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II.5. ASSESSMENT OF ACRYLAMIDE CONTENT IN SOME FOODSTUFFS ON THE ROMANIA MARKET

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Abstract

The paper aimed to present the evaluation of acrylamide (AA) content in foodstuffs during 2018-2019 periods on the Romania market. It is based on the data provided by the National Sanitary Veterinary and Food Safety Authority (ANSVSA). The acrylamide levels were analyzed within ANSVSA by high performance liquid chromatography coupled with a diode array detector (HPLC-UV). The number of foodstuffs samples analyzed in Romania at the national level by ANSVSA was 138 from which 50 were analyzed in 2018 and 88 in 2019. From the total of foodstuffs samples analyzed only in one foodstuff sample was detected the AA level in 2018 and in sixteen foodstuffs samples was detected the AA in 2019.

Key words*: acrylamide, food safety, risk policy.*

INTRODUCTION

Acrylamide (AA) is a food-borne chemical as a result of cooking practices and especially in foodstuffs with high-carbohydrate content which are processed by high temperature (>120°C) (Ghalebi et al., 2019; Pratama & Jacxsens, 2019). Researchers from national food safety authorities, from academia medium, and from food industry have sought to better understand the mechanisms of acrylamide formation and to find ways to minimize its formation in foodstuffs. Maillard reaction, is the main mechanism of acrylamide formation in foodstuffs containing carbohydrates and amino acids, in particular asparagines, but it can be formed and through other pathways, such as the reaction between aspartic acid and reducing sugars, thermal degradation of amino acids and proteins, by the conversion of acrolein, acrylic acid, wheat gluten or by de-amination of 3-aminopropionamide, a result of asparagine enzymatic decarboxylation, as well as decarboxylation and deamination of asparagines (Nachi et al., 2018; Michalak et al., 2019; Maan et al., 2020).

Acrylamide is found in many types of foodstuffs, including breakfast cereals, bakery products, roasted coffee, cappuccino powder, cookies, crackers, snacks, biscuits, wafers, frenchfries, fried potatoes, and potato chips (Ghalebi et al., 2019; Mousa et al., 2019; Kang et al., 2020; Mesías, et al., 2020; Schouten et al.,2020; Wang et al., 2020). For example, acrylamide concentration in potato chips fried in sunflower oil ranged from 525 to 722 μg/kg (Mekawi et al., 2019). It is not found in protein-rich material such as meat and fish and in boiled products (Pratama & Jacxsens, 2019). Acrylamide has also been found in the environment, cosmetics, drinkingwater, as well as cigarette smoke (Tölgyesi & Sharma, 2020).

The toxicity of acrylamide has been acknowledged long ago, since 2002, when it was first reported that acrylamide can be found in foodstuffs by Tareke et al. (Murray et al., 2020). Human exposure to acrylamide may have toxicological effects (neurotoxicity, genotoxicity, carcinogenicity, and reproductive toxicity), and acrylamide has been classified as carcinogenic by the International Agency for Research on Cancer in the 2A group (probably carcinogenic in humans) (Fernandes et al., 2019). Acrylamide is found in various tissues of the body such as liver, kidney, brain, heart and even breast milk (Ghalebi et al., 2019). Several studies using human cells have shown that chronic low-level exposure to acrylamide can lead to progressive degeneration of the peripheral and central nervous systems characterized by cognitive and motor abnormalities (Murray et al., 2020; Wang et al., 2020).

Therefore, many efforts have been made to decrease content of acrylamide in foodstuffs and develop analytical method for the determination of acrylamide. Among the methods of reducing the level of acrylamide in foods may be following: the use of the enzyme asparaginase, which converts asparagine to aspartic acid (Anese et al., 2011); the use of extracts with antioxidant properties (Miśkiewicz et al., 2019); the use of fermentation processes performed with lactic acid bacteria (LAB) for bread manufacturing (Bartkiene et al., 2011); manipulating processing conditions such as time and temperature of the heating process, and including certain preheating treatments such as soaking and blanching(Yıldız et al., 2017); the addition of different phenol standards and olive leaf extract (OLE) during the sterilization and baking process; the identification of potato varieties low in the acrylamide precursors asparagine and reducing sugars that vary with variety, nitrogen (N) fertilizer applications, and other agronomic factors (Johnson et al., 2019; Yu et al*.*, 2019; Fernández et al*.*, 2020; Martín-Vertedor et al*.*, 2020; Rifai & Saleh, 2020).

European Commission started monitoring of acrylamide levels in processed foods since 2007 (Ghalebi et al., 2019). Acrylamide can be analysed using the following methods: gas or liquid chromatography (GC or LC); high performance liquid chromatography mass spectrometry (HPLC-MS), high performance liquid chromatography tandem mass spectrometry (HPLC-MS/MS), the bromination, xanthydrol, and silylation; ultra-performance liquid chromatography (UPLC); liquid chromatography-high resolution mass spectrometry (LC-HRMS); electrochemical biosensor for acrylamide detection based on hemoglobin entrapped in ionic liquid-carbon and other (Fernandes et al., 2019; Michalak et al., 2019; Wawrzyniak et al*.*, 2019; Desmarchelier et al., 2020; Li et al*.*, 2020; Tölgyesi & Sharma, 2020).

The determination of AA levels in different foodstuffs from Romania market was performed by the National Sanitary Veterinary and Food Safety Authority (A.N.S.V.S.A.) by using HPLC-UV method. The aim of this study was to analyze the data, regarding the level of AA in some foodstuffs and the distribution of in Romania, provided by ANSVSA in 2018 and 2019.

MATERIALS AND METHODS

Samples

A total of 138 foodstuffs samples of various types were collected from different counties from Romania market in order to determine their acrylamide level (AA). The foodstuffs samples were analyzed by the National Sanitary Veterinary and Food Safety Authority (A.N.S.V.S.A.). The foodstuffs samples were analyzed in the 2018 and 2019 years namely 50 samples in 2018 and 88 in 2019.

Laboratory tests were performed within the ANSVSA for foodstuffs samples that come from retail-supermarkets, hypermarkets units, specialized stores. The samples were collected by the inspectors of the veterinary sanitary directions and for the food safety from 7 counties in 2018 (Timis, Constanta, Mures, Iasi, Suceava, Prahova, Dolj) and from 16 counties in 2019 (Alba, Constanta, Suceava, Cluj, Timis, Iasi, Mures, Prahova, Bucharest, Dolj, Neamt, Braila, Arges, Vrancea, Bacau, Gorj). The samples were collected under the Surveillance and Control Program in the food safety of non-animal origin field, approved by Order A.N.S.V.S.A. no. 35/2016, with subsequent modifications, or within the self-control program of the production units.

The distribution of the 50 foodstuffs samples analyzed through it AA content by counties in 2018 are shown in Table 1.

Table 1. The AA level in foodstuffs samples analyzed in 2018 from Romania market

Category	**Foodstuffs tested**	**Counties**
Biscuits	4	Dolj (1) Iasi (1) Timis (2)
Fooddishes	1	Iasi (1)
Breakfast cereals	11	Constanta (1) Dolj (2) Iasi (2) Mures (2) Timis (2) Suceava (1) Prahova (1)
Roast coffee	9	Constanta (1) Dolj (1) Iasi (1) Mures (4) Prahova(1) Timis (1)
French fries	6	Constanta (1) Dolj (2) Iasi (1) Timis (1) Prahova(1)
Potato crisps	5	Constanta (1)
Food products based on potato Bread	5 4	Dolj (1) Iasi (1) Prahova(1) Timis (1) Dolj (1) Prahova(1) Timis (2) Iasi(1) Dolj (1) Iasi (1) Prahova (1) Timis (1)
Bakery products	3	Constanta(3)
Cocoa products	1	Prahova (1)
Confectionary products	1	Dolj (1)

The 88 foodstuffs samples analyzed for it AA content in 2019 are shown in Table 2 in which are presented the category of products analyzed and the numbers of collected samples from each Romania county.

Table 2. The AA level in foodstuffs samples analyzed in 2019 from Romania market

Category	**Foodstuffs tested**	**Counties**
Biscuits	6	Dolj (1) Mures (3) Timis (2)
Fooddishes	5	Constanta (1) Cluj (1) Alba (1) Bucuresti (1) Prahova (1)
Breakfast cereals	8	Iasi (1) Dolj (1) Constanta (1) Cluj (1) Bucuresti (1) Suceava (1) Prahova (1) Timis (1)
Roast coffee	18	Iasi (2) Dolj (1) Constanta (1) Cluj (2) Alba (1) Bucuresti (3) Mures (3) Arges (2) Prahova (2) Timis(1)
French fries	9	Iasi (1) Cluj (1) Gorj (5) Timis (2)
Potato crisps	8	Iasi (1), Dolj (1) Constanta(1) Cluj (1) Prahova(1) Bacau (1) Timis (2)
Bread	15	Iasi (1) Dolj (1) Constanta(1) Cluj (1) Braila (3) Bucuresti (2) Vrancea (2) Bacau (2) Timis (2)
Bakery products	7	Iasi (1) Bucuresti (5) Prahova (1)
Cocoa products	1 3	Dolj (1) Iasi (2) Constanta (1)
Confectionary products	8	Bucuresti (2) Bacau (2) Braila (2) Neamt (2)

Acrylamide analysis

The acrylamide levels from the food products were analyzed by high performance liquid chromatography coupled with a diode array detector (HPLC-UV). The method showed a good sensitivity: LOD and LOQ were 20 and 50 $\mu g\ kg^{-1}$ for coffee, potato and potatoes products and 20 and 25 $\mu g\ kg^{-1}$ for cereals, bakery products and bread respectively. The method used was previously described by Wang et al. (2013).

RESULTS AND DISCUSSIONS

The results obtained for the samples analyzed were reported to the guidance levels for acrylamide based on the EFSA monitoring data which came into force on April 11, 2018 admitted by the European Commission (EC) Recommendation no. 2158/2017 established values (Raffan and Halford, 2019).

The benchmark level which were set by EC (2017/2158/EU) for different foodstuffs products are shown in Table 3.

Table 3. Benchmark level for AA in foodstuffs set by the EC (2017/2158/EU)

Food	Benchmark level [µg/kg]
French fries	500
Potato crisps	750
Soft bread (wheat)	50
Soft bread (other)	100
Breakfast cereals: bran products, whole grain cereals, gun puffed grain	300
Breakfast cereals: wheat and rye based	300
Breakfast cereals: maize, oat, spelt, barley and rice based	150
Biscuits	350
Crackers	400
Crispbread	350
Gingerbread	800
Cereal-based baby foods	40
Baby foods (not cereal based) without prunes	-
Baby foods (not cereal based) with prunes	-
Biscuits and rusks for infants and young children	150
Roast coffee	400
Instant coffee	850
Coffee substitute (cereal-based)	500
Coffee substitute (chicory)	4000

Benchmarks means the performance indicators used to verify the effectiveness of mitigation measures and build on experience and the presence of the substance in comprehensive food categories. The reference cannot be used directly as a reference to assess whether a product can be placed on the market or not.

If the reference levels are exceeded, the operators in the sector should review the mitigation measures applied without delay and adjust processes and controls in order to achieve levels of acrylamide as low as possible below the established reference levels.

The foodstuffs category samples analyzed in 2018 are shown in Table 4.

The samples were classified into 11 categories, based on population consumtion and the risk degree establsihed by ANSVSA within the Surveillance and Control Program in the food safety of non-animal origin field.

According to the ANSVSA data in 2018 only in one foodstuff sample the AA was detected. The foodstuff sample in which AA was detected was a roast coffee sample of which value was under the recommandated values set by EC (2017/2158/EU) as it may be seen from Table 3. So, according to the evaluated data in 2018, the foodstuffs samples analyzed from the Romania market, within the surveillance and control program in the field of food safety of non-animal origin were in agreement with the EC recommendations level for AA values.

Table 4. The AA level in foodstuffs products in 2018

Category	Samples	Level of AA (µg/kg)	Benchmark level 2017 (µg/kg)
Biscuits	4	Undetectable	350
Fooddishes	1	Undetectable	500
Breakfast cereals	11	Undetectable	300
Roast coffee	8 1	Undetectable 225	400
French fries	6	Undetectable	500
Potato crisps	5	Undetectable	750
Food products based on potato	5	Undetectable	500
Bread	4	Undetectable	50
Bakery products	3	Undetectable	50
Cocoa products	1	Undetectable	350
Confectionary products	1	Undetectable	350

In 2019 were evaluated by ANSVSA 88 foodstuffs samples which were classified into 10 categories as it may be seen from the Table 5.

Table 5. The AA level in foodstuffs products in 2019

Category	Samples	Level of AA (µg/kg)	Benchmark level 2017 (µg/kg)
Biscuits	6	Undetectabe	350
Fooddishes	4	Undetectabe	500
	1	< 50	
Breakfast cereals	6	Undetectabe	300
	1	32.77	
	1	172.92	
Roast coffee	12	Undetectabe	400
	1	130.89	
	1	79.88	
	1	97.88	
	1	177.56	
	1	243	
	1	< 50	
French fries	7	undetectabe	500
	2	< 50	
Potato crisps	8	undetectabe	750
Bread	12	undetectabe	50
	2	< 25	
	1	25.6	
Bakery products	6	undetectabe	50
	1	25.39	
Cocoa products	3	undetectabe	350
	1 (cocoa)	264.1	
Confectionary products	8	undetectabe	350

As it may be seen from the data analyzed in 2019 by ANSVSA for foodstuffs samples from the Romania market it may be noticed that no foods samples exceeded the AA level recommended by EC (2017/2158/EU) which are shown in Table 3. Only for 16 foodstuffs samples from 88 ones the AA level was detected. Therefore, AA was detected in 18.18% from the total of foodstuffs samples analyzed. From those, the highest AA level was recorded for a cocoa sample. For the foodstuffs categories analyzed the most samples in which was detected AA was roast coffee, namely from 18 samples analyzed in 6 samples AA was detected. From the 6 roast coffee samples analyzed in which AA wasdetected 4 was in a roasted coffee beans form. If we compared the number of foodstuffs samples analyzed in 2018 year compared to 2019 year from the AA level point of view, depending of the category product type, we noticed increased of the samples number analyzed in 2019 compared to 2018 year. In the Table 6 is shown the ratio level increased for the foodstuffs category samples analyzed in 2018 year compared to the 2019 year.

Table 6. Comparative study between the foodstuffs category samples analyzed in 2018 and 2019 years

Category	Samples 2019	Samples 2018	Ratio increase 2019/2018
Biscuits	6	4	1.5
Fooddishes	5	1	5.00
Breakfast cereals	8	11	0.72
Roastcoffe	18	9	2.00
Frenchfries	9	6	1.5
Potatocrisps	8	5	1.6
Bread	15	4	3.7
Bakeryproducts	7	3	2.3
Cacao products	4	1	4.0
Confectionary products	8	1	8.0

CONCLUSIONS

This study compiles the presence of AA from different foodstuffs from the Romania market in order to monitor the evolution of AA presence in different foodstuffs from the last two years, namely, 2018 and 2019. The study has also a descriptive purpose in order to analyze the number of foodstuffs samples analyzed from Romania market from its AA level content. According to the analyzed data the number of analyzed samples was almost twice higher in 2019 compared to 2018 year. If in the year 2018 the foodstuffs collected for analysis were only from 7 counties from the Romania country, in the 2019 year the number of counties from which were collected foodstuffs samples for analysis has been almost doubled reaching to 16. From the total of 138 samples analyzed in 2018 and 2019, acrylamide was detected in 17 samples. The highest AA levels were found in a cocoa product for which was detected a value of 264.1 µg/kg. From the category of foodstuffs samples analyzed the AA was detected the most in roast coffee samples. However, for no foodstuffs sample analyzed on Romania market the AA level did not exceeded the recommended level set by EC (2017/2158/EU). The results show that in Romania a widespread potential risk from AA for public health occurred in cocoa and coffee products. The study supplied important information's for AA level from different foodstuffs from Romania market for food policy makers, public health experts and consumers.

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