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DETERMINATION OF CHROMIUM CONTENT OF FOOD AND BEVERAGES OF PLANT ORIGIN*

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Key words: chromium, fruits, vegetables, wheat, beverages, ZETAAS

Chromium content of some fresh fruits, vegetables and wheat grains collected in 2001 in three various agricultural areas of Poland was determined in the study. Juices, wines and beers purchased on the domestic market were also examined. A total of 272 samples were analysed. Solid samples were microwave digested, and liquid samples were diluted with concentrated nitric acid, without digestion. Electrothermal atomic absorption spectrometry with Zeeman background correction (ZETAAS) was employed for chromium determination. Analytical quality assurance of the method included analysis of certified reference material CTA-VTL-2 (Virginia Tobacco Leaves), and recovery study of real food samples confirmed the reliability of the method. The results show that chromium content varied among and within different food species (<1 to 183 μg/kg). The highest mean values were observed in samples of wheat grains (39 μg/kg), strawberries (32 μg/kg) and cucumbers (19 μg/kg) and the lowest – in juices (<1 μg/L). Some differences in chromium level in plants collected in various regions were also observed.

INTRODUCTION

Chromium is an element occurring in food products of both plant and animal origins. It is regarded as an essential trace element in humans and animals, taking part in various metabolic processes. Chromium, as an essential element, is usually present in food in the trivalent form; the hexavalent form of chromium is toxic and not normally found in food [Noël *et al.*, 2003]. Cr(VI) has been reported to be toxic and carcinogenic to humans owing to its oxidising potential and easy permeation of biological membranes [Schönsleben *et al.*, 1995]. In living organisms, chromium is present in the more stable trivalent state, and its essential biological activities are due to the complexes known as "biologically active chromium" (glucose tolerance factor or GTF) [Anderson, 1981].

Chromium is a component of enzymes which control glucose metabolism and synthesis of fatty acids and cholesterol, and its deficiency leads to severe impairment of glucose tolerance, which finally leads to diabetes and atherosclerotic disease [Cornelis & Wallaeys, 1984]. Insufficiency of chromium is caused mainly by its insufficient supply with food products. The recommended daily intake proposed for chromium by the US RDA [Food and Nutrition Board, 1989] is 50–200 μg/day for a 60-kg person. These levels are not usually reached in the industrialised countries where Cr deficiency is perhaps the principal trace element deficiency, particularly in people above the age of 35. As a result, it is referred to as "geriatric nutrient" [Mateos *et al.*, 2003]. According to literature data, daily Cr intake in European countries ranges from 22 μg to 146 μg, but in most of them it is under 100 μg, and in the USA it is as low as 23 to 62 μg [Van Cauwenbergh *et al.*, 1996]. In Poland, however, chromium content of daily food rations at selected canteens was reported to vary from 52 to 204 μg [Marzec, 1998], 38 to 155μg [Marzec, 1999], 8 to 18 μg [Przysiężna *et al.*, 2002] and from 44 to 124 μg [Marzec *et al.,* 2003].

To assess the dietary intake of chromium with food it is necessary to know its content in different groups of food products. Chromium content of fruits, vegetables and dairy products is relatively low, but their consumption is high, which makes their contribution to chromium intake significant [Schönsleben *et al.*, 1995]. However, scientifically reliable data on the chromium content of different foods and agricultural products are still insufficient [Györi & Prokisch, 1999]. The original data on the chromium content of foods of plant origin in Poland are also scant [Międzobrodzka *et al.*, 1992; Michalak & Buliński, 1995; Mirosławski *et al.*, 1995; Kocjan *et al.*, 2002; Stempin *et al.*, 2002; Sobiech *et al.*, 2003].

Chromium determination in biological matrices and food products, especially at the μg/kg level, is a serious analytical problem, and sample contamination [Van Cauwenbergh *et al.*, 1996; Györi & Prokisch, 1999] or loss of chromium during pre-treatment [Wilplinger *et al.*, 1995;] make it even more difficult. According to Miller-Ihli & Greene [1992], many older references on chromium values are no longer useful because of sample contamination. What is more, in some references the data were obtained with atomic absorption analyses without adequate background correction, or under conditions of significant matrix interferences. These errors resulted in unreasonably high chromium concentrations.

The AAS methods (especially ETAAS) for chromium determination in specific food matrices have been investi-

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gated recently by many authors [Cabrera-Virque *et al.*, 1997; Carlosena *et al.*, 1997; Gallo *et al.*, 1997; Kardjova *et al.*, 1998; Tinggi *et al.*, 1997]. The results of these studies demonstrate that the determination of chromium concentrations in different food products requires special analytical procedures.

The assessment of chromium concentrations in different food products and beverages of plant origin was the objective of the present study. The investigation was performed using a validated method of chromium determination by electrothermal atomic absorption spectrometry with Zeeman background correction (ZETAAS).

MATERIALS AND METHODS

MATERIAL

224 samples of fresh edible plants (strawberries, apples, cucumbers, cabbages, carrots, potatoes, wheat grains) were collected in 2001 in commercial plantations in different regions of Poland. In addition, 30 samples of food products, Polish (juices, beverages and some beers) and European (wines and some beers), were bought on the domestic market.

APPARATUS AND EQUIPMENT

SpectrAA 880 Z spectrometer equipped with Graphite Tube Atomizer (GTA 100) and programmable Sample Dispenser (PSD 100), Varian, Australia; Microwave Sample preparation System MDS 2000 CEM, with equipment for pressure digestion, CEM Corp., Matthews, NC, USA; Analytical balance Medicat 160, Oerlikon, Switzerland; Malaxer HR 2831, Philips; Mixer Power Max 2050, Braun; Sample Mill Cyclotec 1039, Tecator, Sweden; Water still Aquatron A4D, J.Bibby Science Products Ltd, England; Ultrasonic cleaner (bath), Branson, USA; Volumetric flasks, graduated pipette and one mark bulb pipettes, class A were used in the study.

METHODS

Edible parts of fruits and vegetables were prepared as for consumption, *i.e.* washed, surface dried or peeled (if necessary). Samples of fruits and vegetables were homogenised and thoroughly mixed, wheat samples were ground. Solid samples were digested by means of a microwave procedure which was set up experimentally. About 1.0-g sample (fruits and vegetables) or *ca.* 0.3-g (wheat grain and certified reference material) were directly weighed in the Teflon PFA® digestion vessel; after the addition of 4 mL of concentrated nitric acid, (69% Spectrosol BDH Chemical Ltd.); the mixture was allowed to rest at room temperature for *ca.* 16 h. Then it was digested in a microwave unit according to the program set up experimentally, as reported in Table 1. After cooling and removing the excess nitrogen oxides in an ultrasonic bath, the residual solution was diluted with double distilled water taking a volume of 50 mL (for 1-g samples) or 25 mL (for 0.3-g samples). Liquid samples (juices, beers, wines) were thoroughly mixed and degased (beers), then 1 mL of concentrated nitric acid (69% Spectrosol BDH Chemical Ltd.) was added to 1 mL of sample and mixed with water to a volume of 25 mL.

TABLE 1. Microwave digestion program.

Step	Power (%)	Pressure (psi)	Time (min)	TAP* (min)	Fan speed (%)
I	40	40	10	-	100
II	60	80	10	-	100
III	180	120	15	-	100
IV	100	170	10	5	100

*TAP – time at pressure

AA Spectrometer with electrothermal atomization and Zeeman background correction, combined with the method of standard additions and peak area mode were used for chromium measurements. The conditions of furnace operation, set up experimentally, are presented in Table 2. The measurements were performed at a wavelength of 357.9 nm, using pyrocoated partitioned graphite tubes, matrix modifier consisting of a mixture of palladium nitrate and ascorbic acid solutions, and argon as carrier gas. Sample volume was 10 μL.

TABLE 2. Furnace parameters.

Step	Temperature (°C)	Time (sec)	Gas flow (L/min)	Read command
1	85	5	3	-
2	95	10	3	-
3	95	15	3	-
4	120	20	3	-
5	1100	15	3	-
6	1100	10	3	-
7	1100	2	0	-
8	2600	1	0	+
9	2600	2	0	+
10	2700	1	3	-
11	45	20	3	-

Characteristics of the method. Analytical range: 0.05–10 μg/L; sensitivity calculated as triplicate standard deviation of the chromium content of 10 blank samples; 0.01 μg/L; characteristic concentration; 0.40 μg/L; analytical detection limit for chromium determination with regard to sample weight and dilution; *ca.* 1 μg/kg (μg/L) for 1-g (or 1-mL) samples, or *ca.* 2 μg/kg for 0.3-g samples.

Analytical quality assurance. The precision and accuracy of the method was verified by an analysis of certified reference material CTA-VTL-2 Virginia Tobacco Leaves (IChTJ Poland) and by means of recovery assays of food samples (Table 3). The results of chromium determination in CRM were within the limits of certified values, at the coefficient of relative standard deviation of 3%. All determinations were made in six replications, involving the full analytical procedure. During the study a digested sample of CRM was used as a quality control sample.

RESULTS AND DISCUSSION

The ranges and mean chromium concentrations in the food products analysed in the study and some statistical data are reported in Tables 4 and 5.

The results obtained showed that Cr content of food samples ranged from <1 to 183 μg/kg. Chromium content

TABLE 3. Precision and accuracy of chromium determined in food samples.

Sample	Spiking μg/kg or μg/L**	Cr, concentration ± SD* μg/kg or μg/L**	Recovery (%)	RSD (%)
Apples	-	<1	-	-
	100	104±5	104	3.7
	200	191±5	96	2.9
Canned	-	48±1	-	-
potatoes with	100	152±4	104	2.4
carrots	250	284±3	94	1.2
Beer	-	9±1**	-	7.0
	250**	265±9**	102	3.4
	500**	520±14**	102	2.6

*mean±SD for 6 replicates

of edible plants and some alcoholic and non-alcoholic beverages was generally low, but varied between species. There were large differences in Cr levels detected, even between food products of the same brand. The coefficient of variation (RSD), characterizing in this case the dispersion of Cr content in particular groups of foodstuffs, ranged from 13% for beers to 109% for apples (Tables 4 and 5). Each species of edible plants was characterised by different relative distribution of chromium content (Table 6). However, alcoholic beverages (wines, beers), irrespective of country of provenience, contained similar quantities of chromium; chromium content above the limit of detection was not recorded in home-made juices and beverages.

The highest concentrations of chromium were found in wheat grains, strawberries and cucumbers, with mean values ranging from 37 to 32 and 19 μg/kg f.w., respectively. Apples, carrots, cabbages and potatoes contained smaller amounts of this element, mean values being 11, 6, 6 and 4 μg/kg respectively, similarly as for beverages (<1 to 12 μg/L). A different chromium content of plant samples collected in different regions of Poland was also observed (Table 7).

The results obtained in this study were often lower than those reported previously for the same species in Poland, and similar to the current data from other countries.

Chromium levels in wheat grains estimated during this study (<2 to 183 μg/kg, mean 39 μg/kg) are in some cases in accordance with the data reported by other authors. Sobiech *et al.* [2003] found the chromium content of six varieties of Polish wheat grain to range from 36 to 44 μg/kg. Międzobrodzka *et al.* [1992] reported the average chromium content of Polish wheat originating from different regions of southern Poland to vary between 190 and 410 μg/kg (range 0–540 μg/kg, n=30), but Stempin *et al.* [2002] found the chromium level in wheat cultivated in copper-bearing areas to be 190 000 μg/kg (190 mg/kg) d.m.

Chromium content of wheat grains reported by authors from other countries ranged from 23 to 66 μg/kg [Plessi & Monzani, 1990] and from 3 to 90 μg/kg [Cary & Kubota, 1990]. The average chromium content of five Hungarian wheat varieties was found to be 114 μg/kg [Györi & Prokisch, 1999]. According to Mateos *et al.* [2003], who compiled the results of different studies on the topic, chromium content of wheat varies between <15 and 330 μg/kg.

Michalak & Buliński [1995] found that the average chromium level in frozen strawberry fruits was 138 μg/kg, Kocjan *et al.* [2002] reported the chromium content of apples, carrots and potatoes to be 63 μg/kg, 64 μg/kg and

TABLE 4. Chromium content of edible plants (μg /kg f.w.).

Foodstuffs	n	Range	Mean	Median	RSD (%)	90 percentile
Strawberries	42	61–34	32	28	72.5	56
Apples	23	2–63	11	9	109	16
Cucumbers	23	1–74	19	10	105	40
Cabbages	23	<1–15	6	5	66	10
Carrots	43	<1–18	6	4	83	12
Potatoes	24	<1–11	3	4	75	9
Wheat grains	45	<2-183	39	27	95	79

TABLE 5. Chromium content of some beverages of plant origin (μg/L).

Foodstuffs	n	Range	Mean	Median	RSD (%)	90 percentile (μg/L)
Wines	10	10–16	12	12	17	14
Beers	10	6–8	7	8	13	8
Juices, beverages	10	<1	<1	<1		<1

TABLE 6. Relative distribution of chromium content as a percentage of the total number of samples.

Chromium content (μg/kg)	% of samples						
	Strawberries	Apples	Cucumbers	Cabbages	Carrots	Potatoes	Wheat
>100	2.4	0.0	0.0	0.0	0.0	0.0	8.9
50–100	14.3	8.7	0.0	0.0	0.0	4.3	4.4
10–50	76.2	68.0	4.2	4.7	13.0	30.4	64.4
1–10	7.1	34.8	68.0	48.8	65.2	65.2	8.9
<1	0.0	8.7	25.0	46.5	21.7	0.0	13.3*

*<2 μg/kg

TABLE 7. Chromium content of plant samples originating from three provinces of Poland (μg/kg).

Province	n	Range	Mean	Median	90 Percentile	RSD (%)
				Strawberries		
Mazowieckie	18	15–134	36	25	61	78
Lubelskie	16	6–41	21	17	35	57
Kujawsko-Pomorskie	6	32–61	47	50	58	24
				Wheat grains		
Mazowieckie	22	<2–183	31	21	66	109
Lubelskie	17	<2–51	23	25	39	61
Kujawsko-Pomorskie	6	<2–131	62	62	121	82
				Potatoes		
Mazowieckie	23	<1–10	4	3	5	62
Lubelskie	14	<1–10	2	1	5	116
Kujawsko-Pomorskie	6	<1–11	5	4	9	68
				Carrots		
Mazowieckie	23	<1–20	5	3	8	106
Lubelskie	14	<1–18	8	7	12	61
Kujawsko-Pomorskie	6	<1–13	6	2	11	112

66 μg/kg, respectively. These values are higher than the mean chromium concentrations estimated in this study (strawberries – 32 μg/kg, apples – 11 μg/kg, carrots – 6 μg/kg, and potatoes – 4 μg/kg). Chromium contents of food products analysed in this study were often different from those included in the Polish Tables of Trace Elements in Food Products [Marzec *et al.*, 1992], according to which the average chromium content is 35 μg/kg in strawberries, 35 μg/kg in apples, 22 μg/kg in cucumbers, 61 μg/kg in cabbage, 50 μg/kg in carrots, and 40 μg/kg in potatoes.

The mean chromium content of wines available on the Polish market (12 μg/L) is smaller than that reported in the Polish Tables (55 μg/L) [Marzec *et al.*, 1992]. The results obtained are also somewhat lower than those reported for Spanish wine (20 μg/L) by Lendinez *et al.* [1998] and for French wine (mean value for different types of wine was about 22 μg/L), but higher than in the case of Italian wine (8 μg/L) [Cabrera-Vique *et al.*, 1997]. According to these authors, wine can have a significant contribution to the dietary intake of chromium, depending on its intake.

The results of chromium content determination in the food products analysed sometimes appeared to be lower than those presented in literature. According to Wilplinger *et al.* [1995], some of the older values are higher due to contamination. The fact that some of the techniques and methods for chromium determination are not sensitive enough may also be of primary importance. This is confirmed by a general report compiled by the AOAC Committee, promoting official analytical methods. Taking into account the results of previous studies and, progress in ongoing research, it recommends the use of AAS with a graphite furnace for Cr determination in foods [Ihnat, 2003].

The results of this study suggest that, among other factors, the region of sampling may have an influence on the differences noted in Cr content of edible plants in Poland.

CONCLUSIONS

1. The analytical procedure applied in the study, including sample preparation by microwave digestion and chromium content measurement by ZETAAS, is suitable for chromium analysis in foods of plant origin.

2. Chromium content of food samples ranged from <1 μg/kg to 183 μg/kg. The highest chromium concentrations were found in wheat grains, strawberries and cucumbers.

3. There were some differences in chromium level in plants collected in various regions of Poland.

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ZAWARTOŚĆ CHROMU W ŻYWNOŚCI I NAPOJACH POCHODZENIA ROŚLINNEGO

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Przeprowadzono badania zawartości chromu w wybranych owocach, warzywach, ziarnie pszenicy pobranych w 2001 roku w trzech rolniczych regionach Polski oraz w sokach, piwie i winie (ogółem 272 próbki). Badania wykonano przy zastosowaniu techniki spektrometrii absorpcji atomowej z atomizacją elektrotermiczną i korekcją tła Zeemana (ZETAAS), uznanej za najbardziej właściwą do oznaczania chromu. Próbki stałe mineralizowano kwasem azotowym w piecu mikrofalowym w układzie zamkniętym (ciśnieniowym), próbki płynne rozcieńczano kwasem azotowym, z pominięciem etapu mineralizacji. Opracowane doświadczalnie procedury analityczne oznaczania zawartości chromu sprawdzono wykonując analizę zawartości chromu w roślinnym certyfikowanym materiale odniesienia CTA-VTL-2 (Virginia Tobacco Leaves) oraz oceniając dokładność i precyzję oznaczania tego pierwiastka w próbkach rzeczywistych.

Zawartości chromu oznaczone w próbkach badanych produktów spożywczych były na ogół niskie, od <1 do 183 μg/kg. Najwyższe zawartości średnie stwierdzono w ziarnie pszenicy (39 μg/kg) i truskawkach (32 μg/kg), a najniższe w sokach i napojach owocowych (<1 μg/L). Stwierdzono także pewne różnice poziomu zawartości chromu w próbkach badanych surowców pochodzących z różnych rolniczych regionów Polski.