EFFECT OF SOME MANUFACTURING FACTORS ON ACRYLAMIDE FORMATION IN DIFFERENT SOFT BREAD (FENO) FORMULAS

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ABSTRACT

This paper was carried out to investigate the effect of some conditions (baking temperature, baking time, dough pH, moisture content and dough fermentation time) on acrylamide formation in different formulas used for making soft bread (Feno). The principle components used to make the dough of soft bread were wheat flour extraction 72%, water, yeast, salt and baking improvers. The formulas tested in this study were; principle components only (NSM), principle components with powder milk (WM), principle components with sucrose (WS) and principle components with sucrose and powder milk (SM). The obtained results showed that increasing of baking temperature to 250°C, baking time to 25 min and dough pH to 8 led to markedly increase in acrylamide content in each formulas and after that decreased. The effect of baking time on acrylamide formation was somewhat higher than the effect of baking temperature. Increasing of dough moisture and fermentation time led to decrease in acrylamide content of each formulas. The highest level of acrylamide was formed in the formula contained sugar with milk followed by sugar alone while the lowest acrylamide level was found in the formula without sugar and milk under each studied factor. As organoleptic evaluation, baking at 220°C for 15 min was enough to obtain acceptable soft bread.

INTRODUCTION

Acrylamide is a small organic molecule. Its principle use is to manufacture the polyacrylamide, which is a gel like substance used in a variety of purposes such as water treatment, water purification, soil conditioning and food packaging (Allan, 2002). The polyacrylamide is non-toxic. However, acrylamide, before it is turned into polyacrylamide, is a carcinogen and a neurotoxin (Bull *et al.*, 1984). The body itself converts ingested acrylamide to a carcinogenic by-product when attempting to destroy it and this by-product binds and mutates DNA (Paulsson *et al.*, 2003). According to Food and

Agriculture Organization FAO, (2002) the toxicological studies on acrylamide revealed that it causes DNA damage and at high doses neurological and reproductive effects have been observed. Allan, (2002) reported that acrylamide is considered to be genotoxic *in vivo* and carcinogenic in experimental animals. The acute oral LD_{50} for acrylamide in rats is 107-203 mg/kg body weight (Lindsay, 2002). No observed effect level for acrylamide was reported to be up to 2 mg/kg rat body weight per day {World Health Organization (WHO) & Food and Agriculture Organization (FAO) 2002}.

Acrylamide is not being added to food, it is formed during heating of starchy foods at high temperatures. Chemical analysis has shown that acrylamide is presented in a large number of foods and its level differ widely within each food group analyzed, despite it has so far not been detected in raw foods. For example, raw potato has negligible levels of acrylamide (< 0.030 mg/kg of potato) but, if you make potato chips, the level of acrylamide can skyrocket to 1.2 mg/kg of potato chips (Swedish National Food Administration, 2002). Acrylamide does not only form in industrially manufactured foods, but also foods prepared at home are concerned (Glese 2002)

Two different groups studied to clear how acrylamide forms in the foods and discovered that the amino acid asparagines, a building block of proteins, reacts with sugars and starches at high temperature to form the acrylamide. It forms in a process known as a Millard reaction, which is out of the scope of this review (Mottram *et al.*, 2002). Interestingly, they found that the acrylamide formed in significant quantities when the food was heated at about 180°C. Potatoes contain high levels of amino acid asparagines and starch, this may explain why they have the highest acrylamide levels when compared to other chips such as corn (Stadler *et al.*, 2002).

The effect of backing conditions on acrylamide formation in commercial white bread and toast is reported. Levels of acrylamide were found to be relatively low under normal processing conditions; however, high levels could be expected in smaller baked products with more exposed surface area. The highest content of acrylamide in bread was generated in the outer surface, especially the base, and crust. The color is a good indicator of acrylamide levels (CIAA, 2004). The main objectives of this study were to determine the levels of acrylamide in soft bread (Feno) and to investigate the effect of different processing factors on these levels.

MATERIALS AND METHODS

The soft bread ingredients were obtained from local market at Kafr El-Sheikh City, Egypt.

Preparation of soft bread:

The technological procedure that used for soft bread preparation was carried out at home using the method described by Hani *et al.*, (2004) as the following formulas:

Component	Blends			
	WM (%)	WS (%)	SM (%)	NSM (%)
Wheat flour (extraction rate 72%)	60	62	58	64
Water***	30	30	30	30
Sucrose	-	2	2	-
Milk powder	4	-	4	-
Yeast	4	4	4	4
Salt	1.8	1.8	1.8	1.8
Improvers **	0.2	0.2	0.2	0.2

*WM = blend with milk, WS = with adding sugar, SM = with adding sugar and milk, NSM = without adding sugar and milk.

** Improvers = α-amylase, emulsifying agent and ascorbic acid (1:1:1).

***To investigate the effect of dough moisture content on acrylamide formation, another contents of water were added in individual experiment.

The baking oven was supported with digital thermometer and stopwatch to control the baking temperature and time.

Chemical analysis:

Moisture was determined as described in the (A.O.A.C., 1990). The moisture was controlled by water addition to the flour. A 100 g sample of the dough used to prepare the bread was homogenized with 125 ml of distilled water in a blender to form a slurry to measure the dough pH. The pH was measured in the slurry at room temperature using a pH meter. The pH was controlled in the dough with buffer solution (buffer citrate) (Kristel *et al.*, 2006). The acrylamide in samples was extracted using method of (Biedermann *et al.*, 2002). The acrylamide was determined using Gas Chromatography and Mass Spectrophotometer (GC/MS).

Sensory analysis:

In this experiment, the dough blends (WS, WM, SM and NSM) of soft bread were prepared as maintained in table 1 then

fermented for 120 min. The fermented blends were baked at 220^{0}C for various baking time (15, 20, 25 and 30 min). Organoleptic evaluation of soft bread was performed by a semi-trained panel of judges using nine-point hedonic-scale ratings for color, taste, aroma, texture and overall acceptability with 9 being the highest score (extremely liked) and 1 being the lowest score (extremely disliked) (Watts *et al* 1989). This analysis was carried out on soft bread blends baked at 220^{0}C for different baking time (15, 20, 25 and 30 min) to known a low baking temperature and time for production acceptable soft bread as well as known effect of that on acrylamide formation during baking process.

RESULTS AND DISCUSSION

Effect of baking temperature on acrylamide formation:

The correlation between acrylamide formations and different baking temperatures used in baking of soft bread formulas baked for 20 min was shown in Figure1. It was observed that acrylamide content increased markedly in each formula with increasing of baking temperature until 250°C after that it decreased. The increasing of acrylamide content with the increasing of temperature due to the Mailllard reaction, which is the major route to formation of acrylamide in foods, is favoured by conditions of high temperature (Stadler *et al*., 2002). The decreasing occurred in acrylamide content, at baking temperature more than 250^{0}C, may be attributed to the acrylamide could be easily polymerized to give polyacrylamide (Allan, 2002). The highest level of acrylamide was found in formula prepared with adding sugar and milk (SM) followed by that with sugar (WS) followed by that with milk (WM) but the lowest level was in case of formula without adding sugar and milk (NSM) where the levels of acrylamide at 250°C were 415, 308, 240 and 121 µg/kg in mentioned formulas, respectively. These results are in agreement with these obtained by Gilcrest, (2003) who found that the acrylamide arises in food when amino acid asparagines was heated with sugars such as glucose. The cooking processes that produced acrylamide in foods are baking, frying, grilling and toasting, or any cooking method at temperatures greater than 120°C.

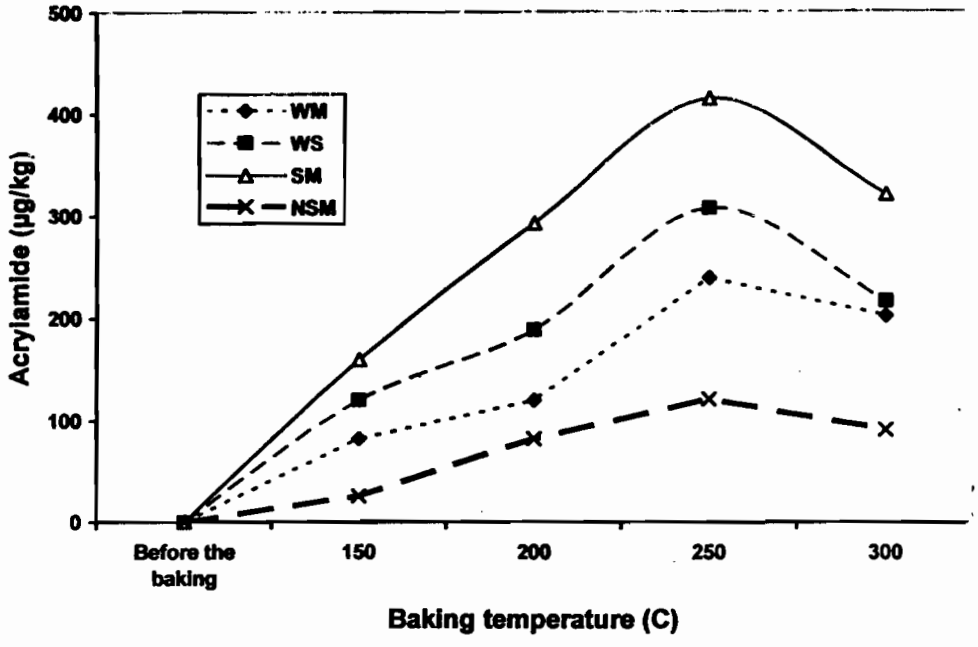

Fig. (1): Effect of baking temperature for 20 min on acrylamide formation (µg/kg) in some formulas of soft bread baked.

Effect of baking time on acrylamide formation:

The effect of baking time on acrylamide formation in different soft bread formulas baked at 220°C was shown in Fig. 2. Increasing of baking time led to high increase in acrylamide level in each formulas until 25 min after that it decreased with increasing of baking time up to 30 min. The recorded levels of acrylamide were 280, 386, 460 and 143 µg/kg after 25 min. at 220°C in formulas named WM, WS, SM and NSM, respectively. These results were consistent with that reported by (Hani *et al.*, 2004 and Eden *et al.*, 2002) they reported that the heat intensity (time and temperature) and all factors enhancing browning and crust formation are the major cause of acrylamide formation in all types of food. The reducing occurred in acrylamide level in the end of baking period may be due to the long time at high temperature causes acrylamide polymerization.

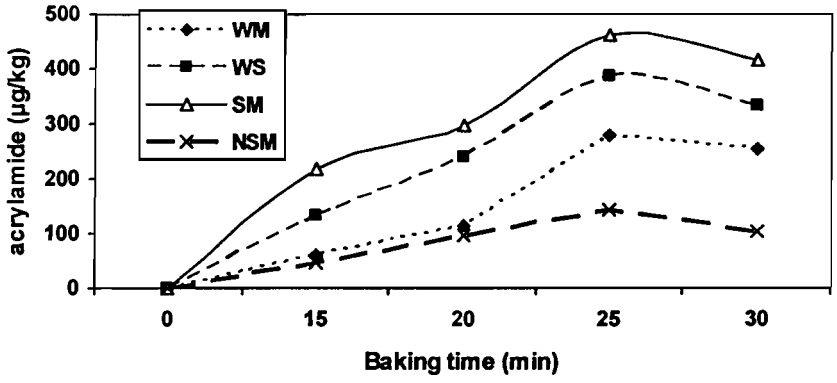

Fig. (2): Effect of baking time on acrylamide formation (µg/kg) in different formulas of soft bread baked at 220° C .

From the results in figure 1 and figure 2, it could be noticed that the effect of baking temperature (for same time 20 min) on acrylamide formation was lower than the effect of baking time (at same temperature 220^{0}C) in formulas WS and NSM, where the increase of baking temperature from 200 to 250^{0}C led to increase of acrylamide content about 1.6 and 1.5 times, while the increase of baking time from 15 to 20 min led to increase of acrylamide content about 1.8 and 2 times respectively. For the formulas WM and SM the effect of increase the baking temperature on acrylamide formation was similar with the effect of baking time, where it was about 2 and 1.4 times, respectively.

Effect of dough pH on acrylamide formation:

Figure 3 shows acrylamide content in different formulas of soft bread baked at 220°C for 20 min at different dough pH values. Increasing pH value of the dough led to increase in acrylamide formation in all formulas till pH 8 then decreased. The formed acrylamide contents were 186, 212, 301 and 90 µg/kg in WM, WS, SM and NSM, respectively when using of dough pH 7 and baked at 220°C for 20 min. Such results were in harmony with these found by (Fredriksson *et al.,* 2004 and Hani *et al.,* 2004) they found that the optimum pH for acrylamide formation is around 7.

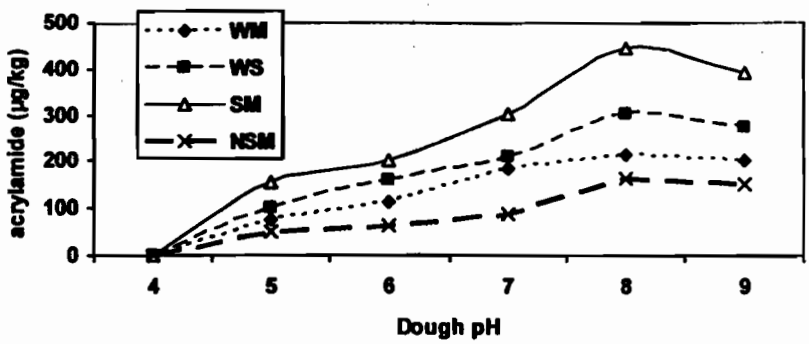

Fig. (3): Effect of dough pH on acrylamide formation (µg/kg) in different formulas of soft bread baked at 220° C for 20 min.

Effect of dough moisture on acrylamide formation:

In this experiment, various dough formulas of soft bread different in their moisture content were prepared to investigate the effect of dough moisture content on acrylamide formation. The results were summarized in Fig. 4. From the results presented in this fig., it could be observed that the formed acrylamide content decreased with increasing the moisture percentage in all dough formulas. Whereas the acrylamide levels in soft bread baked at 220°C for 20 min. were 331, 411, 503 and 235 µg/kg when the dough moisture content was 35% and they decreased to 34, 136, 297 and 38 µg/kg at moisture content of 55% in WM, WS, SM and NSM, respectively. This result can be explained by the high moisture content reduces the effect of high temperature. This finding was in agreement with that obtained by (Pedreschi *et al.,* 2006) who reported that samples with higher moisture content have lower acrylamide level. In addition, Taeymans *et al.*, (2004) said that studies on the mechanism of acrylamide formation within the Maillard reaction have shown that acrylamide formation is favoured in conditions of low moisture content.

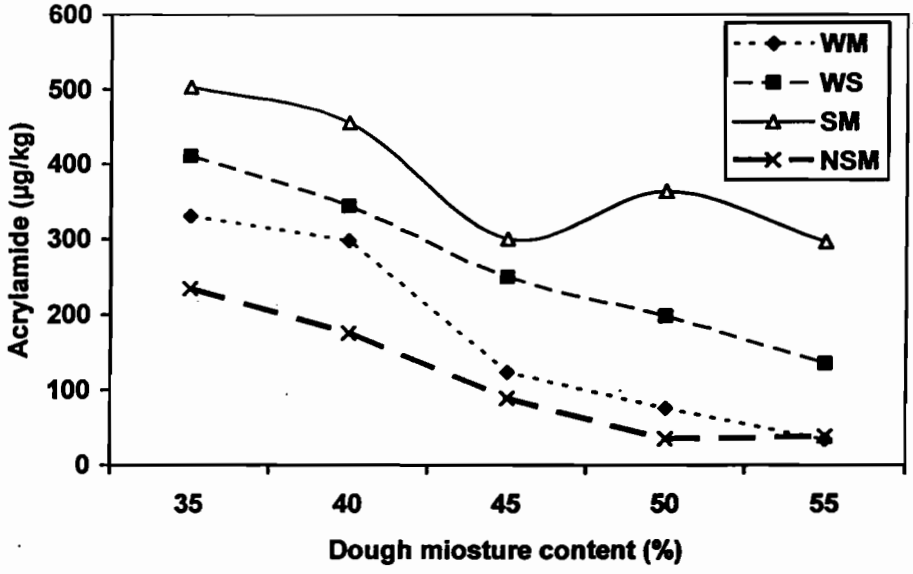

Fig. (4): Effect of dough moisture content on acrylamide formation (µg/kg) in some formulas of soft bread baked at 220°C for 20 min.

Effect of dough fermentation time on acrylamide formation:

Effect of dough fermentation time on acrylamide formation in different soft bread formulas baked at 220° C for 20 min was shown in Fig.5. The acrylamide contents were somewhat increased with increasing of fermentation time in case of formulas SM and WM up to 30 min then steadily decreased. On the other hand, in case of WS formula the acrylamide content linearly decreased with increasing of fermentation time where it was 390 µg/kg after 30 min and decreased to 280 µg/kg after 120 min. In case of NSM formula the acrylamide level slightly increased with increasing of fermentation time since it was 22 µg/kg after 30 min and reached to 97 µg/kg after 120 min then it decreased to 22 µg/kg after 180 min. Fredriksson *et al.* (2004) found that extensive fermentation with yeast may be one possible way to reduce acrylamide content in bread because the free asparagines is consumed by yeast and its content reduced in the fermented dough.

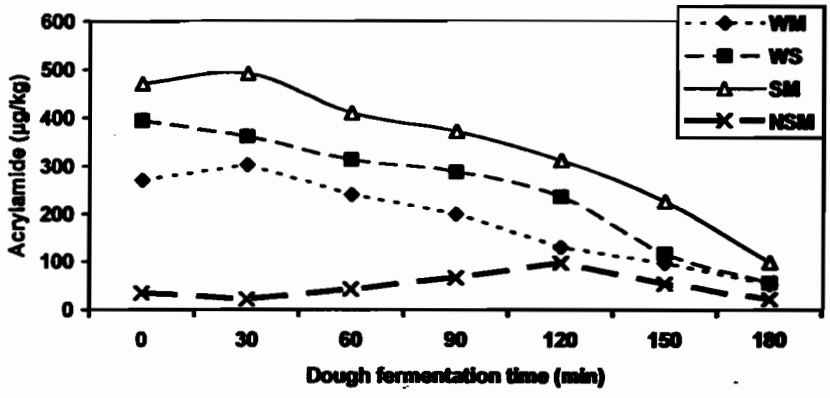

Fig. (5): Effect of fermentation time (min.) on acrylamide formation (µg/kg) in some formulas of soft bread baked at 220° C for 20 min.

Organoleptic evaluation of soft bread:

The organoleptic evaluation of soft bread was performed and the mean of results was recorded in table 2. The results showed that the scores of soft bread characters increased with increasing of baking time from 15 to 20 min after that it decreased. The samples baked for 15 min were, in the mean, liked while the samples baked for 20 min were, in the mean, extremely liked. Unfortunately, the acrylamide contents increased about 2 times in formulas WM, WS and NSM and 1.4 times in formula SM when used baking time 20 min comparing with these baked for 15 min. The results indicated that the soft bread could be baked at 220°C for 15 min with liked acceptability, except the color which was somewhat liked (whitish) and in this case, the low content of acrylamide formed especially in formulas WM and NSM. Stadler *et al.*, (2002) reported that acrylamide in foods is closely linked to the formation of desirable characteristics such as flavor and color. Because the Mailleard reaction is favoured by conditions of high temperature, resulting in the flavors and brown color in roast, baked and fried foods.

Table 2: The relation between organoleptic analysis and acrylamide formed in soft bread formulas baked at 220°C for different baking time.

Blends	Baking time	Color	Taste	Aroma	Texture	Overall acceptability	Acrylamide content*(µg/kg)
WM	15	6	7	7	8	7	61
	20	8	9	9	9	9	116
	25	9	9	9	8	8	280
	30	7	8	8	7	7	254
WS	15	6	7	7	8	7	133
	20	9	9	9	8	9	240
	25	8	8	9	7	8	386
	30	7	8	8	7	7	331
SM	15	7	8	7	9	8	218
	20	9	9	9	9	9	298
	25	8	7	8	7	7	460
	30	6	7	7	6	6	413
NSM	15	6	7	7	7	7	48
	20	8	9	9	9	9	95
	25	8	8	8	7	8	143
	30	7	7	7	6	7	102

*Acrylamide content values were taken from data in figure 2.

Conclusion

Addition of sugar and milk to the dough of soft bread led to a high increasing of acrylamide levels. Addition of sugar alone or milk alone had a middle effect on acrylamide formation. The lowest acrylamide level was in the formula without sugar and milk under all studied conditions. The baking temperature, baking time, dough pH value, dough moisture content and fermentation time have a great effect on the acrylamide formation in soft bread. The effect of baking time on acrylamide formation was somewhat higher than the effect of baking temperature. The increasing of baking temperature, baking time and dough pH led to high increase of acrylamide formation until 250°C, 25 min and pH 8, respectively after such values, the acrylamide level decreased. The increasing of dough moisture content and fermentation time led to reducing of acrylamide levels in all tested formulas of soft bread. As organoleptic evaluation, baking at 220°C for 15 min was enough to obtain acceptable soft bread and in this case, could be reduced the formation of acrylamide approximately to the half comparing with the baking for 20 min at the same baking temperature.

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الملخص العربى

تأثير بعض عوامل التصنيع على تكوين الأكريلاميد في خلطات مختلفة للخبز الفينو

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بدراسة تأثير بعض عوامل التصنيع (درجة حرارة الخبيرو ووقت الخبيز ودرجة pH العجين والنسبة المئوية لرطوبة العجين ووقت تخمير العجين) على محتوى الاكريلاميد فى خلطات مختلفة من الخبز الفينو (مضاف إليها لبن WM مضاف إليها سكر WS و مضاف إليها سكر و لبن SM وبدون سكر و بدون لبن NSM).

النتائج المتحصل عليها أوضحت أنه مع زيادة درجة حرارة الخبيز إلي 250 درجة مئوية ووقت الخبيز إلي ٢٥ دقيقة ودرجة pH للعجين إلي ٨ زادت كمية الأكريلاميد زيادة واضحة ثم انخفضت كمية الأكريلاميد بعد ذلك مع زيادة هذه العوامل وذلك في كل الخلطات.

علي الجانب الأخر مع زيادة نسبة رطوبة العجين إلي٤٥% وزيادة وقت التخمير إنخفض محتوى الخبز الفينو من الاكريلاميد فى كل المعاملات. اضافة السكر مع اللبن أدت الى زيادة معنوية فى محتوى العينات من الاكريلاميد يليها اضافة السكر علي حدة ثم إضافة اللبن على حدة. أقل محتوي من الأكريلاميد كانت في الخلطة التي لم يضف إليها سكر و لبن. إذن نستخلص من هذه الدراسة أن درجة حرارة الخبيز ووقت الخبيز ورقم ال pH ونسبة الرطوبة ووقت التخمير لها تأثير شديد علي تكوين الأكريلاميد في خلطات الخبز الفينو. كذلك إضافة اللبن والسكر أو أحدهما إلي مكونات العجينة يؤدي إلي زيادة ملحوظة في محتوي الخبز الناتج من الأكريلاميد.

لذا ننصح بتصنيع الخبز الفينو علي درجة حرارة لا تزيد عن ٢٢٠ درجة مئوية ووقت خبيز لا يزيد عن ١٥ دقيقة وذلك لخفض كمية الأكريلاميد. كذلك يجب زيادة نسبة الرطوبة ووقت التخمير بقدر الإمكان. بالإضافة إلي ذلك يراعي عدم إضافة اللبن مع السكر إلي المخبوزات خاصة التي تحتاج درجة حرارة عالية ووقت طويل داخل الفرن.