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ORIGINAL PAPER

ASSESSMENT OF THE CONTENT OF CHROMIUM, NICKEL AND COBALT IN CHOCOLATE PRODUCTS WITH DIFFERENT COCOA MASS CONTENT AVAILABLE ON THE POLISH MARKET*

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Abstract

Europe is the world's largest market for chocolate producers and exporters (Germany 17.0%, Belgium 11.0%, Italy 7.3%, the Netherlands 7.0% and Poland with 6.3% of the world market share). Chocolate and chocolate-based products are popular, eagerly eaten and, as research shows, have a beneficial effect on the human body. However, it should be remembered that they are also a source of trace elements, any excess of which can be harmful to the consumer's health. The aim of the study was to determine the content of chromium, nickel and cobalt in chocolate products with different cocoa mass content available on the Polish market. Sixty-three types of chocolate and eight chocolate creams with different cocoa mass content (0% to 90%) were used. The samples were mineralized, and the content of selected elements was analyzed by atomic absorption using a spectrometer. The highest content of the examined elements was recorded for dark chocolate: 1.71, 5.61 and 0.37 mg kg^{-1} of Cr, Ni and Co, respectively. The content of all tested elements, in each interpreted case, increased with the amount of cocoa mass in the tested product. It has been shown that consumption of a daily portion of the analyzed chocolate products suggested by the manufacturer does not cause a risk of exceeding the supply of the tolerated daily intake of the analyzed elements, regardless of the cocoa mass content in the composition of the analyzed products.

Keywords: atomic spectrometry, food, chocolate, cocoa, nutritional value, trace elements.

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INTRODUCTION

Chromium, nickel and cobalt are trace elements that occur both in nature and in living organisms; they are essential for the proper functioning of the human body, but in larger quantities they can have a toxic effect (Iwegbue 2011, Kośla et al. 2018, Drzeżdżon 2019, Salama 2019).

Chromium is important in the metabolism of proteins, carbohydrates and lipids (especially cholesterol). Chromium accelerates blood clotting and causes greater activity of tyrosine kinase and membrane tyrosine phosphatase of insulin receptors, which results in more efficient absorption and use of glucose by the body's cells (Thor et al. 2011, Lewicki et al. 2013, Yasarawan et al. 2013, Lewicki et al. 2014, Ratajczak et al. 2015, Salama 2019).

Chromium also participates in antioxidant processes, RNA and DNA synthesis, and affects the secretion of hormones and some vitamins (Thor 2011, Lewicki et al. 2014, Kośla 2018). Chronic exposure to chromium can cause allergies and damage to the kidneys, lungs and liver, reduce the activity of the immune system, and contribute to the development of carcinogenic diseases (De l'Anses 2011, Dhali et al. 2013, Drzeżdżon et al. 2019).

Nickel is required for the activation of enzymes and lipids, and it is involved in the stabilization of nucleic acid structures and increases hormonal activity. However, in higher doses, it may cause contact allergies or have a carcinogenic effect (Czarnobielska et al. 2011, IARC 2012, Cheong et al. 2014, Wojciechowaska et al. 2016). Nickel and its compounds were classified as a factor causing cancer of the lungs, nasal cavity and paranasal sinuses after inhalation (IARC 2012). Excess nickel in the human body has a negative effect on the digestive, hematological, neurological and immune systems; it penetrates the placental barrier and is toxic to the fetus even at low doses (EFSA 2015).

Estimated Ni consumption by humans is about 300–900 µg per day, of which only 1–10% is absorbed by the body (Dara 2006, Sharma 2007). This element accumulates in the highest concentration in the intestines, lungs, liver and kidneys. The main source of nickel in the diet are plant products, and sweet products mainly based on chocolate (Cubadda 2020, Suomi et al. 2020). According to Sirot et al., chocolate-based products contributed 30-60% of the nickel exposure in the diet of children aged 13-36 months (Sirot et al. 2018).

Cobalt in the human body is a component of many enzymes, and it participates in the metabolism of proteins, folates, fatty acids and in the production of cobalamin – B_{12} (De l'Anses 2011, Winczewska-Wiktor et al. 2012, Reutz et al. 2013). Standards for cobalt consumption for the Polish population depend on age and range from 0.9 µg per day for children aged 1-3 years to 2.8 µg per day for lactating women (Jarosz 2020).

Excess cobalt is mainly accumulated in the liver, kidneys and bones. Eighty percent of consumed cobalt is excreted with urine, and its excess in the body can cause, among other things, lung disease, dermatitis, increased blood pressure, nausea, vomiting, diarrhea, dizziness, cardiomyopathy, slowed breathing, hyperglycemia, neurotoxicological dysfunction, changes in genes and cancer (Said 2012, Jammal et al. 2013, Gavars et al. 2019, Salama 2019, Jarosz 2020, Urbanski 2020). Cobalt deficiency is also dangerous, as a lack of vitamin B_{12} in the body can cause anemia and disorders of the nervous, skeletal and digestive systems. Cobalt is necessary for the metabolism of folates and fatty acids (Carmel 2009, De l'Anses 2011).

Europe is the world's largest market for chocolate producers and exporters (Germany 17.0%, Belgium 11.0%, Italy 7.3%, the Netherlands 7.0% and Poland with 6.3% of the world market share) (CBI 2020). Consumption of just 2 g of dark chocolate a day for 6 months prevents DNA damage, improves the nucleus integrity of cells and blood biochemical parameters (total cholesterol, triglycerides and LDL-cholesterol level in blood) and anthropometric parameters, waist circumference (Leyva-Soto et al. 2018). Chocolate and chocolate-based products are popular, eagerly eaten and, as research shows, have a beneficial effect on the human body. However, it should be remembered that they are also a source of trace elements, any excess of which can be harmful to the consumer's health. In the literature, there are few publications in which trace elements such as Cr, Ni and Co in chocolate products are studied, hence our decision to check how their content in products containing various amounts of cocoa mass in their composition is classified. Therefore, the aim of the research was to determine the content of selected elements (Cr, Ni, Co) in chocolate products available on the Polish market, which contain different amounts of cocoa mass in their composition.

MATERIALS AND METHODS

Sixty-three types of chocolate and eight chocolate creams with different cocoa mass content (from 0% to 90%) were used for the study, in which the content of chromium, nickel and cobalt was determined. The products were divided into subgroups taking into account the content of cocoa mass in the composition, according to Directive 2000/36 /WE of 23 June 2000. The samples fell into six groups: white chocolate with cocoa mass content 0% – 7 samples (made in Poland – 5 and Germany – 2), milk chocolate with cocoa mass content from 25% to 34% – 13 samples (made in Poland – 9, Germany – 1, France – 1, Spain – 1 and Switzerland – 1), dessert chocolate with cocoa mass content from 36% to 50% – 6 samples (made in Poland – 3, Germany – 1, Italy – 1 and Spain – 1), dark chocolate with cocoa mass 60% to 90% – 23 samples (made in Poland – 10, France – 7, Switzerland – 2, Germany – 1, Italy – 1, Ukraine – 1 and Spain – 1), chocolate with nuts and cocoa mass content from 25% to 60% – 14 samples (made in Poland – 10, Germany – 2, Switzerland – 1 and France – 1) and chocolate creams with cocoa content from 4% to 6% – 8 samples (made in Germany – 5, Poland – 2 and United Kingdom – 1). The samples for testing were selected on the basis of data about the amount and types of products most often consumed, containing various amounts of cocoa liquor in the composition. During the research, no differences in the content of chromium, nickel and cadmium in the products were observed relative to their place of production. The research was conducted at the accredited Laboratory of Food Research, which belongs to the Faculty of Biotechnology and Food Sciences at the University of Life Sciences in Wroclaw.

In order to determine the content of selected elements, the samples were mineralized using the dry method. To obtain uniform chocolate samples, each chocolate was thoroughly ground and mixed; as for chocolate cream, between 7.00 and 10.00 g of each product was weighed in duplicate to the nearest 0.001 g. Homogeneous samples were pre-ashed on a heating plate, then mineralized in a muffle furnace for 8 h. The ash was dissolved in about 2 cm^3 of nitric acid (HNO_3; Chemprum, 65%, pure p.a.) and burned up for another 2 h. The minerals were quantitatively transferred to 10 cm^3 measuring vessels with 2 M HNO_3 solution (PN-EN 14082:2004).

The content of selected elements was examined by the atomic absorption method on a spectrometer (SpectrAA 240FS, Varina, Australia). The correctness of the determinations was verified with the certified reference material BCR-191 Brown bread. The uncertainty of measurements did not differ from the reference material by more than 5%. Chromium was determined at a wavelength of λ = 357.9 nm, Ni at λ = 232.0 nm and Co at λ = 240.7 nm, within the range of the standard curve 0-5.0 μg mL^{-1} (Flame Atomic ... 1989).

The results were statistically analyzed in the Statistica 13.3 program. First, the normality of the distribution of the tested samples was checked using the Shapiro–Wilk test. No normal and equal distributions of variances were obtained in groups with different concentrations of Ni, Co and Cr. Therefore, in order to determine the effect of the type of chocolate (grouping variable) on the content of the examined elements (dependent variable), the Kruskal-Wallis and median tests were performed. The significance level was α = 0.05.

RESULTS AND DISCUSSION

The results of the Kruskal-Wallis and median tests together with standard deviations determining the effect of the type of chocolate on the content of chromium, nickel and cobalt in the tested product are presented in Table 1. Based on our analysis of the data in Table 1, it was found that the median of the examined elements in each case was the highest for dark chocolate: 1.71, 5.61 and 0.37 mg kg^{-1} for Cr, Ni and Co, respectively.

Table 1

Content of tested elements in various types of chocolate

Element	Type of chocolate	n	Mean (mg kg^{-1} of the product)	SD	Median	Lower quartile	Upper quartile
Cr	white	14	0.532	0.325	0.436^{ab}	0.371	0.529
	milk	26	0.849	0.643	0.619^{b}	0.451	1.051
	dessert	12	1.309	0.412	1.394^{bc}	1.181	1.555
	dark	46	1.909	0.819	1.714^{c}	1.241	2.446
	with nuts	28	0.336	0.256	0.257^{a}	0.183	0.386
	cream	16	0.263	0.124	0.246^{a}	0.136	0.359
Ni	white	14	0.087	0.065	0.093^{a}	0.038	0.130
	milk	26	0.720	0.305	0.759^{a}	0.484	0.908
	dessert	12	3.111	1.291	3.127^{bc}	2.669	3.912
	dark	46	5.693	1.330	5.609^{c}	4.867	6.369
	with nuts	28	1.731	0.911	1.347^{b}	1.082	2.169
	cream	16	0.858	0.365	0.759^{ab}	0.655	1.093
Co	white	14	0.000	0.000	0.000^{ab}	0.000	0.000
	milk	26	0.000	0.000	0.000^{a}	0.000	0.000
	dessert	12	0.186	0.102	0.197^{cd}	0.144	0.269
	dark	46	0.387	0.168	0.374^{d}	0.293	0.506
	with nuts	28	0.085	0.094	0.056^{bc}	0.000	0.162
	cream	16	0.033	0.029	0.029^{abc}	0.009	0.048

Key: the table shows mean values ± standard deviations (SD); a, b, c, d – statistically homogeneous groups ($p \leq 0.05$); n – number of repetitions.

The relationship between the amount of the analyzed elements and the percentage of cocoa mass in the product composition is shown by linear regression in Figure 1.

Figure 1 shows that the content of the tested elements in the analyzed products increased with the amount of cocoa mass in their composition. This was confirmed by a high correlation of the linear regression coefficient R, which was 0.92 for nickel, 0.79 for cobalt, and 0.64 for chromium.

The median amount of chromium ranged from 0.25 mg kg^{-1} (chocolate cream) to 1.71 mg kg^{-1} (dark chocolate). The lowest median for the chromium content was recorded for chocolate cream (0.25 mg kg^{-1}) and chocolate with nuts (0.26 mg kg^{-1}); these values were significantly lower than the amount of chromium contained in milk (0.62 mg kg^{-1}), dessert (1.39 mg kg^{-1}) and dark (1.71 mg kg^{-1}) chocolate (Table 1).

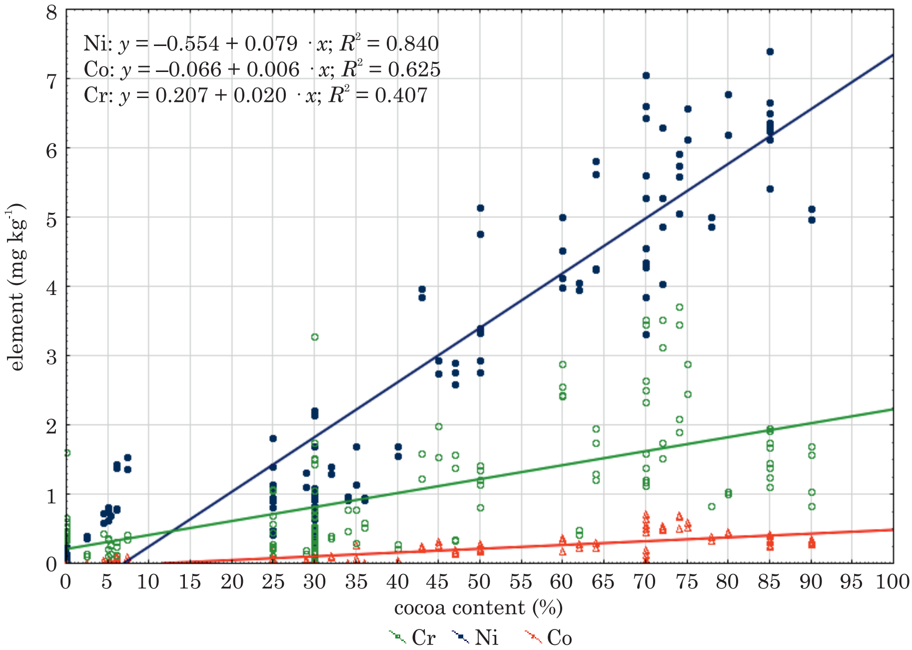

Fig. 1. Content of tested elements in the analyzed products depending on the percentage of cocoa

Sager (2012) obtained similar values for chromium in milk chocolate (0.65 mg kg^{-1} of product), while the value in dessert chocolate was slightly lower (1.03 mg Cr kg^{-1}) than in our experiment. In dark chocolate (up to 70% cocoa content), Sager observed chromium at the level of 1.36 mg kg^{-1}, and in cocoa 1.47 mg kg^{-1} of the product (Sager 2012). The amount of chromium noted by Sager (2012) in dark chocolate and cocoa is similar to the results obtained in our research for dessert chocolate, but lower than the value determined in dark chocolate – which could be due to the different cocoa mass content in the tested dark chocolate. In a report by EFSA (2014), products containing a large amount of cocoa have a chromium content of 1.43 mg kg^{-1} of the product. The legislator does not define the maximum allowable dose of chromium and nickel in food. Due to the toxic effects of Cr on the human body, the CONTAM Panel established the tolerable daily intake (TDI) of chromium for chronic oral exposure at 300 μg Cr kg^{-1} body weight (EFSA 2014, EFSA 2015), while for nickel (III) it is 2.8 Ni μg kg^{-1} body weight (EFSA 2015). Based on dietary and age groups, the EFSA reports that chronic chromium exposure from the diet ranges from 0.6 (minimum lower bound, LB) to 5.6 μg kg^{-1} body weight per day (maximum upper bound, UB), while for nickel it is 2.0 to 13.1 μg kg^{-1} body weight per day. According to the EFSA, children and adolescents who consume significant amounts of chocolate and chocolate-based products are exposed to high levels of nickel intake in food products (EFSA 2014, 2015).

The median amount of nickel in the tested samples ranged from 0.09 mg kg^{-1} (in white chocolate) to 5.61 mg kg^{-1} (in dark chocolate). The lowest average nickel content was observed in white and milk chocolate (0.09 and 0.76 mg kg^{-1}, respectively)and was significantly lower than the nickel content in dark chocolate (62 times), dessert chocolate (35 times), and chocolate with nuts (15 times). A statistically significant difference was observed in the nickel content between dark chocolate and the other products: white chocolate, milk chocolate, chocolate with nuts and chocolate cream (Table 1).

In our research, the highest content of nickel and chromium was recorded in dark chocolate (5.61 and 1.71 mg kg^{-1}, respectively) – Table 1, which means that consumption of about 20 g daily dose (suggested by the producers) of such chocolate daily by a person weighing 60 kg will deliver 114 μg Ni and 38 μg Cr to the body. This amount, in relation to the recommended TDI (Ni = 2.8 μg kg^{-1} body weight; Cr = 300 μg kg^{-1} body weight), does not exceed the quantity that is safe for health (EFSA 2014, EFSA 2015).

Other scientists noted similar amounts of nickel in chocolate (up to 6 mg kg^{-1}), compared to observed 12 mg Ni kg^{-1} of product in cocoa (Sharma 2007, Ščančar et al. 2013). Leblanc et al. (2005) reported an average Ni level in chocolate of 0.63 mg kg^{-1}. Unfortunately, these studies did not provide information about the cocoa mass content in the products analyzed. In this research, white and milk chocolate, which are characterized by low or no cocoa mass, did not contain cobalt, and a significantly lower average cobalt content was observed than in dessert and dark chocolate. In our experiment, an increase in the content of nickel and cobalt was observed along with an increase in the amount of cocoa mass in the composition of the tested products (Figure 1). Sager (2012) noted a Co content of 0.09 mg kg^{-1} in milk chocolate, while in our research a similar cobalt content (0.06 mg) was recorded in chocolate with nuts. Moreover, the highest cobalt content was observed in dark chocolate (0.37 mg kg^{-1}), namely 12 times higher than that measured in chocolate cream (Table 1). Sager (2012) reported a slightly higher Co content in dark chocolate (0.42 mg kg^{-1}), and a slightly lower Co content in dessert chocolate (0.24 mg kg^{-1}). In a French food research report, chocolate has a cobalt content of 0.14 mg kg^{-1} wet weight (De l'Anses 2011). Other scientists reported the cobalt content in chocolate as 0.05 mg kg^{-1} (Biego et al. 1998, Leblanc 2004). The authors of these studies did not provide information on the cocoa mass content in the tested products. In this research, the cobalt content was lower than that reported by other scientists for white chocolate, milk chocolate, chocolate containing nuts and chocolate cream, and higher for dessert and dark chocolate, which could be caused by different cocoa mass content and different origin of the products analyzed.

In our research, a statistically significant difference was found between the cobalt content in dark chocolate and that in white chocolate, milk chocolate, chocolate with nuts and chocolate cream. No statistically significant differences were found between the cobalt content in dark and dessert chocolates and between dark and nut chocolates (Table 1).

Several limitations were observed in this study. One is a lack of comparison of the results obtained with those for a non-matrix reference material. The number of chocolates in the groups and the number of products in each group should be standardized. It also seems justified to test different batches of products from selected producers.

CONCLUSIONS

Based on our analysis of the results, it was found that the content of all tested elements, in each interpreted case, increased with the amount of cocoa mass in the tested product. A statistically significant difference was also observed in the content of Ni, Cr and Co in dark chocolate compared to white chocolate, milk chocolate, chocolate with nuts and chocolate cream. It has been shown that consumption of the daily portion of the analyzed chocolate products, suggested by the manufacturer, does not cause a risk of exceeding the supply of the tolerated daily intake of the analyzed elements, regardless of the cocoa mass content in the composition of the analyzed products.

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