#### ОРИГИНАЛЬНАЯ СТАТЬЯ

# TRACE ELEMENTS (Zn, Cu, Cd) IN SHEEP LIVING IN SEWAGE FARMS OF MARRAKESH CITY

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ABSTRACT. Toxic and essential trace elements (Zn, Cu, Cd) were measured in muscle, bone, liver and kidney of sheep grazing on the sewage farm of Marrakech City, Morocco. These animals were found to be seriously contaminated by metals, especially the cadmium, and the levels were higher in liver and kidney which are the specific target organs for metal bioaccumulation. The high cadmium content seemed to contribute to a reduction in zinc and copper levels. The arithmetic mean concentrations of zinc, copper and cadmium were, respectively, 120, 98.7 and 3.6 mg/kg (dry weight) in the liver; 84.2, 25.8 and 7.4 mg/kg (dry weight) in the kidney.

KEYWORDS: trace elements, sheep, kidney, liver, contamination, metal bioaccumulation.

#### INTRODUCTION

The toxic effects of trace elements released into the environment can be unexpected in the present while it could cause some serious damages in the long term.

The metallic compounds remain indefinitely even if they undergo the nature cycle in the living matter, and along with the amounts newly extracted come adding to their stock in the environment.

These metals deserve a special attention because of their permanence in the living organisms and the concentration phenomena which might happen in some food chains, leading eventually to the humans (Damek-Poprawa, Sawicka-Kapusta, 2003).

The soil is a biochemical reactor that can undoubtedly play a role as an active filter. Its contamination has therefore a dangerous effect in the short and long term on the quality of water and plants destined for human and animal consumption (Pommery et al., 1988).

Cadmium levels observed in animals cover a wide range depending on the species, tissues, nutrition, and other factors.

Transfer studies in pigs, cattle, sheep and other domestic animals have shown that there is apparently a direct correlation between the cadmium content in food and tissues (Hidiroglou, Proulx, 1988; Loska et al., 2005).

Cadmium poisoning can cause bone malformations in mammals including humans (Gale, Ferm, 1973), tubular nephropathy resulting in proteinuria

(Nicholson et al., 1983; Falck et al., 1983; Friberg, 1984), a modification of the blood vessels properties (Nejmeddine et al., 1988).

Vallee and Ulmer (1972) have shown that cadmium can substitute zinc in the active sites of carboxypeptidase A in bovine pancreas, which gives it a significant hydrolysis power towards the esters.

# **MATERIALS AND METHODS**

**Sampling.** 20 sheep aged between 10 and 12 months, reared on sewage farms, were chosen from the study area and 10 from the control area. They were classified under a Moroccan breed called "Sardi" and their grazing method was natural. These animals were chosen in order to determine the degree of trace elements contamination and the risk related to their human consumption.

The liver, the kidney, the ribs (bones and muscles) of the sheep were analyzed. Five samples of each organ were taken and prepared for mineralization.

Mineralization and chemical analysis. The quantification of trace elements transfer and bioaccumulation processes are based on quantitative determination of the contaminants in the environment and in the organisms. For the analysis, samples should be mineralized.

The metals present in animal tissues are either linked to the organic matter (proteins, lipids, etc.) or inorganic complexes. Thus, the mineralization frees these metals and put them in an ionic form.

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The mineralization consisted in putting 100 mg aliquot of a dried sample powder in 2 ml of nitric acid. The mineralization took place in a 25 ml beaker covered with a watch glass and placed on a hot plate at 200°C. Four hours later, the mineralized sample was recovered in a volumetric flask and made up to 25 ml with double distilled water.

Once mineralized, the samples were analyzed by atomic absorption spectrophotometry (Varian 475-AA apparatus) in flame (air-acetylene) for the zinc and copper, or in graphite furnace for the cadmium. Due to equipment availability, the study focused only on these three metals; further work will consider all metals.

#### **RESULTS**

All data on the content of chemical elements in organs and tissues herein are reported for dried samples (dry weight concentrations).

Sheep of the sewage farms. Statistical analysis (Newman-Keuls test at 5% error) showed a significant variation in the metal contents of sheep's organs, depending on the metal analyzed.

**Zinc levels.** The highest levels of Zinc were observed in the liver (Fig. 1).

A significant difference was observed between the levels in the organs analyzed (Fig. 1), with 120 mg/kg in the liver and 50 mg/kg in the bone.

Copper levels. The highest copper concentrations were observed in the liver (Fig. 1). Average copper content for this organ was 98.7 mg/kg. A clear and significant difference was noted between these levels and those of other organs. In the kidney,

the average content was 25.8 mg/kg. As for the muscle, we note that the average copper content was 22 mg/kg (Table 1).

Regarding the bone, the mean levels of copper did not exceed 24 mg/kg (Table 1).

**Cadmium levels.** The highest cadmium content was noted in the kidneys (Fig. 1).

Its levels in the kidneys and liver were respectively 7.4 and 3.6 mg/kg. Statistical analysis showed a significant difference between high levels of cadmium in the kidneys and liver on the one hand, and those of other tissues on the other hand (Table 1).

**Sheep's control samples.** In the controls, the metal contents of the organs were lower than those of the individuals living in the sewage farms. For Zn, Cu and Cd respectively they were 32.5, 10 and 1.3 mg/kg in the kidney; 48, 44 and 1.4 mg/kg in the liver (Fig. 1).

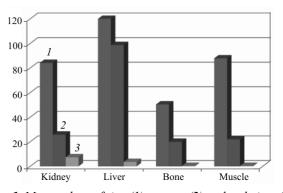


Fig. 1. Mean values of zinc (1), copper (2) and cadmium (3) content in the organs of sheep living in sewage farms (mg/kg dry weight)

**Table 1.** Levels of trace elements in different studied organs in sheep (mg/kg dry weight)

Element		Kidney		Liver		Bone		Muscle	
		S	SC	S	SC	S	SC	S	SC
Zn	M	84.2	32.5	120	47.9	50.4	41.4	87.9	38.6
	SD	13.4	3.6	18.6	7.2	9	4.9	21	3.7
	CV	15.9	11	15.4	15	17.8	11.8	23.9	9.58
	Min	63	28.9	104	40.7	40	36.5	51	34.9
	Max	111	36.1	159	55.1	66	46.4	117	42.4
Cu	M	25.8	9.9	98.7	44.4	20	14.2	22	10.8
	SD	5.4	1.3	22.5	7.6	5.2	1.8	5.2	2.3
	CV	20.9	13.1	22.8	17.1	26	12.6	23.6	21.3
	Min	17.9	8.6	59	36.8	14	12.4	15.2	8.5
	Max	36	11.2	134	52.1	31.5	16.1	31.7	13.1
Cd	M	7.4	1.35	3.6	1.4	0.4	0.1	0.4	0.1
	SD	1.2	0.25	1.2	1.1	0.2	_	0.2	_
	CV	16.2	18.5	33.3	78	50	-	50	_
	Min	4.8	1.1	2.1	0.3	0.2	-	0.15	_
	Max	9.1	1.6	6.1	2.5	0.6	-	0.85	_

N o t e : M – mean value; SD – standard deviation, CV – coefficient of variation; Min – minimum content; Max – maximum level; S – sheep; SC – sheep control.

Element	Kidney	Liver	Bone	Muscle	
Zn	2,59	2,5	1,21	2,27	
Cu	2,6	2,22	1,4	2,03	
Cd	<b>Cd</b> 5,48		4	4	

Table 2. Pollution Factor (PF\*) in the studied organs of sheep living in sewage farms (dry weight)

N o t e : \* PF = Metal concentration in the studied kidney (or liver/bone/muscle) / Metal concentration in the control kidney (or liver/bone/muscle)

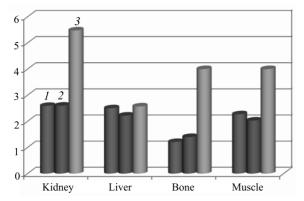


Fig. 2. Pollution Factor in the studied organs of sheep living in sewage farms (dry weight):

1 -zinc, 2 -copper, 3 -cadmium

### **DISCUSSION**

Comparison of metal concentrations in control sheep with those of sheep from the sewage farms revealed that these were very high. The values of pollution factor were significant and greater than 3, especially for cadmium (Table 2; Fig. 2).

Unlike those of zinc and copper, the values of CV (coefficient of variation) of cadmium levels are high for all organs analyzed (Table 1). This shows the heterogeneity of the cadmium contents and consequently the existence of individuals more contaminated by that metal in the group of the animals analyzed.

The comparison of these results with those of other authors showed that the zinc levels in the muscle, liver and kidney are higher than those cited by Leontopoulos et al. (2015), which range respectively from 10.40 to 39.01, 14.90 to 18.44, and 10.04 to 28.98 mg/kg (dry weight) in the pigs from the region of Thessaly.

As for copper, the concentration levels in the muscle, liver and kidney are also higher than those cited by Leontopoulos et al. (2015), which range respectively from 0.3 to 1.23, 2.71 to 5.94 and from 2.44 to 7.02 mg/kg (dry weight) in the pigs from the region of Thessaly.

When it comes to cadmium, the mean levels observed in the muscle, liver and kidney are higher

than those cited by Leontopoulos et al. (2015), which are <0.02 mg/kg (dry weight) in the pigs from the region of Thessaly.

The analysis performed on the medulla (8.6 mg/kg, dry weight) and the cortex (6.9 mg/kg, dry weight) of sheep's kidney from the sewage farms shows a high concentration of cadmium in the cortex which has already been reported by Mogilnicka (Mogilnicka, Piotrowski, 1977).

Our observations along with the bibliographic data confirm that the cadmium preferentially concentrates in the kidney, the copper in the liver and the zinc in the liver as well as in the kidney. This uneven distribution of trace elements in the organs of sheep has been discussed by several authors (Baxter et al., 1983; van der Veen, Vreman, 1986). It mainly depends on the organ function, its physiological properties, including the presence of proteins such as metalloproteins which are capable if fixing the metals.

It also depends on the rate of accumulation in plants which itself depends on the availability of the metal in the soil and their uptake by the plants; this assimilation is easier for the cadmium and zinc (Munshower, 1977; Morel et al., 1986), while the copper is the metal which is more difficult to assimilate (Meeus-Verdinne, 1988), this being due to its strong binding to the soil (Greffard et al., 1985).

#### CONCLUSION

The metal concentrations observed in the sheep kidney and liver samples are higher than those of the controls and those of the bibliographic data, especially for cadmium with the average values are respectively about 7 and 4 mg/kg (dry weight).

Anomalous metal concentrations observed in this analyzed species can affect the entire food chain of which man is the last link and therefore induce a danger to the public health.

Indeed, the experts (FAO/WHO) believe that the human tolerable weekly intake for cadmium should not exceed 0.5 mg (Cumont, 1974).

When the quantities absorbed with food exceed these thresholds, the amounts accumulated can reach some toxic levels and determine damages in the target organs, most often irreversible.

If our results do not provide a definitive answer as to the extent of ecotoxicological consequences of the contamination of the sewage farms population, they however, highlight the non-negligible risk that threatens the species exposed to this contamination.

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# МИКРОЭЛЕМЕНТЫ (Zn, Cu, Cd) У ОВЕЦ, ВЫПАСАЕМЫХ НА ПОЛЯХ ОРОШЕНИЯ СТОЧНЫМИ ВОДАМИ ГОРОДА МАРРАКЕШ

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РЕЗЮМЕ. Содержание токсичных и эссенциальных микроэлементов (Zn, Cu, Cd) было измерено в мышцах, костях, печени и почках овец, выпасаемых на полях орошения сточными водами города Марракеш (Марокко). Было исследовано 20 особей основной группы и 10 особей контрольной группы; овцы имели возраст от 10 до 12 месяцев и принадлежали к местной породе «сарди», выращиваемой на естественном выпасе. Содержание микроэлементов в образцах определялось методом атомно-абсорбционной спектрометрии после минерализации методом мокрого озоления с азотной кислотой. Статистическая обработка данных проводилась с применением критерия Ньюмана–Кейлса. Обнаружено, что биологические образцы исследованных животных значительно загрязнены металлами, особенно кадмием, при этом наибольшее содержание выявлено в печени и почках, представляющих собой основные органы биоаккумуляции металлов. По-видимому, высокое содержание кадмия способствует снижению уровня цинка и меди. Средние концентрации цинка, меди и кадмия в высушенных образцах у животных основной группы составили, соответственно, в печени: 120, 98,7 и 3,6 мг/кг; в почках: 84,2, 25,8 и 7,4 мг/кг. У контрольных животных эти показатели были, соответственно, в печени: 48, 44 и 1,4 мг/кг; в почках: 32,5, 10 и 1,3 мг/кг.

КЛЮЧЕВЫЕ СЛОВА: микроэлементы, овцы, почки, печень, загрязнение, биологическое накопление металлов.