

## Investigation of Acrylamide Levels in Selected Fried Foods in Syria

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### ABSTRACT

This research aims at investigating the acrylamide content in potato chips, fried potato and falafel from local markets and studying the effects of different frying temperatures (130, 150 and 170°C), frying oils (sunflower, cotton and olive oils) and shape of potato pieces (fingers and slices) on the acrylamide content in fried potato processed under controlled conditions. As a result, the average values of acrylamide levels in potato chips, fried potato and falafel samples were 469.33, 56.46 and 153.78 µg/kg, respectively. In the raw potato prepared and fried under controlled conditions, the acrylamide levels (0, 106.62 and 229±5.75 µg/kg) were significantly different ( $P<0.5$ ) at different frying temperatures (130,150 and170°C), respectively and (106.62 and 526.58 µg/kg) for two shapes of pieces (fingers and slices), respectively. The acrylamide level for olive oil frying was nearly undetectable (11.97 µg/kg) and significantly different from both cotton oil and sunflower oil (106.62 and 91.84 µg/kg). respectively.

**Keywords:** Acrylamide, Potato chips, Fried potato, Falafel, Strata X-C cartridge – HPLC-UV.

### INTRODUCTION

Acrylamide was primarily produced as polyacrylamides for industrial purposes such as water and waste-water treatment, crude oil production, paper and paper pulp processing, mineral processing, concrete processing, soil treatment and additives for cosmetics (Eriksson, 2005). According to the International Agency for Research on Cancer (IARC, 1994), acrylamide is defined as a compound with the potential to cause a spectrum of toxic effects. WHO (1996) identified

guidelines for drinking-water quality, with a guideline value (the concentration representing the tolerable risk to the health of the consumer over a lifetime of consumption) of 0.5 microgram per liter in drinking-water. In April 2002, the Swedish National Food Administration and Stockholm University announced their findings that acrylamide, a toxic and potentially cancer-causing chemical, is formed in many types of food prepared/cooked at high temperatures (FAO/WHO, 2002). FDA (2002) announced that potato chips and French fries were found to contain high levels of acrylamide (117-2762, 20-1325 µg/kg), respectively.

Acrylamide ( $\text{CH}_2=\text{CH}-\text{CO}-\text{NH}_2$ ; 2-propenamide) is a white crystalline solid and its solubility in water is extremely high (Eriksson, 2005). The major pathway of formation seems to be the Maillard reaction, in particular in the presence of the amino acid asparagine, which

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directly delivers the backbone of the acrylamide molecule. Reducing sugars, in the case of potatoes mainly glucose and fructose, are required as precursors for the conversion of asparagine into acrylamide (De Vleeschouwer et al., 2008).

It is known that the formation of acrylamide in foods is affected by various factors such as pH, reactant concentrations and the ratio between reactants, temperature and time of processing (De Vleeschouwer et al., 2008). It is well-known that potato products, which are the predominant source of acrylamide exposure, are particularly rich in free asparagine. The chemical composition of the potato is, however, influenced by different factors, such as the variety, the use of fertilizers, the climate, but the most important of all by the storage conditions applied. Storage below 8°C leads to the accumulation of sugars (“low temperature sweetening”), while the level of asparagine does not vary significantly with storage temperature. Consequently, this leads to potatoes with a wide range of sugar concentrations and thus of potatoes with highly variable sugar–asparagine ratios (De Vleeschouwer et al., 2008). Also, protein was effective in decreasing the formation of acrylamide because amino acids (concentration at 35 mmol/kg) other than asparagine, i.e. (glycine, alanine, lysine, glutamine or glutamic acid) competed in reaction with reducing sugars and consequently reduced the amount of formed acrylamide (Eriksson, 2005).

In animals, acrylamide is neurotoxic, mutagenic and carcinogenic, and disturbs fetal development and reproductive functions. Fetotoxic effects of acrylamide include neurotoxicity and other effects such as reduced fetal body weight, decreased number of offspring and infertility. While also neurotoxic in humans,

epidemiologic studies on acrylamide have so far not indicated any carcinogenic effects. However, on the basis of animal studies and genotoxicity (Annola et al., 2008), acrylamide induces mainly mutations through metabolism to glycidamide, while glycidamide is clearly, but moderately mutagenic, the genotoxicity of acrylamide is different: it has been shown to be mainly clastogenic in human cells. Glycidamide can also induce apoptosis and inhibit DNA repair. Glycidamide reacts with DNA forming DNA adducts mainly with N7-guanine and N1 and N3 of adenine (Annola et al., 2008).

Single oral doses produced acute toxic effects only at doses of >100mg/kg of body weight, and reported median lethal doses (LD50s) are generally >150 mg/kg of body weight (JECFA, 2006).

Fried foods are consumed widely all over the world. In Syria, fried potato and falafel are prevailing foods and almost consumed every day. Potato chips are considered one of the favorite foods for children. Regarding the importance of acrylamide formation in these groups of food, this research, therefore, aims at investigating the acrylamide content in potato chips, fried potato and falafel, purchased from local markets in Syria and studying the effects of some factors (frying temperature, shape of potato pieces and type of frying oil) on acrylamide content in fried potato prepared in agriculture collage laboratory.

## MATERIALS AND METHODS

### Food Sample Collection

Twenty samples of locally produced foods that represent two groups of food, fried potato and falafel, were collected from restaurants in Damascus. Ten samples of potato chips were collected from the local markets, the samples were produced in local companies.

Details of the pre-purchase information and processing parameters were not available.

### **Effect of Frying Using Different Temperatures, Frying Oils and Shapes of Potato Pieces on Acrylamide Content of Fried Potato**

Raw potato was purchased from the local market. The samples were prepared and fried under certain conditions. Every sample of 150g of peeled and cut potato was fried in 600ml of frying oil, where the depth of the frying oil was 1.5cm and the frying period was 5 minutes.

The studied variables were :

- Temperature: 130, 150 and 170°C, with considering that 150°C was the control.
- Type of oil: sunflower, cotton and olive oils, with considering that sunflower oil was the control.
- Shape of potato pieces: 5×1×1cm fingers, circular slices with 2-2.5cm radial and 0.5-1mm thickness, with considering that the shape of fingers was the control.

Each time, only one variable was changed and the others were kept at the control.

### **Measurement of Acrylamide**

#### **Sample Preparation and Extraction**

Food samples were prepared and analyzed using Phenomenex method (Peng et al.,2003) which depends on solid phase extraction with a simple modification including repeating centrifugation in step 4 to avoid the presence of fat traces in the final extracted solution of the sample prepared to HPLC analysis.

- 1- 10g of ground sample was added to 50 ml of distilled water, then mixed for 20 minutes.
- 2- The decanted solution was centrifuged at 10,000

rpm for 30min at 4°C.

3- The aqueous phase was extracted from centrifuged tubes by 1ml syringe.

4- The mentioned aqueous phase was centrifuged at 10,000 rpm for 30 min.

5- The aqueous phase was applied to a conditioned Strata-X-C cartridge at a flow rate of less than 0.5ml/min.

6- The cartridge sorbent (after loading with extracted sample) was dried for 30 sec under vacuum (10–12 in. Hg).

7- Ultra pure water (1 ml) was used to elute acrylamide from cartridge sorbent at a flow rate of less than 0.5 ml/min; the eluent was collected in a sample vial, and any residual water was drawn from the sorbent by applying full vacuum.

8- The eluent was then ready for HPLC -UV analysis.

To condition the cartridge: 2 ×1 ml methanol was applied, followed by 2×1 ml water to a strata-X-C tube at a flow rate of 1 ml/min.

Cartridge description: strata X-C with model 8B-S029-ECH and 100mg/6ml capacity.

### **The HPLC Following Conditions**

The column was Inertsil ODS-35µm, 4.6×150mm, 3KI85001, and the mobile phase was 5% CH<sub>3</sub>OH + 95% H<sub>2</sub>O with a flow rate of 0.9ml/min. The injection volume was 25µl and the wave length of UV detection was 210nm (Peng et al.,2003).

### **Calibration Curve of Acrylamide**

The calibration curve was prepared for acrylamide using different concentrations and the respective peak areas: (0.2, 0.4, 0.6 and 0.8 ppm) or (0.5, 1, 1.5 and 2 ppm).

### Determination of % Recovery

A sample of potato chips was disintegrated, homogenized and 10g weighed in two flasks . 50ml of internal standard of acrylamide (0.04ppm) was added to one flask (spike) and 50ml distilled water (blank) was added to the second one. Extraction and analysis were applied on two parts according to the method mentioned above, considering that 15ml of extracted solution was applied on the cartridge . The results were 1.326 and 0.833ppm in the two vials of spike and blank respectively. For explanation, the amount of acrylamide in 50ml of internal standard(IST)with 0.04ppm concentration equals  $50 \times 0.04 = 2 \mu\text{g AA}$  → the amount of acrylamide in 15ml (IST)=  $2 \times 15 / 50 = 0.6 \mu\text{g}$  which was due to the added value.

Recovery =  $(\text{spike-blank}) \times 100 / \text{added value} = (1.326 - 0.833) \times 100 / 0.6 = 82.167\%$ .

### STATISTICAL ANALYSIS

Values shown represent the means  $\pm$  of experiments. Data were considered significantly different ( $P < 0.05$ ) in variables among samples using SPSS 0.7 program.

### RESULTS AND DISCUSSION

Acrylamide contents in the potato chips , fried potato and falafel samples are shown in Tables 1, 2 and 3, respectively. As can be seen from Table 1, the results show that acrylamide contents in the ten samples of potato chips ranged between  $112.115 \pm 10$  and  $1113.585 \pm 10 \mu\text{g/kg}$ . Acrylamide contents in potato chips were reported 752 and 117- 2762  $\mu\text{g/kg}$  by JECFA (2006) and FDA (2002), respectively. The differences in the figures could be attributed to the parameters: a) Agricultural conditions of raw potato (fertilizer, variety and climate) which affect the chemical structure of these products,

especially reducing sugars and asparagine. (Amrein, 2005; De Vleeschouwer et al., 2008). b) Storage conditions of the raw material: storing potato at a temperature below  $8 \text{ }^\circ\text{C}$  increases reducing sugar content. (Amrein, 2005; De Vleeschouwer et al., 2008). c) Treatments before frying: blanching, immersing in citric acid, sodium ascorbate or sodium pyrophosphate solution (Pedreschi, 2005; Eriksson, 2005). d) Processing conditions: heating temperature and time (Amrein, 2005; Eriksson, 2005; Pedreschi et al., 2005; De Vleeschouwer et al., 2008).

**Table 1: Acrylamide content in potato chips.**

<i>Sample</i>	<i>Acrylamide content <math>\mu\text{g/kg}</math> (mean)</i>
1	$1107.5 \pm 12.17$
2	$1113.58 \pm 8.51$
3	$564.31 \pm 43.48$
4	$229.54 \pm 25.56$
5	$322.44 \pm 29.76$
6	$399.37 \pm 12.83$
7	$450.44 \pm 24.07$
8	$189.93 \pm 10$
9	$112.11 \pm 9.17$
10	$204.03 \pm 4.99$

Each value represents three replicates.

Table 2 shows that the level of acrylamide in fried potato obtained from markets ranged from an undetectable value to  $176.53 \pm 10 \mu\text{g/kg}$ . As a matter of fact, during frying, the formation of acrylamide occurs only in the dry surface region of fried potato pieces where temperature may reach the temperature of oil frying ( $150\text{-}190^\circ\text{C}$ ), while the amount of acrylamide is undetectable in the core region where the temperature doesn't exceed ( $103\text{-}104^\circ\text{C}$ ) ( Gökmen et al., 2006). As

a result of the decrease in the surface region weight in comparison with the core region in fried potato pieces, the levels of acrylamide in fried potato were much lower than those in potato chips. It is known that the acrylamide level in French fries was 334 µg/kg (JECFA, 2006) and 20-1325 µg/kg (FDA, 2002). The variation that was noticed in Table 2 and other studies could be attributed to the agricultural practices and storage conditions and the treatments before frying and the processing conditions.

**Table 2: Acrylamide content in fried potato samples.**

Sample	Acrylamide content µg/kg (mean)
1	87.01 ± 7.64
2	176.53 ± 13.76
3	109.29 ± 11.75
4	38.74 ± 6.33
5	43.76 ± 10.45
6	31.39 ± 3.9
7	29.125 ± 1.83
8	ND
9	42.73 ± 0.57
10	6.89 ± 11.94

Each value represents three replicates.

Table 3 illustrates that variation in the acrylamide content in falafel samples was high. It ranged from an undetectable value to 439 µg/kg. Al-Dmooor et al. (2004) showed that acrylamide level in Jordanian falafel reached 3500 µg/kg. The reason for the differences could be attributed to the ingredients of mixture used in preparing and processing falafel, which may be made of chick peas and bread. The level, of acrylamide depends on the proportion of carbohydrate which increases the

level, while protein content enhances (decreases) it. Also, the ingredients may contain citric acid or any other material such as sodium bicarbonate which has a role in changing the pH that affects the acrylamide level (Eriksson, 2005).

**Table 3: Acrylamide content in falafel samples.**

Sample	Acrylamide content µg/kg (mean)
1	312.41 ± 5.36
2	ND
3	439.06 ± 20.70
4	320.35 ± 5.62
5	2.80 ± 4.86
6	55.34 ± 4.90
7	110.1 ± 3.00
8	110.63 ± 2.39
9	ND
10	187.08 ± 1.66

Each value represents three replicates.

Regarding the study of the effect of some factors (shape of pieces, frying temperature, kind of vegetable frying oil) on acrylamide content in fried potato, results are shown in Table 4.

Table 4 showed that the acrylamide content in the potato fingers was 106.62µg/kg; while it was much higher in the potato slices (526.58 µg/kg). This means: the higher the proportion of pieces' surface to weight, the higher the acrylamide content. For interpretation, the temperature of surface pieces equals the temperature of frying which is suitable for acrylamide formation, whereas the temperature of the internal part which represents the most of the weight of pieces remains at 100-104°C during frying and the acrylamide content is undetectable (Gertz et al., 2003; Gökmen et al., 2006).

As noticed from Table 4, the acrylamide contents in fried potato at different frying temperatures (130, 150, 170°C) were (0, 106.62, 229.01 µg/kg), respectively. These results were in agreement with the results of Pedreschi et al. (2005) in that the average of acrylamide content increased 58 times as the frying temperature increased from 120 to 180°C during frying potato slices. In addition, the initial rate of acrylamide formation increases with increasing temperature, from 120 to 200°C, because of increasing the activation energy (De Vleeschouwer et al., 2008). Besides, excessive research on acrylamide formation emphasized that acrylamide formation needs temperatures above 120°C (Gertz et al., 2003; Eriksson, 2005; Amrein, 2005).

Table 4 shows also that the acrylamide level was nearly undetectable with olive oil frying, while it was 106.62 and 91.84 µg/kg with sunflower frying and cotton oil frying, respectively without significant differences between the last two treatments. For demonstration, virgin olive oil contains a high concentration of antioxidants like ortho-diphenolic compounds which are not degraded during frying, and these components are efficiently able to inhibit acrylamide formation in crisps from mild to moderate frying conditions (Napolitano et al., 2008). In addition, olive oil has a high proportion of oleic acid

(monounsaturated acid), while sunflower oil and cotton oil have polyunsaturated fatty acids which produce acrolein during enzymatic and nonenzymatic maturation caused by lipid oxidation processes. In oil and fat, an alternative pathway for the formation of acrylamide through acrolein has been proposed (Eriksson, 2005). On the other hand, acrylamide formation is not significantly influenced by oil oxidation or the presence of any of the hydrolysis products (Mestdagh et al., 2007).

### CONCLUSION

Acrylamide detection was achieved in potato chips, fried potato and falafel samples. It was found that acrylamide level was the highest in potato chips, while it was much lower in fried potato and falafel. Also, a study of the effect of frying temperature, shape of potato pieces and kind of frying oil on the acrylamide content in fried potato was carried out. It was found that the acrylamide level increased significantly by increasing frying temperature from 130 to 170°C, and the acrylamide level was much higher in the slices of fried potato than in fingers. Furthermore, it was shown that the acrylamide level was extremely low in potato fried by using olive oil in comparison with the other oils. The latest result needs more research due to its importance for human health.

**Table 4: Estimated acrylamide levels at variable factors (shape, temperature and oil).**

<i>Variable factor</i>		<i>Acrylamide content µg/kg</i>	<i>Number of replicates</i>
Shape	Fingers	106.62 <sup>a</sup> ±12	3
T 150°C	Slices	526.58 <sup>b</sup> ±20	3
Oil: sunflower			
Temperature	130 °C	00 <sup>a</sup>	3
Shape: fingers	150°C	106.62 <sup>b</sup> ±12	3
Oil: sunflower	170°C	229.01 <sup>c</sup> ±1.855	3
Oil	Olive	11.97 <sup>a</sup> ±20.74	3
T 150°C	Sunflower	106.62 <sup>b</sup> ±12	3
Shape: fingers	Cotton	91.84 <sup>b</sup> ±10.52	3

The different letters in the column indicate a significant difference at a level of P<0.05.

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## التحري عن مستويات الأكريلاميد في أغذية مقلية مختارة في سورية

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### ملخص

يهدف هذا البحث إلى التحري عن محتوى الأكريلاميد في كل من رقائق البطاطا، والبطاطا المقلية، والفلافل المباعة في الأسواق المحلية، وكذلك إلى دراسة تأثير درجات حرارة القلي المختلفة (130، 150، 170°)، وأنواع الزيت المستخدم في القلي (زيت دوار الشمس، زيت القطن، زيت الزيتون)، وشكل قطع البطاطا (أصابع، شرائح) على محتوى الأكريلاميد في البطاطا المقلية المعدة تحت شروط مضبوطة. وبنتيجه الدراسة تبين أن معدل مستوى الأكريلاميد في كل من رقائق البطاطا، والبطاطا المقلية، والفلافل 469.33، 56.46، 153.78 ميكروغرام / كيلو غرام على التوالي. وبالنسبة للبطاطا الطازجة التي تم إعدادها وقليلها تحت شروط مضبوطة في المختبر، لوحظ أن هناك اختلافاً معنوياً ( $P < 0.05$ ) في مستويات الأكريلاميد عند درجات حرارة القلي (130، 150، 170°م) حيث كانت (0، 106.62،  $229 \pm 5.75$  ميكروغرام/كيلوغرام) على التوالي، وكذلك عند استخدام شكلين لقطع البطاطا (أصابع وشرائح) حيث كانت مستويات الأكريلاميد (106.62، 526.58 ميكروغرام/كيلوغرام) على التوالي. وكان مستوى الأكريلاميد عند القلي بزيت الزيتون غير ملحوظ (11.97 ميكروغرام/كيلوغرام) ومختلفاً معنوياً ( $P < 0.05$ ) عما هو عند القلي بزيت دوار الشمس وزيت القطن (106.62، 91.84 ميكروغرام/كيلوغرام) على التوالي.

الكلمات الدالة: الأكريلاميد، رقائق البطاطا، البطاطا المقلية، الفلافل، المصيدة Strata X-C، HPLC-UV.

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