

Bovine meat and offal as a source of human exposure to cadmium in the Czech Republic

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Abstract

An evaluation of cadmium content in the muscle, liver and kidneys of young bovine animals (under two years of age) and cows was conducted in the Czech Republic during the period 1993-2010. The average cadmium content in the muscle of young bovine animals and cows was approximately the same (i.e. 0.007 and 0.008 mg.kg⁻¹ wet weight, respectively) during the aforementioned period. The liver of cows showed a statistically insignificant higher average cadmium content (0.111 mg.kg⁻¹ wet weight) in comparison with the liver of young bovine animals (0.079 mg.kg⁻¹ wet weight). A statistically significant difference was detected in the average cadmium content in cows' kidneys between 1993-1994 and 2009-2010 (0.454 and 0.799 mg.kg⁻¹ wet weight, respectively) at the significance level of $\alpha = 0.05$. A statistically significant trend of an increase in cadmium content in cows' kidneys between 1993-1994 and 2009-2010 at the significance level of $\alpha = 0.05$ was also detected.

Bovine animals, cadmium, kidneys, trends

Introduction

Cadmium (Cd) is a toxic metal occurring in the environment from natural sources (volcanic activity and the weathering of rocks, particularly shales) and as a consequence of anthropogenic activities from industry and agriculture (metallurgy, the electrical industry, phosphate fertilizers and sewage sludge applied to soil) (EFSA 2009).

For a person (non-smoker), who is not occupationally exposed to Cd, foodstuffs represent the source of up to 90% of his total intake of cadmium (EFSA 2009). The tobacco in one cigarette contains 0.5 - 1 µg of cadmium. Approximately 10% of inhaled Cd is deposited in the lungs and a further 30 - 40% is absorbed into the bloodstream. Smokers have 4 - 5 times higher levels of cadmium in the blood and 2 - 3 times greater quantities in the kidneys (Dallongeville et al. 1998). The absorption of Cd from a person's digestive tract is relatively low (3 - 5 %). However, cadmium accumulates in the kidneys and liver and has a very long half-life of excretion from the human body in the range of 10 - 30 years (EFSA 2009), or 20 - 30 years (Kjellström and Nordberg 1978).

Cadmium is primarily toxic to the kidneys, especially to the proximal tubules in the renal cortex, where it accumulates by binding to metallothionein and can cause renal dysfunction and even kidney failure. Exposure to Cd is associated with nephrotoxic, neurotoxic, carcinogenic, genotoxic and teratogenic effects, and also with the development of osteoporosis, or osteomalacia, and damage to the endocrine system and reproductive functions (EFSA 2009). According to the classification of the International Agency for Research on Cancer (IARC) cadmium is classed as a group I human carcinogen (IARC 1993) on the basis of studies on animals and people, particularly in connection with the inhalation of cadmium. The Joint FAO/WHO Expert Committee on Food Additives (JECFA) in 1993, supported by the stance of the European

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Commission's Scientific Committee on Food (SCF) in 1995, established a provisional tolerable weekly intake (PTWI) of Cd of $7 \mu\text{g.kg}^{-1}$ body weight. In 2009 the Panel on Contaminants in the Food Chain (CONTAM) re-evaluated existing knowledge with regard to the highest burden of the target organs, and also with regard to vulnerable groups within the population and the varying degree of Cd absorbability and, taking the safety factor into account, established a tolerable weekly intake (TWI) of Cd of $2.5 \mu\text{g.kg}^{-1}$ body weight (EFSA 2009 and 2011).

Through an analysis of around 140 000 data from 20 member states of the European Union for the years 2003 - 2007, the Panel on Contaminants in the Food Chain determined that the average dietary exposure to Cd of people in EU countries was $2.3 \mu\text{g.kg}^{-1}$ body weight per week with a range of values from 1.9 to $3.0 \mu\text{g.kg}^{-1}$. For vegetarians the weekly exposure to cadmium was as much as $5.4 \mu\text{g.kg}^{-1}$ body weight. The Cd exposure of adult residents of the EU states, as emerged from the aforementioned study, is close to or slightly exceeds the value of TWI $2.5 \mu\text{g.kg}^{-1}$ body weight. For vegetarians, children or residents of regions with a higher environmental burden of Cd (industrial regions, urban agglomerations) Cd exposure is as much as twice the value of TWI (EFSA 2009).

As part of the national "total diet study" in the United Kingdom (UK) in 2009 a total of 119 types of food were analyzed for their cadmium content. If we focus solely on animal products, Cd content was below the detection limit in red meat, poultry, eggs and fat, milk and dairy products. By contrast, the highest Cd content was found in offal (average $-0.084 \text{ mg.kg}^{-1}$). However, in terms of a person's total burden from foodstuffs, in view of the quantity consumed the highest sources of Cd were potatoes (24%), various cereals (21%) and bread (19%) (EFSA 2009). The results of analyses of 880 composite samples of foodstuffs to assess a person's dietary exposure in 2008-2009 in the Czech Republic also showed that significant sources of exposure included potatoes and potato products, various kinds of bread and flour. The contribution of foods of animal origin to Cd exposure was low in comparison with vegetable foods (Ruprich et al. 2009). The main sources of dietary exposure to Cd are therefore cereals and vegetables. Meat and fish are a lesser source, although offal (liver and kidneys) can represent a significant source of Cd due to its accumulation in these organs. However, a person's dietary exposure to Cd does not depend only on the Cd content of foodstuffs, but also on the type of foodstuffs consumed and the degree of their consumption.

The aim of this work was to collect and evaluate the results of an investigation into the content of Cd and relevant trends in samples of the muscle, liver and kidneys of young bovine animals from six months to two years of age and cows over two years of age slaughtered in the Czech Republic, during the period from 1993 to 2010, with the period under observation being divided into units of two years.

Materials and Methods

Animals

The results of the investigation were obtained within the implementation of the national plan for the monitoring of residues and contaminants in accordance with Council Directive 96/23/EC (1996) and Commission Decision 97/747/EC (1997). The minimum number of animals from which tissue samples were taken was calculated according to the methodology of the aforementioned directive and decision. For bovine animals this represented an annual sampling of tissue from at least 0.015% of the animals slaughtered in the previous year. Slaughtered animals destined for tissue sampling were randomly selected by veterinary inspectors at various slaughterhouses in such a way as to cover as large an area of the Czech Republic as possible. Samples were taken of muscle (lean meat), liver (a sample of at least 0.5 kg) and kidneys from each young bovine animal up to the age of two (excluding calves up to six months old) and each cow (over two years old). The average age of cows slaughtered in the Czech Republic was monitored from 2001 and varied in individual years from 69 to 72 months with a median of 65 to 66 months. After collection, tissue samples were frozen and then transported to the laboratories of state veterinary institutes for analysis.

Analytical methods

Homogenized samples of biological material were mineralized in a microwave device after the addition of concentrated nitric acid and hydrogen peroxide. Decomposition was conducted using a programme operated by a control unit. After decomposition was complete, the measurement of the Cd content was carried out using electrothermal atomic absorption spectrometry with Zeeman correction (ET-AAS, or GF-AAS), or in some cases using inductively coupled plasma mass spectrometry (ICP-MS). In the case of the ET-AAS method, the mineralized sample was atomized by heating according to the temperature programme in an electrically heated atomizer (graphite cuvette). The absorbance of Cd was measured at a wavelength of 228.8 nm, which is directly proportional to the concentration of Cd in the sample. In the case of the ICP-MS method, the mineralized sample was nebulized with the help of a conical nebulizer and cooling chamber and introduced into plasma, where the elements are freed from their chemical bonds thanks to the temperature of argon plasma (cca 10 000 K), ionized and directed into the quadrupole. There ions are selected according to their M/Z (mass/charge) and the resultant ion beam is fed into an electron multiplier, which detects the impact of individual ions. The number of ^{111}Cd ions which enter the detector is directly proportional to the concentration of Cd in the sample. The level of quantification (LOQ) in the tissues for the methods used was 0.01 mg.kg^{-1} for the liver and kidneys, and 0.005 mg.kg^{-1} for the muscle.

Statistical evaluation of the results

The statistical evaluation made use of the results of the investigation into Cd content, always divided into two-year units, in the period under observation from 1993 to 2010. In the event of immeasurable values of Cd content, half values of LOQ were used for statistical evaluation. The evaluation of trends in Cd content in the tissue of bovine animals was carried out using statistical testing by comparing two samples of measured values of Cd content in 1993-4 with Cd content in 2009-10. In the kidneys of cows, the significance of the observed trend of an increase in the value of Cd was statistically tested. To test the two samples, compliance testing of variances (F-test) and compliance testing of mean values (Student's t-test of compliance of averages) were used. In order to assess the significance of the observed trend of Cd content in the kidneys of cows from 1993 to 2010, statistical testing of the significance of the direction of the line calculated for this growth trend was used.

Results and Discussion

The results from the investigations are in document comparably low average and median values of Cd concentration in the muscle (meat) of both the observed age groups of bovine animals (Table1). Over the years the average and median values of Cd in the meat of both groups of bovine animals showed no tendency towards a decrease or increase in value. The content of Cd in the liver was comparable over the years, with a slightly higher concentration in the liver of cows in comparison with the Cd content in the liver of younger bovine animals up to two years old.

During the period under observation the average content of Cd for cows was 0.008 mg.kg^{-1} ($0.002 - 0.016 \text{ mg.kg}^{-1}$) in the muscle, 0.111 mg.kg^{-1} ($0.099 - 0.127 \text{ mg.kg}^{-1}$) in the liver and 0.533 mg.kg^{-1} ($0.454 - 0.816 \text{ mg.kg}^{-1}$) in the kidneys, wet weight. The average content of Cd in the muscle of young bovine animals up to two years old was 0.007 mg.kg^{-1} ($0.002 - 0.008 \text{ mg.kg}^{-1}$), in the liver 0.079 mg.kg^{-1} ($0.071 - 0.085 \text{ mg.kg}^{-1}$) and in the kidneys 0.273 mg.kg^{-1} ($0.257 - 0.319 \text{ mg.kg}^{-1}$) wet weight. The average Cd concentrations in cows' kidneys in individual years of the period under observation were from 1.7 to 2.8 times greater than the average Cd concentrations amongst young bovine animals up to two years old. The ratio between the total average Cd content during the period under observation was 1:11:39 for the muscle, liver and kidneys of young bovine animal up to two years old. For cows this ratio was 1:14:67. The absorption and accumulation of Cd in the organs of gestating and lactating cows is more marked when compared with dry cows. The reason for this is the need for more calcium and the subsequent increased absorption of both elements (Bhat-tacharya et al. 1981). The average values of Cd are similar to those shown by Doganoc (1996), who determined the level of Cd for several hundred slaughtered bovine animals to be 0.004 mg.kg^{-1} in the meat, 0.094 mg.kg^{-1} in the liver and 0.373 mg.kg^{-1} in the kidneys (Doganoc 1996). The ratio between the average content of Cd in the muscle, liver and kidneys was therefore 1:23:93. A similar investigation was carried out by Niemi et al. (1991) and they determined cadmium levels in the meat of bovine animals to be 0.013 mg.kg^{-1} , 0.061 mg.kg^{-1} in the liver and 0.350 mg.kg^{-1} in the kidneys (Niemi 1991). Here the ratio of average

Table 1. The cadmium content in the tissues of young bovine animals and cows in the Czech Republic from 1993 to 2010 (mg.kg⁻¹ wet weight)

Tissue	year	Cows (>2 years of age) mg.kg ⁻¹ wet weight				Young bovine animals (>6 months < 2 years of age) mg.kg ⁻¹ wet weight							
		n	n<LOQ	median	average	10% quantile	90% quantile	n	n<LOQ	median	average	10% quantile	90% quantile
muscle	1993-4	367	196	0.005	0.008	0.003	0.017	606	336	0.005	0.008	0.003	0.019
	1995-6	222	119	0.007	0.008	n.d.	0.013	280	192	0.007	0.008	n.d.	0.010
	1997-8	152	98	n.d.	0.007	n.d.	0.014	143	84	n.d.	0.008	n.d.	0.015
	1999-0	186	127	n.d.	0.008	n.d.	0.018	167	128	n.d.	0.008	n.d.	0.010
	2001-2	228	169	n.d.	0.007	n.d.	0.014	235	184	n.d.	0.006	n.d.	0.010
	2003-4	180	135	n.d.	0.010	n.d.	0.015	197	163	n.d.	0.005	n.d.	0.010
	2005-6	77	58	n.d.	0.016	n.d.	0.013	74	61	n.d.	0.004	n.d.	0.013
	2007-8	55	50	n.d.	0.003	n.d.	0.006	23	21	n.d.	0.002	n.d.	0.006
	2009-0	49	49	n.d.	0.002	n.d.	n.d.	31	31	n.d.	0.002	n.d.	n.d.
	1516			0.008			1756			0.007			
liver	1993-4	384	13	0.091	0.111	0.035	0.193	593	11	0.070	0.085	0.032	0.145
	1995-6	225	2	0.089	0.111	0.032	0.198	275	8	0.061	0.071	0.030	0.113
	1997-8	138	5	0.094	0.109	0.039	0.176	139	0	0.071	0.081	0.030	0.130
	1999-0	185	0	0.090	0.127	0.047	0.201	159	0	0.064	0.079	0.031	0.127
	2001-2	229	0	0.080	0.099	0.041	0.175	237	3	0.061	0.074	0.030	0.121
	2003-4	165	0	0.085	0.099	0.043	0.169	194	4	0.068	0.074	0.030	0.120
	2005-6	75	1	0.100	0.122	0.050	0.215	75	0	0.068	0.079	0.031	0.145
	2007-8	53	0	0.099	0.116	0.042	0.233	27	0	0.080	0.081	0.030	0.137
	2009-0	50	0	0.091	0.119	0.038	0.258	31	0	0.064	0.079	0.033	0.150
	1504			0.111			1730			0.079			
kidney	1993-4	388	5	0.380	0.454	0.152	0.860	593	3	0.210	0.264	0.098	0.509
	1995-6	215	1	0.397	0.471	0.139	0.862	272	1	0.206	0.257	0.090	0.497
	1997-8	135	0	0.421	0.513	0.152	1.039	137	0	0.260	0.306	0.094	0.616
	1999-0	183	0	0.457	0.584	0.162	1.095	157	1	0.227	0.270	0.099	0.464
	2001-2	227	0	0.432	0.521	0.161	0.990	235	0	0.207	0.271	0.114	0.475
	2003-4	169	0	0.434	0.557	0.150	1.072	203	0	0.230	0.277	0.105	0.477
	2005-6	79	0	0.520	0.636	0.138	1.340	73	0	0.271	0.319	0.091	0.606
	2007-8	52	0	0.619	0.816	0.227	1.862	24	0	0.200	0.289	0.048	0.789
	2009-0	50	0	0.588	0.799	0.206	2.110	31	0	0.226	0.308	0.092	0.823
	1498			0.533			1725			0.273			

n.d. = not detected

values between the content of Cd in the muscle, liver and kidneys was 1:5:27. In cows which had been given Cd in doses of 0.025 and 0.125 mg.kg⁻¹ of body weight over a long period, Smith et al. (1991) discovered an 8 to 10 times greater concentration of Cd in the kidneys than in the liver. No difference was noted in the muscle between the control animals and the trial animals, which indicates that Cd has no significant affinity to muscle tissue. An increase in the content of Cd in the kidneys of cows is associated with older animals, as was shown by Pechová et al. (1998), who demonstrated the highly significant relationship between the age of cows and Cd content in the kidneys. At the same time they discovered that adult animals had a significantly higher concentration of Cd in the kidneys and liver than calves. This discovery is in accordance with our own results. The higher content of Cd in the liver and kidneys is primarily caused by its attachment to metallothionein, which is synthesized in the liver and kidneys as a reaction to the presence of divalent metal ions, which bind tightly and thus cause detoxification.

Contrary to the kidneys of young bovine animals up to two years old where in the period under observation there was no apparent tendency towards higher Cd concentrations, the kidneys of cows in our study showed a trend towards an increase in Cd concentrations (Fig. 1). The increase in the concentration of cadmium in the kidneys of cows was scientifically validated by statistical testing at the level of significance of $\alpha = 0.05$.

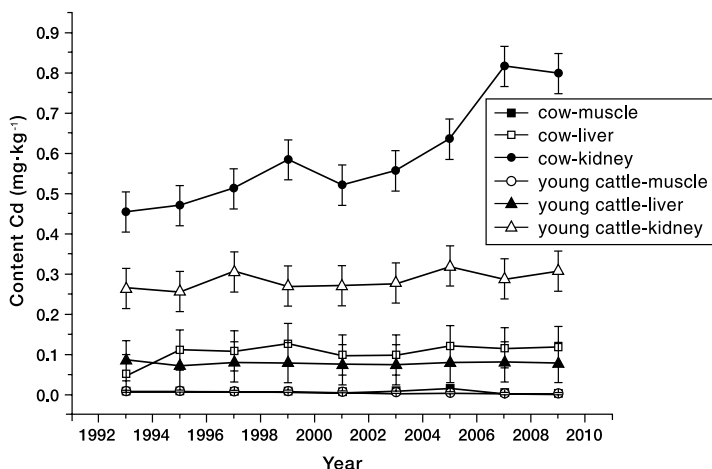


Fig. 1. Average content trends of cadmium in the muscle, liver and kidneys of young bovine animals up to two years old and cows in the Czech Republic from 1993 to 2010

The direction of the regression line expressing the growth trend is also statistically important. The growth trend is very obvious in Fig. 2 where a regression line passes through the measured values of Cd concentrations in individual years.

In one area of Holland with extreme conditions of soil acidity and a high content of cadmium (Cd 2.5 mg.kg⁻¹ and pH 4.5), the fodder crops, in particular the grazing sward, contained more than 1 mg.kg⁻¹ of cadmium in feedstuff with 12% moisture. In the model study of bovine animals up to six years old raised in that area receiving daily 20.5 mg of Cd in their feedstuff, Cd levels in the kidneys ranged from 0.37 to 4.03 mg.kg⁻¹ and in the liver from 0.07 to 0.76 mg.kg⁻¹ wet weight. In the five other tested sites with higher soil pH (pH 5.5), or a lower cadmium content in the soil (Cd 0.5 and 1.0 mg.kg⁻¹), the daily Cd intake in

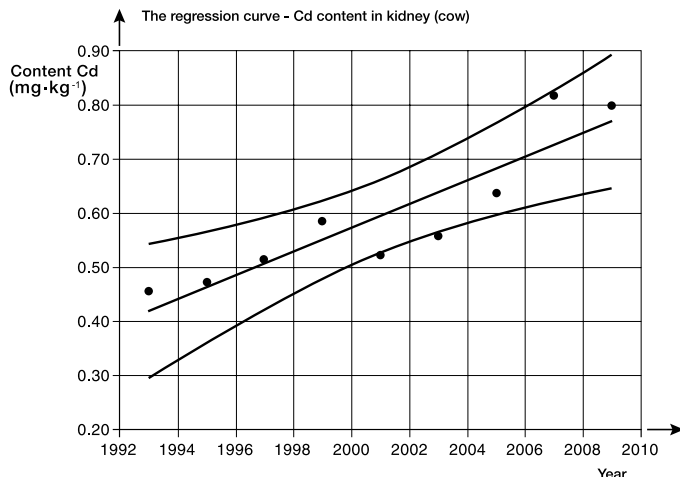


Fig. 2. The growth trend expressed by a regression line passing through the measured points

the feedstuff was lower (1.9 – 10.4 mg) and the values of cadmium in the liver and kidneys were also lower (Fels-Klerx 2011).

Apart from the demonstrable effect of age in bovine animals and the related accumulation of Cd, one of the probable causes of the increased content of Cd in the liver and kidneys of cows in particular is the possible increased intake of Cd through vegetable feedstuffs during the period under observation in the Czech Republic. This supposition is based on data from the Central Institute for Supervising and Testing in Agriculture (CISTA). The mobility of Cd from the soil to the plants increases with decreasing soil pH. Apart from the content of Cd in the soil (substratum) and its form, the utilization of Cd by plants depends on the type of plant, the kind of soil, the content of soluble organic compounds and the presence of other metals such as calcium, zinc, etc. One of the main reasons for the decrease in soil pH, and with it the increased mobility of Cd to plants is the reduction in the total content of calcium in the soil. Liming is used, for example, for the immobilization of cadmium (or lead) in the sewage sludge which is applied to agricultural land. For economic reasons the consumption of lime fertilizer has decreased significantly since 1990. The development of soil reaction was more often directed towards the acidification of soils in the majority of the monitored types of land in the Czech Republic. Since the 1990s, the pH of arable land in the Czech Republic has decreased by 0.1 (to 6.3). Almost 13% of the area which had originally been pH neutral or alkaline transferred to the category of slightly acidic or acidic. For grasslands, the average decrease in pH reached 0.3 degrees and 22% of the acreage was transferred into the two aforementioned categories. Very strongly acidic and acidic soil reactions can be found in 23.5% of agricultural land in the Czech Republic, or roughly 718 000 hectares (Bouma 2005).

Conclusions

The statistical evaluation of the data confirmed, using two independent tests with a 95% probability (significance level $\alpha = 0.05$), the significance of the growth trend in the cadmium concentrations in the kidneys of cows in the Czech Republic during the period under observation from 1993 to 2010. There was no increase in the content of Cd in the

liver and muscle of cows, just as there was no increase in the content of Cd in the kidneys, liver and muscle (meat) of young bovine animals up to two years old during the period under observation from 1993 to 2010.

It was proposed that one of the possible causes of the statistically significant increase in the concentration of Cd in the kidneys of cows between 1993 and 2010 in the Czech Republic could be the increased Cd intake from fodder crops due to the reduction of soil liming and an decrease in soil pH and with it the increased mobility of Cd from the soil to plants. For specific dietary habits associated with a higher consumption of bovine offal, in particular kidneys, this may represent a significant source of exposure to cadmium.

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