VETERINARSKI ARHIV 72 (6), 303-310, 2002

Cadmium in tissues of roe deer (*Capreolus capreolus*) in Croatia

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POMPE-GOTAL, J., A. PREVENDAR CRNIĆ: Cadmium in tissues of roe deer (*Capreolus capreolus*) in Croatia. Vet. arhiv 72, 303-310, 2002.

ABSTRACT

Roe deer tissues (muscle, liver and kidney: 21 samples severally) collected (without regard to sex and age of donors) in the northern region of Croatia in the period 1990-1995 were analysed for cadmium. This toxic heavy metal occurred in low concentration in muscle tissue, although livers and especially kidneys were more heavily contaminated. Forty-three per cent of liver samples and 90% of kidney samples exceeded maximal permissible values and were unfit for human consumption. Concentrations in mg/kg on wet weight basis were: 0.003-0.065, 0.015-2.306 and 0.223-27.686 for muscle, liver and kidney, respectively. The values obtained were compared with available literature data.

Key words: roe deer, cadmium

Introduction

Cadmium (Cd) is recognized as a hazardous environmental pollutant due to its toxicity to many organisms (RUPARELIA et al., 1990). It is a naturally occurring element found in trace quantities throughout the environment (crustal abundance 0.16 mg/kg, EMSLEY, 1989). Its use in industrial processes, such as electroplating, pigment and plastics production and battery

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ISSN 0372-5480
Printed in Croatia

manufacturing increased 400-fold between 1907 and 1976 (STANSLEY et al., 1991). Release of cadmium into the environment occurs from natural sources, industrial waste and from other practices such as coal and fossil fuel combustion, and the land application of sewage sludge and phosphate fertilizer (ROBARDS and WORSFOLD, 1991).

Animals exposed to Cd accumulate the element in their livers and kidneys (SPIERENBURG et al., 1988) as their free proteins-thiol group content leads to a strong fixation of heavy metals. Despite the excretory mechanism for such metals, which is based on low molecular compounds with -SH groups, vertebrae could not develop these mechanisms during the period of evolution to the extent which would be necessary for today's anthropogenic sources of pollution.

The herbivores of terrestrial fauna, birds as well as mammals, demonstrate generally higher renal Cd levels than carnivores since the vegetation is contaminated either by aerial deposition or by absorption of Cd from the soil (PETERSON and ALLOWAY, 1979). Roe deer were found to be suitable as a biomonitor of heavy metal pollution because of their common occurrence, their uniform geographical distribution and their stationariness (FRANK, 1986).

This paper presents our findings on Cd concentrations in the muscle, liver and kidney tissues in roe deer from northern region of the Republic Croatia.

Materials and methods

Tissue samples of animals were taken from randomly selected specimens hunted mainly in the north-western part of Croatia (Medvednica, Vrbovec, Kupčina, Tuhelj - total of 17 animals) and partly in the northern region of Podravina (Đurđevac, Slatina - total of 4 animals) in the period 1990-1995. The animals were not selected according to sex or age but on the acknowledged assumption that they were aged from 2 to 5 years.

After dissection tissues were packed separately and frozen. They were stored at -18 °C and analyzed as soon as possible. The samples were dried and ashed in a thermostated muffle furnace at 450 °C overnight (NIEMI et al., 1991). Ashed samples were dissolved in deionized water and concentrated

nitric acid and then evaporated and heated to 450 °C in a muffle furnace. The ash was redissolved in diluted nitric acid. Cadmium was measured by flame atomic absorption spectrophotometry using Pye Unicam SP192 atomic absorption spectrophotometer fitted with a deuterium lamp for background correction at 228.8 nm with a hollow cathode lamp. The measurements were carried out by comparison with standard solution (Titrisol, Merck art. 9960). For checking accuracy and reproducibility, two blanks and certified reference material - lyophilized bovine liver purchased from the National Institute of Standards and Technology (SRM 1577a) - were analyzed along with tissue samples. The results are given as mg/kg on a wet mass basis. Statistical analysis was performed using the computer program Statgraphics 4.0.

Results

Cadmium concentrations in muscle, liver and kidney tissues are summarized in Table 1. and Figure 1. The arithmetic means with standard deviations and coefficients of variation, followed by median values, minimum and maximum concentrations, as well as the percentage of samples exceeding the permitted values defined by Croatian regulations (Narodne

Table 1. Cadmium in tissues in roe deer shot in the north-western region of Croatia in the period 1990-1995 (mg/kg wet mass)

	Muscle	Liver	Kidney
N° of samples	21	21	21
Average	0.018	0.568	4.905
Standard deviation	0.019	0.502	6.395
Coef. of variation	102 %	88 %	130.4 %
Min. - max.	0.003 - 0.065	0.015 - 2.306	0.223 - 27.686
Median	0.008	0.424	2.278
Lower quartile	0.005	0.321	1.225
Upper quartile	0.023	0.719	5.847
Excess[1]	0 %	43 %	90 %

[1] Percentage of samples exceeding the permitted values defined by Croatian Regulations (NN 46/94 and NN 11/01) given as: Cd: meat (0.1 mg/kg), viscera (0.5 mg/kg).

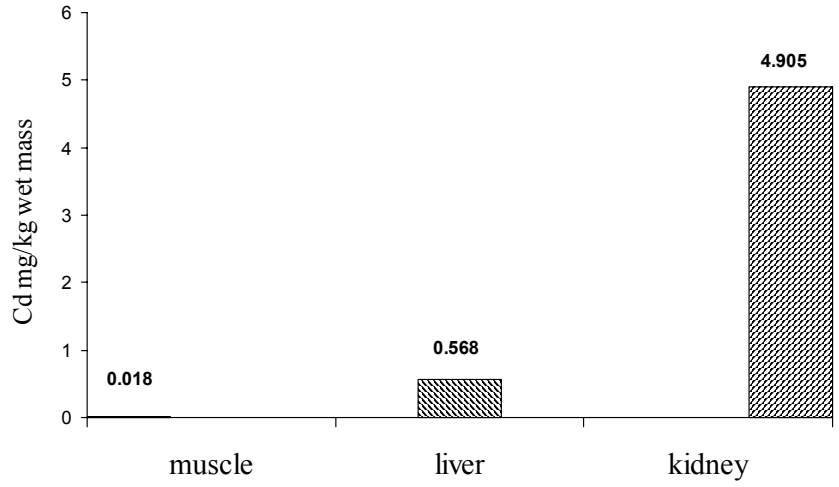

Fig. 1. Cadmium in tissue samples of roe deer (*Capreolus capreolus*) collected in Croatia in the period 1990-1995

novine NN 46/94, pp. 1579-1586 and Narodne novine NN 1/01, pp. 300-302), are shown in Table 1. Fig. 1. represents mean muscle, liver and kidney Cd concentrations for all samples.

Duplicate analysis of certified reference standard bovine liver performed together with deer tissue samples provided results which are both accurate and precise. Certified trace metal concentrations are in good agreement with obtained analytical results:

Certified value: 0.050 ± 0.03 μg/g

Obtained results: 0.053, 0.055 μg/g

Discussion

Cadmium can be easily absorbed by plants and then accumulated in the tissues of herbivores, mostly in the liver and kidneys (WILLOUGHBY, 1979). Primary Cd uptake from soil by plants and secondary Cd intake via

Table 1. Specification of myomorphus mammals examined by renoculture and microscopic agglutination acording to the trapping area with corresponding results

food by animals is the obvious explanation for the higher levels of Cd found in herbivores than in carnivores (FRANK, 1986). The Cd in soil is of natural origin but, as already mentioned, it is also anthropogenic to some extent. Thus, the variation in the environmental Cd burden may be monitored by herbivorous animals.

The cadmium concentrations in 50% of muscle samples were less than 0.005 mg/kg with arithmetic mean 0.018 and median value 0.008 mg/kg, which is about 10 times less than the permitted value (NN, 1994 and NN, 2001).

In comparison with Cd content in deer kidneys and livers from other European regions, our results indicate a marked contamination of these organs with cadmium. The level of 0.568 ± 0.502 mg/kg (mean ± s.d.) or 0.424 mg/kg (median) in livers and 4.905 ± 6.395 mg/kg (mean ± s.d.) or 2.278 mg/kg (median) in kidneys implies the same degree of exposure as in polluted areas. These results are comparable with findings for liver and kidney tissues in industrialised regions of Germany established in the 1980s by RIMKUS and WOLF (1987), indicating 0.450 mg/kg mean value in livers and 3.800 mg/kg mean and 20.000 mg/kg, the greatest value measured in kidney tissue; and by HOLM et al. (1987), who found 0.050- 0.980 mg/kg in liver and 0.700-15.000 mg/kg in kidneys. Despite the broad range of Cd concentrations in our analyzed kidney samples, with an extreme value of 27.7 mg/kg found in roe deer hunted in the locality of Kupčina in the centre of the triangle Zagreb-Sisak-Karlovac, the quantity of 1.225 for the lower quartile means that only 25% of specimens are below this concentration. Ninety per cent of kidneys and 43% of livers (Table 1) exceed maximal permissible concentrations (NN, 1994; NN, 2001) and are unfit for human consumption.

Cd content in kidneys is about ten times higher than in liver. Cadmium tends to accumulate in kidney and, to a lesser extent, in liver. These findings correspond with results obtained from other researches in different regions (HECHT et al., 1984; FROSLIE et al., 1986; MUSANTE et al., 1993; FALANDYSZ, 1994; DOGANOC and GAČNIK, 1995; KOTFEROVA and KORENEKOVA, 1998; POKORNY and RIBARIĆ-LASNIK, 2000). Correlations of Cd concentrations between different organs provide information about the mechanism of

accumulation. In juvenile roe deer liver and kidney Cd concentrations correlate positively (FROSLIE et al., 1986). In general, the liver is thought to be the initial storage site for Cd, where it may be bound to a metallothionein, then transported to the kidney, which is the main and final storage site for Cd (SAMARAAWICKRAMA, 1979). However, upon chronic exposure the kidneys are also considered to take up Cd directly from the blood (FRAZIER, 1982). In older deer there is a relative increase of Cd transport from liver to kidney and/or an increase in the direct uptake of Cd by kidney from the blood, which may be because of insufficient binding capacity in the liver as exposure to Cd persists (WALKERS et al., 1994). These findings, together with information about an extremely long retention period (reaching 10 to 30 years in kidney and livers of mammals) (COOKE and JOHNSON, 1996) is the source of a positive correlation between age of animal and Cd concentration in the viscera. The analyzed specimens, issuing from animals aged two or more years, confirm this correlation.

Average as well as maximum Cd concentrations in liver and kidney in our research exceeded the majority of the levels measured in viscera of European free-living ruminants. Nevertheless, the levels are well below the concentrations dangerous for the animals' health, but confirm the obligation to eliminate kidneys and livers of roe deer as a food source in Croatia. The levels of cadmium in the meat of roe deer do not exceed values permitted by law.

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Received: 12 April 2002
Accepted: 20 December 2002

POMPE-GOTAL, J., A. PREVENDAR CRNIĆ: Koncentracija kadmija u tkivima srneće divljači *(Capreolus capreolus)* u Hrvatskoj. Vet. arhiv 72, 303-310, 2002.

SAŽETAK

Uzorci tkiva srneće divljači (po 21 uzorak mišićnog, jetrenog i bubrežnog tkiva) sakupljeni u sjevernom dijelu Hrvatske (Medvednica, Vrbovec, Kupčina, Tuhelj, te Đurđevac i Slatina) u razdoblju od 1990.-1995. analizirani su na sadržaj kadmija. Uzorci nisu sakupljani sustavno s obzirom na dob i na spol. Kadmij je dokazan u maloj koncentraciji u mišićnom tkivu, ali su jetra i bubrezi bili značajno kontaminirani. Koncentracija je prelazila zakonom maksimalno dopuštene vrijednosti u 43% uzoraka jetrenog tkiva i čak 90% uzoraka bubrežnog tkiva. Raspon koncentracija kadmija izražen u mg/kg vlažne težine uzorka iznosio je: za mišićno tkivo 0,003-0,065; za jetreno tkivo 0,015-2,306; za bubrežno tkivo 0,223-27,686. Dobiveni rezultati uspoređeni su s koncentracijom kadmija u uzorcima srneće divljači u drugim europskim zemljama.

Ključne riječi: srneća divljač, kadmij