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# The presence of acrylamide in various type of food products from the Serbian market

V Koricanac<sup>1</sup>, S Jankovic<sup>1</sup>, D Vranic<sup>1</sup>, I Stankovic<sup>2</sup>, D Nikolic<sup>1</sup>, Z Petrovic<sup>1</sup> and D Milicevic<sup>1</sup>

<sup>1</sup>Institute of Meat Hygiene and Technology, Kaćanskog 13, 11040 Belgrade, Serbia <sup>2</sup> University of Belgrade – Faculty of Pharmacy, Vojvode Stepe 450, 11221 Belgrade, Serbia

E-mail: vladimir.koricanac@inmes.rs

Abstract: Acrylamide forms when some foods are prepared at temperatures usually above 120°C and in low moisture conditions, due to a Maillard reaction between certain amino acids, such as asparagine, and reducing sugars. Acrylamide is carcinogenic to experimental mice and rats, neurotoxic and probably also carcinogenic and genotoxic for humans. The aim of this study was to determine the presence of acrylamide in various groups of food products in which its formation is expected to occur during the production process. In the period December 2017 to March 2021, 529 samples of different types of food products were tested. Samples were collected from the Serbian market. Most of the tested foods, almost half of them (44%), were various types of biscuits. The presence of acrylamide was determined using LC-MS/MS accredited method, with a limit of quantification (LOQ) of 50 µg kg-1and a limit of detection (LOD) of 25 µg kg-1. All samples from the snack product and waffle product groups contained acrylamide. Acrylamide was detected in almost all (98.98%) fine bakery products and biscuits (90.43%). In contrast, only 15.38% of bakery products contained acrylamide. Most of the tested foods contained acrylamide, 83.74% of them.

#### 1. Introduction

Food consumed by human populations is an excellent foundation for various dangers and hazards, whether of biological, chemical or physical origin, with potentially harmful effects on human health. Food can be contaminated at primary and secondary levels. Primary contamination is during all production stages, preparation, processing, treatment and distribution, and secondary contamination occurs due to inadequate and improper storage. Intake of food that contains various hazards can result in diseases in humans [1].

Acrylamide (AA) is a low molecular weight, highly water soluble, organic compound. It forms when some foods are prepared at temperatures usually above 120°C and in low moisture conditions, due to a Maillard reaction between certain amino acids, such as asparagine, and reducing sugars. AA forms in numerous baked or fried carbohydrate-rich foods, including French fries, potato crisps, breads, biscuits and coffee [2, 3, 4].

AA is carcinogenic to experimental mice and rats, causing tumours at multiple organ sites in both species when given in drinking water or by other means [5]. Adverse effects reported in repeated dose

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toxicity studies of AA in rats, mice, monkeys, cats and dogs consisted of body weight loss and effects on the nervous system reflected by hind-limb paralysis, reduction in motor performance and/or histopathological changes in peripheral nerves and nervous system structures. In mice, effects reported, in addition to the neurotoxicity, consisted of effects on the testes, including the degeneration of epithelia in spermatids and spermatocytes, the reduction of spermatozoa, and the presence of multinucleate giant cells, as well as forestomach hyperplasia, hematopoietic cell proliferation of the spleen, preputial gland inflammation, lung alveolar epithelium hyperplasia and cataract and for female mice, ovarian cysts [2]. AA is carcinogenic in multiple tissues in both male and female mice and rats. In rats, the major tumours produced by AA are adenomas, fibroadenomas and fibromas of the mammary gland, thyroid gland follicular cell adenomas or carcinomas, and in F344 rats, testes or epididymis tunica vaginalis mesotheliomas. In mice, the major tumours produced by AA are: Harderian gland adenomas, mammary gland adenoacanthomas and adenocarcinomas, lung alveolar and bronchiolar adenomas, benign ovary granulosa cell tumours, skin sarcomas, and stomach and forestomach squamous cell papillomas in females, and Harderian gland adenomas and adenocarcinomas, lung alveolar and bronchiolar adenomas and carcinomas, and stomach squamous papillomas and carcinomas in males [2].

Cancer is leading cause of mortality worldwide. It is assumed that cancer risk is mainly affected by environmental factors including diet habits [6]. AA is neurotoxic and probably also carcinogenic and genotoxic for humans [7, 8, 9, 10, 11]. Since its discovery in food, AA levels were monitored in various countries and the results indicated a public health concern [12]. The food safety control system in Serbia is based on the examination of foodstuffs during import, self-control by domestic producers and monitoring of foodstuffs in circulation by inspection bodies. The levels of reference values for the presence of AA in food (French fries, potato chips, crackers and other products based on potatoes and potato dough, soft bread, breakfast cereals, biscuits and waffles, gingerbread, roasted and instant coffee, coffee substitutes) are defined in Annex 2 of the Rulebook of maximum concentrations of certain contaminants in food [13].

The aim of this study was to determine the presence of AA in various groups of food products in which AA formation is expected to occur during the production process.

## 2. Materials and Methods

In the period December 2017 to March 2021, 529 samples of different types of food products (biscuits – 230, fine bakery products – 98, snack products – 81, bakery products – 65, waffle products – 29, confectionery – 13, coffee products – 13) were examined. Samples were collected from the Serbian market. The presence of AA was determined using the LC-MS/MS accredited method, after sample extraction and cleanup using the QuEChERS technique [14, 15], with a limit of quantification (LOQ) of 50  $\mu$ g kg<sup>-1</sup>and a limit of detection (LOD) of 25  $\mu$ g kg<sup>-1</sup>. The LC-MS/MS system and software (LabSolutions) used to determine the presence of AA were produced by Shimadzu. All chemicals used were of analytical grade and were used as received without any further purification. The results analysis and graphical presentation of their distribution was performed using Microsoft Office Excel 2016.

#### 3. Results and Discussion

The proportions (numbers and %) of food products in the food groups that contained AA are shown in Table 1. Also, distributions of the results, by groups and in all tested samples, are graphically presented in Figures 1-3.

Most of the tested food samples, almost half of them (44%), were various types of biscuits. On the another side, fewer samples were from the groups of confectionery and coffee products, 2% of all samples for both.

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**Table 1.** Food product samples in defined food groups that were tested for the presence of AA, forthe period December 2017 to March 2021

Food product	Ν	n <sub>d</sub> (%)	n <sub>nd</sub> (%)
Biscuits	230	208	22
Fine bakery	98	(90.45) 97 (98.98)	(3.57) 1 (1.02)
Snack products	81	81 (100.00)	(1.02) 0 (0.00)
Bakery products	65	10 (15.38)	55 (84.62)
Waffle products	29	29	0
Confectionery	13	10 (76.92)	3 (23.08)
Coffee products	13	8 (61.54)	5 (38.46)
Total	529	443 (83.74)	86 (16.26)

N – total number of tested samples;  $n_d$  – number of samples that contained AA;  $n_{nd}$  – number of samples where AA was not detected; LOD = 25 µg kg<sup>-1</sup>

All samples from snack product and waffle product groups contained AA. AA was detected in almost all fine bakery products (98.98%) and biscuits (90.43%). In contrast, only 15.38% of bakery products samples contained AA, which makes this group the only food group within which there were more samples in which AA was not detected (84.62%).

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More than a third (Figure 2) of all tested samples were biscuits containing AA. This was expected, as samples of this food product group were the most numerous (Figure 1).



#### 4. Conclusion

Most of the tested food product samples contained AA, 83.74% of them. In every sample of snacks and waffle products, AA was detected. Only one sample (1.02%) of fine bakery products did not contain AA. Bakery products were the safest group of samples, regarding AA, with only 15.38% of samples containing this compound. All in all, the results of this study confirm [16] that AA, in most food products that are subject to the formation of AA in terms of their chemical composition and production process,

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is unfortunately inevitable. Accordingly, continuous food safety controls are necessary, as well as further research to consider the AA levels in different food groups, and data on the average daily intake of these foods by population groups, in order to integrate these data to assess the health risk of AA intake through food.

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