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# EVALUATION OF THE CONTENT OF SELECTED HEAVY METALS IN PRODUCTS OF PLANT ORIGIN AS A RESULT OF DIFFERENTIATED RATIOS OF FERTILIZERS

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## ABSTRACT

The use of mineral fertilizers helps to obtain higher yields, on the other hand, however, it may lead to the depletion of active forms of nutrients from the soil, which is subsequently conducive to their deficiencies in humans. The content of micronutrients in plants is an important quality characteristic that is evaluated in terms of both human food and animal feeds. Therefore, it becomes essential to determine *the effect of unbalanced mineral fertilization on the micronutrient content in crop plants*. This paper presents the results of an experimental study designed to determine the impact of the absence of N, K, Mg or S *fertilization* and of weather conditions on the iron and manganese content in winter forms of oilseed rape and wheat as well as in spring barley and sugar beet. A four-year, two-factor field experiment was set up in a split-plot design on heavy soil. The content of the micronutrients studied was determined using atomic absorption spectrometry (AAS). Our analysis of the results shows that the experimental factors had a statistically significant effect on the content of the *elements*. The weather conditions caused significant differences in the content of the micronutrients in question, but without a clear direction in these changes. The elimination of nitrogen from a fertilizer dose was associated with a decrease in the iron and manganese content in most of the plants investigated. The absence of individual nutrients in fertilizer doses resulted in significant differences in the iron and manganese content, but these changes were not unambiguous.

**Keywords:** mineral fertilization, iron, manganese, winter rape, winter wheat, spring barley, sugar beets.

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## INTRODUCTION

Heavy metals are considered to be environmental pollutants and their harmfulness is an ever greater problem from the ecological, nutritional, evolutionary and environmental perspective. Out of the ninety naturally occurring elements, fifty-three are considered to be heavy metals, but only some are biologically important. Several are essential micronutrients, e.g. Cu, Zn, Fe, Mn, Mo, Ni and Co, but in excess they can be harmful to the proper plant growth and development (ROUT, SAHOO 2015). According to CZUBA (2000), the micronutrient content is an important quality characteristic that is evaluated in terms of both human food and animal feeds. In Polish soils, the content of micronutrients essential for the proper plant growth varies substantially. The plant available fractions usually account for 10-25% of their total content, and this percentage is determined by the bedrock, soil pH, and particle-size distribution, i.e. light soils contain small amounts of micronutrients, unlike heavy soils, which are richer in micronutrients. Moreover, the availability of Zn, Fe, Cu, Mn and B in acidic soils is much higher than in alkaline soils (KUCCHARZEWSKI, DĘBOWSKI 2001). Low use of organic fertilizers and intensive agricultural production are factors promoting micronutrient deficiencies (CZUBA 1996).

Micronutrient content is an important quality characteristic that is evaluated in terms of both human food and animal feeds (CZUBA 2000, LASER 2007, WOJTKOWIAK et al. 2014). Iron and manganese belong to the most important micronutrients for plants. Iron is the third most limiting micronutrient for plant growth and metabolism. Iron deficiency is the most frequent nutritional problem in many crops and may result in low yields and low nutritional value of crops (ZUO, ZHANG 2011). Manganese is probably an element that has been most thoroughly studied in terms of diseases and plant resistance to diseases (HECKMAN et al. 2003). For plant nutrition, it is therefore important to evaluate the effect of deficiencies in fertilizer nutrients on the plant micronutrient content. Hence, the aim of the present study was to analyze the effect of unbalanced mineral fertilization on the iron and manganese content in the main and by-product yield of winter forms of oilseed rape and wheat as well as of spring barley and sugar beet.

## MATERIAL AND METHODS

A four-year, two-factor field experiment, conducted at the Experimental Station in Zamość (50°42' N, 23°12' E), underlay this study. The experiment was carried out in a split-plot design. The soil on which this experiment was established belonged to soil category IV – heavy soil (41% of <0.02 mm fraction) and soil quality class II. As regards its agricultural suitability, it was

classified as good wheat soil complex. Before the experiment was established, this soil was characterized by slightly acidic pH (pH = 6.02), exchangeable acidity of 0.9 mmol(+) kg<sup>-1</sup>, 1.3 mg(+) kg of exchangeable aluminum, and hydrolytic acidity of 17.1 mmol(+) kg<sup>-1</sup>.

This field experiment employed crop rotation with four plant species: winter oilseed rape (L.) cv. Kana, winter wheat (L.) cv. Sukces, spring barley (L.) cv. Justina, and sugar beet (L.) cv. Deptus. The experimental factors included unbalanced mineral fertilization applied in five treatments and at 4 levels. The applied doses of individual fertilizer nutrients were adjusted to the nutritional requirements of the plants tested. In the control treatments, N:P:K:Mg:S quantities (kg ha<sup>-1</sup>) were respectively 140:31:133:18:60 for winter rape, 150:17:66:18:24 for winter wheat, 100:17:66:18:24 for spring barley, and 120:26:149:36:48 for sugar beet. In fertilizer treatment II, no nitrogen fertilization was applied, in treatment III – no potassium fertilization, in IV – no manganese fertilization, whereas in V – no sulfur fertilization. Fertilizers were applied following good agronomic practice.

Iron and manganese concentrations in plants were determined using atomic absorption spectrometry (AAS) after mineralization of the plant material in concentrated sulfuric acid(VI) with a 30% addition of hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>).

## RESULTS AND DISCUSSION

The weather conditions prevailing during this field experiment varied significantly. In all years of the experiment, plant growth occurred at a temperature higher than the long-term mean (months April – October).

The average monthly rainfall showed high variation in the months of sowing and during the emergence of winter oilseed rape and wheat. Both record high rainfall (August 2006 – 144.8 mm with a long-term mean of 63.9 mm, September 2007 – 144.2 mm and September 2008 – 100.4 mm with a long-term mean of 59.1 mm) and extremely low rainfall (August 2005 – 52.7 mm, 2007 31.6 mm with a long-term mean of 63.9 mm, September 2005 – 15.8 mm, 2006 – 0.8 mm with a long-term mean of 59.1 mm) were recorded. During the spring barley sowing period, the highest rainfall was recorded in 2008 (71.5 mm) and in 2006 (54.8 mm), confronted with the long-term mean of 44.5 mm, whereas the years 2007 and 2009 were characterized by less than half of that rainfall: 21.7 mm and 15.5 mm, respectively. The highest iron content in the main yield of winter rape and wheat was recorded in the 1<sup>st</sup> year of the experiment, and in the by-product yield of these crops – in the 2<sup>nd</sup> year (Tables 1,2). As far as the oilseed rape seed and straw as well as winter wheat straw yields are concerned, the 4<sup>th</sup> year of the experiment was characterized by the lowest iron content in these plant organs. In the main yield of winter forms of oilseed rape and wheat, the highest manganese

Content of iron and manganese in seeds and straw of winter rape

Specification	Mineral fertilization (A)	Year of experiment (B)									
		1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>	mean	1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>	mean
		iron (mg kg <sup>-1</sup> )					manganese (mg kg <sup>-1</sup> )				
Seeds	NPKMgS	98.00	92.00	85.50	88.50	91.00	32.50	35.25	33.00	39.00	34.94
	PKMgS	105.50	90.00	79.00	76.50	87.75	29.50	29.50	28.00	30.50	29.38
	NPMgS	97.00	90.00	87.00	82.00	89.00	33.00	35.75	33.75	37.50	35.00
	NPKS	96.00	87.00	99.75	74.00	89.19	33.25	36.25	36.00	38.00	35.88
	NPKMg	94.00	89.00	84.00	72.50	84.88	32.25	35.25	32.00	37.25	34.19
	mean	98.10	89.60	87.05	78.70	88.36	32.10	34.40	32.55	36.45	33.88
LSD <sub>0.05</sub>		A – n.s. B – 6.289 AxB – 19.70					A – 1.366 B – 1.148 AxB – 3.596				
Straw	NPKMgS	84.50	322.8	61.00	59.50	131.9	19.50	30.00	13.50	20.75	20.94
	PKMgS	67.50	143.5	24.50	51.50	71.75	13.50	16.25	4.250	10.75	11.19
	NPMgS	89.00	265.5	56.50	54.00	116.3	17.00	28.00	13.50	22.00	20.13
	NPKS	62.50	320.3	63.00	54.00	124.9	19.25	25.00	12.75	18.00	18.75
	NPKMg	68.50	154.0	52.00	46.00	80.13	18.00	22.25	13.50	21.50	18.81
	mean	74.40	241.2	51.40	53.00	105.0	17.45	24.30	11.50	18.60	17.96
LSD <sub>0.05</sub>		A – 16.49 B – 13.86 AxB – 43.42					A – 2.292 B – 1.926 AxB – 6.034				

content was found in the 4<sup>th</sup> year, while in the by-product yield falling the lowest in the 2<sup>nd</sup> year. The first year of the experiment was characterized by the lowest manganese content in the main yield of oilseed rape, whereas in the 3<sup>rd</sup> year the lowest manganese content was recorded in rape straw and wheat grain and straw. Regarding spring barley grain (Table 3), a significantly higher iron content was found in the first year of the experiment, while in the second year this content was significantly lower. Straw was characterized by the highest Fe content in the 2<sup>nd</sup> year and the lowest one in the 4<sup>th</sup> year. In the fourth year of the experiment, the manganese content was significantly higher in both grain and straw, while the lowest content of this element was found in grain in the 2<sup>nd</sup> year and in straw in the 3<sup>rd</sup> year. In turn, the first sugar beet root yield was characterized by the lowest iron content (Table 4), while the lowest iron content in leaves was found in the third year. The highest manganese content in both the main yield and by-product yield was recorded in the 2<sup>nd</sup> year, whereas the lowest Mg content occurred in the 1<sup>st</sup> year.

The iron and manganese content (Tables 1 - 4) in the main and by-product yield of the crops studied varied demonstrably depending on the experimental factors and crop species. The concentrations of the micronutrients were within the most frequently reported ranges and below the values con-

Table 2

Content of iron and manganese in grain and straw of winter wheat

Specification	Mineral fertilization (A)	Year of experiment (B)									
		1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>	mean	1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>	mean
		Iron (mg kg <sup>-1</sup> )					manganese (mg kg <sup>-1</sup> )				
Grain	NPKMgS	71.50	38.00	71.75	63.00	61.06	35.50	30.75	29.25	39.50	33.75
	PKMgS	58.50	29.25	60.25	40.25	47.06	29.50	25.50	26.25	31.00	28.06
	NPMgS	64.50	36.00	63.00	52.00	53.88	32.50	30.00	30.00	38.25	32.69
	NPKS	66.00	37.00	63.50	52.50	54.75	36.00	31.50	27.50	40.00	33.75
	NPKMg	86.00	37.50	66.50	56.50	61.63	36.50	31.75	29.75	40.50	34.63
	mean	69.30	35.55	65.00	52.85	55.68	34.00	29.90	28.55	37.85	32.58
LSD <sub>0.05</sub>		A – 2.029 B – 1.705 AxB – n.s.					A – 5.344 B – 4.492 AxB – 14.07				
Straw	NPKMgS	34.50	49.00	31.50	26.00	35.25	29.00	32.50	13.50	25.50	25.13
	PKMgS	24.50	36.50	22.25	18.25	25.38	27.00	27.50	16.00	21.50	23.00
	NPMgS	36.50	50.00	32.50	32.50	37.88	27.50	31.25	19.00	23.50	25.31
	NPKS	36.00	38.00	23.25	31.25	32.13	29.00	37.25	14.25	26.00	26.63
	NPKMg	30.50	49.50	32.00	22.50	33.63	29.50	40.00	16.50	26.50	28.13
	mean	32.40	44.60	28.30	26.10	32.85	28.40	33.70	15.85	24.60	25.64
LSD <sub>0.05</sub>		A – 2.576 B – 2.165 AxB – 13.50					A – 0.020 B – 0.017 AxB – 0.052				

sidered to be toxic (PROŚBA-BIAŁCZYK et al. 2000, KULCZYCKI, GROCHOLSKI 2004, BRODOWSKA, KACZOR 2009).

The supply of iron to plants is an important quality trait since it affects its content in animal and human diet (KOZŁOWSKA-STRAWSKA 2009). Calcium, magnesium, phosphorus, zinc and copper may have a limiting effect on the iron uptake by plants. Moreover, the antagonism between iron and manganese ions may influence the plant content of the investigated micronutrients (BŁAZIAK et al. 1996). In winter wheat straw, spring barley grain and straw as well as in sugar beet roots, the highest iron content was found in the treatments without potassium fertilization. In the cereal plants and in winter rape straw, its lowest content was found in most treatments without nitrogen fertilization. This can be explained by the fact that nitrogen was supplied as ammonium nitrate(V), and the absorption of  $\text{NH}_4^+$  ions is accompanied by release of  $\text{H}^+$  (hydrogen ions) into soil. Moreover, ammonium cations can be nitrified in the soil, which additionally increases the amount of acidic  $\text{H}^+$  ions. As a result of such transformations, the pH may decrease, which affects the amount of iron taken up by plants (BECKER, ASCH 2005, BRODOWSKA, KACZOR 2009). A change in soil pH towards acidic values may bring about an increase in the content of iron in the form that is directly available to plants (KIM, GUERINOT 2007).

Content of iron and manganese in grain and straw of spring barley

Specification	Mineral fertilization (A)	Year of experiment (B)									
		1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>	mean	1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>	mean
		iron (mg kg <sup>-1</sup> )					manganese (mg kg <sup>-1</sup> )				
Grain	NPKMgS	87.00	34.75	54.00	65.25	60.25	12.00	11.25	13.50	15.50	13.06
	PKMgS	49.50	27.00	39.00	43.50	39.75	9.50	9.00	10.00	11.75	10.06
	NPMgS	105.8	35.50	56.25	72.25	67.45	13.00	11.75	13.00	15.25	13.25
	NPKS	88.50	35.00	54.75	65.75	61.00	13.25	12.00	15.50	16.75	14.38
	NPKMg	81.50	33.00	49.00	63.00	56.63	12.50	12.00	13.00	15.25	13.19
	mean	82.46	33.05	50.60	61.95	57.02	12.05	11.20	13.00	14.90	12.79
LSD <sub>0.05</sub>		A – 7.799 B – 6.555 AxB – 20.54					A – 0.892 B – 0.750 AxB – n.s.				
Straw	NPKMgS	69.25	83.75	46.00	33.75	58.19	11.50	11.50	10.50	13.75	11.81
	PKMgS	60.00	63.00	34.75	22.25	45.00	7.000	6.500	7.000	9.000	7.375
	NPMgS	88.00	84.50	47.25	38.50	64.56	18.00	12.50	12.25	18.00	15.19
	NPKS	72.00	84.25	46.25	36.00	59.63	14.50	11.75	10.50	17.50	13.56
	NPKMg	64.00	64.50	37.00	31.50	49.25	16.00	12.00	10.50	15.00	13.38
	mean	70.65	76.00	42.25	32.40	55.33	13.40	10.85	10.15	14.65	12.26
LSD <sub>0.05</sub>		A – 8.061 B – 6.775 AxB – n.s.					A – 2.230 B – 1.874 AxB – n.s.				

In the present study, in most of the winter oilseed rape treatments analyzed, the omission of individual nutrients from a fertilizer dose resulted in reduced Fe content both in seed and straw of this plant (Table 1). In their experiment, BRODOWSKA and KACZOR (2009) found potassium in the chloride form and nitrogen in the nitrate form to have a stimulating effect on the iron uptake by winter oilseed rape.

In this experiment, the iron content in winter wheat grain did not exceed 100 mg kg<sup>-1</sup>, which is in agreement with the results of KABATA-PENDIAS and PENDIAS (after ŚCIGALSKA et al. 2011), who have found that the iron content in cereals ranges widely but rarely exceeds 100 mg kg<sup>-1</sup>. This study demonstrated a significant decrease in the iron content in winter wheat straw and grain harvested from the nitrogen unfertilized plots as well as in grain obtained from the treatments where potassium, magnesium and, in most years, sulfur had been omitted from the fertilizer dose applied. GONDEK et al. (2010) did not find sulfur fertilization to significantly affect the iron content in winter wheat grain. In the present experiment, an increase in the iron content was only found in straw of winter wheat unfertilized with potassium (Table 2).

In spring barley, the omission of nitrogen and sulfur from the fertilizer dose caused a distinct decrease in the iron content in the main yield and by-product yield, whereas the absence of potassium and magnesium in fertil-

Table 4

Content of iron and manganese in root and leaf of sugar beets

Specification	Mineral fertilization (A)	Year of experiment (B)									
		1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>	mean	1 <sup>st</sup>	2 <sup>nd</sup>	3 <sup>rd</sup>	4 <sup>th</sup>	mean
		iron (mg kg <sup>-1</sup> )					manganese (mg kg <sup>-1</sup> )				
Roots	NPKMgS	16.50	39.00	35.75	45.25	34.13	20.50	62.75	45.00	46.50	43.69
	PKMgS	15.00	31.00	35.00	45.25	31.56	20.25	31.50	35.75	43.25	32.69
	NPMgS	17.00	46.25	38.50	37.25	34.75	19.75	44.50	41.00	39.25	36.13
	NPKS	10.50	30.75	32.00	39.00	28.06	21.00	47.50	39.75	46.00	38.56
	NPKMg	16.75	40.00	36.00	36.50	32.31	19.00	40.25	43.50	48.25	37.75
	mean	15.15	37.40	35.45	40.65	32.16	20.10	45.30	41.00	44.65	37.76
LSD <sub>0.05</sub>		A – 4.141 B – 3.481 AxB – 10.90					A – 8.228 B – 6.915 AxB – n.i.				
Leafs	NPKMgS	155.5	144.0	54.00	71.25	106.2	68.50	246.3	101.0	165.0	145.2
	PKMgS	161.0	226.3	71.50	95.50	138.6	69.00	176.0	61.50	111.0	104.4
	NPMgS	164.5	139.5	61.50	130.5	124.0	65.25	133.0	93.00	66.00	89.31
	NPKS	160.0	186.3	55.50	82.75	121.1	78.00	239.0	91.00	136.5	136.1
	NPKMg	176.5	135.0	84.75	149.5	136.4	70.00	218.5	81.00	103.5	118.3
	mean	163.5	166.2	65.45	105.9	125.3	70.15	202.6	85.50	116.4	118.7
LSD <sub>0.05</sub>		A – 12.86 B – 10.81 AxB – 54.00					A – 10.88 B – 9.143 AxB – 28.64				

ization had a contrary effect, as the Fe concentration significantly increased (Table 3).

In sugar beet roots, an increase in the Fe content appeared in the treatments without potassium and sulfur in fertilization (in the first three years), while a lower Fe content occurred in the other treatments. In sugar beet leaves, the absence of nitrogen, potassium, magnesium and sulfur in fertilization was associated with a distinct increase in the iron content. Some literature data indicate that sulfur may have an antagonistic effect on iron availability and metabolism in plant organisms (YOUSFI et al. 2007). In this research, a similar relationship was found only in sugar beet, where the absence of sulfur in crop fertilization caused a significant increase in root and leaf iron content (Table 4).

According to KABATA-PENDIAS and PENDIAS (after KANIUCZAK et al. 2009), manganese is poorly accumulated in plants. This is confirmed by the present experiment. In most treatments with the plants tested, the Mn content was several-fold higher than the iron content. Manganese concentration of about 500 mg kg<sup>-1</sup> dry weight is toxic for most plants (KUCARZEWSKI, DEBOWSKI 2001). The content found in the present study was much lower. The literature data indicate that mineral NPK fertilization results in an increased Mn content in plants, which is associated with the use of physiologically acidic

nitrogen and potassium fertilizers (BŁAZIAK et al. 1996). This is confirmed in our study because the lowest manganese content in most treatments was found at the absence of nitrogen in fertilization of the crops.

In this study, winter oilseed rape was characterized by a reduced Mn content in the main and by-product yields at the absence of nitrogen and sulfur in plant fertilization. In their research, JANKOWSKI et al. (2014) found an increase in the manganese content in winter oilseed rape straw as affected by sulfur soil application. The omission of potassium and magnesium from a fertilizer dose, on the other hand, was associated with an increased Mn content in oilseed rape seeds and a decrease in the content of this element in straw (Table 1).

In winter wheat, the omission of nitrogen and potassium from a fertilizer dose (in the first, second and fourth year) caused a decrease in the manganese content in both grain and straw. These results are consistent with the literature data (RABIKOWSKA, PIŚCZ 1996, KANIUCZAK et al. 2009), which show nitrogen to have a stimulating effect on the manganese uptake by winter wheat. In the present experiment, the absence of magnesium and sulfur fertilization caused an opposite relationship, namely the manganese content increased in both generative and vegetative organs (Table 2). GONDEK et al. (2010), in turn, did not find sulfur fertilization to significantly affect the manganese content.

In spring barley, the omission of nitrogen from the fertilizer dose in this study caused a significant decrease in the straw and grain Mn content, whereas the absence of potassium in fertilization was associated with a distinct increase in the content of this micronutrient in barley straw. KANIUCZAK et al. (2009) found that increasing NPK fertilization doses were associated with an increased Mn content in spring barley. In the present experiment, the manganese content in spring barley grain and straw was found to increase in the case of magnesium deficiency in the environment, whereas BEDNAREK and LIPIŃSKI (1996) did not find magnesium to influence the Mn accumulation in this plant. The absence of sulfur in fertilization in this experiment resulted in an increased manganese content in spring barley straw (Table 3).

In this experiment, the highest manganese content in sugar beet was recorded in optimally fertilized treatments. The omission of any of the nutrients resulted in a decrease in the manganese content in sugar beet roots and leaves (Table 4). In a one-year study, PROŚBA-BIAŁCZYK et al. (2000) found an increase in the manganese content in sugar beet roots and leaves effected by increasing nitrogen fertilization doses.



## CONCLUSIONS

1. Statistically significant differences in the iron and manganese content are found depending on the applied level of mineral fertilization and weather conditions.

2. Weather conditions cause significant differences in the content of the micronutrients studied. However, these changes cannot be identified as following an unequivocal direction.

3. The omission of nitrogen from a fertilizer dose results in the reduced iron and manganese content in most of the plants studied.

4. The absence of potassium in fertilizer doses is associated with a reduced iron content in winter oilseed rape and wheat grain as well as with a reduced manganese content in oilseed rape straw and in the main and by-product yield of winter wheat and sugar beet.

5. In most cases, fertilization without magnesium causes a decrease in the iron content in winter forms of oilseed rape and wheat as well as in sugar beet roots. As far as the manganese content is concerned, the omission of magnesium in fertilizer doses is associated with an increase in the amount of manganese in generative and vegetative organs of cereal plants.

6. The omission of sulfur in a fertilizer dose decreases the iron content in winter forms of oilseed rape and wheat as well as in spring barley. The absence of sulfur in fertilization is associated with a decreased manganese content in winter oilseed rape and sugar beet as well as with its increased content in the cereal plants.

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