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Acrylamide levels in roasted pistachio under various time-temperature conditions and formulations

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ABSTRACT

The most common form of pistachio consumption is the roasted type. This process is done with various additives at different temperatures. Acrylamide could be generated during the heating process of pistachio at high temperatures such as roasting method. It is neurotoxic, genotoxic and probable carcinogenic to humans and can produce due to Maillard reactions. In the current study, the effect of different time-temperature conditions and additives on the acrylamide content of roasted pistachios was investigated. Raw pistachio was formulated with different additive including; NaCl, starch, ascorbic acid, and sodium metabisulphite at three time-temperature; 120 ºC -30 min, 150 ºC -25 min, 180 ºC -20 min by ELISA method. NaCl, sodium metabisulphite and ascorbic acid reduced the amount of acrylamide, and starch had no impact on the amount of acrylamide. The lowest amount of acrylamide was found in time-temperature of 120 ºC- 30 min use each of NaCl, sodium metabisulphite and ascorbic acid.

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1. Introduction

Pistachio is an edible seed of the pistachio trees (Pistacia Vera L.), a plant of the Anacardiaceae family (1). Pistachio is the most important agricultural product cultivated in Iran and Iran is the largest exporter of pistachios (about 86%) in the world (2,3). Pistachio due to large amounts of some nutrients and health promoting compounds, such as unsaturated fatty acids, vitamins, minerals, sterols and polyphenols has been considered (4). One of the most common forms of pistachio's processing is roasting. This process leads to sensory, textural, physical and chemical changes in product, which is mainly a result of the Maillard reactions (2,5). Maillard reaction also plays an important role in the formation of toxic compounds such as acrylamide (6).

Studies have shown that acrylamide of Maillard reactions products was generated during the food heating process at temperatures above 120°C and low humidity, which involves the reaction of especially amino acids (primarily asparagine) and reducing sugars (e.g., glucose) (7-11). Acrylamide (2-propenamide), with the molecular formula of CH2 = CHCONH2, is neurotoxic, genotoxic and probable carcinogenic to humans (12-16). According to neurological, genetic and reproductive toxicities and wide exposure through common foods, such as fried foods, bakery products, fast food and snacks, reduction methods to acrylamide generation in foods around the world have been considered (10). Present procedures to reduce the acrylamide levels involve three basic aspects: modifying raw materials, process optimization

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and the use of additives (8,17).

Little information is available for the reduction of acrylamide in roasted nuts (18). Therefore, the aim of this study was to determine the effect of different time-temperature conditions and additives (NaCl, starch, ascorbic acid, $Na_2S_2O_5$) on the acrylamide levels of roasted pistachios by the ELISA method.

2. Materials and methods

2.1 Materials

Sodium chloride (NaCl), ascorbic acid, sodium metabisulphite ($Na_2S_2O_5$) and potato starch obtained from Merck Company. The ELISA kit was purchased from Morinaga Co, Japan.

2.2. Roasting

Raw pistachio, Fandoghi variety (Ohadi), was prepared from Rafsanjan Pistachio Research Center. Raw pistachio were formulate different additive including; NaCl (15%), NaCl (15%), starch (0.5%), NaCl (15%), ascorbic acid (1%), NaCl (15%) and $Na_2S_2O_5$ (1 %). Then, 30 g of pistachio's samples was placed in a petri dish with a diameter of 14 cm as one layer and roasted in the electric oven (Memmert ule500) at temperatures of 180, 150 and 120 °C at times of 20, 25 and 30 minutes respectively. After that, roasted pistachios were cooled at room temperature and placed in plastic bags, then stored in a temperature at 8 °C.

2.3. Acrylamide analysis

ELISA was used for the determination of acrylamide. Steps of acrylamide determination were performed according to the kit instructions. The kit makes a sensitive and specific assay of acrylamide possible with a detection limit of 0.4 ng/mL (RSD=30%) and a quantification range of 0.7- 90 ng/mL (RSD<20%). The pistachio samples were powered using a blender. One g of powered pistachio samples was placed in a 50 mL centrifugation tube and 19 mL distilled water was added. The mixture was agitated using a vortex mixer. The tubes were centrifuged at 3600 × g for 20 min at 20-30 °C. A solid phase extraction (SPE) cartridges were used for extraction by washings with 1mL aliquots of methanol (once), water (twice). One mL aliquot of the supernatant was applied (the effluent during application was discarded), then it was eluted with 3 mL distilled water. The eluate (3 mL) was collected into a polypropylene tube, it was vortexed thoroughly, and was defined as "pistachio sample extract". After that, serial dilutions of the acrylamide standard (0-500 ng/mL) were prepared for use as ELISA calibration. Then the steps related to the ELISA method were performed according to the instruction of kit.

2.4. Statistical analysis

Statistical analysis was performed using R software (version 3.2.5) and Excel. Linear regression was conducted on the data obtained from the amount of acrylamide for different variables of temperature and time and additives.

3. Results

The results of the present study are given in Fig 2. In this study, the level of acrylamide in different roasting condition was assessed. The level of acrylamide was lower in NaCl, sodium metabisulphite and ascorbic acid groups. The calibration curve was constructed using 4-Parameter Logistic Regression (Fig. 1), and the acrylamide concentration in the "pistachio sample extract" was interpolated from the calibration curve using B/B0 (B0, the absorbance for null acrylamide concentration; B, the absorbance for other concentrations) values of each sample. The acrylamide content in the "pistachio sample" (ng/g) was calculated from the acrylamide concentration in the "pistachio sample extract" (ng/mL) according to the following equation: Acrylamide concentration in the "pistachio sample extract" (ng/mL) × 20 [a]× 3 [b]

[a] Dilution factor (water-extraction step)

[b] Dilution factor (solid-phase extraction step)

4. Discussion

The acrylamide content has been increased with increasing of roasting temperature (Fig. 2). The highest and lowest amounts of acrylamide were found in pistachio samples that were roasted at 180 °C for 20 minutes and 120 °C for 30 minutes, respectively. Schlormann et al. (2015) demonstrated that the amount of acrylamide increases, when the temperature of pistachio roasting increases (5).

NaCl had the most reducing effect on the level of acrylamide in the roasted pistachios. The effect of NaCl on the amount of acrylamide has been investigated in potato (19,20), bakery products (21,22) and model systems (23,24) by various researchers. The reducing effect of NaCl on the amount of acrylamide may be because of the cation inhibition from Schiff base formation of asparagine (25) and/or an increase of polymerization of acrylamide with NaCl (23).

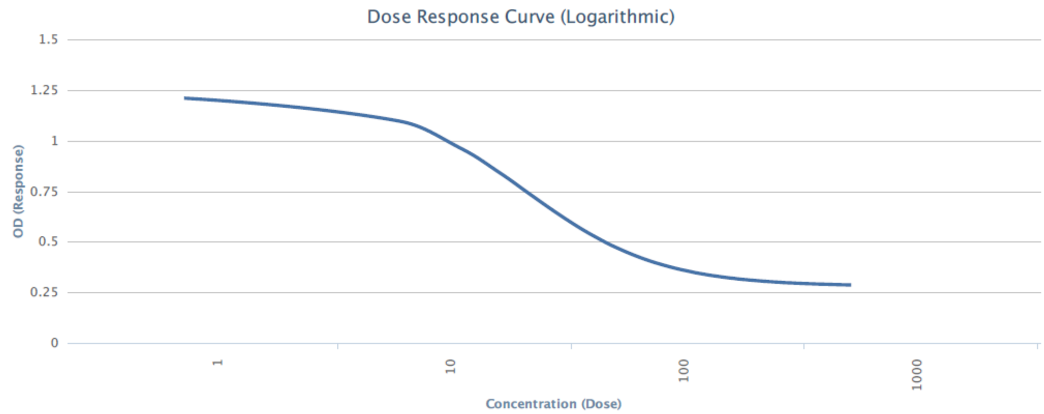

Fig .1. Calibration curve of acrylamide standards (0.69-500 ng/mL), R2=0.9924.

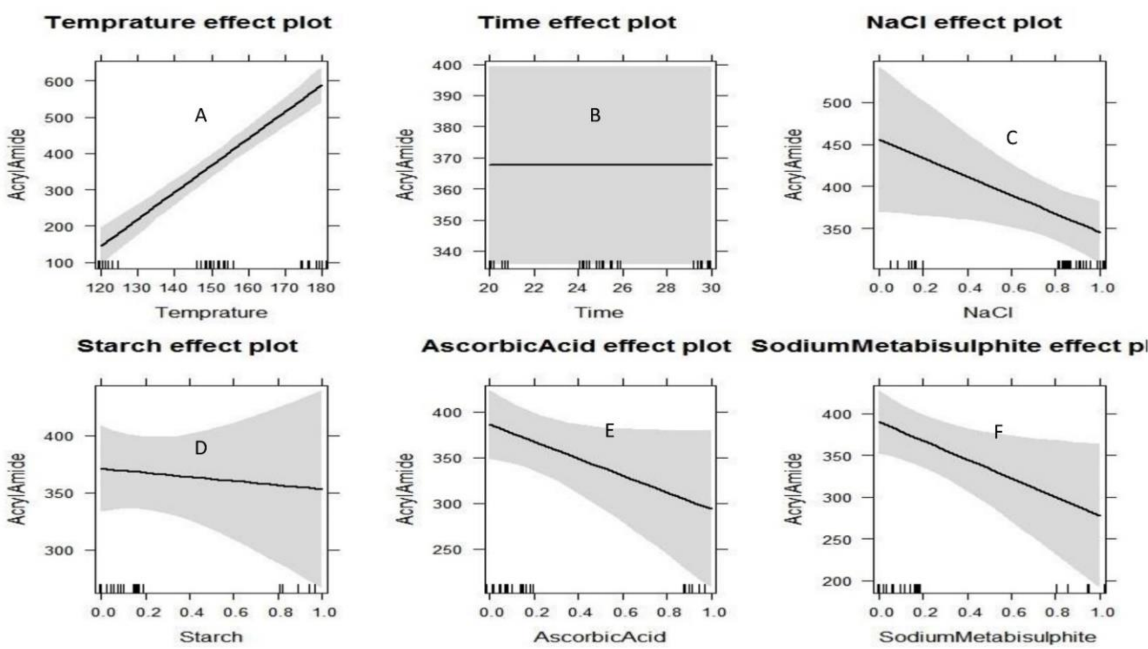

Fig. 2. The effects of (A) temperature (120,150,180 ºC), (B) time (30, 25, 20 min), (C) NaCl, (D) starch, (E) ascorbic acid and (F) sodium metabisulphite on the rate of acrylamide changes (ng/g).

The amount of acrylamide was significantly (p-value <0.05) reduced by sodium metabisulphite in roasted pistachio samples. Inhibition of intermediate compounds responsible in the formation of acrylamide has been expressed as a reaction mechanism for sulphites to reduce acrylamide formation (26-28).

Ascorbic acid caused the lowest reduction in the acrylamide content of the roasted pistachios. The effect of ascorbic acid on the amount of acrylamide also has been investigated in potato products (29,30) and model systems (21,30). Rannou et al. (2016) stated that lowering effect of ascorbic acid is due to the reaction of asparagine with carbonyl groups resulted from oxidation products and/or degradation of ascorbic acid (31). The results showed that starch has no significant impact on the amount of acrylamide and is not effective. The reason may be due to low levels (0.5%) used or method of use (immersion).

5. Conclusion

The application of temperature of 120 °C and use each of NaCl (15%), sodium metabisulphite (1%) and ascorbic acid (1%) could be effective in reducing the amount of acrylamide in roasted pistachios

Conflict of interest

The authors declare no conflicts of interest.

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