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## Cadmium in Animal Feed and in Foodstuffs of Animal Origin

Jasenka Sapunar-Postružnik<sup>1\*</sup>, Davorin Bažulić<sup>1</sup>, Mirela Grubelić<sup>1</sup>,  
Helena Kubala Drinčić<sup>1</sup> and Bela Njari<sup>2</sup>

<sup>1</sup>Croatian Veterinary Institute – Department of Residue Analysis,  
Savska cesta 143, 10000 Zagreb, Croatia

<sup>2</sup>Ministry of Agriculture and Forestry – Department of Veterinary Public Health,  
Ulica grada Vukovara 78, 10000 Zagreb, Croatia

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### Summary

This paper emphasizes the suitability of a systematic residue control, not only how a national monitoring programme may timely detect source of contamination of animal organs with cadmium, but also how a systematic control and procedures may resolve the problem of contamination of foodstuffs. As a result of feeding the pigs with contaminated feed ( $w$  (Cd) = 2.05 to 4.86 mg/kg) the mass fraction levels of this toxic metal in their kidneys ranged between 0.67 and 12.96 mg/kg and those determined in liver ranged between 0.07 and 1.69 mg/kg. The levels of cadmium in all muscle samples ranged between < 0.01 and 0.04 mg/kg. After withdrawal of the contaminated feedmix tissue levels returned to normal ( $w$  (Cd) = 0.19–0.63 mg/kg in kidney, 0.03–0.1 mg/kg in liver and < 0.01 mg/kg in muscle).

*Key words:* cadmium, kidney, liver, feed, monitoring

### Introduction

Long time ago man became aware that food is an important factor of a normal growth and development but also, unfortunately, a cause of many diseases. It doesn't surprise that national governments have always endeavoured to establish a foodstuffs control that would be able to protect consumers against unsafe foodstuffs.

For example, as early as in 1877, in the area of the today's Republic of Croatia, by the Decree of the Royal Government of Croatia, Dalmatia and Slavonia, detailed *ante-mortem* and *post-mortem* inspection of food producing animals was legally mandated (1).

Of course, modern food production technology, where the use of pesticides and veterinary drugs is a *conditio sine qua non*, and the presence of naturally occurring and anthropogenic contaminants, call for daily con-

trol of a large number of new parameters. In the last decade in the Republic of Croatia, various biological and chemical parameters have been controlled on farms, in slaughterhouses, finished product plants, feed mill *etc.*, in target tissues of animals and animal products intended for human consumption, based on hazard analysis and critical control point systems (HACCP) (2). Under the current National Residue Monitoring Programme (3), 70 parameters are controlled, including heavy metals, and one of them is cadmium, an undoubtedly toxic element, having cumulative and irreversible effect on human and animal organisms causing various toxic effects like renal disfunction and osteomalacia (4–7). In view of the fact that it is regularly determined in kidney and liver of bovine and porcine animals (8), at levels

\* Corresponding author; Phone: ++ 385 (0)1 6123 666; Fax: ++ 385 (0)1 6190 841

which are not infrequently at or above the maximum residue level – MRL (9), regular control of this metal is even more justified.

The purpose of this paper is to emphasize that regular monitoring in close cooperation with veterinary inspection contributes to the protection of animal health, as well as of the consumer's health.

## Materials and Methods

Under the Croatian National Residue Monitoring Programme, the levels of cadmium were analysed in muscle tissue, kidney and liver of slaughtered pigs, and in animal feedmixes and premix from September 1995 till March 1996. The samples were separately packed in polyethylene bags and sent either deep frozen or fresh to the laboratory where they were stored at  $-20\text{ }^{\circ}\text{C}$  until the analysis. Prior to analysis the samples were homogenised using Politron blender Model PT2000. The samples were prepared by digestion with  $\text{HNO}_3$  and  $\text{H}_2\text{O}_2$  in high performance microwave digestion unit (Milestone 1200 M). The digestion ( $500\text{--}1000 \pm 0.1\text{ mg}$ ) of the sample to which 3 mL of  $\text{HNO}_3$  solution ( $w=65\%$ ) and 0.5 mL of  $\text{H}_2\text{O}_2$  solution ( $w=30\%$ ) were added, at 300 W for 5 min and then at 600 W for 10 min, proved to be satisfactory. After digestion the samples were washed and diluted to 6 mL into a graduated beaker. Demineralised water and reagents of recognised analytical quality were used. The measurements of cadmium were performed on an atomic absorption spectrophotometer ATI UNICAM 929 using an acetylene/air flame, Cd-hollow cathode lamp (228.8 nm line) and deuterium lamp for background correction. The samples were prepared through various tests combined with blank tests and through application of standard SIGMA solution and aspirated directly to the flame. Under the optimised conditions the detection limit (calculated as three times the SD value of blank) was  $10\text{ }\mu\text{g/kg}$ , and blank assay was  $< 5\text{ }\mu\text{g/kg}$ .

Mean recovery values  $\pm$  SD for muscle, liver, kidney and feed mix were in percent:  $97,94 \pm 1,26$ ;  $95,08 \pm 1,56$ ;  $96,99 \pm 1,27$ ;  $101,7 \pm 4,56$ , respectively (number for each type of sample was 12, and the additional quantity of cadmium was equal to the corresponding MRL's). The calculation of 95 %-confidence levels (10) showed that 95 % of the recovery test results was comprised within the limits of 97.4–98.74 % and 94.18–97.8 % for muscle and liver, respectively, while the values for kidney and feed mix were 96.18–97.80 % and 98.8–104, 6 %, respectively.

## Results

Each cadmium result in the Tables 1–6 represents the mean value of two parallel determinations of the same sample, adjusted for appropriate mean recovery value.

The mass fraction levels of cadmium in pigs muscle, liver and kidney are shown in Table 1. Table 2 contains the data on the levels of cadmium in pigs tissue and organs after an extended sample collection where, in accordance with Guidelines provided by the Croatian Ministry of Agriculture and Forestry – Veterinary Direc-

torate (3), the samples of muscle, liver and kidney from five animals (marked respectively with the indices 1–5) were taken at each farm. Table 3 shows the cadmium levels determined in feedmixes: Grover, ST-1 and ST-2 of different composition and caloric values, and which are used for the feeding of pigs weighing 15–20, 25–60 and 60–100 kg. The indices I and II stand for the farm from which the samples were taken. The same Table includes the results for premix. At the Farm III the same feedmixes Grover, ST-1 and ST-2 of the same supplier, were used but their sampling was not possible during the same sampling procedure which included the Farms I and II because the farmer has ordered a limited quantity of the feedmixes which has run out by the time of the inspector's arrival. Considering that some other sub-suppliers from the same area used the feedmixes coming from the same feed mill, Table 4 shows the cadmium levels in pigs muscle, liver and kidney coming from these farms marked from IV to IX. Tables 5 and 6 show the cadmium levels determined in feedmixes and tissues and organs of the pigs coming from all the nine incriminated farms after successful solution of the problem, *i.e.* after the feeding of animals with a newly prepared mixes using a premix from a different source of supply.

## Discussion

In September 1995, during a routine testing in the context of the Monitoring programme of tissue and organs of slaughtered animals for the presence of residues, the levels of cadmium determined in kidney and liver of pigs from 3 farms (I-III) were far above the permissible levels laid down in Croatian regulations (9), precisely, mass fraction  $w$  (Cd)=1 mg/kg for kidney and 0.5 mg/kg for liver, while the levels determined in muscle were considerably below the respective MRL (0.1mg/kg) (Table 1). Considering that cadmium is a highly toxic element, the objective of this research was to discover and eliminate the source of its intake. As provided in the Guidelines of the Croatian Ministry of Agriculture and Forestry – Veterinary Directorate (3), the next step involved an extended sampling in order to determine the exact number of animals in that particular farms with elevated mass fraction levels of cadmium. Regrettably, out of 15 newly tested animals, 12 in kidney samples and 9 in liver samples contained the unpermissible levels of cadmium. As it can be seen from Table 2, the mass fraction levels of cadmium in muscle are mostly below the MRL while those determined in liver and kidney of the animals from all the three farms (I-III) are above the MRL though still lower than those shown in Table 1.

Table 1. Cadmium mass fraction levels  $w$  (Cd)  $\pm$  SD in kidney, liver and muscle of swine samples collected in September 1995

Farm number	$w$ (Cd) / (mg/kg)		
	Kidney	Liver	Muscle
I	7.93 $\pm$ 0.18	0.87 $\pm$ 0.07	0.020 $\pm$ 0.003
II	9.92 $\pm$ 0.16	1.11 $\pm$ 0.08	0.030 $\pm$ 0.004
III	7.76 $\pm$ 0.18	0.94 $\pm$ 0.03	0.020 $\pm$ 0.003

The only explanation of these results is that between the two samplings (September and October) the pigs had obviously been fed with something else and not only with Grover, ST-1 and ST-2, which was later on confirmed in the interviews with the farmers.

Table 2. Cadmium mass fraction levels  $w$  (Cd)  $\pm$  SD in kidney, liver and muscle of swine samples collected in October 1995

Farm number	$w$ (Cd) / (mg/kg)		
	Kidney	Liver	Muscle
I <sup>1</sup>	2.73 $\pm$ 0.11	0.74 $\pm$ 0.04	<0.01
I <sup>2</sup>	2.82 $\pm$ 0.11	0.64 $\pm$ 0.04	<0.01
I <sup>3</sup>	0.87 $\pm$ 0.04	0.37 $\pm$ 0.01	0.010 $\pm$ 0.002
I <sup>4</sup>	2.10 $\pm$ 0.04	0.63 $\pm$ 0.04	<0.01
I <sup>5</sup>	2.50 $\pm$ 0.13	0.42 $\pm$ 0.01	0.010 $\pm$ 0.002
II <sup>1</sup>	0.71 $\pm$ 0.04	0.10 $\pm$ 0.01	<0.01
II <sup>2</sup>	3.36 $\pm$ 0.08	0.67 $\pm$ 0.03	<0.01
II <sup>3</sup>	5.05 $\pm$ 0.10	0.90 $\pm$ 0.03	<0.01
II <sup>4</sup>	1.24 $\pm$ 0.07	0.25 $\pm$ 0.01	<0.01
II <sup>5</sup>	2.91 $\pm$ 0.08	0.63 $\pm$ 0.02	0.010 $\pm$ 0.001
III <sup>1</sup>	2.89 $\pm$ 0.08	0.63 $\pm$ 0.02	0.010 $\pm$ 0.001
III <sup>2</sup>	0.81 $\pm$ 0.03	0.20 $\pm$ 0.01	<0.01
III <sup>3</sup>	3.15 $\pm$ 0.10	0.48 $\pm$ 0.01	<0.01
III <sup>4</sup>	3.88 $\pm$ 0.10	0.73 $\pm$ 0.01	0.010 $\pm$ 0.001
III <sup>5</sup>	4.06 $\pm$ 0.13	0.83 $\pm$ 0.01	<0.01

On the basis of the records of used feedmixes and the data indicated on the ear tags applied to each animal coming from the mini-farms of the sub-suppliers, and in close and constructive cooperation with a veterinary inspector, it was found out that the animals concerned were fed with the finished feedmixes prepared in the same feed mill and, therefore, the samples of the feedmix and of the components thereof were taken for analysis (Table 3). Table 3 shows that in 4 of 5 feedmixes the cadmium mass fraction levels considerably exceeded the legally mandated MRL (11), *i.e.* 0.5 mg/kg.

In the process of the determination of the cadmium mass fraction levels in feed components, it was found out that the premix for pigs, containing 898 mg/kg (Ta-

Table 3. Cadmium mass fraction levels  $w$  (Cd)  $\pm$  SD in feedmixes and premix

Date of sampling (month/year)	$w$ (Cd) / (mg/kg)	
	Feedmix	Cadmium
10/95	Grover I	0.28 $\pm$ 0.01
10/95	ST-1 <sup>I</sup>	4.05 $\pm$ 0.10
10/95	ST-2 <sup>I</sup>	2.37 $\pm$ 0.08
11/95	ST-1 <sup>II</sup>	4.86 $\pm$ 0.07
11/95	ST-2 <sup>II</sup>	2.05 $\pm$ 0.11
11/95	Premix	898 $\pm$ 14

ble 3) caused the contamination of the feedmix and accumulation of this toxic element in kidney and liver of the tested animals. Through continuing cooperation with the veterinary inspection, the samples of muscle, liver and kidney were taken from the sub-suppliers pigs coming from other farms (IV–IX) using the feed from the same feed mill. The results contained in Table 4 show that all muscle samples meet the requirements laid down by Croatian Regulation (9) while out of six kidney and liver samples from November in as many as five kidney samples and three liver samples the determined mass fraction levels were by many times higher (in kidney even up to 13 times higher) than the MRL's laid down by Croatian Regulation (9). After having prepared the feedmixes with another premix (new batch with 0.48 mg/kg), the analysis showed that in all three tested feedmixes the cadmium levels were considerably below MRL's laid down in Croatian Regulation (11) as shown in Table 5. Subsequently the contaminated feedmixes were withdrawn and the new were distributed to the sub-suppliers and after several months, precisely in March 1996, the sampling of animal tissue and organs was repeated at all farms (I–IX) where the contamination with cadmium had been discovered.

The test results, indicated in Table 6, show that in all the nine tested animals, the levels of cadmium in kidney and liver did not exceed the MRL.

Table 4. Cadmium mass fraction levels  $w$  (Cd)  $\pm$  SD in kidney, liver and muscle of swine after the feeding of animals with contaminated feedmixes on the other farms; samples collected in November 1995

Farm number	$w$ (Cd) / (mg/kg)		
	Kidney	Liver	Muscle
IV	0.67 $\pm$ 0.03	0.070 $\pm$ 0.003	<0.01
V	2.93 $\pm$ 0.04	0.450 $\pm$ 0.003	0.030 $\pm$ 0.003
VI	2.38 $\pm$ 0.06	0.360 $\pm$ 0.002	0.030 $\pm$ 0.003
VII	6.85 $\pm$ 0.08	0.83 $\pm$ 0.04	0.030 $\pm$ 0.002
VIII	12.96 $\pm$ 0.14	1.69 $\pm$ 0.05	0.040 $\pm$ 0.003
IX	7.32 $\pm$ 0.11	1.42 $\pm$ 0.04	0.020 $\pm$ 0.002

Table 5. Cadmium mass fraction levels  $w$  (Cd)  $\pm$  SD in newly prepared feedmixes; samples collected in December 1995

Feedmix	$w$ (Cd) / (mg/kg)
Grover	0.12 $\pm$ 0.01
ST-1	0.11 $\pm$ 0.01
ST-2	0.16 $\pm$ 0.01
premix-new batch	0.48 $\pm$ 0.01

The analytical results clearly show that elevated levels of cadmium in kidney and liver resulted from the feeding the pigs with feedmixes in which the cadmium content was considerably above the permissible level (11). Namely, as it can be seen from Table 3, the absolute source of the feed contamination was the premix, which contained extreme levels of cadmium – almost 900 mg/kg (the source of cadmium was a zinc salt con-

Table 6. Cadmium mass fraction levels  $w$  (Cd)  $\pm$  SD in kidney, liver and muscle of swine after the feeding of animals with newly prepared feedmixes; samples collected in March 1996

Farm number	$w$ (Cd) / (mg/kg)		
	Kidney	Liver	Muscle
I	0.34 $\pm$ 0.01	0.10 $\pm$ 0.01	<0.01
II	0.21 $\pm$ 0.01	0.030 $\pm$ 0.004	<0.01
III	0.63 $\pm$ 0.01	0.09 $\pm$ 0.01	<0.01
IV	0.19 $\pm$ 0.01	0.09 $\pm$ 0.01	<0.01
V	0.49 $\pm$ 0.01	0.06 $\pm$ 0.01	<0.01
VI	0.41 $\pm$ 0.02	0.070 $\pm$ 0.004	<0.01
VII	0.60 $\pm$ 0.01	0.10 $\pm$ 0.01	<0.01
VIII	0.59 $\pm$ 0.02	0.06 $\pm$ 0.01	<0.01
IX	0.54 $\pm$ 0.02	0.04 $\pm$ 0.01	<0.01

taminated with cadmium). This caused direct material damages since the kidneys and livers of all animals slaughtered in that particular slaughterhouse were systematically condemned and destroyed. Timely discovery of the above described contamination with cadmium of feedmixes, liver and kidney of pigs, thanks to efficient functioning of veterinary inspection and residue monitoring under the Croatian National Monitoring Programme (3), harmful effects upon consumer's health were prevented. Possible harmful effects upon public health are indicated by the fact that, taking into account the determined mean mass fraction levels of cadmium in kidney (4.08 mg/kg) and liver (0.67 mg/kg) (Table 1, 2 and 4) and despite the relatively low average consumption of these organs in Croatia (only 26 g/person/week) (12), assuming that the kidneys and livers have been equally consumed (50 % of kidney and 50 % of liver), the weekly intake would have amounted to 61.75  $\mu$ g/person (calculated on 70 kg body weight basis). This is equal to 12.6 % of the provisional tolerable weekly intake PTWI recommended by WHO, *i.e.* 7  $\mu$ g/kg of body weight per week (13). These values would greatly exceed the ones determined in our earlier research when it was estimated that the total dietary weekly intake of cadmium by the Croatian population and the cadmium intake through the consumption of kidney and liver was 121.4 and 5  $\mu$ g/person, respectively, which is equal to 24.4 and 1 % of PTWI (8).

Notwithstanding the fact that the data on the cadmium intake through foodstuffs greatly vary in different countries, it can be concluded that the consumption of the analysed kidneys and livers (expressed in % of PTWI) would result in a cadmium intake which is almost equal to the total human weekly cadmium intake in some countries. For example, total weekly cadmium intake for the population of Germany, Canada and Belgium amounts to 10–13, 6–9 and 7.1 %, respectively, (14–16) which is similar to our possible intake (12.6 % of PTWI).

## Conclusion

This paper shows that the kidneys and livers contaminated with unpermissible levels of cadmium were

not used neither for human nor for animal food, thanks to applicable legal regulations and well-organised national control system.

However, the fact is that the examined organs contained a maximum cadmium mass fraction level of 1.69 mg/kg for liver and as much as 12.96 mg/kg for kidney, as a result of feeding the pigs with cadmium contaminated feed (max. level 4.8 mg/kg), and that the consumption of such organs would have resulted in a cadmium intake equal to 12.6 % of PTWI, which is close to the total human weekly cadmium intake in some countries.

It is believed that this case proves the importance of regular and systematic control, not only of animals and their tissues and organs, but also of animal feed and its ingredients, as provided in the Croatian National Residue Monitoring Programme (3). Adequate control means a sufficient number of representative samples of animal tissue, animal feed and the analysis of all relevant parameters. This is thought to be the best way to provide, within the veterinary public health surveillance system, the animal health protection and of course, protection of ourselves and of the generations to come.

## References

1. Decree of the Royal Government of Croatia, Slavonia and Dalmatia, Department of the Interior, Nr. 2355, Zagreb (1877) pp. 1–41.
2. J. Živković, *Vet. stanica*, 24 (1989) 339.
3. Guidelines of the Croatian Ministry of Agriculture and Forestry – Veterinary Directorate, U.Nr. 525-06-93-1, Zagreb (1993).
4. M. Stoepller: Cadmium. In: *Metals and Their Compounds in the Environment Occurrence, Analysis and Biological Reference*, E. Marian (Ed.), VCH Weinheim (1991) pp. 803–851.
5. K. Roberts, P. Worsfold, *Analyst*, 116 (1991) 549.
6. B. A. Fowler, *Environ. Health Persp.* 100 (1993) 57.
7. R. R. Lauwerys, A. M. Bernard, H. A. Roels, J. P. Buchet, *Clin. Chem.* 40 (1994) 1391.
8. J. Sapunar-Postružnik, D. Bažulić, H. Kubala, L. Balint, *Sci. Total. Environ.* 177 (1996) 31.
9. Regulation of the permissible levels of pesticides, toxins, mycotoxins, metals and histamines and similar substances that may be present in foodstuffs and on other terms and conditions governing the safety of foodstuffs and objects intended for general use, Official Gazette Nr. 46, Zagreb, Croatia (1994) pp. 1579–1586.
10. V. Vranić: *Vjerojatnost i statistika*, Tehnička knjiga, Zagreb (1971).
11. Regulation of the maximum permissible levels of harmful substances and ingredients in animal feed, Official Gazette Nr. 4, Zagreb, Croatia (1994) pp. 65–67.
12. Statistical Yearbook of the Republic of Croatia, Central Bureau of Statistics, Zagreb, Croatia (1992) p. 213.
13. FAO/WHO, Toxicological evaluation of certain food additives and contaminants. Forty-first report of the Joint FAO/WHO Expert Committee on Food Additives (WHO Technical Report Series, No. 837, WHO, Geneva (1993).
14. M. Seifert, M. Ankle, *J. Trace Microbe Tech.* 17 (1999) 101.
15. C. Kim, H. M. Chan, O. Receveur, *Food Addit. Contam.* 15 (1998) 307.
16. Q. Yang, K. Desmit, J. Smeyersverbeke, *J. Agr. Food. Chem.* 43 (1995) 2652.

## Kadmij u krmivu i namirnicama životinjskog podrijetla

### Sažetak

U radu je istaknuta nužnost sustavne kontrole rezidua, kojom se »Nacionalnim monitoring programom« na vrijeme otkriva uzrok onečišćenja životinjskih organa kadmijem, a sustavnom kontrolom i postupcima rješava cjelokupan problem onečišćenja namirnica u relativno kratkom vremenskom razdoblju. Kao rezultat hranjenja svinja krmivom onečišćenim kadmijem ( $w$  (Cd) = 2,05–4,86 mg/kg) u bubrezima je utvrđena količina u rasponu od  $w$  (Cd) = 0,67–12,96 mg/kg, a u jetri od  $w$  (Cd) = 0,07–1,69 mg/kg. Maseni udjel kadmija u uzorcima mišića bio je u rasponu od <0,01–0,04 mg/kg. Nakon uklanjanja onečišćenog krmiva, maseni udjel kadmija u organima i tkivima snizio se na dopuštenu razinu ( $w$  (Cd) = 0,19–0,63 mg/kg u bubrezima;  $w$  (Cd) = 0,03–0,1 mg/kg u jetri i  $w$  (Cd) < 0,01 mg/kg u mišićima).