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Lead and cadmium content in wheat grain from different regions of Poland and risk analysis

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Abstract: The level of wheat grain contamination with lead and cadmium was determined using electrothermal atomic absorption spectrometry with Zeeman background correction (ETAAS) after microwave digestion. The obtained limits of quantification were 0.001 mg·kg^{-1}, for both metals. A total of 300 samples of wheat grain from agricultural regions of Poland were examined, 150 each from the two consecutive harvest years 2017 and 2018.

None of the tested samples exceeded the maximum level of these metals, as specified in the European regulations. The contents of lead and cadmium in wheat grain from both years of harvest ranged from <0.001 to 0.098 mg·kg^{-1} and from 0.006 to 0.098 mg·kg^{-1}, respectively. Despite similar ranges of these metals, the highest lead contents were two times lower than the maximum limit value, while the highest cadmium contents were close to it.

As for lead, a significantly higher ($p < 0.05$) mean content of this metal was found in wheat grain from the 2018 harvest compared to 2017 and in the western compared to eastern regions of the country. However, the cadmium contents did not differ significantly between the two harvest years, but were significantly higher ($p < 0.05$) in wheat grain from the southern regions compared to northern regions of Poland. Additionally, the highest contents of cadmium, close to the maximum limit, were found in the South-West region and in the both years of harvest.

The risk analysis of the occurrence of the excessive contents of toxic metals in wheat grain showed a low risk level for lead in all investigated regions, and a medium level for cadmium, in general.

Keywords: contamination, Poland, risk analysis of excessive content, toxic metals, wheat

INTRODUCTION

Exposure to toxic metals, including lead and cadmium, is of great importance for human health as they are harmful in small doses [Rana *et al.* 2018; Thounwou *et al.* 2012]. Their common feature is the ability to accumulate in the body, a long biological half-life and the related toxicity under long-term exposure. They are responsible for the disruption of the natural life processes, and consequently for the emergence of some civilisation diseases and health disorders. Food has been identified as one of the main source of these metals for humans [EFSA 2012a, b]. However, human exposure is not necessarily associated with food consumption heavily contaminated with toxic metals, but rather to the consumption of large amounts of even slightly contaminated food. The above statement applies mainly to cereals and cereal products, which are considered the staple food in the human diet and consumed in large quantities. They are considered very important in human nutrition because they are the main source of energy for all groups of population and significantly contribute to the consumption of protein, dietary fibre, vitamins (A, B, C, D, E), mineral elements including iron, essential amino acids, fatty acids, phytochemicals [Laskowski *et al.* 2019; Shewry, Hey 2015]. But they can also be contaminated with toxic metals such as lead and cadmium. The content of these metals in cereals, including wheat grain, depends on a number of natural and anthropogenic factors. These include: soil factors as soil abundance in minerals, soil pH, content of organic matter, genotype of plants or climatic conditions, as well as the proximity of highways or large urban agglomerations and industrial plants [Belimov *et al.* 2015; Kabata-Pendias 2011; Kim *et al.* 2017]. The primary source of pollution of the natural environment with toxic metals are volcanic activities, forest fires, and the exploitation of various types of fossil mineral resources and their processing. Toxic metals released during these processes enter the natural cycle of matter and may get into soil as a result of the atmospheric precipitation. An additional source of environmental pollution with these metals is the use of chemicals in agriculture: fertilisers, especially phosphorous, pesticides, and municipal and industrial wastes used for soil fertilisation. The main recipients of various contaminants from the environment, including toxic metal, are plants, which however, are characterised by a different degree of tolerance to their high concentrations, which is closely related to their complicated metabolism. Plants can accumulate these metals and survive even in heavily contaminated soil without any toxic symptoms, to levels already threatening human health [JĘDRZEJCZAK 2013; KHAN *et al.* 2015; RAI *et al.* 2019; WANG *et al.* 2017].

Based on the potentially critical adverse effects, maximum permissible levels for lead and cadmium were specified in two current European regulations, i.e. 0.20 mg·kg^{-1} and 0.10 mg·kg^{-1}, respectively [Commission Regulation (EU) 2021/1317; Commission Regulation (EU) 2021/1323].

This research focused on the assessment of contamination with toxic metals (Pb, Cd) of wheat grain collected from the agricultural regions of the country, divided into four regions and from the two consecutive harvest years 2017 and 2018. The risk analysis of the occurrence of excessive levels of lead and cadmium in wheat grain in various regions of Poland was also considered.

STUDY MATERIALS AND METHODS

STUDY MATERIALS

A total of 300 samples of wheat grain with a mass of approx. 2 kg, from two growing seasons 2017 (n = 150) and 2018 (n = 150) and stored in grain elevators all over Poland, were tested. Sampling was performed in accordance with the PN-EN ISO 24333:2012 standard by companies collecting and storing grain and delivered for analysis. The representative number of samples from individual region were determined on the basis of the total harvest volume in 2016 year, which was 1.08·10^{10} They were respectively: n = 51, 20, 44, and 35 samples each year originated from the North-West, North-East, South-West and South-East regions of Poland (Fig. 1), and each sample represented approx. 7.2·10^{7} kg of wheat grain. All samples were stored in sealed bags at –20°C until analysis.

All chemicals used in this study were of high analytical grade: nitric acid 69%, palladium modifier (10 ±0.02 g·dm^{-3} in % HNO_3), standard solutions of Pb and Cd in concentration of 1000 mg·dm^{-3}, argon 99.999%, double distilled water from Water still AQUATRON A4D (England).

STUDY METHODS

Prior to analysis, the samples were brought to room temperature and ground in a Knife Mill Grindomix GM 200 (Germany) laboratory grinder.

The concentrations of Pb and Cd in wheat samples were determined according to the PN-EN 14084:2004 standard after microwave digestion. About 0.3 g of a ground wheat grain sample

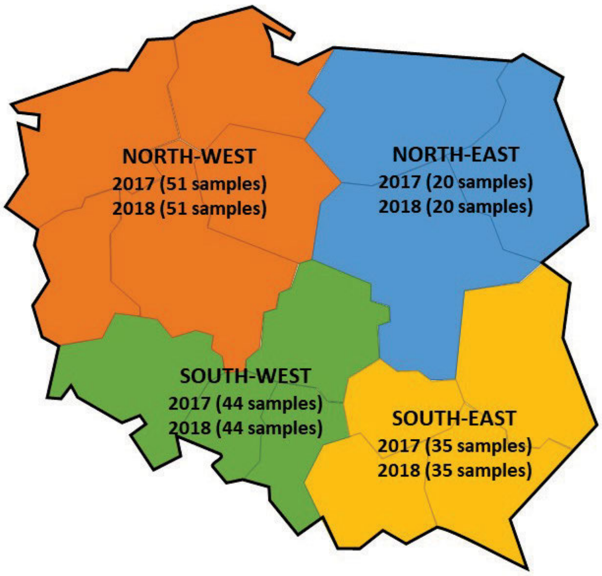

Fig. 1. Investigated regions of Poland from the 2017 and 2018 harvest; source: own elaboration

was weighed with the accuracy of 0.001 g into a Teflon vessel, 4 cm^3 of HNO_3 were added and left until next day. Digestion was performed in a microwave oven (MARS 5) in four steps with power setting of 400–1200 W and during 50 min. After cooling and removing the excess of nitrous oxides in the ultrasonic bath, the sample was transferred into a 25 cm^3 volumetric flask and Pb and Cd were determined using electrothermal atomic absorption spectrometry with Zeeman background correction (ETAAS) – SOLAAR M6.

The limit of quantification (LOQ) was 0.001 mg·kg^{-1} for both metals meeting the criteria of Commission Regulation (EC) No 333/2007. In each analytical run, blank and tested samples in three replicates as well as CRM 189 Wholemeal flour, were carried out.

Risk analysis of lead and cadmium excessive content in wheat grain from various regions of Poland

In order to determine the quality of wheat grain produced in various regions of Poland in terms of Pb and Cd content, and to identify possible regions where excessive contamination of wheat grain with these metals could occur, a risk analysis was performed. Details of the adopted risk levels (high, medium and low) and criteria for risk assessment in relation to the maximum levels of both metals in accordance with EU regulations, are presented in Table 1.

Table 1. The criteria for risk analysis of the tested wheat grain samples

Risk level	Criterion	Level	Content (mg·kg^{-1})	
			Pb	Cd
High	≥10% samples > ML or ≥25% samples > 0.5 ML	ML 0.5 ML	0.20 0.10	0.10 0.05
Medium	≥25% samples > 0.25 ML	0.5 ML	0.10	0.05
Low	≥75% samples < 0.25 ML	0.25 ML	0.05	0.025

Explanations: ML = maximum level.
Source: own study.

Statistical analysis

Statistical analysis and all calculations were performed using TIBCO Software Inc. (2017) and Statistica ver. 13. The mean, standard deviation and median values were calculated for all wheat samples. For the values of lead and cadmium below the LOQ, the half of it (i.e. 0.0005 mg·kg^{-1}) was taken for the calculation. In order to compare the content of toxic metals in wheat grain samples from different regions of Poland, the non-parametric Kruskall–Wallis analysis of variance by ranks and the two-way multiple comparison test were performed. For results comparison between two years of harvest and geographical location the Mann–Whitney U test was used. The adopted level of significance was a = 0.05.

RESULTS AND DISCUSSION

QUALITY CONTROL OF THE RESULTS

The ranges of the obtained results of lead and cadmium content in the certified reference material used in every analytical run are presented in Table 2. They were in a good agreement with certified values of both metals and showed good recoveries.

Table 2. Lead and cadmium contents in the tested certified reference material (CRM)

CRM 189 Wholemeal flour	Pb	Cd
Certified value (mg·kg^{-1})	0.370 ±0.012	0.0713 ±0.0030
Obtained results (mg·kg^{-1})	0.358–0.380	0.0684–0.0740
Recovery (%)	97–103	96–104

Source: own study.

LEAD AND CADMIUM IN WHEAT GRAIN

The results of the lead and cadmium content in the tested wheat grain samples from all over Poland and the four distinguished regions, covering two consecutive harvest years 2017 and 2018 are presented in Table 3.

Most of the tested samples had a measurable content of lead (89%) and cadmium (100%), i.e. above the limit of quantification (0.001 mg·kg^{-1} for Pb and Cd). However, the maximum levels (ML) for lead and cadmium were not exceeded in any of the 300 wheat grain samples tested. Moreover, the mean values of these metals were also well below the ML values, both in the individual regions and all the country, and also from two years of harvest. Only sporadically high levels of lead, and more often in the case of cadmium, were recorded.

From the 2017 harvest, the highest contents of lead were found in 8 samples from 3 regions of Poland: South-West in 3 samples (0.098, 0.084, 0.078 mg·kg^{-1}), South-East in 3 samples (0.088 mg·kg^{-1} of two samples, 0.078 mg·kg^{-1}), and North-West in 2 samples (0.079, 0.076 mg·kg^{-1}). The exception was the North-East region, where the content of this metal in wheat grain was the lowest with 35% results below the LOQ. From the 2018 harvest, the highest contents of lead were found only in 4 samples from the 2 regions: South-West in 2 samples (0.086, 0.073 mg·kg^{-1}) and North-West in 2 samples (0.082, 0.075 mg·kg^{-1}). Nevertheless, even the highest contents of lead found in the tested wheat grain from the 2017 and 2018 harvests, they were at least two times lower than the ML established for this metal (0.20 mg·kg^{-1}).

With regard to cadmium, high contents of this metal, above 0.050 mg·kg^{-1} (0.50 ML) in wheat grain occurred in all regions and in both harvest years, although more often in the southern regions (21 samples from the 2017 and 12 samples from the 2018 harvest) than in the northern regions (3 samples from 2017 and 5 samples from 2018 harvest). Very high contents of this metal, close to the ML value (0.10 mg·kg^{-1}), were found in the

Table 3. Lead and cadmium contents in wheat samples from the 2017 and 2018 harvest

Region	Year	No.	Content (mg·kg^{-1})					
			Pb			Cd		
			range	mean ±*SD*	median	range	mean ±*SD*	median
North-West	2017	51	<0.001–0.079	0.019 ±0.020	0.012	0.014–0.060	0.029 ±0.010	0.028
	2018	51	<0.001–0.082	0.029 ±0.017	0.026	0.012–0.060	0.029 ±0.011	0.026
North-East	2017	20	<0.001–0.031	0.009 ±0.010	0.007	0.016–0.070	0.032 ±0.014	0.030
	2018	20	<0.001–0.051	0.019 ±0.014	0.016	0.008–0.055	0.027 ±0.013	0.023
South-West	2017	44	<0.001–0.098	0.026 ±0.024	0.019	0.015–0.094	0.038 ±0.017	0.035
	2018	44	0.003–0.086	0.024 ±0.016	0.021	0.014–0.098	0.039 ±0.016	0.038
South-East	2017	35	<0.001–0.088	0.019 ±0.024	0.009	0.013–0.080	0.045 ±0.021	0.039
	2018	35	<0.001–0.057	0.018 ±0.015	0.016	0.006–0.088	0.038 ±0.021	0.032
Whole country	2017	150	<0.001–0.098	0.020 ±0.022	0.013	0.013–0.094	0.036 ±0.017	0.032
	2018	150	<0.001–0.086	0.023 ±0.017	0.022	0.006–0.098	0.034 ±0.016	0.030

Explanation: *SD* = standard deviation.
Source: own study.

South-West region from the 2017 (0.094 mg·kg^{-1}) and from the 2018 harvest (0.098 mg·kg^{-1}, 0.094 mg·kg^{-1}).

In another study of wheat grain from the southern region of Poland conducted by Gruszecka-Kosowska [2020], higher lead and a lower cadmium contents were found. They were respectively with the mean values in parentheses: <limit of detection [LOD] – 6.76 mg·kg^{-1} (1.69 ±0.01 mg·kg^{-1}) and <LOD – 0.043 mg·kg^{-1} (0.017 ±0.04 mg·kg^{-1}). However, very similar results to those presented in this work were reported by Jakobsone *et al.* [2015] in Latvia: 0.001–0.097 mg·kg^{-1} Pb and 0.005–0.041 mg·kg^{-1} Cd, as well as by Brizio *et al.* [2016] in Italy (as median values): 0.020 mg·kg^{-1} Pb and 0.040 mg·kg^{-1} Cd. Also EFSA [2009; 2012a] reported the lead and cadmium contents in wheat grains (1,932 samples for Pb and 9,297 samples for Cd) collected from EU countries in 2003–2007, which were very close to the results obtained in this study. Mean lead values ranged from 0.025 to 0.033 mg·kg^{-1}, and cadmium was 0.030 mg·kg^{-1}. A survey conducted in the Great Britain, including 250 wheat grain samples, showed the concentrations of lead and cadmium on the mean level of 0.025 and 0.063 mg·kg^{-1}, respectively [Adams *et al.* 2001]. Khaneghah *et al.* [2020] analysed world data published in 2013–2019, concerning e.g. lead and cadmium content in wheat grain. They included the results of 557 wheat samples from 17 published papers. The ranges of the lead and cadmium contents were respectively: 0.014–1.850 mg·kg^{-1} and 0.010–2.040 mg·kg^{-1}, with the highest contents far exceeding ML values. Very high contents of lead and cadmium which also exceeded ML values were reported from Iran (as a range and mean value in parenthesis): 0.54–4.89 mg·kg^{-1} (1.85 mg·kg^{-1}) of Pb; 0.038–1.99 mg·kg^{-1} (0.41 mg·kg^{-1}) of Cd [Pirsaheb *et al.* 2016]. However, in another study in Iran, such high concentrations of lead and cadmium in wheat grain were not noticed, and they were respectively: 0.007–0.289 mg·kg^{-1} and 0.002–0.036 mg·kg^{-1} [Ghanati *et al.* 2019]. In a study conducted in rural areas in China, the contents of lead and cadmium in wheat grain were relatively low (the mean value in parentheses): 0.013–0.068 mg·kg^{-1} (0.03 mg·kg^{-1}) and 0.005–0.035 mg·kg^{-1} (0.013 mg·kg^{-1}), respectively [Yang *et al.* 2019]. In Bangladesh, the contents of lead and cadmium in wheat grain were determined at the following levels: 0.221 ±0.01 mg·kg^{-1} and 0.011 ±0.008 mg·kg^{-1}, respectively [Ahmed *et al.* 2015]. In general, the variability of the results presented in the literature was related to the geographical location and different conditions of wheat cultivation (soil condition, agricultural practices, climate), and environmental pollution caused by human activities (industry, transport) [Adams *et al.* 2001; Bermudez *et al.* 2012; Kabata-Pendias 2011; Khaneghah *et.al.* 2020; Wierzbowska *et al.* 2018; Young *et al.* 2019].

STATISTICAL ANALYSIS

Statistical analysis showed a significantly higher ($p < 0.05$) content of lead in wheat grain from the 2018 harvest compared to 2017 harvest and no correlation for cadmium (Fig. 2). On the other hand, the comparison of the mean lead and cadmium contents in individual regions showed a significant relationship ($p < 0.05$) between them (Fig. 3). With regard to lead, significantly higher levels in wheat grain ($p < 0.05$) were found in the North-West and South-West regions compared to the other regions, and in general in the western compared to the eastern part of the country. No correlation was found between the northern and the southern parts of the country (Fig. 4).

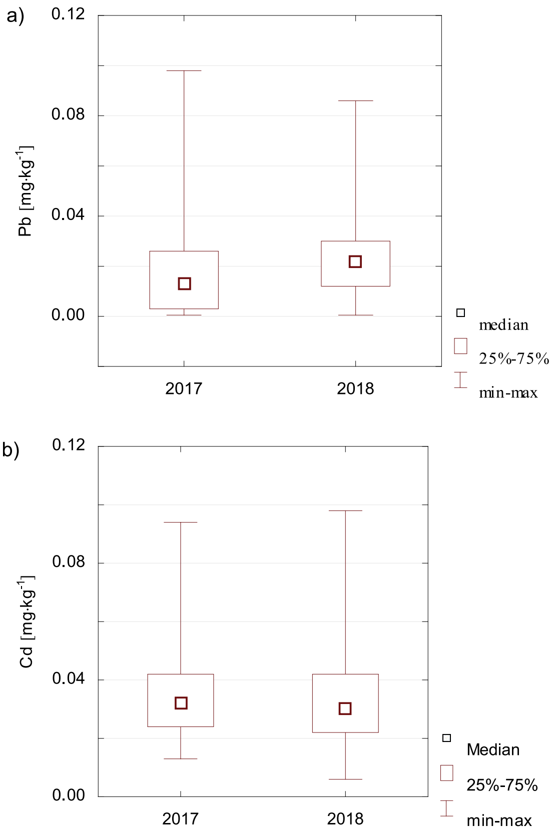

Fig. 2. Content of investigated toxic metals in wheat grain from the 2017 and 2018 years of harvest: a) lead (Pb), b) cadmium (Cd); source: own study

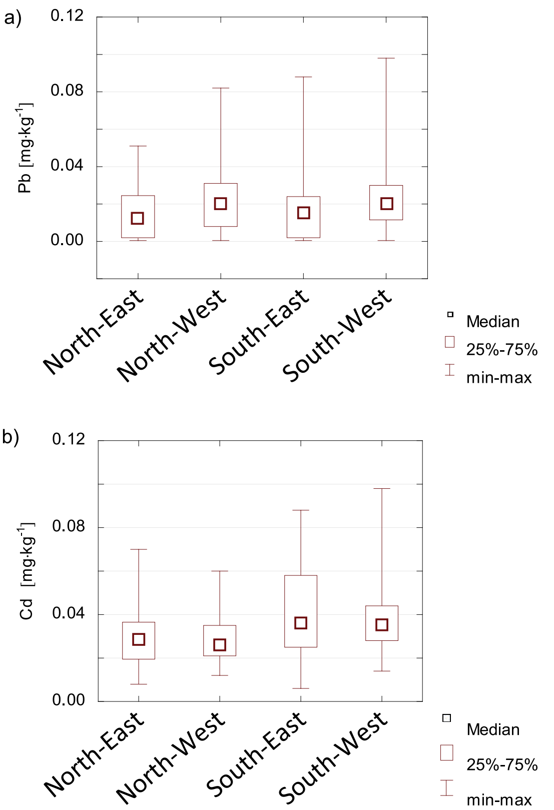

Fig. 3. Content of investigated toxic metals in wheat grain from four individual regions of Poland: a) lead (Pb), b) cadmium (Cd); source: own study

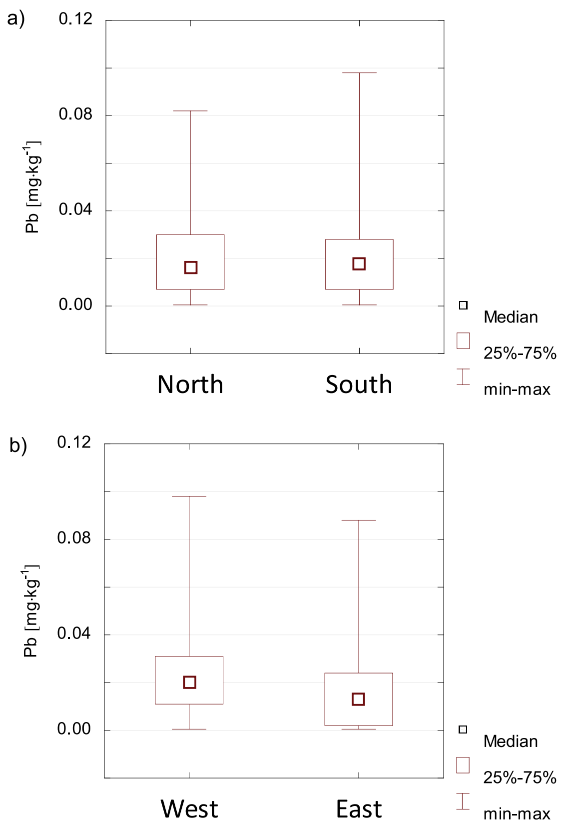

Fig. 4. Lead (Pb) content in wheat grain from: a) the North vs. South regions of Poland, b) from West vs. East regions of Poland (p < 0.05); source: own study

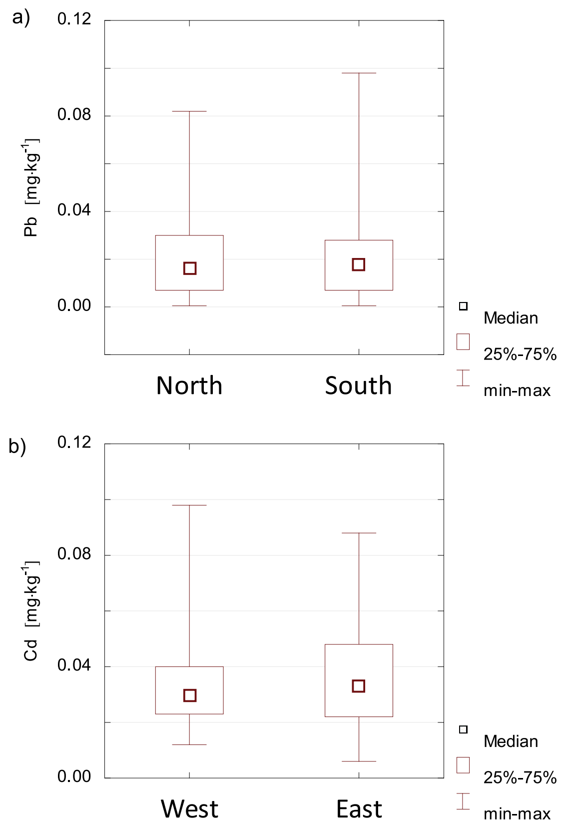

Fig. 5. Cadmium (Cd) content in wheat grain from a) the North vs. South regions of Poland, b) the West vs. East regions of Poland; source: own study

In the case of cadmium, significantly higher ($p < 0.05$) contents of this metal were found in the wheat grain from the South-West and South-East regions compared to the other regions (Fig. 3), and in general in the southern compared to northern part of Poland. There was no correlation between the western and eastern part of the country (Fig. 5).

The statistical relationship between the lead content in wheat grain and the harvest years could be explained by the climatic differences in both harvest years affecting the quality and size of wheat grain yield (drier and warmer 2018 compared to 2017 and water deficit), and the greater industrialisation of the western regions of the country [GUS 2021]. At the same time, no influence of climatic differences in both harvest years on the cadmium content in wheat grain was found. Similar observations on the behaviour of lead and cadmium under the influence of climate changes (temperature, precipitation) were reported by Kirsta *et al.* [2021], in a study conducted over large area and in a changing climate in Russia.

The geographical location of the regions also turned out to be an important factor for the content of both toxic metals in wheat grain. The highest lead contents were found in the western part of the country while cadmium in the southern part of the country. This trend was also confirmed by the research carried out by Gruszecka-Kosowska *et al.* [2019], which showed that cadmium in the soils of the southern Poland occurs in bioavailable forms and therefore it is more easily accumulated in cereals, including wheat. Moreover, the largest amounts of both metals were found in wheat grain from the South-West region. This may be due to greater environmental pollution in this area of the country as a result of the mining and processing of mineral resources [Kabata-Pendias 2011; Gruszecka-Kosowska 2020].

RISK ANALYSIS

The estimated risk level of the possible excessive contents of lead and cadmium in wheat grain from the individual regions and the whole country is presented in Table 4.

Low risk level of excessive lead contamination of wheat grain was found in all four investigated regions and the whole country. In turn, in the case of cadmium, the risk of excessive content of this metal in wheat grain was estimated at a medium level in most regions and years of harvest. Only in wheat grain from the South-East region and 2017 harvest a high risk was found, which was not repeated the following year. This may indicate an incidental phenomenon and the need for a balanced approach to the obtained results. Undoubtedly, however, the largest percentage of wheat grain samples containing cadmium above 0.25 ML (0.025 mg·kg^{-1}) came from the South-West and South-East regions, i.e. 85 and 77%, respectively.

CONCLUSIONS

The results of this study indicated that lead and cadmium contents in wheat grain from all investigated regions of Poland were below the maximum levels specified in the current European regulations. Significantly higher ($p < 0.05$) lead content was found

Table 4. Risk levels of Pb and Cd in wheat grain from different regions of Poland

Region	Year	No. of studied samples	Pb					Cd				
			percentage of $N >$ ML	percentage of $N \in (0.5; 1.0>$ ML	percentage of $N \in (0.25; 0.5>$ ML	percentage of $N >$ 0.25ML	risk level	percentage of $N >$ ML	percentage of $N \in (0.5; 1.0>$ ML	percentage of $N \in (0.25; 0.5>$ ML	percentage of $N >$ 0.25ML	risk level
North-West	2017	51	0	0	12	88	L	0	2	57	43	M
	2018	51	0	0	16	84	L	0	6	59	41	M
North-East	2017	20	0	0	0	100	L	0	10	70	30	M
	2018	20	0	0	5	95	L	0	10	50	50	M
South-West	2017	44	0	0	14	86	L	0	16	82	18	M
	2018	44	0	0	7	93	L	0	9	89	11	M
South-East	2017	35	0	0	11	89	L	0	**40**	86	14	**H**
	2018	35	0	0	6	94	L	0	23	69	31	M
Whole country	2017	150	0	0	13	87	L	0	16	73	27	M
	2018	150	0	0	10	90	L	0	11	69	31	M

Explanations: N = samples number, L = low, M = medium, H = high.
Source: own study.

in wheat grain from the 2018 harvest than from the 2017, and from the western regions compared to the eastern regions of Poland. With regard to cadmium, its contents did not differ significantly in both harvest years, but similarly to lead, it differed between regions i.e. it was significantly higher ($p < 0.05$) in wheat grain from southern regions compared to northern regions of the country. Additionally, in the South-West region of Poland, several samples containing cadmium close to the maximum limit value, were found. Moreover, in this region the contents of both metals were the highest, which may be due to the concentration of industry in this area.

The analysis of the risk of excessive high levels of lead and cadmium in wheat grain showed that in the case of lead, the risk occurred only at a low level in the investigated regions and the whole country, and in both years of harvest. However, for cadmium, a high risk level was estimated in the South-East region and 2017 harvest, what did not happen the next year of harvest. In other regions, the risk of excessive amounts of cadmium in wheat grain from the 2017 harvest and in all regions from the 2018 harvest was at a medium level. This phenomenon may indicate an incidental high degree of wheat grain contamination with cadmium but not as a constant trend. Nevertheless, it may be suggested to put greater emphasis on the quality control of wheat grain produced in Poland in terms of the presence of toxic metals, especially cadmium, mainly when it is intended for human consumption.

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