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SCREENING THE OCCURRENCE OF HEAVY METALS IN BEEF LIVERS AS AFFECTED BY GEOGRAPHICAL LOCATION AND FARMING SYSTEM: THE EU-FUNDED INTAQT PROJECT

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INTRODUCTION

Heavy metals, such as lead (Pb), cadmium (Cd), mercury (Hg), and arsenic (As), can accumulate in the tissues of animals due to environmental pollution, contaminated feed, or exposure to industrial waste [1]. While some heavy metals, like iron (Fe), zinc (Zn), and copper (Cu), are essential for regulating physiological functions in small amounts (e.g., enzyme activity, oxygen transport, and immune response), an excess of these metals, or toxic metals like Pb, Cd, Hg, and As, can cause severe damage to biological systems. The liver, being a primary organ for detoxification, often stores higher concentrations of these metals compared to other tissues, making it particularly important to monitor [2]. Meat and liver from animals are staple food sources for many consumers, and elevated levels of heavy metals in these products can pose serious health risks, including neurological damage, kidney dysfunction, and cancer. Chronic exposure to even low levels of toxic heavy metals can lead to long-term health issues [3]. Therefore, maintaining low levels of harmful metals in meat and liver is crucial for consumer safety. Regulatory bodies, such as the World Health Organization (WHO) and the European Food Safety Authority (EFSA), set maximum permissible limits for these metals in food products to ensure that they do not exceed levels that could harm human health. As global concerns about food safety grow, there is an increasing emphasis on testing and controlling heavy metal contamination in food products, particularly for vulnerable populations such as children and pregnant women. Ensuring the safety of meat and liver for human consumption is essential to protecting public health and maintaining consumer trust in food safety standards. The INTAQT Project is a research initiative co-funded by the European Union's Horizon 2020 program. It aims to unveil the impact of farming systems of meat and milk quality and safety by using innovative methodological approaches. This specific study aimed to address growing concerns over food safety, as consumers are increasingly demanding more reliable and efficient testing methods to ensure that food products are free from harmful substances.

I. MATERIALS AND METHODS

Cattle all over Europe were raised and fed under different feeding regimes and at different locations (which could affect weather conditions, availability of pasture, etc.). **Table 1** summarizes the participating countries, the group of farms (consisting of individual farms) and the feeding regimes. Each farm raised a certain number of cattle from which, after slaughter, the liver was collected. This procedure was done twice, once in summer and once in winter. 300-350 mg of each liver were pooled sorted by season. A 400 mg sample from each pool were placed in individual digestion vessel and, after adding 3mL of a 3:1 mixture of nitric acid (69%) and oxygen peroxide, respectively, the samples were heated and digested in the microwave oven. After digestion, solutions were diluted and brought to 25ml of total volume with MiliQ water and analyzed on an ICP/MS.

Table 1. Farm groups in different European countries, categorized by their respective feeding regimes.

Country	Farm group	Feeding regime
France	B-FR-1	Semi-extensive
	B-FR-2	Semi-intensive
Spain	B-ES-1	Intensive
Switzerland	B-CH-1	Extensive
	B-CH-2	Extensive
	B-CH-3	Semi-extensive
	B-CH-4	Semi-intensive
Italy	B-IT-1	Intensive
	B-IT-2	Intensive
United Kingdom	B-UK-1	Semi-extensive
	B-UK-2	Semi-extensive

A total of six heavy metals were analyzed, namely, Chromium (Cr), Nickel (Ni), Arsenic (As), Cadmium (Cd), Mercury (Hg) and Lead (Pb). All contaminants were measured in µg/Kg. Statistical analyses were performed to test for significant differences in heavy metals in livers from different farming systems and locations (country). Statistics were

carried out using Kruskal-Wallis non-parametric analysis after Shapiro-Wilk confirmation that the samples did not follow a normal distribution. Therefore, median and not mean was taken into consideration to make these determinations.

II. RESULTS AND DISCUSSION

*Table 2. Median values of heavy metals in liver samples from various farm groups, including Switzerland (CH-1 to CH-4), Spain (ES-1), France (FR-1 to FR-2), Italy (IT-1 to IT-2), and the United Kingdom (UK-1 to UK-2). Values are reported in µg/Kg, with '<LOQ' indicating concentrations below the limit of quantification. Statistical significance (P-value) is indicated, with *** representing highly significant differences, ** for moderate significance, • for hints of significance (0.10<pvalue<0.05) and ns for no significant differences. LOQ:25.0 µg/Kg for Hg, 6.0 for the rest.*

Farm ID	CH-1	CH-2	CH-3	CH-4	ES-1	FR-1	FR-2	IT-1	IT-2	UK-1	UK-2	*P*-value
As (µg/Kg)	6.41	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	•
Cd (µg/Kg)	53.87b	27.12d	36.69c	21.04d	20.09d	88.08a	63.41b	19.26d	22.61d	40.41c	37.94c	***
Cr (µg/Kg)	8.56	7.11	11.60	<LOQ	13.11	<LOQ	<LOQ	10.03	<LOQ	17.61	<LOQ	**
Hg (µg/Kg)	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	ns
Ni (µg/Kg)	<LOQ	<LOQ	<LOQ	<LOQ	<LOQ	4.58	<LOQ	<LOQ	<LOQ	10.47	13.35	***
Pb (µg/Kg)	12.09b	24.63a	28.63a	4.87c	12.58b	8.74c	4.73c	6.98c	9.36c	9.54c	21.20a	***

Table 2 shows heavy metal concentrations (As, Cd, Cr, Hg, Ni, Pb) in liver samples from different farm groups across Europe, with varying feeding regimes. Commission Regulation (EU) 2023/915 of 25 April 2023 [4] on maximum levels for certain contaminants in food only provided few liver heavy metal thresholds of the ones we analyzed in this research. The permissible limits for lead and cadmium in liver and meat are set at 200 µg/kg and 500 µg/kg, respectively. In this study, these thresholds were not approached, ensuring that the quality of both liver and meat was maintained with respect to these heavy metal contaminants. Currently, there are no specific European regulations for other heavy metals in liver or meat. However, the concentrations observed were below the limits of quantification, further confirming the safety and quality of the products. The lead results were remarkably lower than other results from samples collected in Iran [2] (273 µg/Kg) or Brazil [5] (231 µg/Kg) suggesting probably a lower environmental contamination but similar to the ones find in Korea [6] (26 µg/Kg). Cadmium and lead concentrations were higher in livers from animals in extensive systems (vs. those from intensive ones), irrespective of the country, which emphasizes the impact of environmental pollution on exposure and heavy metals accretion in animal tissues.

III. CONCLUSION

The permissible limits set by the European Commission for lead and cadmium were not exceeded, assuring the safety and quality of liver and meat. Despite the lack of specific European regulations for other heavy metals in these products, the concentrations measured were well below levels of concern for human health. Additionally, the comparison with studies from other countries highlights that European products exhibit lower contamination levels, further supporting the effectiveness of current food safety measures in Europe.

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