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Health risk assessment of consuming European hare tissues used as an environmental bioindicator*

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Abstract

Environmental pollution with heavy metals is one of the most significant threats to human health. Heavy metals enter the human body through the skin, respiratory tract, and consumption of plant and animal products. The transfer of these elements to subsequent links in the food chain, and consequently into the human body, is limited by the action of biological barriers. However, it should be emphasized that excessive element concentrations reduce the effectiveness of these barriers, which poses a risk of negative impact on the environment and, above all, on human health. The safety and health quality of food is determined, among other things, by the content of undesirable elements such as cadmium, lead, mercury, and arsenic. The health safety of raw materials obtained from game animals is an important element in consumer health protection, especially in the context of environmental degradation. The aim of this study was to assess the health risk (non-carcinogenic and carcinogenic) resulting from the presence of heavy metals in the tissues of the European hare (*Lepus europaeus*) in the context of public health, and to verify the suitability of this species as a bioindicator. The kidneys were selected for analysis as a critical organ for xenobiotic accumulation. Cadmium, lead, and mercury concentrations were analyzed in hares from the western part of the Lublin Upland. Assuming a dietary intake of 0.08 kg person year, the hazard indices (HI) were shown to be low (<0.03) and the carcinogenic risk of lead was negligible (<10-9), indicating no toxic threat to the consumer. At the same time, the statistically significant accumulation of cadmium and mercury with age confirms the hare's role as a sensitive bioindicator of environmental pressure.

Keywords: health risk assessment, heavy metals, kidneys, bioindicator, European hare, food safety

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INTRODUCTION

Heavy metals are a natural and integral element of the natural environment (Gall et al. 2015, Okereafor et al. 2020, Gworek et al. 2024). Human activity leads to their excessive release into the environment, which subsequently results in their incorporation into the food chain. A group of particularly harmful heavy metals that adversely affect human health includes cadmium (Cd), lead (Pb), mercury (Hg), and arsenic (As) – Ochwanowska et al. (2019), Chmielewski et al. (2020), Okereafor et al. (2020), Dietrich et al. (2022). These elements have no known benefits for the human organism; conversely, they can be the cause of numerous negative health effects (Ohiagu et al. 2022). The threat posed by heavy metals results directly from their transfer within the trophic chain: soil – plant – animal – human, and the potential for accumulation in the final link, i.e., the human organism (Enock et al. 2025).

The level of accumulation and the content of heavy metals in a body acquired via food are influenced by one's diet, ingested dose, elimination rate, and routes of absorption. The group of particularly toxic metals includes Cd, Pb, Hg, and As, which can affect the human body through various exposure routes: the respiratory system, the digestive system, and the skin. When absorbed in excessive amounts, they can lead to disorders in the functioning of the human body. Heavy metals in human organisms primarily induce changes in protein synthesis and disturbances in ATP production, which may result in serious pathological changes, including carcinogenic ones. The toxic effect depends largely on the amount of the element entering the body. It should be emphasized that the degree of toxicity also depends on the chemical form in which the metals occur, their solubility in body fluids and lipids, the duration of exposure, and the resistance of the given organism. The negative effects of heavy metals on the human body have been extensively described in the literature (Ochwanowska et al. 2019, Okereafor et al. 2020, Dietrich et al. 2022, Ohiagu et al. 2022, Budi et al. 2024, Jomova et al. 2025).

National legal regulations regarding food safety and nutrition (UOBŻIŻ 2026) define food safety as the aggregate of conditions that must be met, concerning in particular used additives and flavorings, levels of contaminants, pesticide residues, food irradiation conditions, organoleptic characteristics, and the measures that must be taken at all stages of food production or handling in order to protect human health and life. From the consumer's point of view, the health safety of food is the most important quality feature; therefore, food law (both global and European) regulates this issue in detail to ensure that the food consumed meets safety standards (Baba, Esfandiari 2023). However, the concept of food health safety should be viewed in the context of public health, nutrition, food and feed quality, as well as the economic security of the consumer rather than be limited to the safety of the product itself (Walls et al. 2019).

From the consumer's perspective, problems resulting from environmental degradation necessitate determination of heavy metal contamination in meat of free-living animals intended for consumption. Analysis of heavy metal residues, including Cd, Pb, Hg, and As, in internal organs (the liver and kidneys) which perform filtering and detoxifying functions in the animal's body is particularly valuable. The level of meat contamination depends, among other factors, on the species and age of the animals, as well as the degree of environmental degradation (Małová et al. 2019, Rudy et al. 2019).

Among free-living animals, those most exposed to environmental pollutants include grazing species like roe deer, red deer, moose, European bison, and hares. Animals living in a natural ecosystem typically accumulate significantly larger amounts of heavy metals than farm animals, making them potential bioindicators of the state of the environment (Gall et al. 2015, Sevillano Morales et al. 2018, Małová et al. 2019, Rudy et al. 2019, Nkosi et al. 2021, Draghi et al. 2023, Gulin et al. 2023).

Hares are a valuable source of high-quality game meat. Hare meat is appreciated for its tenderness and juiciness, as well as low fat content and the highest protein content compared to the meat of other game and domestic animal species (Mertin et al. 2012, Razmaite, Šiukščius 2023). The availability of game is a significant factor influencing interest among Polish meat consumers. Statistics Poland (GUS) does not provide statistical data on the meat consumption of game species. As indicated by literature data, the consumption of game meat per person per year in Poland is 0.08 kg (Mesinger et al. 2023). In contrast, in such European countries as Bulgaria, Portugal, the United Kingdom, Spain, the Czech Republic, Croatia, Germany, Norway, and Sweden, game meat consumption in recent years has ranged from 0.2 to 1.8 kg/person/year (Sevillano Morales et al. 2018, Czarniecka-Skubina et al. 2022, Mesinger et al. 2023). High consumption of game is recorded in France (5.7 kg/person/year) and Italy (3.8 kg/person/year) (Mesinger et al. 2023). It is estimated that only 2-4% of the population in Europe regularly consume this type of meat. Higher consumption was observed among hunters and their families (Andreotti et al. 2016, Sevillano Morales et al. 2018).

In 2023, Poland exported nearly 30,000 tons of game, mostly to Vietnam (6,465 tons), Germany (4,310 tons), the Ivory Coast (3,528 tons), and the Netherlands (2,201 tons).

The working hypothesis in this study was that consumption of kidneys of wild animals like hares, despite their environmental exposure and tendency to accumulate elements, does not pose a significant heavy-metal toxicological risk at average consumption levels in Poland, while their elemental accumulation supports the role of the hare as a bioindicator. The study aimed to estimate the health risk (non-carcinogenic and carcinogenic) associated with the presence of Cd, Pb, and Hg in hare kidneys, and to assess the degree of contamination in the context of environmental bioindication.

MATERIAL AND METHODS

This study consisted of a systematic review of xenobiotic metal contamination, primarily in the liver and kidneys of hares. The review was conducted using the Scopus database search engine and following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) methodology. Scopus was selected for review as the world's largest database of abstracts and citations, which encompasses a broader spectrum of journals than many alternative sources. The PRISMA methodology ensures a thorough and systematic review, minimizing potential bias. The search included keywords related to the topic of the paper: heavy metals, hare meat contamination, and health risk assessment terms. The search was limited to peer-reviewed articles published in English. The full Scopus query was as follows: (TITLE (hare meat) AND TITLE (Xenobiotic metals in hare liver and kidneys) OR ("heavy metals (and toxic)" OR "potentially toxic elements" OR "heavy metals" OR "toxic elements" OR "trace metals" OR "trace elements" OR "metal content") AND TITLE-ABS-KEY ("health risk" OR "hazard indicator" OR "health hazard"). In the first comprehensive search, a total of 63 articles were retrieved. Table 1 presents the concentrations of xenobiotic metals ($\mu\text{g g}^{-1}$ wet weight) in the tissues of hare species by selected locations.

It is important to remember that anthropogenic environmental pollution affects the quality of plants that constitute hare food. The degree of contamination of the vegetation in the hare's habitat influences the contamination of its meat, therefore the presented assessment results are not representative of the entire population. The health risk assessment applies to meat obtained from a given habitat.

Due to restricted access to official veterinary inspection data concerning laboratory test results of game meat, including heavy metal contamination collected in the CELAB system (Central Database), which is available only to registered users from approved laboratories and inspectorate employees (GIW 2026), available literature data were used for the health risk assessment (Fischer et al. 2025).

The health risk assessment was based on an estimated annual game meat consumption in Poland of 0.08 kg (Mesinger et al. 2023) and results of toxicological assays of heavy metals (Cd, Pb, Hg) in the kidneys of the European hare (*Lepus europaeus*) from the western part of the Lublin Upland, as presented in Table 2 (Fischer et al. 2025).

The standard USEPA methodology was applied for the risk assessment (Vesković, Onjia 2025). The Estimated Daily Intake (EDI) was calculated using the following formula:

$$\text{EDI} = C \times \text{IR} / \text{BW}$$

where: C – the mean metal concentration ($\mu\text{g g}^{-1}$),

IR – the daily ingestion rate (0.08 kg year⁻¹),

BW – the body weight (70 kg).

Table 1

Concentrations of xenobiotic metals ($\mu\text{g g}^{-1}$ wet weight) in hare kidneys in selected locations in 2010-2022

Species	Location	Liver x (min-max)	Reference
As concentrations			
<i>L. americanus</i>	Canada	0.76 (0.19–2.30)	Amuno et al. 2017
<i>L. europaeus</i>	Czech Republic	0.01893 (0.0046–0.097)	Bukovjan et al. 2016
<i>L. europaeus</i>	Ukraine	0.044 (0.032–0.064)	Pilarczyk et al. 2020
Cd concentrations			
<i>L. americanus</i>	Canada	0.49 (0.096–1.092)	Amuno et al. 2017
<i>L. arcticus</i>	Canada	(0.018–3.44)	Amuno et al. 2016
<i>L. europaeus</i>	Croatia	1.054 (0.449–2.453)	Linšak et al. 2014
<i>L. europaeus</i>	Poland	1.21 (0.08–2.75)	Halecki et al. 2017
<i>L. europaeus</i>	Poland	5.7	Wajdzik et al. 2017
<i>L. europaeus</i>	Serbia	0.17 (0.01–0.85)	Petrović 2013
<i>L. europaeus</i>	Serbia	0.208 (0.1–1.414)	Beuković et al. 2022
<i>L. europaeus</i>	Turkey	0.83	Demirbas, Enduran 2017
<i>L. europaeus</i>	Ukraine	0.089 (0.053–0.191)	Pilarczyk et al. 2020
Hg concentrations			
<i>L. americanus</i>	Canada	0.0129 (0.0025–0.0362)	Intrinsik 2015
<i>L. europaeus</i>	Croatia	0.074	Špirić et al. 2012
<i>L. europaeus</i>	Croatia	(0.058–0.189)	Linšak et al. 2013
<i>L. europaeus</i>	Serbia	0.02 (0.006–0.068)	Petrović 2013
<i>L. europaeus</i>	Turkey	0.06	Demirbas, Enduran 2017
Pb concentrations			
<i>L. americanus</i>	Canada	0.0104 (0.0191–0.281)	Intrinsik 2015
<i>L. capensis</i>	Pakistan	28.67 (1.00–116.4)	Ahmed et al. 2016
<i>L. europaeus</i>	Poland	0.83 (0.35–1.41)	Halecki et al. 2017
<i>L. europaeus</i>	Serbia	0.22 (0.06–1.72)	Petrović 2013
<i>L. europaeus</i>	Turkey	2.19	Demirbas, Enduran 2017
<i>L. europaeus</i>	Ukraine	0.674 (0.625–0.780)	Pilarczyk et al. 2020

Non-carcinogenic risk was expressed using the Hazard Quotient (HQ) and Hazard Index (HI):

$$\text{HQ} = \text{EDI/RfD},$$

$$\text{HI} = \sum \text{HQ} = \text{HQ}_{\text{Cd}} + \text{HQ}_{\text{Pb}} + \text{HQ}_{\text{Hg}},$$

where: RfD represents the Reference Dose. The following values were adopted for calculations: $0.001 \mu\text{g g}^{-1} \text{ day}^{-1}$ for Cd, $0.0035 \text{ mg kg}^{-1} \text{ day}^{-1}$ for Pb, and $0.0003 \mu\text{g g}^{-1} \text{ day}^{-1}$ for Hg.

Carcinogenic Risk (CR) for lead was estimated as the product of EDI x CSF, assuming a Cancer Slope Factor (CSF) of 0.0085 ($\mu\text{g g}^{-1}\text{ day}^{-1}$).

RESULTS AND DISCUSSION

Xenobiotic metals are mainly accumulated in the liver and kidneys because these organs play a significant role in detoxification within the organism. Data analysis (Table 2) confirms this dependency, showing particularly

Table 2
Content of heavy metals – cadmium, lead, and mercury in hare kidneys expressed as wet weight in $\mu\text{g g}^{-1}$ ($n=49$)

Value ($\mu\text{g g}^{-1}$)		Mean	Minimum	Maximum	p (for $p \leq 0,05$)
Cadmium	young	3.5542	0.253	11.070	0.01
	old	7.5058	0.448	27.757	
	males	6.1320	0.448	27.757	0.66
	females	7.0700	0.253	26.901	
Lead	young	0.0723	0.018	0.135	0.49
	old	0.0800	0.036	0.178	
	males	0.0727	0.040	0.154	0.22
	females	0.0876	0.018	0.178	
Mercury	young	0.0041	0.001	0.008	0.01
	old	0.0065	0.002	0.020	
	males	0.0061	0.002	0.020	0.46
	females	0.0053	0.001	0.015	

high cadmium concentrations that significantly increase with the age of the animals ($p=0.01$). The mean Cd content in old individuals ($7.5058 \mu\text{g g}^{-1}$) was more than double that in young ones ($3.5542 \mu\text{g g}^{-1}$), indicating strong bioaccumulation of this element in kidneys during the individual's life and confirming the hare's usefulness as a bioindicator of long-term environmental exposure. A similar, statistically significant upward trend was observed for mercury ($p=0.01$), whereas lead concentrations did not differ significantly between age groups ($p=0.49$).

A key element of the study was the health risk assessment. First, the Estimated Daily Intake (EDI) was calculated for each metal based on the assumed consumption of hare kidneys ($0.08 \text{ kg}^{-1} \text{ year}^{-1}$). Additionally, the Carcinogenic Risk (CR) for lead was estimated. These values are presented in Table 3. The highest EDI values were recorded for cadmium, ranging from 1.11×10^{-5} to $2.35 \times 10^{-5} \mu\text{g g}^{-1} \text{ b.w. day}^{-1}$, which results from the specificity of the kidney as a critical organ for this metal.

Table 3

EDI ($\mu\text{g g}^{-1}$ b.w. day $^{-1}$) and health risk indices (HQ, HI, CR) resulting from the consumption of hare kidneys

Group	EDI (Cd)	EDI (Pb)	EDI (Hg)	CR Pb*
Young	1.11×10^{-5}	2.26×10^{-7}	1.28×10^{-8}	1.92×10^{-9}
Old	2.35×10^{-5}	2.50×10^{-7}	2.04×10^{-8}	2.13×10^{-9}
Males	1.92×10^{-5}	2.28×10^{-7}	1.91×10^{-8}	1.93×10^{-9}
Females	2.21×10^{-5}	2.74×10^{-7}	1.66×10^{-8}	2.33×10^{-9}

* values in $\mu\text{g g}^{-1}$ b.w. day $^{-1}$, E-05 denotes $\times 10^{-5}$, CR Pb – Carcinogenic Risk calculated for lead

Regarding carcinogenic risk, the calculated CR values for lead (Table 4) ranged from 1.9×10^{-9} to 2.3×10^{-9} . According to USEPA guidelines, the acceptable risk level falls within the range of 10^{-6} to 10^{-4} . The results are several orders of magnitude lower than the lower limit of this range, allowing the carcinogenic risk to be considered negligible.

Based on the EDI and Reference Doses (RfD), the non-carcinogenic risk was evaluated using the Hazard Quotient (HQ) and Hazard Index (HI). The detailed results with safety interpretation are shown in Table 4.

Table 4

Non-carcinogenic risk assessment (HQ and HI) with safety interpretation

Group	HQ (Cd)	HQ (Pb)	HQ (Hg)	Σ HI	Interpretation*
Young	0.0111	0.00006	0.00004	0.0112	safe
Old	0.0235	0.00007	0.00007	0.0236	safe
Males	0.0192	0.00007	0.00006	0.0193	safe
Females	0.0221	0.00008	0.00006	0.0223	safe

* Interpretation criteria: HQ < 1 or HI < 1 – negligible risk (safe); HQ > 1 – potential health risk exists

As shown in Table 4, the HQ for all analyzed elements were significantly lower than the threshold value of 1. Cadmium had the prevalent contribution to the total risk, with HQ reaching a maximum of 0.0235 in the group of old animals. This implies that the intake of cadmium constituted only about 2.3% of the safe reference dose. HI representing the cumulative risk from the mixture of metals, ranged from 0.0112 to 0.0236. Since HI < 1, the combined exposure to these metals through the consumption of hare kidneys does not pose a non-carcinogenic toxicological threat to the average consumer. However, it is crucial to remember that these metals can inhibit the activity of the liver and kidneys of mammals, and their content depends not only on the season but also on the age of the harvested individuals.

The research by Gulin et al. shows that the hare is a good bioindicator of environmental contamination with xenobiotic metals. Their concentrations

in the liver and kidneys provide the best results reflecting pollution levels. Hare meat is safe for human consumption, except in countries with high levels of pollution. However, eating hare viscera is not recommended (Gulin et al. 2023).

The study conducted by Pilarczyk et al. revealed the presence of relatively high Pb content in wild hares living in western Ukraine. The permissible concentration of this element was exceeded in all meat and offal samples. For Cd, the permissible values were observed in the muscles of gray hares (12% of samples). The highest HQ values were observed for Cd in the ingested liver and meat of the studied animals. However, in none of the cases were HQ or HI values greater than 1, indicating a low probability of adverse health effects associated with hare consumption (Pilarczyk et al. 2020).

Study limitations

Indicators of consumption levels of the analyzed toxic elements should not be treated as final due to existing knowledge gaps, including the need for broader studies on meat contamination and consumption volumes. The results are representative of area-specific, low consumption (national average 0.08 kg year⁻¹) pattern, and may not reflect the risk for groups with high game consumption (e.g., hunters and their families).

CONCLUSIONS

The calculated health risk indices (HQ, HI) for all study groups were significantly lower than one (HI < 0.03). This indicates that at the average consumption level of hare tissues (specifically kidneys) in Poland (0.08 kg year⁻¹), there is no risk of non-carcinogenic negative health effects associated with heavy metals. Furthermore, the estimated carcinogenic risk for lead was negligible.

The analysis of toxic element content demonstrated a statistically significant dependence of cadmium and mercury concentrations on animal age ($p=0.01$). This confirms the role of the European hare as a sensitive bioindicator that effectively accumulates environmental pollutants in parenchymal organs over time.

Continuous improvement of food safety systems and increasing the efficiency of control institutions are necessary to meet dynamically changing threats and consumer expectations. Broadening knowledge regarding trace element contamination in food is particularly important for public health due to their potential harmfulness. Indicators of consumption levels and toxicological reference values for potentially toxic elements (such as BMDL or PTWI) should not be treated as final due to existing knowledge gaps and the need for ongoing research.

Author contributions

Both authors have read and agreed to the published version of the manuscript.

Conflicts of interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

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