



## Mitigation of acrylamide formation in fried potato

Hanaa M. Soliman, Said F. Hamed\*

Fats and Oils Dept., Food Industries and Nutrition Institute, National Research Centre, Dokki, Cairo, Egypt, Giza, Egypt



### Abstract

The objective of this work was to study the effect of different pretreatments of potato slices on acrylamide concentration after frying process. Pretreatments of potato slices included soaking slices in solutions of different temperature (40, 50, 60, 70 °C) for 10 min. Soaking solutions were: (i) tap water, (ii) (0.05M) NaCl aqueous solutions, (iii) (0.05M) CH<sub>3</sub>COOH aqueous solutions or (iv) (0.05) tannic acid solution. The effect of different treatments on asparagine and glucose concentrations in potato slices were investigated by following up their concentration before and after each treatment, while reduction in acrylamide formation after each frying process (at 180°C ± 5°C for 6 min) was measured by liquid chromatography-mass spectrometry (LC-MS) for treated and untreated potato slices. Results showed significant reduction in asparagine and glucose concentrations for all treated potato slices. In spite of causing the least reduction in asparagine or glucose concentration, tannic acid treated potato slices showed the highest significant reduction in acrylamide formation. Results demonstrated the efficiency of all the previously mentioned treatments in reducing acrylamide formation significantly without any significant change of physicochemical analyses or sensory attributes of the produced potato chips except dark colour appearance of tannic acid-soaked potatoes.

Keywords: Acrylamide formation, Maillard Reaction, Asparagine, Glucose, Pretreatments.

### 1. Introduction

Deep fat frying is a popular fast-cooking technique that creates special textures and flavors in foods, despite its many downsides, it is still the most convenient cooking process. Usually, this technique is used to prepare snacks and fast food and has grown tremendously in the past 50 years. In April 2002, Swedish scientists of the Swedish National Food Administration found presence of acrylamide (CH<sub>2</sub>CH<sub>2</sub>CONH<sub>2</sub>) in starchy foods, such as potato chips, French fries, and bread [1]. Then the Center for Food Safety and Applied Nutrition classified acrylamide as carcinogenic and neurotoxic to humans [2], where acrylamide double bond is an electrophilic centre, and consequently it can easily react with nucleophilic groups, consequently, it is possible to covalently bond in vivo to cellular nucleophiles [3]. Moreover, acrylamide has been found to be readily absorbed by the skin and mucosa and diffuse strongly into tissues and fetus [4]. This acrylamide was thought to be formed in foods as a result of reaction between asparagine (an amino acid) and carbonyl-containing compound such as reducing sugars at higher temperatures and low moisture conditions, via Schiff base formation, followed by decarboxylation and imine elimination, such mechanism was

confirmed by using of isotopic substitution [5]. Other studies have been conducted to determine the acrylamide percentage in many foods rich in carbohydrates when cooked at high temperatures [6]. Strong variations in acrylamide levels were observed within each food category, it may be attributed to many factors such as amount of acrylamide precursors in the raw material, food chemical composition, additives, and baking or frying conditions [7, 8]. Thus, several studies have been aimed to reduce the concentration of acrylamide in heated food by changing the heating temperature and / or the pH medium, adding antioxidants, or adding salts [9].

This study is aimed to reduce acrylamide formation in potato chips during deep frying through pretreatment of potato slices by soaking in tap water or acid, salt or antioxidant solutions at different temperatures (40, 50, 60, or 70 °C) in order to hinder Schiff base formation, and diminish acrylamide precursors (asparagine and glucose) and, hence, inhibit acrylamide formation.

### Experimental

**Materials:** Sunflower oil, and potato (*Solanum tuberosum*) were purchased from local market in

\*Corresponding author e-mail: [saidfatouhhamed123@gmail.com](mailto:saidfatouhhamed123@gmail.com); (Said F. Hamed).

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Giza, Egypt. All solvents and reagents were supplied from Merck (Darmstadt, Germany). All experiments and analytical determination were performed at least in duplicate.

#### **Sample preparation:**

Potatoes were peeled then sliced into thickness of 2.2 mm and diameter of 5 cm using a Mandolin Slicer (Matfer model 2000, France). Then they were divided into four groups, each group containing 400 g and soaked individually in 800 ml solution at different temperature (40, 50, 60, 70 °C) for 10 min., the soaking solutions were (i) tap water, (ii) (0.05M) NaCl solution, (iii) (0.05M) CH<sub>3</sub>COOH solutions and (iv) (0.05) tannic acid solution.

#### **Frying process:**

Frying was done in a 500 mL domestic deep-fat fryer. The oil was heated at 180°C ± 5°C, then 100 gm of potato slices were fried for 8 min. Each potato batch was cooled on a tissue paper and then thoroughly homogenized and kept for following analysis. The used oil was exchanged after each frying step.

#### **Determination of sugars content**

Reducing sugar was analyzed [10]. Potato samples were dried at 60°C. And grinded individually, then each sample was suspended in warm distilled water and filtered off through Whatman No1 filter paper. 1ml of the filtrate was added to 3 ml of dinitrophenol solution (0.038 M) and incubated at 65–70°C for 6 min. Then cooled to room temperature. Thus, sugars concentrations could be followed spectrophotometrically at 600 nm [11].

#### **Determination of amino acids content**

Amino acids were extracted using acetate buffer at pH 7.0 in water, the extracted amino acid was then hydrolyzed with 6-aminoquinoline-hemisuccinylcaramin, and the later was quantified using reverse phase HPLC [12].

#### **Extraction and analysis of acrylamide**

Acrylamide was extracted and analyzed [13]. A finely ground or homogenized sample (1.000 g) was weighed into a 10 ml centrifuge tube with a cap, and 50 µl of the internal standard (d3-acrylamide) and 9 ml of acetic acid extraction solution were added. After shaking by a vortex mixer for 30 s, the mixture was sonicated for 5 min. Then, 500 µl of Carrez solution I (15 g of (K<sub>4</sub>[Fe (CN)<sub>6</sub>].3H<sub>2</sub>O in 100 ml of water) and 500 µl of Carrez solution II (30 g of (ZnSO<sub>4</sub>.7 H<sub>2</sub>O) in 100 ml of water) was added and mixed for 1 min. After that, the mixture was centrifuged at

8720 rpm for 10 min. A volume of 5 ml of the clear supernatant was transferred to a separator funnel, 5 ml ethyl acetate was added and mixed well. The ethyl acetate layer was removed and the extraction step was repeated twice with 5 ml of ethyl acetate. Ethyl acetate layers were collected and evaporated in a vacuum rotary evaporator at 35 °C to dryness. The residue was dissolved in 1 ml of acetic acid solution (0.2 mM) and filtered through a 0.45 µm pore size nylon syringe filter. Liquid chromatography-mass spectrometry (LC-MS) analysis was performed with a HPLC system 1200 series (Agilent Technologies, Santa Clara, California, USA) coupled to an Agilent 6410 Triple Quad detector equipped with electrospray ionization (ESI) interface. The analytical separation was performed on a Purospher STAR RP-8ec column (150 mm × 4.6 mm, 3 µm particle size) (Merck, Darmstadt, Germany) using an isocratic mixture of 100 ml of acetonitrile and 900 ml of aqueous solution of PFOA (0.05 mM) at a flow rate of 0.5 ml·min<sup>-1</sup> at ambient temperature. All parameters of the electrospray ionization tandem mass spectrometry (ESI-MS-MS) system were based on in-source generation of the protonated molecular ions of acryl amide and the internal standard (d3-acrylamide), as well as collision-induced production of specific fragment ions for multiple reaction monitoring (MRM) experiments (transition for acrylamide: 72 to 55, for d3-acrylamide: 75 to 58) and the following instrumental parameters were used for acrylamide analysis in the ESI+ mode: drying gas (N<sub>2</sub>) flow of 8 l·min<sup>-1</sup>, gas temperature of 350 °C, nebulizer pressure of 345 k Pa, capillary voltage of 2.5 kV, fragment or of 80 V, collision energy of 5 eV, dwell 50 ms. Calibration was performed by diluting the acrylamide stock solution (0.02 g in 100 ml of methanol) in the range of 50–2000 ng·10 ml<sup>-1</sup> with 50 µl of the internal standard (d3-acrylamide).

#### **Physico-chemical properties of frying oil**

Methods in the A. O. A. C. were used to determine acid value and peroxide value [14-15]. Insoluble oxidized fatty acids and insoluble polymer contents of sunflower oil samples were determined according to the methods of Peled, et. Al [16]. Polar and non-polar components in oil samples were separated by column chromatography according to the method described by previous works [17-18].

#### **Sensory evaluation**

Fried potato chips of different treatments were evaluated for their overall acceptability parameters and results were expressed as a mean value of 10

expert tasters. Sensory quality attributes were evaluated using a point hedonic rating scale with 1 for dislike extremely to 10 for like extremely for each attribute.

### Statistical analysis

The data were analyzed using the statistical package, release 11.0 for windows (SPSS), Chicago, INL, USA). In each assay, the experimental data represent the mean of the ten independent assays  $\pm$  standard deviations (SD).

### Results and Discussion

During food frying, Maillard reaction takes place between the active carbonyl function of the reducing sugars and the free amino group of the amino acids, forming various intermediates that causing foods browning [19] which is mainly related to acrylamide formation [18,20,21].

Hence, our experimental protocol went in following up the effects of pretreatments on minimizing the chemical reactivity of such two important precursors (reduced sugar and amino acid) in potatoes before frying process in order to prevent or diminish acrylamide formation.

### Effect of different treatments of potato slices on acrylamide formation

#### Soaking in hot water

Increasing the temperature of water in which potato slice were soaked tended to increase the intramolecular space within the potato tissue. Thus, it could facilitate water molecule penetration into the potato tissue, and consequently led to increase of sugar and polar amino acids dissolution due to hydrogen bonds formation. Thus, potato slices could get rid of some of its sugar and polar amino acids contents, which are the main precursors for acrylamide synthesis.

As shown in Fig.1 reducing sugar contents of potato slices that were soaked in hot water (40, 50, 60, and 70 °C for 10 min.), decreased progressively from 40.17mg/kg for control sample (without any soaking) to reach 21.28 mg/kg potatoes (for samples soaked at 70 °C). The percentage of decrease resulting from soaking potato slices in hot water (40, 50, 60, 70 °C) compared to the control sample ranged from 10.75 to 47.03% for glucose (Fig. 1). While it was found that the concomitant decrease in the concentration of polar amino acids was from 12.74% to 48.53% for samples soaked in tap water heated to 40°C and 70°C, respectively. The same trend of acrylamide formation was observed in fried potatoes as it decreased from 10.37 mg/kg of fries

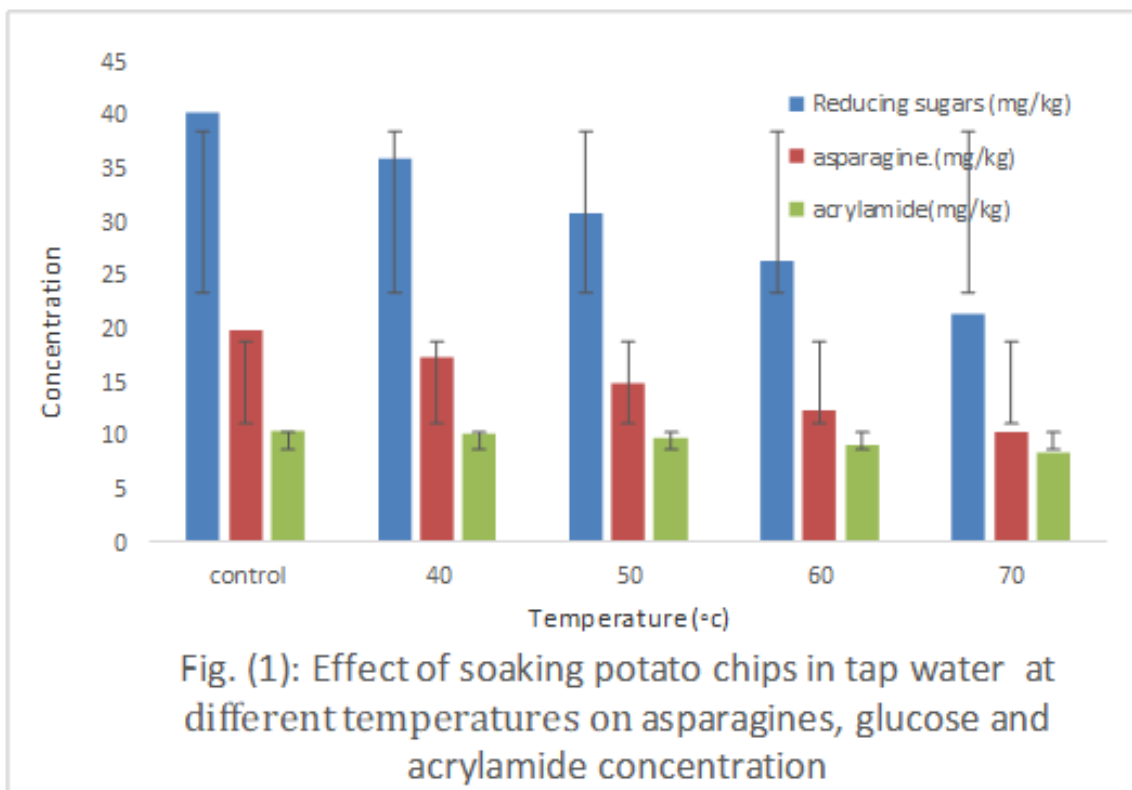
(fried control sample) to 8.38 mg/kg when the potato slices were pre-soaked in hot water (70°C). Significant ( $p < 0.05$ ) reduction in acrylamide formation were from 2.8 % to 19.2 % when samples soaked in hot water (from 40 to 70 °C for 10 min.) compared to a control sample. This decrease in acrylamide can be explained as the result of removing a large amount of acrylamide precursors from the potato tissues during high temperature soaking. In agreement with results Anese et al., suggested that soaking potato chips in tap water or NaCl solution led to a high leaching of important acrylamide precursor such as glucose [22]. Also, Kalyani et al., reported that water soaking and higher temperature conditions lead to greater extraction of acrylamide precursors and decreased production of acrylamide in fried potatoes [23].

#### Soaking in NaCl solution (0.05M)

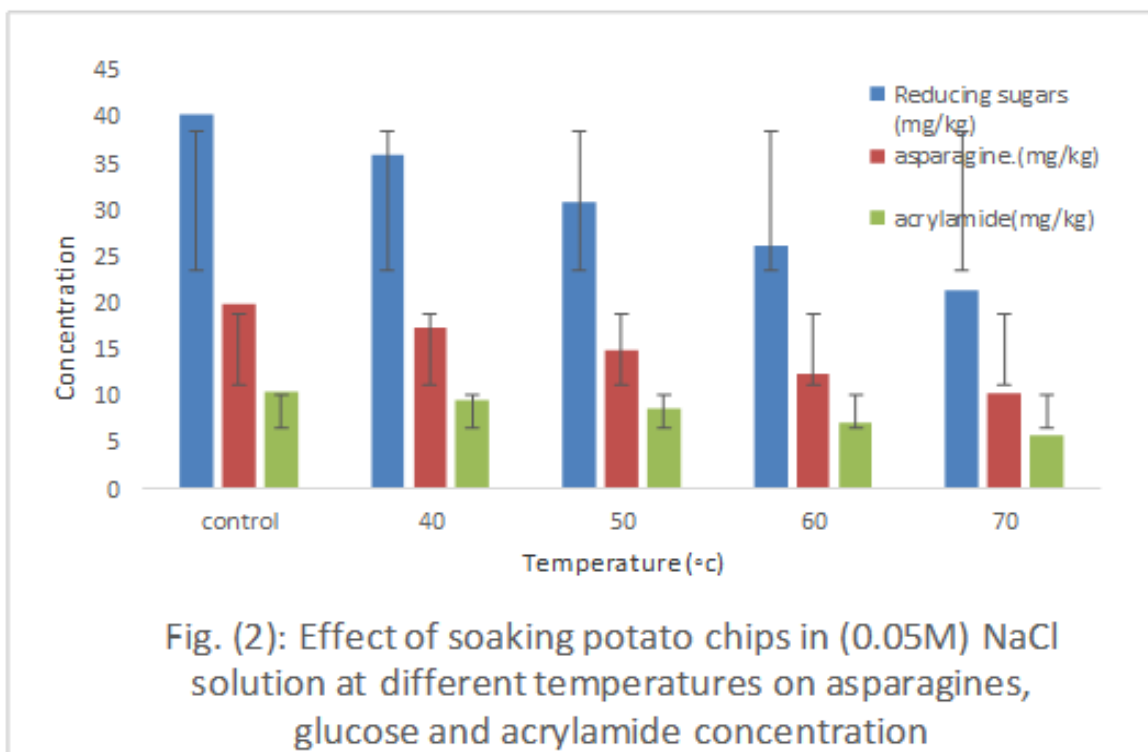
Decreases in glucose, asparagine and acrylamide concentrations were observed when potato chips were soaked in sodium chloride solution (0.05 M) at different temperatures (40, 50, 60 and 70 °C separately) for 10 minutes. Soaking of potato samples at the specified temperature caused reductions ranged from 10.6%, 12.9 % and 8.2 % to 46.9 %, 48.1% and 43.8% for glucose, asparagine and acrylamide, respectively as shown in Fig.2.

#### Soaking in (0.05M) CH<sub>3</sub>COOH Solution

As shown in Fig. 3, soaking potato chips in CH<sub>3</sub>COOH (0.05 M) solution at different temperatures (40, 50, 60 and 70 °C separately) for 10 min. caused the concentration of sugar to decrease from 10.5 % (soaking at 40°C) to 46.9 % (soaking at 70°C) due to dissociation of sugar in water. On the other hand, decrease in asparagine concentration registered 13.2% and 48.7 % at 40 and 70°C, respectively, due to the increases of its dissociation in acid medium. In a concomitant relation, acrylamide formation was going to decrease from 7.0 % to 42.2 % for samples soaked at 40 and 70°C, respectively. Such reduction in acrylamide concentration is assumed to be caused by both actions of water dissolution of acrylamide precursors, in addition to the action of acetic acid anions, which surrounded the partial positively charged carbonyl carbon in sugar, so, they hindered its activity to be attacked by the lone pair of electrons from amino group in asparagine molecule and consequently it hindered Maillard reaction.



The data (values  $\pm$  SE) are the mean values of three measurements for the same sample. L.S.D Least significant differences at  $p \geq 0.05$ .



(Data are expressed as mean  $\pm$  SD values given represent means of three determinations)

### Soaking in (0.05M) tannic acid solutions

Compared to the previous three treatments immersing potato slices in (0.05M) tannic acid solution at different temperatures (40, 50, 60, 70°C separately) for 10 minutes, causing relatively smaller decreases in glucose and asparagine concentrations from 8.5% and 7.8 % (by soaking at 40°C) to 44.7% and 41.0% (by soaking at 70°C), respectively. This may be attributed to the large number of the formed hydrogen bonds between tannic acid and water, which

In agreement with our findings Nems et al. showed that the polyphenols could scavenge the amino source asparagine-derived intermediates and retard the fried food by combined mechanisms such as (1) carbonyl trapping effect, (2) radical scavenging activity, and (3) limitation of sugar degradation through the Maillard reaction. The only drawback encountered in tannic acid treated samples was the dark color of the fried potatoes.

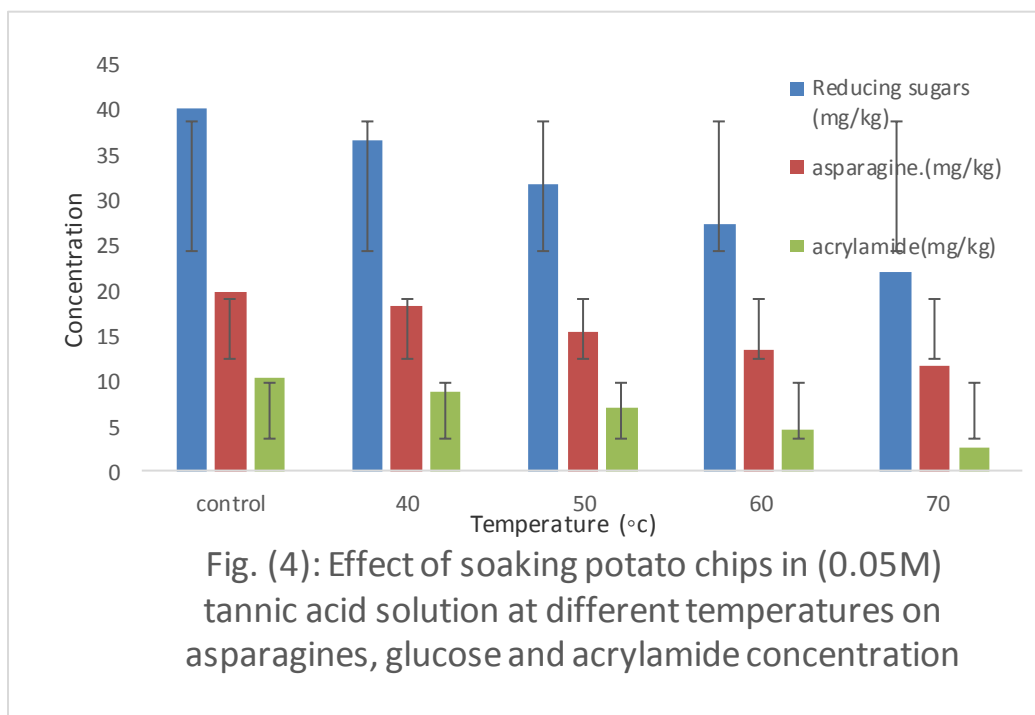
consequently somewhat decrease water ability to leach glucose and asparagine molecules.

On the other hand, acrylamide formation was sharply decreased from 15.4% (by soaking at 40°C) to 73.7% (by soaking at 70°C) as shown in Fig. 4. Reduction of acrylamide concentration in presence of tannic acid is mostly caused by the strong antioxidant activity of tannic acid that has the ability to capture the lone pair of electrons on the amino group of asparagine and inhibited them attacking on the carbonyl group, and consequently it could hinder Maillard reaction.

pathways leading to acrylamide formation [24]. Also, many authors [25-28] reported that polyphenols could efficiently minimize the acrylamide contents in

### Physico-chemical properties of frying oil

As shown in Table (1) there were no significant ( $p < 0.05$ ) changes in physico-chemical properties of frying oil throughout all the experiments because the frying oil was changed after each frying process



(Data are expressed as mean  $\pm$  SD values given represent means of three determinations)

Table 1. Effect of pretreatments on some physico-chemical properties of frying oil

| parameter | Control<br>(Room<br>Temp.) | H <sub>2</sub> O |                 |                 |                 | CH <sub>3</sub> COOH (5ml/l) |                 |                 |                 | NaCl (5g/l)     |                 |                 |                 | Tannic acid     |                 |                 |                 |
|-----------|----------------------------|------------------|-----------------|-----------------|-----------------|------------------------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
|           |                            | 40°C             | 50°C            | 60°C            | 70°C            | 40°C                         | 50°C            | 60°C            | 70°C            | 40°C            | 50°C            | 60°C            | 70°C            | 40°C            | 50°C            | 60°C            | 70°C            |
| F.F.A     | 0.49                       | 0.47<br>±0.05    | 0.49<br>±0.04   | 0.49<br>±0.06   | 0.46<br>±0.04   | 0.5<br>±0.04                 | 0.48<br>±0.04   | 0.47<br>±0.06   | 0.49<br>±0.04   | 0.5<br>±0.04    | 0.55<br>±0.04   | 0.55<br>±0.03   | 0.5<br>±0.04    | 0.51<br>±0.05   | 0.48<br>±0.05   | 0.48<br>±0.04   | 0.49<br>±0.04   |
| P.V       | 1.63                       | 1.87<br>±0.03    | 1.89<br>±0.04   | 1.77<br>±0.05   | 1.91<br>±0.05   | 1.87<br>±0.04                | 1.88<br>±0.04   | 1.86<br>±0.04   | 1.9<br>±0.05    | 1.89<br>±0.03   | 1.77<br>±0.04   | 1.65<br>±0.04   | 1.89<br>±0.05   | 1.71<br>±0.06   | 1.62<br>±0.04   | 1.66<br>±0.04   | 1.64<br>±0.03   |
| O.F.A     | 0.00                       | 0.001<br>±0.004  | 0.001<br>±0.004 | 0.001<br>±0.003 | 0.001<br>±0.003 | 0.001<br>±0.005              | 0.001<br>±0.005 | 0.001<br>±0.005 | 0.001<br>±0.003 | 0.001<br>±0.004 | 0.001<br>±0.003 | 0.001<br>±0.003 | 0.001<br>±0.003 | 0.001<br>±0.004 | 0.001<br>±0.004 | 0.001<br>±0.003 | 0.001<br>±0.005 |
| Polar %   | 0.188                      | 0.188<br>±0.03   | 0.188<br>±0.04  | 0.188<br>±0.05  | 0.188<br>±0.04  | 0.186<br>±0.04               | 0.187<br>±0.05  | 0.186<br>±0.04  | 0.188<br>±0.05  | 0.187<br>±0.05  | 0.187<br>±0.04  | 0.187<br>±0.05  | 0.188<br>±0.04  | 0.185<br>±0.39  | 0.184<br>±0.03  | 0.185<br>±0.03  | 0.188<br>±0.04  |
| Polymer % | 0.149                      | 0.148<br>±0.05   | 0.149<br>±0.03  | 0.149<br>±0.04  | 0.149<br>±0.05  | 0.150<br>±0.05               | 0.150<br>±0.03  | 0.151<br>±0.04  | 0.150<br>±0.05  | 0.149<br>±0.04  | 0.149<br>±0.04  | 0.149<br>±0.04  | 0.149<br>±0.05  | 0.144<br>±0.04  | 0.145<br>±0.05  | 0.146<br>±0.04  | 0.146<br>±0.04  |

(Data are expressed as mean ± SD values given represent means of three determinations)

**Sensory evaluation**

Table 2 illustrates that overall acceptability parameters of the fried potato chips treated with different acrylamide lowering treatments had general acceptability

more than that of control, but the only drawback was the dark color of potato chips that treated with tannic acid.

**Table 2.** Effect of pretreatments on sensory evaluations of fried potato chips

| Parameter   | control       | H <sub>2</sub> O |               |               |               | CH <sub>3</sub> COOH (0.05M) |               |               |               | NaCl (0.05M)  |               |               |               | Tannic acid (0.05M) |               |               |               |
|-------------|---------------|------------------|---------------|---------------|---------------|------------------------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------------|---------------|---------------|---------------|
|             |               | Room Temp.       | 40°C          | 50°C          | 60°C          | 70°C                         | 40°C          | 50°C          | 60°C          | 70°C          | 40°C          | 50°C          | 60°C          | 70°C                | 40°C          | 50°C          | 60°C          |
| Appearance  | 8.71<br>±0.52 | 8.75<br>±0.44    | 8.74<br>±0.50 | 8.77<br>±0.51 | 8.77<br>±0.42 | 9.41<br>±0.42                | 9.40<br>±0.49 | 9.43<br>±0.52 | 9.43<br>±0.46 | 9.51<br>±0.40 | 9.50<br>±0.43 | 9.52<br>±0.50 | 9.52<br>±0.49 | 7.31<br>±0.44       | 7.30<br>±0.53 | 7.30<br>±0.46 | 7.29<br>±0.41 |
| Crust color | 8.90<br>±0.50 | 8.89<br>±0.43    | 8.91<br>±0.50 | 8.90<br>±0.50 | 8.92<br>±0.52 | 9.60<br>±0.52                | 9.64<br>±0.44 | 9.62<br>±0.51 | 9.66<br>±0.47 | 9.71<br>±0.56 | 9.71<br>±0.55 | 9.71<br>±0.48 | 9.73<br>±0.52 | 7.13<br>±0.52       | 7.12<br>±0.50 | 7.12<br>±0.44 | 7.14<br>±0.53 |
| Crumb color | 8.60<br>±0.50 | 8.70<br>±0.45    | 8.66<br>±0.55 | 8.69<br>±0.50 | 8.63<br>±0.57 | 9.57<br>±0.42                | 9.57<br>±0.53 | 9.58<br>±0.40 | 9.58<br>±0.44 | 9.72<br>±0.46 | 9.71<br>±0.53 | 9.72<br>±0.47 | 9.73<br>±0.52 | 8.43<br>±0.46       | 8.45<br>±0.50 | 8.45<br>±0.53 | 8.45<br>±0.44 |
| Flavor      | 7.70<br>±0.53 | 7.71<br>±0.43    | 7.70<br>±0.54 | 7.71<br>±0.48 | 7.69<br>±0.48 | 9.51<br>±0.51                | 9.50<br>±0.47 | 9.53<br>±0.45 | 9.53<br>±0.40 | 9.61<br>±0.50 | 9.62<br>±0.52 | 9.63<br>±0.55 | 9.63<br>±0.52 | 7.22<br>±0.50       | 7.22<br>±0.55 | 7.21<br>±0.47 | 7.22<br>±0.46 |
| Texture     | 8.83<br>±0.53 | 8.85<br>±0.53    | 8.84<br>±0.50 | 8.85<br>±0.44 | 8.83<br>±0.53 | 9.44<br>±0.52                | 9.44<br>±0.50 | 9.44<br>±0.48 | 9.45<br>±0.50 | 9.49<br>±0.44 | 9.51<br>±0.42 | 9.51<br>±0.52 | 9.52<br>±0.48 | 9.21<br>±0.40       | 9.22<br>±0.49 | 9.25<br>±0.53 | 9.24<br>±0.54 |

(Data are expressed as mean ± SD values given represent means of three determinations)

## Conclusion

soaking of potato chips in a solution of NaCl, acetic acid, or tannic acid individually caused significant decrease in acrylamide formation in fried potatoes and also, they could improve the potato chips sensory evaluations. However, soaking in tannic acid although causing the highest acrylamide decrease, it also produced dark colored potato chips which was unacceptable by consumers.

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