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COPPER AND CHROMIUM LEVELS IN CANDIES AND CANDY PACKAGES

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Abstract. *Various plastic materials are used in candies packaging and most of them have inorganic pigments used in inks and plastics that can contaminate the food. Heavy metals, such as copper and chromium can pose health problems. Aim of the study was to determine the levels of Cu and Cr from different samples of candies and also from candy packages using standard addition method and flame atomic absorption spectrometry. The determination of the total metals concentration in the investigated samples was done after sample mineralization step, using two methods of digestion: the acid digestion method and the dry ash method. The calibration curves were obtained using solutions prepared by diluting stock solutions with deionized water to the desired concentrations. The recovery studies were carried out in triplicate for replicates of spiked samples. Results were satisfactory, with recoveries between 98 and 101% indicating the high accuracy of the method. The smaller concentrations of metals were observed in candy packages than in candies. The average highest level of Cu was recorded in apple candies. To appraise the health risk associated with heavy metal contamination of the studied products, estimated daily intake of metals (EDIM) was calculated. Data generated during the study was processed by means of various statistical tests, such as t test and the One-Way ANOVA. Results show that the mean concentration for all the elements was significantly different (α= 0.05) for all samples. The concentrations of Cu and Cr in candies and their packaging do not exceed the permissible limits.*

Keywords: *Cu, Cr, candies, candy packages.*

1. INTRODUCTION

Packaging affects how the food is perceived and experienced during buying, product usage and consumption. During the buying process, packaging design plays a role in identifying the category and brand to which the product belongs and in conferring meaning or in reinforcing existing associations to the product. The shape and color of packaging play an important role on retail shelves, because packaging information influenced food choice [1].

Candies are the favorite food items of children. They are the most vulnerable age group to any kind of contamination in the food chain [2]. Colorful packages of candies have color inks on the outer cover. There are many kinds of metal based inorganic pigments that can migrate from the the surface of the package to the food surface. There are several

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scientific research on migration of contaminants by one of the following mechanism: blocking, rubbing, peeling and diffusion [3-6].

Copper is an essential element required as a structural component of numerous metalloenzymes. There is a lack of information on the potential adverse health effects from chronic exposure to this metal [7-12].

Occupational exposure to chromium has been associated with increased lung cancer incidence in many epidemiological studies [13-15].

Aim of the study was to determine the levels of Cu and Cr from candies made by an international brand and candy packages using the flame atomic absorption spectrometry. In order to increase the sensitivity of the analytical determination was used the method of the standard addition, too [16]. Another goal of the study was to calculate estimated daily intake of studied metals (EDIM) and to process data generated during the study with statistical tests, such as t test and the One-Way ANOVA.

2. MATERIALS AND METHODS

2.1. MATERIALS

The studied samples, purchased from the local market were 5 samples of candies from an international brand with different flavors: strawberry (sample 1), apple (sample 2), cola (sample 3), cherries (sample 4) and orange (sample 5) and their packages (samples 6 to 10).

The determination of the total metals concentration in the investigated samples was done after sample mineralization step, using two methods of digestion: the acid digestion method and the dry ash method.

Candy and candy packages samples were digested by the acid digestion method with a Digesdahl device. Samples were weighted to approximately 0.5 g in a vessel, to which 8.0 ml HNO_3 and 10 ml H_2O_2 were added. The temperature of the instrument was increased from room temperature to 150 °C. Sample blanks were performed with empty vessel, to which 8.0 ml HNO_3 and 10 ml H_2O_2 were added.

The second method was the dry ash method at 500 °C for 6 hours. Samples were weighted to approximately 0.2 g in ceramic crucible. After dry digestion, samples were diluted with HNO_3. Sample blanks were also performed with empty ceramic crucible.

All reagents were of analytical-reagent grade (Merck) and all solutions were prepared using deionized water. The concentration ranges for standard solutions were typically between 50 and 5000 µg/l.

2.2. METHODS

In order to increase the sensitivity of the analytical determination was used the method of the standard addition. The method is applied when the compounds to be determined are in low concentration and the chemical matrix is not constant in the samples to be analyzed. The precision of Cu and Cr determination was evaluated under the optimum conditions; were studied the effects of *p*H, temperature, complexation time, reagent amounts in a previous paper [17]. For the measurement of studied metals absorption spectra was used a DR-2800 HACH LANGE spectrophotometer.

The copper and chromium levels were determined also by the atomic absorption method using a GBC Avanta PM atomic absorption spectrometer. An air-acetylene flame was used for all elements. Hollow cathode lamps were used as radiation source. The acetylene was of 99.999% purity at a flow rate 1.8-2.0 L/min. Analyses were made triplicate, and the mean values were reported.

The calibration curves were obtained using solutions prepared by diluting stock solutions with deionized water to the desired concentrations. The detection and quantification limits were evaluated from calibration curves. LOD and LOQ values for each calibration line were obtained and calculated using the equations: $(3 \cdot s_a - a)/b$ and $(10 \cdot s_a - a)/b$, respectively, where b is the slope of the calibration curve and s_a is the standard deviation of intercept of regression equation. Good linearity was observed with coefficients of determination, R^2, exceeding 0.9992.

The working concentration range was established by analyzing the lowest and the highest concentration values of the proposed concentration range, ten times each of them. The recovery studies were carried out in triplicate for replicates of spiked samples. Results were satisfactory, with recoveries between 98 and 101% indicating the high accuracy of the method. These percentage recoveries ranged within the limits imposed by the Horwitz equation (85-110%) for the established concentration range.

3. RESULTS AND DISCUSSION

3.1. RESULTS

The results for Cu and Cr in candies using standard addition method are presented in Table 1.

Table 1. Levels of Cu and Cr in candies using standard addition method.

Metals [mg/kg] \ Sample no.	1	2	3	4	5
Acid digestion					
Cu	<LOD	<LOD	<LOD	<LOD	<LOD
Cr	0.110	<LOD	0.003	0.013	<LOD
Dry ash digestion					
Cu	0.325	1.651	0.838	<LOD	1.012
Cr	0.208	0.058	0.039	0.138	<LOD

The results for Cu and Cr in candy packages using standard addition method are presented in Table 2.

Table 2. Levels of Cu and Cr in candy packages using standard addition method.

Sample no. / Metals [mg/kg]	6	7	8	9	10
Acid digestion					
Cu	<LOD	<LOD	<LOD	<LOD	<LOD
Cr	0.0009	0.003	0.003	<LOD	<LOD
Dry ash digestion					
Cu	0.058	0.079	0.021	<LOD	<LOD
Cr	0.001	0.144	0.104	0.008	<LOD

The results for Cu and Cr in candies and candy packages using FAAS method are presented in Tables 3 and 4.

Table 3. Levels of Cu and Cr in candies using FAAS method.

Sample no. / Metals [mg/kg]	1	2	3	4	5
Acid digestion					
Cu	0.062	0.825	0.386	<LOD	0.426
Cr	0.271	0.021	0.011	0.020	0.014
Dry ash digestion					
Cu	0.516	1.723	1.142	0.367	1.338
Cr	0.524	0.207	0.117	0.356	0.287

Table 4. Levels of Cu and Cr in candy packages using FAAS method.

Sample no. / Metals [mg/kg]	6	7	8	9	10
Acid digestion					
Cu	<LOD	<LOD	<LOD	<LOD	<LOD
Cr	0.072	0.086	0.053	0.035	0.021
Dry ash digestion					
Cu	0.156	0.107	0.047	0.025	0.038
Cr	0.068	0.193	0.174	0.073	0.039

3.2. DISCUSSION

Scientific results indicate that heavy metals could migrate from the printed outer packages to food. Most of the pigments of the printing inks were based on metallic compounds, therefore harmful metals such as Cu and Cr have been prohibited by law or strictly regulated in most of the countries from being used in food packaging [3].

Cr (III) and Cu are considered as essential trace nutrients; they are required for normal health function and development. Chromium is found at low levels in most biological materials. Chromium (III) works as co-factor for insulin action, so is an essential element for normal glucose metabolism, while Cr (VI) it is mentioned as carcinogenic. Cu is an essential element of several enzymes; it functions as a biocatalyst and it is necessary for body pigmentation.

Therefore, Cu and Cr composition of candies is of interest because of their essential nature. Comparison of the results obtained in this study with other reported studies in literature on metals in candies are difficult, since limited studies are available. The results reveal that concentrations of metals are much lower when the acid digestion was used. The smaller concentrations of metals were observed in candy packages than in candies.

Data generated during the study was processed by means of various statistical tests, such as t test and the One-Way ANOVA. The chosen significance level was 0.05 (equivalent to 5%). Results show that the mean concentration for all the elements was significantly different ($\alpha = 0.05$) for all samples. The Cr concentrations obtained for candies in this study were very small, some were below limit of detection and the maximum concentration for Cr was obtained for strawberry candy (0.524 mg/kg). As it can be seen in Tables 1 and 3, the obtained concentrations were lower than the range of those marketed in Korea (136.9 mg/kg to 1429.3 mg/kg) [3], Turkey (0.74 µg/g – 6.27 µg/g) [13] and similar with those obtained in Mexico (0.088 µg/g to 0.39 µg/g) [14]. From the results, the average highest level of Cu was recorded in apple candies but is lower then the permissible level of Cu in candy (10 µg/g, according to Turkish Standards [20]). Also, are lower than those reported in candies marketed in Mexico [19] and Turkey [18] (2.74 µg/g, respectively 2.5 µg/g).

In candy packages, Cu and Cr concentrations are very small. When for mineralization step was used acid digesion, Cu obtained was under the detection limit. The maximum concentration for Cu was obtained for strawberry candies packages (0.156 mg/kg), when for mineralization step was used dry ash digestion. Cr concentrations in candies packages were in the range: 0.039-0.193 mg/kg using dry ash digestion and FAAS method.

To appraise the health risk associated with heavy metal contamination of the studied products, estimated daily intake of metals (EDIM) was calculated as shown below [21]. The daily intake of metals depends on both the metal concentration in food and the daily food consumption. In addition, the body weight of the human can influence the tolerance of contaminants.

$$EDIM = \frac{C_{metal} x W_{food}}{B_w}$$

where,

- C_{metal} is the concentration of heavy metals in the samples;
- W_{food} represents the daily average weight of the sample consumed (assumed 20 g of each sample is taken by an individual);
- Bw is the body weight assuming that samples are consumed by children between the ages of 3-6 years with average body weight of 30 kg.

The estimated daily intake of metals (EDIM) value for the studied products is shown in Table 5.

Table 5. Estimated daily intake of metals (µg/day) in study samples (n=4)

Sample		Min	Max		Min	Max
1	**Cu**	0.040	0.340	**Cr**	0.180	0.350
2		0.550	1.140		0.010	0.130
3		0.230	0.760		0.007	0.800
4		0.000	0.240		0.010	0.240
5		0.280	0.890		0.009	0.190

The EDIMs values of Cu and Cr showed that all samples are below than 1.4 mg/day recommended limit for Cu, respectively 0.1mg/day recommended limit for Cr in UK [22].

4. CONCLUSIONS

The candy samples from an international brand were collected from the local market in Romania, and the concentration of Cu and Cr were quantified. Small levels of copper and chromium were found in candies and candy packages. The concentrations of Cu and Cr in candies and their packaging do not exceed the permissible limits. Moreover, it could be concluded that our estimated daily intake for the studied elements are below the recommended limits. Thus, consume average amounts of these candies seem not to pose a health-risk for the consumer.

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