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Lead and Cadmium in Red Deer and Wild Boar from Sierra Morena Mountains (Andalusia, Spain)

D. Santiago,[2] M. Motas-Guzmán,[1] A. Reja,[2] P. María-Mojica,[1] B. Rodero,[2] A. J. García-Fernández[1]

[1] Department of Toxicology, School of Veterinary Medicine, University of Murcia, 30071, Murcia, Spain
[2] Department of Pharmacology and Toxicology, School of Veterinary Medicine, University of Córdoba, 14071, Córdoba, Spain

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Lead and cadmium are industrial poisons which may have important consequences for human beings and for domestic and wild animals, either through acute or chronic exposure, since these heavy metals are accumulated in the organism, mainly in liver and kidney. This paper presents the results of an investigation into the levels of lead and cadmium found in the liver and kidney of wild boar and deer in several areas of Andalusia (Southern Spain) as a useful tool for evaluating environmental contamination. In addition, the human and animal health risks are considered.

The Sierra Morena Mountains in the north of Cordoba Province (Andalusia) is an area which derives considerable economic benefits from hunting. Red deer (*Cervus elaphus*) and wild boar (*Sus scofra baeticus*) are pursued in 422 hunting grounds that spread through a vast tract of Mediterranean forest ecosystem. The number of deer and wild boar killed each hunting season is near to ten thousand, and this activity yields more than a thousand tons of extremely edible and well-appreciated meat. Although pesticide and metal contamination in wild fauna species from Spain have been studied (García-Fernández et al. 1995; 1996; 1997; Sierra et al. 1987; Sierra and Santiago 1987) the levels of natural contamination by organometallic and other pesticide residues has not been completely established in wild boar and deer. Some results of an analytical survey of metallic contamination of these two species carried out during the 1993 and 1994 hunting seasons are shown in this paper. The concentrations of lead and cadmium found in the liver and kidney of these species pointed to these metals as being the most frequent toxic metallic pollutants in the Mediterranean forest ecosystem of the area studied.

MATERIALS AND METHODS

Four areas of the Sierra Morena Mountains were selected for sampling according to the agro-biological and natural conditions and the degree of the anthropogenic influence (Figure 1). The first area is the eastern most, which is forestland with the occasional cultivation of olive trees. From here, we obtained samples from 10 deer and 2 wild boar. The second area is the central area, a wooded and uncultivated zone, from which we obtained samples from 10 deer and 13 wild boar. The third area is the most northerly. Mining is carried on near the hunting ground and there is

Correspondence to: A. J. García-Fernández

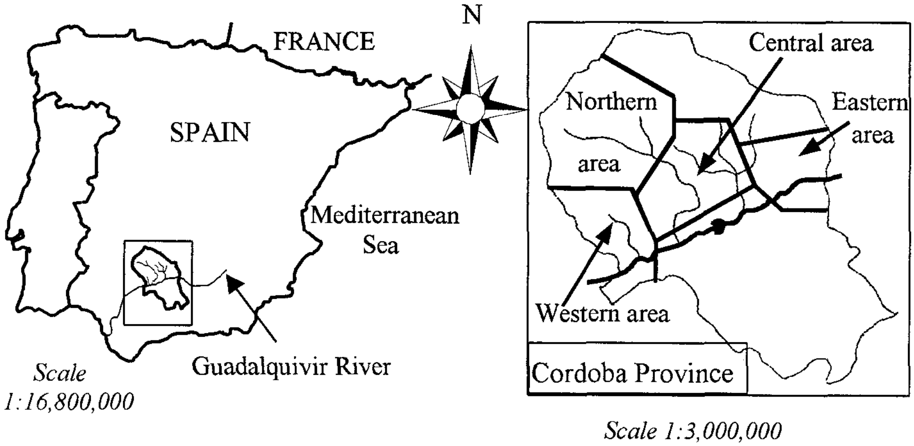

Figure 1. Maps showing the geographical location of Cordoba Province and the studied areas.

a power station, which is closed. From this area we obtained samples from 26 deer and 8 wild boar. Finally, the western most area chosen is in the centre of a highland with extensive grazing of sheep and cattle. Here, we obtained samples from 14 deer and 6 wild boar. In all, then, liver and kidney samples were taken from a total of 67 deer and 29 wild boar. It should be noted that all the waters and tributary streams run from north to south towards the Guadalquivir River, which is located to the South of the Sierra Morena Mountains. The sampled areas are separated by a maximum of 90 Km and each zone has its own sources of possible pollution.

The Anodic Stripping Voltammetry (ASV) analysis method, as described in our previously published paper (García-Fernández et al. 1995) was used to determine the concentrations of lead and cadmium. The method consisted of wet digestion of samples (0.2-0.3 g) using 0.5 ml of an acid mixture (nitric/perchloric/sulfuric, 8:8:1). The samples were then submitted to a progressive thermal treatment. A voltammeter with a VA-646 processor and a VA-647 workstation (Metrohm, Switzerland) was used with analytical standards of cadmium and lead (Sigma, St. Louis, Mo) as references. The detection limits were 0.0025 ppm for cadmium and 0.005 ppm for lead. The repeatability, determined by analyzing 10 identical samples of reconstituted lyophilized blood (European Union Reference Standards) CRM195, was 96.5 ± 1.2% for lead, and 94.2 ± 3.2% for cadmium. To calculate the recovery percentage, we processed 10 samples of each tissue, which had been spiked with known amounts of the lead and cadmium analytical standards. The mean recoveries obtained were 101.2-102.3% (blank), 103.2-100.6% (liver) and 101.9-103.4% (kidney).

Data were grouped according to species, tissue and sampling area. Concentrations were expressed as mean ± standard deviation, minimum and maximum values and median, T tests for independent samples were used to examine differences both between species and between tissues. To examine differences between the sampling areas we used the One-way analysis of variance (ANOVA) test. All statements of significance were based on the 0.05 level of probability.

RESULTS AND DISCUSSION

The concentrations of lead and cadmium found in red deer and wild boar from different areas of the Sierra Morena Mountains are presented in tables 1 and 2. The mean concentrations of cadmium in the kidney of both species were significantly higher than those found in liver, findings which reflect those described by Glooschenko et al. (1988) and Wolkers et al. (1994) in the same species. However, we observed differences between species as regards lead distribution. In deer, the mean lead concentration in liver was very similar to that found in kidneys, while in wild boar the mean hepatic levels of lead were slightly higher than renal levels, although the difference was not statistically significant. Such distributions of lead in deer and wild boar do not resemble those described by the authors cited above, Wolkers et al. (1994), for example, observing higher renal lead concentrations than hepatic levels in both species.

In deer, renal levels of cadmium ranged from 0.40 to 7.61 mg/kg wet wt (mean=1.35 mg/kg) and in wild boar these levels ranged from 0.39 to5.48 mg/kg wet wt (mean=2.16 mg/kg), differences which were statistically significant. However, the concentrations of cadmium in liver were similar in both species (0.28 and 0.21 mg/kg wet wt in wild boar and deer, respectively). On the other hand, the mean concentrations of lead in the livers (0.57 mg/kg wet wt) and kidneys (0.33 mg/kg wet wt) of deer were significantly lower than those in wild boar (2.61 mg/kg in liver and 0.62 mg/kg in kidney). The highest lead concentrations in liver and kidney were found in wild boar (43.06 and 4.34 mg/kg, respectively), while the highest lead levels in deer were very much lower (10.4 1 mg/kg in liver and 2.15 mg/kg in kidney). Swiergosz et al. (1993) did not find differences between red deer and wild boar.

Our results demonstrate that lead accumulates to a greater extent in wild boar whereas cadmium does so in deer. Such findings could be explained by the feeding habits and the environmental presence of these metals (García-Fernández et al. 1995; 1996; 1997). Since both metals are present in all living organisms and are bio-accumulated through the food chain. Wild boar is an omnivorous species and lead is present in all ecosystems at much higher levels than cadmium. However, deer is a herbivore and it is known that the leaves and roots of plants easily assimilate environmental cadmium.

Environmental exposure to cadmium determines a distribution pattern in which the renal levels are much higher than the hepatic levels (Bernard and Lauwerys 1984; Friberg et al. 1974; García-Fernandez et al. 1996; Scheuhammer 1987). The ratio between cadmium concentration in liver and in kidney increases with the intensity of exposure (Friberg et al. 1974). In addition, because cadmium is transported from liver to kidney very slowly, it is reasonable to assume that there has been acute exposure to high cadmium levels when liver concentrations are higher than renal levels (Scheuhammer 1987). In all the animals studied, the hepatic-cadmium/renal-cadmium ratios were much lower than 1, and so it can be concluded that they had been exposed chronically to environmental cadmium. However, these quotients were significantly lower (P<0.001) in deer (mean = 0.09) than in wild boar (mean = 0.18), with Cd-kidney/Cd-liver ratios of 10-11/1 and 4-5/1 in deer and in wild boar, respectively. Since the hepatic levels were similar in both species studied, it is possible that the different feeding habits determined the two different grades of chronic exposure to environmental cadmium.

Table 1. Lead and cadmium concentrations (mg/kg wet wt) in red deer seized in four different hunting areas of the Sierra Morena Mountains (Southern Spain)

		Cd liver	Cd kidney	Pb liver	Pb kidney
All areas	Mean±S.D.	0.21 ± 0.14	2.16 ± 1.05	0.57 ± 1.53	0.33 ± 0.32
	(min-max)	(0.08-0.96)	(0.39-5.48)	(0.05-10.41)	(0.04-2.15)
	median (n)	0.18 (70)	2.04 (63)	0.23 (70)	0.26 (63)
Northern area	Mean±S.D.	0.23 ± 0.18	2.11 ± 0.88	0.56 ± 1.48	0.35 ± 0.23
	(min-max)	(0.10-0.96)	(1.07-4.06)	(0.10-7.79)	(0.08-1.08)
	median (n)	0.19 (26)	1.82 (26)	0.27 (26)	0.31 (26)
Central area	Mean±S.D.	0.22 ± 0.15	2.66 ± 1.33	0.88 ± 2.31	0.36 ± 0.54
	(min-max)	(0.08-0.74)	(0.67-5.48)	(0.05-10.41)	(0.04-2.15)
	median (n)	0.19 (20)	2.34 (14)	0.15 (20)	0.22 (14)
Eastern area	Mean±S.D.	0.16 ± 0.08	1.82 ± 1.30	0.37 ± 0.30	0.19 ± 0.04
	(min-max)	(0.08-0.34)	(0.43-4.22)	(0.15-1.12)	(0.14-0.25)
	median (n)	0.14 (10)	1.37 (9)	0.25 (10)	0.19 (9)
Western area	Mean±S.D.	0.21 ± 0.08	1.98 ± 0.76	0.30 ± 0.22	0.35 ± 0.28
	(min-max)	(0.11-0.40)	(0.39-3.53)	(0.10-0.93)	(0.06-0.93)
	median (n)	0.19 (14)	2.04 (14)	0.23 (14)	0.29 (14)

Chronic exposure to environmental lead and cadmium can produce many toxic effects. These elements can alter the functioning of the kidney tubules and also have toxic effects on the male reproductive system, leading to decreased on spermatogenesis and testicular atrophy (Lu 1991). In a calf showing nervous symptoms, Seimiya et al. (1991) found 1.3 and 9.4 ppm of lead in liver and kidney, respectively. One deer from each of the northern and central areas and two wild boars from the central area had higher hepatic lead concentrations than those cited above, and so it is possible that some of the studied animals hunted could have been suffering from lead poisoning.

Table 2. Lead and cadmium concentrations (mg/kg wet wt) in wild boar seized in four different hunting areas of the Sierra Morena (Southern Spain).

		Cd liver	Cd kidney	Pb liver	Pb kidney
All areas	Mean±S.D.	0.28 ± 0.16	1.35 ± 1.42	2.61 ± 8.35	0.62 ± 0.90
	(min-max)	(0.07-0.73)	(0.40-7.61)	(0.11-43.06)	(0.10-4.34)
	median (n)	0.26 (29)	1.01 (25)	0.43 (29)	0.35 (25)
Northern area	Mean±S.D.	0.28 ± 0.13	1.25 ± 0.68	0.68 ± 0.93	0.93 ± 1.39
	(min-max)	(0.10-0.52)	(0.40-2.07)	(0.15-2.89)	(0.33-4.34)
	median (n)	0.29 (8)	1.39 (8)	0.29 (8)	0.40 (8)
Central area	Mean±S.D.	0.34 ± 0.19	1.78 ± 2.24	5.05 ± 12.25	0.42 ± 0.61
	(min-max)	(0.10-0.73)	(0.52-7.61)	(0.12-43.06)	(0.10-2.02)
	median (n)	0.31 (13)	0.96 (9)	0.46 (13)	0.30 (9)
Eastern area	Mean±S.D.	0.17 ± 0.14	1.09 ± 0.93	0.44 ± 0.18	0.31 ± 0.13
	(min-max)	(0.07-0.26)	(0.43-1.74)	(0.31-0.56)	(0.22-0.40)
	median (n)	0.17 (2)	1.09 (2)	0.44 (2)	0.31 (2)
Western area	Mean±S.D.	0.19 ± 0.12	0.94 ± 0.53	0.64 ± 1.01	0.60 ± 0.61
	(min-max)	(0.07-0.38)	(0.40-1.56)	(0.11-2.68)	(0.16-1.76)
	median (n)	0.15 (6)	0.89 (6)	0.22 (6)	0.39 (6)

Several authors (Crête et al. 1987; Kocan et al. 1980; Stansley et al. 1991) found regional differences in cadmium and lead levels in the liver and kidneys of deer and wild boar. We suspect that lead and cadmium are accumulated in the tissues of wild boar and deer from the northern area with its mining activities to a greater extent than in other areas. The fact that the tributaries of the Guadalquivir River flow towards the south had led us to suspect that the central area would be more polluted than the other zones. The results confirmed these suspicions and significant differences were found between the four areas. Wild animals from the central and the northern area, had the highest mean concentrations of lead and cadmium in both tissues while the lowest mean concentrations were found in tissues of animals from the eastern area. We can conclude that the northern and central areas present more risks than the other two areas of the Sierra Morena Mountains.

In Spain, regulatory agencies have not yet established the maximum concentrations permitted for cadmium and lead in the liver and kidney of animals destined for human consumption. In Czechoslovakia, the limit for lead in tissues is 1 mg/kg in calves and for cadmium in liver and kidney 0.5 and 1 mg/kg, respectively (Cibulka et al. 1989). In Canada, the action level of lead in liver and kidney is 2 mg/kg and for cadmium is 1 mg/kg (Korsrud et al. 1985; Salisbury et al. 1991). In Germany, the values for cadmium are 0.5 mg/kg in liver and 1 mg/kg in kidney (Kreuzer and Rosopulo 1981). In Australia, the maximum cadmium concentration permitted in the liver is 2.5 mg/kg (Kramer et al. 1983). Faced with the lack of regulatory limits for cadmium and lead in these organs in Spain, we propose a limit of 1 mg/kg, above which we suggest there is a reason to investigate the source of contamination.

Because the statistical studies of the lead concentrations found in liver showed a high standard deviation, we used the geometric mean of these levels instead of the arithmetic mean. The geometric means of renal cadmium in deer and wild boar were above 2 and 1 mg/kg, respectively. However, the mean concentrations of lead in liver and kidney in both species were below 0.5 mg/kg. Table 3 shows the frequency distribution of lead and cadmium in these species. The incidence of lead levels above 1 mg/kg were 5.7% and 3.2% in deer livers and kidneys, respectively; and in wild boar they were 17.2% and 12% in livers and kidneys, respectively. No animals had hepatic cadmium levels above 1 mg/kg, although, in kidney 92.1% of deer and 52% of wild boar exceeded this concentration.

Table 3. Frequency distribution (%) of lead and cadmium concentrations in red deer and wild boar from Sierra Morena Mountains (Southern Spain).

		Liver			Kidney		
Lead (mg/kg w.w.)		< 1	1-2	> 2	< 1	1-2	> 2
Cadmium (mg/kg w.w.)		< 0.5	0.5-1	> 1	< 1	1-3	> 3
All areas	Pb in deer	94.3	1.4	4.3	96.8	1.6	1.6
	Pb in wild boar	82.8	3.4	13.8	88.0	4.0	8.0
	Cd in deer	95.7	4.3	-	7.9	71.5	20.6
	Cd in wild boar	86.2	13.8	-	48.0	48.0	4.0
Northern Area	Pb in deer	96	-	4	96	4	-
	Pb in wild boar	88	-	12	88	-	12
	Cd in deer	92	8	-	-	81	19
	Cd in wild boar	88	12	-	38	62	-
Central Area	Pb in deer	90	-	10	93	-	7
	Pb in wild boar	76	8	16	89	-	11
	Cd in deer	95	5	-	7	57	36
	Cd in wild boar	77	23	-	56	33	11
Eastern Area	Pb in deer	90	10	-	100	-	-
	Pb in wild boar	100	-	-	100	-	-
	Cd in deer	100	-	-	22	56	22
	Cd in wild boar	100	-	-	50	50	-
Western Area	Pb in deer	100	-	-	100	-	-
	Pb in wild boar	83	-	17	83	17	-
	Cd in deer	100	-	-	14	79	7
	Cd in wild boar	100	-	-	50	50	-

The concentrations of cadmium and lead found in our survey were greater than those described by other authors in domestic slaughtered species (Cibulka et al. 1989;

Korsrud et al. 1985; Kramer et al. 1983; Salisbury et al. 1991) and these differences were more pronounced for cadmium than for lead. In Holland, Wolkers et al. (1994) found the cadmium and lead concentrations in the organs of deer and wild boar to be higher than in farm animals of the same age, especially in the kidneys.

In our survey, lead and cadmium levels in organs were similar, although slightly lower than those found in game animals in Poland (Falandysz 1994; Swiergosz et al. 1993) Oklahoma (Kocan et al. 1980) Quebec (Crête et al. 1987) and New Jersey (Stansley et al. 1991). In order to preserve the health of wild and domestic animals, we conclude that the lead and cadmium contamination recorded in the Sierra Morena Mountains (Southern Spain) should be further investigated. We also consider that there should be a more rigorous control of metal contamination of organs destined for human consumption.

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