JMBFS
Journal of Microbiology, Biotechnology and Food Sciences
International peer-reviewed scientific online journal
Published by Faculty of Biotechnology and Food Sciences

CONCENTRATIONS OF CHEMICAL ELEMENTS IN SHEEP MILK FROM THREE FARMS IN SLOVAKIA FROM AREAS WITH DIFFERENT ENVIRONMENTAL BURDEN

Simona Almášiová[1], Róbert Toman[1], Martina Pšenková[1], Vladimír Tančin[1], Ivona Jančo[2]*

Address(es): prof. Ing. Róbert Toman, Dr.
[1] Slovak University of Agriculture in Nitra, Faculty of Agrobiology and Food Resources, Institute of Animal Husbandry, Trieda Andreja Hlinku 2, 949 76 Nitra, Slovak Republic.
[2] Slovak University of Agriculture in Nitra, AgroBioTech Research Center, Trieda Andreja Hlinku 2, 949 76 Nitra, Slovak Republic.

*Corresponding author: robert.toman@uniag.sk

https://doi.org/10.55251/jmbfs.9971

ARTICLE INFO

Received 12. 3. 2023
Revised 4. 6. 2023
Accepted 5. 6. 2023
Published 1. 10. 2023

Regular article

OPEN ACCESS

ABSTRACT

Milk is considered as a basic food, highly nutritious, and recommended for everyday consumption during the whole human life. However, due to environmental pollution, milk is not just a source of essential minerals, but potentially can contain heavy metals and trace elements as well which can threaten human health. The study aimed to determine the occurrence of 22 elements: Ag, Al, As, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Li, Mg, Mn, Mo, Na, Ni, Pb, Sb, Se, Sr, Zn in raw sheep milk collected from areas with different environmental burden in Slovakia. Samples were collected from three farms, one from Orava region (potentially undisturbed area) and two from Spiš region (slightly and heavily disturbed area) during the period of lactation. Analysis of the elements was performed using an inductively coupled plasma-optical emission spectrometry with axial plasma configuration and with auto-sampler SPS-3. Statistical significant differences ($P < 0.05$) were found in concentrations of following elements: Al, As, Ba, Ca, K, Li, Sr, and Zn. Calcium, potassium, sodium, and magnesium were most represented in samples from all monitored farms. The highest element variation was found in sheep milk samples from the heavily disturbed area. This fact confirmed the quality of monitored areas according to environmental regionalization by The Ministry of the Slovak Republic and the Slovak Environmental Agency. Heavy metals found in the heavily disturbed area do not represent any concerns according to European legislation, except lead, for which the mean concentration represents 0.02 mg/kg. This value was set by EC Regulation no. 1881/2006 and Codex Alimentarius as the level of the permitted limit of concentration of Pb in milk.

Keywords: sheep milk, essential elements, heavy metals, environmental pollution, Slovakia

INTRODUCTION

Rich nutritional profile, good accessibility, and preferred taste by consumers are the main reasons why milk represents a significant role in human nutrition **(Saribal, 2020; Pompilio *et al*., 2021).** Milk and dairy products contain quality proteins, essential fatty acids, and micronutrients **(Licata *et al*., 2004; Kapila *et al*., 2013)** including several bioactive compounds that play a vital role in biochemical and physiological functions **(Boudebbouz *et al*., 2022).** Children require calcium, magnesium, and potassium for normal metabolism, growth, and development **(Oana *et al*., 2016; Zhou *et al*., 2017)** and milk and dairy products are excellent sources of these elements **(Gaucheron, 2011).** It is beneficial to human health to consume milk regularly **(Srbely *et al*., 2019)**. The intake of milk and dairy products contributes to building bones mass **(Fardelonne, 2019; Ratajczak *et al*., 2021),** positive effects on bone mineral density have been found, as well as reduced fracture risk in some populations (**Melse- Boonstra, 2020**). But sufficient consumption of milk contributes to maintaining muscle mass even in the elderly **(Hanach *et al*., 2019)**, prevents civilization diseases such as cardiovascular diseases (**Guo *et al*., 2017; Thorning *et al*., 2016),** metabolic syndrome **(Lee *et al*., 2018)**, obesity **(Lu *et al*., 2016; Thorning *et al*., 2016)** and cancer (**Aune *et al*., 2017; Zang *et al*., 2015**), and has an overall favorable effect on the life and well-being of a person. **Komada *et al*. (2020)** mentioned that regular consumption of milk even supports quality of sleep. **Thorning *et al*. (2016)** state there is increasing evidence suggesting that especially fermented dairy products, cheese and yogurt, are associated with a reduced risk of diabetes type 2. Dairy foods have a significant role in sports nutrition too **(Beigrezaei *et al*, 2022).** Due to allergy to milk protein or lactose intolerance, skipping milk and dairy products is the relatively current trend in the population. Allergy to milk protein ranges in the population at the level of 2-6 % **(Caffareli *et al*., 2010)** and experts agree in favoring the consumption of lactose-free products over the complete exclusion of milk and dairy products **(Heyman, 2006; Harvey *et al*., 2018).** According to **WHO (2006)**, children younger than 11 years old should consume 2-3 portions of dairy products per day**.** Nonbovine milk products mainly from goats, sheep, camel, and donkeys have recently attracted attention for their nutritional, health-promoting, and processing properties (**Ciliberti *et al*., 2022**). Recently, it is becoming more common for sheep and goat milk and cheese to be consumed locally, mostly bought from small farmers (**Kováčová *et al*., 2021**). As compared to cow milk, sheep milk has higher monounsaturated fatty acids and polyunsaturated fatty acids (PUFA), as well as smaller fat globules (**Balthazar *et al*., 2017).** The levels of Ca, P, Mg, Zn, Mn and are higher in sheep milk than in cow milk, while the opposite trend appears to be the case for potassium and sodium **(Park *et al*., 2007).** The content of macroelements and microelements in milk can vary significantly. Their amount depends on many factors, such as the age of the dairy ewe, health status, lactation period, breeding, season or feed quality, but also the method of milk processing, production process, fermentation, or possible fortification (**Bakircioglu *et al*., 2018; Bilandžić *et al*., 2015; Garcia *et al*., 2006; Wang *et al*., 2014**). Essential elements play an important role in metabolic functions such as maintaining pH homeostasis, osmotic pressure, nerve conduction, and muscle contraction **(Bakircioglu *et al*., 2016)**. Inadequate intake of macroelements (Ca, Mg, K, Na), either insufficient or excessive, is associated with serious health problems such as hypertension, osteoporosis, cardiovascular diseases, and others **(Bilanžić *et al*., 2015).** Milk and dairy products are considered to have little health risk **(Licata *et al*., 2012)** and do not contain heavy metals naturally. However, metals such as cadmium, chromium, nickel, lead, and others can contaminate the environment and thus also the feed and subsequently pass into milk and dairy products and cause various health problems to humans and animals as well sooner or later. Many hazardous elements or compounds accumulate along the trophic chain due to the increase in urban, agricultural, and industrial emissions **(Bansal, 2020; Boudebbouz *et al*., 2022; Khan *et al*., 2014)**. In addition, it has been reported that heavy metals can contaminate raw milk through materials and machines used in milk processing and distribution. **Koyuncu and Alwazeer (2019)** state that processed milk is reported to have higher concentrations of heavy metals compared to raw milk**.** The presence of toxic elements even in low concentrations during regular consumption can pose adverse health effects, lead to metabolic disorders, negatively influence the development or function of organs, and even cause the outbreak of illness **(Girma *et al*., 2014; Khan *et al*., 2014).** As a result of the accumulation of toxic elements, growth disorders, mental retardation, and neurological and cardiovascular diseases can occur **(Jaishankar *et al*., 2014).** High levels of cadmium and lead are determined in many studies dealing with monitoring the content of heavy metals in dairy products **(Totan and Filazi, 2020).** Of all the heavy metals, lead and cadmium have prompted the most concern (**Zhou *et al*., 2017; Rahimi, 2013**). These elements are more often found in samples from areas where there is a higher rate of industrial activities in the vicinity **(Swaileh *et al*., 2009**) Since lead and cadmium have no known beneficial role for animals and plants, they cause adverse health effects, other metals, like Zn, Cr, Cu and Fe, are essentiall elements for the human body; each of them plays a crucial role in metabolism, as well as in biochemical processes. Nevertheless, their presence at high concentrations in animals and subsequently in the human body may harm human health as well **(Boudebbouz *et al*., 2022).** Despite being recommended to consume more milk and dairy product, children are more susceptible to the cumulative effect of heavy metals, because toxic elements can build up in the tissues easier during development. **(Rahimi, 2013).** Accumulation of heavy metals occurs at any age, but the highest predisposition for accumulation is in the first 13 years of life, which may also be because of the development of the organism, children consume more food per unit of body weight compared to adults **(ENHIS, 2007).** It is therefore crucial to monitor the level of toxic elements in milk and dairy products, which are a major source of nutrition, particularly for children. The European Union defines a maximum level of heavy metals in milk (**EC No 1881/2006**). Concentrations of elements are one of factors which determine nutritional value of milk and dairy products. (**Zhou *et al*., 2017**). The absence of heavy metals in milk is considered as a direct indicator of its safety and quality of hygienic processing and an indirect indicator of the level of contamination and environmental pollution **(González-Montana *et al*., 2012).** Since higher content of toxic elements in environment and consequently in animal products such as milk is expected in areas with higher level of contamination, the presented study aimed to determine the content of essential and toxic elements in samples of sheep milk collected from three farms in Slovakia from areas with different environmental burden according to the environmental regionalization of the Slovak Environment Agency and to determine if the usage is suitable for human consumption.

MATERIAL AND METHODS

Collections of samples

The occurrence of 22 chemical elements - essential elements (Ca, Co, Cr, Cu, Fe, K, Li, Mg, Mn, Mo, Na, Se, Zn) and toxic elements (Ag, Al, As, Ba, Cd, Ni, Pb, Sb, Sr) in sheep milk from three selected farms were monitored. The Ministry of the Slovak Republic and the Slovak Environmental Agency has determined the environmental regionalization of Slovakia. In the country, there are three types of environmental quality: regions with potentially undisturbed areas, regions with slightly disturbed areas, and regions with heavily disturbed areas. (**Klinda *et al*., 2016; MESR and SEA, 2018**). Orava region, which is located in the northern part of Slovakia, near the border with Poland, represents a potentially undisturbed area. The area is specific to a mountainous country and lower representation of the industry. Farms from the slightly disturbed area and heavily disturbed area are both from Spiš region. The location is shown in Figure 1. Near the farm from the heavily disturbed area the metallurgical factory, specializing specifically in the production of refined copper is located and it has a huge impact on the quality of the environment. Farms from potentially undisturbed area and slightly disturbed area uses a conventional type of farming, while farm from the heavily disturbed area is an ecological farm. In Orava, there are 750 ewes of the Slovak dairy sheep breed, in Spiš (slightly disturbed area) 300 ewes of the improved valachian breed, and (heavily disturbed area) 450 ewes of the improved valachian breed x lacaune.

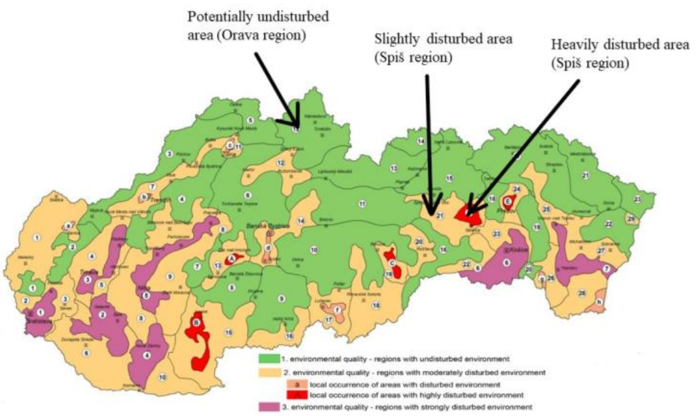

Figure 1 Environmental regional classification of Slovakia **(MESR and SEA, 2018).** Note: Three investigated farms are marked.

Samples preparation

From each farm, samples of sheep milk were collected every two weeks during the period of lactation (from May to October) for one year (in 2021 from the undisturbed and slightly disturbed areas and in 2022 from the heavily disturbed area). In Spiš region, we collected 11 samples from the heavily disturbed area and 12 samples from the slightly disturbed area, and 10 samples from potentially undisturbed area of Orava region were taken, 33 samples in total. Samples were taken from milk tanks as representative of all individuals and stored in freezers at -18℃ until analysis was carried out. First, a homogenization, as preanalytical procedure was performed. The experimental samples weighed between 1.0 and 2.0 g, which was reflected in the measurements.

Analysis of samples

Analysis of the elements was determined using an inductively coupled plasma-optical emission spectrometry with axial plasma configuration and with auto-sampler SPS-3. In the beginning, samples were mineralized in the high-performance microwave digestion system Ethos UP (Milestone Srl, Sorisole, BG, Italy) in a solution of 5 ml $HNO_3 \geq 69.0\%$ (TraceSELECT®, Honeywell Fluka, Morris Plains, USA), 1 ml $H_2O_2 \geq 30\%$, for trace analysis (Sigma Aldrich, Saint–Louis, Missouri, USA), and 2 ml of ultrapure water 18.2 MΩ cm –1; 25°C, Synergy UV, Merck Millipore, France). The method of determination consists of heating and cooling phases. Analysis of the elements was carried out using an inductively coupled plasma–optical emission spectrometer (ICP OES 720, Agilent Technologies Australia (M) Pty Ltd.) with axial plasma configuration and with auto140 sampler SPS-3 (Agilent Technologies, Switzerland). Detections limits (μg/kg) of measured trace elements were followed: Ag 0.3; Al 0.2; As 1.5; Ba 0.03; Ca 0.01; Cd 0.05; Co 0.2; Cr 0.15; Cu 0.3; Fe 0.1; K 0.3; Li 0.06; Mg 0.01; Mn 0.03; Mo 0.5; Na 0.15; Ni 0.3; Pb 0.8; Sb 2.0; Se 2.0; Sr 0.01 and Zn 0.2. and wavelength of determination (nm) follows Ag 328.068; Al 167.019; As 188.980; Ba 455.403; Ca 315.887; Cd 226.502; Co 228.615; Cr 267.716; Cu 324.754; Fe 234.350; K 766.491; Li 670.783; Mg 383.829; Mn 257.610; Mo 204.598; Na 589.592; Ni 231.604; Pb 220.353; Sb 206.834; Se 196.026; Sr 407.771; and Zn 206.200. The legitimacy of the whole method was verified using the certified reference material. Details of the instrumental operating conditions are listed in Table 1. We used the same quality assurance measures, instruments, and laboratory method as in our previous study (**Toman *et al*., 2021**).

Table 1 The operating parameters of determination of elements by ICP-OES

Parameter	Value
RF Power [kW]	1.30
Plasma flow [L/min]	15.0
Auxiliary flow [L/min]	1.50
Nebulizer flow [L/min]	0.85
Replicated read time [s]	5.00
Instrument stabilization [s]	15
Sample uptake delay [s]	25
Pump rate [rpm]	15
Rinse time [s]	10

Statistical analysis

Program Statistica Cz A version 10 (TIBCO Software, Inc., Palo Alto, CA, USA) was used to analyze all results of this study. The obtained results are presented as mean values with standard deviation - descriptive statistics using one-way ANOVA were calculated. Differences in concentrations of the analyzed elements in sheep milk between farms were compared using Sheffe´s test. Statistical significance was defined as $P < 0.05$.

RESULTS AND DISCUSSION

According to available information, most studies dealing with the occurrence of elements in milk in Slovakia are devoted to sheep's milk**.** Table 2 shows detected concentrations of essential and toxic elements in raw sheep's milk collected from farms in areas with different environmental burden in Slovakia. Farm in the slightly and heavily disturbed area are located in Spiš region and farm from Orava represents the region with the potentially undisturbed area. For clarification and to simplify the interpretation of the results (because two farms from areas with different environmental burden come from the same region) nomenclature of environmental regionalization of Slovakia by The Ministry of the Slovak Republic and the Slovak Environmental Agency were used (**MESR and SEA, 2018).**

Calcium (Ca), Sodium (Na), Potassium (K), Magnesium (Mg)

The order of the first four elements according to the occurrence was the same for all monitored farms: Ca > K > Na > Mg. There has been evidence that calcium, potassium, magnesium, and sodium are dominant elements in milk in other studies as well (**Chen *et al*., 2020).** Due to the high level of variability (wide ranges and high standard deviations over the mean values) in mineral contents, comparing milk and dairy products from different species, areas, and farming methods may be more challenging. Diet, breed, individual animal, lactation stage, season of the year, environmental conditions, etc., all mentioned factors affect the mineral content of milk (**Navarro-Alarcón *et al*., 2011).** Among the nutrients that come from dairy, calcium has been the most extensively studied since it is mostly associated with milk consumption. **(Bilandžić *et al*., 2019; Melse – Boonstra, 2020)**. By **Silva and Smetana (2022)** in animal products, the highest concentration of Ca comes from sheep milk, which contain almost 200 mg Ca per 100 g. Normally, 40 % of the calcium from dairy sources is absorbed, with higher absorption in children and lower absorption in the elderly. The absorption of calcium can be influenced by dietary factors. Milk contains phosphopeptides, which are products of enzymatic hydrolysis of casein; alpha-lactalbumin and beta-lactoglobulin, both whey proteins and lactose, which help to increase calcium absorption during digestion (**Melse-Boonstra, 2020**). Concentrations of calcium dominated among all elements in raw sheep's milk from tanks from all monitored areas. We found the highest concentrations of Ca (1654.98 mg/kg) in samples from the heavily disturbed area from a farm with ecological management. However, this concentration was significantly higher ($P < 0.05$) only in comparison with the concentration of Ca (1426.24 mg/kg) in samples from the potentially undisturbed area and not in comparison with the concentration of Ca (1541.56 mg/kg) in samples from the slightly disturbed area**. Pšenková *et al*. (2022)** and **Pšenková and Toman (2020)** found in their studies higher calcium concentrations in samples of sheep's milk from the undisturbed area, in the Orava region, compared to our results. The concentrations they found are more in line with the average concentration in our samples from the heavily disturbed area. **Antunović *et al*., (2011, 2020)** detected even higher concentrations in sheep's milk in Croatia. The highest concentrations of Ca were found in three regions in Austria as well **(Mayer and Fiechter, 2012**). The consumption of raw sheep milk is rare in Slovakia, but hypothetically with average consumption of milk (0.13 kg/day), our samples fulfil daily recommended dose of calcium in the following percentage and order: 26 % from potentially undisturbed area, 29 % slightly disturbed area and 31 % heavily disturbed area for children and 14 %, 15 % and 17 % for physically hard-working men. Recommended doses for Slovak population are listed in table 3.

Table 2 Mean concentrations of chemical elements in sheep's milk from three areas in Slovakia with the different environmental burden (mg/kg).

Element	Region with potentially undisturbed area	Region with slightly disturbed area	Region with heavily disturbed area
	n= 10	n= 12	n=11
	$\bar{x}$+ SD	$\bar{x}$+ SD	$\bar{x}$+ SD
Ag	0.004 ± 0.002	ND	ND
Al	1.38 ± 0.86[a]	ND	0.53 ± 0.56[a]
As	0.06 ± 0.08[a]	ND	0.42 ± 0.15[a]
Ba	0.01 ± 0.02[a, b]	0.15 ± 0.04[a, c]	0.28 ± 0.12[b, c]
Ca	1426.24 ± 136.01[a]	1541.56 ± 69.04	1654.98 ± 92.94[a]
Cd	ND	ND	ND
Co	ND	ND	ND
Cr	ND	ND	ND
Cu	ND	0.15 ± 0.05	0.14 ± 0.14
Fe	1.57 ± 0.28	1.98 ± 0.75	2.05 ± 0.39
K	858.38 ± 104.94[a]	946.35 ± 110.89 [b]	1062.08 ± 87.83[a, b]
Li	0.01 ± 0.002[a]	0.03 ± 0.02[b]	0.07 ± 0.03[a, b]
Mg	135.80 ± 11.67	140.29 ± 21.40	151.53 ± 14.07
Mn	ND	0.06 ± 0.02	0.06 ± 0.01
Mo	ND	ND	0.002 ± 0.01
Na	405.74 ± 38.45	450.00 ± 153.76	457.86 ± 99.04
Ni	ND	ND	0.09 ± 0.07
Pb	ND	ND	0.02 ± 0.03
Sb	ND	ND	0.07 ± 0.06
Se	0.19 ± 0.14	ND	0.15 ± 0.16
Sr	0.90 ± 0.12[a]	0.94 ± 0.18	1.11 ± 0.20[a]
Zn	4.19 ± 0.46[a]	ND	4.97 ± 0.79[a]

SD - standard deviation; [a, b, c] same index in the row for the same element indicates statistically significant difference between compared regions (P<0.05);
ND - concentrations are below the LOD (limit of detection)

Table 3 Recommended daily intake of essential elements for population of the Slovak Republic (group with the lowest and highest requirement) (**Kajaba *et al*., 2015**).

	Ca	Cu	Fe	K*	Mg	Mn*	Na*	Se	Zn
Children (4-7 years old)	700 mg	800 µg	9 mg	1300 mg	120 mg	1,5- 2 mg	500 mg	25 µg	5 mg
Hard-working men (19-34 years old)	1300 mg	1800 µg	12 mg	4000 mg	420 mg	2-5 mg	1500 mg	75 µg	16 mg

*values from **DACH** recommendation, no official recommended daily intake is set for these elements in Slovak Republic

Goat and sheep milk contain higher levels of potassium than other cattle, concentration of K in different types of milk is in decreasing order: goat > sheep > cow > camel > buffalo > human (**Miedico *et al*., 2016).** The potassium content (1062.08 mg/kg) was significantly highest in samples from ecological farms compared to other monitored regions. Higher concentrations of element K were found in samples of sheep's milk from Croatia **(Antunović *et al*., 2011, 2020**) and Austria **(Mayer and Fiechter, 2012**) while **Toman *et al*. (2021)** report lower values in Slovakia, in the Orava region. In the comparison of K level in goat milk from an ecological farm from region Orava, the concentration of K was higher than in all sheep milk samples in the present study, but in goat samples from a conventional farm from region Stredné Považie, the mean concentration of K was higher than in our samples from the farm from a region with the potentially undisturbed area but lower than in our samples from slightly and heavily disturbed areas **(Almášiová *et al*., 2023).** Average consumption of milk from potentially undisturbed area would provide 8 % of recommended dose of K for children, milk from slightly disturbed area would provide 9 % and from heavily disturbed area would provide 10 % for children, while for hard-working men it would be 3 % from all farms. According to some studies, potassium has a preventive effect against hypertension **(O´Halloran *et al*., 2016).** Monitoring the potassium and sodium content in milk also has a preventive character within the health status of animals **(Capcárová *et al*., 2019).** Together with calcium, magnesium is the most important element of bone health **(Capcárová *et al*., 2019). Oh and Deeth (2017)** state that milk and milk products are one of the main sources of magnesium in the diet today. Recent meta-analyses also signal the importance of adequate magnesium intake in the prevention of metabolic syndrome **(La *et al*., 2016; Sarrafzadegan *et al*., 2016.)** Concentrations of Mg in our samples (135.80 mg/kg, 140.29 mg/kg, 151.53 mg/kg are higher than from previous Slovak studies (**Pšenková and Toman, 2021; Pšenková *et al*., 2022**), but also lower in comparison with samples from Austria (**Mayer and Fiechter, 2012**) and Croatia (**Antunović *et al*., 2020**). According to our results, when consuming 0.13 kg milk, children receive 15 % of recommended daily intake of Mg from potentially undisturbed and slightly disturbed area and 16 % from heavily disturbed area, while adult hard-working male receives 4 % of recommended daily intake from all monitored areas.

Iron (Fe), Copper (Cu), Manganese (Mn), Selenium (Se)

Iron is essential for maintaining proper cell function and it is involved in the transport of respiratory gases (**Lieu *et al*., 2001**). Milk is not considered as a good source of iron and its low concentrations in milk do not meet human needs, but its presence in milk has been shown to have a positive effect on inhibiting the growth of bacteria (**Bailey *et al*., 2011**). The measured concentrations of Fe in samples of sheep milk were as follows: 1.57 mg/kg in samples from the potentially undisturbed area, 1.98 mg/kg in samples from the slightly disturbed area, and 2.05 mg/kg in samples from the heavily disturbed area. **Kandhro *et al*. (2022**) describe that the levels of Fe were significantly lower in milk samples of all selected cattle (cow, buffalo, goat, sheep, and camel) used in his study than in other food commodities. **Raynal-Ljutovac *et al*. (2008**) mentions that ewe milk has the highest Fe level among the types of milk. The contents of Fe in our samples were higher than in goat milk from Italy (**Miedico *et al*., 2016**), China (**Zhou *et al*., 2017; Chen *et al*., 2020**), and Croatia (**Antunovic *et al*., 2020**). Concentrations of iron in goat milk from Slovakia were lower only in comparison with our samples from farms from slightly and heavily disturbed areas (**Almášiová *et al*., 2023**). Since iron levels are lower than the recommended dose, there is no risk of iron toxicity for humans **(Bigucu *et al*., 2016).** Elements Cu, Mn and Se are esssential for the human body, but their excessive consummation through diet could be toxic and cause adverse health effects **(ATSDR, 2004; ATSDR, 2012; Nogueira and Rocha, 2011)**. Presence of Cu was detected in samples of sheep milk from region with slightly and heavily disturbed area in concentrations which are in agreement with those reported in the literature **(Chen *et al*., 2020, Bilandžič *et al*., 2015; Kodrik *et al*., 2011)**. Same situation happened in the case of Mn **(Qin *et al*., 2009; Enb *et al*., 2009),** while Se was detected in samples from region with potentially undisturbed area and region with heavily disturbed area. Concentrations of Se were lower than **Chen *et al*. (2020)** found in his study but higher than **Pšenková *et al*. (2022)** found in Orava region in Slovakia.

Silver (Ag), Cadmium (Cd), Cobalt (Co)

Concentrations of toxic elements Ag, Cd, and Co were found in all samples below the limit of detection. In the study by **Tunegová *et al*. (2018)** in samples of sheep's milk from Orava region (potentially undisturbed area) and Horná Nitra (heavily disturbed area) in Slovakia, the content of As, Ni, Pb, and Cd was below the limit of detection as well, although the mentioned elements were present in soil samples from the given environment. In another Slovak study, the investigated Cd content in sheep's milk was below the limit of quantification (**Pšenková *et al.*, 2022**).

Aluminium (Al), Arsenic (As), Molybdenum (Mo), Nickel (Ni), Chromium (Cr), Antimony (Sb), Lead (Pb), Lithium (Li)

In our study Al and As occurred below the limit of detection only in samples from a slightly disturbed area, while concentrations of Mo, Ni, Cr, Sb, and Pb were found only in samples from a heavily disturbed area. Nickel concentrations were also below the LOD in previous studies in Orava (**Pšenková and Toman, 2020; Pšenková *et al*., 2022**). Various range of monitored elements was found in samples of ewe milk from the farm from a heavily disturbed area, which is confirmation of environmental regionalization. A significant difference *(P* < 0.05) between the concentration of Al (1.38 mg/kg) in samples from the potentially undisturbed area and samples from the region with the heavily disturbed area (0.53 mg/kg) was found. A significant difference *(P* < 0.05) between the concentration of As (0.06 mg/kg) in samples from the potentially undisturbed area and samples from the region with the heavily disturbed area (0.42 mg/kg) was found. Concentrations of only three elements, Ag, Cd, and Co were lower than the limit of detection in those samples. Trace amounts of mentioned elements (Al, As, Mo, Ni, Cr, Sb) in samples of sheep´s milk from the heavily disturbed area were captured, but in the case of lead, the average concentration of 0.02 mg/kg in samples reached the level of the permitted limit set by **EC Regulation no. 1881/2006** and **Codex Alimentarius (2007).** We found the presence of lead in 54% of samples, of which in five samples its concentration exceeded the permitted limit 0.02 mg/kg. Evidence that even regular and long-term consumption of low amounts of Cd and Pb can pose health problems exists, as it is highly cumulative, causes pathological changes and these elements are carcinogenic (**Castro-Gonzáles *et al*., 2019; Amegah *et al.*, 2021**). In previous studies, the above-limit lead concentration was also determined by **Toman *et al*. (2022)** in the Orava region, although in other studies from Orava, the lead content was below the detection limit **(Pšenková and Toman, 2020; Pšenková *et al*., 2022).** Generally, sheep's milk contains higher levels of lead and cadmium than cow's and goat's milk. **Oana *et* al. (2016)** confirmed the hypothesis that cadmium and lead are mostly associated with the protein fraction (casein fraction). A high level of lead in the soil can cause an increase in its content in feed and subsequently in milk (**Bocquet *et al*., 2021; Koyuncu and Alwazeer, 2019). Patra *et al.* (2006**) based on the significant negative correlation of the lead level with the concentration of copper, cobalt, and iron in the blood of cows in the vicinity of industrial units, assumed that the increased level of lead in milk can also affect the resulting profile and concentrations of trace elements of milk. Subsequently, in another study, higher concentrations of lead, but also copper, cobalt, and iron were determined in milk samples from an area near an operational lead-zinc smelter and steel mill compared to the control group **(Patra *et al.*, 2008).** There are many studies where higher lead content was detected than this limit as well, for example from Pakistan, Mexico, Croatia, and Hungary (**Kazi *et al*., 2009; Suturović *et al*., 2014; Pompilio *et al*., 2021; Póti *et al*. 2012).** A significant difference *(P* < 0.05) between the concentration of Li was found in samples from region with heavily disturbed region (0,07 mg/kg) and both remaining regions – 0,01 mg/kg in samples from potentially undisturbed region and 0,03 mg/kg from region with slightly disturbed area.

Barium (Ba), Strontium (Sr)

A significant difference (P < 0.05) in the concentration of Ba were found in samples from all regions. Concentrations of Ba were as follows: 0.01 mg/kg in samples from potentially undisturbed areas, 0.15 mg/kg in samples from the slightly disturbed area, and 0.28 mg/kg from the heavily disturbed area. Barium is a physiological antagonist of potassium. In foods of animal origin, the concentration of barium is relatively low (**Oskarsson, 2015**). To characterize the danger of barium, the value of the total daily intake of 20 μg/kg of body weight was selected as an indicative value for health (**WHO, 1990**). The dose at which adverse effects could be expected has not been determined. According to **Pearson and Ashmore** (**2019**), due to the conservatism in setting this value, barium is not considered to be an element that affects human health through the food chain. The content of barium and strontium in milk and milk products has not been determined in a large number of available studies. The concentration of strontium (1.11 mg/kg) in samples from the heavily disturbed area was significantly higher (*P* < 0.05) only in comparison with the concentration of Sr (0.90 mg/kg) in samples from the potentially undisturbed area and not in comparison with the concentration of Sr (0.94 mg/kg) in samples from the slightly disturbed area. As a calcium antagonist, strontium accumulates in the bones, which is particularly unpleasant if it occurs in childhood. In adults, it adheres to the surface of the bones, in children it can be used to form the mineral part of the bone and thus it is stored in the body for many years. If calcium and protein are deficient in a child's diet, strontium can cause poor bone growth The presence of strontium in samples is recommended to take into account with sufficient consumption of calcium. **(ATSDR, 2020; Saribal, 2020).** To characterize the danger of strontium to health, the tolerable daily intake value of 130 μg/kg of body weight, established by the **WHO (2010**), was chosen as an indicative value.

CONCLUSION

According to the results of the sample analysis of raw sheep milk, we can conclude that there are marked differences between the samples from areas with different environmental burden, as expected. This fact confirms the quality of environmental regionalization by The Ministry of the Slovak Republic and the Slovak Environmental Agency. The very low concentrations (below the limit of detection) of toxic elements in sheep milk from potentially undisturbed area and slightly disturbed area proved that the level of environmental contamination in the selected area of Slovakia is low, and milk produced in this part of Slovakia has a high nutritional value, whether consumed directly or further processed, it is safe for consumption and helps to ensure adequate nutrition. On the other hand, more heavy metals in samples from the heavily disturbed area were found in comparison with others farms. Their concentrations were in accordance with permitted limits by European Union, or relevant limit was not established yet, except lead, for which the mean concentration represents 0.02 mg/kg. This is the most fundamental finding in the present study because this value represents the level of the permitted limit set by EC Regulation no. 1881/2006 and Codex Alimentarius General Standard for Contaminants and Toxins in Food and Feed (Codex Stan 193-1995). The next results of this study is content of essential elements such as Ca, K, and Mg which was higher in samples from the region with the heavily disturbed area. Concentration of Ca was significantly higher in farm from area with heavily disturbed area in comparison with farm from potentially undisturbed area, while concentration of K was significantly higher in comparison with both remaining farms. According to these results, further monitoring for the presence of heavy metals in milk is recommended, followed by consideration of regular consumption of sheep's milk and dairy products from heavily disturbed area, if appropriate. Considering the results, we can assume that regular long-term consumption can be even more dangerous for vulnerable groups, but further health risk assessment could be beneficial and more informative.

Acknowledgments: This work was supported by the Slovak Research and Development Agency under Contract no. APVV-18-0227**.**

REFERENCES

Swaileh, K. M., Abdulkhaliq, A., Hussein, R. M., & Matani, M. (2009). Distribution of Toxic Metals in Organs of Local Cattle, Sheep, Goat and Poultry from the West Bank, Palestinian Authority. *Bulletin of Environmental Contamination and Toxicology*, *83*(2), 265–268. https://doi.org/10.1007/s00128-009-9704-x

Almášiová, S., Toman, R., Pšenková, M., Tančin, V., Mikláš, Š., & Jančo, I. (2023). Chemical elements content in goat milk, whey, cheese and yogurt from an ecological and conventional farm in Slovakia. *Journal of Central European Agriculture*, *24*(1), 43–52. https://doi.org/10.5513/JCEA01/24.1.3733

Amegah, A. K., Sewor, C., & Jaakkola, J. J. K. (2021). Cadmium exposure and risk of adverse pregnancy and birth outcomes: A systematic review and dose–response meta-analysis of cohort and cohort-based case–control studies. *Journal of Exposure Science & Environmental Epidemiology*, *31*(2), Art. 2.https://doi.org/10.1038/s41370-021-00289-6

Antunović, Z., Mioč, B., Klir, Ž., Širić, I., Držaić, V., Lončarić, Z., Bukvić, G., & Novoselec, J. (2020). Concentrations of mercury and other elements in ewes' milk: Effect of lactation stage. *Chemosphere*, *261*, 128128. https://doi.org/10.1016/j.chemosphere.2020.128128

Antunović, Z., Bogut, I., Senčić, D., Katić, M., & Mijić, P. (2005). Concentrations of selected toxic elements (cadmium, lead, mercury and arsenic) in ewe milk in dependence on lactation stage. Czech Journal of Animal Science, 50(8), 369-375. https://doi.org/10.17221/4179-cjas

ATSDR (Agency for Toxic Substances and Disease Registry). (2004). Toxicological profile for copper U.S. Department of Health and Human Services, Atlanta, GA.

ATSDR (Agency for Toxic Substances and Disease Registry). (2012). Toxicological profile for manganese. U.S. Department of Health and Human Services, Atlanta, GA.

ATSDR (Agency for Toxic Substances and Disease Registry) Public Health Summaries Strontium.2020. Available at https://www.atsdr.cdc.gov/toxprofiles/tp159.pdf [Accessed 31 August 2022].
Aune, D., Giovannucci, E., Boffetta, P., Fadnes, L. T., Keum, N., Norat, T., Greenwood, D. C., Riboli, E., Vatten, L. J., & Tonstad, S. (2017). Fruit and vegetable intake and the risk of cardiovascular disease, total cancer and all-cause mortality—A systematic review and dose-response meta-analysis of prospective studies. *International Journal of Epidemiology*, *46*(3), 1029–1056. https://doi.org/10.1093/ije/dyw319
Bailey, J.R., Probert, C.S., & Cogan, T.A. 2011. Identification and characterisation of an iron-responsive candidate probiotic. *PLOS One*. 6, e26507. https://doi.org/10.1371/journal.pone.0026507
Bakircioglu, D., Topraksever, N., Yurtsever, S., Kizildere, M., & Kurtulus, Y. B. (2018). Investigation of macro, micro and toxic element concentrations of milk and fermented milks products by using an inductively coupled plasma optical emission spectrometer, to improve food safety in Turkey. *Microchemical Journal*, *136*, 133–138.https://doi.org/10.1016/j.microc.2016.10.014
Balthazar, C. F., Pimentel, T. C., Ferrão, L. l., Almada, C. N., Santillo, A., Albenzio, M., Mollakhalili, N., Mortazavian, A. M., Nascimento, J. S., Silva, M. C., Freitas, M. Q., Sant'Ana, A. S., Granato, D., & Cruz, A. G. (2017). Sheep Milk: Physicochemical Characteristics and Relevance for Functional Food Development. *Comprehensive Reviews in Food Science and Food Safety*, *16*(2), 247–262. https://doi.org/10.1111/1541-4337.12250
Bansal, O. P. (2020). Health impacts of the potentially toxic metals present in milk, dairy products, chocolates, alcoholic and non-alcoholic beverages: A review. *Journal of Environmental Science, Toxicology and Food Technology*, 14 (7), 35-46.
Beigrezaei, S., Forbes, S. C., Kaviani, M., Roy, B. D., & Salehi-Abargouei, A. (2022). Chapter 15—Role of dairy foods in sport nutrition. A. G. da Cruz, C. S. Ranadheera, F. Nazzaro, & A. M. Mortazavian (Ed.), *Dairy Foods* (339–364). Woodhead Publishing. https://doi.org/10.1016/B978-0-12-820478-8.00009-2
Bigucu, E., Kaptan, B., Palabiyik, İ., & Oksuz, O. (2016.). The Effect of Environmental Factors on Heavy Metal and Mineral Compositions of Raw Milk and Water Samples. *Journal of Tekirdag Agricultural Faculty*, 13(4), 61- 70.
Bilandžić, N., Sedak, M., Djokic, M., Bozic, D., Solomun-Kolanovic, B., & Varenina, I. (2015). Differences in macro- and microelement contents in milk and yoghurt. *Archives of Biological Sciences*, *67*(4), 1391–1397.https://doi.org/10.2298/ABS140312117B
Bilandžić, N., Sedak, M., Čalopek, B., Đokić, M., Kolanović, B. S., Varenina, I., Luburić, B., Đurđica, & Varga, I. (2019). Macro- and microelement contents in sterilized milk of different manufacturers from Croatia and EU. *Veterinarska stanica*, 50 (1), 1-10.
Bocquet, A., Barouki, R., Briend, A., Chouraqui, J.-P., Darmaun, D., Feillet, F., Frelut, M.-L., Guimber, D., Lapillonne, A., Peretti, N., Rozé, J.-C., Simeoni, U., Turck, D., & Dupont, C. (2021). Potential toxicity of metal trace elements from food in children. *Archives de Pédiatrie*, *28*(3), 173–177.https://doi.org/10.1016/j.arcped.2021.03.001
Boudebbouz, A., Boudalia, S., Bousbia, A., Gueroui, Y., Boussadia, M. I., Chelaghmia, M. L., Zebsa, R., Affoune, A. M., & Symeon, G. K. (2022). Determination of Heavy Metal Levels and Health Risk Assessment of Raw Cow Milk in Guelma Region, Algeria. *Biological Trace Element Research*., 201, 1704-1716.https://doi.org/10.1007/s12011-022-03308-1
Caffarelli, C., Baldi, F., Bendandi, B., Calzone, L., Marani, M., & Pasquinelli, P. (2010). Cow's milk protein allergy in children: A practical guide. *Italian Journal of Pediatrics*, *36*, 5.https://doi.org/10.1186/1824-7288-36-5
Capcarova, M., Binkowski, L. J., Stawarz, R., Schwarczova, L., & Massanyi, P. (2019). Levels of Essential and Xenobiotic Elements and Their Relationships in Milk Available on the Slovak Market with the Estimation of Consumer Exposure. *Biological Trace Element Research*, *188*(2), 404–411.https://doi.org/10.1007/s12011-018-1424-9
Castro – González, N. P., Calderón – Sánchez, F., Pérez – Sato, M., Soní – Guillermo, E., & Reyes – Cervantes, E. (2019). Health risk due to chronic heavy metal consumption via cow's milk produced in Puebla, Mexico, in irrigated wastewater areas. *Food Additives & Contaminants: Part B*, *12*(1), 38–44.https://doi.org/10.1080/19393210.2018.1520742
Ciliberti, M. G., Santillo, A., Caroprese, M., & Albenzio, M. (2022). Chapter 5—Nonbovine milk products. V A. G. da Cruz, C. S. Ranadheera, F. Nazzaro, & A. M. Mortazavian (Ed.), *Dairy Foods* (s. 91–115). Woodhead Publishing. https://doi.org/10.1016/B978-0-12-820478-8.00001-8
Deutsche Gesellschaft für Ernährung (DGE) *DACH-Referenzwerte für die Nährstoffzufuhr. 1. Aufl.* Frankfurt am Main: Umschau/Braus; 2000.
EC (2015) Commission regulation (EC) no. 488/2014 of 12 May 2014 amending Regulation (EC) No 1881/2006 as regards maximum levels of cadmium in foodstuffs. OJEU 49:5-24.
Enb, A., Donia, M. A., Abd-Rabou, N. S., Abou-Arab, A. A. K., & El-Senaity, M. H. (2009). Chemical composition of raw milk and heavy metals behavior during processing of milk products. *Global Veterinaria*, *3*(3), 268–275.
European Environment and Health Information System (ENHIS). (2007). Exposure of children to chemical hazards in food. Fact Sheet No. 4.4, CODE: RPG4_Food_EX1. World Health Organization.
Fardellone, P. (2019). The effect of milk consumption on bone and fracture incidence, an update. *Aging Clinical and Experimental Research*, *31*(6), 759–764.https://doi.org/10.1007/s40520-019-01192-9
García, M. I. H., Puerto, P. P., Baquero, M. F., Rodríguez, E. R., Martín, J. D., & Romero, C. D. (2006). Mineral and trace element concentrations of dairy products from goats' milk produced in Tenerife (Canary Islands). *International Dairy Journal*, *16*(2), 182–185. https://doi.org/10.1016/j.idairyj.2005.01.011
Gaucheron, F. (2011). Milk and Dairy Products: A Unique Micronutrient Combination. *Journal of the American College of Nutrition*, *30*(sup5), 400S-409S.https://doi.org/10.1080/07315724.2011.10719983
Girma, K., Tilahun, Z., Haimanot, D. (2014). Review on milk safety with emphasis on ıts public health", *World Journal of Dairy & Food Sciences* 9 (2): 166-183, 2014. https://doi:10.5829/idosi.wjdfs.2014.9.2.85184
González-Montaña, J. R., Senís, E., Gutiérrez, A., & Prieto, F. (2012). Cadmium and lead in bovine milk in the mining area of the Caudal River (Spain). *Environmental Monitoring and Assessment*, *184*(7), 4029–4034.https://doi.org/10.1007/s10661-011-2241-1
Guo, J., Astrup, A., Lovegrove, J. A., Gijsbers, L., Givens, D. I., & Soedamah-Muthu, S. S. (2017). Milk and dairy consumption and risk of cardiovascular diseases and all-cause mortality: Dose–response meta-analysis of prospective cohort studies. *European Journal of Epidemiology*, *32*(4), 269–287.https://doi.org/10.1007/s10654-017-0243-1
Hanach, N. I., McCullough, F., & Avery, A. (2019). The Impact of Dairy Protein Intake on Muscle Mass, Muscle Strength, and Physical Performance in Middle-Aged to Older Adults with or without Existing Sarcopenia: A Systematic Review and Meta-Analysis. *Advances in Nutrition*, *10*(1), 59–69.https://doi.org/10.1093/advances/nmy065
Harvey, L., Ludwig, T., Hou, A. Q., Hock, Q. S., Tan, M. L., Osatakul, S., Bindels, J., & Muhardi, L. (2018). Prevalence, cause and diagnosis of lactose intolerance in children aged 1-5 years: A systematic review of 1995-2015 literature. *Asia Pacific Journal of Clinical Nutrition*, *27*(1), 29–46.https://doi.org/10.6133/apjcn.022017.05
Heyman, M. B. (2006). Lactose Intolerance in Infants, Children, and Adolescents. *Pediatrics*, *118*(3), 1279–1286. https://doi.org/10.1542/peds.2006-1721
Chen, L., Li, X., Li, Z., & Deng, L. (2020). Analysis of 17 elements in cow, goat, buffalo, yak, and camel milk by inductively coupled plasma mass spectrometry (ICP-MS). *RSC Advances*, *10*(12), 6736–6742.https://doi.org/10.1039/D0RA00390E
Jaishankar, M., Tseten, T., Anbalagan, N., Mathew, B. B., & Beeregowda, K. N. (2014). Toxicity, mechanism and health effects of some heavy metals. *Interdisciplinary Toxicology*, *7*(2), 60–72.https://doi.org/10.2478/intox-2014-0009
Joint FAO/WHO Codex Alimentarius Commission. (2007). Codex alimentarius: cereals, pulses, legumes and vegetable proteins. Rome: World Health Organization: Food and Agriculture Organization of the United Nations.
Kajaba, I., Štencl, J., Ginter, E., Šašinka, M., Trusková, I., Gazdíková, K., Hamade, J., Bzdúch, V. (2015). New recommended dietary allowances (RDA) of the population from 2015 –practice implementation. In. New health recommendations in the nutrition of the Slovak population –And transfer to practical nutrition. Lužianky: National agricultural and food industry center, pp. 10-26. ISBN 978-80-89162-64-2
Kandhro, F., Kazi, T. G., Afridi, H. I., & Baig, J. A. (2022). Compare the nutritional status of essential minerals in milk of different cattle and humans: Estimated daily intake for children. *Journal of Food Composition and Analysis*, *105*, 104214.
https://doi.org/10.1016/j.jfca.2021.104214
Kapila, R., Kavadi, P. K., & Kapila, S. (2013). Comparative evaluation of allergic sensitization to milk proteins of cow, buffalo and goat. *Small Ruminant Research*, *112*(1), 191–198.https://doi.org/10.1016/j.smallrumres.2012.11.028
Kazi, T. G., Jalbani, N., Baig, J. A., Kandhro, G. A., Afridi, H. I., Arain, M. B., Jamali, M. K., & Shah, A. Q. (2009). Assessment of toxic metals in raw and processed milk samples using electrothermal atomic absorption spectrophotometer. *Food and Chemical Toxicology*, *47*(9), 2163–2169.https://doi.org/10.1016/j.fct.2009.05.035
Khan, N., Jeong, S., Hwang, I. M., Sung kim, J., Sung hwa, CH., Yeong nho, E., Yeon choi, J., Kyung Su Park, Kyong Su Kim. (2014). Analysis of Minor and Trace Elements in Milk and Yogurts by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS). *Food Chemistry*, 147, 220–24.https://doi.org/10.1016/j.foodchem.2013.09.147
Klinda, J. Mičík, T. Nemethova, M. Slamkova (2016). Environmental regionalization of the Slovak republic 2016. ISBN: 978 -80 – 89503 – 48 -3.
Kodrik, L., Wagner, L., Imre, K., Polyak, K. F., Besenyei, F., & Husveth, F. (2011). The Effect of Highway Traffic on Heavy Metal Content of Cow Milk and Cheese. *Hungarian Journal of Industry and Chemistry*, 15–19. https://doi.org/10.1515/375
Komada, Y., Okajima, I., & Kuwata, T. (2020). The Effects of Milk and Dairy Products on Sleep: A Systematic Review. *International Journal of Environmental Research and Public Health*, *17*(24), 9440.https://doi.org/10.3390/ijerph17249440
Kováčová, M., Výrostková, J., Dudriková, E., Zigo, F., Semjon, B., & Regecová, I. (2021). Assessment of Quality and Safety of Farm Level Produced Cheeses from

Sheep and Goat Milk. *Applied Sciences*, *11*(7), Art. 7.https://doi.org/10.3390/app11073196
Koyuncu, M., & Alwazeer, D. (2019). Determination of trace elements, heavy metals, and antimony in polyethylene terephthalate–bottled local raw cow milk of Iğdır region in Turkey. *Environmental Monitoring and Assessment*, *191*(11), 666. https://doi.org/10.1007/s10661-019-7851-z.
La, S. A., Lee, J. Y., Kim, D. H., Song, E. L., Park, J. H., & Ju, S. Y. (2016). Low Magnesium Levels in Adults with Metabolic Syndrome: A Meta-Analysis. *Biological Trace Element Research*, *170*(1), 33–42.https://doi.org/10.1007/s12011-015-0446-9
Lee, M., Lee, H., Kim, J. (2018). Dairy food consumption is associated with a lower risk of the metabolic syndrome and its components: A systematic review and meta-analysis. *British Journal of Nutrition*. vol. 120,373-384.https://doi.org/10.1017/S0007114518001460
Licata, P., Di Bella, G., Potortì, A. G., Lo Turco, V., Salvo, A., Dugo, G. (2012). Determination of trace elements in goat and ovine milk from calabria (Italy) by ICP-AES. *Food Additives and Contaminants: Part B Surveillance*, 5 (4), 268-271. https://doi.org/10.1080/19393210.2012.705335
Licata, P., Trombetta, D., Cristani, M., Giofrè, F., Martino, D., Calò, M., & Naccari, F. (2004). Levels of "toxic" and "essential" metals in samples of bovine milk from various dairy farms in Calabria, Italy. *Environment International*, *30*(1), 1–6. https://doi.org/10.1016/S0160-4120(03)00139-9
Lieu, P.T., Heiskala, M., Peterson, P.A., & Yang, Y. (2001). The roles of iron in health and disease. *Molecular Aspects of Medicine.* 22. 1-87.
Lu, L., Xun, P., Wan, Y., He, K., & Cai, W. (2016). Long-term association between dairy consumption and risk of childhood obesity: A systematic review and meta-analysis of prospective cohort studies. *European Journal of Clinical Nutrition*, *70*(4), 414–423. https://doi.org/10.1038/ejcn.2015.226
Mayer, H. K., Fiechter, G. (2012). Physical and chemical characteristics of sheep and goat milk in Austria. *International Dairy Journal*, *24*(2), 57–63.https://doi.org/10.1016/j.idairyj.2011.10.012
Melse-Boonstra, A. (2020). Bioavailability of Micronutrients From Nutrient-Dense Whole Foods: Zooming in on Dairy, Vegetables, and Fruits. *Frontiers in Nutrition*, *7*, 101.https://doi.org/10.3389/fnut.2020.00101
MESR and SEA. 2018. State of the Environment Report Slovak Republic 2018. Bratislava, Banská Bystrica: Ministry of Environment of the Slovak Republic, Slovak Environmental Agency. ISBN 978-80-8213-007-5.
Miedico, O., Tarallo, M., Pompa, C., & Chiaravalle, A. E. (2016). Trace elements in sheep and goat milk samples from Apulia and Basilicata regions (Italy): Valuation by multivariate data analysis. *Small Ruminant Research*, *135*, 60–65. https://doi.org/10.1016/j.smallrumres.2015.12.019
Navarro-Alarcón, M., Cabrera-Vique, C., Ruiz-López, M. D., Olalla, M., Artacho, R., Giménez, R., Quintana, V., & Bergillos, T. (2011). Levels of Se, Zn, Mg and Ca in commercial goat and cow milk fermented products: Relationship with their chemical composition and probiotic starter culture. *Food Chemistry*, *129*(3), 1126–1131.https://doi.org/10.1016/j.foodchem.2011.05.090
Nogueira, C. W., & Rocha, J. B. (2011). Toxicology and pharmacology of selenium: emphasis on synthetic organoselenium compounds. *Archives of toxicology*, *85*(11), 1313–1359. https://doi.org/10.1007/s00204-011-0720-3
O'Halloran, S. A., Grimes, C. A., Lacy, K. E., Campbell, K. J., & Nowson, C. A. (2016). Dietary Intake and Sources of Potassium and the Relationship to Dietary Sodium in a Sample of Australian Pre-School Children. *Nutrients*, *8*(8), Art. 8.https://doi.org/10.3390/nu8080496
Oana, C., Tănaselia, C., Miclean, M., Levei, E., Șenilă, M., & Șenilă, L. (2016). Analysis of Minor and Trace Elements in Cow, Goat and Sheep Milk in the NW Part of Romania. *ProEnvironment* 9, 87 – 90.
Oh, H. E., & Deeth, H. C. (2017). Magnesium in milk. *International Dairy Journal*, *71*, 89–97.https://doi.org/10.1016/j.idairyj.2017.03.009
Oskarsson, A. (2015). Chapter 29—Barium. V G. F. Nordberg, B. A. Fowler, & M. Nordberg (Ed.), *Handbook on the Toxicology of Metals (Fourth Edition)* (s. 625–634). Academic Press.https://doi.org/10.1016/B978-0-444-59453-2.00029-9
Park, Y.W., Juarez, M., Ramos, M., & Haenlein, G.F.W. (2007). Physico-chemical characteristics of goat and sheep milk. *Small Ruminants Res*, 68, 88–11. https://doi.org/10.1016/j.smallrumres.2006.09.013
Patra, R. C., Swarup, D., Kumar, P., Nandi, D., Naresh, R., & Ali, S. L. (2008). Milk trace elements in lactating cows environmentally exposed to higher level of lead and cadmium around different industrial units. *Science of The Total Environment*, *404*(1), 36–43. https://doi.org/10.1016/j.scitotenv.2008.06.010
Pearson, A. J., & Ashmore, E. (2019). Risk assessment of antimony, barium, beryllium, boron, bromine, lithium, nickel, strontium, thallium and uranium concentrations in the New Zealand diet. *Food Additives & Contaminants: Part A*, *37*(3), 451–464. https://doi.org/10.1080/19440049.2019.1704445
Pompilio, C.G. N., Francisco, C.S., Tulio, F. de M.T. M., Samuel, S.-M. S., & Elisa, G.J. F. (2021). Heavy metals in blood, milk and cow's urine reared in irrigated areas with wastewater. *Heliyon*, *7*(4).https://doi.org/10.1016/j.heliyon.2021.e06693
Póti, P., Pajor, F., Bodnár, Á., & Bárdos, L. (2012). Accumulation of some heavy metals (Pd, Cd and Cr) in milk of grazing sheep in north-east Hungary. *Journal of Microbiology, Biotechnology and Food Sciences*, 2 (1) 389-394.
Pšenková, M., & Toman, R. (2020). Essential and toxic elements concentrations in animal tissues of sheep from two different regions of Slovakia. *Acta Fytotechnica et Zootechnica*, *23*(4), Art. 4. https://afz.fapz.uniag.sk/legacy/journal/index.php/on_line/article/view/774/../774.html
Pšenková, M., & Toman, R. (2021). Determination of Essential and Toxic Elements in Raw Sheep's Milk from Area of Slovakia with Environmental Burden. *Biological Trace Element Research*, *199*(9), 3338–3344.https://doi.org/10.1007/s12011-020-02452-w
Pšenková, M., Toman, R., & Almášiová, S. (2022). Analysis of concentrations of risk and toxic elements in sheep milk from area of Slovakia with potentially undisturbed environment. *Journal of Microbiology, Biotechnology and Food Sciences*, e5422–e5422. https://doi.org/10.55251/jmbfs.5422
Qin, L.-Q., Wang, X.-P., Li, W., Tong, X., & Tong, W.-J. (2009). The minerals and heavy metals in cows milk from China and Japan. *Journal of Health Science*, *55*(2), 300–305. https://doi.org/10.1248/jhs.55.300.
Rahimi, E. (2013). Lead and cadmium concentrations in goat, cow, sheep, and buffalo milks from different regions of Iran. *Food Chemistry*, *136*(2), 389–391.https://doi.org/10.1016/j.foodchem.2012.09.016
Ratajczak, A. E., Zawada, A., Rychter, A. M., Dobrowolska, A., & Krela-Kaźmierczak, I. (2021). Milk and Dairy Products: Good or Bad for Human Bone? Practical Dietary Recommendations for the Prevention and Management of Osteoporosis. *Nutrients*, *13*(4), Art. 4. https://doi.org/10.3390/nu13041329
Raynal-Ljutovac, K., Lagriffoul, G., Paccard, P., Guillet, I., & Chilliard, Y. (2008). Composition of goat and sheep milk products: An update. *Small Ruminant Research*, *79*(1), 57–72.https://doi.org/10.1016/j.smallrumres.2008.07.009
Saribal, D. (2020). ICP-MS Analysis of Trace Element Concentrations in Cow's Milk Samples from Supermarkets in Istanbul, Turkey. *Biological Trace Element Research*, *193*(1), 166–173https://doi.org/10.1007/s12011-019-01708-4
Sarrafzadegan, N., Khosravi-Boroujeni, H., Lotfizadeh, M., Pourmogaddas, A., & Salehi-Abargouei, A. (2016). Magnesium status and the metabolic syndrome: A systematic review and meta-analysis. *Nutrition*, *32*(4), 409–417. https://doi.org/10.1016/j.nut.2015.09.014
Silva, B. Q., & Smetana, S. (2022). Review on milk substitutes from an environmental and nutritional point of view. *Applied Food Research*, *2*(1), 100105.https://doi.org/10.1016/j.afres.2022.100105
Srbely, V., Janjua, I., Buchholz, A. C., & Newton, G. (2019). Interventions Aimed at Increasing Dairy and/or Calcium Consumption of Preschool-Aged Children: A Systematic Literature Review. *Nutrients*, *11*(4), Art. 4.https://doi.org/10.3390/nu11040714
Suturović, Z., Kravić, S., Milanović, S., Đurović, A., & Brezo, T. (2014). Determination of heavy metals in milk and fermented milk products by potentiometric stripping analysis with constant inverse current in the analytical step. *Food Chemistry*, *155*, 120–125. https://doi.org/10.1016/j.foodchem.2014.01.030
Thorning, T. K., Raben, A., Tholstrup, T., Soedamah-Muthu, S. S., Givens, I., & Astrup, A. (2016). Milk and dairy products: Good or bad for human health? An assessment of the totality of scientific evidence. *Food & Nutrition Research*, *60*, 32527. https://doi.org/10.3402/fnr.v60.32527
Toman, R., Psenkova, M., Tancin, V., & Miskeje, M. (2022). Mutagens in raw ewe milk in Orava region, northern Slovakia: Metals. *Environmental Science and Pollution Research*, *29*(41), 62259–62271.https://doi.org/10.1007/s11356-022-20871-1
Toman, R., Pšenková, M., Imrich, I., Hluchý, S., & Almášiová, S. (2021). Essential and non-mutagenic elements in raw ewe milk. *Science, Technology and Innovation*, *14*(3), Art. 3.https://doi.org/10.55225/sti.316
Totan, F. E., & Filazi, A. (2020). Determination of some element levels in various kinds of cow's milk processed in different ways. *Environmental Monitoring and Assessment*, *192*(2), 112.https://doi.org/10.1007/s10661-020-8088-6
Tunegová, M., Toman, R., Tančin, V., & Janíček, M. (2018). Occurrence of selected metals in feed and sheep´s milk from areas with different environmental burden. *Potravinarstvo*, *12*(1), 454–460.https://doi.org/10.5219/920
Wang, H., Liu, Z., Liu, Y., Qi, Z., Wang, S., Liu, S., Dong, S., Xia, X., & Li, S. (2014). Levels of Cu, Mn, Fe and Zn in Cow Serum and Cow Milk: Relationship with Trace Elements Contents and Chemical Composition in Milk. *Acta Scientiae Veterinariae*, 42 (1190), 1-15.
WHO. Environmental health criteria 107: Barium. United Nations Environment Programme, International Labour Organization and World Health Organization: Geneva, Switzerland, 1990; 148 s.
WHO. Strontium and strontium compounds. WHO: Geneva, Switzerland, 2010; 63 s.
World Health Organization. Regional Office for Europe. Food and nutrition policy for schools: a tool for the development of school nutrition programmes in the European Region. Copenhagen: WHO Regional Office for Europe. 2006.
Zang, J., Shen, M., Du, S., Chen, T., & Zou, S. (2015). The Association between Dairy Intake and Breast Cancer in Western and Asian Populations: A Systematic Review and Meta-Analysis. *Journal of Breast Cancer*, *18*(4), 313–322. https://doi.org/10.4048/jbc.2015.18.4.313
Zhou, X., Qu, X., Zhao, S., Wang, J., Li, S., & Zheng, N. (2017). Analysis of 22 Elements in Milk, Feed, and Water of Dairy Cow, Goat, and Buffalo from Different

Regions of China. *Biological Trace Element Research*, *176*(1), 120–129. https://doi.org/10.1007/s12011-016-0819-8