 37.1% and (P/L) ratio by 46.3% and 51.9%, respectively. However, treating wheat flour with 7.5 and 10 ppm α-amylase increased (L) value by 16.1% and 29.0%, respectively. Similar results were reported by Hassan *et al*. (2014) for treating wheat flour with protease enzyme.

The deformation energy of dough (W) representing the energy necessary to inflate the dough bubble to the point of rupture decreased by treating wheat flour with α-amylase enzyme. Treating wheat flour with 7.5 and 10 ppm α-amylase enzyme reduced (W) values by 34.9% and 26.2%, respectively. The lowest (W) value was obtained by treating flour with 7.5 ppm α-amylase enzyme (149×10^{-4} J). This value indicated that the flour is more suitable for betifore making (Hassan *et al.*, 2014). Weak gluten flour with low (P) value (strength of gluten), long (L) value (extensibility) and (W) value ranged from 120 to 160 is preferred for cakes and other confectionery products. Strong gluten flour will have high P values and is preferred for breads, (Al-Dmoor, 2013).

Farinograph properties

The farinograph properties of wheat flour treated with different levels of α-amylase are presented in Table (5) and Figure (1). Water absorption was not affected by treating wheat flour with α-amylase enzyme. These results are supported by the results previously reported in Tables 2 and 3. Dough stability time was decreased by treating wheat flour with α-amylase. The reduction in dough stability varied from 15 to 25%. This suggests a reduction in the strength of the dough. The dough tolerance index, dough weakening and degree of softening values were increased by treating wheat flour with α-amylase. Inrani and Venkateseara Rao (2006) reported that dough prepared with 20 ppm α-amylase showed decreases in farinograph stability from 3.5 to 2.5 min when compared to the control. Khalil *et al.* (2000) reported that addition of 1% malt to wheat-cassava composite flours reduced the weakening values of dough however, malt addition at 2% levels or higher resulted in marked increase of dough weakening.

Sensory properties of betifores:

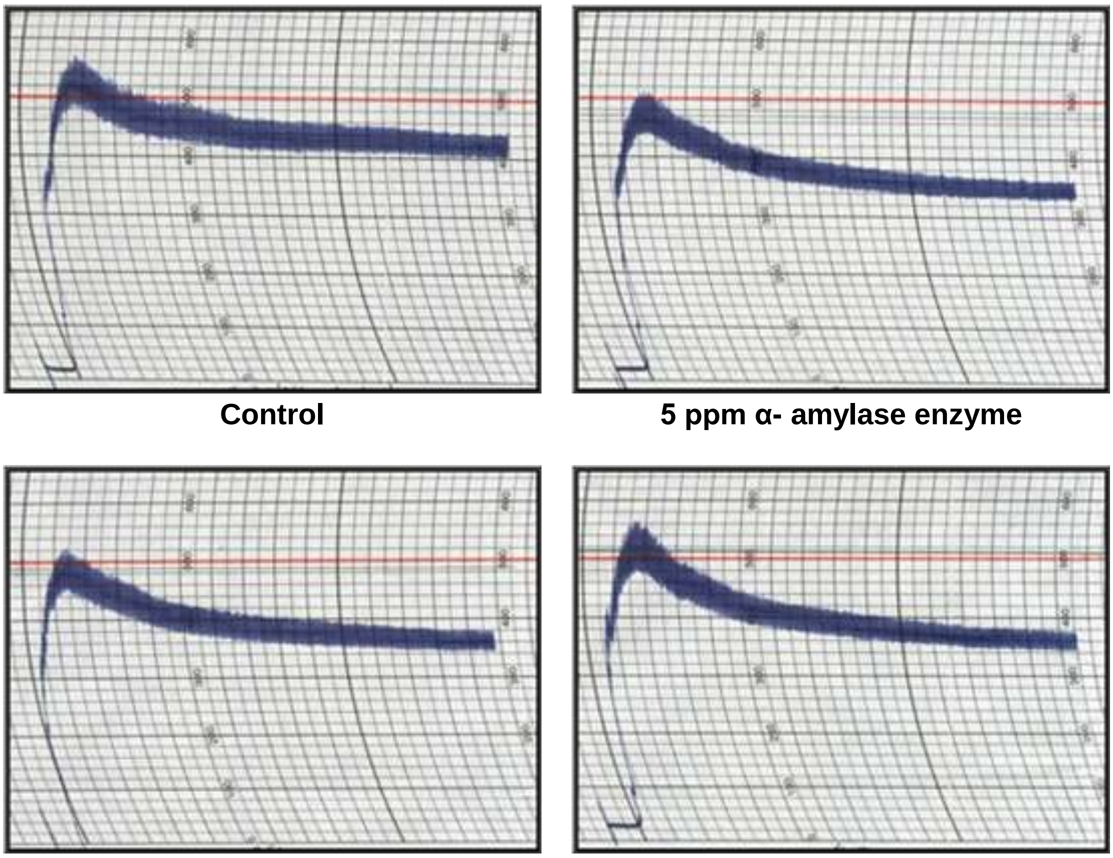
Data presented in Table (6) show the sensory properties of betifores prepared with different levels of α-amylase enzyme. Betifores prepared with α-amylase had higher ($p \leq 0.05$) rating scores than the control for all sensory properties. Betifores prepared with 7.5 ppm α-amylase had higher ($p \leq 0.05$) mean scores for all sensory properties compared to betifores prepared with 5 and 10 ppm α-amylase. These results are

Sensory properties	Levels of α-amylase (ppm)				
	0	5	7.5	10	LSD
Appearance	$5.89^c \pm 1.00$	$7.00^b \pm 0.00$	$9.00^a \pm 0.00$	$7.33^b \pm 0.58$	1.017
Texture	$5.17^d \pm 0.29$	$6.00^c \pm 0.00$	$8.67^a \pm 0.58$	$7.00^b \pm 0.00$	0.608
Crispness	$4.67^c \pm 0.58$	$6.33^b \pm 0.58$	$9.17^a \pm 0.00$	$7.00^b \pm 1.00$	1.245
Colour	$5.33^d \pm 0.29$	$6.83^c \pm 1.04$	$9.67^a \pm 0.29$	$8.00^b \pm 0.00$	1.053
Flavour	$6.67^c \pm 0.29$	$7.83^b \pm 0.58$	$9.17^a \pm 0.29$	$8.17^b \pm 0.29$	0.544
Mouth feel	$5.17^c \pm 0.29$	$7.17^b \pm 1.04$	$9.50^a \pm 0.50$	$8.00^b \pm 0.00$	1.121
Overall acceptability	$6.17^d \pm 0.29$	$7.17^c \pm 0.29$	$9.33^a \pm 0.58$	$8.00^b \pm 0.00$	0.666

Table 6: Sensory properties of betifores prepared with different levels of α-amylase enzyme. *Means in the same row with different letters are significantly different (p < 0.05).*

Physical properties	Control	α-amylase	LSD
Weight (g)	$5.08^a \pm 0.28$	$4.94^a \pm 0.18$	0.469
Width (cm)	$3.70^a \pm 0.10$	$3.67^a \pm 0.06$	0.156
Thickness (cm)	$0.75^a \pm 0.01$	$0.60^b \pm 0.01$	0.033
Spread ratio[1]	$4.93^b \pm 0.10$	$6.12^a \pm 0.04$	0.294
Spread factor	$100^b \pm 0.00$	$124.14^a \pm 1.97$	3.607

Table 7: Physical properties of betifores prepared with 7.5 ppm α-amylase enzyme.
[1]width/thickness. Means in the same row with different letters are significantly different ($p \leq 0.05$).



Control — 5 ppm α- amylase enzyme — 7.5 ppm α-amylase enzyme — 10 ppm α-amylase enzyme

Figure.1: Farinograms of wheat flour treated with different levels of α-amylase enzyme

supported by the results previously reported in Tables 2 and 4. These results agree well with those reported by Hassan *et al.* (2014) for betifores prepared with 25 ppm protease enzyme. Therefore, Betifores prepared with 7.5 ppm α-amylase was selected for physical properties study.

Physical properties of betifores

Data presented in Table (7) show the physical properties of betifores prepared with 7.5 ppm α-amylase enzyme. Betifores weight and width were not significantly ($p > 0.05$) affected by α-amylase enzyme. However, betifores thickness, spread ratio and spread factor were significantly ($p \leq 0.05$) affected by α-amylase enzyme. No significant ($p > 0.05$) differences were observed in weight and width between the betifores prepared with α-amylase and control. Betifores prepared with α-amylase had a lower ($p \leq 0.05$) thickness as compared to the control. Betifores prepared with α-amylase had higher ($p \leq 0.05$) spread ratio and spread factor than the control. The betifores spread ratio values were in the range of 4.93-6.12. These values are lower than those (7.00-9.73) reported by Ozturk *et al.* (2008). Cauvain and Young (2006) and Goesaert *et al.* (2009) reported that α-amylase can improve the gas-retention properties of fermented dough and reduce dough viscosity during starch gelatinization, with consequent improvements in product volume and softness. These results agree well with those reported by Hassan *et al.* (2014) for betifores prepared with 25 ppm protease enzyme.

CONCLUSION

The dough stability time, the resistance of the dough to deformation (P) and deformation energy of dough (W) were decreased by treating wheat flour with α-amylase enzyme. This suggests a reduction in the dough strength and the wheat flour become more suitable for betifores making.

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