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## CONTENT OF MINERALS IN SOYBEAN SEEDS AS INFLUENCED BY FARMING SYSTEM, VARIETY AND ROW SPACING\*

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### ABSTRACT

Soybean is an important source of protein and fat in the food and animal feed industries. Moreover, soybean also contains many other compounds, including minerals, which are beneficial for health and reduce the risk of many diseases. This study was conducted at the Czesławice Experimental Farm near Lublin (51°18'23"N, 22°16'02"E) in 2016. The soil on which the experiment was set up belongs to typical Luvisols. It was characterized by high phosphorus and potassium availability as well as very high magnesium availability. The aim of the present study was to determine the effect of cultivation (organic and conventional system) and row spacing (22.5 cm and 35 cm) on the level of minerals in seeds of two soybean varieties: Aldana and Merlin. The study was conducted using a split-plot design, in three replicates. Seeds were sown on May 5, whereas harvest was carried out on September 5, 2016. After seeds were harvested, the content of the following major minerals was determined in them: phosphorus, potassium, magnesium, calcium, molybdenum, manganese, copper and nickel. Variety Aldana seeds were characterized by a significantly higher content of potassium and copper, while in var. Merlin seeds the calcium concentration was higher. The contents of phosphorus, potassium, calcium, copper and nickel were found to be significantly higher in conventional system grown soybean seeds. Seeds of soybean grown at a row spacing of 22.5 cm contained more copper and nickel. Row spacing was not found to significantly affect the macronutrient content in the studied material.

**Keywords:** organic system, conventional system, varieties, row spacing, *Glycine max* L., macronutrients, trace elements.

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## INTRODUCTION

Soybean (*Glycine max* (L.) Merrill) is a leguminous plant that is most frequently grown in the world and it is an important source of protein and fat in the food and animal feed industries (SHARMA et al. 2014). Soybean seed contains from 38 to 42% of protein and from 19 to 22% of fat in dry matter (BELLALOU, GILLEN 2010). Moreover, soybean also contains many other compounds, including minerals, which are beneficial for health and reduce the risk of many diseases (KUMAR et al. 2014). Humans require at least 22 mineral elements for their well-being (WELCH, GRAHAM 2002, WHITE, BROADLEY 2005, GRAHAM et al. 2007). Food and water are primarily a source of minerals for humans and animals, and the content of essential elements in them is largely determined by their concentrations both in soil and in the plant.

The level of minerals in plant raw materials is dependent on environmental and varietal factors, but it is also possible to influence mineral content through agronomic practices (WANG et al. 2008, DEVI et al. 2013). Numerous breeding programs are committed to developing new varieties with improved agronomic characteristics or nutritional profiles. Although the chemical composition of seed material is genetically controlled, it is known to be affected not only by a genotype but also by the geographic location and agronomic practices (WILCOX, SHIBLES 2001). Optimization of agricultural practices with variable environmental factors, in particular temperature, drought and soil conditions, is necessary to maintain high quality of food grade seeds. Many authors have determined the effects of crop rotation on yield, weed and disease control, soil fertility, and soil structure (VARVEL 2000, TEMPERLY, BORGES 2006, KOŁODZIEJCZYK et al. 2017). Studying the effect of row spacing, BOYDAK et al. (2002) and BELLALOU et al. (2015) found row spacing to significantly affect seed nutrient content (including protein and fat content), and the magnitude of this effect depended on cultivar and environmental factors, especially temperature and drought (BELLALOU et al. 2014).

Moreover, it is worth noting that soybean cultivation under the organic system is preferred compared to the conventional system, which uses pesticides, because this will reduce the risk of accumulation of pesticide residues in plants and soil (AZIZ et al. 2011). Among the benefits of organic technologies are higher soil organic matter and nitrogen, lower fossil energy inputs, yields similar to those of conventional systems, and conservation of soil moisture and water resources (PIMENTEL et al. 2005).

This valuable leguminous plant attracts an ever greater interest and therefore research on its agronomy and nutritional value is fully justified. The aim of the present study was to determine the effect of cultivation (organic and conventional system) and row spacing (22.5 cm and 35 cm) on the level of minerals in seeds of two soybean varieties (Aldana and Merlin).

It was hypothesized that agricultural practices (cultivation system, row spacing) and the genetic factor (variety) affect macro- and micronutrient content in soybean seed.

## MATERIAL AND METHODS

The experiment was conducted in 2016 at the Czesławice Experimental Farm (51°18'23"N, 22°16'2"E), belonging to the University of Life Sciences in Lublin. Soybean seeds were the experimental material. The soil on which the experiment was established belongs to the luvisolic order, typical Luvisol type, typical subtype (PWt) (Soil Classification of Poland 2011), whereas according to the IUSS Working Group WRB (2015) it is classified as Haplic Luvisol (LV-ha) with neutral reaction (pH in 1 M KCl = 7.1). The soil was characterized by high availability of phosphorus (129.5 mg P kg<sup>-1</sup> soil) and potassium (177.6 mg K kg<sup>-1</sup> soil) as well as very high magnesium availability (68.6 mg kg<sup>-1</sup> soil). The humus content was 11 g kg<sup>-1</sup> soil. The study was conducted using a split-plot design in three replicates, on plots with an area of 21.6 m<sup>2</sup> (2.7 × 8 m).

The experimental factors analyzed were as follows: cultivation system (organic and conventional), row spacing (22.5 cm and 35 cm), and soybean variety (Aldana and Merlin). Soybean was sown on May 5, at a depth of 3 cm and at a planned density of 100 plants per 1 m<sup>2</sup>. General information on soybean cultivation under the organic and conventional systems is shown in Table 1.

Weather conditions during the growing season are shown in Table 2. The year in which this study was conducted (2016) proved to be warmer and drier compared to the long-term mean (1963-2010). The average air temperature was higher than the long-term mean by 0.9°C, while the total rainfall was lower than the long-term by 8.4 mm. The month in which soybean was sown (May) was warmer by 0.2°C, whereas the total rainfall was lower by 26 mm compared to the long-term mean for this month. The humidity conditions improved in July, when the total rainfall was more than double the long-term mean (the total rainfall was higher by 113 mm than the long-term mean).

Harvest was carried out on September 5 (var. Aldana) and September 14 (var. Merlin).

Soybean seeds ground in a KNIFETEC 1095 laboratory mill (Foss Tecator, Sweden) were used for chemical analysis. To determine dry weight, samples were dried at 105°C to constant weight. In dried meal, the contents of some minerals were determined. The concentration of phosphorus was determined by the Egner-Riehm colorimetric method with ammonium molybdate, at a wavelength of 660 nm, on a Specol 221 apparatus. An Atomic Absorption Spectrometer apparatus (iCE 3000 Series, Thermo Fisher Scientific) was used to determine calcium and potassium by emission flame spectroscopy, whereas magnesium, copper, manganese, molybdenum, nickel were determined by absorption flame spectroscopy. The material for determination of macronutrients was subjected to mineralization in concentrated sulfuric acid (H<sub>2</sub>SO<sub>4</sub>) and perchloric acid (HClO<sub>4</sub>). The material for analyses of the concen-

Table 1

Characterization of soybean cultivation under organic and conventional systems

Previous crop: spring wheat			
Cultivation system		O	C
Tillage			
Autumn tillage operations	skimming	+	+
	harrowing × 2	+	+
	autumn ploughing – 25 cm	+	+
Spring tillage operations	harrowing	+	+
	cultivating	+	+
	harrowing	+	+
	sowing	+	+
	post-sowing harrowing	+	+
	harrowing (3 days after sowing)	+	+
	harrowing (1-leaf stage)	+	-
	harrowing (3-leaf stage)	+	-
	interrow weed control × 2	+	-
Fertilization			
Bio-algeen S90 (1x 2.0 ml per 1.0 l water): sowing; lateral branch development		+	-
NPK (30-40-80) kg ha <sup>-1</sup> ; beginning of flowering: N - 20 kg ha <sup>-1</sup> : before sowing		-	+
Crop protection			
Nitragina (bacteria <i>Bradyrhizobium japonicum</i> ), 300 g ha <sup>-1</sup> : seed dressing		+	+
Vitavax 200 FS (a.i. carboxin, thiuram), 400 ml 100 kg <sup>-1</sup> seed, with water addition (1:1); immediately after sowing: Afalon Dyspersyjny 450 SC (a.i. linuron) + Dual Gold 960 EC (a.i. metolachlor-S), 1 l + 1.8 l·ha <sup>-1</sup>		-	+

O – organic, C – conventional

trations of micronutrients was mineralized in a mixture of nitric acid (HNO<sub>3</sub>) and perchloric acid (HClO<sub>4</sub>). Prior to assays, the content of calcium, potassium and magnesium trials was diluted appropriately. The other mineral compounds were determined in concentrated samples.

The results were statistically analyzed by three-way analysis of variance, in accordance with the experimental design with three replicates. Confidence half-intervals were calculated using the Tukey's test at a significance level of  $P = 0.05$ . Due to the absence of interactions between the factors, this paper presents the main effects for the factors studied. Calculations were made using Statistica 12 software.

Table 2

Distribution of rainfall and temperatures during the 2016 growing season compared to the long-term means (1963-2010) (according the Czeslawice Meteorological Station)

Month	Temperature (°C)		Rainfall (mm)	
	2016	long-term data	2016	long-term data
May	13.8	13.6	37.0	63.5
June	18.0	16.5	51.6	72.7
July	18.7	18.3	193.3	80.0
August	17.7	17.7	38.1	69.5
September	15.1	13.1	16.8	59.5
Mean/sum (May-September)	16.7	15.8	336.8	345.2

## RESULTS AND DISCUSSION

Human activity causes excessive amounts of metals to be released into the environment, which to some extent also results in their incorporation into the food chain. Essential minerals that are of great importance for the proper functioning of the organism include macronutrients such as magnesium, calcium and potassium as well as the following micronutrients: iron, zinc, copper, selenium, cobalt, nickel, vanadium, chromium, manganese and molybdenum (STANIAK 2014). Seeds of leguminous plants are a valuable element of everyday diet due to their high healthfulness. Soybean seeds are a rich source of not only protein and fat, but also minerals.

Existing research on the quality of legume seeds shows large divergence in their mineral content (KOZAK et al. 2008, MACÁK et al 2015, KAHRAMAN 2017), which can be attributed to the following factors: cultivation system, variety or climate and soil conditions. The analysis performed in the present study reveals significant variation in the content of macronutrients, depending on the cultivated variety and cultivation system (Tables 3-4).

Among the soybean varieties studied, var. Aldana seeds were characterized by a significantly higher potassium content (by 10%, i.e. 2.15 g kg<sup>-1</sup>). The variety Merlin seeds contained significantly more calcium (by 12%, i.e. 0.09 g kg<sup>-1</sup>) (Table 3). The average potassium and calcium content in the seeds studied was as follows: 21.5 g kg<sup>-1</sup> for potassium and 0.78 g kg<sup>-1</sup> for calcium. The calcium content of legume seed, an important component of vegan diets, is low and its availability is reduced by oxalate and to a lesser extent phytate (MORAGHAN et al. 2006). KOZAK et al. (2008) report that seeds of soybean grown under the conditions of southwestern Poland contained, respectively, 16 g kg<sup>-1</sup> of potassium and 1.7 g kg<sup>-1</sup> of calcium, which is 26% less potassium and almost twice as much calcium in relation to the results of the present study. Despite the lack of statistically significant differences in the phosphorus and magnesium content in seeds of the soybean varieties analy-

Table 3

The mineral content in soybean seeds, depending on a variety

Element	Variety		LSD <sub>0.05</sub>
	Aldana	Merlin	
(g kg <sup>-1</sup> )			
Phosphorus	10.32	10.37	n.s.
Potassium	22.63	20.48	1.182
Magnesium	2.02	2.09	n.s.
Calcium	0.74	0.83	0.041
(mg kg <sup>-1</sup> )			
Molybdenum	28.73	28.91	n.s.
Manganese	16.30	17.45	n.s.
Copper	5.09	4.27	0.355
Nickel	5.83	6.00	n.s.

n.s. – not significant

Table 4

The mineral content in soybean seeds, depending on a cultivation system

Element	Cultivation system		LSD <sub>0.05</sub>
	O	C	
(g kg <sup>-1</sup> )			
Phosphorus	9.88	10.80	0.591
Potassium	20.78	22.33	1.182
Magnesium	2.03	2.08	n.s.
Calcium	0.74	0.82	0.041
(mg kg <sup>-1</sup> )			
Molybdenum	28.56	29.08	n.s.
Manganese	17.53	16.23	n.s.
Copper	4.30	5.06	0.355
Nickel	5.61	6.22	0.296

n.s. – not significant, O – organic, C – conventional

zed, var. Merlin was characterized by their higher content. These differences ranged from 0.5% for phosphorus to 3% for magnesium. Phosphorus in soybean seeds is stored primarily as phytic acid, which is nutritionally unavailable (WILCOX et al. 2000). However, the adverse properties of phytic acid can be mitigated by using various culinary techniques, such as grinding, thermal treatment or soaking (BIEŻANOWSKA-KOPE et al. 2006). The soybean seeds studied contained on average 10.3 g kg<sup>-1</sup> of phosphorus. This is a significantly higher amount than that reported by KOZAK et al. (2008) who found

5.2 g kg<sup>-1</sup> of phosphorus in var. Aldana seeds, while Nawiko seeds contained 5.7 g kg<sup>-1</sup> of this element. In twelve soybean genotypes, MORAGHAN et al. (2006) found the seed magnesium content to range from 1.67 to 2.23 g kg<sup>-1</sup>, which correlates with the results of the present study (on average 2.05 g kg<sup>-1</sup>).

As far as the macronutrients determined in soybean seeds are concerned, a cultivation system significantly affected phosphorus, potassium and calcium content (Table 4). Seeds of soybean grown under the conventional system contained significantly more of these macronutrients, respectively 9% (0.92 g kg<sup>-1</sup>) more for phosphorus, 7% (1.55 g kg<sup>-1</sup>) for potassium, and 11% (0.08 g kg<sup>-1</sup>) for calcium. In a study by MACÁK et al. (2015), a significantly higher content of potassium and magnesium was detected under the ecological system in comparison to the conventional system. In the soybean seeds analyzed, no statistically significant difference in magnesium concentration was found between the organic and conventional systems, as it was on a similar level, respectively 2.03 and 2.08 g kg<sup>-1</sup>.

Row spacing was not found to have a statistically significant effect on the content of the macronutrients in soybean seed (Table 5). The differences in the seed content of the elements analyzed in soybean grown at different row spacings was from 1% for calcium, 2% for potassium, to 3% for phosphorus and magnesium.

Many metals, such as Cu, Mn, Fe, Zn and Ni, are essential micronutrients, but can become toxic at concentrations higher than the amount required for normal growth. Moreover, manganese (Mn), copper (Cu) and nickel (Ni) are included on a list of representative trace metals whose levels in food samples represent a reliable index of environmental pollution (GHAEDI et al. 2005). The concentration of the metals studied in soybean seeds did not exceed the WHO limits for food (NKANSAH, AMOAKO 2010). In the descending order by quantity, these were as follows: Mo > Mn > Ni > Cu (Table 3).

Table 5

The mineral content in soybean seeds, depending on row spacing

Item	Row spacing (cm)		LSD <sub>0.05</sub>
	22.5	35	
(g kg <sup>-1</sup> )			
Phosphorus	10.18	10.51	n.s.
Potassium	21.78	21.33	n.s.
Magnesium	2.02	2.09	n.s.
Calcium	0.78	0.79	n.s.
(mg kg <sup>-1</sup> )			
Molybdenum	29.0	28.64	n.s.
Manganese	17.33	16.43	n.s.
Copper	4.95	4.41	0.355
Nickel	6.14	5.70	0.296

n.s. – not significant

Molybdenum is an important factor in human diet and its content in food products, including plants, primarily depends on soil properties and plant species. Leguminous plants and leafy vegetables are usually characterized by a high content of this element. According to GAD, KANDIL (2013), the average content of molybdenum in cowpea seed was found to be 0.45-8.24 mg kg<sup>-1</sup>. The soybean seeds tested contained several times more of this element (on average 28.82 mg kg<sup>-1</sup>). Neither of the agronomic factors used caused significant differences in the molybdenum concentration in soybean seeds (Tables 3-5).

AKINYELE, SHOKUNBI (2015) found soybean seeds to be the richest source of Mn among legumes. Generally, the data reported in this study (the average manganese content 16.88 mg kg<sup>-1</sup>) fall within the range found in literature (MOTTAGHIAN 2008, KAHRAMAN 2017). No significant differences in the manganese content attributable to a cultivation system and row spacing were detected between the studied varieties (Tables 3-5).

Copper and nickel are considered to be micronutrients that are harmful only in excess, but in smaller amounts they are necessary for proper functioning of the organism.

The present study on the content of heavy metals in soybean seed found the copper content to vary and to be significantly dependent on the factors studied. The seeds contained on average 4.68 mg kg<sup>-1</sup> of copper, which is from 2- to 5-fold less compared to previous scientific reports (KAHRAMAN 2017, KRESOVIĆ 2017). Such a large discrepancy is probably due to genotype differences as well as climate and soil conditions. In the study discussed, seeds grown in the conventional system contained most copper, almost 18% (0.76 mg kg<sup>-1</sup>) more compared to the organic system. The variety and row spacing also significantly impacted the level of this nutrient. Var. Aldana contained 19% (0.82 mg kg<sup>-1</sup>) more copper than var. Merlin. Soybean grown at a row spacing of 22.5 cm was characterized by a 12% higher amount (0.54 mg kg<sup>-1</sup>) of copper than soybean harvested from the plots with the larger row spacing (35 cm).

Nickel is an element necessary for the proper growth and functioning of plants, but in higher concentrations it exhibits toxic effects (ANTONKIEWICZ et al. 2016). Opinions on the role and importance of nickel for human organism are divided. Some reports show that nickel, similarly to cadmium and lead, belongs to useless elements and moreover some nickel bonds are considered to be carcinogenic (GUO et al. 2016). Other papers indicate the physiological role of nickel, which consists in activation of some enzymes, increased hormonal activity, stabilization of nucleic acid structures and also participation in lipid metabolism (MULROONEY, HAUSINGER 2003). The permissible nickel content in agricultural produce, depending on its intended use, is 10 mg kg<sup>-1</sup> DW in the case of human consumption and animal feed use (KABATA-PENDIAS et al. 1993, ŚCIGALSKA et al. 2011). In the present study, the average nickel content in soybean seed was 5.92 mg kg<sup>-1</sup>. Although a variety



as a factor was not found to have a statistically significant effect on the nickel content, its higher amounts were noted in var. Merlin (by 3%, i.e. 0.17 mg kg<sup>-1</sup>). Significantly more nickel was found in seeds originating from the conventional system compared to the organic system (by 11%, i.e. 0.61 mg kg<sup>-1</sup>). Seeds harvested from plants grown at a row spacing of 22.5 cm contained 8% more nickel, i.e. 0.44 mg kg<sup>-1</sup>, compared to those grown at a spacing of 35 cm.

## CONCLUSIONS

1. Among the varieties compared, var. Aldana seeds were characterized by a significantly higher content of potassium and copper. Var. Merlin seeds contained significantly more calcium.

2. The cultivation of soybean using the conventional system resulted in a significant increase in the content of phosphorus, potassium, calcium, nickel and copper in the seeds.

3. Row spacing did not have a significant effect on the macronutrient content in soybean seed. Seeds of soybean grown at a row spacing of 22.5 cm contained more copper and nickel.

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