# OCCURRENCE OF ACRYLAMIDE IN SELECTED FOOD PRODUCTS

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**SUMMARY**

*Objective:* Acrylamide is a toxic compound found in occupational and non-occupational environment. It originates from industrial use, technological processes in construction as well as food production, water purification, and laboratory use. Adverse effects including neurotoxicity, probably carcinogenicity, genotoxicity, teratogenicity, and reproductive toxicity were reported in many studies. Our study is focused on acrylamide content in selected food products in the Czech Republic.

*Method:* In this study, we determined acrylamide content in selected samples of commonly consumed food products (potato chips, biscuits, popcorn, corn flakes, breakfast cereals, and baked muesli).

*Results:* The concentration of acrylamide in most samples of potato chips, biscuits and popcorn exceeded the benchmark limits. The acrylamide content in samples of potato chips from one manufacturer in 4 cases out of 5 analysed samples exceeded benchmark limit. On the other hand, no corn flakes sample acrylamide content from the same manufacture exceeded the benchmark limit. With 20 of all analysed samples exceeding the benchmark levels for acrylamide, it can be concluded that 47.6% of samples did not comply with the benchmark level.

*Conclusion:* Dietary intake of frequently consumed food products with the acrylamide content exceeding the recommended comparative value could have adverse effects on the human health. Our study confirmed high amount of acrylamide in some selected food samples. The reduction of dietary burden with acrylamide is possible by complying with the correct technological principles in food preparation.

*Key words:* acrylamide, process contaminants, food technology, dietary burden

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## INTRODUCTION

Acrylamide (ACR) is a food contaminant formed during the Maillard reaction of carbohydrate-rich foods at a temperature above 120 °C (1–4). This compound is classified as a process contaminant with negative effects on human health. Acrylamide affects negatively the nervous and reproductive systems, as well as the prenatal and postnatal human development (5). ACR presence in heat-treated food products was published by many authors (2, 3, 6, 7). The concentration of acrylamide in various types of foods depends on the composition of food (high content of starch, reduced saccharides, presence of amino acids, especially asparagine, content of water), cooking methods, temperature and length of heating processes (baking, frying, and grilling) (4, 5). The highest amount of ACR is formed by Maillard reaction in raw materials with high content of asparagine and glucose when heated to 180 °C. This reaction creates 368 μmol of acrylamide per 1 mol of asparagine. With higher water content in the mixture, the concentration of ACR is almost three times higher. However, at the same temperature only one-sixth of the amount of ACR is formed from methionine (8). Moderate amounts of ACR, 5–50 $\mu g.kg^{-1}$ have been found in heated protein-rich foods, and higher levels 150–4,000 $\mu g.kg^{-1}$ in carbohydrate-rich foods such as beetroot and potatoes. The amount of ACR in food depends on its precursors and on the method of heat treatment, especially temperature and time used for heating. Romani et al. demonstrated how an increase in temperature combined with a longer heating time leads to a rapid increase of ACR content in foods (9).

A large proportion of population is exposed to acrylamide from consumed food. One third of human energy intake is from food contaminated by ACR (10). According to the WHO data, the estimated average daily intake of ACR is 0.3–0.8 $\mu g.kg^{-1}$ per day (10). Mean and 95th percentile dietary ACR exposures across surveys and age groups were estimated at 0.4 to 1.9 $\mu g.kg^{-1}$ body weight (b.w.) per day and 0.6 to 3.4 $\mu g.kg^{-1}$ b.w. per day, respectively (11).

Joint FAO/WHO Expert Committee on Food Additives (JECFA) clarifies the details of the effects on the population's health by publishing data from 17 countries regarding food consumption in these countries. According to this report, the exposure of the whole population to ACR is 1 $\mu g.kg^{-1}$ b.w. per day. For highly exposed group of consumers, who are young people and kids with low body weight, the value can be higher, going up to 4 $\mu g.kg^{-1}$ b.w. per day (12). In most countries, food products such as French

fries, potato chips, coffee, crackers, pastry, and bread are the main contributors to the daily ACR intake (12). The main contributor to total dietary exposure is generally the category "potato fried products" (except potato crisps and snacks) (11).

ACR is a toxic and potentially danger chemical compound. The contamination of the environment by ACR and its occurrence in high concentration in some heat-treated food products can have harmful effects on human health.

The International Agency for Research on Cancer (IARC) classified acrylamide as probably carcinogenic to human, group 2A (13). Friedman reported three major adverse effects in animals, neurotoxicity, developmental toxicity, and carcinogenicity (5). The neurotoxic effects were observed in human, especially from the occupational environment (14). EFSA experts stated ACR in food is a processed contaminant (15). Based on the findings of the calorie content of the US diet, the risk assessment suggests that up to 40% of all foods contain acrylamide (5). Friedman reached the conclusion that children may be more at risk than adults since the amount of ACR consumed per kilogram of body weight is more important in this category (5). Infants, toddlers and other children are the most exposed groups (11).

The populations are chronically exposed, via their diets, to ACR and its harmful effects. Limiting and monitoring the usage of ACR as well as reducing its level in products have therefore been included amongst the priorities of health authorities such the World Health Organization (WHO), the European Food Safety Authority (EFSA), the Food Agriculture Organization (FAO), and others.

The aim of our study was to determine the acrylamide content in the selected samples of commonly available food products from market chain in the Czech Republic (CR).

## MATERIALS AND METHODS

### Experimental Work

In our experimental work we focused on common food products from the market chain in the Czech Republic. The 42 food samples were analysed to determine their ACR content included potato chips, corn flakes, popcorns, biscuits, breakfast cereals for kids, and backed muesli.

The representative part of each sample was analysed by HPLC method. All samples (2 g) were homogenised, 40 µl inner standard of acrylamide-$d_3$ (Sigma-Aldrich) and 40 ml of deionised $H_2O$ were added to each homogenate. After shaking (60 minutes) the samples were transferred to a centrifuge tube (polypropylene) and centrifuged for 20 minutes at 3000 rpm at 10 °C temperature. A volume of 10 ml of supernatant was used for purification (multimode column activated by methanol and water), the eluate was applicated on column ENV + then eluted with a 2 ml mixture methanol: water (60 : 40) and concentrated by evaporation of methanol under nitrogen stream at 40 °C. Approximately 500 µl of each sample was transferred into a glass vial and analysed by LC/MS/MS method, LOD = 3 $\mu g.kg^{-1}$, LOQ = 10 $\mu g.kg^{-1}$. For quantification of the acrylamide content in the samples, a 9-point standard calibration curve was used (0, 5, 10, 50, 100, 500, 1000, 2000, and 5000 µg/L standard of Acrylamide, Fluka).

### Conditions for Analysis

HPLC chromatograph with MS/MS detector (AB Sciex Triple Quad 4500), column Hypercarb 5 µm, 50 × 2.1 mm with precolumn 5 µm, 10 × 2.1 mm.

### Conditions for Detection

Particles 5 µm, mobile phase 0.1% acetic acid in water, 25 °C, flow 0.4 $ml.min^{-1}$, injection 10 µl MS/MS detection: ionization mode ESI+, curtain gas 24, collision gas 8, ion spray voltage 5500, temperature 400 °C, ion source gas 1–30, ion source gas 2–20. Chromatogram record was evaluated by Analyst 1.6.1 (AB Sciex) software.

## RESULTS AND DISCUSSION

The content of ACR in the analysed samples is given in Table 1 and Table 2. In Table 1 are given the results of all analysed food samples. The results confirmed the lowest content of ACR in the breakfast cereals (mean 89 $\mu g.kg^{-1}$). The low ACR content was found in roasted muesli (mean 126 $\mu g.kg^{-1}$) and corn flakes (mean 115 $\mu g.kg^{-1}$). The highest ACR values were determined in potato chips (mean 982 $\mu g.kg^{-1}$), biscuits (mean 679 $\mu g.kg^{-1}$) and popcorns (mean 761 $\mu g.kg^{-1}$).

In 2018, new regulations were accepted in the CR – Commission Regulations (EU) 2017/2158, valid from 11thApril 2018 – establishing mitigation measures and benchmark levels as well as new rules for manufacturers to decrease the acrylamide content in some groups of risky foods (16). According to these regulations, the manufacturers shall monitor the ACR value in their products and adjust the technological conditions to comply with the benchmark levels of ACR. When the content of ACR exceeds the benchmark levels, higher frequency of samplings and adjustments of the production conditions are recommended to achieve the benchmark limit.

According to these new Commission Regulations (16), 20 of all our analysed samples exceeded the benchmark levels for acrylamide, accounting for 47.6% non-compliant samples (Table 1). The benchmark limits accepted and valid from 11 April 2018 (16) are stricter than indicative values for the same food products (Table 1), according to Commission Recommendation (2013/647 /EU) of 8 November 2013, valid earlier (17).

Our results are consistent with the results widely published by other authors for the same food products (18). Results of our samples from 1 manufacturer (Table 2) in 4 cases out of 5 samples of potato chips exceeded the benchmark limit, on the other hand, no sample of corn flakes exceeded limit for this food product. Potato chips are included amongst risky foods with high content of ACR precursors. Technological conditions for the production of corn flakes from the same manufacturer (the composition of raw materials, temperature, and time for heating) are probably well balanced since ACR values did not exceed the benchmark limit for this food product. It is important to state that all samples were taken and analysed earlier than the new regulations (16) were accepted (Table 1 and 2).

Sorgel et al. reported that 10 to 50% of dietary acrylamide in pregnant women is transferred via blood through the placenta to the foetus. Breast milk was found to contain up to 18.8 µg/L of

***Table 1.*** *Acrylamide content in food samples (N = 42)*

| Samples (n) | Acrylamide (μg.kg$^{-1}$) | Mean ACR (μg.kg$^{-1}$) | SD (μg.kg$^{-1}$) | Minimum (μg.kg$^{-1}$) | Maximum (μg.kg$^{-1}$) | *Indicative value (μg.kg$^{-1}$) | **Benchmark level (μg.kg$^{-1}$) | Limit exceeded (%) |
|---|---|---|---|---|---|---|---|---|
| Potato chips (8) | 790 | 982 | 424.4 | 83 | 1,550 | 1,000 | 750 | 87.5 |
| | 930 | | | | | | | |
| | 986 | | | | | | | |
| | 1,480 | | | | | | | |
| | 83 | | | | | | | |
| | 921 | | | | | | | |
| | 1,120 | | | | | | | |
| | 1,550 | | | | | | | |
| Biscuits (2) | 571 | 679 | 108.0 | 571 | 787 | 500 | 350 | 100 |
| | 787 | | | | | | | |
| Popcorn (8) | 554 | 761 | 304.4 | 433 | 1,410 | 200 | 150 | 100 |
| | 433 | | | | | | | |
| | 460 | | | | | | | |
| | 785 | | | | | | | |
| | 797 | | | | | | | |
| | 1,410 | | | | | | | |
| | 1,010 | | | | | | | |
| | 638 | | | | | | | |
| Corn flakes (9) | 117 | 115 | 42.2 | 50 | 191 | 200 | 150 | 33.3 |
| | 102 | | | | | | | |
| | 129 | | | | | | | |
| | 103 | | | | | | | |
| | 50 | | | | | | | |
| | 55 | | | | | | | |
| | 157 | | | | | | | |
| | 127 | | | | | | | |
| | 191 | | | | | | | |
| Breakfast cereals (10) | 136 | 89 | 64.1 | < 5 | 253 | 400 | 300 | 0 |
| | 55 | | | | | | | |
| | 45 | | | | | | | |
| | 72 | | | | | | | |
| | 69 | | | | | | | |
| | 112 | | | | | | | |
| | < 5 | | | | | | | |
| | < 5 | | | | | | | |
| | 147 | | | | | | | |
| | 253 | | | | | | | |
| Roasted muesli (5) | 50 | 126 | 82.5 | 50 | 279 | 400 | 300 | 0 |
| | 73 | | | | | | | |
| | 84 | | | | | | | |
| | 142 | | | | | | | |
| | 279 | | | | | | | |

*Commission recommendation (2013/647/EU) – indicative values
**Commission regulation (EU) 2017/2158 – benchmark levels

***Table 2.*** *Acrylamide in food samples produced by one company*

| Food sample | Sample number | Acrylamide (μg.kg$^{-1}$) | Mean ACR (μg.kg$^{-1}$) | SD (μg.kg$^{-1}$) | Minimum (μg.kg$^{-1}$) | Maximum (μg.kg$^{-1}$) | *Benchmark level (μg.kg$^{-1}$) |
|---|---|---|---|---|---|---|---|
| Potato chips | 1 | 790 | 853.8 | 450.25 | 83 | 1,480 | 750 |
| | 2 | 930 | | | | | |
| | 3 | 986 | | | | | |
| | 4 | 1,480 | | | | | |
| | 5 | 83 | | | | | |
| Corn flakes | 19 | 117 | 112.75 | 11.10 | 102 | 129 | 150 |
| | 20 | 102 | | | | | |
| | 21 | 129 | | | | | |
| | 22 | 103 | | | | | |

Samples from the same manufacturer, different batches of foodstuffs.
*Commission regulation (EU) 2017/2158

acrylamide. Because it is water-soluble, acrylamide can cross both placental and blood-brain barriers, the authors suggest protecting the foetus. The pregnant woman should not consume food products with high acrylamide content (19). According to Kromerová (20), the highest average levels of acrylamide in food samples collected in the Slovak Republic (SR) between 2006 and 2014 and analysed by accredited laboratories (database of the Food Research Institute) were found in roasted coffee (707.8 μg.kg$^{-1}$), potato products (409.8 μg.kg$^{-1}$), durable pastry (318.6 μg.kg$^{-1}$), and other cereal products (302.6 μg.kg$^{-1}$). The category "durable pastry" includes e.g., biscuits, rusks, sponge cakes and the category "other cereal products" includes e.g., breakfast cereals, crisps, puffed rice, and cereal porridge. As regards food intended to infant and young children, in total, for the period 2013–2015, 164 foods for infants and young children were laboratory analysed by the Regional Republic Health Authorities for acrylamide in the SR. Arithmetic mean of the amount of ACR (middle bound) in cereal-based foods and baby food intended for infants and young children aged 6 month to 3 years for the period 2013–2015 is 7.5 μg.kg$^{-1}$. The indicative values laid down in Commission Recommendation 2013/647/EU (17), for individual categories of food for infants and young children aged 6 month to 3 years for the period 2013–2015 were not exceeded and range from 4 to 15% of these indicative values. For the calculation of the risk assessment of exposure to acrylamide from food for infants and young children, the results of food samples from 2013–2015 were used. The average daily intake of acrylamide from food for the average citizen in the SR for the period 2006–2014 is 249 μg.kg$^{-1}$ b.w. per day. The average daily intake of acrylamide from cereal-based foods for infants and young children (average consumption) and specified foods for infants is 0.6 μg.kg$^{-1}$ b.w.per day, for young children 0.2 μg.kg$^{-1}$ b.w. per day (20).

ACR is potentially dangerous to human health. It is necessary to permanently reduce its amount in food. This would be possible through strict compliance with the regulations and adequate production conditions in both manufacturers and home-made preparations. Non-legislative measures can also contribute to this, e.g. "acrylamide toolbox" with widely available information how to reduce ACR content in food preparation (21).

## CONCLUSION

Our results were an example of commonly consumed foods and some popular delicacies from the market chain in the Czech Republic. Compared to the current legislation almost one half of these samples did not comply with benchmark levels. The newly adopted legislation is designed to motivate food manufacturers to reduce acrylamide content in their food products. This is a way to reduce the dietary burden of the population with this process contaminant.

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**Conflict of Interest**
None declared

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