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Oczapowskiego Street 8, 10-719 Olsztyn, Poland

phone: +48 89 5233231

jadwiga.wierzbowska@uwm.edu.pl

### **Webmaster**

slawomir.krzebietke@uwm.edu.pl

www.uwm.edu.pl/jelementol

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ORIGINAL PAPERS

# INFLUENCE OF DIFFERENT POTASSIUM FERTILIZATION LEVEL ON NUTRITIONAL STATUS OF WINTER WHEAT AND ON YIELD DURING CRITICAL GROWTH STAGE

**Renata Gaj**

**Chair of Agricultural Chemistry  
Poznań University of Life Sciences**

## Abstract

An optimum plant crop nutrition status during critical stages of growth is one of the most important factors shaping their ability to yield. It has been assumed that plant nutrition during the spring wheat stem elongation stage has a significant influence on the growth and yield of this crop. In Poland, potassium is a critical nutrient for plant growth and yielding. In order to verify this hypothesis, three series of one factorial experiment were conducted in 2003-2005 that involved reduced rates of potassium applied to cv. *Zyta* winter wheat. The following rates of potassium were applied: 0, 25, 50 and 100 kg ha<sup>-1</sup>. The assessment of wheat nutritional status was conducted at the beginning of stem elongation (BBCH30/31) using a German model called PIPPA. It had been hypothesized that this particular stage was decisive for wheat growth and yielding. Wheat plants showed deficiency in Ca, K, P and N irrespective of the applied potassium rate. Calcium and potassium deficiencies were crucial for the final grain yield. However, the relationships that occurred between nitrogen and main nutrients, i.e. pairs of nutrients such as N: P, N: K, N: Ca, showed a much better prognostic value, i.e. the relationship with grain yield, than the levels of nutrients in separation. The nitrogen content in leaves at the beginning of shooting showed its limiting effect on grain yield, provided that Ca was deficient, an event which appeared when N: Ca was wider than 34: 1. The same correlation was noticed for potassium, but there the excess of nitrogen revealed its harmful effect when the N:K ratio was above 1.0.

**Key words:** winter wheat, potassium rate, nutritional status, the beginning of shooting.

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dr hab. Renata Gaj, Chair of Agricultural Chemistry, Poznań University of Life Sciences, Wojska Polskiego str. 71F, 60-625 Poznań, Poland, e-mail: grenata@up.poznan.pl

## WPLYW ZRÓŻNICOWANEGO POZIOMU NAWOŻENIA POTASEM NA STAN ODŻYWIENIA W KRYTYCZNEJ FAZIE WZROSTU I PLONOWANIE PSZENICY OZIMEJ

### Abstrakt

Optymalne odżywienie roślin uprawnych w krytycznych fazach wzrostu jest jednym z najważniejszych czynników realizacji ich potencjału plonotwórczego. Przyjęto założenie, że odżywienie roślin w fazie BBCH31 istotnie wpływa na wzrost i plonowanie pszenicy. W Polsce składnikiem krytycznym dla wzrostu i plonowania roślin uprawnych jest potas. Celem sprawdzenia tej hipotezy w latach 2003-2005 przeprowadzono 3 serie jednoczynnikowych doświadczeń polowych ze zróżnicowanymi dawkami potasu stosowanymi w uprawie pszenicy ozimej odmiany Zyta. Potas aplikowano w dawkach: 0, 25, 50 i 100 kg ha<sup>-1</sup>. Stan odżywienia pszenicy ozimej oceniano na początku strzelania w źdźbło (BBCH31) na podstawie niemieckiego programu PIPPA. Rośliny, niezależnie od zastosowanej dawki potasu, wykazały stan niedożywienia Ca, K, P i N. Niedobory wapnia i potasu okazały się kluczowe dla kształtowania plonu ziarna. Jednakże relacje między zawartościami dla par składników: N : P, N : K oraz N: Ca wykazały znacznie większą wartość prognostyczną, czyli związek z plonem ziarna, niż zawartości składników rozważane oddzielnie. Nadmiar azotu w liściach w fazie początku strzelania w źdźbło wykazał ujemny wpływ na plon ziarna w warunkach niedoboru wapnia, który ujawnił się dopiero dla N:Ca > 34. Tę samą prawidłowość zanotowano dla potasu, lecz nadmiar azotu ujawnił swe hamujące działanie, gdy stosunek N:K przyjął wartości większe od 1,0.

Słowa kluczowe: pszenica ozima, dawki potasu, odżywienie roślin, początek strzelania w źdźbło.

## INTRODUCTION

During the early stage of growth of cereal plants, their nutritional status decides about the future yields. The nutritional status of grain plants in the early stages of their vegetation period enables to predict the yield level. In a given growth phase, plants need proper levels of nutrients. For obtaining the expected grain yield (1), the nutrient content in crops should be higher than critical (2); moreover, nutrients should be available in proper ratios. The proper ion ratios of nutrients in soil and in plants is one of the most important factors determining the metabolism and yield quality or level. Mineral fertilization may be more effective if we have more information about the crop's nutritional status. The agricultural extension service for plant producers relies on critical values proposed in the 1970s by BERGMANN (1986). These values may be less valid for present cultivars and yields. It seems that new critical values enabling prediction of plant nutritional status given in the German model PIPPA are much better (SCHNUG, HANEKLAUS, 2008).

In this paper, the potassium nutritional status of winter wheat was studied in a field experiment in respect of the optimal potassium fertilization. It was assumed that the prognostic PIPPA model may be useful for winter wheat in Polish soil and climatic conditions.



## MATERIAL AND METHODS

Field experiments were conducted within years 2003-2005 in the Brody Agricultural Experimental belonging to the Poznań University of Life Sciences. Winter wheat (*Triticum aestivum* L., Zyta) was the tested plant. The experiment was conducted in one-factor trial in four replications, and the following potassium treatments: 0, 25, 50, 100 kg K ha<sup>-1</sup>. As background was applied the following mineral fertilization: N-180, P-22, Mg-15 kg ha<sup>-1</sup>. The highest potassium dose 100 kg ha<sup>-1</sup> was assumed as optimal for chosen N P Mg doses. The control without potassium (NPMg) and any fertilizers (CA) were also regarded. The field experiment has been started on podsolic soil containing 15 to 20% of clay particles – bonitation class IVa, which was characterized by a light acid reaction, high content of available phosphorus 105 mg P kg<sup>-1</sup> and medium availability of potassium 125 mg K kg<sup>-1</sup> and magnesium 37 mg Mg kg<sup>-1</sup>. In every year the winter wheat was cultivated after winter oilseed rape.

The PIPPA program (Professional Interpretation Program for Plant Analysis) as developed by SCHNUG and HANEKLAUS (2008) in the Institute of Plant Nutrition and Soil Science (FAL) in Braunschweig was used for plant nutritional level evaluation.

As the base of model PIPPA is used the boundary line expressing the dependence between the relative yield and the nutrients content in particular plant organs. The boundary line concept was originated more than 30 years ago (WEBB 1972) however its use is developing slowly. EVANYLO, SUMNER (1987) and SCHNUG, HANEKLAUS (1998) have developed the critical levels of different nutrients in winter wheat and have described them in mathematical way as curve line. The points sprayed behind the line show the effect of factors not connected with the nutrient. The maximal possible yield at given nutrient content is calculated.

The weather conditions in the years of the field experiment were different. The precipitations in 2003 year were especially unadequate; in the months April-July were 20% below average (Table 1). The statistical analysis according to Anova calculation for one factor experiment was used at the  $p < 0.05$ .

## RESULTS AND DISCUSSION

The nutritional status evaluation was done for the beginning of straw shooting stage BBCH30/31 described in the literature as critical stage (BERGMANN 1992, REINER 1992). The increasing potassium dose had real effect on the content of nitrogen, copper and iron in the plant (Table 2). The nitrogen

Table 1

Weather conditions during vegetation of winter wheat

Vegetation season	Months											
	Aug	Sept	Oct	Nov	Dec	Jan	Feb	March	Apr	May	June	July
	Temperature (°C)											
2002/2003	20.9	13.8	7.5	3.7	-3.4	-1.6	-3.2	3.4	8.2	16	19.8	19.6
2003/2004	20.8	14.8	6.1	5.3	2.0	-3.5	2.2	5.1	10.0	13.6	16.3	17.3
2004/2005	19.1	13.7	9.6	4.1	1.9	2.1	-1.5	1.8	8.8	12.8	16.4	19.7
Multi-year	17.4	13.1	8.5	3.4	-0.2	-1.8	-0.6	2.8	7.7	13.1	16.3	17.8
Rainfall (mm)												
2002/2003	129	49	127	69	13	60	74	20	21	20	35	97
2003/2004	9	31	40	28	42	73	32	21	23	44	59	60
2004/2005	57	43	49	69	54	49	66	23	19	86	40	127
Multi-year	62	50.3	41.8	44.7	47.1	36.5	30.6	38.8	38	54.7	65.7	56

Table 2

Content of macro- and micronutrients in winter wheat during BBCH 31 stage versus the level of potassium fertilization (mean for 3 years)

Treatments	Nutrients								
	N	P	K	Mg	Ca	Zn	Mn	Cu	Fe
	(g kg <sup>-1</sup> )					(mg kg <sup>-1</sup> )			
Absolute control	25.3	3.27	21.03	0.99	1.05	21.62	47.30	3.934	129.4
NPMgK0	31.3	3.36	22.80	1.06	1.29	25.69	49.43	4.617	192.7
NPMg 25% K	30.5	3.27	23.10	1.05	1.21	23.48	50.40	3.845	101.3
NPMg 50% K	34.7	3.35	22.59	0.98	1.15	24.08	47.77	3.875	142.9
NPKMg100% K	32.7	3.40	23.54	1.01	1.16	22.48	47.93	4.255	150.9
LSD <sub>0.05</sub>	2.5	ns	ns	ns	ns	ns	ns	0.5091	17.19

content in the plant was increasing only to potassium dose responding to 50% of maximal potassium dose. Every potassium dose caused the decrease of copper and iron content in winter wheat leaves. At different potassium doses the potassium content in wheat leaves was similar ~23 g in kg. The obtained nutrients content in the wheat was much lower than critical levels of BERGMANN (1992) 45 g of potassium, 4-10 g of calcium, 1.2-2.5 g of magnesium, 6 mg of copper in 1 kg of wheat. Some authors as critical potassium content in the wheat regard 20 to 50 g kg<sup>-1</sup> (LEIGH, JONES 1984).

The real correlations are stated for next nutrients pairs: N: K, N: P, N: Ca (Table 3). The mechanisms of these correlations for N: K and N: P described in the literature (MARSCHNER et al. 1996). The uptake and the transport in the plant of nitrate ions depends on potassium supply. The correlation analysis of N: K pair for control object without potassium fertilization

Table 3

Correlation coefficient between nutrients content in leaves during BBCH 31 stage and grain yield of winter wheat ( $n = 60$ )

Variable	Nutrients (g kg <sup>-1</sup> )				
	N	P	K	Ca	Mg
Yield (t ha <sup>-1</sup> )	-0.307*	-0.536*	0.132	-0.576*	0.306*
N	1	0.479*	0.476*	0.827*	-0.109
P	-	1	0.259*	0.748*	-0.264*
K	-	-	1	0.400*	0.360*
Ca	-	-	-	1	-0.165
Mg	-	-	-	-	1

\*correlation significant at  $p < 0.05$

shows the grain yield decrease (Figure 1). Similar trends show other objects. It means the yield decrease may be expected if N: K ratio increases above 1.0. This supports the MARSCHNER et al. (1996) hypothesis that only plants well supplied with potassium may uptake sufficient nitrogen amounts necessary for growth. It means both potassium and nitrogen supply must be adequate simultaneously. The level of potassium fertilization had also real effect on the N: Ca and N: P indexes of grain yield (Table 4). Special attention may be paid to N: Ca pair for 25% potassium dose in comparison to maximum 100% dose. The optimal N: Ca ratio enabling the yield 7.8 t from ha as calculated on regression equation is 34 to 1 (Figure 2). The correlation analysis (Table 4) shows that pairs N: P; N: K; N: Ca are more usefull for predicting the winter wheat grain yielding than relation between each nutrient content and grain yields.

The plant index parts analysis and proper interpretation of the results enable the evaluation of plants nutritional status. In this study plant nutritional status of winter wheat in BBCH31 phase was evaluated with the aid

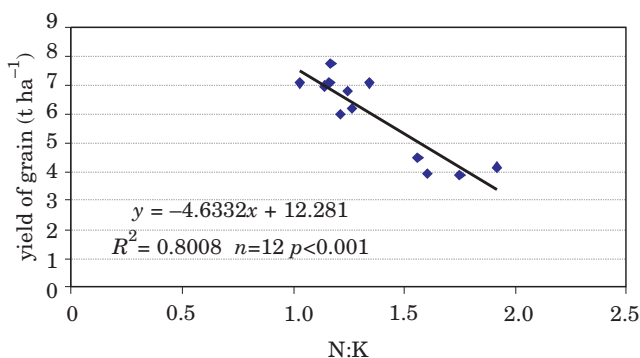


Fig. 1. Correlation between N:K concentration indices in leaves during BBCH 31 stage and grain yield of winter wheat in  $K_0$  treatment

Table 4

Correlation coefficients between nutrient indices during the critical stage BBCH 31 and grain yield of winter wheat ( $n = 12$ )

Treatments	Indices of nutrients		
	N: P	N: K	N: Ca
Absolute control	0.689*	0.612*	-0.773*
NPMg – $K_0$	-0.849*	-0.894*	0.877*
NPMg – 25% K	-0.535	-0.740*	0.713*
NPMg – 50% K	0.642*	-0.746*	0.685*
NPMg – 100% K	0.350	-0.851*	0.735*

\*correlation significant at  $p < 0.05$

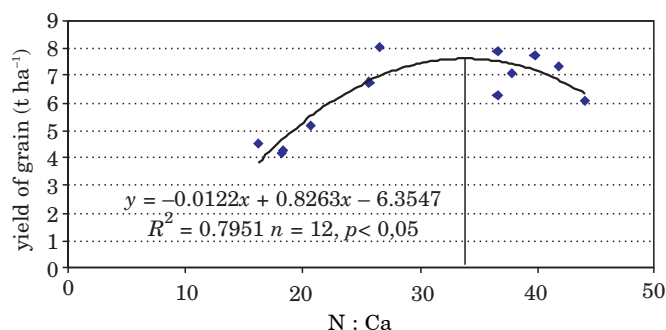


Fig. 2. Correlation between N : Ca indices in leaves during BBCH 31 stage and grain yield of winter wheat in treatment K25%

of PIPPA program. In contrast to evaluation with the aid of critical levels developed by BERGMANN (1992) it was stated that plant yield was restricted by insufficiency of many nutrients (Table 5). It is obvious that winter wheat for high yielding demands adequate supply of all nutrients. In all years of the field experiment the winter wheat yields were restricted by inadequate calcium supply meanwhile the restricting affect of other nutrients (K, N, P, Mg, Zn) was slighter. Thus the PIPPA program enabled to find the nutrients in unadequate level and simultaneously to evaluate their share in restricting the yields expressed in percentage (Figure 3).

Table 5

Rating of nutrients significantly limiting yield of winter wheat

Treatments	Elements									
Absolute control	Ca	Mg	K	N	Zn	P	Cu	Fe	Mn	
K <sub>0</sub> , NPMg	Ca	K	P	N	Mg	Zn	Cu	Fe	Mn	
25%K, NPMg	Ca	K	P	N	Zn	Mg	Cu	Fe	Mn	
50%K, NPMg	Ca	K	Mg	Cu	P	Zn	N	Fe	Mn	
100%K, NPMg	Ca	K	Mg	Zn	P	N	Cu	Fe	Mn	

K<sub>0</sub> – potassium control

— nutrients limiting yield

— nutrients not limiting yield

The reasons of Ca and Mg deficiency in plant may be different and may be caused by Ca Mg deficiency in the soil and by difficulties in uptake of these nutrients by plant and in their transport in the plant during the growth. Both Ca and Mg are uptaken by the youngest root tissue (RUSSELL 1977). Their uptake may be disturbed by toxic level of Alluminium ions. Such explanation may be true as soil pH after winter wheat harvest in all objects was 5 to 6 meanwhile the winter wheat for optimal growth demands neu-

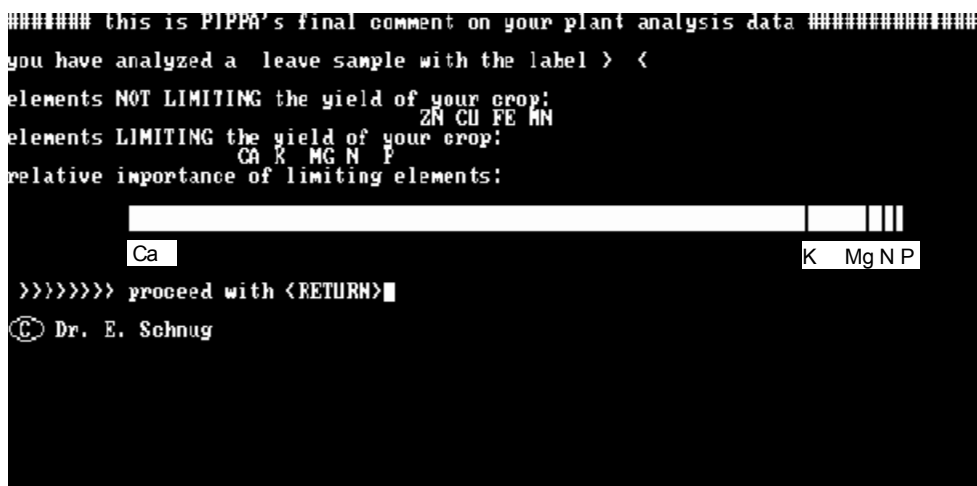


Fig. 3. Structure of share of nutrients in winter wheat at BBCH 31stage, as the index of grain yield limitation

tral soil. The soil acidity (pH below 5; 0.01 mol  $\text{CaCl}_2$  in 1  $\text{dcm}^3$  of soil) is regarded as main factor restricting the yields (GRZEBISZ et al. 2006) and the knowledge about soil pH is very important for proper plant production. Although winter wheat showed potassium deficiency level in BBCH31 phase the influence of potassium doses on wheat yields was insignificant (Figure 4). According to SZCZEPANIAK (2004) the real effect of potassium fertilization on the plant yields may be expected at low K available content in the soil and at water stress during plant growth. The results of MERBACH et al.

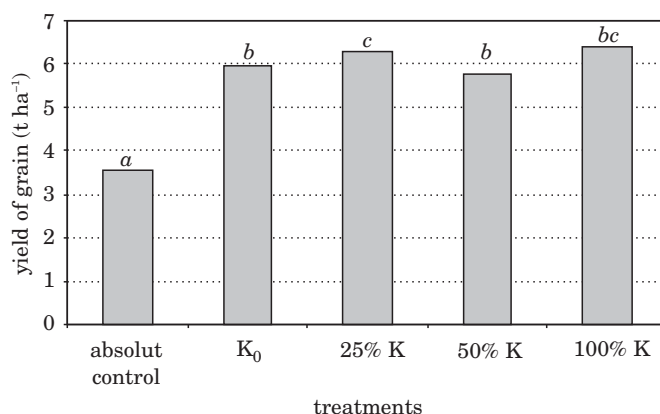


Fig. 4. Effect of the level of potassium application on grain yield of winter wheat,  $\text{t ha}^{-1}$ , *a, b, c* – letters indicate statistical differences between treatments,  $p < 0.05$

(1999) show that the decrease of potassium doses at medium K available level in the soil causes the yield decrease and this decrease is the highest in root plants and the lowest in cereal plants.

## CONCLUSIONS

1. The evaluation of nutritional status of winter wheat in strow shooting stage has shown at first place the insufficient level of calcium in the plant and then potassium and magnesium.

2. Pairs ratio content N: K, N: Ca, N: P in the plant enables better prediction of the yields than the content of each nutrient.

3. The winter wheat yields were restricted by Ca and K supply but calcium deficiency was more important factor.

4. Ca and K was not equilibrated with N. N excess was observed when ratio N to Ca was higher than 34 to 1 and ratio N to K higher than 1.

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# **ZINC AND CADMIUM ACCUMULATION IN MAIZE (*ZEA MAYS* L.) AND THE CONCENTRATION OF MOBILE FORMS OF THESE METALS IN SOIL AFTER APPLICATION OF FARMYARD MANURE AND SEWAGE SLUDGE**

**Krzysztof Gondek**

**Chair of Agricultural and Environmental Chemistry  
University of Agriculture in Krakow**

## **Abstract**

The total content of heavy metals in soil does not indicate bioavailability of these elements after introduction of sewage sludge to soil. Determination of the rate of heavy metal mobilization from sewage sludge after its application to soil is very important for agricultural practice since it allows us to assess the rate at which these elements pass into the soil solution, which conditions their uptake by plants. This research has been conducted to assess the effect of farmyard manure and sewage sludge fertilization on the amounts of zinc and cadmium absorbed by maize and the content of their mobile forms in soil. The research consisted of a three-year pot experiment. Farmyard manure and sewage sludge fertilization resulted in larger total yields of maize biomass (for the 3-year period of the investigations) than harvested from the treatments where only mineral compounds were used as fertilizers. Irrespectively of the fertilizer dose, the total (for 3 years) amounts of zinc and cadmium absorbed by maize were the highest in the treatments where only mineral salts were added and Zn and Cd quantities taken up by maize in the first year of the experiment contributed to this result. Fertilization with farmyard manure and sewage sludge did not cause any significant mobilization of mobile zinc and cadmium forms in soil after the first year of the research. As a result of organic matter mineralization and progressing soil acidification, in the second and third year of the research, the content of mobile forms of the elements in soil increased, although to a lesser degree than under the influence of exclusively mineral treatment.

**Key words:** zinc, cadmium, maize, soil, sewage sludge, farmyard manure.

## **AKUMULACJA CYNKU I KADMU W KUKURYDZY (*ZEA MAYS* L.) ORAZ ZAWARTOŚĆ MOBILNYCH FORM TYCH PIERWIASTKÓW W GLEBIE PO ZASTOSOWANIU OBORNIKA I OSADÓW ŚCIEKOWYCH**

### **Abstrakt**

Kryterium ogólnej zawartości metali ciężkich nie ujmuje ich biologicznej przyswajalności po wprowadzeniu osadu ściekowego do gleby. Określenie tempa uruchamiania metali ciężkich z osadów ściekowych po ich wprowadzeniu do gleby jest bardzo ważne dla praktyki rolniczej, pozawala bowiem ocenić szybkość przechodzenia tych pierwiastków do roztworu glebowego, co warunkuje ich pobranie przez rośliny. Celem badań była ocena wpływu nawożenia obornikiem i osadami ściekowymi na pobranie cynku i kadmu przez kukurydzę oraz zawartość mobilnych form tych pierwiastków w glebie. Badania prowadzono w 3-letnim okresie w warunkach doświadczenia wazonowego. W wyniku nawożenia obornikiem i osadami ściekowymi osiągnięto większe sumaryczne plony biomasy kukurydzy (w 3-letnim okresie badań) w porównaniu z zebranymi w obiektach, w których zastosowano wyłącznie nawożenie mineralne. Sumaryczne (z 3 lat) pobranie cynku i kadmu przez kukurydzę, niezależnie od dawki nawożenia, było największe w obiektach, w których zastosowano wyłącznie sole mineralne, do czego przyczyniły się ilości Zn i Cd pobrane przez kukurydzę w pierwszym roku badań. Nawożenie obornikiem i osadami ściekowymi nie spowodowało istotnego uruchomienia mobilnych form cynku i kadmu w glebie po pierwszym roku badań. W wyniku mineralizacji materiałów organicznych oraz postępującego zakwaszenia gleb w drugim i trzecim roku badań, zawartość mobilnych form badanych pierwiastków w glebie się zwiększyła, chociaż w mniejszym stopniu niż pod wpływem wyłącznego nawożenia mineralnego.

Słowa kluczowe: cynk, kadm, kukurydza, gleba, osad ściekowy, obornik.

## **INTRODUCTION**

A considerable content of fertilizer components and organic substance in sewage sludge justifies their environmental use. The fertilizer value of sewage sludge goes beyond supplying nutrients to plants which ensure their proper growth and development. It also advantageously affects physical, chemical and biological properties of soil (KLASA et al. 2007, GŁAB, GONDEK 2008).

In Poland, the use of municipal sewage sludge in agriculture, beside microbiological concerns, raises questions about heavy metal content. The content of these elements in sewage sludge depends on the sorption capacity of solid particles in sewage from which heavy metals are transferred to sludge (ROSIK-DULEWSKA et al. 2007). The total content of heavy metals in sewage sludge is a primary albeit insufficient criterion in assessment of the usability of sewage sludge in the environment, including agriculture. The total content of heavy metals does not diminish their bioavailability after introducing sewage sludge to soil. Determination of the rate of mobilization of heavy metal from sewage sludge after its application to soil is very important for agricultural practice since it allows us to assess the rate at which they pass into the soil solution, which conditions their uptake by plants (FUENTES et al. 2006, RENOUX et al. 2007).

The aim of this research has been to assess the effect of fertilization with farmyard manure and sewage sludge on amounts of zinc and cadmium absorbed by maize and the abundance of their mobile forms in soil.

## MATERIAL AND METHODS

The effect of fertilization with farmyard manure and sewage sludge on zinc and cadmium amounts absorbed by maize and the content of their mobile forms in soil has been investigated in a three-year pot experiment. The experimental design comprised the following treatments (each in four replications): 0 – without fertilization; NPK-I – mineral treatment; NPK-II – mineral treatment; FYM-I – farmyard manure, FYM-II – farmyard manure; SSA-I – sewage sludge A, SSA-II – sewage sludge; SSB-I – sewage sludge B and SSB-II – sewage sludge B. The treatment symbols marked with I or II indicate the fertilization levels. The research was conducted on soil of the granulometric composition corresponding to sandy silt loam (determined using Casagrande's method modified by Prószyński) collected from the arable layer (0-20 cm) of an arable field in southern Poland (10 km west of Krakow). Before the experiment started, basic physical and chemical properties of the soil material were determined using methods described by OSTROWSKA et al. (1991). The results are presented in Table 1. The experiment was conducted in PVC pots with 5.0 kg of air-dried soil.

Table 1

Some properties of soil before the establishment of the experiment

Determination		Value
pH KCl		5.69
Hydrolitic acidity	(mmol(+) kg <sup>-1</sup> d.m.)	23.4
Total N	(g kg <sup>-1</sup> d.m.)	1.25
Organic C		13.36
Total Zn	(mg kg <sup>-1</sup> d.m.)	78.3
Total Cd		0.65

Sewage sludge originated from two municipal treatment plants (a mechanical and biological system) from Krzeszowice (SSA) and Niepołomice (SSB) situated in the Province of Malopolska. Before collecting samples, sewage sludge was subjected to aerobic stabilization in isolated open tanks, where aeration process went on constantly at ambient temperature. The aeration process lasted for 5 days for sludge from Krzeszowice (SSA) and 8

days for sludge from Niepołomice (SSB). Afterwards, sewage sludge was dewatered on filtration beds. The process lasted for 2 months for SSA sludge and 3 months for SSB sludge. The final stage of the stabilization of sludge from Krzeszowice was hygienization using hydrated calcium. After dewatering, sewage sludge from Niepołomice was removed to a windrow and left.

Farmyard manure used for the experiment originated from pigs fed with commercial concentrates. The experiment made use of farmyard manure after 6-month storage on a manure plate.

In fresh samples of farmyard manure and sludge, pH was measured with a potentiometer. Dry mass content (at 105°C for 12 h) and total nitrogen content after mineralization of samples in concentrated sulphuric acid were determined using Kjeldahl's method. In dried and ground material, organic matter concentration was assessed after calcination of samples in a muffle furnace (at 550°C for 8 hours). The concentrations of phosphorus, potassium, zinc and cadmium were assessed after mineralization of samples in a muffle furnace (at 450°C for 5 h) and the content of ash was measured by dissolving in diluted (1:2) nitric acid (V). The concentration of phosphorus was determined using the vanadium-molybdenum method in a Backman DU 640 spectrophotometer. Potassium was assessed by flame photometry (FES). Zinc and cadmium were assessed using the ICP-AES method in a JY 238 Ultrace apparatus. The determinations were performed using the methods described by BARAN and TURSKEI (1996). Chemical characteristics of farmyard manure and sewage sludge can be found in Table 2.

Table 2

Chemical composition of organic materials and farmyard manure

Determination		FYM	Sewage sludge A (SSA)	Sewage sludge B (SSB)
Dry matter	(g kg <sup>-1</sup> )	189	311	418
pH H <sub>2</sub> O		6.22	6.12	5.73
Total N	(g kg <sup>-1</sup> d.m.)	25.1	17.2	42.4
Total P		22.6	5.48	19.3
Total K		26.7	2.71	2.81
Organic matter	(g kg <sup>-1</sup> d.m.)	679	353	553
Total Zn	(mg kg <sup>-1</sup> d.m.)	531	899	1683
Total Cd		1.28	2.71	2.25

In order to meet the restrictions regulating application of sewage sludge to soil as described in the Regulation of the Minister for the Natural Environment (*Regulation...* 2002), prior to the experiment, soil material was limed with chemically pure CaO. The dose was determined on the basis

of the soil's total hydrolytic acidity. After four weeks, farmyard manure, sewage sludge and mineral salts (NPK) were supplied to soil. Doses of farmyard manure and sewage sludge were calculated on the basis of their nitrogen concentrations. Two levels of nitrogen fertilization were applied. A single dose of nitrogen supplied with fertilizers was  $0.109 \text{ g N kg}^{-1}$  soil d.m. and a double dose was  $0.218 \text{ g N kg}^{-1}$  soil d.m. Phosphorus and potassium were supplemented at an equal level to the amounts introduced with fertilizers in soil of all the treatments (except the control):  $0.114 \text{ g}$  and  $0.228 \text{ g P kg}^{-1}$  soil d.m. as aqueous solution of  $\text{Ca}(\text{H}_2\text{PO}_4)_2 \cdot \text{H}_2\text{O}$  and  $0.134 \text{ g}$  and  $0.269 \text{ g K kg}^{-1}$  soil d.m. in the form of aqueous solution of KCl. In the second and third year of the experiment, supplementary fertilization with chemically pure salt solutions was applied: nitrogen as  $\text{NH}_4\text{NO}_3$ , phosphorus as  $\text{Ca}(\text{H}_2\text{PO}_4)_2 \cdot \text{H}_2\text{O}$  and potassium as KCl. The quantities of the elements supplied in the second and third year of the experiment were identical:  $0.40 \text{ g}$  and  $0.80 \text{ g N}$ ;  $0.10 \text{ g}$  and  $0.20 \text{ g P}$  and  $0.70$  and  $1.40 \text{ g K pot}^{-1}$ , respectively for the single (I) and double (II) dose.

Maize, c.v. San (FAO 240), was grown as 5 plants per pot. Maize (for green fodder) was harvested at the stage of 7-9 leaves. The period of plant vegetation was 47 days in the first, 66 days in the second and 54 days in the third year. During the experiment, the plants were watered with distilled water to 50% of soil maximum water capacity.

After the harvest, the plants were dried (at  $70^\circ\text{C}$ ) to constant weight and dry mass yield of aerial parts and roots was determined. After drying, the biomass was crushed in a laboratory mill and subjected to chemical analysis.

In order to determine the concentrations of zinc and cadmium content, crushed plant material, aerial parts and roots separately, was mineralized in a muffle furnace (at  $450^\circ\text{C}$  for 5 h) and the remains were dissolved in diluted (1:2) nitric acid. The concentrations of Zn and Cd were determined using the ICP-AES method in a JY 238 Ultrac apparatus.

The soil material collected after the growing season from each pot was brought to air-dried state, thoroughly mixed and sifted through a sieve with 1 mm mesh. The soil's pH was measured with a potentiometer in  $1 \text{ mol dm}^{-3}$  KCl suspension (OSTROWSKA et al. 1991) and the total and mobile forms of zinc and cadmium were determined. Concentrations of total forms of these elements were assessed after mineralization of samples in a muffle furnace (at  $550^\circ\text{C}$ , 8 h) and dilution of the remains in a mixture of concentrated nitric and chlorous acids (2:1) (OSTROWSKA et al. 1991). Mobile Zn and Cd forms were extracted from the soil by means of  $1 \text{ mol dm}^{-3}$   $\text{NH}_4\text{NO}_3$  solution (DEL CASTILHO, RIX 1992). In the solutions and filtrates thus obtained, the concentrations of the analysed elements were assessed using the ICP-AES method in a JY 238 Ultrac apparatus.

The analyses of plant and soil material were conducted in four replications, but organic material and initial soil samples were tested in two repli-

cations and a plant reference sample NCS DC73348 (China National Analysis Center for Iron & Steel) or soil reference AG-2 (*AgroMAT*) were added to each series of the analysed material. A result was regarded reliable if the relative standard deviation (RSD) error did not exceed 5%.

The results were verified statistically according to the constant model where the investigated factor was fertilization. One-way ANOVA was conducted and the significance of differences was estimated using Fisher's LSD test at the significance level  $p < 0.01$  (STANISZ 1998).

## RESULTS AND DISCUSSION

Total maize yields (aerial parts and roots) for the three years from the fertilized treatments were significantly larger (by between 45 and 92%) than the amount of biomass harvested from the non-fertilized treatment (Table 3). Farmyard manure and sewage sludge fertilization resulted in larger yields of maize biomass (during the three year period) than mineral fertilizers. Biomass yields, higher than gathered on the treatments where farmyard manure and municipal sewage sludge were used, were produced on the treatment receiving mineral fertilizers only in the first year of the experiment (Table 3). In the subsequent years, maize yielded better in the treatments where farmyard manure or sewage sludge were used, particularly in bigger doses. Such an effect did not occur when plants were fertilized with mineral salts.

In the treatment receiving mineral fertilisers, the highest share of maize biomass in the total yield for three years (50%) was noted in the first year

Table 3

Yields biomass of maize in each year of the experiment (g d.m. pot<sup>-1</sup> ± standard error;  $n = 4$ )

Treatment	1st year	2nd year	3rd year	Total yield
0	55.2 <sup>def</sup> ± 1.7	27.1 <sup>a</sup> ± 0.3	27.4 <sup>a</sup> ± 1.2	109.7 <sup>a</sup> ± 2.5
NPK-I	85.0 <sup>j</sup> ± 2.9	40.6 <sup>bc</sup> ± 1.6	49.6 <sup>cd</sup> ± 0.8	175.2 <sup>c</sup> ± 3.5
NPK-II	76.0 <sup>hij</sup> ± 5.4	31.2 <sup>ab</sup> ± 0.6	52.7 <sup>cde</sup> ± 1.1	159.9 <sup>b</sup> ± 5.6
FYM-I	61.8 <sup>defg</sup> ± 1.8	56.5 <sup>def</sup> ± 1.5	60.8 <sup>defg</sup> ± 1.7	179.1 <sup>c</sup> ± 3.2
FYM-II	57.5 <sup>defg</sup> ± 2.1	74.8 <sup>hij</sup> ± 3.1	64.3 <sup>efgh</sup> ± 3.0	196.6 <sup>d</sup> ± 2.8
SSA-I	57.0 <sup>defg</sup> ± 2.0	55.3 <sup>def</sup> ± 0.5	69.4 <sup>ghi</sup> ± 2.3	181.7 <sup>c</sup> ± 2.9
SSA-II	59.0 <sup>defg</sup> ± 1.1	75.7 <sup>hij</sup> ± 1.4	64.1 <sup>efgh</sup> ± 1.2	198.8 <sup>d</sup> ± 1.9
SSB-I	58.9 <sup>defg</sup> ± 2.9	63.4 <sup>efgh</sup> ± 1.4	52.5 <sup>cde</sup> ± 1.0	174.8 <sup>c</sup> ± 3.9
SSB-II	64.6 <sup>efgh</sup> ± 2.2	78.8 <sup>ij</sup> ± 2.4	67.4 <sup>fghi</sup> ± 1.9	210.8 <sup>e</sup> ± 2.8

Means followed by the same letters in a column did not differ significantly at  $p < 0.01$  according to Fisher's LSD test.

of the investigations, irrespective of the fertilizer dose (Table 4). A reverse tendency was observed for the treatments where farmyard manure and sewage sludge were applied, since a higher proportion of maize biomass in the total (for three years) yield was recorded in the second or third year of the experiment.

Table 4

Contribution (%) of biomass yield of maize in each year of the experiment

Treatment	1 <sup>st</sup> year	2 <sup>nd</sup> year	3 <sup>rd</sup> year
0	50.3	24.8	24.9
NPK-I	48.5	23.2	28.3
NPK-II	47.4	19.6	33.1
FYM-I	34.5	31.5	34.0
FYM-II	29.2	38.0	32.7
SSA-I	31.3	30.5	38.2
SSA-II	29.7	38.1	32.2
SSB-I	33.7	36.3	30.0
SSB-II	30.6	37.4	32.0

The effect of mineral fertilization on shaping the amount of crop yields is more thoroughly documented than the effect of natural or organic fertilizer application, particularly organic waste materials (RAUN, JOHNSON 1999, BLECHARCZYK, MAŁECKA 2005, IDZIAK 2005, STĘPIEŃ et al. 2005). The present experiment demonstrated an advantageous effect of farmyard manure and sewage sludge fertilization on the amount of maize biomass yields at the optimal water access. The investigations conducted by SIENKIEWICZ (2003) showed that maize produces higher yield w receiving farmyard manure fertilization than exclusively mineral fertilization. The result of FYM, according to the author, intensifies itself in the subsequent crop rotations. Positive effect of farmyard manure on crop yields after a prolonged period of its application was observed in other experiments (BLECHARCZYK, MAŁECKA 2005, SCHMIDT et al. 2000). In their research comparing the fertilizer effect of farmyard manure and sewage sludge, KALEMBASA and KUZIEMSKA (1991, 1992) observed that in the first year of sewage sludge application its fertilizer value expressed by maize crop yield was lower than the farmyard manure value; it was approximately identical in the second and third year and only in the fourth year it was better than that of farmyard manure. In the present investigations, the yield forming effects of farmyard manure and sewage sludge were comparable and the differences between the biomass amounts from individual treatments were not significant. A beneficial effect of sewage sludge fertilization on biomass yield was also reported by KRZYWY and WOŁOSZYK (1997) and SKOWROŃSKA et al. (1999, 2001) observed a decrease in maize biomass

after farmyard manure and sewage sludge application in relation to mineral fertilization. The author obtained the lowest maize yield after the application of farmyard manure and sewage sludge in the first year of the experiment, which coincides with the results presented in this work. Most probably, maize response in PATORCZYK-PYTLIK's study, expressed as a decline in biomass yield, was caused by a considerable heavy metal burden in the sewage sludge.

Zinc plays an important role in plants, mainly by activating many enzymes. It should also be emphasized that when municipal sewage sludge is used for fertilization, Zn is supplied to soil in relatively larger amounts than other trace elements. Total amounts of zinc taken up by maize aerial parts and roots were the highest in the first year of the experiment in the treatments where mineral fertilizers were used (Table 5). In the second and third year maize absorbed more zinc in the treatments where farmyard manure

Table 5

Uptake of zinc and cadmium with maize biomass ( $\mu\text{g pot}^{-1} \pm$  standard error;  $n = 4$ )

Treatment	1 <sup>st</sup> year	2 <sup>nd</sup> year	3 <sup>rd</sup> year	Total uptake
	Zn			
0	825 <sup>a</sup> $\pm$ 123	353 <sup>a</sup> $\pm$ 8	426 <sup>a</sup> $\pm$ 18	1604 <sup>a</sup> $\pm$ 120
NPK-I	1745 <sup>b</sup> $\pm$ 84	516 <sup>ab</sup> $\pm$ 15	999 <sup>b</sup> $\pm$ 26	3260 <sup>c</sup> $\pm$ 90
NPK-II	1902 <sup>b</sup> $\pm$ 140	914 <sup>cd</sup> $\pm$ 50	1861 <sup>d</sup> $\pm$ 67	4677 <sup>e</sup> $\pm$ 150
FYM-I	618 <sup>a</sup> $\pm$ 47	551 <sup>ab</sup> $\pm$ 18	1001 <sup>b</sup> $\pm$ 29	2170 <sup>b</sup> $\pm$ 66
FYM-II	657 <sup>a</sup> $\pm$ 19	1122 <sup>d</sup> $\pm$ 97	1450 <sup>c</sup> $\pm$ 68	3229 <sup>c</sup> $\pm$ 105
SSA-I	697 <sup>a</sup> $\pm$ 41	647 <sup>bc</sup> $\pm$ 13	1121 <sup>b</sup> $\pm$ 27	2465 <sup>b</sup> $\pm$ 69
SSA-II	792 <sup>a</sup> $\pm$ 64	1533 <sup>e</sup> $\pm$ 106	1813 <sup>d</sup> $\pm$ 28	4138 <sup>d</sup> $\pm$ 115
SSB-I	668 <sup>a</sup> $\pm$ 22	772 <sup>bc</sup> $\pm$ 38	1066 <sup>b</sup> $\pm$ 43	2506 <sup>b</sup> $\pm$ 42
SSB-II	879 <sup>a</sup> $\pm$ 60	1548 <sup>e</sup> $\pm$ 135	1717 <sup>d</sup> $\pm$ 56	4144 <sup>d</sup> $\pm$ 190
	Cd			
0	6.37 <sup>a</sup> $\pm$ 1.14	3.28 <sup>a</sup> $\pm$ 0.61	1.73 <sup>a</sup> $\pm$ 0.37	11.38 <sup>a</sup> $\pm$ 1.62
NPK-I	15.24 <sup>b</sup> $\pm$ 1.28	3.71 <sup>a</sup> $\pm$ 0.29	10.32 <sup>b</sup> $\pm$ 0.86	29.27 <sup>bc</sup> $\pm$ 0.75
NPK-II	24.23 <sup>c</sup> $\pm$ 3.51	13.78 <sup>cd</sup> $\pm$ 0.37	33.66 <sup>d</sup> $\pm$ 2.07	71.67 <sup>e</sup> $\pm$ 3.82
FYM-I	4.62 <sup>a</sup> $\pm$ 0.29	3.17 <sup>a</sup> $\pm$ 0.32	7.44 <sup>ab</sup> $\pm$ 0.56	15.23 <sup>a</sup> $\pm$ 0.69
FYM-II	4.30 <sup>a</sup> $\pm$ 0.60	11.60 <sup>bc</sup> $\pm$ 1.56	17.44 <sup>c</sup> $\pm$ 2.36	33.34 <sup>cd</sup> $\pm$ 1.71
SSA-I	5.13 <sup>a</sup> $\pm$ 0.35	4.23 <sup>a</sup> $\pm$ 0.47	6.61 <sup>ab</sup> $\pm$ 0.96	15.97 <sup>a</sup> $\pm$ 0.75
SSA-II	6.32 <sup>a</sup> $\pm$ 0.66	14.47 <sup>cd</sup> $\pm$ 2.20	20.68 <sup>c</sup> $\pm$ 2.54	41.47 <sup>d</sup> $\pm$ 4.93
SSB-I	5.33 <sup>a</sup> $\pm$ 0.67	6.48 <sup>ab</sup> $\pm$ 0.56	7.92 <sup>ab</sup> $\pm$ 0.84	19.73 <sup>ab</sup> $\pm$ 1.31
SSB-II	7.34 <sup>a</sup> $\pm$ 1.59	18.46 <sup>d</sup> $\pm$ 2.92	18.96 <sup>c</sup> $\pm$ 2.77	44.76 <sup>d</sup> $\pm$ 6.19

Means followed by the same letters in a column did not differ significantly at  $p < 0.01$  according to Fisher's LSD test.



or municipal sewage sludge were applied, particularly in larger doses. Also WOŁOSZYK et al. (2009) point to higher quantities of zinc taken up by plants fertilized with bigger doses of sewage sludge. On the other hand, BOWSZYS et al. (2009) indicated the form of the applied sewage sludge (dried or granulated sludge) as another factor, apart from a sludge dose, which may condition the content and amounts of the element absorbed by plants.

Cadmium is a crucial element for plant development. Because of its considerable mobility in soil, it is relatively easily absorbed by plants. Maize absorbed the highest quantities of cadmium in the first year in the treatment with a double dose of mineral salts (Table 5). In the second and third year of the experiment, generally greater uptakes of cadmium were found in the treatment where maize was fertilized with double doses of farmyard manure or sewage sludge. Total (for three years) amounts of cadmium taken up by maize, irrespective of the fertilizer dose, were the highest in treatments where exclusively mineral fertilizers were used (Table 5). The results disagree with the research conducted by PATORCZYK-PYTLIK (2001), who revealed higher total amounts of cadmium absorbed by maize in treatments where processed sewage sludge was applied. However, it should be emphasized that the organic materials used by the author had considerably bigger cadmium concentrations. The research conducted by PIOTROWSKA and GAŁCZYŃSKA (1990) indicated that excessive accumulation of cadmium in plants resulted from application of high doses of sewage sludge which were considerably loaded with this element.

Soil pH is one of the most important indicators of soil fertility, most rapidly changing under the influence of fertilization. After three years, a negative effect of particularly bigger dose of mineral fertilizers (Table 6)

Table 6

Soil reaction (pH KCl  $\pm$  standard error;  $n = 4$ ) after harvest of plants

Treatment	1 <sup>st</sup> year	2 <sup>nd</sup> year	3 <sup>rd</sup> year
0	5.90 <sup>defgh</sup> $\pm$ 0.01	5.94 <sup>efgh</sup> $\pm$ 0.02	5.81 <sup>cde</sup> $\pm$ 0.01
NPK-I	5.78 <sup>cd</sup> $\pm$ 0.01	5.69 <sup>bc</sup> $\pm$ 0.09	5.59 <sup>b</sup> $\pm$ 0.01
NPK-II	5.77 <sup>cd</sup> $\pm$ 0.01	5.70 <sup>bc</sup> $\pm$ 0.03	5.42 <sup>a</sup> $\pm$ 0.01
FYM-I	5.98 <sup>gh</sup> $\pm$ < 0.01	5.90 <sup>defgh</sup> $\pm$ 0.03	5.71 <sup>bc</sup> $\pm$ 0.01
FYM-II	6.02 <sup>h</sup> $\pm$ 0.01	5.84 <sup>def</sup> $\pm$ 0.01	5.63 <sup>b</sup> $\pm$ 0.02
SSA-I	5.99 <sup>h</sup> $\pm$ < 0.01	5.92 <sup>efgh</sup> $\pm$ < 0.01	5.69 <sup>bc</sup> $\pm$ 0.01
SSA-II	6.00 <sup>h</sup> $\pm$ 0.01	5.96 <sup>fgh</sup> $\pm$ 0.01	5.69 <sup>bc</sup> $\pm$ 0.01
SSB-I	5.93 <sup>efgh</sup> $\pm$ 0.02	5.92 <sup>efgh</sup> $\pm$ 0.01	5.69 <sup>bc</sup> $\pm$ < 0.01
SSB-II	5.96 <sup>fgh</sup> $\pm$ 0.01	5.86 <sup>defgh</sup> $\pm$ 0.02	5.61 <sup>b</sup> $\pm$ 0.01

Means followed by the same letters in a column did not differ significantly at  $p < 0.01$  according to Fisher's LSD test.

on soil pH was most pronounced. The acidifying effect of mineral fertilizers has been confirmed in papers by STRĄCZYŃSKA (1998) and KOPEĆ (2000). Fertilization with farmyard manure and sewage sludge decreased the soil acidification rate, mainly owing to introducing a considerable amount of alkalizing elements and organic matter. According to JARECKI (1991), treatment with natural or organic fertilizers stabilizes soil reaction. BARAN et al. (1996) also noticed a beneficial effect of fertilization, particularly with bigger doses of sewage sludge, on soil pH.

Application of waste materials as fertilizers rises some doubts, especially due to a threat of natural environment pollution with heavy metals (MOSQUERA-LOSADA et al. 2010). Beside the total heavy metal content in waste materials, also the rate of release of their mobile forms to the soil solution

Table 7

Content of mobile forms of zinc and cadmium in soil ( $\text{mg kg}^{-1} \pm$  standard error;  $n = 4$ ) after harvest of plants

Treatment	1 <sup>st</sup> year	2 <sup>nd</sup> year	3 <sup>rd</sup> year
	Zn		
0	$0.403^{abc} \pm 0.01$	$0.610^{ab} \pm 0.08$	$0.593^a \pm 0.02$
NPK-I	$0.514^e \pm 0.01$	$0.757^{cd} \pm 0.02$	$0.976^d \pm 0.02$
NPK-II	$0.507^{de} \pm 0.02$	$0.824^d \pm 0.03$	$1.288^e \pm 0.02$
FYM-I	$0.356^{ab} \pm 0.01$	$0.547^a \pm 0.02$	$0.708^b \pm 0.02$
FYM-II	$0.349^a \pm 0.02$	$0.664^{abc} \pm 0.01$	$0.867^c \pm 0.02$
SSA-I	$0.433^{bcd} \pm 0.02$	$0.671^{abc} \pm 0.01$	$0.856^c \pm 0.04$
SSA-II	$0.448^{cde} \pm 0.01$	$0.703^{bcd} \pm 0.01$	$0.996^d \pm 0.02$
SSB-I	$0.458^{cde} \pm 0.03$	$0.600^{ab} \pm 0.02$	$0.845^c \pm 0.02$
SSB-II	$0.481^{cde} \pm 0.02$	$0.849^e \pm 0.03$	$1.267^e \pm 0.02$
Treatment	Cd		
0	$0.014^c \pm < 0.001$	$0.014^a \pm < 0.001$	$0.014^a \pm < 0.001$
NPK-I	$0.018^d \pm < 0.001$	$0.019^{bc} \pm < 0.001$	$0.023_f \pm < 0.001$
NPK-II	$0.017^d \pm < 0.001$	$0.023^c \pm < 0.001$	$0.034^g \pm < 0.001$
FYM-I	$0.011^{ab} \pm < 0.001$	$0.013^a \pm < 0.001$	$0.015^{ab} \pm < 0.001$
FYM-II	$0.010^a \pm < 0.001$	$0.013^a \pm < 0.001$	$0.019^{cde} \pm < 0.001$
SSA-I	$0.013^{bc} \pm < 0.001$	$0.014^a \pm < 0.001$	$0.017^{bc} \pm < 0.001$
SSA-II	$0.013^{bc} \pm < 0.001$	$0.013^a \pm < 0.001$	$0.021^{def} \pm < 0.001$
SSB-I	$0.014^c \pm < 0.001$	$0.014^a \pm < 0.001$	$0.019^{bcd} \pm < 0.001$
SSB-II	$0.014^c \pm < 0.001$	$0.015^{ab} \pm < 0.001$	$0.022^{ef} \pm < 0.001$

Means followed by the same letters in a column did not differ significantly at  $p < 0.01$  according to Fisher's LSD test.

and their arresting in soil are vary important. The concentrations of zinc mobile forms in soil were diverse, not only between the applied fertilization treatments but it also between the years of the experiment (Table 7). The lowest zinc concentrations extracted with  $\text{NH}_4\text{NO}_3$ , irrespective of the applied fertilization, were assessed in the soil after the first year of the investigations. After three years, the content of mobile forms of zinc generally increased in soil of all treatments, and the share of mobile zinc forms in the total content also grew, especially after the application of a double dose of the fertilizer (Figure 1). Also BARAN et al. (2001) associated an increase in zinc soil concentrations, including soluble zinc compounds, with doses of sewage sludge, but at the same time pointed to a dependence between the rate of mobilization of this element into the soil solution and the date of sewage sludge application. PATORCZYK-PYTLIK, SPIAK (1996) observed considerable changes in the content of zinc extracted with  $1 \text{ mol dm}^{-3} \text{ HCl}$  solution from soil fertilized with sewage sludge, which they interpreted as conditioned by a considerable amount of this element supplied with sewage sludge. In the author's own investigations, progressing soil acidification has a significant influence on increasing mobile zinc forms.

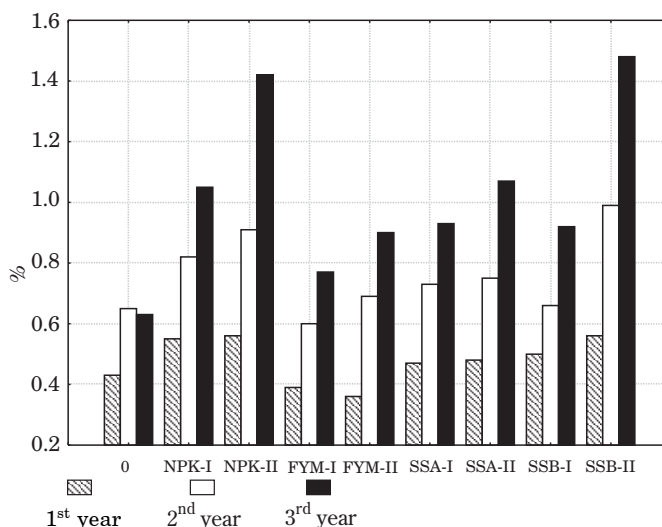


Fig. 1. Contribution (%) of mobile forms in the total content of zinc in soil

Irrespective of the applied fertilization, the greatest quantities of cadmium extracted using  $\text{NH}_4\text{NO}_3$  were found in the soil from the treatment where a double dose of mineral salts was applied (Table 7). Farmyard manure or sewage sludge supplied to soil, irrespective of their dose, did not lead to any marked diversification of the contents of mobile cadmium forms in soil, no matter which dose of the fertilizers was applied. Observations of

the dynamics of concentrations of cadmium mobile forms in soil reveal both greater diversification and higher content of this Cd form after the third year of the experiment. Also the share of mobile cadmium forms in its total content raised visibly (Figure 2).

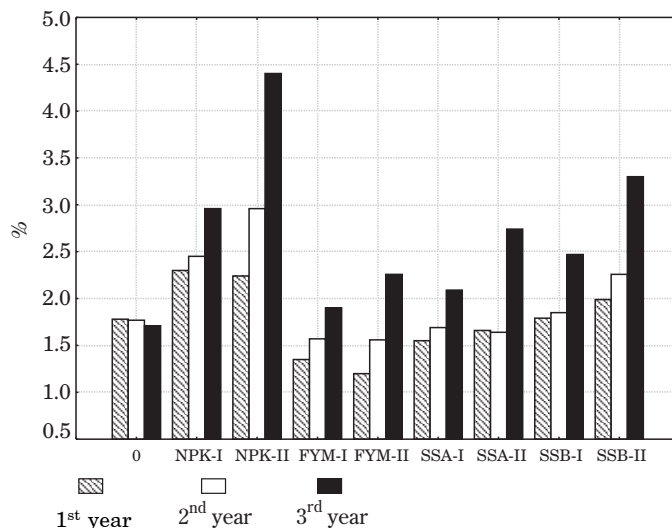


Fig. 2. Contribution (%) of mobile forms in the total content of cadmium in soil

In soil, cadmium belongs to highly mobile elements. Its mobility is conditioned by several factors, including soil pH (BASTA et al. 2005). This research revealed gradual acidification of soil, irrespective of the applied treatment, which favoured increase in mobile forms of this element. Also IŻEWSKA et al. (2006) demonstrated considerably higher concentrations of soluble cadmium forms in soil fertilized with sewage sludge.

## CONCLUSIONS

1. Fertilization with farmyard manure and sewage sludge allowed us to obtain larger yields of maize biomass (for the 3-year period of investigations) than in the treatments where exclusively fertilization with mineral compounds was used.

2. Total (for 3 years) amounts of zinc and cadmium absorbed by maize, irrespective of a fertilization dose, were the highest in the treatments where only mineral salt fertilization was applied, an effect to which quantities of Zn and Cd absorbed by maize in the first year of the experiment greatly contributed.

3. Fertilization with farmyard manure and sewage sludge did not cause any significant mobilization of mobile zinc and cadmium forms in soil after the first year of the experiment. As a result of mineralization of organic materials and progressing soil acidification in the second and third year of the investigations, the content of the mobile forms of the examined elements in soil raised, although to a lesser degree than under the influence of exclusively mineral fertilization.

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# **EFFECT OF SOIL CONTAMINATION WITH ANTHRACENE AND PYRENE ON YIELD AND ACCUMULATION OF MACRONUTRIENTS IN BUTTER LETTUCE (*LACTUCA SATIVA* L.)**

**Sławomir Krzebietke, Stanisław Sienkiewicz**

**Chair of Agricultural Chemistry and Environmental Protection  
University of Warmia and Mazury in Olsztyn**

## **Abstract**

Toxic compounds which belong to PAHs are generated during all types of combustion of fuels and other substances as well as a result of natural processes (mineralisation). Products which appear during the above processes eventually reach soil, where they accumulate. The objective of this study has been to evaluate the effect of anthracene and pyrene accumulated in soil on yield, concentration of macronutrients (N, P, K, Mg, Ca, Na) and their uptake by cv. Vilmorin butter lettuce (*Lactuca sativa* L.) grown under the minimum and 3-fold enriched abundance of substrate. A pot experiment in four replicates was carried out twice in a greenhouse at the University of Warmia and Mazury in Olsztyn, in the spring of 2007 and 2008. Supplementary fertilisation was applied before planting butter lettuce. At the first rate of fertilisation, nitrogen was introduced in a full dose before planting lettuce, but at the triple rate of nutrients in substrate, it was divided into 2/3 of the dose before planting and the remaining 1/3 applied to soil 10 days after planting. Soil contamination with anthracene (ANT) and pyrene (PYR) or their mixture started 10 days after planting lettuce. Soil application of the two PAHs was performed 5 times at five-day intervals until the end of the growing season of lettuce. Determination of macronutrients (N, P, K, Mg, Ca, Na) was accomplished with standard methods after mineralisation ( $\text{H}_2\text{SO}_4 + \text{H}_2\text{O}_2$ ) of the plant material dried at 60°C. The determinations were performed in comparison to certified material (CTA-VTL-2).

Increasing the abundance of substrate in nutrients (N, P, K, Mg, Na, Cl) by three-fold led to a 13.7% increase in yield of lettuce heads. The PAH compounds present in the substrate depressed lettuce yield. This tendency was more evident when anthracene rather than pyrene contaminated soil. ANT and PYR depressed the concentration of nitrogen but raised the concentration of calcium in substrate less abundant in nutrients.

**Key words:** *Lactuca sativa* L., macronutrients, anthracene, pyrene, soil, fertilizer rates.

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dr inż. Sławomir Krzebietke, Chair of Agricultural Chemistry and Environmental Protection, University of Warmia and Mazury in Olsztyn, Oczapowskiego Street 8, 10-719 Olsztyn, Poland, e-mail: slawomir.krzebietke@uwm.edu.pl

## WPLYW SKAŻENIA GLEBY ANTRACENEM I PIRENEM NA PLONOWANIE I KUMULACJĘ MAKROSKŁADNIKÓW W SAŁACIE MASŁOWEJ (*LACTUCA SATIVA* L.)

### Abstrakt

Toksyczne związki z grupy WWA powstają nie tylko w wyniku szeroko pojętych procesów spalania paliw, lecz również wskutek procesów naturalnych (mineralizacji). Produkty powstałe w trakcie tych procesów ostatecznie trafiają do gleby i ulegają kumulacji. Celem badań była ocena wpływu antracenu oraz pirenu zakumulowanych w glebie na plonowanie oraz zawartość makroskładników (N, P, K, Mg, Ca, Na) i ich pobranie przez sałatę masłową (*Lactuca sativa* L.) odmiany Vilmorin uprawianą w warunkach minimalnej i 3-krotnie zwiększonej zasobności podłoża. Doświadczenie wazonowe w 4 powtórzeniach prowadzono 2-krotnie wiosną w hali vegetacyjnej UWM w Olsztynie, w latach 2007-2008. Uzupełniające nawożenie zastosowano przed posadzeniem sałaty masłowej. W przypadku minimalnego poziomu nawożenia azot zastosowano w całości przed sadzeniem, natomiast w przypadku potrojonej ilości składników pokarmowych w podłożu – 2/3 dawki przed posadzeniem rozsady, a 1/3 dawki doglebowo po 10 dniach od posadzenia. Skażenie gleby antracenenem (ANT) oraz pirenem (PYR) i ich mieszaniną rozpoczęto po 10 dniach od posadzenia sałaty. Wybrane WWA aplikowano doglebowo 5-krotnie w odstępach 5 dni. Zabieg stosowano do końca wegetacji. Oznaczenia makroskładników (N, P, K, Mg, Ca, Na) dokonano standardowymi metodami po mineralizacji ( $H_2SO_4+H_2O_2$ ) wysuszonego w 60°C materiału roślinnego. Oznaczenia przeprowadzono wobec materiału certyfikowanego (CTA-VTL-2).

Zwiększenie 3-krotnie zasobności składników pokarmowych (N, P, K, Mg, Na, Cl) w podłożu przyczyniło się do wzrostu plonu główek sałaty o 13,7%. Obecne w podłożu związki z grupy WWA zmniejszyły plon sałaty. Wyraźniej ta tendencja występowała w przypadku antracenu niż pirenu. ANT i PYR doprowadziły do spadku zawartości azotu oraz wzrostu koncentracji wapnia, w warunkach niskiej zasobności podłoża w składniki pokarmowe.

Słowa kluczowe: *Lactuca sativa* L., makroskładniki, antracen, piren, gleba, dawki nawozu.

## INTRODUCTION

Due to the disturbed balance between their decomposition in soil and the influx from different sources (both natural and anthropogenic ones), polycyclic aromatic hydrocarbons (PAHs) are present in all compartments of the environment (KIPOPOULOU 1999, TSIBULSKY 2001). Most of these compounds eventually accumulate in soil (MALISZEWSKA-KORDYBACH 1992). These substances reach soil from anthropogenic sources, such as man-made air and water contamination, but they also originate from organic and natural fertilisers and are produced as a result of mineralisation of organic matter, either present or introduced to soil (BLUMER 1976). Polycyclic aromatic hydrocarbons can either intensify or inhibit life processes in plants (HUANG et al. 1997, WIECZOREK et al. 2001, 2004).



This paper discusses the effect of soil contamination with PAHs (anthracene and pyrene or their mixture) on the yield as well as the concentration and uptake of N, P, K, Ca, Mg and Na by cv. Vilmorin butter lettuce (*Lactuca sativa* L.) grown on substrate differently abundant in nutrients.

## MATERIAL AND METHODS

A pot experiment was conducted in 2007 and 2008 in a greenhouse of the University of Warmia and Mazury in Olsztyn. It comprised four replicates. Butter lettuce (*Lactuca sativa* L.) of cv. Vilmorin was used for the trials. The lettuce was planted in Kick-Brauckmann pots containing 10 dm<sup>3</sup> mineral substrate. The mineral soil used for the experiment, of pH 6.5 and EC 0.11, contained (in mg dm<sup>-3</sup>) 4.1. N-NO<sub>3</sub>, 5.5 N-NH<sub>4</sub>, 44.2 P, 173.3 K, 60.9 Mg, 921.9 Ca, 8.3 Na, 13.4 Cl, 71.7 S-SO<sub>4</sub>. Soil in pots was brought to the following level of abundance (in mg dm<sup>-3</sup>) (N-P-K-Mg-Na-Cl – level I): 60-50-50-40-20-30.8, (N-P-K-Mg-Na-Cl – level II): 180-150-150-120-60-92.4) – factor I. In order to control weeds, the substrate was sprayed with propyzamide, the only active substance allowed to be used for weed control in lettuce cultivation. The amount of propyzamide applied was 0.65 mg dm<sup>-3</sup> of substrate. The spraying treatment was performed according to the recommendations, i.e. a day before planting lettuce. One gram of anthracene (ANT) or pyrene (PYR) was dissolved in 10 cm<sup>3</sup> of acetonitrile (ACN) and filled up to the volume of 100 cm<sup>3</sup> with deionised water. Working solutions were diluted to the concentration of 100 mg dm<sup>-3</sup> of ANT, PYR or ANT+PYR. Control trials were watered with the solution of ACN of the same concentration or with deionised water. Soil application of the PAHs (ANT, PYR or ANT+PYR) was performed every 5 days, in the amount of 9 cm<sup>3</sup> of the concentration 100 mg dm<sup>-3</sup> of anthracene, pyrene or their mixture, starting on day 10 after planting butter lettuce. The contaminants were spread over the surface of soil 5 times, in the volume of 100 cm<sup>3</sup> of deionised water. In total, 4.5 mg of each tested PAH (450 mg kg<sup>-1</sup> of soil) was introduced to soil – factor II.

Lettuce was harvested after 6 weeks and fresh matter yield of lettuce heads was determined. After wet mineralisation in H<sub>2</sub>SO<sub>4</sub>, the concentration of macronutrients (N<sub>og</sub>, P, K, Ca, Na and Mg) was determined with standard methods. In order to verify the correctness of the analytical determinations, certified material (CTA-VTL-2) was used and the measurements obtained were loaded with the following error: P – 4.5%, K – 2%, Ca – 2.8%, Mg – 1.5%, Na – 7%. The results of the determinations underwent statistical analysis using the software application Statistica 7.0.

## RESULTS AND DISCUSSION

Butter lettuce responded to the experimental factors, particularly to the increased abundance of substrate in N, P, K, Mg, Na and Cl (Table 1). The three-fold increase in the content of macronutrients in soil caused an increase in the lettuce head fresh matter yield by an average of 23.2 g. The experiment demonstrated a negative effect of the soil contamination with anthracene and pyrene on the amount of produced lettuce biomass. Anthracene had a more adverse influence because its application depressed the mass of lettuce heads by 8.2% versus the control treatment. Similar impact of compounds belonging to the PAH group on lettuce and other plants has been indicated by CHAINEAU et al. (1997), MALISZEWSKA-KORDYBACH and SMERCZAK (1999). The results of our trials suggest that the two polycyclic aromatic hydrocarbons produced different results. Anthracene reduced the mass of lettuce heads irrespective of the substrate abundance whereas the soil application of pyrene stimulated the growth of lettuce heads when the soil abundance in nutrients was low. The mixture of these compounds had an indirect influence on the quantity of fresh mass of lettuce. KUMMEROWA et al. (1995), who examined the influence of another PAH (benzo(a)pyrene), obtained higher growth of roots of lettuce when concentrations of this compound were low (0-50 mg dm<sup>-3</sup>), while higher rates of this contaminant in the medium retarded the growth.

Concentrations of nitrogen, potassium and calcium in butter lettuce were modified mainly by the abundance of substrate in nutrients (Figure 1). When it was increased three-fold, the dry matter of lettuce contained more N (by

Table 1

Effect of fertilisation and soil application of PAHs on fresh mass of butter lettuce (*Lactuca sativa* L.) heads

Treatments	Control	ANT	PYR	ANT + PYR	<i>X</i> ± SD
	g head <sup>-1</sup>				
Fertilisation level I	171.0	164.3	177.2	166.5	169.7 ± 16.27
Fertilisation level II	203.0	182.8	196.6	189.2	192.9 ± 36.63
<i>X</i> ± SD	187.0 ± 21.1	173.5 ± 24.7	186.9 ± 34.5	177.8 ± 38.7	-

16.5%), K (by 10.7%) and Ca (by 22.5%) in comparison to the average concentrations of these elements when the abundance of substrate was low. Polycyclic aromatic hydrocarbons also affected the concentrations of N, K and Ca, but these modifications were not confirmed statistically. The influence of PAHs was more evident when the substrate was less abundant in nutrients. Anthracene, pyrene as well as their mixed application caused a higher concentration of calcium in lettuce and a lower one of nitrogen. Anthracene contributed to depressing the concentration of nitrogen by 2.3 g kg<sup>-1</sup> d.m., but slightly raised the concentration of potassium in lettuce leaves. The results indicating the concentration of calcium are in accord

with the report by WIECZOREK et al. (2006), who found out that anthracene introduced to soil increased the concentration of calcium in leaves of yellow lupine.

In the experiment presented in this paper, significant influence of the level of fertilisation on the concentration of phosphorus, magnesium and sodium was demonstrated (Figure 2). Lettuce grown on less abundant sub-

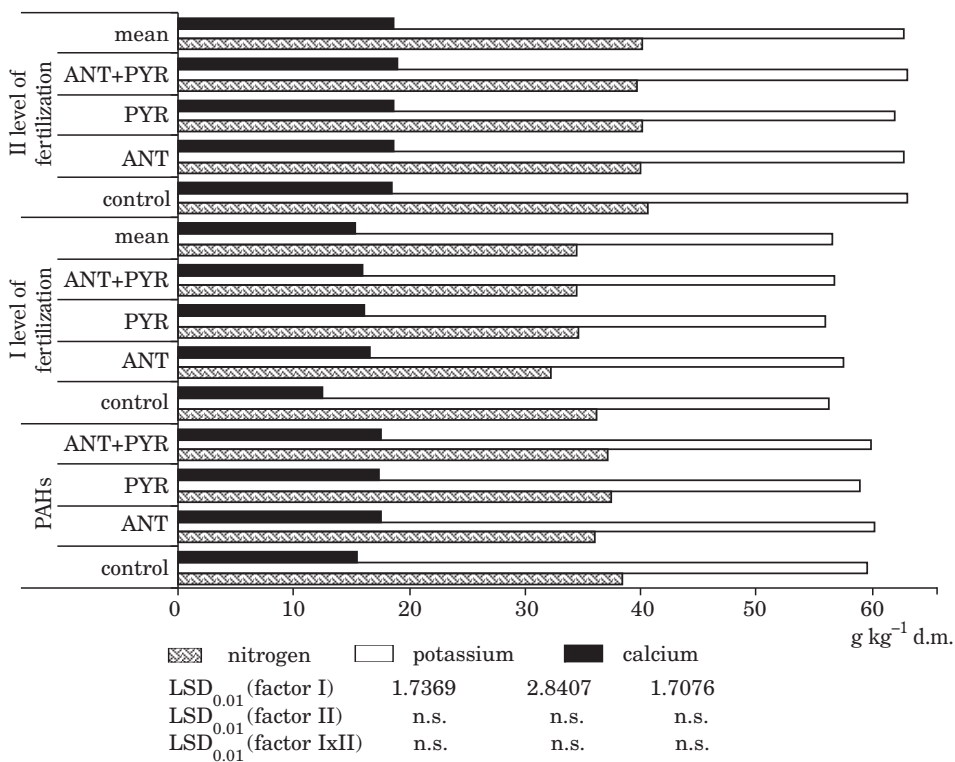


Fig. 1. Effect of fertilisation and soil application of PAHs on the concentration of nitrogen and calcium in butter lettuce

strate contained 1.74-fold more phosphorus than magnesium and 2.2-fold more than sodium. Lettuce cultivated on more abundant soil contained just 1.42-fold more phosphorus and 1.12-fold more sodium. Contamination of soil with ANT and PYR did not have any considerable effect on the concentration of the above nutrients. In turn, WIECZOREK et al. (2006) demonstrated that anthracene introduced to soil increased the concentration of magnesium in leaves of yellow lupine. In the authors' own experiment, no such effect appeared. Contrary to that, the concentration of magnesium in lettuce grown on less abundant substrate was lower. Contamination of soil more abundant in nutrients with pyrene increased the concentration of P, K and Na in edible parts of butter lettuce.

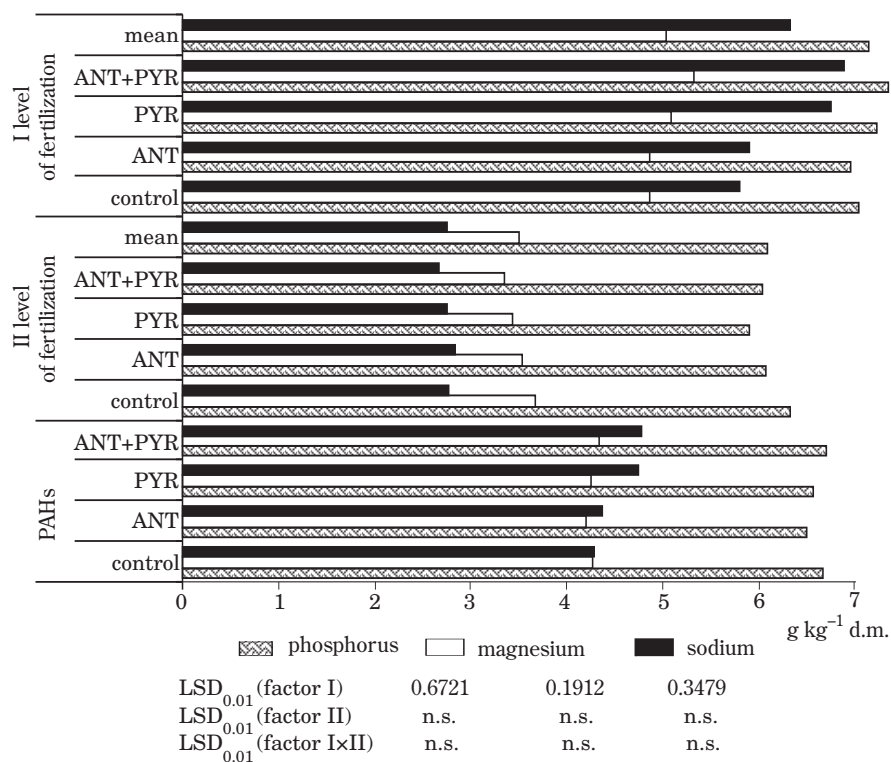


Fig. 2. Effect of fertilisation and soil application of PAHs on the concentration of phosphorus, magnesium and sodium in butter lettuce

The tests revealed a significant effect of the applied PAHs on the concentration and competitiveness in the uptake of calcium and potassium ions by butter lettuce (Figure 3). The PAHs modified to a larger extent the ratios between the concentration of Ca to K when soil was less abundant in nutrients. As the concentration of calcium increased, the concentration of potassium decreased, which was further depressed by the PAHs present in soil. The high correlation coefficients obtained in our tests indicate that these compounds may have a negative effect on the uptake of nutrients, especially when soils are less abundant in nutrients.

The amounts of macronutrients absorbed by butter lettuce depended primarily on the abundance of substrate in N, P, K, Mg, Na and Cl (Table 2). As a result of the increased concentrations of the nutrients in soil, their uptake by lettuce increased. The uptake of N, P, K and Ca rose by 6% (K) up to 19% (Ca) and in the case of magnesium by 41% while the uptake of sodium was two-fold higher. Contamination of soil with anthracene reduced the uptake of nitrogen, phosphorus and magnesium but stimulated the accumulation of potassium, calcium and sodium. Pyrene applied to soil caused

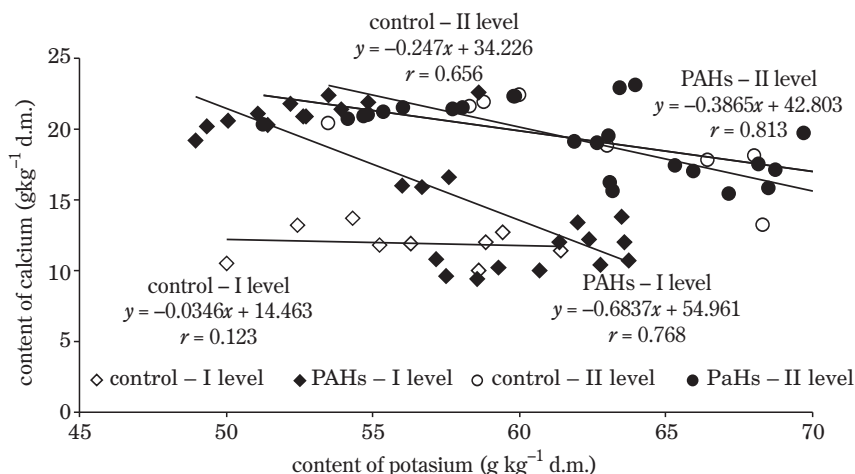


Fig. 3. Correlation between the concentration of calcium and potassium in butter lettuce depending on fertilisation and soil application of PAHs

an increased uptake of K, Ca, Mg and Na by lettuce. When soil contained larger quantities of PAHs (ANT+PYR), the accumulation of nitrogen, phosphorus and potassium in edible parts of lettuce was negatively affected.

## CONCLUSIONS

1. Anthracene and pyrene, if present in soil, can cause depressed yields of butter lettuce.

2. The concentration of nutrients and their uptake by butter lettuce depended on the abundance of substrate in N, P, K, Mg, Na and Cl and, to a lesser degree, on the presence of anthracene and pyrene in soil.

3. Anthracene and pyrene caused an increased concentration and uptake of calcium and sodium by lettuce.

4. Low abundance of substrate in conjunction with the soil contamination with ANT and PYR may considerably modify the concentration of nutrients in butter lettuce.

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# **EFFECT OF NITROGEN, PHOSPHORUS AND POTASSIUM FERTILIZATION ON YIELDING AND BIOLOGICAL VALUE OF FRUITS OF AUBERGINE (*SOLANUM MELONGENA* L.)**

**Bartosz Markiewicz, Anna Golcz**

**Chair of Horticultural Crop Fertilization  
Poznan University of Life Sciences**

## **Abstract**

A plant growing experiment was conducted in 2002-2003 on the aubergine cultivars Epic F<sub>1</sub> and Solara F<sub>1</sub> grown in an unheated polyethylene tunnel greenhouse at the Experimental Station in Marcelin, the University of Life Sciences in Poznań. Seedlings were planted on May 15 on beds at a 0.5 × 0.5 m spacing, i.e. 4 plants m<sup>-2</sup>, into 6 dm<sup>3</sup> cylinders filled with a mixture, limed to pH<sub>H<sub>2</sub>O</sub> = 6.5, of mineral soil (light loamy sand containing 12% clay fraction – deposited on medium-heavy loam) with highmoor peat from Lithuania (v : v = 4 : 1). Basic fertilization – pre-vegetation and top dressing with macronutrients, based on an analysis of the substrate using the universal method in 0.03 M CH<sub>3</sub>COOH, was determined to attain the assumed levels: L (N – 200, P – 175, K – 330 mg dm<sup>-3</sup>), S (N – 300, P – 265, K – 500 mg dm<sup>-3</sup>), H (N – 400, P – 350, K – 665 mg dm<sup>-3</sup>), while maintaining the N : P : K ratio at 1 : 0.9 : 1.7. The aim of this study has been to determine the effect of a fertilization level and cultivar on the yield and biological value of fruits of aubergine grown on a mixture of mineral soil with highmoor peat (v : v = 4 : 1). The total yield, number of fruits and weight of individual fruits were determined. Significant effect was found for the fertilization level and cultivar on the total yield, mean number of fruits and weight of a single aubergine fruit. Fruits of cv. Epic F<sub>1</sub> aubergine contained more vitamin C than fruits of cv. Solara F<sub>1</sub>. In both years, the solids content in fruits of the two aubergine cultivars ranged from 4.0 to 5.5 %. A higher mean dry matter content in aubergine fruits was recorded in cv. Solara F<sub>1</sub>.

**Key words:** aubergine, fertilization, biological value.

## WPŁYW NAWOŻENIA AZOTEM, FOSFOREM I POTASEM NA PLONOWANIE I WARTOŚĆ BIOLOGICZNĄ OWOCÓW OBERŻYNY (*SOLANUM MELONGENA* L.)

### Abstrakt

W latach 2002-2003 przeprowadzono doświadczenia wegetacyjne z uprawą oberżyny odm. Epic F<sub>1</sub> i Solara F<sub>1</sub> w nieogrzewanym tunelu foliowym w Stacji Doświadczalnej Marcecin Uniwersytetu Przyrodniczego w Poznaniu. Rośliny sadzono 15 maja na zagonach w rozstawie 0,5 × 0,5 m, tj. 4 rośliny m<sup>-2</sup>, w cylindrach o obj. 6 dm<sup>3</sup> wypełnionych zwapnowaną do pH<sub>H<sub>2</sub>O</sub> = 6,5 mieszaniną gleby mineralnej (piasek gliniasty lekki o zawartości 12% części ilowych – zalegający na glinie średniej) z torfem wysokim z Litwy (v : v = 4 : 1). Nawożenie podstawowe – przedwegetacyjne i pogłówne makroskładnikami, oparte na analizie podłoża wykonanej metodą uniwersalną wg NOWOSIELSKIEGO (1988) w 0,03 M CH<sub>3</sub>COOH – ustalono do założonych poziomów z zachowaniem proporcji makroskładników N : P : K = 1 : 0,9 : 1,7 : N (N – 200, P – 175, K – 330 mg dm<sup>-3</sup>), S (N – 300, P – 265, K – 500 mg dm<sup>-3</sup>), W (N – 400, P – 350, K – 665 mg dm<sup>-3</sup>). Celem pracy było określenie wpływu poziomów nawożenia na plon i wartość biologiczną owoców oberżyny uprawianej w mieszaninie gleby mineralnej z torfem wysokim (v : v = 4 : 1). Określono plon ogólny owoców, liczbę owoców, średnią masę pojedynczego owocu oraz wartość biologiczną owoców. Stwierdzono istotny wpływ poziomu nawożenia i odmiany na plon ogólny, średnią liczbę owoców oraz masę pojedynczego owocu oberżyny. Owoce oberżyny odmiany Epic F<sub>1</sub> zawierały więcej witaminy C niż owoce odmiany Solara F<sub>1</sub>. We wszystkich latach badań zawartość ekstraktu w owocach obu odmian oberżyny wynosiła od 4,0 do 5,5%. Większą średnią zawartość suchej masy w owocach oberżyny oznaczono u odmiany Solara F<sub>1</sub>.

Słowa kluczowe: oberżyna, nawożenie, wartość biologiczna.

## INTRODUCTION

The most suitable soils for aubergine are the ones with pH close to neutral (pH 6.5-7.0), friable, medium-heavy, rich in humus and nutrients, with a sufficient amount of water, but not water-logged. Such soils are chernozems, alluvial soils and sandy loams rich in humus, with good air and water relations.

The research conducted by Polish authors on aubergine cultivation in polyethylene tunnel greenhouse mostly deals with organic substrates, particularly peat (BUCZKOWSKA 1998 MICHAŁOJC, BUCZKOWSKA 2008, 2009). Aubergine may also be grown on substrates enriched with peat and on mixtures of peat and bark (GAJEWSKI, GAJC-WOLSKA 1998). The high cost of crop cultivation in tunnels stimulates the search for more economical solutions.

The aim of this study has been to determine the effect of a fertilization level and cultivar on the yield and biological value of fruits of aubergine grown on a mixture of mineral soil with highmoor peat (v:v = 4:1).



## MATERIAL AND METHODS

In 2002-2003, a plant growing experiment was conducted on aubergine cultivars Epic F<sub>1</sub> and Solara F<sub>1</sub> grown in an unheated polyethylene tunnel greenhouse at the Experimental Station in Marcelin, the University of Life Sciences in Poznań. Seedlings were planted on May 15 on beds at a 0.5 x 0.5 m spacing, i.e. 4 plants m<sup>-2</sup>, into 6 dm<sup>3</sup> cylinders filled with a mixture, limed to pH<sub>H<sub>2</sub>O</sub> = 6.5, of mineral soil (light loamy sand containing 12% clay fraction – deposited on medium-heavy loam) with highmoor peat from Lithuania (v : v = 4 : 1). Basic fertilization – pre-vegetation and top dressing with macronutrients, based on an analysis of the substrate using the universal method in 0.03 M CH<sub>3</sub>COOH, was designed so as to attain the assumed levels: low (N), standard (S) and (W) – Table 1, while maintaining the N : P : K ratio at 1 : 0.9 : 1.7. The other macro- and micronutrients constituted the background of the experiment.

Table 1

Nutrient levels in pre-vegetation fertilization and top dressing of aubergine

Nutrient	Fertilization			
	pre-vegetation		top dressing	
	(mg dm <sup>-3</sup> )			
	LSH	L	S	H
N	250	200	300	400
P	220	175	265	350
K	415	330	500	665
Ca	1500 - 2000			

Top dressing was performed 3 times at 4-week intervals. Deficits of nitrogen, phosphorus and potassium were supplemented to the assumed levels. In the experiments, mineral fertilizers, i.e. NH<sub>4</sub>NO<sub>3</sub>, KH<sub>2</sub>PO<sub>4</sub> and K<sub>2</sub>SO<sub>4</sub>, were applied.

The total yield, number of fruits and weight of individual fruits were determined.

The following quality parameters were determined in fresh aubergine fruits (FORTUNA et al. 2003):

- 1) dry weight – with the oven-dry method,
- 2) vitamin C – according to Tillmans (PN – 90/A – 75101/11),
- 3) total solids – by refractometry (PN – 90/A – 75101/02).

The total yield, number of fruits and the weight of individual fruits were analyzed statistically using Duncan's test for three-factorial experiments at a significance level  $\alpha = 0.05$ . Factor A was the year of the study (2), factor B

– fertilization level (3) and factor *C* – cultivar (2). In total, the experiment consisted of 12 combinations 4 replications, while each replication was composed of 5 plants.

## RESULTS AND DISCUSSION

In the experiment, the mean yield of fruits in the second year of the experiment ( $3.86 \text{ kg m}^{-2}$ ) was significantly higher than in the first year ( $2.83 \text{ kg m}^{-2}$ ) – Table 2. The difference between the years in terms of the mean total yield of fruits may have been caused by the lower mean daily temperature and insolation in the first 6 weeks of the experiment.

Table 2

The effect of fertilization level and cultivar on the total yield of aubergine fruits

Year	Nutrition level	Total yield ( $\text{kg m}^{-2}$ )			
		cultivar ( <i>C</i> )		mean ( <i>A</i> x <i>B</i> )	mean ( <i>A</i> )
(A)	(B)	Epic	Solara		
I	L	2.15 <sup>b*</sup>	1.60 <sup>a</sup>	1.88 <sup>a</sup>	2.83 <sup>a</sup>
	S	2.73 <sup>c</sup>	2.20 <sup>b</sup>	2.46 <sup>b</sup>	
	H	4.58 <sup>f</sup>	3.74 <sup>de</sup>	4.16 <sup>d</sup>	
	mean ( <i>A</i> x <i>C</i> )	3.15 <sup>b</sup>	2.51 <sup>a</sup>		
II	L	3.51 <sup>d</sup>	2.97 <sup>c</sup>	3.24 <sup>c</sup>	3.86 <sup>b</sup>
	S	3.89 <sup>de</sup>	3.95 <sup>e</sup>	3.92 <sup>d</sup>	
	H	4.88 <sup>f</sup>	4.00 <sup>e</sup>	4.44 <sup>e</sup>	
	mean ( <i>A</i> x <i>C</i> )	4.09 <sup>d</sup>	3.64 <sup>c</sup>		
Mean ( <i>C</i> )		3.62 <sup>b</sup>	3.08 <sup>a</sup>		
Mean ( <i>B</i> x <i>C</i> )		2.83 <sup>b</sup>	2.28 <sup>a</sup>	L L. S. H – fertilization level L – low S – standard H – high	
		3.31 <sup>c</sup>	3.07 <sup>bc</sup>		
		4.73 <sup>e</sup>	3.87 <sup>d</sup>		
Mean ( <i>B</i> )		2.56 <sup>a</sup>			
		3.19 <sup>b</sup>			
		4.30 <sup>c</sup>			

\* Means marked with the same letters do not differ significantly at a level of  $\alpha = 0.05$ .

The total yield of fruits in case of plants growing on the mixture of mineral soil with highmoor peat in the years 2002-2003 ranged from 1.60 to  $4.88 \text{ kg m}^{-2}$ . A significant effect of the fertilization level on the total yield of aubergine fruits was observed. The lowest mean yield was harvested at the low fertilization level ( $2.56 \text{ kg m}^{-2}$ ), while the highest – at the high fertilization level ( $4.30 \text{ kg m}^{-2}$ ). Yielding of plants was significantly affected by the aubergine cultivar. The mean total yield harvested in both years was higher from cv. Epic F<sub>1</sub> ( $3.15$  and  $4.09 \text{ kg m}^{-2}$ ) than from cv. Solara F<sub>1</sub> ( $2.51$  and

3.64 kg m<sup>-2</sup>). The range of the harvested yield was lower than that given by CEBULA and AMBROSZCZYK (1999), who reported that the yield of aubergine fruits grown in a tunnel ranged from 6.83 to 10.17 kg m<sup>-2</sup>.

A significantly higher mean number of fruits (10.77 fruits m<sup>-2</sup>) was harvested in the second year of the experiment in comparison to the first year (8.75 fruits m<sup>-2</sup>) – Table 3. Moreover, a significant effect of the fertilization level on the mean number of fruits – from 7.78 fruits m<sup>-2</sup> at the application of the low fertilization level to 11.70 fruits m<sup>-2</sup> at the high fertilization level. The number of fruits in the experiment ranged from 5.10 fruits m<sup>-2</sup> to 12.60 fruits m<sup>-2</sup> depending on the year, cultivar and fertilization level. The mean number of fruits in this study was similar to those reported by other authors (CEBULA 1996).

Table 3

The effect of the fertilization level and cultivar on the mean number of aubergine fruits

Year	Nutrition level	Number of fruits (pcs. m <sup>-2</sup> )			
		cultivar (C)		mean (A x B)	mean (A)
(A)	(B)	Epic	Solara		
I	L	7.20 <sup>b</sup>	5.10 <sup>a</sup>	6.15 <sup>a</sup>	8.75 <sup>a</sup>
	S	8.40 <sup>bc</sup>	7.80 <sup>bc</sup>	8.10 <sup>b</sup>	
	H	13.35 <sup>h</sup>	10.65 <sup>e</sup>	12.00 <sup>d</sup>	
	mean (A x C)	9.65 <sup>b</sup>	7.85 <sup>a</sup>		
II	L	9.03 <sup>cd</sup>	9.78 <sup>de</sup>	9.41 <sup>c</sup>	10.77 <sup>b</sup>
	S	10.98 <sup>ef</sup>	12.00 <sup>fg</sup>	11.49 <sup>d</sup>	
	H	12.60 <sup>gh</sup>	10.21 <sup>de</sup>	11.40 <sup>d</sup>	
	mean (A x C)	10.87 <sup>c</sup>	10.66 <sup>c</sup>		
Mean (C)		10.26 <sup>b</sup>	9.26 <sup>a</sup>		
Mean (B x C)		8.11 <sup>a</sup>	7.44 <sup>a</sup>		
		9.69 <sup>b</sup>	9.90 <sup>b</sup>		
		12.98 <sup>c</sup>	10.43 <sup>b</sup>		
Mean (B)		7.78 <sup>a</sup>			
		9.80 <sup>b</sup>			
		11.70 <sup>c</sup>			

Key: see Table 2

The weight of a single fruit differed significantly between the years (Table 4). Moreover, a significant effect of the fertilization level was observed on this yield parameter. The recorded mean weight of a single fruit ranged from 303.52 g to 390.32 g depending on the year, cultivar and fertilization level. The recorded fruit weight was higher than reported by CEBULA (1996). Moreover, CEBULA and AMBROSZCZYK (1999), in their study on yielding of several aubergine cultivars, obtained the mean weight of a single fruit within the range from 346 to 475 g, depending on the cultivar.

Table 4

The effect of the fertilization levels and cultivar on the weight of a single aubergine fruits

Year	Nutrition level	Mass of single fruits (g)			
		cultivar (C)		mean (A x B)	mean (A)
(A)	(B)	Epic	Solara		
I	L	316.47 <sup>ab</sup>	319.70 <sup>ab</sup>	318.07 <sup>ab</sup>	327.33 <sup>a</sup>
	S	326.37 <sup>ab</sup>	305.80 <sup>a</sup>	316.08 <sup>a</sup>	
	H	343.85 <sup>ab</sup>	351.80 <sup>bc</sup>	347.82 <sup>b</sup>	
	mean (A x C)	328.9 <sup>a</sup>	325.76 <sup>a</sup>		
II	L	389.00 <sup>c</sup>	303.52 <sup>a</sup>	346.26 <sup>b</sup>	358.79 <sup>b</sup>
	S	353.95 <sup>bc</sup>	328.52 <sup>ab</sup>	341.23 <sup>ab</sup>	
	H	387.45 <sup>c</sup>	390.32 <sup>c</sup>	388.88 <sup>c</sup>	
	mean (A x C)	376.80 <sup>c</sup>	340.79 <sup>a</sup>		
Mean (C)		352.85 <sup>b</sup>	333.27 <sup>a</sup>		
Mean (B x C)		352.73 <sup>bc</sup>	311.61 <sup>a</sup>		
		340.16 <sup>ab</sup>	317.16 <sup>a</sup>		
		365.65 <sup>bc</sup>	371.06 <sup>c</sup>		
Mean (B)		332.17 <sup>a</sup> 328.66 <sup>a</sup> 368.35 <sup>b</sup>			

Key: see Table 2

The dry matter content in the analyzed fruits ranged from 7.27 % to 10.01 % d.m. (Table 5). The effect of a cultivar and fertilization level on the mean dry matter content in aubergine fruits was observed. A higher mean dry matter content was determined in cv. Solara F<sub>1</sub> (9.42% d.m.) than in cv. Epic F<sub>1</sub> (8.72% d.m.). The mean content of dry matter in fruits increased

Table 5

The effect of the fertilization level and cultivar on the dry matter content in aubergine fruits

Year	Nutrition level	Dry matter (%)			
		cultivar (C)		mean (B)	mean (A)
(A)	(B)	Epic	Solara		
I	L	7.27	8.98	8.22 9.43 9.57	8.87
	S	8.76	9.52		
	H	9.24	9.46		
II	L	7.35	9.28	8.22 9.43 9.57	9.28
	S	9.71	9.74		
	H	10.01	9.59		
Mean (C)		8.72	9.42		

Key: see Table 2

with an increasing fertilization level from 8.22 to 9.57% d.m. The mean dry matter content in fruits determined in this study did not exceed  $\pm 10\%$ . A low dry matter content in fruits of aubergine grown in organic substrates was obtained by CEBULA (1996) and CEBULA and AMBROSZCZYK (1999).

The total solids content in fruits of aubergine grown on mineral soil with highmoor peat ranged from 4.00 to 5.50% depending on the year, fertilization level and cultivar (Table 6).

Table 6

The effect of the fertilization level and cultivar on the total solids content in aubergine fruits

Year	Nutrition level	Extract (%)			
		cultivar (C)		mean (B)	mean (A)
(A)	(B)	Epic	Solara		
I	L	5.5	4.0	5.00 4.62 4.75	4.66
	S	4.5	4.0		
	H	5.0	5.0		
II	L	5.5	5.0		4.91
	S	4.5	5.5		
	H	5.0	4.0		
Mean (C)		5.00	4.58		

Key: see Table 2

ESTEBANA et al. (1992) claimed that aubergine fruits contained the biggest amounts of ascorbic acid on day 42 from their setting. In this study, aubergine fruits were collected for analyses about 40 days after their setting. The content of vitamin C in aubergine fruits ranged from 12.9 mg% to 23.7 mg%, only slightly exceeding the values reported by CEBULA, AMBROSZCZYK (1999) – Table 7. The investigations conducted by WIERZBICKA, KUSOWSKA

Table 7

The effect of the fertilization level and cultivar on the vitamin C content in aubergine fruits

Year	Nutrition level	Vitamin C (mg%)			
		cultivar (C)		mean (B)	mean (A)
(A)	(B)	Epic F <sub>1</sub>	Solara F <sub>2</sub>		
I	L	17.2	17.2	17.2 16.1 16.1	14.7
	S	15.1	12.9		
	H	12.9	12.9		
II	L	17.2	17.2		18.31
	S	21.6	15.1		
	H	23.7	15.1		
Mean (C)		17.96	15.06		

Key: see Table 2

(2002) showed that the vitamin C content in vegetables depended primarily on the species as well as the cultivar. The biosynthesis of this compound in fruits is also significantly affected by insolation (WO•NIAK et al. 2002).

## CONCLUSIONS

1. Significant effect was found of the fertilization level and cultivar on the total yield, mean number of fruits and weight of a single aubergine fruit.

2. It is recommendable to provide aubergine growing on mineral soil mixed with highmoor peat (v:v – 4:1) with pre-vegetation supplementation consisting of nitrogen, phosphorus and potassium brought in the substrate to the following levels ( $\text{mg dm}^{-3}$ ): N – 250, P – 220, K – 415, while the top dressing treatment (from the third week of the growing season) should maintain the content of the nutrients in the substrate at: N – 400, P – 350 and K – 665.

3. Fruits of aubergine cv. Epic F<sub>1</sub> contained more vitamin C than fruits of cv. Solara F<sub>1</sub>.

4. In fruits of both aubergine cultivars in the two years of the study, the total solids content ranged from 4.0 to 5.5%.

5. A higher mean dry matter content in aubergine fruits was recorded in cv. Solara F<sub>1</sub>.

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# **ASSESSMENT OF METAL HAZARD TAKING HAIR AS AN INDICATOR OF TRACE ELEMENT EXPOSURE TO WORKERS IN OCCUPATIONAL ENVIRONMENT**

**Rita Mehra, Amit Singh Thakur**

**Department of Pure and Applied Chemistry  
Maharshi Dayanand Saraswati University Ajmer, India**

## **Abstract**

The use of biological tissues as diagnostic tools of trace element level for recognising the disease status of an exposed human population is an important area of investigation. Quantitative analysis of 10 trace elements viz lead, calcium, magnesium, chromium, manganese, iron, nickel, copper, and zinc in the human body was performed using hair as biopsy material. In the present investigation, workers in a roadways workshop, locomotive workshop and Pb battery units were included as subjects. Head hair samples were collected from the nape region of the skull. Subsequent to sampling of hair, a questionnaire recommended by the World Health Organization was filled in order to obtain details about the subjects regarding occupation, duration of exposure, medical history, etc. Concentration of elements in hair was determined using an atomic absorption spectrophotometer. Workers under mental stress were treated as cases and workers without any symptoms of mental stress were chosen as controls. Significant difference in concentration of lead, calcium, magnesium, manganese, nickel, copper and zinc was found in hair of subjects with mental stress than those of controls. Concentration of chromium and iron was found higher in hair of subjects with mental stress but this difference was not significant with respect to controls.

**Key words:** hair, mental stress, trace elements, occupational environment, health hazards.

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dr Rita Mehra, Associate Professor & Head, Department of Pure and Applied Chemistry,  
Maharshi Dayanand Saraswati University Ajmer 305009, Rajasthan India,  
e-mail.: mehra\_rita@rediffmail.com, thakuramit000111@gmail.com

## OCENA ZAGROŻENIA ZATRUCIEM METALAMI NA PODSTAWIE ANALIZY WŁOSÓW JAKO WSKAZNIKA WYSTAWIENIA ROBOTNIKÓW NA DZIAŁANIE PIERWIASTKÓW ŚLADOWYCH W ŚRODOWISKU PRACY

### Abstrakt

Ważnym obszarem badań jest wykorzystywanie tkanek biologicznych do określania zawartości pierwiastków śladowych w celu diagnozowania chorób wśród populacji narażonych na skażenie środowiskowe. Analizowano zawartość 10 pierwiastków śladowych, m.in. ołowiu, wapnia, magnezu, chromu, manganu, żelaza, niklu, miedzi oraz cynku, w ciele ludzkim na podstawie włosów pobranych od robotników z warsztatów drogowych, kolejowych oraz mających do czynienia z akumulatorami zawierającymi rtęć. Próbkę włosów pobrano z okolicy potylicznej czaszki. Po pobraniu próbek włosów, uczestniczący w badaniu wypełnili ankiety Światowej Organizacji Zdrowia, których celem było zebranie szczegółowych danych dotyczących miejsca pracy, czasu trwania ekspozycji na czynniki ryzyka, historii przebytych chorób, itp. Zawartość pierwiastków we włosach określono za pomocą spektrofotometru absorpcji atomowej. Robotnicy doświadczający stresu psychicznego zostali ujęci jako przypadki chorobowe, natomiast nie przejawiający objawów napięcia zostali potraktowani jako przypadki kontrolne. Stwierdzono istotne różnice w zawartości ołowiu, wapnia, magnezu, manganu, niklu, miedzi i cynku we włosach osób z objawami napięcia psychicznego w porównaniu z włosami pobranymi od osób bez takich symptomów. Zawartość chromu i żelaza we włosach osób z objawami stresu była także wyższa, ale różnice nie były istotne w porównaniu z kontrolą.

Słowa kluczowe: włosy, stres psychiczny, pierwiastki śladowe, zagrożenie zdrowia.

## INTRODUCTION

Rapid industrialization with discharges from metallurgical and chemical plants has redistributed many elements in the environment. Chemical and metallurgical plants in India continue to be among the world's top ten polluters. This has increased the risk of human exposure to toxic elements, in particular metals, through ingestion, inhalation, injection and absorption through exposed parts of the body. The level of these elements in humans can be determined by using human tissues as biopsy material (NOWAK, CHMIELNICKA 2000). Hair is easily available and gives significant information about element levels in the body as compared to other biopsy material (MEHRA, BHALLA 2000, MEHRA, JUNEJA 2003, SELA et al. 2007). Biomonitoring of hair in recent times has been gaining significance in various fields of science such as environmental science, medical science, forensic science and archaeological science (MEHRA, JUNEJA 2005 a,b,c, MEHRA et al. 2010 a,b, VALKOVIC, LIMIC 1986, POZEBON et al. 2008, DANIEL et al. 2004, DU et al. 1996, SEMPLE 2005). Accumulation of trace elements leads to various adverse effects on health of an organism. It is therefore desirable to evaluate the trace element concentrations, so that necessary measures can be taken to prevent the trace element pollution in the environment as also caused by undue exposure of human population to trace elements. This simple, non-inva-

sive biopsy materials will help in identifying human populations in the environment at risk of elemental pollution so that necessary preventive actions can be taken (MEHRA, JUNEJA 2004, 2005 a,b,c,d, QUANDT et al. 2010, YORIFUGI et al. 2010).

Monitoring biological tissues is considered as an ideal step of integrated exposure monitoring for health risk assessment and also for use in forensic science (DANIEL et al. 2004). Human hair and nails are considered to be reliable indicators of elemental exposure including trace and toxic elements in natural and work environment and their concentration can be linked to parameters characterizing and conditioning human life such as nutrition, environment, race, health, age, sex etc. In occupational exposure, health ailments have been observed including neurological and respiratory problems such as fatigue, poor memory, dizziness, asthma and other risks (DURAND, WILSON 2006, MEHRA 2002, MEHRA, JUNEJA 2005 a,b,c, TELISMAN et al. 2001, CHIEN et al. 2010, FREIRE et al. 2010, GALLAGHER et al. 2010, ANDERSON 1986, GUSTAFSON et al. 2007).

The present study is a case control study in which workers of roadways workshops, locomotive workshops, and Pb-Cd battery units reporting mental stress were included as case subjects and without mental stress - as control subjects. In total, 40 subjects of the same age, sex and eating habits were included as subjects, among which 20 subjects were cases with mental stress and the other 20 subjects were controls without mental stress. Head hair samples were collected, which were analysed for concentration of lead (Pb), calcium (Ca), magnesium (Mg), chromium (Cr), manganese (Mn), iron (Fe), nickel (Ni), copper (Cu), and zinc (Zn). It was hypothesised that workers reporting mental stress have higher concentration of trace elements in their hair as compared to workers without mental stress. It was also the goal of this study to verify whether hair could be an indicator of health hazards caused by accumulation of trace elements. The main aim of this study is to identify a population at risk of exposure to trace and toxic elements in the environment. This study will also help in evaluating metal body burden and its relation to health.

## **MATERIAL AND METHODS**

In the present study, workers of roadways workshop, locomotive workshop, and Pb-Cd battery units presenting mental stress and without mental stress were taken as subjects. A total of 40 subjects of the same age, sex and eating habits were included in this study. Out of these 40 subjects, 20 were cases complaining of mental stress and the remaining 20 were controls without mental stress. All the workers included in the study worked for 8 hours a day, six days a week. Head hair samples were personally collected from all subjects using stainless steel scissors and stored in air-

tight polythene bags. Other information about the subjects regarding the age, sex, occupation, nature of occupation, medical history etc. was obtained from a questionnaire recommended by the World Health Organisation, which was filled in while sampling.

Hair samples were cut into small pieces of 1 cm to make washing feasible. Pretreatment of hair samples was done to decontaminate them using non-ionic detergent (Triton X-100), acetone and deionised water and kept for drying at 110°C in an oven for one hour. Pretreated hair samples were then digested using nitric acid and perchloric acid in a 6:1 ratio in order to obtain colourless clear solution. The acid is now evaporated to obtain white residue, which was then dissolved in 0.1 N nitric acid (BABU et al. 2009, MEHRA, BHALLA 1996, MEHRA, JUNEJA 2003, 2004). The quantitative analysis of lead (Pb), calcium (Ca), magnesium (Mg), manganese (Mn), nickel (Ni), copper (Cu), zinc (Zn), chromium (Cr) and iron (Fe) was performed in an Atomic Absorption Spectrophotometer (AAS) Perkin Elmer Model-5000 using air acetylene flame. Cathode lamps were used, set at different wavelengths separately for individual elements. The data thus obtained were then analysed to achieve mean standard deviation and the test of significance was performed using Student *t* test.

## RESULTS

Mean and standard deviation of the level of all the tested trace elements in hair samples of workers with mental stress and controls have been calculated and summarized in Table 1. On applying student *t* test for significance, it was revealed that the concentration of Pb, Ca, Mg, Mn, Ni, Cu, and Zn in hair of subjects with mental stress differ significantly from the controls. However, concentration of Cr and Fe in hair of subjects with mental stress was found higher but this was not significantly different from the control subjects.

Table 1 reveals that the concentrations of all the nine elements in hair of subjects with mental stress are higher than in the hair of controls. It becomes evident from the given data that the case subjects are exposed to these elements at their work place and these elements are accumulated in their hair. Figure 1 summarises the total concentration of all nine elements in hair of subjects and controls. The sum of the total and mean concentration of all the nine elements in hair of subjects with mental stress and without mental stress was 3,349.84 mg g<sup>-1</sup> and 2,078.95 µg g<sup>-1</sup> respectively.

Table 1

Concentration of lead, calcium, magnesium, manganese, nickel, copper, zinc, chromium and iron in hair of workers with mental stress and controls

Trace Elements	Mental stress		Controls	
	range ( $\mu\text{g g}^{-1}$ )	mean ( $\pm\text{SD}$ )	range ( $\mu\text{g g}^{-1}$ )	mean ( $\pm\text{SD}$ )
Pb	6.58-22.91	19.33* ( $\pm 1.86$ )	3.12-13.43	10.91 ( $\pm 2.12$ )
Ca	1428.57-3458.42	2237.01* ( $\pm 738.07$ )	775.13-1963.64	1372.48 ( $\pm 584.60$ )
Mg	252.63-614.73	465.95* ( $\pm 201.39$ )	180.25-460.52	223.14 ( $\pm 86.06$ )
Mn	6.76-9.30	8.16* ( $\pm 1.01$ )	1.88-10.46	6.71 ( $\pm 2.34$ )
Ni	28.44-41.47	34.44* ( $\pm 3.28$ )	10.54-35.76	25.58 ( $\pm 13.85$ )
Cu	7.19 - 47.68	26.66* ( $\pm 15.03$ )	3.52-13.07	9.66 ( $\pm 4.55$ )
Zn	131.53 - 392.56	291.97* ( $\pm 96.13$ )	101.82 - 216.91	198.94 ( $\pm 50.37$ )
Cr	27.55-112.06	80.73 ( $\pm 16.89$ )	18.11-85.70	65.90 ( $\pm 24.65$ )
Fe	104.57-255.43	185.59 ( $\pm 47.38$ )	77.23-210.72	165.63 ( $\pm 70.68$ )

\*significant difference ( $P < 0.05$ )

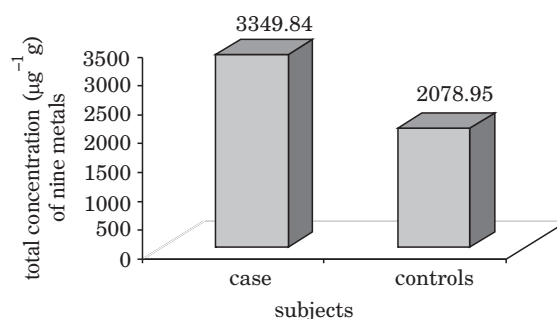


Fig. 1. Total concentration ( $\mu\text{g g}^{-1}$ ) of all the nine metals under in hair of case and control subjects

## DISCUSSION

Exposure to trace elements lead to their accumulation in the biological tissues, which evokes various adverse responses in organisms. The balance of trace elements in the body is very much required for proper functions of body systems. Any imbalance in the concentration of elements in the body leads to various disorders; likewise, maintaining a correct balance of trace elements is required in the body for the proper functioning of the nervous system. Various mental disorders relating to elements have been studied, as illustrated in Table 2.

Table 2

Different mental disorders related with elements

Disorder	Reason	Reference
Behaviour disturbances	elevated Cr, Mo, Pb depressed Co, Pb	RIMLAND et al. 1983
Learning disability	elevated Cd, Cu, Mg, Pb	RIMLAND et al. 1983
Mental retardation	elevated Pb depressed Na	RIMLAND et al. 1983
Behaviour problems	elevated Ab	MOON et al. 1986
Manic depression	elevated V	NAYLOR et al. 1984
Mental retardation (in children)	elevated Ca depressed Fe, Cu, Mg	SHRESTHA et al. 1988

The result of this work indicate that the concentrations of lead, calcium, magnesium, manganese, nickel, copper and zinc in scalp hair of workers with mental stress were significantly different from those without mental stress. Because both groups of workers – with and without mental stress – are from the same community, within the same age bracket, of the same sex and experienced similar socioeconomic conditions, the difference in hair trace elements between these two groups could not have resulted from these variables. It is clear that the difference in the lead, calcium, magnesium, manganese, nickel, copper and zinc concentrations in scalp hair of workers with mental stress and controls, being a significant one, is not accidental. The reported results support our finding that the level of elements affect the metal status in a human organism (RIMLAND, LARSON 1983, MOON, MARLOWE 1986, NAYLOR et al. 1984, SHRESTHA, CARRERA 1988).

## CONCLUSIONS

In this study, some trace elements in scalp hair of workers with mental stress showed quite a different pattern of concentrations than in controls, which indicates that such differences might be related to exposure to these metals. Although the levels of these elements in human hair are a function of many complex variables, as is also the cause of mental stress, information on some essential element imbalance might provide clues for further studies in this area.

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# **CONTENT OF ZINC IN PLANTS FERTILIZED WITH MUNICIPAL SOLID WASTE AND URBAN GREEN WASTE COMPOSTS**

**Wiera Sądej, Anna Namiotko**

**Chair of Chemistry Environment  
University of Warmia and Mazury in Olsztyn**

## **Abstract**

In a vegetation experiment, the effect of composts made from unsorted municipal solid waste and urban green waste on the content of zinc in plants has been analyzed. The municipal waste composts matured in heaps for 1, 3 and 6 months. They were applied in three rates: 10, 20 and 30 g kg<sup>-1</sup> of soil. The compost made of urban green waste aged in a pile for 6 months and was added to soil in the amount of 10 g kg<sup>-1</sup> of soil. It has been determined that the content of zinc in plants was varied and depended on the type and rates of compost used as well as the species of crops. Application of higher rates of composts did not invariably result in an increased concentration of zinc in plant tissues; on the contrary, in some cases the level of zinc determined in fertilized plants was lower than in the control. Much more elevated concentrations of zinc were found after the application of fresh compost made of municipal waste (maturing for one month in a heap). Fertilization of maize and barley with urban green waste compost produced a more favourable result than the application of analogous rates of municipal solid waste compost, as the plants contained less zinc. With respect to the other test plants, such as sunflower, mustard and phacelia, the application of composted green matter caused a considerable increase in the content of zinc, as compared with the treatments fertilized with composted municipal waste, at the same fertilization rates.

**Key words:** municipal solid waste, urban green waste, compost, zinc, plant, soil.

## ZAWARTOŚĆ CYNKU W ROŚLINACH NAWOŻONYCH KOMPOSTAMI Z ODPADÓW KOMUNALNYCH I ZIELENI MIEJSKIEJ

### Abstrakt

W doświadczeniu wegetacyjnym analizowano wpływ kompostów wyprodukowanych z niesegregowanych odpadów komunalnych oraz z odpadów zieleni miejskiej na zawartość cynku w roślinach. Użyte komposty z odpadów komunalnych dojrzywały w pryzmach odpowiednio przez 1, 3 i 6 miesięcy. Zastosowano je w 3 dawkach: 10, 20 i 30 g kg<sup>-1</sup> gleby. Kompost z odpadów zieleni miejskiej dojrzywał w pryzmie 6 miesięcy, użyto go w ilości 10 g kg<sup>-1</sup> gleby. Stwierdzono, że zawartość cynku w roślinach była różnicowana w zależności od rodzaju użytego kompostu, wielkości dawek oraz gatunku uprawianej rośliny. Działanie zwiększonych dawek kompostów nie zawsze skutkowało wzrostem zawartości cynku w roślinach, a w niektórych przypadkach jego zawartość była niższa od stwierdzonej w obiekcie kontrolnym. Znaczny wzrost zawartości cynku w większości analizowanych roślin nastąpił po zastosowaniu świeżego kompostu z odpadów komunalnych po miesiącu dojrzewania w pryzmach. Nawożenie kompostem z odpadów zieleni miejskiej w przypadku kukurydzy i jęczmienia okazało się korzystniejsze niż identyczna dawka kompostu z odpadów komunalnych, ponieważ rośliny z tego obiektu zawierały mniej cynku. U pozostałych roślin: słonecznika, gorczycy i facelii zastosowanie tego kompostu spowodowało znaczący wzrost zawartości omawianego pierwiastka, w porównaniu z obiektami użyznanymi kompostem z odpadów komunalnych, w przypadku tego samego poziomu nawożenia.

Słowa kluczowe: odpady komunalne, odpady z zieleni miejskiej, kompost, cynk, roślina, gleba.

## INTRODUCTION

Elevated content of trace elements in soil may be a result of human activity. Progressing industrialization and increasing use of chemicals in agriculture contribute to soil pollution with trace elements. Waste of all types, including large amounts of organic waste, is another by-product of man's activity. It seems that the optimal form of organic waste utilization should be their introduction into natural processes (GONDEK, FILIPEK-MAZUR 2005, 2006). However, some organic waste may contain excessive quantities of heavy metals (FRITZ, VENTER 1988, RUTKOWSKA et al. 2003 a, b, SADEJ et al. 2003, 2004, RAMOS et al. 2004), which is their greatest disadvantage. Among such waste is municipal solid waste. Composts made from this type of waste used in overtly large rates can lead to an excessive concentration of heavy metals in soils (KABATA-PENDIAS, PIOTROWSKA 1987, CHWASTOWSKA et al. 1993).

These elements, once they have penetrated soil, are included in a food chain and can end up in a human organism (FERGUSON 1990). Zinc is an element counted as a heavy metal. It is used as a component of metal alloys, for making paints and batteries and in the poligraphic industry. The main source of zinc emission are colour metal smelting plants as well as combustion of coal and crude oil.

SALGUEIRO et al. (2000) emphasize that zinc is one of the major elements which influence functions of a human body. On a global scale, this microelement is deficient in a daily diet of people, which is a serious problem due to the role zinc plays in a human body at different stages of its development. SZLEGIEL-ZAWADZKA (2001) points to the fact that zinc deficit in a man's body can lead to a variety of disorders, both concerning body systems and single organs. As a micronutrient, zinc is necessary for proper metabolism of proteins and carbohydrates. It is also involved in insulin synthesis and in maintaining the balance between acids and alkalis in an organism.

Regarding its influence on soil, zinc is considered to be a metal producing unwanted burden on this non-renewable element of the natural environment (GORLACH, GAMBUŚ 2000). It is among the most mobile elements in soil and its plant availability depends on the soil's reaction, decreasing in proportion to an increase in the pH values of soil. In contaminated soils, this element will accumulate mainly in roots of plants. Chlorosis, listed among some typical signs of excessive amounts of zinc in plants, appears due to depressed efficiency of photosynthesis, leading to inhibited growth of affected plants (KABATA-PENDIAS, PENDIAS 1999). When excessive levels of zinc appear in soil, the yield of crops can be largely depressed (BARAN et al. 2008).

The objective of this study has been to evaluate the content of zinc in plants fertilized with composts made from unsorted municipal solid waste and urban green waste.

## MATERIAL AND METHODS

A pot experiment was conducted in a greenhouse of the University of Warmia and Mazury in Olsztyn. Kick-Brauckman pots were used for the trials, each filled with 10 kg of typical brown soil containing 1.12% organic matter. The soil had a texture of light loamy sand. The content of available phosphorus, potassium and magnesium was: 77 mg P, 170 mg K and 66 mg Mg per 1 kg of soil. The pH of the soil in 1 mol KCl was 5.50. The experiment consisted of 4 replicates.

The experiment compared the effects produced by two types of compost: made from mixed municipal solid waste generated according to the biothermic technology MUT-DANO, which matured in compost heaps for 1, 3 and 6 months, and made from urban green waste, which aged in a heap for 6 months. The composts for the experiment were taken on the same day, but they differed in the ageing time, therefore the initial material from which they were made was not identical but had similar characteristics. The agrochemical evaluation of the value of the municipal waste composts in different maturity stages had been presented in our previous paper (SADEJ et al. 2003). The composts made from municipal waste were applied in three rates:

10, 20 and 30 g kg<sup>-1</sup> of soil, which corresponds to 30, 60 and 90 t per 1 ha, whereas the green waste compost was introduced to soil in the amount of 10 g kg<sup>-1</sup> (30 t ha<sup>-1</sup>). The experiment was carried out for three years. In the first year, maize (*Zea mays* L.) and sunflower (*Helianthus annuus* L.) were grown, followed by spring barley (*Hordeum vulgare* L.) and white mustard (*Sinapis alba* L.) in the second year and lacy phacelia (*Phacelia tanacetifolia* Benth.) in the third year. The plants were harvested in the following maturity phases: maize during the panicle emergence phase, sunflower in the early inflorescence, spring barley in the phase of shooting, white mustard and lacy phacelia during the full inflorescence phase. In the third year of the experiment, mineral fertilization was applied to all treatments, in the following rates (in mg kg<sup>-1</sup> of soil): 74 N, 31 P and 109 K.

Immediately after the plant harvest, the yield and dry matter content of the plants were determined. The content of zinc in the plant material, composts and soil was determined using the atomic emission spectrophotometric method with inductively induced plasma, in an ICP-AES apparatus (manufactured by Leeman Labs), having previously mineralized the material in a 5 : 4 mixture of HNO<sub>3</sub> and HClO<sub>4</sub> in a heating block (manufactured by VEL) (OSTROWSKA et al. 1991).

The content of zinc in the soil used for the experiments (31.92 mg kg<sup>-1</sup> d.m.) was assessed as natural according to the criteria elaborated by the IUNG in Puławy (KABATA-PENDIAS et al. 1993). The content of this element was however variable. The fresh compost made from municipal waste contained 934.5 mg Zn kg<sup>-1</sup> d.m., but after a month's maturation its content rose to 1041.2 mg Zn kg<sup>-1</sup> d.m. The highest concentration of zinc was found in the compost maturing for three months (1340.60 mg Zn kg<sup>-1</sup> d.m.). After the next three months in a heap, the compost contained over 300 mg Zn kg<sup>-1</sup> d.m. less zinc, whose concentration fell to 1031.35 mg Zn kg<sup>-1</sup> d.m. The compost made from urban green waste which was kept in a heap for 6 months contained 649.55 mg Zn kg<sup>-1</sup> d.m., i.e. about 1/3 less than the municipal waste composts. The above differences were most probably an effect of the differences in the properties of the initial material used for production of the composts.

## RESULTS AND DISCUSSION

The lowest rates of the composts tested in the trials led to an increase in the dry matter yield of maize, sunflower and barley but depressed that of white mustard and phacelia (Table 1). The response of these plant species to increased rates of municipal waste composts was similar. It was demonstrated that the crops tended to respond more positively to the fertilization with the one- and three-month-old composts than with the compost which aged

Table 1

Yield of dry mass of aerial parts of plants (g pot<sup>-1</sup>)

Dose of compost (g kg <sup>-1</sup> soil)		Species of cultivated plant				
		maize	sunflower	spring barley	white mustard	phacelia tanacetifolia
		year of cultivation				
		I		II		III
Control 0		58.8	15.9	9.9	4.4	47.2
Municipal solid waste compost heap-stored for 1 month						
10		69.5	16.4	14.1	3.7	43.5
20		87.6	19.9	14.3	3.2	46.3
30		85.2	17.9	14.6	3.1	48.8
Municipal solid waste compost heap-stored for 3 months						
10		72.9	16.1	14.3	4.4	45.8
20		86.7	14.3	15.4	4.5	44.9
30		81.9	28.3	16.4	3.8	45.6
Municipal solid waste compost heap-stored for 6 months						
10		71.0	16.0	12.9	3.8	42.9
20		74.6	21.3	15.7	3.2	42.7
30		84.4	19.6	14.5	3.2	48.9
Green waste compost						
10		63.2	23.3	13.4	3.1	47.0
LSD <sub>0.05</sub>	dose of compost (d)	2.6	1.2	1.2	0.3	1.5
	phase of maturing (f)	1.5	0.7	n.s.	0.2	n.s.
	interaction: d·f	4.6	2.1	n.s.	0.6	2.6

for 6 months, and the fertilization with the compost maturing for one month produced the strongest response in maize and phacelia, whereas sunflower, barley and mustard were more responsive to the three-month-old compost. Fertilization treatments with six-month-old compost produced a weaker response in maize, mustard and phacelia compared to the other composts, while sunflower and barley responded better to the fertilization with six-month-old compost.

In respect of the yield of aerial parts of sunflower, barley and phacelia, the green waste compost was less effective than the municipal waste compost which matured for the same period of time. In the case of maize and mustard, the effect produced by the former compost was less favourable.

The results obtained in the present experiment are similar to those reported by JANKOWSKI (1997), although in his study the positive effect of compost application was also observed in the subsequent years of the trials. Increased dry matter yield of crops fertilized with municipal waste compost was also noticed by ZHELJAZKOV and WARMAN (2003). ŁABĘTOWICZ et al. (2002) concluded that dry matter yields rose as the applied rates of compost were higher, a tendency that was also observed in the present study. In turn, LEKAN et al. (1997) proved that dry matter yield of plants was significantly higher when the rate of municipal waste compost was as high as 200 t ha<sup>-1</sup>.

Content of heavy metals is an important indicator of the heavy metal contamination of composts, especially the ones which are to be used in agriculture. With respect to zinc, the analyzed composts were characterized by very high quality as in none of the tested material the permissible quantity of zinc established by the Industry Standards for class I composts, i.e. 1500 mg Zn kg<sup>-1</sup>, was exceeded.

Fertilization treatments involving the analyzed composts caused changes in the content of zinc in the test crops, which depended on the species of a grown crop and, to a lesser extent, on the type of compost applied.

The effect of municipal and urban green waste composts in the first year after their application led to an increase in the content of zinc, both in maize (Figure 1) and in sunflower (Figure 2). The content of zinc in the aerial biomass of both crops increased as the applied rates of composts rose, but the relative changes were smaller in sunflower. The effect of the compost maturity phase on the content of this metal was varied. Using the municipal waste compost that had the shortest maturation time (1 month) caused the highest increase in the content of zinc, both in maize and in sunflower. Application of composts which matured in heaps for longer time

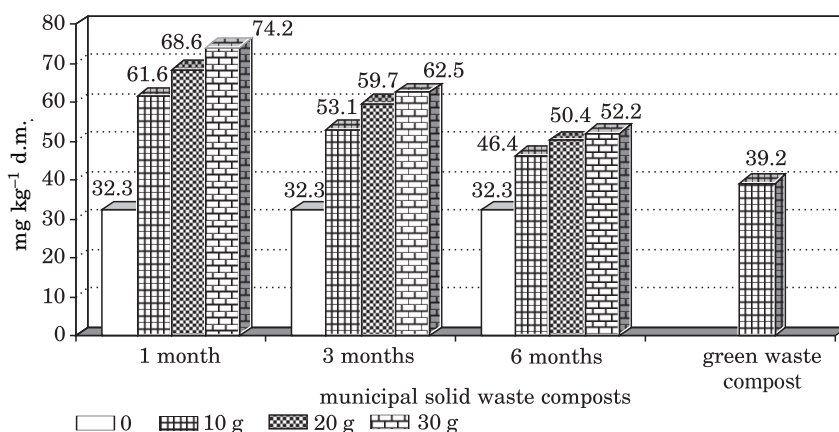


Fig. 1. Content of zinc in aerial parts of maize (*Zea mays* L.)

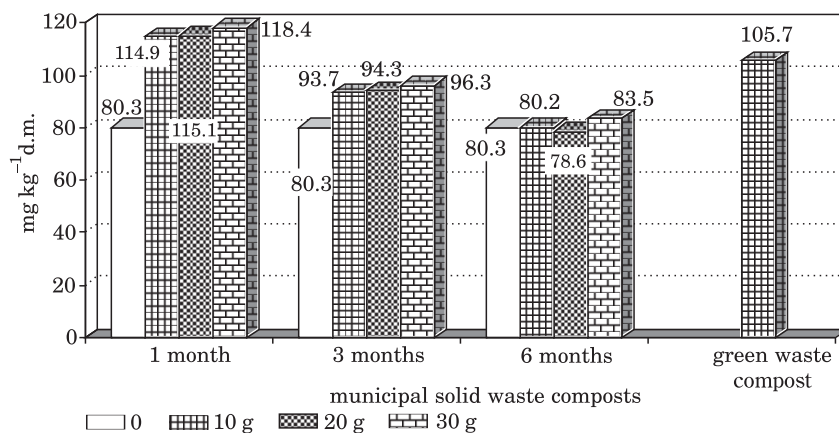


Fig. 2. Content of zinc in aerial parts of sunflower (*Helianthus annuus* L.)

led to a successive decrease in the concentration of zinc in both crops, which was a result of the fact that the yield of these plants was lower when older composts had been introduced to soil. Of the two test plants, much more zinc was determined in sunflower than in maize, and this tendency was observed in all fertilization treatments. The results of the tests are in accord with the proven thesis that dicotyledons contain more heavy metals than monocotyledons (GAMBUŚ, GORLACH 1996). PETRUZZELLI et al. (1989) demonstrated that compost fertilization led to altered levels of zinc in maize, with its concentration being dependent on plant organs. More zinc was found in roots than in kernels.

Maize fertilized with urban green waste compost accumulated less zinc than maize nourished with an identical rate of municipal waste compost. In respect of sunflower, the former compost caused an increase in zinc concentration in plants relative to all the rates of composts made from municipal waste and maturing for 3 and 6 months.

In the subsequent year of the experiment, the effect of composts was different than in the first year. Application of municipal waste composts tended to depress the content of zinc in the biomass of spring barley, below the level observed in the control pots (Figure 3). The most profound effect, reaching 20%, was recorded after the lowest rate of composts was applied. Slightly more zinc than in the control was found only in the object fertilized with 1-month-old compost added to soil at a rate of 20 g kg<sup>-1</sup> soil. Analogously, application of compost made from green waste depressed the content of zinc in spring barley biomass compared to its concentration in control plants. This effect was similar to the results of using identical doses of municipal waste composts maturing in heaps for 1 and 6 months.

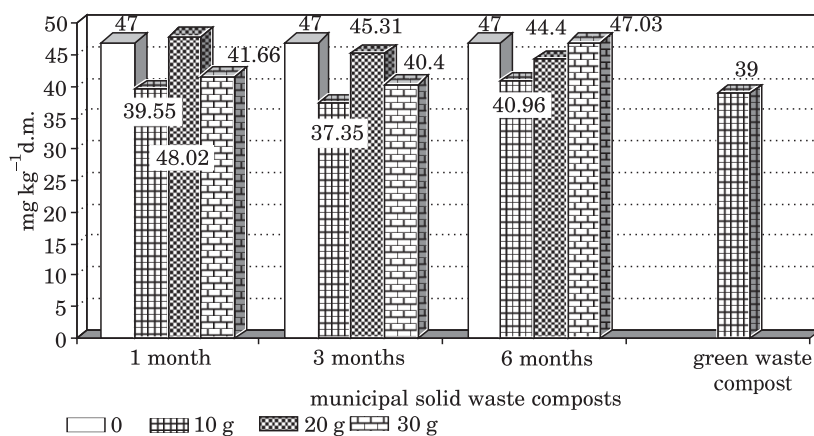


Fig. 3. Content of zinc in aerial parts of spring barley (*Hordeum sativum* L.)

Regarding white mustard, fertilization with municipal waste composts introduced to soil in a rate of 10 g kg<sup>-1</sup> of soil depressed the content of zinc by about 24 to 54% compared to the content of this metal in the control plants (Figure 4). No unidirectional changes in the content of Zn caused by the larger rates of composts were found, except higher doses of 1-month-old compost depressing the content of zinc in plants to a similar extent, by about 30% compared to the lowest rate. The effect of the maturation period on the content of zinc in plants was in general similar to that observed in the first year of the tests, namely the more mature the compost, the lower the average content of zinc in the biomass of white mustard. Application of urban green waste compost produced an almost identical

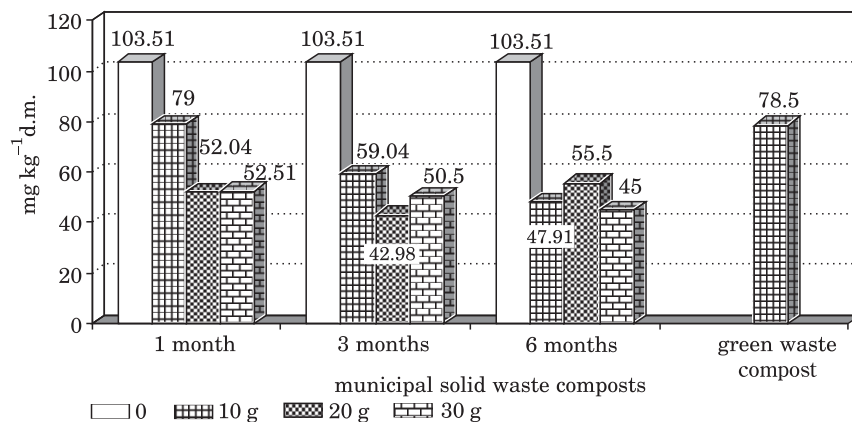


Fig. 4. Content of zinc in aerial parts of white mustard (*Sinapis alba* L.)



effect to that of the lowest rate of municipal waste compost, remaining in a heap for one month, i.e. it lowered the content of zinc in mustard by about 24% versus the control plants, but the decrease was much smaller than produced by the same rate of six-month-old compost. This response of both plant species was observed although fertilization with composts had a different effect on their yields. Sunflower, for example, produced 30 to 60% higher yields whereas the yields of mustard did not change much or else dropped by 14 to 30% under the influence of composts.

In the third year of the experiment, when composts had been added to soil, the response of one of the test plants, namely lacy phacelia, was different from that observed in other plants grown immediately after the introduction of composts to the substrate or in the second year. Fertilization with municipal waste composts generally depressed the content of zinc in the biomass of lacy phacelia, but the differences were smaller than in the plant species discussed earlier in this paper (Figure 5). The largest decline in the concentration of zinc in plant tissues, reaching 18%, followed a fertilization treatment with 1-month-old compost. Among the objects fertilized with municipal waste compost, it was only the three-month-old compost applied in the lowest rate that caused an increase in the zinc concentration in phacelia biomass by about 6% compared to the plants from the control pot. Among all the objects fertilized with the lowest compost doses, the highest content of zinc, 18% higher than in the control plants, was observed when urban green waste compost had been applied, and the yields from both treatments were on a similar level. This response was different than the one noticed in the previous years for maize, sunflower and barley, but the same as occurred in white mustard.

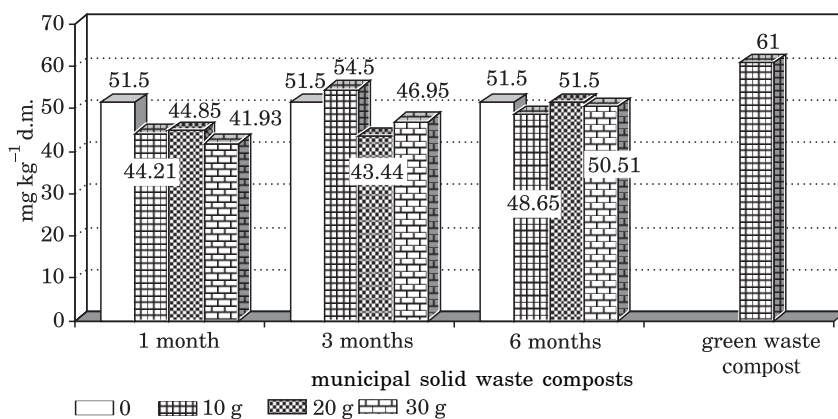


Fig. 5. Content of zinc in aerial parts of phacelia (*Phacelia tanacetifolia* L.)

Many authors (JANKOWSKI 1997, ŁABĘTOWICZ et al. 2002) suggest that, in general, application of municipal waste composts significantly increased the content of heavy metals in plants, in proportion to the increasing rates of such fertilizers. The authors' own results are partly in agreement with the cited references, as the increasing doses of composts did not invariably lead to an increase in the zinc content in plants. LEKAN et al. (1997) claim, however, that fertilization with municipal waste composts leads to higher concentrations of zinc in plants. Similar conclusions are formulated by STĘPIEŃ et al. (2006).

CASTRO et al. (2009) proved that fertilization with municipal waste composts significantly increased the content of zinc in lettuce versus the control, with the highest content of this element found in the second year of growing lettuce. GAJ and GÓRSKI (2004) demonstrated that composts introduced to soil raised the content of zinc in sugar beet but did not deteriorate the quality of the yield. The content of this element was varied and depended on the applied dose of compost. These authors determined that more Zn accumulated in sugar beet roots than in leaves. SCHUMAN et al. (2001) found out that compost fertilization led to a decrease in the content of zinc in plants, which meant that its toxicity declined, too.

The present experiments have clearly pointed to the need of including the content of heavy metals in composts into calculations of applicable rates of composts. This conclusion is confirmed by a simplified zinc balance, worked out during the study, which suggests that application of larger rates of composts leads to increased amounts of zinc in soil, even several years after a given fertilization treatment (Table 2). Zinc uptake by plants depended also on the type and rate of applied composts, species-specific characteristics of plants and volume of the yield they produced. The highest uptake of zinc was observed for the plants grown on soil fertilized with composts left in heaps for the shortest period of time. When the three-year-long experiment was terminated, most zinc was found remaining in the soil fertilized with 3-month-old compost, which was a consequence of the initial content on zinc in this compost (Table 2).

WEBER et al. (2002) believe that the content of heavy metals in soil depends on the soil's type and the type of compost applied. Soils poor in organic matter are susceptible to pollution with heavy metals, but the organic matter introduced to such soils with the applied compost contributes to decreasing the amounts of heavy metals assimilated by plants growing on the fertilized soil. Moreover, quantities of heavy metals taken up by plants, apart from the type of soil and compost, depend on the species or even an organ of a cultivated crop.

GIGLIOTTI et al. (1996) demonstrated that compost fertilization caused a significant increase in the content of zinc in soil compared to an unfertilized treatment. Similar conclusions were drawn by GAJ and GÓRSKI (2004). SMITH (2009) states that heavy metals in composts are strongly associated

Table 2

Simplified balance of zinc for three years (mg pot<sup>-1</sup>)

Dose of compost (g kg <sup>-1</sup> soil)	Zn added to soil with composts (mg pot <sup>-1</sup> )	Zn uptake by plants (mg pot <sup>-1</sup> )	Difference of balance (mg pot <sup>-1</sup> )
Control 0	-	6.53	-6.53
Municipal solid waste compost heap-stored for 1 month			
10	104.12	8.94	95.18
20	208.24	11.23	197.01
30	312.36	11.26	301.10
Municipal solid waste compost heap-stored for 3 months			
10	134.06	8.67	125.39
20	268.12	9.40	258.72
30	402.18	10.84	391.34
Municipal solid waste compost heap-stored for 6 months			
10	103.14	7.37	95.77
20	206.28	8.51	195.77
30	309.42	9.34	300.08
Green waste compost			
10	64.96	8.57	56.39

with organic matter, which reduces the share of their soluble forms and plant availability. Composting is a process which leads to reducing the bioavailability of heavy metals, therefore the environmental threat to human health or such as polluting fertilized soil and cultivated plants is minimal.

The results of the experiment have proven that plants fertilized with the tested composts did not contain excessive amounts of zinc and could be used as fodder plants, as the concentration of zinc attested in these crops was within the permissible range values as stated in the Framework Guidelines for Agriculture (KABATA-PENDIAS et al. 1993).

## CONCLUSIONS

1. With respect to the amount of zinc, the analyzed composts were characterized by high quality, as none of them contained the amounts of zinc that would exceed the quantities established by the Industrial Standards for composts classified as class I. Green waste compost contained 30% less zinc than municipal waste composts.

2. The content of zinc in plants depended on a plant species and volume of dry matter yield as well as the type and rate of the applied fertilizer. The highest increase in the content of this bioelement in the biomass of most of the test plants was observed as a result of the application of 1-month-old municipal solid waste compost. The effect produced by the higher rates of composts did not invariably cause an increase in the zinc content.

3. Fertilization with urban green waste compost caused smaller accumulation of zinc in the biomass of maize and barley, but in the other crops (sunflower, white mustard, lacy phacelia) it typically led to a considerable increase in the content of this metals compared with identical rates of municipal waste composts.

4. Having completed the cycle of our trials, most zinc remaining in soil was found in the treatment consisting of fertilization with three-month-old municipal waste compost. In the soil fertilized with urban green waste compost there was about 50% less zinc than in the soil to which an identical rate of 6-month-old municipal waste composts was introduced. This was a consequence of the fact that there was less zinc in green waste compost.

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# **USEFULNESS OF CALCIUM-MAGNESIUM PRODUCTS IN PARTURIENT PARESIS IN HF COWS**

**Przemysław Sobiech<sup>1</sup>, Krzysztof Rypuła<sup>2</sup>,  
Barbara Wojewoda-Kotwica<sup>2</sup>, Sylwester Michalski<sup>2</sup>**

**<sup>1</sup>Chair of Internal Diseases**

**University of Warmia and Mazury in Olsztyn**

**<sup>2</sup>Department of Epizootiology and Veterinary Administration  
with Clinic of Infectious Diseases**

**University of Environmental and Life Sciences in Wrocław**

## **Abstract**

The increased milk yield in dairy cattle has made it increasingly difficult to balance accurately the feed ration. This results in energy and mineral deficiencies as well as various homeostasis disorders including calcium and magnesium balance disorders, which in turn lead to metabolic disorders. In our study, we chose to assess the therapeutic efficacy of two most popular products of this type i.e. Glucalvet (Vetoquinol Biowet) and Antiparen-N (Vetoquinol Biowet). The study was conducted on 33 HF cows displaying symptoms of post-parturient paresis that occurred between 2 and 4 day postpartum. The cows were divided into two groups: group A and group B, which were administered Glucalvet and Antiparen-N, respectively. Blood samples were taken twice from the jugular vein of each animal – just before and 24 hours after drug administration. The samples were used to determine serum concentrations of Ca, Mg, P, Cl and glucose. The average body temperature in both groups was slightly below normal and statistically significantly increased 24 hours after the intravenous infusion. The same tendency was observed for the pulse rate. The magnesium levels in the first group improved significantly after drug administration but its level was still lower than the physiological range. In the second group of animals, a slight drop in the Mg level after the Antiparen-N administration. Levels of Ca and P increased in both group of animals after the treatment. The average chloride concentration was similar for both assays. The serum glucose concentration increased significantly in both groups of animals after drug administration. Summing up, the study presented in this paper indicates that Glucalvet and Antiparen-N prove to be useful in the treatment of post-parturient paresis conditioned by Ca and Mg deficiency.

**Key words:** cow, parturient paresis, calcium-magnesium, Glucalvet, Antiparen-N.

## PRZYDATNOŚĆ STOSOWANIA PREPARATÓW WAPNIOWO-MAGNEZOWYCH W ZALEGANIU OKOŁOPORODOWYM KRÓW

### Abstrakt

Wysoka produkcyjność krów mlecznych sprawia trudności we właściwym zbilansowaniu dawki pokarmowej. Stosowanie nieadekwatnej do potrzeb dawki pokarmowej powoduje wiele zaburzeń w homeostazie ustroju, z których najczęstsze są problemy energetyczne i zaburzenia mineralne, w tym niedobory wapnia i magnezu. Celem badań było określenie skuteczności terapeutycznej dwóch najbardziej popularnych infuzyjnych leków stosowanych w zaburzeniach mineralnych – Glucalvetu i Antiparenu-N. Badania wykonano na 33 krowach rasy HF wykazujących objawy zalegania poporodowego pojawiającego się 2-4 dni po porodzie. Krowom podzielonym na 2 grupy A i B podawano odpowiednio Glucalvet i Antiparen-N. Od wszystkich zwierząt pobrano 2-krotnie krew do badań – tuż przed podaniem leków i 24 h po ich aplikacji. Zwierzęta poddano badaniom klinicznym, natomiast w zakresie parametrów laboratoryjnych oznaczono surowiczy poziom glukozy, wapnia, magnezu, fosforu, chlorków. Średnia temperatura ciała i liczba tętna przed podaniem leków była nieznacznie obniżona, natomiast po aplikacji statystycznie wzrosła. Koncentracja Mg wzrosła istotnie u krów z grupy pierwszej po podaniu leku, ale pozostawała nadal poniżej wartości referencyjnych. W grupie drugiej, po podaniu preparatu Antiparen-N, zaobserwowano nieznaczny spadek poziomu Mg. Stężenie Ca i P wzrosło u wszystkich zwierząt po aplikacji preparatów, natomiast koncentracja chlorków nie uległa zmianom w ciągu badanego okresu. U wszystkich krów zaobserwowano wzrost poziomu glukozy w 24 h po podaniu preparatów. Wyniki wskazują na to, że stosowanie preparatów Glucalvet i Antiparen-N jest skuteczne w leczeniu porażenia poporodowego i niedoborów wapnia i magnezu u krów mlecznych.

Słowa kluczowe: krowy, zaleganie poporodowe, wapń-magnez, Glucalvet, Antiparen-N.

## INTRODUCTION

Higher milk yield in dairy cattle has made it increasingly difficult to balance accurately the feed ration. The result is energy and mineral deficiencies as well as various homeostasis disorders, including calcium and magnesium balance disorders. These disorders in turn lead to metabolic disorders, such as pre-parturient, parturient and post-parturient paresis caused by prolonged calcium deficiencies, spring grass tetany, winter tetany and whole milk tetany of calves caused by reduced magnesium levels (STEC 1985). One of the problems occurring most often in dairy cattle in the perinatal phase is subclinical and clinical hypocalcemia. Apart from typical symptoms, reduced humoral and cellular resistance can be observed. Other symptoms include decreased rumen motor activity and abomasum motor activity and lower feed intake which increases the risk of abomasum displacement (KIMURA et al. 2006, DEGARIS, LEAN 2008). GOFF (2008) points to a decrease in muscle contractility including the teat sphincter muscle.



There are a lot of medicinal products available that are used to remove the symptoms of parturient paresis in cows. They all differ in their composition and therapeutic properties. The study described in the paper are an attempt at assessing the therapeutic efficacy of two most popular products of this type i.e. Glucalvet (Vetoquinol Biowet) and Antiparen-N (Vetoquinol Biowet). Apart from this, changes in the mineral element concentrations, glucose and chloride concentrations were examined in the animals afflicted with post-parturient paresis following intravenous infusion of the products in question.

## MATERIALS AND METHODS

The study was conducted on 33 HF cows displaying symptoms of post-parturient paresis which occurred between 2 and 4 day postpartum. The cows were divided into two groups: group A and group B, which were administered Glucalvet and Antiparen-N respectively. A total of 15 cows received Glucalvet whereas Antiparen-N was administered to 18 cows.

The products used in the treatment are both solutions for intravenous use. Their composition is as follows:

- Glucalvet – hypertonic solution containing: calcium chloride (8.0 g), magnesium chloride (3.0 g), anhydrous glucose (5.0 g), disodium edetate (0.012 g), ferrous sulfate (0.00675 g), copper sulfate (0.0016 g), cobaltous chloride (0.00027 g) and water for injection (up to 100.0 cm<sup>3</sup>);
- Antiparen-N – hypertonic solution containing: calcium chloride (8.0 g), magnesium chloride (1.0 g), sodium chloride (2.0 g), glucose (4.0 g) and water for injection (up to 100.0 cm<sup>3</sup>).

Both products were given in a single administration in accordance with the manufacturer's recommendations, i.e. after warming the solution to body temperature and determining the dose at 1 cm<sup>3</sup> kg<sup>-1</sup> bw. The rate of infusion was around 20-50 cm<sup>3</sup> min<sup>-1</sup>.

All the animals were put under observation, during which the following functions were assessed: appetite and thirst, motor activity, severity of paresis (standing position, sternal recumbent position – animals are conscious, lateral recumbent position – animals are unconscious), body temperature, pulse rate and breathing rate.

Two blood samples were taken from the jugular vein of each animal – the first one directly after the diagnosis and prior to drug administration and the second one 24 hours after drug administration. The samples were used to determine the concentrations of calcium, magnesium, phosphorus, chlorides and glucose in the serum. The assays were done in a Cobas Mira S chemical analyzer (Roche, France).

The results were analyzed statistically with Wilcoxon signed-rank test. The arithmetic average, standard deviation and significance level with the threshold  $p$  value of = 0.05 ( $*p < 0.05$ ,  $**p < 0.01$ ,  $***p < 0.001$ ) were determined.

## RESULTS

Within a few hours following drug administration, noticeable improvement in the general state of health was observed in all group A animals. The cows started to assume standing position, the feed intake increased and the milk yield improved. On the second or third day, the treated animals started to recover to normal health.

Prior to the treatment, the average body temperature in this group was slightly below normal and 24 hours following the intravenous infusion of Glucalvet, a statistically significant increase to 38.8°C was observed. Similarly, the breathing rate rose from 13 min<sup>-1</sup> prior to treatment to 17 min<sup>-1</sup> following treatment. The same tendency was observed for the pulse rate where a significant increase from 42 to 54 heart beats/minute was noted (Figures 1-3).

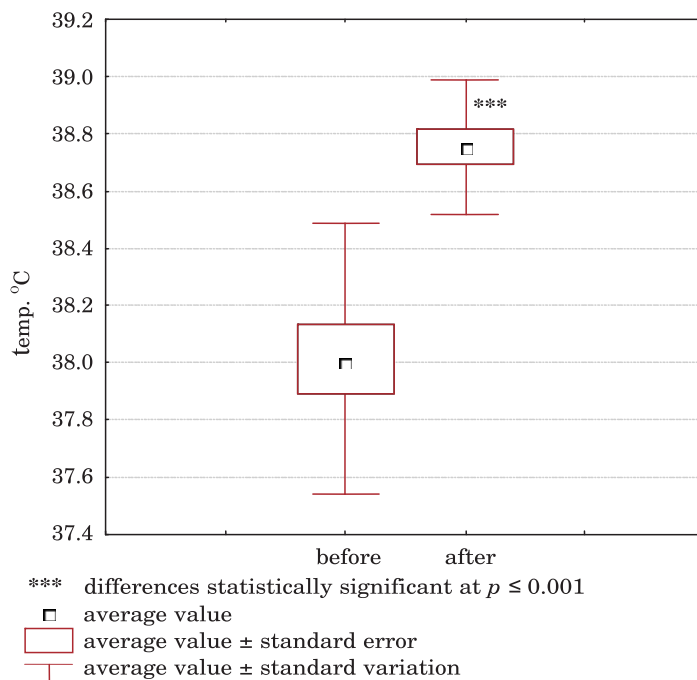


Fig. 1. Body temperature dynamics in cows before and after administration of Glucalvet

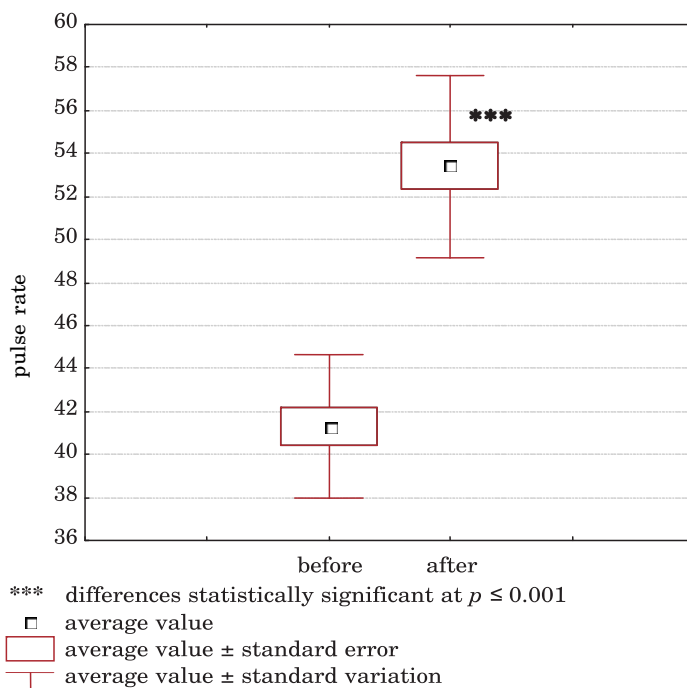


Fig. 2. Pulse rate dynamics in cows before and after administration of Glucalvet

Prior to the administration of Glucalvet, the calcium concentration in blood serum was  $1.33 \text{ mmol dm}^{-3}$ , whereas afterwards it rose to  $1.77 \text{ mmol dm}^{-3}$ . In both cases, the assayed calcium concentration was very low and even after the infusion it failed to achieve the normal physiological value. The phosphorus concentration also increased following the infusion of Glucalvet and achieved the value of  $0.87 \text{ mmol dm}^{-3}$ . This increase was not statistically significant, unlike the calcium concentration increase. The magnesium levels, in turn, improved significantly from  $0.77 \text{ mmol dm}^{-3}$  to  $0.92 \text{ mmol dm}^{-3}$ , i.e. they were at the lower end of the normal physiological range (Table 1).

The average chloride concentration was similar for both assays. It remained within the normal physiological range and was  $100.23 \text{ mmol dm}^{-3}$  following the drug infusion (Table 1).

Similarly, the glucose concentration in blood serum increased significantly from  $1.81 \text{ mmol dm}^{-3}$  to  $3.04 \text{ mmol dm}^{-3}$ . Before the cows were administered Glucalvet, their average glucose concentration was below the normal physiological value but following the drug infusion it was within the physiological range (Table 1).

Similar observations were made in group B. A few hours following the administration of Antiparen-N, noticeable improvement in the general health

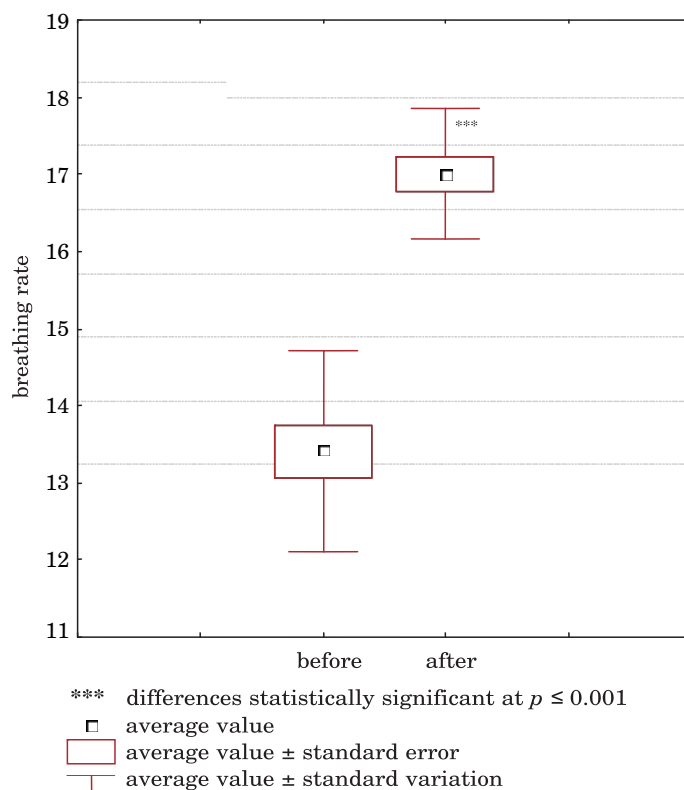


Fig. 3. Breathing rate dynamics in cows before and after administration of Glucalvet

state of the animals was noted. Recovery to the normal condition was observed 2 to 3 days following the infusion. Following treatment with Antiparen-N, the improvement of the clinical indicators such as body temperature, breathing rate and pulse rate was as significant as that observed in the cows treated with Glucalvet (Figures 4-6).

The calcium concentration in these cows prior to treatment was very low i.e.  $1.24 \text{ mmol dm}^{-3}$ , whereas following the treatment, it rose substantially to achieve  $1.77 \text{ mmol dm}^{-3}$ . The phosphorus concentration showed a similar increase from  $0.75 \text{ mmol dm}^{-3}$  to  $1.11 \text{ mmol dm}^{-3}$ . However, in this group a slight drop in the magnesium concentration from  $0.79$  to  $0.74 \text{ mmol dm}^{-3}$  was observed, in contrast to the Glucalvet-treated group. Another similarity between groups B and A was that in both groups the concentration of chlorides remained the same. It was  $101.18 \text{ mmol dm}^{-3}$  following the administration of Antiparen-N. The glucose concentration in turn increased considerably from  $1.51 \text{ mmol dm}^{-3}$  prior to treatment to  $2.43 \text{ mmol dm}^{-3}$  following the Antiparen-N administration. This increase, however, was not as high as that observed in group A animals (Table 1).

Table 1

Average values and standard deviation of calcium, phosphorus, magnesium and chloride concentrations and glucose levels in animals treated with Glucalvet and Antiparen-N

Assay	Ca (mmol dm <sup>-3</sup> )	Pin (mmol dm <sup>-3</sup> )	Mg (mmol dm <sup>-3</sup> )	Cl <sup>-</sup> (mmol dm <sup>-3</sup> )	Glucose (mmol dm <sup>-3</sup> )
Glucalvet					
Before administration	1.33 ± 0.30	0.72 ± 0.58	0.77 ± 0.28	100.25 ± 0.96	1.81 ± 1.13
After administration	1.77* ± 0.59	0.87 ± 0.45	0.92** ± 0.27	100.23 ± 0.83	3.04** ± 0.24
Antiparen-N					
Before administration	1.24 ± 0.44	0.75 ± 0.45	0.79 ± 0.29	100.51 ± 2.32	1.51 ± 0.85
After administration	1.77* ± 0.78	1.11* ± 0.57	0.74 ± 0.22	101.18 ± 3.26	2.43** ± 0.91

\* differences statistically significant at  $p \leq 0.01$

\*\* differences statistically significant at  $p \leq 0.001$

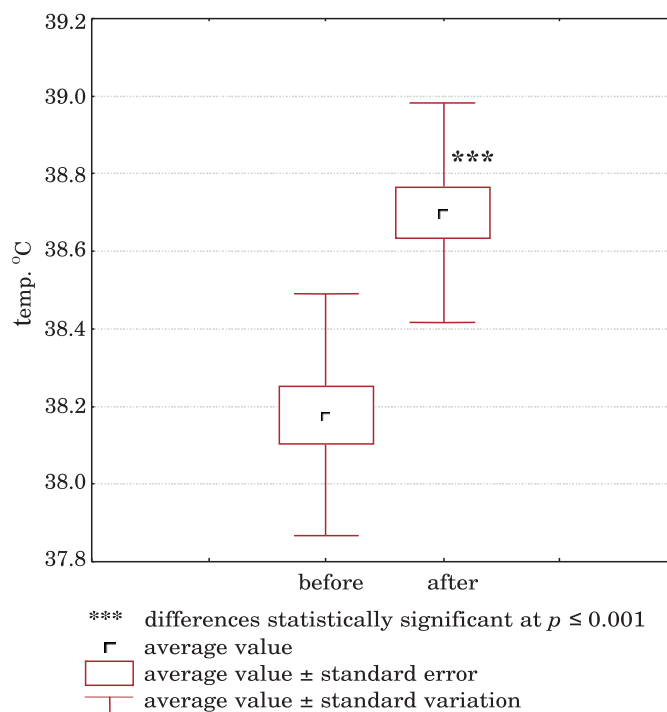


Fig. 4. Body temperature dynamics in cows before and after administration of Antiparen-N

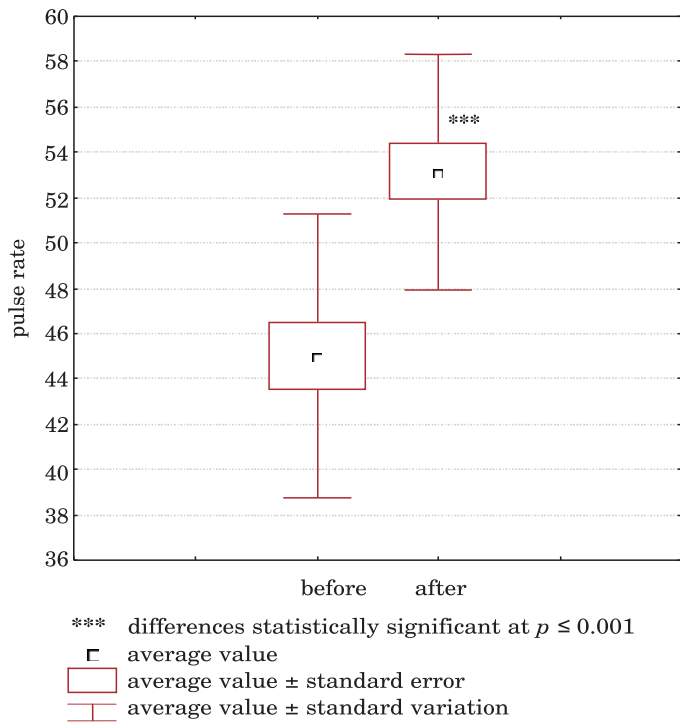


Fig. 5. Pulse rate dynamics in cows before and after administration of Antiparen-N

DISCUSSION

Based on the clinical data collected during the study, the body temperature of all the animals suffering from parturient paresis was below normal. In both groups, the average temperature was around 38.1°C. Lower body temperature is a characteristic symptom of parturient paresis and is observed in 60% of animals afflicted with this condition (BRAUN et al. 2004). An increase in body temperature observed after the treatment with Glucalvet and Antiparen-N is a symptom of recovery and proves the health benefits of both products. Significantly higher breathing and pulse rates, albeit within the normal physiological range, can also be observed in cows treated with calcium products (HOUE et al. 2001).

The levels of mineral elements (calcium, phosphorus, magnesium) in the blood serum of animals with symptoms of post-parturient paresis from both groups were significantly low. The concentration of these elements was practically half the physiological level. Similar concentrations were observed by GOFF (2007) and LEAN et al. (2006). BRAUN et al. (2006) demonstrated that

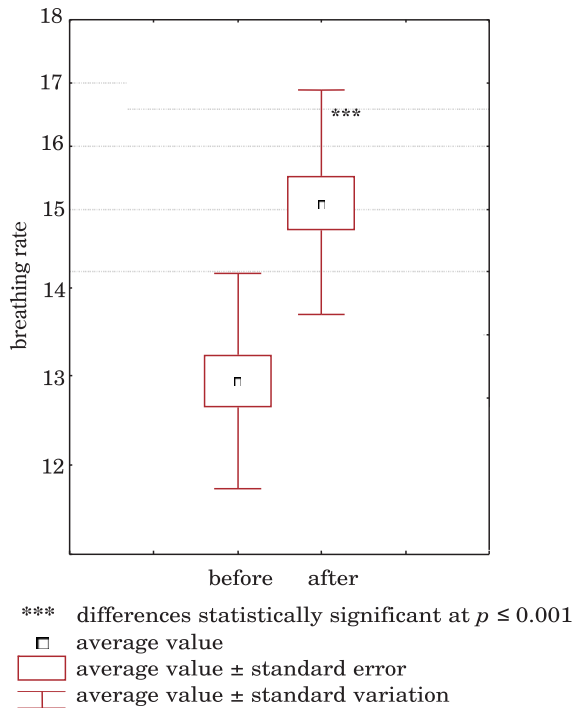


Fig. 6. Breathing rate dynamics in cows before and after administration of Antiparen-N

such drastically low concentrations of calcium and phosphorus in the blood serum can be observed in 90% of cows with post-parturient paresis. Intravenous infusion of Glucalvet and Antiparen-N brought about a significant increase in calcium levels. However, these concentrations were still low and remained below the normal physiological levels. Higher blood serum Ca concentration observed after the infusion of both products is attributed to calcium chloride they contained. However, low Ca levels observed after 24 hours following the administration indicate that the animals were still calcium-deficient. This suggests that Ca re-supplementation might be required. BRAUN et al. (2004) demonstrate that only repeated parenteral administration of calcium products to cows suffering from post-parturient paresis helps to achieve the normal Ca levels in body fluids.

After treatment with either of the products, phosphorus levels were higher although the increase was statistically significant only after the infusion of Antiparen-N. Higher serum concentration of phosphorus in the cows from both groups was probably linked to its greater mobilization from the bones, which in turn brought about an increase of serum concentration. Full investigation of these mechanisms would however require some further studies conducted on more animals.

The magnesium concentrations in the blood serum of cows suffering from post-parturient paresis fall far short of normal levels, which is confirmed by the findings of other authors (ANDRESEN et al. 1999, BEDNAREK et al. 2000). These authors believe that magnesium deficiencies play a prominent role in the development of the condition in question. Hypomagnesaemia has direct effect on calcium metabolism by decreasing the secretion of parathyroid hormone in hypocalcaemia and making tissue receptors less sensitive to parathyroid hormone (RUDE 1998). Additionally, magnesium deficiency causes impairment of intestinal absorption of calcium since Ca absorption is promoted by the active metabolite of vitamin D<sub>3</sub>. This metabolite is formed during hydroxylation, which depends on an appropriate concentration of magnesium ions (GOFF 2008). The data obtained from the present experiment showed a statistically significant increase in the serum concentration of magnesium following the administration of Glucalvet and its slight decrease following the infusion of Antiparen-N. In the first case, the increased magnesium concentrations are due to high Mg supplementation (100 cm<sup>3</sup> of Glucalvet contains 3 g of magnesium chloride). No such increase was observed following the treatment with Antiparen-N, which can be attributed to its lower content in the product. The study conducted by DAUNORASA et al. (2008) demonstrated that intravenous administration of magnesium compounds causes the serum magnesium concentration to rise in the first 2 hours following the administration, after which its levels go down. In the present study, magnesium was assayed 24 hours following supplementation and it appears that after the infusion of both products a temporary increase in the serum magnesium concentration occurred. These increased levels continued to be observed in the blood samples collected later only in the Glucalvet-treated animals because this product is richer in magnesium. BRAUN et al. (2007) demonstrated that even a tiny amount of intravenously infused magnesium significantly increases its serum concentration in cows. Therefore, better results of treatment of parturient paresis are achieved when calcium and magnesium are administered parallel than if only the former is administered (LEAN et al. 2006).

The concentration of chlorides both prior and following the administration of the products was similar in the treated cows. The chloride anion is stable in the body and its levels rarely fluctuate. HORST et al. (1997) found out that feed supplemented with chlorine and sulphur compounds boosted the treatment of parturient paresis and helped to prevent its development. On the other hand, higher concentration of sodium and potassium is one of the factors contributing to the development of this condition. The beneficial mechanism of higher concentrations of chloride ions in the treatment of hypocalcaemia consists in inducing slight ketosis, which promotes calcium and phosphorus mobilization from the bones (GOFF et al. 2007). Both prior and following the administration of the products, the concentration of chlorides in the cows remained within normal physiological limits, which seems



to suggest that in this particular case it did not affect the pathogenesis of parturient paresis.

BEDNAREK et al. (2000) demonstrated that energy deficiencies play a vital role in the pathogenesis and development of parturient paresis. Low glucose levels were also observed in the animals included in the present studies. Glucose concentration rose significantly following the administration of both products. Glucose facilitates the use of macro- and micronutrients by the body and provides extra energy, whose deficiency contributes to the development of metabolic diseases e.g. ketosis. Increased post-partum milk production in cows leads to excessive use of energy reserves. This in turn can cause the limited reserves of magnesium to be depleted and lead to chronic lesions manifested by Mg deficiencies and ketosis. Glucose levels in the cows we studied were decidedly higher following the administration of both products, which indicates that the energy supply was sufficient.

Summing up, the study presented in this paper indicate that Glucalvet and Antiparen-N prove to be useful in the treatment of post-parturient paresis conditioned by Ca and Mg deficiency. Thanks to their complex composition, these products have a beneficial effect on the electrolyte equilibrium and energy metabolism of animals afflicted with this condition.

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# CHANGES IN THE QUALITY OF WATER IN BRDOWSKIE LAKE IN 1997-2006

**Ryszard Staniszewski, Józef Szoszkiewicz**

**Chair of Ecology and Environmental Protection  
Poznań University of Life Sciences**

## Abstract

Most lakes in Poland are shallow and vulnerable to degradation mostly due to lake morphology and landscape structure. Other factors, like discharged sewage, internal loading and human activities in the watershed are also important. During studies on Brdowskie Lake, water samples were taken twice a year (spring and summer season) from the surface layer and analyses of soluble reactive phosphates, total phosphorus, nitrates, conductivity, five-day biochemical oxygen demand, chlorophyll *a*, dry mass of seston and Secchi depth were undertaken.

Brdowskie Lake is situated in Kujawskie Lakeland and its catchment basin covers about 155.3 km<sup>2</sup>. The littoral vegetation is dominated by reed bed with a minor presence of other taxa. The lake is very susceptible to degradation (morphology, agricultural lands, housing) and has several potential sources of pollution, e.g. Noteć River, a nameless stream and summer houses. In general, water quality of the lake was better during the spring season, especially in terms of chlorophyll *a* and dry mass of seston concentrations. In 1997-2006, the level of conductivity was very changeable with the maximum values observed in 1999-2001.

Significant correlations between some parameters in certain seasons of the year were found, e.g. between chlorophyll *a* and Secchi depth during spring and total phosphorus and phosphates in summer. The impact of water level fluctuations on water quality parameters, like total phosphorus and chlorophyll *a* was observed.

According to the results, the quality water in Brdowskie Lake is improving. After a biological wastewater treatment plant in the catchment had opened, the rate of salts flowing into the lake with sewage was reduced.

**Key words:** water quality indicators, phosphorus, Brdowskie Lake, Kujawskie Lakeland, Province of Wielkopolska, water level fluctuations.

## ZMIANY JAKOŚCI WÓD JEZIORA BRDOWSKIEGO W LATACH 1997-2006

### Abstrakt

Większość jezior w Polsce to jeziora płytkie i podatne na degradację. Jej głównym powodem są na ogół morfologia zbiorników oraz niekorzystna struktura użytkowania zlewni. Inne czynniki, takie jak dopływ ścieków, ładunek wewnętrzny i działalność gospodarcza, mają również istotne znaczenie. Podczas badań Jez. Brdowskiego próby wody pobierano 2 razy w roku (wiosną i jesienią) z warstwy powierzchniowej i wykonywano oznaczenia takich wskaźników, jak: fosfor reaktywny, fosfor ogólny, azot azotanowy, przewodność elektrolityczna właściwa, pięciodobowe zapotrzebowanie tlenu, chlorofil *a*, sucha masa sestonu i widzialność krążka Secchi'ego.

Jezioro Brdowskie usytuowane na Pojezierzu Kujawskim ma zlewnię o pow. ok. 155,3 km<sup>2</sup>. Litoral zdominowała trzcina, a udział innych taksonów roślin jest mały. Akwen bardzo podatny na degradację (warunki morfologiczne, duży udział terenów wykorzystywanych rolniczo, zabudowania wiejskie) ma wiele potencjalnych źródeł zanieczyszczenia wody, jak np. rzeka Noteć, ciek bez nazwy, domki letniskowe z nieuregulowaną gospodarką wodno-ściekową oraz pobliskie zabudowania wiejskie. Zazwyczaj jakość wody była wyższa na wiosnę, szczególnie odnośnie do stężeń chlorofilu *a* oraz suchej masy sestonu. W latach 1997-2006 poziom przewodności elektrolitycznej był bardzo zmienny, a maksimum przypadało na lata 1999-2001.

Stwierdzono istotne korelacje między niektórymi wskaźnikami w zależności od pory roku, jak w przypadku chlorofilu *a* i widzialności krążka Secchi'ego wiosną i fosforu ogólnego i fosforanów rozpuszczonych latem. Zaobserwowano wpływ wahań poziomu lustra wody na niektóre wskaźniki jakości (fosfor ogólny, chlorofil *a*).

Uzyskane wyniki świadczą o poprawie jakości wód Jez. Brdowskiego. Po uruchomieniu oczyszczalni ścieków w Poloniszu obniżył się np. poziom przewodności elektrolitycznej, co świadczy o ograniczeniu dopływu różnych soli do wód akwenu.

Słowa kluczowe: wskaźniki jakości wody, fosfor, Jezioro Brdowskie, Pojezierze Kujawskie, Wielkopolska, wahania poziomu lustra wody.

## INTRODUCTION

Most lakes in Poland (about 60%) are shallow. They often lie in lowlands and are susceptible to degradation mostly due to lake morphology and landscape structure (CHOIŃSKI 1995). In shallow lakes, both depth and volume of water can strongly affect trophic conditions (HAKANSON 2005, STANISZEWSKI et al. 2009). Other factors that influence water quality are discharged sewage, internal loading, agricultural activities, industry, climatic conditions, vegetation structure, soil conditions, elevation and even longitude (HEATHWAITE 1995, SAPEK 1998, SRINDERGAARD et al. 1999, NÖGES et al. 2003, HAKANSON 2005 and others). The influence of some other factors on Brdowskie Lake, such as inflow of wastewater from houses (the western shore), wastewater after rainfall (the eastern shore) and from summer houses is difficult to assess.

## MATERIAL AND METHODS

Water samples were taken from surface layer of the lake twice a year (spring and summer 1997-2006).

The following analyses were undertaken:

- soluble reactive phosphates – samples filtered using 0.45 µm pore size, Ascorbic Acid Method;
- total phosphorus – Acid Persulfate Digestion Method;
- nitrates – samples filtered using 0.45 µm pore size, Cadmium Reaction Method;
- conductivity – electrometrically;
- five-day biochemical oxygen demand (BOD 5) – Winkler method, summer only;
- chlorophyll *a*;
- dry mass of seston;
- Secchi depth.

Water quality was evaluated in accordance with the regulations imposed by the Ministry for the Environment (*Rozporządzenie Ministra Środowiska* 2008). Statistical evaluation was made using the programme Statistica (Statsoft Inc. 2004).

## RESULTS AND DISCUSSION

Brdowskie Lake is a shallow lake (maximum depth of 5 m, average depth of 2.2 m and the catchment basin area of 155.3 km<sup>2</sup>) situated in Kujawskie Lakeland (the Noteć River watershed) and covers an area of 198.2 ha; its catchment basin covers about 155.3 km<sup>2</sup> (*Raport o stanie środowiska...* 2005). Due to its morphology and landscape structure of the catchment basin (agricultural lands 74%, housing 10%) as well as prevalence of arable lands in the vicinity of the lake shores, Brdowskie Lake is very susceptible to degradation. Potential sources of pollution are tributaries (eg. the Noteć River in the northern part of lake, a nameless stream flowing from the south-western direction) and summer houses on the western and eastern lake shore.

Littoral vegetation is dominated by reed bed (class *Phragmitetea*, Tx. et Preisg. 1942) with a minor presence of perfoliate pondweed (*Potamogeton perfoliatus* L.) and other plant species from the class of *Potametea* (Tx. et Preisg. 1942). Phytoplankton is sometimes strongly dominated by blue-green algae, especially during summer, when they cause algal blooms (STANISZEWSKI, SZOSZKIEWICZ 2000, *Raport o stanie środowiska...* 2005, STANISZEWSKI et al. 2005). It is a lake with good conditions for carp bream, whose the potential

fish yield is about 35 kg per 1 ha, but other species like common bleak, roach and perch are also present.

In general, water quality was better during spring season (Tables 1a, b). The highest concentrations of total phosphorus in Brdowskie Lake were ob-

Table 1a

Characteristics of selected water quality parameters of Brdowskie Lake in 1997-2006, spring measurements

Parameter	Mean values	Standard deviation
Total phosphorus (mg P dm <sup>-3</sup> )	0.08	0.04
Chlorophyll <i>a</i> (µg Chl <i>a</i> dm <sup>-3</sup> )	24.69	12.42
Secchi depth (m)	0.97	0.40
Dry mass of seston (mg DMS dm <sup>-3</sup> )	10.40	5.31
Soluble reactive phosphates (mg PO <sub>4</sub> dm <sup>-3</sup> )	0.11	0.06
Nitrates (mg N-NO <sub>3</sub> dm <sup>-3</sup> )	0.09	0.07
Conductivity (mS cm <sup>-1</sup> )	0.714	0.227

Table 1b

Characteristics of selected water quality parameters of Brdowskie Lake in 1997-2006, summer measurements

Parameter	Mean values	Standard deviation
Total phosphorus (mg P dm <sup>-3</sup> )	0.10	0.05
Chlorophyll <i>a</i> (µg Chl <i>a</i> dm <sup>-3</sup> )	39.83	15.79
Secchi depth (m)	0.79	0.16
Dry mass of seston (mg DMS dm <sup>-3</sup> )	17.85	7.73
Soluble reactive phosphates (mg PO <sub>4</sub> dm <sup>-3</sup> )	0.17	0.10
Nitrates (mg N-NO <sub>3</sub> dm <sup>-3</sup> )	0.08	0.11
Conductivity (mS cm <sup>-1</sup> )	0.685	0.267
BOD 5 (mg O <sub>2</sub> dm <sup>-3</sup> )	5.0	2.0

Table 1c

Water quality categories of Brdowskie Lake in 1997-2006 evaluated on the basis of average results (spring and summer surveys) of total phosphorus, chlorophyll *a*, Secchi depth and conductivity in the surface layer of water (five categories according to Rozporządzenie Ministra Środowiska 2008)

Year	Total phosphorus	Chlorophyll <i>a</i>	Secchi depth	Conductivity	The worst category assigned
1997	I-II	III	III-V	III-V	III-V
1998	I-II	III	III-V	III-V	III-V
1999	I-II	III	III-V	III-V	III-V
2000	I-II	II	III-V	III-V	III-V
2001	I-II	III	III-V	III-V	III-V
2002	I-II	IV	III-V	I-II	III-V
2003	I-II	III	III-V	I-II	III-V
2004	I-II	III	I-II	I-II	III
2005	III-V	IV	III-V	I-II	III-V
2006	I-II	II	I-II	I-II	II

served in summer 2005 but in spring they were lower (Tables 1a, b). Chlorophyll *a* and dry mass of seston behaved similarly. During the tests, Secchi depth was sometimes lower than 60 cm (spring 1998, summer 2003 and 2005) and once was over 180 cm (182 cm, spring 2006), which was attributable to an unusually low concentration of chlorophyll in water ( $4.28 \mu\text{g Chl } a \text{ dm}^{-3}$ ).

Concentration of soluble reactive phosphates in lake water ranged from 0.01 to 0.36  $\text{mg PO}_4 \text{ dm}^{-3}$  and showed slightly increasing tendency with maximum in summer 2006. It was partially related to water level fluctuations but it was not statistically significant.

The results showed, that presence of nitrates in surface layer was generally higher in spring before part of nitrogen was incorporated in biomass of phytoplankton and macrophytes. The highest concentrations of nitrates were observed in the year 2003 during spring (about  $0.20 \text{ mg N-NO}_3 \text{ dm}^{-3}$ ) and summer ( $0.40 \text{ mg N-NO}_3 \text{ dm}^{-3}$ ) measurements were concentration of seston was also very high. Results obtained in summer surveys were very diverse and it gave high value of standard deviation for nitrates (Table 1b).

In the years 1997-2006 the conductivity of lake waters was very changeable. During spring the highest values ( $> 1.000 \text{ mS cm}^{-1}$ ) were observed in the years 1999-2001 and the lowest in the year 2006 ( $< 0.500 \text{ mS cm}^{-1}$ ). According to summer surveys the highest value ( $> 1.000 \text{ mS cm}^{-1}$ ) was measured in the year 1999 and very low in the years 2002-2006, one year

after sewage treatment plant in Polonisz had started-up (31<sup>st</sup> of March 2001). Conductivity showed positive but not statistically significant correlation with water level fluctuations.

Analyses of biochemical oxygen demand were made during summer period and results ranged from 2.0 mg O<sub>2</sub> dm<sup>-3</sup> to 8.0 mg O<sub>2</sub> dm<sup>-3</sup>.

According to physico-chemical parameters from the year 2006 the Brdowskie Lake waters can potentially fit standards of second category of water quality. Taking into account actual criteria, during at least eight years of surveys the situation was far from this. Main problem was low values of Secchi depth and high conductivity (Table 1c).

Presence of significant correlations between some trophic parameters in certain year season were found (Tables 2, 3). During spring season strong correspondence between concentration of chlorophyll *a* and Secchi depth was

Table 2

Brdowskie Lake – linear correlation among trophic parameters for the years 1997-2006, spring measurements ( $p < 0.05$ ,  $N = 10$ )

Parameter	Total phosphorus	Soluble reactive phosphates	Chlorophyll <i>a</i>	Secchi depth	Dry mass of seston
Total phosphorus	1.00	0.58	-0.47	0.54	-0.51
Soluble reactive phosphates		1.00	-0.43	0.38	-0.61
Chlorophyll <i>a</i>			1.00	-0.84*	0.62
Secchi depth				1.00	-0.50
Dry mass of seston					1.00

\* statistically significant

Table 3

Brdowskie Lake – linear correlation among trophic parameters for the years 1997-2006, summer measurements ( $p < 0.05$ ,  $N = 10$ )

Parameter	Total phosphorus	Soluble reactive phosphates	Chlorophyll <i>a</i>	Secchi depth	Dry mass of seston
Total phosphorus	1.00	0.70 *	0.61	-0.64*	0.52
Soluble reactive phosphates		1.00	0.05	-0.34	0.08
Chlorophyll <i>a</i>			1.00	-0.61	0.58
Secchi depth				1.00	-0.65*
Dry mass of seston					1.00

\* statistically significant



found. In summer time relations between total phosphorus and phosphates, total phosphorus and water clarity and also between Secchi depth and dry mass of seston. Strong correlations between water clarity and trophic parameters were found by other authors in earlier studies (CARLSON 1977, MAN-DAVILLE 2000 and others).

In surface waters of Brdowskie Lake the impact of water level fluctuations on water quality parameters was not strong and only in case of trophic parameters significant correlations were found. Especially in case of chlorophyll a concentration in spring (positive correlation) and total phosphorus concentration in summer (negative correlation) – Table 4.

Table 4

Linear correlation between water table level and trophic parameters, spring ( $p < 0.05$ ,  $N = 8$ ) and summer measurements ( $p < 0.065$ ,  $N = 8$ ), Brdowskie Lake in the years 1999-2006

Parameter	Total phosphorus	Soluble reactive phosphates	Chlorophyll <i>a</i>	Secchi depth	Dry mass of seston
Water table level – spring	-0.57	-0.34	0.71 *	-0.53	0.32
Water table level – summer	-0.72*	-0.70	-0.09	0.44	-0.63

\* statistically significant

## CONCLUSIONS

1. Brdowskie Lake waters studied in the period 1997-2006 have had rather poor quality but results from years 2004 and 2006 proved, that according to physico-chemical parameters the lake has a potential to achieve second category of water quality.

2. Since the year 2002 the decrease of water conductivity has been found and it was one year after start-up of biological waste water treatment plant in lake catchment, which probably limited the rate of salts inflowing to lake with sewages.

3. Influence of water level fluctuations on water quality parameters was observed but only in case of total phosphorus concentration and water clarity it was significant.

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## SEASONAL VARIABILITY OF MINERAL NITROGEN IN GROUNDWATER OF HYDROGENIC SOILS\*

**Sławomir Szymczyk<sup>1</sup>, Jan Pawluczuk<sup>2</sup>,  
Arkadiusz Stępień<sup>3</sup>**

<sup>1</sup>Chair of Land Reclamation and Environment Management,

<sup>2</sup>Chair of Soil Science and Soil Protection,

<sup>3</sup>Chair of Agricultural Systems

University of Warmia and Mazury, Olsztyn

### Abstract

Agricultural use of land on hydrogenic soils is associated with lowering the groundwater level, which intensifies the processes of organic matter mineralisation and, in effect, releases large amounts of mineral nitrogen. The aim of the study was to determine the seasonal variability of the concentrations of nitrogen mineral compounds (N-NO<sub>2</sub>, N-NO<sub>3</sub> and N-NH<sub>4</sub>) in groundwater under extensively managed peat-muck soils, situated in Wrocikowo in the Olsztyn Lakeland and in the Dymerskie Meadows in the Mrągowo Lakeland. Peat-muck soil, found at the study sites, had varied physical properties. The highest content of mineral particles (69.3%) in the muck layer was found in soil marked as MtI 120gy under turf-covered wasteland, and in the peat layer (51.5%) – in soil marked as MtII 60gy under extensively managed meadow. Groundwater taken from piezometers installed in hydrogenic soils was used as the study material. Water for chemical determinations was taken during four seasons: spring – in May, summer – in August, autumn – in November and winter – in January. The concentration of mineral forms of nitrogen in groundwater of peat-muck soil was found to depend on its type, the depth where the groundwater was found and the type of soil use. The highest concentration of mineral nitrogen was found in summer (1.62 mg dm<sup>-3</sup> on average), and the lowest (1.11 mg dm<sup>-3</sup> on average) was found in winter. The concentration of mineral nitrogen in groundwater of extensively managed peat-muck soil ranged from 0.81 to 2.27 mg dm<sup>-3</sup> and was found to be dependent on the type of soil rather than its use. Lowering the level of groundwater in peat-muck soil increases the concentration of mineral forms of nitrogen, especially N-NH<sub>4</sub>. Ammonium nitrogen was the dominant form of nitrogen in the groundwater of peat-muck soil (MtII 60gy and MtI 120gy) of non-managed and extensively managed meadows, whe-

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dr inż. Sławomir Szymczyk, Chair of Land Reclamation and Environment Management, University of Warmia and Mazury, Olsztyn, Plac Łódzki 2, 10-718 Olsztyn, e-mail: szymek@uwm.edu.pl

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reas nitrate nitrogen was the dominant form of the element in the MtIc 35gy soil under the meadow and the MtII bb under the pasture.

**Key words:** mineral nitrogen, groundwater, meadow, pasture, biogenic substances, peat-muck soil.

## SEZONOWA ZMIENNOŚĆ STĘŻENIA MINERALNYCH FORM AZOTU W WODACH GRUNTOWYCH GLEB HYDROGENICZNYCH

### Abstrakt

Zagospodarowanie rolnicze terenów położonych na glebach hydrogenicznych wiąże się z obniżeniem poziomu wody gruntowej, co powoduje nasilenie procesów mineralizacji substancji organicznej, a w efekcie uwalnianie dużych ilości azotu mineralnego. Celem badań było określenie zmienności sezonowej stężenia mineralnych związków azotu ( $\text{N-NO}_2$ ,  $\text{N-NO}_3$  i  $\text{N-NH}_4$ ) w wodzie gruntowej pod ekstensywnie zagospodarowanymi glebami torfowo-murszowymi położonymi na obiektach Wrocikowo – Pojezierze Olsztyńskie i Łąki Dymerskie – Pojezierze Mrągowskie. Występujące na obiektach badań gleby torfowo-murszowe charakteryzowały się zróżnicowanymi właściwościami fizycznymi. Największą zawartość części mineralnych (69,3%) w warstwie murszowej stwierdzono w glebie MtI 120gy pod zadarnionym nieużytkiem, a w warstwie torfowej (51,5%) – w glebie MtII 60gy pod ekstensywnie zagospodarowaną łąką. Materiał badań stanowiła woda gruntowa pobierana z piezometrów zainstalowanych w glebach hydrogenicznych. Wodę do oznaczeń chemicznych pobierano w czterech porach roku: wiosną – maj, latem – sierpień, jesienią – listopad i zimą – styczeń. Wykazano, że stężenie mineralnych form azotu w wodzie gruntowej gleb torfowo-murszowych jest uzależnione od ich rodzaju, głębokości występowania wód gruntowych oraz sposobu zagospodarowania gleb. Największe stężenie azotu mineralnego stwierdzono latem (średnio  $1,62 \text{ mg dm}^{-3}$ ), a najmniejsze (średnio  $1,11 \text{ mg dm}^{-3}$ ) zimą. W wodzie gruntowej gleb torfowo-murszowych zagospodarowanych ekstensywnie zawartość azotu mineralnego wahała się od  $0,81$  do  $2,27 \text{ mg dm}^{-3}$  i była bardziej uzależniona od rodzaju gleby niż sposobu jej użytkowania. Obniżenie poziomu wody gruntowej w glebach torfowo-murszowych powoduje w niej zwiększenie stężenia mineralnych związków azotu, szczególnie  $\text{N-NH}_4$ . W wodzie gruntowej gleb torfowo-murszowych (MtII 60gy i MtI 120gy) nieużytkowanych i ekstensywnie użytkowanych łąk, dominującą formą azotu mineralnego był  $\text{N-NH}_4$ , a w glebach MtIc 35gy pod łąką i MtII bb pod pastwiskiem  $\text{N-NO}_3$ .

**Słowa kluczowe:** azot mineralny, woda gruntowa, łąka, pastwisko, składniki biogenne, gleby torfowo-murszowe.

## INTRODUCTION

There are many marshes and swamps in the young glacial areas of northeastern Poland (KIRYLUK 2004, PIĄŚCIK et al. 2000, SZYMCZYK, GLIŃSKA-LEWCZUK 2007). Peatlands are a dominant form among hydrogenic habitats. They are not only natural reservoirs of water in the environment but also places where organic matter accumulates. Owing to these properties, peat retards the flow of biogenic substances in the environment, including nitrogen compounds, released in the process of mineralisation of organic matter

(BRANDYK et al. 1996, SAPEK, SAPEK 2004). Mineral nitrogen compounds, which are formed in the processes of organic matter decomposition, are a very important element of nutrient circulation, especially in marshy ecosystems. Released in the process of mineralisation of organic nitrogen compounds, ammonium nitrogen, followed by nitrate nitrogen, form the available supply of mineral nitrogen, whose amount determines the abundance of the nutrient in soil. Released from hydrogenic soils, mineral nitrogen can be taken up by plants, migrate down the soil profile to groundwater or become denitrified. The amount of mineral nitrogen leached from hydrogenic soil to groundwater depends mainly on the dynamics of its release from soil and may vary from year to year; its intensity may be different in different seasons (PAWLUCZUK, GOTKIEWICZ 2003, SAPEK, SAPEK 2004). The organic matter mineralisation rate and, in consequence, the concentration of mineral forms of nitrogen in water depend mainly on soil humidity, on the genetic type of organic matter in which decomposition processes take place, and on the weather conditions and the manner of soil use (PAWLUCZUK, SZYM CZYK 2008). Much of the peatlands in Poland have been drained and are used agriculturally, which has precipitated water and the outflow biogenic substances, including considerable amounts of mineral nitrogen compounds (KOC, SZYM CZYK 2003, KOC et al. 2005, SAPEK, SAPEK 2004). The aim of the study was to determine the seasonal variability (four seasons of the year) of concentration of mineral compounds of nitrogen ( $\text{N-NO}_2$ ,  $\text{N-NO}_3$  and  $\text{N-NH}_4$ ) in groundwater of peat-muck soil depending on their use (meadow, pasture, turf-covered wasteland) against the background of habitat conditions.

## MATERIAL AND METHODS

Seasonal variability in concentration of mineral compounds of nitrogen ( $\text{N-NO}_2$ ,  $\text{N-NO}_3$  and  $\text{N-NH}_4$ ) in groundwater of peat-muck soil was examined in 2004-2008 at the following sites: Wrocikowo (2004-2006), a village situated in the Olsztyn Lakeland, and the Dymerskie Meadows (2006-2008) in the Mragowo Lakeland. The study on the concentrations of  $\text{N-NO}_2$ ,  $\text{N-NO}_3$  and  $\text{N-NH}_4$ , presented in this paper is a continuation of our earlier research into the dynamics of mineralisation of organic matter of peat soil at the same sites (PAWLUCZUK 2006, PAWLUCZUK, SZYM CZYK 2008, SZYM CZYK, SZYPEREK 2005). The peat bog in Wrocikowo was formed at the site of a former kettle lake. The 142.66 ha drainage basin which feeds the peat bog has been reclaimed and is used as agricultural land. The water which drained off from the upper areas flows into the peat bog, from which it is channelled off by a drainage ditch. Only part of the peat bog (extensively managed meadow – MtI 120gy soil) was used agriculturally during the study period, while the other (MtII 60gy soil) was turf-covered wasteland, an ecological site (SZYM CZYK, SZYPEREK 2005). Hydrogenic soils of the Dymerskie

Meadows were put to agricultural use by draining Dymers Lake (242 ha) in 1876. This also drained the peat soil (*ca* 111 ha), situated on the lake's edge. Currently, only part of the muck soil in the Dymerskie Meadows is under extensively managed grassland (meadow and pasture), where the air-water balance is not properly regulated due to negligence in the maintenance of the drainage system (PAWLUCZUK 2006, PAWLUCZUK, SZYMCZYK 2008). The physical properties of the peat-muck soil at the study sites varied (Table 1). The highest concentration of minerals (69.3%) in the muck layer was found in MtI 120gy soil, and in the peat layer (51.5%) – in MtII 60gy soil in Wrocikowo. The hydrogenic soils – MtIc35gy under the extensively managed meadow and MtIIbb under the heavily exploited pasture in the Dymerskie Meadows – were less depelted. Lower concentrations of mineral substances were determined both in the upper muck layers (37.9% and 58.9%, respectively) and in peat (25.8% and 32.1%, respectively). Ash content in the analyzed soils decreased in deeper organic layers. Hydrogenic soils in Wrocikowo and the Dymerskie Meadows were slightly acidic (Table 1).

Table 1

Physicochemical properties of hydrogenic soils in Wrocikowo and in the Dymerskie Meadows

Soil	Layer (cm)	Level	Ash content (%)	Volumetric density (g cm <sup>-3</sup> )	pH <sub>KCl</sub>
MtII 60gy	do 20	Mt	67.3	0.65	6.7
	20-60	Otnitu	51.5	0.33	6.5
MtI 120gy	do 20	Mt	69.3	0.46	6.2
	20-120	Otnitu	41.6	0.23	6.1
MtIc 35gy	do 20	Mt	37.9	0.27	5.7
	20-35	Otnitu	25.8	0.15	6.0
MtII bb	do 20	Mt	58.9	0.39	5.9
	20-60	Otnitu	32.1	0.26	5.9

Two piezometers were placed at the depth of 150 cm in organic soil near the pits at each study site, with their filtration part at the depth of 120-150 cm below the surface. The groundwater level in the piezometers was measured once a month. Water samples for chemical determinations were taken in four seasons (spring – in May, summer – in August, autumn – in November, winter – January). The following were determined in the water samples: N-NO<sub>2</sub> – colorimetrically with sulphanilic acid, N-NO<sub>3</sub> – colorimetrically with disulphonic acid, N-NH<sub>4</sub> – colorimetrically with Nessler's reagent. Subsequently, the concentration of mineral nitrogen (N-NO<sub>2</sub> + N-NO<sub>3</sub> + N-NH<sub>4</sub>) was calculated. The results were analysed statistically.

Analysis of variance was performed (test  $F$ ) and variability coefficients ( $V\%$ ) were calculated, taking into account entire sets of data for each soil.

## RESULTS AND DISCUSSION

The use of each of the study sites depended mainly on the soil humidity, which was in turn determined by the groundwater level. Typical seasonal variability of groundwater levels was recorded at both the study sites – the minimum in summer (July–September) and the maximum in winter (January–March). The highest levels of groundwater were recorded in peat soils with underlying organic gyttja MtIc 35gy – the Dymerskie Meadows (average – 38 cm below the surface) and in MtI 120gy – Wrocikowo (average – 42 cm below the surface), where they were significantly higher compared to the other two types of soil (Figure 1).

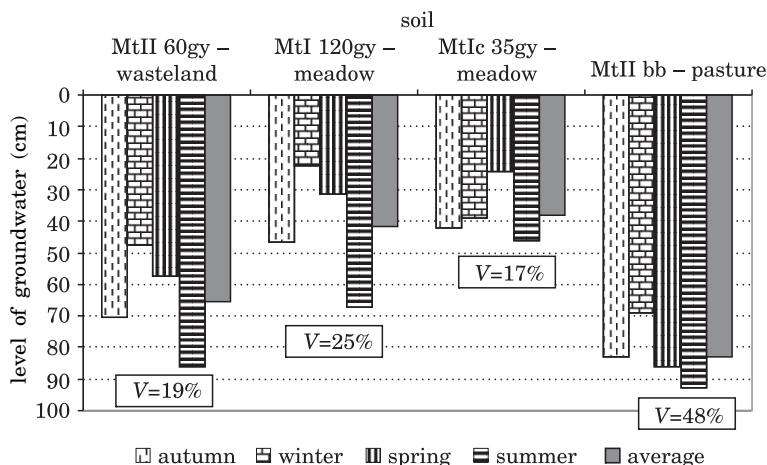


Fig. 1. Seasonal average levels of groundwater in hydrogenic soils

Where gyttja was found in shallower layers (MtIc 35gy soil), a clear tendency for the ground level to remain high with low variability was found throughout the study period ( $V = 17\%$ ). This was the consequence of the meadow being situated in the vicinity of gyttja-containing soil and the lack of proper maintenance of the draining ditch. The lowest levels of groundwater with the highest variability during the study period ( $V = 48\%$ ) were recorded under extensively managed pasture, in MtIIbb soil. Such high variability of the groundwater surface level as compared to the other study sites was due to atmospheric precipitations in the consecutive years of study.

It could also be of some importance that the grassland was used as pasture, where higher biomass production resulted in greater evapotranspiration.

Higher concentration of  $\text{N-NO}_3$  was found in groundwater in the Dymerskie Meadows (0.54 and 0.43  $\text{mg dm}^{-3}$  on average – Figure 2); the values found in the peat-muck soil in Wrocikowo were much more variable (MtI 120gy soil –  $V = 76\%$ , MtII 60gy soil –  $V = 45\%$ ).

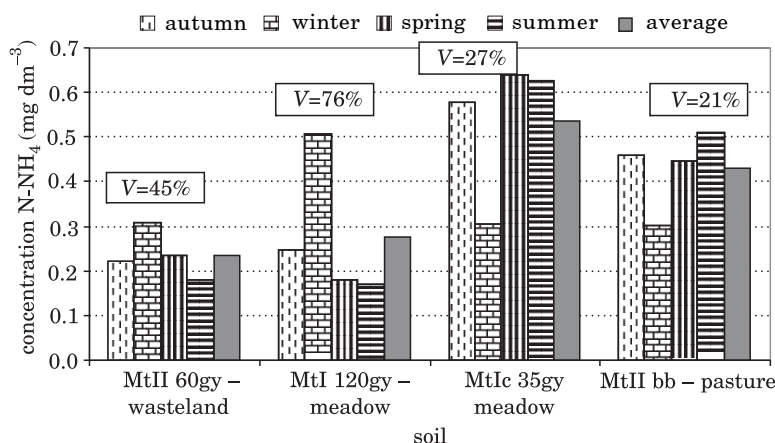


Fig. 2. Seasonal average concentrations of  $\text{N-NO}_3$  in groundwater of hydrogenic soils

The highest concentration of  $\text{N-NO}_3$  (0.54  $\text{mg dm}^{-3}$  on average) was found in groundwater under the extensively managed meadow in peat-muck soil (MtIc 35gy), formed on shallow organic gyttja, whereas the lowest values (0.24  $\text{mg dm}^{-3}$  on average) were found in peat-muck soil (MtII 60gy) formed on carbonate gyttja under wasteland meadow (Figure 2). This was associated both with the type of substratum and with the groundwater stagnation depth. As compared to the results reported by other authors, these values are up to several dozen times lower (SAPEK, SAPEK 2004). Over 20-fold higher variability was recorded when the results attained in this study were compared with the values found in groundwater under intensively managed grassland (meadows and pastures) (JASZCZYŃSKI et al. 2006).

Lowering the groundwater level in peat soils favours an increase in organic matter mineralisation rate, with increasing nitrogen concentration as a result (PAWLUCZUK, GOTKIEWICZ 2003, SAPEK, SAPEK 2004). The correlations calculated for the results (from  $R = 0.13$  in MtII bb soil to  $R = 0.51$  in MtII 60gy soil) have shown that the lowering of the groundwater level at the study sites was accompanied by a decrease in nitrate(V) concentrations, which increased when the groundwater level rose (Table 2). This may be attributed to more intensive mineralisation of organic nitrogen compounds, and, in effect, releasing higher amounts of nitrates(V) in the upper layer of peat-muck soils when the groundwater level lowers, which then migrate to groundwater after it rises.



Table 2

Correlation between the groundwater level and mineral nitrogen compounds

Nitrogen form	Soil			
	MtII 60gy	MtI 120gy	MtIc 35gy	MtII bb
N-NO <sub>3</sub>	0.51	0.40	0.32	0.13
N-NO <sub>2</sub>	0.28	-0.52	0.61*	0.06
N-NH <sub>4</sub>	-0.68*	-0.44	0.11	0.04

\* correlation significant at  $p < 0.05$ 

Seasonal variability in the soil humidity level, which depends on the depth of the groundwater level, resulted in a considerable variability of N-NO<sub>3</sub> concentrations groundwater (V from 21 to 76%). The concentration of N-NO<sub>3</sub> in groundwater of MtII 60gy and MtI 120gy soils decreased from spring to summer and subsequently started to increase in autumn, attaining the highest values in winter, whereas the lowest concentrations in MtIc 35gy and MtII bb soils were found in winter, and the highest were in spring and summer, respectively.

As compared to the extensively-managed meadows on MtI 120gy and MtII 60gy soils (Wrocikowo), significantly higher concentrations of N-NO<sub>3</sub> were found in groundwater in MtII bb and MtIc 35gy soils (the Dymerskie Meadows). However, the groundwater in MtIc 35gy soil under the extensively-managed meadow contained more nitrates (15% more on average) than that under pasture in MtII bb soil. This may have been caused by less intensive nitrate uptake by the vegetation covering the more humid meadow habitat.

The concentration of N-NO<sub>2</sub> in the analyzed groundwater varied (from 0.008 to 0.067 mg dm<sup>-3</sup>); it was particularly high in Wrocikowo (MtI 120gy soil – V=143% and MtII 60gy soil – V = 138%), which may be attributed both to the nature of the substratum and to the variability of groundwater levels (Figure 3).

They were also significantly higher than in the Dymerskie Meadows. Low and the least varied concentrations of N-NO<sub>2</sub> in groundwater in MtIc 35gy soil may be attributed to high and stable levels of groundwater. Extensive management of the pasture on MtII bb soil, formed from low rush peat, resulted in a nearly twofold increase in the concentration of N-NO<sub>2</sub> in groundwater as compared to MtIc 35gy soil, formed on shallow organic gytja. Except for MtI 120gy soil ( $R = -0.52$ ), a similar tendency to increase with rising groundwater levels was observed for concentrations of N-NO<sub>2</sub> and N-NO<sub>3</sub>. In MtIc 35gy soil, the concentration of MtIc 35gy was found to decrease significantly with increasing saturation of the soil profile with water ( $R = 0.61$ ).

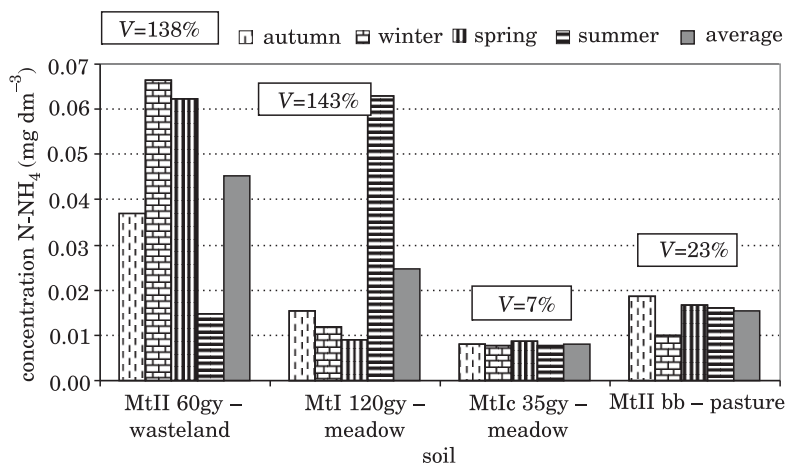


Fig. 3. Seasonal average concentration of  $\text{N-NO}_2$  in groundwater of hydrogenic soils

The concentration of ammonium nitrogen in groundwater of turf-covered hydrogenic soils depended on their type and was subject to high seasonal variability. A considerably higher load of  $\text{N-NH}_4$  in groundwater in MtII 60gy soil was recorded in summer ( $2.49 \text{ mg dm}^{-3}$ ) and in autumn ( $2.47 \text{ mg dm}^{-3}$ ), in MtI 120gy soil – mainly in summer ( $1.52 \text{ mg dm}^{-3}$ ), and in MtIc 35gy soils ( $0.53 \text{ mg dm}^{-3}$ ) and MtII bb ( $0.52 \text{ mg dm}^{-3}$ ) – in autumn. However, it was significantly higher in groundwater in Wrocikowo than in the Dymerskie Meadows (Figure 4).

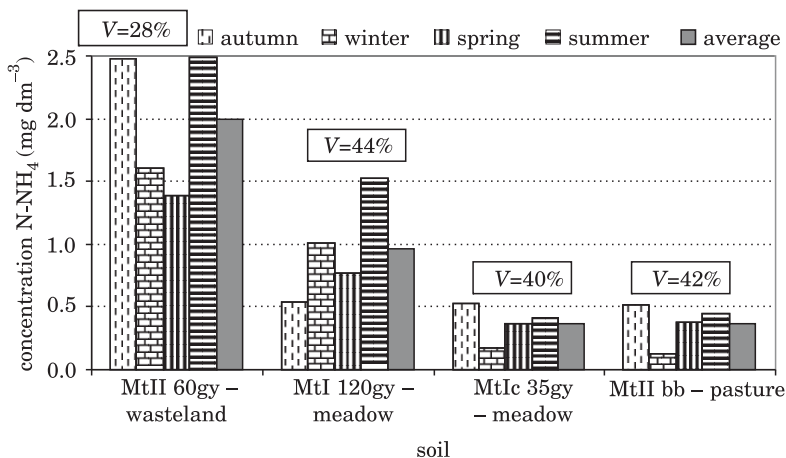


Fig. 4. Seasonal average concentration of  $\text{N-NH}_4$  in groundwater of hydrogenic soils

This may have been associated with higher variability of water levels, especially with a clear minimum during the summer, which favoured more intensive mineralisation of organic matter, especially in MtII 60gy. This is confirmed by high correlation coefficients ( $R=-0.68$ ) of  $\text{N-NH}_4$  concentrations with water levels (Table 2). Very low correlations between these features were determined in the Dymerskie Meadows.

It should be stressed that the concentrations of  $\text{N-NH}_4$  (from 0.37 to 1.99  $\text{mg dm}^{-3}$  on average) in groundwater under extensively-managed or unused hydrogenic soils were 2- to 8-fold lower than those determined by JASZCZYŃSKI et al. (2006) for intensive meadow or pasture use of peat-muck soils.

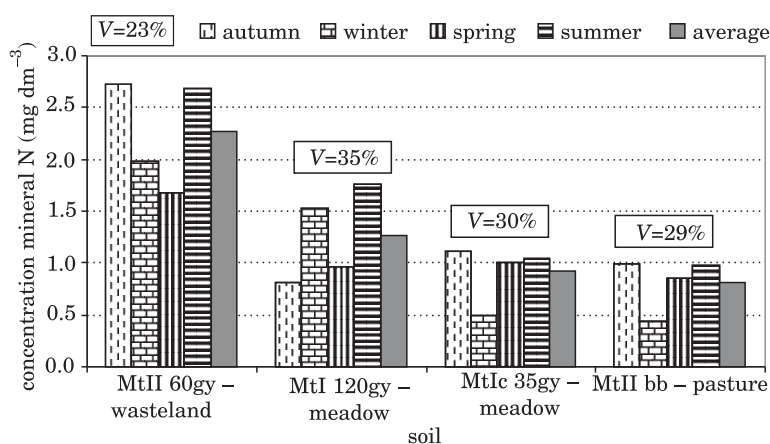


Fig. 5. Average seasonal concentration of mineral nitrogen in groundwater of hydrogenic soils

The concentration of mineral nitrogen (from 0.81 to 2.27  $\text{mg dm}^{-3}$  on average) in groundwater of muck soils (total  $\text{N-NO}_3$ ,  $\text{N-NO}_2$  and  $\text{N-NH}_4$ ) was more dependent on the type of soil than on its use (Table 5). Significantly higher concentrations of mineral nitrogen were found in groundwater in Wrocikowo, where they were 4-fold higher (MtII 60gy) and twice higher (MtI 120gy) than in the Dymerskie Meadows. Ammonium nitrogen dominated the total mineral nitrogen in groundwater in Wrocikowo. This indicates higher mineralisation of organic matter at the site, which may result from more intensive draining than in the Dymerskie Meadows. Extensive use of hydrogenic soils for meadows (MtI 120gy soil) as well as fallowing (MtII 60gy) results in loading groundwater with mineral nitrogen compounds, especially the ammonium species. Unlike Wrocikowo,  $\text{N-NO}_3$  dominated over  $\text{N-NH}_4$  in groundwater in the Dymerskie Meadows. The difference between these two compounds varied considerably from season to season. The concentration of  $\text{N-NO}_3$  in groundwater of MtIc 35gy soil was 49% higher on

average, and only 8% higher in MtII bb soil, which was mainly affected by the concentrations in winter. Despite better conditions for mineralisation of organic matter, which resulted from lower levels of groundwater and oxygenation of the soil profile, which facilitates more intensive nitrification of ammonium nitrogen, the ammonium form accounted for a greater portion of the total mineral nitrogen in groundwater under the pasture than in that under the extensively-managed meadow. This may have resulted from more intensive bioaccumulation of  $\text{N-NO}_3$  by vegetation, which grows more intensively on the pasture than on the meadow. The highest concentration of mineral nitrogen in groundwater of peat-muck soil is found in summer ( $1.62 \text{ mg dm}^{-3}$  on average), and the lowest ( $1.11 \text{ mg dm}^{-3}$  on average) is found in winter.

## CONCLUSIONS

1. Concentration of mineral forms of nitrogen in groundwater of peat-muck soils depends on their type, the depth at which groundwater is found and the soil use; the highest concentration of mineral nitrogen is found in summer and the lowest is found in winter.

2. The concentration of mineral nitrogen (total  $\text{N-NO}_3$ ,  $\text{N-NO}_2$  and  $\text{N-NH}_4$ ) in groundwater of peat-muck soil under meadow, pasture of turf-covered wasteland ranges from  $0.81$  to  $2.27 \text{ mg dm}^{-3}$  and depends more on the type of soil than on its use.

3. Ammonium species is the dominant form of nitrogen in groundwater of unused peat-muck soil (MtII 60gy soil) and extensively-managed meadow (MtI 120gy soil) –  $1.52$  and  $2.49 \text{ mg dm}^{-3}$ , respectively – whereas nitrates were the dominant form of nitrogen in MtIc 35gy soil under meadows and in MtII bb soil under pasture –  $0.64$  and  $0.51 \text{ mg dm}^{-3}$ , respectively.

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# **AN ATTEMPT TO ASSESS THE IMPACT OF ANTHROPOPRESSURE ON THE ECOLOGICAL STATE OF URBANISED WATERCOURSES OF KRAKOW CONURBATION AND THE DIFFICULTIES ENCOUNTERED\***

**Marta Wardas<sup>1</sup>, Urszula Aleksander-Kwaterczak<sup>1</sup>,  
Szymon Jusik<sup>2</sup>, Beata Hryc<sup>2</sup>, Tomasz Zgoła<sup>2</sup>,  
Marcin Sztuka<sup>1</sup>, Magdalena Kaczmarska<sup>1</sup>, Michał Mazurek<sup>3</sup>**

<sup>1</sup>University of Sciences and Technology in Krakow

<sup>2</sup>Poznan University of Life Sciences

<sup>3</sup>Institute of Meteorology and Water Managment

## **Abstract**

Rivers and streams in cities are treated as urbanised watercourses because of their significant transformation. Their load, channeling and incorporation into the water-sewerage infrastructure are often so considerable that such watercourses can hardly be recognised as an intrinsic component of surface waters. Anthropopressure, as reflected in quantitative and qualitative degradation caused by flow regulation and economic development in the drainage basin area, makes evaluation of the impact of human activity on the aquatic environment somewhat difficult. Based on the recommendations of the Water Framework Directive, an attempt has been made to assess the ecological state of selected tributaries of the Prądnik-Białucha River within the Krakow Conurbation. Aquatic environment sampling of the Sudół Dominikański (Rozrywka) watercourse was performed (September 2005) in order to determine some physicochemical, chemical and biological parameters, paying particular attention to macrophytes. The parameters measured on site: pH, electrolytic conductivity and Eh of water and bottom sediments, and zoological observations, were subjected to analysis. In the laboratory, concentrations of heavy metals, both in water and in solid particles (sediments and suspended matter) and anions in water were determined. The environmental state of the Sudół Dominikański watercourse was compared with that

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dr Marta Wardas, al. Mickiewicza 30, 30-059 Kraków, e-mail: mw@geol.agh.edu.pl

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in the area of the Prądnik-Białucha River valley, for which an assessment had been conducted in the previous year (September 2004).

Key words: ecological state, watercourse, sediments, macrophytes.

## **PRÓBA I TRUDNOŚCI WYKONANIA OCENY WPLYWU ANTROPOPRESJI NA STAN EKOLOGICZNY W CIEKACH ZURBANIZOWANYCH KRAKOWSKIEGO ZESPOŁU MIEJSKIEGO (KZM)**

### **Abstrakt**

Rzeki i strumienie w miastach, wskutek ich znacznego przekształcenia, traktuje się jako cieki zurbanizowane. Często ich obciążenie i zabudowa infrastrukturą wodno-kanalizacyjną są tak znaczne, że trudno nadal uznawać je za jednolite części wód powierzchniowych. Antropopresja przejawiająca się degradacją ilościową i jakościową w wyniku regulacji przepływu i zagospodarowania zlewni powoduje, że ocena skali wpływu działalności człowieka na środowisko wodne nie jest prosta. Na podstawie zaleceń Ramowej Dyrektywy Wodnej podjęto próbę oceny stanu ekologicznego wybranych dopływów rzeki Prądnik-Białucha, w obrębie Krakowskiego Zespołu Miejskiego. Wykonano opróbowanie środowiska wodnego cieku Sudół Dominikański (Rozrywka – wrzesień 2005 r.), w kierunku określenia niektórych elementów fizykochemicznych, chemicznych oraz biologicznych, szczególnie zwracając uwagę na makrofity. Analizie poddano mierzone w terenie wskaźniki: pH, PEW i Eh wody i osadów dennych oraz obserwacje sozologiczne. W laboratorium zmierzono zawartość metali ciężkich zarówno w wodzie, jak i cząstkach stałych (osadach i zawiesinach) oraz zawartość anionów w wodzie. Stan środowiska cieku Sudół Dominikański porównano ze stanem w rejonie doliny rzeki Prądnik-Białucha, dla której ocenę wykonano rok wcześniej (wrzesień 2004 r.).

Słowa kluczowe: stan ekologiczny, ciek wodny, osady wodne, makrofity.

## **INTRODUCTION**

Achieving good ecological state of waters, both qualitatively and quantitatively, means that surface waters should stay as shaped by nature. At the same time, in defined sections or areas, they should be suitable for public water supply, bathing, sustaining fish life of the *Salmonidae* or at least the *Cyprynidae* family and they should meet appropriate requirements in protected areas (M.P.03.33.433 (Act)). Activities serving to achieve this objective will include limiting pollution from communal and industrial point sources by constructing, expanding and modernising sewerage systems and wastewater treatment plants; in the case of point sources and agricultural area sources of pollution, the overall objective should be reached through appropriate storage and use of natural fertilisers, and the use of organic and mineral fertilisers in accordance with Good Agricultural Practices (Dz.U.04.176.1827 (R)). The recommendations contained in the Ecological Policy of Poland and the rules which ensure coherence of the European Community's environmental protection policy prove that without eliminating



direct pollution sources in drainage basins it will not be possible to achieve the priority aim, i.e. improved quality of surface waters, by 2010 nor even by 2015 (2000/60/EC). For the sake of water management, uniform parts of surface waters are distinguished as separate and significant elements of surface waters, such as a stream, brook, river, channel or their parts. Artificial or strongly altered waters are distinguished from intrinsic components of surface waters (Dz.U.05.239.2019 (Act)).

The problems encountered while evaluating the effect of anthropopressure on the ecological state of urbanised watercourses are due to the lack of defined reference conditions. These watercourses are omitted from regional monitoring studies – sometimes their mouth area is sampled just in order to determine the impact of pollution carried into bigger rivers. The area drained by these watercourses and the quality of waters they carry are rarely a subject of studies, as the local monitoring of surface waters has been abandoned. Some scattered and non-standardised studies of urbanised watercourses have been carried out in the Krakow Conurbation, called the Krakow Metropolitan Area in current planning documents, which is territorially larger than the recognized boundaries of the Krakow Urbanised Area and much bigger than the City of Krakow. Such studies were conducted, for instance, to prepare the City of Krakow Atlas (TRAFAS 1998), the Geochemical Atlas of the City of Krakow and its Surroundings (LIS, PASIECZNA 1995), the Sozological Map, Local Land Development Plans and the Small Retention Programme of the Province of Małopolska ([www.wrotamalopolski.pl](http://www.wrotamalopolski.pl)) together with detailed studies concerning the regulation of individual streams where there is a flood risk.

The Study of Determinants and Land Development Plans of Krakow (2003) contained a category of River Parks, which should be established in the surroundings of river valleys because of their special conditions and natural resources. The ranking list for Krakow is the following: *Urban Parks*: Błonia Węgrzynowickie (Kościelnicki Stok), Fort Mistrzejowice – Park Complex „Mistrzejowice”, Mydlniki Quarry, Kliny-Zacisze, Rakowski Forest, Płaszów-Camp, Ruczaj, Rząka, Skotniki, Tetmajera, Wróblowicki; *River Parks*: the Dłubnia (with the Baranówka), the Drwinka and the Serafa with the Malinówka, the Kościelnicki Stream, the Prądnik with tributaries (including the Białucha, the Sudół from Modlnica, the Bibiczanka and the Sudół Dominikański), the Rudawa (with the Młynówka Królewska), the Wilga (including the Cyrkówka and the Potok Siarczany), the Vistula and *Sections of River Parks*: Aleksandra (part of River Park of the Drwinka and the Serafa with the Malinówka), Dębicki (fragment of the Vistula River Park), the Drwinka (part of the River Park of the Drwinka and the Serafa with the Malinówka), John Paul II (part of the Wilga River Park), the Młynówka Królewska (the Rudawa River Park area), Nadwiślański (part of the Vistula River Park), Płaszów – the Gardens (part of the Vistula River Park), the Potok Siarczany (part of the Wilga River Park), the Rozrywka (part of the

Prądnik and Tributaries River Park), the Wilga-Rydlówka (the Wilga River Park area), Zakrzówek (BPTK area; refers to the Vistula River Park) ([www.krakow.pl/prasowka](http://www.krakow.pl/prasowka)). These areas, due to their significant transformation, are strongly urbanised. It is necessary to make their inventory, to eliminate pollution sources and to monitor them periodically. Some watercourses in towns, not only formally but also actually, due to their the load, culverting and incorporation into the water-sewerage infrastructure, being piped into a sewer system, losing hydrological continuity of some sections, and being subjected to periodical flow shortages etc., can hardly be recognised as uniform parts of surface waters or be treated as their significant elements. However, according to the implementation rules of the Water Framework Directive and in compliance with the Water Law standards, it is necessary to undertake actions to gradually eliminate pollution with priority substances and to stop or gradually eliminate the emission, draining and losses of priority hazardous substances (Dz.U.04.126.1318 (R)).

Anthropopressure, as reflected by quantitative and qualitative degradation, being the result of flow regulation and economic development of the drainage basin, makes an assessment of the scale of the human impact on the aquatic environment somewhat difficult. The survey of areas through which small watercourses flow, both within the district boundaries of the Krakow Urbanised Area and in the District of Krakow, shows that the areas drained by such streams are largely neglected and untidy and the described state is recognised only in critical and extraordinary situations, at times of environmental hazard of anthropogenic nature or during natural disasters, i.e., high water or flooding. The Water Framework Directive demands that particular attention be paid to biological elements such as environmental status indices or – in the case of urbanised watercourses – the ecological potential.

Based on the recommendations of the Water Framework Directive, an attempt has been made to assess the ecological status of selected tributaries of the Prądnik-Białucha River within the Krakow Conurbation. In this particular region, as reported by the media, the plans to create River Parks are at the highest risk of being abandoned. One reason is that the land allocated for this project is in part private property. The area within the park boundaries around the Sudół Dominikański (from Lublańska Road to the administrative boundaries of Krakow) and in the Prądnik Valley (from the administrative boundaries to Opolska Road) is planned for multi-storey development, which is confirmed by applications submitted by land developers to the Department of Architecture ([www.biznespolska.pl/wiadomosci/prasa](http://www.biznespolska.pl/wiadomosci/prasa)).

The objective of the study has been to verify the following hypothesis: the current method of land development and water and sewage management in the Sudół Dominikański Valley and in the area of the Prądnik-Białucha River lead to their quantitative and qualitative degradation.

## THE AREA UNDER STUDY

### Site studies

The Sudół Dominikański Stream has its source in the area of Zielonki District and flows to the City of Krakow from the north, where it loses its hydrological continuity with the natural channel and joins the Prądnik-Białucha River by means of a collecting pipe, thus becoming its highly urbanised left tributary (Figure 1). The length of the stream is 7 km. As it flows, it drains Basutów and Batowice, passes behind Batowicki Cemetery, and appears in Czerwony Prądnik from the east. On the website of the Prądnik Association, the following can be read: "Here, every couple of years, following a bigger rainfall, it gives us a hard time, flooding Majora Street and basements of nearby blocks of flats. It disappears into underground pipes near the junction of Majora Street and Dobrego Pasterza Road, to reappear once more for a couple of hundred meters between Dobrego Pasterza and Lublańska Roads. Some time ago, in the neighbourhood of the Dominican Friars Mill, it joined the waters of the Mill Stream (flowing from a weir on the Biały Prądnik). Before the roadway of Lublańska Road, it disappears again in a concrete underground collector, near the old slaughterhouse, by Olszecka Road. It joins the Białucha near Olszyny Road." ([www.republika.pl/towpradnik](http://www.republika.pl/towpradnik)).

In its upper section, the stream is an insignificantly transformed watercourse, flowing through an area densely covered by vegetation. It cuts across farmland, meadows and idle green areas. There are only private farmsteads on the land along the channel. In the City of Krakow, it flows among dense urban settlements, transport routes and near many local pollution sources. The watercourse encounters places of significant environmental transformation. Its channel has been subject to multiple, often "wild" regulations and in places its flow totally disappears. In its final section, the stream has been piped twice and where its waters are directed to the Prądnik-Białucha, the watercourse is a concrete collector, which functions as a storm-water drain.

The drainage basin of the Sudół Dominikański covers an area within two geological structures: the Silesian-Krakow Anticlinorium and the Pre-Carpathian Sink. Its south-eastern part, called Krakow Upland, is built of rocky and shoal limestone (with flints) and plate limestone, which form characteristic outliers and slope rocks emerging from a layer of loess clay and loess (GRADZIŃSKI 1972). Along its nearly entire length, it flows on Holocene floodplain terraces (clays, fen soil, sand and pebbles) lying on Quaternary loess (thickness *ca* 15 m), into which it cuts in places. In the valley slopes, some older formations appear, e.g. Upper Jurassic, Middle and Upper Cretaceous and Miocene sediments (BUKOWY 1956).

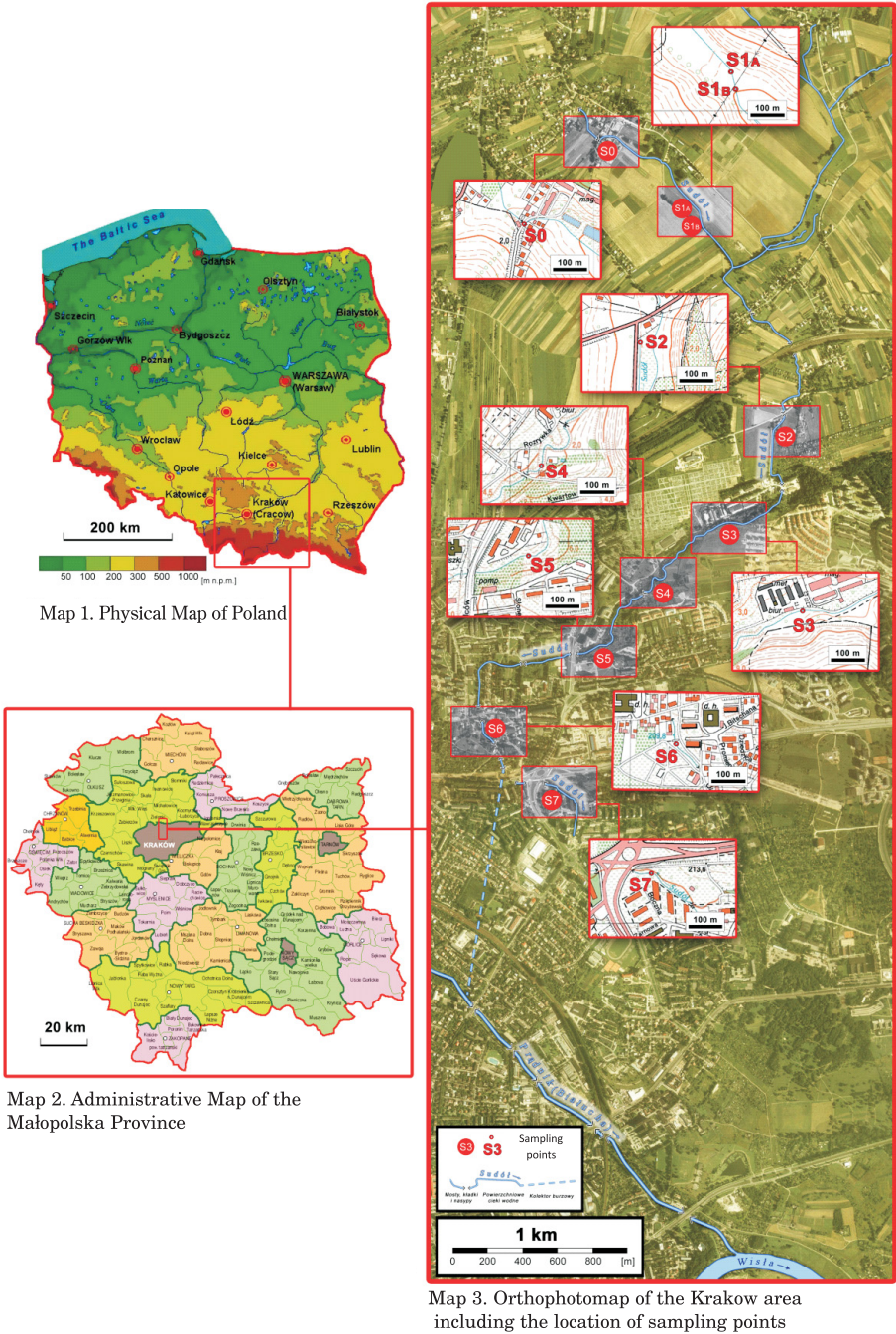


Fig. 1. Location of environmental sampling sites along the Rozrywka (Sudół Dominikański) Stream

The pollution sources of the stream's aquatic environment are:

- general domestic sewage (no sewerage systems, leaking septic tanks) and communal waste;
- disturbed hydro-relations;
- farming, gardens and allotments (plant pesticides, fertilisers, organic waste);
- industry (dry and wet atmospheric deposition, surface run-offs from storehouses and workshops);
- transport (heavy metal and oil product pollution, run-offs from transport bases and filling stations);
- civilisation pollution (corrosion and degradation of industrial infrastructure remains, area degradation, e.g. pits of a former brickyard in Zielonki filled illegally with communal waste, from which the run-offs infiltrate together with rainfall water and groundwater to surface and underground waters.

## SCOPE AND METHODS

### Site studies

During the site studies (September 2005), a detailed zoological-ecological mapping of the Sudół Dominikański Stream Valley was performed. Particular attention was paid to the existing or potential pollution sources of the stream, shape and conservation of the natural state of the channel, type of economic development and the green cover, presence of macrophytes growing in the watercourse or partly submerged ones, type of water flow, disappearance or loss of hydrological continuity. Environmental samples were taken for laboratory analyses at 8 points (Figure 1), the exact position of which was determined using a GPS receiver. They covered the following material:

- water (6 samples, S1-S6);
- bottom sediments, surface layer (8 samples, S1-S7);
- bottom sediment cores (2 cores, S2 and S7);
- macrophytes, in most cases waterside, single specimens growing in the watercourse (evenly along the entire sampling route).

After performing tests for indices sensitive to transport or storage conditions (pH, Eh and electrolyte conductivity), the samples were protected and stored cooled, isolated from light and oxygen until analysed in the laboratory. Water samples were filtered through 0.45 µm pore membrane filters. The samples for metal cation concentration assays were acidified.

The state of the aquatic environment of the Sudół Dominikański watercourse was compared with that of the Prądnik-Białucha River studied in the previous year (September 2004).

### Laboratory analyses

Water samples were analysed for the concentration of heavy metals: Cd, Cu, Pb and Zn, with the use of inductively coupled plasma atomic emission spectroscopy (ICP-AES Perkin-Elmer Plasma 40 instrument). The anion content ( $\text{Cl}^-$ ,  $\text{NO}_3^-$ ,  $\text{SO}_4^{2-}$ ,  $\text{PO}_4^{3-}$ ) was determined using an ion chromatograph (Dionex DX-100 Ion Chromatograph). The suspension density in the water samples was determined. In the aqueous suspension (fraction  $> 0.45 \mu\text{m}$ ) and bottom sediments (fraction  $< 0.063 \text{ mm}$ ), concentration of the metals was determined after solubilising them by extraction with concentrated nitric acid (65%) at  $130^\circ\text{C}$  and using flame atomic absorption spectrometry (AAS-PU 9).

## RESULTS ANALYSIS AND DISCUSSION

### Water samples

The water in the Sudół Dominikański Stream showed a high degree of turbidity, especially in its upper section, where engineering works were being carried out and the channel was mostly free from crust vegetation. The pH values of water samples did not vary essentially along the stream course, ranging between 7.60 and 7.98. Oxidative conditions were found with the Eh (mV) values from +135 to +190, whereas the electrolytic conductivity EC ( $\text{mS cm}^{-1}$ ) was within the 0.96-1.47 range (average value 1.25) – Table 1.

Table 1

Parameters determined for water samples from the Sudół Dominikański Stream

Sample	Parameter		
	pH	Eh (mV)	EC ( $\text{mS cm}^{-1}$ )
S1a	6.90	+125	0.47
S1b	7.30	+98	0.52
S2	7.60	-135	0.59
S3	7.55	-95	0.98
S4	7.53	-85	1.03
S5	7.41	-35	0.58
S6	7.65	-71	0.39
S7	7.55	+30	0.73
Min	6.91	-135	0.39
Max	7.65	+125	1.03

The concentration of anions determined in water samples varied, generally increasing downstream. Concentrations ( $\text{mg dm}^{-3}$ ) of  $\text{Cl}^-$ ,  $\text{NO}_3^-$ ,  $\text{SO}_4^{2-}$ ,  $\text{PO}_4^{3-}$  varied within the ranges 35-76 (average value 58), 1.5-23 (average value 10), 55-92 (average value 80) and 6.7-10 (average value 9.4), respectively (Table 2).

Table 2

Concentration of some anions in water samples from  
the Sudół Dominikański Stream

Sample	$\text{Cl}^-$	$\text{NO}_3^-$	$\text{SO}_4^{2-}$	$\text{PO}_4^{3-}$
	$(\text{mg dm}^{-3})$			
S1	34.8	1.47	54.6	<1.2*
S2	74.1	<0.3*	91.6	6.74
S3	41.1	<0.3*	75.9	7.02
S4	54.8	3.04	79.4	6.73
S5	66.2	12.0	89.3	10.3
S6	76.2	23.2	87.0	9.26
Min	34.8	1.47	54.6	6.73
Max	76.2	23.2	91.6	10.3
Mean	57.8	9.9	79.6	9.41

\* threshold level acc. to WÓJCIK et al. (1999)

Maximum amounts ( $\mu\text{g dm}^{-3}$ ) of Cd, Cu, Pb and Zn were at the level of 0.045, 5.00, 0.30 and 24.2, respectively (Table 3).

Table 3

Concentration of some metals in water samples from  
the Sudół Dominikański Stream

Sample	Cd	Cu	Pb	Zn
	$(\mu\text{g dm}^{-3})$			
S1	0.045	2.30	0.20	11.7
S3	0.036	4.30	0.30	24.2
S6	0.020	5.00	0.20	10.0

### Bottom sediment and water suspension samples

Bottom sediments from the Sudół Dominikański showed insignificant differences in the pH value, which varied in the range 6.90-7.65, and highly varying redox potentials: in the initial and final sections the conditions were

oxidative, whereas in the middle section they were reductive. The electrolytic conductivity (EC) values ( $\text{mS cm}^{-1}$ ) of sediments of similar degree of hydration ranged between 0.39 and 1.03 (average 0.66) – Table 4.

Table 4

Parameters of bottom sediment samples from the Sudół  
Dominikański Stream

Sample	Parameter		
	pH	Eh (mV)	EC ( $\text{mS cm}^{-1}$ )
S1a	6.90	+125	0.47
S1b	7.30	+98	0.52
S2	7.60	-135	0.59
S3	7.55	-95	0.98
S4	7.53	-85	1.03
S5	7.41	-35	0.58
S6	7.65	-71	0.39
S7	7.55	+30	0.73
Min	6.91	-135	0.39
Max	7.65	+125	1.03

In the case of bottom sediment cores sampled at the point S2, both in its top zone (-211 mV) as well as in its bottom zone (-134 mV), reductive conditions were found (Figure 2). Both depth profiles show a difference with respect to lithology. The first one, sampled in the upper section of the stream (S2) down to 20 cm depth, is composed of dark sediments, highly nourished, turning into brown sandy sediments in the lower zones. A sediment core taken in the downstream section of the stream (S7) is formed of uniform, dark, sandy-silt with an insert of clay sediments appearing only at the depth of 13-14 cm.

Bottom sediments were characterised by variable particle size distribution. The finest grained samples, for which 90 wt.% was the powdery-clay fraction, were collected in the upper section of the stream. The other samples contained 2–89 wt. % of the powdery-clay fraction. The content of metals found in the bottom sediments generally showed a rising tendency downstream. The amounts ( $\text{mg kg}^{-1}$ ) of Cu, Pb and Zn ranged between 7-73 (average 27), 16-124 (average 46) and 47-943 (average 257), respectively. The values that exceeded the geochemical background (also local) were mostly found for lead (Table 5).





Fig. 3. Changes in lithology in the bottom sediment cores from the Sudół Dominikański Stream

Table 5

Concentration of some metals in bottom sediment samples  
from the Sudół Dominikański Stream

Sample	Parameters			
	fr.<0.063 mm (%)	Cu (mg kg <sup>-1</sup> )	Pb (mg kg <sup>-1</sup> )	Zn (mg kg <sup>-1</sup> )
S1a	97.3	12	16	47
S1b	88.6	19	22	102
S2	42.6	24	33	161
S3	63.7	12	23	58
S4	43.7	35	38	263
S5	77.8	7	28	55
S6	1.6	73	124	943
S7	50.3	33	85	426
Min	1.63	7	16	47
Max	97.30	73	124	943
Mean	41.80	27	46	257
Local background for the Vistula River (HELIOS-RYBICKA 1986)		40	45	110
Geochemical background (TUREKIAN, WEDEPOHL 1961)		45	20	95

The suspension density ranged between 0.001 and 0.078 (g dm<sup>-3</sup>). The most polluted suspension samples were found, as in the case of bottom sediments, in the lower section of the stream, and the Pb and Zn content (mg kg<sup>-1</sup>) there reached 8600 and 2800, respectively.

Strong linear correlations were found between the analysed metals. The linear correlation coefficient ( $R^2$ ) takes values >0.8 (Figure 3).

The results were compared to the current Polish regulations and, in the case of sediment samples, also the German ones.

Water assessment:

- Minister for the Environment Regulation of 11 February on the classification for presenting the state of surface and underground waters, the method for conducting the monitoring, the method for interpretation of the results, and the presentation of the state of these waters (Dz.U. 04.32.284(R)) (cancelled – none currently in force);

Bottom sediment assessment:

- classification of aqueous sediments based on geochemical criteria (BOJAKOWSKA, SOKOŁOWSKA 1998),

- German classification of sediments and aqueous suspension LAWA (Imer 1997, LAWA 1998),
- classification based on the geoaccumulation index – Igeo (MÜLLER 1981).

With respect to the five-degree classification of surface waters (Dz.U. 04.32.284(R)), no cases of exceeded concentration of  $\text{Cl}^-$  or  $\text{SO}_4^{2-}$  anions were found (Class I), whereas the amount of  $\text{NO}_3^-$  anions was slightly elevated in the downstream section (Class II). Phosphates occurred in the highest concentrations and therefore the tested water samples were classified as Class V. According to the LAWA (1998) classification, the sediments in the upper section of the stream may be recognised as non-polluted with respect to the concentration of the concerned metals (Class I and I-II), in the mid-

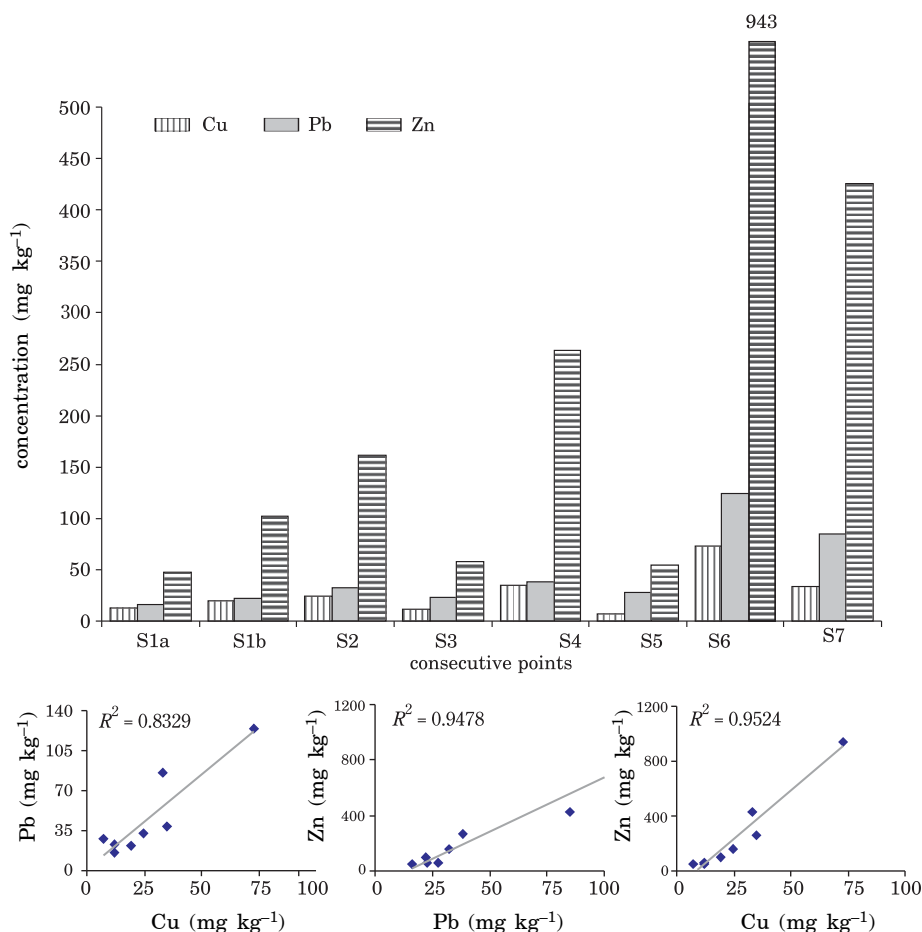


Fig. 3. Variability in Cu, Pb, and Zn concentrations in sediments sampled downstream of the Sudół Dominikański Stream and linear dependences between concentrations of individual metals

dle section as slightly polluted and in the lower section as highly polluted. In the case of the two other classifications, the situation looks similar (Table 6).

Table 6

Classification of bottom sediments from the Sudół Dominikański Stream with respect to their Cu, Pb and Zn concentration

Sample	Parameters									General assessment		
	Cu (mg kg <sup>-1</sup> )			Pb (mg kg <sup>-1</sup> )			Zn (mg kg <sup>-1</sup> )					
	1	2	3	1	2	3	1	2	3	1	2	3
S1a	I	I	0	I	I	0	I	I	0	I	I	0
S1b	I	I	0	I	I	0	I-II	I	0	I-II	II	1
S2	I-II	II	0	I-II	I	1	I-II	I	1	I-II	I	1
S3	I	I	0	I	I	0	I	I	0	I	I	0
S4	I-II	II	0	I-II	I	1	II	II	1	II	II	1
S5	I	I	0	I-II	I	0	I	I	0	I-II	I	0
S6	II	II	1	II-III	II	3	III	II	3	III	II	3
S7	I-II	II	0	II	II	2	II-III	II	2	II-III	II	2

1) acc. to LAWA classification (1998)

2) acc. to geochemical classification by BOJAKOWSKA, SOKOŁOWSKA (1998)

3) acc. to geoaccumulation index Igeo (MÜLLER 1981)

## Vegetation

The studies of aquatic vegetation, as indices of the changes in the ecological state of the Sudół Dominikański Stream, should be treated at this stage as preliminary ones, i.e. making an inventory. Studies on aquatic vegetation should be integrated into the monitoring of the state or the ecological potential of the watercourse, which includes assays of the pH value, concentration of salts and oxygen content in the sediments, as well as the content of heavy metals. In a relatively simple way, with the aid of hydrobotany experts, it will be possible to complement the studies with biological elements recommended by WFD (SZOSZKIEWICZ et al. 2002).

In the Sudół Dominikański Valley, at sections (S1-S6) adjunct to the sampling points, the following plants were found:

- S1. Reed canary grass (*Phalaris arundinaceae*), marshpepper knotweed (*Polygonum hydropiper*), hedge bindweed (*Calystegia sepium*), stinging nettle (*Urtica dioica*), common amaranth (*Amaranthus retroflexus*);
- S2. Hairy willow herb (*Epilobium hirsutum*), tansy (*Tanacetum vulgare*), marshpepper knotweed (*Polygonum hydropiper*), mugwort (*Artemisia vulgaris*), nodding beggartick (*Bidens cernua*), burning nettle (*Urtica urens*), greater celandine (*Chelidonium majus*);

- S3. Creeping buttercup (*Ranunculus repens*), wood avens (*Geum urbanum*), bluegrass (*Poa sp.*), shepherd's purse (*Capsella bursa pastoris*), bishop's goutweed (*Aegopodium podagraria*), tansy (*Tanacetum vulgare*), hedge woundwort (*Stachys sylvatica*);
- S4. Water chickweed (*Myosoton aquaticum*), marshpepper knotweed (*Polygonum hydropiper*);
- S5. Brome grass (*Bromus sp.*), reed canary grass (*Phalaris arundinaceae*), unbranched bur-reed (*Sparganium emersum*);
- S6. Creeping bentgrass (*Agrostis stolonifera*).

Among 21 species found on the bank slopes during the studies, as many as 11 belong to apophytes (native taxons preferring transformed sites), and further three are characterised by a high degree of hemerophily (positively respond to the growth of transformation). Among the identified apophytes, typically ruderal species dominate, including celandine (*Chelidonium majus*), burning nettle (*Urtica urens*), common amaranth (*Amaranthus retroflexus*) and tansy (*Tanacetum vulgare*). Most of the species tolerant to transformation occurred at site S2. All the 7 species found at this site respond positively to anthropogenic pressure. The species found at site S3 indicate its significant shading and a potential forest site.

### Comparison of the cleanliness of the Sudół Dominikański and the Prądnik-Białucha River

Based on the results of the studies on the Sudół Dominikański Stream (September 2005) and the Prądnik-Białucha River fed by this stream (September 2004), a comparison was made between the cleanliness of both watercourses. It allowed us to state whether the Sudół Dominikański Stream could be a source of heavy metal pollution of the Prądnik-Białucha River. First, the ranges of the parameters determined for these watercourses, pH, Eh and electrolytic conductivity, were compared for both watercourses (Table 7). The pH values vary in both cases in the range from slightly acidic to slightly alkaline. It may be said that the aquatic environment of the

Table 7

Comparison of some parameter value ranges (averages) for the Sudół Dominikański Stream and the Prądnik-Białucha River

Parameters	The Sudół Dominikański	The Prądnik - Białucha
pH	6.91: 7.65	6.74: 7.34
Eh (mV)	+125 : -135	178 : - 350
Electrolytic conductivity (mS cm <sup>-1</sup> )	0.391: 1.030 (0.660)	0.418 : 1.080 (0.810)
Cu (mg kg <sup>-1</sup> )	7 : 73 (27.0)	151 : 729 (321.7)
Pb (mg kg <sup>-1</sup> )	16 : 124 (46.1)	27 : 108 (64.7)
Zn (mg kg <sup>-1</sup> )	47 : 943 (257)	148 : 675 (331.7)

sediments from the Prądnik-Białucha River are more reduced than those from the stream flowing into it. This might result from the presence of sediments of greater thickness in the bed of the Prądnik-Białucha River than in the smaller and shallower Sudół Dominikański. In both cases, the salt concentration, expressed by electrolytic conductivity, may be described as relatively small. However, in the case of heavy metals, the average Cu concentration is more than 10-fold higher in the sediments of the Prądnik-Białucha River than in those from the Sudół Dominikański. For Pb, both the concentration plots, and the mean concentration are at a level close to 50 ( $\text{mg kg}^{-1}$ ). A greater span of values was observed for Zn concentration ranges in the sediments from the Sudół Dominikański, whereas this range for the sediments from the Prądnik-Białucha was narrower but its average value was higher.

By comparing the results of the analysis of the above parameters for two sites located at the Prądnik-Białucha River, before and after the outlet of the Sudół Dominikański collector, we looked for any significant differences (Table 8). Taking into account the fact that the differences in metal concentrations in sediments are generated by many factors (such as grain size distribution and mineral composition of the sediments, sampling site, other 'enriching' sources or factors making the sediments lean), it is thought that the outlet of the storm water drain, such as the Sudół Dominikański collector, slightly cleans the sediments from heavy metals. The phenomenon of washing out metals from sediments in an acidic environment, characteristic of storm waters in the City of Krakow, is also confirmation of this thesis. It seems that such a remarkable difference in the pH values in the bottom sediment environments, as well as the oxidation of bottom sediments from the value of -67 mV to +142 mV, may be explained by the action of storm sewers. It was also checked if there was a relationship between the cleanliness of both watercourses by comparing the results of measurements made at two sites localised at the Prądnik River, upstream and downstream of the place where the Sudół Dominikański joins the Prądnik (Table 8).

Table 8

Summary of some parameter values for the Prądnik-Białucha River upstream (I) and downstream (II) of the Sudół Dominikański Stream inflow

Parameters	I	II
pH	7.80	4.16
Eh (mV)	-67.0	142
Electrolytic conductivity ( $\text{mS cm}^{-1}$ )	no data	0.32
Cu ( $\text{mg kg}^{-1}$ )	42	22
Pb ( $\text{mg kg}^{-1}$ )	56	44
Zn ( $\text{mg kg}^{-1}$ )	211	148

ADYNKIEWICZ-PIRAGAS and DRABIŃSKI (2001) assessed the impact of hydrotechnical development projects on the environmental state of the Smortawa River. The channel regulation carried out in the lower section of the river and the construction of dam type weirs and fall stages generally caused a decrease in species diversity and changes in the species composition of macrophyte assemblages (aquatic, off-bankside and bankside zones), plankton and periphyton, in comparison to the non-regulated section. Also the ecomorphologic valorisation, performed by Ilnicki method (ILNICKI, LEWANDOWSKI 1997) showed a fall in the naturalness of the hydrotechnically transformed section.

## SUMMARY AND CONCLUSIONS

The waters of the Sudół Dominikański, because of their pH value and the Zn, Pb and Cu content, correspond to Class I purity waters with respect to the current standards. However, because of their high phosphate content in all the studied sites, the waters are classified as Class V, which means non-potable water. This low assessment with regard to water purity class is also affected by the concentration of river suspension.

The Cu, Pb and Zn content in the sediments and the suspension increase downstream, which is certainly affected by the rise in the urbanisation of the valley. According to the classification assumed for the purposes of assessment of the pollution of sediments with heavy metals, the sediments in the upper section of the stream may be recognised as non-polluted, in the middle section as slightly polluted and in the lower section as highly polluted.

The aquatic environment of the Prądnik-Białucha sediments is more reductive than the Sudół Dominikański Stream flowing into it, with the pH values in both cases varying from slightly acidic to slightly alkaline, while the salt content of both watercourses may be described as relatively low. In the Prądnik-Białucha sediments, the average concentration of Cu is over 10-fold higher but and that of Pb is similar in both samples (*ca* 50 mg kg<sup>-1</sup>). A larger span of values characterises the Zn concentration ranges in the sediments of the Sudół Dominikański. The Zn range for the Prądnik-Białucha is narrower but shows a higher average concentration.

A positive process of sediment purification by storm waters is observed downstream the collector outlet, where it is compounded by the washing out of the metals with the acidic environment characteristic of storm waters in Krakow. In the vicinity of both watercourses, particularly the Sudół Dominikański, the presence of exceptionally adverse factors was observed (incorrect constructions around the banks, no access to the channel, the presence of illegal rubbish dumps, non-regulated water and sewage management).

Our preliminary survey of aquatic vegetation (small total abundance of macrophytes and a large share of ruderal species of apophytes) points to very strong anthropopressure associated with morphological transformations and eutrophication. In the places where the morphological stressor exceeded the limits of ecological tolerance of macrophytes, we observed total disappearance of tracheophytes and the appearance of resistant, macroscopic structural algae of the *Cladophora* genus.

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# **EFFECT OF GROWTH REGULATORS ON THE MINERAL BALANCE IN SPRING TRITICALE**

**Jadwiga Wierzbowska, Stanisław Sienkiewicz,  
Teresa Bowszys**

**Chair of Agricultural Chemistry and Environmental Protection  
University of Warmia and Mazury in Olsztyn**

## **Abstract**

It is assumed that progress in cultivar breeding corresponds to over 50% of the increase in cereal yields. Among possible ways of improving the success of fertilisation and, consequently, increasing yields and enhancing the mineral balance in crops is the application of growth regulators.

The purpose of this study has been to assess the effect of growth regulators and their doses on the mineral balance in spring triticale. The research was based on a two-factor pot experiment, completed in four replicates. The experiment was carried out in Kick-Brauckmann pots filled with light soil of slightly acidic reaction and highly abundant in available forms of P, moderately abundant in K and poor in Mg. Mineral fertilisation per pot consisted of 1.5 g N, 2.0 g K and 0.25 g Mg. Nitrogen and potassium were applied in two rates – half the complete dose before sowing and the remaining amount as top dressing during the stem elongation phase. All the rate of magnesium was introduced to soil before sowing.

Prior to sowing the tested cereal, grain of cv. Migo spring triticale was soaked for 24 hours in water (control) or in aqueous solutions of growth regulators: IBA (indole butyric acid), NAA ( $\alpha$ -naphthaleneacetic acid), BAP (benzyl adenine): 5, 10, 20 mg dm<sup>-3</sup>; GA<sub>3</sub> (gibberellin acid): 20, 40, 80 mg dm<sup>-3</sup> and Tria (triacontanol): 0.1, 0.2, 0.2 mg dm<sup>-3</sup>. Spring triticale was harvested in the full maturity stage.

An increase in the total nitrogen concentration in grain ranged from 6% following an application of GA<sub>3</sub> to 15% after using NAA. Owing to its increased concentration and higher yield of triticale, the accumulation of nitrogen in grain increased by about 20% in plants treated with NAA and by 15-17% when the seed material was dressed with Tria, BAP or GA<sub>3</sub>. IBA produced the weakest effect, increasing the accumulation of this nu-

trient in grain by just 8%. This effect was obtained mainly by improved remobilisation and transport of N from vegetative parts to grain. The contribution of grain to nitrogen accumulation varied from 63% (control) to 71-73% after an application of the tested growth regulators. Under the influence of IBA, the concentration of potassium (by 14%) and the share of straw in potassium accumulation increased. In respect of the other macronutrients, the influence of growth regulators was less evident.

Key words: spring triticale, mineral balance, growth regulators.

## WPŁYW REGULATORÓW WZROSTU NA GOSPODARKĘ MINERALNĄ PSZENŻYTA JAREGO

### Abstrakt

Szacuje się, że o wzroście plonów zbóż w ponad 50% decyduje postęp odmianowy. Jedną z możliwości zwiększenia efektywności nawożenia, a tym samym zwiększenia plonowania i poprawienia gospodarki mineralnej roślin, jest stosowanie regulatorów wzrostu.

Celem pracy była ocena wpływu regulatorów wzrostu i ich dawek na gospodarkę mineralną pszenżyta jarego. Podstawę badań stanowiło dwuczynnikowe doświadczenie wazonowe, wykonane w 4 powtórzeniach. Badania prowadzono w wazonach typu Kick-Brauckmanna napełnionych glebą lekką o odczynie lekko kwaśnym oraz bardzo wysokiej zasobności w przyswajalne formy P, średniej w K oraz niskiej w Mg. Nawożenie mineralne na wazon wynosiło odpowiednio: 1,5 g N, 2,0 g K oraz 0,25 g Mg. Azot i potas zastosowano w 2 dawkach – połowę przedsięwzięcia, a resztę pogłównie w fazie strzelania w źdźbło, natomiast magnez w całości przedsięwzięcia.

Przed siewem ziarno pszenżyta jarego odmiany Migo moczone 24 h w wodzie (kontrola) lub w wodnych roztworach regulatorów wzrostu: IBA (kwas indolilomasłowy), NAA (kwas  $\alpha$ -naftylooctowy), BAP (benzyladenina) – 5; 10; 20 mg dm<sup>-3</sup>; GA<sub>3</sub> (kwas gibereliny) – 20; 40; 80 mg dm<sup>-3</sup> oraz Tria (triacontanol) – 0,1; 0,2; 0,2 mg dm<sup>-3</sup>. Pszenżyto jare zebrano w fazie dojrzałości pełnej.

Przyrost zawartości azotu ogółem w ziarnie wahał się od 6% do 15% po zastosowaniu odpowiednio GA<sub>3</sub> i NAA. Na skutek zwiększonej koncentracji i przyrostu plonu, akumulacja tego składnika w ziarnie wzrosła o ok. 20% na obiektach traktowanych NAA, a 15-17% po zaprawianiu materiału siewnego Tria, BAP lub GA<sub>3</sub>. Najslabiej działał IBA, bowiem przyrost akumulacji tego składnika w ziarnie wynosił tylko 8%, co uzyskano głównie dzięki zwiększonej remobilizacji i odprowadzaniu N z części wegetatywnych do ziarna. Udział ziarna w akumulowaniu azotu wahał się od 63% (kontrola) do 71-73% po zastosowaniu badanych regulatorów wzrostu. Pod wpływem IBA zwiększyła się zawartość potasu (o 14%) oraz udział słomy w jego gromadzeniu. W przypadku pozostałych makroskładników działanie regulatorów wzrostu było mniej widoczne.

Słowa kluczowe: pszenżyto jare, gospodarka mineralna, regulatory wzrostu.

## INTRODUCTION

The economic importance of spring triticale, compared to other cereals, is small. Observations made for many years have demonstrated that on poorer soils, triticale can produce higher yields than spring wheat. Grain of spring triticale, like that of winter forms, is considered as one of the most

valuable fodder cereals owing to its high protein concentration and metabolic energy, comparable to, and sometimes even higher than that in wheat grain (SOBKOWICZ, PODGÓRSKA 2006). The fact that triticale contains less of anti-nutritive substances creates many possibilities of using its grain for feeding purposes (KONDRACKI 2000). Moreover, triticale is the cereal whose grain can be used for human consumption (HABER, LEWCZUK 1990, SOBCZYK et al. 2009). The soil and climatic conditions as well as agronomic practice are the factors which condition the yield and amino acid composition of proteins, which in turn shape the fodder and nutritional values of triticale grain (SPYCHAJ-FABISIAK et al. 2005, STANKIEWICZ 2005). Spring cultivars of triticale are characterised by a higher concentration of proteins than winter forms (SPYCHAJ-FABISIAK et al. 2005, GIL 1995, GIL, NARKIEWICZ-JODKO 1997).

Application of growth regulators is one of the ways of improving yields and mineral balance of crops. The effect of using such substances depends on many factors, e.g. a crop species or even its cultivar, environmental conditions or fertilisation (CZAPLA et al. 2007, CZAPLA, NOGALSKA 2000a,b, WIERZBOWSKA 2006a,b, WIERZBOWSKA, BOWSZYS 2008a,b, WIERZBOWSKA et al. 2007).

The objective of this study has been to evaluate the influence of growth regulators on mineral balance in spring triticale.

## MATERIAL AND METHODS

The study was based on a two-factor pot experiment, performed in four replicates. The trials were set up in Kick-Brauckmann pots, filled with 9.5 kg of light soil, slightly acidic in reaction (pH 6.4 in 1 mol KCl dm<sup>-3</sup>) and very highly abundant in the available form of P (92.6 mg kg<sup>-1</sup>), moderately abundant in K (104.2 mg kg<sup>-1</sup>) and poor in Mg (30 mg kg<sup>-1</sup>). Mineral fertilisation per pot consisted of 1.5 g N in the form of urea, 2.0 g K in the form of KCl and 0.25 g Mg as MgSO<sub>4</sub>·H<sub>2</sub>O. Nitrogen and potassium were applied in two rates: 50% before sowing and the remaining amount as top dressing during the stem elongation phase; in contrast, all magnesium was introduced to soil before sowing.

In each pot, 25 plants of cv. Migo spring triticale were grown. Before sowing, seeds of spring triticale were soaked for 24 hours in water (control) or in aqueous solutions of growth regulators (Table 1). Triticale was harvested in the full maturity phase. The plant material, having been mineralised in concentrated sulphuric (VI) acid with an addition of hydrogen peroxide, was assayed to determine the concentration of nitrogen (spectrophotometrically with the hypochlorite method), phosphorus (with the vanadium-molybdenene method), potassium and calcium (with the flame photometric method, ASA) and magnesium (with the atomic absorption method, AAS).

Table 1

Design of the experiment			
Variant	Dose of growth regulators (mg dm <sup>-3</sup> )		
	I	II	III
Control (aqua destillata)			
IBA	5	10	20
NAA	5	10	20
GA3	20	40	80
BAP	5	10	20
Tria	0.1	0.2	0.4

The results of the chemical analyses underwent analysis of variance. Tukey's test at the level of significance  $\alpha=0.01$  was performed in order to verify the data.

## DISCUSSION OF THE RESULTS

The concentration of macronutrients in cereal grains changes depending on the weather conditions, soil abundance and agronomic practice. Growth regulators had a significant effect on the total nitrogen concentration in spring triticale grain (Table 2). The highest increase (by 14.8% in the concentration of this nutrient was obtained when the seed material had been treated with synthetic auxin NAA). The other phytohormones raised the concentration of nitrogen by 6.2-7.8% versus the control. Triacontanol increased the concentration of phosphorus and potassium in grain by *ca* 7% and that of magnesium by over 18%. Compared to the control, the concentration of nitrogen in husks decreased under the influence of phytohormones by 56-61%. Moreover, the auxins (IBA and NAA) and gibberillin depressed the concentration of phosphorus by about 25% while NAA and BAP significantly decreased the concentration of P in straw. In turn, the concentration of potassium in straw rose significantly after treating the seed material with the growth regulators.

BRZOZOWSKA (2006) points to the fact that some herbicides can cause an increase in the concentration of total nitrogen in winter triticale grain but do not have any effect on the concentration of other macronutrients. CZAPLA et al. (2007) concluded that growth regulators and their mixtures with magnesium sulphate VI differentiated the concentration of nitrogen and magnesium in spring triticale. Triticale grain was characterised by a higher concentration of these nutrients after an application of any of the growth regulators (except Tria); however, when the growth regulators were applied together with magnesium sulphate (VI), the concentration of nitrogen gen-

Table 2

Concentration of macronutrients ( $\text{g kg}^{-1}$ )

Growth regulator (pgr)	Concentration (mg dm <sup>-3</sup> )	Grain					Husk					Straw				
		N	P	K	Ca	Mg	N	P	K	Ca	Mg	N	P	K	Ca	Mg
Control		25.60	3.80	3.60	0.20	1.10	13.70	1.40	8.50	1.10	0.70	11.40	0.80	18.70	3.80	1.10
	5	27.20	3.80	3.60	0.30	1.20	4.00	1.00	8.30	1.10	0.60	8.80	0.60	20.70	4.00	1.20
	10 20	27.40 27.60	3.80 3.70	3.60 3.60	0.30 0.30	1.20 1.00	6.20 8.00	1.20 1.00	8.30 8.70	1.30 1.20	0.60 0.60	10.80 12.40	0.80 0.80	21.20 22.00	4.80 6.40	1.30 1.70
Mean for IBA		27.40	3.77	3.60	0.30	1.13	6.07	1.07	8.43	1.20	0.60	10.67	0.73	21.30	5.07	1.40
NAA	5	28.80	4.10	3.80	0.20	1.30	8.00	1.00	8.70	1.20	0.60	10.20	0.80	20.00	4.40	1.20
	10	30.60	3.80	3.80	0.20	1.30	5.40	1.00	9.40	1.20	0.60	11.00	0.60	20.00	4.20	1.20
	20	28.80	3.60	3.40	0.20	1.00	4.00	1.00	9.40	1.20	0.50	10.80	0.80	20.00	4.00	1.10
Mean for NAA		29.40	3.83	3.67	0.20	1.20	5.80	1.00	9.17	1.20	0.57	10.67	0.73	20.00	4.20	1.17
GA <sub>3</sub>	20	26.40	3.90	3.80	0.20	1.20	4.00	1.00	8.50	1.20	0.60	10.20	0.80	18.50	3.20	1.10
	40	27.20	3.90	3.60	0.20	1.20	6.20	1.00	8.10	1.20	0.60	11.00	0.90	19.40	4.00	1.20
	60	28.00	4.00	3.80	0.30	1.30	7.00	1.10	9.20	1.30	0.60	11.00	0.80	19.80	4.00	1.30
Mean for GA <sub>3</sub>		27.20	3.93	3.73	0.23	1.23	5.73	1.03	8.60	1.23	0.60	10.73	0.83	19.23	3.73	1.20
BAP	5	27.60	3.90	3.60	0.20	1.20	5.40	1.30	9.40	1.40	0.60	11.00	0.60	20.00	5.00	1.20
	10	28.00	3.80	3.60	0.20	1.20	5.40	1.60	9.20	1.20	0.60	12.00	0.70	18.40	3.30	1.10
	20	27.20	3.80	3.80	0.20	1.20	4.00	1.10	9.00	1.20	0.60	10.20	0.60	19.80	3.80	1.10
Mean for BAP		27.60	3.83	3.67	0.20	1.20	4.93	1.33	9.20	1.27	0.60	11.06	0.63	19.40	4.03	1.13
Tria	0.1	27.20	3.80	3.60	0.20	1.30	4.00	1.30	8.50	1.20	0.60	11.00	0.80	20.00	4.60	1.30
	0.2	28.80	4.20	4.00	0.30	1.30	5.80	1.30	9.00	1.20	0.60	12.80	0.80	20.00	4.40	1.30
	0.4	26.80	4.20	4.00	0.20	1.30	5.40	1.30	8.30	1.10	0.60	11.00	0.90	20.00	4.10	1.30
Mean for Tria		27.60	4.07	3.87	0.23	1.30	5.07	1.30	8.60	1.17	0.60	11.60	0.83	20.00	4.37	1.30
LSD <sub>0.01</sub> for pgr		1.263	0.258	0.265	n.i.	0.157	2.530	0.288	0.606	n.i.	n.i.	n.i.	0.139	0.928	n.i.	n.i.

erally declined, both in grain and in vegetative parts. Mixtures of the growth regulators with magnesium sulphate increased the content of Mg in the flag leaf.

In another study on spring wheat, growth regulators, especially auxin and gibberellin, depressed the concentration of nitrogen in grain and vegetative of wheat except the second leaf (WIERZBOWSKA, BOWSZYS 2008a). Some other investigations have demonstrated that under the influence of kinetin and auxin, the concentration of potassium in grain and vegetative organs of spring wheat increased but decreased under the effect of gibberellin (WIERZBOWSKA 2006a). In turn, the concentration of calcium in wheat plants was more strongly dependent on growth regulators than on the rate of potassium fertilisation. Growth regulators only slightly raised the concentration of magnesium in grain, but distinctly decreased its level in stems and leaves (WIERZBOWSKA 2006b). In turn, foliar application of NAA and magnesium sulphate (VI) increased the concentration of macronutrients in spring barley grain and modified their concentration in vegetative parts of this crop (CZAPLA, NOGALSKA 2000b).

In the present study, the highest protein yield per pot (8.53 g) was obtained after an application of NAA and the lowest one (7.68 g) when seeds were treated with another synthetic auxin, namely IBA. It was found out that the value of protein yield per pot rose correspondingly to doses of growth regulators, but it was only when the seed material had been treated with IBA that the highest protein yield was obtained as a result of the application of the second rate of this phytohormone (Figure 1).

Compared to the control, when IBA had been applied, the uptake of nitrogen with the spring triticale's biomass slightly rose, in contrast to the other growth regulators, which caused its small decline (Figure 2). Under the influence of the phytohormones, the accumulation of nitrogen in grain increased, especially when NAA and GA<sub>3</sub> had been applied as they raised the accumulation of this nutrient by about 20%. Except IBA, the growth regulators caused enhanced accumulation of phosphorus in the biomass and in grain (from 6% by NAA and BAP to 12% by GA<sub>3</sub> and Tria). IBA and triacontanol increased by 7% the accumulation of potassium in the aerial biomass of triticale, mainly in straw.

By treating the seed material with the growth regulators, it was possible to increase the contribution of grain to nitrogen accumulation in triticale's aerial biomass, mainly by decreasing (2.5- to 3-fold) the share of husks in the accumulation of this element. Compared to the control, NAA limited the distribution of potassium, calcium and magnesium to grain, but led to an increased accumulation of these elements in straw. A reverse tendency was observed under the influence of gibberellin.

WIERZBOWSKA and BOWSZYS (2008a) demonstrated that growth regulators caused a decrease in the accumulation of nitrogen in aerial parts of wheat, especially in grain, but under the effect of gibberellin, the accumulation of this nutrient in husks and stems rose. The contribution of grain to the



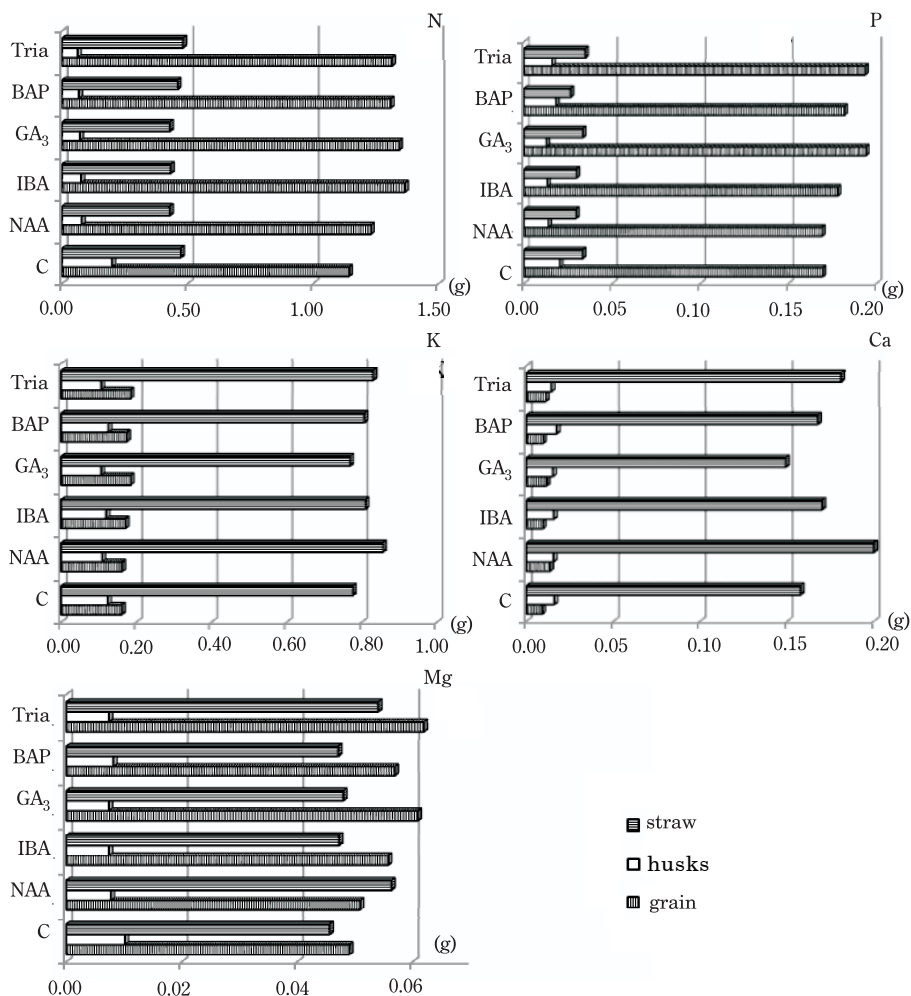


Fig. 1. Accumulation of macronutrients in organs of spring triticale

accumulation of nitrogen ranged from 62 to 68%. The growth regulators, especially gibberellin, depressed the contribution of grain in the accumulation of this nutrient. A study conducted by KLIKOCKA and KOMISARCZUK (2000) proved that as the nitrogen fertilisation level rises, so does the concentration and accumulation of magnesium in spring triticale grain. In contrast, simplifications in soil tillage lead to depressed concentrations of Mg in grain.

Under the effect of the growth regulators (except GA<sub>3</sub>), the K:(Ca+Mg) ratio in grain and straw of triticale became narrower but the ratio of these ions in husks broadened (Figure 4). Under the influence of the tested phytohormones, the Ca:P mol ratios in vegetative organs of triticale became broad-

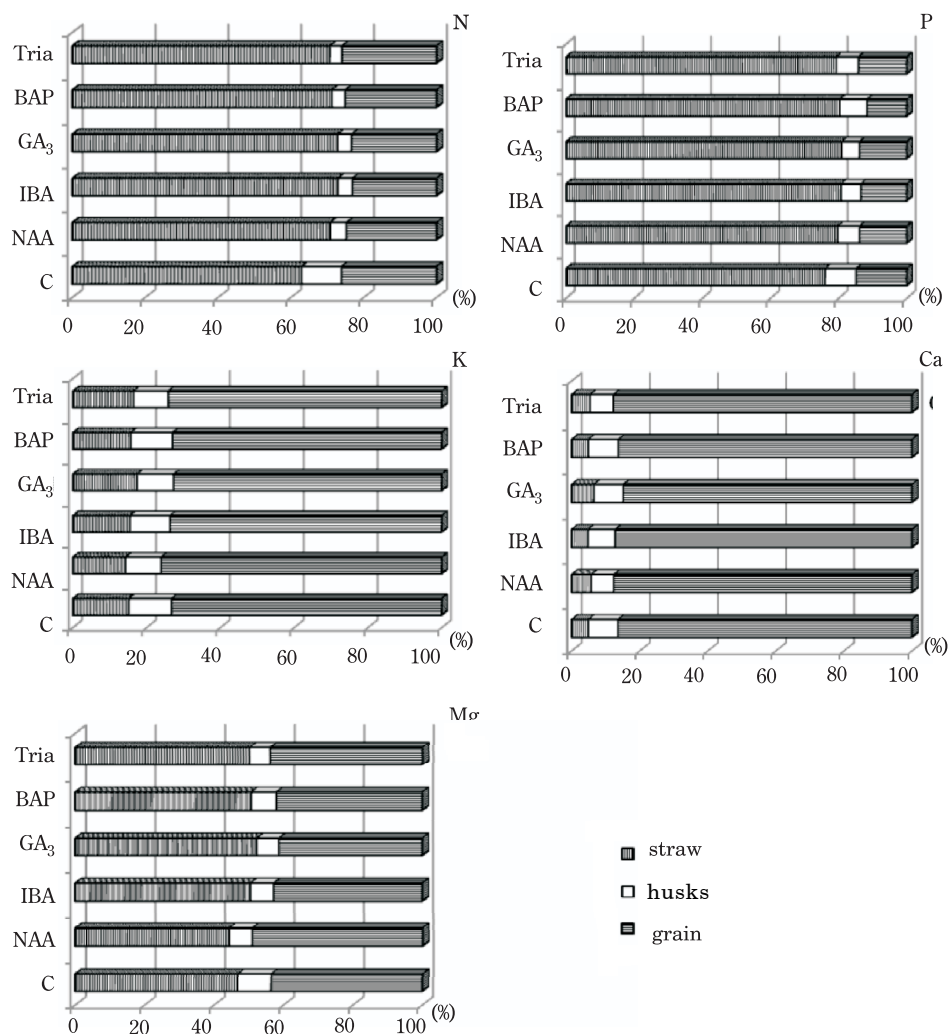


Fig. 2. Distribution of nutrients in spring triticale

er. In addition, IBA caused analogous changes both in vegetative parts and in grain (Figure 5). This ratio became most distinctly broader in husks under the effect of NAA and GA<sub>3</sub>, and in straw – mostly under the effect of IBA. PILEJCZYK et al. (2004) found out that the value of mol ratios of K:Ca and K:Mg in grain and straw of triticale became narrower as the rate of the applied nitrogen rose; under the same conditions, the Na:K ratio became broader. It was also demonstrated that as the concentration of phosphorus in grain rose, so did the concentration of potassium and calcium. In our previous studies on spring wheat, the K:(Mg+Ca) proportions in grain and in

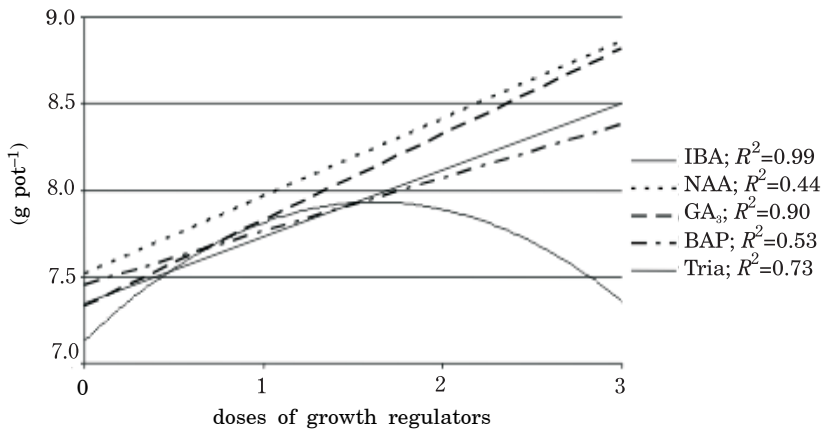


Fig. 3. Protein yield in triticale grain

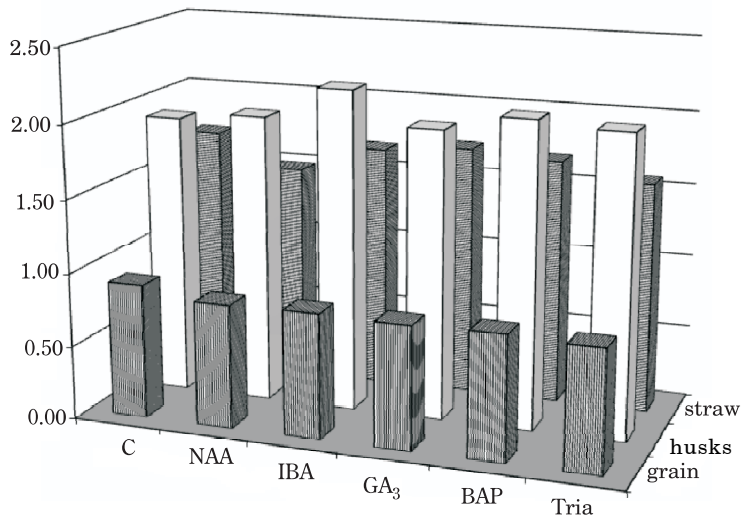


Fig. 4. K:(Ca+Mg) ionic ratios

vegetative organs were narrower under the influence of gibberellin but broader when kinetin or auxin (NAA) had been applied. Auxin broadened this ratio in grain and leaves but narrowed it in husks and in the stem (WIERZBOWSKA, BOWSZYS 2008a,b, WIERZBOWSKA 2006b).

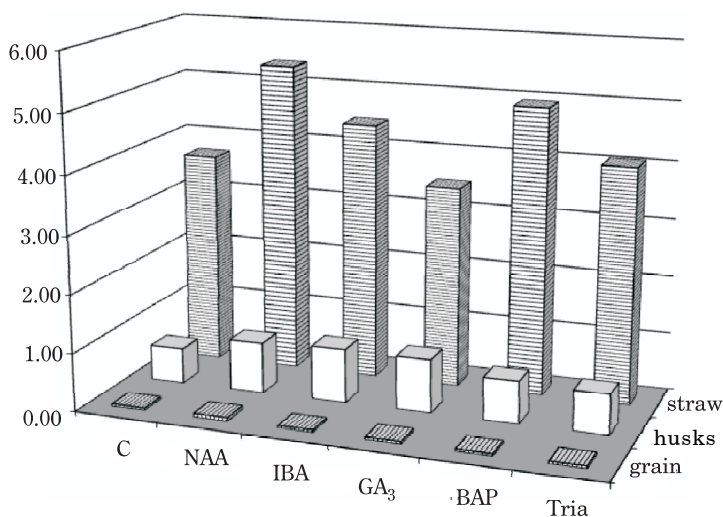


Fig. 5. Ca:P mol ratios

## CONCLUSIONS

1. The growth regulators, especially NAA, significantly affected the concentration of nitrogen in grain and depressed its amount in husks. Triacantanol caused an increase in the concentration of phosphorus, potassium and calcium in grain. The phytohormones increased the concentration of potassium and depressed that of phosphorus in triticale straw.

2. The highest protein yield in triticale grain was obtained when seed material had been treated with NAA before sowing, and generally increased proportionately to the concentration of the growth regulators.

3. The growth regulators, especially NAA and GA<sub>3</sub>, raised the uptake and accumulation of nitrogen and phosphorus in grain; IBA and triacantanol increased the uptake of potassium and its accumulation on straw.

4. The phytohormones increased the contribution of grain to the accumulation of phosphorus and potassium.

5. The growth regulators caused the narrowing of the K:(Ca + Mg) ionic ratios in grain and straw, but broadened it in husks. After the application of the phytohormones, the Ca:P ratio in vegetative organs, and under the influence of IBA also in grain, became broader.

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## **YIELDS AND QUALITY OF GREEN FORAGE FROM RED CLOVER DI- AND TETRAPLOID FORMS**

**Krystyna Żuk-Gołaszewska<sup>1</sup>, Cezary Purwin<sup>2</sup>,  
Barbara Pysera<sup>2</sup>, Jadwiga Wierzbowska<sup>3</sup>,  
Janusz Gołaszewski<sup>4</sup>**

<sup>1</sup> Chair of Agrotechnology and Plant Production Management

<sup>2</sup> Chair of Animal Nutrition and Feed Science

<sup>3</sup> Chair of Agricultural Chemistry and Environmental Protection

<sup>4</sup> Chair of Plant Breeding and Seed Science

University of Warmia and Mazury in Olsztyn

### **Abstract**

The purpose of the study has been to determine the yield, chemical composition and nutritive value of green forage from di- and tetraploid forms of red clover grown in different seed density regimes. The yield of green forage was analysed in the first year of full use, in 2003 and 2004, having cut the plants in the early inflorescence phase (1<sup>st</sup> cut). In green forage samples, the following were determined: basic chemical composition, concentration of water soluble carbohydrates (WSC), fibre fractions (NDF, ADF and ADL) and macronutrients (P, K, Mg, Ca, Na). The nutritive value of green forage was expressed according to the INRA 1988 system.

The yield of green matter from the tetraploid cultivars Bona and Jubilatka was higher than from the diploid cultivars Krynica and Parada. The cultivar Bona distinguished itself by its highest average yield of green matter, dry matter, total protein and net energy. The dry matter content was the most variable green forage quality characteristic. In both years, green forage from the diploid cultivars contained more dry matter than the one made from the tetraploid varieties. At the same time, seed density did not have any significant influence on the differentiation of green forage yields.

Green forage from the two diploid cultivars was characterised by a similar energy and protein value and a better fill unit value (better potential intake by ruminants). The concentrations of phosphorus, potassium and calcium in green forage from the diploid forms were slightly higher compared to the tetraploid forms, unlike the level of magnesium, which was slightly lower. Irrespectively of the polidy level, the concentration of macronu-

trients, except phosphorus, was higher in the first year of the study. Considering the nutritional requirements of animals, green forage from the analysed red clover cultivars was characterised by an inadequate level of magnesium, deficient amount of sodium and an improper Ca:P ratio. The results suggest that diploid forms of red clover can potentially generate a higher nutritive value than tetraploid ones.

Key words: red clover, forage quality.

## PLONY I JAKOŚĆ ZIELONKI DI- I TETRAPLOIDALNYCH FORM KONICZYNY CZERWONEJ

### Abstrakt

Celem badań było określenie plonowania oraz składu chemicznego i wartości pokarmowej zielonki di- i tetraploidalnych odmian koniczyny czerwonej uprawianej z zastosowaniem różnej gęstości siewu nasion. Plon zielonki analizowano w pierwszym roku pełnego użytkowania w latach 2003 i 2004, po skoszeniu roślin w fazie początku kwitnienia (I pokos). W próbach zielonki oznaczono podstawowy skład chemiczny, zawartość cukrów rozpuszczalnych w wodzie (WSC), frakcje włókna (NDF, ADF i ADL) i makroelementy (P, K, Mg, Ca, Na). Wartość pokarmową zielonki wyrażono według systemu INRA 1988.

Plon zielonej masy odmian tetraploidalnych Bona i Jubilatka był wyższy niż odmian diploidalnych Krynja i Parada. Wyróżniała się odmian Bona, dla której stwierdzono najwyższy średni plon zielonej masy, suchej masy, białka ogólnego i energii netto. Cechą jakościową zielonki o największej zmienności była zawartość suchej masy. W obu latach użytkowania zielonka odmian diploidalnych miała wyższą zawartość suchej masy niż zielonka odmian tetraploidalnych. Jednocześnie gęstość siewu nasion nie miała istotnego wpływu na zróżnicowanie plonów zielonki.

W przypadku zielonki odmian diploidalnych stwierdzono zbliżoną wartość energetyczną i białkową oraz korzystniejszą wartość wypełnieniową (większa możliwością pobrania przez przeżuwacze). Poziom fosforu, potasu i wapnia w zielonkach odmian diploidalnych był nieznacznie wyższy, a magnezu niższy w porównaniu z odmianami tetraploidalnymi. Niezależnie od poziomu ploidalności odmian koncentracja makroelementów, z wyjątkiem fosforu, była wyższa w pierwszym roku badań. Uwzględniając wymagania pokarmowe zwierząt, zielonka badanych odmian koniczyny miała odpowiedni poziom magnezu, niedoborową ilość sodu oraz niewłaściwy stosunek Ca:P. Uzyskane wyniki wskazują, że odmiany diploidalne koniczyny czerwonej mają możliwości osiągania wyższej wartości pokarmowej niż odmiany tetraploidalne.

Słowa kluczowe: koniczyna czerwona, jakość zielonki.

## INTRODUCTION

Next to grasses, red clover is a leading forage crop grown in the moderate climate zone (LEE et al. 2004). The advantages of cultivating red clover are such as its high fodder value, mixed cultivation with grasses, barley, oat or wheat, its positive role in crop rotation systems by binding nitrogen, restricting the development of cereal diseases ("disease break") and providing natural protection from monocot weeds. Moreover, red clover has bene-



ficial influence on the natural environment as it prevents erosion, acts as a phytomelioration plant and enhances the landscape's aesthetic value (BAWOLSKI, ŚCIBOR 1982, DUER 1999, HOGH-JANSEN, SCHJOERRING 1997, KOSTUCH 1998, SPANCER, TODD 2003, STEINER et al. 1995).

The main storage components accumulated by red clover are total protein, which can make up as much as 21% of the dry matter, and fibre (between 18-26% of d.m.). Both di- and tetraploid forms of red clover are grown commercially. The two forms have different morphological traits and agricultural characteristics. In a study conducted by TOMASZEWSKI Junior (1989), the yield of grain from diploid forms was 30-60% higher than from tetraploid ones. Likewise, JARANOWSKI and BRODA (1986) as well as BRU•DZIAK et al. (1989) concluded that diploid cultivars are more productive. However, tetraploid cultivars can possess superior qualitative composition. ŻUK-GOŁASZEWSKA et al. (1999), who analysed yields of di- and tetraploid cultivars of red clover, demonstrated that red clover tetraploid forms were superior in production of total protein yields. At present, the quality-orientated breeding of clover focuses on improving the content and quality of proteins (the amino acid composition, sensitivity to proteolysis, PPO activity), delaying the lignification process as plants mature or increasing the share of carotens and decreasing the concentration of anti-nutritional compounds, e.g. isoflavones (LEE et al. 2004).

The objective of this study has been to determine the level of yields and composition of green forage, including the feed value, obtained from red clover di- and tetraploid cultivars.

## MATERIAL AND METHODS

The research was based on plant material such as green forage made from di- and tetraploid cultivars of red clover, grown in strict field experiments conducted in 2003 and 2004 on experimental fields at the Experimental Station in Bałcyny. Two-factor experiments with different forms of red clover (diploid cultivars Krynica and Parada, tetraploid cultivars Bona and Jubilatka) sown in different seed density regimes (4, 8, 12, 16 kg ha<sup>-1</sup>) were set up in a split-plot design. The yield of green forage was determined in the first year of full use, during the early inflorescence phase (1<sup>st</sup> cut). Samples of green forage weighing 0.5 kg were taken from ten places on each plot and then an average analytical sample was extracted. Next, the samples were dried up for 24 h at 60°C in a forced air-flow dryer (manufactured by BINDER) and comminuted in a high-speed grinding mill (made by FOSS). The content of dry matter, crude ash, total nitrogen, ether extract, crude fibre was determined according to the AOAC procedures (2005). The concentration of water soluble carbohydrates (WSC) was determined with the an-

throne method (THOMAS 1977) whereas NDF, ADF and ADL fibre fractions were assayed using GOERING and VAN SOEST'S method (1970) in an ANKOM 220 apparatus. The content of true protein was determined using trichloroacetic acid (LICITRA et al. 1996). For determination of the content of ash components, comminuted material was mineralised in concentrated sulphuric acid with addition of chlorine dioxide as an oxidising agent. The concentration of phosphorus was determined by colorimetry using the vanadium-molybdenum method. Potassium, calcium and sodium were determined with the photoflame method (ESA) and the concentration of magnesium was assayed using the absorption atomic spectrophotometry (AAS).

The feed value of green forage was derived from the chemical composition, according to the INRA's fodder evaluation system (WINWAR programme, KOWALSKI, KAŃSKI 1993) and expressed in the following units: UFL (unit of energy for milk production), PDIE (protein digested in the small intestine depending on rumen fermented organic matter), PDIN (protein digested in the small intestine depending on rumen degraded protein) and FUC (fulfilment unit for cows).

The results of the green forage yield volumes and chemical analyses underwent statistical analysis using the correlation and regression methods as well as analysis of variance. For comparison of the treatment means, T Tukey's test was applied and HSDs (honest significant differences) were established. The evaluation of groups of means was performed using orthogonal contrasts (ELAND 1964). Statistical hypotheses were verified at the level of significance  $p < 0.05$ .

## RESULTS AND DISCUSSION

The first year of the experiment was warm and dry. The average of the temperatures during the plant growing season was higher than the mean long-term temperature and the precipitation in the spring months (MARCH, April) was lower or close to the multi-year average. The weather conditions in 2004 were more favourable to the vegetative development of red clover. After a wet winter, profuse rainfall appeared in April, becoming more and more intensive in all the consecutive months of the growing season. In May and June, the rainfall was 87.1 and 90.6 mm, respectively, thus being higher than the long-term average (56.7 and 68.3 mm) ŻUK-GOŁASZEWSKA et al. 2006a.

The yield of red clover green matter was significantly different between the cultivars in each year; likewise, the year x cultivar interaction was significant (Table 1). The weather conditions, being more favourable to the vegetative growth of plants in 2004, raised the green forage yield by an average  $12.3 \text{ t ha}^{-1}$  compared to 2003.

Table 1

Green forage yield of di- and tetraploid red clover cultivars grown in different sowing density regimes in the two years of the experiment ( $\text{t h}^{-1}$ )

Cultivars (C)	Sowing density (S) ( $\text{kg ha}^{-1}$ )	Year (Y)		Mean
		2003	2004	OxG
Krynia	4	38.5	52.3	45.4
	8	42.7	51.4	47.0
	12	40.1	49.8	44.9
	16	43.5	51.0	47.2
Parada	4	43.6	53.7	48.6
	8	44.1	45.6	44.8
	12	44.6	44.8	44.7
	16	44.3	47.1	45.7
Bona	4	48.7	61.2	54.9
	8	45.9	63.2	54.5
	12	49.2	61.0	55.1
	16	46.7	63.9	55.3
Jubilatka	4	42.3	55.4	48.8
	8	43.4	62.9	53.2
	12	32.6	61.8	47.2
	16	42.6	64.6	53.6
		YxC		O
Krynia		41.2	51.1	46.1
Parada		44.1	47.8	46.0
Bona		47.6	62.3	55.0
Jubilatka		40.2	61.2	50.7
		YxS		G
	4	43.2	55.6	49.4
	8	44.0	55.8	49.9
	12	41.6	54.4	48.0
	16	44.3	56.7	50.5
		L		
		43.3	55.6	

HSD<sub>0.05</sub> for: Years (Y) – 5.79; Cultivars (C) – 3.77, YxC – 6.42;  
Sowing density(S) – n.s.; LxG – n.s.; OxG – n.s.; LxOxG – n.s.

In general, the tetraploid cultivars Bona and Jubilatka yielded higher than the diploid cultivars Krynja and Parada. The highest average yield of green matter, found for cv. Bona, was significantly higher than generated by all the other varieties. However, the response in yield of green matter produced by the analysed red clover cultivars depended on the weather conditions in the two years of the tests (Figure 1). In 2003, the cultivar Bona produced a higher yield than all the other cultivars but in 2004 yielded like the tetraploid cultivar Jubilatka. Stable albeit the lowest yields were produced by the diploid cultivar Parada. In the present study, differences in the

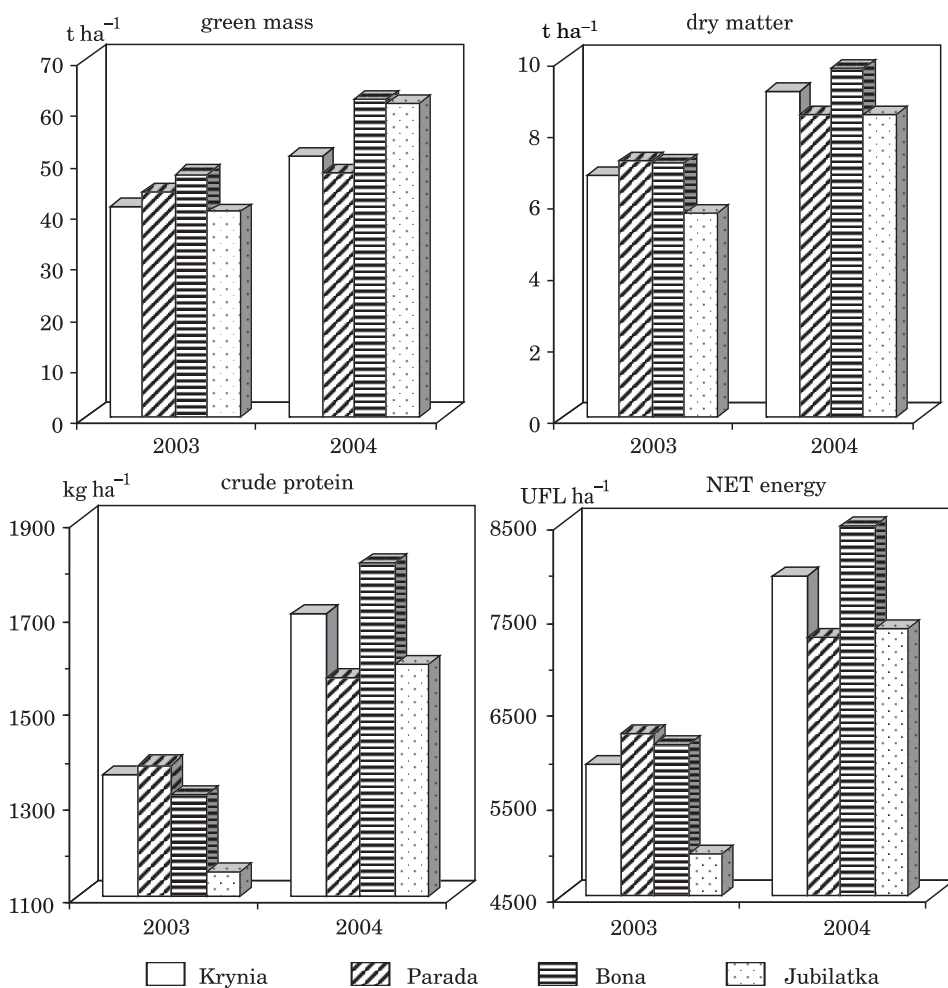


Fig. 1. Green mass, dry matter, crude protein and net energy in di- and tetraploid red clover cultivars

yields of green matter found between the years may have been caused by the difference in the rainfall during the growing season of red clover. Analogously to the study reported by LETO et al. (2004), differences in yields produced by di- and tetraploid cultivars did not exceed 10%. In turn, GRAMAN (1988) attained a 10-60% higher green matter yield from tetraploid cultivars than from diploid ones in years characterised by very high precipitation. The correlation between green and dry matter in diploid cultivars in both years of the trials was significant, which is confirmed by results of some previous studies conducted by MUNTEAN and SAVATTI (2003).

The net energy yields were correlated with yields of dry matter, which was associated with approximately the same energy value of dry matter in all the cultivars. The smallest differences in protein yields may have been caused by a higher content of protein in the cultivars producing lower yields (Figure 1).

The chemical composition of the analysed green forage produced from red clover was differentiated, although statistically proven differences appeared only for the content of dry matter, neutral detergent fibre (NDF) and acid detergent lignin (ADL) – Tables 2 and 3.

Among the analysed traits, dry matter was most variable. The concentration of this yield component was found to differ between the years of harvest, ploidy status and between the two diploid cultivars Krynja and Parada. In both years, more dry matter was found in green forage from the diploid cultivars than from the tetraploid ones. At the same time, the tetraploid cultivars responded differently to the weather conditions prevailing during the experiment. The cultivar Bona, similarly to the diploid cultivars, generated less dry matter in green forage in 2003, whereas cv. Jubilatka, which generally contained low amounts of dry matter, produced more of this component in 2004 (significant comparison *years x tetraploid cultivars*).

Significant differences in the content of structural sugar fractions appeared in the groups of di- and tetraploid cultivars. In both years of the trials, the NDF fraction in the green forage from the diploid cultivar Parada was higher than in the green forage from cv. Krynja; by analogy, the tetraploid cv. Jubilatka contained more NDF than cv. Bona. The higher level of NDF in the two cultivars mentioned above may be a result of their higher content of polysaccharides (hemicelluloses), which ensure high digestibility of fodder by ruminants (PURWIN 2007). The ADL fraction in green forage, which indicates the lignification degree, was different in the two tetraploid cultivars. Likewise, the diploid cultivars were characterised by a different response in terms of the content of the ADL fraction in the years of the experiment.

The differences in the content of total protein and true protein were relatively small (within the error limit), but noteworthy was the differentiation in the concentration of protein in the diploid cultivars depending on the course of the weather conditions during the tests.

Table 2

Means for the chemical composition of di- and tetraploid red clover cultivars in the years of the experiment (g kg<sup>-1</sup> d.m.)

Cultivars	Year	Dry matter	Ash	Organic matter	Crude protein	True protein	True protein/ /crude protein	Ether extract	WSC	NDF	ADF	ADL
Krynja	2003	165	111	888.6	200	136	70.0	23.8	94.1	407	337	45.6
	2004	178	111	889.0	187	139	74.7	21.9	103.7	454	321	41.0
	mean	172	111	888.8	193	138	72.3	22.8	98.9	430	329	43.3
Parada	2003	163	111	889.2	193	146	76.0	18.9	76.6	445	333	31.0
	2004	177	113	886.8	185	139	75.2	21.0	99.8	500	336	45.5
	mean	170	112	888.9	189	143	75.6	19.9	88.2	472	334	38.2
Bona	2003	150	114	885.7	185	119	64.6	21.1	78.1	392	325	31.7
	2004	156	112	888.0	186	140	74.9	21.8	94.9	454	325	38.4
	mean	153	113	886.8	185	130	69.7	21.4	86.5	423	325	35.0
Jubilatka	2003	142	111	888.6	202	144	71.4	21.2	108.8	489	299	42.8
	2004	138	111	888.8	188	141	75.2	20.3	99.0	499	332	52.9
	mean	140	111	888.7	195	143	73.3	20.7	103.9	494	316	47.8
SEM		5.71	5.62	6.26	9.32	12.52	4.97	4.21	16.65	31.74	14.61	5.38

Table 3  
Estimates of contrasts between treatments in analyses of the chemical composition of green mass

Contrasts	Dry matter	Ash	Organic matter	Crude protein	True protein	True protein/crude protein	Ether extract	WSC	NDF	ADF	ADL
Between years	-6.2*	0.7	-0.5	-3.8	-3.7	-0.3	12.6	-2.5	3.6	4.4	-1.3
Diploid cvs vs. tetraploid cvs	-49.7*	2.2	-2.0	-0.5	-2.9	-1.5	11.0	-2.2	10.7	-13.7	-2.0
Diploid cvs: Krynja vs Parada	1.0	-2.4	2.4	-1.5	-4.4	-1.6	1.9	5.4	-39.8*	-7.4	3.1
Tetraploid cvs: Jubilatka vs Bona	-11.1*	-0.8	1.1	4.4	2.8	-0.2	15.4	1.1	44.7*	5.1	9.8*
Years vs ploidy	21.1*	6.4	-5.9	-5.1	-10.6	-3.4	9.8	15.0	24.0	-27.2	-13.7
Years vs diploid cvs	-1.3	-0.4	0.4	-6.0	-8.5	-2.2	17.4	3.0	13.1	14.8	15.0*
Years vs tetraploid cvs	13.8*	-0.1	0.6	4.7	2.0	-1.0	-11.0	-5.9	-0.4	-3.6	-9.7

\* significant at  $p < 0.05$

PURWIN (2007) reports that the concentration of water soluble carbohydrates (WSC) is a factor that determines the suitability of green forage for ensiling and their energy value. It is assumed that tetraploid cultivars are better regarding the nutritive value compared to diploid forms. In general, they contain more protein and water soluble carbohydrates but less fibre (BIENIASZEWSKI FORDOŃSKI 1996). In the present study, the results are ambiguous. However, we noticed a certain tendency towards a higher concentration of water soluble carbohydrates in the second year of the tests (2004), which in general was more favourable to the growth of the plants (Table 3).

The feed value of green forage is verified by the energy and protein value, relations between the concentration of crude fibre and water soluble carbohydrates as well as the fulfilment value. The results presented in Table 4 demonstrate that the tested green forage has a similar energy value (UFL) irrespectively of the cultivar and year. The protein value of green forage was more evidently differentiated between the years than between the cultivars, assuming lower values in 2004. A higher value of PDIN compared to PDIE in green forage from all the analysed cultivars is typical of green forage made from papilionaceous plants, which contain relatively little crude fibre and a high level of soluble sugars.

Table 4

Nutritive value of di- and tetraploid red clover cultivars according to INRA (1988)

Cultivars	Year	UFL kg <sup>-1</sup> d.m.	PDIN (g kg <sup>-1</sup> d.m.)	PDIE (g kg <sup>-1</sup> d.m.)	FUC kg <sup>-1</sup> d.m.
Krynica	2003	0.87	125.0	98.67	0.85
	2004	0.87	117.2	96.42	0.78
	mean	0.87	121.1	97.54	0.82
Parada	2003	0.87	120.9	97.88	0.86
	2004	0.86	115.3	95.17	0.78
	mean	0.86	118.1	96.52	0.82
Bona	2003	0.86	116.0	95.86	0.93
	2004	0.87	116.9	96.69	0.90
	mean	0.86	116.5	96.28	0.92
Jubilatka	2003	0.87	127.1	99.58	0.99
	2004	0.87	118.2	96.88	1.01
	mean	0.87	122.6	98.23	1.00

Noteworthy are big differences between the di- and tetraploid forms in the fulfilment value of the green forage, a parameter which characterises possible intake of bulk feeds by ruminants. A much higher value of fulfilment units (FUC) in green forage from tetraploid forms of red clover is



generally an undesirable parameter as it implies potentially lower capacity for the intake during direct feeding of animals (INRA 1988). A lower fulfilment value of green forage from diploid cultivars is associated with their lower content of dry matter. Similar energy and protein values alongside a lower fulfilment value of diploid cultivars indicate that it is possible to produce good quality fodder from diploid cultivars of red clover. Our results correspond to the conclusions drawn by DROBNA and JANČOVIČ (2006), who suggested that it should be possible to introduce diploid forms of red clover to commercial production owing to their nutritive value being higher than that of tetraploid cultivars.

The weather conditions during the growth of plants and the form of red clover differentiated the concentration of mineral components in green forage (Table 5).

Table 5

Content of macronutrients in di- and tetraploid red clover cultivars (g kg<sup>-1</sup> d.m.)

Cultivars	Year	P	K	Mg	Ca	Na
Krynia	2003	3.41	38.36	3.54	14.67	0.60
	2004	4.46	37.64	3.15	14.79	0.46
	mean	3.93	38.00	3.35	14.73	0.53
Parada	2003	3.64	43.04	3.37	15.15	0.43
	2004	3.82	39.60	3.14	15.08	0.41
	mean	3.73	41.32	3.26	15.11	0.42
Bona	2003	3.61	40.56	3.85	14.05	0.46
	2004	3.67	38.66	3.23	13.79	0.40
	mean	3.64	39.61	3.54	13.92	0.43
	2003	3.45	39.92	3.80	16.12	0.82
	2004	3.71	39.18	3.07	13.29	0.43
	mean	3.58	39.55	3.43	14.70	0.62

The concentration of phosphorus in green forage in the first year of the tests ranged between 3.41 and 3.64 g in 1 kg d.m., with more phosphorus found in the green forage from cv. Parada (2n) and Bona (4n). In the second year, the concentration of phosphorus was higher, and the effect exerted by the form of red clover became more evident. The biggest difference (over 30%) between the two years of the tests was found for the diploid cultivar Krynia, whereas the smallest one (1.7%) appeared for the tetraploid cv. Bona. In general, the diploid forms were characterised by a slightly higher concentration of phosphorus than the tetraploid ones. Contrary relations in the concentration of phosphorus in di- and tetraploid cultivars were determined by BIENIASZEWSKI and FORDOŃSKI (1996), who found out that tetraploid forms

contained more phosphorus and potassium than diploid ones. In our experiment, the content of potassium was shaped differently. In both years, cv. Parada had the highest concentration of potassium and, irrespectively of the cultivar, the concentration of potassium was higher in 2003. The concentrations of magnesium were shaped analogously, but the tetraploid cultivars contained more of this element. In these cultivars, in the second year of the tests, a large difference as found, namely 18.9% for cv. Bona and 23.5% for cv. Jubilatka. This, however, does not undermine the finding that the content of magnesium in all the types of green forage analysed, regardless the type of red clover or the year of the experiment, fully covered the nutritional demands of animals, especially dairy cows, which – according to the INRA – demand 1.5 to 2.0 g Mg kg<sup>-1</sup> d.m. of fodder. When analysing the concentration of calcium in the two consecutive years of the experiment, it was demonstrated that the tetraploid forms (cv. Bona and Jubilatka) were characterised by a higher level of this nutrient in 2003 (1.8 and 21.3% more, respectively) than in 2004. No effect of the year on the level of calcium in the diploid cultivars was discovered. Generally, the average concentration of calcium was the highest in green forage from cv. Parada (15.11 g kg<sup>-1</sup> d.m.) and the lowest one – in cv. Bona (13.9 g kg<sup>-1</sup> d.m.).

Considering the nutritional usability of forage, the concentration of calcium in red clover was twice as high as required by animals, which confirms the suggestion expressed by PLAZA et al. (1009) that cultivation of red clover in a pure sowing regime does not provide full quality fodder in terms of the content of mineral components. It is so because the quality of fodder is determined by mutual ratios between particular mineral elements (Figure 2). In the present study, the Ca:P ratio in both years of the tests was widened and ranged within 2.57-3.62:1, while the optimum ratio is 1.5-2.0:1 (the INRA). At the same time, it was demonstrated that cv. Jubilatka in the first year of the experiment and cv. Krynja in both years were characterised by the most favourable K:(Ca+Mg). As SABA et al. (2000) claim, high content

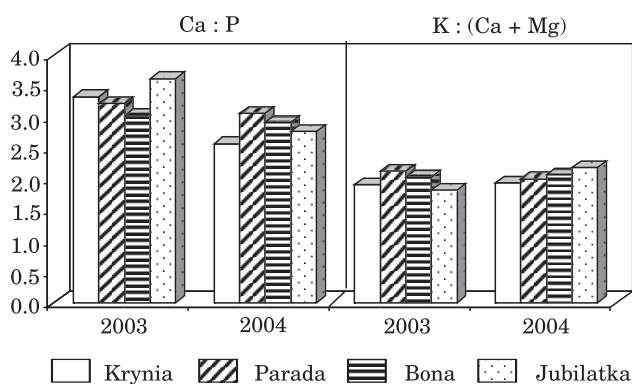


Fig. 2. The ratios of Ca:P and K:(Ca + Mg) in di- and tetraploid red clover cultivars

of potassium, due to the antagonistic metabolic effect of this element on magnesium, may constitute an additional factor which depresses the bioavailability of magnesium. Sodium deficit acts in a similar fashion. In the analysed types of green forage (Table 5), the level of sodium was deficient compared to the nutritional demand of animals, because the optimum values according to the INRA are 1.7-2.0 g kg<sup>-1</sup> d.m. Higher concentrations of this element were found in the green forage made in the first year of the experiment. The average content of sodium was the lowest in cv. Parada (2n) and Bona (4n).

## CONCLUSIONS

1. Yields of green forage from the first cut of red clover of the tetraploid cultivars are 15% higher than produced by the diploid varieties. The highest average yields of green matter, dry matter, total protein and net energy were obtained from cv. Bona red clover.

2. In the green forage from the diploid cultivars, the concentration of phosphorus, potassium and calcium was higher in the tetraploid cultivars, unlike the content of magnesium, which was lower. Irrespective of the ploidy level, the concentration of macronutrients, except phosphorus, was lower in the year which favoured the vegetative development of plants.

3. The above results suggest that diploid forms of red clover can potentially achieve a higher nutritive value than tetraploid cultivars.

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**REVIEW PAPER**

**MAGNESIUM AS A NUTRITIONAL  
TOOL OF NITROGEN EFFICIENT  
MANAGEMENT – PLANT PRODUCTION  
AND ENVIRONMENT**

**Witold Grzebisz, Katarzyna Przygocka-Cyna,  
Witold Szczepaniak, Jean Diatta, Jarosław Potarzycki**

**Chair of Agricultural Chemistry and Environmental Biogeochemistry  
Poznan University of Life Sciences**

**Abstract**

Nowadays, the main objectives of plant crop growers aim at two targets (i) increasing food production and (ii) simultaneously, reducing the environmental impact of increasing fertilizer nitrogen consumption. On a global scale, fertilizer nitrogen recovery ranges from 33 to 50%. The required efforts stimulating production but protecting the environment focus on increasing unit productivity of fertilizer N. Magnesium, owing to its biological functions in plants, should play a much more important role in modern agriculture controlling N economy of crop plants and, consequently, nitrogen dispersion in the environment. In Poland, arable soils are generally poor in total and available magnesium. This state can be considered as indicating the necessity of applying magnesium and then maintaining a well-fed plant nutritional status of growing crops. Crops well supplied with magnesium since the beginning of their growth, as seen from studies on the response of sugar beets and maize, are in a position to increase nitrogen unit productivity. Cereals respond to Mg supply when a dressing treatment takes place just before the onset of flowering. Another factor significantly affecting fertilizer nitrogen recovery in Poland is soil pH. Acid soils fertilized with Mg contain increased soil exchangeable Mg levels, which in turn depress the pressure of toxic aluminum on growing crops. Improvement of the plant Mg nutritional status enables plants to incorporate some of potentially residual N into biomass, increasing

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prof. dr hab. Witold Grzebisz, Chair of Agricultural Chemistry and Environmental Biogeochemistry, Poznan University of Life Sciences, Wojska Polskiego 71F street, 60-625, Poznań, e-mail: witegr@up.poznan.pl

biomass yield. It can therefore be concluded that magnesium, owing to its ameliorating function in arable soils, meets the main requirement of sustainable nitrogen management, both in agriculture and in the environment.

**Key words:** sustainable agriculture, nitrogen productivity, crop plants, magnesium, soil fertility, aluminum bio-toxicity, environment protection.

## **MAGNEZ JAKO CZYNNIK ŻYWIENIOWY EFEKTYWNEGO GOSPODAROWANIA AZOTEM – PRODUKCJA ROŚLINNA I ŚRODOWISKO**

### **Abstrakt**

Główne zadania stawiane współcześnie producentom roślin uprawnych skupiają się na dwóch celach: (i) zwiększeniu produkcji żywności, i (ii) jednocześnie zmniejszeniu ujemnego wpływu wzrastającego poziomu nawożenia azotem na środowisko. W skali globalnej wykorzystanie azotu szacuje się od 33 do 50%. Wymagane działania proprodukcyjne i proekologiczne skupiają się na zwiększeniu jednostkowej produkcji azotu stosowanego w nawozach. W nowoczesnym rolnictwie magnez, ze względu na funkcje biologiczne w roślinie, powinien odgrywać dużo większą rolę w kontroli gospodarki azotowej rośliny, a tym samym rozproszenia azotu w środowisku. W Polsce gleby uprawne są ogólnie ubogie w całkowity i przyswajalny magnez, co stwarza konieczność stosowania nawozów magnezowych w sposób zabezpieczający odpowiedni poziom odżywienia rośliny. Rośliny dobrze zaopatrzone w magnez od początkowych faz rozwoju, jak wynika z reakcji buraków lub kukurydzy, są w stanie istotnie zwiększyć jednostkową produktywność azotu. Zboża reagują na nawożenie magnezem wówczas, gdy zabieg odbywa się tuż przed kwitnieniem. Drugim istotnym czynnikiem ograniczającym wykorzystanie azotu przez rośliny uprawiane w Polsce jest odczyn gleb. Traktowanie gleb kwaśnych nawozami magnezowymi, w następstwie wzrostu koncentracji wymiennego magnezu w glebie, istotnie zmniejsza presję toksycznego glinu na rosnącą roślinę. Poprawa stanu odżywienia roślin magnezem umożliwia włączenie części potencjalnie niewykorzystanego N (azot rezydualny) w biomasę, co zwiększa jej plon użytkowy. Można zatem stwierdzić, że stosowanie magnezu, mającego wpływ na poprawę funkcjonowania gleb uprawnych, wypełnia tym samym zadania związane z realizacją zrównoważonej gospodarki azotowej w rolnictwie i środowisku.

**Słowa kluczowe:** zrównoważone rolnictwo, produktywność azotu, magnez, zasobność gleb, fitotoksyczność glinu, ochrona środowiska.

## **SUSTAINABLE AGRICULTURE**

Despite the enormously high rate of technological progress, food production remains the main concern the world is currently facing. Food production should not only eradicate famine but also eliminate malnutrition. In about 40 years, from 1960 to 1999, the world population doubled from 3 to 6 billion people. At present, however, the number of food consumers suffering insufficient supply of high food quality and experiencing malnutrition-related health problems, ranges from 0.5 to 0.8 billion (ALEXANDRATOS, 1999; DYSON 1999). Short-term demographic prognoses assume the world population will reach 7.5 to 7.8 billion people in 2025. The food demand growth results in

two facts: (i) a higher number of people and (ii) shifting consumption patterns boosted by higher incomes of economically growing up societies, especially in Central-Eastern Europe, South America and Asia. Food supply data published at the beginning of the 21<sup>st</sup> century assumed a 33 and 57% increase (1997 baseline values) in demand for cereals and meat, respectively (ROSEGRANT et al., 2001). In the case of total cereal production, this scenario was achieved in 2008. Within the 10-year period, yields of total cereals increased from 2.994 in 1998 to 3.539 t ha<sup>-1</sup> in 2008, i.e., by 18.2%, but the harvested area rose just by 1.7% (from 700 to 712 billions ha) (FAOSTAT 2010).

The above statistical data clearly indicate that the global food supply strategy can be successfully realized relying on the yield increase and not on the harvested area increase. However, even this strategy of food supply growth is subjected to criticism due to two aspects. The present pattern of intensive agriculture, immanently resulting from the *Green Revolution* concept (new dwarf varieties, irrigation, pesticides and fertilizers), has reached its *saturation level*. This strategy, dominating in the last 60 years, is the main reason for deep environmental disturbance, caused by uncontrollable inputs of nitrogen and phosphorus to ecosystems near farmlands (SOCOLOW, 1999, TILMAN 1999, TOWNSEN et al. 2003).

The negative environmental consequences of intensification of agriculture observed in the last decades of the 20<sup>th</sup> century began in western societies (North America, West Europe). This has been pointed out by different ecological organizations, which exert strong pressure on governments. In the 1980s, the first UN global studies on the environment status started. The final report, known as *Our Common Future*, was presented in 1987 during the General Assembly of the UN ([www.un-documents.net/a42r187.htm](http://www.un-documents.net/a42r187.htm)). This document soon became a basis, or even a milestone of the economic and political doctrine nowadays known as sustainable development. In agriculture, this doctrine is referred to as sustainable agriculture. In the European Union, the most important document, significantly affecting the current agricultural development, is the Nitrate Directive (91/676/EEC), which aims at protecting waters against excess of nitrates originating from farming practices.

The present paper deals with magnesium as a factor increasing the efficiency of fertilizer nitrogen in crop production technology. The main hypothesis of the paper assumes that magnesium is part of sustainable development of plant production, considered as a significant component of the food production branch.

## MECHANISMS OF NITROGEN UPTAKE BY CROP PLANTS

Harvested yields are generally a function of the quantity and dynamics of nitrogen accumulation by the growing crop during its vegetation. The

quantitative requirements of currently grown crops for fertilizer nitrogen are a result of two interacting factors:

1. Crop requirements, including the most important production factors:
  - critical growth periods with respect to development of yield forming components,
  - dynamics of dry matter accumulation,
  - expected yield.

2. Soil potential to supply nitrogen in rates sufficiently high to cover crop plant requirements at critical stages of growth and yield formation.

The first aspect helps to quantify the amount of fertilizer nitrogen required to cover crop plant needs over the growth season and the second one determines the soil capacity to cover these requirements from soil resources. Both create a diagnostic basis for assessing quantitative needs of currently grown crops for fertilizer nitrogen. Plants take up nitrogen in two main inorganic forms, as nitrates ( $\text{NO}_3^-$ ) and ammonium ( $\text{NH}_4^+$ ). The main metabolic core of nitrogen uptake by plant roots is through the transportation of ions from the soil solution through the plasma membrane into the cytoplasm. However, the mechanism of the uptake of each nitrogen form is ion specific. The first difference is the diffusion rate, a prerequisite of the absorption rate, which is *ca* 100-times higher for nitrate ( $2.7 \cdot 10^{-6} \text{ cm}^2 \text{ s}^{-1}$ ) as compared to ammonium ions ( $6.1 \cdot 10^{-8} \text{ cm}^2 \text{ s}^{-1}$ ) (RAYNAUD, LEADLEY 2004). Because of such a big difference, much more nitrate than ammonium ions reach the root surface. Consequently, a plant exploiting the nitrate pool much faster creates a much larger depletion zone in the rhizosphere. With respect to ammonium, four main processes govern ion concentration in the soil solution: (i) plant N demand, (ii) N immobilization/mineralization, (iii) ammonium nitrification rate and (iv)  $\text{NH}_4^+$  ion adsorption/desorption by soil particles. This form of nitrogen effectively affects the crop growth rate provided that it is present in small quantities. However, excess of ammonium disturbs significantly the plant growth due to the impairment of basic biochemical and physiological processes such as ATP synthesis, and the uptake of  $\text{K}^+$ ,  $\text{Mg}^{2+}$  and  $\text{Ca}^{2+}$  ions (BRITTO, KRONZUCKER et al. 2002, KUBIK-DOBOSZ 1998).

Crop plant nutrition with nitrate nitrogen depends on its concentration in the soil solution, that is the higher the concentration the higher its uptake. Depletion of nitrate from the soil solution is a soil induced signal for a plant to change the pattern of partitioning assimilates in order to increase the roots system size. The response of aerial organs of a plant to insufficient supply of nitrogen is signaled by the appearance of classical symptoms of nitrogen deficiency. This phenomenon is important for a farmer as a diagnostics indicator for an extra plant canopy N requirement, in practice covered by fertilizer application. However, the uptake of negatively charged nitrate ions in comparison to ammonium ions require a double energy expense by a growing plant (CANNEL, THORNLEY 2000). Transport of nitrate ions through the plasma membrane takes place in the protonated form:  $2\text{H}^+:\text{NO}_3^-$ ,



i.e., a nitrate ion requires preceding protonation, an energy-dependent mechanism. This physiological process is directly related to the activity of  $H^+$ -ATP-ase, which in turn depends on the plant nutritional status in phosphorus and magnesium. Hypothetically, every cation may participate as a counter-ion for nitrate, but based on the plant nutrient content, the crucial role is played by potassium ( $K^+$ ), which is accumulated by high yielding crops in the highest amounts (GRZEBISZ 2005). Magnesium ions ( $Mg^{2+}$ ) do not compete with potassium ones. The latter as well as excessive amounts of protons are antagonistic to the uptake of  $Mg^{2+}$  ions (OCHAŁ and MYSZKA, 1984). However, any rise in the  $Mg^{2+}$  concentration in the growth solution does not limit the uptake of potassium ions. In spite of these weaknesses, magnesium is considered as a crucial nutrient for the uptake nitrogen, especially nitrates, by high yielding crop plants. The main reason, as mentioned earlier, is the hydrogen pump efficiency, which requires high  $H^+$ -ATP-ase activity, depending in turn on  $Mg^{2+}$  as an enzyme cofactor. Therefore, the vigorous growth of any plants, particularly crops, requires a good supply of at least three basic elements: phosphorus, magnesium and potassium (FERNANDES, ROSSIELLO 1995, IMSANDE, TOURAINE 1994).

## NITROGEN: PLANT PRODUCTION AND ENVIRONMENT

Total fertilizer nitrogen consumption increased many folds between 1960 and 2010, from *ca* 12 in 1960 to 90 mln t  $y^{-1}$  at present. This tremendous N use resulted in a significant increase of food production. For example, total wheat production rose from 200 to 700 mln t  $y^{-1}$  in this period and its mean yields went up almost three-folds, from 1.1 to 3.1 t  $ha^{-1}$  (FAOSTAT, available online, 28.08.10). However, the high rate of wheat yield increase is not directly related to the rate of fertilizer nitrogen consumption. This discrepancy is the main reason to formulate two important questions:

- what part of the applied fertilizer nitrogen does the crop plant use in fact?
- does the non-consumed part of the applied fertilizer nitrogen create any threat to the environment's functionality?

Actually, there is no easy and true answer to the first question. Theoretically, fertilizer nitrogen recovery may reach 100% and even higher. Scientific papers about N fertilizer recovery, presenting field experiment data, are mostly in the broad range from 33 to 75%. However, very high maximal values of N recovery appear seldom, mostly under conditions of extremely low N rates, which do not ensure plant production profitability. The data concerning field conditions show much lower N fertilizer recovery, ranging only from 33% to 50% (DOBERMANN, CASSMAN 2002). Hence, from 45 to 60 mln t of the annually applied fertilizer N is not transformed into plant biomass, but becomes dispersed in soil or in ecosystems neighboring agricultural land (EICKHOUT et al. 2006, GALLOWAY, COWLING 2002).

Nitrate ions move freely both in the soil solution and/or in a stream of transpiration water. Their mobility does not depend on soil pH in the range from 2 to 10. Under conditions of soil water saturation and lack of plant cover, nitrates undergo translocation in accordance with the water gravity movement. The second type of nitrate movement induced physiologically is known as mass flow, and is a process transporting N to roots of growing plants. Therefore, nitrates are mostly subjected to leaching during autumn or winter. In addition, nitrate ions under insufficient oxygen concentration or its low movement in the soil are subjected to reduction, yielding  $N_2$  or N oxides such NO and  $N_2O$ . Due to their biological and chemical activity, nitrates and N oxides are termed as active forms of nitrogen (GALLOWAY, COWLING 2002, STRONG, FILLERY 2002, TOWNSEND et al. 2003).

The negative effects of agriculture on the environment, presented above, caused by the low fertilizer nitrogen efficiency are the basic reason for making a significant correction to the current production systems. The main target of fertilization practices is to cover nitrogen requirements of crop plants in the course of their growth, with respect to amounts, forms and time of fertilizer N application. Fulfilling this production induced paradigm is a key to elaborate an efficient system of fertilizer nitrogen management.

Nitrogen applied into soil as N fertilizer ( $N_f$ ) can be divided into two main fractions (pools): first taken up and consumed by currently growing plants ( $N_{up}$ ) and second, named as residual nitrogen ( $N_r$ ), i.e., not taken up by plants. The latter one ( $N_r$ ) undergoes further biogeochemical changes. Based on the distribution of final products, two sub-groups of residual nitrogen may be distinguished:

- fixed ( $N_s$ ),
- active ( $N_a$ ),

$$N_r = N_s + N_a \quad (1)$$

The fixed nitrogen fraction ( $N_s$ ) represents a pool of applied  $N_f$  temporarily unavailable to currently growing crop plants. This part of nitrogen occurs in soil in its primary form or is physically fixed by clay particles and/or incorporated into microbial biomass (immobilized). Potential availability of this previously applied nitrogen depends on the intensity of biogeochemical processes taking place in soil from harvest of the previous crop to the sowing of the subsequent one. The second form of residual nitrogen, ( $N_a$ ), relates to the part of the fertilizer nitrogen which is lost due to nitrates leaching to underground waters and nitrate oxides and ammonium volatilization to atmosphere. These forms of nitrogen create a real threat to natural ecosystems and in turn to human health (GALLOWAY, COWLING 2002, TOWNSEND et al. 2003).

The classical formula of the apparent fertilizer nitrogen recovery ( $E_N$ ) by the growing crop plant is as follows:

$$E_N = (N_u/N_f) \cdot 100\% \quad (2)$$

The above formula presents only a general relationship, but its application requires special experimental methodology of gathering data. The main target of any farmer, adhering to the sustainable agriculture rules, is to diminish losses of nitrogen to the environment. Therefore, equation No 2 should be transformed into an index of nitrogen losses ( $L_N$ ) as presented below:

$$L_N = (1 - N_u/N_f) \cdot 100\% \quad (3)$$

In the light of the ways of fertilizer nitrogen transformation in the soil, as presented above, the key step for farmers to undertake is to work out an effective fertilizing system – a strategy of soil and fertilizer nitrogen management. The production objective of an efficient N application strategy is to cover crop plant nitrogen requirements in critical stages of yield formation by (i) maximizing its yielding potential and (ii) minimizing N losses to neighboring ecosystems. The components of the fertilization system are: (i) crop and its nitrogen requirement, (ii) crop and nutrient requirements others than nitrogen, (iii) soil and its potential to supply nitrogen and other nutrients, (iv) N dressing patterns (amounts, rates, timing), and other nutrients, supporting N efficiency.

## ROLES OF MAGNESIUM IN CROP PLANT NITROGEN MANAGEMENT

There are many well recognized biochemical and physiological functions of magnesium in plants. However, farmers have to pay special attention to those which are directly or indirectly related to crop plant N management. The most important functions of magnesium are as follows:

- nitrogen uptake,
- photosynthesis, including:
  - synthesis of chlorophyll,
  - phosphorylation,
  - CO<sub>2</sub> fixation and reduction,
- phloem load of assimilates,
- distribution of carbohydrates among plant parts,
- control of the amounts of reactive oxygen substances (ROS),
- synthesis of organic compounds – starch, proteins, crude fats (CAKMAK, KIRKBY 2008, SHAUL 2002).

The first function of magnesium in the plant body related to plant production is nitrogen uptake (see part 2 of the article). The capacity of the currently cultivated crops to fix CO<sub>2</sub> is generally related to their canopy (expressed as the Leaf Area Index (LAI) or Green Area Index (GAI)), active

in solar energy absorption over the growth season. The rate of CO<sub>2</sub> fixation also depends on nitrogen concentration in the aerial biomass (GASTAL, LE-MAIRE 2002).

Sugar beet is the most classical crop in which the importance of both indices is evident. This crop is able to express its maximum photosynthetic capacity provided that its LAI is equal 3.0 and the nitrogen concentration in leaves is not lower than 40 g kg<sup>-1</sup> dry matter. In Europe, nitrogen uptake rate by sugar beet seedlings is the most decisive factor affecting radiation use efficiency by the crop canopy and in turn controlling the rate of aerial biomass growth (ANDRIEU et al. 1997). Phosphorus, magnesium and potassium are of great importance but should be considered as nutrients supporting nitrogen uptake by plants and its further transformation into plant biomass (RUBIO et al. 2003). The same considerations hold true for other crops but maize presents a special case. This crop is extremely sensitive to nitrogen supply during two distinct stages of growth, i.e., the stage of 5(6)-leaves, when plants form the primary yield structure. Insufficient supply of nitrogen negatively affects the potential number of leaves and cobs. The second crucial stage appears from the middle to the end of tasseling. During this particular period insufficient supply of nitrogen decreases the rate of biomass accumulation by the stem, hence depressing the potential number of fertile ovules (GRZEBISZ et al. 2008, SUBEDI, MA 2005).

Biochemical and physiological functions of magnesium in higher plant photosynthetic activity are fairly well recognized (CAKMAK, KIRKBY 2008, SHAUL 2002). However, yield-forming functions of this nutrient are not easily defined, due to its indirect effect, i.e., it can be assessed only *via* nitrogen plant management. The basic features of magnesium, as presented earlier, comprise three consecutive steps. The primary one is the synthesis chlorophyll molecules, which depends on the activity of magnesium chelatase, responsible for the incorporation of Mg<sup>2+</sup> ion into a proto-porphyrin ring. Any other divalent ions such as Cu<sup>2+</sup>, Zn<sup>2+</sup> or Ni<sup>2+</sup> are not able to replace Mg<sup>2+</sup> in this specific function. In the first phase of photosynthesis, known as “the light step”, magnesium is the main nutrient limiting the gaining of metabolic energy through the synthesis of compounds such as ATP and NADPH. In the second phase of photosynthesis, termed as “the dark step”, the CO<sub>2</sub> molecule is fixed by the enzyme Ribulose 1,5-bisphosphate carboxylase/oxygenase (RuBisCO). This enzyme requires some kind of preoperational activation and is dependent on Mg<sup>2+</sup> supply. The key step of CO<sub>2</sub> fixation, i.e., its reduction, is strongly affected by the level of metabolic energy supply, as again related to ATP plant resources. It is well known that the enzyme RuBisCO is strongly related to both CO<sub>2</sub> and O<sub>2</sub>. The main enzyme preference does not only depend on the CO<sub>2</sub> concentration in the growth environment but also on the plant magnesium nutritional status. Insufficient plant magnesium content is the main reason for increasing the rate of primary carbon decomposition due to the process known as photorespiration (SHAUL 2002).

The second group of processes, directly resulting from the nutritional status of a given crop plant, but indirectly affecting its nitrogen economy, refers to the partitioning of assimilates among roots and aerial biomass. There are only few experiments, conducted mostly under controlled conditions (hydroponics), reporting this very important process. However, these reports allow us to explain the negative outcome of a limited supply of magnesium to growing plants. As a rule, amounts of assimilates transported to roots significantly decrease, evoking a spiral of plant degradation symptoms. First to appear is the root system size reduction, negatively affecting the water and nutrients uptake; next, the growth rate of aerial parts of plants is impaired (Table 1). The same type of plant behavior has been observed for potassium but not for phosphorus (CAKMAK, KIRKBY 2008, HERMANS et al. 2005).

Table 1

Effect of insufficient supply of magnesium to mulberry plant (*Morus alba* L.) on biomass distribution between shoots and roots and magnesium concentrations (KUMAR TEWARI et al. 2006)

Plant parts	Control		Low Mg supply	
	mean	± SD*	mean	± SD*
Dry matter yield, g plant <sup>-1</sup>				
Shoots (S)	78.6	4.9	14.5	0.27
Roots (R)	8.2	0.81	1.1	0.01
Shoots/roots ratio (S/R)	9.7	0.37	13.7	0.17
Magnesium concentration, mg g <sup>-1</sup> dry weight				
Young leaves	1.8	0.0	0.3	0.9
Old leaves	2.9	0.0	0.2	0.1

\*SD – standard deviation

The first, mild symptoms of magnesium deficiency are related to accumulation of extra starch and reductive sugars in leaves. The main physiological reason for this storage of compounds in leaf tissue is the low activity of energy compounds such as ATP, responsible for the loading of phloem assimilates and finally for their transport to other plant parts (CAKMAK, KIRKBY 2008, HERMANS et al. 2005). Visible symptoms of magnesium deficiency in an advanced stage, as shown in Photo 1 for a grape plant, such as green veins on the background of yellowing areas, are striking. Emergence of these symptoms on leaves is attributed to the low capability of green tissues to transform the excess of solar radiation into energy compounds. The direct cause of visible symptoms of Mg deficiency is, however, extra production of reactive oxygen substances (ROS) and the generation of compounds such as H<sub>2</sub>O<sub>2</sub> (CAKMAK, KIRKBY 2008, TEWARI KUMAR et al. 2006). Under conditions of severe deficiency of magnesium, the production of ROS increases, causing



Photo 1. Classical symptoms of magnesium deficiency on grapevine leaves at ripening  
(author: W. Grzebisz; no of photo DSCF0036)

leaf tissue damage. As a consequence, crop plants may suffer due to Mg deficiency and are not able to take up enough nitrogen for building their assimilation area. Secondly, this area is permanently reduced under ROS action. Leaf chlorosis, as a particular symptom of advanced magnesium deficiency, appears first on older leaves, exposed to light, i.e., energy saturated. The classical examples are crop plants exposed to light such as potatoes, sugar beets and especially grapes and sunflower.

## **IMPROVEMENT OF FACTORS LIMITING NITROGEN PRODUCTIVITY BY MEANS OF MAGNESIUM SUPPLY**

The dynamics of nitrogen uptake from soil by crop plants depends on their nutritional status with respect to phosphorus, potassium and magnesium. Transportation of the first two nutrients from any point of the soil body to the root surface is related to their diffusion gradients, which are determined not only by physical properties of a given element but also by soil water content and its temperature. Magnesium ions are transported towards roots in a water transpiration stream (mass flow) and therefore the plant supply is related to magnesium soil concentration. Hence, total and especially available magnesium resources (water soluble) are crucial for crop

plant nutrition with magnesium as well as nitrogen. An analysis of the total soil magnesium resources in Europe ([www.gtk.fi/publ/foregsatlas](http://www.gtk.fi/publ/foregsatlas)) underlines low capacity of soils in Poland with respect to this element. The topsoil total magnesium content ranges from 1 to 2.5 g kg<sup>-1</sup> and for the subsoil it varies from 1 to 3.4 g kg<sup>-1</sup>. For comparison, the total content of magnesium in the topsoil in the Czech Republic ranges from 7.7 to 10.8 g kg<sup>-1</sup>. Hence, the presented data stress a significant depletion of topsoil magnesium resources as compared to subsoil ones, which poses a risk of potential deficiency of available magnesium forms. This hypothesis is fully supported by data periodically published by Regional Agrochemical Stations for whole Poland. According to LIPIŃSKI (2005), more than 50% of Polish soils are poor in soil available magnesium.

Under the production conditions in Poland, any choice of a well-defined magnesium fertilization system should be preceded by evaluation of three factors which determine a potential crop plant magnesium nutritional status. They are as follows: (i) quantitative crop plant requirements, (ii) content of soil available magnesium – magnesium content in the soil solution, (iii) timing and form of applied magnesium fertilizer (HUNDT, KERSCHBERGER 1991). Plant magnesium requirements are crop specific, but as a rule dicotyledonous crops are much more sensitive to magnesium deficiency than cereals. Each plant crop accumulates magnesium progressively in the course of vegetation, reaching top values just before full ripening. It is necessary to point out a close relationship between magnesium and phosphorus uptake patterns (Figure 1). In spite of some resemblances among different types of crop plants, they respond differently to fertilizer magnesium application timing. The expected pattern of the response of crop plants to magnesium supply is a prerequisite for selecting a fertilizer magnesium system. Root and tuber crops are best responsive to soil magnesium level. The growth rate of

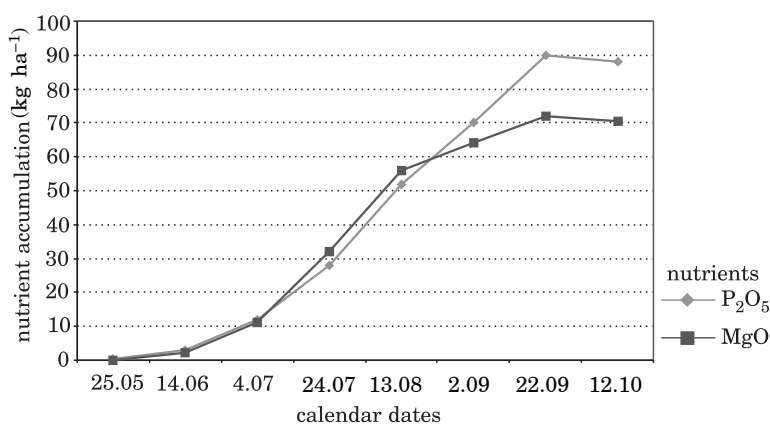


Fig. 1. Magnesium and phosphorus accumulation by sugar beet plantation over the growing season (based on: GRZEBISZ et al. 1998)

beet seedlings required to intercept sufficiently high amount of solar energy at the beginning of the growth season is the main explanation of this type of response (ANDRIEU et al. 1997). Therefore, it should not be surprising that this crop responds to magnesium foliar application performed at early stages of growth but before canopy closing, i.e. the LAI of 3.0 (BARŁÓG, GRZEBISZ 2001). As presented in Figure 2, interactions of soil and foliar magnesium application to sugar beet resulted in a huge increase of fertilizer nitrogen

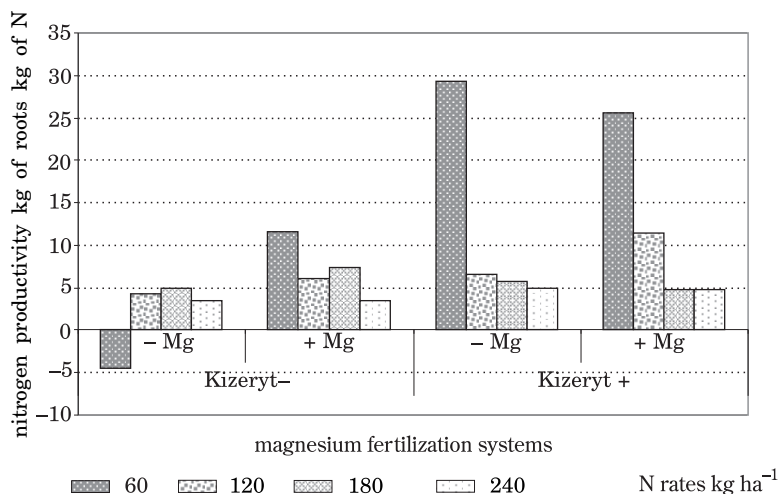


Fig. 2. Partial factor productivity of fertilizer nitrogen index response to systems of magnesium application (based on: GRZEBISZ et al. 2001)

efficiency (one of the factors in fertilizer nitrogen productivity), increasing the final yield of taproots and recoverable sugar. However, the described phenomenon, which is also noticeable in other crops, e.g. maize as presented by SZULC (2010), is most pronounced under the conditions of low nitrogen supply (soil + fertilizer nitrogen). It could therefore be supposed that under low N supply, any extra-added magnesium stimulates two independent processes. The first hypothesis assumes an extended growth of plant roots due to raised activity of auxins caused by low N and easily available magnesium. The second hypothesis relies on a higher rate of nitrate uptake, accelerating the canopy growth rate (ANDRIEU et al. 1997). As a result of a higher total uptake of nitrogen, which takes place at the beginning of the vegetative season, some crops are able to produce higher yield, using the applied fertilizer nitrogen more efficiently. Slightly different responses are observed for cereals, which show an increase of magnesium uptake at ripening. Therefore, this group of crops responds significantly to foliar application of fertilizer magnesium, but carried out just before anthesis (Table 2).



Table 2

Effect of the magnesium rate and its timing on grain yield and nitrogen economy of winter cereals

Species Nitrogen economy features	Control	Tillering BBCH28 Mg rates (kg ha <sup>-1</sup> )		HeadingBBCH55 Mg rates (kg ha <sup>-1</sup> )	
		2.5	5.0	2.5	5.0
Rye					
Yield**, t ha <sup>-1</sup>	4.30	4.50	4.75	4.40	4.75
N in grain, g kg <sup>-1</sup>	12.8 ± 0.4	12.8 ± 0.4	13.8 ± 0.1	13.3 ± 0.1	14.0 ± 0.2
N uptake, kg ha <sup>-1</sup>	89.0	94.7	106.2	98.0	128.5
Unit nitrogen uptake*, kg N t <sup>-1</sup> ****	20.7	21.0	22.4	22.3	27.1
Wheat					
Yield***, t ha <sup>-1</sup>	5.30	5.50	5.75	5.55	5.90
N in grain, g kg <sup>-1</sup>	19.0 ± 1.0	19.2 ± 1.1	19.7 ± 0.8	19.5 ± 0.8	20.0 ± 0.7
N uptake, kg ha <sup>-1</sup>	142.9	149.8	161.6	152.8	168.4
Unit nitrogen uptake*, kg N t <sup>-1</sup> ****	27.0	27.2	28.1	27.5	28.5

\*based on MATŁOSZ (1992), \*\*LSD<sub>0.05</sub> = 0.15, \*\*\*LSD<sub>0.05</sub> = 0.12,

\*\*\*\*grain + respective straw biomass.

The key is the root system, although it can be considered as just a sufficient factor conditioning the uptake of nutrients by crops from the soil body. The size and architecture of the root system are limited by many factors. Soil reaction is one of the main plant production limiting factors (aluminum toxicity) in Poland, where *ca* 60% of arable soils are acid and very acid (LIPIŃSKI 2005b). This level of soil pH is a prerequisite to aluminum bio-toxicity. This phenomenon is the main cause of the reduction of the root system size as it inhibits:

- the division of root cap cells,
- the growth of elongation root zone.

Excess of aluminum in root apoplast, resulting from insufficient supply of calcium, is the main reason for sequentially induced processes such as: (i) synthesis of cellulose – the main component of cell walls, (ii) synthesis of callose (TERAOKA et al. 2002). This organic compound is produced by plant tissues in order to separate healthy part from ill or wounded ones. Allocation of this polysaccharide into the root meristeme of a plant root means the end of its growth, decreasing then the vertical extension of the root system. As a result of these processes, crop plant functions become restricted, with the prevalence of:

- root system size reduction (Figure 3),

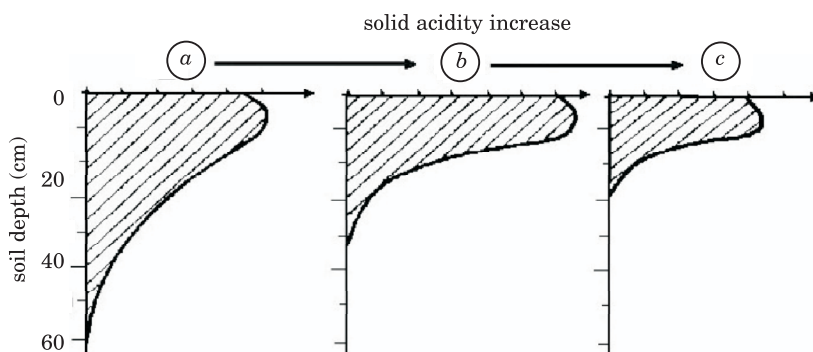


Fig. 3. Size of crop plant root system under growing soil acidification (MARSCHNER 1991):  
*a* – neutral, *b* – acid, *c* – very acid

- increased threat of nitrate leaching,
- increasing amounts of leached calcium and magnesium (SCHWEIGER, AMBERGER, 1979);
- fixation of available phosphorus by iron and aluminum cations.

The main criterion of water and nutrients uptake, including nitrate-nitrogen uptake, by crop plants is the vertical distribution of their roots and the coefficient of diffusion of nutrients in the soil solution. Plants are able to take up mobile nutrients in the zone extending below 20 cm of the root maximal length (MARSCHNER 1991). Therefore, it can be concluded that the reduced root system of crop plants due to aluminum toxicity is not able to take up nutrients moving downwards, such as nitrates or sulfates. Under acid soil conditions, a substantial amount of fertilizer nitrogen is not taken up by crop plants, thus enlarging the pool of residual nitrogen.

Toxic symptoms caused by aluminum emerge as deficiency symptoms typical of phosphorus, calcium and even magnesium, as presented in Photo 2 (KELTJENS, KEZHENG TAN 1993, SILVA et al. 2010). The deficit of available magnesium appears not only in arable soils, but also in forest and perennial grasses, leading to their degradation (PANNATIER et al. 2004). In fertile soils, the content of exchangeable aluminum should not exceed  $36 \text{ mg Al}_{\text{ex}} \text{ kg}^{-1}$  soil (SKUBISZEWSKA, DIATTA 2008), but negative effects of elevating the content of toxic aluminum ions was detected even at a level of  $9 \text{ mg Al}_{\text{ex}} \text{ kg}^{-1}$  soil (DIATTA et al. 2010).

In the light of the above consequences of elevated toxic aluminum levels, a question arises about ameliorative effects of magnesium. So far, scientific reports have been scanty but the main aspects of magnesium presence may be summarised as follows:

- 1) decreasing accumulation of toxic  $\text{Al}^{3+}$  ions by crops in the presence of magnesium ions (KINRAIDE et al. 2004, SKUBISZEWSKA, DIATTA 2008);



Photo 2. Maize plants with visible symptoms of magnesium deficiency – aluminum toxicity induced (author: W. GRZEBISZ, no of photo P1010035)

- 2) a crop well supplied with magnesium excretes to its rhizosphere higher amounts of organic acids and other organic substances exhibiting chelating properties for  $\text{Al}^{3+}$  (YANG et al. 2007);
- 3) a crop well supplied with magnesium excretes to its rhizosphere divalent cations, such  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$ , in turn rising up root apoplast pH above 5.0 supposed to protect a plant root against  $\text{Al}^{3+}$  toxicity (SILVA et al. 2010).

In today's agricultural practice, magnesium fertilizers represent two main chemical compounds:

- carbonates (dolomite, magnesite);
- sulfates (Kieserite, Epsom salt).

Agrochemical results of carbonate application are easily defined owing to their well-known geochemical transformation pathways. This type of magnesium fertilizers allows us to reach basic targets after a single treatment. The first type of fertilizers causes neutralization of toxic aluminum and the second one is associated with to a huge increase of available magnesium content. Magnesium fertilizers based mostly on sulfates reveal a much more complicated geochemical action, because both ions are involved in toxic  $\text{Al}^{3+}$  soil solution control (DIATTA et al. 2010).

Yield increase is a result of the amounts of nitrogen taken up by the cultivated crops and incorporated into yield during the growing season. Therefore, as presented in Table 3, yield increase can be considered as an increased temporary potential of crop canopy to accumulate nitrogen. The final production effect of magnesium application, i.e., increased yields, is a result of the agrochemical action of this nutrient, which enables plants to recover some of residual nitrogen in soil, thus diminishing its active pool.

## FINAL CONCLUSIONS

Owing to its influence on the plant growth and distribution of assimilates, nitrogen is a yield-forming nutrient (RUBIO et al. 2003). However, today's production technologies cannot exceed a certain level of productivity due to a relatively low fertilizer nitrogen recovery and low unit productivity (DOBERMANN, CASSMAN 2002, GRZEBISZ et al. 2009). One of the most limiting factors is the current system of crop fertilization, guided by three main rules of nitrogen application: the right amount, right form and right timing (JANSSEN 1998). In many parts of the world, including Poland, the applied nitrogen rates are generally badly balanced with phosphorus and potassium (GRZEBISZ et al. 2009). The yield-forming potential of magnesium as a nutrient affecting basic biochemical and physiological plant processes including nitrogen economy is not fully exploited. In Poland, plant production as a natural base for food production is carried out under low natural soil fertility conditions, which does not allow farmers to exploit the yielding potential of crops. The main reasons for low yields harvested by farmers are: (i) low soil pH, a prerequisite for aluminum toxicity, and (ii) low total and available magnesium contents. Application of fertilizer magnesium in agriculture should be therefore considered as one of the most important factors ameliorating both groups of the limiting factors, thus improving the efficiency of the applied fertilizer nitrogen. Therefore, it can be concluded that magnesium is an important component in the sustainable system of farmland management, because it ensures sufficiently high-level plant production and environmental sustainability.

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**Katedra Chemii Rolnej i Ochrony Środowiska**  
**ul. Oczapowskiego 8, 10-719 Olsztyn-Kortowo**  
**jadwiga.wierzbowska@uwm.edu.pl**

**dr hab. Katarzyna Gliška-Lewczuk**  
**University of Warmia and Mazury in Olsztyn**  
**Pl. Łódzki 2, 10-759 Olsztyn, Poland**  
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