

# **EFFECT OF PLANT BIOSTIMULATION WITH PENTAKEEP V FERTILIZER AND NITROGEN FERTILIZATION ON THE CONTENT OF MACRO- AND MICRONUTRIENTS IN SPINACH\***

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## **Abstract**

The aim of the research was to determine the influence of foliar nutrition with Pentakeep V as well as differentiated nitrogen fertilisation on the content of macro- (Ca, K, Mg, Na, P and S) and micronutrients (B, Cu, Fe, Mn and Zn) in spinach leaves. Pentakeep V is a fertilizer containing 5-aminolevulinic acid (5-ALA), which is the direct precursor of chlorophyll in plants. In 2006-2007, a pot experiment with spinach *Spinacia oleracea* L. cv. Spinaker F<sub>1</sub> was carried out. The plants were cultivated in 60 × 40 × 20 cm containers placed in an open field under a shade-providing fabric. Containers were filled with loamy clay soil (35% of sand, 28% silt, 37% clay) with the organic matter content of 2.44% in 2006 and 2.52% in 2007. The experiment design included 2 sub-blocks: with and without foliar nutrition. The plants were sprayed twice with Pentakeep V fertilizer in a dose of 0.02% w/v (16 ml 100 dm<sup>-3</sup> – 3000 dm<sup>3</sup> per 1 ha). In each sub-block, soil fertilization with nitrogen was applied: 1 – control (without N fertilization), 2 – 25 mg N dm<sup>-3</sup> of the soil (50% of N dose), 3 – 50 mg N dm<sup>-3</sup> of the soil (100% of N dose). Nitrogen fertilization was applied in the form of ammonium nitrate prior to seed sowing. Among all of the determined nutrients, a significant interaction between foliar nutrition and soil application of nitrogen was observed in the case of Ca and Fe content in spinach leaves. Foliar application of Pentakeep V decreased the content of Ca in plants without N fertilization as well as increased the amount of this element in plants fertilized with full dose of nitrogen (100% of N dose). These observations were further verified by the changes of Ca content in soil after plant cultivation. Increased uptake of Ca from soil was observed for plants treated with Pentakeep V and fertilized with the full dose of nitrogen. A higher content of this

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\*The research was financed by Cosmo Seiwa Agriculture Co., LTD. Japan.

element in soil was noted after cultivation of plants receiving only Pentakeep V (without N fertilization). Specific interaction of Pentakeep V on the increased content of Fe in spinach fertilized with 50% dose of N was observed. In comparison with the control, N fertilization in both doses (regardless of the foliar nutrition) led to the decrease of the plant content of Ca, Na and Fe as well as resulted in the increased concentration of K in spinach leaves. Plants fertilized with half-dose of N were characterized by lower content of Mn but plants treated with 100 % of nitrogen had higher concentration of this element in comparison to the control. Foliar application of Pentakeep V (considered independently of N fertilization) did not significantly influence the content of these nutrients in spinach leaves. Weather conditions throughout both years of cultivation had no effect on the interaction between foliar nutrition with Pentakeep V and N fertilization on the content of analyzed nutrients.

Key words: biostimulation, 5-aminolevulinic acid, macronutrients, micronutrients, mineral nutrition.

### WPLYW BIOSTYMULACJI ROŚLIN NAWOZEM PENTAKEEP V ORAZ NAWOŻENIA AZOTEM NA ZAWARTOŚĆ MAKRO- I MIKROSKŁADNIKÓW W SZPINAKU

#### Abstrakt

Celem badań było określenie wpływu dokarmiania dolistnego nawozem Pentakeep V oraz zróżnicowanego pod względem dawki nawożenia azotem na zawartość makro- (Ca, K, Mg, Na, P and S) i mikroskładników pokarmowych (B, Cu, Fe, Mn i Zn) w szpinaku. Nawóz Pentakeep V zawiera kwas 5-aminolewulinowy (5-ALA), który w roślinach jest m.in. bezpośrednim prekursorem cząsteczek chlorofilu. W latach 2006-2007 przeprowadzono doświadczenie wazonowe z uprawą szpinaku *Spinacia oleracea* L. Spinaker F<sub>1</sub>. Szpinak uprawiano w pojemnikach ażurowych o wymiarach 60×40×20 cm umieszczonych na terenie otwartym pod cieniówką. Pojemniki wypełniono gliną średnią pylastą (35% piasku, 28% pyłu i 37% ilu) zawierającą 2,44% i 2,52% materii organicznej odpowiednio w 2006 i 2007 roku. Badaniem objęto dwa podbloki z dolistnym i bez dolistnego dokarmiania roślin. Rośliny dokarmiano dolistnie dwukrotnie nawozem Pentakeep V w dawce 0,02% m/o (16 ml 100 dm<sup>-3</sup> – stosując w przeliczeniu 3 000 dm<sup>3</sup> wody na 1 ha. W obrębie podbłoków zastosowano doglebowe nawożenie azotem: 1 – kontrola (nienawożona azotem), 2 – 25 mg N dm<sup>-3</sup> gleby (50% dawki N), 3 – 50 mg N dm<sup>-3</sup> gleby (100% dawki N). Nawożenie azotem zastosowano przedsięwzięcie w formie saletry amonowej. Spośród wszystkich oznaczonych pierwiastków istotny wpływ współdziałania dokarmiania dolistnego z doglebowym nawożeniem azotem stwierdzono jedynie w odniesieniu do zawartości Ca i Fe w szpinaku. W roślinach nienawożonych azotem dokarmianie dolistne Pentakeep V powodowało zmniejszenie zawartości Ca w szpinaku, w roślinach zaś nawożonych 100% dawką N zwiększenie. Wykazane zmiany zawartości Ca w szpinaku pod wpływem Pentakeep V i nawożenia azotem znajdują uzasadnienie w kierunku zmian zawartości tego pierwiastka w glebie wykazanych po zakończonej uprawie. Świadczą one (odpowiednio w przypadku kontroli i nawożenia 100% dawką N) o zmniejszonym i zwiększonym pobieraniu Ca z gleby przez rośliny dokarmiane dolistnie Pentakeep V. Wykazano specyficzne oddziaływanie Pentakeep V na zwiększenie zawartości Fe w roślinach szpinaku nawożonych 50% dawką N. W porównaniu z kontrolą nawożenie dwiema zastosowaniami dawkami azotu (rozpatrywane niezależnie od dokarmiania dolistnego) powodowało zmniejszenie zawartości Ca, Na i Fe, a wzrost zawartości K w szpinaku. Nawożenie 50% dawką N powodowało zmniejszenie zawartości Mn w szpinaku, a 100% dawką N zwiększenie. Zabieg dokarmiania dolistnego Pentakeep V, rozpatrywany niezależnie od nawożenia azotem, nie powodował istotnych zmian w zawartości badanych pierwiastków w szpinaku. Przebieg warunków klimatycznych w obydwu latach

badań nie miał istotnego wpływu na oddziaływanie dokarmiania dolistnego Pentakeep V i nawożenia doglebowego azotem na zawartość badanych pierwiastków w szpinaku.

Słowa kluczowe: biostymulacja, kwas 5-aminolewulinowy, makroskładniki, mikroskładniki, żywienie mineralne.

## INTRODUCTION

Plant biostimulation has recently become an increasingly more common treatment in modern agricultural production, carried out to intensify the quantity and improve the quality of crop yield. This procedure can be performed independently or along with foliar nutrition. Biostimulation is carried out using growth stimulators (bioactivators, biostimulators). According to the Act of 10 July, 2007 on Fertilizers and Fertilization (Journal of Laws, 2007 no 147, item 1033), a growth stimulator is an organic or mineral compound or its mixture, which has positive impact on plants' growth or other metabolic processes in plants, excluding a growth regulator, which is a plant protection product in the sense defined in the provisions on plants protection. One of the compounds used in plant biostimulation is 5-aminolevulinic acid (ALA), a common precursor to tetrapyrrole compounds found in chlorophyll and hemes. ALA is also a natural organic acid presented in all living organisms (TANAKA et al. 2005). HOTTA et al. (1997) suggest that ALA has plant growth regulating properties at low concentrations and may enhance agricultural productivity. Foliar application of this chemical compound leads to an increased content of photosynthetic pigments in leaves and higher photosynthetic activity. YARONSKAYA et al. (2006) found a positive relation between ALA content and carbon dioxide assimilation in barley seedlings. Foliar application of ALA resulted in a higher content of chloroplast pigments as well as increased photosynthetic and antioxidant activity in pakchoi (MEMON et al. 2009). The effect of ALA or fertilizers containing this compound (e.g. Pentakeep<sup>®</sup> fertilizers) on mineral nutrition is still vaguely defined and only few reports concern this question. Diverse effects of foliar application of Pentakeep<sup>®</sup> on the content of N, Cu and Zn in *Phoenix dactylifera* L. palm leaves were described (AWAD 2008). Studies by WATANABE et al. (2000) showed that ALA improves salt tolerance in cotton seedlings through the reduction in sodium uptake. The aim of this research was to determine the influence of foliar nutrition with Pentakeep V and different nitrogen fertilization on the content of macro- (Ca, K, Mg, Na, P and S) and micronutrients (B, Cu, Fe, Mn and Zn) in spinach leaves.

## MATERIAL AND METHODS

Spinach (*Spinacia oleracea* L.) cv. Spinaker F<sub>1</sub> was cultivated in 2006-2007 in open containers 60×40×20 cm in size, placed in an open field under a shade-providing fabric. The containers were filled with silt loam soil (35% sand, 28% silt and 37% clay) with the content of organic matter of 2.44% in 2006 and 2.52% in 2007, and the following concentrations of the available nutrient forms soluble in 0.03 M acetic acid (for 2006 and 2007, respectively): N (NO<sub>3</sub>-N+NH<sub>4</sub>-N) – 16.6-86.3 mg, P – 16.6-64.8 mg, K – 37.6-53.1 mg, Mg – 121.4-158.3 mg and Ca – 1032.2-2342.9 mg dm<sup>-3</sup> soil. In 2006 and 2007, soil pH<sub>(H<sub>2</sub>O)</sub> was 6.38-6.99, while the total concentration of salt in soil (EC) was 0.19-0.41 EC mS cm<sup>-1</sup>, respectively. The content of available forms of phosphorus and potassium was supplemented before the cultivation to the following levels: 60 mg P (in 2006) and 200 mg K dm<sup>-3</sup> (in 2006 and 2007) of soil.

The research comprised two sub-blocks: with and without plant foliar nutrition. In the sub-block with foliar application, plants were sprayed twice (on 5 and 12 September 2006 as well as on 3 and 14 September 2007) with Pentakeep V in a dose of 0.02% w/v (16 ml 100 dm<sup>-3</sup>). The solution was applied in the amount of 3 000 dm<sup>3</sup> per hectare according to the manufacturer's recommendation (Cosmo Seiwa Agriculture Co., LTD. Japan). The following combinations with soil fertilized with nitrogen were distinguished within the sub-blocks: 1 – control (without N fertilization), 2 – 25 mg N dm<sup>-3</sup> of the soil (50% of N dose: equal 50 kg N ha<sup>-1</sup> in field fertilization), 3 – 50 mg N dm<sup>-3</sup> of the soil (100% of N dose: equal 100 kg N ha<sup>-1</sup> in field fertilization). Nitrogen fertilization was carried out prior to seed sowing using ammonium nitrate. Pentakeep V contains (in gravimetric percent): 9.5% N (3.8% NO<sub>3</sub>-N, 5.7% NH<sub>4</sub>-N), 5.7% MgO, 0.14% B, 0.02% Cu, 0.6% Fe-DTPA, 0.23% Mn, 0.02% Mo, 0.16% Zn and 5-aminolevulinic acid in concentration not declared by the producer.

The experiment was carried out using a split-plot method in four replicates. Each replicate (one container) consisted of 4 rows of plants. In both years of the experiment, seeds were sown on 1 August using 15 seeds in a row. After germination plants were thinned out leaving 10 seedlings in one row (40 plants per one container). Spinach plants were harvested on 19 and 18 September in the subsequent years.

In each year, shredded plant material (spinach leaves) was dried at 70°C, ground and mineralized in 65% super pure HNO<sub>3</sub> (Merck no. 100443.2500) in a CEM MARS-5 Xpress microwave oven (PASŁAWSKI, MIGASZEWSKI 2006). Concentrations of Ca, K, Mg, Na, P, S, B, Cu, Fe, Mn and Zn were determined in the mineralized plant material using the ICP-OES technique with the use of a Prodigy Teledyne Leeman Labs USA spectrometer.

In both years, prior to the experiment, organic matter concentration in soil was determined using Tiurin method modified by Oleksynova. Soil  $\text{pH}_{(\text{H}_2\text{O})}$  was assessed with a potentiometer, while the total concentration of salt in soil EC was measured conductometrically. Prior to the experiment and after the harvest, the content of Ca, K, Mg and P in soil was determined after extraction with 0.03M  $\text{CH}_3\text{COOH}$ . Concentrations of Ca, K and Mg were assessed by the AAS method and P was measured by the vanadium-molybdenum method. It was only after the harvest in 2007 that the concentrations of Na and S (after extraction with 0.03 M  $\text{CH}_3\text{COOH}$ ) as well as B, Cu, Fe, Mn and Zn after extraction with 0.01 M  $\text{CaCl}_2$  in soil were determined by the ICP-OES method.

The results were verified statistically using the ANOVA module of Statistica 8.0 PL programme at the significance level  $P < 0.05$ . The significance of changes was assessed with the use of variance analysis. Whenever significant changes were detected, homogenous groups were determined by Duncan's test.

## RESULTS AND DISCUSSION

The research conducted by AWAD (2008) indicated that during acclimatization of young plants from *in vitro* conditions, soil application of Pentakeep V in concentrations of 0.02%, 0.04% and 0.08% improved growth capacity and increased biomass of *Phoenix dactylifera* L. plants. Higher concentrations of chlorophyll a, accompanied by an increased content of N, Cu and Zn, were observed in palm leaves. In contrast, no significant effect of Pentakeep V on the leaf content of P, K, Fe and Mn was found. It was only in the case of Zn that its higher accumulation in plants correlated with an increasing concentration of this element in soil after Pentakeep V application. As no differences were observed in the soil concentrations of N and Cu, the increase in leaf content of these nutrients was caused exclusively by stimulating the activity of ALA.

Foliar treatment, among other physiological responses, can induce more effective root uptake of mineral nutrients (ADAMEC 2002, BARCZAK et al. 2007). In the present study, the results of statistical analysis indicated significant interaction of foliar nutrition with Pentakeep V and soil fertilization with nitrogen on the Ca and Fe content in spinach leaves (Tables 1 and 2). In both sub-blocks, the concentration of calcium in plants fertilized with 50% dose of N remained at the same level and was lower in comparison to the control plants (Table 1). Foliar nutrition with Pentakeep V caused a significant decrease in the Ca content in control plants without N fertilization, which can indicate a reduced calcium accumulation in plant tissues. This assumption can be supported by the increased amount of this element in

Table 1

The effect of Pentakeep V foliar nutrition and N fertilization on the content of Ca, K, Mg, Na, P and S in spinach

Combinations		(% d.w. – means from 2006-2007)					
Means for interaction: foliar nutrition × nitrogen fertilization		Ca	K	Mg	Na	P	S
Without foliar nutrition	control	1.59 <i>c</i>	6.10 <i>a</i>	1.02 <i>a</i>	0.22 <i>a</i>	0.75 <i>a</i>	0.59 <i>a</i>
	25 mg N dm <sup>-3</sup>	1.29 <i>a</i>	6.70 <i>a</i>	1.05 <i>a</i>	0.17 <i>a</i>	0.73 <i>a</i>	0.63 <i>a</i>
	50 mg N dm <sup>-3</sup>	1.26 <i>a</i>	6.79 <i>a</i>	0.95 <i>a</i>	0.15 <i>a</i>	0.77 <i>a</i>	0.58 <i>a</i>
Pentakeep V	control	1.43 <i>b</i>	6.00 <i>a</i>	0.99 <i>a</i>	0.21 <i>a</i>	0.74 <i>a</i>	0.63 <i>a</i>
	25 mg N dm <sup>-3</sup>	1.29 <i>a</i>	6.62 <i>a</i>	0.99 <i>a</i>	0.17 <i>a</i>	0.72 <i>a</i>	0.62 <i>a</i>
	50 mg N dm <sup>-3</sup>	1.38 <i>b</i>	6.71 <i>a</i>	1.08 <i>a</i>	0.19 <i>a</i>	0.69 <i>a</i>	0.66 <i>a</i>
Means for nitrogen fertilization							
control		1.51 <i>b</i>	6.05 <i>a</i>	1.00 <i>a</i>	0.21 <i>b</i>	0.75 <i>a</i>	0.61 <i>a</i>
25 mg N dm <sup>-3</sup>		1.29 <i>a</i>	6.66 <i>b</i>	1.02 <i>a</i>	0.17 <i>a</i>	0.73 <i>a</i>	0.63 <i>a</i>
50 mg N dm <sup>-3</sup>		1.32 <i>a</i>	6.75 <i>b</i>	1.02 <i>a</i>	0.17 <i>a</i>	0.73 <i>a</i>	0.62 <i>a</i>
Means for foliar nutrition							
without foliar nutrition		1.38 <i>a</i>	6.53 <i>a</i>	1.01 <i>a</i>	0.18 <i>a</i>	0.75 <i>a</i>	0.60 <i>a</i>
Pentakeep V		1.37 <i>a</i>	6.44 <i>a</i>	1.02 <i>a</i>	0.19 <i>a</i>	0.72 <i>a</i>	0.64 <i>a</i>
Test <i>F</i> for interaction: foliar nutrition × nitrogen fertilization × year of study		n.s.	n.s.	n.s.	n.s.	n.s.	n.s.

Means followed by the same letters are not significantly different for  $P < 0.05$ .

Test *F*: n.s. – means are not significant different.

soil from this combination after spinach harvesting (Table 3), particularly in 2006 (Figure 1). Therefore, it is more interesting to have observed how Pentakeep V increased the Ca uptake and Ca accumulation in spinach plants fertilized with the full dose of nitrogen. The results of chemical analysis of soil from this combination showed a lower calcium concentration in comparison to the combination with 50 mg N dm<sup>-3</sup> fertilization and without foliar nutrition. However, this dependence was observed only in 2006 (Figure 1). It should also be mentioned that differences in K concentration noted in soil after spinach cultivation in 2007 (Figure 1) were not correlated with the spinach leaf content of potassium in either year of the experiment (Table 1).

Spinach leaves of plants fertilized with the N dose of 25 mg dm<sup>-3</sup> and not treated with Pentakeep V were characterized by the lowest accumulation of Fe (Table 2). Foliar nutrition with Pentakeep V contributed to a significant increase in the content of this element in plants. In other N combinations, the Fe concentration in leaves was not dependent on foliar

Table 2

The effect of Pentakeep V foliar nutrition and nitrogen fertilization on the content of B, Cu, Fe, Mn, Zn in spinach

Combinations		(% d.w. – means from 2006-2007)				
Means for interaction: foliar nutrition × nitrogen fertilization		B	Cu	Fe	Mn	Zn
Without foliar nutrition	control	30.6 a	11.5 a	468.2 b	224.8 a	195.0 a
	25 mg N dm <sup>-3</sup>	29.0 a	12.7 a	393.3 a	191.0 a	191.8 a
	50 mg N dm <sup>-3</sup>	31.5 a	12.5 a	446.5 b	246.1 a	209.7 a
Pentakeep V	control	29.0 a	12.5 a	453.5 b	202.3 a	191.8 a
	25 mg N dm <sup>-3</sup>	28.7 a	11.8 a	443.0 b	199.5 a	182.5 a
	50 mg N dm <sup>-3</sup>	30.2 a	12.5 a	424.3 ab	234.2 a	203.7 a
Means for nitrogen fertilization						
control		29.8 a	12.0 a	460.9 b	213.5 b	193.4 a
25 mg N dm <sup>-3</sup>		28.8 a	12.3 a	418.2 a	195.3 a	187.2 a
50 mg N dm <sup>-3</sup>		30.9 a	12.5 a	435.4 ab	240.1 c	206.7 b
Means for foliar nutrition						
without foliar nutrition		30.4 a	12.2 a	436.0 a	220.6 a	198.8 a
Pentakeep V		29.3 a	12.3 a	440.3 a	212.0 a	192.7 a
Test <i>F</i> for interaction: foliar nutrition × nitrogen fertilization × year of study		n.s.	n.s.	n.s.	n.s.	n.s.

Means followed by the same letters are not significantly different for  $P < 0.05$ .

Test F: n.s. – means are not significant different.

application, which makes the results obtained for the combination of 25 mg dm<sup>-3</sup> N dose difficult to interpret. The concentration of Fe in soil from all combinations remained at the same level (Table 3). Higher content of this element in plant tissues can result from specific interaction of Pentakeep V with plants nourished with the lower dose of nitrogen. Nitrogen fertilization (regardless of foliar nutrition) decreased the Fe content in spinach leaves, especially in the case of 25 mg dm<sup>-3</sup> N dose (Table 2).

In the present study, no significant effect of the interaction between Pentakeep V foliar nutrition and N fertilization on the content of K, Mg, Na, P, S, B, Cu, Mn and Zn in spinach leaves was observed (Tables 1 and 2). This finding is supported by the results of our previous study on foliar nutrition and N fertilization (SMOLEŇ, SADY, 2009), where only a slight interaction of foliar nutrition (alternately with 2% of urea solution, 1% of Supervit R fertilizer solution and again with 2% urea solution) and N fertilization on the mineral composition of carrot roots was found. Among all of the nutrients taken into consideration in that study, i.e. Al, As, B, Ba, Be, Bi, Ca, Co, Cr, Fe, Ga, In, K, Li, Mg, Mn, Mo, Na, Ni, Pb, Sb, Se, Sr, Ti and V, it

Table 3

The content of Ca, K, Mg, Na, P, S, B, Cu, Fe, Mn and Zn in soil after spinach cultivation – means from 2006-2007

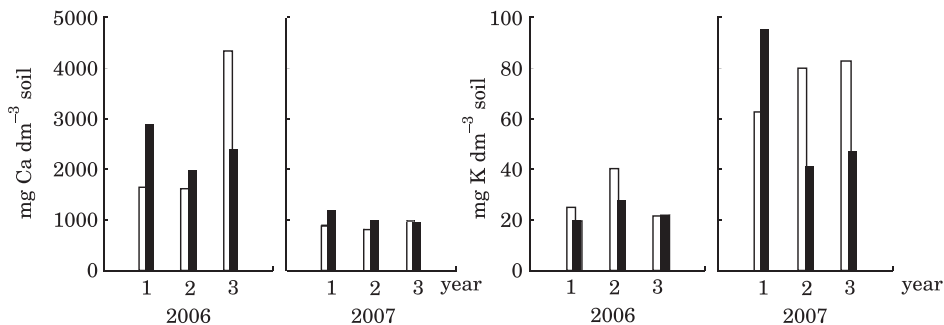
Combinations		(mg dm <sup>-3</sup> soil)						(mg kg <sup>-1</sup> soil)					
Means for interaction: foliar nutrition × nitrogen fertilization		Ca	K	Mg	Na	P	S	B	Cu	Fe	Mn	Zn	
Without foliar nutrition	control	1271.2 a	43.8 ab	83.0 a	6.9 a	38.0 a	4.5 a	<0.0013	2.05 a	277.7 a	170.5 a	56.9 a	
	25 mg N dm <sup>-3</sup>	1218.2 a	60.2 b	84.4 a	6.8 a	44.3 a	4.0 a	<0.0013	2.06 a	264.8 a	160.6 a	55.5 a	
	50 mg N dm <sup>-3</sup>	2667.3 c	52.3 ab	120.6 b	7.6 a	39.8 a	7.8 a	<0.0013	2.31 a	274.6 a	174.0 a	48.4 a	
Pentakeep V	control	2049.8 bc	57.6 b	122.7 b	19.8 a	37.8 a	5.0 a	<0.0013	1.83 a	283.2 a	190.4 a	46.0 a	
	25 mg N dm <sup>-3</sup>	1501.5 ab	34.7 a	90.4 ab	8.6 a	41.0 a	5.3 a	<0.0013	1.87 a	279.7 a	183.9 a	47.3 a	
	50 mg N dm <sup>-3</sup>	1690.0 ab	34.4 a	97.7 ab	7.4 a	36.0 a	9.4 a	<0.0013	2.02 a	279.3 a	177.5 a	50.4 a	
Means for nitrogen fertilization													
control	control	1660.5 a	50.7 a	102.9 a	13.4 a	37.9 a	4.7 a	<0.0013	1.94 a	280.4 a	180.5 a	51.5 a	
	25 mg N dm <sup>-3</sup>	1359.9 a	47.5 a	87.4 a	7.7 a	42.6 a	4.7 a	<0.0013	1.97 a	272.3 a	172.3 a	51.4 a	
	50 mg N dm <sup>-3</sup>	2178.6 b	43.4 a	109.2 a	7.5 a	37.9 a	8.6 b	<0.0013	2.17 a	276.9 a	175.7 a	49.4 a	
Means for foliar nutrition													
without foliar nutrition	control	1718.9 a	52.1 a	96.0 a	7.1 a	40.7 a	5.4 a	<0.0013	2.14 a	272.3 a	168.4 a	53.6 a	
	Pentakeep V	1747.1 a	42.3 a	103.6 a	11.9 a	38.3 a	6.6 a	<0.0013	1.91 a	280.8 a	183.9 b	47.9 a	
Test F for interaction: foliar nutrition × nitrogen fertilization × year of study		*	*	n.s.	-	n.s.	-	-	-	-	-	-	

Means followed by the same letters are not significantly different for  $P < 0.05$ .

Test F:- means are significantly different. n.s. - not significant, "-" for Na, S, B, Cu, Fe, Mn and Zn results only for 2007



LSD<sub>foliar nutrition × nitrogen fertilization × year of study</sub> = 1015.3 LSD<sub>foliar nutrition × nitrogen fertilization × year of study</sub> = 32.1



1 – control, 2 – 25 mg dm<sup>-3</sup> dose of N, 3 – 50 mg dm<sup>-3</sup> dose of N.

□ – sub-block without foliar nutrition, ■ – sub-block with Pentakeep V.

Fig 1. Ca and K content in soil after spinach cultivation in 2006 and 2007

was only the concentration of Na in cv. Kazan F<sub>1</sub> carrot that was affected by the interaction of the tested factors.

The effect of N fertilization on mineral nutrition depends on the form of applied N, its dose as well as the cultivated species (JURKOWSKA et al. 1981, SORENSEN 1999). JURKOWSKA et al. (1981) demonstrated that increasing N fertilization led to a higher content of N, S, Ca, Na and Mg as well as a decreased concentration of P, Cl and K in oat and sorrel plants. In a study conducted by SORENSEN (1999), higher doses of nitrogen nutrition resulted in lower (P and K) or higher (Na) accumulation of macronutrients in cabbage and carrot. In the present research, nitrogen fertilization (in both doses) contributed to the reduction in the leaf content of Ca, Na and Fe (Tables 1 and 2) as well as increased plant concentration of potassium. Variable interaction of N nutrition with Mn accumulation was observed as the application of 25 mg N dm<sup>-3</sup> decreased and that of 50 mg dm<sup>-3</sup> N dose increased the Mn concentration in leaves. The demonstrated changes in the Na and Mn content in plants did not coincide with the results of soil analysis (Table 3), as the content of these nutrients in soil remained at the same level.

In the research conducted by SMOLEŃ and SADY (2009), foliar nutrition (analyzed independently of nitrogen fertilization) considerably increased Bi and Be concentrations, although it did not affect the content of the other twenty-three nutrients in storage roots. The results from the present study indicate that foliar application of Pentakeep V (regardless of N nutrition) had no significant effect on the content of all the analyzed nutrients in spinach leaves (Tables 1 and 2). Except for manganese, the mean values of concentrations of elements in soil from both sub-blocks (with and without foliar nutrition) remained at the same level (Table 3).

AWAD (2008) informed that the effectiveness of ALA influence on plants is closely related to weather conditions during cultivation. In our study, the total rainfall in 2006 was 2.6-fold lower than in 2007 but its distribution was

much more uniform throughout the growing season (Table 4). The first decade of August 2007 was characterized by relatively small rainfall and nearly twice as many sunshine hours as in 2006. In the first decade of September 2007, the number of sunshine hours was 4.6-fold lower and the rainfall was 18.8-fold higher than in 2006. The average relative humidity of air in the first decade of August 2006 was higher, and in the third decade lower, in comparison to the respective periods in 2007. In both years, the total number of sunshine hours remained at the same level. Despite variable climatic conditions in both years, the content of the analyzed nutrients in spinach leaves was not related to this factor. This observation suggests that there is no significant interaction between foliar nutrition, nitrogen fertilization versus the cultivation years, the third factor included in our statistical analysis.

The influence of Pentakeep V application and nitrogen fertilization on yield, nitrogen metabolism and the content of heavy metals and trace nutrients in spinach plants will be presented in a separate publication.

Table 4

Meteorological data from the spinach cultivation period in 2006 and 2007

Month	Decade	2006				2007			
		average air temperature (°C)	rainfall (mm)	sunshine (h)	air humidity RH (%)	average air temperature (°C)	rainfall (mm)	sunshine (h)	air humidity RH (%)
August	1	18.8	35.1	36.4	83.4	20.9	0.2	71.0	61.0
	2	20.2	10.3	63.1	73.7	20.4	10.4	55.1	78.2
	3	16.4	58.7	55.5	82.1	19.2	14.0	81.0	68.2
September	1	16.8	15.3	68.4	76.5	13.1	288.0	14.6	86.7
	2	16.8	1.0	64.5	76.1	12.1	0.8	56.7	79.9
Total		-	120.4	287.9	-	-	313.4	278.4	-

## CONCLUSIONS

1. Among all of the tested nutrients (Ca, K, Mg, Na, P, S, B, Cu, Fe, Mn and Zn), a significant effect of interaction between foliar nutrition and nitrogen fertilization was found only for the content of Ca and Fe in spinach leaves.

2. Pentakeep V application on plants unfertilized with N resulted in a decrease in the Ca content, although foliar nutrition increased the leaf concentration of this element in plants fertilized with 50 mg N dm<sup>-3</sup>.

3. Foliar nutrition of plants fertilized with the 25 mg N dm<sup>-3</sup> dose led to an increase in the accumulation of Fe in spinach.

4. Nitrogen fertilization at both levels (regardless of foliar nutrition) caused a reduction in the Ca, Na and Fe content as well increased accumulation of K in spinach leaves.

5. Soil application of 25 mg N dm<sup>-3</sup> resulted in a decrease, while the 50 mg dm<sup>-3</sup> N dose increased the Mn concentration in plants.

6. Foliar application of Pentakeep V (regardless of N nutrition) had no significant effect on the content of Ca, K, Mg, Na, P, S, B, Cu, Fe, Mn and Zn in spinach leaves.

7. The weather conditions throughout the cultivation period had no significant impact on the interaction Pentakeep V with nitrogen fertilization on the content of Ca, K, Mg, Na, P, S, B, Cu, Fe, Mn and Zn in spinach plants.

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